

PROCEEDINGS



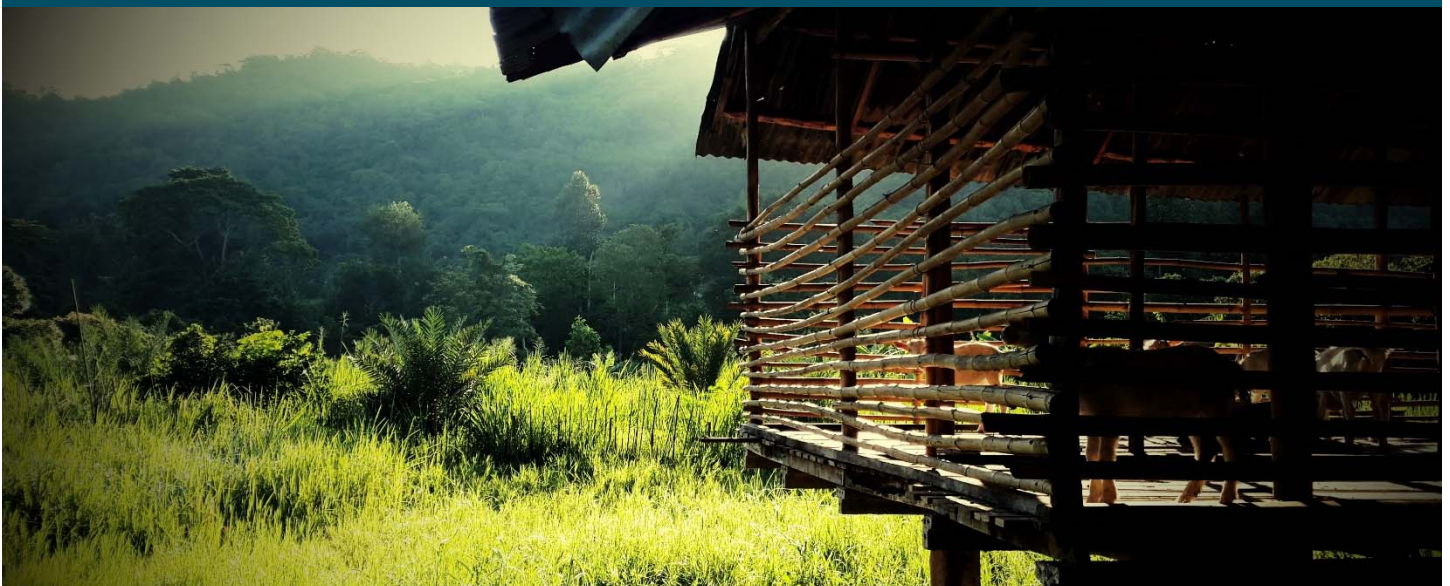
5th

SAADC 2015

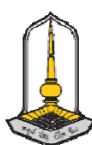
The 5th International Conference on
Sustainable Animal Agriculture for Developing Countries

**“CLIMATE SMART SUSTAINABLE ANIMAL AGRICULTURE FOR FOOD SECURITY
AND LIVELIHOOD IMPROVEMENT IN THE DEVELOPING COUNTRIES”**

October 27-30, 2015, Dusit Thani Pattaya Hotel, THAILAND



Jointly organized by



PROCEEDINGS
of
The 5th International Conference on
Sustainable Animal Agriculture for Developing Countries
(SAADC 2015)
October 27-30, 2015
Dusit Thani Pattaya Hotel, Thailand

Jointly Organized by:



Faculty of Sciences and Liberal Arts, Rajamangala University of Technology Isan



Institute of Agricultural Technology, Suranaree University of Technology



Faculty of Technology, Mahasarakham University



Faculty of Veterinary Medicine, Mahanakorn University of Technology



Faculty of Animal Sciences and Agricultural Technology, Silpakorn University Phetchaburi IT Campus



Faculty of Science and Technology, Nakhon Ratchasima Rajabhat University



Faculty of Technology, Udon Thani Rajabhat University



Tropical Feed Resources Research and Development Center (TROPREC)



Department of Livestock Development
Thailand



The Animal Husbandry Association of Thailand under the Royal Patronage of
H.R.H. Princess Maha Chakri Sirindhorn

Message from the President of RMUTI

Dear Participants,

It is my great honor to welcome all of participants to attend the 5th Sustainable Animal Agriculture for Developing Countries (SAADC) conference which held at the Dusit Pattaya Hotel, Chonburi, Thailand during 27-30 October 2015. It is also 10 years Anniversary of Rajamangala University of Technology Isan (RMUTI), which established depending on Rajamangala University of Technology Act B.E. 2548 (2005). On behalf of RMUTI, I would like to welcome about 350 participants from 40 countries to participate at the conference. The principal objective of SAADC is to provide a venue for animal scientist, agriculturist, farmers and private sectors to build up the relationship and to exchange their experiences.



The 5th SAADC 2015 is organized by seven institutes such as Rajamangala University of Technology Isan (RMUTI); Suranaree University of Technology (SUT); Mahasarakham University (MSU); Silpakorn University (SPU); Mahanakorn University of Technology (MUT); Nakhon Ratchasima Rajabhat University (NRRU) and Udon Thani Rajabhat University (UDRU).

All sponsors are highly appreciated to make the conference more successful. Last but not least, all partners who contributed to this conference are deeply thanks without your fully supports this conference would never be accomplished.

With best wishes,

A handwritten signature in black ink, appearing to be 'V. Limkaisang', written in a cursive style.

Assistant Professor Dr. Viroj Limkaisang

President of RMUTI

27 October 2015

Message from President SAADC International Advisory Committee

Ladies and Gentlemen,

First and foremost, I would like to thank the Organising Committee of the 5th International Conference on Sustainable Animal Agriculture for Developing Countries (SAADC2015) for inviting me to pen a few words in this Souvenir Programme.




I would like to take this opportunity to share with you, especially those who are attending the SAADC series of conferences for the first time that SAADC has grown steadily since the inaugural SAADC2007 organised by Yunnan Agricultural University in Kunming, China. The numbers of participants and countries involved have increased from less than 200 from seven countries in the inaugural conference to more than 300 from 40 countries in this conference. This reflects the relevance of SAADC in providing a platform for animal scientists and producers especially from the developing countries to share experience and network to promote sustainable animal agriculture in our respective countries.

This week we are here again to present our research findings and ideas for promotion of sustainable animal agriculture. I congratulate the Organising Committee for their hard work throughout the last two years to make it possible for us to meet in one of the world renowned beach resorts in Thailand. I would like to thank members of the SAADC2015 International Advisory Committee and the SAADC2015 in-house editors for their input and hard work to support the local organising committee of this conference. Special thank goes to Dr Chris Anderson of the CSIRO Publishing for his help to create the SAADC2015 special issue in the journal of *Animal Production Science* for publication of selected papers presented by the participants of this conference.

Most of all, I thank each and every one of you for your participation in making this conference a great success. I would like to encourage all participants, particularly the younger ones to take this opportunity to make new friends and to create new opportunities to foster cooperation towards promotion and enhancement of sustainable animal agriculture in our respective countries.

Yours sincerely,

A handwritten signature in black ink, appearing to read 'Juan Boo Liang', written in a cursive style.

Professor Dr. Juan Boo Liang

President
SAADC2015 International Advisory Committee
27 October 2015

Message from the Chairman of the 5th SAADC Conference

On behalf of Rajamangala University of Technology Isan (RMUTI), I would like to express my deeply thanks to the SAADC International Advisory Board (IAB) for their agreement to permit RMUTI to organize the 5th SAADC conference together with our co-hosts institutes, these are Suranaree University of Technology (SUT), Mahasarakham University (MSU), Silpakorn University (SPU), Mahanakorn University of Technology (MUT), Nakhon Ratchasima Rajabhat University (NRRU) and Udontani Rajabhat University (UDRU).



The 5th SAADC 2015 consists of scientific session, private sector demonstration, social and cultural activities. The scientific session offers plenary session, invited session, symposium and graduate course. The symposium is an entitled on “Understanding of Biological Product: The role for sustainable Animal Production” by Associate Professor Dr. Kriengsak Poonsuk (K.M.P. BIOTECH CO., LTD). The workshop is an established on “Publishing Your Research Findings in International Journals” by Dr. Thomas J. Schonewille (Utrecht University, The Netherlands). The cultural activities are Thai regional dancing (Fon Ram) with the contributing of Rajamanagala University of Technology Tawan-ook. Field trips are based on two routes: Route I is a “Dairy Buffalo Farm: Runjaun Farm” and Route II is a tropical garden so called “Saun Nongnooch”.

I would like to express my sincerely thanks for the keynote, plenary, invited speakers and participants who had been fully supported to make the conference more success and fruitful.

I deeply appreciate to the International Advisory Board (IAB) and the local organizing committee for their great effort and dedication to make the proceeding in time.

Last but not least, I would like to sincerely thanks to President of RMUTI for his fully supports to make this conference successful.

Wish best wishes.

A handwritten signature in blue ink, appearing to be 'Chalermpon Yuangklang'.

Assistant Professor Dr. Chalermpon Yuangklang

Chairman of the 5th SAADC 2015
Dusit Thani Hotel, Pattaya, Chonburi, THAILAND

Message from Academic Committee Chairman

As the host of the 5th International Conference on Sustainable Animal Agriculture for Developing Countries (SAADC2015), Rajamangala University of Technology Isan do realize the significance of research, innovation and application in terms of international development of economics and society. The SAADC 2015 conference has its objectives to provide a chance for researchers in field of animal science, agriculture and related fields including academicians, researchers, administrators and private sectors both in developing and developed countries to share their own experiences, to develop collaborative networks among institutions and to strengthen research quality of staff and students for sustainable animal agriculture production.



From the number of oral and poster presentations submitted in this conference in Pattaya, I do impress your participation and have confidence that you all are the scientists with very great enthusiasm to solve problems as well as to share valuable information and knowledge for people prosperity.

I would like to particularly thank all guest speakers and participants who make this conference such a valuable collaborative and successful forum. My sincere thanks go to our co-organizing committee form Suranaree University of Technology, Nakhon Ratchasima Rajabhat University, Mahasarakham University, Mahanakorn University of Technology, Silpakorn University, and Udon Thani Rajabhat University. Special thanks to the scientific committee, reviewers and editorial boards for their great contribution to make the conference successfully organized.

I believe all delegates will benefit substantially from the conference through the presentations of expert speakers and exchanges of ideas with one another. I wish you all have most pleasant and most wonderful time in the conference in Pattaya, Thailand and a safe journey home.

A handwritten signature in blue ink, which appears to read "Kraisit Vasupen".

Assistant Professor Dr. Kraisit Vasupen

Chairman of Academic Committee
SAADC 2015

INTERNATIONAL ADVISORY COMMITTEE:

Chairman:

Prof. Dr. Liang Juan Boo (Malaysia)

Committee Members:

Prof. Dr. Peter Wynn (Australia)

Dr. Hiroyuki Konuma (FAORAP)

Prof. Dr. Harinder Makkar (FAO)

Prof. Dr. Long Ruijin (China)

Assist. Prof. Dr. Chalermpon Yuangklang (Thailand)

Prof. Dr. Liang Chou Hsia (Taiwan, ROC)

Prof. Dr. Pietro Celi (Australia)

Prof. Dr. Junichi Takahashi (Japan)

Dr. Vo Thi Lam Thanh (Vietnam)

Assoc. Prof. Dr. Pramote Paengkoum (Thailand)

Dr. Elizabeth Wina (Indonesia)

ORGANIZING COMMITTEE:

Advisors:

Assist. Prof. Dr. Viroj Limkaisang (President of RMUTI)

Prof. Dr. Charan Chantalakhana

Prof. Dr. Metha Wanapat

President of the Animal Husbandry Association of Thailand under the Royal Patronage of H.R.H. Princess Maha Chakri Sirindhorn

Chairperson:

Assist. Prof. Dr. Chalermpon Yuangklang

Vice chairperson:

Assoc. Prof. Dr. Pramote Paengkoum

Secretary:

Dr. Benya Saenmahayak

Fund raising committee:

Chairperson: Assist. Prof. Dr. Sasiphan Wongsuthavas

Secretary: Dr. Siwaporn Paengkoum

Public Relations:

Dr. Weerawat Namanusart

Social Culture:

Assoc. Prof. Dr. Opart Pimpa

Dr. Siwaporn Paengkoum

International Scientific Committee:

Chairman: Assist. Prof. Dr. Kraisit Vasupen (Thailand)

Co-Chairman: Dr. Vincenzo Tufarelli (Italy)

Secretary: Assist. Prof. Dr. Smerjai Bureenok (Thailand)

Members:

Publishing Advisor: Dr. Chris Anderson (Australia)

Assoc. Prof. Dr. Songsak Chumpawadee (Thailand)

Prof. Dr. Pietro Celi (Australia)

Prof. Dr. Ermias Kebreab (USA)

Prof. Dr. Mohd Ariff Omar (Malaysia)

Prof. Dr. Norhani Abdullah (Malaysia)

Prof. Dr. Ariff Omar (Malaysia)

Prof. Dr. Christopher McSweeney (Australia)

Prof. Dr. Peter Daniels (Australia)

Dr. Elizabeth Wina (Indonesia)

Prof. Joaquim Balcells (Spain)

Dr. James Chin (Australia)

Prof. Dr. Thomas J. Schonewille (The Netherlands)

Registration:

Dr. Siwaporn Paengkoum

Dr. Nittaya Pitiwittayakul

Dr. Totsapron Srisaphoomi

Financial/Treasurer:

Mrs. Ekarat Vasupen

Website:

Mr. Ekkachai Sae-jueng

Mr. Ekkalak Chimjarn

Mr. Thanin Rabiabpho

Mrs. Chonlada Chimjarn

Mr. Soravit T.Siriwattana

List of Reviewers:

Prof. Dr. Yangqing Lu
Prof. Dr. Kazuaki Takehara
Prof. Dr. Anton C. Beynen
Prof. Dr. Kawamoto Yasuhiro
Prof. Dr. Wahid Haron
Dr. Jothi Panandam
Dr. Chen Wei Li
Dr. Tomoyuki Suzuki
Dr. Eko Widodo
Dr. Abdoreza Soleimani
Dr. Awis Q. Sazili
Assoc. Prof. Dr. Boonlom Cheva-Isarakul
Assoc. Prof. Dr. Chalong Wachirapakorn
Assoc. Prof. Dr. Songsak Chumpawadee
Assoc. Prof. Dr. Jatuporn Kajaysri
Assist. Prof. Dr. Araya Seubkhamphet
Assist. Prof. Dr. Jamlong Mitchaothai
Assist. Prof. Dr. Sakchai Reunphet
Assist. Prof. Dr. Walaiporn Tonpitak
Assist. Prof. Rachakris Lertpatarakomol
Assist. Prof. Dr. Pattaraporn Poommarin
Assist. Prof. Dr. Skorn Koonawootrittriron
Dr. Wittawat Molee
Dr. Danai Sangthong
Dr. Darsaniya Punyadarsaniya
Dr. Grisada Khumpool
Dr. Pannigan Chaichanasak
Dr. Sunisa Sirimongkolvorakul
Dr. Sunpetch Sophon
Dr. Thuchadaporn Chaikhun-Marcou
Dr. Umaporn Rungroekrit
Mr. Patipan Jaipeng
Mr. Pichai Jirawatthanapong
Mrs. Kornkamon Padiporn
Dr. Khanitta Ruangwittayanusorn
Dr. Atra Chaimongkol
Dr. Daranee Sookying
Dr. Chatchawan Singhapol
Dr. Narin Preyavichyapugdee
Dr. Nittaya Pitiwittayakul
Mr. Pichet Sriboonyoung
Dr. Juan Boo Liang
Prof. Dr. Thomas J. Schonewille
Prof. Dr. Eric A. Decker
Prof. Dr. Pietro Celi
Prof. Dr. Peter Wynn
Dr. Hassan Sadri
Dr. Devika Saddul
Dr. Tan Hui Yin
Dr. Irfan H. Djunaidi
Dr. Mohd Ariff Omar
Dr. Elizabeth Wina
Assoc. Prof. Dr. Surintorn Boonanuntanasarn
Assoc. Prof. Dr. Anut Chantiratikul
Assoc. Prof. Dr. Opart Pimpa
Assoc. Prof. Dr. Pantipa Na Chiangmai
Assoc. Prof. Dr. Pramote Paengkoum
Assist. Prof. Dr. Chalermpon Yuangklang
Assist. Prof. Dr. Kraisit Vasupen
Assist. Prof. Dr. Smerjai Bureenok
Assist. Prof. Dr. Sasiphan Wongsuthavas
Assist. Prof. Dr. Charunee Kasornpikul
Assist. Prof. Dr. Sutisa Khampaka
Assist. Prof. Dr. Chaweng Sarnklong
Dr. Chakrapong Chaikong
Dr. Seksan Wongsiri
Dr. Kanok-on Nugboon
Dr. Benya Saenmahayak
Dr. Totsapron Srisaphoomi
Dr. Weerawat Namanusart
Miss Sirinun Lasrichan
Dr. Pornpan Saenphoom
Dr. Chaowadee Laosutthipong
Dr. Wantanee Polviset
Dr. Peerapot Nitipot
Dr. Huang Xiaodam
Dr. Anan Chaokaur
Dr. Suphavadee Chimtong
Dr. Janjira Sittiya
Dr. Nattiya Chumnanka
Dr. Chomphoonuch Khongla
Dr. Saowaluck Yammeun-art

CONTENTS

Aquaculture		
A-338	Aquaculture in a protected gulf: The case of Amvrakikos (Greece)	1
A-369	Meat and bone meal as an alternative for fish meal in diets for black carp (<i>Mylopharyngodon piceus</i>)	4
A-524	Growth performance of carrageenan-producing seaweeds of <i>Kappaphycus</i> and <i>Eucheuma</i> in Sumbawa	11
A-531	Advantages of environmentally sound poly-eco-aquaculture in fish farms	15
Animal Biotechnology		
AB-125	Study of humoral immune response using IBD Blen® and Vaxxitek HVT-IBD® vaccines even high maternal derived antibody in broiler chickens	17
AB-169	Fertility status of local PO cattle and its crosses with Limousin and Simental bull in Situbondo Regency, East Java, Indonesia	20
AB-319	Reliable monoclonal antibodies for immunodiagnosis of fasciolosis in both animal and human	23
AB-342	Vitrification of mouse embryos: comparison of Cryotop and hemi straw closed system methods	26
AB-343	Impact of complete feed silage from sugar cane waste product on Bali beef cattle performance	29
AB-437	Development of a rapid immunochromatography test for detecting antibodies after anthrax vaccination in cattle: A preliminary study	32
AB-481	The effect of <i>Cinnamomum burmannii</i> extract as an immunomodulator on the increase of GR-1 expressing IFN γ and macrophage	36
AB-555	Genetic variation of MHC Class II DRB3 gene in local goat from South Sulawesi Indonesia	39
Animal Genetic/Breeding		
AG-080	The relationship between longevity and reproductive efficiency in Lori-Bakhtiari ewes	43
AG-095	The Interleukin-8 gene polymorphism and its association with milk production traits in Holstein Cows	46
AG-100	Genetic improvement of production performance for Yorkshire	49
AG-104	Genetic parameters and trends for growth and carcass traits of Landrace in Korea	52
AG-105	Effect of sex and carcass weight on pork belly characteristics of Large White	55
AG-107	Influence of sex and carcass weight on pork belly muscle of Large White	58
AG-130	Production and reproduction characteristics of tegal and magelang ducks	61
AG-149	Breeding potency of Bali cattle as indigenous beef cattle breed in Sumbawa Island Indonesia	64
AG-161	Institutional development on conservation of Madura cattle	67
AG-164	Annual trend of genetic improvement for production performance of three swine breeds	70
AG-182	Phenotypic and chromosome band intensity characters of swamp buffalo in very isolated area of East Java, Indonesia	73
AG-211	The study of reproductive efficiency over the lifetime of Lori-Bakhtiari ewes	76
AG-263	Performance testing of Kamphaeng Saen bulls	80
AG-356	Modelling genomic selection strategies to improve genetic gain in swine breeding programs using ZPLAN+	84
AG-431	Effects of different mating methods on hatchability and embryonic mortality of indigenous chicken eggs	87
AG-530	Proximity of genetic cross Boer goat with Local goat to parent based on gene DNA <i>Capra hircus</i> growth hormone (ChGH)	91

AG-535	Effect of numbers of day of progesterone intra-vaginal device insertion on oestrus rate pregnancy rate and litter size in Thai-native cross breed goats	94
AG-560	Genetic parameter estimation for prolificacy trait of local Ettawah crossbreed goat	97
Animal Nutrition		
AN-007	Effect of microbial mixture on survival of fermented juice of epiphytic lactic acid bacteria (FJLB)	100
AN-012	Effects of storage time on external and internal characteristics of lutein eggs	102
AN-014	Efficacy of probiotic <i>Enterococcus mundtii</i> in dried form in broilers	106
AN-021	Truly absorbed protein in the small intestine content of alfalfa hay harvested at various blooming	110
AN-026	Growth response of purebred Merino and crossbred prime lambs from feed supplemented with canola and flaxseed oils	113
AN-033	Vitamin E and C effect on meat of Muscovy duck	117
AN-036	<i>In vitro</i> nutrients digestibility and fermentation characteristics of king grass combined with concentrate containing mixed microbes	121
AN-037	Periodic changes in chemical composition and <i>in vitro</i> digestibility of Gramineae feed resources in the Philippines	125
AN-038	Chemical composition of forages and browses offered to stall fed goats on smallholder farms in Mauritius	128
AN-047	Feed intake and serum metabolite of goats fed crude glycerin from waste vegetable oil	131
AN-056	Nutritive value of oil palm fronds treated with white rot fungi	135
AN-057	Effect of chemical treated shrimp meal on growth performances of broilers	139
AN-068	Effect of bee pollen as a natural antioxidant on the performance, carcass and antioxidant status of V-line rabbits	142
AN-073	Nutritive value of grower pig ration using local feeds in West Manokwari District, Manokwari	145
AN-078	The effect of chemical processing of soybean meal on <i>in vitro</i> ruminant intestinal available protein	149
AN-086	The effect of Mao pomace supplementation in diets on blood parameters of meat ducks	152
AN-116	Effects of cassava treated lactic acid supplementation on dry matter degradability and rumen fermentation in beef cattle	155
AN-121	Feeding value of dried cashew nut testa in finishing pigs: effects on growth performance, economic return and carcass characteristics	158
AN-124	Effect of oral administration of red ginger extract on performances of hybrid ducks	164
AN-143	The inclusion effect of mangosteen (<i>Garcinia Mangostana</i> L.) peel as feed additive on blood profile and testosterone level of Mojosari male duck	167
AN-144	The effects of quantitative restricted feeding on performances and internal organ weight broiler	170
AN-146	Evaluation of mulberry leave as a functional feed additive of laying hens	174
AN-147	Effects of performance and intestinal morphology by supplementation with a functional feed additive in poultry diet	177
AN-151	Nitrogen balance and carcass quality in broilers given a low-protein diet in the grower period and higher-protein diets in the finisher period	180
AN-153	Effect of a heat stress reducing additive on meat production and quality in Hanwoo heifers	183
AN-154	Effect of fermented plant extracts on enteric methane production in the rumen	186
AN-171	Enhancement of rice straw nutrient value by solid state fermentation with <i>Trichoderma</i>	189
AN-174	Allometric productivity forage and goat foraging behaviour in rangeland at Ebelo Amboasary in Southern of Madagascar	191

AN-177	Supplementation of recombinant lycopene on egg quality and blood characteristics in quail diet	198
AN-184	Effect waste of cabbage on rabbit meat	201
AN-187	Effect of propolis as a natural antioxidant on the performance, carcass and antioxidant status of V-line rabbits	206
AN-193	The effects of phenological stages on forage quality of four rangeland species for the sheep nutrition in Sari plain, Iran	210
AN-202	Effect of ginger root powder supplementation on growth performances and carcass characteristics of broiler chickens as rearing in hot climate	214
AN-205	Fermentation patterns of alfalfa hay and <i>Ulva Fasciata</i> using gas production technique	218
AN-206	To compare intestinal available protein of <i>Ulva Fasciata</i> with alfalfa hay using a new gas technique	221
AN-213	Effect of <i>Curcuma domestica</i> stock solution on layer performance, egg quality, and antioxidant activity	224
AN-215	Effect of yeast fermented fresh cassava root fed beef cattle on digestibility	228
AN-229	Changes in metabolic hydrogen flow on bovine rumen fermentation in response to cashew nut shell liquid	231
AN-231	Utilization of cassava pulp and corncob fermented with <i>Aspergillus niger</i> for animal feed: effect on protein levels	234
AN-256	Nutrient composition and in vitro ruminal degradability of selected local plants used as goat feed in Malaysia	236
AN-269	Effect of crude glycerin supplementation on performance of dairy heifers	240
AN-271	Utilization of fresh cassava with ruzi grass fermented by microbes from Pangkhaomark as diets in swine	244
AN-284	Evaluation of urea-yeast fermented fresh cassava root as protein source by using <i>in vitro</i> gas production technique	247
AN-291	The effects of soybean meal treated with green tea marc crude extract on oxidative status and milk production of dairy cows	251
AN-294	Utilization of <i>Samanea saman</i> pod meal as protein source in diet on voluntary feed intake and digestibility of goats	255
AN-301	Digestibility and nitrogen balance of growing goats fed different <i>Mimosa pigra</i> (L.) meal levels	258
AN-303	Effects of banana stem supplemented on productive performance of finishing pigs	261
AN-305	Effect of cultivation time on populations of yeast and lactic acid bacteria co-cultures in fermented milk	264
AN-309	Effect of dietary supplementation of bioceramic powders on production performance of broiler chickens	268
AN-314	Effect of cutting age and ensilage on chemical composition of Pak Chok1 and King giant napier grasses	272
AN-324	Effects of levels of dried leucaena (<i>Leucaena leucocephala</i>) supplementation on nutritive value of milk in organic dairy cows	276
AN-348	Potential of seaweed as feed to make a healthy broiler meat chicken	280
AN-354	Metabolic imprinting improves rumen development via modulation of epigenetic gene expression including histone modification	283
AN-357	Effect of rumen content on the performance and external body measurement of Sudanese desert kids	287
AN-358	Potassium iodate supplementation in layer drinking water for iodine enriched egg and laying performance	291
AN-367	Efficacy of dry powdered <i>Enterococcus italicus</i> on immune responses to <i>Mycoplasma hyopneumoniae</i> vaccination	294
AN-370	Effect dietary energy concentrate on birth weight and weaning weight of Sudanese Taggar goat kid's	297
AN-398	Feeding standard for Hanwoo cattle: past, present and future	300
AN-400	Estimation of the TDN of spent mushroom substrates used as Hanwoo feed	303

AN-402	Feed intake, nutrient digestibility and rumen parameters in goats as affected by mao (<i>Antidesma thwaitesianum</i> Muell. Arg.) seed meal supplementation	306
AN-410	Effects of spineless cactus feeding on milk production, milk quality and antioxidant capacity in dairy goat	309
AN-416	Effects of wet soya milk waste supplementation on feed intake and growth performance of goats fed corn stubble silage	316
AN-421	Effect of high choline levels supplementation on phosphatidylcholine concentration in egg yolk of laying hen	319
AN-429	Effect of coconut oil supplementation on meal pattern, feed intake, and milk yield in early lactating crossbred dairy goat	324
AN-432	The effect of the use of cassava leaves silage in concentrate on goat performance	331
AN-434	Effects of CNCPS fraction-enriched protein feeds on ruminal fermentation in Holstein Steers fed TMR containing low protein as a basal	334
AN-440	Effects of dietary pomegranate by-products on performance, immunity, intestinal microbiology and odorous gas emissions from excreta in broilers	338
AN-442	Effects of synbiotic supplemented in broiler diet on carcass and meat quality	341
AN-444	Feed intake, digestibility, nitrogen retention and daily weight gain of steers fed on sugarcane stalk based complete diet silage	344
AN-445	Effect of nitrate addition to cassava chip on In vitro gas production	348
AN-447	Effects of TDN value in TMR on ruminal fermentation characteristics and effective dry matter degradability by rumen microbes	351
AN-451	Utilization of Paprika (<i>Capcicum annum</i>) by product for ruminants feeding	354
AN-452	Performance of Lohi sheep and Beetal goats fed various fodders	357
AN-453	Intake digestibility of summer fodders fed sheep and goats	360
AN-458	Feed intake and nutrient digestibility in goats of silages prepared from Stylo legume (<i>Stylosanthes guianensis</i> CIAT184) treated with dried mao pomaces (DMP) and lactic acid bacteria	363
AN-461	Effect of combination acidifiers-garlic-Phyllanthus niruri L. powder and encapsulated form in feed on production performance and egg quality of laying hens	366
AN-464	Characteristics of fermentation kinetics and digestibility of PUFA saponification and aldehyd protected as cattle feed supplement by in-vivo	371
AN-470	Effect of usage probiotics powder as feed additive on the eggs quality of laying hens	374
AN-475	Supplementation of different sources of nitrogen and its effects on rumen microbial biomass and <i>in vitro</i> feed degradability parameters	379
AN-480	Rumen adaptation for urea on feed intake, nutrient digestibility and microbial protein syntheses of swamp buffaloes	383
AN-484	Study on rumen ecology of swamp buffaloes as affected by urea as protein source in concentrate mixture fed on rice straw based	387
AN-485	Effect of <i>Flemingia macrophylla</i> (FLM) as a protein source on rumen fermentation and microbial population in dairy steers	391
AN-486	Effect of rice straw treat menton feed intake and nutrient digestibility in swamp buffaloes	394
AN-491	Efficacy of endogenous emulsifier in broilers diets on growth performance	398
AN-494	Effects of fattening length and energy levels on meat characteristics of Iranian native lamb	402
AN-496	Comparative efficacy of herbal methionine and synthetic DL-Methionine on performance in laying hen	405
AN-498	In Situ evaluation of heat treated vegetable protein sources	409
AN-499	Efficacy of probiotics (Sanizyme) on performance in broiler diets	413
AN-501	Efficacy of probiotics (Sanizyme) on performance and digestibility in weaning piglets diets	417

AN-504	Effects of condensed tannins of some tropical plants on ruminal gas production in vitro	421
AN-505	Dietary fat sources on growth performance and body composition in broiler chickens	425
AN-506	Influence of rice straw treated on rumen fermentation and microbial population in swamp buffaloes	428
AN-512	A study on nutrient intake and digestibility, rumen environment and nitrogen retention of sheep fed different levels of ensiled water hyacinth in diets	431
AN-513	Effect of dried cassava chips in growing rabbit diets on meat performance and economic returns	434
AN-517	Effects of wet soya milk waste supplementation on feed intake and growth performance of goats fed corn stubble silage	438
AN-519	Productive performance and production cost of different cross bred meat goats fed high levels of OPL fermented TMR	441
AN-522	The effect of <i>S. rarak</i> microparticles on blood profile and productivity of broiler chickens raised on litter system inoculated with <i>E. tenella</i>	445
AN-532	Effect of different Chinese herbs on antioxidant capacity and immune function in Sansui laying duck	448
AN-534	Enhancing the nutritional value of soybean through supplementation with new-generation feed enzymes for poultry –Review	452
AN-539	The effects of feed fermented by Azotobacter microbes culture on milk production and its feed efficiency at dairy cattle	456
AN-552	Evaluation of nutrient digestibility of mixed cassava pulp and Napier Pakchong grass for use as an alternative feedstuff in laying hens	459
AN-564	Effect of extraction methods for krabok oil on milk production, compositions and fatty acids in milk of dairy cows	462
AN-569	Supplementation of different sources of nitrogen and its effects on rumen microbial biomass and <i>in vitro</i> feed degradability parameters	466
Animal Physiology		
AP-008	Effects of lycopene on hepatic metabolic and immune-related gene expressions in chickens	467
AP-173	The effect of different altitude to adaptability, feed consumption and weight gain's lactating ettawa's cross bred	470
AP-435	Differential level of plasma nesfatin-1, ghrelin and leptin for onset of puberty in Murrah buffalo heifers	473
AP-472	The effect of teat seal on milk microorganism number in postpartum cows	477
AP-526	Growth performances of PO cattle and its crossbred with European cattle (POE) maintained in different environmental conditions	480
AP-536	Morphological studies of thyroid gland of Saidi rams fed mannan oligosaccharide supplemented diet	483
AP-538	The study of extracellular hsp70 & physiological parameters, the effect of feeding improvement of the Ongole crossbred & it's crossing breed	486
Animal Reproduction		
AR-023	Breeding soundness evaluation in Garut ram	489
AR-024	Individual variation on the success of garut ram frozen semen production	492
AR-066	Efficiency of difference doses of pregnant mare's serum gonadotropin on superovulations in meat goats	496
AR-082	Noni effect on goat sperm motility after cooling	499
AR-119	Productive and reproductive performance of indigenous Lime and Parkote buffaloes in the Western hills of Nepal	502
AR-120	The Oocyte parthenogenesis stimulation by protein extract of goat spermatozoa	505
AR-131	Supplementation of vascular endothelial growth factor (VEGF) increases the maturation of porcine COCs derived from small follicles	508
AR-194	Supplementation of L-Carnitine on matured goat oocyte <i>in vitro</i>	511
AR-254	The acceptability of reduced sperm concentration in frozen buffalo semen	514

AR-255	Reproductive performance of cattle and buffaloes treated with prostaglandin F2 α and gonadotropin releasing hormone in Thailand and Philippines	518
AR-310	Morphometric dimensions of the spermatozoa in Thai native boar depend on the ejaculates	522
AR-383	Effects of addition juice date palm to the extender on the percentage of live and motility of frozen thawed bull spermatozoa	526
Basic Veterinary Science		
BVS-148	Antibacterial activity of wood vinegar against <i>Salmonella</i> Enteritidis and <i>Salmonella</i> Typhimurium	529
BVS-207	Virucidal efficacy of <i>Clinacanthus nutans</i> and <i>Houttuynia cordata</i> extract against virulent Newcastle Disease Virus	532
BVS-313	Ciliary activity and life span of swine precision-cut lung slices: Comparison between changed and unchanged medium	536
BVS-331	Factors influencing the incidence of Babesiosis in sheep of district Toba Tek Singh, Pakistan	541
BVS-340	The <i>in vitro</i> antibacterial activity of <i>Muntingia calabura</i> against <i>Staphylococcus aureus</i> and <i>Streptococcus agalactiae</i>	546
BVS-399	Study of the optimal condition of RNA in situ hybridization for HER-2/neu in canine mammary gland	549
Food Science and Technology		
FST-267	Process development of cooking wine from whey of buffalo milk	553
FST-302	Study of differential protein composition of raw milk and processed milk by using SDS-PAGE and Native-PAGE	556
FST-476	Development and production of Shrikh and by utilization of pomegranate fruit	559
FST-483	Physico-chemical quality of kefir of etawah crossbred goat milk	563
FST-551	Histological liver of mice (<i>Mus musculus</i>) consumption collagen extract of broiler's bone in South Sulawesi, Indonesia	566
FST-578	Effect of jelly addition on kefir quality	574
Feed Technology		
FT-099	Crude triterpenoid, phenolic compounds and enzyme activities of fermented soybean hull by <i>Antrodia cinnamomea</i>	577
FT-111	Effect of performance and intestinal characteristics on supplemented with protease in the broiler diet	579
FT-299	The levels of TBARs in pellet fish feed mixed with different astaxanthin levels and packaging methods	582
FT-355	Standardized total tract digestibility of phosphorus of various meal diets as protein source in growing-finishing pigs	587
FT-406	Application of feed technology of quail (<i>Coturnix-coturnix Japonica</i>) using waste of Skipjack (<i>Katsuwonus pelamis</i>)	592
FT-430	Quality of protein concentrate from <i>Jatropha curcas</i> seed cake produced by chemical and biological processing	595
FT-544	Serum testosterone, testes size and semen quality of rams fed sugarcane bagasse treated with urea or pronifer	600
Livestock and Environment		
LE-375	Effects of storage on pathogenic bacteria content of layer manure extract	604
LE-408	Effect of heat stress environment on the blood parameters and behavior pattern in Korean native calves	608
LE-460	Study of local feeds potency for pig farming development in Manokwari, West Papua, Indonesia	612
LE-510	A response of <i>in vitro</i> and <i>in vivo</i> methane production, nutrient digestibility and rumen parameters of sheep by Cat fish oil (CFO) supplementation	615
LE-548	Water footprint of milk production in Thailand	618
LE-550	Biogas unit, an alternative solution for reducing green house gas effect of animal waste	621

Livestock Farming System		
LF-022	Development of farmer champions and their role in progressing smallholder beef production in Vietnam	624
LF-072	The assessment of cattle and palm oil plant integration system in West Sumatera, Indonesia	628
LF-162	Characterization of native pig raisers and their current production systems in the integrated sweet potato –native pig production system in Baliem valley, Jayawijaya regency, Papua province, Indonesia	634
LF-218	Smallholder identified constraints to adoption of new forage options in South Central Coast Vietnam	638
LF-220	Studies on socio-economical profile of the dairy farmers in Latur district of Maharashtra state	645
LF-234	Economic impact of spatial development on goat farming in Banjarnegara district Indonesia	644
LF-349	Pasture management and supplemented feed enhanced the performance of farmed buffaloes in Sabah, Malaysia	649
LF-361	Developing technology and husbandry skills required for efficient animal production in villages in the highlands of Papua Indonesia	654
LF-378	Partnership in broiler farm closed house system (case study at Tuban, East Java, Indonesia)	657
LF-381	Moving families from subsistence animal production to small commercial production using a participatory approach with a multidisciplinary team	660
LF-389	Development of local cattle with sustainable in North Sulawesi	663
LF-393	Analysis of the resource potential of the coconut crop-cattle in the district of East Likupang	667
LF-404	Utilization of cattle waste as compost fertilizer	671
LF-465	The Study of nutritive value of plant for goat in Pattani province of Thailand	674
Livestock Management		
LM-044	Production performance of laying hen housing on litter system with different temperature	677
LM-046	Use of agricultural by-product in pig ration to reduce feed cost in Manokwari Regency, West Papua Province, Indonesia	680
LM-079	Evaluation of goat milk quality to support dairy goat development	683
LM-155	Insight into broiler development in East Java	687
LM-203	Association among fat, protein, lactose and total solid of milk produced by farmers in central part of Thailand	690
LM-215	Conventional and deep - litter pig production system: income over feed cost of three breed cross fattening pigs	693
LM-306	Application of PCR technique to detect <i>Staphylococcus aureus</i> that causes mastitis in dairy cows	696
LM-345	Enhancing goat farm performance thru the farmer livestock school –goat enterprise management modality	699
LM-443	Constraints to improved productivity of smallholder cow-calf systems in South Central Coast Vietnam – insights from recent surveys	703
LM-553	Efforts to increase production of cow's milk through the cooperation empowerment in Sinjai Regency	707
LM-589	Why poultry welfare in Kuwait is an obstacle to trade?	711
Meat Science		
M-045	Nutrition, fatty acid and cholesterol content of Garut lamb meat at different ages fed with diet containing mungbean sprouts waste	715
M-163	Some functional properties of beef liver protein concentrates	718
M-245	Meat quality assessment in goats subjected to conscious halal slaughter and slaughter following minimal anaesthesia	722
M-270	Effects of dietary blend of canola oil and palm oil on fatty acid composition, color and antioxidant profile of <i>biceps femoris</i> muscle in goats	726

M-298	Meat quality of crossbred fattening pigs sired by Pakchong 5 boars and commercial boars	733
M-412	Comparison muscle fiber size, sarcomere length and tenderness between chicken and duck meat	737
M-413	Effect of immunocastration on myosin heavy chain isoform expression	741
M-441	Meat composition, fatty acid profile and oxidative stability of meat from broilers supplemented with pomegranate (<i>Punica granatum</i> L.) by-products	744
M-448	Effect of dried fermented juice of epiphytic lactic acid bacteria on broiler meat quality	747
M-459	Prediction of meat and carcass quality traits by real-time Ultrasound in swine	751
M-489	Drip and cooking loss of longissimus dorsi muscle under different levels of energy and protein in Iranian kid native breed	754
Others		
O-084	Microbiological and chemical characteristics of probiotic goat cheese with mixed cultures of <i>L. rhamnosus</i> and <i>L. plantarum</i>	758
O-092	Production elasticity of broiler farming in blitar regency, East Java, Indonesia	762
O-141	The differences of milk density and fat content in Tawang Argo Village compared with Indonesian National Standard	766
O-196	Marketing analysis of broiler farming system on partnership scheme in Ponorogo Regency, East Java Province, Indonesia	769
O-204	Physical properties and microstructure of dangke pipening, a traditional cheese of Enrekang Sulawesi Indonesia	772
O-221	Studies in urban geography of latur city of maharashtra state of India with special reference to demographic features	777
O-252	Body growth of Thai wild boar (<i>Sus scrofa jubatus</i>) within a yearling age in a deep litter pig production system	780
O-266	Consumer attitude toward meat consumption in Ghana	783
O-352	Effect of herbal immunostimulant formulation with Newcastle disease oral pellet vaccine on immune response in native chicken	787
O-377	Effects of probiotics supplementation on nutrient digestability in captive asiatic elephants in temples of Tamilnadu in India	790
O-384	A retrospective investigation of co-endoparasitism of dog and cat patients in Nongchok subprovince, Thailand	795
O-463	Quality of chicken eggs in the flea market, KamphaengSaen district, NakhonPathom province, Thailand	780
O-477	Effect of carbon and nitrogen sources on growth of <i>Alternaria solani</i> (Ell. & Mart.) Causing early blight of Tomato	803
Veterinary Public Health		
VPH-249	Prevalence of parasite infection of puppy dogs to 2 months under animal hospital institute, Udon Thani Rajabhat University	806
INVITED PAPERS		
Invited speaker	Is small animal farming system of low technology?	809
Invited speaker	Family poultry farming system and their characteristics in developing countries	813
Invited speaker	Two types of grazing system under tropical pasture in South western islands of Japan	819
Invited speaker	Value added technology for cattle hide quality improvement in sustainable animal production	824
Invited speaker	Human behaviours as determinants of animal disease: meeting the challenge of a complex mix of changing production systems	845
Invited speaker	Challenges for sustainability of beef cattle production in Thailand	848
Invited speaker	Capacity building and services to assist local farmers to improve aquaculture management in Vietnam	852

Aquaculture in a protected gulf: The case of Amvrakikos (Greece)

Perdikaris¹, C., E. Gouva², A. Chantzaropoulos³, A. Chatzopoulos², I. Skoufos² & I. Paschos²

¹Department of Fisheries, Region of Epirus, Igoumenitsa, 46100, Greece, ²Department of Agricultural Technology, Technological Educational Institute of Epirus, Arta, 47100, Greece, ³Municipal Hatchery of Lake Pamvotis, Ioannina, 45500, Greece

Aquaculture sector profile

A. Marine aquaculture

The established finfish cage farms (Figure 1) in the Amvrakikos Gulf are 23, producing mainly sea bass and sea bream and minor volumes of pagrus, snapsnout sea bream and meagre (Anonymous, 2012). The majority of the final product (over 90%) which is estimated to approximately 7,000 tons annually is exported to the western European markets, of which 80-90% to the Italian market. The estimated overall income from fish farming exceeds €30,000,000 in annual base, which is an important contribution to the regional fisheries balance. The gulf's 13 shellfish culture farms mostly produce Mediterranean mussels with annual production capacity of over 3,000 tones, which are also exported predominantly to the Italian market. However, shellfish production capacity is underutilized as it is seriously affected by the loggerhead sea turtle preying on mussels and by eutrophication. Finally, two marine hatcheries are established in the area producing annually 50,000,000-60,000,000 fish fry.



Figure 1. Typical marine finfish farm with rectangular and circular floating cages in the Amvrakikos Gulf.

B. Lagoons

The rivers Louros and Arachthos which are flowing into the northern part of the gulf have created a series of lagoons (Rodia, Tsoukalio, Logarou, Tsopeli, Mazoma) (Figure 2). These lagoons are organized in the Tsoukalio complex over an area of 6000 ha and the Logarou complex over an area of 4000 ha (Mpasiouli, 2010). These lagoons are managed by cooperatives of professional fishermen and contribute most of the fisheries production of the gulf (i.e. mainly in seabream, various species of mullets and salted roe, seabass and eel).

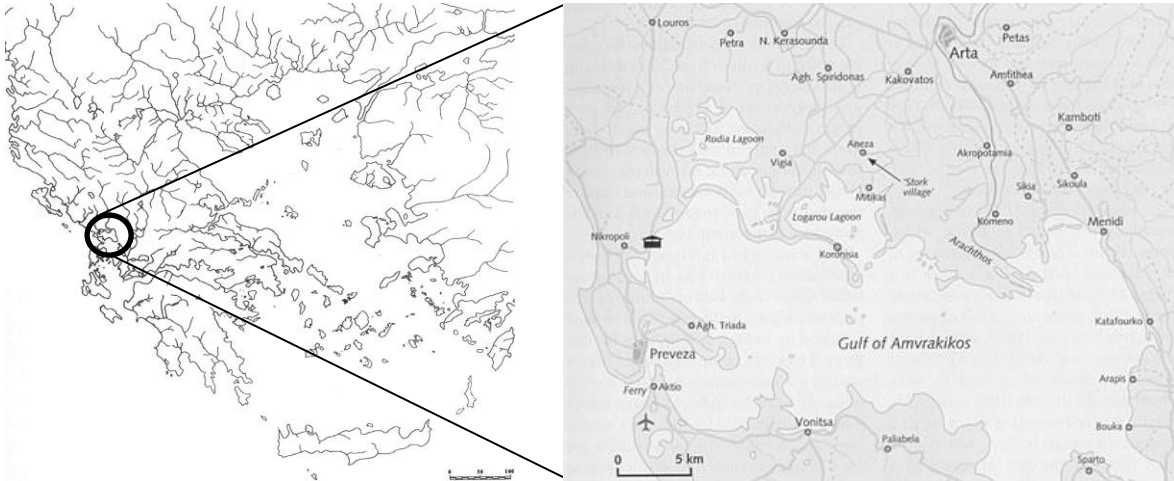


Figure 2. Map of Greece with and the Amvrakikos Gulf on the enlarged section.

Fisheries and processing sector profile

The overall number of fishing vessels is approximately 460 (Anonymous, 2012), most of which are small motorized boats. There are three major commercial harbours and seven small fishing vessel refuges. Shrimp (Figure 3a) annual landings vary annually from 50 to 140 tones, sardine from 100 to 300 tones, shellfish from 10 to 50 tones, cuttlefish from 50 to 120 tones, while striped mullet from 20 to 130 tones. All the above products are highly appreciated as top-quality delicacies of high nutritional value; they are closely related to the local cousin and highly valued by the tourists. Probably the most valuable product in the area is the salted mullet-roe (Figure 3b) with annual production of 350-600 kg. Concerning the processing facilities in the area, there are 11 packaging plants, four processing plants for smoked products, one processing plant for frozen products and one shellfish depuration centre (Anonymous, 2012).



Figure 3. Delicatessen products from the Amvrakikos gulf: (a) local shrimp (*Melicertus kerathurus*) and (b) salted grey mullet (*Mugil cephalus*) roe.

Constrains and proposals

In a nut-shell, the main problems/constrains for further growth of the aquaculture and fisheries sectors in the Amvrakikos Gulf are the following:

- pollution (agriculture chemicals, domestic sewage and inflows from livestock units)
- overfishing, illegal fishing and ill-practised fishing activities (Conides, 2001)
- formation of an anoxic zone below the depth of 35 m (N.C.M.R., 1989)
- fresh water flow reduction and irregular inflows (water management for upstream hydro-electric power production and irrigation)
- increase of mean water temperature
- problems in the implementation of spatial planning in various economic sectors

Therefore, sensible directions for future sustainable production and growth of the aquaculture and fisheries sectors in the gulf could be the:

- continuous environmental monitoring (e.g. pollution, endangered species)
- implementation of effective fisheries assessment and socio-economic studies
- establishment of fisheries and aquaculture production database
- strict implementation of fisheries laws through improved surveillance
- promotion of organic aquaculture and fishing-based tourism initiatives
- promotion of applied research and sensitization of the local communities on the natural, economic and cultural value of the area.

References

- Anonymous, 2012. 'Basket' of agricultural products. Region of Epirus, Greece.
- Mpasiouli, V., 2010. The coastal zone of Amvrakikos Gulf: Assessment of the ecological status and methods of integrated management. M.Sc. thesis, Departments of Veterinary Medicine, University of Thessaly and Aquaculture & Fisheries, Technological Educational Institute of Epirus, Greece.
- Conides, A., 2001. Study of the current state of fishery of the native prawn *Penaeus (Melicertus) kerathurus* population in north Mediterranean. Final Report, Project 037/98, DG XIV-Fisheries. pp. 349.
- ET.AN.AM., 1999. Monitoring of surface waters of Amvrakikos Gulf, 1997-1999. Technical Report, ET.AN.AM. SA, Preveza, June 1999.
- N.C.M.R. 1989 Oceanographic study of Amvrakikos Gulf. Ministry of Environment, Greece.

Meat and bone meal as an alternative for fish meal in diets for black carp (*Mylopharyngodon piceus*)

Tran Thi Nang Thu* & Tran Quang Hung

Vietnam National University of Agriculture, Hanoi, Vietnam

Abstract

A growth experiment was conducted to determine growth performance, feed utilization of black carp, *Mylopharyngodon piceus* fed three isonitrogenous (35%) and isolipidic (10%) diets, adding meat and bone meal (MBM) levels of 5, 10, and 15%, (abbreviated MBM5, MBM10, and MBM15, respectively) as replacement for fish meal (FM). Each diet was randomly fed to three replicate groups of homogenous black carp (initial average weight of 8.43 ± 0.21 g), held in 500L composite tanks with 10 fish/tank. Fish were fed by hand to apparent satiation twice daily at 08:00 h and 15:00 h for 45 days. Fish fed diets, MBM5, MBM10 and MBM15 showed no significant difference in growth and feed utilization parameters ($P > 0.05$). The findings propose that FM in black carp diets can be replaced by MBM without negative effects on growth performance, feed utilization.

Keywords: black carp, meat bone meal, fish meal, growth performance, feed utilization

*Corresponding author: trannangthu@vnua.edu.vn

Introduction

Black carp (*Mylopharyngodon piceus*) is an important farmed fish species in term of economic and nutritional value in China and Northern of Vietnam (Leng & Wang, 2003; Van et al., 2010). In the former, *M. piceus* ranks the fourth most famous farmed fish species and has been cultured for over 2,000 years in traditional farming ponds. The annual cultured production of black carp reaches almost 380,000 tons (Hu et al., 2014). In the latter, black carp has been widely farming in Northern provinces, with the average net profit was 54.2 million VND/ha/year (2,555 USD) and 68.0 million VND/ha/year (3,205 USD) in poly-culture and mono-culture system, respectively (Phuong et al., 2009), and up to 464.0 million VND/ha/year (21,871 USD) for intensive system (Van et al., 2010).

Being carnivorous feeding habits, black carp requires high dietary protein levels ranging from 35% to 40% (Leng & Wang, 2003; Van & Thu, 2013; Hu et al., 2014). Phuong et al. (2009) and Van et al. (2010) reported that black carp farmed in Northern provinces of Vietnam are mainly fed with commercial feed sources which are dedicated for other farmed fish species rather than black carp. It is well known that these feeds largely depend on fish meal (FM), the major dietary protein source, commonly accounting for 20 - 60% of aquafeeds (Watanabe, 2002; Glencross et al., 2007; FAO, 2012). Recently, global aquafeed price has undergone a sharp increase by 73% since 2005, driven by rising price of fish meal (FM) (FAO, 2012). Therefore, in order to reduce feed costs and the use of FM in aquafeeds, more extensive use of alternative feed ingredients is needed (Glencross et al., 2007; Hardy, 2010; Burr et al., 2012).

Meat and bone meal (MBM) is considered as a potential alternative ingredient over other plant proteins due to its economically rendered and available animal protein sources (Ai et al., 2006). The successful use of MBM as replacement for FM in diets has been reported in hybrid

striped bass, *Morone chrysops* x *M. saxutilis* (Bharadwaj et al., 2002), gibel carp, *Carassius auratus gibelio* (Xue & Cui, 2001; Yang et al., 2004), African catfish, *Clarias gariepinus* (Goda et al., 2007), sutchi Catfish, *Pangasius hypophthalmus* (Kader et al., 2011) without major adverse effects on growth. However, results have been less encouraging on some freshwater fish, Nile tilapia, common carp and rainbow trout (Xue & Cui, 2001). On the other hand, recently research of Hung et al. (2015a) showed that nutrients, dry matter, crude protein, crude lipid, ash and gross energy of MBM are well digested by black carp, an freshwater fish species, with apparent digestibility values ranging 57 - 80% (Hung et al., 2015a). In addition, study on hybrid striped bass and Sutchi catfish confirmed that the increased MBM inclusion could reduce cost of fish feed by 17 - 20% (Bharadwaj et al., 2002; Kader et al., 2011).

Therefore, the utilization of MBM in diets for black carp could be a promising way to produce least cost feeds and reduce the use of fish meal. The aim of present study was to evaluate growth performance and feed utilization of black carp fed diets where FM was replaced by MBM.

Methods and Materials

Diets preparation

Table 1. Composition and chemical analysis of three experimental diets (% dry matter).

Ingredients	Diets 1 (MBM5)	Diet 2 (MBM10)	Diet 3 (MBM15)
Fish meal	36	32	28
Meat bone meal	5	10	15
Hipro70 ^a	3	3	3
Soybean meal	15	15	15
Wheat gluten	3	3	3
Cassava powder	13.41	12.41	11.41
Maize meal	12	12	12
Fish oil	10	10	10
Vitamin & Mineral premix ^b	2	2	2
Antifungal	0.07	0.07	0.07
Antioxidant ^c	0.02	0.02	0.02
Binder	0.5	0.5	0.5
<i>Proximate analyses</i>			
Dry matter	96.46	96.71	95.89
Crude protein	35.27	35.08	35.64
Crude lipid	10.1	10.8	10.2

^aPurchased from RT Chemtronics GmbH, Germany

^bProduct name: Customix 5199, purchased from Bayer Vietnam.

^cAntioxidant (mg/g): butylated hydroxytoluene, 12.5.

Three iso-nitrogenous (crude protein, 35%) and iso-lipidic (crude lipid, 10%) experimental diets were formulated to satisfy nutrient requirements of black carp, in which the added levels of MBM in three experimental diets were 5, 10, and 15 (% in DM) (abbreviated MBM5, MBM10, and MBM15). The chemical compositions of the experimental diets are presented in Table 1. Nutrient contents of protein and lipid in experimental diets were within the range required for normal growth of black carp (*M. piceus*) (Leng & Wang, 2003; Van & Thu, 2013; Hu et al., 2014).

Fish and fish culture

The experiment was conducted at the Laboratory of Aquaculture, Vietnam National University of Agriculture, Vietnam. Black carp were obtained from Research Institute for Aquaculture No.1, BacNinh province, Vietnam and were acclimated to the experimental conditions for one month. During this stage, fish were fed with commercial feed to apparent satiety. At the start of the experiment, a total of 90 fish (initial mean weight, 8.43 ± 0.21 g) were randomly held into 500L composite tanks. Each experimental diet was fed to three trial tanks. In all nine tanks, water was flowed at rate of 2L/min and equally aerated 24h/day. Water temperature, dissolved oxygen and pH were checked daily and ranged from 22.5 to 30.1°C (mean temperature, 26.3°C), 4.9 - 6.3 mg/L, and 6.9 - 7.9, respectively. Reportedly, the closed re-circulating culture system used in this experiment was capable of maintaining suitable water quality parameters for experimental fish (Wheaton et al., 1994).

Fish were carefully fed to visible satiety, twice daily at 08:00 h and 15:00 h for 45 days.

Calculations

The following variables were calculated:

$$\text{Survival rate (\%)} = 100 \times \frac{\text{TFf}}{\text{TFi}}$$

$$\text{Specific growth rate (SGR, \%)} = 100 \times \frac{\text{Ln(Wf)} - \text{Ln(Wi)}}{\text{T}}$$

$$\text{Weight gain (WG, \%)} = (\text{Wf} - \text{Wi}) \times 100/\text{Wi}$$

$$\text{Daily weight gain (DWG, g/fish/day)} = \frac{\text{Wf} - \text{Wi}}{\text{T}}$$

$$\text{Protein efficiency ratio (PER)} = \frac{\text{Wet weight gain (g)}}{\text{Total protein intake (g)}}$$

$$\text{Protein intake (g/fish/day)} = \frac{\text{Feed intake (g)}}{\text{Percent protein in diet}}$$

$$\text{Total feed intake per fish (FI, g/fish)} = \frac{\text{Total feed intake (g)}}{\text{Number of fish}}$$

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Total feed intake (g)}}{\text{Total wet weight gain (g)}}$$

Where:

TFfis total number of fish at terminal

TFiis total number of fish at initial

Wfand Wiare the average final weight and the average initial weight, respectively

T is the experimental period (day)

Chemical analyses

Chemical composition of diets samples, FM and MBM were analyzed according to AOAC (1995) methods.

Statistical analyses

All data were analyzed by one-way analysis of variance (ANOVA) with Tukey's post hoc test for individual comparisons. A significance level of 0.05 was used for all comparisons.

Results and discussions

FM and MBM were obtained from commercial sources in Vietnam and were analyzed for chemical composition at Central Laboratory, Faculty of Animal Science, Vietnam National University of Agriculture, Vietnam. The data on nutrient composition of these ingredients was presented in Table 2.

Table 2. Chemical composition (%DM) and gross energy (Kcal/kg DM) content of FM and MBM.

	FM	MBM
Dry matter	91.00	91.70
Crude protein	60.10	49.10
Crude lipid	8.73	9.27
Ash	25.70	32.60
Gross energy	4,360	3,850

During the experimental period, growth was not affected significantly among all dietary treatments, while feed utilization parameters were similar for all treatments ($P > 0.05$) (Table 3). In particular, final body weight, weight gain (WG), daily weight gain (DWG), specific growth rate (SGR), protein efficiency ratio (PER), feed intake (FI) and protein intake tended to decrease with increasing dietary MBM. Survival rate over the experimental period were very high, at above 96% for all groups. These results indicated that 20% of FM could be replaced by MBM without significant effects on growth and feed utilization of black carp.

Table 3. Growth, feed utilization and survival rate of black carp (*M. piceus*) fed the test diets.

	MBM5	MBM10	MBM15
Initial BW (g)	8.40 ± 0.07	8.50 ± 0.04	8.40 ± 0.26
Final BW (g)	11.17 ± 0.05	11.00 ± 0.24	10.77 ± 0.26
WG (%)	33.07 ± 1.45	29.30 ± 3.28	28.37 ± 0.47
DWG (g)	0.061 ± 0.002	0.055 ± 0.005	0.053 ± 0.001
SGR (%)	0.63 ± 0.02	0.57 ± 0.06	0.56 ± 0.01
FCR	3.35 ± 0.10	3.94 ± 0.32	3.98 ± 0.06
PER	0.84 ± 0.02	0.73 ± 0.06	0.73 ± 0.12
PI (g/fish/day)	0.073 ± 0.001	0.075 ± 0.002	0.073 ± 0.001
FI (g/fish/45days)	9.28 ± 0.09	9.64 ± 0.20	9.45 ± 0.02
SR (%)	100	96.67 ± 3.33	100

Values (Mean ± SE) are mean of triplicate.

BW: body weight; WG: weight gain; DWG: daily weight gain; SGR: specific growth rate; FCR: feed conversion ratio; PER: protein efficiency ratio; PI: protein intake; FI: total feed intake per fish; SR: survival rate.

Means without superscript letters are not significantly different ($P > 0.05$).

Growth parameters, WG, DWG were relatively lower than those reported with the same fish species (Sun et al., 2011; Hu et al., 2014; Wang et al., 2014; Hung et al., 2015b), while SGR was comparable with data registered by Hung et al. (2015b) and considerably lower than that of Sun et al. (2011).

Reportedly, the limitations in use of alternative protein sources may be due to lower acceptance and palatability with replacement of FM by alternative proteins (Domingues et al., 2003). MBM was reported to be poorer palatability compared to fish meal, and increase dietary MBM caused decreasing FI which in turn accounted for growth reduction (Xue & Cui, 2001; Hossain & Islam, 2007). However, our study indicated that there was no significantly difference on FI in all tested diets, and to some extent, FI increased in correspondence with MBM inclusion levels, implying that all experimental diets were well accepted by black carp.

FI varied in marginal range of 9.28 - 9.64 (g/fish/45days) and was significant lower than previous published data for same species (Wang et al., 2014; Hung et al., 2015b). FCR and PER values were not significantly affected by the replacement of FM with MBM. FCR in present study was similar with that reported by Van et al. (2010) for black carp of 300g/individual, held at stocking density of 1 fish/m², and by Van and Thu (2013) for same fish species fed commercial diets of 25% protein content. On the other hand, this value was relatively higher than previous studies (Sun et al., 2011; Hung et al., 2015b). Data on PER was in good agreement with that registered for black carp of same growth stage (Hu et al., 2014). The present study indicated that MBM is a potential alternative protein source for black carp farming and this has been supported by growth performance, feed utilization. MBM is a less expensive and its dietary inclusion is therefore promising.

Acknowledgement

The authors are grateful to International Foundation for Science (IFS) for financial support and to the Faculty of Fisheries, Vietnam National University of Agriculture for providing facilities during this experiment.

References

- Ai, Q., K. Mai, B. Tan, W. Xu, Q. Duan, H. Ma and L. Zhang, 2006. Replacement of fish meal by meat and bone meal in diets for large yellow croaker, *Pseudosciaena crocea*. *Aquaculture*. 260(1-4): 255-263
- Abdul Kader, M., Bulbul, M., Yokoyama, S., Ishikawa, M., Koshio, S., Sakhawat Hossain, M., Arshad Hossain, M. (2011). Evaluation of meat and bone meal as replacement for protein concentrate in the practical diet for Sutchi Catfish, *Pangasius hypophthalmus* (Sauvage 1878), reared under pond condition. *Journal of the World Aquaculture Society*. 42(3): 287-296.
- Bharadwaj, A. S., W. R. Brignon, N. L. Gould, P. B. Brown and Y. V. Wu, 2002. Evaluation of Meat and Bone Meal in Practical Diets Fed to Juvenile Hybrid Striped Bass *Morone chrysops* × *M. saxatilis*. *Journal of the World Aquaculture Society*. 33(4): 448-457
- Burr, G. S., W. R. Wolters, F. T. Barrows and R. W. Hardy, 2012. Replacing fishmeal with blends of alternative proteins on growth performance of rainbow trout (*Oncorhynchus mykiss*), and early or late stage juvenile Atlantic salmon (*Salmo salar*). *Aquaculture*. 334-337(0): 110-116
- Domingues, P., A. Sykes, A. Sommerfield and J. Andrade, 2003. Effects of feeding live or frozen prey on growth, survival and the life cycle of the cuttlefish, *Sepia officinalis* (Linnaeus, 1758). *Aquaculture International*. 11(5): 397-410
- FAO, 2012. The State of World Fisheries and Aquaculture 2012. FAO-Food and Agriculture Organization of the United Nation 00153 Rome, Italy. 230p

- Glencross, B. D., M. Booth and G. L. Allan, 2007. A feed is only as good as its ingredients – a review of ingredient evaluation strategies for aquaculture feeds. *Aquaculture Nutrition*. 13(1): 17-34
- Goda, A. M., E. R. El-Haroun and M. A. Kabir Chowdhury, 2007. Effect of totally or partially replacing fish meal by alternative protein sources on growth of African catfish *Clarias gariepinus* (Burchell, 1822) reared in concrete tanks. *Aquaculture Research*. 38(3): 279-287
- Hardy, R. W., 2010. Utilization of plant proteins in fish diets: effects of global demand and supplies of fishmeal. *Aquaculture Research*. 41(5): 770-776
- Hossain, M. A. and S. F. Islam, 2007. Meat and bone meal as partial substitute for fish meal in nursery diet for giant freshwater prawn, *Macrobrachium rosenbergii* (de Man). *Journal of the World Aquaculture Society*. 38(2): 272-280
- Hu, Y., Y. Huang, F. Feng, L. Zhong, Q. Ai, T. Xiao and H. Wen, 2014. Effect of soybean meal replacement by cottonseed meal on growth, feed utilization and some blood physiological/biochemical indices of juvenile black carp, *Mylopharyngodon piceus*. *Aquaculture Research*. n/a-n/a
- Hung, T. Q., S. A. Mintra and T. T. N. Thu, 2015a. Apparent digestibility of feed ingredients for black carp (*Mylopharyngodon piceus*). *Chiang Mai University Journal of Natural Science*. 14(3): August - December
- Hung, T. Q., T. T. N. Thu and S. A. Mintra. (2015b). Effect of rice bran replacement by corn meal on growth, feed utilization of black carp (*Mylopharyngodon piceus*). Paper presented at the The 53rd Kasetsart University Annual Conference: Smart Agriculture: The Future of Thailand, Bangkok, Thailand.
- Kader, M. A., M. Bulbul, S. Yokoyama, M. Ishikawa, S. Koshio, M. S. Hossain, G. U. Ahmed and M. A. Hossain, 2011. Evaluation of Meat and Bone Meal as Replacement for Protein Concentrate in the Practical Diet for Sutchi Catfish, *Pangasius hypophthalmus* (Sauvage 1878), Reared under Pond Condition. *Journal of the World Aquaculture Society*. 42(3): 287-296
- Leng, X. J. and D. Z. Wang, 2003. Black carp nutrition and feed formulating technology. *Journal of Shanghai Fisheries University*. 12: 265-270
- Millamena, O. M., 2002. Replacement of fish meal by animal by-product meals in a practical diet for grow-out culture of grouper *Epinephelus coioides*. *Aquaculture*. 204(1–2): 75-84
- Phuong, N. T. D., N. V. Trung and K. V. Van, 2009. Status of black carp culture in Red river Delta, Vietnam. *Journal of Science and Technology (Ministry of Agriculture and Rural Development, Vietnam)*. 2: 80-85
- Robaina, L., F. J. Moyano, M. S. Izquierdo, J. Socorro, J. M. Vergara and D. Montero, 1997. Corn gluten and meat and bone meals as protein sources in diets for gilthead seabream (*Sparus aurata*): Nutritional and histological implications. *Aquaculture*. 157(3–4): 347-359
- Sun, S., J. Ye, J. Chen, Y. Wang and L. Chen, 2011. Effect of dietary fish oil replacement by rapeseed oil on the growth, fatty acid composition and serum non-specific immunity response of fingerling black carp, *Mylopharyngodon piceus*. *Aquaculture Nutrition*. 17(4): 441-450
- Van, K. V. and T. T. N. Thu, 2013. Effect of dietary protein levels on black carp (*Mylopharyngodon piceus*) growth performance in semi-intensive. *Journal of Science and Technology (Thai Nguyen University, Vietnam)*. 101(1): 81-84
- Van, K. V., T. A. T. Tuyet, T. D. Hoai and K. T. Dung, 2010. Initial Results of Black Carp Monoculture in Ponds in Haiduong Province. *Journal of Science and Development (Vietnam National University of Agriculture)*. 8(3): 481-487
- Wang, C., X. Zhu, D. Han, J. Jin, Y. Yang and S. Xie, 2014. Responses to fishmeal and soybean meal-based diets by three kinds of larval carps of different food habits. *Aquaculture Nutrition*. n/a-n/a

- Watanabe, T., 2002. Strategies for further development of aquatic feeds. *Fisheries Science*. 68(2): 242-252
- Wheaton, F. W., J. N. Hochheimer, G. E. Kaiser, R. F. Malone, M. J. Krones, G. S. Libey and C. C. Easter. (1994). Nitrification filter design methods [Press release]
- Xue, M. and Y. Cui, 2001. Effect of several feeding stimulants on diet preference by juvenile gibel carp (*Carassius auratus gibelio*), fed diets with or without partial replacement of fish meal by meat and bone meal. *Aquaculture*. 198(3-4): 281-292
- Yang, Y., S. Xie, Y. Cui, W. Lei, X. Zhu, Y. Yang and Y. Yu, 2004. Effect of replacement of dietary fish meal by meat and bone meal and poultry by-product meal on growth and feed utilization of gibel carp, *Carassius auratus gibelio*. *Aquaculture Nutrition*. 10(5): 289-294

Growth performance of carrageenan-producing seaweeds of *Kappaphycus* and *Eucheuma* in Sumbawa

Nunik Cokrowati*¹, A.Nikmatullah², Sunarpi³, Z. Abidin¹

¹ Aquaculture study program, Mataram University

² Agriculture Faculty, Mataram University

³ Mathematic and Science Faculty, Mataram University

Jl. Majapahit No. 62 Telp. (0370) 633007 Mataram 83125

Abstract

This research purpose is to address growth performance of carrageenan-producing seaweeds of *Kappaphycus* and *Eucheuma*. The research was conducted in coastal area of Kaung Island, Buer sub-district, Sumbawa Regency from August to September 2012. Importance of *Eucheuma/Kappaphycus* spp. for Indonesian economy and farmer livelihood are main source of hydrocolloids and main source of income. West Nusa Tenggara is one of the production center for *Eucheuma/Kappaphycus* in Indonesia. The method used for this study was planting seaweed using long-line system involving 25 farmers. The species planted were *Kappaphycus alvarezii* Tembalang, *Kappaphycus alvarezii* Maumere, *Kappaphycus striatum* and *Eucheuma spinosum*. The growth of *Eucheuma* spp was measured every 7 days until harvesting time which is 45 days. Initial seed weight was 100 g and the number of seeds perline were 200. Purphoses sampling done by 5 samples per line for analysis of fresh weight, dry weight, carragenan content and incident disease. The result of this research shows that the increase in weight *Kappaphycus alvarezii* Tembalang is 0,82 grams/day, *Kappaphycus alvarezii* Maumere is 0,06 grams/day, whereas that of *Kappaphycus striatum* is 0,97 grams/day and *Eucheuma spinosum* is 5,59 grams/day. It can be concluded that *K. alvarezii*, *K. striatum* and *E. spinosum* can grow in Kaung, *E. spinosum* is more adaptable to Kaung ecology, and can be grown throughout the year.

Keyword: cultivation, long line, fresh weigth, dry weight, adaptable.

*Corresponding author: n_cokrowati@icloud.com

Introduction

The development if seaweed cultivation to improve community's prosperity in NTB is supported by economic potency and large cultivation area. As an illustration, data of economy survey in 2010 show that seaweed farmer revenue in NTB ranged from Rp. 26.538.000 to Rp. 60.458.500 per year, in which the potency of cultivation area reached 25.206 hectares (Pemda NTB, 2011). However, the use of this area just reached 44% with total production of dry seaweed was 221.047 tonnes (Pemda NTB, 2011).

Economically, seaweed cultivation is one of productive businesses within coastal area which brings benefit. Besides accelerating seaweed harvest time (30 to 50 days) and cultivation that does not need a complex maintenance, seaweed (*Eucheuma/Kappaphycus*) cultivation in NTB is also an important source of livelihoods that improves community's revenue along the coastal area. This is because the cultivation of seaweed can bring revenue which is 26 to 60 million rupiahs per year and it depends on cultivation method used and the total area of cultivation.

In the other hand, dry seaweed production in NTB just reached 220,000 tonnes, and this value is less than the potency of production which is more than 1 million tonnes per year (Pemda NTB, 2011). Seaweed is not only an important source of alternative livelihood for farmers, but also a source of hydrocolloid used in many industries. Some of these industries are pharmacy, food, paint, and cosmetic that uses carrageenan as one of raw materials.

According to those facts, the local government of NTB has taken strategic steps including extension and intensification of seaweed cultivation in order to improve seaweed production in NTB and increase locally-generated revenue. This effort also aims to grow economic business which is productive that benefits coastal community that generally has low income. From the perspective of NTB, extension of seaweed cultivation directed to Sumbawa Island has a big potency because of the extent of potential area that has not been used which is more than 50% out of 20.200 hectares of potential area for seaweed cultivation in Sumbawa Island. Kaung Island is a potential area to develop cultivation of many species of seaweed such as *Kappaphycus* and *Eucheuma*. According to this fact, there is a need of a research to address the growth of and carrageenan from *Kappaphycus* and *Eucheuma* cultivated in marine area of Kaung Island.

Material and Methods

The method used for this study was planting seaweed using long-line system involving 25 farmers. The species planted were *Kappaphycus alvarezii* Tembalang, *Kappaphycus alvarezii* Maumere, *Kappaphycus striatum* and *Eucheuma spinosum*. The growth of *Eucheuma* spp was measured every 7 days until harvesting time which is 45 days. Initial seed weight was 100 g and the number of seeds per line were 200. Purphoses sampling done by 5 samples per line for analysis of fresh weight, dry weight, carrageenan content and incident disease.

Seaweed was planted from August to September 2012. The seed used was introduced from seed bed in Gerupuk Bay area and Mid-Lombok Regency. Data of weight increase was calculated using following formulas:

$$\text{Increase of weight} = \text{weight on } t \text{ (time)} - \text{initial weight}$$

$$\text{Growth rate (\% per day)} = ((\ln W_t - \ln W_o) / t) \times 100$$

Analysis of carrageenan was done by extracting seaweed, cooked with pressure on temperature of 100°C for 2 to 3 hours until seaweed turned into a gel, with alcohol. The analysis of carrageenan was done in Immunology Lab, Faculty of Math and Sciences, Universitas Mataram.

Results and discussion

The result of measurement of four 4 seaweed species cultivated on four ropes on this study is shown on the table 1.

Table 1. Average weight of seaweed (g) cultivated between August and September 2012.

No.	Species	Day-							
		0	7	14	21	28	35	42	49
1.	<i>E. cottonii</i> (tembalang)	100	146	178	207	156	112	130	140
2.	<i>E. cottonii</i> (maumere)	100	148	166	120	103	82	99	103
3.	<i>E. spinosum</i>	100	146	178	213	262	300	337	374
4.	<i>E. striatum</i> (sacol)	100	146	179	210	169	126	138	148

Weighing was done for each clump with 180 times of repeat per unit

According to the data, decreasing weight is experienced by *E. cottonii* Tembalang, *E. cottonii* Maumere, and *E. striatum*. This condition generally happens on day 28. The decreasing weight is caused by *ice ice* disease that caused seaweed thallus to break. The data of weight increase shows that *E. spinosum* production reaches 74 Kg per 100 m².

Seaweed species of *E. spinosum* constantly experiences increasing weight although this species is attacked by *ice ice* disease. *Eucheuma spinosum* also has the highest growth compared with other species of seaweed. The data also show that there are some seaweed clumps from species of *E. cottonii* tembalang and maumere, and *E. striatum* gone because of *ice ice* attack.

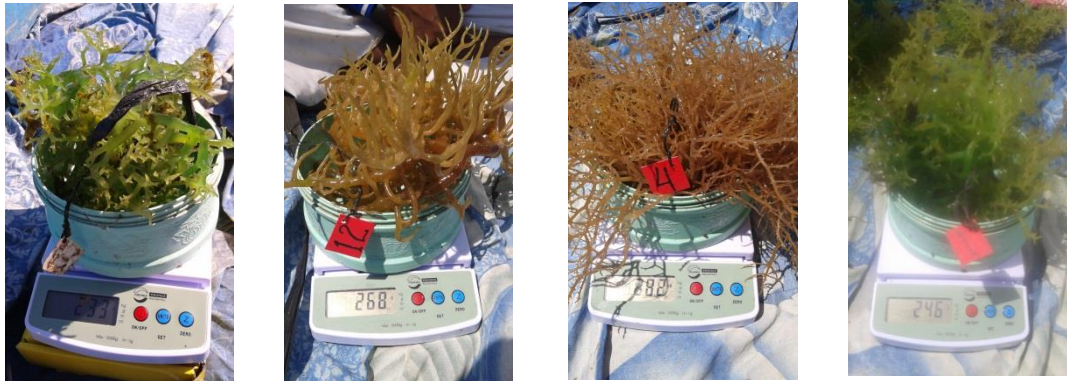


Figure 1. Seaweed weighing

Following are the ratio between wet and dry weights of each seaweed species on different planting days. Sample was taken from ropes for research purpose. Drying was done on the top of *para-para* for 3 days.

Table 2. The ratio between wet and dry weights of seaweed.

No.	Seaweed	Weight measurement (g)											
		Day 28			Day 35			Day 42			Day 49		
		W	D	D/W	W	D	D/W	W	D	D/W	W	D	W/D
1	<i>E. cottonii</i> (tembalang)	156	20	0.12	112	15	0.13	130	13	0.1	140	13	0.09
2	<i>E. cottonii</i> (maumere)	103	14	0.13	82	11	0.13	99	9	0.09	103	9	0.08
3	<i>E. spinosum</i>	262	42	0.16	300	49	0.16	337	54	0.16	374	60	0.16
4	<i>E. striatum</i> (sacol)	169	21	0.12	126	17	0.13	138	16	0.11	148	15	0.10

Weighing was done for each clump with 180 times of repeat per unit

According to table above, it can be seen that the highest ratio between wet and dry weights is experienced by *E. spinosum*. The ratio between wet and dry weights can represent water content in seaweed. The higher the ratio, the lower water content in seaweed, a fact which means that the higher dry seaweed production.

The result of measurement of seaweed sample done in Immunology Lab, Faculty of Match and Sciences, Universitas Mataram is shown on following figure 2.

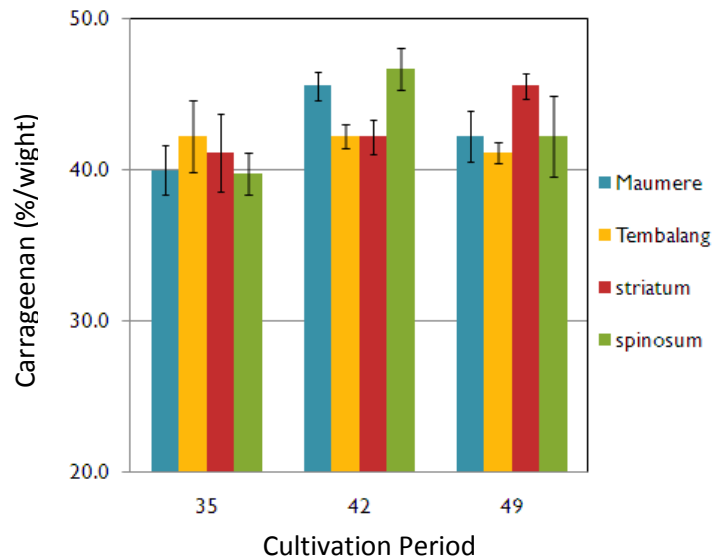


Figure 2. Percentage of Carrageenan in many Species and Cultivation Periods

The content of seaweed carrageenan in *E. spinosum* and *K. alvarezzi* strain maumere cultivated for 42 days tends to be higher compared with that of cultivated for 35 and 49 days, whereas the carrageenan content in *E. striatum* tends to increase gradually until day 49 of cultivation. Carrageenan content of *K. alvarezzi* (tembalang) tend to be stabile on the three cultivation periods.

Conclusion

K. alvarezii, *K. striatum* and *E. spinosum* can grow in Kaung. *E. spinosum* is more adaptable to Kaung ecology, and can be grown throughout the year.

References

- Anggadireja, J.T.S. Irawati, Kusmiyati., 2006. *Potensi dan Manfaat Rumput Laut Indonesia Dalam Bidang Bidang Farmasi*. Deputi Bidang Pengkajian Ilmu Dasar dan Terapan BPPT, Jakarta.
- Blink, L.R., 2004. *Physiology and Biochemistry of Algae*. In Manual of Physiology. Academic Press. New York.
- BPS 2010. Nusa Tenggara Barat Dalam Angka.
- Ditjenkan Budidaya, 2005. *Profil Rumput Laut Indonesia*. Direktorat PerikananBudidaya, Departemen Kelautan dan Perikanan. Jakarta.
- Pemda NTB, 2011. Evaluasi dan Pencapaian Program Pijar NTB.
- Sunarpi, Jupri A., dan Julisaniah NI, 2009. Pengembangan Pupuk Cair Ramah Lingkungan Berbasis Rumput Laut Alam Nusa Tenggara Barat Untuk Mendukung Sistem Pertanian Berkelanjutan. Laporan Hasil Penelitian Hibah Startegis Nasional. Lemlit Universitas Mataram.

Advantages of Environmentally Sound Poly-Eco-Aquaculture in Fish Farms

S. Kadowaki*¹ & Y. Kitadai²

¹ Aquaculture Science, Faculty of Fisheries, Kagoshima University, 4-50-20 Shimoarata, Kagoshima 890-0056, Japan, ²“Hakase” Cramming School, 1-1 Minobayashi, Anan. Tokushima 774-0017, Japan

Abstract

Environmentally sound poly-eco-aquaculture enables the preservation of aquatic environments to be compatible with that of sustainable aquaculture. With this method, not only healthy fish can be cultured in purified water, but also the productivity will increase by recycling seaweed to feed fish. The maximum nitrogen uptake rate of each seaweed per square meter of seaweed area was 2.9 mg N/m²/day for *Laminaria japonica*, 3.1 mg N/m²/day for *Undaria pinnatifida* and 3.6 mg N/m²/day for *Ulva pertusa*. The maximum phosphate uptake rate was 0.43 mg P/m²/day, 0.54 mg P/m²/day, and 0.19 mg P/m²/day. The calculated values of nitrogen and phosphate uptake rates, obtained by integrating the nutrient concentrations, light intensity, and water temperatures, corresponded well with each observed value. The minimum seaweed cultural density necessary per unit area of *Seriola quinqueradiata* farm was calculated using the values of the maximum nitrogen uptake rate. The maximal production rates were 0.75 mg O₂/g wet/h for *L. japonica*, 0.83 mg O₂/g wet/h for *Un. pinnatifida*, and 6.39 mg O₂/g wet/h for *Ul. pertusa*. The minimal weight of cultured seaweeds necessary to accommodate the oxygen consumption of an individual *S. quinqueradiata* was calculated as 1.17 kg wet/a fish, 0.83 kg wet/a fish, and 0.21 kg wet/a fish.

Introduction

The industry of marine aquaculture in the 21st is expected to be practiced in harmony with the environment. It is our responsibility to hand down a blue and abundant sea for our future generations to inherit. Environmentally sound poly-eco-aquaculture is a technical innovation of aquaculture used to purify water and promote a balanced ecosystem by breeding seaweed and shellfish around coastal fish farms. The seaweed is used to feed for fish and shellfish. ‘Eco’ in the word “eco- aquaculture” means a harmony of ecology for nature and economy for humanity.

In order to establish coastal fish farms which enable sustainable aquaculture, there is a recent requirement for concrete measures to be taken to improve water quality in the farms. Biological water purification is necessary for preventing eutrophication of water and for reducing oxygen deficiency in water (Kadowaki 1994, 2001, 2004). Seaweed cultivation is currently attracting much attention as a plausible measure in this plight. Aiming to improve the water quality of coastal yellowtail *Seriola quinqueradiata* farms in warm water zone over a year long period, different seaweed species were cultivated in the farm during each season. ‘Wakame’ *Undaria pinnatifida* was grown during the winter months, ‘Konbu’ *Laminaria japonica* in the spring, and sea lettuce *Ulva pertusa* in the summer and autumn. Following this, the relationship between nitrogen and phosphate uptake rates, oxygen production rate by the different seaweeds, and the nutrient concentration, light intensity, and temperature of the water in the farm was estimated (Kitadai & Kadowaki 2003, 2004a, 2004b, Kitadai 2005). Next, the improvement in nitrogen uptake in relation to nitrogen load by yellowtail aquaculture, as well as the cultivation scale of seaweeds necessary for oxygen production in relation to oxygen consumption per

individual yellowtail and per cubic meter of the net cage, was estimated. This study exemplifies the extent to which water quality of feeding fish farms was improved by cultivating seaweeds.

Environmentally Sound Poly-Eco-Aquaculture

In order to create a truly rich production by cultured fish, we would like to propose that in the part where the balance of ecosystem has been broken, the balance of ecosystem is restored by the introduction of poly-eco-aquaculture which directly utilizes solar energy as shown in Figure. The primary principle of poly-eco-aquaculture is breeding seaweeds, such as *Un. pinnatifida*, *L. japonica*, and *Ul. pertusa* throughout the year for the artificial formation of sea forest around cultured fish cages. The seaweed will uptake nutrients, such as nitrogen, and phosphate from fish feces and remaining feed. The seaweed also inhibits pathogenic bacteria (Nagahama & Hirata 1990) and red tide (Hirata et al., 1986). Grown seaweeds will be fed to abalone (*Haliotis discus hannai*, *H. discus discus*, *H. gigantea*), sea urchin (*Stichopus japonica*, *Holothuria pervicax*), yellowtail and red sea bream *Pagrus major*. Sea cucumber *Stichopus japonica* is grown in symbiosis with abalone in aquaculture net cages. Feces of abalone are fed to sea cucumber. Scallop *Chlamys nobilis* can be cultured because they eat organic suspended substances, such as remaining feed and the fish feces.

When cultured abalone and sea cucumber are dried, they can be stored for long periods and can be shipped long distance at room temperature. It was also found that half pearls could be grown in cultured, giant abalone *H. gigantea* in five months after a nucleus was inserted into it. The shells can also be used for mother of pearl work. With poly-eco-aquaculture, there is a higher additional value, as well as expectation of increasing job opportunities. The capability at which seaweeds can uptake nitrogen has been researched, and it was found that the purification of aquatic environments to allow the amount of cultured fish became feasible when the area of seaweed breeding was more than the area of fish aquaculture. Based on the research results, Azuma-cho Fisheries Co-operative Association decided to employ seaweed breeding near the marine aquaculture farm in an effort to increase the proportion of the area of seaweed to the area of fish aquaculture, aiming at safe, sustainable aquaculture. The fishermen themselves are practicing poly-eco-aquaculture.

Conclusion

From the results, it was obvious that all of the seaweed species had the capacity to take in nitrogen and phosphate loads and that they fulfilled the role as oxygen producers. However, it may be difficult for seaweed to completely take in nitrogen and phosphate loads alone. Even with *Ul. pertusa* which uptakes nitrogen and phosphate loads most effectively, it would take an area of *Ul. pertusa* two and a half times of area of a fish farm in order to take in the loads completely. Still, it is considered important to cultivate effective seaweed for the eutrophication of each fish farm and improve the water quality. It was also shown that seaweed worked effectively in supplying oxygen. This indicates that it is both possible to reduce environmental load and supply oxygen necessary for feeding fish, in addition to managing the water environment, using seaweed.

The time to fully implement poly-eco-aquaculture is now ! In order to reproduce marine aquaculture farms, concrete measures need to be taken to promote year-round seaweed breeding inside and outside of fish farms, artificial formation of seaweed beds, reuse and circulation of output biomass of seaweed. Environmentally sound poly-eco-aquaculture answers the needs of both environment and industry, because it will enable environmental conservation through water purification, compatible with sustainable aquaculture that would culture healthy fish. When the sea is supported by the biodiversity, the productivity of fish farms will be developed, and a rich sea having sustainable productivity might be realized. It is our hope that those in the aquaculture industry will try this eco-friendly approach to promote sustainable aquaculture.

Study of humoral immune response using IBD Blen® and Vaxxitek HVT-IBD® vaccines even high maternal derived antibody in broiler chickens

Kornkamon Paditporn^{1*}, Jamlong Mitchaothai² & Sakchai Ruenphet³

¹Clinic for Poultry Department, ²Clinic for Swine Department, ³Immunology and Virology Department, Faculty of Veterinary Medicine, Mahanakorn University of Technology, 140 Cheum-Sampan Rd., Nong Chock, Bangkok 10530 Thailand.

Abstract

Infectious bursal disease (IBD) also known as Gumboro disease is an important viral disease in poultry industry due to significant economic losses resulting from high mortality and immunosuppression. The disease can only be controlled and prevented by proper vaccination and biosecurity. Nine hundred broilers were divided into 3 groups for present study. The first group was not vaccinated with IBD vaccine, the second was vaccinated using IBD-Blen® (Merial, USA) by drinking water at 2-week-old. Another was vaccinated using VAXXITEK® HVT+IBD (Merial, USA) by subcutaneous route at 0-day-chick. Broiler sera of thirty birds were collected at 1, 2, 3, 4 and 5-week-old, antibody titer was measured using IBD enzyme-linked immunosorbent assay (ELISA) test kit by ProFLOK® PLUS (Synbiotics Corp, USA). Antibody titers of VAXXITEK® HVT+IBD vaccinated group was not significant difference with IBD-BLEN® vaccinated groups. However, both vaccinated groups were significant difference with unvaccinated group since 3-week-old. These results indicated that VAXXITEK® HVT+IBD and IBD Blen® could provide better response to broiler chicken, in term humoral immunity, even in the presence of high maternal derived antibody.

Keywords: Infectious bursal disease, VAXXITEK® HVT+IBD, IBD-Blen®

**Corresponding author: kornkamo@mut.ac.th*

Introduction

Infectious bursal disease (IBD) also known as Gumboro disease is an important viral disease in poultry industry due to significant economic losses resulting from high mortality and immunosuppression especially in young chickens. Generally, prevention of avian diseases including IBD, can be achieved by biosecurity. However, IBD virus is highly resistant to adverse environmental conditions, as a result the pathogens persist in chicken houses, therefore, the control of this disease depends mainly on vaccination (Al-Natour et al., 2004).

Generally, the commercial vaccines can be divided into 3 groups depending on the virulence such as mild, intermediate and intermediate-plus or hot vaccines. Disadvantage of mild and intermediate vaccine are maternal derived antibodies (MDA) destroying in the chicks, so these vaccine should be used in the chicks that have a low level of MDA (Tsukamoto et al., 1995) and should have a booster dose in next 2 weeks to get the optimum antibody. On the other hand the intermediate-plus or hot vaccines are suitable for the high MDA chickens (Haddad et al., 1997) because the vaccines virus can break through the MDA and stimulate the active antibodies against the IBDV in vaccines.

The objective of this study was to evaluate the immunological response of 2 types of vaccine in commercial broiler chickens even high maternal derived antibody.

Materials and Methods

Nine hundred, 0-day-old Cobb 500 and Ross 308 broiler chickens were obtained from a commercial hatchery. The chickens were fed *ad libitum*. All of chickens were divided into 3 groups. The first group, the chickens were not vaccinated with IBD vaccine. The second group, the chickens were vaccinated with IBD-Blen[®] (Merial, USA) vaccine by drinking water at 2-week-old. Another group, the chickens were vaccinated with VAXXITEK[®] HVT+IBD (Merial, USA) by subcutaneous injection at 0-day-chick. Sera were collected at 0 day of age for maternal antibody determination. In addition broiler sera of thirty birds were collected at 1, 2, 3, 4 and 5-week-old, and analyzed for IBD antibodies using enzyme-linked immunosorbent assay (ELISA) test kit by ProFLOK[®] PLUS (Synbiotics Corp, USA).

All data were statistical analysis into a spreadsheet programmes (Excel 2000; Microsoft Corporation) and transferred to SPSS 10.0 software. Data of different parameters were analyzed using analysis of variance (ANOVA) as repeated measurement with Tukey HSD's multiple range test. The difference between parameters was regarded as significant when the p value was less than 0.05.

Results

Fig. 1 showed the average IBD antibody titers graph of the present study. At 0 day chick, antibody titers was observed in all group that were passive immunity from breeding parents. The antibody titers decreased slowly during 1week post vaccination in the VAXXITEK[®] HVT+IBD groups at 1-week-old, while IBD-Blen[®] group also increased during 1week post vaccination at 3-week-old. The antibody titers of both vaccinated groups were significantly ($P < 0.05$) higher than the unvaccinated group since 3-week-old (Table 1).

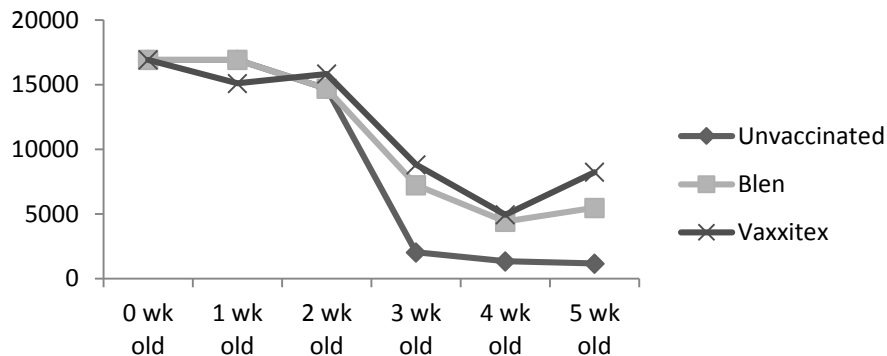


Figure 1. Average IBD antibody titers at 0-5 week old

Table 1. IBD antibody titer (\log_{10}) of the chicken throughout the experiment.

Titer	0 wk old	1 wk old	2 wk old	3 wk old	4 wk old	5 wk old
Unvaccinated	4.23±0.05a	4.23±0.03a	4.16±0.08a	3.21±0.33b	2.86±0.44b	2.67±0.68b
Blen [®]	4.23±0.05a	4.23±0.03a	4.16±0.08a	3.76±0.33a	3.57±0.30a	3.67±0.25a
VAXXITEK [®]	4.23±0.05a	4.17±0.09b	4.20±0.05a	3.90±0.24a	3.64±0.26a	3.88±0.20a

^{a,b} Values within the same column with different superscripts mean statistically significant difference ($p < 0.05$), Data represent mean \pm SD.

Summary and Discussion

The commercial live attenuated IBD vaccines such as IBD-Blen[®], were intermediate plus as winterfield 2512 IBD virus strain. Generally, the intermediate-plus or hot vaccines are suitable for the high MDA chickens because the virus in vaccines can break through the MDA and stimulate the active antibodies against the IBDV in vaccines but do not recommended for chickens younger than 10 days of age (Chansiripornchai and Wanasawaeng, 2009; Haddad et al., 1997). The major problem with active immunization is the interference with MDA, which might neutralize the vaccine. Thus the timing of IBD vaccine administration is crucial (Saif, 1998; van den Berg, 2000; Müller *et al.*, 2003).

VAXXITEK[®] HVT+IBD is the recombinant turkey Herpesvirus (HVT) expressing the VP2 gene of the Infectious Bursal Disease virus. This recombinant vaccine could prevent Marek's disease and IBD of chickens at the same time after vaccinated by the subcutaneous route at 1-day old (Rashid et al, 2013). Present study indicated that active immune response could be induced at day-old vaccination with VAXXITEK[®]HVT+IBD vaccine.

In conclusion, the humoral immune response of the VAXXITEK[®] HVT+IBD group that vaccinated at 0-day chick, was not significant difference with IBD-BLEN[®] groups that vaccinated at 14-day old. However, both vaccinated groups were significant higher than unvaccinated group since 3-week-old, even in the presence of high maternal derived antibody.

Reference

- Al-Natour, M. Q., Ward, L. A., Saif, Y. M., Stewart-Brown, B. and Keck, L. D., 2004. Effect of Different Levels of Maternally Derived Antibodies on Protection Against Infectious Bursal Disease Virus. *Avian disease*. 48: 177-182
- Chansiripornchai, N. and Wanasawaeng, W., 2009. Avian immune system. In: *Avian Immune System and Serum Interpretation*. Chansiripornchai, N. (ed), Trionasarn Co. Ltd., Bangkok. pp 27-74
- Haddad, E.E., Whitfill, C.E., Avakian, A.P., Ricks, C.A., Andrews, P.D., Thoma, J.A. and Wakenell, P.S., 1997. Efficacy of a novel infectious bursal disease virus immune complex vaccine in broiler chickens. *Avian disease*. 41(4): 882-889
- Müller H, Islam MR, Raue R., 2003. Research on infectious bursal disease--the past, the present and the future. *Veterinary Microbiology*. 97(1-2):153-65
- Rashid, M.H., Luo, H., Akhter, J., Islam, M.T., Islam, M.R., Rahman, M.M., Cao, Y. and Xue, C., 2013. Protective effect of Vaxxitek HVT+IBD vaccine against Infectious Bursal Disease in broiler chickens. *Progress Agriculture*. 24(1 & 2): 69-78
- Saif YM, 1998. Infectious bursal disease and hemorrhagic enteritis. *Poultry Science*. 77(8):1186-1189
- Tsukamoto, K., Tanimura, N., Kakita, S., Ota, K., Mase, M., Imai, K. and Hihara, H., 1995. Efficacy of three live vaccines against highly virulent infectious bursal disease virus in chickens with or without maternal antibodies. *Avian Disease*. 39(2): 218-229
- Van den Berg TP, 2000. Infectious bursal disease in poultry: a review. *Avian Pathology*. 29:175-194.

Fertility status of local PO cattle and its crosses with Limousin and Simental bull in Situbondo Regency, East Java, Indonesia

S. Suyadi, S Wahjuningsih and Kuswati*

*Faculty of Animal Husbandry, University of Brawijaya
Jl. Veteran Malang, Indonesia 65145*

Abstract

Fertility status or fertility index is a simple indicator to measure the fertility conditions of cow or bull in herd. This study was to evaluate the reproductive performances of local Peranakan Ongole (PO) cattle and its crosses with Limousin and Simental at Parity 3 and 4. There were no significant differences between parities and breeds for the reproductive performances and fertility status (FS) in this location. The reproductive performances and fertility status of PO cows and its crosses with Limousin and Simental bull conducted artificially insemination were in normal range, and there were no significant difference between breeds and parities.

Keywords: local cattle, service per conception, days open, conception rate, calving rate.

**Corresponding author: suyadi2008@yahoo.com*

Introduction

Improving productivity of beef cattle can be achieved by crossing between local cattle and exotic cow that has the advantage of production far above the average production of local cattle. In Indonesia, we know Peranakan Ongole (PO, Ongole grade) cattle as one of the local cattle that spread in overall of Indonesia regions. This cattle is well adaptive to the tropical condition with high risk of tick infection, high environmental temperature and humidity, and tolerant to poor quality of grass with high fibre for their feed, although they show low growth rate and productivity.

Nowadays, the farmers have high interest to cross this cattle with temperate breeds to get higher growth and productivity, although some reports showed that this crosses resulted low reproductive performances (Nugroho, 2013). Our previous studies (Suyadi et al. 2013; Suyadi and Nugroho, 2014) showed that the reproductive performances was considered influenced by the breed of animal resulting between PO- x Limousin cattle and affected by the altitudes (lowland or highland areas) where the animal were reared. In the pure breed of cattle (PO-cattle) the service per conception (value=1.64 - 1.65) was lower than those for LimPo (Limousin x PO-) cattle with value 1.90 – 2.00 for both at lowland and highland areas, whereas the conception rate of PO cattle were higher than those for LimPo cattle. Finally, the calving interval (in month) was similar between the two breeds of cattle.

In Bali cattle, the productive and reproductive performances were not only influenced by the breed but the altitudes. The crosses between Bali cattle with Simental bull via artificial insemination improved the the birth weight, weaning weight and pre-pubertal growth, although these decreased the reproductive performances i.e. reduced pregnancy rate and calving rate as well as prolonged days open period and calving interval (Pribadi, et al. 2014, 2015). This study was to evaluate the reproductive performances of local PO cows and its crosses with Limousin and Simental cattle.

Materials and methods

This study was conducted as a Survey Study, and to analyze the fertility status on 112 cows of local Peranakan Ongole (PO) cattle and its crosses with Limousin (LIMPO) 118 cows and with Simental semen bull (SIMPO) 113 cows by measuring the reproductive performances including Service per Conception (S/C), Days Open (DO), Conception Rate (CR) and Calving Interval (CI) at third and fourth parity. The study was conducted at farmer condition in Situbondo Regency, East Java, Indonesia and the data were collected from the records by inseminator. Data were analysed using analysis of variance according to the group of breeds and parities.

Results and discussion

This study was emphasized to analysis the reproductive performances of local cattle and its crosses with Limousin and Simental cattle though artificial insemination breeding. The population structure of cattle at the location was shown in Table1.

Table 1. Population structure of cattle in location of study with different breeds

Breed	Mature		Young Animal		Calf	
	Bull	Cow	Male	Female	Male	Female
PO	0	126	0	2	0	0
LIMPOCrossbred	0	311	2	27	5	18
SIMPOCrossbred	1	325	3	8	7	23
Total	1	762	5	37	12	41

Service per conception (S/C) was not different between breeds, and all these values were in normal standard (Table2). The S/C values in all breeds and parities of 1.38 – 1.43 were

Table 2. Service per conception (S/C) of the cows at Parity 3 and 4 of different crossbred cows.

Breed	N	Parity -3	Parity-4	Mean \pm SD
PO	112	1.42 \pm 0.58	1.41 \pm 0.59	1.42 \pm 0.01
LIMPO Crossbred	118	1.39 \pm 0.51	1.37 \pm 0.58	1.38 \pm 0.01
SIMPO Crossbred	103	1.40 \pm 0.58	1.46 \pm 0.59	1.43 \pm 0.04

Coception rate (CR) is an simple indicator to determine the fertility status of cows in herd. The CR of PO and its crosses with Limousin and Simental in the location of study were no significant difference among groups (Table 3). The value of CR in this study ranging 62-64% were catagorized as high value, since in othe location this ranged 25-38% (Noercahyo et al. 2015).

Table3. Conception rate of different crossbred cows at Parity-3 and Parity-4.

Breed	N	Parity -3 (%)	Parity-4 (%)	Mean (%)
PO	112	64.28	65.17	64.37
LIMPO Crossbred	118	61.86	67.79	64.83
SIMPO Crossbred	103	65.04	60.19	62.62

Days open (DO) and calving interval (CI) period for PO cattle were significantly longer than those for Limpo and Simpo cattle,, and not significant different between parities (Table 4 and Table 5). This data were contradicted to the recent study at other location, where the reproductive effieency of PO cattle was higher than its crosses with Limousin cattle (Noercahyo, 2015).

Table4. Days Open (DO) of different crossbred cows at Parity-3 and Parity-4

Breed	N	Parity -3 (%)	Parity-4 (%)
PO	112	125.3±24.2 ^a	123.9±21.5 ^a
LIMPO Crossbred	118	114.2±16.3 ^b	116.5±20.5 ^b
SIMPO Crossbred	103	113.1±19.1 ^b	112.8±16.1 ^b

a,b superscript within same column was significant difference (P<0.05)

Table5. Calving interval (CI) of different crossbred cows at Parity-3 and Parity-4

Breed	N	Parity -3 (%)	Parity-4 (%)
PO	112	411.1±24.1 ^a	409.5±22.0 ^a
LIMPO Crossbred	118	399.8±16.5 ^b	402.4±21.2 ^b
SIMPO Crossbred	103	398.3±18.9 ^b	398.6±16.6 ^b

a,b superscript within same column was significant difference (P<0.05)

Conclusion

The reproductive performances and fertility status of PO cows and its crosses with Limousin and Simmental bull conducted artificially insemination were in normal range, and there were no significant different between breeds and parities, although the reproductive efficiency for PO cattle tend to lower than those for its crosses with Limousin and Simmental cattle.

References

- Noercahyo MRD, Nuryadi and Suyadi, 2015. Reproductive efficiency of PO and its crosses with Limousin at different Parity in Bondowoso Regency. Research Report (unpublished data).
- Nugroho H. 2013. Production and reproduction performances of local PO cattle and its crosses with Limousin cattle reared under different climates in East Java, Indonesia. PhD. Thesis. University of Brawijaya. Malang, Indonesia.
- Pribadi LW., S. Maylinda, Moch. Nasich and S. Suyadi. 2014. Prepubertal growth rate of Bali cattle and its crosses with Simmental breed at lowland and highland environment. IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS), e-ISSN: 2319-2380, p-ISSN: 2319-2372. Volume 7, Issue 12 Ver. II (Dec. 2014), PP 52-59. www.iosrjournals.org
- Pribadi LW., S. Maylinda, Moch. Nasich and S. Suyadi. 2015. Reproductive efficiency of Bali cattle and its crosses with Simmental breed in the lowland and highland areas of West Nusa Tenggara Province, Indonesia. Livestock Research for Rural Development 27 (2) 2015
- Suyadi and H Nugroho, 2014. Analysis of reproductive efficiency in Peranakan Ongole (PO)- and its crosses with Limousin (Limpo) cattle in East Java, Indonesia. Proceeding 16th-AAAP, Yogyakarta, Indonesia.
- Suyadi S., L. Hakim, S. Wahjuningsih and H. Nugroho, 2013. Reproductive Performance of Peranakan Ongole (PO)- and Limousin x PO crossbred (Limpo) cattle at different altitude areas in East Java, Indonesia. Journal of Applied Science and Agriculture, 9(11) Special 2014, Pages: 81-85.

Reliable monoclonal antibodies for immunodiagnosis of fasciolosis in both animal and human

Anuracpreeda ^{*1,3}, P., R. Chawengkirtikul² & P. Sobhon³

¹Division of Agricultural Science, Mahidol University, Kanchanaburi Campus, Saiyok, Kanchanaburi 71150, Thailand, ²Department of Microbiology, Faculty of Science, Mahidol University, Rama VI Rd., Bangkok 10400, Thailand, ³Department of Anatomy, Faculty of Science, Mahidol University, Rama VI Rd., Bangkok 10400, Thailand

Abstract

Tropical fasciolosis is a disease caused by two major species of trematode parasites, i.e., *Fasciola hepatica* and *F. gigantica*. Although fasciolosis is primarily a ruminant disease, human infections have been also reported by the World Health Organization. The current diagnosis of fasciolosis is often unreliable and has low sensitivity, as fluke's eggs are not found during the pre-patent period and shedding of parasitic eggs is intermittent. The antibody detection for fasciolosis in animals has been developed and used for a number of years. This antibody detection is not a direct indicator of active infection, and cross-reactivity with other parasites' antigens is often difficult to differentiate. Therefore, antigen detection assay is considered to be better as it can identify animals with pre-patent or occult infection, which could not be detected by the usual parasitological test. Furthermore, the antigen detection can give a more accurate indication of current infection. In the present study, we have produced the monoclonal antibodies (MoAbs) specific against *F. gigantica* somatic antigens using the *in vitro* hybridoma technique by fusion of the immunized spleen cells and myeloma cells. Reactivity and specificity of these MoAbs were examined by ELISA and immunoblotting assays. Seven stable clones, namely PA01, PA02, PA03, PA04, PA05, PA06 and PA07 were obtained from the hybridoma cells. It was found to be IgG₁ and λ light chain isotypes. All of these MoAbs exhibited no cross-reactions with other parasites' antigens. For immunolocalization, *F. gigantica* somatic antigens were present in the caecal epithelium and caecal lumen of the fluke. Thus, it is possible that these MoAbs could be a good candidate for immunodiagnosis of fasciolosis in both ruminant and human.

Keywords: *Fasciola gigantica*, monoclonal antibody, ELISA, immunoblotting, immunolocalization

*Corresponding author: Panat1@yahoo.com

Introduction

Tropical fasciolosis in ruminants caused by *Fasciola gigantica* is an economically important disease that causes a worldwide loss to the livestock industry at more than 3 billion U.S. dollars per annum (Spithill et al., 1999). Although fasciolosis is primarily a ruminant disease, human infections have been also reported by the World Health Organization. The current method of diagnosis for fasciolosis is based on the microscopic detection of fluke's eggs in feces. It is often unreliable and has low sensitivity, as fluke's eggs are not found during the pre-patent period and shedding of parasitic eggs is intermittent. The antibody detection for fasciolosis in animals has been developed and used for a number of years. This antibody detection is not a direct indicator

of active infection, and cross-reactivity with other parasites' antigens is often difficult to differentiate. Therefore, antigen detection assay is considered to be better as it can identify animals with pre-patent or occult infection, which could not be detected by the usual parasitological test. Furthermore, the antigen detection can give a more accurate indication of current infection (Anuracpreeda et al., 2013). Up to now, the main sources of potential immunodiagnostic antigens in fasciolosis are the somatic antigens of the parasites. The somatic products are known to modulate the immune response of the host and thus the parasite antigens invoke the host protective immune responses which result in the death and expulsion of the parasites during the development of concomitant immunity. Since this protein is released into the host fluid in a fairly large amount, it could be a good candidate for immunodiagnosis of fasciolosis by *F. gigantica*, especially the early phase of infection. In the present study, the monoclonal antibodies (MoAbs) against *F. gigantica* somatic antigens were produced and characterized for their binding with these antigens in various parasite's tissues. The cross reactivities with other trematode and non trematode parasite antigens were also tested in order to verify their specificity for possible applications in immunodiagnosis.

Materials and Methods

In the present study, we have produced the monoclonal antibodies (MoAbs) specific against *F. gigantica* somatic antigens using the *in vitro* hybridoma technique by fusion of the immunized spleen cells and myeloma cells. The positive clones were selected and expanded according to Anuracpreeda et al. (2014). Reactivity and specificity of these MoAbs were examined by ELISA and immunoblotting assays. Proteins in somatic fraction will be separated using 12.5% SDS-PAGE according to the method of Laemmli (1970). After electrophoresis, the resolved polypeptide bands will be either revealed by silver staining (Anuracpreeda et al., 2013) or electrophoretically transferred onto nitrocellulose membranes for immunoblotting. Specific MoAb class and subclass are determined by enzyme immunoassay using Mouse Typer Sub-Isotyping Kit (Bio-Rad, USA). MoAbs were used for localization of the antigenic molecules in the parasites' tissues by immunoperoxidase technique as previously described by Anuracpreeda et al. (2014).

Results and Discussion

Seven stable clones, namely PA01, PA02, PA03, PA04, PA05, PA06 and PA07 were obtained from the hybridoma cells. It was found to be IgG₁ and λ light chain isotypes. All of these MoAbs exhibited no cross-reactions with other parasites' antigens. For immunolocalization, *F. gigantica* antigens were present in the caecal epithelium and lumen of the fluke (Figure 1). The MoAbs that we produced is quite specific to *F. gigantica* somatic antigens and exhibited no cross-reactivity with antigens in other trematode and non trematode parasites. This implied that these MoAbs might bind only to a common epitope, which is present exclusively in *F. gigantica*.

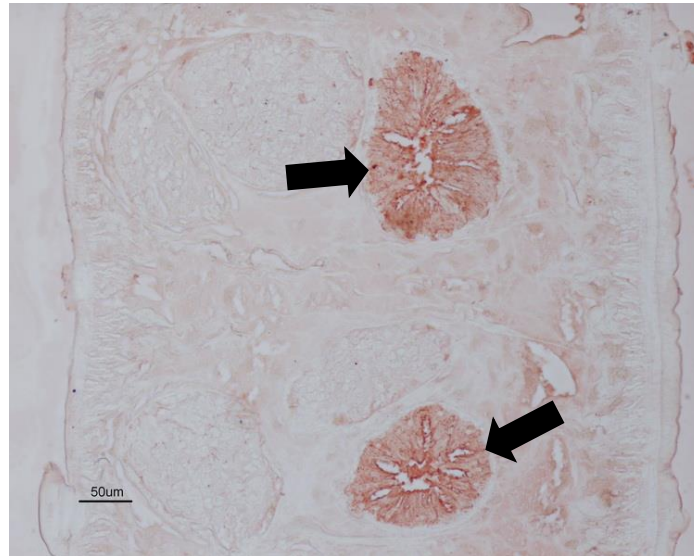


Figure 1. Anatomical localization of the *F. gigantica* antigenic target recognized by MoAb showed intense staining in the caecal epithelium and in the lumen of the caecum (arrows).

Conclusions

Hence, it is possible that these MoAbs could be a good candidate for immunodiagnosis of fasciolosis in both ruminant and human.

References

- Anuracpreeda, P., R. Chawengkirtikul, Y. Tinikul, J. Poljaroen, C. Chotwiwatthanakun and P. Sobhon, 2013. Diagnosis of *Fasciola gigantica* infection using a monoclonal antibody-based sandwich ELISA for detection of circulating cathepsin B3 protease. *Acta. Trop.* 127: 38-45.
- Anuracpreeda, P., T. Srirakam, S. Pandonlan, N. Changklungmoa, C. Chotwiwatthanakun, Y. Tinikul, J. Poljaroen, K. Meemon and P. Sobhon, 2014. Production and characterization of a monoclonal antibody against recombinant cathepsin L1 of *Fasciola gigantica*. *Acta. Trop.* 135: 1-9.
- Laemmli, U. K., 1970. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. *Nature* 227: 680-685.
- Spithill, T. W., P. M. Smooker and D. B. Copeman, 1999. *Fasciola gigantica*: epidemiology, control, immunology and molecular biology. In: Dalton, J.P. (Ed.), *Fasciolosis*. CABI Publishing, Oxon, pp. 465-525.

Vitrification of mouse embryos: comparison of Cryotop and hemi straw closed system methods

Aniruth, A.¹, Y. Liang¹, T. Yoisungnern¹, T. Vutyavanich² & R. Parnpai¹

¹Embryo Technology and Stem Cell Research Center and School of Biotechnology, Suranaree University of Technology, Nakhonratchasima 30000, Thailand.

²Department of Obstetrics and Gynecology, Faculty of Medicine, Chiang mai University, Chaingmai 50200, Thailand.

Introduction

Vitrification is a process by which the cells are frozen rapidly without the formation of intracellular ice crystals and represent a milestone in cryopreservation techniques. Disruption of intracellular organelles, as well as formation of intracellular ice crystal, is a major hazard of the freezing process, which may result in loss of cell viability. In 2005, Cryotop was reported and showed high survival rates after warming, at 90.8% survival rate, 89.6% fertilization rate, a pregnancy rate of 41.4% per transfer and a live-birth and ongoing-pregnancy rate per transfer of 10/29 (34.5%). Cryotop vitrification method is deemed as a “new benchmark” for mammalian embryos cryopreservation. However, the rapid cooling rates of vitrification are achieved by placing the embryos in small volumes of media containing cryoprotectants and exposing them directly to liquid nitrogen (LN₂) as opened system. There was the potential risk of disease transmission through contaminated LN₂ or metal surfaces of the instruments used during freezing and storage. In 2009, Vutyavanich and colleagues reported a modified, low cost, closed-system solid surface vitrification (SSV) that showed high survival rate (95.7%), blastocyst formation rate (79.3%). The objective of this study was to compare the survival rate and the developmental potential of mouse embryos in various stages when vitrified-warmed by Cryotop and hemi straw closed system (HS-CS) methods.

Materials and methods

Piezo- ICSI procedure

Motile sperm were suspended in 50 µL of 7 %PVP medium, in a 90 mm culture dish (as ICSI dish) covered with mineral oil. Mature eggs (MII) of outbred ICR mice, were loaded into KSOM medium drop in ICSI dish. The ICSI method by Piezo micromanipulator was performed as previously described (Yoshida et al., 2007).

In vitro embryo culture

After ICSI, injected fresh oocytes were cultured for 96-100 h in KSOM medium, at 37°C with an atmosphere of 6 %CO₂ in air as control group. Embryo development was recorded at 26 h (2-cell), 38 h (4-cell), 50 h (8-cell), 62 h (morula), 80 h (early blastocyst) and 100 h (expanded blastocyst) after ICSI. Part of the 2-cell, 4-cell, 8-cell, morula and early blastocyst stage ICSI-derived embryos from fresh oocytes were vitrified by either Cryotop or HS-CS methods as described below.

Embryos vitrification

Five good quality embryos were selected and transferred into the equilibration solution of D-PBS containing 7.5% EG and 7.5% Me₂SO for 5 min and then exposed to the vitrification solution of D-PBS containing 15% EG, 15% Me₂SO and 0.6 M sucrose for 30 s. then 5 embryos per groups were loaded by either Cryotop or HS-CS groups.

Embryos warming

The vitrified embryos from either Cryotop or HS-CS groups were warmed by directly immersing the tip of Cryotop into 3 mL of 1.0 M sucrose in D-PBS supplemented with 20% FBS for 5 min, and then transferring to 0.5 M, 0.25 M and 0 M sucrose in D-PBS supplemented with 20% FBS for 5 min interval. The survived embryos were washed 5 times in KSOM medium and cultured in KSOM medium at 37°C in humidified atmosphere of 6 %CO₂ in air.

Differential staining of embryos

Differential staining of the expanded blastocysts was performed as previously described (Thouas et al., 2001), with slight modifications. Briefly, expanded blastocysts were treated with 0.1 mg/mL propidium iodide (PI) and 0.2% Triton X-100 dissolved in D-PBS for 35 s. After that, the expanded blastocysts were treated with 25 µg/mL Hoechst 33342 in 99.5 %ethanol for 4 min, and mounted on glass slides in a glycerol droplet.

Results and discussion

This study was conducted to determine the effect of vitrification at various embryo stages (2-cell, 4-cell, 8-cell, morula and early blastocyst stages) on embryo survival and further development. While vitrified embryos were compared with two different methods of Cryotop and HS-CS methods.

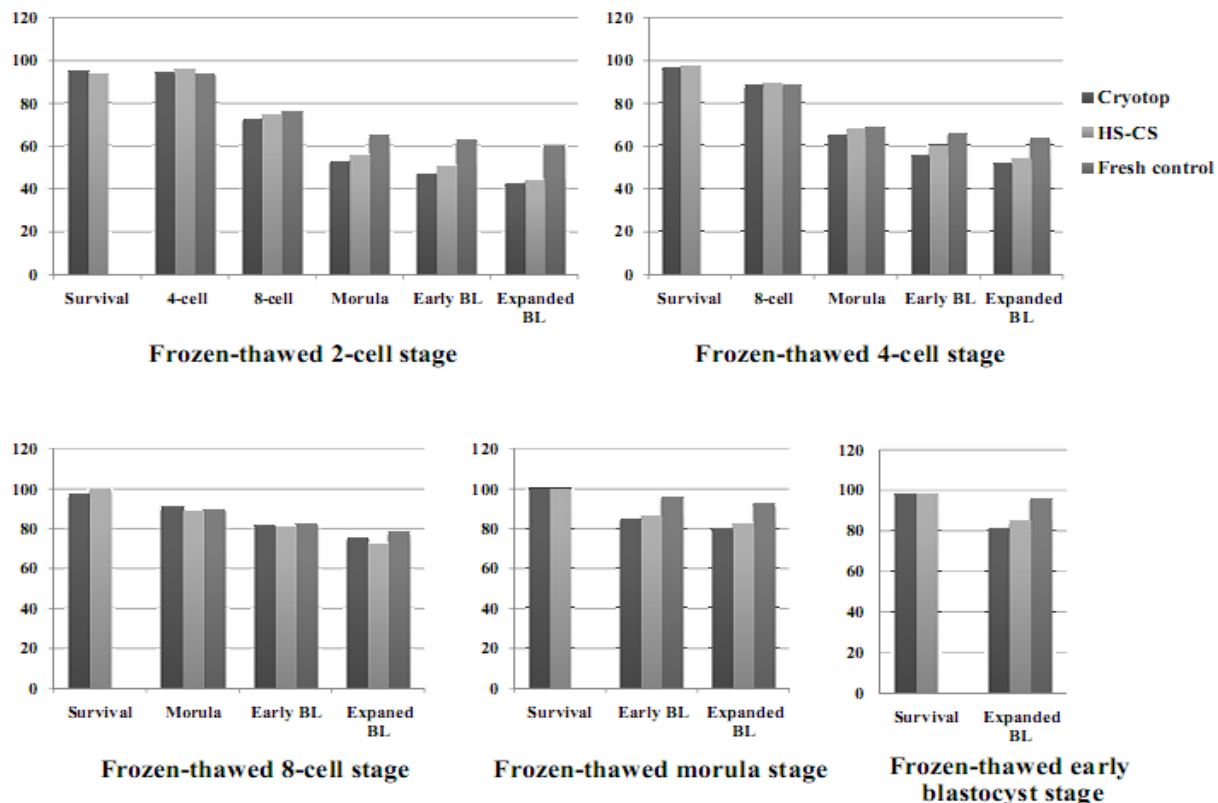


Figure 1. The survival rate and embryos development from frozen-thawed embryos at various stages by compared Cryotop method, HS-CS method with fresh control.

Our data showed no significant difference in survival rate and embryo development rate after thawing between the Cryotop and the HS-CS methods from different stage of vitrified embryos (Figure 1). The total cell (TC) and the ICM/TE ratio are well-established parameters for evaluating the quality of blastocyst. Our results showed that the quality and developmental competence of vitrified-warmed derived expanded blastocysts were not significantly different among embryos vitrification groups at 2-cell, 4-cell, 8-cell, morula and early blastocyst stages by compared Cryotop method, HS-CS method with fresh control (Table 1). In conclusion, The HS-CS method is an effective method for mice embryo vitrification in various stages as well as Cryotop, which is simple to operate, inexpensive to establish and also eliminates or minimize the risk of contamination.

Table 1. Total cell number, Trophectoderm (TE) and Inner cell mass (ICM) cell counts and their ratios (ICM/TE) in expanded blastocyst embryos after vitrification at various embryo stages and in non-vitrified fresh controls.

Groups	Stage of embryo frozen/thawed	Number of blastocyst	TE (Mean±S.D.)	ICM (Mean±S.D.)	TC (Mean±S.D.)	ICM/TE (Mean±S.D.)
Cryotop	2-cell	10	47.1±10.6	21.0±4.7	68.1±12.1	0.46±0.13
HS-CS	2-cell	10	50.1±18.1	21.2±4.2	71.3±19.9	0.47±0.17
Control	2-cell	10	57.3±17.3	28.2±5.3	85.5±20.6	0.52±0.14
Cryotop	4-cel	10	45.4±10.6	20.8±4.9	66.2±14.1	0.46±0.10
HS-CS	4-cell	10	46.4±13.8	21.5±5.4	67.9±19.0	0.47±0.07
Control	4-cell	10	57.3±17.5	28.1±5.5	85.4±20.6	0.52±0.14
Cryotop	8-cell	10	48.6±13.2	22.4±4.9	71.0±17.1	0.48±0.12
HS-CS	8-cell	10	47.4±8.8	22.8±6.6	70.2±13.2	0.48±0.12
Control	8-cell	10	57.8±17.2	28.6±5.5	86.4±20.6	0.52±0.14
Cryotop	Morula	10	58.2±18.6	25.8±5.8	84.0±22.3	0.47±0.13
HS-CS	Morula	10	56.3±9.3	27.4±6.8	83.7±15.0	0.49±0.09
Control	Morula	10	58.3±17.1	29.2±5.3	87.5±20.4	0.53±0.14
Cryotop	Blastocyst	10	59.3±17.4	28.2±5.3	87.5±21.3	0.50±0.15
HS-CS	Blastocyst	10	60.4±16.6	29.7±6.7	90.1±22.3	0.51±0.12
Control	Blastocyst	10	58.3±17.3	29.1±5.5	87.4±20.8	0.53±0.14

Four replications were performed.

There was no significant difference among these groups, $P \geq 0.05$, (ANOVA).

References

- Kuwayama, M., Vajita, G., Ieda, S., and Kato, O. 2005. Comparison of open and closed method for vitrification of human embryos and the elimination of potential contamination. *Reproductive Biomedicine Online* 11: 608-614.
- Thouas, G.A., Korfiatis, N.A., French, A.J., Jones, G.M., and Trounson, A.O. 2001. Simplified technique for differential staining of inner cell mass and trophectoderm cell of mouse and bovine blastocysts. *Reproductive Biomedicine Online* 3: 25-29.
- Vutyavanich, T., Sreshthaputra O., Piromlertamorn, W. and Nunta, S. 2009. Closed-system solid surface vitrification versus slow programmable freezing of mouse 2-cell embryos. *Journal of Assisted Reproduction and Genetics* 26: 285-290.
- Yoshida, N. and Perry, A.C.F. 2007. Piezo-actuated mouse intracytoplasmic sperm injection (ICSI). *Nature Protocol* 2: 296-304.

Impact of complete feed silage from sugar cane waste product on Bali beef cattle performance

Adrizal¹, Montesqrit¹ & Asmi Abas²

¹Animal Science Faculty, Andalas University, Kampus Limau Manis Padang Tel (0751)71464.
E-mail: faterna@unand.ac.id

²Economic Faculty, Andalas University, Kampus Limau Manis Padang, Tel (0751)71088.
E-mail: fekon@unand.ac.id

Abstract

The research objective was to study the impact of using complete feed silage from sugar cane waste product which supplemented by other feed stuffs on Bali beef cattle performance. The experiment tested two kinds of diet ie: the existing diet that has been applied by the farmers (A) and complete feed silage from sugar cane waste product (B). It was used 14 heads of Bali bull with 1.5 to 2.0 years old, body weight 135 to 155 kg. The experiment was conducted on CV Gemini farm in Sijunjung Regency, West Sumatra Province Indonesia on 5 July to 27 September 2014. The result show that the average daily gain (ADG) of B (0.62 kg) was higher than A (0.24 kg). Daily feed intake (DFI) as fed basis for A and B was 17 kg and 13 kg respectively, so the feed efficiency (FE) was 5.8% and 16%. ADG, DFI and FE were significantly difference ($P \leq 0.01$) between the treatments. The technical performance improved the income over feed cost (IOFC). IOFC of A only Rp 1500/head/day, while the B can provide much higher i.e. Rp 8729/head/day. It is concluded that the using of the silage provide good impact on Bali beef cattle fattening.

Keyword: *complete feed silage, Bali cattle, sugar cane waste product*

*Corresponding author: faterna@unand.ac.id

Introduction

Background

In order to improve efficiency of fattening cattle in the rural, it is recommended to use the local feed resources. The alternatives of the local feed resource are waste product of sugar cane that comprise of top sugar cane, bagasse and molasses. One of the sugar cane plantations in West Sumatra (Indonesia) locate in Talang Babungo village, Hiliran Gumanti district, Solok Regency. Sugar cane plantations are managed traditionally since the days of colonization of the Netherlands. At this time the extensive sugar cane plantations in the area approximately 515 Ha. Estimation of the production of sugarcane tops, bagasse and molasses in the area was 4387 tons/year, 11498 tons/year, and 486 tonnes/yr respectively. Based on the availability of waste are expected to be able to support the requirement of fodder for 1500 cattle every year (Adrizal et al., 2012). The problems faced in the sugar cane waste utilization is the fluctuations in the availability, in line with the fluctuations in demand for sugar. Based on the problem, the waste need to be stored in the form of complete feed silage.

Objective of research

The purpose of this research was to examine the effectiveness, efficiency and improvement of farmers income using the complete feed silage made from sugar cane waste product for Bali cattle fattening compared to existing feed.

Materials and Methods

The materials were used 14 Bali bulls 1.5 to 2 years old. The initial weight of the bulls ranged between 143 kg up to 152 kg. The feed materials used are natural grass, sugar cane top, bagasse, molases, palm kernel cake, *Thitonia diversifolia*, *Calliandra eriophylla*, ultra mineral and salts. The feed formula presented in Table 1. The feed A was the existing feed which applied by the farmers, while the B that want to be introduced to increase the income of farmers. Feed B was processed by ensilage procedure and keep it in a plastic bag lined with polyethylene. Each sack is filled with the complete feed of 50 kg.

Experiment on bulls were conducted by dividing the bulls randomly, 7 heads got treatment A and the rest the treatment B. Bulls were placed in random order in 14 compartments of the enclosure. Feed A fed freshly, while B in silage form. Feeding and drinking water were carried out *adlibitum*. Evaluation of feed intake, average daily gain and feed efficiency between the treatments was conducted by testing the comparison of the average of of both sample groups with statistic t test instrument. The economic impact of both the treatment seen by analyzing income over feed cost.

Table 1. The formula of feed treatments.

No	Feed stuff	A		B	
		DM	Asfed	DM	Asfed
1	Sugarcane top			20.0%	23%
2	Bagasse			17.3%	16%
3	Molases			5.0%	14%
4	Natural grass	100%	100%	0.0%	0%
5	<i>Thitonia diversifolia</i>			15.0%	25%
6	<i>Calliandra eriophylla</i>			20.0%	15%
7	Palm kernel cake			19.7%	7%
8	Ultra mineral			2.0%	0.6%
9	salts			1.0%	0.3%
	Total	100%	100%	100%	100%

Results and Discussion

Performance of Beef Cattle

The treatment results are presented in table 2. The table show that average daily gain (ADG) on treatment B highly significant ($P < 0.01$) higher than A, nevertheless dry matter (DM) intake was not different significantly. ADG of bulls that fed traditional feed only 0.24 kg, whereas with the complete feed silage can be reached 0.62 kg. Shelton (2012) also stated that Indonesian local beef cattle that fed traditionally can be reached ADG about 0.15 kg to 0.25 kg, in terms of according to Mastika (2003) it is potential up to 0.76 kg.

Income over feed cost

Cost of traditional feed was much lower than complete feed silage, but the lower ADG cause the income over feed cost decrease. Thus the feed cost savings do not cause income over feed cost better. Otherwise the complete feed silage, although higher feed costs, but the ADG is higher, causing income over feed cost is much higher than on traditional feed.

Table 2. Impact of treatments on performance and income over feed cost of Bali beef cattle.

No	Performance	Treatments	
		A	B
1	The average initial weight (kg)	149	147
2	The average final weight (kg)	169	199
3	The average daily gain (ADG) (kg)*	0.24	0.62
4	The average daily feed intake (DM basis)(kg)	4.2	3.9
5	The average daily feed intake (DM basis)(% body weight)	2.8%	2.7%
6	The average of feed efficiency (%)*	5.8%	16%
7	Feed price (Rp/kg as fed basis)	500	1250
8	The average daily feed cost (Rp)	8,549	16,071
9	Average daily Income over feed cost (Rp)	1,051	8,729

Note:

* Highly significant ($P < 0.01$)

** assuming the price of meat Rp 40.000/kg live weight

Conclusion

Complete feed silage made of sugar cane waste product gives good impact on Bali beef cattle performance both technically and economically.

Acknowledgement

We would like to say thank you very much to DP2M Dikti which funded this research through RAPID program in 2013-2014.

References

- Adrizal, A Suprpto, Mirzah. 2012. *The Potency of Sugarcane Waste Product for Supporting Beef Cattle Feed Resources at Integrated Farming Center in Solok Regency, West Sumatra*. 2nd International Seminar on Animal Industry, Jakarta 5-6 July, 2012; pp 389-393.
- Mastika, I.M. (2003). Feeding Strategies to Improve the Production Performance and Meat Quality of Bali Cattle (*Bos sondaicus*) in Shelton, HM. 2012. *Tropical Forages in Indonesia: Past experience and Future Opportunity*. Proceeding of the 2nd International Seminar on Animal Industry, 5-6 July 2012, Jakarta. pp 64-74
- N.R.C.1976. *Nutrient Requirements of Beef Cattle*. 5th revised edition. National Academy of Science. Washington DC.
- Shelton, HM. 2012. *Tropical Forages in Indonesia: Past experience and Future Opportunity*. Proceeding of the 2nd International Seminar on Animal Industry 5-6 July 2012, , Jakarta pp 64-74.

Development of a rapid immunochromatography test for detecting antibodies after anthrax vaccination in cattle: A preliminary study

Sriasih, M^{*}, A. Rosyidi, E. Yulianti & S. N. Depamede

Microbiology & Biotechnology Laboratory, Faculty of Animal Science, University of Mataram, Jalan Majapahit No. 62. Mataram – Indonesia

Abstract

Anthrax is a zoonotic disease of strategic importance with high mortality rate in ruminants. Vaccination and monitoring of antibody titer after anthrax vaccination have been carried out in an effort to prevent anthrax disease in cattle. Enzyme linked immunosorbent assay (ELISA) is a method commonly used in the monitoring of antibody titer after vaccination. However, the use of the ELISA method requires skilled laboratory personnel, specialized laboratory equipment, and relatively more expensive. This study aimed to develop a method to detect antibodies after anthrax vaccination in cattle using rapid immunochromatography detection. Colloidal gold as a marker were conjugated with protective antigen (PA), with a concentration of 0.2, 0.4 and 0.8mg/ml, then put on the conjugate pads as part of the immunochromatography test strip. A total of 13 serum samples of cattle after vaccination was used in this study. The sample consisted of seven positive and six negative sera samples based on the results of the ELISA test to detect the presence or absence of anti-PA antibodies in serum. The results using the rapid immunochromatography test indicate that the anti-PA antibodies in the serum can be detected within 10min. Antigen concentration of 0.2mg/ml in the conjugate pads showed the same sensitivity test as other antigen concentrations used in this study.

Keywords: anthrax, rapid test, immunochromatography, cattle, protective-antigen

**Corresponding author: madesriasih@yahoo.co.nz*

Introduction

Anthrax, which is caused by the bacterium *Bacillus anthracis*, is one of five strategic contagious animal diseases in ruminants in Indonesia. Siregar (2002) reported that there are still many anthrax endemic areas in Indonesia such as Jakarta, West Java, Central Java, Yogyakarta, West Nusa Tenggara (NTB), East Nusa Tenggara (NTT), West Sumatra, Jambi, Southeast Sulawesi, Central Sulawesi, South Sulawesi and Papua. The mortality rate due to anthrax is very high (at 80%), especially in ruminants such as cattle, sheep and goats. In NTB particularly the islands of Sumbawa, and in particular Flores island of NTT province, the case of anthrax almost repeated every year so that the two islands are classified as areas with high endemicity (Putra et al., 2011).

In an anthrax endemic areas, vaccination and good surveillance are considered as the most appropriate control for anthrax. Antibody responses after vaccination are routinely monitored in vaccine recipients. A quantitative enzyme-linked immunosorbent assay (ELISA) has been developed and used for the serological detection of protective antigen (PA)-specific IgG in cattle. However, the ELISA is impractical in field settings, as it requires laboratory equipment and highly trained personnel.

With advancing technology, the development and commercialization of screening tests are moving toward rapid point-of-care assays (Bienek et al., 2008). Rapid immunochromatography detection is a semiquantitative colorimetric test that is well established for

detecting diseases or monitoring vaccination status. A specimen is suspended in sample buffer and added to the sample pad. The samples flow via capillary action through a conjugate pad and across nitrocellulose membrane that has a test strip and a control strip. If disease-specific antibodies are present in the specimen tested, they will react with the test strip containing the diagnostic target molecule and the result can be visualised after 10 to 15min with the unaided eye.

This study was conducted to determine whether positive reaction occurs in an immunochromatography test strip developed using the serum of cattle following anthrax vaccination.

Materials and Methods

Specimens

Blood samples were collected from 13 cattle after anthrax vaccination from Palibelo (PL) and Labuan Badas (LB) districts in Sumbawa island. Serum was then separated by centrifugation at 5,000 rpm for 15min. Serum samples used for the immuassays were frozen at -20°C until needed. As a gold standard for comparison, an ELISA was used to characterise the specimens by their amounts of PA-specific IgG. Based on ELISA test, seven samples (PL1, PL4, PL22, PL23, PL15, PL55 and PL63) were tested positive and six samples (LB43, LB12, LB6, PL3, PL70 and negative control) were negative.

Antigen source

Recombinant PA (rPA) purchased from Indonesian Research Centre for Veterinary Science was used as the target molecule in the immunoassay. The antigen was then run on 12% acrylamide gel electrophoresis in order to determine its purity.

Preparation of colloidal gold and Rapid Immunochromatography

Colloidal gold probes measuring 40nm were prepared with citric acid by using a modified citric acid reduction method. The rapid immunochromatography test device consists of nitrocellulose membrane, conjugate pad and adsorbent pad. The rPA, with the concentration of 0.2, 0.4 and 0.8mg/ml, were striped in the line test position while a goat anti-mouse antibody was applied on the control test position on nitrocellulose membrane. The membrane was then dried in a controllable drying chamber (Heraeus Instruments, USA) for 60min at 60°C. Colloidal gold conjugated with the rPA (2.5µg/ml) were dispensed onto a conjugate pad, and the pad was then affixed to the test strip by overlapping the nitrocellulose membrane by its proximal end. The addition of a sample pad completed the assembly by overlapping onto the conjugate pad. Test strip, with size of 6cm by 4mm, was produced using Matrix 2360-programable shear (Kinematic Automation, USA). All samples were tested by placing 4µl of sera onto the sample delivery pad. The fluid was drawn across the antigen-coated membrane and within 10min a visible pink dot formed in the control line. A positive result, when anthrax anti-PA antibody is present, is evident by the formation of an additional dot in the test line.

Results and Discussion

With regard to the gel electrophoresis analysis, the rPA used for coating the nitrocellulose membrane in this study were relatively pure (data not shown). Gel electrophoresis analysis also showed that protein with size of approximately 60kDa was the major component. Seven positive and six negative sera, based on the ELISA test, were tested using the rapid immunochromatography developed. The rPA concentration of 0.2mg/ml on the solid phase seems to be sufficient to detect anthrax anti-PA IgG present in the vaccinated cattle. Figure 1 shows the representative of negative (negative control) and positive (sample #PL1) immunochromatography test results.

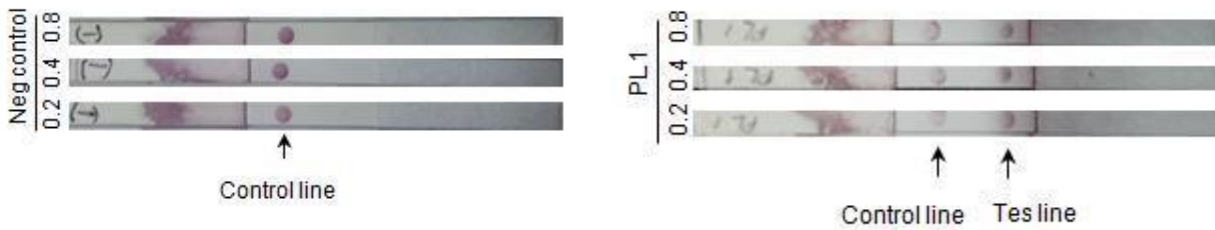


Figure 1. Representative immunochromatography test results

All sera tested positive on the ELISA test showed positive results on the rapid test (Table 1). Whilst, 67% (4 out of 6) sera showed negative on the rapid test were also negative on the ELISA test. The sensitivity and specificity of the rapid test were 100% and 67% respectively.

Table 1. Diagnostic performance of rapid immunochromatography test to PA-specific IgG present in vaccinated cattle in comparison to ELISA test.

ELISA	Sample No	rPA conc on immunochromatography test		
		0.2mg/ml	0.4mg/ml	0.8mg/ml
Negative	Negative cont	-	-	-
	LB43	-	-	-
	LB12	-	-	-
	LB6	-	-	+/-
	PL3	+	+	+
	PL70	+/-	+/-	-
Positive	PL1	++	++	++
	PL4	+	+	+/-
	PL22	+/-	+/-	+/-
	PL23	+	+	+
	PL15	+	+	+
	PL55	++	++	+
	PL63	+	+	+

Note: - (negative); +/- (weak positive); + (strong positive); ++ (very strong positive)

When estimating the specificity of diagnostic test, it is important to ensure that samples should represent the population for which the test is going to be used (Peruski & Peruski Jr., 2003; Muller et al., 2004). The limited number of samples tested in this study may contribute to relatively low level of specificity. The use of the ELISA as a gold standard in this study should also be taken into account as it is also a screening test method. This suggests that further works on validating a good gold standar is necessary.

Anti-PA antibody titres are not frequently measured after anthrax vaccination both in human (Bienek et al., 2008) and animals. The major reason for this is that anti-PA IgG measurements are laboratory based, necessitating transport material to be tested to laboratory, skilled personnel and specific equipment. Therefore, a simple and rapid method would provide an important tool to screen or measure anti-PA antibody after anthrax vaccination. This study indicates that we have successfully developed a strip tes kit that can rapidly detect the anti-PA IgG on cattle sera following vaccination. The results imply that the strip assay is highly sensitive and sufficiently specific to support the detection. The test is easy to operate, may suit for on-site testing and is much quicker than the time required for ELISA. Further study to examine the accuracy of the rapid test with respect to its specificity still needs to be undertaken.

Acknowledgements

This study was funded by the Indonesian Ministry of Research, Technology and Higher Education and supported by Prof. Mulyanto, Director of NTB Hepatitis Laboratory.

References

- Bienek, D.R., R.E. Biagini, D.G. Charlton, J.P. Smith, D.L. Sammons and S.A. Robertson. 2008. Rapid point-of-care test to detect broad ranges of protective antigen-specific immunoglobulin G concentrations in recipients of the US-licensed anthrac vaccine. *Clin Vaccine Immunol.* 15(4): 644-649.
- Muller, J.D., C.R. Wilks, K.J. O'Riley, R.J. Condron, R. Bull and A. Mateczun. 2004. Specificity of an immunochromatographic test for anthrax. *Aust Vet J.* 82(4):220-222.
- Peruski, A.H. and L.F. Peruski Jr. 2003. Immunological methods for detection and identification of infectious diseases and biological warfare agents. *Clin Diagn Lab Immunol.* 10(4): 506-513.
- Putra, A.A.G., H. Scott-Or, H. Widowati. 2011. Anthraks di Nusa Tenggara. Direktorat Jenderal Peternakan dan Kesehatan Hewan Kementerian Pertanian bekerjasama dengan Australian Center for International Agricultural Research.
- Siregar, E. A. 2002. Anthraks: Sejarah Masa Lalu, Situasi Pada Saat Ini, Sejarah Diagnosa dan Kecenderungan Perkembangan Ilmu di Masa Depan. Paper disampaikan pada Simposium Sehari Penyakit Anthraks. Balitvet Bogor 17 Juli 2002.

The Effect of *Cinnamomum burmannii* extract as an immunomodulator on the increase of GR-1 expressing IFN γ and macrophage

Pratiwi TS^{1,*}, Putri A², Murwani S²

¹Animal Husbandry Faculty, University of Brawijaya, Malang, Indonesia, ² Medical Veterinary School, University of Brawijaya, Malang, Indonesia

Abstract

Cinnamomum burmannii was known as one of herbal medicine that has been used traditionally as an ingredient of traditional medicine extract, contains *cinnamaldehyde*, which is a naturally trigger of the body's immune response. This study aimed to evaluate the immunostimulant effect of *C. Burmannii* and the increase of Granulocyte Receptor - 1 (GR-1) that can be recognized from neutrophil expressing IFN γ and the increase of macrophage phagocytosis activities. Thirty wistar mouse were appropriately infected with *Salmonella enteridis* and then orally treated with *C. burmannii* alcohol extract at different dosages. The mouse were observed for the increase of GR-1 expressing IFN γ using flow-cytometry and the increase of macrophage phagocytosis activities using Giemza staining. The result showed that increasing dosage of *C. burmannii* extract treatment increased the GR-1 level and the IFN γ by 97.7% and macrophage phagocytosis activity by 98.1% (P<0.05). Thus, the observations reflected clearly that *C. burmannii* ethanol extract can be utilized as immunomodulator that increased immune response.

Keywords: *Cinnamomum burmannii*, GR-1, IFN γ , macrophage, phagocytosis

*Corresponding author: pratiwi_trisunuwati@yahoo.com

Introduction

Several researches on herbs to enhance immune responses have been carried out, for example research on the effect of extracts of *Hedyotis corymbosa*, *Cassia alata* L leaf extract, *Blumea balsamifera* on phagocytic activity of macrophages *Mus musculus* strain BALB/c that were infected by *Salmonella typhimurium*, *Listeria monocytogenes* on the effect of immunomodulatory on macrophage phagocytic potential (Kusmardi, 2007; Munawaroh, 2008) However, research on the immunomodulatory effects of ethanol extract *Cinnamomum* of *Cinnamomum burmannii* to increase phagocytic activity of interferon gamma and macrophages on *Mus musculus* strain BALB/c has not yet been done before and needs further research. *Burmannii cinnamon* of *Cinnamomum* species is one of the medicinal plants that are often found in regions of Indonesia (Gunawan and Mulyani, 2004), contains some compounds that are beneficial to health including drug efficacious for gout, high blood pressure. Arrar (2009) reported that *C. burmannii* was also proven to be antibacterial in *Bacillus cereus*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Helicobacter pylori*, *Salmonella typhimurium*, and *Escherichia coli*. Active compounds contained in cinnamon is essential oil, safrole, sinamadehid, eugenol, tannins, resins, calcium oxalate, tanning agents, flavonoids, saponins and other nutritional content such as sugar, protein, crude fat and pectin which allegedly helped in immune response (Gunawan and Mulyani, 2004; Wang, 2009). Ramchandra (2006) reported that cinnamon of *Cinnamomum zeylanicum* species at a dose of 100 mg/kg had immunostimulatory effects in animal models of *Mus musculus*.

Material and Methods

Thirty male wistar BALB/c (SPF) were appropriately infected with *Salmonella enteritidis* at a dose of $0.2 \text{ ml} \times 10^8 \text{ ml/CFU}$ and then orally treated along 14 days with *C.burmannii* ethanol (high polar) extract at different dosages i.e 0, 50, 100,150 and 200 mg/kg body weight (P0, P1, P2,, P3, P4). Samples of peritoneal fluid and spleen tissue were collected, one drop of peritoneal fluid sample as a thin smear Giemza staining test to evaluate the number of active macrophages in activity phagocytosis. While the mouse were observed for the increase of *GR-1* expressing *IFN γ* using flow-cytometry (Rifa'i et al., 2004). Analysis of data on the number GR-1 that express *IFN γ* and phagocytic activity of macrophages using one-way ANOVA.

Results and Discussion

Average number of CD4 that expressed *IFN γ* (bound to the extracellular antibody GR-1) and active macrophage cells that performed phagocytosis in *Salmonella enteritidis* non-infected and infected and cinnamon (*C. burmanii*) ethanol extract non-treated and treated animals were presented in Table 1. It was shown that mice treated using *C. burmanii* ethanol extract showed significantly higher ($p < 0.05$) average number of CD4 that expressed *IFN γ* compared to those in either the negative control treatment or positive control treatment (Table 1 and Figure 1). While, positive control treatment (P0+) showed also significantly higher ($p < 0.05$) average number of CD4 that expressed *IFN γ* than negative control treatment (P0-). The data were in line with research conducted by Abbas et al, (2003). They indicated that number of neutrophil cells (CD4) and *IFN γ* was higher in animals infected with *Salmonella enteritidis* than in non-infected.

Table 1. Average number of CD4 and active macrophage cells that performed phagocytosis in *Salmonella enteritidis* non-infected and infected and cinnamon (*C. burmanii*) ethanol extract non-treated and treated animals

Treatment	CD4 (cell)	Active macrophage (cell)
P0-	306.081,25 \pm 5.690,46 ^a	43.50 \pm 3.69 ^a
P0+	1.520.512,50 \pm 67.915,16 ^b	57.50 \pm 2.64 ^b
P1+	1.888.450,00 \pm 272.411,00 ^b	65.25 \pm 2.62 ^c
P2+	2.458.481,25 \pm 30.0534,49 ^c	76.75 \pm 3.86 ^d
P3+	3.105.168,75 \pm 301.001,89 ^d	84.50 \pm 1.29 ^e
P4+	3.938.531,25 \pm 65.507,98 ^e	91.75 \pm 3.77 ^f

In addition, interferon gamma (*IFN γ*) regulate gene expression in a number of neutrophils including complement receptor regulator, B lymphocyte stimulator, dendrites chemotactic factors, chemokines receptors, neutrophil chemotactic factors and pro-inflammatory cytokines (Rifa'i, et al., 2004). Sinamaldehyd inhibits the release of ROS and activates MAPKs as an expression of pro-inflammatory cytokines. Finally express highly increased against GR-1(*IFN γ*) and the number of active macrophage cells and regulate gene expression against complement regulator B cells and proinflammatory cytokines. It was also shown in Table 1 that average active macrophage cells that performed phagocytosis increased linearly ($P < 0.05$) with the increase of level of cinnamon (*C. burmanii*) ethanol extract treatment. Phagocytic activity of macrophages from peritoneal fluid was also tested using Giemsa staining smears test, showed a different colour and structures.



Figure 1. IFN γ as measured (flowcytometry) of mice treated *C. Burmannii* ethanol extract showed significantly higher ($p < 0.05$) than those in either the negative control treatment or positive control treatment

Conclusion

C. burmannii extract treatment on *S. enteritidis* infected M musculus BALB/c have potential as immunostimulatory as characterized by the increase of GR-1 expressing IFN γ and the number of active phagocyte macrophage cells.

References

- Abbas, A.K., K.M. Murphy and A. Sher. 2003. Functional diversity of helper T lymphocytes. *Nature*. 383: 787 – 793.
- Araar, H. 2009. Cinnamon Plant Extracts : a Comprehensive Physico-Chemical And Biological Study For Its Potential Use as a Biopesticide. Master of Sci in Mediterranean Organic Agriculture. Istituto Agronomico Mediterraneo di Bari
- Gunawan, D dan S. Mulyani. 2004. Komponen Penyusun Minyak Atsiri Beberapa cinnamomum. Prosid Pengembangan Riset dan Teknologi Bahan Obat Alami dan Rangka Peningkatan Sumber Daya Manusia. Simposium Penelitian Bahan Obat Alami VIII. Perhimpunan Penelitian Bahan Obat Alami dan Balitro. Bogor.
- Kusmardi., S. Kumala dan E. E. Triana. 2007. Efek Imunomodulator Ekstrak Daun Ketepeng Cina (*Cassia alata* L.) Terhadap Aktivitas dan Kapasitas Fagositosis Makrofag. *Makalah Kesehatan*, (11)2 : 50-53.
- Munawaroh, F., Sudarsono dan A. Yuswanto. 2008. Pengaruh Pemberian Ekstrak Fenolik Daun Sembung untuk Melihat Adanya Peningkatan Aktifitas Makrofag pada Mencit Jantan yang Diinfeksi dengan *Listeria monocytogenes*. Fakultas Farmasi. Universitas Gadjah Mada Yogyakarta.
- Ramchandra, N. S. 2006. Immunomodulatory Activity of Cinnamon Bark (Dissertation). School of Pharmacology. Rajiv Gandhi University of Health Science Karnataka Bangalore.
- Rifa'i, M., Y. Kawamoto., I. Nakashima and H. Suzuki. 2004. Essential Roles of CD8 CD122 Regulatory T Cells in the Maintenance of T Cell Homeostasis. *Journal Exp. Med.* 200(9):1123-1134
- Wang, R and B. Yang. 2009. Extraction of Essential Oils from Five Cinnamon Leaves and Identification of Their Volatile Compound Compositions. *Innovative Food Science and Emerging Technologies*, 10 : 289–292

Genetic variation of MHC Class II DRB3 gene in local goat from South Sulawesi Indonesia

Muhammad Ihsan Andi Dagong¹, Lellah Rahim¹, Rr Sri Rachma Aprilita Bugiwati¹ & Nurul Purnomo²

¹Animal Breeding and Genetic Laboratory, Animal Husbandry Faculty, Hasanuddin University, Tamalanrea Makassar 90245, Indonesia, ²Integrated Biotechnology Laboratory, Animal Husbandry Faculty, Hasanuddin University, Tamalanrea Makassar 90245

Abstract

Major histocompatibility complex (MHC) class II proteins are primarily restricted to the surface of immune cells and are responsible for immune regulation. The objective of this study is to identify the genetic variation of MHC class II DRB3 genes in Indonesian local goats from South Sulawesi province. A total of 113 blood samples were collected from three local goats (Boerawa, Kacang and Peranakan Ettawa) in South Sulawesi province. The genomic DNA was extracted by using Genomic DNA extraction Kit and then MHC Class II DRB3 gene were amplified by PCR with predicted amplicon length 285 bp. To identify alleles variation of MHC gene, the PCR products were cut with *Hae*III restriction enzymes. Genetic variation between populations calculated based on genotypic and allelic frequencies, observed heterozygosity (H_o), expected heterozygosity (H_e) and the Hardy-Weinberg equilibrium. There were 6 alleles found in local goat population (A, B, C, D, E and F alleles). C and F were the common alleles found in this population (0.278 and 0.269 respectively), while the rare allele was E allele (0.022). Genotype frequencies of this gene were respectively AA (0.018), AB (0.053), BB (0.009), AC (0.142), BC (0.088), CC (0.106), AD (0.080), BD (0.018), CD (0.009), DD (0.009), AE (0.018), BE (0.0), CE (0.009), DE (0.0), EE (0.0), AF (0.177), BF (0.053), CF (0.097), DF (0.0), EF (0.018) and FF genotype (0.097). The observed heterozygosity value (H_o) was 0.761 while the expected heterozygosity (H_e) was 0.771. Allelic variation found in local goat population could be used as genetic information in selection program for disease resistant.

Keywords: genetic variation, MHC Class II DRB3, local goat, disease resistant, South Sulawesi

Introduction

Goats have an important role in moving the economy in rural areas. Local goats commonly maintained by rural communities in Indonesia are Kacang and Peranakan Ettawa, as well as some types of goats from crossbreed as Boerawa which is a hybrid between Peranakan Ettawa and Boer goats. Kacang and Peranakan Ettawa goats as local genetic resources, is very important because of their economic characteristics and capabilities to diseases resistance.

The major histocompatibility complex (MHC) is an important group of genes that play a vital role in immune systems. MHC genes are divided into three classes; class I and II genes exhibit most genetic variation (Trowsdale, 1996; Baghizadehet al., 2009). The polymorphism of these genes could affect each individual to respond differently to specific antigens. Therefore, the MHC gene may play a role in determining the durability and vulnerability of each individual in a population against specific diseases. The objective of this research was to identify polymorphisms of the MHC DRB3 gene at exon 2 region of local goats from South Sulawesi province Indonesia.

Materials and Methods

Samples Collection and DNA Extraction

A total of 113 samples were collected from Boer, Kacang and Peranakan Ettawa goats from South Sulawesi Province, Indonesia. Blood samples were collected from the jugular vein, and DNA was isolated with Genomic DNA extraction kit (Gene JET Genomic DNA purification kit)

Gene Amplification

The following primers were used to amplify a 285 bp of MHC DRB3 gene in exon 2 region of Caprine (CLA): F: 5'-TATCCCGTCCTCTGCAGCACATTTC-3' and R: 5'-TCGCCGCTGCACACTGAAACTCTC-3'. PCR reaction were performed in 25 µl aliquots containing ~100 ng of DNA template, 0.25 mM of each primer, 150 µM dNTP mix, 2.5 mM MgCl₂ and 0.5 U Taq DNA polymerase with 1x buffer. The PCR condition was performed with an initial denaturation 94°C for 2 min, followed by 35 cycles of denaturation at 94°C for 45 s, annealing at 64°C for 60 s, extension at 72°C for 60 s and final extension at 72°C for 5 min. The PCR products checked by electrophoresis in 1.5% agarose gel, and stained with EtBr then visualized under UV light.

MHC DRB3 Alleles Identification

The PCR products were digested with *Hae*III restriction endonucleases for 18 h at 37°C. Restriction products were subjected to electrophoresis in 8% of Polyacrylamide gel at 250 V for 2 h in 0.5x TBE buffer. Gels were stained based on the method of Byun et al., 2009 with modification in staining solution (0.1% AgNO₃, 0.04% NaOH 10 N and 0.4% NH₃).

Data Analysis

The genotype and allele frequencies were calculated based on Nei & Kumar (2000) formulation.

$$X_{ii} = \frac{n_{ii}}{N} \times 100\% \text{ and } X_i = \frac{\left(2n_{ii} + \sum_{j \neq i} n_{ij}\right)}{2N},$$

where X_{ii} = i^{th} genotype frequency, X_i = i^{th} allele frequency, n_{ii} = number of sample of i^{th} genotype, n_{ij} = number of sample of ij^{th} genotype, and N = total sample.

The test of Hardy-Weinberg equilibrium (HWE) with chi-square test (Kaps & Lamberson, 2004). $\chi^2 = \sum (\text{Obs} - \text{Exp})^2 / \text{Exp}$.

where χ^2 = chi-square, Obs = number of observation of i^{th} genotype, and Exp = number of expected of i^{th} genotype.

Observed (H_o) and Expected heterozygosity (H_e) based on Nei's heterozygosities (1973) and computed using PopGene32 software version 1.31 (Yeh et al., 1999).

$$H_o = \sum_k^s w_k \sum_{i \neq j}^q X_{kij} \text{ and } H_e = 1 - \sum_k^s w_k \sum_i^q x_{ki}^2.$$

where H_o = observed within-population heterozygosity, H_e = expected within-population heterozygosity, w_k = relative population size, X_{kij} ($i \neq j$) = the frequency of $A_i A_j$ in the k^{th} population.

Results and Discussion

This study shows a highly genetic diversity in the MHC locus DRB3 on local goat in Indonesia. Amplification of MHC DRB3 gene was targeted an amplicon with approximately 285 bp in length and based on PCR-RFLP results, identified about 6 types of alleles and 21 genotypes in the MHC DRB3 gene. The six types of alleles were allele A with restriction pattern 146 bp/75 bp/64 bp, B (163 bp/75 bp/47 bp), C (146 bp/120 bp/19 bp), D (154 bp/131 bp), E (220 bp/65 bp)

and F (154 bp/75 bp/37 bp/19 bp). This finding was similar to the other reports on domestic goat breeds (Li et al., 2006 and Zhao et al., 2011). Li et al (2004) found a total of 6 alleles and 18 genotypes in 12 Chinese indigenous goat population, while Zhao et al (2011) reported 8 alleles and 13 genotypes in 10 domestic goats in Southwest China.

Table 1. Alleles Frequency, Observed (H_o) and Expected (H_e) Heterozygosity of MHC DRB3 in Indonesian Local Goat.

Population (n)	Alleles Freq.						Heterozygosity		X^2
	A	B	C	D	E	F	H_o	H_e	
Indonesian Local Goat (113)	0.252	0.115	0.278	0.061	0.022	0.269	0.761	0.771	27.90

note :n = number of samples (head); H_o = Observed heterozygosity, H_e = Expected heterozygosity; X^2 = chi-square for H-W equilibrium.

Based on the alleles frequencies, the C (0.278), F (0.269) and A (0.252) allele were the common alleles in Indonesian local goat. The total amount frequency of these three alleles were 0.799. While the rare allele were E (0.022) and D (0.061) alleles (Table 1). AF, AC and CC were the common genotypes found in this population, and the rare genotypes were BB, CD, DD and CE. While, BE, DE, EE and DF genotypes were not found in this study (Table 2). The data presented in this study contain informative finding on the diversity of MHC gene in Indonesian local goat from South Sulawesi province. This finding also indicated that local goat in this area may vary in their genetic potential for the immune response or diseases resistance.

Table 2. Genotype Frequency of MHC DRB3 in Indonesian Local Goat.

Locus MHC DRB3		
Genotype	Observed (O)	Expected (E)
AA	1.80	7.09
AB	5.30	6.58
BB	0.90	1.44
AC	14.20	15.96
BC	8.80	7.28
CC	10.60	8.68
AD	8.00	3.54
BD	1.80	1.61
CD	0.90	3.92
DD	0.90	0.40
AE	1.80	1.26
BE	0	0.57
CE	0.90	1.40
DE	0	0.31
EE	0	0.04
AF	17.70	15.45
BF	5.30	7.04
CF	9.70	17.08
DF	0	3.79
EF	1.80	1.35
FF	9.70	8.13

The observed heterozygosity (H_o) of Indonesian local goats were 0.761, slightly lower compared with expected heterozygosity (H_e). Based on chi-square test, indicated that alleles frequencies of MHC DRB3 gene in this population were not in Hardy-Weinberg equilibrium. This suggests that selection process has altered the MHC pool in local goat population in South Sulawesi province.

Conclusion

Results of this study indicate that the allele variation in MHC DRB3 gene found in the local goat from South Sulawesi province Indonesia, could be used as genetic information. Further research can be done to find an association of the MHC DRB3 with immune response or disease resistant for selection program.

References

- Baghizadeh, A., M. Bahaaddini, M. R. Mohamadabadi and N. Askari, 2009. Allelic variations in exon 2 of Caprine MHC Class II DRB3 Gene in Raeini Cashmere goat. *Am-Eurasian J Agric. Environ. Sci.* 6: 454-459.
- Byun, S.O., Q. Fang, H. Zhou and J.G.H. Hickford, 2009. An effective method for silver-staining DNA in large numbers of polyacrylamide gels. *Anal. Biochem.* 385: 174–175.
- Kaps, M. and W.R. Lamberson, 2004. *Biostatistic for Animal Science*. CABI Publishing, London.
- Li, M.H., K. Li, J. Kantanen, Z. Feng, B. Fan and S.H. Zhao, 2006. Allelic variations in exon 2 of caprine MHC class II DRB3 gene in Chinese indigenous goats. *Small Rumin. Res.* 66: 236-243.
- Nei, M. and S. Kumar, 2000. *Molecular Evolution and Phylogenetics*. Oxford University Press, New York.
- Trowsdale, J., 1996. Molecular genetics of HLA class I and class II regions. In: Browning MJ, McMichael AJ (eds) *HLA and MHC: genes, molecules and function*. BIOS Scientific, Oxford. pp: 23-38.
- Yeh, F.C., R.C. Yang and T. Boyle, 1999. *POPGENE version 1.31 : Microsoft Window-based Freeware for Population Genetic Analysis*. Edmonton, AB. Canada : University of Alberta Canada.
- Zhao, Y., H. Xu, L. Shi and J. Zhang, 2011. Polymorphisms in Exon 2 of MHC Class II DRB3 Gene of 10 Domestic Goats in Southwest China. *Asian-Australas. J. Anim. Sci.* 24: 752–756.

The relationship between longevity and reproductive efficiency in Lori-Bakhtiari ewes

M. Vatankhah¹

¹ P. O. Box 415, Department of Animal Science, Agriculture and Natural Resources Research Center, Shahrekord, IRAN.

Abstract

The phenotypic, genetic and environmental correlations between longevity and reproductive efficiency traits over a lifetime in Lori-Bakhtiari ewe's were estimated using the data set consisted of 8202 records of reproductive traits in a lifetime of 2478 ewes collected from 1989 to 2012 inclusive from a Lori-Bakhtiari research flock at Shooli station in the southwestern part of Iran (Shahrekord). Reproductive efficiency over all consecutive lambing opportunities were calculated by adding the total reproductive efficiency traits per ewe joined for all of the parturition opportunities. The reproductive efficiency included, average conception rate (ACR), number of parity (NP), total number of lambs born (TNLB), total number of lambs weaned (TNLW), total of lambs birth weight (TLBW) and total of lambs weaning weight (TLWW). The data set analyzed with multi-trait animal model included the fixed effects birth year of ewe, number of parturition in ewe's lifetime, ewe body weight as covariate and random effects direct additive genetic and residual effects. The estimates of genetic correlations between longevity and ACR, NP, TNLB, TNLW, TLBW and TLWW were 0.17 ± 0.13 , 0.96 ± 0.01 , 0.86 ± 0.03 , 0.88 ± 0.03 , 0.89 ± 0.03 and 0.90 ± 0.02 , respectively. The estimates of phenotypic correlations between longevity and ACR, NP, TNLB, TNLW, TLBW and TLWW were -0.01 ± 0.03 , 0.92 ± 0.01 , 0.77 ± 0.01 , 0.78 ± 0.01 , 0.84 ± 0.01 and 0.82 ± 0.01 , respectively. The environmental correlations between longevity and total reproductive efficiency traits over ewe's lifetime were lower than phenotypic correlations. Thus, improving longevity by genetic selection could make significant improvements in reproductive efficiency traits over ewe's lifetime.

Keywords: Reproductive efficiency, Ewe's lifetime, Genetic parameters.

Introduction

Reproduction and survival rate are key factors determining the efficiency of lamb production in any environment (Snyman et al., 1997). Improving reproductive performance increases productivity of the breeding ewe unit, more efficient use is made of available feed, more surplus animals are available for sale and greater selection pressure is available, increasing the potential for genetic gains. Different strategies may be applied to increase sheep productivity: frequent lambing and high prolificacy of ewes or rapid growth rate of lambs. The lifetime ewe efficiency reported as different indices such as total of lambs born and weaned in a lifetime, total of weight of lambs weaned in a lifetime and the ratio of lamb's weight to ewe weight in some breed of sheep (Qureshi, 1997; Duguma et al., 2002; Eküz et al., 2004; Lee et al., 2009; Mishra et al., 2009). Thus, reproductive performance per ewe exposed and ewe's lifetime are economically important and when selection of breeding animals takes place, both should be considered in a breeding program. However, to incorporate aspects of lifetime into a breeding objective, data must be available to measure the length of time a sheep stays in the flock in order to obtain

estimates of genetic parameters for longevity. Furthermore, any genetic or environmental relationships that exist between ewe's life time and reproductive performance should be incorporated into a multiple-trait evaluation program for optimal response to selection for economically important traits in a breeding program. The objective of this study was to evaluate the relationships between longevity and ewe's reproductive performance to improve profitability in rearing sheep under village system.

Material and Methods

The data set used in this study consisted of 8202 records of reproductive traits in a lifetime of 2478 ewes collected from 1989 to 2012 inclusive from a Lori-Bakhtiari research flock at Shooli station in the southwestern part of Iran (Shahrekord). The flock is managed under semi-migratory or village system (Vatankhah, 2013). The traits evaluated in this study were ewe's longevity and reproductive efficiency over the lifetime of ewes, by adding up the total number of lambs weaned per ewe joined on all of consecutive lambing parturition opportunities (1 to 8). Ewes that failed to a lamb or did not wean a lamb, received zero for that year. The cumulative reproduction traits were evaluated for each ewe exposed to the ram during her lifetime included, average conception rate (ACR), number of parity (NP), (0 to 8), total number of lambs born per ewe (TNLB), (0 to 10), total number of lambs weaned per ewe (TNLW), (0 to 10), total of lamb birth weight (TLBW) and total of lamb weaning weight (TLWW). Covariance components and correlations between longevity and reproductive traits in a ewe's lifetime were estimated using the restricted maximum likelihood method (WOMBAT program of Meyer, 2013), fitting a two-trait animal model as follows:

$$\mathbf{y}_i = \mathbf{X}_i \mathbf{b}_i + \mathbf{Z}_i \mathbf{a}_i + \mathbf{e}_i$$

Where, \mathbf{y}_i , \mathbf{b}_i , \mathbf{a}_i and \mathbf{e}_i are the vectors of observations, fixed effects, direct additive genetic effects, and residual random effects for the i th trait, respectively. Incidence matrices \mathbf{X}_i and \mathbf{Z}_i related the observations of the i th trait to the respective fixed effects and additive genetic effects, respectively. The average information (AI) REML algorithm was used to maximize the likelihood (convergence criterion was 10^{-8}) and additional restarts were performed until no further improvement in log likelihood occurred.

Results and Discussion

The estimate of correlations between ewe's longevity and ewe's lifetime reproductive traits are shown in Table 1. The genetic, environmental and phenotypic correlations between average conception rate (ACR) and longevity were weak positive, negative and negative, respectively.

The estimates of genetic, environmental and phenotypic correlations between longevity and other traits (NP, TNLB, TNLW, TLBW and TLWW) were positive and high. However, the estimates genetic correlations were higher than phenotypic, and phenotypic higher than environmental correlations. The estimates of genetic correlations imply that there is not any antagonistic between longevity and reproductive traits over ewe's lifetime, and selecting animals on the basis of any reproductive traits except average conception rate will result in a correlated response in longevity and vice versa. The genetic correlations between longevity with number of lambs born and number of lambs weaned over of the ewe's lifetime in Dorper sheep were 0.38 and 0.51 respectively (Zishiri, et al., 2013) which lower than results obtained in this study. The genetic correlation between longevity and NP, TNLB, TNLW, TLBW and TLWW in Karakul sheep were 0.99, 0.84, 0.53, 0.68 and 0.68, respectively (Bahri Binabaj, 2013). Also, this researcher reported that the genetic correlation between longevity and TNLB, TNLW, TLBW and TLWW in Baluchi sheep were 0.65, 0.53, 0.72 and 0.56, respectively. According to the results of this study genetic improvement in longevity leading to improvement of reproductive traits and consequently reduced maintenance cost in ewe replacement. On the other hand, ewe

with larger longevity, produced more lambs over her lifetime, and automatically will have a more contribution to the next generation. Thus, this study demonstrated that it is possible to genetically improve longevity and reproductive traits In Lori-Bakhtiari sheep through genetic selection.

Table 1. Correlation estimate (\pm s.e) between longevity and ewe's lifetime reproductive traits

Trait	Genetic	Environmental	Phenotypic
ACR	0.17 \pm 0.13	-0.11 \pm 0.06	-0.01 \pm 0.03
NP	0.96 \pm 0.01	0.89 \pm 0.01	0.92 \pm 0.01
TNLB	0.86 \pm 0.03	0.73 \pm 0.03	0.77 \pm 0.01
TNLW	0.88 \pm 0.03	0.72 \pm 0.02	0.78 \pm 0.01
TLBW	0.89 \pm 0.03	0.82 \pm 0.02	0.84 \pm 0.01
TLWW	0.90 \pm 0.02	0.77 \pm 0.02	0.82 \pm 0.01

References

- Bahri Binabaj, F. 2013. Genetic study of stayability traits in different ages their association with reproductive traits in Karakul and Baluchi sheep breeds. Ferdowsi University of Mashhad, Ph.D Dissertation, 175 p.
- Duguma, G., Schoeman, S. J., Cloete, S. W. P. and Jordaan, G. F. 2002. Genetic and environmental parameters for ewe productivity in Merinos. *S. Afr. J. Anim. Sci.* 32: 154-159.
- Eküz, B., Zcan, M. and Yilmaz, A. 2005. Estimates of Phenotypic and Genetic Parameters for EweProductivity Traits of Turkish Merino (Karacabey Merino) Sheep. *Turk. J. Ve.t Anim. Sci.*29 : 557-564.
- Lee, G. J., Atkins, K. D. and Sladek, M. A. 2009. Heterogeneity of lifetime reproductive performance, its components and associations with wool production and liveweight of Merino ewes. *Anim. Prod. Sci.* 49 (7): 624-629.
- Meyer, K. 2013. WOMBAT- A program for mixed model analyses by restricted maximum likelihood. User notes, Animal Genetics and Breeding Unit, Armidale, 113 pp.
- Mishra, A. K., Arora, A. L., Prince, L. L. L., Gowane, G. R. and Kumar, S. 2009. Lifetime litter size and ewes productivity efficiency of Garole \times Malpura crossbred sheep. *Indian Journal of Animal Sciences.*79(10): 1075–1077.
- Qureshi, M. A., Nawaz, M. and Khan, M. A. 1997. Lifetime production of Kajli ewes at Khushab and Khizerabad: Reproduction and lamb production as affected by ewe longevity. *Asian Australas. J. Anim. Sci.*10 (4): 408-415.
- Snyman, M. A., Olivier, J. J., Erasmus, G. J. and Van Wyk, J. B. 1997. Genetic parameter estimates for total weight of lamb weaned in Afrino and Merino sheep. *Livest. Prod. Sci.* 48, 111–116.
- Vatankhah, M. 2013. Estimation of the genetic parameters for survival rate in Lori-Bakhtiari lambs using linear and Weibull proportional hazard models. *J. Agr. Sci. Tech.* 15: 1133-1143.
- Zishiri, O.T., Cloete, S.W.P., Olivier, J.J. and Dzama, K. 2013. Genetic parameters for growth, reproduction and fitness traits in the South African Dorper sheep breed. *Small Ruminant Research.* 112: 39-48

The Interleukin-8 gene polymorphism and its association with milk production traits in Holstein Cows

Ridhowi, A*¹, C.L. Liang², H.L.Chang², M.C Wu³ & Suyadi¹

¹University of Brawijaya, Indonesia

²National Pingtung University of Science and Technology, Taiwan

³Livestock Research Institute, Council of Agriculture, Taiwan

Abstract

Genomic selection on individual genes is a promising method to genetically improve economically important traits in dairy cows. The objective of this study was to identify polymorphism of Interleukin-8 (IL8) gene and its association with milk production traits in Holstein dairy cows. Sixty-eight Holstein cows with 171 milking records were genotyped and were evaluated for the impact of polymorphism of IL8 gene on milk production traits. There were 56, 50, 31, 20, 11, and 3 cows with 1st, 2nd, 3rd, 4th, 5th, and 6th lactation records, respectively, which were further classified as primiparous and multiparous records. Fixed effects model was employed for the analysis with genotype of IL8 gene. Traits evaluated included 305-2X-ME, daily milk yield, fat%, protein%, lactose%, total solid%, somatic cell count (SCC), and somatic cell score (SCS). Genotypic frequencies of CC, CT and TT were 0.22, 0.47, and 0.31, respectively. No significant difference was found among genotypes for 305-2X-ME, fat%, protein%, total solid%, and SCS. However, polymorphism of IL8 gene did show significant effects on daily milk yield and SCC ($P < 0.05$) and lactose% ($P < 0.01$). These results confirm that IL8 gene plays main role in phenotypic traits, and it might be used in marker assisted selection to improve milk production traits.

Keywords: interleukin-8 gene, milk production traits, polymorphism, Holsteincows

*Corresponding email: dhowi_fapetub@yahoo.co.id

Introduction

The intramammary infections among dairy animals persist for longer periods of time, associated with elevated SCC, neutrophils activities, and affect milk production in dairy animals. The proportion of neutrophils as the percentage of the SCC has been proposed as a mastitis indicator (Sharif & Muhammad, 2008). According to Fonseca et al. (2011), it is hard to produce effective vaccines against mastitis disease due to the large variety of microorganisms causing it and its multifactor character. Therefore, one of the most promising ways to reduce the problems caused by mastitis, besides adequate sanitary conditions, is the selective breeding of resistant animals.

Genes associated with neutrophil function are potential genetic markers for mastitis and immune response, as neutrophil migration from blood to the site of infection is essential for resolution of most mastitis pathogens (Paape et al., 2000). The ability of neutrophils to migrate into infected tissues is dependent upon recognition of inflammatory mediators by neutrophil cytokine, chemokine, and complement receptors (Yougerman et al., 2004). Among various chemokines, one of the important chemokine associated with leukocyte migration particularly neutrophils is IL8 (Yougerman et al., 2004; Mahesh et al., 2011). IL8 is an important activator and chemoattractant for neutrophil and has been implicated in a variety of inflammatory diseases immune reaction, and organism defense (Zhang & Chen, 2002; Nishimura, 2003). According to

Mahesh et al. (2011) IL8 is also one of the ligands that binds chemokine receptor-1 (CXCR1) and chemokine receptor-2(CXCR2) with high affinity and it is responsible for the massive influx of neutrophil to the mammary gland during bacterial infection. The objectives of this study were to identify the polymorphism of IL8 gene in Holstein cows and to evaluate associations between these polymorphisms with milk production traits in Holstein cows.

Materials and Methods

Sixty-eight Holstein cows from two dairy farms were genotyped and 171 lactation records were collected with 56, 50, 31, 20, 11, and 3 records for the 1st, 2nd, 3rd, 4th, 5th, and 6th lactation animals, respectively. Milking data were extracted from the DHI databank provided by Taiwan Livestock Research Institute (http://www.angrin.tlri.gov.tw/menue_all.htm). Traits evaluated were 305-2X-ME, daily milk yield, fat%, protein%, lactose%, total solid%, somatic cell count (SCC), and somatic cell score (SCS) with $SCS = \log_2(SCC/100,000) + 3$. The gDNA was extracted using Pure gene TM Quick Tips (Gentra USA) and Gene quant II UV-spectrophotometer was used to assess the quantity and quality of DNA samples at 260/280 nm. The polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) technique was used for genotyping. The primer was designed according to the sequences of *IL8* (NC_007304) gene on Gen Bank. Effects of *IL8* gene polymorphism on traits studied were evaluated using the General Linear Models (GLM) procedure of SAS 9.2 software. In addition to random residual, fixed effects of farm, parity and genotype were included in the model. Least-squared means of genotypes were then calculated and compared with each other at 5% significant level.

Results and Discussion

Three genotypes, CC, CT and TT, were obtained for the SNP of IL8.2862T>C detected with genotypic frequencies being 22% (15/68), 47% (32/68), 31% (21/68) respectively, which C and T alleles were evenly distributed with 0.45 and 0.55 frequencies in the herds studied. The corresponding χ^2 statistics for Hardy-Weinberg equilibrium test was 0.17 (Table 1).

Table 1. The genotypic and allelic information of IL8 gene in Holstein cows.

No. of cows genotype	Genotypic frequency			Allelic frequency		χ^2 -value	P-value
	CC	CT	TT	C	T		
68	0.22%	0.47%	0.31%	0.45%	0.55%	0.17	0.67

Association between IL8 genotypes and milk production traits were evaluated. IL8 genotypes was not significantly associated with 305-2X-ME, fat%, protein%, and total solid%, nevertheless it had significant association with daily milk yield ($P < 0.05$), lactose% ($p < 0.01$) and SCC ($P < 0.05$). The highest value on daily milk yield and lactose were obtained in cows with CT genotype followed by TT and CC genotype. While the highest value on SCC was obtained by TT genotype followed by CT and CC genotype. Decreasing SCC on cows with CC and CT genotype was implying that CC and CT genotype cows have high potency to activate neutrophil than TT genotype. As known, IL8 is an important chemokine involved in the attraction of neutrophil to site of inflammation. Moreover IL8 has direct effects on bovine neutrophil and it was significantly correlated with SCC (0.62, $P < 0.05$) (Bhupal, 2007).

The fact that milk production traits in this study was closely related with SNPs of IL8.2862T>C is in line with Chen et al. (2011). He reported that that SNPs of IL8.2862T>C and IL8.2789A>G had significant association ($P < 0.01$) with milk production, fat, protein and SCS.

This study found that an increasing daily milk yield in CT genotype accompanied with lower SCC was observed in cows with CT genotype ($33.26 \pm 1.52 \times 10^4$ cells/mL) compared with TT genotype ($34.77 \pm 1.96 \times 10^4$ cells/mL). Interestingly, this condition did not happen in cows

with CC genotype. Although cows with CC genotype showed lowest SCC ($28.27 \pm 1.84 \times 10^4$ cells/mL), the daily milk yield in this genotype was still lower compared with other genotypes (Table 2). We predict might be this is because the number of cells secreting milk in cows with CC genotype was lower than other genotypes. Therefore, CT genotype had higher in number of cells secreting milk and much more healthy condition compared with CC and TT genotype. Hence, more milk production was observed in CT cows. This statement is supported by Boutinaud et al. (2004) who reported that the ability of ruminant mammary glands to produce milk is determined by the number of cells secreting milk and their level of activity.

Table 2. The effect of *IL8* genotype on 305-2X-ME, daily milk yield, SCC and SCS and milk component (fat, protein, lactose, total solid) in Holstein cows.

Trait	n	Genotype			P-value	
		CC	CT	TT		
Milk (kg)	305-2X-ME	171	7.826 ± 215	8.079 ± 175	8.123 ± 225	NS
Daily milk yield (kg/d)		171	26.63 ± 0.61^b	28.56 ± 0.50^a	28.03 ± 0.64^{ab}	*
Fat (%)		171	3.75 ± 0.07	3.75 ± 0.05	3.78 ± 0.07	NS
Protein (%)		171	3.24 ± 0.03	3.25 ± 0.03	3.25 ± 0.03	NS
Lactose (%)		171	4.71 ± 0.02^b	4.81 ± 0.02^a	4.76 ± 0.02^{ab}	**
Total solid (%)		171	12.41 ± 0.10	12.52 ± 0.82	12.50 ± 0.10	NS
SCC (10^4 cells/mL)		168	28.27 ± 1.84^b	33.26 ± 1.52^a	34.77 ± 1.96^a	*
SCS		168	3.62 ± 0.16	3.88 ± 0.13	3.99 ± 0.17	NS

LSM \pm SE; **: $P < 0.01$; *: $P < 0.05$; NS: $P \geq 0.1$.

NS = Not significant; n = Number of records.

These values confirm that *IL8* gene plays main role in phenotypic traits, and it can be used as gene assisted selection to improve milk production traits.

References

- Bhupal, G. 2007. Cytokine expression in milk somatic cells during mastitis in cattle and buffaloes. Ms. Thesis. Anand Agricultural Univ., Anand.
- Chen, R., Y. Zhangping, J. Dejun, M. Yongjiang, C. Yin, L. Yunlong, W. Haitao, W. Xiaolong, and C. Lingling. 2011. Polymorphisms of *IL8* gene correlate with milking traits, SCS and mRNA level in Chinese Holstein. *Mol. Bio. Rep.* 38:4083-4088.
- Fonseca, I., G. R. Antunes, D. S. Paiva, C. C. Lange, S. E. F. Guimares, and M. F. Martins. 2011. Differential expression of genes during mastitis in Holstein-Zebu crossbreed dairy cows. *Gen. Mol. Res.* 10:1295-1303.
- Mahesh. S. D., G. R. Gowane, L. S. Kokate, S. B. Kumar, and B. Bhushan. 2011. CXCR Gene and its Association with Mastitis Resistance in Cattle- a Review. *Int. J. Stress Manage.* 2:444-447.
- Nishimura, T. 2003. Expression of potential lymphocyte trafficking mediator molecules in the mammary gland. *Vet. Res.* 34:3-10.
- Rambeaud, M., and G.M. Pighetti. 2005. Impaired neutrophil migration associated with specific bovine CXCR2 genotypes. *Infect. Immun.* 73:4955-4959.
- Rambeaud, M., R. Clift, and G.M. Pighetti. 2006. Association of a bovine CXCR2 gene polymorphism with neutrophil survival and killing ability. *J. Vet. Imm.* 111:231-238.
- Sharif A., and G. Muhammad. 2008. Somatic cell count as an indicator of udder health status under modern dairy production: a review. *Pakistan Vet. J.* 28:194-200.
- Youngerman, S. M., A. M. Saxton, S. P. Oliver, and G. M. Pighetti. 2004. Association of CXCR2 polymorphisms with subclinical and clinical mastitis in dairy cattle. *J. Dairy Sci.* 87:2442-2448.
- Zhang, W., and H. Chen. 2002. The study on the interleukin-8 (*IL8*). *Pub. Med. NCBI.* 9:697-702 (Abstr.).

Genetic improvement of production performance for Yorkshire

Viterbo*, V. S., H. S. Kang, B. M. Lopez, Y. H. Kim, J. H. Hong, T. H. Kim, K. C. Nam & K. S. Seo

Department of Animal Science and Technology, Sunchon National University, Suncheon, 540-742, South Korea

Abstract

The study was conducted to assess heritability estimates and genetic correlations of growth and carcass traits of Yorkshire, as well as genetic improvement of those traits in the past ten years. A total of 120,161 heads of Yorkshire from commercial farm in South Korea were used in this study. Basic statistical analysis was done using SAS version 9.3 and genetic traits were calculated using *remlf90* and *blupf90* according to mixed animal model. Production parameters included days at 90 kg (D90), average daily gain (ADG), lean percentage (LP), eye muscle area (EMA) and Backfat thickness (BF). Heritability estimates were 0.41, 0.39, 0.27, 0.27 and 0.43 D90, ADG, LP, EMA and BF respectively. Moreover, genetic correlations were significantly high between ADG and BF with -0.96, and between ADG and EMA with -0.662. Growth parameters (D90 and ADG) showed negative correlations with carcass traits (LP, EMA and BF). Genetic improvement was recorded in ADG with increase of 5g per day, followed and in D90 with -0.88 d/year genetic trend. Moreover almost no trend was noted to EMA, LP and BF. Heritability and correlation is useful to improve growth traits and carcass traits especially those with low genetic improvement.

Keywords: genetic improvement, heritability, production performance, Yorkshire

**Corresponding author: viterbo.vanessa@gmail.com*

Introduction

Improvement of animal models and accessibility of computer programs lead to more efficient and accurate estimation of animal genetic components. Imboonta et al. (2007) reported positive and negative correlations between growth traits and reproductive traits which affect the over-all profit. Growth traits and carcass traits are both economically significant and the genetic estimation and monitoring of genetic trend are useful in attaining breeding goals. Genetic analysis using recent data helps assess the status of production and determine possible areas of improvement. This study was conducted to assess heritability estimates and genetic trends for average daily gain (ADG), number of days at 90kg (D90), backfat thickness (BF), eye muscle area (EMA) and lean percentages (LP) in Yorkshire produced in commercial farms in South Korea for the past ten years.

Materials and Methods

Animal Records

This study was conducted from 2004 to 2013 using 120,161 heads of Yorkshire from commercial farm in South Korea. Production traits consist of ADG, D90, BF, EMA and LP. The LP, EMA and BF were measured using PIGLOG (ultrasound technology).

Statistical Analysis

Parameters were evaluated to assess factors that have fixed effect and were included in the animal model. Basic statistical analysis was computed using Statistical package SAS version 9.3. Genetic estimation and breeding value were analyzed using *remlf90* and *blupf90* (Misztale et al., 2002) following mixed animal model:

$$Y_{ijklm} = \mu + \text{SEX}_j + Y_k + S_i + bD_{ijk} + a_{ijklm} + e_{ijklm}$$

Where Y_{ijklm} = observation of the studied traits; μ = overall mean; SEX_j = j^{th} sex effect; Y_k = k^{th} year of production in the farm; S_i = i^{th} season during production in the farm; b = regression of y for end of production in the farm by age (days); D = end of production (days); a_{ijklm} = animal random effect; e_{ijklm} = residual effect.

Result and Discussion

Heritability Estimates

The heritability estimates and components of variance of Yorkshire are given in Table 1. Data revealed moderately to highly heritable value for five traits. The heritability estimates for D90, ADG, LP, EMA, and BF were 0.41, 0.39, 0.27, 0.27, and 0.43, respectively. Results were comparable to estimations reported by different researcher where the range of heritability estimates for D90, ADG, LP, EMA and BF were 0.29, 0.13 to 0.29, 0.42 to 0.43, 0.13 to 0.42, and 0.44 to 0.60, respectively (Van Wijk et al., 2005; Akanno et al., 2013). Moderately to highly heritable traits are expected to improve more efficiently in response to selection.

Table 1. Estimates of heritability, genetic and residual variance of Yorkshire.

Estimates	Production Parameters				
	D90	ADG	LP	EMA	BF
h^2	0.41	0.39	0.27	0.27	0.43
σ_g^2	45.48	1178	3.16	2.22	2.62
σ_r^2	65.72	1774	3.71	6.12	3.37

h^2 = heritability, σ_g^2 = genetic variance, σ_r^2 = residual variance

Genetic correlations

Genetic correlations of five parameters are shown in Table 2. Based on the data, growth rate parameters such as D90 and ADG showed negative to low positive correlation with carcass traits, LP, EMA and BF. The ADG were noted for high negative correlations with EMA (-0.663) and BF (-0.962) while BF was inversely correlated to other carcass traits. According to Miar et al. (2014) BF was negatively correlated to EMA with correlation coefficient of -0.34. The result suggests that simultaneous improvement of these five parameters will be difficult due to negative correlations between these traits.

Table 2. Genetic Correlation of five production parameters of Yorkshire.

Parameters	D90	ADG	LP	EMA	BF
D90	1	0.193	-0.007	-0.355	-0.259
ADG, g/d		1	0.094	-0.662	-0.962
LP, %			1	-0.233	-0.145
EMA, mm				1	-0.233
BF, mm					1

D90=days at 90 kg, ADG=average daily gain, LP=lean percentage, EMA=eye muscle area, BF=backfat

Genetic Trend

Standardized breeding value of growth and carcass traits of Yorkshire are illustrated in figure 1 and regression coefficient of breeding value were shown in Table 3. Based on the graph below highest genetic gain was drawn in ADG, and the graph for this trait tended to increase continuously, while almost no trend were recorded on EMA, BF and LP. In addition advantageous decrease in D90 was also recorded. Merks (2000) were able to document a 290g/day improvement of ADG from 550 g/day in 1930 to 840 g/day in 1990 while Chen et al. (2002) reported genetic improvement of -0.40d/yr to reach 113.5 kg body weight. Further improvement on traits with no genetic trend will be beneficial in animal production.

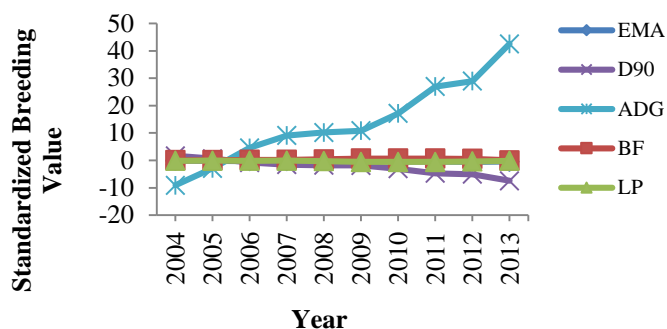


Figure 1. Standardized breeding value of growth and carcass traits of Yorkshire.

Table 3. Regression of EBV for production performance parameter of Yorkshire.

	Production Parameters				
	Age at 90 kg	ADG	LP	EMA	BF
r	-0.88	5.00	-0.04	-0.04	0.04

r= regression coefficient

References

- Akanno, E., F. Schenkel, V. Quinton, R. Friendship and J. Robinson, 2013. Meta-analysis of genetic parameter estimates for reproduction, growth and carcass traits of pigs in the tropics. *Livest. Sci.* 152: 101-113.
- Chen, P., T. Baas, J. Mabry, J. Dekkers and K. Koehler, 2002. Genetic parameters and trends for lean growth rate and its components in US Yorkshire, Duroc, Hampshire, and Landrace pigs. *J. Anim. Sci.* 80: 2062-2070.
- Imboonta, N., L. Rydhmer and S. Tumwasorn, 2007. Genetic parameters and trends for production and reproduction traits in Thai Landrace sows. *Livest. Sci.* 111: 70-79.
- Merks, J. W., 2000. One century of genetic changes in pigs and the future needs. *BSAS occasional publication*: 8-19.
- Miar, Y., G. Plastow, S. Moore, G. Manafiazar, P. Charagu, R. Kemp, B. Van Haandel, A. Huisman., C. Zhang and R. McKay, 2014. Genetic and phenotypic parameters for carcass and meat quality traits in commercial crossbred pigs. *J. Anim. Sci.* 92: 2869-2884.
- Misztal, I., S. Tsuruta, T. Strabel, B. Auvray, T. Druet and D. Lee, 2002. BLUPF90 and related programs (BGF90). In: *Proceedings of the 7th world congress on genetics applied to livestock production*. p 21-22.
- Van Wijk, H., D. Arts, J. Matthews, M. Webster, B. Ducro and E. Khol, 2005. Genetic parameters for carcass composition and pork quality estimated in a commercial production chain. *J. Anim. Sci.* 83: 324-333.

Genetic parameters and trends for growth and carcass traits of Landrace in Korea

Hong*, J. H., V.S.Viterbo, B. M.Lopez, H. S. Kim, T. H. Kim, Y. H. Kim, K. C. Nam & K.S.Seo

Department of Animal Science and Technology, Suncheon National University, Suncheon, 540-742, South Korea

Abstract

The objectives of this study were to obtain heritability estimates, genetic correlations and genetic trend for economic traits using data from 2004 to 2013. Animal records of 37,984 heads of Landrace from commercial farm in South Korea were used in this study. Data included age at 90 kg (A90), average daily gain (ADG), lean percentage (LP), eye muscle area (EMA), and backfat thickness (BF). Statistical analysis was performed using SAS version 9.3 and genetic estimates were calculated using *gremlf90*, *blupf90* following mixed animal model. Results revealed heritability estimates (\pm SE) of 0.45 ± 0.02 , 0.43 ± 0.04 , 0.47 ± 0.03 , 0.24 ± 0.03 and 0.48 ± 0.02 for A90, ADG, LP, EMA, and BF, respectively. Furthermore, genetic correlations revealed low positive correlation to highly negative correlation between five traits. Highest negative correlations were found between age at 90 kg and ADG of -0.95 ± 0.01 , likewise high negative genetic correlations were observed between LP and BF with -0.66 ± 0.01 , and between ADG and EMA with -0.32 ± 0.02 . Genetic trend observed were -0.35 ± 0.10 d/year, 2.07 ± 0.08 g/day, $-0.05\pm 0.007\%$ /yr, -0.12 ± 0.02 mm/yr and 0.007 ± 0.01 mm/year for A90, ADG, LP, EMA, and BF, respectively. Findings suggest that genetic improvement favored age ADG trait that showed high heritability estimates.

Keywords: genetic trend, genetic estimates, economic traits, Landrace

*Corresponding authors: boayo486@nate.com

Introduction

Growth and carcass parameters are important factors that are given attention in the genomic selection of swine. Assessments of improvement on these traits are necessary to illustrate effectiveness of selection, and to guide direction of breeding goals to improve traits with high economic importance. Recent computer programs that analyze genetic estimations and relationship, helps in evaluating selection procedures to achieve breeding goal more efficiently and accurately.

This study utilized data compiled in recent years using Landrace breed to assess genetic estimates, and to evaluate genetic trend for age at 90 kg (A90), average daily gain (ADG), lean percentage (LP), eye muscle area (EMA) and backfat thickness (BF).

Materials and Methods

Experimental Animals

The total number of Landrace used in this study was 37,984 heads (2004 to 2013). The economic traits were age at 90kg (A90), average daily gain (ADG), lean percentage (LP), eye muscle area (EMA), and backfat thickness (BF). The PIGLOG (ultrasound technology) was utilized to measure LP, EMA and BF.

Statistical Analysis

All parameter were evaluated to assess factors that have fixed effect and were included in the animal model. Basic statistical analysis was computed using Statistical package SAS version 9.3. The genetic estimation and genetic gain were analyzed using airemlf90 and blupf90 (Misztal, 2002), following animal mixed model:

$$Y_{ijkl} = \mu + \text{SEX}_j + Y_k + S_i + bD_{ijkl} + a_{ijkl} + e_{ijkl}$$

Where Y_{ijkl} = observation of the studied traits; μ = overall mean; $\text{SEX}_j = j^{\text{th}}$ sex effect; $Y_k = k^{\text{th}}$ year of production in the farm; $S_i = I^{\text{th}}$ season during production in the farm; b = regression of Y_{ijklm} for end of production in the farm by age (days); D = end of production (days); a_{ijkl} = animal random effect; e_{ijkl} = residual effect. The genetic trend was calculated by regression of the average estimated breeding value on the year of birth of the animals with records.

Result and Discussion

Heritability Estimates

Heritability estimates and components of variance were shown in Table 1. Heritability estimates were high for A90, ADG, LP and BF except for EMA which is moderately heritable. The result were higher than the data reported by Akanno et al. (2013) wherein the heritability estimates were 0.28, 0.28, 0.42, 0.33 for D90, ADG, LP and BF, excluding EMA with higher estimate of 0.51.

Table 1. Estimates of heritability, genetic variance and residual variance.

Estimates	Production Parameters				
	A90	ADG	LP	EMA	BF
h^2	0.45	0.43	0.47	0.24	0.48
σ_g^2	48.48	1340	2.80	2.11	2.36
σ_r^2	58.57	1771	3.14	6.62	2.54
SE	0.02	0.04	0.03	0.03	0.02

h^2 = heritability, σ_g^2 = genetic variance, σ_r^2 = residual variance, SE = standard error.

Genetic Correlations

Results of genetic correlation on production traits were given in Table 2. Highest negative correlation was recorded between D90 and ADG, as the ADG increases then shorter days to reach 90 kg was expected, this negative correlation was practically beneficial. Low to moderate correlations were observed between EMA and other traits except between EMA and ADG which shows negative correlation of -0.32. Another high negative correlation was between %LP and BF.

Akanno et al. (2013) conducted a meta-analysis of genetic parameters using tropical swine which revealed comparable result of correlation between ADG and A90 (-1), and lower correlation between leanness and loin eye area of 0.41. The results suggest that selection improvement on one trait will be advantageous on some traits and disadvantageous on the others

Table 2. Genetic correlations (\pm SE) of Landrace for Production traits.

Parameters	D90	ADG	LP	EMA	BF
A90	1	-0.96 \pm 0.01	-0.01 \pm 0.04	0.26 \pm 0.02	0.06 \pm 0.01
ADG, g/d		1	0.04 \pm 0.01	-0.32 \pm 0.02	-0.09 \pm 0.03
LP, %			1	0.25 \pm 0.01	-0.66 \pm 0.03
EMA, mm				1	-0.04 \pm 0.03
BF, mm					1

A90 = days at 90 kg, ADG = average daily gain, LP = lean percentage, EMA = eye muscle area, BF = backfat.

Genetic Trend

Estimated breeding value (EBV) of five economic traits is shown in Figure 1, while the regression coefficients are shown in Table 3. Based on the graph, the EBV of ADG exhibits the largest increase in genetic trend of 2.07 g/d while very low to negative trend were illustrated on the remaining traits. Merks (2000) documented a total genetic improvement of ADG of 5.67g/year from 1930 to 1990. Holl & Robison (2003) reported positive genetic trend of 0.013mm/year for BF. Therefore, the result of this study suggested that selection in swine favors the improvement of ADG which is highly heritable, while possible improvements on other heritable traits might be hindered by other factors, such as monitoring and proper evaluation of traits, costing and time frame required.

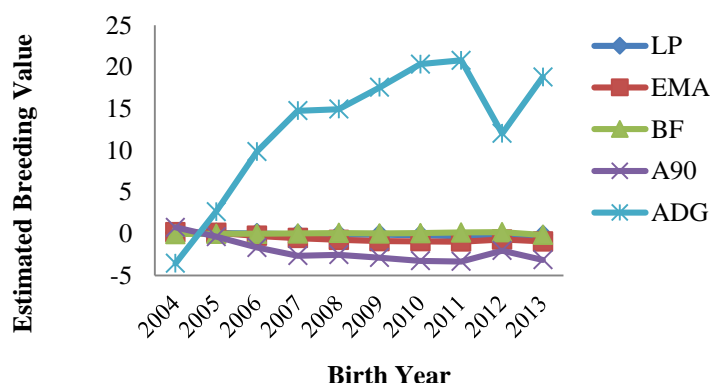


Figure 1. The Estimated Breeding Value of economic traits of Landrace.

Table 3. Regression coefficient(\pm SE) of estimated breeding value from 2004 to 2013.

	Production Parameters				
	A90	ADG	LP	EMA	BF
r	-0.35 \pm 0.10	2.07 \pm 0.08	-0.05 \pm 0.007	-0.12 \pm 0.02	0.007 \pm 0.01

r= regression coefficient, A90=days at 90 kg (d/yr), ADG=average daily gain(g/d), LP=lean percentage(%/yr), EMA=eye muscle area(mm/yr), BF=backfat(mm/yr).

References

- Akanno, E., F. Schenkel, V. Quinton, R. Friendship, and J. Robinson. 2013. Meta-analysis of genetic parameter estimates for reproduction, growth and carcass traits of pigs in the tropics. *Livest. Sci.* 152: 101-113.
- Chen, P., T. Baas, J. Mabry, J. Dekkers, and K. Koehler. 2002. Genetic parameters and trends for lean growth rate and its components in US Yorkshire, Duroc, Hampshire, and Landrace pigs. *J. Anim. Sci.* 80: 2062-2070.
- Holl, J. W., and O. Robison. 2003. Results from nine generations of selection for increased litter size in swine. *J. Anim. Sci.* 81: 624-629.
- Merks, J. W. 2000. One century of genetic changes in pigs and the future needs. *BSAS occasional publication*: 8-19.
- Misztal, I., S. Tsuruta., T. Strabel., B. Auvray., T. Druet., and D. Lee. 2002. BLUPF90 and related programs (BGF90). In: *Proceedings of the 7th world congress on genetics applied to livestock production.* p 21-22.

Effect of sex and carcass weight on pork belly characteristics of Large White

Kim*, H. S., B. M. Lopez, V. S. Viterbo, J.H.Hong, T.H.Kim, Y.H.Kim, K.C.Nam & K. S. Seo

Department of Animal Science and Technology, Suncheon National University, Suncheon, 540-742, South Korea

Abstract

The objective of this study was to evaluate the effect of sex and carcass weight on pork belly traits as well as to assess phenotypic correlations of pork belly traits. A total of 676 heads Large White were used in the study. Data were recorded immediately after slaughtering which included carcass weight, backfat thickness, belly weight, belly thickness, belly length, belly width and muscle percentage. Traits were analyzed using SAS version 9.3. Barrow showed statistically higher carcass weight (90.11 kg) and backfat thickness (21.06 mm) ($P>0.01$) than the gilt with 97.55 kg and 17.63 mm for carcass weight and backfat thickness, respectively. Furthermore, belly weight of 7.47 kg and belly length of 559.29 mm ($P>0.01$) of barrow were significantly higher than 6.86 kg belly weight and 546.74 mm belly length of gilt. However, muscle percentage of belly was recorded statistically higher in gilt with 54.89% ($P>0.01$) than barrow with 48.72%. Moreover, carcass weight has significant effect on belly characteristics. Carcass weight of 100 kg and above ($P>0.01$) showed the highest value for backfat, belly weight, belly thickness, belly length, and belly width however highest muscle percentage of belly ($P>0.01$) was recorded to carcass weighing below 79 kg. Positive correlations were observed between carcass weight and belly traits except for muscle percentage of belly with -0.32 phenotypic correlations ($P>0.01$).

Keywords: sex, carcass weight, pork belly, Large White

**Corresponding author: yummy6640@naver.com*

Introduction

Pork belly, one of the primal cuts, has high market value because Koreans enjoys eating grilled pork belly known as “Samgyeopsal”. According to D'Souza et al. (2004), consumers are willing to pay higher amount for pork belly that contains higher amount of lean and lower intermuscular fats and subcutaneous fat deposits. The improvement of pork belly is an opportunity for local producers to meet the demand of the local market. Selection trend is towards lower carcass fat and improve or maintain intramuscular fat that is associated to eating quality (Yang et al., 2010). This study was conducted to assess the effect of sex and carcass weight on pork belly characteristics of Large White produced commercially in the local farm in South Korea.

Materials and Methods

Data

The effect of sex and carcass weight on pork belly characteristics were investigated using 676 heads of Large White, which consisted of 334 heads of barrow (castrated male swine) and 342 heads of gilt. Data were collected from year 2011 to 2013. Pork belly measurements were

collected using only the right side of belly. All the evaluations were supervised by Korea Institute for Animal Products Quality Evaluation (KAPE), an association authorize to grade animal products according to South Korean government rule. Data includes carcass weight, backfat, belly weight, belly thickness, belly length, belly width and belly muscle percentage. To evaluate the effect of carcass weight, carcass was divided into four categories, group A carcass weighting below 79 kg, group B carcass weighting 80-89kg, group C carcass weighting 90-99 kg, and group D carcass weighting 100 kg and above.

Statistical analysis

Basic statistics were analyzed using Statistical package SAS version 9.3. The effect of sex, carcass weight and correlation on pork belly characteristics were evaluated using proc GLM following mixed animal model. Statistical differences were considered at an alpha level 0.01. The model was as follows:

$$Y_{ijkl} = \mu_i + \text{SEX}_{ij} + \text{YS}_{ik} + a_{ijkl} + \beta \text{Age}_{ijkl} + e_{ijkl}$$

Where Y_{ijkl} = observation for a traits i^{th} ; μ_i = overall mean of i^{th} trait; SEX_{ij} = j^{th} sex effect of i^{th} trait; YS_{ik} = k^{th} year-season effect of i^{th} trait; a_{ijkl} = animal random effect; β = covariate of number of test day; Age_{ijkl} = number of test day; e_{ijkl} = random error.

Results and Discussions

Effect of sex on carcass and belly traits

The effect of sex on carcass weight and pork belly traits is shown in Table 1. Result revealed that carcass weight, backfat (BF) thickness, belly weight, belly length were significantly higher in barrow than in gilt, however muscle percentage was significantly higher in gilt than in barrow. Scramlin et al. (2008) did not find significant differences between barrow and gilt in terms of carcass weight, belly length and belly yield while Correa et al. (2006) suggested that barrow yields higher belly weight and back fat thickness compared to gilt.

Table 1. Effects of sex on carcass traits and pork belly traits of Large White.

Traits	Sex	
	Barrow	Gilt
Carcass weight kg	90.11±0.77 ^a	87.55±1.06 ^b
BF mm	21.06±0.43 ^a	17.63±0.59 ^b
Belly weight kg	7.47±0.08 ^a	6.85±0.11 ^b
Belly thickness mm	47.26±0.61 ^{ns}	46.85±0.84 ^{ns}
Belly length mm	559.29±2.48 ^a	546.74±3.41 ^b
Belly width mm	281.0±1.06 ^{ns}	278.54±1.46 ^{ns}
Muscle percentage, %	48.71±0.29 ^b	54.89 ±0.49 ^a

^{a, b} different superscript on the same row is significantly different ($P < 0.01$), ns- not significant.

Effect of carcass weight on pork belly

The effect of different carcass weight on belly characteristics is presented in Table 2. Carcass weight showed significant influence on pork belly characteristics. Based on the data, highest backfat and belly characteristics, except belly muscle percentage, were recorded on carcass weighting 100 kg and above while lowest value was recorded on carcass weighting below 79 kg. The result emphasize the direct relationship between slaughter weight and belly traits such as belly weight, belly percentage belly thickness (Correa et al., 2006; Correa et al., 2008).

Table 2. Belly characteristics of Large White as influenced by carcass weight.

Traits	Carcass weight			
	A Below 79 kg	B 80-89kg	C 90-99 kg	D 100 kg and
BF	15.99 ±0.54 ^d	18.15±0.44 ^c	20.75±0.47 ^b	23.44±0.62 ^a
Belly weight	5.97 ±0.07 ^d	6.76±0.05 ^c	7.62±0.06 ^b	8.63±0.08 ^a
Belly thickness	42.47 ±0.79 ^d	45.53±0.654 ^c	49.16±0.68 ^b	51.57±0.9 ^a
Belly length	532.62±3.04 ^d	544.77±2.51 ^c	562.65±2.64 ^b	578.48±3.49 ^a
Belly width	272.14±1.38 ^d	277.61±1.14 ^c	282.71±1.2 ^b	287.97±1.59 ^a
Muscle percentage, %	52.38±0.63 ^a	51.62±0.52 ^a	50.79±0.55 ^b	48.29±0.72 ^c

a, b, c, d different superscript on the same row is significantly different ($P<0.01$).

Phenotypic Correlations

Phenotypic correlation of pork belly traits of Large White is presented on Table 3. Carcass weight was moderately to highly correlated to backfat (0.54), belly weight (0.85), belly width (0.51), belly length (0.38), and belly thickness (0.33) thus simultaneous enhancement on these traits are economical. Additionally, it was noted that belly weight was highly correlated with other belly traits like belly width (0.56), belly length (0.48), and moderately correlated with belly thickness (0.23). However, negative correlations were prominent for belly muscle percentage and other traits such as backfat, belly weight, and belly width. Correlations revealed simultaneous improvement of carcass and pork belly traits, except muscle percentage, were useful.

Table 3. Phenotypic correlations of carcass traits and pork belly of Large White.

	CW	BF	Bwt	BWd	BL	BT	% M
CW	1	0.54	0.85	0.51	0.38	0.33	-0.32
BF		1	0.29	0.39	0.13	0.29	-0.57
Bwt			1	0.56	0.48	0.23	-0.45
BWd				1	-0.01	-0.97	-0.42
BL					1	0.10	-0.12
BT						1	-0.14
% M							1

CW=carcass weight, BF=backfat, Bwt=belly weight BWd=belly width, BL=belly length, BT=belly thickness, %M=muscle percentage, highlighted values were significant at level $P<0.01$.

References

- Correa, J., L.Faucitano, J. Laforest, J. Rivest, M. Marcoux, and C. Gariépy. 2006. Effects of slaughter weight on carcass composition and meat quality in pigs of two different growth rates. *Meat Sci.* 72: 91-99.
- Correa, J., C. Gariépy, M. Marcoux, and L. Faucitano. 2008. Effects of growth rate, sex and slaughter weight on fat characteristics of pork bellies. *Meat Sci.* 80: 550-554.
- D'Souza, D., D. Pethick, F. Dunshea, D. Suster, J. Plucke, and B. Mullan. 2004. The pattern of fat and lean muscle tissue deposition differs in the different pork primal cuts of female pigs during the finisher growth phase. *Livest. Prod. Sci.* 91: 1-8.
- Scramlin, S., S. Carr, C. Parks, D. Fernandez-Dueñas, C. Leick, F. McKeith, and J. Killefer. 2008. Effect of ractopamine level, gender, and duration of ractopamine on belly and bacon quality traits. *Meat Sci.* 80: 1218-1221.
- Yang, K., J. Ma, Y. Guo, T. Guo, Y. Zhao, N. Ding, M. Betti, G. Plastow and L. Huang. 2010. Correlations between fat depot traits and fatty acid composition in abdominal subcutaneous adipose tissue and longissimus muscle: Results from a White Duroc × Erhualian intercross F population. *J. Anim. Sci.* 88: 3538-3545.

Influence of sex and carcass weight on pork belly muscle of Large White

Y.H. Kim*, V.S. Viterbo, B. M. Lopez, J.H. Hong, T. H. Kim, K. C. Nam and K. S. Seo

Department of Animal Science and Technology, Suncheon National University, Suncheon, 540-742, South Korea

Abstract

A total of 676 heads of Large White were used to assess the effect of sex and carcass weight on belly muscle characteristics. Statistical analysis was performed using SAS version 9.3. Parameters included carcass weight and pork belly muscle traits such as deep pectoral muscle area, latissimus dorsi muscle area, cutaneous trunci muscle area, rectus abdominis muscle area, internal and external abdominal oblique muscle area, total muscle and total fat. Effect of sex was significant on total belly muscle characteristics. Total muscle area (2307.53 cm²) and muscle percentage (54.89%) of pork belly were statistically higher in gilt than barrow. On the other hand total fat area of 2384.51 cm² of pork belly was statistically higher in barrow than gilt with 1928.78 cm². Moreover, effect of carcass weight on the pork muscle characteristics was also significant. Result showed highest muscle area were found in carcass weighting 100 kg and above, however statistical analysis revealed that total muscle percentage in the pork belly was decreasing as the carcass weight increases. Highest muscle percentage of 52.38% was recorded on carcass weighting below 79 kg while lowest muscle percentage of 48.29 % was noted on 100 kg and above carcass weight.

Keywords: pork belly muscle, sex, carcass weight, Large White

Introduction

The swine industry is the second largest contributor to the country's agriculture-livestock industry. Also, pork has become a major source of protein for the people. The cost of pork is determined by the market value of its primal cuts. Currently, the importation of pork fills the demand in the local market (Smith *et al.*, 2014) and imported pork are normally lean cuts such as loin and ham. Pork belly is one of the most important cuts of pork and considered as Koreans' favorite primal cut. Phenotypic evaluation of pork belly muscle was limited, thus this study was conducted to assess the influence of sex and carcass weight on pork belly characteristics of Large White swine.

Materials and Methods

Data

A total of 676 heads Large White were being utilized in this study. Assessment of meat quality traits were conducted under the supervision of Korea Institute for Animal Products Quality Evaluation (KAPE). Meat quality traits included carcass weight and pork belly muscles such as the deep pectoral, latissimus dorsi, cutaneous trunci, rectus abdominis, and the external and internal abdominal oblique muscles. Carcass weight was immediately recorded after slaughtering while belly muscle characteristics were evaluated 24 hours post-slaughtering. Only the right side of the pork belly cuts was used to measure pork belly muscle. Belly muscle measurements were conducted using image scanning. To evaluate the effect of carcass weight, carcass was divided

into four categories, group A carcass weighting below 79 kg, group B carcass weighting 80-89kg, group C carcass weighting 90-99 kg, and group D carcass weighting 100 kg and above.

Statistical Analysis

Elementary statistics were performed using GLM procedure of SAS version 9.3. To determine the effect of sex and carcass weight of the animals on pork belly muscle, least square means were performed according to the following mixed animal model:

$$Y_{ijkl} = \mu_i + \text{SEX}_{ij} + \text{YS}_{ik} + a_{ijkl} + \beta \text{Age}_{ijkl} + e_{ijkl}$$

where Y_{ijkl} = observation for a traits i^{th} ; μ_i = overall mean of i^{th} trait; SEX_{ij} = j^{th} sex effect of i^{th} trait; YS_{ik} = k^{th} year-season effect of i^{th} trait; a_{ijkl} = animal random effect; β = covariate of number of test day; Age_{ijkl} = number of test day; e_{ijkl} = random error.

Results and Discussions

Effect of sex on pork belly muscle

The effect of sex on pork belly muscle traits were shown in Table1. Statistical analysis revealed that sex has significant influence on carcass weight, total belly and total belly muscle trait. Barrow showed significant advantage over gilt in terms of carcass weight, total belly area and total fat area of 90.12 kg, 4616 cm² and 2384.51 cm², respectively. Previous studies showed that sex has no significant effect on carcass weight (Cisneros et al., 1996) and belly weight (Cisneros et al., 1996; Stupka et al., 2004; Scramlin et al., 2008). In contrast, Correa *et al.* (2006) reported heavier belly weight of barrow at different slaughter weight (115 kg and 125 kg) than gilts. Although pork belly muscle traits are comparable on both gender, individual pork belly muscle traits of female swine tended to become higher which resulted to significantly higher total muscle area (2307.053 cm²) and muscle percentage (54.89%) compared to barrow.

Table1. Least square means for Sex on carcass weight and pork belly muscle characteristics.

Traits	Sex	
	Barrow	Gilt
Carcass weight (kg)	90.12 ^a	87.56 ^b
Total belly area (mm ²)	4616.83 ^a	4236.23 ^b
Deep pectoral m., (mm ²)	104.48	108.04
Latissimus dorsi m., (mm ²)	214.93	219.34
Cutaneous trunci m., (mm ²)	473.01	486.96
Rectus abdominis m., (mm ²)	220.54	225.86
External abdominal oblique m., (mm ²)	354.39 ^b	374.35 ^a
Internal abdominal oblique m., (mm ²)	92.02	96.99
Total muscle (mm ²)	2228.71 ^b	2307.53 ^a
Total fat (mm ²)	2384.51 ^a	1928.78 ^b
Belly muscle (%)	48.72 ^b	54.89 ^a

^{a, b} different superscript on the same row is significantly different ($P < 0.01$).

Effect of carcass weight on pork belly muscle

Influence of carcass weight on pork belly characteristics is illustrated in Table2. Data suggest that carcass weighing 100 kg and above yielded highest total belly area and pork belly muscle areas while significant decrease of the corresponding traits were observed as the carcass weight

decreases. Furthermore, lowest pork belly muscle traits were observed in carcass weighting 79 kg below. Some authors noted belly weight was observed to increase as the slaughter weight increases (Correa et al., 2006; Correa et al., 2008).

Data suggest that as the carcass weight increases, pork belly and pork belly muscle area also increases; however observations showed that muscle percentage has indirect relationship with carcass weight. Highest muscle percentage was recorded in carcass weighting 79 kg below and 80-89 kg with 52.28% and 51.62% respectively, while lowest muscle percentage were comparable on carcass weighing 90-99 kg and above 100kg. Muscle percentage is an important factor in processing meat as according to Person *et al.* (2005) belly that contains muscle cutaneous trunci more than 50% yields highest percentage of bacon processed from pork bellies.

Table 2. Effect of carcass weight on pork belly traits.

Traits	Carcass weight			
	A	B	C	D
	<79kg	80-89kg	90-99kg	>100kg
Total belly(mm ²)	3734.24 ^d	4212.32 ^c	4734.08 ^b	5369.05 ^a
Deep pectoral m., (mm ²)	90.26 ^c	100.55 ^b	111.49 ^a	118.00 ^a
Latissimus dorsi m., (mm ²)	193.92 ^d	207.43 ^c	226.86 ^b	245.57 ^a
Cutaneous trunci m., (mm ²)	405.07 ^d	456.92 ^c	510.40 ^b	546.66 ^a
Rectus abdominis m., (mm ²)	194.81 ^c	213.86 ^b	239.00 ^a	242.74 ^a
External abdominal oblique m., (mm ²)	313.67 ^d	347.14 ^c	381.17 ^b	411.05 ^a
Internal abdominal oblique m., (mm ²)	820.86 ^d	92.43 ^c	101.24 ^b	110.05 ^a
Total muscle (mm ²)	1959.39 ^d	2172.33 ^c	2381.43 ^b	2533.86 ^a
Total fat (mm ²)	1785.80 ^d	2074.10 ^c	2369.46 ^b	2836.72 ^a
Belly muscle (%)	52.38 ^a	51.62 ^a	50.79 ^b	48.29 ^c

^{a, b, c, d} different superscript on the same row is significantly different ($P < 0.01$).

The results of the study showed that Sex and Carcass weight has significant effect on pork belly muscle characteristics; this can help in improving overall pork belly characteristics.

References

- Cisneros, F., M. Ellis, F. McKeith, J. McCaw, and R. Fernando. 1996. Influence of slaughter weight on growth and carcass characteristics, commercial cutting and curing yields, and meat quality of barrows and gilts from two genotypes. *J. Anim. Sci.* 74: 925-933.
- Correa, J., L. Faucitani, J. Laforest, J. Rivest, M. Marcoux, and C. Gariépy. 2006. Effects of slaughter weight on carcass composition and meat quality in pigs of two different growth rates. *Meat Sci.* 72: 91-99.
- Correa, J., C. Gariépy, M. Marcoux, and L. Faucitano. 2008. Effects of growth rate, sex and slaughter weight on fat characteristics of pork bellies. *Meat Sci.* 80: 550-554.
- Person, R., D. McKenna, D. Griffin, F. McKeith, J. Scanga, K. Belk, G. Smith, and J. Savell. 2005. Benchmarking value in the pork supply chain: Processing characteristics and consumer evaluations of pork bellies of different thicknesses when manufactured into bacon. *Meat Sci.* 70: 121-131.
- Scramlin, S., S. Carr, C. Parks, D. Fernandez-Dueñas, C. Leick, F. McKeith, and J. Killefer. 2008. Effect of ractopamine level, gender, and duration of ractopamine on belly and bacon quality traits. *Meat Sci.* 80: 1218-1221.
- Smith, K. N., Yong K.B., and W. S.L. 2014. USDA GAIN: Republic of Korea Livestock and Products Semi-annual 2014. The Pig Site.
- Stupka, R., M. Sprysl, and M. Pour. 2004. Analysis of the formation of the belly in relation to sex. *Czech J. Anim. Sci.* 49: 64-70.

Production and reproduction characteristics of Tegal and Magelang ducks

Dattadewi Purwantini¹⁾, Ismoyowati¹⁾ & Setya Agus Santosa¹⁾

¹⁾ Faculty of Animal Science, University of Jenderal Soedirman, Jl. Dr. Soeparno No. 60, Purwokerto 53122, Indonesia

Abstract

The objective of the present research was to describe and compare the production characteristics of Tegal and Magelang duck, comprising vital body measure (body weight, abdominal circumference, breast circumference, shank length, and neck length), egg production (egg weight, hatching weight and egg production), and reproduction characteristic (fertility, hatchability, and embryo mortality). Research materials were 196 Tegal ducks and Magelang ducks, each consisted of 16 males and 80 females. Experimental method was applied by calculating the mean and standard deviation, and comparing the characteristics of production and reproduction of Tegal and Magelang duck with one sample t-test. The results of mean and standard deviation of Tegal duck comprised vital body measure were (1392,74±117,99 g; 26,97±2,71 cm; 26,25±1,33 cm; 20,26±1,03 cm; 6,31±0,35 cm and 20,27±1,63 cm, respectively), egg production (67,25±5,71 g; 44,19±3,33 g and 66,41±12,84 %, respectively) and reegg production (86,87±9,12 %; 43,02±14,15 % and 56,95±16,16%, respectively). While in Magelang duck the result demonstrated vital body measurement (1612,18±122,74 g; 27,65±0,88 cm; 27,75±1,44 cm; 22,28±1,75 cm; 6,53±0,47 cm and 21,80±2,08 cm, respectively), egg production (67,91±2,56 g; 46,43±4,37 g and 65,08±11,80 %, respectively) and reproduction characteristics (87,14±1,54 %, 50,53±11,45 % 49,43±2,09%, respectively). T-test result showed that vital body measurement, egg production and production characteristics between Tegal duck and Magelang duck was significantly different, in which the latter showed relatively higher performance than the former, but egg production percentage of the former outperformed the latter. Tegal duck and Magelang duck crossbreed was viable to obtain offspring with more superior vital body measure and egg production percentage.

Key words: characteristics, production, reproduction, Tegal duck, Magelang duck

Introduction

Magelang and Tegal duck are two of 15 native Indonesian ducks bred in Central Java Province with relatively high population among the other ducks. In 2010, duck population in Central Java reached 4.848.263 birds and continued to increase to 5.006.163 birds in 2011 (Statistics Indonesia and Central Java Regional Development Agency, 2012). Purwantini et al. (2013) reported that Tegal duck and Magelang duck have a closer genetic relation and share common maternal inheritance with *Anas platyrhynchos* and *Anas zonorhyncha* shown by the genetic distance of 0.000 – 0,019.

Egg production of Magelang ducks are specific and superior with relatively large body, high egg production and more varied plume color than the other native ducks. Purwantini et al. (2013) reported 11 plume colors of Magelang duck, Subiharta et al. (2013) stated that Tegal duck with *Branjangan* plume color (dominant dirty brownish white with dark brown spots) has the highest egg production compared to the other plume color. The plume color of Tegal ducks are relatively similar, namely 99.44% *Branjangan*, 0.24% white and 0.32% mixed black-brown.

Chavez and Lasmini (1978) reported that 50% of Tegal ducks population have egg production above 60%. Subiharta et al. (2013) stated that only 25% ducks have 65% egg production and 50% of the population has less than 50% egg production.

The objective of this research was to describe and compare the production characteristics of Tegal duck and Magelang duck, including vital body measure, egg production and reproduction characteristic. The significance of the research was to provide the basis of projected selection and crossbreeding policy.

Materials and methods

Research materials were 196 birds consisted of Tegal and Magelang duck, each comprised 16 male and 60 female. Ducks were kept in 16 group cages with 1 male and 5 female, measuring 250 cm long, 250 cm wide and 70 cm high, completed with individual cage to manage individual production record.

Experimental method was applied by calculating the mean and standard deviation and comparing the production and reproduction characteristics of Tegal and Magelang duck using one sample t-test according to Steel and Torrie (1998).

Result and discussion

Production characteristics. Research results reported mean and standard deviation of the characteristics of vital body measure and egg production of Tegal and Magelang duck as presented in Table 1.

Tabel 1. Mean and standard deviation of characteristics of vital body measure and egg production of Tegal and Magelang ducks.

Production Characteristics	Mean and standard deviation	
	Tegal duck	Magelang duck
Body weight (g)	1392,74 ± 117,99 ^a	1612,18 ± 122,74 ^b
Abdominal circumference (cm)	26,97 ± 2,71 ^a	27,65 ± 0,88 ^b
Breast circumference (cm)	26,25 ± 1,33 ^a	27,75 ± 1,44 ^b
Body length (cm)	20,26 ± 1,03 ^a	22,28 ± 1,75 ^b
Shank length (cm)	6,31 ± 0,35	6,53 ± 0,47
Neck length (cm)	20,27 ± 1,63 ^a	21,80 ± 2,08 ^b
Egg weight (g)	67,25 ± 5,71	67,91 ± 2,56
Hatching weight(g)	44,19 ± 3,33 ^a	46,43 ± 4,37 ^b
Egg production (%)	66,41 ± 12,84 ^b	65,08 ± 11,80 ^a

1Footnote: superskrip a, b different in the same rows indicate significant in HSD test (P <0.05)

Tabel 1. demonstrated that vital body measure of Magelang duck were relatively larger than those of Tegal duck, while egg production of Tegal duck was relatively higher than that of Magelang duck. According to Subiharta et al. (2013) Tegal ducks have above 70% egg production and belong to the category of early fast laying than the other native ducks. The suggested initial age of native ducks to lay egg is 150-170 days old. Ducks were kept in uniformed environment either the feed or management so that the similar characteristics shown indicated genetic variation. Observation on egg production was performed during the first 156 days of initial production, body measurement was done three times and the measured values were averaged.

Reproduction characteristics. The mean and standard deviation of reproduction characteristics of Tegal and Magelang ducks are presented in Table 2.

Table 2. Mean and standard deviation of reproduction characteristics of Tegal and Magelang duck

Reproduction characteristics	Mean and standard deviation	
	Tegal duck	Magelang duck
Fertility (%)	86,87 ± 9,12 ^a	87,14 ± 1,54 ^b
Hatchability (%)	43,02 ± 14,15 ^a	50,53 ± 11,45 ^b
Embryo mortality (%)	56,95 ± 16,16 ^a	49,43 ± 2,09 ^b

1Footnote: superskrip a, b different in the same rows indicate significant in HSD test (P <0.05)

Tabel 2. demonstrated that fertility and hatchability of Magelang ducks were relatively higher than those of Tegal duck, but the embryo mortality was lower. High hatchability in Magelang ducks was related to the high fertility, laying condition, and maternal and paternal genetic factor. Relatively high embryo mortality in Tegal duck was affected of relatively low hatchability compared to Magelang duck. Yuwanta et al. (2001) stated that Turi duck in Yogyakarta had 74,51 – 75,42% fertility. The high fertility in Tegal and Magelang duck compared to Turi duck was assumedly due to the relatively optimum ratio of male and female, namely 1:6-8.

Result of t test. The significance of production and reproduction characteristics of Tegal and Magelang duck was performed by means of t test. Result showed that t count value for vital body measure, it indicated that t count was higher than t. Accordingly, vital body measure of Tegal duck was significantly different from that of Magelang duck. Result of t count in egg production characteristics, it indicated that reproduction characteristics between Tegal duck and Magelang duck was significantly different, in which Magelang duck was relatively more favorable than Tegal duck.

Conclusion

Conclusively, production and reproduction characteristics of Magelang ducks were superior to those of Tegal duck. Crossbreeding Tegal and Magelang ducks was viable to obtain offspring with more superior vital body measure and egg production percentage.

References

- Chavez and Lasmini, 1978. Comparative performance of native Indonesia egg laying duck. Center Report No. 6. Center for Animal Research and Development, Bogor.
- Purwantini, D., T. Yuwanta, T. Hartatik and Ismoyowati, 2013. Morphology and Genetic Diversity of Mitochondrial DNA D-Loop Region Using PCR-RFLP Analysis in Magelang Duck and Other Native Duck. The Journal of The Indonesian Tropical Animal Agriculture, Vol 38 No 1.
- Purwantini, D., T. Yuwanta, T. Hartatik and Ismoyowati, 2013. Polymorphism of D-Loop Mitochondrial DNA Region and Phylogenetic in Five Indonesian Native Duck Population. Int. J. Poult. Sci., 12 (1): 55 - 63.
- Statistics Indonesia and Central Java Regional Development Agency, 2012. Jawa Tengah in Number 2012. Central Java BPS and BPJ Cooperation
- Steel, R.G.D and J.H. Torrie, 1998. Principle Procedure of Statistics Indeks. Translated by Bambang Sumantri. Statistics Principle and Procedure: A Biometric Approach. Third Edition. PT. Gramedia Pustaka Utama, Jakarta
- Subiharta, D.M. Yuwono and P. Sudrajad, 2013. Characteristics of Tegal (*Anas platyrhynchos javanicus*) as Superior Native Layers in Central Java and the Production Optimizing Effort. National Seminar: Projecting the Rise of Local Agricultural and Marine Superior Seedling Commodity. Aculty of Agriculture Trunojoyo University Madura.
- Yuwanta, T., Zuprizal, A. Musofie dan N.K. Wardani. 2001. Production and Reproduction of Turi Bantul Ducks on Sex Ratio, Old Mix and Maintenance System Different. Tropical Livestock Development. University of Diponegoro, Semarang, pp:68-78.

Breeding of Bali cattle as indigenous beef cattle breed in Sumbawa island Indonesia

Nurgiartiningsih, V. M. A., L. Hakim & A. Budiarto

University of Brawijaya, Faculty of Animal Husbandry, Jl. Veteran, Malang 65145, Indonesia

Abstract

Study on breeding potency of Bali cattle was conducted in station of Bali cattle breeding and forage in Serading, Sumbawa, West Nusa Tenggara, Indonesia. Data on birth weight, population structure and mortality during 2009 to 2013 were collected. Data were analyzed using analysis of variance applying software of Genstat version 16 (Anonymous 2014). Result showed that mean of birth weight of Bali cattle during 2009–2013 was 14.93 ± 1.90 kg. During 3 years of study, birth weight of Bali cattle increased, which was 14.45 kg in 2009 to be 16.19 kg in 2011. However, it then reduced to be 13.47 kg in 2013. It was found that birth weight of male was significantly different to those of female, which were 15.38 ± 2.26 kg and 14.20 ± 1.83 kg for male and female, respectively. The male to female ratio was 3 to 78. Proportion of adult, young and calves were 34.62%, 17.95% and 47.43%, respectively. The result indicated that there are improvement of performance and genetic quality of Bali cattle; although it increased only 16.85% during 12 years. This ratio of male to female cattle was categorized ideal. Considering the availability of feed resources in West Nusa Tenggara and high adaptability of Bali cattle in Sumbawa Island, there is high potency of improving genetics and performance of Bali cattle in Sumbawa, West Nusa Tenggara, Indonesia.

Keywords: birth weight, mortality, population structure

**Corresponding author: vm_ani@yahoo.com*

Introduction

Bali cattle are one of the important beef cattle breeds contributing to the development of livestock in Indonesia, and are the most predominant genotype within Sumbawa island Indonesia. Taxonomical names of Bali cattle are *Bossondaicus*; *Bosjavanicus*; *bos/Bibosbanteng*. This indigenous beef cattle breed plays an important role in increasing beef meat production to meet the national demand. Bali cattle has superiorities including high fertility, high tolerance to heat stress and having dual purpose functions for draught and beef (Anonymous, 2010). Bali and Nusa Tenggara have the third largest population of beef cattle in Indonesia. Population of Bali cattle in those two areas in 2013 was 2.1 million, which was approximately 43.75% of the total number of Bali cattle in Indonesia. To support the development of Bali cattle in West Nusa Tenggara and national program in self sufficient of cattle meat, the local government has launched a slogan “West Nusa Tenggara as One million cattle area”.

Recently, artificial insemination using exotic breed have been executed in West Nusa Tenggara Island. Application of crossbreeding for long period will lead to the extinction of Bali cattle. In order to enhance the improvement of Bali cattle in Nusa Tenggara Island, apart from the Centre of Bali cattle breeding station in Bali that belonged to Direktorat General of Livestock and Animal Health Republic of Indonesia, there is also a station of Bali cattle breeding and forage in Serading, Sumbawa island that belonged to Department of Livestock and Animal Health of West

Nusa Tenggara. Both institutions have responsibility in improvement and distribution of high genetic potency of Bali cattle.

Adapted breeding strategy for Bali cattle in Sumbawa should be applied to improve the genetic potency in the existing condition and increase Bali cattle population. This research was conducted to explore and evaluate the performance and genetics potency of Bali cattle as local genetic resources of Indonesia.

Materials and Methods

Research was conducted in station of Bali cattle breeding and forage in Serading, Sumbawa West Nusa Tenggara. Two hundred and eighty data on birth weight, population structure and mortality during 2009 to 2013 were collected and analyzed. Data were analyzed using software GENSTAT 16 (Anonymous, 2014).

Results and Discussion

Population Structure

Population structure of Bali cattle in Serading are presented in Table 1, as follows:

Table 1. Population of Bali cattle in Bali cattle breeding and forage Serading 2014.

Composition	Number of Population		
	Male	Female	Total
Adult	3	78	81
Young	9	33	42
Calves	59	52	111
Total	71	163	234

The male to female ratio was 3 to 78, which equal 1 to 26. This ratio was categorized as ideal according to Gary (2014), who stated that the ideal male to female ratio was a 4 to 5 male per 100 females. Proportion of adult, young and calves were 34.62%, 17.95% and 47.43%, respectively. Calving rate of female calves was 64-67% which was categorized good and can be used as selected population for candidate dam as long as those calves were offspring of best dam.

The proportion of calves was high with the value of 47.43% of total population. The high number of male calves (53.15%) showed that male replacement stock was in good condition, which meant that replacement stocks are available for maintaining the sustainability of population structure. On the other hand, replacement stock for female was not sufficient for the shorter time due to the low proportion of young female cow in the population which was only 20.25%. If they will be used as replacement stock, it will still take long time to do selection for production performance and phenotypic characteristic.

Birth Weight of Bali Cattle

Mean of birth weight of Bali cattle in Serading during 2009-2013 was presented in Table 2. Mean of birth weight was lower than birth weight of Bali cattle in Bali reported by Budiarto et al (2011) and Martojo (2003) which were 17.75 kg and 16.8 respectively. Mean of birth weight of Bali cattle in Serading was higher compared to mean of birth weight in West Nusa Tenggara in year 2002, which was 12.7 kg. It indicated that there are improvement of performance and genetic quality of Bali cattle, although it increased only 16.85% during 12 years.

Table2. Mean of birth weight, number of sire, dam and offspring of Bali cattle.

Year	Number of Sire	Number of Dam	Number of Offspring	Birth Weight (kg)	SEM
2009	3	40	40	14.45 ^a	0.25
2010	4	65	65	15.51 ^b	0.26
2011	1	54	54	16.19 ^b	0.24
2012	1	43	43	15.02 ^a	0.33
2013	1	79	79	13.47 ^c	0.21
Mean				14.93	0.13

^{abc}Means in the same column without common letter are different at $P=0.01$.

Statistical analysis showed that there are significant differences of birth weight in different year. During 3 years of study, birth weight of Bali cattle increased, which was 14.45 kg in 2009 to be 16.19 kg in 2011. However, it then reduced to be 13.47 in 2013. The reason for this condition seemed to be due to the increased number of calves in year 2013, which only 43 calves was in 2012 and increased 79 calves in 2013. Another reason might be due to the low genetic quality of sire. Since 2011, Bali cattle breeding of Serading has applied specific program in which only one sire per year has been allowed to breed female. This breeding program should be evaluated genetically and also economically, considering that other sires have to wait for long time to be able to breed. This condition may cause decreasing the ability of productive sire to produce more offsprings. Further affect will reduce genetic quality of sire because the reproductive and genetic potency of older sire must be decreasing with increasing age.

Reference

- Anonymous, 2010. Program Swasembada Daging Sapi 2014. Kementerian Pertanian Direktorat Jenderal Peternakan. Jakarta
- Anonymous, 2014. GENSTAT Release 16. VSN International Ltd.
- Budiarto, A., L. Hakim, S. Maylinda, V. M. A. Nurgartiningih and G. Ciptadi, 2011. *Output of Bali Cattle in Base Population as Genetic Resources*. Research Report. Faculty of Animal Husbandry. Universitas of Brawijaya. Malang.
- Gary, L. A., 2014. *Diagnosing Reproductive Problems in Beef Cattle Herds*. Cattle Research and Education Center
- Martojo, H., 2003. *Indigenous Bali Cattle: The Best Suited Cattle Breed for Sustainable Small Farm in Indonesia*. Laboratory of Animal Breeding and Genetics, Faculty of Animal Science. Bogor Agricultural University. Indonesia

Institutional development on conservation of Madura cattle

*Siswijono**, S. B., Z. Fanani, B. A. Nugroho & V. M. A. Nurgiartiningsih

University of Brawijaya, Faculty of Animal Husbandry, Jl. Veteran, Malang 65145, Indonesia

Abstract

Study on institutional development on conservation of Madura cattle was aimed to empowerment of farmers, to improve production performance and to conserve Madura cattle as genetic resource of Indonesia. Survey method was applied to collect data using conversation technique and observation participation. Result showed that most of the farmers managed 1-4 cattle as their own, but there were 32.5% farmers managed cattle using profit sharing system. Reproductive performance of Madura cattle was categorized good. service per conception (S/C) was 1.5 and calving interval was 18 months. Artificial insemination using frozen semen of exotic breed has been applied institutionally in small holder farmer and farmer tended to prefer exotic breed than Madura cattle as local breed. Economic aspect was the main reason for choosing crossbreeding. Without good breeding program, cross breeding could threaten the existence of Madura cattle as genetic resource of Indonesia. Government policy to determine Sapudi island as breeding centre for Madura cattle was an institutional change in conservation of Madura cattle. In order to conserve Madura cattle, education and supervision to the farmers should be done continuously. Economic incentive as a reward for Madura cattle farmer should be given for developing of Madura cattle conservation model.

Keywords: institutional, conservation, genetic resource

**Corresponding author: suprih_bs@yahoo.com*

Introduction

Population of Madura cattle in Madura Island in 2012 was 905,271, which occupied 18.26% of total beef cattle population in East Java. Increasing the number of Madura cattle in Madura was tended to be lower than increasing in East Java. It was reported that performance of Madura cattle in Madura Island was lower than those in East Java.

Implementation of artificial insemination using high quality beef bull breed expected to improve performance of Madura cattle. Farmer in Madura Island showed high preference in crossbreeding system in order to improve performance of their Madura cattle (Nurgiartiningsih, 2011). This condition has negative effect on conservation of Madura cattle as local genetic resources of Indonesia.

This research was done to determine institutional model of Madura cattle conservation in Madura Island as a strategy in conservation of local beef cattle breed and in improvement of Madura cattle performance.

Material and Method

Research was done using survey method in Bangkalan and Sampang districts, Madura Island. Primary and secondary data were collected using technique of interview and participation observation.

Respondents as source of data were randomly sampled farmer. Data were analyzed descriptively, which were evaluated and correlated the result and social and economic aspects of technical parameter in beef cattle breeding.

Result and Discussion

Characteristics of cattle farming system

Result of survey showed that most of household farmer (97.5%) kept 1 to 4 cattle, in which 55% has 1 to 2 cattle and 42.50% has 3 to 4 cattle.

Reproductive performance showed that service per conception was 1.5 and calving interval was 18 months. Those results indicated that reproductive performance of Madura cattle was categorized good. Nurgiartiningsih (2011) reported that reproductive performance of Madura cattle in four districts in Madura Island was 1.37.

Institutional conservation of Madura cattle

Data in Table 1 presented the information on perception of cattle farmer on crossbreeding of Madura cattle with exotic breed bull.

Table 1. Distribution of farmer's perception on crossbreeding of Madura cattle.

Perception	Number of Respondent	Percentage
Disagree	11	22.50
Neutral	19	47.50
Agree	10	25.00
Total	40	100

Research indicated that 72.50% of cattle farmer tended to agree about crossbreeding as a solution to improve Madura cattle performance. Only 22.50% of farmer didn't agree about crossbreeding. In fact, crossbreeding of Madura cattle with could increase income of farmer. Nurgiartiningsih, dkk (2011) reported that the higher the blood percentage of Limousine the higher the price of crossbred.

Data on cattle farmer that choose bull breed as source of frozen semen was presented in Table 2.

Table 2. Distribution of farmer choosing bull breed as source of frozen semen in Madura.

Bull breed	Number of Respondent	Percentage
Madura cattle	7	26.90
Limousine	19	73.10
Total	26	100

Data on Table 2 showed that 73.10% of cattle farmer choosing crossbreeding using exotic breed Limousine based on economic consideration. Crossbreeding Madura cattle with Limousine increased body weight of 850 kg during 3-4 years (Kustiyah, 2012). Information on crossbreeding in Madura indicated that Madura was changed to be an opened area. One effective

solution in conservation of genetic resources was through developing institutional regulation which guarantee the protection of biodiversity (McNeely, 1992). Therefore, in order to conserve Madura cattle, government has already determined Madura cattle as indigenous breed of Indonesia and Sapudi Island as area of Madura cattle conservation.

Government policy to determine Sapudi island as breeding centre for Madura cattle was an institutional change in conservation of Madura cattle. Issue in the future would appear when farmer realized that economic rewards of Madura cattle breeding was lower than those of crossbreeding. It is an interesting case because farmer prefer crossbreeding due to economic aspect. Base on the situation, application of crossbreeding should be managed properly in order to prevent extinction of Madura cattle. Based on economic aspect as main reason of agree in crossbreeding, institutional of Madura cattle conservation should be fulfilled the economic aspect.

Conclusion and Suggestion

It could be concluded that 1. Economic aspect was the main reason of farmer to agree about crossbreeding using exotic breed 2. Determination of Sapudi Island as breeding centre of Madura cattle was an institutional change in conservation of Madura cattle 3. Development of institutional conservation of Madura cattle should considered economic aspect.

In order to conserve Madura cattle, education and supervision to the farmers should be done continuously. Economic insentive as a reward for Madura cattle farmer should be given for developing of Madura cattle conservation model. Madura society should be involved in development of institutional conservation of Madura cattle.

References

- Keputusan Menteri Pertanian Nomor: 3735/Kpts/HK.040/11/2011 tentang penetapan Rumpun Sapi Madura.
- Keputusan Direktur Jenderal Peternakan Nomor: 18020/Kpts/PD.420/F2.3/02/2013 tentang Pedoman Pelaksanaan Pembibitan Sapi Potong di Pulo Raya, Pulau Sapudi, Pulau Nusa Penida.
- Kutsiyah F, 2012. Analisis Pembibitan Sapi Potong Di Pulau Madura. *Wartazoa* Vol. 22 No. 3.
- McNeely, J. A., 1992. *Ekonomi dan Memanfaatkan Perangsang Ekonomi Untuk Melestarikan Sumberdaya Hayati*. Edisi Pertama Yayasan Obor Indonesia. Jakarta.
- Nurgiartiningsih, V. M. A., S. B. Siswijono, Kusmartono, Hartutik and I. Subagiyo, 2011. *Pemetaan Hasil Persilangan Sapi Madura Di Pulau Madura*. Laporan Penelitian Fakultas Peternakan kerjasama dengan Dinas Peternakan Jawa Timur.

Annual trend of genetic improvement for production performance of three swine breeds

Kim, T. H. *, Y. H. Kim, H. S. Kim, J. H. Hong, V. S. Viterbo, B. M. Lopez, M. T. Makumbe
K. C. Nam & K. S. Seo

Department of Animal Science and Technology, Suncheon National University, Suncheon,
540-742, South Korea

Abstract

This study was conducted to determine the annual trends of improvement for Korean swine test traits. Loin depth (LD), meat percentage (MP), eye muscle area (EMA), days at 90kg (D90), and modified backfat at 90kg (BF) were used for Landrace, Yorkshire and Duroc breeds from South Korea Stock Farms. Basic statistical analysis was done using SAS version 9.1 on 592,965 animal records collected over a 25year period (1991 to 2014). Highest LD was 60.68 in 1992 and decreased to 48.36 in 2015. MP and EMA values showed similar to the LD. Highest value of EMA was 39.0 in 1992 and this decreased every year. D90 showed an increasing trend. There was no clear trend for BF with big differences among years. Least squares means for all other test traits decreased except for D90 and BF. D90 had acubic equation trend. LS means for D90 decreased from 1991 to 1999. D90 increased until 2000 then decreased the following years. LS means for BF followed a decreasing trend. LS means for BF were 16.17 and 13.17 in 1991 and 2014. D90 and average daily gain improved over the period under study, contributing to increase in per capita pork consumption. Consumers prefer belly to loin meat portions leading to decrease in LD, EMA and BF. Korean swine breeders' goals need to be focused on improving LD or EMA and meat quality to meet consumer demands.

Keywords: annual trend, test traits, production performance, least square means

Corresponding author: popgen@daum.net

Introduction

In Korea, per capita consumption of meat is persistently increasing. Per capita consumption of meat in 2010 was 38.6kg (8.8kg for beef, 19.1kg for pork and 10.7kg for chicken). This has since doubled for beef. Pork consumption has increased 1.4 times in comparison to the 1990s. The swine industry constitutes 42% (US\$ 48 million) in Korean livestock industry. Commonly raised include Landrace, Large White and Duroc, with increased interest in Berkshire because of its meat quality traits. The portions of pork preferred by Korean consumers are tenderloin, loin, blade shoulder and belly (Kim, 2009).

Materials and Methods

Animals

Landrace, Large White and Duroc boars and gilts data from 6 Korean Great Grand Parent (GGP) farms collected from 1991 to 2014 was used. Total heads of pigs in the data set were 592,965. Large White breed constituted the largest portion of heads (64.15%). Three quarters of test pigs

were females. Because data was from different farms, it was checked and abnormal data which was beyond $\pm 4\sigma$ was removed.

Table 1. The frequency of breeds and sex.

Breeds	Heads	%	Sex	Heads	%
Landrace	122,483	20.66	Male	150,277	25.34
Large White	380,398	64.15	Female	442,688	74.66
Duroc	90,084	15.19			
Total	592,965	100	Total	592,965	100

Traits

The traits investigated were Loin Depth at the end of trial (LD), meat percentage (MP), eye muscle area (EMA), days at 90kg (D90), and modified backfat at 90kg (BF). LD and EMA were measured using an A-mode ultrasound device (PIGLOG 105) between the 3rd and 4th last rib and 3 inches away from the midline. Lean MP was acquired by getting the percentage weight of the whole carcass. The individual body weight, age and average daily gain (ADG) of pigs were obtained to calculate D90kg. BF was measured using an A-mode ultrasound device (PIGLOG 105). BF thickness measurements were taken at three points: scapula posterior position around the 4th or 5th rib, last rib and waist (last lumbar vertebrae) at 4cm to the dorsal line; and the average of the three readings calculated for live pig body. BF was calculated using the formula below.

$$BF = \text{BFt measurement} + \frac{(90 - \text{test end weight}) \times \text{backfat measurements}}{(\text{test end weight} - 11.34)}$$

Analysis

All traits were analyzed using SAS ver 9.1 software and Microsoft Office Excel program.

Results and Discussion

From 1991 to 1997, LD decreased but showed no changes after 1997. There was decreasing trend every year for MP. EMA and LD had almost similar trends. There was a decreasing trend in D90. BF thickness trend decreased until 2006 and then increased in the subsequent years. The reduction in D90 over the study period can be attributed to genetic gains in ADG through selection, and improvement in feed and feeding programs. This increased pork turnover rates and per capita pork consumption in Korea. Annual deviations in trend for traits analyzed could largely be attributed to the swine breeding programs and to the genetics-environment interactions associated with the possibility of disease occurrence and or other factors such as management and feed across the six GGP farms.

Conclusion

D90 and ADG in swine were improved from 1991 to 2014 in Korea. Meat quality and production traits were not improved over the same study period. Consumers still demand high quality meat and the Korean swine breeders' goals need to be focused at meeting these demands by improving LD or EMA and meat quality.

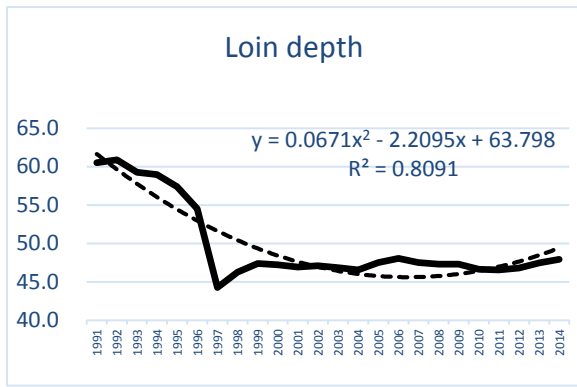


Figure 1. Trend of annual loin depth.

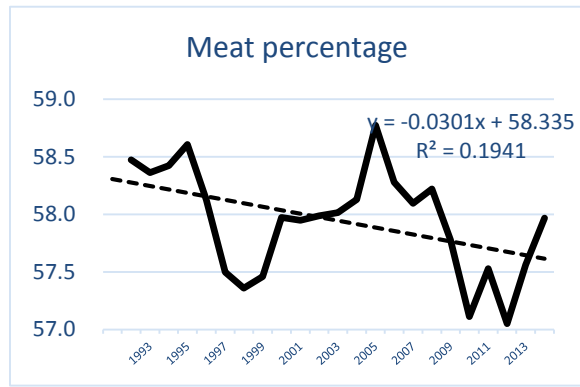


Figure 2. Trend of annual meat percentage.

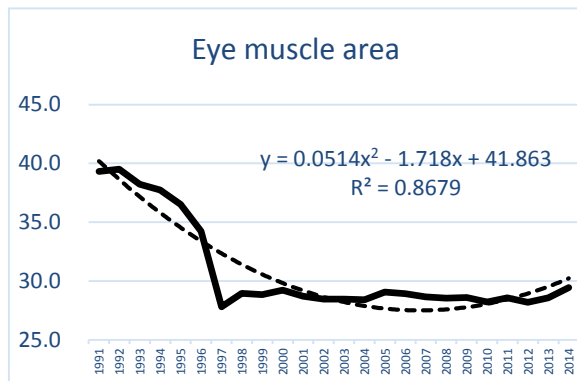


Figure 3. Trend of annual eye muscle area.

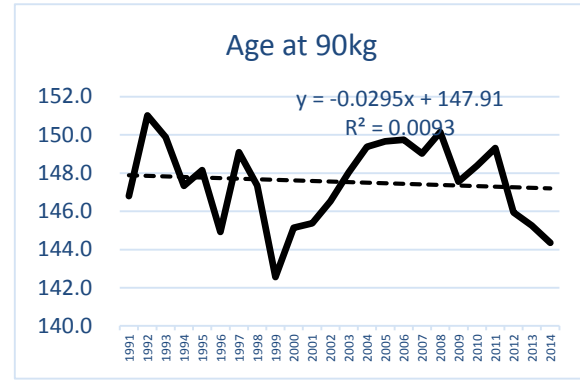


Figure 4. Trend of annual average daily gain

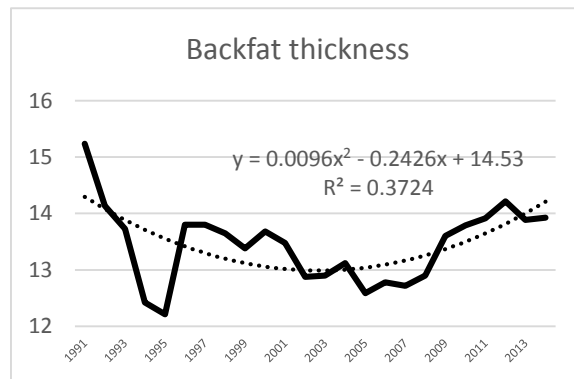


Figure 5. Trend of annual backfat thickness.

References

- Kim, Eun-Mi, S. H. Seo, M. A. Lee, K. H. Kwon, G. H. Jun, 2010. Preference and Consumption Patterns of General Consumers of Meat Dishes. *J. Korean Soc. Food Cult.* 25: 251-261.
- Korean National Statistical Office, 2014. Korean Agricultural Statistical Information Services. http://kosis.kr/statisticsList/statisticsList_01List.jsp.
- Ministry of Agriculture, Food and Rural Affairs, 2014. Agricultural main statistical references. <http://library.mafra.go.kr/skyblueimage/14566.pdf>.

Phenotypic variations and chromosome analysis of swamp buffalo from isolated area in Kangean Island East Java Indonesia.

G. Ciptadi,^{*1,2}, V.M.A.Nurgiartiningsih^{*1}, Marjuki^{*1}, S. Wahjuningsih^{*1}, Moh. N.Ihsan^{*1} and Mudawamah^{*3}

¹Faculty of Animal Husbandry, UB, Malang, Indonesia, ². Bioscience Laboratory, UB, Indonesia. ³ Faculty of Animal Sci., Islamic University of Malang.

Abstract

The aims of this research are to identify the phenotypic variation and to analyze the chromosome characters of the swamp buffalo in Kangean Island, East Java Indonesia. Chromosome analysis was performed to the buffalo with high level of inbreeding. The 0.5 ml of blood sample per animal was added to 5 ml chromosomal medium (Karyo MAX, Gibco). After 70 hours add to 1 ml colchicines, centrifuge at 1000 RPM for 10 minute and added then by fixative solution. Slides were stained with Giemsa. Analysis have been done by cytovision image (Genetix,USA). Phenotypic variations were observed base on the skin color. Result showed that the 2 N diploid chromosome were observed normal, male/female buffalo was 48 (46 autosomes and 2 sex chromosomes: XY/XX). All the autosomes and sex chromosomes were found normal. The phenotypic character in isolated area with high inbreeding was varied in their color of skin from grey (normal) to red and white (abnormal). Analysis of banding quality of each chromosome showed different intensity. It was recommended to performed chromosomal investigation of breeding buffalo, especially for male buffalo for commercial semen production in the near future.

Keywords: Buffalo breeding, Chromosome, AI, Genetic, Inbreeding.

**Corresponding author: ciptadi6@gmail.com and ciptadi@ub.ac.id*

Introduction

The potential production and reproduction of Indonesian buffalo is relatively limited. The main function of buffalo is generally as a working animals and meat producer. Buffalo in Kangean Island has another specific function, particularly related to tourism in the district of Arjasa as buffalo racing. Total buffalo in this island is estimated at about 5,187 heads (Nurgiartiningsih et al, 2012). The fact of the variability in skin color aberrant phenotype especially the red color is also a concern related to the Madura culture.

The purpose of this activity is to identify the genetic (chromosome) of buffalo with specific characteristics of skin color (white, grey, and red) that exist in the island Kangean, East Java. Activities carried out include two main activities: (1) phenotypic identification /morphometric and (2) karyotyping of buffalo chromosome.

Materials and Method

Research was conducted to characterize phenotypic and genetic (karyotyping) of swamp buffalo. The data was collected by purposive sampling which focused on the buffalo with abnormalities or deviations skin color (white and red) that exist in this island. Blood samples were taken as much as 6 buffaloes derived from Kangean Island for chromosome analysis with standard G-banding and analysis using software cytovision assistance. As a comparison (control) of blood samples taken is four (2-head) buffalo gray outside this island. The output of the research is the data

performance (phenotypic) and chromosome data (normality/genetic abnormalities). Data obtained could be subsequently used for the consideration of the development strategies of local Indonesian buffalo.

Results and Discussion

Phenotypic Variation of Buffalo

The buffalo that was existed in Kangean Island was gray (dark and light), which reached 81.25% of the total sample observed. White buffalo and red appear in very small percentages, respectively 8.33% and 10.42% (n=48). White color, although the price is relatively expensive at present conditions are not favored by farmers because based on past experiences of the white buffalo that require special care (Figure 1.). Until now, white buffalo is used to show an annual ritual and custom events. White and red buffalo is one of the specific characters that appear in Kangean island that may be used as a phenotypic trade mark Madura Buffalo. These new traits that can be identify, characterize and then can be used as the basic of inheritance.

Newborn buffalo colored is white and then in line with increasing of age it will be varied such as black, gray or red. Observations in the field indicate that the gray buffalo are red on a particular body part that is on the abdomen, thighs, and there are white on the ankle down (Figure 1.). It is still needed further analysis of morphological forms chromosomes and DNA analysis in order to more accurately ascertain the pattern of inheritance between phenotypic characters color of gray, red and white. Actually this black color were exist, but their color origin is grey then treated by farmer to be black, especially for the racing buffalo. For the near future, it need to do continuous recording to provide more evidence about the inheritance pattern of red buffalo leather character as well as to avoid inbreeding.

An interesting finding that traditional buffalo race became one of the driving households to buy a buffalo which was then kept as a special buffaloes are maintained for the benefit of the race. Research result of Nurgiartiningsih et al, (2012.) reported an information obtained through interviews respondents that the public does not know the history of the origin of buffalo livestock farming in this region. Genetic Resources of local buffalo, especially in Kangean, Madura has not been widely described or analyzed in a clear and specific, particularly at the cellular and molecular level. Government policies for the preservation of germ plasm native Indonesia requires basic data necessary support to mapping the real conditions and potential Madura buffalo that exist today.

Chromosome Analysis (band Intensity)

The results showed that buffalo is genetically normal, based on the level of chromosomes are diploid chromosome $2N = 48$ standard karyotyping for swamp buffalo. The inheritance pattern tends to codominant phenotype associated with emergence of red or characters also may be a polygenic nature of qualitative character. Male buffalo is used to practice of natural mating/service without good recording which is occurrence of inbreeding with the possible emergence of characters homosizygotic harmful recessive.

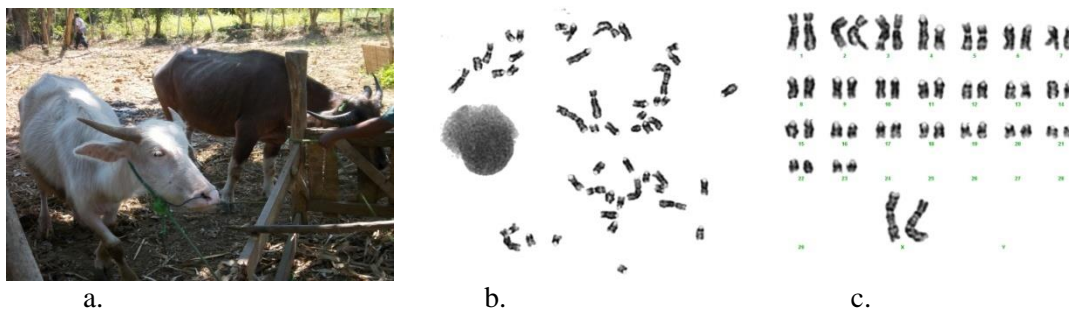


Figure 1. Phenotypic character of swamp buffalo in Kingman Island, Madura : a. Normal skin grey color and white skin color. b, and c . Spreading chromosome staining by G- banding and karyotyping.

Based on the number of chromosomes are genetically Kangean buffalo is categorized in swamp buffalo. The local water buffalo in Asia are generally classified into two main sub species based on the size of the body, appearance, the biological characteristics and types chromosome namely river buffalo ($2N = 50$) and swamp buffalo ($2N = 48$). General characteristics buffalo skin body is gray color, drooping neck and has a large horn that leads to the rear. In general, Kangean swamp buffalo is often found in swamp, river and beach area or rice field area. Meanwhile river buffalo has a body color grey-black or dark gray and horn elongated circular or straight back.

There are several possible causes of the characters of red and white phenotype on Kangean buffalo. The first cause is may be the inheritance pattern of codominance. The second estimation, character of a new phenotype white or red may also causes of the influence of multiple genes/polygenic. Red and white characters on a buffalo Kangean possibility controlled by polygenic effect that works in the relay in accordance with the development of age and hormonal expression. Supanuam et al (2009), appearance of the white buffalo is not an albino character, but the effect could not be identified genetic cause. The Kangean also found the existence of dark shiny black buffalo, found in the arena of racing animal. Based on the research results through in depth interviews to several farmers, it was resumed that the shiny black color is modified by means of greasing racing buffalo skin with oil.

Generally not found Kangean buffalo with abnormalities of chromosome number. It is understood that in all groups there were no buffalo several chromosomal abnormalities such as translocations Robertson ($2N = 47$) (Gupta and Chadhri, 1978, Guvtavsson 1979) or another chromosome abnormality. Kangean Buffalo is suspected there is no genetic abnormalities. However, with the problems in the field concerning reproduction performance were interesting to study the sex chromosomes (XX and XY chromosomes), the chromosome morphology of sex chromosomes is necessary to be done in the future, especially for commercial breeding purposes (Ahmad, 2014). The results showed that in all buffaloes based on the size and sharpness of images of chromosomes between cells with one another shows the same result that everything is normal, although there are variations. At all buffalo were observed for N diploid chromosome of 48 normal. The X chromosome is sub metacentric with the largest size, otherwise the Y chromosome is the smallest size. Autosomal of chromosomal Kangean buffalo is acrosentric.

Acknowledgements

This work was supported by Research Grant of HIKOM (Hibah Kompetensi-2014/2015) from Direktorat General of High Education-DIKTI, Ministry of Education Republic of Indonesia. No Contract in accordance DIPA UB No. 023.04.2.414989/2014. and the DIKTI No. 0261 / E5 / 2014, January 2014.

References

- Ahmad, I, K. Javed1 and A. Sattar. 2004. Screening of breeding Bull of Different Breeds through Karyotyping. *Pakistan Vet. J.*, 24(4)
- Gatot Ciptadi. 2014. *Genetics and Animal Livestock Breeding*. Brawijaya University.
- Gupta, P. and S. P R. Chaudhri, 1978. Robertsonian changes in the chromosomes of Indian Murrah buffalo, *Bubalus bubalis*. *The Nucleus*, 21: 90-97.
- Gustavsson, I., 1979. Distribution and effects of the 1;29 Robertsonian translocation in cattle. *J. Dairy Sci.*, 62: 825-835
- Nurgiartiningsih, VMA. S.Bambang, G, Ciptadi, and Kusmartono. 2012. Early studies of white-albino buffalo in Kingman Island. Madura, East Java. UB-University, Research Report.

The study of reproductive efficiency over the lifetime of Lori-Bakhtiari ewes

M. Vatankhah

P. O. Box 415, Department of Animal Science, Agriculture and Natural Resources Research Center, Shahrekord, IRAN

Abstract

The data set used in this study consisted of 8202 records of reproductive traits in a lifetime of 2478 ewes collected from 1989 to 2012 inclusive from a Lori-Bakhtiari research flock at Shooli station in the southwestern part of Iran (Shahrekord). Reproductive efficiency over all consecutive lambing opportunities were calculated by adding the total reproductive efficiency traits per ewe joined for all of the parturition opportunities. The reproductive efficiency included, number of parity (NP), total number of lambs born (TNLB), total number of lambs weaned (TNLW), total of lambs birth weight (TLBW), total of lambs weaning weight (TLWW), total of lambs weaning weight per kg ewe body weight (TLWW/EBW) and total of lambs weaning weight per kg metabolic ewe body weight (TLWW/MEBW). The overall mean reproductive efficiency traits were 3.31, 3.31, 3.21, 16.51 kg, 87.31 kg, 1.51 and 4.16 for NP, TNLB, TNLW, TLBW, TLWW, TLWW/EBW and TLWW/MEBW, respectively.

Keywords: Reproductive efficiency, Ewe's lifetime, Genetic parameters

Corresponding author: vatankhah_mah@yahoo.com

Introduction

Improved ewe productivity is a major objective in the sheep industry and could be achieved by increasing the number of lambs weaned and weight of lambs weaned per ewe per year (Duguma et al., 2002). Evaluating the cumulative lifetime production of a group of ewes presents a good measure of the entire flock operation and improving the reproductive rate of the ewe flock offers one of the greatest single opportunities for increasing the efficiency of lamb production (Iman and Slyter 1996). Due to the fact that reproductive traits were the most important traits in all sheep production systems (Vatankhah, 2005), increasing the efficiency of sheep production in total lifetime by improving reproductive traits is economically important. The study of reproductive ewe efficiency traits in total lifetime are necessary to improve profitability in rearing sheep under village system.

Material and Methods

The data set used in this study consisted of 8202 records of reproductive traits in a lifetime of 2478 ewes collected from 1989 to 2012 inclusive from a Lori-Bakhtiari research flock at Shooli station in the southwestern part of Iran (Shahrekord). The flock is managed under semi-migratory or village system (Vatankhah, 2013). The traits evaluated in this study were reproductive efficiency over the lifetime of ewes, by adding up the total number of lambs weaned per ewe joined on all of consecutive lambing parturition opportunities (1 to 8). Ewes that failed to a lamb or did not wean a lamb, received zero for that year. The cumulative reproduction traits were evaluated for each

ewe exposed to the ram during her lifetime included, number of parity (NP), (0 to 8), total number of lambs born per ewe (TNLB), (0 to 10), total number of lambs weaned per ewe (TNLW), (0 to 10), total of lamb birth weight (TLBW), total of lamb weaning weight (TLWW), total lamb weaning weight per kg of ewe body weight (TLWW/EBW) and total of lamb weaning weight per kg metabolic ewe body weight ($EBW^{0.75}$) in ewe's lifetime (TLWW/MEBW). The GLM procedure of SAS (2004) was applied to identify important fixed effects affecting on reproductive efficiency traits in a ewe's lifetime using the following model.

$$y_{ijk} = \mu + A_i + B_j + b(EBW_{ijk} - EBW_{000}) + e_{ijk}$$

In this model, y_{ijk} was each of the observations; μ was the overall mean; A_i was the i th year of ewe birth ($i = 1988$ to 2011); B_j was the effect of j th parturition in a ewe's lifetime ($j = 1$ to 8) for all of the traits except number of parities in total ewe's lifetime; b the linear regression coefficient of ewe body weight; EBW_{ijk} was the mean body weight of each ewe (kg); EBW_{000} was the overall mean for ewe body weight and e_{ijk} was the residual effects.

Results and Discussion

The overall mean of NP (No.) and TNLB (head) over the lifetime of the ewes (Table 1) were similar (3.31), due to survival rate below one between birth and weaning, whereas the TNLW decreased by 0.1 and was 3.21. The overall mean for TLWW as net reproductive efficiency was 87.3 kg. The ratio of kg of TLWW per kg ewe body weight was 1.5 and per kg metabolic ewe body weight calculated was 4.2. The variability (standard deviation) for all of the variables assessed was high, but was greatest for especially TLWW. All the ewe efficiency traits assessed were affected ($P < 0.01$) by non-genetic factors such as year of birth, number of parity (except for NP) and body weight as covariate. All the ewe efficiency traits increased with increasing parity number. TNLB did not differ between ewes in their sixth and seventh, and seventh and eighth parities. Regression coefficients indicated that body weight of the ewes significantly ($P < 0.01$) increased all the variables measured with the exception of TLWW/EBW and TLWW/MEBW (Table 1). The greatest coefficient of variation occurred in NP (45.6%) ranging from 28.2% to 33.8%, while the fixed model itself accounted for 30% of the variation in NP and between 74% and 82% of the variation in all other variables. Thus, the reproductive efficiency traits were low due to low lambing and the twinning rates during the lifetime of ewes. Factors, other than genetic variation, accounted significant variation in reproductive efficiency and improvement in nutrition and management (environmental effects) could increase reproductive efficiency significantly. However, high standard deviation indicated that improvement reproductive efficiency traits especially NP could be improve TLWW.

Table 1. Least squares means of different levels of some fixed factors in different ewe's reproductive efficiency traits.

Trait	NP	TNLB	TNLW	TLBW	TLWW	TLWW/EBW	TLWW/MEBW
Overall Mean \pm SD	3.31 \pm 1.79	3.31 \pm 2.17	3.21 \pm 2.20	16.51 \pm 11.04	87.31 \pm 63.93	1.51 \pm 1.06	4.16 \pm 2.95
Year of birth	**	**	**	**	**	**	**
No. of parity	-	**	**	**	**	**	**
1	-	0.99 \pm 0.06 ^g	0.98 \pm 0.05 ^g	5.19 \pm 0.24 ^g	24.37 \pm 1.51 ^g	0.35 \pm 0.03 ^g	1.02 \pm 0.07 ^g
2	-	1.82 \pm 0.06 ^f	1.69 \pm 0.05 ^f	8.86 \pm 0.24 ^f	44.74 \pm 1.51 ^f	0.78 \pm 0.03 ^f	2.13 \pm 0.07 ^f
3	-	3.11 \pm 0.06 ^e	2.98 \pm 0.06 ^e	15.18 \pm 0.24 ^e	80.62 \pm 1.54 ^e	1.43 \pm 0.03 ^e	3.92 \pm 0.07 ^e
4	-	4.11 \pm 0.06 ^d	3.96 \pm 0.06 ^d	20.12 \pm 0.24 ^d	108.40 \pm 1.54 ^d	1.90 \pm 0.03 ^d	5.22 \pm 0.07 ^d
5	-	5.32 \pm 0.06 ^c	5.11 \pm 0.06 ^c	26.02 \pm 0.26 ^c	140.25 \pm 1.63 ^c	2.46 \pm 0.03 ^c	6.75 \pm 0.08 ^c
6	-	6.02 \pm 0.07 ^b	6.08 \pm 0.07 ^b	31.46 \pm 0.30 ^b	171.70 \pm 1.92 ^b	2.96 \pm 0.03 ^b	8.16 \pm 0.09 ^b
7	-	6.28 \pm 0.13 ^b	7.30 \pm 0.13 ^a	37.58 \pm 0.55 ^a	209.86 \pm 3.47 ^a	3.55 \pm 0.06 ^a	9.84 \pm 0.16 ^a
8	-	6.97 \pm 0.32 ^a	7.42 \pm 0.31 ^a	40.07 \pm 1.35 ^a	220.69 \pm 8.50 ^a	3.77 \pm 0.15 ^a	10.42 \pm 0.40 ^a
EBW Covariate	0.121 \pm 0.005 ^{**}	0.017 \pm 0.004 ^{**}	0.019 \pm 0.004 ^{**}	0.164 \pm 0.017 ^{**}	0.733 \pm 0.110 ^{**}	-0.010 \pm 0.002 ^{**}	-0.012 \pm 0.005 [*]
R ²	0.30	0.74	0.77	0.82	0.79	0.78	0.78
%CV	45.56	33.79	33.13	28.18	33.46	33.09	33.06

NP, Number of parity; TNLB, Total number of lambs born in ewe's lifetime; TNLW, Total number of lambs weaned in a ewe's lifetime; TLBW, Total of lamb birth weight in a ewe's lifetime; TLWW, Total of lambs weaning weight in a ewe's lifetime; TLWW/EBW; Total of lambs weaning weight per ewe body weight (EBW) in a ewe's lifetime; TLWW/MEBW; Total of lambs weaning weight per metabolic EBW (EBW^{0.75}) in a ewe's lifetime. *, ** Significant at P<0.05 and P<0.01, respectively. The differences between LSM of traits for different levels of No. of parity indicated with the same letters were not significant at P<0.05.

References

- Duguma, G., Schoeman, S. J., Cloete, S. W. P. and G. F. Jordaan, 2002. Genetic and environmental parameters for ewe productivity in Merinos. *S. Afr. J. Anim. Sci.* 32: 154-159.
- Iman, N. Y. and A. L. Slyter, 1996. Lifetime and wool production of Targhee or Finn-Dorset-Targhee ewes managed as farm or rangeflock. 2. Cumulative lamb and wool production. *J. Anim. Sci.* 74: 1757-65.
- Vatankhah, M., 2005. Defining a proper breeding scheme for Lori-Bakhtiari sheep in village system. Ph.D. Thesis. University of Tehran, 207 pp.
- Vatankhah, M., 2013. Estimation of the genetic parameters for survival rate in Lori-Bakhtiari lambs using linear and Weibull proportional hazard models. *J. Agr. Sci. Tech.* 15: 1133-114

Performance Testing of Kamphaeng Saen Bulls

Raungprim T, S. Majarune, & W. Maitreejet*

*Animal Science Department, Faculty of Agriculture at Kamphaeng Saen
Kasetsart University (KU) Kamphaeng Saen Campus, Nakhon Pathom 73140, Thailand*

Abstract

Kamphaeng Saen (KPS) beef cattle found since 1969 by a group of animal scientists from Kasetsart University (KU) is the first registered in Thailand. With The long-term research genetic improvement and performance testing of KPS bull project, the KPS breed is a composite of 25% Thai native cattle breed, 25% Brahman and 50% Charolais breed. The young bull was selected after weaning at 7-8 months old from beef farms across the country. The KPS bulls were brought to testing station from farm members with specific criteria of the project. From 1999 to 2012, there were sever performance testing lots, 40,50,50,50,50,48 and 50 weaning young bull station-tested. The bulls in each lot were placed into individual pen under the same management and feeding systems for 120 days. These traits were weighted according to relative importance in selection in order to calculate grade point average (GPA) of each bull for kept frozen semen. The KPS bulls had 30.02+4.03 kg for average birth weight, 193.09+45.19 kg for adjusted 205-day weights and 344.31+77.95 kg for adjusted 365-day weight. When the KPS bulls was one year of age, there were measured 136.24+9.67 cm for body length, 125.70+6.06 cm for hip height and 28.00+3.82cm for average testicle circumference. The KPS bulls in feed lot was 1.34+0.20 kg for average daily gain (ADG), 5.57+1.03 for feed conversion ratio (FCR) and 3.06+0.46 for GPA. The KPS breed are indicated in this paper the results of potential genetic growth under local farm condition in Thailand.

Keywords: Kamphaeng Saen beef cattle, performance testing, ADG, FCR

**Corresponding author: swktp@ku.ac.th*

Introduction

Thai indigenous or native breed have relatively small body size, slow grow rate, but high calving percentage under sub-optimal feeding. For this reason, Thai native cattle have not been used for commercial fattening system. Since the demand for quality beef has been increasing in last 3 decades, Brahman crossbreed was imported (Chantalakhana, 1984). Later on the exotic breeds like Charolais were introduced, mostly in the form of frozen semen for crossbreeding. Other new breeds resulted from *Bos taurus* and *Bos indicus* cross such as Santa Gertrudis, Charbray, and Droughtmaster were beef development in some countries (Hardjosubroto 1984 & Tumwasorn 1996). The first KPS breed can reduce imported quality beef. The KPS breed is the long-term effort research results of the Department of Animal Science staffs, Faculty of Agriculture, KU, which was initially through the research project on the improvement of cattle for draught and meat since 1969.

Subsequent to the KPS breed establishment, genetic improvement of various crossbreeding systems was critically evaluated (Chantalakhana et al. 1977a, 1977b; Tumwasorn et al., 1982; Tumwasorn et al., 1993; Bunyavejchewin, 1996), the composite breed of 25% Thai native cattle 25% Brahman and 50% Charolais breeds showed the superior genetic potential under the local Thai farming environment. Thai native is good for fertility and adaptability while Brahman is good for growth and longevity, and Charolais is good for growth,

meat quality. Consequently, the KPS breed (25%Thai Native x 25% Brahman x 50% Charlorias) benefits has large size, rapid.

Materials and Methods

Since 1997 selection of young bulls though performance testing is the main activity at this stage in order to continued genetic improvement and breed stabilization. Operational procedures are fifty crossbred calves at 8-10 months old having the genetic composition of 25% Thai native, 25% Brahman and 50% Charolais are selected annually from the farmers' herds based on weaning weight which has to be above herd average of each farm. These bulls must have a pedigree (sire, dam, paternal and maternal grand dams and sires as well as and birth weight. From 1997 to 2012 under same management and feeding systems annually at beef farm KU, KPS. The KPS bull was tested. The testing KPS bull calves were kept in individual pens and were fed total mixed ration of 17% protein and 77% TDN for 120 days. The KPS bulls were recorded ADG, FCR, testicle circumference, hip height, body length and general appearance. These traits were recorded according to relative importance in selection index in order to calculate GPA of each bull and 205-day adjusted weight. All these traits were analyzed by a method of covariance analysis for unequal subclass numbers using SAS software (SAS, 2003)

Results and Discussion

Performance testing of KPS young bulls of seven lots showed in Table 1. Birth weight and adjusted 205-day weight were significant different ($P<0.05$) average group 30.02 ± 4.03 kg and 193.09 ± 45.19 kg respectively. Feeding trial weight gain, daily gain (ADG) and feed conversion ratio (FCR) of were significant different ($P<0.05$) average lot 161.27 ± 24.27 kg, 1.34 ± 0.20 kg and 5.57 ± 1.03 kg respectively. Yearling Adjusted 356-day weight, body length, hip height, and testicle circumference of all lots were significant different ($P<0.05$) average lot 344.31 ± 77.95 kg, 136.24 ± 9.67 cm, 125.70 ± 6.06 cm and 28.00 ± 3.86 cm, respectively. These traits were weighted according to relative importance for selection in order to calculate grade point average (GPA) of each bull and grade point average lot 1 to 7 had 3.06 ± 0.46

After the first performance testing results each lot selected top 10 bulls had collected semen and kept in frozen in order to use for sire. Top 10 KPS young bulls of seven lots showed in Table 2. From lot 1 to 7, birth weight, adjusted 205-day weight gain, daily gain (ADG), feed conversion ratio (FCR) yearling Adjusted 356-day weight, body length, hip height, testicle circumference and grade point average (GPA) of all lots were significant different ($P<0.05$) average lot 30.43 ± 3.89 kg, 215.23 ± 49.73 kg, 179.85 ± 20.48 kg, 1.50 ± 0.17 , 5.27 ± 0.23 kg, 387.30 ± 83.82 kg, 140.79 ± 8.80 cm, 128.34 ± 4.72 cm, 29.57 ± 3.39 cm and 3.40 ± 0.24 respectively.

All top 10 KPS young bulls of seven lots were best compared to the first performance testing, birth weight and adjusted 205-day weight, weight gain, daily gain (ADG) and feed conversion ratio (FCR), Adjusted 356-day weight, body length, hip height, testicle circumference and grade point average (GPA) all groups. In building up a composite beef breed.

Table 1. Production performance of bulls.

Traits	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Average
Number of bull calves	40	50	50	50	50	48	50	
Associated traits								
Birth weights (kg)	30.34±4.62 ^{ab}	30.32±4.03 ^{ab}	30.01±4.36 ^{ab}	31.51±5.19 ^a	29.11±5.77 ^b	30.18±4.00 ^{ab}	28.72±3.80 ^b	30.02±4.03
Adjusted 205-day weight (kg)	194.38±36.04 ^b	171.79±35.87 ^c	188.50±28.50 ^b	198.34±41.25 ^b	158.98±37.61 ^c	244.96±53.89 ^a	197.01±25.67 ^b	193.09±45.19
Feeding trial weight Gain (kg)	167.88±25.26 ^b	177.31±20.25 ^a	167.98±20.82 ^b	160.84±27.69 ^b	141.12±18.88 ^d	151.52±21.27 ^c	162.75±17.66 ^b	161.27±24.27
Average daily gain (kg)	1.40±0.21 ^b	1.48±0.17 ^a	1.40±0.17 ^b	1.34±0.23 ^b	1.18±0.16 ^d	1.26±0.18 ^c	1.36±0.15 ^b	1.34±0.20
Feed conversion ratio(kg)	5.58±0.91 ^c	5.21±0.65 ^d	6.00±0.75 ^b	5.84±0.64 ^{bc}	5.01±0.54 ^d	4.91±0.89 ^d	6.42±1.50 ^a	5.57±1.03
Yearling								
Adjusted 365-day weight (kg)	382.07±50.79 ^b	374.23±53.08 ^b	334.92±47.64 ^c	327.97±72.06 ^c	260.34±65.77 ^d	412.59±94.60 ^a	328.35±45.70 ^c	344.31±77.95
Body length (cm)	132.86±6.40 ^c	134.03±5.94 ^c	132.24±8.37 ^c	139.67±7.85 ^b	127.36±7.17 ^d	148.44±8.15 ^a	138.94±7.18 ^b	136.24±9.67
Hip height (cm)	124.06±4.36 ^d	125.02±4.58 ^{cd}	126.45±5.58 ^{bc}	126.22±4.66 ^{bcd}	120.28±7.08 ^e	130.25±5.03 ^a	127.46±5.69 ^b	125.70±6.06
Testicle circumference (cm)	30.08±2.85 ^a	28.66±2.90 ^a	26.56±3.22 ^b	29.06±3.54 ^a	23.54±3.53 ^c	29.50±3.28 ^a	29.10±2.98 ^a	28.00±3.82
Grade point average	3.20±0.41 ^{ab}	3.29±0.24 ^a	3.04±0.29 ^c	3.13±0.47 ^{bc}	2.58±0.35 ^e	3.34±0.25 ^a	2.88±0.27 ^d	3.06±0.46

^{abc} Means±SD with different superscript in the same row differed significantly (P<0.05)

Table 2. Top 10 bulls for collected semen.

Traits	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Average
Number of bull calves	10	10	10	10	10	10	10	
Associated traits								
Birth weights (kg)	32.50±4.79 ^a	30.20±3.49 ^{ab}	31.00±3.89 ^{ab}	31.08±3.51 ^{ab}	29.30±3.74 ^{ab}	30.75±3.26 ^{ab}	28.20±4.02 ^b	30.43±3.89
Adjusted 205-day weight (kg)	227.59±32.97 ^{ab}	197.71±41.64 ^b	194.57±35.42 ^b	215.60±35.88 ^b	188.00±40.86 ^b	260.70±84.14 ^a	222.43±26.11 ^{ab}	215.23±49.73
Feeding trial weight Gain (kg)	195.84±11.34 ^{ab}	200.28±16.36 ^a	171.72±12.32 ^{cd}	183.54±14.05 ^{bc}	161.16±10.90 ^d	163.60±23.58 ^d	182.83±16.35 ^{bc}	179.85±20.48
Average daily gain (kg)	1.63±0.09 ^{ab}	1.67±0.14 ^a	1.43±0.10 ^{cd}	1.53±0.12 ^{bc}	1.34±0.09 ^d	1.36±0.20 ^d	1.52±0.14 ^{bc}	1.50±0.17
Feed conversion ratio (kg)	4.86±0.33 ^d	5.02±0.47 ^{cd}	5.71±0.56 ^{ab}	5.55±0.33 ^{bc}	4.91±0.75 ^d	4.61±0.97 ^d	6.22±0.93 ^a	5.27±0.23
Yearling								
Adjusted 365-day weight (kg)	441.02±28.86 ^a	424.99±45.59 ^{ab}	359.40±38.56 ^{bc}	359.62±63.17 ^{bc}	311.86±72.90 ^c	440.17±48.07 ^a	374.03±45.91 ^{abc}	387.30±83.82
Body length (cm)	137.50±2.79 ^{cd}	138.10±8.40 ^{cd}	135.30±6.33 ^d	145.30±7.65 ^{ab}	135.30±3.86 ^d	150.60±9.35 ^a	143.40±9.48 ^{bc}	140.79±8.80
Hip height (cm)	126.51±3.33 ^b	128.00±3.56 ^{ab}	126.80±2.85 ^b	128.50±3.72 ^{ab}	126.00±6.55 ^b	131.30±3.37 ^a	131.30±6.24 ^a	128.34±4.72
Testicle circumference (cm)	30.90±3.11 ^a	30.70±3.40 ^a	27.70±2.67 ^{bc}	31.00±3.71 ^a	26.10±2.77 ^c	29.90±2.42 ^{ab}	30.70±2.79 ^a	29.57±3.39
Grade point average	3.68±0.11 ^a	3.60±0.07 ^{ab}	3.22±0.10 ^c	3.50±0.08 ^b	3.07±0.14 ^d	3.51±0.20 ^b	3.23±0.14 ^c	3.40±0.24

^{abc} Means±SD with different superscript in the same row differed significantly (P<0.05)

Conclusion

The KPS breed between exocotic breeds and Thai native breeds can improve commercial fattening system in Thailand, The KPS beef cattle is appropriated for Thai famer interms of low cost and high profit. The further research is to investigate genetic progress, the conventional molecular

References

- Bunyavejchewin P. 1996. Potential of beef production in tropical Asia. In: *Proceedings of the 8th AAAP Animal Science Congress, Vol. 1*, Japan Society of Zootechnical Science, Tokyo, Japan. pp 404-403.
- Chantalakhana, C., 1984. Beef cattle and buffalo breeding in Thailand. In: J.W. Copland, ed. *Evaluation of Large Ruminants for the Tropics*, ACIAR, Canberra, Australia. 29-36.
- Chantalakhana C., B. Rengsirikul, P. Prucasri and S.Tumwasorn 1977b. *Performance of Thai indigenous cattle and their crossbred from American Brahman and Charolais sires*. Department of Animal Science, Kasetsart University, Bangkok, Thailand.
- Chantalakhana C. K, Markvichitr, P. Prucasri and S.Tumwasorn 1977a. *Preliminary report on the performance of one fourth American Brahman Crossbreds in villages* Department of Animal Science, Kasetsart University, Bangkok, Thailand.
- Hardjosubroto W. 1984. Breed evaluation of large ruminants in Indonesia. In: Copland J.W.(ed.), *Evaluation of Large Ruminants for the Tropics*.ACIAR, Canberra, Australia. pp.74-81.
- SAS. 2003. SAS Intitute. SAS Procedure Gyide for Personal Computer, Version 6. SAS Institute, Cary, NC. USA. 584 pp.
- Tumwasorn S. 1996. *Beef cattle production: Means for skill development of Thairfarmers*. Augsornsiam Press, Bangkok, Thailand. 442 p.
- Tumwasorn S., K.Markvichitr, P. Innuraksa, P. Prucasri and C. Chantalakhana 1993a. Heterosis and additive breed effects on growth traits from crossing among Thai local, Charolais and American Brahman under Thai conditions. *Thai J. Agric. Sci.* 28: 27-41.
- Tumwasorn S., P. Prucasri, K. Markvichitr, B. Rengsirikul, P. Innuraksa and C. Chantalakhana, 1982b. Comparative performances of Thai Indigenous Native, Brahman half-bred and Charolais half-bred cattle at Kamphaengsaen Animal Research Station. In: *Proceedings of Animal Science Research. The 20th Annual Conference*. Kasetsart University, Bangkok, Thailand. pp. 363-376.

Modelling genomic selection strategies to improve genetic gain in swine breeding programs using ZPLAN+

Lopez*, B. M., T. H. Kim, V. S. Viterbo, H. S. Kim, J. H. Hong, Y. H. Kim, K. C. Nam & K. S. Seo

Department of Animal Science and Technology, Suncheon National University, Suncheon, 540-742, South Korea

Abstract

The objective of this study were to evaluate the present conventional selection program and to compare with the new selection strategy that uses genomic enhanced breeding value (GEBV) as selection criteria to improve the genetic gain in swine breeding program. The software ZPLAN+ was used to calculate and compare the generation intervals, genetic gain, total cost, return and profit of each selection strategy. The first strategy reflected the current conventional breeding program which was a progeny test system (CS). The first genomic selection strategy was a selection scheme strictly based on genomic information (GS). The second genomic selection scenario was a mixture of genomic information and progeny tests (MS). The results showed that the mean generation intervals of CS and MS were the same, which was 1.88; while GS was 1.67. The annual monetary genetic gain of GS and MS were 30 percent and 20 percent higher, respectively, when compared to CS. Additionally, the discounted profit per year of GS was 21 percent higher than CS. However, MS was less profitable than CS. In comparison among genomic breeding scenarios, GS was advantageous than MS based on profit. The genomic selection schemes especially GS were clearly superior to conventional scheme in terms of generation interval, annual monetary genetic gain and profit.

Keywords: breeding program, ZPLAN+, genomic selection, genetic gain

Introduction

Animal breeding schemes need well-designed breeding plans to maximize long-term genetic gain from genomic information (Henryon et al., 2014). To optimize the design of breeding programs, a full understanding of selection index theory to predict the outcome of performance recording, genetic evaluation and subsequent selection is required (Toghiani, 2012). With advances in molecular technology or new biological tools, researchers have established ways to incorporate variation at the very basic DNA level into breeding programs. Genomic tools, such as single nucleotide polymorphism (SNP), have led to a new method of selection called “genomic selection” developed by Meuwissen (2007), in which dense SNP genotypes covering the whole genome are used to predict the breeding value. Nowadays, genomic selection is discussed as a potential method to improve genetic gain for all livestock species and it has been successfully implemented for dairy cattle. Such an improvement is tempting in other species. To date, a powerful tool that can evaluate and optimize breeding programs of large complexity is ZPLAN+ (Täubert et al., 2010). It is therefore essential to assess the potential of using genomic information via software like ZPLAN+. The main objective of this study is to design a swine breeding program with high genetic gains and low breeding cost. Also, this paper aims to compare conventional from genomic breeding programs.

Materials and Methods

Modelling software

The computer program ZPLAN+ (Täubert et al., 2010), a user-friendly interfaces software was used to simulate and evaluate the different breeding schemes. This deterministic software allows modeling all related breeding structures, genetic and economic parameters to account complex breeding programs with special emphasis on genomic information. ZPLAN+ was developed based on the gene flow method (Hill, 1974), selection index procedure for predicting reliabilities by Hazel and Lush (1942), and as well as on a complex economic modeling. Breeding schemes were compared in terms of generation interval, monetary genetic gain, breeding costs, returns and discounted profit.

Breeding goal

The breeding goal traits considered in this simulating breeding programs were average daily gain (ADG), back fat thickness (BF), feed conversion rate (FCR) and number of born alive (NBA). Heritability, phenotypic standard deviations and economic weights of the breeding goal traits in this simulating breeding scheme were obtained from National Swine Improvement Federation (NSIF) Guidelines for Uniform Swine Improvement Programs website (NSIF, 1987) that can also be found in Canadian Centre for Swine Improvement (CCSI) website.

Breeding scenarios

Three selection strategies were modeled in ZPLAN+ as shown in Table 1. The first strategy reflected the current conventional breeding program which was a progeny test system (CS). The second strategy was a scheme strictly based on genomic information alone (GS). The third scenario was a mixture of two scenarios (MS) where the selections were based on pedigree, GEBV of the animals, own performance and progeny information.

Table 1. Selection schemes modelled in ZPLAN+.

Scenario	Information Sources
CS	Pedigree, Own performance, Progeny
GS	Pedigree, GEBV
MS	Pedigree, GEBV, Own performance, Progeny

Results and Discussion

Generation intervals

The mean generation intervals in the different selection strategies are shown in Table 2. ZPLAN+ calculates the mean generation interval directly from the gene flow-matrix. The CS and MS schemes have an average of 1.88 generation interval. The GS has shorter generation interval than CS. The generation interval was reduced to 11 percent, resulting in 1.67 year. The reduction is due to the early use of genotype sires without waiting for the progeny information.

Table 2. Generation intervals of different selection schemes.

Breeding Program	CS	GS	MS
Generation interval	1.88	1.67	1.88
relative	100	89	100

Monetary genetic gain, breeding costs, returns and profit

The main target in each breeding program is to maximize the genetic gain per generation and per year. The overall annual monetary genetic gain (AMGG) of the breeding goal per year is shown in Table 3. Applying the conventional selection strategy may create an annual monetary genetic gain of 8.90\$. In comparison with genomic selection strategies, GS and MS can increase the annual monetary genetic gain to 30 percent and 20 percent, respectively. This confirms the results of former studies that estimate higher genetic gain when incorporating genomic information (Schaeffer, 2006; Täubert et al., 2011).

The costs, returns and profit with relative percentage in each selection schemes are shown also in Table 3. The variable costs in every selection schemes in this study are not accounted. Thus, profits are higher than expected. The discounted costs of genomic selection strategies are higher than CS as expected. The costs of GS and MS are 44 percent and 70 percent higher than CS, respectively, assuming a genotyping cost of 120\$. Conducting CS may result to 67.58\$ as the discounted return per animal. Such return increases to 31 percent (GS) or 19 percent (MS) when adopting the genomic selection schemes, assuming 1000 genotyped male candidates and a reference population size of 1000. Furthermore, the relative profit of GS is 21 percent higher than CS. However, the profit in MS is 19 percent lower than CS.

Table 3. Annual monetary genetic gain, costs, returns and profit with relative percentage of different selection schemes.

Parameter	Unit	CS	GS	MS
Overall AMGG	\$	8.90	11.56 (130)	10.65 (120)
Discounted return	\$	67.58	88.36 (131)	80.15 (119)
Discounted costs	\$	28.32	40.83 (144)	48.27 (170)
Profit	\$	39.27	47.53 (121)	31.88 (181)

AMGG = Annual monetary genetic gain.

References

- Hazel, L. N., and J. L. Lush. 1942. The Efficiency of Three Methods of Selection. *J. Hered.* 33: 393-399.
- Henryon, M., P. Berg, and A. C. Sørensen. 2014. Animal-breeding schemes using genomic information need breeding plans designed to maximise long-term genetic gains. *Livest. Sci.* 166: 38-47.
- Hill, W. G. 1974. Prediction and evaluation of response to selection with overlapping generations. *Anim. Sci.* 18: 117-139.
- Meuwissen, T. 2007. Genomic selection : marker assisted selection on a genome wide scale. *Journal of Animal Breeding and Genetics* 124: 321-322.
- NSIF. 1987. Guidelines for Uniform Swine Improvement Programs.
- Schaeffer, L. R. 2006. Strategy for applying genome-wide selection in dairy cattle. *Journal of Animal Breeding and Genetics* 123: 218-223.
- Täubert, H., F. Reinhardt, and H. Simianer. 2010. ZPLAN+ A new software to evaluate and optimize animal breeding programs. In: Proceedings of the 9th World Congress on Genetics Applied to Livestock Production, Leipzig, Germany.
- Täubert, H., S. Rensing, and F. Reinhardt. 2011. Comparing Conventional and Genomic Breeding Programs with ZPLAN+. In: *Interbull Bulletin*. p 162-168.
- Toghiani, S. 2012. Quantitative Genetic Application in the Selection Process for Livestock Production.

Effects of different mating methods on hatchability and embryonic mortality of indigenous chicken eggs

Lamidi¹, A.W., S.S. Abiola², M.O. Ozoje³ & O.A. Adeyemi²

¹Department of Agricultural Education, Federal College of Education (Technical), Akoka-Lagos, Nigeria²Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, Nigeria.³Department of Animal Breeding and Genetics, Federal University of Agriculture, Abeokuta, Nigeria

Abstract

The study was conducted to determine the effects of mating methods on hatchability and embryonic mortality of indigenous chicken eggs. Ninety, 17 week-old normal feathered indigenous chicken breeders comprising 72 hens and 18 cocks were allotted to three mating methods: pen mating (PM), alternate males (AM) and stud mating (SM). Each group, comprising 24 hens and 6 cocks, was replicated thrice with 8 hens and 2 cocks per replicate. A total of 387 hatchable eggs were obtained from the hens for determination of hatchability and embryonic mortality. Data obtained were subjected to analysis of variance in a completely randomized design. Mating methods significantly ($P < 0.05$) influenced hatchability and embryonic mortality of eggs as hens in SM produced more chicks (76.48%) and less embryonic deaths (23.52%) than PM (66.75% and 33.25%). There was a strong positive ($P < 0.01$) correlation between egg weight and chick hatching weight. It was concluded that SM and AM resulted in better hatchability and lower embryonic mortality of normal feathered indigenous eggs and therefore could be considered as better alternatives to PM in poultry breeding programmes.

Keywords: mating methods, hatchability, embryonic mortality, indigenous chickens

Corresponding author: lammiwise2000@yahoo.com

Introduction

Hatching characteristics are major parameters of reproductive success in poultry. The process of producing fertile and hatchable eggs in chicken is preceded by successful mating activity of the breeders. Mating in poultry is of several kinds and it includes flock mating, pen mating, stud mating, alternating males and artificial insemination.

Avian embryos develop within the confines of an egg shell independent of maternal physiological functions (Tazawa, 2004). Christensen & Bagley (1989) described embryonic mortality as a non-random event with the chance of an embryonic death occurring not being equal on all days of incubation. Artificial insemination, as a mating method, was widely reported in literature for commercial strains of chicken. However, there is dearth of information on the effects of natural methods of mating on fertility and hatchability of chicken eggs. Therefore, the present work is aimed at determining the effects of different mating methods on fertility and hatchability of indigenous chicken eggs.

Materials and Methods

Ninety, 17 week-old normal feathered indigenous chicken breeders (fifth generation progenies) were sourced from Teaching and Research Farms, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. The birds, comprising 72 hens and 18 cocks were allotted to three mating methods: pen mating (PM), alternate males (AM) and stud mating (SM). Each group, comprising 24 hens and 6 cocks, was replicated thrice with 8 hens and 2 cocks per replicate. The breeders were intensively managed in a deep litter pen. The site falls within the rain forest vegetation zone of South-Western Nigeria on Latitude 7° 13'; 49. 46" N, Longitude 3° 26 ' 11.98" E and altitude of 76 inches above sea level. The climate is humid with a mean annual rainfall of 1037 mm. The annual mean temperature and humidity are 34.7° C and 82% respectively (Google Earth, 2010). The birds were allowed unrestricted access to breeder ration and clean water.

A total of 387 hatchable eggs were obtained from the hens over a 7-day period for determination of hatchability and embryonic mortality. The eggs were pre-numbered and weighed before being transported to the hatchery in egg trays. In the hatchery, the eggs were stored at 17°C for 7 days while they were fumigated before being set in the incubator on treatment and replicate basis. Temperature in the incubator was maintained at 37°C for 18 days. Fertility was determined by candling on the 18th day of incubation while hatchability was based on the number of fertile eggs. The chicks were weighed individually on the day of hatching. Embryonic mortality was determined when the unhatched incubated eggs were opened up. Embryos that died in the early stage of incubation were classified as dead-in-germ (DIG) while those that died at a later stage as dead in shell (DIS).

Data obtained were subjected to analysis of variance (ANOVA) in a Completely Randomized Design. Pearson correlation estimates for hatchability traits was also performed. Significant means were separated using Duncan Multiple Range Test of SAS (1999).

Results and Discussion

Table 1 revealed that hatchability, embryonic mortality, percentage male and female as well as ratio of male to female, were all significantly ($P < 0.05$) influenced by mating methods. Hatchability of fertile eggs was higher in SM (76.48%) and AM (73.08%) than PM (66.75%). Thicker egg shell which hens produced at different stages of lay could partly be responsible for the poor hatchability associated with their eggs (Lamidi, 2014). Therefore, eggs obtained from the SM group could be considered as the best in terms of economic returns since the number of chicks hatched matters most in poultry breeding enterprise. This occurrence could be premised on (aside from sound management, genetic background and good quality semen) lower frequency of mating which characterized the SM practised in this study. However, the findings of this study contrasted to that of Jones & Mench (1991) who demonstrated that higher frequency of mating was reflected in a higher proportion of offspring.

More embryonic deaths ($P < 0.05$) occurred in eggs obtained from PM (33.25%) and AM (26.92%) than those collected from SM (23.52%). Stale sperm in the oviduct has been associated with poor chick quality and early embryonic mortality (Bakst & Howarth, 1997). With respect to this study, more embryonic deaths occurred towards the end of incubation. Such late embryonic mortality is not uncommon and may be due to non-genetic factors (Munir & Mohammad, 2010). The number of cockerel chicks hatched from eggs produced by hens in AM and PM was significantly ($P < 0.05$) higher than those derived from hens in SM. The values recorded for percentage female (pullet chicks) were in sharp contrast to those obtained for percentage male. Eggs from SM and PM yielded higher ($P < 0.05$) pullet chicks than those from AM. This is not unexpected as previous researchers (Oyebimpe & Abiola, 2001) had reported a higher male: female chicks at hatching.

Table 2 showed that initial egg weight had high significant ($P<0.01$) positive correlation with final egg weight and chick hatching weight. The same trend was observed between final egg weight and chick hatching weight. The present finding is in agreement with that of Malago & Baitilwake (2009) who reported a positive strong correlation between egg weight and chick weight in chickens.

Table 1. Effect of different mating methods on hatchability and embryonic mortality of indigenous chicken eggs.

Parameters	Mating methods		
	Pen mating	Alternate males	Stud mating
No of eggs set	129.00±0.00	129.00±0.00	129.00±0.00
AEW (g)	49.89±1.38	52.24±2.24	52.74±0.70
Fertility (%)	89.64±3.60	90.19±1.67	85.94±5.55
Hatchability (%)	66.75±1.28 ^b	73.08±3.20 ^{ab}	76.48±2.03 ^a
Hatching yield (%)	59.69±3.49	65.86±2.57	65.50±2.56
EM (%)	33.25±1.28 ^a	26.92±3.20 ^{ab}	23.52±2.03 ^b
DIG (%)	14.53±3.94	25.33±5.27	25.83±5.83
DIS (%)	85.47±3.94	74.67±5.27	74.17±5.83
Chick weight (g)	34.66±0.66	35.43±1.44	35.96±0.40
Egg : chick	1.40±0.02	1.47±0.02	1.47±0.00
Male chick (%)	55.13±1.28 ^{ab}	60.25±2.56 ^a	51.28±1.28 ^b
Female chick (%)	44.87±1.28 ^{ab}	39.75±2.56 ^b	48.72±1.28 ^a
Male : female	1.23±0.06 ^{ab}	1.54±0.18 ^a	1.06±0.06 ^b

Means on the same row with different superscripts differ significantly ($P<0.05$).

AEW = Average egg weight, EM = Embryonic mortality, DIG = Dead-in-germ, DIS = Dead-in-shell

Table 2. Correlation coefficients on fertility and hatchability parameters.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	1.00													
2	0.98**	1.00												
3	0.40	1.21	1.00											
4	0.23	0.42	0.80	1.00										
5	0.12	0.04	0.64	0.57	1.00									
6	0.47	0.54	-0.19	-0.50	-0.40	1.00								
7	0.58	0.53	0.42	0.07	0.53	-0.57	1.00							
8	-0.47	-0.54	0.19	0.50	0.40	-0.00**	-0.57	1.00						
9	0.39	0.41	0.06	-0.17	-0.28	0.63	0.37	-0.63	1.00					
10	0.36	0.30	0.41	0.20	0.02	0.37	0.38	-0.38	0.35	1.00				
11	-0.46	-0.44	-0.26	0.02	0.19	-0.64	-0.45	0.64	-0.88**	-0.75*	1.00			
12	0.94**	0.92**	0.35	-0.23	0.18	0.54	0.66	-0.54	0.41	0.53	-0.55	1.00		
13	0.36	0.25	0.64	0.45	0.28	-0.78	0.20	0.08	0.18	-0.27	-0.27	0.33	1.00	
14	-0.36	-0.25	-0.04	-0.45	-0.28	-0.08	-0.20	-0.08	-0.18	-0.27	0.27	-0.33	-1.00**	1.00

** $P<0.01$

* $P<0.05$

1 = Initial egg weight 2 = Final egg weight 3 = Egg weight loss (g) 4 = % Weight loss 5 = % Fertility
 6 = % Hatchability 7 = % Hatching yield 8 = Embryonic mortality 9 = Dead-in-germ 10 = Dead-in-shell
 11 = Weak-in-shell 12 = Chick hatching weight 13 = % Male 14 = % Female

Conclusion

The findings of this study reveal that stud mating could be employed as a more productive mating method in terms of hatchability and percentage pullet chicks than the conventional method.

References

- Bakst, M. R. and B. Howarth, 1997. Hydrolysis of the hen's perivitelline layer by cock sperm. *Bio.of Rep.*17: 370 – 379
- Christensen, V. L., and L. G. Bagley. 1989. Embryology of the turkey. In: Recent advances in turkey Science (Nixey, C and Grey, I C. eds) Butterworth and Co. London. 69 – 90
- Google Earth. 2010. [Http//www.googleearth](http://www.googleearth).
- Jones, M.E.J. and Mench, J.A. 1991. Behavioural correlates of male mating success in a multisire flock as determined by DNA fingerprinting. *Poult. Sci.*70: 1493-1498.
- Lamidi, A. W., 2014, Effects of different mating methods on fertility and hatchability of indigenous chicken eggs and post-hatch performance of chicks. Ph.D Thesis. Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.
- Malago, J.J. and M.A. Baitilwake, 2009. Egg traits, fertility, hatchability and chick survivability of Rhode Island Red, local and crossbred chickens. *Tanzania Vet.J.* 26(1) 24-36
- Munir, M.A. and S.A. Mohammad, 2010. Effects of ambient temperature flock age and breeding stock on egg production and hatchability of broiler hatching eggs. *J.Biolo.Sci.*2(3): 55-66.
- Oyebimpe, K and S.S. Abiola, 2001. Effect of egg incubation position in electric table incubator on hatchability and post-hatching performance of chicks. *Bul.Ani.Health& Prod. Africa.* 49:122-128.
- SAS. 1999. *SAS/STAT User's Guide*. Version 8 for Windows. AS Institute, Cary, North Carolina, USA.
- Tazawa, H., 2004. Embryonic carcass variables during incubation. *World's Poul. Sci.* 60: 478 – 488

Proximity of genetic cross Boer goat with Local goat to parent based on gene DNA *Capra hircus* growth hormone (*ChGH*)

Moch Nasich

Brawijaya University, Veteran, Malang 65145, Indonesia

Abstract

The aims of this study were to identify and analyze the closeness of the relationship between parent (Boer and Local) with their kid by the DNA of the gene *Capra hircus* Growth Hormone (*ChGH*). The results of this study can be used as the initial information in an effort to develop a goat farm with a certain growth rate for the purpose of meat production. The materials used in this study were respectively 5 head blood samples from Boer, local and cross-bred kid result. Blood samples of DNA isolated by salting out method. Total DNA was amplified with primers that in the design of the growth hormone gene into exon 2 that F-primer →5'-AGG TAT CTG CAC CCA GAC ATT TGG-3' and R-primer →5'-CCT GAC CAC ATC CTT ACT TGG ATA -3'. PCR results in pieces with RFLP method using restriction enzyme *HaeIII*. Amplification and restriction results in electrophoresis using 1.5% agarose gel and documented with a Polaroid camera. Data were analyzed descriptively and Multi-Variate statistical Packed (MVSP) 3.1. The results showed that the primers used to amplify the gene can *ChGH* specifically, to produce a 426 bp DNA fragment size. GH gene amplification products can be cut with restriction enzymes and DNA that produces 5 fragment in sizes 400, 300, 200, 100 and 50 bp. Based on gene DNA *Capra hircus* Growth hormone, the percentage Boer buck genetic closeness with kid was 51.42%, while the local doe parent with kid was 48.57%. These results indicate that the Boer goat stud genetic contribution was slightly higher (2.85%) of the doe Local goat.

Keywords: GH gene, PCR-RFLP, polymorphism

Corresponding author: nasich@ub.ac.id or m.nasich@yahoo.com

Introduction

In Indonesia, goats give a very small contribution to the national meat needs of around 5% (Directorate General of Livestock, 2005). However, when viewed from the potential need for meat, livestock each year about 5.6 million head is slaughtered to the needs of "Idul Adha" celebration, not including the need for aqiqoh, restaurant until sate stalls, growth of the goats population is not comparable with the growing demand, so it is very possible if one day there will be a scarcity of mutton in the country, so that in the future Indonesia must be imported from Australia or New Zealand.

In 1996 Indonesia has import a new variant of Boer goats. Boer goat is known as a breed that has the ability to fast grow or meat production are high. Goat is intended to be able to improve the production of meat goats Local in some areas. Up to this time has a lot of goats from crosses between buck of Boer goats with local goat in the community.

Crosses according Hardjosubroto (1994) is a mating of animal from different breed with the main purpose of combining two or more different properties into one of the breed crosses. By considering the advantages and disadvantages of each properties of the two breed, the results of a cross can have advantages (hybrid vigor) is higher or lower than the average ability of the parents.

Growth is one important indicator in determining the level of meat production in animal. One of the factors that play a role in the growth of an individual animal is Growth Hormone (GH). In mammals, GH biological effect on the growth of the individual after birth, growth rate and milk production (Ge, et al., 2002).

On the basis of this background, it is on this occasion was observed for genotype closeness between Boer goat, local and cross-bred based gene results *Capra hircus* Growth Hormone (CHGH).

Materials and Methods

The material was Boer goat blood from buck, the doe (PE) and the kid, each breed 5 sample of blood. This research method is a laboratory method to work the following way:

1. Sampling of blood; 2. Isolation of DNA; 3. Measurement of DNA Qualitative by an electrophoresis with 1% of agarose gel; 4. Amplification of DNA by PCR; 5. Restriction DNA by RFLP method; 6. Data Analysis

The data were analyzed by Un weight pair group method with arithmetic averages (UPGMA), clustering analysis using Multivariate Statistic Program (MVSP) version 3.1 (Kovach Computing Service) by determining the similarity value by using simple matching coefficient.

Results and Discussion

Amplification results were confirmed using 1.5% agarose gel electrophoresis. From the agarose gel electrophoresis, it appears that the DNA bands PCR results showed at a position above 400 bp. Position of bp can be known of the presence of the marker in electrophoresis on the left side. The bp size after adjusting for gene sequencing *Capra hircus* Growth hormone (ChGH), it is known that the size of 426 bp exon 2. From these circumstances, it can be said that by using the PCR primers were designed as expected.

From the agarose gel electrophoresis known that ChGH gene primers can amplify the specific gene ChGH Boer goats, local and cross-bred to produce the results of the bands/DNA bands corresponding in size to it exon. In *Capra hircus* and other ruminants, based on the gene sequences obtained from research ChGH (Khan et al. (2006).

Specificity of the primer according to Hyndman and Mitsunashi (1996) viewed from the ability to perform hybridization on the target fragments, so that the results of the specific primer amplification product fragment is the target that forms a ribbon when electrophoresis. Given all living beings have the fat component in the cell, then the GH gene can be found in virtually all living creatures, including goats (Lardizibal et al. 2001).

Similarity of Boer goats, Local and offspring

From the results of gene amplification *Capra hircus* Growth hormone (ChGH) using restriction enzymes HaeIII, then made relationships are based on UPGMA clustering analysis (Unweight pair group method with arithmetic averages) by using the program MVSP ver 3.1 (Kovach Computing service) to determine the similarity value through using simple matching coefficient. From the similarity value, then the genetic closeness between Boer goats (buck) and Local goats (doe) to offspring can be seen in Table 1 below.

Table 1. Values of genetic closeness between buck, doe and kid based gene ChGH.

Parents	Kid	Genetic Closeness with	
		Buck	Doe
P4	19.1	0.4	
19			0.8
P4	20.1	0.4	
20			0
P3	21.1	0.6	
21			0.8
P3	40.1	0.8	
40			0.6
P2	22.1	0.6	
22			0.6
P2	41.1	0.8	
41			0.6
Total		3.6	3.4
Percentage		51.42	48.57

From Table 1, it can be seen that the proximity of kid from crosses between Boer goats with local goats are nearer to males (Boer goat), although the difference is only 0.03, but this differences are in this expression could be greater, given the genetic influence on the growth or weight gain descendants of Boer goats ranges between 15-25% (Nasich, 2009).

From these results, it can be concluded that: 1. The design of primer pairs of growth hormone gene sequencing results of Beetal goat and other ruminants in exon 2 can be managed in accordance with the size of the exons. 2. There is a polymorphism in the gene amplification product ChGH on Boer goats, both local and from crosses. 3. Genetic closeness between Boer goat with local goats is nearer to the Boer goat.

References

- Ge, W., M.E. Davis, H.C. Hines, K.M. Irvin and R.C.M. Simmen. 2003. Association of Single Nucleotide Polymorphism in the Growth Hormone and Growth Hormone Receptor Genes with Blood serum Insulin-like growth Factor I Concentration and Growth Traits in Angus Cattle. *J. Anim. Sci.* 81 : 641-648.
- Hyndman, D.L. dan M. Mitsuhashi. 1996. PCR Primer Design. Bartlett, J.M.S. dan Stirling, D, Ed. PCR Protocols. Humana Press. pp 81-88.
- Khan, M.A., Sadaf, F. and Akhtar, M.W. 2006. Sequence analysis of the growth hormone gene of beetal goat and other ruminants. University of the Punjab, New Campus, Lahore, Punjab 54590, Pakistan.
- Lardizibal, K.D., J.T. Mai, N.W. Wagner, A Wyrick, T. Voelker dan D.J. Hawkins. 2001. Purification, Cloning and Expression in Insect Cells of Two Polypeptides from *Mortierella ramanniana* with Diacylglycerol Acyltransferase Activity. *Journal Biol Chem.* 276: 38862-38869.
- Nasich, M. 2009. Performan Produksi Kambing Hasil Persilangan antara Pejantan Boer dengan Induk Kambing Lokal. Kumpulan Abstrak Seminar Nasional Stem Cell dan Hasil-hasil Penelitian Ilmu Hayati. Lab Sentral U B.

Effect of numbers of day of progesterone intra–vaginal sponge insertion: estrus rate pregnancy rate and litter size in Thai-native cross breed goats

Juggrid Jugsumrit,¹ Chalermpon Yuangklang,² Jakrit Yaeram,² Kraisit Vasupen²
& Benya Saenmahayak²

¹Nakhonratchasima a.i research center, ²Rajamagala University of Technology Isan

Abstract

The aim of this study was to determine the effect of estrus synchronization protocols on estrus, pregnancy rate and litter size in Thai-native cross breed goats. Sixty nanny goats were randomly assigned into two experiments to select the efficient method applied for goat estrous synchronization. In experiment I, a total of 40 does were divided into two treatment with a completely random design (CRD) and does of treatment I were synchronized by intravaginal sponge impregnated for 5 days combine medroxyprogesterone acetate (sponge) was inserted on day 0, PGF_{2α} 1.25 μg; PMSG 150 IU; (I/M) and sponge removal on day 5 and treatment II was treated the same as the treatments I, except sponge was inserted for 7 days. The trial II is same as the both group except sponge was inserted for 16 days. Estrus signs and standing heat were observed in all does of both groups by deviated bucks for 4 days after sponge removal. All does were inseminated with frozen semen. TAI was two times performed 48 and 72 hr after sponge removal. Pregnancy was checked by ultrasonographic scanning at 60 days after TAI. The results of this study showed the treatment I had higher conception rate than the treatment II and the trial II (60.40 and 25%) (P < 0.05). All does in both groups expressed estrus (60/60). There was no significant difference of litter size in both groups (1.6, 1.5 and 1.4), respectively. This study was revealed that 5 days sponge insertion estrus synchronization improves conception rate and litter size in goats and is beneficial for reproductive herd management in Thai-native crossbreed goats.

Keyword: synchronization of estrus, estrus rate, pregnancy rate, goats

Introduction

Thailand is land of agriculture and livestock. The products from agriculture is very important to the Thai economy. Goats are small ruminants, which have a high potential to utilize economic and new trend raised in Thailand for meat and milk. The demand for livestock products has been increasing yearly, partly as a consequence of a continuous increase in population. The production has become important involving the use of high yielding breeds subjected to selection programs and artificial insemination (AI) as an essential production strategy.

The methods to goat synchronization of estrus are 5~16 days of treatment with fluorogestone acetate (FGA) or medroxyprogesterone acetate (MAP) impregnated vaginal sponges and intramuscular injection of pregnant mare serum gonadotrophin (PMSG) at the time of sponge removal (Motlomelo et al. 2002; Amarantidis et al. 2004; Baldassarre and Karatzas 2004; Fonseca et al. 2005; Saribay et al., 2008 and Karaca et al., 2011) treatment with FGA or MAP impregnated intravaginal sponges and intramuscular injection of PMSG and PGF_{2α} or its analogs 48 h before sponge withdrawal (Amarantidis et al. 2004; Baldassarre and Karatzas 2004; Khanum et al. 2006; Dogan et al. 2008). The efficiency and ovarian response of goats to estrous synchronization varies with the type of intravaginal sponge, kind of progestagen (Romano 1996), breed, nutritional status (Mani et al. 1992), stress, environment, male effect (Lopez-Sebastian et al. 2007), and procedures of administration of hormone (Chao et al. 2008; Dogan et al. 2008). At present, progesterone

intravaginal sponges gave a response of only 35~50% in goats in the tropic area (Rosnina et al. 1992). Therefore, the possibility to apply estrous synchronization in changing situation of goat production induce of estrus will increase productivity of small ruminants and the income of small holder farmers.

This trial was performed to test the efficiency of different of number days progesterone intra-vaginal sponge insertion combine with PMSG and PGF_{2α} in the synchronization of estrus farm animal breeding programs to the percent of estrus conception rate and little size in Thai-native cross breed goats.

Material and Method

This study was conducted in the Nakhonratchasima province of Thailand which has the geographic coordinated of 14° 58' 28' N latitude and 102° 5' 53' E longitude with an average annual temperature of 27 C°. Totally 60 nanny Thai-native cross breed goats, 2-5 aged with body weight ranges of 25-35 kg and BCS 2.5-4. All does maintained as one flock under field conditions. The goats were kept indoors at night and had access to grazing outdoors for most of the day. Indoors, the does were fed concentrate and rice straw supplemented which assigned into two experiment at experiment I, a total of 40 does were divided into II treatment with a completely random design (CRD), and the does of treatment I were synchronized by intravaginal sponges impregnated for 5 days and does of treatment II impregnated for 7 days combine medroxyprogesterone acetate (sponge) was inserted on day 0, PGF_{2α} 125 µg; PMSG 150 IU; (I/M) and sponge removal on day 5 and treatments II protocol was treated the same as the treatments I except sponge was inserted for 7 days and The trial II is same as the both group except sponge was inserted for 16 days. Estrus signs and standing heat were observed in all does of both groups by deviated bucks for 4 days after sponge removal. All does was inseminated with frozen semen. TAI was two time performed 48, 72 hr after sponge removal and pregnancy was checked by ultrasonographic scanning 60 days after TAI.

Results

The time from sponge withdrawal to induced estrous rate following the short and long sponge treatments for the Thai-native cross breeds goats are shown in Table 1 and a summary of the distribution of animals showing estrus is set out in the time from sponge withdrawal to the onset of estrus was 100% (60/60). Sponge treated groups. The mean pregnancy rate following A.I for all animals in this study was 60, 40 and 25%, and little size was 1.6, 1.5 and 1.4, respectively.

Table 1. Estrus response, conception rate, pregnancy rate and litter size.

day	n	Estrus response (%)	Conception rate	Pregnancy rate	Litter size
5	20	100	60	60	1.6
7	20	100	40	40	1.5
16	20	100	26	26	1.4

Discussion

This study showed that of the progestagen (P4) treatment. Total estrus response 100% after sponge withdrawal. The pregnancy rate among groups was observed in the flock. The lower pregnancy rate observed after the long-term treatment with P4 is in agreement with a previous reported (Saribay et al., 2011). In two trials, a 40, 60 and 26% decrease in pregnancy rate was observed in short time compared with long time. Recently, it was found in cows that when follicle dominance was prolonged more than 9 d, a reduction in follicle health occurred (Minh et al., 1999). Although the ovulatory capability was maintained and the oocytes were competent to be fertilized, the ability to reach the 16-cell stage was compromised and early embryonic death occurred.

In the ST group, where P4 levels were higher due to the endogenous progesterone secretion of exogenous P4 absorption, a normal follicular turnover was promoted and a newly formed follicle ovulated. This could explain the higher pregnancy rate observed in this group. It is interesting to note that in the short-term treated goats, the pregnancy rate was better than usually achieved after traditional synchronization protocols (Motlomelo et al., 2002). In a breeding program for goats, many of the advantages of using artificial insemination depend on the control of estrous behavior. Although long-term P4 treatment offers an efficient technique for synchronizing estrus in does, the low fertility attained is a major limitation. However, under conditions where natural mating is used frequently, short-term treatment could be useful to assure good reproductive performance in the flock. Despite the lack of high estrus synchrony, a concentrated period within one week could improve goat management under field conditions. Alternatively, a combined 5 d MAP treatment associated with prostaglandin injection at sponge withdrawal could be applied to induce CL regression during the breeding season.

Conclusion

This study reveals that 5 days sponge insertion estrus synchronization improves conception rate and litter size in goats and is beneficial for reproductive herd management in Thai-native crossbred goats.

Reference

- Amarantidis, I., Karagiannidis A., Saratsis P., and Brikas P., 2004. Efficiency of methods used for estrous synchronization in indigenous Greek goats. *Small Rumin. Res.* 52(3):247–252.
- Baldassarre, H., and Karatzas C.N., 2004. Advanced assisted reproduction technologies (ART) in goats. *Anim. Reprod. Sci.* 82–83:255–266.
- Chao, L.M., Takayama K., Nakanishi Y., Hamana K., Takagi M., Kubota C., and Kojima T., 2008. Luteal lifespan and fertility after estrus synchronization in goats. *J. Vet. Sci.* 9(1):95–101.
- Dogan, I., Konlyali A., Gunay U., and Yurdabak S., 2008a. Comparison of the effect of cronolonesponges and PMSG or cloprostenol on estrous induction in Turkish Saanen goats. *Polish J. Vet. Sci.* 11(1):29–34.
- Fonseca, J.F., Bruschi J.H., Santos I.C.C., Viana J.H.M., and Magalhaes A.C.M., 2005. Induction of estrus in non-lactating dairy goats with different estrous synchrony protocols. *Anim. Reprod. Sci.* 85:117–124.
- Khanum, S.A., Hussain M., and Kausar R., 2006. Manipulation of estrous cycle in dwarf goat (*Capra hircus*) using gestrumate under different management conditions. *Anim. Reprod. Sci.* 92:97–106.
- Lopez-Sebastian, A., Gonzalez-Bulnes A., Carrizosa J.A., Urrutia B., Diaz-Delfa C., Santiago-Moreno J., and Gomez-Brunet A., 2007. New estrus synchronization and artificial insemination protocol for goats based on male exposure, progesterone and cloprostenol during the non-breeding season. *Theriogenology* 68:1081–1087.
- Mihm M, Curran N, Hyttel P, Knight PG, Boland MP, Roche JF. 1999. Effect of dominant follicle persistence on follicular fluid oestradiol and inhibin and on oocyte maturation in heifers. *Reprod Fertil* 116:293-304.
- Motlomelo, K.C., Greyling J.P.C., and Schwalbach L.M.J., 2002. Synchronisation of oestrus in goats: the use of different progestagen treatments. *Small Rumin. Res.* 45(1): 45–49.
- Romano, J.E., 1996. Comparison of fluorogestone and medroxyprogesterone intravaginal pessaries for estrus synchronization in dairy goats. *Small Rumin. Res.* 22:219–223.
- Rosnina, V., Jainudeen M.R., and Nihayah M., 1992. Superovulation and egg recovery in goats in the tropics. *The veterinary record.* 130(5): 97–99.
- Saribay, M.K., Karaca, F., Dogruer G., Ergun, y., Yavas, I., and Ates, C.T., 2011. Oestrus synchronization by short and long term intravaginal sponge treatment in lactating goats during the breeding season: The effects of GnRH administration immediately after mating on fertility. *J. Anim. Vet. Adv.* 10 (23):3134-3134

Genetic parameter estimation for prolificacy trait of Indonesian local Ettawah Goat

Mudawamah*¹, G. Ciptadi², V.M. A. Nurgiartiningsih², C. D.K. Bottema³

¹Islamic University of Malang, MT Haryono 193, Malang, 65155, Indonesia, ²Brawijaya University of Malang, Malang, 65145, Indonesia, ³University of Adelaide, Roseworthy, SA 5371, Australia.

Abstract

The reproductive characters especially prolificacy trait variation in Indonesian Local Ettawah Goat (ILEG) was considered as an excellent aspect to increase ILEG goat production even though this also depends on the management of the production system and prevailing environmental and economic conditions. The objective of this research was to estimate genetic parameter of prolificacy trait in village breeding center of ILEG goat in East Java. The data included the performance of 520 Does in 14 breeding centers of ILEG goat in East Java with 1347 prolific records. Genetic parameter estimation was performed by calculating variance component, repeatability and heritability estimation using paternal half sib correlation. The data were analyzed using ANOVA with SPSS 15 version. The least square means of prolificacy trait was 1.72 ± 0.53 . The heritability and variance component for prolificacy trait was 0.2 and 0.18. The repeatability and variance component for prolificacy trait was 0.89 and 2.18. It was concluded that the prolificacy trait of ILEG goat in East Java was medium categorized. Heritability and repeatability of prolificacy trait was considered medium and high.

Keywords: prolificacy, goat, genetic parameter, heritability, repeatability

Introduction

ILEG is one of the potential local goat in Indonesia that maintained by the majority of the Indonesian small holder farms. Most goats are kept as savings and the family business as a model farm in Africa (Otuma and Onu, 2013), as well as in Indonesia. ILEG business is a priority in the Indonesian government program that highlights the welfare independence of food sovereignty based on agribusiness populist.

One important factor for the success of agribusiness goats are the properties of prolificacy. To implement the breeding program at the ILEG based on the nature of prolificacy. It is necessary to be considered genetic parameters such as heritability (h^2) and repeatability (r). Heritability is a very important genetic parameter to measure how much additive genetic variation of a trait inherited from parent to kids. While repeatability is to measure how much repetition on each character, include the litter size of does.

The value of heritability and repeatability was varied depending on the breeds. Some studied estimate the heritability and repeatability prolific trait of the sheep was 0.025-0.17 and 0.029 to 0.28 at the low category (Dariusz et al., 2011; Janssens et al., 2004; Vatankhah and Talebi, 2008). While the research results of heritability estimation on Ripollesa goat was 0.13 (Cassellas et al., 2007), Alpine goats was 0.039 (Kasab et al., 2012), Black Bengal Goat was 0.08 (Mia et al., 2013), goat D'man and Africa was 0.09 and 0.39 while the repeatability value was 0.11 and 0.12 (Boujenane et al., 1999; Adele et al., 2010).

Therefore, it was necessary to do research of the genetic parameter estimations on the prolificacy breeding program of PE goats in order to increase the welfare independence of food sovereignty based on populist agribusiness.

Materials and Methods

Prolificacy traits

The prolific data used in this study were taken from 520 Does from 14 breeding centers of PE goat in East Java. The location research is about temperatured 19-34°C, the climate is considered to be tropical. The number data analysis were 1347 records .

Data analyses

Heritability and repeatability was only estimated for the prolific potency of PE Does that had more than 3 times kidding because the data from the the does resulted were expected true potential of prolific potency. The data included the performance of estimation was performed for calculating variance component, repeatability and heritability estimates using paternal half sib correlation. The data were analyzed using by least square method outlined by Akesson et al. (2008) with SPSS version 15.

Variance component σ_s^2 was estimated by equating the expected mean squares to the observed mean squares with ANOVA's (Becker, 1992). Paternal half-sib estimate of heritability was calculated according the following formulas:

$$h^2 = \frac{4 \sigma_s^2}{\sigma_e^2 + \sigma_s^2}$$

Results and Discussion

The least square means of prolificacy trait was 1.72 ± 0.53 . Heritability and repeatability was only estimated for the prolific potency of ILEG Does that had more than 3 times kidding because the data from the the does resulted data were expected true potential of prolific potency. Heritability and repeatability estimates were based on dam-daughter relationships estimates were based on anova with SPSS 15 version (Table 1).

Table 1. Heritability and Variance Component of Prolificacy potency.

Trait	Value	Genetic Variation	Pphenotypic Variation	Variance Component of dams	Variance component between progeny within dams
Heritability	0.20	0.10	0.02	0.18	0.02
Repeatability	0.89			2.18	0.27

Conclusion

The prolific value of ILEG Does was 1.72 that was categorized medium. The heritability and repeatability estimate of prolific potency were medium and high categorized.

Acknowledgements

The Authors thank The Director of High Education for funding this research. Thanks are also due to goat breeding center in East Java.

References

- Adele, N.K., M.A. Dilala and A.O. Abdulyeken, 2010. Phenotypic and genetic parameter estimates of litter size and body weights in goats. *Int. J. Sci. Nat.*, 1 (2): 262-266.
- Akesson, M., S.Bensch, D. Hasselquist, M. Tarka and B. Hansson, 2008. Estimating Heritability and Genetic Correlations: Comparing the Animal Model with Parent-Offspring Regression Using Data from a Natural Population. *PLoS ONE* 3 (3):e1739 (1-10).
- Becker, W.A., 1992. *Manual of Quantitative Genetics*. The fifth edition. Pullman. USA: Academic Enterprises. pp. 5-35.
- Otuma, M.O. and P.N. Onu, 2013. Genetic effects, relationships and heritability of some growth trait in Nigeria crossbreed goats. *Agric. Biol. J. North America*: 4 (4); 388-392.
- Boujenane, I., M. Kerfal and M. Khallouk, 1999. Genetic and phenotypic parameters for litter traits of D'man ewes. *Anim. Prod.* 52:127-132.
- Casselas, J., G. Caja, A. Farret, and J. Piedrafta., 2007. Analysis of litter size and days to lambing in the Ripollesa ewe. Estimation of variance components and response to phenotype selection on litter size. *J. Anim. Sci.* 85:625-631.
- Dariusz, P., B. Kowaliszyn and S. Mroczkowski, 2011. Heritability and repeatability of the number of lambs born and reared estimated using linear and threshold models. *Archiv Tierzucht*: 54 (3): 271-279.
- Janssens S., W. Vandepitte, L. Bodin, 2004. Genetic parameters for litter size in sheep natural versus hormone-induced oestrus. *Gen. Sel. Evol.* 36:543-562.
- Kasab, A., B. Mioc, D. Jurkovic, D. Kompan, K. Plotonik, 2012. Estimates of environmental effects, genetic parameters and genetic trends for reproductive traits in Alpine goat. *Acta Agriculturae Slovenica, Supplement* 3:193-198.
- Mia, M.M., M.A.M.Y. Khandoker, S.S. Husain, M.O. Faruque, D.R. Notter, 2013. Estimation of genetic and phenotypic parameters of some reproductive traits of Black Bengal does. *Iranian J. Appl. Anim. Sci.* 3 (4): 829-837.
- Vatankhah, M. and M. A. Talebi, 2008. Estimates and correlations between production and reproductive traits in Lori-BaSouth Akhtiari sheep in Iran. *South African J. Anim.Sci.* 38 (2): 110-118.

Effect of microbial mixture on survival of fermented juice of epiphytic lactic acid bacteria (FJLB)

*Kraisit Vasupen**, *Benya Saenmahayak*, *Ekarat Vasupen* and *Smerjai Bureenok*

Faculty of Sciences and Liberal Arts, Rajamangala University of Technology Isan, Nakhon Ratchasima, 30000 Thailand

Abstract

This study was conducted to evaluate the effect of microbial mixture on viability of fermented juice of epiphytic lactic acid bacteria after freeze drying. Three types of mixed FJLB cultures were prepared as microbial mixture with skim milk, maltodextrin and starch. Those were freeze dried and stored at 4 °C for 30 days. FJLB with all microbial mixtures were determined viability, immediately after freeze drying and at day 30 during storage by counting number of colony forming unit per milliliter (CFU/ml) on MRS agar medium. FJLB mixed with skim milk and maltodextrin had higher survival of FJLB after freeze drying than FJLB mixed with starch (89%, 88% and 75%, respectively). These results suggested that, skim milk and maltodextrin are good microbial mixtures for freeze dried FJLB.

Keyword: Lactic acid bacteria, Freeze dried, Protectants

**Corresponding author: ks_vasupen@yahoo.com*

Introduction

In some parts of the world, antibiotics used to treat animals and added to feed as growth promoters may have adverse effects when associated resistant bacteria are taken into account. As the resistance of pathogens to antibiotics and the possibility of antibiotic residues in animal products attract increasing attention, the interest in the use of alternatives to in-feed antibiotics has been growing. Recent research with Lactic acid bacteria (LAB) in pigs suggests that LAB provide a potential alternative to antibiotic strategies (Partanen and Mroz, 1999, Yang et al, 2015). LAB from grass was a source of probiotic. Methodology for growing and increasing LAB population was easy by incubated grass juice with liquid sugar over night (Bureenok et al., 2005). However, LAB in liquid form was not suitable to use as feed additive. Freeze drying is usually used in the preservation of LAB. This technique has side effects on the physical state of membrane lipids and structure of sensitive proteins and decreasing of cell viability. Drying medium such as sugars, polysaccharides, skim milk, and egg yolk for preservation and decrease the loss of viability of bacteria cultures due to freezing stress are commonly used (Schoug, 2009, Hubalek, 2003). Alternative drying medium such as maltodextrin is a polysaccharide that is used as a food additive also can be used. This work were determined the effect of maltodextrin, skim milk, and starch on survival of LAB after freeze drying and during storage at 30 days.

Materials and Methods

Fermented juice of epiphytic lactic acid bacteria (FJLB) prepared from Napier grass. Napier grass 200 g was mixed with sterile water 1 L and sugar 2% (w/v) (Bureenok et al., 2005) incubated 2 days. FJLB were contained lactic acid bacteria 10^8 - 10^9 CFU/ml. FJLB suspension was prepared and mixed with three different mixtures (maltodextrin, skim milk, and starch) at 20% w/v. Three

replicates for each suspensions were frozen at -40°C for 24 hours and freeze dried for 4 hours in freeze dryer (CHAIST, model BETA 1-8 plus). After freeze dried immediately and during storage 30 days, dried FJLB with different mixtures were prepared in sterile distillation water (serial dilution 10^{-1} - 10^{-9}) and determined number of viable lactic acid bacteria by pour plating on MRS agar medium, incubated at 30°C for 18 hour. Lactic acid bacteria colony was counting as colony forming unit per milliliter (CFU/g).

Results and discussion

Table 1 Lactic acid bacteria counts on dried fermented juice of epiphytic lactic acid bacteria

	LAB (log ₁₀ CFU/g)		
	Before freeze-drying	After freeze-drying	Stored at 30 d
Skim milk	7.98	7.13 ^a	7.14 ^a
Maltodextrin	7.91	7.01 ^a	6.98 ^a
Starch	7.88	5.92 ^b	5.14 ^b
SEM	0.01	0.11	0.38
P-value	0.314	0.0004	0.018

^{a, b} Means within the same column with difference superscripts differ significantly ($P < 0.05$)

Ability of microbial mixtured to protect the viability of FJLB and during storage at 4°C for 30 days after freeze drying was studied. After freeze dried immediately, FJLB mixed with skim milk and maltodextrin had 7.13 and 7.01 log₁₀ CFU/g. Survival rate of FJLB higher than FJLB mixed with starch (5.92 log₁₀ CFU/g survival). The same result found after stored dried FJLB for 30 days. Loss viable lactic acid bacteria were higher when using starch as microbial mixture. Carvalho et al. (2004) reported that most LAB cultures in commercial selected skim milk powder as drying medium because it prevents cellular injury, creates a porous structure in freeze dried product that makes rehydration easier and contain proteins that provide a protective coating for cells. The degree of polymerization of dextran can affect its cryoprotective effectivity in *Pseudomonas* F8 (Hubalek, 2003) as a possible reason for protection by maltodextrin during freeze drying.

Conclusion

In conclusion, skim milk and maltodextrin are good microbial mixtures for freeze dried FJLB. This result will be useful in production of probiotic cultures. Thereby, it will aid to the development of new probiotic feed products which are important in infectious disease prevention.

References

- Bureenok, S., T. Namihira, Y. Kawamoto and T. Nakada. 2005. Additive effects of fermented juice of epiphytic lactic acid bacteria on the fermentative quality of guineagrass (*Panicum maximum* Jacq.) silage. *Grassland Science*. 51: 243-248.
- Carvalho., A. S., J. Silva, P. Ho, P. Teixeira, F. X. Malcata, and P. Gibbs. 2004. Relevant factors for the preparation of freeze-dried lactic acid bacteria. *International Dairy Journal* (14) 835-847.
- Hubalek, Z., 2003. Protectants used in the cryopreservation of microorganisms. *Cryobiology* (46) 205-229.
- Partanen, K. H., and Z. Mroz. 1999. Organic acids for performance enhancemant in pigs diets. *Nutrition Research Reviews*. 12. 117-145.
- Schoug, A., 2009. A Dry Phase of Life Freeze-drying and storage stability of *Lactobacillus coryniformis* Si3 in sucrose-based formulations, Doctoral Thesis, Faculty of Natural Resources and Agricultural Sciences, Department of Microbiology, Swedish University of Agricultural Sciences, Uppsala
- Yang, F., C. Hou, X. Zeng and S. Qiao. 2015. The Use of Lactic Acid Bacteria as a Probiotic in Swine Diets. *Pathogens* (4) 34-45.

Effects of storage time on external and internal characteristics of lutein eggs

K. Lokaewmanee* & P. Meesrri

Department of Agriculture Resources, Faculty of Natural Resources and Agro-Industry, Kasetsart University, Chalermphrakiat Sakon Nakhon Province Campus, Sakon Nakhon 47000, Thailand

Abstract

The effects of egg storage time were studied on the external and internal characteristics of lutein eggs. A total of 330 lutein eggs were stored at room temperature (30°C) for periods of 0, 3, 5, 7, 9, 11, 13, 15, 17, 19 and 21 days, respectively. At the end of each storage period, 30 lutein eggs were evaluated for egg quality. Storage time from 0 to 21 days did not affect the eggshell thickness, shell ratio or yolk color ($P>0.05$). The effect of days in storage was statistically significant on the albumen ratio and Haugh unit ($P<0.05$) which both decreased with increasing days of storage. Conversely, egg water loss and the yolk ratio increased with increasing days of storage ($P<0.05$). At the end of storage, the lutein value in the eggs was close to the value at the beginning of storage. The results indicated that storage time did not produce an adverse effect on the eggshell thickness, shell ratio, yolk color and concentration of lutein in lutein eggs.

Keywords: storage time, characteristics, lutein eggs

**Corresponding author: csnkdp@ku.ac.th*

Introduction

Lutein is considered a nutrient that prevents age-related macular degeneration (Snodderly, 1995) by aiding in filtering damaging blue light and sunlight (Sperling et al., 1980). Macular degeneration is the leading cause of blindness, resulting in progressive and irreversible loss of central region vision (Lesson and Caston, 2004). As lutein is not synthesized in the body, dietary ingestion is the only source to meet the requirements of lutein to prevent macular degeneration (Lim et al., 1992). The intake of lutein in adults is less than 1 mg/d, which is much less than the preventive levels being tested for these nutrients (Grando et al., 2003). Goodrow et al., (2006) reported that egg normally contains 0.14 to 0.16 mg of lutein. Moreover, Lokaewmanee et al., (2012) found that layers fed 50 mg/kg dietary lutein increased the lutein concentration in their eggs. However, there is limited information on the storage time of lutein eggs with regard to egg quality and the lutein concentration in the egg. Therefore, the present study was conducted to investigate the effect of storage time on the external and internal characteristics of lutein eggs.

Materials and Methods

Lutein eggs were collected after the hens had been fed dietary lutein (50 mg/kg of feed) for 3 weeks. In total, 330 lutein eggs were stored at room temperature (about 30°C). According to storage time, eggs were weighed, broken and evaluated after 0, 3, 5, 7, 9, 11, 13, 15, 17, 19 and 21 days of storage.

Thirty lutein eggs from each the period of storage were measured for egg water loss and egg quality. At first, the egg weight was recorded by using an electronic digital balance. Each egg was broken onto a metal plate and the height of the albumen was measured as the distance between the metal plate and the electrode placed on top of the thick part of the egg. Then, the weights of the albumen and egg yolk were measured using an electronic digital balance. The shell was dried at room temperature and weighed according to Scott and Silversides (2000). The shell thickness was measured at three different parts (equator, top and truncated edge) of the shell in each egg using a micrometer and the mean was recorded as the shell thickness. Yolk color was measured by a subjective method using the Roche yolk color fan (Roche Ltd., Basel, Switzerland). At the start (day 0 of storage) and the end (day 21 of storage), lutein in the eggs was analyzed by the Institute of Food Research and Product Development, Kasetsart University, Bangkok, Thailand. Then, the egg water loss, shell ratio, albumen ratio, yolk ratio and Haugh unit were calculated as follows:

$$\text{Egg water loss} = \frac{(\text{Egg weight} - \text{Egg broken weight})}{\text{Egg weight}} \times 100$$

$$\text{Shell ratio} = \frac{(\text{Shell weight})}{\text{Egg weight}} \times 100$$

$$\text{Albumen ratio} = \frac{(\text{Albumen weight})}{\text{Egg weight}} \times 100$$

$$\text{Yolk ratio} = \frac{(\text{Yolk weight})}{\text{Egg weight}} \times 100$$

$$\text{Haugh unit} = 100 \log(H - 1.7W^{0.37} + 7.6)$$

Where

H=Observed height of the albumen (mm)

W=Weight of egg (g)

Statistical Analysis

All data were statistically analyzed using one-way analysis of variance with the Statistical Analysis System computer software (SAS, 1996). Differences among the treatment groups were tested by

Tukey's Studentized range test, with differences considered significant at $P < 0.05$ (Steel and Torrie, 1980).

Results and Discussion

A summary of the external and internal characteristics of lutein eggs after storage is presented in Table 1. Storage of 0 to 21 days did not affect the eggshell thickness, shell ratio and yolk color ($P > 0.05$). The effect of storage time was statistically significant on the egg water loss, albumen ratio, yolk ratio and Haugh unit ($P < 0.05$). The egg water loss and yolk ratio significantly increased with increased storage time. Our findings are in agreement with those of Silversides and Villeneuve (1994), who reported a significant egg water loss increase of 1.94 g after storage for 10 days at 29°C. Tilki and Saatci (2004) reported a significant yolk ratio increase of 3.34% after storage for 35 days at room temperature (about 15-18°C). Dramatic deterioration was observed in both the albumen ratio and Haugh unit due to the storage time. Tilki and Saatci (2004) reported a significant albumen ratio decrease of 5.40% after storage for 35 days at room temperature (about 15-18°C). In general, the storage time and temperature appear to be the most critical factors affecting the Haugh unit (Samli et al., 2005). These results are in agreement with those of Scott and Silversides (2000), who reported a significant ($P < 0.05$) decrease from 91.4 to 40.6 in the Haugh unit in eggs after 10 days of storage at 29°C. Interestingly, the concentration of egg lutein was not significantly different throughout 21 days of storage. The reason for this is not clear and further research is needed to clarify this point.

Table 1 Effects of egg storage time on external and internal characteristics in lutein eggs (Mean±SD, n=30).

Days of Storage	0	3	5	7	9	11	13	15	17	19	21	P-value
Egg water loss,%	-	1.06±0.02 ^d	1.11±0.02 ^d	1.71±0.01 ^d	3.87±0.02 ^c	4.12±0.01 ^{bc}	4.67±0.04 ^b	5.81±0.06 ^b	7.95±0.03 ^a	8.24±0.05 ^a	9.24±0.02 ^a	<0.01
Eggshell thickness,mm	0.40±0.01	0.41±0.01	0.40±0.01	0.41±0.01	0.40±0.01	0.39±0.01	0.38±0.01	0.40±0.01	0.41±0.01	0.41±0.01	0.39±0.01	0.59
Shell ratio,%	10.37±0.22	10.69±0.24	10.02±0.14	10.26±0.24	9.80±0.18	10.03±0.16	11.03±0.18	10.77±0.14	10.88±0.20	11.41±0.21	10.94±0.10	0.24
Albumen ratio,%	65.53±0.48 ^a	64.89±0.47 ^a	63.46±0.27 ^a	60.62±0.49 ^{ab}	60.37±0.55 ^{ab}	58.10±0.60 ^b	53.10±0.60 ^b	50.25±0.42 ^{bc}	47.23±0.46 ^c	46.62 ±0.28 ^c	46.40±0.48 ^c	<0.01
Yolk ratio,%	27.37±0.35 ^c	27.88±0.42 ^c	29.64±0.22 ^b	30.39±0.37 ^{ab}	30.26±0.46 ^{ab}	31.01±0.59 ^a	31.37±0.41 ^a	31.93±0.40 ^a	31.45±0.25 ^a	33.88±0.30 ^a	34.99±0.41 ^a	<0.01
Haugh unit	76.81±0.48 ^a	60.02±0.35 ^b	52.83±0.25 ^c	51.02±0.73 ^c	40.68±0.39 ^d	39.68±0.38 ^d	37.19±0.40 ^d	33.70±0.50 ^e	26.04±0.27 ^f	14.70±0.28 ^g	10.15±0.20 ^g	<0.01
Yolk color	12.40±0.20	12.35 ±0.20	12.46±0.20	12.01±0.20	12.40±0.20	12.40±0.20	12.40±0.23	12.20±0.20	12.22±0.20	12.10±0.20	12.40±0.20	0.61
Lutein, µg/100g	2,944.90	-	-	-	-	-	-	-	-	-	2,791.00	0.72

^{a-g} Means in the same row with different superscripts differ significantly (P<0.05).

Conclusion

The results suggested that the eggshell thickness, shell ratio, yolk color and concentration of egg lutein were not affected during 0 to 21 days of storage at 30°C.

References

- Goodrow, E.F., T.A. Wilson, S.C. Houde, R. Vishwanathan, P.A. Scollin, G. Handelman and R.J. Nocolas, 2006. Consumption of one egg per day increase serum lutein and zeaxanthin concentration in adults without altering serum lipid and lipoprotein. *J. Nutr.* 136:2519-2524.
- Grando, F., B. Olmedilla and I. Blanco. 2003. Nutritional and Clinical relevance of lutein in human health. *Br. J. Nutr.* 90:487-502.
- Leeson, S. and L. Caston, 2004. Enrichment of egg with lutein. *Poult. Sci.* 83:1709-1712.
- Lim, B.P., A. Nagao, J. Terao, K. Tanaka, T. Suzuki and K. Takama, 1992. Antioxidant activity of xanthophylls on peroxy radical-mediated phospholipid peroxidation. *Biochimica. et. Biophysica. Acta.* 1126:178-184.
- Lokaewmanee, K., K. Yamauchi, T. Komori and K. Saito, 2012. Effects of dietary lutein on raw and boiled egg yolk color and lutein concentration in egg yolk. The 15th AAAP Animal Science Congress. 26-30 November 2012, Thannasat University, Rangsit Campus, Thailand.
- Samli, H.E., A. Agma and N. Senkoylu. 2005. Effects of storage time and temperature on egg quality in old laying hens. *J. Appl. Poult. Res.* 14:548-553.
- SAS Institute, 1996. SAS User's Guide: Statistical Version 7.0. SAS Institute Inc., Cary, NC.
- Scott, T.A. and F.G. Silversides, 2000. The effect of storage and strain of hen on egg quality. *Poult. Sci.* 79:1725-1729.
- Silversides, F.G. and P. Villeneuve. 1994. Is the Haugh unit correction for egg weight valid for egg stored at room temperature. *Poult. Sci.* 73:50-55.
- Snodderly, D.M., 1995. Evidence for protection against age-related macular degeneration by carotenoids and antioxidant vitamins. *Am. J. Cli. Nutr.* 62:1448S-1461S.
- Steel, R.G.D. and J.H., 1980. Principle and Procedures of Statistics A Biometrical Approach. 2nd ed., McGraw-Hill Inc., New York.
- Sperling, H.G., C. Johnson and R.S. Harwerth, 1980. Differential spectralphotic damage to primate cones. *Vision Res.* 117-1125.
- Tilki, M. and M. Saatci. 2004. Effects of storage time on external and internal characteristics in partridge (*Alectoris graeca*) eggs. *Revue. Med. Vet.* 15(11):561-564.

Efficacy of probiotic *Enterococcus mundtii* in dried form in Broilers

Kasornpikul, C.¹, & S.Chalorsuntisakul

Faculty of Animal Science and Agricultural Technology, Silpakorn University, Phetchaburi Information Technology Campus, Sam Praya, Cha-am, Phetchaburi, 76120, THAILAND

Abstract

Enterococcus mundtii is a beneficial microorganism found in the faeces of the poultry raised in the western part of Thailand. If its easy powder form can be developed, this will allow farmers to get an easy-use and -access to the product. The objective of this research was to examine the safety of this probiotic for broilers and the efficacy of its powder form on growth and immunity boosting after vaccination in the animal. In the production procedure of the probiotic powder, the probiotic was prepared in Man, Rogosa and Sharpe (MRS) broth. The precipitate obtained was dissolve in NaCl solution (0.9%) and then mixed with methylcellulose 1.25% and broken milled rice decontaminated with steam sterilization. The mixture was then dehydrated in a decontaminated hot air oven at 45°C during 3 days and tested with 60 male broilers, which were raised separately into 2 groups with 2 replications for each groups and 15 birds per replications. The research was used completely randomized design (CRD). The broilers were fed according to their nutrition requirement as follows; the commercial feed group and the commercial feed supplemented with powdered probiotic of 10⁶cfu/g. The experimental period was done for 28 days with a vaccination against Newcastle disease. 3 broilers (13.34%) in the control group were found dead. The average daily gain was statistically different. However, feed intake, feed conversion ratio (FCR), and the immunity level against Newcastle disease were not difference. In conclusion, although the average daily gain was higher in the control group, but no broiler was dead in the probiotic group. This allows the farmers to earn more from selling benefits and implies that the powdered probiotic is safe and can be used to feed broilers.

Keywords: *Enterococcus mundtii*, *beneficial microorganism*, *poultry*, *broilers*, *probiotics*

Corresponding author: makemerry@gmail.com

Introduction

Enterococcus mundtii is a useful probiotic obtained from poultry faeces raised in the western part of Thailand. It has been tested in broilers and found efficient in growth boosting and against the development of pathogenic bacteria (Chalorsuntisakul et al., 2010). If it can be developed into an easy probiotic powder, it will be beneficial and more accessible to the farmers. The development of the powdered probiotic is done by using rice as component. More than 60% of Thai population are farmers, and the main crop is rice. Thailand is the biggest exporter in the world rice market, more than 7 million tons a year. Thai rice gains a reputation worldwide and brings a great amount of income to the country each year. Thus, rice is the country's most important economic crop (Thai Rice Foundation under Royal Patronage, 2014). Therefore, the powdered probiotic development

by using rice is adding value to broken rice or broken milled rice which is the residue and has low price. However, the product should be tested on the safety for the animal before use and examined for its efficacy in boosting growth and enhancing the immunity against vaccination of the animal before its distribution to the farmers.

Materials and Methods

To prepare the probiotic powder, probiotic *Enterococcus mundtii* (Surawat et al., 2010) was bred in MRS broth and then spinned in a centrifuge to obtain the precipitate which was dissolved in the solution of NaCl 0.9% and then mixed with methylcellulose 1.25%. This mixture was then mixed with broken milled rice sterilized with steam in a ratio of 1:2 and then dehydrated in a decontaminated oven at 45°C during 3 days (modified from Suthamma & Taweechai, 2014). The number of survival bacteria in the product was then determined using the pour plate method. The product was mixed with industrial poultry feed to obtain the volume of 10⁶cfu/g. In vivo, the test was performed on broilers. A total, 60 male broilers were selected and kept together in a house during 7 days before being separated into 2 groups with 2 replications for each groups and 15 birds per replications in the area of 1 m² for each replication. A completely randomized design (CRD) was used for all experiments. Control group (Cont.), fed on commercial feed and the treatment group (Treat.) fed on commercial feed supplemented with probiotic powder of 10⁶ cfu/g. and determined weighed and mortality rate. At day 28 a vaccination against Newcastle disease was done. Then a blood test was performed 7 days after the vaccination in order to determine the immunity level.

Results

In the preparation of probiotic powder, 1.06 X 10¹²cfu/ml. of powered probiotic could be obtained and kept in a well-sealed container during 3 months. After the application, at the end of the test period, 3 broilers (13.34%) in the control group were found dead, but no broiler dead in the treatment group. The difference in terms of average daily gain was statistically different. However, feed intake, feed conversion ratio (FCR), and the immunity level against Newcastle disease were not difference, as showed in the Table 1.

Table 1: Growth performance values and the immunity level against Newcastle disease vaccine.

	ADG		FI		FCR		HI – Titer (log2)	
	Cont.	Treat.	Cont.	Treat.	Cont.	Treat.	Cont.	Treat.
Mean	62.09	59.75	105.83	101.50	1.704	1.698	2.79	2.75
Variance	2.339	0.003	3.437	0.242	0.00014	0.00004	0.423	0.125
F	779.666*		14.202		3.5		3.384	

*The difference is statistically significant at F (0.05, 1, 1).

Discussions and Conclusions

Enterococcus mundtii is a genus of bacteria that produce the bacteriocin type mundtacin. Bennik et al. (1998) reported that mundtacin belonged to the class II bacteriocins of lactic acid bacteria. It was showed to inhibit the growth of *Listeria monocytogenes*, *Clostridium botulinum* and a variety

of lactic acid bacteria. Therefore, it is expected that *Enterococcus mundtii* should be appropriate to be used as probiotic. Chalorsuntisakul et al. (2010) isolated this kind of bacteria from the poultry faeces in the western part of Thailand. As they can grow at the temperature of 30 – 45 °C, the method of Suthamma&Taweechai (2014) has been adapted to this experiment in which the probiotic was dehydrated not at 37 °C but at 45 °C in order to obtain well-dried rice and prevent it from mold. The probiotic cells were protected by Methyl celluloses, which pharmaceutical grades have been used as thickeners, binders, emulsifiers, and stabilizers in a variety of cosmetic and food products (Sigma Aldrich, 2014).

In this present experiment, however, it was found that the average daily gain of the control group was significantly higher than the probiotic group, but no difference was found in feed intake, FCR, and the immunity level. The higher average daily gain of the control group could be explained by the stocking density of the broilers. As those in the 13.34% of control group were found dead, the population density of this group was lower than the treatment group. On this, Feddes et al. (2002) explained that there was a significant effect of stocking density on broiler performance and carcass traits. They carried out an experiment to observe the effects of four stocking and water nipple densities on broiler performance and carcass traits which were measured in two trials. The stocking densities of 23.8, 17.9, 14.3, and 11.9 birds/m² corresponded to 260, 195, 156, and 130 birds per pen, respectively. It was found that water nipple density had no effect on broiler performance or carcass quality. Maximum growth was observed at a stocking density of 14.3 birds/m² and FCR did not significantly differ among treatments. This finding is consistent with this present experiment.

In terms of the immunity level against Newcastle disease, no difference was found in both groups. This is consistent with Alkhalf et al. (2010) findings of their study that there were no significant differences between samples taken at 21 and 28 days of age (7 and 14 days post immunization) while probiotic had a significant enhancement effect on antibody titres against NDV at 42 days of age (28 days post immunization). In this present experiment, no blood sample was collected at 14 and 28 days after the vaccination. As the result, the difference in terms of the immunity level could not be determined. This will be a suggestion for the next experiment. In conclusion, although the average daily gain of the control group in this experiment was higher than the probiotic group, but the higher survival rate of the latter allowed the farmers to earn more from selling benefits, and its survival rate at 100% signified the safety of using the powdered probiotic in raising broilers.

References

- Alkhalf, A., M. Alhaj., and I. Al-Homidan. 2010. Influence of probiotic supplementation on immune response of broiler chicks. *Egypt. Poult. Sci.* 30: 271-280.
- Bennika M.H.J., B. Vanloob, R. Brasseur, L.G.M. Gorris and E. J. Smida . 1998. A novel bacteriocin with a YGNGV motif from vegetable-associated *Enterococcus mundtii*: full characterization and interaction with target organisms. *BBA–Biomembranes.* 1373: 47–58.
- Chalorsuntisakul S., C. Kasornpikul and M. Preyavichyapugdee. 2010. Isolation of Beneficial Microorganism Isolated from Poultry Feces in Western Region of Thailand. Available online http://dric.nrct.go.th/bookdetail.php?book_id=247634.
- Feddes J.J., E.J. Emmanuel and M.J. Zuidhoft. 2002. Broiler performance, body weight

variance, feed and water intake, and carcass quality at different stocking densities. *Poultry Sci.* 81(6): 774-779.

Suthamma K. and N.Taweechai. 2014. Powder production of bacterial antagonistic *Bacillus Subtilis* (Bs). Available online http://www.rdi.ku.ac.th/kasetresearch52/04-plant/gaigaew/plant_00.html.

Sigma aidrich. 2014. Product information Methyl Cellulose. Available at https://www.sigmaaldrich.com/content/dam/sigma-aldrich/docs/Sigma-Aldrich/Product_Information_Sheet/m0262pis.pdf.

Thai Rice Foundation under Royal Patronage .2014. History of Thai Rice Foundation . Available online <http://www.thairice.org/html/foundation/foundth01.html> (17December 2014)

Truly absorbed protein in the small intestine content of alfalfa hay harvested at various blooming

Kheyrandish, P., M. Danesh Mesgaran & A. Vakili

Department of Animal Science, Faculty of Agriculture, Ferdowsi University of Mashhad, P. O. Box 91775-1163, Mashhad, Iran

Abstract

The present study was conducted to determine chemical composition and truly absorbed protein in the small intestine (DVE) content of alfalfa hay sampled during different stage of the plant maturation. Samples of alfalfa hay were taken at initial, half and full bloom stages, then dried at 65 °C 48 h, using air forced oven, then analyzed to determine crude protein, neutral detergent fiber and acid detergent fiber. Ruminant incubations for test feeds and laboratory techniques were performed according to protocol for in situ rumen incubations published by central veevoeder bureau standards. Dry matter content of the samples were increased with the plant maturation and varied from 260 (g/kg) at the initial bloom to 280 (g/kg) in the full bloom. The NDF and ADF content of alfalfa at the initial bloom, half bloom and in full bloom were 381 and 292; 432 and 308; 511 and 327 g/kg DM, respectively. However, crude protein was decrease with the stage of growth and the value for the initial, half and full bloom was 198, 192, and 190g/kg, respectively. The value of the DVE of the samples was decline with the stage of the blooming and were 162.9, 161.3 and 156.1 g/kg for initial, half and full bloom, respectively. The present results indicated that the stage of maturity might impact on the nutritional value of alfalfa hay by altering the chemical composition and especially the protein content evaluated as truly absorbed protein in the small intestine.

Keywords: DVE/OEB, alfalfa hay, growth stage

**Corresponding author: danesh@um.ac.ir*

Introduction

Attention to forage evaluation will be one of the most critical step to successful rearing. France et al. (2000) who noted “the description of a feed for its ability to sustain different types and levels of animal performance” as feed evaluation. Several investigators noted that changing in chemical composition of forage is affected by growth stage, forage species (Griffin et al., 1994), climate (Mathison et al., 1996), planting (Hintz & Albrecht, 1994), and growing conditions (Cox et al., 1994). The objectives of this study were to determine truly absorbed protein in the small intestine content of alfalfa hay harvested at various blooming and the newly developed Dutch protein evaluation system: the DVE/OEB model (Tamminga et al., 1994) were employed.

Material and Methods

Alfalfa hays were cut and assigned to one of three stage of maturity including 1) initial blooming 2) half blooming and 3) full blooming. The DM, ash, EE and crude protein were determined. Netural detergent fiber (NDF) and acid detergent fiber (ADF) values were analyzed according to the procedures of Van Soest et al. (1991). The DVE value is calculated as follows: $DVE = DVBE + DVME - DVMFE$, where DVME is the microbial protein synthesized in the rumen and digested in the intestine, DVBE is the feed protein not degraded in the rumen but digested in small intestine, and DVMFE is the endogenous protein losses associated with digestion. Data were analyzed as a complete randomized design by MIXED procedures of SAS.

Results and Discussion

Chemical compositions

The effects of growth stage of alfalfa hay on chemical composition are presented in Table 1.

Table 1.chemical composition.

Item	DM	CP	NDF	ADF	Ash
			g/kg DM		
Initial bloom	260 ^b	198.20 ^a	381 ^c	292 ^c	103.80 ^a
Half bloom	280 ^a	192.60 ^a	432 ^b	308 ^b	92.80 ^b
Full bloom	280 ^a	189.80 ^a	511 ^a	327.3 ^a	91.20 ^b
SEM	1.15	4.2	3.5	3.7	1.4

a, b and c indicate significance among treatments ($P < 0.05$).

Dry matter content increased with plant maturation ($P < 0.05$) and was varied from 260 (g/kg) at the initial bloom to 280 (g/kg) in the full bloom. Both NDF and ADF contents were affected by advancement of age. Orloff and Putnam (1998) reported by delaying each day in harvesting of second cut of alfalfa, ADF and NDF increased approximately 0.4 and 0.37 percent. No maturation effect on crude protein content was observed ($P > 0.05$). The lack of a significant effect of advancing maturity on crude protein content in this study may be related to alfalfa species or growing conditions. As alfalfa maturity advanced ash content declined ($P < 0.05$) and decreased from 103.80 in initial blooming to 91.20 g/kg DM in full blooming.

Truly absorbed protein and degraded protein balance (DVE/OEB)

Throughout the course of the maturity, there were significant differences among truly absorbed protein in the intestine by advancement of age (Table 2). The DVE value markedly reduced during different stages of growth ($P < 0.05$). The OEB that is conclude the imbalance between microbial protein synthesis from available rumen degradable CP and potential energy from anaerobic fermentation in the rumen. The optimum OEB value in a dairy ration is zero or slightly above (Tamminga et al., 1994). The OEB content reduced by advancement of age and was lowest in full blooming (-17.14). When OEB is negative, inclusion of addition fermentable OM such as

degradable starch may improve microbial protein. Stage of maturity might impact on the nutritional value of alfalfa hay by altering the chemical composition.

Table 2. Prediction of truly digested and absorbed rumen synthesized microbial (MCP) and undergraded feed protein (RUP) in the small intestine according to the new Dutch DVE/OEB.

Items	Truly digested and absorbed MCP					Truly digested and absorbed MCP			
	FOM	MCP ^{DVE} _{RDP}	MCP _{FOM}	DVME	DVMFE	TPSI	DVBE	DVE	OEB
	g/kg DM								
Initial bloom	517.53 ^a	69.25 ^a	77.63 ^a	55.15 ^a	2.72 ^a	174.36 ^a	116.14 ^a	168.56 ^a	-8.38 ^a
Half bloom	564.88 ^a	70.30 ^a	84.73 ^a	54.02 ^a	2.44 ^a	173.27 ^a	108.77 ^a	161.16 ^{ab}	-14.30 ^a
Full bloom	584.75 ^a	79.10 ^a	87.71 ^a	52.92 ^a	2.39 ^a	167.83 ^a	105.58 ^a	156.14 ^b	-17.41 ^a
SEM	41.37	5.13	6.20	1.42	0.038	4.76	2.82	2	6.92

FOM: organic matter fermented in the rumen, MCP^{DVE}_{RDP}: microbial protein synthesized in the rumen based on rumen degraded feed crude protein, MCP_{FOM}: microbial protein synthesized in the rumen based on available energy, DVME: Rumen synthesized microbial protein digested in the small intestine, DVMFE: endogenous protein losses in the digestive tract, TPSI: true protein supplied to the small intestine, DVBE: Digestion in small intestine of the undergraded feed protein, DVE: truly absorbed protein in the small intestine, OEB: reflects the difference between the potential microbial protein syntheses based on rumen degraded feed CP and that based on energy (rumen fermented OM) available, for microbial fermentation in the rumen

Conclusion

The present results indicated that the stage of maturity might impact on the nutritional value of alfalfa hay by altering the chemical composition and especially the protein content evaluated as truly absorbed protein in the small intestine.

References

- Cox, W. J., J. H. Cherney, D. J. R. Cherney, and R. D. Pardee, 1994. Forage quality and harvest index of corn hybrids under different growing conditions. *Agron J.* 86: 277–282.
- France, J., M. K. Theodorou, R.S. Lowman, D.E. Beever, 2000. Feed evaluation for animal production. In: Theodorou, M.K., France, J. (Eds.), *Feeding Systems and Feed Evaluation Models*. CAB International, Wallingford, UK, pp. 1-9.
- Griffin, T. S., K. A. Cassida, O. B. Hesterman, and S. R. Rust, 1994. Alfalfa maturity and cultivar effects on chemical and in situ estimates of protein degradability. *Crop Sci.* 34 : 1654–1661.
- Hintz, R. W. and K. A. Albrecht, 1994. Dry matter partitioning and forage nutritive value of soybean plant components. *Agron J.* 86: 59–62.
- Mathison, R. D., C. C. Sheaffer, D. L. Rabas, D. R. Swanson, and J. H. Halgerson, 1996. Early spring clipping and herbicide treatments delay alfalfa maturity. *J. Prod. Agric.* 9 : 505–509.
- Orloff, S. B. and D. H. Putnam, 1998; Selecting cutting schedules -The yield and quality tradeoff. Proceedings, 28 California Alfalfa Symposium, 3-4 December, 1998, Reno, Nevada, UC Cooperative Extension, University of California, Davis.
- Tamminga, S., Van W.M. Straalen, A.P.J. Subnel, R.G.M. Meijer, A. Steg, C.J.G. Wever, M.C. Block, 1994. The Dutch protein evaluation system: the DVE/OEB-system. *Livestock Prod. Sci.* 40, 139±155.

Growth response of purebred Merino and crossbred prime lambs supplemented with canola and flaxseed oils

Nguyen V. D.^{*1,2}, S. W. Ives¹, R. W. Smith¹ & A. E.O. Malau-Aduli^{1,3}

¹University of Tasmania, Private Bag 98, Hobart, TAS 7001, Australia, ²National Institute of Animal Science, Ha Noi, Viet Nam, ³James Cook University, Townsville, QLD 4781, Australia.

Abstract

The objective of this study was to evaluate the influence of flaxseed and canola oil supplementation on the growth of genetically divergent prime lambs. Seventy-two weaned lambs from Corriedale sires mated to Merino dams (CxM), White Suffolk sires mated to Corriedale dams (WxC) and purebred Merino (MxM) were randomly distributed into six treatment groups. Each group were daily supplemented with 1 kg pellet per lamb either flaxseed or canola oil at no oil (control), 25ml/kg (low) and 50ml/kg (high) oil levels. Lambs had ad libitum access to lucerne hay and water over a ten-week period. Results demonstrated that while gender had an inconsequential impact, significant differences in average daily gain (ADG) were attributable to oil supplementation. Furthermore, the appropriate level of these oils should be 50ml/kg over a ten-week supplementation. Lamb breed exerted the most significant impacts on liveweight and body condition score with WxC lambs elicited the best growth performance in this study.

Keywords: flaxseed oil, canola oil, supplementation, prime lambs, growth.

**Corresponding author: V.D.Nguyen@utas.edu.au*

Introduction

Crossbreeding and intensive supplementation are methods to improve lamb growth and slaughter weights (Fogarty and Mulholland 2014). Australian sheep producers are seeking the best combination of genetics and dietary supplementation options for producing superior lambs. Intake of omega-3 long-chain polyunsaturated fatty acids (ω -3 LC PUFA) has been related to a lower risk of cardiac diseases in humans (Mozaffarian and Wu 2011). Lamb contains a higher fat level, but it has lower contents of ω -3 LC PUFA (Enser et al. 1996). Incorporating vegetable oils into lamb diets can increase the ω -3 LC PUFA content of lamb. Oils from flaxseed and canola are rich sources of healthy ω -3 LC PUFA (Gillingham et al. 2011). These oils are the sources of energy-dense supplements. Despite many research investigations on improving the ω -3 LC PUFA content in animal products, on-farm research on the appropriate supplementary levels of these oils in prime lambs has received little attention. Therefore, the study investigated the impact of supplementing ω -3 LC PUFA enriched diets on the growth of prime lambs from genetically divergent backgrounds under the same management.

Materials and Methods

A total of 72 seven-month-old first cross lambs (average live weight (LW) 33.4 kg and body condition score (BCS) 2.7) from Corriedale sires mated to Merino dams (CxM), White Suffolk sires mated to Corriedale dams (WxC) and purebred Merino (MxM) were randomly allocated to one of 6 treatment groups (12 lambs per group) and supplemented with 1kg of pellets per day for 10 weeks including 3 weeks of adjustment at the following levels: High = 50mL/kg, Control = no

oil and Low = 50% high and 50% control pellets. All lambs were dewormed and raised in a partially roofed 4x12m pen and had unlimited access to lucerne hay and clean water. Feed intakes were recorded daily while BCS and LW were taken fortnightly. Feed samples were dried in a fan-forced oven to a constant weight at 65°C to determine dry matter (DM) content. Total nitrogen (N) was quantified using an elemental analyser (PE2400 Series II; Perkin-Elmer Corp, USA), and crude protein (CP) content was estimated by multiplying N by 6.25. Ether extract (EE) was determined using an Ankom fat/oil extractor (ANKOMXT15; ANKOM Technology, USA). Acid detergent fibre (ADF) and neutral detergent fibre (NDF) contents were measured by utilising an Ankom fibre analyser (ANKOM220; ANKOM Technology, USA). The samples were combusted in a furnace at 550°C for 5 hours to quantify ash content. Near infrared reflectance spectroscopy method was used to estimate metabolisable energy (ME). The nutritive compositions of feedstuff are presented in Table 1. General linear model procedures (PROC GLM) in Statistical Systems Analysis (SAS 2009) were used to fit the fixed effects of ω -3 supplementation level, gender and breed on lamb growth traits. Separation of significant differences between means was performed using Tukey's tests at the $P < 0.05$ threshold.

Results and Discussion

Oil rich in ω -3 LC PUFA had no significant ($P > 0.05$) impact on total dry matter intake (DMI) (Table 2). Our results were consistent with Manso et al. (2009) who fed lambs with dietary oil levels at 40 g/kg DMI. Conversely, Francisco et al. (2015) reported that lipid supplementation levels of up to 80 g/kg DMI caused a decrease in DMI due to reduced feed palatability and digestive nutrient flows (Annett et al. 2011). The effects of lipid supplementation may be dependent on feeding levels (Francisco et al. 2015). In this study, as dietary lipid contents were below 5% of total DM, oil supplementation had no deleterious effect on DMI of lambs.

Table 1. Feed nutrient compositions

Items	Unit	Control pellet	Canola pellet	Flaxseed pellet	Lucerne hay
DM	g/kg	898	879	894	896
OM	g/kgDM	920	918	936	928
CP	g/kgDM	145	147	140	174
EE	g/kgDM	30	57	41	15
NDF	g/kgDM	238	239	230	465
ADF	g/kgDM	92	89	98	309
Ash	g/kgDM	64	80	82	72
ME	MJ/kgDM	10.7	11.1	11.1	–

Table 2. Daily feed intake (kg DM/head)[‡]

Treatment	Pellet	Lucerne hay	Total
Control	0.828±6 ^b	0.911±24	1.761±31
Canola Low	0.844±6 ^{ab}	0.892±32	1.734±38
Canola High	0.828±6 ^b	0.915±28	1.743±41
Flaxseed Low	0.845±6 ^{ab}	0.981±14	1.840±20
Flaxseed High	0.861±5 ^a	0.865±37	1.724±47
<i>P-value</i>	0.01**	0.09 ^{ns}	0.17 ^{ns}

Table 3. Impact of flaxseed oil supplementation, breed and gender on lamb growth[‡]

	n	LW (kg)	ADG (g/day)	BCS (0-5)
Treatment				
Control	24	44.1±0.9	152±11 ^a	3.3±0.1
Canola Low	12	44.9±1.1	176±12 ^{ab}	3.3±0.1
Canola High	12	44.5±1.2	189±11 ^b	3.4±0.1
Flaxseed Low	12	44.1±0.8	165±10 ^{ab}	3.3±0.1
Flaxseed High	12	44.8±1.0	188±13 ^b	3.3±0.1
<i>P value</i>		0.07 ^{ns}	0.04 [*]	0.10 ^{ns}
Gender				
Ewes	36	45.1±0.6	186±12	3.3±0.1
Wethers	36	43.7±0.7	166±10	3.3±0.1
<i>P value</i>		0.08 ^{ns}	0.09 ^{ns}	0.10 ^{ns}
Breed				
CxM	24	44.5±0.9 ^{ab}	182±13 ^b	3.3±0.1 ^a
MxM	24	43.1±0.7 ^a	156±7 ^a	3.1±0.1 ^a
WxC	24	45.7±0.7 ^b	189±11 ^b	3.5±0.1 ^b
<i>P-value</i>		0.02 [*]	0.01 [*]	0.04 [*]

[‡]Column means bearing different superscripts differ significantly

Oil-rich ω -3 supplementation resulted in significant ($P < 0.05$) differences in average daily gain (ADG), while it did not cause any marked variation in final liveweight and BCS (Table 3). Lamb growth did not significantly differ between genders. The results are in agreement with Malau-Aduli et al. (2014) who supplemented degummed canola oil at the same levels to prime lambs. The absence of negative influences on the growth traits could be attributed to the similarity of DMI and ME content of the diets. Growth significantly ($P < 0.05$) differed among breeds (Table 3). WxC lambs had markedly heavier liveweight (45.7 kg) and higher BCS (3.5) than purebred Merinos (43.1 kg and 3.1 respectively). Purebred Merino lambs recorded the smallest ADG (156 g/day). The differences could be due to variations in genetic disposition towards muscle, wool, fat and diversity in feed conversion efficiency between breeds (Hegarty et al. 2006). Lewis et al. (2006) concluded that lambs from high growth breeds grew larger than their counterparts from low growth pedigree. Therefore, when prime lambs are fed identical nutritional regimes, liveweight and ADG are under genetic regulation (Cronjé and Boomker 2000).

Conclusions

Gender and dietary supplementation of prime lambs with canola or flaxseed oil did not affect the growth traits excluding ADG. Lamb breed significantly influenced lamb growth with WxC lambs eliciting the best growth performance. In conclusion, both canola and flaxseed oils can be effectively used in feedlot regimes in prime lamb industry. It is suggested that the suitable level of these oils should be 50 ml/kg pellet to achieve improved growth over a ten-week period.

References

- Annett, R., Carson, A., Dawson, L., Kilpatrick, D., 2011. Effects of dam breed and dietary source of n-3 PUFAs on the growth and carcass of lambs sourced from hill sheep flocks. *Animal* 5: 1023-1035
- Cronjé, P.B., Boomker, E.A., 2000. Ruminant physiology: digestion, metabolism, growth, and reproduction. (CABI publishing: Wallingford, UK)
- Enser, M., Hallett, K., Hewitt, B., Fursey, G., Wood, J., 1996. Fatty acid content and composition of English beef, lamb and pork at retail. *Meat Sci.* 42: 443-456

- Fogarty, N., Mulholland, J., 2014. Seasonal reproductive performance of crossbred ewes in intensive lamb-production systems. *Anim. Prod. Sci.* 54: 791-801
- Francisco, A., Dentinho, M., Alves, S., Portugal, P., Fernandes, F., Sengo, S., Jerónimo, E., Oliveira, M., Costa, P., Sequeira, A., 2015. Growth performance, carcass and meat quality of lambs supplemented with increasing levels of a tanniferous bush and vegetable oils. *Meat Sci.* 100: 275-282
- Gillingham, L.G., Gustafson, J.A., Han, S.-Y., Jassal, D.S., Jones, P.J., 2011. High-oleic rapeseed (canola) and flaxseed oils modulate serum lipids and inflammatory biomarkers in hypercholesterolaemic subjects. *Brit. J. Nutr.* 105: 417-427
- Hegarty, R., Shands, C., Marchant, R., Hopkins, D., Ball, A., Harden, S., 2006. Effects of available nutrition and sire breeding values for growth and muscling on the development of crossbred lambs. 1: Growth and carcass characteristics. *Crop Pasture Sci.* 57: 593-603
- Lewis, R.M., Emmans, G.C., Simm, G., 2006. Describing effects of genetic selection, nutrition, and their interplay in prime lambs using growth and efficiency functions. *Aust. J. Agr. Res.* 57: 707-719
- Malau-Aduli, A., McEvoy, P., Parsons, D., Lane, P., 2014. Breed variation in wool quality, growth and plasma metabolites of prime lamb fed degummed canola, *Proceedings of the 10th World Congress of Genetics Applied to Livestock Production*. Vancouver, Canada, 17-22 August 2014
- Manso, T., Bodas, R., Castro, T., Jimeno, V., Mantecon, A., 2009. Animal performance and fatty acid composition of lambs fed with different vegetable oils. *Meat Sci.* 83: 511-516
- Mozaffarian, D., Wu, J.H., 2011. Omega-3 fatty acids and cardiovascular disease Effects on risk factors, molecular pathways, and clinical events. *J. Am. Coll. Cardiol.* 58: 2047-2067
- SAS (2009) Statistical analysis system, version 9.2. Cary, USA

Vitamin E and C Effect on Meat of Muscovy Duck

Elly Tugiyanti ¹, Tri Yuwanta ², Zuprizal ² & Rusman ²

¹Faculty of Animal Science, University of Jenderal Soedirman, Purwokerto 53123, Indonesia

²Faculty of Animal Science, Gadjah Mada University, Fauna 3 street, Bulaksumur 55281 Yogyakarta – Indonesia

Abstract

This research objective was to study effect of vitamin E and C supplementation in feed on body weight, feed conversion, colour value (L*, a*, and b*) and Fe (Ferrum) level of Muscovy duck meat. The supplementation of vitamin E and C has not significantly affected on body weight and feed conversion of muscovy duck, a* and b* of Muscovy duck meat, but significantly affected on Fe level and L*.

Keyword : antioxidant, meat colour, performance

Corresponding author: tugiyanti.elly@gmail.com

Introduction

Muscovy duck meat is less favorable in society because it is dark-colored and less appetizing. Among factors influencing meat color are myoglobin level and status. Myoglobin molecules consist of globin protein molecules and heme cluster. Fe levels could serve as catalyst to accelerate oxidation in meat (Nelson and Cox, 2008). Muscovy duck meat contains unsaturated fatty acid, thereby prone to autooxydation which produces free radicals, then become peroxide which under decomposition will release aldehyde, alcohol and ketone compounds, each of which induces typical aroma in meat. Fat oxidation process can be inhibited by antioxidant, among which are the well-known vitamin E and C, the former is lipophilic and the latter is hydrophilic. Vitamin C can also regenerate radical vitamin E.

Material and Methods

Eighty four male Muscovy duck aged 9 weeks old were utilized in experimental research under Completely Randomized Design (CRD) with seven treatments namely basal feed (21% protein and 3100 kcal/kg feed metabolic energy) without vitamin E and vitamin C, basal feed + 400IU vitamin E, basal feed + 600IU vitamin E, basal feed + 400mg/kg feed vitamin C, basal feed + 600mg/kg feed vitamin C, basal feed + combined 200IU vitamin E + 200mg/kg feed vitamin C, basal feed + combined 300IU vitamin E+300mg/kg feed vitamin C, given from 10 weeks to 14 weeks old. Each treatments was subject to 4 replications, continued by Honestly Significant Difference Test (HSD) in case of difference among treatments.

Results and Discussion

Body weight

The feed supplemented with vitamin E and C did not significantly affect on body weight of 14-week-old Muscovy duck because vitamin E and C prevented free radicals formation and oxidative stress that affected body health, but could not increase body weight. Surai&Dvorska (2002) said that vitamin E and C are the primary antioxidant chain that prevents the damage of biological system in poultry. Vitamin E and C synergize, in which vitamin E prevents free radicals and emit the hydrogen ion so that vitamin E becomes radical. Then vitamin C regenerates the radical vitamin E into normal form and the body becomes healthier (Surai, 2003). As stated by Iqbal et al. (2004) and Nobakht et al. (2012) that vitamin E and C supplementation affects the other vitamins and nutrients, i.e. vitamin A. Body weight of 14-week-old Muscovy duck in this research was 2210.42 – 2358.46 g.

Feed conversion

The feed supplemented with vitamin E and C did not significantly affect on feed conversion of 14-week-old Muscovy duck because the supplementation did not affect feed intake and body weight. This result showed that antioxidant intake did not induce negative effect on Muscovy duck physiological function, and Muscovy duck can well convert feed into meat. Purba & Ketaren (2010) reported that supplementing vitamin E and santonin did not negatively affect feed conversion of Mojosari and Alabiocrossbredduck. Vitamin E serves more effectively as scavenger of free radicals to minimize cell dysfunction and effectively overcomes stress and essential for body immune(Kusnadi et al., 2006). Lacy & Vest (2004) argued that the primary factors influencing feed conversion were genetic, poultry condition, temperature, ventilation, sanitation, water quality, disease, medication, and management. Feed conversion in this research was 3.89 ± 0.11 – 4.15 ± 0.17 . Arifah et al. (2013) reported 3.03 ± 0.21 feed conversion of 10 week old muscovy duck.

Color value

The feed supplemented with vitamin E and C significantly affected ($P < 0.01$) on L^* (meat brightness) because vitamin C serves in extracellular fluid, water soluble and reactive with ferritin heme myoglobin. As oxygen scavenger, vitamin C keeps Fe^{2+} ion in reduction form (Raharjo, 2004), while vitamin E is fat-soluble, very effective in cell membrane, preventing lipid peroxide and maintaining color stability through receptor mediation of Low Density Lipoprotein(LDL) (Li et al., 2009; Gaytan et al., 2010). Feed supplemented with vitamin E and C did not significantly affect a^* value (reddish color) and b^* (yellowish color). Nelson & Cox (2008) and Winarsi (2011) observed chambers inside globulin molecule for four water molecules, and the polar cluster on the outer surface; therefore, myoglobin significantly determines meat color. L^* value of 14-week-old male Muscovy duck was 21.81 ± 1.67 – 50.92 ± 1.66 , a^* value was 8.14 ± 2.79 – 9.27 ± 1.93 , and b^* value was 6.83 ± 1.54 – 10.77 ± 3.24 .

Table 1. Colour value and Fe level of Muscovy duck meat.

	Basal feed (Bf)	Bf + vit. E 400 IU	Bf + vit. E 600 IU	Bf + vit. C 400 mg/kg	Bf + vit. C 600 mg/kg	Bf + vit. E 200 IU+ vit. C 200 mg /kg	Bf + vit. E 300 IU + vit C 300 mg /kg
L*	21,81 ^a ±1,67	27,12 ^b ± 2,85	31,82 ^b ± 2,43	36,94 ^c ± 1,31	44,84 ^d ± 3,17	47,23 ^e ± 0,84	50,92 ^e ± 1,66
a* ns)	8,14 ± 2,79	8,60 ±1,78	9,27 ± 1,93	9,06 ± 2,64	8,88 ± 2,31	8,35 ± 1,04	8,96 ± 0,53
b* ns	9,14 ± 2,26	8,67 ± 2,83	7,54 ± 0,17	7,12 ± 1,44	7,74 ± 1,65	7,60 ± 1,34	7,69 ± 1,77
Fe level (mg/100g)	1,81 ^c ± 0,09	1,80 ^c ± 0,09	1,64 ^c ± 0,08	1,23 ^a ± 0,11	1,25 ^a ± 0,07	1,45 ^b ± 0,06	1,54 ^{bc} ± 0,04

Ferrum (Fe) level

The feed supplemented with vitamin C and E significantly affected ($P < 0.01$) meat's Fe level of 14-week-old Muscovy duck. Fe level in this research was 1.23–1.80 mg/100g. HSD test showed that Fe level in feed without vitamin E and C supplementation was not different from that supplemented vitamin E 400IU and 600 IU, because vitamin E is effective to prevent cell membrane damage due to free radicals attack from oxidation (Khan et al., 2011). Fe levels in treatment supplemented with 400mg/kg or 600mg/kg vitamin C was similiarly each other. Similarly, Fe level in combined 200IU vitamin E and 200mg/kg vitamin C was not different from 300IU vitamin E and 300mg/kg vitamin C. Ridwan (2012) state that vitamin C contributed positive effect on iron status because it turned feri into fero. Meat would get darker as Fe level increased, as stated by Min et al. (2010). Russel et al. (2004) reported that thigh meat of Manila duck contained higher Fe than breast meat.

References

- Arifah, N., Ismoyowati, N. Iriyanti. 2013. Growth and feed conversion rate of various of local male duck (*Anas platyrhynchos*) and Manila male duck (*Cairrina Moschata*). Journal Ilmiah Peternakan 1(2): 718-725
- Gaytan, C.N , D. Shin , A. R. Sams , J. T. Keeton , R. K. Miller , S. B. Smith and M.X. Sanchez-Plata. 2010. Dietary lipid source and vitamin E effect on lipid oxidation stability of refrigerated fresh and cooked chicken meat. Poul. Sci. 89: 2726 – 2734
- Iqbal K., A. Khan and M.M.A.K. Khattak. 2004. Biological significance of ascorbic acid(vitamin C) in human health – A Review. Pakistan J. Nutr. 3 (1): 5 -13
- Khan, R.U., S. Naz, Z. Nikousefat, V. Tufarelli, M. Javdani, N. Rana and V. Laudadio. 2011. Effect of vitamin E in heat-stressed poultry. World's Poul. Sci.J. 67 : 469 – 478
- Kusnadi, E; R. Widjajakusuma, T. Sutardi, P.S. Hardjosworo and A. Habibie. 2006. Pemberian antanan (*Centella asiatica*) dan vitamin C sebagai upaya mengatasi efekcekaman panas pada broiler. Media Peternakan, 2006 : 133 – 140
- Lacy, M. and R. Vest, 2004. Improving feed conversion in Broiler: A Guide for grower. <http://www.agrocoat.nedfeedconversion.htm> (December, 22, 2012)

- Li, W.J., G.P. Zhao, J.L. Chen, M.Q. Zheng and J. Wen. 2009. Influence of dietary vitamin E supplementation on meat quality traits and gene expression related to lipid metabolism in the beijing-you chicken. *Brit. Poult. Sci.* 50(2): 188 -198
- Min, B., J.C. Cordray and D.U. Ahn. 2010. Effect of NaCl, myoglobin, Fe(II), and Fe(III) on lipid oxidation of raw and cooked chicken breast and beef loin. *J. Agric. Food Chem.* 58:600 – 605.
- Nelson, D.L. and M.M. Cox. 2008. *Lehninger: Principles of Biochemistry*. 5th ed.. W.H.Freeman And Company. USA.
- Nobakht, A. A. Ariyana and F. Mazlum. 2012. Effect of different levels of canola oil with vitamin E on performance and carcass traits of broilers. *Int. Res. J. Appl. Basic Sci.* Vol., 3 (5), 1059 -1064,
- Purba, M. dan P.P. Ketaren. 2010. Performa itik MA jantan umur enam minggu dengansuplementasi santoquin dan vitamin E dalam pakan. *Proseding Seminar NasionalTeknologi Peternakan dan Veteriner*: 696 - 706
- Raharjo, S. 2004. *Kerusakan Oksidatif Pada Makanan*. Pusat Studi Pangan dan Gizi.Universitas Gadjah Mada. Yogyakarta
- Ridwan, E. 2012. Kajian interaksi zat besi dengan zat gizi mikro lain dalam suplementasi. *Penel. Gizi Makan* 35(1): 49 – 54
- Russell, E.A., P.B. Lynch, K. O'Sullivan and J.P. Kerry1. 2004. Dietary supplementation of α -tocopheryl acetate on a-tocopherol levels in duck tissues and its influence on meatstorage stability. *Int. J. Food Sci. Tech.* 39: 331 – 340
- Surai, P.F. 2003. *Natural Antioxidants in Avian Nutrition and Reproduction*. Nottingham University Press. Thrumpton
- Surai, P.F. and J.E. Dvorska. 2002. Effect of selenium and vitamin E content of the breeder's diet on lipid peroxidation in breast muscles during storage. *Proceedings of Australian Poultry Science Symposium*, Sydney, pp. 187-192
- Winarsi, H. 2011. *Antioksidan Alami & Radikal Bebas. Potensi dan Aplikasinya dalam Kesehatan*. Cetakan ke-5. Penerbit Kanisius. Yogyakarta

***In vitro* nutrients digestibility and fermentation characteristics of king grass combined with concentrate containing mixed microbes**

Santoso B.*¹, M.N. Lekitoo¹, B. Tj. Hariadi¹, H. Abubakar² & T.W. Widayati³

¹Department of Animal Nutrition, ²Department of Biology, ³Department of Animal Production, University of Papua, Manokwari, West Papua, 98314, Indonesia

Abstract

The main feed for ruminants is forage which is composed by polysaccharides. Feed digestion in the rumen is done by microbes, thus type and population of microbes are important factor which affect the digestibility of nutrients. The objective of this study was to evaluate *in vitro* nutrient digestibility and fermentation characteristics of king grass combined with concentrate containing mixed microbes. *Lactobacillus plantarum*, one strain yeast of *Saccharomyces cerevisiae* and two strains of cellulolytic bacteria *i.e.* *Acinetobacter baumannii* and *Pseudomonas aeruginosa* was added in concentrate. The cellulolytic bacteria was isolated from rice straw waste and palm oil seeds waste, respectively. The concentrate was mainly formulated from agricultural and food industry wastes *i.e.* cassava waste, tofu waste and rice bran. The bacteria and yeast was added in concentrate at 10^6 - 10^7 cfu/g. The *in vitro* nutrient digestibility was conducted according to Tilley and Terry procedure using 250 mg of substrate comprised king grass and concentrate (70: 30, DM). The results showed the concentrate containing 7.2×10^6 cfu/g of *L. plantarum*, 3×10^8 cfu/g of *S. cerevisiae*, 8.6×10^7 cfu/g of *A. baumannii* and *P. aeruginosa*. The OM digestibility was higher ($P < 0.01$) in grass substrate with concentrate containing *L. plantarum* and *S. cerevisiae* than concentrate without addition of microbe. However, NDF digestibility was higher ($P < 0.01$) in grass substrate combined with concentrate containing mixed microbes. Addition of mixed microbes increased ($P < 0.05$) $\text{NH}_3\text{-N}$, acetic acid and total VFA concentration. It was concluded that addition of mixed microbes in concentrate improved fermentation activity and *in vitro* nutrient digestibility.

Keywords: by-products, concentrate, digestibility, rumen, cellulolytic

Corresponding author: santosob@lycos.com

Introduction

The main feed for ruminants is forages including grasses, legumes, browseable trees and fibrous crop byproduct. The fermentative digestion of fibre in the rumen is carried out by a mixture bacteria, protozoa and fungi, thus type and population of microbes are important factor which affect the digestibility of nutrients. King grass (*Pennisetum purpureophoides*) is a tropical grass that is often used as the main feed ruminants. This grass contains 14.8, 75.1 and 40.4%, respectively for crude protein, NDF and ADF (Santoso et al., 2013b). Seo et al. (2010) stated that microorganisms such as *Lactobacillus*, *Streptococcus* and *Enterococcus* are commonly used in probiotic for ruminants. Furthermore, *Saccharomyces cerevisiae* and *Aspergillus oryzae* are two primary fungal direct-fed microorganism (DFM) that have been supplemented to diet in ruminants. Newbold (1995) revealed that addition of *S. cerevisiae* in ruminant could improve animal

production through increasing mechanism of bacteria viability. Meanwhile, feeding combination of probiotics of buffaloes rumen microbes and catalyst supplement increased NDF digestion and proportion of acetate (Krisnan et al., 2009). In the previous *in vivo* study, most of researchers directly fed microorganism to the animal. However, this method is less efficient when it is applied to a number of animal. Therefore, the objective of the present study was to evaluate *in vitro* nutrient digestibility and fermentation characteristics of king grass combined with concentrate containing mixed microbes.

Materials and Methods

The concentrates were mainly formulated by agricultural and food industry by-products such as rice bran, tofu waste and cassava waste. *Lactobacillus plantarum* that isolated from *Pennisetum purpureophoides* was cultured using MRS broth at 30 °C for 48 h (Santoso et al. 2013a), meanwhile *S. cerevisiae* was cultured using malt extract broth at 30 °C for 48 h (Newbold, 1995). Two kind of cellulolytic bacteria *i.e.* *Pseudomonas aeruginosa* and *Acinetobacter baumannii* were isolated from seed of palm oil waste and rice straw, respectively and cultured in Carboxyl Methyl Cellulase at 29 °C for 48 h. The materials of concentrate were manually mixed by hand and then sprayed on top with culture of LAB, yeast and cellulolytic bacteria.

Five treatments used in this study namely (G) Grass; (G+A) grass + concentrate without microbes; (G+B) grass + concentrate containing *L. plantarum* and *S. cerevisiae*; (G+C) grass + concentrate containing *L. plantarum*, *S. cerevisiae* and *P. aeruginosa*; (G+D) grass + concentrate containing *L. plantarum*, *S. cerevisiae* and *A. baumannii*. The ratio of grass of and concentrate was 70: 30 on DM basis. The feeds were incubated in glass tube containing rumen liquor-buffer mixtures at 39 °C for 96 h (Tilley and Terry, 1963) for *in vitro* nutrients digestibility measurement. In separate experiment, the feeds were incubated in the syringes containing rumen liquor-buffer mixtures at 39 °C for 48 h (Menke and Steingass, 1988) for measurement pH, ammonia-N and volatile fatty acids (VFAs) concentrations.

Results and Discussion

Chemical composition of feeds

The chemical composition of king grass and concentrate are presented in Table 1.

Table 1. The chemical composition of grass and concentrates.

	G	Concentrates			
		A	B	C	D
Dry matter (%)	84.3	89.9	86.1	83.3	83.3
Organic matter (%)	91.8	94.3	93.8	94.7	94.1
Crude protein (%)	11.4	15.0	13.5	14.0	13.9
NDF (%)	79.3	61.7	61.4	61.9	59.4
ADF (%)	38.9	16.2	16.0	17.8	16.5
<i>L. plantarum</i> (cfu/g)	-	-	7.2×10 ⁶	-	-
<i>S. cerevisiae</i> (cfu/g)	-	-	3.0×10 ⁸	3.0×10 ⁸	3.0×10 ⁸
<i>P. aeruginosa</i> (cfu/g)	-	-	-	8.6×10 ⁷	-
<i>A. baumannii</i> (cfu/g)	-	-	-	-	1.9×10 ⁸

G: grass; A: concentrate without microbes; B: concentrate containing *L. plantarum* and *S. cerevisiae*; C: concentrate containing *L. plantarum*, *S. cerevisiae* and *P. aeruginosa*; D: concentrate containing *L. plantarum*, *S. cerevisiae* and *A. baumannii*

Grass used in the present study had 11.4% of CP, which is above to the threshold value of 7% as suggested by Minson and Milford (1966). Concentrates contained CP varied from 13.9 to 15%. The NDF and ADF contents in concentrates were similar, varied from 59.5 to 61.9% and 16.0 to 17.8%, respectively. The number of LAB, yeast and cellulolytic bacteria ranged from 7.2×10^6 to 3.0×10^8 cfu/g. The population of LAB and yeast in the concentrate was lower than in probiotic of 5×10^9 cfu/g as used by Lila et al. (2004).

Fermentation characteristics

The pH value in substrate R+C and R+D were lower ($P<0.01$) than control feed (Table 2). The lower pH value could be due to higher total VFA concentration as result of cellulolytic bacteria activity thus suppressed pH value. However, the pH value in all treatments ranged from 6.82 to 6.91, which is in the optimal range pH 6.7 ± 0.5 required to maintain normal cellulolysis (Van Soest, 1994). Concentration of $\text{NH}_3\text{-N}$ in substrate G+C and G+D was higher ($P<0.05$) compared to substrate G and G+B. Concentration of $\text{NH}_3\text{-N}$ ranged from 25.9 to 29.6 mg/100 ml, and were above the threshold value for both maximum fiber digestion as recommended by Abdulrazak et al. (1997). The proportion of acetic acid (C2) and total VFA concentration was higher ($P<0.05$) in concentrate with addition of cellulolytic bacteria (G+C and G+D) than other treatment feeds.

Table 2. *In vitro* fermentation characteristics in supernatant after 48 h of incubation.

	Treatments					SEM	P
	G	G + A	G + B	G + C	G + D		
pH	6.91 ^A	6.87 ^A	6.86 ^{AB}	6.83 ^B	6.82 ^B	0.01	0.01
$\text{NH}_3\text{-N}$ (mg/100 ml)	25.9 ^b	26.3 ^b	28.5 ^{ab}	29.6 ^a	29.5 ^a	0.82	0.02
C2 (mol/100 mol)	72.7 ^b	75.3 ^{ab}	80.8 ^{ab}	86.0 ^a	86.5 ^a	3.37	0.05
C3 (mol/100 mol)	29.3	32.0	30.7	32.6	36.4	1.83	0.14
C4 (mol/100 mol)	8.3	9.7	10.7	11.6	9.4	1.15	0.39
Total VFA (mM)	110.2 ^{ab}	117.1 ^{bc}	122.2 ^{ab}	134.0 ^a	128.6 ^{ab}	4.55	0.03

Mean values with different superscript letters ^{a-c} within the same row are significantly different ($P<0.05$)

Mean values with different superscript letters ^{A-B} within the same row are significantly different ($P<0.01$)

Nutrients digestibility

The IVDMD in substrate consisted of grass and concentrate was higher ($P<0.01$) compared to control feed (Table 3). Concentrate containing LAB and yeast had the highest ($P<0.01$) OM digestibility as compared to other treatments. Combination of LAB, yeast and cellulolytic bacteria in concentrates (G+B, G+C, G+D) enhanced ($P<0.01$) IVNDFD by 19.7% as compared to concentrate without mixed microbes. This result was comparable to study of Lila et al. (2004) that addition of *S. cerevisiae* increased *in vitro* DM degradability.

Table 3. *In vitro* digestibility (%) of dry matter, organic matter and neutral detergent fiber of feeds.

	Treatments					SEM	P
	G	G + A	G + B	G + C	G + D		
IVDMD	39.2 ^b	44.1 ^a	44.5 ^a	45.4 ^a	46.1 ^a	0.82	<0.01
IVOMD	49.4 ^c	53.8 ^{bc}	59.7 ^a	57.7 ^{ab}	58.2 ^{ab}	1.17	<0.01
IVNDFD	25.5 ^c	35.4 ^b	43.8 ^a	43.4 ^a	45.1 ^a	1.47	<0.01

Mean values with different superscript letters within the same row are significantly different ($P<0.01$)

In other study, Krisnan et al. (2009) concluded that addition of probiotic collected from buffalo rumen in catalytic supplement increased NDF digestibility in sheep. Moreover, Chaucheyras et al. (1995) noted that *S. cerevisiae* had ability to provide growth factors, such as organic acids or vitamins, thereby stimulating ruminal populations of cellulolytic bacteria.

Conclusions

Concentrate containing mixture microbes namely LAB, yeast and cellulolytic bacteria changed *in vitro* ruminal fermentation patterns by increasing NDF digestibility, total VFA and NH₃-N concentrations, and decreasing pH value.

References

- Abdulrazak, S. A., T. Fujihara, J. K. Ondiek and E. R. Ørskov, 2000. Nutritive evaluation of some Acacia tree leaves from Kenya. *Anim. Feed Sci. Technol.* 85:89-98.
- Chaucheyras, F. G. Fonty, G. Bertin, J. M. Salmon and P. Gouet, 1995. Effect of *Saccharomyces cerevisiae* (Levucell SC), a microbial additive for ruminants, on lactate metabolism *in vitro*. *Can. J. Microbiol.* 42:927-933.
- Krisnan, R., B. Haryanto dan K. G. Wiryawan, 2009. The effect of combined probiotics with catalyst supplementation on digestion and rumen characteristics in Priangan sheep Indon. *J. Anim.Vet. Sci.* 14 (4):262-269.
- Lila, Z. A., N. Mohammed, T. Yasui, Y. Kurokawa, S. Kanda and H. Itabashi, 2004. Effects of a twin strain of *Saccharomyces cerevisiae* live cells on mixed ruminal microorganism fermentation *in vitro*. *J. Anim. Sci.* 82:1847-1854.
- Menke, K. H. and H. Steingass, 1988. Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Anim. Res. Develop.* 28:7-55.
- Minson, D. J. and R. Milford. 1966. The energy values and nutritive value indices of *Digitaria decumbens*, *Shorghum alnum* and *Phaseolus atropurpureus*. *Aust. J. Agric. Res.* 17:411-423.
- Newbold, C. J., 1995. Probiotics for ruminants. *Ann. Zootech.* 45: 329-335.
- Santoso, B. A. Maunatin, B. Tj. Hariadi and H. Abubakar, 2013a. Isolation and identification of lactic acid bacteria originated from king grass (*Pennisetum purpureophoides*) as candidate of probiotic for livestock. *Indon. J. Anim.Vet. Sci.* 18(2):131-137.
- Santoso, B., E.W. Saragih and B. Tj. Hariadi, 2013b. Effect of water extract of plants containing tannin on *in vitro* methanogenesis and fermentation characteristics of the grass. *J. Indonesian Trop. Anim. Agric.* 38(1): 47-54.
- Seo, J. K., S. W. Kim, M. H. Kim, S. D. Upadhaya, D. K. Kam and J. K. Ha, 2010. Direct-fed microbials for ruminant animals. *Asian-Aust. J. Anim. Sci.* 23:1657-1667.
- Tilley, J. M. and R. A. Terry, 1963. A two stage technique for the *in vitro* digestion of forage crops. *J. Brit Grassl. Soc.* 18:104-111.
- Van Soest, P. J., 1994. *Nutritional Ecology of The Ruminant*. (2nd ed.). Comstock Publishing Associates a Division of Cornell University Press, Ithaca, NY, USA and London, UK. p. 476.

Periodic changes in chemical composition and *in vitro* digestibility of Gramineae feed resources in the Philippines

Hayashi¹, Y., E. Takeya¹, Y. Ikeno¹, H. Kumagai², C. Soriano³, E. M. Cruz⁴, N. P. Garcia⁵, D. L. Aquino⁵ & T. Fujihara⁵

¹Faculty of Agriculture, Meijo University, Kasugai, 486-0804, Japan, ²Graduate School of Agriculture, Kyoto University, Kyoto, 606-8502, Japan, ³Malaybalay Stock Farm, Department of Agriculture, Malaybalay, Bukidnon, 8700, Philippines, ⁴Small Ruminant Center, Central Luzon State University, Science City of Munoz, Nueva Ecija, 3120, Philippines, ⁵Philippine Carabao Center, Department of Agriculture, Science City of Munoz, Nueva Ecija, 3120, Philippines

Abstract

Many tropical grasses have already been studied for its agronomic characteristics and quality. However, reports on chemical composition and *in vitro* digestibility of Gramineae feed resources in the Philippines are meager. In addition, periodic changes in nutritive value of Gramineae feeds in the country are still obscure. Hence, the present study is conducted to identify the chemical composition and *in vitro* digestibility of Gramineae feed resources in four different periods divided by the temperature and rainfall in the Philippines. Nueva Ecija and Bukidnon Provinces in the Philippines were selected for the collection of Gramineae feed resources in the hot-wet-period (HWP), the cool-wet-period (CWP), the cool-dry-period (CDP) and the hot-dry-period (HDP). The chemical composition and *in vitro* digestibility of the resources were analyzed. The digestibility of dry matter (DMD), organic matter (OMD) and neutral detergent fiber (NDFD) was calculated. Fourteen feed resources were collected in each period. The most resources tended to have lower dry matter (DM) contents in HWP and CWP than in CDP and HDP. The crude protein (CP) contents of most resources tended to show the lowest value in CDP or HDP. The CP concentrations of *Brachiaria brizantha*, *Setaria incrassata* and *Coix lacryma-jobi* tended to be higher than those of the other resources. The DMD of *Brachiaria brizantha*, *Paspalum atratum* and *Pennisetum purpureum* in the four periods assumed to be stable and higher than that of the other resources. The most resources tended to show the highest DMD, OMD and NDFD in HWP or CWP. Although the digestibility of the most resources was over 30%, the digestibility of *Imperata cylindrica* was below 25%. The periods divided by the temperature and rainfall caused the variance of chemical composition and digestibility.

Keywords: chemical composition, feed, Gramineae, in vitro digestibility, Philippines

Introduction

The production of milk and meat in tropical Asian countries is continuously increasing. Efficiency of utilization of available feed resources is one of the critical issues in livestock productions; however, the scarce supply and insufficient quality of feed materials in the countries are still constraints due to the water deficiency in dry season and the unstable feed productivity. On the

other hand, there are many herbaceous species as ruminant feed resources in the tropical area. The Gramineae form a very large family and can be utilized as main feed resources for ruminants. Thus, the identification of periodic changes in chemical composition and digestibility of Gramineae feed resources is required.

Materials and Methods

Nueva Ecija and Bukidnon Provinces located in Luzon and Mindanao Islands, respectively, in the Philippines were selected for the collection of Gramineae feed resources in July, 2013 during the HWP, November, 2013 during the CWP, February, 2014 during the CDP and May, 2014 during the HDP. The concentrations of DM, OM, CP and neutral detergent fiber (NDF) of the resource samples were analyzed. *In vitro* digestibility was measured using the solution of artificial saliva and rumen fluid orally collected from four Japanese Saanen goats. The DMD, OMD and NDFD were calculated after the digestion at 38° C for 48 hours in an anaerobic condition.

Results and Discussion

Twelve species of feed resources were collected in each period. Most of the feed resources tended to have lower DM in HWP and CWP than in CDP and HDP (Figure 1 (A)). On the other hand, the CP contents of most feed resources tended to show lowest values during CDP or HDP (Figure 1 (B)). The lower contents of CP in dry season than in rainy season were in agreement with the results for grasses in West Sumatra, Indonesia (Evitayani et al., 2004). The grasses might have matured in CDP or HDP and decreased CP content in these periods. However, some grass species such as *Brachiaria brizantha*, *Setaria incrassate* and *Paspalom atratum* showed the maximum CP contents in HDP. The CP concentrations of *Brachiaria brizantha*, *Setaria incrassate* and *Coix lacryma-jobi* tended to be higher than those of the other resources (10.5~25.7%, 9.2~24.2% and 13.3~15.8%, respectively). There were wide variables or changes in OM and NDF of the feed resources during the four periods of evaluation. On the average, the DMD of *Brachiaria brizantha*, *Paspalom atratum* and *Pennisetum purpureum* in the four periods assumed to be stable and higher than that of the other resources with 45.9~51.4%, 43.3~50.1% and 47.4~53.7%, respectively (Figure 1(C)). Majority of the feed resources tended to show the highest DMD, OMD and NDFD in HWP and CWP (Figure 1 (C-E)). Nasrullah et al. (2003) reported that the higher DMD in rainy season compared to dry season in South Sulawesi, Indonesia of which this report supported the results of the present study. The digestibility of the most resources was over 30% while the digestibility of *Imperata cylindrical* was below 25%.

Although the positive correlations between the CP contents and the digestibility were identified (DMD: $r=0.43$, OMD: $r=0.57$ and NDFD: $r=0.36$, $P<0.01$), the negative correlations between the NDF contents and the digestibility was shown (DMD: $r=-0.51$, OMD: $r=-0.61$ and NDFD: $r=-0.30$, $P<0.05$).

The periods divided by the temperature and rainfall caused the variance of chemical composition and digestibility. The appropriate choice of better nutritious and digestible feed resource in each period can contribute to the efficient livestock production in tropical Asia.

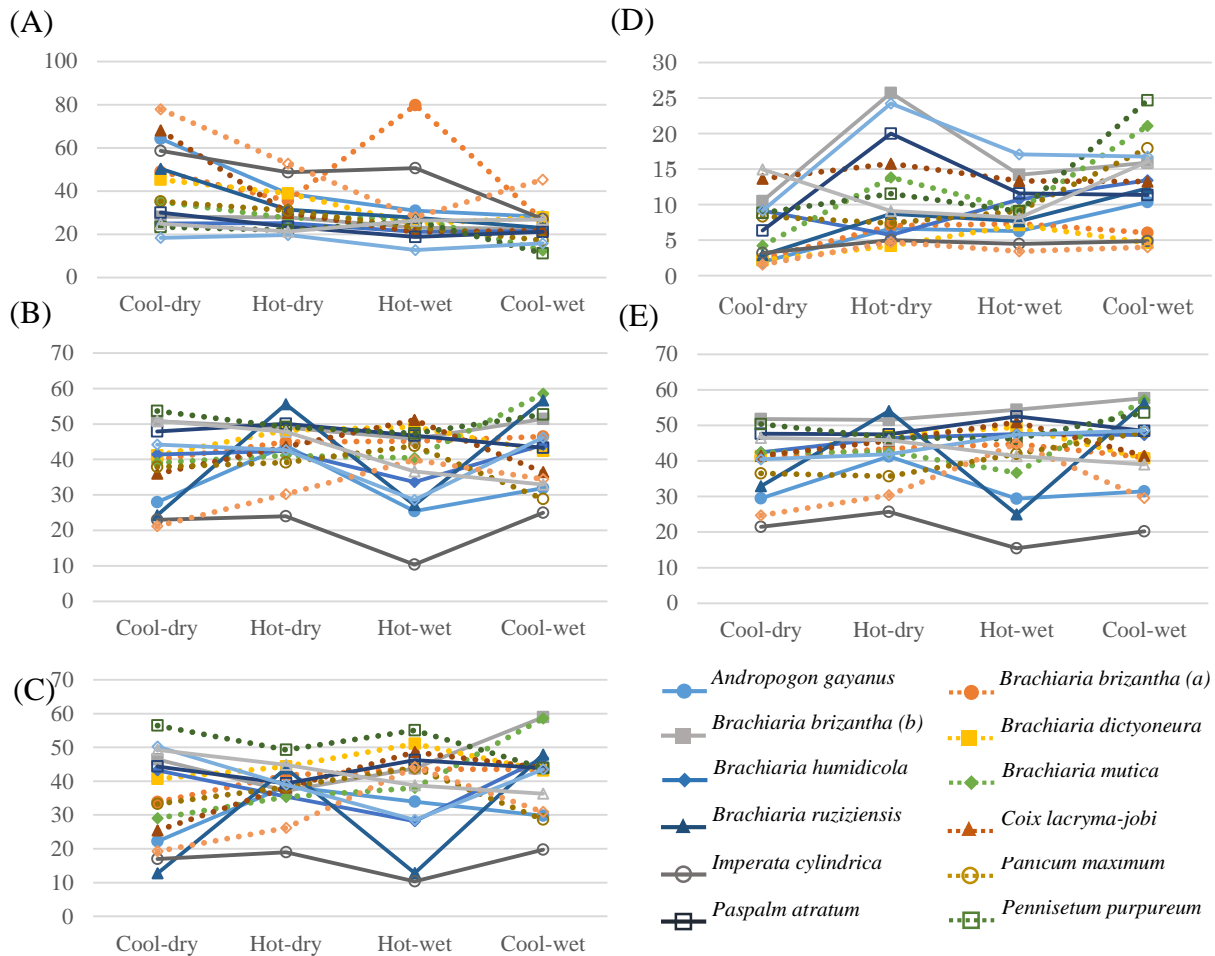


Figure 1. Changes of chemical composition and *in vitro* digestibility of Gramineae feed resources in the Philippines (%). (A) dry matter contents, (B) crude protein contents, (C) dry matter digestibility, (D) organic matter digestibility, (E) neutral detergent fiber digestibility

References

- Evitayani, L. Warly, A. Fariani, T. Ichinohe and T. Fujihara, 2004. Seasonal changes in nutritive value of some grass species in West Sumatra, Indonesia. *Asian-Aust. J. Anim. Sci.* 17: 1663-1668
- Nasrullah M., R. Niimi, R. Akashi and O. Kawamura, 2003. Nutritive evaluation of forage plants in South Sulawesi, Indonesia. *Asian-Aust. J. Anim. Sci.* 16: 693-701

Chemical composition of forages fed to stalled goats in smallholder farms in Mauritius

Saddul^{*1}, D., R.K., Ramnauth, G. Saraye, B. Ramlugun & J. Ramtohol

¹Livestock Research Dept., Food and Agricultural Research & Extension Institute, Reduit, Republic of Mauritius

Abstract

A survey was conducted on the types of forages fed to stalled goats on smallholder farms in two agroclimatic zones (humid and subhumid) during the wet and dry seasons in Mauritius. Forages were grouped as trees/shrubs, grasses, climbers and other herbaceous plants including sugarcane tops. Trees forages were used on all farms and included *Leucaena leucocephala* (Acacia) and *Litsea glutinosa* (Ti feuilles), *Albizia lebbek* (Bois Noir), *Melia azedarach* (Lilas), *Santalum album* (Chandan) and *Litsea monopelata* (Gros feuilles). Some farms used tree species as sole diet while most farms use it in combination with grasses, climbers and other herbaceous species. There was wide variation in the chemical composition of the forages, namely, crude protein (CP) 3.2 to 34.5%, ash 4.2 to 12.8%, acid detergent fibre (ADF) 23.7 to 66.2%, neutral detergent fiber (NDF) 45.0 to 88.6% and lignin (ADL) 1.6 to 44.1%. The average CP content of climbers, grasses, trees and other herbaceous plants was 15.7, 9.2, 16.7 and 13.2%, respectively. The total phenolics (TP), total tannins (TT) and condensed tannins (CT) contents ranged from 0.57 to 12.8, 0.27 to 10 and 0.02 to 2.5 %, respectively. Highest TP content (12.8%) was found in *Syzygium cuminis* (Jamblon) while highest TT and CT were found in *Schinus terebenthifolius* (Poivrier marron) (10.8% and 2.5%, respectively) and Jamblon (10.0% and 0.93%, respectively). Tree species were categorized according to phenolic content: low to negligible levels (<1%) (Lilas), intermediate level (1-5%) (Acacia, Bois Noir, Ti feuilles and Gros Feuilles), high levels (>5%) (Poivrier marron and Jamblon). Trees like Acacia, Lilas, Bois Noir, Ti Feuilles and Gros Feuilles are available throughout the year and are good sources of protein that can be exploited as alternatives to partly replace concentrates and improve productivity of goats.

Key words: smallholder farms, goats, forages, tannins, crude protein

**Corresponding author: dsaddul@yahoo.com*

Introduction

Smallholder goat farmers in Mauritius generally collect forages from road sides, river sides and marginal lands and choose species that are relished by goats without awareness of their nutritional worth. In general, trees and shrubs constitute a major component of the forage diet under the cut and carry system, typical of traditional practices and only few farms use commercial concentrates as supplement. The low carcass weight of goats under the traditional systems is an indication of their low performance due probably to inadequate nutrition and remedial measures require knowledge of nutritional worth of forages offered to the goats. This study aimed at determining the types and nutritive values of fodder that were being fed to goats.

Materials and Methods

Mauritius (20°10'S, 57°31'E) has a sub-tropical climate throughout the year, with two distinct seasons, namely, a wet summer (November to April) and a dry winter (May to October). Forages were collected from 17 goat farms during the wet and dry seasons located in the humid (1250-3000mm rain) and sub humid zone (<1250mm rain) where goat rearing is concentrated. The forages were sorted over a 12-month period and identified according to their common name. Samples of parts normally eaten by goats were taken and dried at 52°C (Makkar, 2003) and ground to 1mm and 0.5mm for proximate analysis (AOAC, 1990; Van Soest et al. 1991) and TP, TT, CT (Makkar, 2003). Data were analysed using descriptive statistics of SPSS 16.0 (SPSS, 1992).

Results and Discussion

Forages fed to goats consisted of a combination of native grasses, climbers, leguminous and non-leguminous trees and shrubs, and other herbaceous plants totaling to 41 species in vegetative, flowering and seed production stages. Trees and shrubs were collected throughout the year and form the bulk of the diet on all farms. Climbers and grasses were used on some farms throughout the year, in addition to forages from trees. On other farms, climbers and grasses were fed during the dry season only when trees and shrub were less abundant. Sugarcane tops, muguet (*Convallaria* spp), herbe watoe (*Tristema mauritianum*), brede malbar (*Amaranthus*) and herbe siflette (*Paspalidium geminatum*) were used during the dry season on some farms in humid regions to ensure that the goats were fed sufficient roughage.

Among the trees, Ti Feuilles, Acacia and Poivrier marron were used on all farms throughout the year, indicative of their island-wide non-seasonal availability while Gros Feuilles were mostly available in the humid region. Bois Noir, Chandan, and Lilas were available throughout the year in the subhumid region, while twigs and leaves from fruit trees such as Jamblon, Jackfruit (*Artocarpus heterophyllus*), Prune (*Prunus* sp.) and Bilimbi (*Averrhoa bilimbi*) were used during the dry season. Lilas was also used for its antihelmintic properties as reported by Orwa et al. (2009).

Table 1 shows the average proximate composition of forages consumed by goats. Average values ranged as follows: DM 10.9 to 54.0%, CP 3.2 to 34.5%, ash 4.2 to 12.8%, Phosphorus 0.16 to 1.5%, Ca 0.06 to 4%, ADF 23.7 to 66.2%, NDF 45.0 to 88.6% and ADL 1.6 to 44.1%. Trees and climbers had high CP content compared to native grasses which had high NDF and lignin content. High CP content was recorded for trees (Acacia and Bois Noir: 22.9%) and climbers (20.4%), while among grasses, Herbe sikkin (*Bothriochloa pertusa*) and Herbe d'argent (*Ischaemum aristatum*) had lowest CP (4.4 and 4.9%, respectively).

Table 1. Mean values for chemical composition (% DM) of forages offered to goats.

Forage type	DM	CP	Ash	ADF	NDF	ADL
Trees/Shrubs	35.8±0.96	16.7±0.5	8.3±0.2	43.8±0.8	63.6±0.74	17.3±0.7
Grasses	28.8±0.99	9.2±0.6	10.5±0.3	45.9±1.2	79.5±0.88	7.4±1.1
Climbers	21.1±0.96	15.1±0.7	10.5±0.3	41.1±1.3	58.9±1.1	11.6±1.1
Herbaceous spp.	28.4±1.4	13.2±1.1	10.9±0.5	35.2±1.9	57.5±1.2	8.3±1.7

The contents of TP, TT and CT in tree forages ranged from 0.57 to 12.8, 0.27 to 10 and 0.02 to 2.5%, respectively (Table 2). *Cordia macrostachya* (Herbe conde), Bilimbi and Lilas had negligible amounts of TP, TT and CT while Jamblon and Poivrier marron had TP and TT contents exceeding 5%, but with CT less than 2.5%. Acacia, Bois noir, Ti Feuilles and Gros Feuilles formed the bulk of the forage diet of goats on most farms and is the sole forage source on some farms without any adverse effects on the animals as the CT content is below the threshold value of 5%.

Table 2. Total phenolics (TP), total tannins (TT) and condensed tannin (CT) (% DM) of tree species.

Common name	Species	TP	TT	CT
Condé	<i>Cordia macrostachya</i>	0.57	0.27	-
Bilimbi Leaves	<i>Averrhoa bilimbi</i>	0.64	0.22	0.02
Lilas	<i>Melia azedarach</i>	0.67	0.17	0.03
Condé	<i>Cordia macrostachya</i>	1.00	0.29	0.03
Coqueluche	<i>Pongamia globra</i>	1.00	0.29	0.03
Prune	<i>Prunus sp.</i>	1.20	0.61	0.14
Chandan	<i>Santalum album</i>	1.30	0.22	0.91
Feuille lacol	<i>Cordia sp</i>	1.30	0.25	0.01
Ti feuilles	<i>Litsea glutinosa</i>	1.90	0.75	0.82
Bois Noir	<i>Albizia lebbek</i>	3.00	1.80	0.23
Acacia	<i>Leucaena leucocephala</i>	4.20	1.60	1.10
Poivre marron	<i>Schinus terebenthifolius</i>	10.50	10.50	2.50
Jamblon	<i>Syzygium cuminis</i>	12.80	10.00	0.93
Mean		3.3	1.9	0.88
SEM		0.48	0.06	0.76

SEM: Standard error of the means

Conclusions and recommendations

Farmers fed their goats with a mixture of different species of that actually provided a mix of approximately 14% CP which is acceptable if fed in adequate amounts. Locally available trees and climbers are good sources of protein. Trees are readily available throughout the year unlike climbers and are promising species that are comparable to commercially available concentrates.

Acknowledgements

Funding from The International Foundation for Science, contribution of institutions for chemical analyses and identification of fodder and the collaboration of farmers are gratefully acknowledged.

References

- AOAC, 1990. Official Methods of Analysis of the Association of Official Analytical Chemists. 15th ed. Washington, DC, USA. 30 pp.
- Makkar, H.P.S. 2003. Quantification of tannins in tree and shrub foliage: A laboratory manual. Kluwer Academic Publishers.
- Van Soest, P.J., Robertson, J.B. and Lewis, B.A. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74: 3583-3597.
- Orwa, C., A. Mutua, Kindt, R., Jamnadass R. and S. Anthony. 2009. Agroforestry Database: a tree reference and selection guide version 4.0 (<http://www.worldagroforestry.org/sites/treedbs/treedatabases>)
- SPSS Inc. Released 2007. SPSS for Windows, Version 16.0. Chicago, SPSS Inc.

Feed intake and serum metabolite of goats fed crude glycerin from waste vegetable oil

Chanjula, P¹, S. Pongprayoon¹ & S. kongpan¹

¹Department of Animal Science, Faculty of Natural Resources, Prince of Songkla University, 90110, Thailand

Abstract

This study aimed to evaluate the effects of crude glycerin from waste vegetable oil (CGWVO) supplementation on feed intake, blood metabolites, and hormone concentrations of goats. Four-Thai Native x Anglo Nubian crossbred growing male goats with an average body weight of 31.5±1.9 kg were randomly assigned according to a 4x4 Latin square design with four consecutive 21-d periods. Treatment diets contained 0, 2, 4, and 6% of dietary DM of CGWVO. Goats were fed unlimited amounts (*ad libitum*) as a total mixed ration (TMR). Based on this experiment, there was no significant difference ($p>0.05$) among treatment groups regarding daily DMI (total DMI, % BW, and g/kg BW^{0.75}), except DMI of goat fed 6% of CGWVO in the diets which was the lowest ($p<0.05$) as compared with other treatments. The blood glucose, BHBA and packed cell volume (PCV) were similar among treatments ($p>0.05$), whereas plasma insulin was significantly ($p<0.05$) higher as higher levels of CGWVO were incorporated into diets. The data suggest that CGWVO (63.42% of glycerol, 4.38% methanol, and 47.78% of crude fat) may be used in diets of goats with concentrations up to 4% without negative effects on feed intake and blood.

Keywords: By-product, crude glycerin, waste vegetable oil, serum metabolite, goat.

Introduction

Recent increases in biofuel production have led to increasing prices of traditional feedstuffs that make livestock producers search for alternative feeds to lower production costs without sacrificing animal performance. Recent efforts have evaluated the effects of the inclusion of crude glycerin (above 86% of glycerol and <0.64% of methanol) in diets on intake, digestibility, performance, carcass, and meat quality traits of sheep with reporting of acceptable inclusion of 21% and 15% respectively on diet dry matter (Gunn et al., 2010; Avila-Stagno et al., 2013). However, a limit number of studies have evaluated the effects of crude glycerin from waste vegetable oil (CGWVO) contaminated with high crude fat and methanol contents in diets fed to goats. This study hypothesized that CGWVO containing 63.42% of glycerol, 47.78% of crude fat, and 4.38% of methanol in dry matter (DM) basis may be used as an energy source in diet of goats at concentrations up to 6% on DM basis without compromising feed intake and serum metabolite. Thus, the objectives of this study were to evaluate the effects of CGWVO (containing 63.42% of glycerol, 47.78% of crude fat, and 4.38% of methanol in DM basis) on feed intake, blood metabolites, and hormone concentrations in goats.

Materials and Methods

Four male crossbred (Thai Native x Anglo Nubian) goats, about 18 months old and 31.5 ± 1.9 kg body weight, were randomly assigned according to a 4x4 Latin square design to investigate the effects of CGWVO on feed intake and blood metabolites. The 4 corn-based dietary treatments consisted of 0, 2, 4, and 6% of CGWVO (DM basis) were formulated to be isonitrogenous and isocaloric to meet or exceed the requirements of growing goats. The CGWVO used in this study originated from waste vegetable oils of palm, soybean, rice bran and sunflower. CGWVO was produced by methylic route contained 63.42% of glycerin, 23.93% of water, 0.47% of sodium, 47.78% of crude fat, and 4.38% of methanol obtained from Specialized Research and Development Center for Alternative Energy from Palm Oil and Oil Crop, Faculty of Engineering, Prince of Songkla University, Thailand. CGWVO from single batch was added to the total mixed ration (TMR) as liquid.

All goats were kept individually in pens (0.50x1.20m) under well-ventilated sheds where water and mineral salt were available at all time. The experiment was conducted for 4 periods, and each period lasted for 21 days. During the first 14 d of each period, all animals were fed by respective diets for *ad libitum* intake, whereas during the last 7 d, the animals were moved to metabolism crates for total collection during the time goats with restriction to 90% of the previous voluntary feed intake to ensure total feed intake. Feeds were provided twice times in two equal portions daily at 0800 and 1600 h. For determination of daily DMI, refusals were collected and weighed daily before feeding. Feed samples obtained each time were oven dried at 60°C for 72 h and grounded to pass through a 1-mm sieve, and composited by period on an equal weight basis, and analyzed for DM, ether extract, ash, CP content (AOAC, 1995). Goats were individually weighed before the morning feeding at the beginning and end of each experimental period. At the end of each period, blood samples (about 10 mL) were collected from a jugular vein into tubes containing 12 mg of EDTA. Plasma was separated by centrifugation at $2500 \times g$ for 15 min at 5 °C and stored at -20 °C until analysis. Plasma glucose, insulin, BHBA, and packed cell volume (PCV) were measured by using commercial kits (No. 640, Sigma Chemical Co., St. Louis, USA). All data were subjected to the analysis of variance using Proc. GLM and treatment means were compared using Duncan's New Multiple Range Test.

Results and Discussion

The results showed that (Table 1) overall mean feed intakes (total DMI, %BW, and g/kgW^{0.75}) were similar for all treatments ($p > 0.05$), except DMI of goat fed 6% of CGWVO in the diets which was the lowest ($p < 0.05$) as compared with other treatments. The results of this study were in agreement with a study by Lage et al. (2014) reported DM intakes particularly decreased when fed diets containing different contents of crude glycerin contaminated with high concentrations of crude fat and methanol (up to 12% of DM) to finishing lambs. In the present study, CGWVO had 63.42% of glycerin, 47.78% of crude fat, and 4.38% of methanol. The greater concentration of lipid in diets of animals fed diets with the higher concentration of CGWVO was likely to be the main factor that contributed for a reduction of dry matter intake. Ruminant animals are relatively intolerant to high concentrations of fat and feed intake usually decrease as fat content of the diets

exceeds 6% DM basis (Palmquist and Jenkins, 1980). Thus, the association of CGWVO with higher content of crude fat in diets decreased the DMI by the animals.

Table 1. Effects of 0, 2, 4, and 6% dietary CGWVO on feed intake of goats.

Item	Dietary CGWVO, %				SEM
	0	2	4	6	
Total DMI, kg/d	1.107 ^a	1.167 ^a	1.146 ^a	1.008 ^b	0.02
DMI, %BW	3.12 ^a	3.27 ^a	3.25 ^a	2.80 ^b	0.05
DMI, g/kg W ^{0.75}	76.05 ^a	79.79 ^a	75.22 ^a	68.74 ^b	1.21
BW change, kg/d	0.135	0.130	0.142	0.105	0.02
BW change, %	8.27	7.80	8.63	5.95	1.61

^{a-b}Means within rows followed with different superscript letters are statistically different (p<0.05).

SEM = Standard error of the mean (n = 4).

No significance (p>0.05) of CGWVO inclusion was detected for blood glucose, BHBA, and PCV (Table 2), and all were within the normal range of 50-75 mg/dL and 22-38 mg/dL, respectively (Jain, 1993). The plasma insulin was lower for the diet 0% of CGWVO than for the diets 4% of CGWVO, while the difference between the diets 2%, 4% and 6% of GC were not significant. Our results showed that goats fed diets containing glycerin (4%) had higher DMI and BW change, a finding that was associated to a tendency for higher insulin levels. However, it remains unclear whether this was due to the dietary treatment. However, insulin concentrations were not correlated with glucose concentrations. Circulating insulin concentrations usually corresponds to change in circulating glucose concentrations (Jenny and Polan, 1975). However, insulin secretion is a result of many factors, and some case has been shown to have a low correlation with blood glucose concentrations (McAtee and Trenkle, 1971). Although insulin secretion responds to circulating glucose concentrations, a lag between increased concentrations of glucose and insulin is often reported (Lake et al., 2006), whereas Gunn et al. (2010) reported that insulin concentrations also increased linearly relative to time of sampling around feeding (P<0.001). Based on the experimental data, substituting corn grains with CGWVO (63.42% of glycerin, 47.78% of crude fat, and 4.38% of methanol.) up to 4% of DM in the diets of goats had no effect on feed intake and blood metabolites. It could be effectively used as an alternative energy source to substitute for cereals in the diets. Thus, in the case of a competitive price, CGWVO may be effectively used as a partial energy source in the diets of goats.

Table 2. Effects of 0, 2, 4, and 6% dietary CGWVO on blood metabolites in goats.

Item	Dietary CGWVO, %				SEM
	0	2	4	6	
Glucose, mg/dL	75.37	73.97	77.72	79.38	3.78
Insulin, μ U/mL	2.51 ^b	6.14 ^{ab}	8.10 ^a	5.94 ^{ab}	1.25
BHBA, mg/dL	4.60	5.06	4.66	4.73	0.42
PCV, %	32.12	31.13	30.62	32.37	0.58

^{a-b} Means within rows followed with different superscript letters are statistically different (p<0.05).

SEM = Standard error of the mean (n = 4).

References

- AOAC. 1995. Official Methods of Analysis. 16th ed. Assoc. Off. Anal. Chem., Arlington, VA.
- Avila-Stagno, J., A. V. Chaves, M. L. He, O. M. Harstad, K. A. Beauchemin, S. M. McGinn and T. A. McAllister. 2013. Effects of increasing concentrations of glycerol in concentrate diets on nutrient digestibility, methane emissions, growth, fatty acid profiles, and carcass traits of lambs. *J. Anim. Sci.* 91:829-837.
- Gunn, P. J., A. F. Schultz, M. L. Van Emon, M. K. Neary, R. P. Lemenager, C. P. Rusk and S. L. Lake. 2010. Effects of elevated crude glycerin concentrations on feedlot performance, carcass characteristics, and serum metabolite and hormone concentrations in finishing ewe and wether lambs. *Prof. Anim. Sci.* 26:298-306.
- Jain, N. C. 1993. *Essential of Veterinary Hematology*. Lea & Febiger. Philadelphia.
- Jenny, B. F. and C. E. Polan. 1975. Postprandial blood glucose and insulin in cows fed high grain. *J. Dairy Sci.* 58:512-514.
- Lage, J. F., P. V. R. Paulino, L. G. R. Pereira, M. S. Duarte, S. C. Valadares Filho, A. S. Oliveira, N. K. P. Souza and J. C. M. Lima. 2014. Carcass characteristics of feedlot lambs fed crude glycerin contaminated with high concentrations of crude fat. *Meat Sci.* 96:108-113
- Lake, S. L., E. J. Scholljegerdes, D. M. Hallford, G. E. Moss, D. C. Rule and B. W. Hess. 2006. Effects of body condition score at parturition and postpartum supplemental fat on metabolite and hormone concentrations of beef cows and their suckling calves. *J. Anim. Sci.* 84:1038-1047.
- McAtee, J. W. and A. Trenkle. 1971. Metabolic regulation of plasma insulin levels in cattle. *J. Anim. Sci.* 33:438-442.
- Palmquist, D. L. and T. C. Jenkins. 1980. Fat in lactation rations: Review. *J. Dairy Sci.* 63:1-14.

Nutritive value of oil palm fronds treated with white rot fungi

Chanjula, P.^{1*}, V. Petcharat², & C. Promkor³

¹Department of Animal Science, Faculty of Natural Resources, Prince of Songkla University, Hat Yai Campus 90110, Thailand, ²Department of Pest Management, Faculty of Natural Resources, Prince of Songkla University, Hat Yai Campus 90110, Thailand, ³Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology Isan, Sakon nakhon Campus, 47160, Sakon nakhon, Thailand

Abstract

The effects of white rot fungi (WRF) treated biologically on chemical composition of oil palm fronds (OPF) were investigated using a completely randomized design with a 6x3 factorial arrangement consisting of eighteen treatments and four replicates. Fungal treatments decreased ($p < 0.05$) the amount of neutral detergent fiber (NDF) and acid detergent fiber (ADF) but increased ($p < 0.05$) crude protein (CP) content of OPF. As a result of this experiment, treatment of OPF with six WRF particularly *Lentinus sajor-caju* (LSc) and *Schizophyllum commune* (SC) resulted in reducing of its cell wall components and increasing of CP and adding a high amount of urea (0.5-1%) intended to increase CP in OPF.

Keywords: Fungal treatment, oil palm fronds, nutritive value, white rot fungi

Introduction

Oil palm, *Elaeis guinensis*, is an important crop of Thailand and other tropical countries. The production of palm oil has expanded rapidly, associated with production of a variety of agricultural co-products. Among those are leaves and petioles from oil palm trees, also called oil palm fronds (OPF), are an important co-product. In particular, OPF has been given emphasis lately as it has great potential to be utilized as a roughage source or as a component in complete feed for ruminants. However, the utilization of these materials as a feed source for ruminants is limited for their complex biological structure and low protein content (Ishida and Abu Hassan, 1997). This is consistent with low OPF digestibility in ruminant. Also, OPF have a low energy value, varying between 4.9 and 5.6 MJ metabolisable energy (ME)/kg DM (Zahari and Alimon, 2005). To degrade the lignocellulosic bonds and increase bioavailability of nutrients, various physical and chemical delignification methods have been examined in agricultural co-products such as rice and wheat straw (Hamed and Elimam, 2010). Although these methods have advantages, they are costly, low in effectiveness, not environmentally friendly and also require application of technology. One of the promising techniques is colonization with white rot fungi (WRF) is considered to be a promising technique because of its preferential degradation of lignin (Okano et al., 2005). Due to the existence of many species and strains of WRF in nature and their possible different effects on the nutritive value of the substrates, there is an increased research interest on the characteristics of the species and strains including the ability of their growth on the OPF and

their effects the nutritive value of the OPF. This study was conducted to assess the effect of six WRF for their ability and selectivity to degrade lignin and their effect on chemical composition.

Materials and Methods

Preparation of the white rot fungi inoculum: Stock culture of six WRF (*Pleurotus ostreatus*, PO; *Pleurotus djamor*, PD; *Lentinus squarrosulus*, LS; *Schizophyllum commune*, SC; *Lentinus polychrous*, LP; *Lentinus sajor-caju*, LSc) was provided by the Division of Applied Microbiology, Department of Agriculture, Ministry of Agriculture and Cooperative, Thailand and kept at 4 °C. Before being used, each WRF was separately grown in Petri dishes containing 25 ml potato dextrose agar (200 g potato, 20 g dextrose and 12 g agar) at 25 °C for 5-7 days. Then each WRF was transferred into sterile sorghum seeds incubated for 7-10 days, and it was used as inoculum.

Treatment of oil palm fronds (OPF): This study was based on randomly assigned OPF to a completely randomized design with a 6x3 factorial arrangement to evaluate six white rot fungi (WRF) (coded: PO, PD, LS, SC, LP, and LSc) with three levels of urea (0, 0.5, and 1% DM basis). Fresh OPF were chopped into 1–2 cm lengths, packed in cotton bags (30x45 cm), and then autoclaved twice at 120 °C for 30 min with cooling between cycles. The autoclaved OPF were inoculated with spawns (at a rate of 3-4% w/w) of six WRF cultures with three levels of urea and repacked in plastic bags (17.8x30 cm). Before inoculating with the WRF, the moisture content of OPF was adjusted to 600 g/kg by adding 45 ml of distilled water to 40-45 g OPF directly in each inoculation recipient. The chemical composition of fresh OPF is presented in Table 1. The bags incubated in the fermentation chamber where temperature was automatically adjusted to 25°C, and relative humidity was kept at 78±5% by spraying water.

Chemical analyses: After 21 days of incubation, the OPF were removed from the fermentation chamber and allowed to dry at 60 °C for 96 h. Samples were weighed after drying to determine the DM. Sub-samples (about 20 g) were taken from each experimental unit, milled through 1 mm sieve, and analyzed for DM, ash, CP content (AOAC, 1995), and NDF and ADF (Van Soest et al., 1991). All data were subjected to the analysis of variance using Proc. GLM and treatment means were compared by using Duncan's New Multiple Range Test.

Results and Discussion

Average chemical composition of fresh oil palm frond (OPF) is presented in Table 1. OPF contained 62.95% DM, 5.30% ash, 3.57% CP, 86.02% NDF, and 71.69% ADF. Nevertheless, the nutritive value of OPF may depend on the combined effects of age and environmental factor including cultivar, growth stage, plant density, plant part, soil fertility, harvesting frequency, season and climate.

Table 1. Chemical composition of fresh oil palm fronds (OPF).

Item	% of DM basis
Dry matter (DM)	62.95
Organic matter (OM)	94.70
Ash	5.30
Crude protein (CP)	3.57
Neutral detergent fiber (NDF)	86.02
Acid detergent fiber (ADF)	71.69
Hemicellulose	14.33

As shown in Table 2, there were no A×B interactions ($p>0.05$) with respect to growth rate, density, and chemical composition of OPF, but there were significantly affected ($p<0.05$) by cultures of WRF. After 21 days of incubation, the maximum growth and density were observed in species LSc and SC compared with other treatment groups and the minimum for species PD. Likewise, growth rate was affected ($p<0.05$) by levels of urea; species of WRF receiving 1% of urea had greater growth rate than that of 0% of urea, whereas 0.5 and 1% of urea showed a similar growth. The chemical compositions of OPF are shown in Table 2. The CP, NDF, and ADF contents ranged from 5.49-6.58, 62.28-77.50, and 55.55-67.14% respectively and were significant ($p<0.05$) among the treatments. Biological treatment of the OPF with WRF had significantly ($p<0.01$) increased the CP content. Among the treated WRF, CP was higher for the LSc, LS, and SC. Similarly, increases in these CP levels were reported by Ragunathan et al., (1996) for straw. It may be related to the amount of growth and cultural differences. The biological treatment of OPF with six species of WRF significantly ($p<0.05$) reduced the NDF and ADF contents of OPF. Similarly, decreasing in these fiber fraction levels were reported by Jalç et al. (1996) for wheat straw. However, the ability of the WRF to degrade these components varied among the species. LS and PO showed significantly ($p<0.05$) lower ability than the others to decreasing the NDF and ADF contents. The ash content ranged from 4.20-6.52 % and was not significant ($p>0.05$) among the treatments. At all urea levels, CP increased significantly with increasing urea levels. Urea addition of 1% increased CP significantly compared with 0%. At all urea levels, NDF decreased significantly with increasing urea levels. Urea addition of 1% decreased NDF significantly when compared with 0 and 0.5%, whereas ADF was affected ($p<0.05$) by levels of urea. The decrease in NDF was probably due to swelling of hemicellulose and cellulose, breaking of lingo-cellulosic bonds. In conclusion, treatment of OPF with WRF particularly for LSc and Sc resulted in changing growth and reduction of its cell wall components, increasing of CP, and adding a high amount of urea (0.5-1%) intended to increase CP in OPF.

Table 2. Number of days for full colonization and chemical composition of fungal treated oil palm frond at 3 weeks (% of DM basis).

Treatment	Number of days			Chemical composition			
	Urea (%)	Growth ²	Density ³	CP	NDF	ADF	Ash
PO	0	7.02 ^b	++	5.86 ^{abcd}	73.45 ^a	56.65 ^f	5.89
	0.5	7.00 ^b	++	5.77 ^{bcd}	73.91 ^a	56.46 ^{ef}	4.25
	1	7.00 ^b	++	6.20 ^{abcd}	71.45 ^{abc}	55.55 ^f	5.28
PD	0	7.10 ^a	+	5.49 ^d	70.96 ^{abc}	58.16 ^{cdef}	6.52
	0.5	7.10 ^a	++	6.05 ^{abcd}	73.19 ^a	60.12 ^{bcd}	5.09
	1	7.02 ^b	++	5.70 ^{cd}	62.28 ^c	57.61 ^{def}	5.47
LS	0	5.02 ^c	++	5.84 ^{abcd}	74.90 ^a	65.17 ^{ab}	5.71
	0.5	5.00 ^c	++	6.10 ^{abcd}	77.50 ^a	67.14 ^a	6.03
	1	5.00 ^c	++	6.49 ^{ab}	75.29 ^a	64.07 ^{abcd}	5.45
SC	0	4.00 ^d	++	6.09 ^{abcd}	74.06 ^a	59.12 ^{bcd}	4.91
	0.5	4.00 ^d	++	5.86 ^{abcd}	71.09 ^{abc}	57.73 ^{def}	5.59
	1	4.00 ^d	++	6.30 ^{abc}	70.98 ^{abc}	57.88 ^{def}	4.87
LP	0	5.03 ^c	+	5.50 ^d	70.78 ^{abc}	63.40 ^{abcde}	4.56
	0.5	5.00 ^c	+	5.82 ^{abcd}	69.39 ^{abc}	64.63 ^{abc}	4.20
	1	5.00 ^c	+	5.92 ^{abcd}	68.31 ^{abc}	62.26 ^{abcde}	5.31
LSc	0	4.00 ^d	+++	6.13 ^{abcd}	71.80 ^{ab}	57.39 ^{def}	4.50
	0.5	4.00 ^d	+++	6.58 ^a	68.14 ^{abc}	56.51 ^f	5.28
	1	4.00 ^d	+++	6.50 ^{ab}	63.53 ^c	55.85 ^f	5.69
SEM		0.02	-	0.22	2.55	2.03	0.66
P-value	A	*	-	**	**	**	NS
	B	*	-	*	*	NS	NS
	AxB	NS	-	NS	NS	NS	NS

^{a-d} Within rows not sharing a common superscripts are significantly different (p<0.05).

¹White rot fungi: PO = *Pleurotus ostreatus*, PD = *Pleurotus djamor*, LS = *Lentinus squarrosulus*, SC = *Schizophyllum commune*, LP = *Lentinus polychrous*, LSc = *Lentinus sajor-caju*.

²No. of days for full colonization of substrate.

³+ denote the density of mycelium. More + indicates more dense growth of mycelium

*, ** Significantly different at p<0.05 and p<0.01, respectively. NS: Not significant.

References

- AOAC. 1995. Official Methods of Analysis. 16th ed. Assoc. Off. Anal. Chem., Arlington, VA.
- Hamed, A. H. M. and M. E. Elimam. 2010. Performance and digestibility in Nubian goats fed stream treated sorghum stover. Pak. J. Nutr. 9:298–301.
- Ishida, M. and O. Abu Hassan. 1997. Utilization of oil palm frond as cattle feed. Japan Agric. Res. Quart. 31:41–47.
- Jalc, D., F. Nerud, R. Zitnan and P. Sikora. 1996. The effect of white-rot basidiomycetes on chemical composition and in vitro digestibility of wheat straw. Folia. Microbiol. 41:73–75.
- Okano, K., M. Kitagawa, Y. Sasaki and T. Watanabe. 2005. Conversion of Japanese red cedar (*Cryptomeria japonica*) into a feed for ruminants by white-rot basidiomycetes. Anim. Feed Sci. Technol. 120:235–243.
- Ragunathan, R., R. Gurusamy, M. Palaniswamy and M. Swaminathan. 1996. Cultivation of *Pleurotus* spp. on various agro-residues. Food Chem. 55:139-144.
- Van Soest, P. J., J. B. Robertson and B. A. Lewis. 1991. Methods of dietary fibre, neutral detergent fibre and non starch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74:3583-3597.
- Zahari, M.W., and A.R. Alimon. 2005. Use of palm kernel cake and oil palm by-products in compound feed. Palm Oil Developments, vol. 40. Malaysian Palm Oil Board, Kuala Lumpur, Malaysia, pp. 5–8.

Effect of chemical treated shrimp meal on growth performances of broilers

Mustanur Rahman¹ & Katsuki Koh²

¹Interdisciplinary Graduate School of Science and Technology, Shinshu University, 399-4598 Nagano, Japan, ²Faculty of Agriculture, Shinshu University, 399-4598 Nagano, Japan

Abstract

The present study was performed to evaluate whether formic acid treated shrimp meal (SM) is more suitable protein source for broilers than untreated one. Seven dietary groups (5 chicks each) were allocated to one of the 7 experimental diets (control diet and diets containing 5%, 10% and 15% of untreated SM (SMu) and treated SM (SMt). Their growth performance was measured from 8 to 35 days of age with free access to diets and water.

The growth performance data exhibited that body weight gain (BWG) decreased with increasing level of SMu and SMt, and this tendency was prominent in untreated groups, although 5% SMu and SMt groups showed similar and greater BWG, respectively, comparing with control group. Feed intake decreased slightly with increasing level of SMu, but increased in 5% SMt group and unchanged in 10% and 15% SMt groups. Feed conversion ratio (FCR) deteriorated with increasing level of SMu and SMt, except 5% SMu and SMt groups showing similar FCR to control group. The deteriorating tendency was prominent in untreated groups. Similarly, nitrogen retention in SMu and SMt groups showed reduction with increasing level of SM and the reduction effect was somewhat greater in SMu groups. Chitin digestibilities in SMt groups were greater than corresponding values in SMu groups. In conclusion, it is suggested that formic acid treated SM may be more suitable protein source for broiler diets than untreated one.

Keywords: broiler, growth performance, shrimp meal, treatment

Corresponding author: mustanur2010@gmail.com

Introduction

SM has been tried to use as a potential protein source for broiler diets, although its performance is not enough for commercial use (Khempaka et al., 2006a, b). In addition, the inclusion level of SM is still low when compared with conventional protein sources, which might be attributed to the high levels of chitin and/or ash present in SM. For instance, chitin is considered to have low protein digestibility in rats (Oduguwa et al., 1998), and low DM digestibility in broilers (Khempaka et al., 2006b). In this connection, our previous *in vitro* study (Rahman & Koh, 2014) clarified that formic acid treatment was able to reduce such undesirable compounds in SM, which led us to the hypothesis that the nutritional quality of SM can be improved by the treatment. Unfortunately, there are no reports concerning the effect of formic acid treated SM in broiler diets, to our knowledge.

In order to clarify this hypothesis, we investigated the effect of formic acid treatment of dietary SM on growth performance in broilers in the present study.

Materials and Methods

SM made of heads with hulls of black tiger shrimp (*Penaeus monodon*) was treated with formic acid as summarized as follows: 100 g of SM was soaked in 300 ml of 3% formic acid for 20 min, sun-dried, ground to pass through 1.0 mm size and then used as a treated SM.

A total of 35 male broiler chicks (8 days of age) were randomly assigned into 7 dietary groups having similar body weight (5 chicks each). Each group received one of the 7 experimental diets (control diet and diets containing 5%, 10% and 15% of SMu and SMt). Metabolizable energy and CP were adjusted to about 3,180 kcal/kg and about 235 g/kg, respectively (Table 1), and other nutrients were formulated to meet or slightly exceed the requirements of broilers defined by Japanese feeding standard for poultry (2011). Diet and water were provided *ad libitum* during the experimental period (8 to 35 days of age). Body weight and feed intake were recorded weekly and daily, respectively. Digestibility and nitrogen retention studies were also performed.

Table 1. Ingredients and chemical composition of experimental diets.

	Control	Untreated SM			Treated SM		
		5%	10%	15%	5%	10%	15%
Ingredients(g/kg)							
Commercial diet ¹	550	550	550	550	550	550	550
Soybean meal	185	135	88	42	130	78	25
Corn	239	225	210	193	234	222	214
Shrimp meal	0	50	100	150	50	100	150
Corn oil	10.5	24.5	36.5	49.5	20.5	34.5	45.5
Vit. + min. premix ²	15.5	15.5	15.5	15.5	15.5	15.5	15.5
Analyses							
Crude protein (%)	23.6	23.4	23.4	23.4	23.5	23.6	23.6
Chitin (%)	-	0.9	1.7	2.6	0.8	1.5	2.3
ME (kcal/kg, calculated)	3180	3180	3174	3173	3182	3180	3175

¹Broiler starter diet (CP≥23.5%, ME≥3050 kcal/kg, Nippon Formula Feed Mfg. Kanagawa, Japan); ²NRC (1994).

The obtained data were analyzed by one way ANOVA. The Tukey's multiple comparison tests was performed at a significance level of 5% among the dietary groups.

Results and Discussion

BWG and feed intake showed similar results (Fig. 1): comparing with the data in control group, these values decreased with increasing level of SMu, but unchanged in 10% and 15% SMt groups: exceptional increases of these values were found in 5% SMt group. FCR deteriorated with increasing level of SMu and SMt, except 5% SMu and SMt groups which showed similar FCR to control group. These results may be

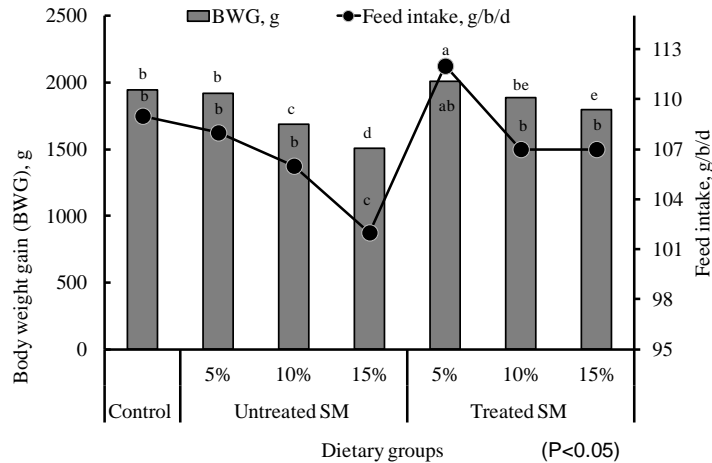


Figure 1. Body weight gain and feed intake of broilers given dietary untreated and treated SM

attributed that formic acid has the ability to degrade chitin which is in agreement with our previous *in vitro* study (Rahman & Koh, 2014). The similar results have been reported by Fox et al. (1994) who found improved growth and better survival rate in shrimps when formic acid treated SM was given. Nitrogen retention was greater in 5% SMt group, although this did not differ significantly with control and 10% SMt groups, whereas this decreased significantly in 15% SMu ($P < 0.05$). This decreasing tendency was prominent in untreated SM groups,

which supports our previous findings (Rahman & Koh, 2014). Chitin digestibility was greater in SMt than that of the corresponding values of SMu. This may be noted that chitin in shrimp is recognized to be slightly soluble in dilute formic acid.

In conclusion, the results obtained here suggest that formic acid treated SM may be more suitable protein source for broiler diets than untreated one.

References

- Fox, C. J., P. Blow, J. H. Brown and I. Watson, 1994. The effect of various processing methods on the physical and chemical properties of shrimp heads meal and their utilization. *Aqua*. 122: 209-226.
- Khempaka, S., K. Koh and Y. Karasawa, 2006a. Effect of shrimp meal on growth performance and digestibility in growing broilers. *J. Poult. Sci.* 43: 250-254.
- Khempaka, S., M. Mochizuki, K. Koh and Y. Karasawa, 2006b. Effect of chitin in shrimp meal on growth performance and digestibility in growing broilers. *J. Poult. Sci.* 43: 339-343.
- Oduguwa, O. O., A. O. Fanimu, E. A. Lyayi, O. O. Kalejaiye and O. A. Oyekola, 1998. Preliminary studies on the effects of different processing methods on the nutritive value of shrimp waste meal. *Nig. J. Anim. Prod.* 25: 139-144.
- Rahman, M. and K. Koh, 2014. Improvement in nutritional quality of shrimp meal with autoclave and chemical treatments. 16th AAAP Animal Science Congress, Yogyakarta, Indonesia.

Effect of bee pollen as a natural antioxidant on the performance, carcass and antioxidant status of V-line rabbits

Zeweil, H. S., M. H. Abd El-Rahman, S. M. Zahran, Y. M. El-Gendy & E. M. AlHemaley

Department of Animal and Fish Production, Faculty of Agriculture (Saba Basha), University of Alexandria, Alexandria, Egypt

Abstract

The present study was conducted to determine the effect of supplementing bee pollen (BP) as a natural growth promoter and antioxidant on growth performance, carcass traits, lipid profile and lipid peroxide (Malondialdehyde) of growing V-line rabbits. A total number of 27 V-line rabbits at 5 weeks old with average initial live body weight of 731.18 ± 26.86 g were divided into three groups (n=9 rabbits/group). Each group was subdivided into three replicates with three rabbits each in a completely randomized design. The first group received basal diet free of BP. The second and third groups were fed diets containing 0.1 and 0.2 g BP/kg diet, respectively. The results indicated that body weight and feed intake were not affected by treatments; however, feed conversion ratio was significantly improved by Bp supplementation. Carcass traits were not affected by treatments. Bee Pollen diets significantly ($P \leq 0.01$) lowers the level of serum total lipids and low density lipoprotein, however, it had insignificant decreasing effect on total cholesterol and triglycerides and had numerically increasing effect on high density lipoprotein as compared with the control group. Serum malondialdehyde significantly ($P \leq 0.01$) decreased by Bp supplementation in blood serum as compared to the control group. It is well demonstrated in the present study that the consumption of BP has positive effects on rabbits' performance, had a beneficial effect on blood lipid regulation which was demonstrated to be ascribed to their antioxidant activity and on lipid peroxide of growing V-line rabbits.

Keywords: rabbit, bee pollen, performance, carcass, blood constituents.

Introduction

In rabbit production, the ban of antibiotic growth promoters poses a serious challenge as because of the very complex and peculiar digestion (caecotrophy, microbial fermentation), the rabbit is susceptible to enteric diseases, particularly after weaning. For this reason, several studies have been conducted to find alternatives to replace dietary antibiotics (Eiben et al., 2008). Bee pollen enhances immunizing function of poultry, promotes animal growth, protects intestinal tract health and improves animal products quality and security (Liu et al., 2010). Also, bee pollen has ability to protect kidney, and can decrease the level of triglycerides, cholesterol, creatinine and blood urea nitrogen in rats (Hu et al., 2003). Therefore, the aim of the present study was to determine the effect of bee pollen as natural growth promoters on growth performance, carcass traits, lipid profile and lipid peroxide (Malondialdehyde) of growing V-line rabbits

Materials and methods

A total number of 27 V-line rabbits at 5 weeks old with an average initial live body weight of 731.18 ± 26.86 g were divided into three groups (n=9 rabbits/group). Each group was subdivided into three replicates with three rabbits each in a completely randomized design. Rabbits fed a basal

diet containing 17.0 % crude protein and 2650 kcal digestible energy / kg diet. The first group received basal diet free of bee pollen (BP). The second and third groups were fed diets containing 0.1 and 0.2 g BP /kg diet, respectively. The experiment lasted for eight weeks. All cages were provided with a manual feeder, and clean fresh water was available continuously through an automatic system of nipple drinkers. Rabbits were kept under the same hygienic and environmental conditions during the experimental period, they provided with 14 h of light daily. All biochemical traits of blood serum were determined using commercial kits (Diamond Diagnostics, Halliston, MA, USA). Data were analyzed using one-way ANOVA of GLM procedure of SAS® (SAS Institute, 2000). Significant differences between means were detected using new Duncan multiple range test (Duncan, 1955).

Results and discussions

Results presented in Table 1 indicated that body weight and feed intake were not affected by treatments; however, feed conversion ratio was significantly ($P \leq 0.01$) improved by Bp supplementation as compared with the control group. Carcass traits were not affected by treatments. Bee Pollen diets significantly ($P \leq 0.01$) lowers the level of serum total lipids and low density lipoprotein, however, it had insignificant decreasing effect on total cholesterol and had numerically increasing effect on high density lipoprotein as compared with the control group. Serum malondialdehyde significantly ($P \leq 0.01$) decreased by Bp supplementation in blood serum as compared to the control group. Attia *et al.* (2011) observed that the feed efficiency was improved in growing rabbits due to the supplementation of 200 - 500 mg BP kg⁻¹ body weight compared to the unsupplemented control group, during moderate and hot seasons, from weaning to mature age. Also, Ke *et al.* (2009) found that the use of 0.2% BP polysaccharides in broiler diets did not influence the carcass yield of the birds. The decrease in serum total lipids may indicate a depletion of energy source to maintain body temperature. The decrease in serum total lipids, and cholesterol and low density lipoprotein could be due to phospholipids and PUFA particularly linolenic fatty acid which represented 1.19% in BP (Xu *et al.*, 2009). Hajkova *et al.* (2013) indicated that BP possesses a noticeable source of compounds with health protective potential and antioxidant activity. Attia *et al.* (2014b) reported that the addition of BP caused a significant increased of total antioxidants capacity as compared to control diet of rabbits. Also, treated with flavonoids, polyphenolic compounds and certain alkaloids resulted to reduce cholesterol level and play an important role in the prevention of a number of chronic diseases such as cancer and cardiovascular disease in rabbits. The decrease of cholesterol levels may be directly related to the influence of BP on lipid metabolism. It is well demonstrated in the present study that the consumption of BP has positive effects on rabbits' performance, had a beneficial effect on blood lipid regulation which was demonstrated to be ascribed to their antioxidant activity and on lipid peroxide of growing V-line rabbits.

Table 1: Effect of different levels of bee pollen on V-line rabbits

Items	Control	Bee pollen level (g / kg diet)	
		0.1	0.2
<u>Rabbits performance:</u>			
Initial body weight, g	714.38±36.59	746.67±22.02	732.50±21.98
Final body weight, g	2251.88±79.91	2431.67±62.53	2455.63±69.50
Daily body weight gain, g	25.20±0.90	27.62±0.76	28.25±1.06
Feed intake, g	111.18±1.46	105.40±0.31	108.51±1.57
Feed conversion ratio	4.45±0.17 ^a	3.84±0.10 ^b	3.87±0.12 ^b
<u>Carcass characteristics:</u>			
Pre-slaughter weight, g	2066.67±14.81	2241.67±19.22	2330.00±40.72
Carcass %	51.42±1.98	51.96±0.37	49.81±1.04
Liver, %	3.20±0.28	3.07±0.13	4.04±0.41
Heart, %	0.34±0.03	0.33±0.02	0.32±0.02
Spleen, %	0.08±0.02	0.08±0.02	0.07±0.01
Kidneys, %	0.78±0.12	0.69±0.08	0.64±0.01
Kidneys fat, %	0.36±0.20	0.37±0.10	0.29±0.04
Lungs, %	0.54±0.01	0.56±0.11	0.46±0.02
<u>Serum blood lipids profile:</u>			
Total lipids	167.10 ± 0.55 ^a	159.90±2.40 ^b	155.50±1.87 ^b
Total cholesterol	52.00±1.66	49.00±1.35	48.93±0.93
High density lipoprotein (HDL)	25.07±0.48	26.37±0.58	27.23±0.50
Low density lipoprotein (LDL)	19.27±0.57 ^a	15.23±1.11 ^b	16.83±0.27 ^b
Serum malondialdehyde	19.78±0.95 ^a	15.77±0.38 ^b	13.53±0.66 ^c

Means within row bearing different superscripts are significantly different.

References

- Attia, Y. A., A. M. El-Hanoun, F. Bovera, G. Monastra W. S. El-Tahawy and H. I. Habiba, 2014. Growth performance, carcass quality, biochemical and haematological traits and immune response of growing rabbits as affected by different growth promoters. *J. Anim. Physiol. Anim. Nutr.* 98: 128–139.
- Attia, Y. A., A. Al-Hanoun, and F. Bovera, 2011. Effect of different levels of bee pollen on performance and blood profile of New Zealand White bucks and growth performance of their offspring during summer and winter months. *Journal of Animal Physiology and Animal Nutrition*, Vol. 95: 17-26.
- Duncan, D.B., 1955. Multiple range and multiple *F* tests. *Biometrics*, 11: 1-42.
- Hajkova, Z., R. Toman, S. Hluchý, B. Gálik, D. Bíro, M. Martiniaková, R. Omelka and I. Boboňová, 2013. The effect of pollen on the structure of the small intestine in rats after an experimental addition in diet. *Anim. Sci. and Bio.*, 46: 232-237.
- Ke, P., H. Sun and Z. Gao, 2009. Effect of bee pollen polysaccharide on growth performance of broilers and chemical compositions of muscle. *Feed Industry*, Vol. 27: 36-38.
- Xu, X., L. Sun, J. Dong and H. Zhang, 2009. Breaking the cells of rape bee pollen and consecutive extraction of functional oil with supercritical carbon dioxide. *Innovative Food Sci. and Emerging Tech.*, 10: 42–46.

Nutritive value of grower pig ration using local feeds in West Manokwari District, Manokwari

Hartini, S., M.N.Lekitoo, S.Lumatauw, D.D.Rahardjo, S.Y.Randa, T.W.Widayati & R.Siregar
University of Papua, Gunung Salju Amban, Manokwari 98314, Indonesia

Abstract

A study was carried out to observe the nutritive value of grower pig ration using local feeds applied by farmers in West Manokwari district, Manokwari, Indonesia. Thirty-five pig farmers from 3 different places at West Manokwari district were used as respondents. Descriptive method with technical survey was used to know the followings: 1. Information on flock size of grower pigs owned by farmer, 2. Identification of local feeds used to formulate a ration, 3. Information on the quantity of each local feed used in the ration, and 4. Information on the frequency of grower pig feeding in a day. In addition, the chemical analysis of each local feed was applied. Data obtained was used to calculate the nutritive value (protein and energy contents) of the rations. Supporting data obtained were education background, age, and job preference of the farmer. Results showed that 80% of the grower pig rations using local feeds applied by the farmers in West Manokwari district had achieved the standard of protein requirement, but had not achieved the standard of energy requirement suggested by NRC. The majority of the farmers (49%) only had a high school education background. Approximately 86% of the farmer was in productive age, and about 63% said that pig farming was their primary job. In conclusion, local feeds were potential to be used as basic ingredient for the grower pig ration in West Manokwari district, however due to less knowledge of the farmer about feed formulation therefore the nutritive value (energy content) of the rations still below the nutritive standard suggested by the NRC.

Keywords: grower pig ration, local farmer, nutritive value

Introduction

In Manokwari, West Papua, Indonesia, pigs are kind of animal that favorable by indigenous or local people to be raised. Pigs in Papuan tribes are usually kept for the purpose of consumption, generating income, ceremony, or social events (Iyai et al., 2013). Pigs are used for bride prices; pigs are also used as payments to resolve social disputes. The killing of pigs is also tied to important events such as cremation, marriage and initiation rites. Pigs population in Manokwari was 2614 pigs (BPS Papua Barat, 2013), with distribution at West Manokwari 792 pigs, South Manokwari 407 pigs, East Manokwari 1030 pigs, and North Manokwari 385 pigs.

Ration in pig farming holds an important factor since about 60-85% of total production cost is feed. To raise pigs, farmers in Manokwari depend mostly on the use of local feed resources since a commercial feed was very expensive. The farmers mixed their pig ration without

calculating the nutrient content of the ration. According to NRC (1998), feed intake for grower pig was 1.855kg DM/day or 8.09kg (as fed)/day, whereas the protein and metabolizable energy requirements were 18% and 3256kkal/kg, respectively. The study was carried out to evaluate the nutritive value of grower pig ration using local feed resources. Pig farmers located at West Manokwari district was chosen as respondents for this study.

Materials and Methods

Thirty-five pig farmers from 3 different places at West Manokwari district (Padarni, Sanggeng, and Wosi villages) were used as respondents in this study. The method used in the study was descriptive method with survey and interview techniques. Data collected from respondents were: 1) flock size of grower pigs, 2) identification of local feeds used in a ration, 3) the quantity of local feeds used in the ration, and 4) frequency of feeding per day. In addition, proximate analysis (AOAC, 2002) of each local feeds was applied. Data collected were used to calculate total protein and energy content of the rations. As supporting data were education background, age, and job preference of the farmer.

To calculate total Protein or Metabolizable Energy (ME) in the ration the formula used was;

$$\text{Total Protein or ME ration} = a_i P_i + a_j P_j + \dots + a_n P_n,$$

where;

i, j, ...n = type of local feed used

a = total of each local feed used

P = Protein or ME content of feed used

To calculate total Protein or ME needed based on total animal the formula used was;

$$\text{Total Protein or ME needed} = a \times i \times j,$$

where;

a = total animal (pigs)

i = feed intake of grower pig/day, which was 1.855kg DM (NRC, 1998)

j = Protein or ME requirement for grower pig. CP 18%, ME 3265 kkal/kg DM (NRC, 1998)

Analysed Data

All data were tabulated and then compared to nutrient requirement for grower pig based on NRC (1998).

Results and Discussion

Flock size of grower pigs owned by farmers was ranged between 3 to 11 pigs. The majority of the farmers (63%) had 3 to 4 grower pigs, while those who had 8 to 11 grower pigs only 9% (3 respondents). Feed given per pig per day was less than 4.5kg. This feed offered by the farmers was far below the standard intake for a grower pig by NRC (1998) which was 8.09kg/pig/day. The low intake will definitely affect growth rate. The pig farmers were paid strictly on a live-weight basis; it was their loss when their pigs did not achieve maximum growth. There were ten kinds of local feeds used as feed ingredient in the pig grower ration (Table 1). The local feed mostly used in the ration was vegetable leftovers (66%), followed by taro peelings (60%), cassava peelings (49%),

tofu waste and restaurant waste (37%, respectively), fish waste (26%), kitchen scraps (11%), sweet potato peelings (9%), mung bean peelings and stem plantain (3%, respectively). The protein content (CP) of local feeds was ranged between 2.4 to 31.21 % DM, whereas the energy contents (GE) was ranged between 2649 to 4951 kkal/kg DM (Table 1). The proximate analysis showed that some of local feeds were potential to be used as basic ingredients for ration formulation either as protein or energy sources. Roots and tubers (Machin 1992 in González et al., 2003), kitchen scraps (swill) (Iyai et al., 2013) are kind of feeds that could potentially substitute the conventional animal feeds. Based on the calculation of nutritive contents (protein and energy), it was found that 28 out of 35 respondents (about 80%) had their protein content in their ration above the standard requirement stated by NRC, but all the energy content still below the NRC's standard. The education background of the farmer in the West Manokwari district mostly high school (49%). It was likely that the farmers had less knowledge on feed formulation.

Table 1. The identification, percentage and nutritive contents of local feeds used by pig farmers in West Manokwari district.

No	Type of local feed	% Feed used by farmers	Nutritive contents							
			Dry Matter basis				As fed basis			
			DM ¹⁾ (%)	CP ¹⁾ (%)	GE ¹⁾ (kkal)	ME ²⁾ (kkal)	DM ¹⁾ (%)	CP ¹⁾ (%)	GE ¹⁾ (kkal)	ME ²⁾ (kkal)
1.	Fish waste	26	92.60	31.21	3433	2709	29.41	9.91	1090	860
2.	Tofu waste	37	91.77	23.85	4951	3906	14.13	3.67	763	602
3.	Mung bean peelings	3	87.80	15.10	4022	3174	16.80	2.89	769	607
4.	Taro peelings	60	88.85	4.26	3649	2879	26.72	1.28	1098	866
5.	Vegetable leftovers	66	93.94	15.80	3684	2907	9.84	1.65	385	304
6.	Stem plantain	3	91.81	2.91	3805	3002	24.88	0.79	1032	814
7.	Restaurant waste	37	91.32	13.72	4202	3315	35.85	5.43	1649	1301
8.	Kitchen scraps	11	91.32	14.20	2665	2103	35.85	5.2	1648	1300
9.	Sweet potato peelings	9	91.76	4.64	2649	2879	23.10	1.26	1091	861
10	Cassava peelings	49	91.76	2.40	4204	3317	23.60	1.21	864	682

¹⁾DM: Dry Matter, CP: Crude Protein, GE: Gross Energy, ME: Metabolizable Energy.

²⁾ME= 78.9%GE (Sihombing, 1997).

In conclusion, the nutritive value of the grower pig rations in West Manokwari district still below the nutritive standard suggested by the NRC and training pig farmers on feed formulation will be the strategy that can be applied to overcome the problem. With proper knowledge on feed formulation, formulating pig rations using local feeds would help to increase pig production in West Manokwari district and to give top profit return to the farmer.

References

- AOAC. 2002. Official Methods of Analysis of the Association of Analytical Chemist. Association of Official Analytical Chemist, Arlington, VA, USA.
- BPS Papua Barat, 2013. Papua Barat Dalam Angka (West Papua in Numbers). Badan Pusat Statistik, Manokwari.
- González, C., I.Díaz, H.Vecchionacce and J. Ly. 2003. Performance traits of pigs fed sweet potato (*Ipomoea batatas L.*) foliage *ad libitum* and graded levels of protein. Livestock Research for Rural Development 15 (9). Retrieved August 31, 115, from <http://www.lrrd.org/lrrd15/9/gonz159.htm>
- Iyai, D.A., O. Marani, T. Marjen and L. Usior. 2013. Pig farming performances of three Papuan tribes: case study of Byak, Onate and Arfak tribes in Papua Barat. J. Indonesian Trop. Anim. Agric. 38(1): 55-64.
- NRC. 1998. Nutrient Requirements of Swine 10th revised edition. National Academic Press. Washington, D.C.
- Sihombing, D.T.H. 1997. Ilmu Ternak Babi. GadjahMada University Press, Yogyakarta.

The effect of chemical processing of soybean meal on *in vitro* ruminant intestinal available protein

Danesh Nesgaran, M., J. Falahati zowe & S. A. Vakili

Department of Animal Science, Ferdowsi University of Mashhad, Mashhad, Iran

Abstract

The aim of this study was to investigate the effects of chemical processing of soybean meal on the amount of available crude protein at the duodenum (uCP) using a new modified gas technique. Evaluated samples were two samples of unprocessed soybean meal and one sample of processed soybean meal (heat processing with xylose) containing 495, 480 and 446 g/kg crude protein (CP), respectively. To perform the gas test, rumen fluid was collected before the morning feeding from two rumen fistulated Holstein dairy cows (640 ± 38 kg, body weight). Feed samples (200 mg) and blanks (only 30 ml of buffered rumen fluid) incubated simultaneously in three repeats and three runs for 8 and 24 hours. At the end of the each incubation time, the uCP was calculated as non-ammonia N which was calculated by subtracting the amount of ammonia N released in the incubation medium of the total incubated N (sum of N content of feed sample and ammonia N in blanks). Effective uCP (EuCP) was calculated via an exponential equation using the estimated uCPs at 8 and 24 h post incubation. Ratio of effective uCP (at the passage rate of 0.1/h) to the CP content of feed samples for processed soybean meal and two untreated soybean meals were 0.873, 0.747 and 0.785 (SEM=0.0355), respectively. These results showed that the processing had no significant effect on the amount of EuCP of soybean meal ($P > 0.05$). However, the amount of relative EuCP for processed soybean meal was 10-15 percent greater than untreated soybean meals.

Keywords: soybean meal, protein

*Corresponding author: danesh@um.ac.ir

Introduction

Nitrogen efficiency in ruminants, especially in high yielding dairy cows is low (Calsamiglia et al., 2010) and environmental nitrogen pollution through dairy farms is becoming a serious challenge. There are several methods to estimate ruminal protein degradability; between these methods, recently Edmunds et al. (2012) described a new gas production technique that facilitates to estimate sum of microbial protein and ruminally undegraded protein (that is approximately equivalent with the utilizable crude protein at the duodenum (uCP)) in the incubation medium. This technique eliminate the confounding effects of various parameters such as *de novo* synthesis of microbial protein, microbial markers, loss of the feed particles through bag pores, soluble proteins and endogenous nitrogen on evaluation of feed proteins. Moreover, this new gas test estimated the nutritional value of feed proteins using CP content of feed sample and concentration of ammonia nitrogen in the incubation medium which are easily measurable. Therefore, the aim of this study

was to evaluate the effects of chemical processing of soybean meal on the amount of uCP using this new modified gas technique.

Materials and Methods

Commercial untreated soybean meals (soybean meal 1 and 2) and a processed soybean meal (heat processing with xylose) were used as feed samples. These three feed samples were incubated simultaneously in complete randomized design. The experiment was conducted with six replicates in three runs. In each run, a blank (buffered rumen fluid without feed sample) was incubated in eight replicates. A new gas production technique according to Edmunds et al. (2012) was followed. The rumen fluid was collected manually before the morning feeding from two rumen fistulated lactating Holstein dairy cows (640 ± 38 kg, body weight) that were fed a diet that containing corn silage (250 g/kg DM), alfalfa hay (250g/kg DM), and concentrate (500g/kg DM consisted of 30% barley grain, 20% corn grain, 10% cotton seed meal, 10% soybean meal, 10% sugar beet pulp, 18% wheat bran and 2% vitamin and mineral supplement). Rumen fluid was filtered through four layers of cheesecloth and then was mixed with buffered mineral solution (Menke and Steingass, 1988) in ratio of 1:2. Incubations were performed in 125 ml serum bottles. 200mg of each feed sample was weighed (0.001g Precision Balance) into incubation bottles then; 30 ml of buffered rumen fluid (BRF) was added to each bottle and oxygen was removed from the bottles headspace using CO₂ stream. Then, each bottle was sealed with rubber stopper and aluminum cap. The incubations were performed in a water bath at 39 °C for 8 and 24 h. Blanks were contained only BRF and treated in the same manner as the samples. Half of the replicates of each treatment (and blanks) at 8 h and remained bottles at 24 h post incubation were transferred to a water-ice mixture to stop fermentation. Afterwards, the bottles were opened and 5 ml of liquid phase was pipetted into 50 ml serum bottles that were contained 5 ml of 0.2 N HCl. Then, these bottles were sealed with rubber stopper and aluminum cap and stored in refrigerator at 4 °C for measurement of ammonia concentration. For each of feed samples the uCP value was calculated according to the equations proposed by Edmunds et al. (2012). Data were analyzed using GLM procedure of SAS (9.2, 2002).

Results and discussion

The amounts of CP and uCP values of the feed samples are shown in the Table 1. The two untreated soybean meals had higher CP content than processed soybean meal. The uCP value of processed soybean meal was greater than untreated soybean meals at 8 h post incubation but, differences were not significant ($P > 0.05$). Also, the differences between uCP values of feed samples at 24 h post incubation were not significant ($P > 0.05$). The processed soybean meal had higher EuCP value compared with two untreated soybean meals however; differences were not significant ($P > 0.05$). Ratio of EuCP to the CP content of feed sample for processed soybean meal was greater than untreated soybean meals but, differences were not significant ($P > 0.05$). The amounts of uCP decreased as the incubation time increased. This study was conducted to evaluate the effects of heat processing with xylose on the amount of utilizable crude protein applied by soybean meal at the duodenum using a new gas production technique. Our results indicated that heat processing with xylose cannot significantly affect uCP and EuCP values of soybean meal. However, uCP and EuCP value of processed soybean meal were numerically greater than untreated soybean meals. It has been demonstrated that an increase in rumen undegradable protein at the expense of ruminally

degradable protein can reduced production of microbial protein. But, the used technique in our experiment and the nature of feed samples provided the conditions that N was not limiting factor for producing microbial protein (Edmunds et al., 2012). In the current study, higher CP content of untreated soybean meals may affect the uCP estimates. It has been concluded that at the case of replacing soybean meal with high RUP sources, the lack of an overall increase in non-ammonia nitrogen flow to the small intestine despite increasing in non-ammonia non-microbial nitrogen flow was a result of decreasing in microbial nitrogen flow. Heat processing temperature and xylose supplementation level are other factors that affecting ruminal protein degradability. In the current study, the heat processing temperature might be not enough to affect soybean meal protein degradability. Vaga et al. (2014) examined the effects of various heat processing methods on uCP values of field beans and lupines using gas production technique. They used processed protein feeds in a total mixed ratio and shown that heat processing of field beans and lupines effectively increased uCP values. Therefore, measuring the uCP values of processed soybean meals when those are used in a total mixed ratio may be useful for determining possible changes occurred with more accuracy.

Table 1. Effects of heat processing with xylose on CP, uCP and EuCP (g/kg DM) values of soybean meal and its relative amounts of EuCP.

Feed sample	CP	uCP, 8 h	uCP, 24 h	EuCP	EuCP/CP
processed soybean meal	446.281 ^c	429.273	230.064	388.841	0.873
untreated soybean meal 1	480.780 ^b	417.084	233.562	379.911	0.785
untreated soybean meal 2	495.723 ^a	404.699	232.446	369.808	0.747
SEM	1.578	9.329	19.236	16.912	0.035

SEM: standard error of the mean. Within columns, means followed by different letters are significantly different by Tukey's test at 5% probability.

Conclusion

This study indicated that the heat processing with xylose had no significant effect on uCP and EuCP values of soybean meal but, the amounts of them were increased numerically. The used new gas production technique provided good, fast and inexpensive estimates of uCP value of soybean meal products and shown that it can be used to evaluate protein feeds without interference effects caused by endogenous nitrogen and de novo synthesis of microbial protein. Nevertheless, more researches are needed to identify factors that affecting the accuracy of the technique.

References

- Calsamiglia, S., A. Ferret, C. K. Reynolds, N. Kristensen, N., and A. M. van Vuuren, 2010. Strategies for optimizing nitrogen use by ruminants. *Anim.* 4:1184–1196.
- Edmunds, B., K.-H. Südekum, H. Spiekers, M. Schuster and F.J. Schwarz, 2012. Estimating utilisable crude protein at the duodenum, a precursor to metabolisable protein for ruminants, from forages using a modified gas test. *Anim. Feed Sci. Technol.* 175: 106-113.
- Vaga, M., M. Hetta and P. Huhtanen, 2014. Effects of heat treatment on protein feeds evaluated in vitro utilizing the method utilisable protein. *Proceedings*

The effect of Mao pomace supplementation in diets on blood parameters of meat ducks

Srisuda Sirilaophaisan*¹, Pongsatorn Gunun¹, Chamnanwit Promkote¹, Kitsanathon Sintala¹, Paiwan Punyakaew¹ & Kessara Ampaporn¹

¹Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology Isan Sakon Nakhon Campus 47160

Abstract

The objective of this study was to determine the effects of dietary supplementation with Mao pomace on blood parameters in meat duck. One hundred and twenty 1-d-old Cherry Valley duck were allocated to three treatments with four replicates based on a Completely Randomized Design. Dietary treatments were included 0% (Control), 0.5% and 1.0% Mao pomace powder. Blood parameters of ducks aged 54 days were determined for hematocrit, red blood cell count, white blood cell count, heterophil, lymphocytes, cholesterol and triglyceride. The results showed that ducks fed with Mao pomace at level of 0.5% significant decrease ($P < 0.05$) in cholesterol and triglyceride concentrations.

Keywords: Mao pomace, meat duck, blood parameters

**Corresponding author: sreesuda88@hotmail.com*

Introduction

Mao pomace (MP), a residue that remains after juice extraction by pressing Mao in the wine and juice industry, consists mainly of seeds and marcs, accounts for approximately 25% of the fresh weight of the Mao. Phytochemicals in wastes from Mao wine and juice industry, both seeds and marcs have benefits, including antioxidant, anti-inflammatory, anti-apoptotic and anti-carcinogenic activities (Puangpronpitag et al., 2011). Mao seeds and Mao marcs to be an abundant source of polyphenols (97.32 to 130 mg gallic acid equivalents/g) and proanthocyanidins (Puangpronpitag et al., 2008). Previous study revealed that Mao pomace is an excellent potential feed source for ruminant animal (Suphanphuwong et al., 2012; Gunun et al., 2014). Moreover, Sirilaophaisan et al. (2015) suggested that diet with 0.5% MP can improve growth performance of Cherry Valley ducks. Even though several studies have focused on the productive performance, little is known about the effects of MP on the blood parameters. Therefore, the objective of the current study was to evaluate the addition of MP on blood parameter in meat ducks.

Materials and Methods

A total of 120 one day old Cherry Valley ducks were weighed and allotted into 3 treatment groups consisting of 4 replicates with 10 birds per replicate using Completely Randomized Design. All birds were raised in open house system. The experimental basal diets were based mainly on broken

rice and soybean meal as control. MP powder supplemented diets were prepared by adding 0.5 and 1.0%. At 54 days old, blood samples were used instantly to measure ratio of heterophil to lymphocytes, hematocrit, red blood cells count (RBC) and white blood cell count (WBC). Serum lipids were assessed by enzymatic colorimetric tests (CHOD-PAP method). Data were statistically analyzed by polynomial contrasts and Duncan's New Multiple-Range Test.

Results and Discussion

The effects of MP on blood parameters in Cherry Valley duck were shown in Table 1. There were no differences in hematocrit, RBC, heterophil, lymphocyte and the H/L ratio among the treatments. However, WBC, cholesterol and triglyceride were decreased in the groups fed the diets supplemented MP compared to the control group ($P < 0.05$). Little information is available about dietary MP on blood hematology and biochemistry. The lower cholesterol and triglyceride in duck fed with MP diets might be due to its effects on lipid metabolism. It was found that MP could depress intestinal lipid absorption, intestinal chylomicron and liver very low density lipoprotein secretions (Osakabe and Yamagishi, 2009; Ngamukote et al., 2011), inhibit intestinal lipoprotein secretion (Vidal et al., 2005) and inhibit cellular cholesterol. These findings agree with Zhai et al. (2014), who fed tilapia (*Oreochromis Niloticus*) with grape seed proanthocyanidins, which levels of triglyceride and total cholesterol were significantly lowered than control. Furthermore, Kukongviriyapan et al. (2013) reported that dietary supplementation of MP may be useful to prevent oxidative stress and hypertension in rats. In conclusion, the current study demonstrated that supplementation of MP powder at level of 0.5% in diets can decrease WBC and blood cholesterol and triglyceride of meat ducks.

Table 1. Effect of supplementation Mao pomace on blood parameters of Cherry Valley ducks.

Items	Control	Mao pomace (%)		SEM	<i>P</i> -value	
	No added	0.5	1.0		Linear	Quadratic
Hematocrit (%)	36.25	30.00	32.00	2.139	0.194	0.149
RBC ($\times 10^6/\text{mm}^3$)	2.40	2.25	2.18	0.118	0.209	0.800
WBC ($\times 1000/\text{mm}^3$)	17425 ^a	16025 ^{ab}	15200 ^b	643.665	0.037	0.724
Heterophil (%)	15.50	15.75	16.25	0.858	0.552	0.908
Lymphocyte (%)	84.50	84.25	83.75	0.858	0.552	0.908
H/L Ratio	0.183	0.188	0.194	0.012	0.557	0.981
Cholesterol (mg/dl)	176.50 ^a	160.00 ^b	180.00 ^a	2.814	0.402	0.001
Triglyceride (mg/dl)	213.25 ^a	163.75 ^b	218.75 ^a	3.343	0.275	<.0001

^{a-b} Mean with different superscripts within the same row differ significantly ($P < 0.05$).

Acknowledgement

The authors wish to thank the Faculty of Natural Resources, Rajamangala University of Technology Isan Sakon Nakorn Campus, Sakon Nakhon for the facility during the experiment.

References

- Gunun P., Anantasook, N., Wanapat, M., Sirilaophaisan, S., Kaewwongsa, W., Sakunkhu, S., Ampaporn, K. and Yuangklang, C. 2014. Influence of Mao pomace supplementation on degradability and *in vitro* gas production. *Khon Kaen Agr J.* 42 (suppl 1): 295–300.
- Kukongviriyapan U., Donpunha, W., Pakdeechote, P., Kukongviriyapan, V., Pannangpetch, P., Sripui, J. and Sae-Eaw, A. 2013. Effect of Mamao Pomace on the reduction of blood pressure in L-Name-induced hypertensive rats. *Srinagarind Med J.* 28(suppl.):266-270.
- Osakabe, N. and Yamagishi, M. 2009. Procyanidins in theobroma cacao reduce plasma cholesterol levels in high cholesterol-fed rats. *J. Clin. Biochem. Nutr.* 45:131-136.
- Ngamukote, S., Makynen, K., Thilawech, T. and, Adisakwattana, S. 2011. Cholesterol-lowering activity of the major polyphenols in grape seed. *Molecules* 16:5054-5061.
- Puangpronpitag, D., Areejitranusorn, P., Boonsiri, P., Suttajit, M. and Yongvanit, P. 2008. Antioxidant activities of polyphenolic compounds isolated from *Antidesma thwaitesianum* Mull. Arg. Seeds and marcs. *J. Food Sci.* 73(9):648-653.
- Puangpronpitag, D., Yongvanit, P., Boonsiri, P., Suttajit, M., Areejitranusorn, P., Na, H.K. and Surh, Y.J. 2011. Molecular mechanism underlying anti-apoptotic and anti-inflammatory effects of Mamao (*Antidesma thwaitesianum* Mull.Arg.) polyphenolics in human breast epithelial cells. *Food Chem.* 127:1450-1458.
- Sirilaophaisan, S., Gunun, P., Ampaporn, K., Punyakaew, P. and Hongladdaporn, C. 2015. Effects of Mao pomace supplementation on the growth performance of Cherry Valley duck. *Khon Kaen Agr J.* 43 (suppl 1):386–391.
- Suphanphuwong D., Asa, U.N., Bureenok, S., Vasupen, K., Wongsuthavas, S., Paengkoum, P., Wachirapakorn, C., Wanapat, M. and Yuangklang, C. 2012. Effects of mao pomace supplementation on feed intake and nutrients digestibility in goat. *Khon Kaen Agr J.* 40 (suppl 2):222–229.
- Vidal, R., Hernandez-Vallejo, S., Pauquai, T., Texier, O., Rousset, M., Chambaz, J., Demignot, S., Lacorte, J.M. 2005. Apple procyanidins decrease cholesterol esterification and lipoprotein secretion in Caco-2/TC7 enterocytes. *J. Lipid Res.* 46:258-268.
- Zhai, S.W., Lu, J. J. and Chen, X.H. 2014. Effects of dietary grape seed proanthocyanidins on growth performance, some serum biochemical parameters and body composition of tilapia (*Oreochromis Niloticus*) fingerlings. *Italian J. Ani. Sci.* 13(3).

Effects of cassava treated lactic acid supplementation on dry matter degradability and rumen fermentation in beef cattle

Pilachai R., W. Thongdee, Y. Chumpol, N. Anantasook & P. Napasirth

Faculty of Technology, Udon Thani Rajabhat University, 41000, Thailand

Abstract

Objectives of this study were to investigate the effect of lactic acid (LA) treated cassava in rations with low or high rice straw on dry matter degradability and rumen fermentation in beef cattle. Four experimental rations contained either high-forage (HF, 30% DM of rice straw) inclusion either 35% of cassava treated water (UTC) or LA treated cassava (LA-TC) ration or low-forage (LF, 15% DM of rice straw) inclusion either 50% of UTC or LA-TC ration. The effect of the experimental rations on dry matter degradability in rumen fluid was performed using gas technique. The in vivo trial, four fistulated beef cattle were each fed the four experimental rations with a 4×4 Latin square with 2×2 factorial arrangement treatment. Results of the in vitro trial indicated that the level forage, but not the LA-TC affects the cumulative gas production. Irrespective of LA-TC, gas production was significantly higher ($P<0.05$) for the LF rations compared with the HF rations. The in vivo trial showed that DM intake was not affected ($P=0.32$) by dietary treatments. Mean rumen pH was lower ($P=0.04$) for the LF rations compared to the HF rations. The supplementation of LA-TC significantly ($P=0.01$) increased rumen pH compared to UTC rations. Total VFA concentrations in the rumen fluid were significantly greater ($P=0.05$) for the LF rations compared to the HF rations, but was not affected by the supplemental LA-TC. All dietary treatments had no effect on the concentration of lactate in rumen fluid. In conclusion, both supplemental low and high LA-TC did not affect cumulative gas production and rumen fermentation. However, feeding a low forage ration containing high LA-TC was shown to improve rumen pH. The results reveal that the amount of forage in the ration is an important element in improving rumen pH.

Keywords: cassava treated lactic acid, gas production, rumen fermentation, beef cattle

Introduction

Feeding high rapidly fermentable starch such as cassava in combination with low forage content may increase the risk of rumen acidosis in dairy cattle. Reducing the rate of ruminal degradation starch (undegradable starch) using a chemical method may prevent rumen acidosis. It has been shown by Liljeberg et al. (1995) that lactic acid modified starch degradation by decreasing the rate of amylolysis, thereby decreasing the rate of fermentation in the rumen. Furthermore, treating cassava with lactic acid decreased the degradation rate of dry matter and starch contents in the rumen (Rittichai et al., 2015). However, it is unknown whether cassava treated lactic acid supplementation in different forage to concentrate ratio rations would improve rumen pH. Therefore, two experiments were conducted to investigate the effect of LA-TC supplementation on dry matter degradability and rumen fermentation in beef cattle fed rations containing two level of forage to concentrate ratios.

Materials and Methods

Experimental designs and Animals

An in vitro gas technique was carried out to determine the effect of the experimental rations on dry matter degradability in rumen fluid according the method described by Menke & Steingass (1988). In vivo trial, four, rumen-fistulated beef cattle were randomly assigned to four experimental rations in a study with a 2 × 2 factorial arrangement in a 4 × 4 Latin square with 21-periods. Each period consisted of a 16 days adaptation period, followed by a 5 days experimental period.

Experimental rations and data collection

Prior to mix the ration, cassava chip was finely ground, thereafter cassava was treated with 1% of lactic acid (DL-lactic, 85%, wt/wt) or water (untreated cassava) in a ratio of 1 to 1 wt/vol. for 48 h. During the first 16 days of each run-in/wash out period, the cattle were offered a total mixed ration diet (TMR) in a ratio of 50:50 of rice straw to concentrate *ad libitum*. During the 5 days of each experimental period, cattle were offered 4 types of experimental TMR *ad libitum*. The rations contained high-forage (HF) inclusion either low UTC or LA-TC (F:C=30:70, 35% UTC or LA-TC) or low-forage (LF) inclusion either high UTC or LA-TC (F:C=15:85, 50% UTC or LA-TC).

On day 5 of each experimental period, pH of rumen contents was recorded 15 min before feeding and 9 times hourly after feeding and rumen fluid samples were taken from the ventral sac of the rumen, 15 min before feeding and 3, 6 and 9 h after the morning feeding for analysis of VFA and lactic acid.

Statistical analysis

Cumulative gas production data were calculated according to the model of Ørskov & McDonald (1979). All data were subjected to analysis of variance (ANOVA) with cow, experimental period and dietary treatments as factors and interaction between rice straw and LA treated cassava using SPSS version 20. Significant differences between treatments were considered when $P < 0.05$.

Results and Discussion

The effect of dietary treatments on DM intake and selected indices of rumen fermentation in cattle are shown in Table 1. Results of the in vitro trial indicated that the level forage, but not the LA-TC affects the cumulative gas production. The cumulative gas production of the LF rations were significantly higher ($P < 0.05$) compared with the HF rations.

The in vivo trial showed no difference ($P = 0.32$) in DM intake. Mean DM intake of the ration ranged from 8.2-9.6 kg/day. An interaction of F:C ratio and LA-TC was observed for rumen pH ($P = 0.031$). Mean rumen pH was lower ($P = 0.04$) for the LF rations compared to the HF rations. The supplementation of LA-TC significantly ($P = 0.01$) increased rumen pH compared to UTC rations. Mean postprandial rumen pH below 5.6 for at least 3 h was observed only in the cattle fed the LF rich in UTC ration. Total VFA concentrations in the rumen fluid were significantly greater ($P = 0.05$) for the LF rations compared to the HF rations, but was not affected by the supplemental LA-TC. All dietary treatments had no effect on the concentration of lactate in rumen fluid.

The present study shows that the dietary supplementation of LA-TC and high level of rice straw improved rumen pH. The increase in rumen pH is most likely that the reduction rate of LA-TC fermentation in the rumen. These results were consistent with the reported by Iqbal et al. (2009) that LA treated barley modulated rumen fermentation and increased rumen pH. The underlying mechanisms by which lactic acid reduce the degradation of cassava starch is not clear. However,

it was hypothesized that LA has the potential to modified the starch granules by which limited the action of amylases (Liljeberg et al., 1995) and provided barrier for enzymatic degradation (Östman et al., 2002), thereby decreasing the rate of digestibility in the rumen.

Table 1. Effect of dietary treatments on DM intake and selected indices of rumen fermentation in cattle¹.

	High forage		Low forage		SEM	P-value		
	UTC ³	LA-TC ⁴	UTC	LA-TC		R ⁵	C ⁶	R×C
Gas production (ml/g OM) ²	115.6	117.8	123.4	127.3	5.41	0.01	0.22	0.84
DMI (kg/d)	9.1	9.6	8.1	8.7	1.14	0.12	0.35	0.95
Rumen pH	6.0	6.1	5.5	6.1	0.18	0.04	0.01	0.03
Total VFA (mM)	78.5	81.6	93.9	84.1	6.54	0.05	0.41	0.13
Lactate (mM)	2.1	1.8	1.9	2.2	0.37	0.15	0.51	0.43

¹ DMI values are means in five consecutive days and selected indices of rumen fermentation are means of 3 post-prandial values in each period. ²Gas production data were calculated according to the model of Ørskov & McDonald (1979). ³UTC=untreated cassava, ⁴LA-TC= lactic acid treated cassava, ⁵R= Effect of rice straw, ⁶C= Effect of cassava treatment.

Conclusion

Supplemental low and high LA-TC did not affect cumulative gas production and rumen fermentation. However, feeding a low forage ration containing high LA-TC was shown to improve rumen pH. The results reveal that the amount of forage in the ration is an important element in improving rumen pH and may be the most significant factor in preventing rumen acidosis rather than rumen undegradable starch.

Acknowledgment

The study was financially supported by Udon Thani Rajabhat University, Thailand.

References

- Iqbal, S., Q. Zebeli, A. Mazzolari, G. Bertoni, S. M. Dunn, W. Z. Yang and B. N. Ametaj, 2009. Feeding barley grain steeped in lactic acid modulates rumen fermentation patterns and increases milk fat content in dairy cows. *J. Dairy Sci.* 92: 6023–6032.
- Liljeberg, H. G. M., C. H. Lonner and I. M. E. Bjorck, 1995. Sourdough fermentation or addition of organic acids or corresponding salts to bread improves nutritional properties of starch in healthy humans. *J. Nutr.* 125: 1503–1511.
- Menke, K. H. and H. Steingass, 1988. Estimation of the energetic feed value obtained from chemical analysis and gas production using rumen fluid. *Anim. Res. Dev.* 28: 7-55.
- Ørskov, E. R. and I. McDonald, 1979. The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. *J. Agric. Sci.* 92: 499-503.
- Östman, E. M., N. Nilsson, H. G. Lilberg, M. Molin and I. M. E. Bjorck, 2002. The effect of lactic acid on blood glucose and insulin responses to cereal products: mechanistic studies in healthy subjects and *in vitro*. *J. Cereal Sci.* 36: 339–346.
- Rittichai Pilachai, Walailuck Kaewwongsa, Anan Petlum, Yauvapol Chumpol and Weerachai Thongdee, 2014. Rumen digestibility of lactic acid steeped cassava using nylon bag technique. *Khon Kaen Agric. J.* 42 (4): 567-57.

Feeding value of dried cashew nut testa in finishing pigs: Effects on growth performance, economic return and carcass characteristics

*P. Poommarin**¹, *R. C. Sulabo*², *R. A. Vega*², *D. P. T. Depositario*³ & *C. C. Sevilla*²

¹*Faculty of Animal Sciences and Agricultural Technology, Silpakorn University, Thailand,*

²*Animal and Dairy Sciences Cluster, College of Agriculture, University of the Philippines Los Baños, Philippine,* ³*College of Economics and Management, University of the Philippines Los Baños, Philippines*

Abstract

A total of 48 LYD finishing pigs (initial BW of 82.1 kg) were used to determine the effect of dried cashew nut testa (DCNT) on growth performance, economic return, and carcass characteristics in finishing pigs. Pigs were randomly allotted to 8 treatments using a 4 × 2 factorial arrangement in a randomized complete block design. Diet and gender were the factors. Dietary treatments were: corn-SBM diet with no DCNT and corn-SBM diet with 5% unsoaked DCNT (unsoaked DCNT), 5% unsoaked DCNT with added fat (unsoaked DCNT + fat), and 5% soaked DCNT (soaked DCNT). No interactions ($P > 0.23$) were observed on growth performance. There were no differences in ADG, ADFI, F/G, final BW, and caloric efficiency among pigs fed the different diets. Barrows were heavier (106 kg; $P < 0.05$) and had greater (1.04 kg/d; $P < 0.01$) ADG than gilts (98 kg and 0.87 kg/d, respectively). Margin over feed cost ranged from 649.8 to 711.6 baht/pig, with pigs fed the unsoaked DCNT having the least net margin while pigs fed the soaked DCNT having the greatest net margin. There were no ($P > 0.20$) interaction and diet effects on carcass characteristics. However, barrows had heavier (80 kg; $P < 0.05$) hot carcass weight than gilts (73.82 kg). Back fat thickness at P2 and LSQ of gilts were better ($P < 0.05$) than barrows. Therefore, in areas where DCNT is abundant, readily available, and cheap, inclusion of 5% soaked DCNT may be used in finishing pig diets.

Keywords: alternative feedstuff, dried cashew nut testa, growth performance, carcass characteristics, pigs

**Corresponding author: noopoommarin@hotmail.com*

Introduction

Feed cost accounting for about 60 to 80% of the total cost of producing finishing pigs, attaining profitability is the basic challenge of pig production. The major feed ingredients used as energy sources in swine diets in Thailand are corn and broken rice. Price of corn also increased 22.31% from 2009 to 2011 (Thai Feed Mill Association, 2011). Pig producers frequently seek low cost feed alternatives, especially when traditional feeds are expensive. One of the by-products of cashew nut process is dried cashew nut testa (DCNT), which is the skin of nut kernel, normally used either as fertilizer or disposed as agricultural waste. However, an alternative use for DCNT may be as feedstuff for pigs. The cost of DCNT is also less than other oilseed co-products (Pawatsak, 2012). Currently, there are no studies evaluating DCNT as a feed ingredient in swine diets. Hence, the feasibility of using DCNT as an ingredient in swine diets should be studied to optimize its utilization toward effects of growth performance, economic return and carcass characteristics.

Material and methods

Animals and treatments: A total of 48 LYD finishing pigs (24 barrows and 24 gilts) with an initial BW of 82.1±2.1 kg were used. Pigs were blocked by weight and randomly allotted to 1 of 4 treatment diets (Table 1). There were 6 replication pigs per treatment with 1 pig per pen. Treatments were arranged in a 4×2 factorial with treatment diet (corn-SBM, unsoaked DCNT, unsoaked DCNT+fat and soaked DCNT) and gender (barrows and gilts) as factors. Diets were formulated as follow nutrient requirement of NRC (2012) for finishing pigs (80-95 kg). Soaked DCNT using in diets was soaked by water for 24 hr. Diets and feed ingredients were analyzed for DM, CP, EE, ADF and NDF (AOAC Int., 2005). Analysis of GE was using isoperibol bomb calorimetry (CAL2K). Condensed tannins in diets and DCNT were analyzed by following Terrill et al., (1992).

Table 1. Diet composition (as-fed basis)

Ingredient (%)	corn-SBM	Unsoaked DCNT	Unsoaked DCNT+FAT	Soaked DCNT
Corn	67.51	62.51	62.51	62.51
Soybean meal, CP 44%	19.65	19.69	15.73	19.69
Cassava meal	10.00	10.00	10.00	10.00
Unsoaked DCNT	-	5.00	5.00	-
Soaked DCNT	-	-	-	5.00
Palm oil	-	-	3.70	-
Monocalcium phosphate, P 21%	1.05	1.05	1.06	1.05
Limestone	0.80	0.78	0.80	0.78
Salt	0.40	0.40	0.40	0.40
L-Lys HCl	0.20	0.19	0.31	0.19
L-Threonine	0.07	0.06	0.12	0.06
DL-Methionine	0.02	0.02	0.05	0.02
L-Tryptophan	0.00	-	0.02	-
Choline choride	0.02	0.02	0.02	0.02
Vitamin-mineral premix ^{1/}	0.25	0.25	0.25	0.25
Cr ₂ O ₃	0.03	0.03	0.03	0.03
Total	100.00	100.00	100.00	100.00
<i>Calculated composition</i>				
Standardized ileal digestible (SID) AA				
Lys, %	0.80	0.80	0.80	0.80
Met, %	0.24	0.24	0.24	0.24
Thr, %	0.51	0.51	0.51	0.51
Trp, %	0.15	0.16	0.15	0.16
Met+Cys, %	0.51	0.49	0.44	0.49
DM, %	88.22	88.42	85.16	88.42
ME, kcal/kg	3,264	3,096	3,264	3,096
CP, %	14.50	14.89	13.15	14.89
CF, %	3.38	3.89	3.63	3.89
Ca, %	0.55	0.55	0.55	0.55
total P, %	0.41	0.40	0.41	0.40
available P, %	0.25	0.25	0.25	0.25
Condensed Tannins (g/kg DM)	2.28	5.04	4.82	3.87

^{1/}The vitamin-mineral premix provided the following quantities of vitamins and micro minerals per kilogram of complete diet: Vitamin A, 3,200,000 IU; vitamin D3, 600,000 IU; vitamin E, 3,000 g. vitamin K3, 0.5000 g.; thiamin, 0.304 g.; riboflavin, 0.960 g.; pyridoxine, 0.656 g.; cyanocobalamin, 6.00 mg.; niacin, 6.000 g.; D-pantothenic acid, 4.8000 g.; folic acid, 0.2000 g.; biotin, 6.000 mg; ascorbic acid 4.000 g.; choline choride 20.00 g.; Cu, 40.000 g; Fe, 50.000 g.; Mn 16.000 g.; Zn 40.000 g.; Co 0.240 g.; Se 0.040 g.

Sampling and measurements: Pigs were allotted to consume feed and water *ad libitum* from feeder and an automatic waterer. Pigs and diets were weighted on beginning and final of experiment to calculate average daily gain (ADG), average daily feed intake (ADFI), feed/gain (F/G) and caloric efficiency.

Analyses of economic return: The economic return analyses of finishing diets were separately done for both male and female pigs. Diet costs used in these analyses were based on ingredient prices (Baht) in Thailand when the study was undertaken on March, 2013 and live weight price of pigs were based on August, 2013. The economic return such as feed cost per gain (FCG), value of gain per pig, feed cost per pig and margin over feed cost were determined.

Carcass characteristics and entrails weight: The final body weight was about 95 to 100 kg. Pigs were fasted for 24 hr prior to slaughter (Ibarra, 1983), weighed, and then slaughtered. Carcass characteristics such as slaughter weight, hot carcass weight, dressing percentage, carcass length, P2 back fat thickness, loin eye area (LEA), Lenden-Speck-Quotient (LSQ) (Mitchothai et al, 2007) and entrails weight (large intestine (LI), small intestine (SI), stomach, heart, lungs, liver, spleen and kidneys) were evaluated.

Statistical analysis: Data were analyzed as a randomized complete block design using the MIXED procedure of SAS (SAS Inst., Inc. Cary, NC) with pig as the experimental unit. The model included diet, gender and diet \times gender interaction as fixed effects, and block as the random effect. Least square means were calculated for each independent variable and means were separated using the PDIFF option of SAS. Backfat thickness at P2, loin eye area and LSQ were adjusted to a common carcass weight by using hot carcass wt as a continuous covariate in the statistical model. A level of significance was set at $P \leq 0.05$ for all statistical tests and considered a trend at $P \leq 0.10$.

Results and discussion

Growth performance: There were no diet \times gender interaction in any of the growth performance parameters measured; therefore, only the main effect of diet and gender will be discussed (Table 2). Previously, Donkoh et al. (2012) evaluated the nutritional value of DCNT (0.18% of condensed tannins) by using laboratory rats as a model for pigs. Using DCNT at 0, 5, 10, and 15% by replacing corn in the rat diet. They found the ADG in rats fed with 10 and 15% DCNT was reduced. The diet containing 5% DCNT had the best feed efficiency among the treatments. Pour and Edriss (1997) was reported the effects of dietary sorghum with different CTs (1.05 to 6.02%) and tallow supplementation (3% and 6%) on the performance of broiler chicks was evaluated. Live weights were not significantly affected when 3% tallow was added to the diets containing different concentrations of tannin. However, addition of 6% tallow increased BW and improved F/G significantly at the highest intake of tannin. Barrows had greater ($P < 0.04$) ADG and final BW and tended ($P = 0.10$) to have better F/G than gilts, which agrees with previous reports (Augspurger et., 2002; Latorre et al., 2003).

Table 2. Effect of dried cashew nut testa (DCNT) and gender on growth performance and economic return of finishing pigs (Main effects)

Item	Diet ^{1/}				SEM	Gender			P-value		
	corn-SBM	Unsoaked DCNT	Unsoaked DCNT+FAT	Soaked DCNT		Barrows	Gilts	SEM	Diet	Gender	Diet × Gender
Initial BW, kg	81.7	82.5	82.3	81.9	2.1	84.4	79.9	2.9	0.80	0.27	0.81
ADG, kg	0.97	0.91	0.94	0.98	0.04	1.04	0.87	0.03	0.64	<0.001	0.55
ADFI, kg	2.60	2.60	2.62	2.61	0.09	2.72	2.49	0.10	0.99	0.12	0.33
F/G	2.74	2.93	2.81	2.77	0.15	2.67	2.98	0.13	0.68	0.10	0.23
Final BW, kg	102.1	101.7	102.1	102.1	2.1	106.1	97.9	2.7	0.99	0.04	0.86
Caloric efficiency, Mcal ME:kg of gain	8.93	9.17	9.18	8.59	0.50	8.49	9.45	0.41	0.77	0.10	0.26
Cost of feed, Baht	13.54	13.12	13.54	13.27		-	-				
Feed cost per pig, Baht/pig	738.8	715.1	745.8	728.0		764.6	699.3				
Value of gain ^{2/} , Baht/pig	1,422.6	1,336.5	1,381.3	1,435.3		1,517.49	1,270.35				
Feed cost per kg gain, Baht/kg	37.06	38.88	38.10	36.82		35.66	39.77				
Margin over feed cost ^{3/} , Baht	683.8	649.8	635.5	701.35		752.91	568.07				

^{1/}corn-SBM = corn-soybean meal based diet; Unsoaked DCNT = corn-soybean meal based diet with 5% untreated DCNT; Unsoaked DCNT+FAT = corn-soybean meal based diet with 5% untreated DCNT and 3.7% added palm oil; Soaked DCNT = corn-soybean meal based diet with 5% DCNT pre-treated by soaking in distilled water for 24 h

^{2/}Value was determined by using a live weight price of 69.85 Baht/kg.

^{3/}Margin over feed cost = value of gain - feed costs during trial period.

Table 3. Effect of dried cashew nut testa (DCNT) on carcass quality of finishing pigs (Main effects)

Item	Diet ^{1/}				SEM	Gender			P-value		
	corn-SBM	Unsoaked DCNT	Unsoaked DCNT+FAT	Soaked DCNT		Barrows	Gilts	SEM	Diet	Gender	Diet × Gender
Live wt, kg	101.3	101.0	100.4	101.8	2.2	104.9	97.4	1.5	0.97	0.01	0.94
Dressing percentage, %	75.9	75.9	76.7	75.6	0.51	76.2	75.8	0.43	0.51	0.43	0.85
Carcass length, cm	76.8 ^a	74.2 ^{ab}	73.1 ^b	75.9 ^{ab}	0.99	75.2	74.7	0.69	0.05	0.61	0.91
Back fat thickness at P2 ^{2/} , cm	2.37	2.20	2.37	2.23	0.21	2.48	2.10	0.19	0.76	0.03	0.27
LEA ^{2,3/} , cm ²	52.7	54.4	53.5	55.4	2.8	51.9	56.1	2.3	0.90	0.17	0.20
LSQ ^{2,4/}	0.17	0.16	0.13	0.15	0.01	0.17	0.13	0.01	0.30	0.01	0.88
Intestinal wt, % of hot carcass											
SI	2.75	2.71	2.80	2.71	0.11	2.72	2.76	0.08	0.93	0.70	0.72
LI	2.33	2.43	2.49	2.37	0.10	2.41	2.40	0.07	0.65	0.95	0.52
Stomach	0.79	0.75	0.76	0.84	0.02	0.79	0.78	0.02	0.07	0.52	0.46
Organ wt, % of hot carcass											
Heart	0.48	0.49	0.46	0.46	0.02	0.47	0.47	0.01	0.52	0.52	0.86
Liver	2.12	2.09	2.01	2.17	0.07	2.07	2.12	0.05	0.46	0.54	0.52
Spleen	0.36	0.33	0.38	0.36	0.26	0.35	0.36	0.02	0.62	0.75	0.87
Kidneys	0.46 ^a	0.44 ^a	0.40 ^b	0.42 ^{ab}	0.02	0.41	0.45	0.02	0.04	0.01	0.77

^{1/}corn-SBM = corn-soybean meal based diet; Unsoaked DCNT = corn-soybean meal based diet with 5% untreated DCNT; Unsoaked DCNT+FAT = corn-soybean meal based diet with 5% untreated DCNT and 3.7% added palm oil; Soaked DCNT = corn-soybean meal based diet with 5% DCNT pre-treated by soaking in distilled water for 24 h

^{2/}Data were analyzed using hot carcass weight as a covariate

^{3/}LEA = Loin Eye Area

^{4/}LSQ = Lenden-Speck-Quotient

^{a,b} Values within a row lacking a common superscript are different ($P < 0.05$)

Economic return: Because ADFI was unaffected by treatment and adding DCNT to the diets reduced cost of feed, feed cost per pig was 23.7 baht less for pigs fed the unsoaked DCNT diet compared with the corn-SBM diet. Adding fat to the unsoaked DCNT, however, increased feed cost per pig by 30.7 baht. Value of gain was 86.1 and 41.3 baht less per pig for those fed the unsoaked DCNT diet and the unsoaked DCNT+fat diet compared with those fed the corn-SBM diet. However, the value of gain increased by 98.8 baht when pigs were fed the soaked DCNT rather than the unsoaked DCNT diet (Table 2). Feed cost per kg of gain was greatest in pigs fed the unsoaked DCNT diet. However, pigs fed the soaked DCNT diet had the least feed cost per kg gain compared to all the diets. Margin over feed cost ranged from 649.8 to 711.6 baht/pig, with pigs fed the unsoaked DCNT having the least net margin while pigs fed the soaked DCNT having the greatest net margin.

Carcass characteristics and entrails weight: There was no diet \times gender interaction effects, therefore, only main effects will be discussed (Table 3). No differences were observed on carcass characteristics among pigs fed the different diets. Lizado et al. (2005) and Gardiner et al. (2008) also found no negative effects of feeding high-tannin sorghum on carcass weight, carcass yield, and lean percentage compared with growing pigs fed corn. However, carcass length of pigs fed the unsoaked DCNT+fat was shorter ($P < 0.05$) than pigs fed the corn-SBM diet. The LSQ quotient is a carcass grading system used in Thailand (Sethakul et al., 2002). In the present experiment, the average LSQ value indicate that carcass of pigs fed the different diets corresponded to a grade of “A”, which indicates excellent carcass grading. Therefore, the results of the current study indicate that adding 5% DCNT to finishing pig diets did not negatively affect carcass characteristics and grading. Barrows had greater ($P < 0.02$) live weight and hot carcass weight than the gilts but there were no differences in dressing percentage and carcass length. Gilts had thinner ($P = 0.03$) back fat than the barrows but there was no difference in loin eye area between barrows and gilts. As a result, gilts had a lower ($P = 0.01$) LSQ value than the barrows. Testosterone promotes muscle growth. Testosterone levels are related to muscle mass and play a key role in regulating muscle mass, which is lacking in barrows (Jaturasitha et al., 2006). Protein accretion is therefore less in barrows compared with gilts, energy is then transferred to fat accretion at a higher rate (Blatzler et al., 1954). This may explain the thicker backfat and higher LSQ value in barrows compared with gilts among pigs fed the different diets and among barrows and gilts, however, there was a tendency ($P = 0.07$) for an effect of diet on stomach weight. The relative weight of the kidneys of pigs fed the unsoaked DCNT+fat diet was smaller ($P = 0.04$) than those fed the corn-SBM and unsoaked DCNT diets (Table 3).

References

- AOAC international. 2005. Official method of analysis, 18th Edition. Association of Office Analytical Chemists, Washington, D.C., USA.
- Augspurger, N.R., M.Ellis, D.N. Hamilton, B.F. Wolter, J.L. Beverly and E.R.Wilson. 2002. The effect of sire line on the feeding patterns of grow-finish pigs. *Appl. Anim. Behav. Sci.* 75:103–114
- Donkoh, Armstrong, Victoria Attoh-Kotoku, Reginald Oseikwame. 2012. Evaluation of nutritional quality of dried cashew nut testa using laboratory rat as a model for pigs. *The Scientific World Journal*. 2012, Article ID 984249 : 1-5.
- Gardiner, G.E., A.J. Campbell, J.V.O’Doherty, E. Pierce, P.B. Lynch, F.C. Leonard, C. Stanton, R.P. Ross and P.G. Lawlor. 2008. Effect of *Ascophyllum nodosum* extract on growth performance, digestibility, carcass characteristics and selected intestinal microflora populations of grower–finisher pigs. *Anim Feed Sci. Technol.* 141: 259-273.
- Ibarra, I. Perlito. 1983. Meat processing for small and medium scale operations. College of Agriculture, University of the Philippines at Los Baños, College, Laguna, Philippines.

- Jaturasitha, S., S. Kamopas, T. Suppadit, R., Khiaosa-Ard and M. Kreuzer. 2006. The effect of gender of finishing pigs slaughtered at 110 kilograms on performance, and carcass and meat quality, *ScienceAsia* 32: 297-305.
- Latorre, M.A., Medel, P., Fuentetaja, A., Lázaro, R., Mateos, G.G., 2003. Effect of gender, terminal sire line and age at slaughter on performance, carcass characteristics and meat quality of heavy pigs. *Anim. Sci.* 77: 33-45.
- Lizardo, R, Jany Peiniau and A. Aumaitre. 2005. Effect of sorghum on performance, digestibility of dietary components and activities of pancreatic and intestinal enzymes in the weaned piglet. *Anim Feed Sci. Technol.*, 56: 67-82.
- Mitthaotai, J., C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsutthavas, P. Srenanul, R. Hovenier, H. Everts and A.C. Beynen. 2007. Effect of dietary fat type on meat quality and fatty acid composition of various tissues in growing–finishing swine. *Meat Science.* 76: 95–101.
- NRC. 2012. National Research Council. Nutrient Requirements for Swine, 11th ed. National Academy Press, Washington, DC.
- Pawatsak, K. 2012. Lummanow cashew nuts factory, Chonburi, Thailand. The interviewed information (Unpublished)
- Pour-Rezal, I and M.A.Edriss. 1997. Effects of dietary sorghum of different tannin concentrations and tallow supplementation on the performance of broiler chicks. *British Poultry Science* 38: 512–517.
- Sethakul, J., R. Sitthigripong, K. Tuntivisoottikul, S. Lerdsuwon and K. Muangmusit. 2002. Grading and development of pig carcass in Thailand. Project report of Thailand Research Fund (TRF) under contract No. RDG 4220004. Bangkok, Thailand
- Terrill, T.H., A.M. Rowan, G.B. Douglas and T.N. Barry. 1992. Determination of extractable and bound condensed tannin concentration in forage plants, protein concentrate meals and cereal grains. *Journal of the Science of Food and Agriculture.* 58, p.321-329.
- Thai feed mill association. 2011. The situation of corn feed livestock. *J. of Thai feed mill business* 29(139): 27

Effect of oral administration of red ginger extract on performances of hybrid ducks

Eko Widodo¹, Bondan Prakoso² & Edhy Sudjarwo³

¹Department of Animal Nutrition, Faculty of Animal Husbandry, University of Brawijaya, Indonesia, ²Master Student in Faculty of Animal Husbandry, University of Brawijaya, Indonesia, ³Department of Animal Production, Faculty of Animal Husbandry, University of Brawijaya, Indonesia

Abstract

Red ginger is one of traditional herbs popular among people in Asia for healing and disease prevention. It has been reported to have antimicrobial effect, but some reports indicated improving performances of broiler. The purpose of the research was to evaluate and find the best level of oral administration of turmeric water extract use on performances of hybrid duck. The result was expected to contribute knowledge on the use of red ginger for hybrid duck performance improvement. In this experiment, there were 5 treatments P0: basal feed, P1: basal feed+antibiotic, P2: basal feed+red ginger extract 0.7 ml/duck/day, P3: basal feed+ red ginger extract 1.4 ml/duck/day and P4: red ginger extract 2.1 ml/duck/day with oral treatment on performances of hybrid duck. Variables in this research were feed consumption, body weight gain, feed conversion ratio and IOFC. The method of this research was experiment arranged in Completely Randomized Design and if there was a different effect among the treatments would be further tested by Duncan's Multiple Range Test. The result of this research showed that the use of red ginger extract with oral treatment highly significantly affected on feed consumption, body weight gain, feed conversion ratio and IOFC ($P < 0,01$). The conclusion of this research was oral administration of red ginger extract would improve performances of hybrid duck and it is suggested to use 2.1 ml oral administration of red ginger water extract per duck per day.

Keywords: Red Ginger, Oral Administration, Performances, and Hybrid Duck

Introduction

Duck raising in Indonesia plays an important role due to the dependence of thousands of small farmers on raising ducks. In fact, imported duck is even getting popular as roasting Peking duck becomes part of wedding party menu. An attempt was made of crossing between Campbell and Peking duck, known as hybrid duck. Though the growth rate does not as fast as Peking duck, economic consideration is more dominant so that raising hybrid duck becomes one of the alternatives for farmers. For economic success in raising hybrid duck largely depends on the price of feed and expenses for medicine, vaccine and premix. Effort was made to reduce by the use of red ginger water extract rather than antibiotic-free diet than commercial diet to reduce feed cost.

The use of red ginger has been reported to improve broiler performances (Herawati and Marjuki, 2011), and Agustina (2010) reported that combination of ginger with other herbs at 2.5 ml per liter of drinking water was able to improve body weight gain in broiler. Alternatively, a simple way of extraction by squeezing shredded *Curcuma longa* was proposed to be implemented for hybrid duck. The objective was to examine the effect of constant concentration of *Curcuma longa* extract administered orally on hybrid duck performances.

Materials and Methods

Bird management

One hundred and twenty 2 weeks old ducks were purchased from local hatchery were from crossing between female Cambell and male Peking ducks. They were distributed to 5 dietary treatments. Each treatment was repeated 4 times, and each replication contained 6 ducks. The ducks were raised in a litter floor pen of 1.25x1.00x1.00 m. Each pen was equipped by feeder and drinker, lamp and brooder facilities to maintain room temperature to meet the requirement.

Dietary treatments

The dietary treatments consisted of:

P0 = basal feed (without treatment),

P1 = basal feed added with antibiotic (containing 300mg tetracycline/kg feed)

P2 = basal feed plus orally administered with sput of 0.7 ml red ginger extract/day

P3 = basal feed plus orally administered with sput of 1.4 ml red ginger extract/day

P4 = basal feed plus orally administered with sput of 2.1 ml red ginger extract/day

Feed and water were provided *ad libitum*. The length of experiment was 5 weeks, and basal diet used as presented in Table 1

The variables measured included feed consumption, body weight gain, feed conversion, production number, mortality and Income Over Feed Cost (IOFC). IOFC was calculated by the different between the price of meat per kg multiply by weight gain and expenses of feed cost.

Statistical analysis

The data obtained were expressed as mean \pm SD. Data were then subjected to analysis of variance of Completely Randomized Design, and if the significant effect among the treatments existed then being tested by using Duncan Multiple Range Test (Yitnosumarto, 1993).

Table 1. Basal diet and its composition

Feed ingredients	Basal Diet (2 -7Weeks)
Yellow corn (%)	70.54
Rice polishing (%)	9.62
Soybean meal (%)	12.83
Fish meal (%)	5.13
Bone meal (%)	0.86
Coconut oil (%)	0.64
Premix (%)	0.21
Lysine (%)	0.09
Methionine (%)	0.09
Total (%)	100
Nutrient contents	
Metabolizable energy (kcal/kg)*	3101
Crude protein (%)	18.18
Crude fat (%)	5.43
Crude fibre (%)	3.75
Calcium (%)*	0.60
Phosphorus (%)*	0.37
Lysine (%)*	0.60
Methionine (%)*	0.35

*calculated

Results and discussion

Effect of treatments on performances of hybrid ducks are showed in Table 2.

Table 2. Effect of dietary red ginger treatment on performances of hybrid duck.

Variables	Perlakuan				
	P0	P1	P2	P3	P4
Feed Consumption (g/bird/28 days)	2429 ± 60 ^a	2415 ± 26 ^a	2637 ± 31 ^b	2558 ± 73 ^b	2581 ± 37 ^b
Body weight gain (g/bird/28 days)	685 ± 6 ^{ab}	639 ± 55 ^a	726 ± 52 ^{bc}	764 ± 15 ^{cd}	820 ± 34 ^d
Feed Conversion	3.55 ± 0.09 ^{bc}	3.80 ± 0.30 ^c	3.65 ± 0.26 ^{bc}	3.35 ± 0.11 ^s	3.15 ± 0.16 ^a
IOFC (IDR/bird/28 days)	6692 ± 290 ^a	5593 ± 1348 ^a	6892 ± 1259 ^{ab}	8112 ± 479 ^{ab}	9418 ± 937 ^{bc}

Superscript a-d in the same raw indicates highly significant effect ($P < 0.01$)

1 US\$ = IDR 11,500 and the price of live muscovy duck was Rp 25,000 /bird at the time of experiment

Feed consumption, body weight gain, feed conversion and IOFC were highly significantly affected ($P < 0.01$) by the treatments. The results indicated that there was a tendency of increasing feed consumption of hybrid duck due probably to gingerol content of red ginger which may stimulate secretion of digestive enzymes as reported by Mide (2009). Darwiset *al* (1991) also mentioned that the use of curcuma stimulate appetite of animal, of which red colour of ginger may represent curcumin content. Herawati and Marjuki (2011) mentioned that the use of 1.0 to 1.5% red ginger powder improved broiler performances. As the feed intake increase and enzyme production stimulated, body weight was then found significantly improved. Feed conversion was significantly affected ($P < 0.05$) by the treatment. Results obtained for body weight gain was consistent with feed conversion which suggest that the use of 2.1 ml curcuma/day of oral administration showed the best feed conversion. Consistent result was also obtained with IOFC was obtained in P4, suggested that oral administration of red ginger may be good alternative to be implemented by farmers.

Conclusion

Based on the result of current study, it might be concluded that the use of oral administration of 2.1 ml curcuma/bird/day could be beneficial to be implemented for hybrid duck raising.

References

- Agustina, L. 2010. Penggunaan Ramuan Herbal Sebagai *Feed Additive* Untuk Meningkatkan Performans *Broiler*. Lokakarya Nasional Inovasi Teknologi Dalam Mendukung Usaha Ternak Unggas Berdayasaing. Kota? Bogor?
- Darwis SN, Madjo Indo ABD, and Hasiyah S. 1991. Tumbuhan obat family zingiberaceae. Badan Penelitiandan Pengembangan Pertanian. Pusat Penelitiandan Pengembangan Tanaman Industri, Bogor.
- Herawati and Marjuki. 2010. The effect of feeding red ginger (*Zingiber officinale* Rosc) as phytobiotics on broiler slaughter weight and meat quality. International Journal Poultry Science 10(12): 983-986.
- Mide, M. Z. 2009. Penampilan Broiler yang Mendapatkan Ransum Mengandung Tepung Daun Katuk dan Rimpang Kunyit. <http://repository.unhas.ac.id/handle/123456789/2383>.
- Yitnosumarto, S. 1993. Percobaan Perancangan Analisa dan Interpretasinya. Gramedia Pustaka Utama. Jakarta.

The inclusion effect of mangosteen (*garcinia mangostana l.*) peel as feed additive on blood profile and testosterone level of Mojosari male duck

Irfan H. Djunaidi¹, Edhy Sudjarwo¹, & Dini Hardini²

¹Fac. of Animal Science, University of Brawijaya-Malang Indonesia

²The Assessment Institute for Agriculture Technology (BPTP) East Java, Indonesia

Abstract

The research was aimed to investigate the effect of Inclusion of Mangosteen peel meal (MPM) as feed additive on blood lipid profile and testosterone hormone level of Mojosari male duck. The material used in this study was 40 males Mojosari duck at the age of 10 months. The method used an experiment with a completely randomized Design with 4 level mangosteen inclusion on basal diet : were P0 = basal feed (without MPM), P1 = 1 , P2 = 2, and P3 = 3 g MPM/day/bird; with 5 replications (@2 male ducks each).

The variables measured were total cholesterol, triglycerides, HDL and LDL, and testosterone hormone level. The results of this research showed that inclusion of MPM significantly affected to testosterone level, but not significant effect on blood serum lipid profile. The average value of cholesterol, triglyceride, HDL and LDL and testosterone level of blood were: cholesterol (P0 = 188.0 , P1 = 162.4, P2 = 170.8 and P3 = 167.2 mg/dl), triglycerides (P0 = 182.8, P1 = 140.8, P2 = 179 and P3 = 161.8 mg/dl), HDL (P0 = 103.3, P1 = 144.28, P2 = 99.94 and P3 = 106.56 mg/dl), LDL (P0 = 45.52, P1 = 26.66, P2 = 31.04 and P3 = 28.28 mg/dl), and testosterone level (P0 = 232.8, P1 = 262.0, P2 = 372.8, and P3 = 349.4 ng/dl), respectively. The inclusion of MPM as feed additive tended to improve the lipid blood profile and testosterone level of Mojosari male duck.

Keywords: Mangosteen peel meal, Mojosari duck

Corresponding author: irjuna@ub.ac.id

Introduction

In East Java, recently a duck local culinary food improving and require more local duck meat supply. This phenomena was increase due to the friendly taste and price for most of the community people. But, concern about duck meat fortunately a high fat and cholesterol content still as awareness. Some practical effort has been done to reduce fat content, but nutritional manipulation technique can be apply with utilization of natural feed additive to reduce the fat and cholesterol of duck meat. The mangosteen peel meal (MPM) containing active compound such as α -mangosteen and xanthone is expected to decrease the levels of cholesterol, low-density lipoprotein (LDL) in blood and meat of duck. A high cholesterol and LDL blood level indicated tend to coronary heart disease and stroke. Normal cholesterol should be below 200 mg/dl.

The previous research Dachriyanus (2006) showed that addition of MPE extract caused lower cholesterol blood levels of neonatal mice at different doses. The α -mangosteen improved the lipoprotein lipase enzyme activity for hydrolysis of low density lipoprotein into fatty acids and glycerol, LDL levels down, HDL increase, and blood vessels more clear. The research was aimed

to investigate the effect of MPM as feed additive to the blood characteristic and testosterone level of the Mojosari male duck.

Material And Methods

Research was executed in Sumberseka rResearch Centre, Faculty of Animal Science Brawijaya University and blood and testosterone level analysis in medical Clinic laboratories Pattimura, Malang. A 40 male of 10 month old of Mojosari duck (*Anas platyrhynchos javanicus*) was used as material with average BW 1.5 kg. The mangosteen peel meal was prepared with low temperature drying, and the duck were fed by commercial duck feed.

The research was feeding trial experiment and designed with completely randomized design using 4 treatments 5 replications (@ 2 male ducks). The treatments were P0 = basal feed (without MPM), P1 = 1, P2 = 2, and P3 = 3 g MPM/day/bird. The basal feed given was a commercial duck feed produced by local feed industry and met the nutrient requirement, and feed treatment offered individually and restricted about 160 g/day, and the MPM added by mixed with their daily feeding ration. The water drinking was *ad libitum*. The feeding treatment was done for 6 weeks and the blood sample was taken on week 4 and 6. The blood sample was taken from the *vena pectoralis* in the duck wing. The blood lipid profile analysis was done using COD PAP method and testosterone level analyzed by *radioimmunoassay* (RIA). The data recorded to calculated average blood lipid profile (mg/dl) and Testosterone level ($\mu\text{g/dl}$) of Mojosari male duck. The collected data were subjected to statistical Anova analysis and compared by least significance difference test (Steel and Torrie, 1994).

Result and Discussions

The result of MPM treatment effect on the blood profile and testosterone level are shows on Table 1. The result showed that the MPM additive showed a positive effect both to the lipid blood profile and testosterone level. The Cholesterol, triglyceride, and LDL content tend to decreased and HDL and testosterone level increased, even statistically only testosterone level was statistically showed a significant differences.

Alfa mangosteen and xanthone as a Phenol compound can inhibit the formation of intestinal absorption of micelle site of bile acids which is one of its functions to dissolve cholesterol through bile duct into the intestine, so that in the end the body cholesterol decreased. Because of their large size so it can't be absorbed by the digestive tract and excreted through the feces straight, other than by flavonoids, reduced blood cholesterol levels allegedly is influenced also by the plant fat called phytosterol.

Table 1. The Inclusion Effect of MPM Treatment on Blood Lipid Profile (mg/dl) and Testosterone level ($\mu\text{g/dl}$) of Mojosari Male Duck.

Variables	Treatments			
	P0	P1	P2	P3
Cholesterol	188 \pm 18.11	162.4 \pm 10.43	170.8 \pm 28.67	167.2 \pm 10.66
Triglyceride	182.8 \pm 66.16	140.8 \pm 51.55	179 \pm 82.40	161.8 \pm 75.48
HDL	103.3 \pm 21.13	114.28 \pm 22.10	99.94 \pm 7.58	106.56 \pm 8.09
LDL	45.52 \pm 9.27	26.66 \pm 14.90	31.04 \pm 13.23	28.28 \pm 7.83
Testosterone	232.8 \pm 13.64 ^a	262.0 \pm 83.86 ^a	372.8 \pm 81.49 ^b	349.4 \pm 24.04 ^c

Note: a different superscript on same row means a significantly differences ($P < 0.05$).

An antioxidant needed to lower the oxidation reactions and serves to prevent or stop the ravages of free radicals into the safe bonds stop lipid peroxide (Surai, 2003). Mayes *et al.* (1997) stated that a high HDL cholesterol and low cholesterol was a normal because HDL was transported and converted to bile acids. HDL particles serve carries cholesterol from adipose tissue to the liver organ. Approximately 75-80% of the cholesterol is converted into HDL particles by the enzyme cholesterol acyl transferase lecithin (LCTA) to be transported to the liver and circulated again, so that blood cholesterol levels decrease. An increase in HDL and LDL correlated negatively and positively to risk suffering from atherosclerosis. The xanthone and alfa mangosteen can protect and neutralized the free radical effect and testosterone production increased. A high testosterone level indicate a high libido and supporting spermatogenesis (Zirkin et al., 1989)

Conclusion

The inclusion of Mangosteen peel meal as feed additive increased the testosterone level and have no effect to the total cholesterol, triglycerides, HDL, and LDL of blood of Mojosarimale duck.

References

- Dachriyanus, D.O., R.Oktarina, O.Ernas, Suhatri, and H.M. Mukhtar. 2006. Test of α -Mangostin to total cholesterol, Triglyceride, HDL, and LDL male white Mice and Lethal Dose of 50 (LD₅₀). Faculty of Sci and Math. Andalas University. Padang.
- Geca, T. O. 2010. Antibacterial activity of α -Mangosteen of Mangosteen fruit (*Garcinia mangostana* L.) to *Staphylococcus aureus* and *Aeruginosa* Multiresistent. Fac. Pharmacy. Univ. Muhammadiyah Surakarta.
- Lehninger, A. L. 1982. Introduction of Biochemistry, Part I. Translated: M. Thenawidjaja. Erlangga. Jakarta.
- Mayes, P. A., R. K. Murray, D. K. Granner, and V. W Rodwell. 1997. Biochemistry Harper. 24th Ed. Translated: Hartono, A. EGC. Jakarta.
- Nurusyifah. 2010. Analysis Marker content of α -Mangostin on boiled and dried Mangosteen peel (*Garcinia mangostana* L). Fac. Farmasi. Univ. Muhammadiyah Surakarta.
- Pavlovic V, Cekic S, Rankovic G & Stoiljkovic N. 2005. Antioxidant and Pro-oxidant Effect of Ascorbic Acid. Acta Medica Medianae. 44 (1): 65-69.
- Steel, R. G. D. dan J. H. Torrie. 1993. Principle and Procedure of Statistic. PT Gramedia Pustaka. Jakarta.
- Surai, P. F. 2003. Natural antioxidants in Avian Nutrition and Reproduction. Nottingham University Press. England.
- Zirkin, B. R. , R. Santulli, C.A. Awoniyi, and L.L. Ewing. 1989. Maintenance of advanced spermatogenic cells in the adult rat testis: quantitative relationship to testosterone concentration within the testis. Endocrinol. 124 (6):3043-3049.

The effects of quantitative restricted feeding on performances and internal organ weight broiler

Dini Hardini¹ and Irfan H. Djunaidi²

¹The Assessment Institute for Agriculture Technology (BPTP) East Java

²Faculty of animal Science, Brawijaya University, Malang-East Java Indonesia (irjuna@ub.ac.id)

Abstract

This research was aimed to evaluate the effects of quantitative restricted feeding in different time period on production performance, carcass percentage, and internal organ. One hundred and sixty unsexed broiler (Lohmann) were used in this research and designed by Completely Randomized design. The treatments was 80 % feed restriction for normal recommended feed consumption. Four levels of feeding were P0 (*ad libitum*), W3 (feed restricted on week 3), W4 (feed restricted on week 4), W5 (feed restricted for week 5). Results showed that there were no negative effects of quantitative restricted feeding on performance in broiler and internal organ weight. The treated chicks tends to have lower feed consumption and conversion, low abdominal fat carcass percentage, and no differences of growth performance, internal organ weight and physical carcass properties compared to control.

Keywords : broiler performance, quantitative restricted feeding

Corresponding author: irjuna@ub.ac.id

Introduction

Restricted feeding is one of the primary management tools currently used to reduce the incidence of metabolic disorders. Various methods of feed restriction such as intermittent feeding, skip-a-day feeding, appetite suppression, time of restriction (Yu & Robinson, 1992) and quantitative feed restriction (Lee & Leeson 2001) are used in broilers to improve their efficiency of feed utilization and weight gain. The feed restriction for reasonable periods may produce effective method to achieve the benefits of compensatory growth. In many physical feed restriction or diet dilution studies, there are reports of body fat deposition, (Zhonget al., 1995).

Khetani et al. (2009) found that restricted feeding in short period produced a better broiler performances. The broiler chickens with restricted feeding in short term have time to catch up growing after re feeding. Previous study showed that the best performance of restricted feeding was shown on 80% of the total recommended feed (Djunaidi & Hardini, 2010). This study aimed to examine the effect of quantitative feed restriction in different time period on the broiler performances, internal organ weight, and physical meat properties.

Material and Methods

The research executed in Poultry house unit of the Assessment Institute for Agricultural technology (BPTP) East Java, Malang. One hundred sixty unsexed day old MB 202 chicks purchased local poultry shop. The chicks were weighed individually and reared in group for 10 days (adaptation period). At day 11 the chicks were randomly divided into 16 experimental units of 10 chicks each. These units were further allotted randomly to 4 treatments groups W₀, W₃, W₄ and W₅, with 4 replication each in pen (80 x 100 sq.m) rice husk litter. The chicks in group T₀ were fed *ad libitum* and served as control. The bird in groups W₃, W₄ and W₅ were kept on feed restriction on 80% of recommended consumption at week 3 (day 14 -20), week 4 (day 21- 27) and week 5 (day 28 – 35), respectively.

The commercial broiler starter mash was used and birds were fed *ad libitum* until day 14. After that, the chicks were treated according to treatment using grower crumble up to the age of 5 weeks. The vitamin mix served and mixed with fresh and clean water drinking.

The data on initial body weight, weekly feed consumption, body weight were collected during experimental period. At the end of experimental period, 3 of birds from each replicate were selected randomly, weighted and slaughtered for their carcass dressed, abdominal fat, organ weight, and physical meat properties. The data recorded to calculated carcass percentage and relative weight (g/100 g BW) of abdominal fat and organ weight. The collected data were subjected to statistical Anova analysis and compared by least significance difference test (Steel & Torrie, 1994).

Result and Discussion

Table 1 showed that feed restriction no significant effect ($P > 0.05$) to the feed consumption, weight gain, feed conversion and carcass percentage. The amount of feed consumption depend on the feed energy density, body size, activity, and environment temperature.

The body weigh gain of P₀ was highest due to no feed restriction both on the starter and grower phase. Compared to the treated group, the differences tend to be lower with early feed restriction time, the average daily gain of W₃ and W₄ were higher than W₅, although it lower than W₀. This happens could be effected by the chick have time recovery after restriction feed treatment.

The average value of feed conversion of the P₃ treated chicks was lower compared to the control and other treatment groups. The lower feed conversion value derived from lower feed consumption and higher body weight gain. This could be happen due to the feed restriction on W₃ the chick was done on day 21-28, so in the last week (day 28 – 35) the animal increase their consumption and there was any compensatory growth in these period. Its means that chicks lower feed to have similar body weigh growth.

Table 1. The effect of quantitative feed restriction on production performance, internal organ weight and physical meat properties of broiler.

Parameters	Treatments			
	W ₀	W ₃	W ₄	W ₅
Production performances				
Feed consumption (g/d)	142.9±49.5	134.2±57.9	132.3±56.9	119.1±21.0
Weight gain (g/d)	73.5±10.0	68.5±20.5	69.1±19.3	66.0±10.4
Feed conversion	1.92±0.42	1.93±0.43	1.88±0.34	1.83±0.39
Carcass, %	68.49±6.14	63.62±2.54	64.17±2.77	69.23±0.94
Abdominal fat (%)	1.78±0.17 ^b	1.86±0.21 ^b	1.31±0.17 ^a	1.32±0.41 ^{ab}
Internal organ weight (%)				
Liver	1.97±0.04 ^{ab}	2.44±0.39 ^b	1.42±0.10 ^a	2.32±0.41 ^{ab}
hearth	0.48±0.11	0.49±0.02	0.48±0.10	0.54±0.11
Pancreas	0.21±0.20	0.18±0.01	0.22±0.05	0.19±0.04
Lung	0.56±0.12	0.50±0.10	0.62±0.07	0.63±0.06
Proventriculus	0.54±0.13	0.49±0.03	0.55±0.11	0.46±0.07
Gizzard	1.30±0.15	0.97±0.15	1.12±0.16	1.12±0.23
Physical meat properties				
pH	5.40±0.07	5.41±0.05	5.44±0.06	5.43±0.06
WHC (%)	33.04±1.96 ^a	33.92±0.28 ^b	36.42±1.06 ^{ab}	34.48±0.55 ^b
Water content (%)	76.74±2.54	75.45±0.77	75.3±0.75	75.76±0.66

Notes: the different superscript on the same row means significantly differences (P<0.01).

The carcass percentages have positive correlation with the final body weight. No much differences of final body weight of the treated group chicks compared to the control group. The carcass percentage of the treated chick were decrease in W3 and W4 but increase in W5. Amaefule et al., (2006) found that carcass percentage linear to final body weight and body weight gain, the higher body weight will be produce higher carcass percentage.

The other positive effects of feed restriction was decreasing of carcass fat deposition. The abdominal fat percentages of the treatment chicks were lower compared to the control group. The value of abdominal fat of W4 and W5 statistically very significantly lower compared to the control group chick. These data indicated that the fat deposition of the broiler was produce in the last period of the finisher phase. If the chicks have limited access to the feed, they will be produce lower body fat.

There was no significant effect of the feed restriction to the internal organ weight, except the liver weight. The liver weight of the treatment group chick were tended to be higher compared to the control group. The normal average of liver weight was 3 % of BW (Moran, 1982), but in this research less than 2.43 % BW, the other internal organ also have normal weight. The internal organ weight of the W5 group tend to have higher value compared to the other treatments groups. Even thought statistically, no differences effect happens among treatments.

Statistical analysis result showed that the feed restriction have significantly effect to WHC value and no effect to the pH and water content of the meat. The average pH value of the meat was 5.4 that mean still in normal pH value (5.5-5.8). The meat pH value affected to tenderness and texture, higher pH to have better tenderness and hard/solid texture, sticky, and higher WHC tend to be media of microorganism growth (Soeparno, 1998).

Conclusion

The feed restriction on the level 80 % of recommended consumption have no differences production performance, internal organ weight and physical meat properties if applied at week 3 until 5 of their life period. The best broiler performance will be produce by feed restriction on week 4 (day 22-28) without any negative effect to internal organ weight or physical meat properties.

References

- Amaefule, K.U., F.C. Iheukwumere., A. S. Lawal and A. A. Ezekwonna,2006. The effect of treated rice milling waste on performance, nutrient restriction, carcass and organ characteristics of finisher broiler. *Int. J. Poult Sci.* 5(1): 51-55.
- Djunaidi, I. and D.Hardini, 2010. Effect of quantitative feed restriction on broiler performances under hot condition. Research report. Brawijaya University. Malang. Indonesia.
- Khetani, T.I.,T.T. Nkukwana, and N. Chimonyo, 2009. Effect of quantitative feed restriction on broiler performance. *Trop. Anim. Health. Prod.* 41: 379-384.
- Lee,K.H.andS. Leeson, 2001. Performance of broilers fed limited quantities of feed or nutrients during 7-14 days of age. *Poult. Sci.*, 80: 446-454.
- NRC, 1994.*Nutrient Requirement of Poultry*.National Research Council.National Academy of Sciences, Washington D.C. Diaksestanggal 23 November 2011.
- Soeparno, 1998. *Science and Technology of Meat*. GadjahMada University Press.Yogyakarta.
- Steel, R.G.D. dan J.D. Torrie. 1994. *Principle and Procedures of statistic*. 3rd Ed.. PT. Gramedia. Jakarta
- Yu, M.W. and F.E. Robinson, 1992. The application of short term feed restriction to broiler chicken production: a review. *J.Appl. Poult. Res.*1: 147-153.
- Zhong, C., H. S. Nakaue, C.Y. Hu and L.W. Mirosh, 1995.Effect of full feed and early feed restriction on broiler performance, abdominal fat level, cellularity and fat metabolism in broiler chickens. *Poult. Sci.*, 74: 1636-1643.

Evaluation of mulberry leave as functional feed additive

Lin*, W. C. , B. Yu & T. T. Lee

Department of Animal Science, National Chung Hsing University, 250 KuoKuang Road, Taichung 402, Taiwan

Abstract

The purpose of this study is to evaluate the antioxidant activity ability of Taisung No.3 mulberry leaf (ML) as functional feed additive by a sequence of *in vitro* and *in vivo* experiments. The results presented that the total phenolic compounds were 7.4 mg GAE (gallic acid equivalent)/g dry weight for its aqueous extracts. The chelating capacity of Fe²⁺ was 71.6% when the concentration of extracts was 1.0 mg/ml. The scavenging ability of 1, 1-diphenyl-2-picrylhydrazyl (DPPH) free radical was 45.9% when added 0.1 mg/mL of extracts. At the concentration of 25 mg/ml, the extracts have approximate 84.0% reducing power compared with ascorbic acid at the concentration of 0.5 mg/ml. In 2.5 mg/mL of ML aqueous extracts have approximate equal equivalent antioxidant capacity (TEAC) compared with 0.05 mg/mL of ascorbic acid. The liposome oxidation inhibiting ability was 43.9% when added 50 mg/ml extracts. In vivo experiment, a total of 96 brown laying hens (*HENDRIX*) were assigned into 4 treatment diets including dry mulberry leaves at 0% (control), 0.5%, 1% or 2%, respectively, for 12 weeks. Each treatment had eight replicates with three hens each. The results indicated that 0.5% and 1% ML supplemented groups had significantly higher mRNA concentration of antioxidant-regulated genes and significantly lower ROS modulator 1 (ROMO1) expression. Serum malondialdehyde (MDA) content was lower and catalase activity was higher as well as superoxide dismutase (SOD) activity in all ML supplemented groups compared to the control group. Egg mass and feed conversion rate (FCR) were significantly improved, while 1% ML supplementation had overall best effects. The Egg yolk weight, egg shell weight, egg shell strength, shell thickness, egg yolk color and Haugh unit were increased among all the groups supplemented with ML powder. The highest ML supplemented dose (2%) in this study did not result in the best effect. Instead, 0.5% ML supplementation had exerted the overall best effect. In conclusion, the mulberry leaves could be used as a functional feed additive to enhance performance and oxidant status of laying hens.

Keywords: Mulberry leaves, antioxidants, laying hens, feed additives.

Introduction

Oxidative damage is one of the major concerns in poultry production, it occurs when exogenous and/or endogenous reactive oxygen species (ROS), which is associated with free radical formation and oxidative stress. (Sosa et al., 2013). These factors would depress health status and adversely affects production parameters leading to heavy economic losses (Zaviezo, 1999). Increasing

literatures demonstrated that mulberry leaves exhibit high antioxidant activity attributed to abundant and various kinds of (poly) phenolic substances (Jeszka-Skowron et al., 2014). Due to the decreasing application in silkworms in Taiwan, the mulberry leaves have been regarded as a valuable low-cost material with proven biological activities. These facts pave the way to the revalorization of this underused botanical material in order to conduct further nutritional application with powerful antioxidant property.

Material and Methods

Bioactive components and antioxidants of ML extracts were tested *in vivo*. *In vivo* experiment, a total of 96 brown laying hens (*HENDRIX*) were assigned into 4 treatment diets including dry mulberry leaves at 0% (control), 0.5%, 1% or 2%, respectively, for 12 weeks. Laying performance, egg quality, serum traits and relative RNA concentration of antioxidant-regulated genes in chicken PBMCs were then determined.

Results

In vitro results presented that the total phenolic compounds were 7.4 mg GAE (gallic acid equivalent)/g dry weight for its aqueous extracts. The chelating capacity of Fe^{2+} was 71.6% when the concentration of extracts was 1.0 mg/ml. The scavenging ability of DPPH free radical was 45.9% when added 0.1 mg/mL of extracts. At the concentration of 25 mg/ml, the extracts have approximate 84.0% reducing power compared with ascorbic acid at the concentration of 0.5 mg/ml. In 2.5 mg/mL of ML aqueous extracts have approximate equal equivalent antioxidant capacity (TEAC) compared with 0.05 mg/mL of ascorbic acid. The liposome oxidation inhibiting ability was 43.9% when added 50 mg/ml extracts.

The results indicated that 0.5% and 1% ML supplemented groups had significantly higher mRNA concentration of antioxidant-regulated genes and significantly lower ROMO1 gene expression. Serum MDA content was lower by maximum at 60% and catalase activity of 0.5% ML supplemented group was 5.6% higher as well as SOD activity was 92.1% higher compared to the control group. FCR of the 0.5%, 1%, 2% ML supplemented groups were significantly improved by 3.5%, 4.6% and 4.0%, respectively, while 1% ML supplementation had overall best effects. Supplementation with ML showed effects on egg quality. Egg yolk weight was improved by 3.9%, 4.3% and 4.4%, respectively. Egg shell weight was increased by 3.3%, 4.4% and 2.6%, respectively. Dietary ML (0.5%, 1%, 2%) increased shell strength by 5.1%, 4.8 and 8.5% respectively. Egg yolk color of ML supplemented group was increased at 4.6-9.1% and Haugh unit of ML supplemented group was improved by 3.7-7.5% than the control group. The highest ML supplemented dose (2%) in this study did not result in the best effect. Instead, 0.5% ML supplementation had exerted the overall best effect. Our study suggests that the optimum supplementation of ML is 0.5% to 1.0% in the basal diet, which exerted the best effect on laying performance, egg quality and antioxidant capacity on laying hens.

Conclusion

Based on the results, the mulberry leaves could be used as a functional feed additive to enhance performance and oxidant status of laying hens.

Acknowledgments

The authors thank the Ministry of Science and Technology for financially supporting this study.

References

- Jeszka-Skowron, M., E. Flaczyk, J. Jeszka, Z. Krejpcio, E. Król, and M. S. Buchowski. 2014. Mulberry leaf extract intake reduces hyperglycaemia in streptozotocin (STZ)-induced diabetic rats fed high-fat diet. *J. Funct. Food*8:9-17.
- Sosa, V., T. Moline, R. Somoza, R. Paciucci, H. Kondoh, and M. E. L. Leonart. 2013. Oxidative stress and cancer: an overview. *Ageing Res. Rev.* 12:379-390.
- Zaviezo, R. 1999. Nutritional management of birds affected by heat. *Rev. Indust. Avi.*46(Suppl. XII):42-46.

Effects of performance and intestinal morphology by supplementation with a functional feed additive in poultry diet

Lai¹, L. P., M. T. Lee¹, B. Yu¹ & T. T. Lee¹

¹Department of Animal Science, National Chung Hsing University, 250 Kuo Kuang Road, Taichung 402, Taiwan.

Abstract

The present study was to produce a functional feed additive by solid-state fermentation of soybean hulls (100%, SBH) and soybean hulls partially replaced of *Pleurotus eryngii* stalk residue (75:25, SBHP) with *Aureobasidium pullulans* SH-218 that owns hemicelluloses secreting activities. The solid-state fermentation enhanced xylanase and mannanase activities as well as antioxidant capacity in terms of total phenolic content and trolox equivalency of both substrates; while the antioxidant properties were more noticeable in SBHP which showed higher levels of xylotriose, mannose and mannobiose as well. A total of 400 broilers were randomly assigned to one of the four dietary treatments including basal diet (control) and basal diet supplemented with 0.5% fermented SBH (0.5% FSBH), 0.5% fermented SBHP (0.5% FSHP) or 1.0% fermented SBHP (1.0% FSHP) until 35 day of age respectively. Body weight gain, body weight along with villus height/crypt depth ratio of jejunum and ileum were much more pronounced in broilers fed with 0.5% FSHP. The highest lactic acid bacteria/*Clostridium perfringens* ratio in ileum was observed in birds receiving 0.5% FSBH. In conclusion, solid-state fermentation of soybean hulls with *A. pullulans* could efficiently degrade anti-nutritional hemicellulose that subsequently increased oligosaccharides content, which accompanied by improved broiler intestinal microflora. Further supplementation of *Pleurotus eryngii* stalk in diet not only improved intestinal morphology, but had bird optimal growth performance by exerting its bioactive metabolite properties.

Keywords: soybean hulls, *Pleurotus eryngii* stalk residue, *Aureobasidium pullulans*, broilers

Introduction

Rearing costs in poultry industry keep rising in terms of advancing prices of feedstuffs encourage the exploitation of agricultural by-product as alternatives in recent years. However, most of them contain large amount of poorly digestive non-starch polysaccharides that limit the utilization in birds despite the virtue of low-priced availability and considerable quantity. Soybean hull, generating about 18-20 million tons annually, contains 20 to 25% of hemicellulose that exerts adverse effects on physical and chemical properties in gut that subsequently compromise avian performance (Choct, 2006).

Oxidative damage is associated with free radical formation and oxidative stress induced by endogenous and exogenous stimuli causes health and performance deterioration (Giannenas et al.,

2010); hence, high antioxidant status has been regarded as one of the major factors positively affecting avian performance in the concentrated poultry industry. Moreover, the poultry industry would greatly appreciate natural antioxidants replaced of synthetic ones and satisfy consumer demands for safe and environmentally friendly.

Pleurotus eryngii, commonly called the King Oyster mushroom has been shown to accumulate a variety of bioactive compounds considered to be important antioxidants and synergists that can improve the oxidative status of an animal (Reis et al., 2012). Moreover, the by-product of edible portion of fruiting body, *Pleurotus eryngii* stalk residue (*PESR*), possesses abundant metabolites which may not only provide the requirements for biological metabolism, but also exhibit potential antioxidant properties (Lee et al., 2012).

Aureobasidium pullulans is a black yeast-like microfungus that occurs frequently in varied environments. Different strains of *A. pullulans* can be applied to different fields that rely on the variant functional components they possess, such as the capability of secreting distinct enzymes (Rajeeva et al., 2010); nevertheless, scant research has been conducted. The objective of the present study was to produce a beneficial feed additive by solid-state fermentation of soybean hulls and *PESR* with *A. pullulans*, and to further evaluate the effects of fermented soybean hulls and *PESR* by *A. pullulans* supplemented in broiler diets.

Materials and methods

Four hundred 1-day-old broilers (Ross 308) were randomly and evenly allocated into 4 groups, including the control group (corn-soybean meal diet), 0.5% FSBH, 0.5% FSHP or 1.0% FSHP group for 5 weeks. The feed consumption, weight gain, feed conversion rate, intestinal microbiome and morphology as well as serum antioxidant capacity of broiler were determined.

The data was statistically analyzed using the general linear model procedure (GLM) of SAS software (2004) following a random arrangement. The mathematic model was: $Y_{ijk} = T_i + B_j + T_i \times B_j + \varepsilon_{ijk}$, where, Y_{ijk} = observed response of enzyme in a cage; T_i = fixed effect of feed or ferment daytreatment; B_j = fixed effect of batchtreatment; $T_i \times B_j$ = Interaction of feed or ferment daytreatment; ε_{ijk} = residual error when cage was regarded as experimental unit, $\varepsilon_{ijk} \sim N(0, \sigma_\varepsilon^2)$. The mean values were compared between the two feed or four ferment day groups using the LSMEANS with the significantly level at $P < 0.05$.

Results and discussion

Our findings demonstrated that exogenous hemicellulase effectively produced by the co-fermentation of soybean hull and *PESR* with *A. pullulans* SH-218, further eliminated partial anti-nutritional factors than FSBH presented in substrates. Avian intestinal microbiome could be affected by diet composition and dietary manipulations mainly through the mode of action such as creating an environment favored by specific bacteria in gut or/and competitive exclusion. In the present study, dietary supplementation of fermented products significantly increased cecal lactic acid bacteria counts as well as the lactic acid bacteria/*C. perfringens* ratio compared with the corresponding control group. Additionally, enhanced growth performance was obtained by dietary administration of *PESR* in response to optimal intestinal morphology.

SSF of soybean hull and a small amount of PESR with *A. pullulans* SH-218 could effectively increase total phenolic content further proved by results shown in the TEAC analysis; besides, serum catalase activity in birds fed 0.5% FSHP is the most pronounced among treatments. Results showed above indicating that promising antioxidant potential presented in PESR effectively elaborate in broilers receiving FSHP.

Table 1. Effect of fermented soybean hull (FSBH) and soybean hulls with *Pleurotus eryngii* (75:25, FSHP) supplemented in diet on intestinal morphology of 35 days old broilers.

Item	Experimental diets				SEM
	Control	0.5% FSBH	0.5% FSHP	1.0% FSHP	
Jejunum					
Villus height/Crypt depth	6.71 ^b	6.66 ^b	8.62 ^a	6.53 ^b	0.27
Ileum					
Villus height/Crypt depth	5.65 ^b	7.45 ^a	7.51 ^a	8.42 ^a	0.34

References

- Choct, M. 2006. Enzymes for the feed industry: past, present and future. *World Poultry Sci. J.* 62:5-16.
- Giannenas, I., I. S. Pappas, S. Mavridis, G. Kontopidis, J. Skoufos and I. Kyriazakis. 2010. Performance and antioxidant status of broiler chickens supplemented with dried mushrooms (*Agaricus bisporus*) in their diet. *Poult. Sci.* 89:303-311.
- Lee, T. T., J. Y. Ciou, C. J. Chiang, Y. P. Chao and B. Yu. 2012. Effect of *Pleurotus eryngii* stalk residue on the oxidative status and meat quality of broiler chickens. *Agric. Food Chem.* 60:11157-11163.
- Rajeeva, G., S. Ranjan, G. Monika and K. G. Manogya. 2010. *Aureobasidium pullulans*, an economically important polymorphic yeast with special reference to pullulan (Review). *Afr. J. Biotechnol.* 9(47):7989-7997.
- Reis, F. S., A., Martins, L. Barros and C. F. R. Ferreira. 2012. Antioxidant properties and phenolic profile of the most widely appreciated cultivated mushrooms: A comparative study between *in vivo* and *in vitro* samples. *Food Chem.* 50: 1201–1207.

Nitrogen balance and carcass quality in broilers given a low-protein diet in the grower period and higher-protein diets in the finisher period

*Koh*¹, K., K. Takeuchi², M. Rahman³, S. Yonekura¹ & A. Adrizal⁴*

¹Shinshu University, 399-4598 Nagano, Japan, ²Graduate school of Agriculture, Shinshu University, 399-4598 Nagano, Japan, ³Interdisciplinary Graduate School of Science and Technology, Shinshu University, 399-4598 Nagano, Japan, ⁴University of Jambi, Jambi 36361, Indonesia

Abstract

In the present study, growth performance, nitrogen (N) balance and body composition were measured in broilers given a low-protein diet (LPD, CP 17.6%) during 3-week grower period and then LPD, a standard- (SPD, CP 19.6%) or a high- (HPD, CP 22.0%) protein diets during 1-week finisher period. A total of 24 male broilers (21 d of age) were divided into control and L groups. Control group (6 birds) was given SPD throughout the experimental periods. L group (18 birds) was given LPD during the grower period, and then this group was divided into 3 groups of 6 birds, which were L group given LPD, S group given SPD and H group given HPD during finisher period. During the whole experimental period, there were no significant differences in body weight gain (WG), feed intake (FI) and feed conversion ratio (FCR) among dietary groups, and N excretion decreased and N retention improved significantly in H group, comparing with control group. Body fat deposition increased in L group, but such tendency was not found in H group. In conclusion, it is suggested that feeding LPD during the grower period followed by feeding HPD during the finisher period can decrease N excretion during the whole experimental period and prevent excessive fat deposition in broilers.

Keywords: body fat deposition, broiler, growth performance, nitrogen retention

Introduction

In poultry production, the use of low protein diet is recognised to be one of the solutions to reduce negative environmental impact due to ammonia emission from manure, and therefore, many studies have been conducted. The rationale of such studies is to improve the efficiency of N utilisation by feeding low protein diets supplemented with essential amino acids (Aftab et al., 2006). Unfortunately, such dietary manipulation led to excessive fat deposition which impairs carcass quality of broilers, although N excretion was decreased successfully (Yamazaki et al., 1996; Aletor et al., 2000). Taking practical use into account, we have tried to develop a solution using usual feed resources. In this context, it is well known that body fat in broilers inversely relates to dietary CP level, and therefore it may be possible that excessive body fat deposited during which LPD is given can be eliminated by the subsequent feeding of HPD. The purpose of the present study was to measure growth performance, N balance and body composition in broilers given LPD during 3-week grower period and then LPD, SPD or HPD during 1-week finisher

period, and to discuss this feeding program can decrease N excretion without deteriorating their growth performance and carcass quality.

Materials and Methods

This research was conducted in accordance with guidelines for regulation of animal experimentation of Shinshu University, Japan.

Table 1. Composition of experimental diets.

	LPD	SPD	HPD
Ingredient (g/kg)			
Commercial finisher diet	500.0	500.0	500.0
Maize	369.0	285.0	235.0
Soybean meal	95.0	170.0	170.0
Corn oil	18.2	29.0	29.0
Fish meal	0.0	0.0	50.0
Vitamin premix	3.0	3.0	3.0
Mineral premix	12.0	12.0	12.0
Amino acid premix	2.8	1.0	1.0
Calculated composition			
CP (%)	17.6	19.6	22.0
ME (MJ/kg)	13.38	13.38	13.38

A total of 24 male broilers (21 d of age) were divided into 2 dietary groups, such as control and L groups. Control group (6 birds) was given SPD throughout the experimental period (from 22 to 49 d of age) (Table 1). L group (18 birds) was given LPD during the grower period (from 22 to 42 d of age). At the end of the grower period, L group was divided again into 3 groups of 6 birds having similar body weight, which were L group given LPD successively, S group given SPD and H group given HPD during the finisher period (from 43 to 49 d of age). Diets and water were provided ad libitum throughout the experimental period. Body weight and feed intake were recorded weekly and daily, respectively. Faecal samples were collected daily and used for N balance calculations. At the end of finisher period, birds were fasted for 24 h with free access to water and sacrificed, and body composition was measured. Data were analysed to one-way ANOVA. Statistical significances among the dietary groups were determined with Tukey's multiple comparison tests.

Results and Discussion

During the grower period, L group showed numerically smaller WG, similar FI, significantly impaired FCR ($P < 0.05$), comparing with control group. Intake, excretion and retention of N were smaller in L group than in control group ($P < 0.05$), and N efficiency in L group tended to be improved. During the finisher period, there was no significant difference in WG, FI and FCR among groups, although FI in H group tended to decrease. N intake increased with increasing level of dietary CP level, but the value in H group was not greater than that in S group, which was due to decreased FI in H group. N excretion was around 20 g/bird/wk, excepting L group in which the

value decreased significantly ($P < 0.05$). There was no significant difference in N retention among groups. Comparing with control group, N efficiency increased in 3 experimental groups.

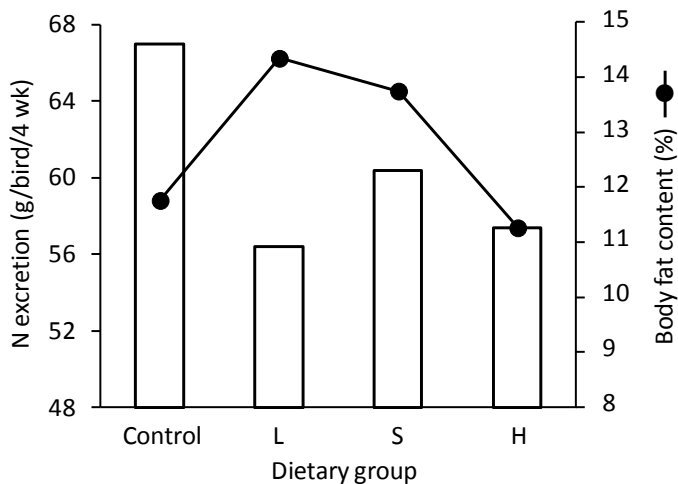


Figure 1. N retention and body fat content in broilers.

During the whole experimental period, WG, FI and FCR showed similar trends to those during finisher period. N intake was greatest in control group and smallest in L group. N excretion decreased in 3 experimental groups. There was little difference in N retention. N efficiency increased significantly in L and H groups and tended to increase in S group, comparing with control group. There was no difference in carcass weight, and CP and crude ash contents among groups. On the other hand, body fat in control group was about 12%, which increased in L group but not in H group. The similar trend was found in abdominal fat deposition. In conclusion, it is suggested that feeding LPD in the grower period followed by feeding HPD in the finisher period can decrease N excretion without deteriorating growth performance and carcass quality.

Acknowledgement

The present study was supported by the Grant from the JSPS-DGHE Bilateral Joint Research Project (2013-2015 FY), entitled "Upstream and downstream strategy of ammonia mitigation in curtain-sided poultry farm".

References

- Aftab, U., M. Ashraf and Z. Jiang, 2006. Low protein diets for broilers. *World's Poult. Sci.* 62: 688-701.
- Aletor, V.A., I. I. Hamid, E. Nieß and E. Pfeffer, 2000. Low protein amino acid-supplemented diets in broiler chickens: effects on performance, carcass characteristics, whole-body composition and efficiencies of nutrient utilization. *J. Sci. Food Agric.* 80:547-554
- Yamazaki, M., A. Murakami, M. Yamazaki and M. Takemasa, 1996. Reduction of nitrogen excreted from broiler chicks feeding low-protein, amino acid supplemented diets. *Jpn. Poult. Sci.* 33:249-255.

Effect of a heat stress reducing additive on meat production and quality in Hanwoo heifers

Lee¹, A-Leum, D. T. Mbiriri¹, C. H. Ryu¹, W. S. Song¹, M. S. Kim¹, S. Cho² & N. J. Choi¹

¹Department of Animal Science, Chonbuk National University, Jeonju, 561-756, Korea, ²Institute of CALS Co., Ltd, Seongnam, 463-741, Korea

Abstract

Following the successful development of a feed additive that has the potential of alleviating heat stress in beef cattle, we designed this study to assess the effects of the additive on growth performance and meat quality in Hanwoo (Korean beef breed) cattle finished during the summer season in Korea. The additive contained fat, choline and yeast at 0.05%, 0.05% and 0.025%, respectively. Thirty two heard of Hanwoo fattening heifers were randomly assigned to the control group (no additive) and treatment group (with additive). The additive was added to the animal's diets at 100 g/ d/ head for 120 days. At the end of the study period, the final weights between the two groups did not differ. However, the additive resulted in greater carcass yield of leaner meat than the control group. The treatment group also had better meat color – a major determinant in the choice of beef consumers. We concluded that the additive is beneficial in Hanwoo beef production.

Introduction

Heat stress in cattle can lead to huge losses in productivity and consequently profits (Williams et al., 2009). In an early study, we developed a feed additive to alleviate heat stress in beef cattle. We used the response surface methodology – a statistical tool – to optimize fat, choline and yeast quantities in a blend that could alleviate heat stress without negatively affecting animal productivity. The optimum quantities of fat, choline and yeast in the blend were determined to be 0.05%, 0.05% and 0.025%, respectively.

Fat increases the energy density – which would normally increase metabolic heat production, defeating the intended goal – but is not extensively degraded by rumen microbes (Beede & Shearer, 1991). It also protects rumen degradation of other nutrients, lowering metabolic heat load. Choline improves the growth of finishing cattle (Bryant et al. 1999; Bindel et al. 2000) whereas yeast cultures boost volatile fatty acid (VFA) production (Gray & Ryan, 1989).

We verified the optimized blend *in vitro* and *in vivo*. It lowered ruminal temperatures and did not depress rumen fermentation both *in vitro* and *in vivo*. This is a follow-up study to assess the effects of the additive on growth performance and meat quality in finishing Hanwoo beef heifers during the summer season in Korea – June to September.

Materials and methods

Thirty two Hanwoo heifers were randomly assigned to a diet supplemented or non-supplemented with the formulated additive. The main ingredients of the additive were protected fat, choline and yeast culture. Their blending conditions were optimized using the fractional factorial design. The additive was supplemented at 100 g/d/herd for 120 days until slaughter. We determined the following carcass traits: carcass weight, back fat thickness, loin area, carcass index and carcass grade. Meat quality was determined by marbling score, meat color, fat color, maturity, texture and grade. Meat grades were classified into five grades (1⁺⁺:1⁺:1:2:3) and one of three carcass grades (A, B or C). Each carcass was fabricated, meat characteristics were adjusted by pH, meat color (CIE L, a, b), Warner-Bratzler shear force (WBs) and cooking loss.

Results

Table 1. Effect of additive on carcass characteristics of Hanwoo heifer.

	Control	Treatment	SEM ¹⁾	p-value
Carcass characteristics				
Carcass weight (kg)	381.67	415.11	6.440	<0.05
Backfat thickness(mm)	21.33	15.00	1.192	<0.05
Loin area (cm ²)	85.44	94.33	1.613	<0.05
Carcass index	60.02	64.34	0.785	<0.05
Carcass grade(%), A:B:C	0:44:55	22:33:44	-	-
Meat quality				
Marbling	4.22	5.11	0.315	0.315
Meat color	4.78	4.67	0.102	0.694
Fat color	2.89	2.89	0.057	1.000
Maturity	3.11	3.11	0.159	1.000
Texture	1.56	1.11	0.086	<0.05
Grade (%), 1 ⁺⁺ :1 ⁺ :1:2:3	0:22:33:44:0	0:55:33:0:11	-	-

¹⁾Standard error of mean.

Table 2. Effect of additive on meat characteristics of Hanwoo heifer.

	Control	Treatment	SEM ¹⁾	p-value	
pH	5.54	5.56	0.008	0.552	
Meat Color	CIE L* ²⁾	36.04	38.63	0.472	<0.05
	CIE a*	19.14	21.75	0.497	0.006
	CIE b*	14.09	16.51	0.427	<0.05
WBs ³⁾ (kgf)	3.97	2.50	0.165	<0.05	
Cooking loss (%)	17.82	19.63	0.647	0.185	

¹⁾Standard error of mean.

²⁾Commission Internationale de Leclairage); L:lightness; a:redness, b:yellowness

³⁾Warner-Bratzler shear force.

Conclusion

The additive showed beneficial effect on beef production in Hanwoo heifers. It increased carcass weight, reduced backfat thickness and increased meat quality. We therefore recommend the incorporation of this additive in fattening Hanwoo cattle diets, especially during the hot summer season.

References

- Beede, D. K. and J. K. Shearer, 1991. Nutritional management of dairy cattle during the hot weather. *Agric. Pratic.* 12: 5-13.
- Bindel, D. J., J. S. Drouillard, E. C. Titgemeyer, R. H. Wessels and C. A. Loest, 2000. Effects of ruminally protected choline and dietary fat on performance and blood metabolites of finishing heifers. *J. Anim. Sci.* 78: 2497-2503.
- Bryant, T. C., J. D. Rivera, M. L. Galyean, G. C. Duff, D. M. Hallford and T. H. Montgomery, 1999. Effects of dietary level of ruminally protected choline on performance and carcass characteristics of finishing beef steers and on growth and serum metabolites in lambs. *J. Anim. Sci.* 77: 2893-2903.
- Gray, W. R. and J. P. Ryan, 1989. Effect of yeast culture on volatile fatty acid levels in ovine rumen fluid incubated with oats, barley and hay. *Biochem. Soc. Trans.* 17: 390-392.
- Williams, J. E., D. E. Spiers, L. N. Thompson-Golden, T. J. Hackman, M. R. Ellersieck, L. Wax, D. P. Colling, J. B. Corners and P. A. Lancaster, 2009. Effects of Tasco in alleviating heat stress in beef cattle. *Prof. Anim. Sci.* 25: 109-117

Effect of fermented plant extracts on enteric methane production in the rumen

Shin¹, S. J., B. Choi¹, A. L. Lee¹, S. Cho² & N. J. Choi¹

¹Department of Animal Science, Chonbuk National University, Jeonju, 561-756, Republic of Korea

²Institute of CALS, CALS Co. Ltd., Seongnam, 463-741, Republic of Korea

Abstract

Methane emissions from ruminants negatively affect the environment and animal productivity. Methane is a more potent greenhouse gas than atmospheric carbon dioxide. Its emission also represents a loss feed energy that could be available to the host animal. The present study was conducted to investigate effect of plant extracts that were fermented by different starter cultures on the reduction of enteric methane production in the rumen. A total of 8 strains, *Pichia anomala*, *Weissella confusa*, *W. cibaria*, *Saccharomyces cerevisiae*, *Lactobacillus brevis*, *L. plantarum*, *L. curvatus* and *L. sakei* were used as starter culture strains. 10 plants, *Eucommia ulmoides* OLIV., *Allium hookeri*, *Dioscorea quinqueloba*, *Asarum sieboldii*, *Hovenia dulcis* Thunb, *Morus alba* L., *Acanthopanax senticosus*, *Leonurus japonicas* Houtt, *Glycyrrhiza uralensis* and *Platycodon grandiflorum* Nakai were fermented. The dried plant powder was included in MRS broth, and starter culture was inoculated. Fermentation was conducted at 30°C for 48 h and then broth was dried at 60°C. Extraction was performed using ethanol, and the extract was prepared after concentration using rotary evaporation. Fermented plant extracts (FPEs) was supplemented in diet at 0.1%. Rumen fermentation parameters were investigated using rumen simulated *in vitro* fermentation. FPEs did not alter ruminal pH. *P. grandiflorum* fermented by *L. curvatus* and *L. plantarum* showed significantly greater ammonia nitrogen concentration than the control (P<0.05). Fermented *A. hookeri* by all starter cultures had greater ammonia nitrogen concentration than the control (P<0.05). *A. hookeri* fermented by *L. curvatus* and *L. plantarum* significantly increased volatile fatty acid production (P<0.05). These treatments also significantly reduced methane production (P<0.05). As a conclusion, the present study suggests that fermentation of *A. hookeri* using *L. curvatus* and *L. plantarum* can improve its biological activity and supplementation of these extracts could improve animal productivity via reduction of enteric methane emissions.

Keywords: animal productivity, fermentation, plant extracts, ruminal methane emission, starter culture

Introduction

Recently the increased greenhouse gas (CO₂, CH₄ and N₂O) in the troposphere has been involved in the consistent increase in atmospheric temperature and global warming over the last few decades (IPCC, 2001). The concentration of methane (CH₄) has increased at a rate of 10 µL/L per yr (1 µL = 10⁻⁹ L) since the preindustrial revolution (Moss et al., 2000). Domesticated ruminants are

estimated to produce about 80 Tg of CH₄ annually (1 Tg = 1 million metric tons) (NRC, 2000). Johnson & Johnson (1995) estimated that the energy lost as CH₄ in ruminants can range from 2 to 12% of the gross energy intake. According to many different research, plants mainly contain one or some predominant active molecules (secondary metabolites), which are responsible for specific biological effects about anti-nutritional factors than interfere with an animal's ability to maximize utilization of ingested nutrients. Particularly, plant extract effects evaluated anti-oxidant, anti-bacterial and methanogenesis reduction. However, investigation and result for plant extracts were insufficient. This study was conducted to improvement effect of plant extracts that were fermented by different starter cultures on the reduction of enteric methane production in the rumen.

Materials and Methods

Plant extract

All starter strains were maintained on MRS broth (Difco, USA) and incubated in 30°C with agitation (150 rpm). Medicinal Plants were ground by cutter miller (MF 10.1, IKA, Stanfen, Germany). Aliquot 3 g of fine powder of 10 plants were added into 30 mL MRS broth to prepare fermentation culture and then it was autoclaved (121°C, 15 min). Previously prepared seed culture was added into the fermentation culture at 1 mL inoculation rate. The inoculated culture was incubated at 30°C for 48 h. After incubation, broth was placed on aluminum dish and then dried at 60°C until completely drying (about 24 h). The dried culture broth was ground using mortar and 1 g of powder was suspended in 99.9% ethyl alcohol for extraction. Then extraction was performed at 30°C shaking incubator with agitation (150 rpm) for 20 h. The extract was filtered thorough filter paper (Whatman No 1) and the ethanol in filtrate was evaporated using rotary evaporator (N-1110, EYELA, Japan). The concentrated extract was re-dissolved in 2 mL of ethanol and stored in -20°C freezer until use.

***In vitro* ruminal fermentation**

Rumen fluid for *in vitro* fermentation was collected from cannulated Hanwoo steers (400 ± 5 kg) fed commercial concentrate and rice straw. pH analysis was used topH meter (Sevneasy, Mettler Toledo®, Switzerland). Total gas production was measured using a glass syringe connected to a needle. Ammonia nitrogen was determined according to Chaney & Marbach (1962). Volatile fatty acid analysis was performed according to Erwin et al. (1961). It was analyzed on a gas chromatograph (GC-7890A, Agilent Technologies, Inc., USA) using following the operating conditions; oven 150°C, injector 200°C, and FID 250°C and equipped with an auto sampler and FUSED SILICA Capillary (30 m × 2.25 mm, 0.25 µm film thickness) Column (NuKol™, Supelco, USA). Dry matter disappearance was analyzed as outlined by (Moore, 1970). Methane and Hydrogen production were analyzed on gas chromatograph (GC-7890A, Agilent Technologies, Inc., USA) using following the operation condition; oven 100°C, inlet 150°C and TCD 150°C and equipped with an auto sampler and Carboxen™, fused silica capillary column (0.53 mm i.d x 30 m length, SUPELCO, USA)

Statistical analysis

The data generated were subjected to analysis of variance (ANOVA) using the general linear model procedures in SPSS (version 18, IBM, USA) and multiple comparisons were performed using Duncan's posthoc test.

Results

Table 1. *In vitro* rumen fermentation parameters of ethanol extract of fermented *A. hookeri* using different starter culture strain at 24 h.

Contents	Treatments ¹				SEM ²
	Control	<i>A. hookeri</i>	<i>A. hookeri</i> NJ40	<i>A. hookeri</i> NJ45	
pH	6.60	6.60	6.59	6.60	0.00
NH ₃ -N, mg/100 mL	6.24	6.66	6.66	6.93	0.10
Total VFA, mM	60.35 ^b	40.53 ^a	39.97 ^a	41.32 ^a	2.63
Acetate, mM	36.21 ^b	13.37 ^a	13.47 ^a	13.86 ^a	2.97
Propionate, mM	12.76 ^b	0.72 ^a	0.75 ^a	0.75 ^a	1.57
iso-Butyrate, mM	0.68 ^a	8.23 ^b	8.44 ^b	8.55 ^b	1.01
n- Butyrate, mM	7.83 ^b	2.10 ^a	2.23 ^a	2.15 ^a	0.74
iso-Valerate, mM	1.69 ^b	1.17	1.22 ^a	1.22 ^a	0.06
n-Valerate, mM	1.18 ^a	3.03 ^b	2.97 ^b	2.98 ^b	0.24
A/P ratio	3.47 ^c	3.03 ^b	2.97 ^a	2.98 ^{ab}	0.06
Total gas, mL	76.67 ^b	71.33 ^a	67.33 ^a	70.00 ^a	1.20
Hydrogen, mL	0.04	0.03	0.03	0.03	0.00
Methane, mL	6.84 ^c	6.36 ^b	5.80 ^a	5.65 ^a	0.15

¹ Control, no additive; *A. hookeri*, only fermented; *A. hookeri* NJ40, *L. curvatus* as starter culture strain; *A. hookeri* NJ45, *L. plantarum* as starter culture strain.

² Standard error of the mean.

a, b, c, d, e Different superscript in same row means significantly different (P <0.05).

References

- Chaney, A. L. and E. P. Marbach, 1962. Modified reagents for determination of urea and ammonia. Clin. Chem. 8: 130-132.
- Erwin, E., G. Marco and E. Emery, 1961. Volatile fatty acid analyses of blood and rumen fluid by gas chromatography. J. Dairy Sci. 44: 1768-1771.
- IPCC (Intergovernmental Panel on Climate Change). 2001. Climate change 2001: The scientific basis. Cambridge, UK; Cambridge University Press.
- Johnson, K. A. and D. E. Johnson, 1995. Methane emissions from cattle. J. Anim. Sci. 73:2483-2492.
- Moore, J. 1970. Procedures for the two-stage *in vitro* digestion of forages. Nutrition Research Techniques for Domestic and Wild Animals 1: 5001-5003.
- Moss, A. R., J. -P. Jouany and J. Newbold, 2000. Methane production by ruminants: Its contribution to global warming. Ann. Zootech. 49:231-253.
- NRC, 2002. The scientific basis for estimating air emissions from animal feeding operations. National Academy PRESS, Washington, DC.

Enhancement of rice straw nutrient value by solid state fermentation with *Trichoderma*

Pan*, Y. G¹., C. T. Lo², B. Yu¹ & T. T. Lee¹

¹Department of Animal Science, National Chung Hsing University, Taichung, Taiwan

²Department of Biotechnology, National Formosa University, Yunlin, Taiwan

Abstract

Rice straw has been a common agricultural waste by rice production. The usage of rice straw to be feed ingredient is limited since its high proportion of cellulose and hemicellulose mainly as xylan, which is difficult to degrade by gastrointestinal enzymes of animals. The present study was to enhance of rice straw nutrient value by solid state fermentation with *Trichoderma*. The results showed that the fermented rice straw for 6 days could produce higher cellulase (24.82 U/g DM) and xylanase (409.67 mg/g DM) than that of unfermented rice straw. Total saccharide as hexose and pentose increased approximately 55.7 mg/g DM and 34.35 mg/g DM, respectively. Transmission electron microscopy observation presented that the lignocellulose structure showed degradation and fragmentation after rice straw fermented by *Trichoderma*. In conclusion, this study found that the fermented rice straw by *Trichoderma* could increase cellulase and xylanase contents as well as cell wall structure was apparently crushed. Hence, the fermented rice straw may potential as feedstuff for ruminants.

Keywords: rice straw, Trichoderma, cell wall.

Introduction

The price of animal feedstuff have been rising since the highly usage of biofuel production worldwide. There is a trend to pursue new uncostly source of animal feedstuff. Rice straw is a common agricultural waste, since combustion of rice straw in open air was an environmental hazard, there is an urge to solve the problem by fast and environmentally friendly method. Rice straw has high gross energy content, but it needs cellulase and hemicellulase to release its sugar content which limited its usage as animal feedstuff. *Trichoderma* spp are Ascomycota fungi which have been used as enzyme producer for cellulase and hemicellulase. Rostika and Safitri (2012) fed corn cob fermented by *T. viride* and *T. reesei* to Java barb and elevate the growth rate. Belewu et al. (2008) fed *T. harzanium* fermented cassava to lactating goat and enhanced the milk quantity and quality. Mohame and Abou-Zeina (2008) fed *T. viride* fermented sugar beet pulp to growing goat kid and improved the growth performance while serum biochemical parameters unaffected, which demonstrated the save usage of *Trichoderma* spp.

Materials and Methods

***Trichoderma* cultivation and treatment**

30g of rice straw was sterilized by immersed in 500c.c. of 100°C hot water for 30 minute. Then adjust the moisture to 80%. Three *Trichoderma*(NTLg S5-1, NTLg S9-1 and Tien Chung Ca6-1) was cultured on potato dextrose agar (PDA) for 5 days. Inoculum was prepared by adding culture medium to each PDA and 1.2mL of the spore suspension (~10⁷ spores/ml) was used as inoculum.

Fermentation

After inoculation, the rice straw was kept at 25°C for 6 days, and agitated the culture at day 4 and day 5 to ferment the inside of the rice straw.

Analysis of the endoglucanase and xylanase

Enzyme was recovered by adding 50 ml of distilled water to 5g of fermented substrate and stirred at 500rpm for 30 minute on the ice bath. Endocellulase assay was carried out by DNS method with 1% CMC in 0.05M pH4.8 citrate buffer as substrate and 50°C as incubation temperature. Xylanase assay was carried out by DNS method with 1% beech wood xylan in 0.05M pH5.0 citrate buffer as substrate and 50°C as incubation temperature.

Analysis of the total saccharide

Total saccharide was analyzed by phenol–sulfuric acid method while pentose was measured by 480nm absorbance and hexose was measured by 490nm absorbance after adding 5 % phenol and 95% sulfuric acid.

Results

The cellulase content of fermented rice straw by NTLg S5-1, NTLg S9-1 and Tien Chung Ca6-1 was 24.82±0.34, 10.24±0.26, 17.94±0.2 respectively. The xylanase content of fermented rice straw by NTLg S5-1, NTLg S9-1 and Tien Chung Ca6-1 was 409.67±3.36, 68.27±15.82, 135±12.28 respectively. The hexose content of fermented rice straw by NTLg S5-1, NTLg S9-1 and Tien Chung Ca6-1 was 4.57±2.54, 60.27±7.4, 46.44±0.16, 32.85±4.79 respectively. The pentose content of fermented rice straw by NTLg S5-1, NTLg S9-1 and Tien Chung Ca6-1 was 3.83±2.44, 38.18±4.56, 31.57±0.6, 18.16±1.4 respectively.

Conclusion

Trichoderma NTLg S5-1 has the most potential between the three *Trichoderma* as the candidate for rice straw fermentation since it produced the highest cellulase and xylanase and elevated the total saccharide content which is important parameter when introducing fermented rice straw as feedstuff for ruminant.

Acknowledgments

The authors thank the National Science Council for financially supporting this study.

References

- Belewu, M. A., A. A. Asifat, and M. B. Yousuf, 2007. Evaluation of *Trichoderma harzanium* treated cassava waste on the quality and quantity of milk of goat. *Afr. J. Biotechnol.* 6: 2193-2196
- Dubois M., K. A. Gilles, J. K. Hamilton, P. A. Rebers and F. Smith, 1956. Colorimetric method for determination of sugars and related substances. *Anal. Chem.* 28: 350–356
- Mohamed, M. I. and Hala A. A. Abou-Zeina, 2008. Effect of dietary supplementation with biologically treated sugar beet pulp on performance and organs function in goat kids. *Am.-Eurasian J. Agric. Environ. Sci.* 4: 410-416
- Rostika, R. and R. Safitri, 2011. Influence of fish feed containing corn-cob was fermented by *Trichoderma* sp, *Aspergillus* sp, *Rhizopus Oligosporus* to the rate of growth of Java barb (*Puntius Gonionitus*) *APCBEE Proc.* 2:148-152

Allometric productivity forage and goat foraging behaviour in rangeland at Ebelo Amboasary in Southern of Madagascar

Randrianariveloseheno AJM,^{1*} Rakotozandriny JN¹ and Daccord R²

¹High School of Agronomical Science - Department of Animal Science - Laboratory of Animal Nutrition - PO Box 175 Antananarivo Madagascar

²Swiss Federal Research Station for Animal Production and Dairy Products - Agroscope Liebefeld Posieux – CH 1725 Posieux, Switzerland

Abstract

Foraging activities rhythms of ruminants depend on biomass availability assessed by destructive and/or no destructive processes in rangelands. In Southern of Madagascar, relationships between forage availability (dry matter DM) and goat foraging behaviour with six goat adults such as bite rate have been analysed with allometric parameters such as leaf area index and branches number during two periods. In four identified pastureland vegetation groups, four dominant forage species such as *Acacia farnesiana*, *Poupartia caffra*, *Rhigozum madagascariense* and *Ziziphus mauritiana*. *Poupartia caffra* presented best value of leaf area index with $26.47 \pm 3.54 \text{ m}^2$ ($p < 0.05$). Available biomass was higher with significant difference ($p < 0.05$) such as *Acacia farnesiana* with $14.70 \pm 9.40 \text{ kg DM per plant}$; *Poupartia caffra* with $71.52 \pm 47.38 \text{ kg DM per plant}$; *Rhigozum madagascariense* with $6.04 \pm 3.41 \text{ kg DM per plant}$ and *Ziziphus mauritiana* with $5.66 \pm 1.65 \text{ kg DM per plant}$. Branches number parameter has been better for biomass estimation ($r^2 = 0.92$) than leaf area index ($r^2 = 0.04$). *Acacia farnesiana* range presents a best foraging and persistent daily bites ($7\text{-}8 \text{ bites} \cdot \text{mn}^{-1}$, $p < 0.05$) with high availability of forages species. Primary productivity effects have been determinate by seasonality, rangeland ecology and defoliation. Plant – animal interactions allow selecting, developing best autochthonous forage species and restoring semi-arid rangelands.

Keywords: allometric, biomass, bite, goat, Madagascar.

**Corresponding author: sehenorajm1@yahoo.fr or sehenorajm@univ-antananarivo.mg*

Introduction

Foraging activities rhythms of ruminants depend on biomass availability in pastureland. In shrubland, many processes have been used to determinate fodder production such as destructive and/or no destructive assessment. At semi-arid rangeland in Southern of Madagascar, relationships between forage availability and goat foraging behaviour have been analysed with allometric parameters such as leaf area index and branches number, and bite rates for preference in rangelands. This study aims to consider the dominant and the preferred browses species in rangelands implied to estimate the autochthonous browses productivity for restoring and rehabilitating the pastured ecosystems, for sustainable development in agro ecologic zones.

Materials and methods

In rangelands dominate by shrubs and browses species, six adults goats have been selected within mix composition of herd for vegetation monitoring, floristic assessment in ruminants foraging

behaviour. During six successive days at two foraging periods (Period 1 during end of dry season and Period 2 during end of wet season), foraging behaviour have been recorded during five minutes per animal every thirty minutes at the dominant foraging moment (from 08:30 to 11:00 and from 13:00 to 15:30). Goat foraging behaviour have been involved in pasture activities rhythms such as browsing, ruminating, moving or resting mentioned by Bourbouze (1981), Le Houérou (1980) and Meuret *et al.* (1985). And plant species intake and bite number were recorded and analyzed with the dominant fodder species in native pastureland. In sub arid rangelands, the higher availability and preferred browses species have been selected in sufficient number such as 15 browses plants for statistical precision (Cissé, 1980, Rasolohery, 2000), for allometric measurement with aerial organs (Norton, 1975). Two allometric parameters have been considered such as Leaf Area Index, secondary branches numbers from branches lower diameter than 5 cm (Bille, 1980). According to the horizontal projection of the summit by Prade *et al.* (1988), El Hassani *et al.* (1994); Rondeux (1999), the Leaf Area Index (LAI) has been calculated with 8 moved at 45° and concentrated rays on a point O close to geometric center as follow:

$$S_8 = \frac{\Pi}{8} \sum_{i=1}^8 R_i^2$$

The destructive method corresponds to direct quantification of biomass DW (Dry Weight) expressed with kg Dry Matter for establishing mathematical relationships as follow:

$$DW = ax + b$$

with a and b corresponding to coefficients of regression and x as allometric parameter such as branches number or Leaf Area Index.

Results and discussions

Allometric character

Considering floristic density and specific frequency, four dominant forage species such as *Acacia farnesiana*, *Poupartia caffra*, *Rhigozum madagascariense* and *Ziziphus mauritiana*, within sixty plants have been implied in physic parameters for biomass assessment as dry matter. *Poupartia caffra* has been especially characterized by around canopy for leaf area index measurement. Available biomass was high with significant difference ($p < 0.05$) with 5.66-71.52 kg DM per plant such as *Acacia farnesiana* with 14.70±9.40 kg DM per plant; *Poupartia caffra* with 71.52±47.38; *Rhigozum madagascariense* with 6.04±3.41 kg DM per plant and *Ziziphus mauritiana* with 5.66±1.65. About Leaf Area Index, an uniform canopy has been recorded with 26.47±3.54 ($p < 0.05$). Available canopy volume with leaf area index has been lower used for biomass prediction with lower coefficient of determination ($r^2 = 0.040$) than estimation with canopy volume with $r^2 = 0.99$ found by Mulubrhan *et al.* (2012); and branches number has been better parameter for assessing the biomass production with $r^2 = 0.92$.

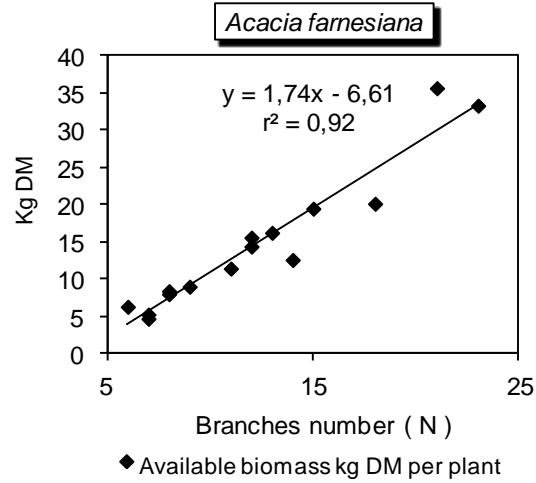
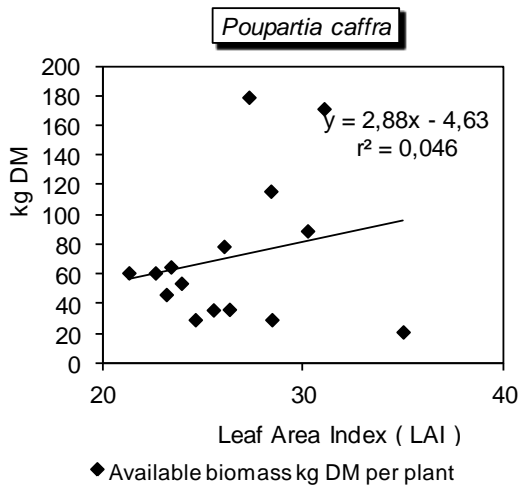


Figure 1: Available biomass of *Poupartia caffra* in function of leaf area index.

Figure 2: Available biomass of *Acacia farnesiana* in function of branches number.

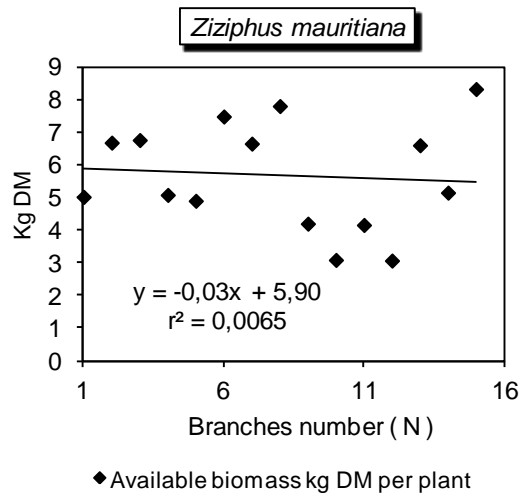
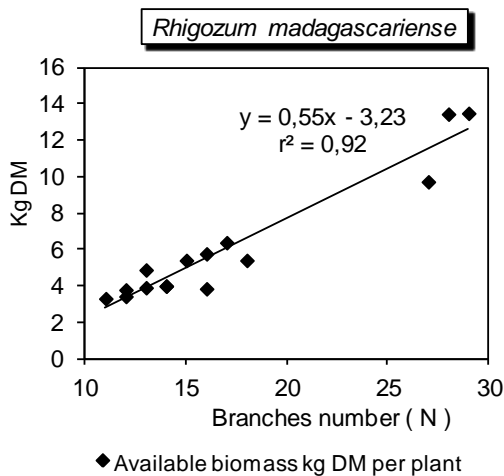


Figure 3: Available biomass of *Rhigozum madagascariense* in function of branches number.

Figure 4: Available biomass of *Ziziphus mauritiana* in function of branches number.

At this agro ecological zone, the fodder productions of 4 browses species (*Acacia farnesiana*, *Poupartia caffra*, *Rhigozum madagascariensis* and *Ziziphus mauritiana*) have been involved in significant production ($p < 0.05$) such as 6 to 71 DM kg per plant and higher productivity than 2 to 21 DM kg per plants with 7 fodder species in dry dense forests mentioned by Le Hou  rou (1980) and Randriamahaleo (2000). Among others, a Leaf Area Index parameter has been involved in low coefficient of determination. In fact, some factors have determined the browses species production: the climatic parameters have limited the available phytomass by photosynthetic activities; individuals plants have benefited some favourable ecological conditions such as soil, water and solar radiance for increasing the capacity of biomass and leaf production. And hydric reserves have contributed to modify dry matter content, especially the relation between leaf surface and trunk section plants in tropical zone mentioned by Ovenbird (1995). Pastoral environment, floristic density and physiological defoliation have influenced the biomass production per plant confirmed by Wilson *et al.* (1980).

Foraging behaviour and bite rate

Acacia farnesiana range has presented a best forage, available during periods and more consumed with daily bites ($7-8 \text{ bites.mn}^{-1}$, $p < 0.05$). In rangeland groups, climatic seasonality, rangeland ecology and defoliation model have been significant for foraging behaviour effects ($p < 0.05$) at two periods and primary productivity (figure 5).

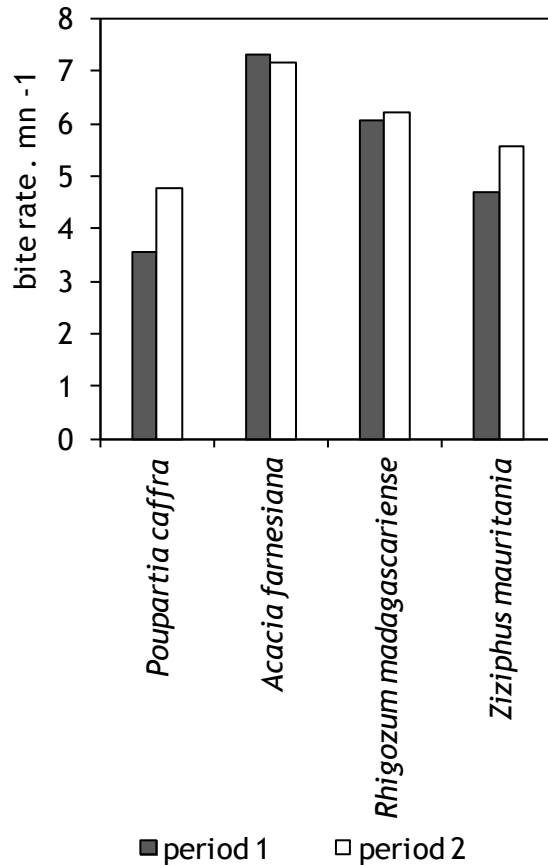


Figure 5: Bite rate and four dominant browses species.

In rangeland dominated by shrubs and browse species, fodder accessibility has been determined by vegetation structure. In functional foraging, bite rate value has been better than $2.76 \pm 0.6 \text{ bites.mn}^{-1}$ found by Nyamukanza and Scogings (2008) but lower with 1 to 9 bites.mn^{-1} than 57 bites.mn^{-1} found in grassland by Dicko – Touré (1980) and from 50 to 75 bites.mn^{-1} by Bocquier et al. (1987). Browsing and intake have been influenced by plant height, which is different between grasses and browse species. Structural vegetation such as canopy cover and floristic density have been considered as important factors for ruminants during selecting forage species (Wilson, 1978). Among others, floristic compositions such as Legumes species have influenced the intake rate and have improved the ruminant behaviour (Weldemariam, 2015). Foraging behaviour has been affected by floristic density and distances between shrub and browse species trunk. Herbivores must cover some long distances to find edible biomass in degraded woodlands and Ruminants species have compensated for small bite sizes by increasing bite rates (Fleurance et al., 2009). In fact, spatial factors have affected the result for decrease the bites per minute number as found by Wilson and Harrington (1980) and Van Soest (1982). In sub arid rangeland, pastoral vegetation has been dominated by browse species. Annual climatic variation have involved in fodder availability and in animal performance with natural forage supplementation (Decruyenaere et al., 2009. Kassa and Mekasha, 2014). In interactions between rangeland type and phenology, bite rate was better during period of maximum biomass resource. Daily bite rate increased during the wet season and the beginning of dry season. A decrease in

kinetic average intake has been observed in less accessible pastoral areas such as *Poupartia caffra* and *Kigelianthe madagascariensis* rangeland types. Consequently, ruminants have increased time spent on foraging activities. Intake rate decreases if width leaf and fibre content increase (Meuret 1997). And phenological stages did not appear to affect the fodder intake mentioned by Decruyenaere et al. (2009). Among others, anti-quality factors improve selected and refused forage and decrease palatability (Devaux 1973, Meuret 1997) such as some molecules, synthesised and favoured by climatic conditions in arid and subarid environments (Wilson, 1978). And preference ratio decreases with increasing numbers of unpalatable species in browse plant with higher anti-quality substance contents such as alkaloids, mimosin and tannins as mentioned by Bruneton (1999). Forage morphological aspect have affected intake rate such as aerial organs (leaf, branches) mentioned by Mosavat and Chamani (2013) and canopy parameters such as leaf area index mentioned by Gibb and Orr (1997). During foraging period, physiologic activities have affected eating rhythms and foraging behaviour such as satiation factors related to ruminal function mentioned by Baumont et al. (2000).

Conclusions

Various methods of investigation can be used in the study of the fodder system to goats and / or to ruminant. The diachronic method in analytical and systematic approaches was used to clarify the relations animal - plant in the pastoral areas. In rangelands, floristic monitoring and assessment allow considering the abundant and dominant species in different agro ecological forms. With four selected browses species, allometric measurements have been considered among approaches to estimate fodder biomass without destructive process within leaf area index ($r^2=0.046$) and branches number ($r^2=0.92$). In fact, the estimation models have been determinate by more factors such as botanic form and ecological conditions. The kinetic intake allows observing the nuances in rangeland and pastoral quality. In high floristic value situation, herbivores have tended to decrease the intake rate characterized by bite rate. Consequently, selected browses species have been involved in better bite rate value with higher availability and nutritive value for satisfying goat requirements and Ruminants. And foraging behaviour assessment has been interested with floristic, allometric and intake parameter to manage the fodder resources in natural pastureland. The relationships between phytosociological and nutritional parameters are implied for range management and for improving rangeland floristic with autochthons species in semi-arid environment and rangelands.

Acknowledgments

Authors gratefully acknowledge International Fund for Agriculture Development for funding this Project and Smallholders and livestock herder for their assistance during fodder plant monitoring and parameters measurements in pasturelands.

References

- Balehegn M., Eniang E A and Hassen A., 2012. Estimation of browse biomass of *Ficus thonningii*, an indigenous multipurpose fodder tree in northern Ethiopia. *African Journal of Range and Forage Science*, 29 (1): 25-30.
- Baumont R., Prache S., Meuret M. and Morand-Fehr P., 2000. How forage characteristics influence behaviour and intake in small ruminants: a review. *Livestock Production Science* 64: 15–28.
- Bille J. C., 1980. Measuring the primary palatibility production of the browse plants. *Proceeding of the International Symposium on the Browse in Africa / ILCA* - pp. 185 - 196.
- Bocquier F., Theriez M. and Brelurut A., 1987. Ewe food requirements in dairy period. *Bull. Tech. INRA - CRZV Théix*, 70: 199 – 211.

- Bourbouze A., 1981. Pasturelands use in different production system. *International Symposium. ITOVIC, INRA*. pp. 429 - 457.
- Bruneton J., 1999. Toxic plants. Dangerous vegetation for man and animals. 3rd. ed, *Tec. and Doc.* pp 529.
- Cissé M. I., 1980. The browse production of some trees of the Sahel . Relationships between maximum foliage biomass and various physical parameters. *Proceeding of the International Symposium of the browse in Africa*. pp. 205 - 210.
- Decruyenaere V., Buldgen A. and Stilmant D., 2009. Factors affecting intake by grazing ruminants and related quantification methods: a review. *Biotechnol. Agron. Soc. Environ.* 2009 13(4) : 559-573.
- Devaux C., 1973. Toxic plants or famous toxic for livestock in Western of Africa. *IEMVT / Service of Agrostology*. pp 148 + appendix.
- Dicko - Toure M. S., 1980. Measuring the secondary of pasture and applied example in the study of an extensive production system in Mali. - *Proceeding of the International Symposium on the Browse in Africa* . pp. 247 - 253.
- El Hassani T. A., Persoon E., 1994. Physiological and agronomical basis in vegetation production. *Modern Agronomy*. pp 544.
- Fleurance G., Fritz H., Duncan P., Gordon I. J., Edouard N. and Vial C., 2009. Instantaneous intake rate in horses of different body sizes: Influence of sward biomass and fibrousness. *Applied Animal Behaviour Science*, 117 : 84–92.
- Fournier C., 1995. Hydric fonction of six coexistant browses species in sahelian savannas (Ferlo Region – Northern of Senegal). *Works and Document, ORSTOM*, 132, pp 165.
- Gibb M. and Orr R., 1997. Grazing behaviour of Ruminants. *IGER Innovations*, 54-57.
- Gounot M., 1969. Quantitative study of vegetation methods. *Masson et Cie, Paris*. pp 314.
- Le Houérou H. N., 1980. Browse in Africa. The current state knowledge. *Proceeding of the International Symposium on the Browse in Africa, ILCA*, pp 491.
- Meuret M., Bartiaux N., Bourbouze A., 1985. Dairy goat consumption assessment in woodlands. Direct methods of bite rate – Chrom oxyd marker method. *Ann. Zoot.* 32 (2): 159 - 180.
- Meuret M., 1997. Small Ruminants forage intake in lands and woodlands areas. *Prod. Anim.* 10: 391-401.
- Mosavat N. and Chamani M., 2013. A Review: Comparison between Grazing Behavior of Cattle and Sheep. *Global Journal of Biodiversity Science And Management*, 3(2): 138-140.
- Mulubrhan Balehegn, EA Eniang and Abubeker Hassen, 2012. Estimation of browse biomass of *Ficus thonningii*, an indigenous multipurpose fodder tree in northern Ethiopia. *African Journal of Range and Forage Science*, 29 (1): 25-30.
- Norton B. E., 1975. Primary production simulation within models. *ILCA. Symposium acts, Bamako Mali*. pp. 313 - 321.
- Nyamukanza C.C. and Scogings P.F., 2008. Sprout selection and performance of goats fed *Acacia karroo* coppices in the False Thornveld of the Eastern Cape, South Africa. *African Journal of Animal Science*, 38 (2): 1-8.
- Prade J. and Bouchon J., 1988. Dendrometry. 2nd ed., *ENGREF*, pp 328.
- Rabarison H ., 2000. Phytological study at principal types of vegetation formations in Tsingy de Bemaraha (Classic methods and multidimensional analysis). Forest ressources use. *Dissertation of Doctorat Thesis, EESSciences, Dep. Biology and Ecology of Vegetation*. pp 167.
- Randriamahaleo T. R., 2001. Negatif effect study of farming in Beza Mahafaly Forest. *Engineering dissertation, ESSAgronomiques, Animal Science*, pp 74.
- Rasolohery A., 2000. Epiged phytomasse in post cultural successions in South Western of Madagascar. *Dissertation DEA, SBA – EESSciences, Biology and Ecology of Vegetation - Ecology of Vegetation*. pp 75.
- Rondeux J., 1999. Woody measurement and forest population - *Gembloux Agronomical Presses* - pp. 521

- Thuresson T., 2002. Ground sampling value with low density for national forest monitoring. *Unasyva* 210, 53 (3): 19 -23.
- Van Soest P. J., 1982. Nutritional ecology of the ruminant: ruminant metabolism, nutritional strategies, cellulolytic fermentation and the chemistry of forage and plants fibers. *O & B. Books Corenl University*. pp 373.
- Wilson C., 1978. Plants realtions and pastures. *CAB*. pp 428.
- Wilson A. D., Harrington G. N., 1980. Nutritive value of australian browse plants. *Proceeding of the International Symposium on the Browse in Africa*. pp. 291 - 297.

Supplementation of recombinant lycopene on egg quality and blood characteristics in quail diet

Chang^{1*}, C. H., W. T. Hsu¹, C. J. Chiang², Y. P. Chao³, T. T. Lee¹ & B. Yu¹

¹Department of Animal Science, National Chung Hsing University, Taichung, Taiwan

²Department of Medical Laboratory Science and Biotechnology, China Medical University, Taichung, 404, Taiwan

³Department of Chemical Engineering, Feng Chia University, Taichung, 407, Taiwan

Abstract

Lycopene has inhibited *in vivo* low-density lipoprotein (LDL) oxidation and HMG-CoA reductase activity, affect cholesterol biosynthesis and reduce blood triglyceride and cholesterol content, thereby reducing cardiovascular and chronic diseases. This study was to determine the effect of dietary supplementation of bacterial lycopene (BL) produced by *Escherichia coli* on the egg quality and blood characteristics of laying quails. A total of 70 100-day-old quails (*Coturnix coturnix japonica*) were fed with the basal diet supplemented with bacterial lycopene (BL), commercial lycopene (CL) or canthaxanthin for 4 weeks. The results showed that the yolks' triglyceride content was significantly lower for the group with BL and CL supplement. The group with BL supplement also had the lowest level of serum triglyceride. In conclusion, the results indicated that the BL produced by *E. coli* has potential as a feed additive in the diet of laying quails to decrease levels of serum lipid and triglyceride.

Keywords: recombinant lycopene, laying quail, feed additive

Introduction

Studied has noted that lycopene has a strong antioxidant capacity and can inhibit *in vivo* low-density lipoprotein (LDL) oxidation and HMG-CoA reductase activity, affect cholesterol biosynthesis, and reduce blood triglyceride and cholesterol content, thereby reducing human cardiovascular and chronic diseases. Moreover, addition of lycopene to the rat diet could reduce the serum malondialdehyde (MDA) content and inhibit lipid peroxidation and oxidized LDL generation. Leal et al. (1999) added lycopene to broiler's diet, and resulted in reduce serum MDA content of those birds. To our best knowledge, little information is available regarding the effect of recombinant lycopene in quail diets. In this work, lycopene was produced by *Escherichia coli* and used as a supplement to the quail diet. A systematic study was then conducted to investigate blood lipid traits and serum antioxidant enzymes of quails and the lipid content of the yolks. Consequently, the result indicates that recombinant lycopene decrease levels of serum lipid and triglyceride.

Materials and Methods

Laying quails, management and egg sample collection

A total of 70 100-day-old laying quails (*Coturnix coturnix japonica*) were randomly allocated into 7 dietary groups including the control group (basal diet), received a corn-soybean meal diet and

the basal diet supplement with BL, commercial lycopene (CL) or canthaxanthin (CA) for 6 or 18 mg/kg, respectively. Two eggs were collected randomly in each replicate per group (total 10 eggs) every Sunday to measure the egg quality (egg yolk weights, egg albumin weights and egg shell weights).

Blood collection for serum lipids and antioxidant enzyme determination

Five mL of blood was also collected from one layer per replicate (5 birds per group) at week 4 from the birds' left wing veins using a sterilized syringe and needle for measuring the total lipid, triglyceride, cholesterol and antioxidant enzymes content.

Results

Table 1. Effect of dietary supplementation of bacterial lycopene, commercial lycopene and canthaxanthin on yolk triglyceride and cholesterol content in laying quail at week 4

Item	C1		BL2, mg/kg		CL3, mg/kg		CA4, mg/kg		SEM	P value
	0	6	18	6	18	6	18			
Triglyceride, mg/g yolk	197a	198a	154d	154cd	131e	177ab	175bc	3.3	<0.001	
Triglyceride, mg/yolk	814a	772ab	611bc	677abc	529c	715abc	735ab	30.96	<0.001	
Cholesterol, mg/g yolk	13.6	13.3	13	12.2	12	12.7	12.2	0.75	0.13	
Cholesterol, mg/yolk	56.6	53.9	53	54.2	50.5	55.8	57.3	2.31	0.81	

n=10, ¹C=control group (basal diet). ²BL: basal diet supplemented with bacterial lycopene produce.

³CL: basal diet supplemented with commercial lycopene. ⁴CA: basal diet supplemented with canthaxanthin.

^{a, b, c} Means within the same row without the same superscripts are significantly different ($P < 0.05$).

Table 1 presents the effect of dietary supplementation of BL, CL and CA on yolk triglyceride and cholesterol concentrations in laying quails at week 4. The results showed that the yolks' triglyceride content in the BL and CL groups was significantly lower when supplemented at 18 mg/kg than in the control group. However, the yolk cholesterol concentrations were no different among the groups at week 4.

Table 2. Effect of dietary supplementation of bacterial lycopene, commercial lycopene and canthaxanthin on blood characteristics in laying quail at week 4

Item	C1		BL2, mg/kg		CL3, mg/kg		CA4, mg/kg		SEM	P value
	0	6	18	6	18	6	18			
Total lipid, mg/dl	1176a	1109ab	944ab	895b	872b	1151a	950ab	65.4	<0.001	
Triglyceride, mg/dl	1217a	937ab	737b	923ab	895ab	964ab	881ab	75.4	0.15	
Total Cholesterol, mg/dl	119	131	127	128	114	108	128	5.77	0.17	
HDL, mg/dl	69.8ab	81.5a	83.0a	70.3ab	64.4b	63.5b	62.7b	3.97	0.02	
LDL, mg/dl	21.7	22	22.7	25.5	24.3	22.5	22.8	1.56	0.88	

n=10, ¹C=control group (basal diet). ²BL: basal diet supplemented with bacterial lycopene produce.

³CL: basal diet supplemented with commercial lycopene. ⁴CA: basal diet supplemented with canthaxanthin.

^{a, b, c} Means within the same row without the same superscripts are significantly different ($P < 0.05$).

Dietary supplementation of BL, CL and CA on serum lipids, triglycerides and cholesterol content in laying quails at week 4 is shown in (Table 2). Compared to the other groups, the BL group had the lowest content of serum triglyceride when supplemented at 18 mg/kg. The CL groups had significantly lower serum lipid content than the control group. The HDL content was higher in the

BL groups than in other groups. However, the serum total cholesterol and LDL content showed no significant difference among the treatments. There are two majority sources of blood triglyceride: the protein-bound particles after the intestines' digestion and absorption of lipids in the diet that enter into the blood's circulation; the other is an endogenous one synthesized by the liver before entering into the blood as the major source of the body's triglycerides. Hu et al. (2008) added 12 mg/kg lycopene and 12 mg/kg fluvastatin to the male rabbits' high-fat diets. After eight weeks, the results of blood traits suggested that both treatments could reduce the triglyceride and increase HDL content, while lycopene group could also increase the blood antioxidant capacity and inhibit lipid peroxidation. Currently, treatment groups can reduce the blood triglyceride level, indicating that lycopene mainly accelerates lipid metabolism discharge and reduce triglyceride content of serum and eggs. In conclusion, the results indicated that the BL produced by *E. coli* has potential as a feed additive in the diet of laying quails to decrease levels of serum lipid and triglyceride.

References

- Hu, M.Y., Y.L. Li, C.H. Jiang, Z.Q. Liu, S.L. Qu and Y.M. Huang, 2008. Comparison of lycopene and fluvastatin effects on atherosclerosis induced by a high-fat diet in rabbits. *Nutrition*. 24: 1030–1038
- Leal, M., A. Shimada, F. Ruíz and E. González de Mejía, 1999. Effect of lycopene on lipid peroxidation and glutathione-dependent enzymes induced by T-2 toxin *in vivo*. *Toxicol. Lett.* 109: 1–10

Effect waste of cabbage on rabbit meat

Sri Minarti¹ and Osfar Sjoftjan²

¹ Department of Animal production, Faculty of Animal Husbandry, Brawijaya University, Malang

² Department of Animal Nutrition, Faculty of Animal Husbandry, Brawijaya University, Malang

Abstract

The research aims to study the pesticide residues in agricultural waste leaves of cabbage and meat rabbits consuming agricultural wastes cabbage leaf. The materials used are 12 New Zealand White rabbits weaning age of 1.5 months stratified by body weight. Rabbits fed cabbage waste maintained until the age of 3 months with the addition of concentrate feed in the form of concentrate. The method used is the exploration of the observed and variables are residues in meat, fat and water content of the meat. Results showed that there is some kind of pesticide residues in cabbage leaves that endosulfan, profenofos and klorprifos respectively of 0.0017; 0.0028; 0.0012 ppm. Rabbit meat contained pesticide residues, namely endosulfan, profenofos and klorprifos. The amount of pesticide residues in rabbits each major group (B) 0.00043; 0.00091; 0.0029 ppm. Groups are (S) .00035; 0.00040; 0.0015 ppm. Small groups (K) 0.00012; 0.0014; 0.0006 ppm The content of the fat content of meat rabbits fed forage kale waste is a large group (B) $3.28 \pm 0.17\%$; moderate group (S) $2.93 \pm 0.24\%$; and a small group (K) was $2.80 \pm 0.08\%$. Moisture content of meat from rabbits fed cabbage agricultural waste from a large group (B) of $66.78 \pm 0.73\%$; moderate group (S) $65.78 \pm 0.44\%$; and a small group (K) $65.55 \pm 0.39\%$. The conclusion of this study is commonly used cabbage leaves farmers as the primary forage feed rabbits are some pesticide residues, but still below the specified threshold so as not to endanger livestock that consume. Giving waste cabbage leaves do not affect the levels of fat and water content in meat rabbits. Advice can be given that agricultural waste which will be given to rabbits should be withering / inn a few days to lower the value of the level of pesticide residues.

Keywords: Agricultural waste cabbage, New Zealand white rabbit, residual meat

Corresponding author: mienunibraw@yahoo.com

Introduction

Cabbage leaf vegetables have grown in Indonesia commodity throughout the year. Harvest time enough short namely cabbage leaves for 100 days. The national crop productivity production of cabbage at 20.51 ton / ha (anonymous, 2012). Cabbage plants against pests susceptible. As far as the husband's business handle still using pesticides. Stone is the largest prayer one cabbage leaf producing region in east java, Indonesia (anonymous, 2010). Farmers pesticide applications use extremely intensive pesticide can because they consider pest problem cabbage plants. Farmers often use pesticides exceeding the dosage. Intensity of pesticide use in plant cabbage it was the first time one week on spraying heart of the rainy season and the first time 2 weeks spraying liver in drought. Currently arrived harvest, cabbage leaf waste many wasted. Husband wastes used as feed them rabbits. Cabbage leaves as forage granted up to 100% of the total feed took the rabbit. On pesticide residues are very worrying for rabbit meat waste consuming and produced. Husband study examined the effects of residual waste given cabbage in quality rabbit meat.

Materials and Methods

Materials and Methods

The material used is 12 New Zealand White rabbits weaning age of 1.5 months were maintained up to the age of 3 months with cabbage leaves fed forages and concentrates

The method used is a method of exploration, to assess the real situation in the field, which is the habit of cabbage farmers and ranchers rabbits in the study site.

Research procedures

This study is divided into two phases: the preparation and implementation phase or data collection, as well as the observed variables and weights and measures. Research stage is as follows:

1. Preparation phase include:

- 1.1. Preparing rabbit tail with an estimated total of 12 rabbits weaning age of 1.5 months
- 1.2. The division of the rabbit groups according to body weight as follows:
 - a. Small rabbits (K) with a weight of 400-500 g
 - b. Medium rabbits were (S) with a weight of 500-600 g
 - c. Large rabbit (B) with a weight of 600-700 g
- 1.3. Surveys and interviews to farmers on pesticides commonly used, spraying, and how much time and how many times the use of spraying until harvest time in planting cabbage.
- 1.4. Preparation waste collection cabbage leaves as the main forage. Waste cabbage taken each day at the time of harvest and given to rabbits next day
- 1.5. Drug preparation in the form of wormectin, medoxi-LA, and vitamine B complex.

2. Step of implementation includes:

1. Observations of temperature and humidity Temperature and humidity every 07.00 am and 15.00 pm.
2. Placement of cattle in a cage that has been provided.
 3. Feeding 08.00-09.00 WIB o'clock in the morning with milk concentrate feed PAP and afternoon 15.00-16.00 pm with waste feed cabbage leaves.
4. Cutting rabbit done at the age of 3 months simultaneously.
5. Sampling of meat rabbits selected in the legs (hind leg).
6. Samples were analyzed at the Laboratory of rabbit meat to measure the concentration of pesticide residues, moisture content, and the fat content to follow the procedures of the Association of Official Analytical Chemist (AOAC, 2005).

Variable Observations

The variables were observed in this study is

1. Testing of pesticide residues in meat rabbits
2. Testing the fat content of meat rabbits
3. Testing the water content of rabbit meat.

Data Analysis

Data were analyzed descriptively based on mean of the results.

Results and Discussion

Pesticide Type Uses and Applications

Based on observations and the results of interviews with 10 farmers cabbage in 3 (three) District in Batu, pesticides used by farmers cabbage is Lantis, Daconil 75 WP, SC Prevathon 50, Curacron 500 EC, Dursban 200 EC, Sevin 85 WP, Dharmasan 600 EC, Diazinon 60 EC, Fastrin 100 EC. Farmers do control against pests and plant diseases from planting to harvest. Farmers use pesticides as much as 12 times in the rainy season and 6-8 times in the dry season. Last Spraying is done 4 days prior to harvest.

Content of Pesticide Residues in Cabbage

Based on the analysis of pesticide residues it is known that the leaves of cabbage contain three (3) kinds of pesticide residues, namely endosulfan, profenofos, and klorfifos. On average residues can be seen in Table 1.

Table 1. Content of Pesticide Residues in Cabbage (ppm)

Pesticide Active Ingredients	Ingredients Pesticide Residues (ppm)
Endosulfan	0.0017
Profenofos	0.0028
Klorpirifos	0.0019
Total	0.0064

Source: Results Analysis Laboratory Faculty of Agriculture, University of Brawijaya (2013).

Cabbage leaves are still detectable pesticide residues because the waxy coating on the leaves of cabbage that is as a pesticide accumulation. According Tarumingkeng (1992) in Nuraini (2002) that the pesticides tend to accumulate in a layer of wax and fats of plants, especially in the skin layers that are stable and persistent. Results of analysis of pesticide residues found in cabbage leaves showed that the active ingredient greatest profenofos abortion on samples cabbage leaves are used as feed containing pesticide residues 0.0028 ppm. Other residues were detected among other pesticides containing the active ingredient chlorpyrifos and endosulfan. When compared to the standard ISO 7313: 2008 (Anonymous, 2008) then the pesticide residues were detected is still below the threshold of pesticide residues allowed in food. Standard endosulfan pesticide residue, chlorpyrifos, profenofos row is 0.1; 1; 1 ppm. The small residues of this pesticide for pesticide residue analysis were not performed on fresh cabbage leaves, but the leaves of cabbage that has been lodged 1-2 days. The high content of profenofos in cabbage leaves are used as feed in this study because of the use of organophosphate class of pesticides that are currently commonly used by farmers in Indonesia, in lieu of organ chlorine pesticides restricted group. Based on research conducted by Munarso, Miskiyah and Broto (2009) in Yusnani and Anwar (2013) found residues of pesticides endosulfan, klorfifos, profenofos, malathion, karbil as much as 0.0074, 0.0079 0.001, 0.0056, 0.0005 ppm the cabbage plants from samples taken from some of the existing market in Malang and Cianjur. Washing can reduce pesticide residues especially those that are systemic. Research Indraningsih (2006) stated that agricultural waste will be given to cattle should be done first before washing is given, in order. Pesticide residues can be lost or decompose, and sometimes this process takes place at a constant rate. According Tarumingkeng (1992) in Nuraini (2002) factors influencing this change is evaporation, washing, enzymatic degradation and translocation. Insecticide in crops can be lost altogether because of the metabolic processes associated with plant growth process itself. The rate of disappearance of insecticide residues of pesticides, follow the law kinetic first order is associated with a given amount of insecticide (deposit) and the factor of time, which will take place two (2) phases: the beginning of the residue which will disappear quickly called disipilasi process, the next stage where slow disappearance of residual insecticide called persistence. The occurrence of these two processes, among others, due to the deposit of insecticide can be absorbed and moved (translocated). High or low pesticide residues are also affected by the amount of use of the pesticide.

Content of Pesticide Residues in Meat Rabbits

The average yield of pesticide residues in meat rabbit weight based groups are presented in Table 2.

Table 2. Content of Pesticide Residues in Meat Rabbits (ppm)

Group	CompoundActivePesticide (ppm)		
	Endosulfan	Profenofos	Klorpirifos
large	0,00043	0,00091	0,0029
medium	0,00035	0,00040	0,0015
Kecil	0,00012	0,00014	0,0006

Source: Results Analysis Laboratory, Faculty of Agriculture, University of Brawijaya (2013).

In meat rabbits fed cabbage waste, discovered the active ingredient endosulfan, profenofos and klorfirifos. The active ingredient is thought to have come from feed contaminated with pesticide residues so that the resulting products still contain pesticide residues. Indraningsih research results (2004) indicate that waste cabbage leaves given to cattle can cause pesticide residues in livestock meat such as pesticide contamination at the time of planting cabbage and contamination of agricultural land. Lowest pesticide residues found in the group of rabbits rabbit small body weight, because the consumption of cattle to feed depending on the weight. The smaller the body weight, the less feed consumed. Based on the Indonesian National Standard (Anonymous, 2008), residues in meat is still safe to eat because it is still below the specified limits, that endosulfan 0.2 mg / kg; profenofos 0.05 mg / kg; chlorpyrifos 0.05 mg / kg. The effects of organochlorine pesticide residues generated class is chronic that can cause disturbances in liver function, adrenal and also can pose a carcinogenic effect. The use of pesticides is thought to inhibit the body's metabolism in rabbits through the nervous system. Potential and toksitas pesticide basically associated with inactivity of enzymes kolineterase. These enzymes cut kolineterase neurotransmitters (nerve conductor) acetylcholine. According Tarumingkeng (1992) mammalian nervous system is one part of the body systems most sensitive to toxins. Possible mechanisms that pesticides spread to organs and tissues of the body through the circulatory system as well as through the movement of cross-shaped barrier membrane. According to research Indraningsih (2004), agricultural waste organic cabbage and corn as animal feed contained pesticide residues less than the non-organic; pesticide residue still is because the soil is still contaminated by previous pesticide residues.

Fat Content of Meat Rabbits

Table 3. Content of Fat in Meat Rabbits (%)

Group Body Weight of Rabbit	content Fat content (%)
Large	3.28±0,17
Medium	2.93±0,24
Kecil	2.80±0,08

Source: Results of laboratory analysis of UB's Faculty of Mathematics and Natural Sciences (2013).

Results of analysis of the average fat content in meat rabbits highest of 3.28 + 0.17% of the group of rabbits with big body weight. The lowest fat content of 2.80 + 0.08% obtained from rabbits with a small body weight. Fat depot is a physiological process of livestock, with the function is as a backup to maintain homeostasis of the body heat (De Blass., Tores., Fraga., Perez and Calves., 1977). Fat growth in rabbits took place when more than two months old which is at approximately 1.5-2.0 kg of weight, but the fat contained still smaller when compared to other livestock. Placement of fat in the body rabbits occurred around the ribs, along the spine, thigh area, around the neck, kidney and heart (Bogart, 1981). De Blas (1997) states that the New Zealand White rabbits aged 6, 12 and 24 months had a fat each by 3.69%; 9.88% and 15.16%. Based on these data, the results showed that the pesticide residues in rabbit feed does not affect the fat content of rabbit carcass.

Water Content on Meat Rabbits

The average of the test results of water content in meat rabbits with treatment grouping different body weights are presented in Table 4.

Table 4. Percentage of Water Content on Rabbit Meat (%)

Group Body Weight of Rabbit	Water Content (%)
Large	66.78 \pm 0,73
Medium	65.78 \pm 0,44
Small	65.55 \pm 0,39

Source: Results of laboratory analysis of UB's Faculty of Mathematics and Natural Sciences (2013).

Based on the results of Table 4 shows that the average water content at the highest rabbit meat is indicated by a rabbit out of a large group of body weight that is equal to 66.78 \pm 0.73% while the lowest was showed by a rabbit out of a small group of body weight, which amounted to 65.55 \pm 0.39%. According Soeparno (1992) that the factors that influence the water holding capacity of meat is the difference between the water holding capacity of muscle, such as the species, age, and muscle function, as well as feed, transportation, temperature, humidity, storage, and preservation, gender, health, treatment for cuts and intramuscular fat, the effect of age and breed of cattle will affect the body composition animals. There is an inverse relationship between the level of water and fat in the animal body. If the animals get older, then decrease the moisture content in weight gain, and conversely the addition of fat. The water content of rabbit meat in this study is consistent when compared to the water content of rabbit meat by Soeparno (1992), which ranges between 68-75%. These results indicate that administration of cabbage in the feed did not affect the water content in meat rabbits.

Conclusion

Rabbits were fed contaminated cabbage waste pesticides in the area of Batu in East Java, located on the safe limit for pesticide residues in meat with fat and water content in the normal range.

References

- Anonymous, 2008. SNI. The Maximum Limit of Pesticide Residues In Agricultural Produce. ICS 65.100.01. National Standardization Agency. ISO 7313: 2008. [Http: // Horticulture. MOA. Go.Id/Admin/Services/SNI_Batas_Maksimum_Pestisida.Pdf](http://Horticulture.MOA.Go.Id/Admin/Services/SNI_Batas_Maksimum_Pestisida.Pdf). Accessed on February 5, 2013.
- Anonymous. 2010. The Central Bureau of Statistics East Java. Area Of Planted Harvested Of Productivity Cabbage East Java. Available at [Http: //Www.Jatim Prov.Go.Id /Index.Php? Option= Com_ Databank & Task = Tag & Tag = Agriculture &Itemid= 94 & Limit = 10 & Limit Start = 150](http://Www.Jatim.Prov.Go.Id/Index.Php?Option=Com_Databank&Task=Tag&Tag=Agriculture&Itemid=94&Limit=10&LimitStart=150). Accessed On March 26, 2013.
- Anonymous. 2012. Department Of Agriculture Batu. Village Profile at Bumiaji Batu. Department Of Agriculture, Kota Batu
- A.O.A.C. 2005. Official Methods of Analysis Of The Association of Official Analytical Chemists. Editor Kenneth Helrich, 15th Virginia, USA: The Official Association of Analytical Chemists, Inc.
- De Blass, JC, A. Torres, MJ Fraga, E. Perez and JF Calves. 1977. Influence of Weight and Age on The Body Composition Of Young Doe Rabbits. *J. Animal Sci.* 45 (1): 48-53.
- Indraningsih. 2006. Source Contaminants and Countermeasures Pesticide Residues in Food Animal Products: An Overview. *Wartazoa Vol. 16 No. 2 Years Old.* 2006.
- Nuraini. 2002. Pesticide Residue Levels In Liver And Local Fat Tailed Sheep Meat Males Which Were Fed Leaves Onion. Thesis. Pertanian Bogor Institute: Bogor.
- Soeparno. 1992. Meat Science and Technology. Gadjah Mada University Press, Yogyakarta
- Tarumingkeng, RC 1992. The Nature Insecticide Working Of Mechanism and Its Use Impacts. Christian University in Jakarta.
- Yusnani. Anwar, D., Anwar. 2013. Identification Group Organophosphate Pesticide Residues in Vegetables Potatoes In Supermarket Lott Mart And Eggplant Market Makasar 2013. Engineering Center of Disease Control And Environmental Health. Makasar

Effect of propolis as a natural antioxidant on the performance, carcass and antioxidant status of V-line rabbits

Zeweil, H. S., S. M. Zahran, M. H. Abd El-Rahman, Y. M. El-Gendy & M. Al-Taib

Department of Animal and Fish Production, Faculty of Agriculture (Saba Basha), University of Alexandria, Alexandria, Egypt

Abstract

The present study was conducted to determine the effect of supplementing propolis as a natural growth promoter and antioxidant on growth performance, carcass traits, lipid peroxide (Malondialdehyde) and the antioxidative status of growing V-line rabbits. A total number of 27 V-line rabbits at 5 weeks old with an average initial live body weight of 791.6 ± 21.86 g. were divided into three groups (n=9 rabbits/group). Each group was subdivided into three replicates with three rabbits each in a completely randomized design. The first group received basal diet free of propolis. The second and third groups were fed diets containing 400 and 800 mg propolis /kg diet, respectively. The results indicated that body weight, weight gain and feed conversion ratio were improved ($P \leq 0.01$) in the group given 400 mg propolis diet as compared to the control and the other experimental group, however, feed intake and carcass traits were not affected by different treatments. Propolis diets lowers the level of serum total lipids, total cholesterol and low density lipoprotein, however, it had insignificant effect on blood serum HDL of rabbits as compared to the control group. Total antioxidant capacity increased in blood serum by propolis supplementation as compared to the control group. It is well demonstrated in the present study that the consumption of propolis had positive effects on rabbits' performance, had a beneficial effect on blood lipid regulation which was demonstrated to be ascribed to their antioxidant activity and the antioxidative status of growing V-line rabbits.

Keywords: Rabbit, propolis, performance, carcass, blood constituents

Introduction

Stock health and welfare management are key factors in animal health and food safety. The application of in-feed antibiotic growth promoters in livestock diet threatens consumer health and has arisen into a controversial issue worldwide. Many countries tend to prohibit the use of antibiotics as growth promoters due to their effects and their residual problems in tissue of animals. Supplementation of natural components in poultry diet is now widely distributed in the world. These components are served as growth promoters, which are healthful and help to improve the production performance of animals and poultry, also it is considered as an excellent immune system booster (Abd El-Rahman & Mosaad, 2013). Propolis is one of these components. It is an adhesive, dark yellow to brown coloured exudates. It has a strong antibacterial activity in addition to antifungal, antiviral and antiprotozoal properties (Scheller *et al.*, 1999). Propolis supplementation used as antioxidant, antimicrobial and antimutagenic based on its rich flavonoid, phenolic acid terpenoid contents (Prytyk *et al.*, 2003; Wag *et al.*, 2003). The purpose of the present study was to determine the possible beneficial effects of dietary propolis on performance, carcass traits and serum lipid profile in V-line rabbits.

Materials and Methods

A total number of 27 V-line rabbits at 5 weeks old with an average initial live body weight of 791.6 ± 21.86 g. were divided into three groups (n=9 rabbits/group). Each group was subdivided into three replicates with three rabbits each in a completely randomized design. Rabbits fed a basal diet containing 17.3 % crude protein and 2690 kcal digestible energy / kg diet. The first group received basal diet free of propolis. The second and third groups were fed diets containing 400 and 800 mg propolis /kg diet, respectively. The experiment lasted for eight weeks. All cages were provided with a manual feeder, and clean fresh water was available continuously through an automatic system of nipple drinkers. Rabbits were kept under the same hygienic and environmental conditions during the experimental period, they provided with 14 h of light daily. All biochemical traits of blood serum were determined using commercial kits (Diamond Diagnostics, Halliston, MA, USA). Data were analyzed using one-way ANOVA of GLM procedure of SAS® (**SAS Institute, 2000**). Significant differences between means were detected using new Duncan multiple range test (**Duncan, 1955**).

Results and Discussions

Results presented in Table 1 indicated that body weight, daily body weight gain and feed conversion ratio were improved ($P \leq 0.01$) in the group given 400 mg propolis diet as compared to the control and the other experimental group, however, feed intake and carcass traits were not affected by different treatments. Propolis diets lowers the level of serum total lipids, total cholesterol and low density lipoprotein, however, it had insignificant effect on blood serum HDL of rabbits as compared to the control group. Total antioxidant capacity increased in blood serum by propolis supplementation as compared to the control group. The effect of propolis on total lipids is in agreement with Kolankaya *et al.* (2002) found that propolis significantly decreased cholesterol and triglycerides in rats. In addition, Fuliang *et al.* (2005) revealed that ethanol extract propolis and water extract propolis (1 ml/100 g BW) decreased blood total cholesterol and triglycerides in plasma of fasting rats, and this is directly related to the influence of propolis on lipid metabolism (Matsui *et al.*, 2004). It is well demonstrated in the present study that the consumption of 400 mg propolis / kg diet had positive effects on rabbits' performance, had a beneficial effect on blood lipid regulation which was demonstrated to be ascribed to their antioxidant activity and the antioxidative status of growing V-line rabbits.

Table 1: Effect of different levels of propolis on V-line rabbits

Items	Control	Propolis level (mg / kg diet)	
		400	800
<u>Rabbits performance:</u>			
Initial body weight, g	774.2 ± 23.4	782.9 ± 20.1	817.9 ± 23.1
Final body weight, g	2345.8 ± 19.7 ^b	2468.8 ± 25.9 ^a	2389.6 ± 15.4 ^b
Daily body weight gain, g	24.95±0.27 ^b	26.76±0.35 ^a	24.95±0.37 ^b
Daily feed intake, g	91.22±0.49	91.08±0.27	90.13±0.64
Feed conversion ratio	3.66±0.05 ^a	3.41±0.05 ^b	3.62±0.05 ^a
<u>Carcass characteristics:</u>			
Pre-slaughter weight, g	2325.0±40.7	2456.3±54.9	2372.5±34.4
Carcass %	55.04±0.26	53.98±0.62	55.68±0.20
Liver, %	2.93±0.25	3.19±0.08	2.88±0.16
Heart, %	0.32±0.02	0.31±0.02	0.29±0.02
Spleen, %	0.09±0.00	0.09±0.00	0.11±0.01
Kidneys, %	0.59±0.03	0.59±0.02	0.55±0.03
Kidneys fat, %	0.34±0.01	0.34±0.02	0.35±0.02
Lungs, %	0.53±0.03	0.61±0.03	0.58±0.04
<u>Serum blood lipids profile:</u>			
Total lipids	282.50±7.22 ^a	241.25±6.57 ^b	261.50±9.02 ^{ab}
Total cholesterol	70.28±1.32 ^a	55.85±0.84 ^b	60.18±2.56 ^b
High density lipoprotein (HDL)	29.25±2.21	34.00±1.58	33.25±0.85
Low density lipoprotein (LDL)	22.00±1.29 ^a	17.50±0.87 ^b	17.25±0.63 ^b
Total antioxidant capacity	1.71±0.03 ^b	2.18±0.10 ^a	1.96±0.11 ^{ab}

Means within row bearing different superscripts are significantly different.

References

- Abdel-Rahman, M. A. and G. M. Mosaad, 2013. Effect of Propolis as Additive on Some Behavioural Patterns, Performance and Blood Parameters in Muscovy Broiler Ducks. *Journal of Advanced Veterinary Research* 3: 64-68.
- Duncan, D.B., 1955. Multiple range and multiple *F* tests. *Biometrics*, 11: 1-42.
- Fuliang, H. U., H. R. Hepburn, H. Xuan, M. Chen, S. Daya and S. E. Radloff, 2005. Effects of propolis on blood glucose, blood lipid and free radicals in rats with diabetes mellitus. *Pharmacology Research* 51: 147– 152.
- Kolankaya, D., G. Selmanoglu, K. Sorkun and B. Salih, 2002. Protective effects of Turkish propolis on alcohol induced serum lipid changes and liver injury in male rats. *Food Chemistry* 78: 213–217.
- Matsui, T., S. Ebuchi, T. Fujise, K. J. Abesundara, S. Doi, H. Yamada and K. Matsumoto, 2004. Strong antihyperglycemic effects of water-soluble fraction of Brazilian propolis and its bioactive constituent, 3,4,5-tri-O-caffeoylquinic acid. *Biological & Pharmaceutical Bulletin* 27: 1797–1803.
- Prytyk, E., A. Dantas, K. Salomao, V. Periera, S. Bakova, D. Castro and A. Eto, 2003. Flavonoids ad trypanocidal activity of Bulgarian propolis. *Journal of Ethnopharmacology* 88: 189–193.
- Scheller, S., S. Dworniczak, K. Waldemar-Klimmek, M. Rajca, A. Tomczyk and J. Shani, 1999. Synergism between ethanolic extract of propolis and anti-tuberculosis drugs on growth of mycobacteria. *Zeitschrift für Naturforschung* 54: 549 – 553.
- Wag, B., Y. Lien and Z. Yu, 2003. Supercritical fluid extractive fractionation study of the antioxidant activities of propolis. *Food Chemistry* 86: 237 – 243.

The effects of phenological stages on forage quality of four rangeland species for the sheep nutrition in Sari plain, Iran

Sara Mohammadi¹, Mohammadreza Tatian*², Reza Tamartsh³ & Hossein Aghajantabar⁴

^{1,4} M.Sc. Student of Range Management, Sari Agricultural Sciences and Natural Resources University, ^{2,3} Faculty Members, Sari Agricultural Sciences and Natural Resources University Department of Range Management, Natural Resources Faculty, Badeleh, Sari, Postal Code; 4844174111, Iran

Introduction

Forage quality is a series of plant factors that affect the yield and the general concept is that all nutrition properties of forage in relation to animal feed needs and the amount of energy available for animal is determined (Arzani et al., 2006). Therefore, the suitable grazing time and consequently the readiness of rangeland can be determined by understanding the changes in forage quality indices of rangeland plants. This study, which had compared three different phenological stages including vegetative growth, flowering and seeding of four important rangelands species, aimed to determine forage quality factors such as energy Metabolizable Energy, dry matter digestibility, crude protein, percentage of Acid Detergent Fiber in order to identify the best and the most appropriate species in terms of providing energy for the sheep.

Materials and methods

The study area

The study area was located at the Sari Agricultural Sciences and Natural Resources University in northern plain lands of Sari, Iran. The elevation is 11 meters below sea level and has a humid and temperate climate with an average annual rainfall of 580 mm and an average annual temperature of 17.2 ° C. In order to perform this research, after preparing the seed bed in the field, seeds of four palatable rangeland species including *Trifolium repens*, *Onobrychis sativa*, *Medicago scutellata*, and *Medicago sativa* were separately planted in one square-meter plots with three replications. After growth beginning and the emergence of leaves, the first randomly full sampling of 5 individuals in each plot was carried out. Subsequent samplings were done at the beginning of the flowering period and the beginning of the seeding in the same way. The collected samples were transferred to the laboratory and after the final drying, forage quality parameters such as crude protein, acid detergent fiber, dry matter digestibility, and metabolizable energy were determined in Lab. DMD and ME were calculated based on $DMD\% = 83.58 - 0.824ADF\% + 2.626N\%$ (Oddy et al., 1983) and $ME (Mj/Kg) = 0.17DMD - 2$ (Australian Standing Committee on Agriculture, 1990) formulas respectively. Data were analyzed using analysis of variance method and the mean comparisons in SPSS 16 software.

Results

The evaluation results of the forage quality of *Onobrychis sativa* and *Trifolium repens* showed that parameters of acid detergent fiber (ADF) and digestibility (DMD) for both species and Metabolizable Energy for the second species were under the influence of metabolic energy and were significant at 5% level; however the protein had no significant change (Figure 1).

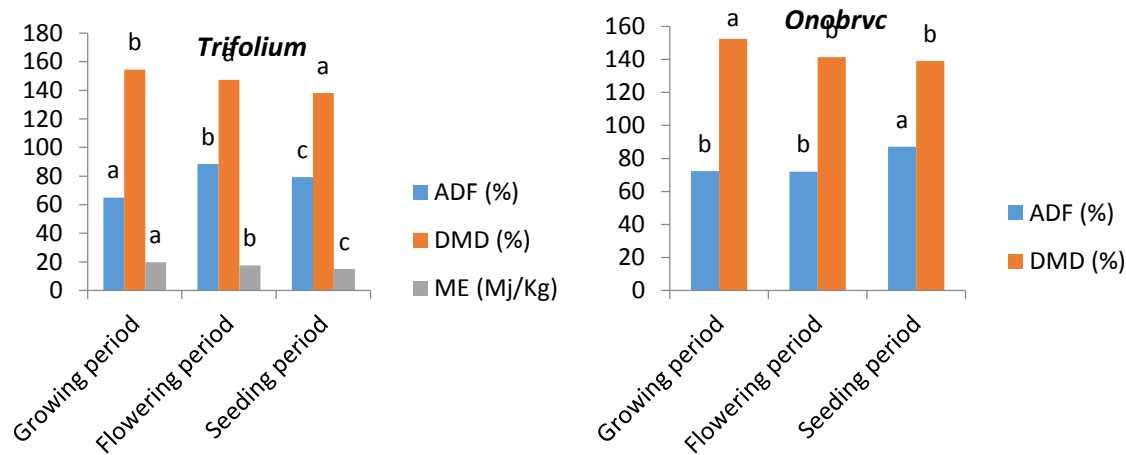


Figure 1. Mean comparison of forage quality parameters at different stages.

By studying the forage quality of *Medicago sativa* and *Medicago scutellata*, it was observed that from flowering to seeding stages features such as protein and digestibility for both species and Metabolizable Energy for the second species had declined, but the acid detergent fiber had increased (Table 1).

Table 1. Analysis of variance of forage quality parameters at different stages.

Seeding period	Flowering period	Growing period	F statistics	Feature's name	Species
4.9	5	5.56	^{ns} 0.82	CP (%)	<i>Medicago sativa</i>
14.93	15.14	15.92	^{ns} 0.12	ME (Mj/Kg)	
82.33	79.66	78.66	^{ns} 2.31	ADF (%)	
145.93	147.36	149.33	^{ns} 2.51	DMD (%)	
4.53	4.6	4.73	^{ns} 0.07	CP (%)	<i>Medicago scutellata</i>
15.52	16.60	18.49	3.10 ^{ns}	ME (Mj/Kg)	
83.66	83.65	79.33	0.54 ^{ns}	ADF (%)	
144.72	145.92	147.15	0.28 ^{ns}	DMD (%)	

Discussion

The results showed that the effect of phenological stage on forage quality parameters including acid detergent fiber and dry matter digestibility were more specific than the other indices, So that the percentage of ADF for *Onobrychis sativa* and *Trifolium repens* had increased at ending stages

compared to the beginning and the DMD value had shown a significant reduction for these species. This issue suggests that for these species, the above indices have been more affected by vegetative changes than the other ones. In fact, by plant growth, the maintenance and strength tissues would increase; these tissues are built of structural carbohydrates such as cellulose, hemicellulose, and lignin; therefore, with the completion of growth period and increasing structural carbohydrates, the percentage of fiber would increase in plants (Chen et al., 2001). By increasing the amount of lignin and cellulose materials during plant aging, the Dry Matter Digestibility had also been reduced. Hoffman et al. (2003) stated that increasing in structural tissues would decrease the DMD in stems. In addition to the above parameters, the Metabolizable Energy had significantly decreased in *Trifolium repens*, because by plant aging the added amount of lignin would reduce the digestibility of animals (Mut et al., 2006). In fact, the metabolizable energy content in the leaves and stems of all species had decreased with the development of phenological stages and leaves and stems of plants in the first stages of growth have the highest metabolizable energy (Kellums and Church, 2002). Despite the lack of significant changes in forage quality, *Medicago sativa* and *Medicago scutellata* had a similar trend to the other study species. Moore and Undersander (2002) stated that the forage digestibility had a direct relationship with the characteristics of the cell wall. For this reason, if the contents of a plant cell are fully digestible, then the digestibility may not change by plant aging (Albayrak and Turk, 2013). The results showed that the protein content between different species had increased from beginning to the end of growth. The importance of *Onobrychis sativa* and *Trifolium repens* was noteworthy, which had the least and highest crude protein in the active growth stage, respectively. In addition, *Trifolium repens* had higher Metabolizable Energy and Dry Matter Digestibility than any other study species at the flowering stage; while *Onobrychis sativa* had the lowest amounts. Considering all the above, it can be said that among the study species, these two species had the highest and lowest quality indices, respectively, during active growth and flowering compared to the seeding stage. Two species of *Medicago* had moderate changes in most cases; this could be due to the proximity of these two species and also their difference with the two other species, which had led to statistical differences between them. However, the only significant difference of these two species was observed for ADF. Due to the fact that with the development of phenological stages the proportion of plant organs would change (leaves, shoots, stems and flowers) and since the plant organs are different in physiological characteristics as well as each organ has a different weight ratio at each different stages, hence it could create a different value at each phenological stage that could be effective in this case (Wramit et al., 2012). In general it can be said that because there was a difference between the studied species in terms of forage quality indices; thus without regard to the composition of the plant, a consistent basis cannot be considered in order to calculate the nutritional value. Therefore, it is suggested to consider relative forage quality and indices such as the crude protein, metabolizable energy and dry matter digestibility as the basis for calculating livestock's nutritional needs.

References

- Albayrak, S., and M. Turk, 2013. Changes in the forage yield and quality of legume-grass mixtures throughout a vegetation period. *Turkish J. of Agriculture Forage* 37: 139-147.
- Arzani, H, M. Basiri, F. Khatibi and G. Ghorbani, 2006. Nutritive value of some Zagros mountain rangeland species. *Small Ruminant Research* 65: 128- 135.
- Chen, C.S., S.M. Wang, and Y.K. Chang, 2001. Climatic factors, acid detergent fiber, natural detergent fiber and crude protein contents in digit grass. *Proceedings of the XIX International Grassland Congress, Brazil*.
- Hoffman, P.C., K.M. Lundberg, L.M. Bauman, and R.D. Shaver, 2003. The effect of maturity on NDF digestibility. *Focus on Forage* 5: 1-3.
- Kellums, R.O., and D.C. Church, 2002. *Livestock feeds & feeding*, Prentice Hall, 217p.
- Mut, Z., I. Ayan, and H. Mut, 2006. Evaluation of forage yield and quality at two phenological stages of Triticale genotypes and other cereals grown under rained conditions. *Botany* 35(1): 45-53.
- Oddy, V.U., G.E. Roberts, and S.G. Low, 1983. Prediction of In-vivo dry matter digestibility from the Fiber and Nitrogen content of a feed, In *Feed Information and Animal production*, Packham, Common wealth Agricultural Bureau., Australia, 298.
- Standing Committee on Agriculture, 1990. *Feeding standards for Australian livestock ruminants*, CSIRO, Australia, 39.
- Wramit, N., M. Kenneth and S. Fales, 2012. Forage quality of native warm-season grasses in response to Nitrogen fertilization and harvest date. *Animal Feed Science and Technology*, 2:46-59.

Effect of ginger root powder supplementation on growth performances and carcass characteristics of broiler chickens as rearing in hot climate

K. Soisuwan and N. Chauyuchuwong

Department of Animal Science , Faculty of Agriculture, Rajamagala University of Technology Srivijaya, Nakhon Si Thammarat, Thailand, 80110

Abstract

This study was conducted to investigate the effect of dietary supplementation with ginger root powder as a natural growth promoter on performances and carcass characteristic of broiler chickens as rearing in hot climates. A total of 250 one-day-old male broiler chicks (Ross 308) were allocated to five treatments with five replicates. The dietary treatments consisted of the basal diet with no supplement as control, basal diet containing 100 mg/kg vitamin E as positive control, basal diet containing 8, 16 and 24 g/kg of ginger root powder as dietary treatment. The curcumin content of the ginger powder was $1.29 \pm 0.04\%$ by weight. Body weight gain and feed intake of chickens were not influenced by the dietary treatments. Broilers fed ginger root powder supplemented diets exhibited better feed efficiency over the entire experimental periods in comparison with control group ($P < 0.05$). A significant decrease ($P < 0.05$) in abdominal fat pad was observed in chickens fed the supplementation of ginger root powder. The obtained results from this study could be concluded that dietary inclusion of ginger root powder have no significant to improve broiler performance, however, it has significantly improved ($P < 0.05$) feed efficiency. It was also found that supplementation with ginger root powder has significantly improved ($p < 0.05$) carcass characteristic of broilers in term of reduction abdominal fat percentage.

Keywords: ginger root powder, broiler performances, carcass characteristic, hot climate

**Corresponding author: ksoisuwan52@gmail.com*

Introduction

Heat stress causes oxidative stress in broiler chickens (Lin et al., 2006) and oxidative stress has been recorded as a primary factor to decrease performance and carcass characteristics (Mujahid et al., 2007). Previous research has shown that detrimental effects of heat stress could be alleviated by antioxidants. Many synthetic antioxidant compounds have shown to be used in broiler diets, however, to have toxic or mutagenic effect in poultry, so that there is growing interest in source of natural antioxidants (Nagulendran et al., 2007). One source that has been investigated is ginger root (Zhang et al., 2009). Ginger (*Zingiber officinale Roscoe*) root contain active chemical including volatile oils such as zingiberene and curcumene and pungent compounds such as gingerols and shogaols (Ravindran and Babu, 2005). All of these compounds have the activity of antioxidant. The aim of the present study was to investigate the suitability of ginger root powder on growth performances and carcass characteristics of broiler chickens as rearing in hot climates.

Materials and methods

Fresh ginger root purchased from local market were cleaned and sliced into small pieces and dried for two days. The samples were further dried in a hot-air oven at $50\text{ }^{\circ}\text{C}$ for 24h and then ground

into powder. The determined curcumin content of the sample used in this study was averaged $1.29 \pm 0.04\%$ by weight. The obtained powder was incorporated into the experimental diets. A total of 250 one-day-old Cobb-500 male chicks provided by a local commercial hatchery were randomly allocated to one of 5 experimental treatments. Each treatment condition consisted of 5 replicate pens with 10 each. Each pen was $140 \times 110 \text{ cm}^2$ with one hanging tube feeder and one suspended drinker. The birds were reared on concrete flooring covered with wood shavings as litter material. The feeding trials lasted 56 d. The experiment was a completely randomized design and dietary treatments were as follows: a maize-soya bean meal based) basal (diet as control (C(basal diets supplemented with 100 mg/kg vitamin E) DL- α -tocopherol acetate (8, 16 and 24 g/kg ginger root powder. The dietary treatments were formulated with isonitrogenous and isocaloric nutrients while another nutrient contents were met by the recommendation of NRC) 1994(. Ingredient and nutrient composition of experimental diets were shown in Table 1. Feed and water were provided *ad libitum* throughout the experimental period)56 days of age(. The lighting programs consisted of 24 h light and birds were reared in open-side house as temperature maintained between 28-31 C° which during the summer season in the south of Thailand)April-May,2014(.

Chicken 3 were individually weighed at arrival and weekly throughout the experimental period on a pen basis. Feed intake was measured per pen and feed conversion ratio (FCR ; feed intake / weight gain) was calculated at the same intervals. All pens were checked for mortality daily and the birds that died during the experiment from each group were weighed and feed intake was adjusted accordingly. At 56 d of age, three birds per replicate randomly chosen were slaughtered and eviscerated carcass, abdominal fat, live, heart were collected, weighed and calculated as a percentage of live body weight. Data were subjected to ANOVA using GLM procedures as suggested by Khunthum (2006). Least significant difference comparison were made between treatment means for main effects when there was a significantly different at $P < 0.05$.

Results and discussion

It was found that the experimental diets had no significant effect ($P > 0.05$) on feed intake and body weight gain of chickens at the whole period of the experiment (Table 2.). Although supplementation of ginger root powder significantly improved ($P < 0.05$) feed conversion ratio and it was also found that there has no significant differences ($P > 0.05$) due to dietary treatments were observed on mortality. These results were consistent with the observations were made by Emadi and Kermanshashi) 2006 (who reported that an inclusion of 2.5 to 7.5 g/kg of diet with ginger root powder had no effect on feed intake and weight gain of broiler chickens. The most efficient feed conversion ratio)FCR (in broiler chickens fed diet supplemented with ginger powder has been due to the impact of growth promoter substances, such as phytogetic products, on performance could be related to a more efficient use of nutrients, which in turn results in an improved FCR)Durrani et al, 2006(. Ahmadi) 2010 (also reported that alike antibiotics, ginger powder could control and limit the growth and colonization of numerous pathogenic and non-pathogenic species of bacteria in chickens gut resulting in balanced by improved feed conversion ratio.

Table 2 Effect of ginger root powders on growth performances and carcass characteristics* of broiler chickens in hot climates)56 days of age(

Treatments	Body weight gain	Feed intake	Feed conversion ratio ^{1/}	Mortality	Eviscerated carcass	Abdominal fat ^{1/}
	(g)	(g)	(g/g)	(%)	(%)	(%)
Control	2,862	5,080	2.01 ^a	3.27	74.81	2.07 ^a
Vitamin E (100mg/kg)	2,866	5,087	1.98 ^{ab}	3.06	73.96	1.98 ^{ab}
Ginger powder (8g/kg)	2,887	5,064	1.84 ^b	3.15	74.62	1.64 ^b
Ginger powder (16g/kg)	2,891	5,070	1.81 ^b	3.09	73.81	1.58 ^b
Ginger powder (24g/kg)	2,880	5,061	1.83 ^b	3.14	74.57	1.52 ^b
SEM	28.7	59.1	0.03	0.97	4.23	0.08
<i>P-value</i>	<i>0.49</i>	<i>0.61</i>	<i>0.04</i>	<i>0.33</i>	<i>0.41</i>	<i>0.03</i>

^{1/a-b} Mean within a column and under each main effect with no common superscripts differ significant (P<0.05)

* mean values and SEM are based on 5 replicates

Table 1 Ingredient composition and calculated chemical analysis of the experimental diets (% as fed basis)

Item	Stater (1-21d)					Grower(22-42d)					Finisher(43-56d)						
	Contr ol	Positive Control (VitE100mg/k g)	8		24	Contr ol	Positive Control (VitE100mg/k g)	8		16	24	Contr ol	Positive Control (VitE100mg/k g)	8		16	24
			ginger	root	Powder (g/kg)			ginger	root	Powder (g/kg)	ginger			root	Powder (g/kg)		
Yellow corn	54.52	54.51	53.72	52.92	52.12	61.24	61.23	60.44	4	59.6	58.84	55.9	55.9	55.1	54.3	53.5	
Rice bran	-	-	-	-	-	-	-	-	-	-	-	7.0	7.0	7.0	7.0	7.0	
Soybean meal, 44% CP	39.0	39.0	39.0	39.0	39.0	31.0	31.0	31.0	31.0	31.0	31.0	28.0	28.0	28.0	28.0	28.0	
Palm oil	2.5	2.5	2.5	2.5	2.5	3.5	3.5	3.5	3.5	3.5	3.5	5.0	5.0	5.0	5.0	5.0	
Ginger powder	-	-	0.8	1.6	2.4	-	-	0.8	1.6	2.4	-	-	0.8	1.6	2.4	-	
Vitamin E	-	0.1	-	-	-	-	0.1	-	-	-	-	0.1	-	-	-	-	
Oyster shell	1.13	1.13	1.3	1.13	1.13	1.03	1.03	1.03	1.03	1.03	0.98	0.98	0.98	0.98	0.98		
Dicalcium phosphate Premix	1.5 0.5	1.5 0.5	1.5 0.5	1.5 0.5	1.5 0.5	1.93 0.5	1.93 0.5	1.93 0.5	1.93 0.5	1.93 0.5	1.78 0.50	1.78 0.50	1.78 0.50	1.78 0.50	1.78 0.50		
Sodium chloride	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35		
Lysine - HCL	0.16	0.16	0.16	0.16	0.16	0.17	0.17	0.17	0.17	0.17	0.20	0.20	0.20	0.20	0.20		
DL-Methionine	0.34	0.34	0.34	0.34	0.34	0.28	0.28	0.28	0.28	0.28	0.28	0.28	0.28	0.28	0.28		
Total	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0		
Calculated analysis (%)									0	100.0							
ME (kcal/kg)	3,080	3,080	3,080	3,080	3,080	3,180	3,180	3,180	0	3,180	3,200	3,200	3,200	3,200	3,200		
Crude Protein	23	23	23	23	23	20	20	20	20	20	18	18	18	18	18		
Calcium	1.09	1.06	1.07	1.09	1.07	0.98	0.97	0.97	0.98	0.96	0.08	0.81	0.08	0.81	0.08		
Avai phosphorus	0.45	0.46	0.45	0.43	0.44	0.4	0.41	0.4	0.4	0.41	0.35	0.34	0.35	0.34	0.35		
Lysine	1.3	1.31	1.3	1.31	1.3	1.1	1.11	1.09	1.1	1.11	0.93	0.92	0.93	0.92	0.91		
Methionine	1	1.01	1.01	1.0	1.01	0.78	0.77	0.78	0.78	0.77	0.66	0.65	0.66	0.65	0.66		
Threonine	0.87	0.86	0.85	0.86	0.85	0.81	0.8	0.81	0.8	0.81	0.68	0.69	0.68	0.67	0.68		
Tryptophane	0.25	0.25	0.24	0.23	0.24	0.20	0.19	0.20	0.19	0.19	0.17	0.17	0.18	0.17	0.18		

I Provided per kilogram of diet: retinyl acetate 3,500 IU; cholecalciferol, 1,000 ICU, DL- α -tocopherol acetate 4.5 IU; menadione sodium bisulfate complex, 2.8 mg; vitamin B12, 5.0 mg; riboflavin, 2.5 mg; pantothenic acid, 4.0 mg; niacin, 15.0 mg; choline, 172 mg; folic acid, 230 mg; ethoxyquin, 56.7 mg; manganese, 65 mg; iodine, 1 mg; iron, 54.8 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg

Fermentation pattern of alfalfa hay and *Ulva Fasciata* using in gas production technique

Paktinat*, M., Eyni, B. & M. Danesh Mesgaran

Department of Animal Science, Faculty of Agriculture, Ferdowsi University of Mashhad, P. O. Box 91775-1163, Mashhad, Iran

Abstract

The objective of this study was to evaluate fermentation characteristics of alfalfa hay and *Ulva Fasciata*. Samples of particle size of 2 mm were oven dried at 65°C for 48 h., then 200 mg of each were weighed and placed in four replicates into 125-ml capacity serum bottles. The gas production was continuously measured by incubating samples in buffered rumen from cow for 96hr. Cumulative gas production was recorded at 2,4,6,8,12,24,48,72 and 96 hr of incubation periods. In all of incubation time, gas volume (ml / 200 mg DM) of Alfalfa hay was higher than *Ulva fasciata*. The fractional rates (c) of gas production were highest (0/08) in Alfalfa hay ($p < 0.01$). Gas production from the insoluble fraction (b) in Alfalfa hay was higher (59/96) than *Ulva Fasciata* (46/93).

Keywords: Gas production, Alfalfa hay, Ulva fasciata

Introduction

The volume of produced gas should reflect the fermentation profile of a feed in the rumen. This led to development of in vitro gas production techniques (IVGPT), which simulate the rumen environment and allows estimation of kinetics of rumen fermentation by measuring cumulative gas production (Menke & Steingass, 1988). Algeria is a country with a littoral stretching over 1200 Km. This ecosystem is not well known, despite that it constitutes a reservoir of rich biodiversity. High quantities of seaweeds are available and very little valued; some species are washed-up and become a source of bad smell and pollution after decomposition (Hind et al. 2014). Over the past fifty years, the use of seaweeds has increased considerably, with the consequent increase in applied research in various related fields (Jiménez-Escrig et al., 2000). As known, seaweeds are used as sources of food for human nutrition in many countries because these natural resources are rich in soluble dietary fibers, proteins, minerals, vitamins, antioxidants and polyunsaturated fatty acids, with a low calorific value (Mohamed et al., 2012). They are also exploited in industry for agar, alginate and carrageenan productions. Their use as fertilizer, as fuel and cosmetics products has been also pointed (McHugh, 2003). *ulva* sp This is a small genus of marine and brackish water green algae. It is edible and is often called 'Sea Lettuce'. Species with hollow, one-layered thalli were formerly included in *Enteromorpha*, but it is widely accepted now that such species should be included in *Ulva* (Algaebase. 2010).

Material and Methods

This experiment was accomplished at the Mashhad Ferdowsi University. alfalfa hay at 50% blooming stage and *Ulva fasciata* from salty water of boshehr were harvested. All feed samples were ground to pass through a 2 mm screen and then oven dried (Behdad Co., BC Oven 70, 3493,

Iran) at 65°C for 48 h [AOAC]. The gas production procedure was performed as described by (Menke & Steingass, 1988). Rumen inoculum was collected from three ruminally fistulated steers (580 ± 4.5 kg, body weight) prior to offering the morning feeding. Animals were fed 10.4 kg DM, a diet containing alfalfa hay (50%), wheat straw (20%), barley grain (15%), soybean meal (14%) and mineral-vitamin premix (1%). Ruminal content was immediately blended and strained through four layers of cheesecloth to eliminate large feed particles and transferred to the laboratory in a prewarmed thermos. A sample of 200 mg was weighed into a 125-ml serum bottles in 3 runs and 4 replicates. Under continuous flushing of CO₂, 30 ml of buffered rumen fluid (ratio of buffer to rumen fluid was 2:1) was dispensed with pipetor pump into a 125 ml serum bottle. Gas production was measured at 2, 4, 6, 8, 10, 12, 16, 24, 48, 72 and 96 h. Cumulative gas production data were fitted to the exponential equation $Y=b(1-e^{-ct})$, where b is the gas production from the fermentable fraction (ml), c is the gas production rate constant (ml h⁻¹), t is the incubation time (h) and Y is the gas produced at time t. Data was analysis by completely randomized design using the general linear model procedure of SAS.

Results and Discussion

The results of this experiment showed (Table 1 & figure 1) that gas production parameters of alfalfa hay was higher than *ulva fasciata*. Maybe lower gas production and lower part of b and c in *ulva fasciata* is due to higher amount of Ash in this treatment higher amount of organic matter is result to higher gas production (Blummel et al, 1997). The low gas production constant rate might be due to the slow releasing of nitrogen content of *ulva fasciata* in the incubation bottles. Furthermore, the high nitrogen content of the feedstuff caused an elevated amount of gas. The positive correlation between crude protein content and gas production is in agreement with the study of Labri et al who reported a positive correlation between crude protein content and potential gas production.

Table 1- Gas production parameters of alfalfa hay and *ulva fasciata*

Treat	b	c	SEM
alfalfa hay	59.96 ^a	0.08 ^a	0.697
<i>ulva fasciata</i>	46.93 ^b	0.07 ^b	0.006

b : gas production from fermentable part (ml), c: gas production rate constant (ml h⁻¹), SEM: Standard error of means, Means with letters within each columns differed significantly (P<0.01).

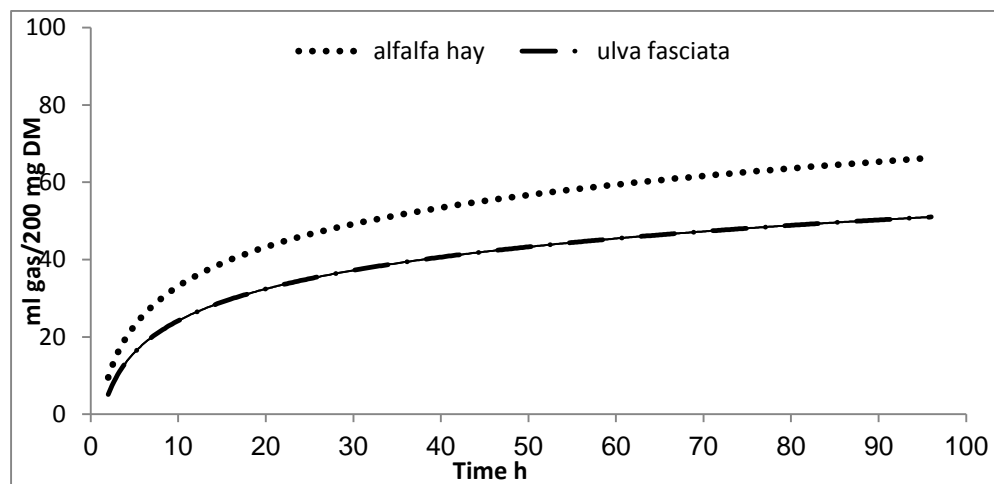


Fig. 1. Gas production profile of different treatments. As the standard errors were the same for all of the treatments, they are not shown in the figure. (SEM=1.46, R²=0.96)

References

- Association of Official Analytical Chemists. Official Methods of Analysis. 15th ed. AOAC, Arlington, VA; 1990.
- Blummel, M., P. Bullerdieck. 1997. The need to complement gas production measurements with residue determination from *in sacco* degradability to improve the prediction of voluntary intake of hays. *Animal Science* 64: 71 – 75.
- Guiry M.D., Guiry G.M. (2010) AlgaeBase. World-wide electronic publication, National University of Ireland, Galway.<http://www.algaebase.org>; searched on 20 October 2010.
- Jiménez-Escrig, A. and Sanchez-Muniz, F.J. (2000). Dietary fibre from edible seaweeds: chemical structure, physicochemical properties and effects on cholesterol metabolism. *Nutr Res.* 20 (4): 585- 598.
- Labri A, Smith JW, Kurdy IO, Adeknlé IO, Raji AM, Ladipo DO. Chemical composition, rumen degradation, and gas production characteristics of some multipurpose fodder trees and shrubs during wet and dry season in the humid tropics. *Anim Feed Sci Technol.* 1998;72:81-96.
- McHugh, D. J. (2003). A guide to the seaweed industry. FAO fisheries technical paper 441. Rome: Food and Agricultural Organisation of the United Nations.
- Menke KH, Steingass H. Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Anim Res Dev.* 1988;28:7–25.
- Mohamed, S., Hashim, S.N. and Rahman, H.A. (2012). Seaweeds: A sustainable functional food for complementary and alternative therapy. *Trends Food Sci Tech.* 23:83-96.
- Zitouni Hind 1, 2, Arhab Rabah 3, Boudry Christelle 2, Bousseboua Hacène 1, Beckers Yves. 2014. Chemical and biological evaluation of the nutritive value of Algerian green seaweed *Ulva lactuca* using *in vitro* gas production technique for ruminant animals.

To compare intestinal available protein of *Ulva Fasciata* with alfalfa hay using a new gas technique

Eyni, B., Paktinat*, M. & M. Danesh Mesgaran

Department of Animal Science, Faculty of Agriculture, Ferdowsi University of Mashhad, P. O. Box 91775-1163, Mashhad, Iran

Abstract

This study was carried out in to compare the amount of ruminant intestinal available crude protein (uCP) of *Ulva Fasciata* (seaweed) with alfalfa hay. using a new modified gas technique. The plant samples of particle size of 2 mm were oven dried at 65°C for 48 h. To perform the gas test, rumen fluid was collected before the morning feeding from two rumen fistulated Holstein dairy cows (640 ± 38 kg, body weight). Feed samples (200 mg) and blanks (only 30 ml of buffered rumen fluid) incubated simultaneously in three repeats for 8, 24 and 48 hours. At the end of the each incubation time, the uCP was calculated as non-ammonia N which was calculated by subtracting the amount of ammonia N released in the incubation medium of the total incubated N (sum of N content of feed sample and ammonia N in blanks). Effective uCP (EuCP) was calculated via an exponential equation using the estimated uCPs at 8, 24 and 48 h post incubation. Effective uCP (at the passage rate of 0.06/h) of *Ulva fasciata* (122/91) were significantly ($P < 0.01$) higher than that of alfalfa hay (152/67). The result showed that uCP in 24 and 48 h was higher in *Ulva Fasciata* (149.58, 125/1) than the value obtained from alfalfa hay (105/43, 49/91) respectively ($P < 0.05$).

Keywords: *Ulva fasciata*, alfalfa hay, protein

*Corresponding author: Paktinat55@yahoo.com

Introduction

Seaweeds could be a potentially valuable resource for ruminants feeding but not yet valued by the Algerian scientific community, although their utilization as feed supplements for livestock is not new. Arieli et al (1993) showed that seaweed *Ulva lactuca* is an interesting feed supplement for sheep, but not for poultry. Ventura & Castanon (1998) pointed out that this species represents medium quality forage for goats, with high protein content. Hansen et al (2003) concluded that seaweeds *Laminaria digitata* and *Laminaria hyperborea* have the potential to be used as an alternative feed source for small ruminants under some conditions. Mora castro et al (2009) suggested that marine algae *Macrocystis pyrifera* represents a good unconventional feeding as a nutritional supplement for goats. Rjiba ktita et al (2010) concluded that seaweeds *Ruppia maritima* and *Chaetomorpha linum* could be used as alternative feed resources for growing lambs during drought periods. The objective of this study was determining utilizable crude protein of *ulva fasciata* and alfalfa hay, and investigating possibility of replacement of alfalfa with this seaweed.

Material and methods

In our study rumen fluid was collected from three ruminally fistulated steers (580 ± 4.5 kg, body weight) prior to offering the morning feeding. Animals were fed 10.4 kg DM, a diet containing alfalfa hay (50%), wheat straw (20%), barley grain (15%), soybean meal (14%) and mineral-vitamin premix (1%). The fluid was extracted before the morning feeding and transported in a pre-warmed thermos, which was completely filled, and immediately sealed. The rumen fluid was filtered through two layers of cheesecloth into a warm flask and then added to the reduced buffer solution. After allowing 15 min to acclimatise, 30 ml of the solution was added to a pre warmed syringe containing 200 ± 30.0 mg substrate. Syringes were immediately placed in a rotary incubator which had been prewarmed to 39°C . The starting time of the incubation was recorded after all syringes had been filled. Each feedstuff was analysed in duplicate (analytical replicates) and over two runs using different batches of rumen fluid (statistical replicates). At the end of each incubation time (8 and 48 h) gas volume was recorded and syringes put on ice to stop microbial activity. Syringes remained in the ice for a minimum of 2 h and until required for ammonia analysis. Gas production (GP) was also recorded at 24 h for use in calculation of ME. At both the 8 and 24 h readings, the plunger was set back to 30 ml (not for the blank). A blank, containing rumen fluid/buffer solution without added substrate ($\text{NH}_3\text{N}_{\text{blank}}$), was also incubated in duplicate alongside the samples for 8 and 48 h. Ammonia-N (mg $\text{NH}_3\text{-N}/30$ ml) from both the blank and from the syringes containing substrate ($\text{NH}_3\text{N}_{\text{sample}}$) was measured by distillation and used in the following calculation:

$$\text{uCP (g/kg DM)} = \frac{\text{NH}_3\text{N}_{\text{blank}} + \text{N}_{\text{sample}} - \text{NH}_3\text{N}_{\text{sample}}}{\text{weight (mg DM)}} \times 6.25 \times 1000$$

to calculate effective uCP to assumed passage rates (K_p) of 0.02, 0.04 and 0.06/h using the formula:

$$\text{effective uCP} = y + a \times \ln \left(\frac{1}{K_p} \right)$$

Data analysis was carried out with the SAS GLM procedures (SAS, 1996).

Results and Discussion

Results of this experiment are in tables 1 and 2. amount of uCP, relative uCP, Effective uCP and relative effective uCP of *ulva fasciata* was higher than alfalfa hay. The uCP is the sum of rumen undegraded feed protein (UDP) and microbial protein available at the duodenum; and effective uCP is the uCP value accounting for a specific passage rate (melesse et al, 2013). In contrast to metabolisable protein (AFRC, 1993), uCP is based on crude protein not taking into account true protein and its intestinal digestibility maybe lower amount of ammonia production is result to high amount of CP flow in to deudenum that could be Advantageous for ruminant. this result show that *ulva fasciata* is a good replacement for alfalfa hay although further investigation is required.

Table 1- utilizable crude protein (uCP), Amoniacontent and Relative utilizable crude protein (uCP/CP%) of *ulva fasciata* and alfalfa hay (g/kg DM)

	ulva fasciata			Alfalfa hay		
	8	24	48	8	24	48
Amonia	0.03 ^b ±0.12	0.6 ^b ±0.14	1.38 ^b ±0.15	0.96 ^a ±0.12	2.98 ^a ±0.14	4.89 ^a ±0.15
uCP	167.5±4	149.58 ^a ±4.4	125.1 ^a ±4.5	168.4±4	105.43 ^b ±4.4	49.91 ^b ±4
Relative uCP	99.7 ^a ±2.04	89.03 ^a ±2.2	74.46 ^a ±2	84.75±2.04	53.06 ^b ±2.2	25.12 ^b ±2

Means with letters within each raw differed significantly ($P < 0.05$).

Table 2- Effective utilizable crude protein (EuCP) and Relative Effective utilizable crude protein (EuCP/CP%) of ulva fasciata and alfalfa hay (g/kg DM)

	ulva fasciata	Alfalfa hay	SEM
EuCP (g/kg)	152.67 ^a	122.91 ^b	5.6
EuCP (uCP/CP%)	90.87 ^a	61.85 ^b	2.9

Means with letters within each raw differed significantly ($P < 0.05$), SEM: Standard error of means.

References

- AFRC, 1993: Energy and Protein Requirements of Ruminants. CAB International, Wallingford, UK.
- Arieli, A., D. Sklan and G. Kissil, 1993. A note on the nutritive value of *Ulva lactuca* for ruminants. *Anim. Prod.* 57: 329-331.
- Hansen, H.R., B.L. Hector and J.Feldmann, 2003. A qualitative and quantitative evaluation of the seaweed diet of North Ronaldsay sheep. *Anim. Feed. Sci. Technol.* 105: 21-28.
- Melesse A., H. Steingass, J. Boguhn and M. Rodehutschord. 2013. In vitro fermentation characteristics and effective utilisable crude protein in leaves and green pods of *Moringa stenopetala* and *Moringa oleifera* cultivated at low and mid-altitudes. *Journal of Animal Physiology and Animal Nutrition.* 97: 537–546.
- Mora Castro, N., M. Casas Valdez. A. Marín Álvarez. R N. Águila Ramírez. I. Sánchez Rodríguez. H. Hernández Contreras and L.Sanginés García, 2009. The kelp *Macrocystis pyrifera* as nutritional supplement for goats. *Revista Científica.* XIX. 1: 63-70.
- Rjiba Ktita, S., A.Chermi and M.Mahouachi, 2010. The use of seaweeds (*Ruppia maritima* and *Chaetomorpha linum* for lamb fattening during drought periods. *Small Ruminant Res.* 91:116-119.
- Ventura, M.R. and J.I.R. Castanon, 1998. The nutritive value of seaweed (*Ulva lactuca*) for goats. *Small Ruminant Res.*, 29: 325-327.

Effect of *Curcuma domestica* stock solution on layer performance, egg quality, and antioxidant activity

Yuli Frita N^{1,2}, H. L. Chang¹, M. J. Lin¹, and E. Widodo²

¹Department of Animal Science - National Pingtung University of Science and Technology, Pingtung, Taiwan

²Animal Husbandry Faculty - Brawijaya University, Malang, Indonesia

tyasfrita@yahoo.co.id, hlachang@mail.npust.edu.tw, meijen@mail.npust.edu.tw,

ekowidodo.ub@gmail.com

Abstract

The aim of this research was to evaluate the effect of *Curcuma domestica* stock solution on layer performances, egg quality, and antioxidant activity of egg yolk. A total number of 32 *Lohman LSL-lite* white laying hens were divided into 4 treatments; there were 8 replication birds in individual cages. Laying hens were fed with 4 experimental diets, basal diet and diets with 1%, 2%, and 3% of *Curcuma domestica* 10% stock solution. Eggs were collected daily and analyzed of the eggs which were divided into three age stages, namely stage 1 (week 22 - 25), stage 2 (26 - 29), and stage 3 (30 - 33), respectively. The respective layer performances and egg quality were determined every week at every age stage. Antioxidant activity and color stability were determined every week at second age stage. The data was analysed using GLM in a windows-based software package, SAS version 9.1. Data was obtained from different level, age stage, and interaction among level and age stage. The differences were tested by LSM. Significant level used in the group comparisons was set at $p < 5\%$. The addition of *Curcuma domestica* 10% stock solution did significantly improve laying performances, egg quality, and antioxidant activity. Addition of 3% *Curcuma domestica* stock solution increased layer performances including water consumption, hen day egg production, and egg weight as well as antioxidant activity including FRAP, iron chelating, and DPPH on egg yolk. Moreover, addition of 2% *Curcuma domestica* 10% stock solution increased egg shell thickness and egg shell strength. In summary, the use of 3% addition of *Curcuma domestica* 10% stock solution had ability to improve layer performances and antioxidant activity of egg yolk on laying hens.

Keywords: antioxidant activity, Curcuma domestica, egg quality, layinghen, performance

Introduction

Recently, feed additive has a high chemical substance that affected on animal health. However, the addition of chemicals substance in feed would bring a high risk to health (Dibner & Buttin, 2002). The addition of chemical substances will not only have negative effect in animals, however will also have negative impact on livestock production. The use of plant herbs as a natural supplement in livestock feeds has beneficial effect on the health animals (Radwan, 2003). Natural herbs have

no risk on accumulated residues of chemical substances in livestock product and it has no harmful effect in animal health. *Curcuma domestica* contains bioactive compound of curcumin. It was commonly used as flavoring, coloring, and preservative agents. The important natural substance of *Curcuma domestica* can be used as food colorant and spice with the high antioxidant and antimicrobial properties (Pruthi, 1980).

Materials and Methods

The experimental was carried out at the Poultry Research Farm and Dairy Laboratory, Department of Animal Science, National Pingtung University of Science and Technology (NPUST). Thirty-two (32) *Lohman LSL-Lite* white laying hens were divided into 4 treatments in which each treatment had 8 replications with 1 laying hen per replication. Individual battery cages (40 x 30 x 40) cm was used and equipped with feeder and bottle drinker. In this experimental were used 4 treatments during 12 weeks, consist of control (basal diet + drinking water), T1 (basal diet + 1% of *Curcuma domestica* stock solution), T2 (basal diet + 2% of *Curcuma domestica* stock solution), T3 (basal diet + 3% of *Curcuma domestica* stock solution). All treatments were measured into analysis layer performance, egg qualities, and antioxidant activity.

Layer performances were recorded daily in every treatment at the first stage, second stage, and third stage. The layer performances include feed intake, water consumption, hen day egg production (HDP), egg weight and egg mass, and feed conversion ratio (FCR).

Egg qualities were recorded weekly by two eggs in each treatment at the first age stage, second age stage, and third age stage. Two eggs in each treatment were broke up to analyzed egg quality. The egg quality includes Haugh unit (HU), eggshell thickness, eggshell strength, eggshell percentage, and egg shape index. Antioxidant activity of egg yolk was measured by DPPH (1,1-Diphenyl-2-picrylhydrazyl), FRAP (Ferric Reducing Antioxidant Power), and iron chelating. Two eggs yolk per treatment were collected weekly at second age stage.

The data was analyzed by GLM (General Linear Model) in a windows-based software package, SAS version 9.1. Data was obtained from different level, age stage, and interaction among level and age stage. The differences between level and age stage were tested by least squares mean. Significant level used among treatment comparisons was set at $p < 5\%$.

Results and Discussion

Different level of *Curcuma domestica* 10% stock solution improved water consumption, HDP, and egg weight ($p < 0.01$) compared control group. Results showed that the addition of 3% *Curcuma domestica* 10% stock solution increased 8.15 mL of water consumption, 6.4% of HDP, and 3.77 g of egg compared control group. Agreed with statement El-Sheikh (2006) that herbal plants influenced appetite and water-consumption. *Curcuma domestica* increased enzymatic activity in the digestive tract resulting in improving nutrient utilization. Therefore, *Curcuma domestica* improved HDP and egg weight.

The effect of different levels of layer fed *Curcuma domestica* 10% stock solution on egg quality has significantly ($P < 0.05$) improved 0.05 mm of egg shell thickness and 0.84% of egg shell percentage. Egg quality could be measured by increasing the number fertilized of eggs as well as improved quality of the embryos examined by microscopic examination. The organic

material from herbal plants has calcium binding properties and its organization during shell formation influences the strength of the shell.

Curcuma domestica 10% stock solution increased FRAP of egg yolk. The addition of 3% *Curcuma domestica* 10% stock solution improved 18% FRAP compared control group. The increasing of FRAP has the ability as antioxidant, anti-inflamantory, and anti-tumor (Radwan, 2008). In this case, *Curcumin* as an antioxidant activity in human and animals. Increase of curcumin has positive impact on antioxidant activity of animal production, i.e. egg yolk of layer hens.

Curcuma domestica 10% stock solution significantly ($p < 0.05$) affected the iron chelating. The addition of 3% *Curcuma domestica* 10% stock solution improved 16% of the stability of iron chelating compared with those observed in control group. Antioxidant properties of the yolk were evaluated based on iron ions chelation effect. Egg yolk has been recognized have strong antioxidant (Sakanaka, 2006). Free radical scavenging assay of iron chelating confirmed that the higher hydrolysates shown the higher of antioxidant properties on egg yolk. Addition of *Curcuma domestica* 10% stock solution significantly ($p < 0.05$) improved in DPPH. The addition of 3% *Curcuma domestica* 10% stock solution increased DPPH compared control group. DPPH measured indicated the ability of antioxidants to inhibit free radicals by donating a hydrogen atom. Antioxidant activity contributes as free radical scavenging nature to prevent atom hydrogenate (Sathisa et al., 2011). Addition of *Curcuma domestica* 10% stock solution has ability of increasing DPPH of egg yolk

Table 1. Different level and age stage of addition *Curcuma domestica* 10% stock solution on layer performances.

Variable	Feed intake (g/bird/day)	Water consumption (mL/bird/day)	HDP (%)	Egg weight (g)	Egg mass	FCR
Control	100.90	305.81 ^b	91.96 ^c	49.23 ^b	48.42	2.15
T1 (1%)	101.09	312.32 ^a	94.56 ^b	52.65 ^a	49.87	2.04
T2 (2%)	101.18	313.88 ^a	95.93 ^b	52.88 ^a	50.70	2.01
T3 (3%)	101.20	313.96 ^a	98.36 ^a	53.00 ^a	49.20	2.05
SEM	0.97	8.03	3.87	3.59	4.18	0.19

SEM: Standard error of mean.

^{a-c} Means within row with different superscripts were significantly different ($P < 0.05$).

Table 2. Effect of different level and age stage of *Curcuma domestica* 10% stock solution on egg quality.

Items	Egg shape index (%)	Egg shell thickness (mm)	Egg shell percentage (%)	Egg shell strength (kgf)	Haugh unit
Control	75.89	0.37 ^c	13.33 ^b	3.28	85.70
T1 (1%)	75.78	0.38 ^{bc}	14.10 ^a	3.33	86.81
T2 (2%)	75.84	0.41 ^{ab}	14.23 ^a	3.41	86.71
T3 (3%)	76.01	0.42 ^a	14.17 ^a	3.38	85.65
SEM	2.65	0.06	1.16	0.43	4.09

SEM: Standard error of mean.

^{a-c} Means within row with different superscripts were significantly different ($P < 0.05$).

Table 3. Effect of addition level of *Curcuma domestica* 10% stock solution on antioxidant activity of egg yolk.

Variable	Treatment				SEM
	Control	T1 (1%)	T2 (2%)	T3 (3%)	
FRAP (%)	0.866 ^c	0.879 ^c	0.924 ^b	1.020 ^a	0.014
Iron chelating (%)	94.371 ^b	103.612 ^a	104.338 ^a	109.603 ^a	2.269
DPPH (%)	32.003 ^b	34.809 ^{ab}	37.216 ^{ab}	39.602 ^a	2.087

SEM: Standard error of mean.

^{a-c} Means within row with different superscripts were significantly different (P <0.05).

References

- Dorman, H. J., P. Surai, and S. G. Deans. 2000. In vitro antioxidant activity of a number of plant essential oils and phytoconstituents. *J. Essent. Oil Res.* 12: 241-248.
- El-Sheikh, H. M. M., 2006. Effect of probiotic supplementation to broiler chicken diets on its performance, carcass characteristics and blood constituents, M. Sc. Thesis, Fac. of Agric. (Saba Basha), Alexandria Univ., Egypt.
- Genedy, S. and G. H. S. Zeweil, 2003. Evaluation of using medicinal plants as feed additives in growing Japanese quail diets. The 68th Scientific Conference of Polish Animal Production Society, Krakov, Poland.
- Hernandez, F., J. Madrid, V. Garcia, J. Orengo, and M. D. Megias, 2004. Influence of two plant extracts on broilers performance, digestibility and digestive organ size. *Poult. Sci.* 83:169-174.
- Kermanshi, H. and A. Riasi. 2006. Effect of turmeric rhizome powder (*Curcuma longa*) and soluble NSP degrading enzyme on some blood parameters of layer hens. *Int. J. Poult. Sci.* 5:494-498.
- Malekizadeh, M., M. M. Moeini, and S. Ghazi, 2012. The effects of different levels of ginger (*Zingiberofficinalerosc*) and turmeric (*Curcuma longalinn*) rhizomes powder on some blood metabolites and production performance characteristics of layer hens. *J. Agr. Sci. Tech.* 14:127-134.
- Radwan, N. L. 2003. Effect of using some medicinal plants on performance and immunity of broiler chicks. Ph.D. Thesis. Poult.Nutr.Dept. Fac. Agric. Cairo University.
- Radwan, N. L., R. A. Hassan, E. M. Qota, and H. M. Fayek, 2008. Effect of natural antioxidant on oxidative stability of eggs and productive and reproductive performance of layer hens. *Int. J. Poult. Sci.* 7:134-150.

Effect of yeast fermented fresh cassava root fed beef cattle on digestibility

Chamnanwit Promkot^{*1}, Peerapothn Nitipothn², Nonthasak Piampol³, Somjit Kanthaprom² and Sarawut Doungmawong²

¹ Faculty of Natural resources, Skon Nakhon Campus ² Faculty of Agro-Industrial Technology, Kalasin Campus ³ Faculty of Science and Art, ^{1,2,3} Rajamangala University of Technology Isan

Abstract

The objective of this study was to evaluate effects of level of yeast-fermented fresh cassava root (0, 10, 20 and 30% dry matter) in concentrate diet on dry matter intake (DMI) and nutrient digestibility. Four, non-pregnant female Brahman beef cattle (body weight 256 ± 11.5 kg) were randomly allocated to 4 treatments in 4 x 4 Latin square design with 21-d periods. Fresh cassava root was girded and fermented for 21 days then fermented with yeast as yeast-fermented fresh cassava root. Cattle was fed rice straw as roughage. Dry matter intake was not difference among the treatments. However, ADG was trend to increase when yeast-fermented fresh cassava root was fed at 20 % in concentrate diet. Nutrients digestibilities were not differ among the treatments. However fiber digestibility (NDF and ADF) were trend to increase when yeast-fermented fresh cassava root was fed. It is concluded that yeast-fermented fresh cassava root could be used as protein source in cattle diet up to 30% with trend to enhance the dry matter intake and NDF digestibility.

Keywords: Cassava root, Beef cattle Feed, Yeast, Yeast-fermented fresh cassava root

Introduction

Incorporation of microbial additives such as a culture of *Saccharomyces cerevisiae* to the diet has become a common practice in ruminant nutrition. Yeast supplementation for ruminants by the use of dry cassava chips and fresh cassava root fermented with pure culture of *S. cerevisiae* had been studied and could increase its protein content from 3 % in non-fermented cassava root to 21-30 % in fermented products (Boonnop et al., 2009). Boonnop et al. (2010) studied the effects of yeast fermented cassava chips as a protein source replacement of soybean meal (SBM) in a concentrate diet and fed to beef cattle, found that yeast fermented cassava chips could fully replace soybean meal and improved rumen YEFECAP has a potential for use as a protein source for improving rumen fermentation efficiency in ruminants fermentation efficiency and nutrient digestibilities. Recently research by our team that No data is available for feeding of fresh cassava root fermented with pure culture of *S. cerevisiae* to beef cattle. The objective of this study was to evaluate effects of level of yeast-fermented fresh cassava root (0, 10, 20 and 30% dry matter) in concentrate diet on dry matter intake (DMI) and nutrient digestibility.

Materials and methods

Animals, design: Four, non-pregnant female Brahman beef cattle (body weight 256 ± 11.5 kg) were randomly allocated to 4 treatments in 4 x 4 Latin square design with 21-d periods.

Experimental feeds and management: The cattle were individually penned and clean fresh water and mineral blocks were offered free choice. Four dietary treatments were concentrate ration with four levels of yeast-fermented fresh cassava root (YEFECAR) (0, 10, 20 and 30% dry matter). Ingredient compositions of concentrate feed and roughage (rice straw; RS) are shown in Table 1.

All concentrate mixtures had similar CP and were given to animals in two equal parts (1.5% of body weight per day) while rice straw was given *ad libitum*.

Sampling procedure, data collection and chemical composition analysis: Rice straw and concentrate were sampled daily during the collection period and were composited by period prior to analyses. Data of BW was recorded on the day of 14 and 21 of each period. Feed and fecal samples were collected during the last five days of each period. Fecal samples were collected by rectal sampling during the last 2 days of sampling. Composited samples were dried at 60°C and ground (1 mm screen) and then analysed for DM, ash and CP content (AOAC, 1990), NDF and ADF (Goering and Van Soest, 1970) and acid-insoluble ash (AIA) (Van Keulen and Young, 1977). The AIA was used to estimate digestibility of nutrients (Van Keulen and Young, 1977). Rumen fluid (sampling by stomach tube) samples were collected at 0 and 4 h-post feeding. Rumen fluid was immediately measured for pH using a portable pH meter.

Statistical analysis: Statistical analyses were performed using the GLM procedure of SAS. Data were analyzed using the model $Y_{ijk} = \mu + M_i + A_j + P_k + \epsilon_{ijk}$, where Y_{ijk} is the observation from animal j , receiving diet i , in period k ; μ , the overall of mean, M_i , the mean effect of level of concentrate ($i = 1, 2, 3, 4$), A_j , the effect of animal ($j = 1, 2, 3, 4$), P_k , the effect of period ($k = 1, 2, 3, 4$), ϵ_{ijk} the residual effect. Mean differences with a significant F value ($p < 0.05$) for treatment were statistically compared using Duncan's New Multiple Range Test. Trends were declared at $0.05 < P < 0.10$.

Results and discussion

Total intake (concentrate and RS intakes) was no significant differences among the treatments. However intake was trend ($P < 0.1$) to increase when animal fed YEFECAR diet. This finding is consistent with the observation of previous works (Boonnop et al., 2010) that while using yeast-fermented cassava chip as protein sources in concentrate diets for dairy steers, it could improve DMI. The DMI tended to be higher for animal fed YEFECAR diet compared to controlled diet in this study may be due to higher fiber (NDF, ADF) digestibility.

Nutrients digestibilities were not differ among the treatments. However fiber digestibilities (NDF and ADF) were trend ($P < 0.01$) to increase when YEFECAR was added to the concentrate diet. Improvement of NDF digestibility in animals fed a YEFECAR diet may be due to feeding yeast-fermented cassava product had increased numbers of cellulolytic, amylolytic and proteolytic rumen bacteria (Boonnop et al., 2010; Wanapat et al., 2011a, 2011b) resulted in improved fiber digestibility (Boonnop et al., 2010; Wanapat et al., 2011a, 2011b). ADG was trend ($P < 0.1$) to increase when yeast-fermented fresh cassava root was fed at 20 % in concentrate diet. This finding could be due to higher intake and nutrients digestibility in animal fed 20 % YEFECAR.

It is concluded that yeast-fermented fresh cassava root could be used as protein source in cattle diet up to 30 % dry matter in concentrate feed which not have any negative effect on dry matter intake and nutrients digestibility.

Table 1 Ingredient compositions (% of dry matter) of concentrate treatments (difference level of yeast fermented cassava root, YEFECAR) and roughage: rice straw (RS) and ADG, dry matter intakes, rumen pH and nutrient digestion coefficients in beef cattle fed differences level of YEFECAR

Item	YEFECAR in concentrate feed (%DM)				RS
	0 (control)	10	20	30	
Ingredient: Cassava chip	67.7	63.1	58.1	53.1	
YEFECAR	0.0	10.0	20.1	30.0	
Soybean meal	15.2	9.9	5.1	0.0	
Rice bran	10.1	10.1	10.1	10.0	
Molasses	3.3	3.3	3.3	3.3	
Urea	1.2	1.3	1.3	1.3	
Salt	1.1	1.1	1.1	1.1	
Sulphur	0.2	0.2	0.2	0.2	
Mineral mix ¹	1.1	1.1	1.0	1.1	
Dry matter (%)	88.3	78.8	71.1	64.9	90.4
Crude protein (%)	10.1	10.1	10.0	10.0	2.6
Neutral-detergent fiber (%)	33.3	34.1	34.9	36.0	80.4
Acid-detergent fiber (%)	10.9	13.1	13	15.2	67.8
Ash (%)	10.7	11.7	11.2	14.5	11.3
					SEM
Dry matter intake, kg/hd/d	8.8	8.4	8.6	8.5	0.3
% body weight	2.7	2.8	2.9	2.8	0.1
ADG, kg/hd/d	0.792	1.016	0.961	0.724	0.2
Rumen pH, 0 h, post-feeding	6.9	6.9	6.8	6.8	0.04
4 h	6.8	7.0	7.2	6.8	0.06
mean	6.9	7.0	7.0	6.8	0.05
Digestion coefficients (%)					
Dry matter	53.5	54.4	54.6	54.1	1.3
Crude protein	58.5	59.2	59.1	58.8	1.2
Neutral-detergent fiber	45.3	46.2	46.4	46.0	1.4
Acid-detergent fiber	40.2	41.2	41.5	41.7	1.6

¹Mineral mix (each kg contains): Iron 2.14 g; Iodin 0.15 g; Sulphur 11.82 g; Copper 0.23 g; Magnesium 0.96 g; Sodium 2.68 g; Manganese 7.21 g; Cobalt 0.03 g; Phosphorus 19.60 g; Selenium 0.003 g; Zing 0.16; Calcium 204.03 g.

References

- AOAC. 1990. Official Methods of Analysis. 15th edn. Association of Official Analytical Chemists, Virginia.
- Boonnop, K., M. Wanapat and C. Navanukraw. 2010. Replacement of soybean meal by yeast fermented-cassava chip protein (YEFECAP) in concentrate diets fed on rumen fermentation, microbial population and nutrient digestibilities in ruminants. *J. Anim. Vet. Adv.* 9:1727–1734.
- Boonnop, K., M. Wanapat, N. Nontaso and S. Wanapat. 2009. Enriching nutritive value of cassava root by yeast fermentation. *Sci. Agric.* 66:616–620.
- Goering, H. K. and P. J. Van Soest. 1970. Forage Fiber Analysis (Apparatus, Reagent, Procedures and Some Application): *Agric. Handbook No. 379.* ARS, USDA, Washington, DC.
- Van Keulen, J. and B. A. Young. 1977. Evaluation of acid insoluble ash as a neutral marker in ruminant digestibility studies. *J. Anim. Sci.* 44:282–287.
- Wanapat, M., K. Boonnop, C. Promkot and A. Cherdthong. 2011a. Effects of alternative protein sources on rumen microbes and productivity of dairy cows. *Maejo Int. J. Sci. Technol.* 5:13–23.
- Wanapat, M., S. Polyorach, V. Chanthakhoun and N. Sornsongnern. 2011b. Yeast-fermented cassava chip protein (YEFECAP) concentrate for lactating dairy cows fed on urea–lime treated rice straw. *Livest. Sci.* 139:258–263.

Changes in metabolic hydrogen flow on bovine rumen fermentation in response to cashew nut shell liquid

Mitsumori*¹, M., O. Enishi¹, T. Shinkai¹, K. Higuchi¹, Y. Kobayashi¹, A. Takenaka¹, K. Nagashima², M. Mochizuki² & Y. Kobayashi³

¹National Institute of Livestock and Grassland Science, Tsukuba, Ibaraki 305-0901, Japan,

²Idemitsu Kosan Co. Ltd, Sodegaura, Chiba 299-0293, Japan, ³Hokkaido University, Sapporo, Hokkaido 060-8589, Japan

Abstract

Analysis of the metabolic hydrogen flow estimated from concentrations of short-chain fatty acids (SCFAs) and methane was applied to evaluate effects of cashew nut shell liquid (CNSL), a methane inhibitor, on bovine rumen fermentation. Three cows were fed a concentrate and hay diet without or with a CNSL-containing pellet, which was blended with only silica (trial 1) or with several other ingredients (trial 2). Methane production was measured in a respiration chamber system, and energy balance and nutrient digestibility were monitored. Estimation of metabolic hydrogen demonstrated that a part of metabolic hydrogen was used for hydrogen gas production, and a large amount of it flowed into production of methane and SCFA in both trial 1 and 2, when CNSL was administered to the bovine rumen. The results obtained by regression analyses showed that the effect of CNSL supply on methane reduction was coupled with a significant ($P < 0.01$) decrease of acetate and a significant ($P < 0.01$) increase of propionate and hydrogen gas. These findings reveal that CNSL is able to reduce methane and acetate production, and to increase hydrogen gas and propionate production *in vivo* rumen fermentation.

Keywords: cashew nut shell liquid, hydrogen gas, metabolic hydrogen, methane, rumen

*Corresponding author: mitumori@affrc.go.jp

Introduction

Methane production by ruminants results in the contribution to global warming, which accounts for 28 to 33% of global anthropogenic methane emissions (FAO 2007). Energy loss through methane production in ruminants is thought to be 2 to 15% of the gross energy (GE) obtained from feed (Wright et al. 2004). Therefore, many attempts, directed toward mitigation of methanogenesis in the rumen with methane inhibitors or by controlling diet composition, have been reported (Mitsumori & Sun 2008; McSweeney & Mackie 2012).

Cashew (*Anacardium occidentale*) nut shell liquid (CNSL), a byproduct of cashew nut production in tropical countries, contains a large amount of anacardic acids, which shows antibacterial activities against gram-positive bacteria (Kubo et al. 1993). In our previous study (Shinkai et al. 2012), we demonstrated that addition of CNSL to diets of cows led to reduction of number of methanogens and methane production up to 38.3%, and enhancing propionate production in the rumen.

The present study, therefore, aimed to estimate metabolic hydrogen balance in the rumen and to investigate the flow of metabolic hydrogen when methanogenesis was inhibited by CNSL. The data analyzed in this study was collected in our previous study (Shinkai et al. 2012), in which CNSL was added to the diet and methane production in the rumen of Holstein cows was decreased.

Materials and Methods

Three ruminally fistulated, non lactating Holstein cows were repeatedly used in 2 feeding trials (mean body weight of 547 ± 31 kg for trial 1 and 548 ± 27 kg for trial 2). Procedures of the feeding trials were described in details in our previous report (Shinkai et al. 2012). Briefly, two types of CNSL pellets, pellet A formed with silica powder for the use in trial 1 and pellet B formed with alfalfa meal, defatted rice bran, silica powder, crude sugarcane molasses and tapioca flour for the use in trial 2.

The movement of metabolic hydrogen (2H) in the form of reduced protons was calculated from 2H utilised (2HU) and 2H produced (2HP) in rumen fermentation. The 2HP as a fermentation intermediate and 2H utilised in SCFA (2HUS) during a control period (2HUS_{Cont}) were estimated from the amount and molar proportions of acetate (C2), propionate (C3), butyrate (C4), isovalerate (Ci5) and valerate (C5), as previously reported (Demeyer 1991; Goel et al. 2009; Mitsumori et al. 2012), in the following equations:

$$2HP \text{ (mM)} = 2 \times C2 + C3 + 4 \times C4 + 2 \times Ci5 + 2 \times C5,$$

$$2HUS_{Cont} \text{ (mM)} = 2 \times C3 + 2 \times C4 + C5.$$

2HUS of the rumen administered with CNSL (2HUS_{CNSL}) was calculated from 2HUS_{Cont} and concentrations of total short-chain fatty acids (TSCFA) during a control period and CNSL period (TSCFA_{Cont} and TSCFA_{CNSL}, respectively), as follows:

$$2HUS_{CNSL} \text{ (mM)} = 2HUS_{Cont} \times TSCFA_{CNSL} / TSCFA_{Cont}.$$

The relationship among 2HU, 2HP, 2H recovery in methane (2HUM), and 2H recovery in H₂ (2HUH) is expressed by the following equation (Mitsumori et al. 2012):

$$2HU \text{ (mM)} = 0.9 \times 2HP = 2HUS + 2HUM + 2HUH.$$

Since methane formation was not inhibited in the controls, there was minimal H₂ production and thus 2HU was calculated according to follows (Goel et al. 2009; Mitsumori et al. 2012):

$$2HU = 2HUS + 2HUM = (2 \times C3 + 2 \times C4 + C5) + (4 \times \text{methane}).$$

Thus, methane production in controls (M_{cont}) was:

$$M_{cont} \text{ (mM)} = (2HU_{Cont} - 2HUS_{Cont}) / 4 = [(0.9 \times 2HP_{Cont}) - 2HUS_{Cont}] / 4$$

(Mitsumori et al. 2012). Methane production in CNSL treatments (M_{CNSL}) was calculated by using M_{cont} and methane production in controls and methane reduction rates (MR; %).

Results and Discussion

Acetate concentrations were significantly reduced ($P < 0.05$) in both trials 1 and 2. Whereas the acetate: propionate ratio remained unchanged between control and CNSL 1 period of trial 2, those of CNSL period in trial 1 and CNSL 2 period in trial 2 significantly differed from each control period. Methane production, total SCFA concentration, and *i*-valerate concentration of CNSL period in trial 1 were significantly lower than those of control. The 2HP of CNSL period in trial 1 was significantly different ($P < 0.05$) from that of control in trial 1. Ninety percent of 2HP was distributed into SCFA and methane in the case of control in both trials, but that was distributed into SCFA, methane, and hydrogen gas by the addition of CNSL. There was a tendency for 2HP to decrease with increasing MR. There were significant ($P < 0.01$) linear regressions of MR level and 2H flowing into methane (2HUM) or into hydrogen gas (2HUH). No significant relationship between 2HUS and MR level was observed. Simple regression analyses revealed the strong correlation ($P < 0.01$) between MR level and the concentration of acetate or propionate, and no significant relationship between that and the concentration of butyrate. There were significant ($P < 0.01$) relationships between MR level and methane, hydrogen gas, or propionate, although no significant relationship between that and SCFA.

The flow of metabolic hydrogen in rumen fermentation was estimated from concentrations of SCFA and methane. The estimation demonstrated that a part of metabolic hydrogen was used for hydrogen gas production, and a large amount of it flowed into production of methane and

propionate, when CNSL was administered to the bovine rumen. This study reveals that CNSL is able to reduce methane and acetate production, and to increase hydrogen gas and propionate production. Further study is required to validate the efficacy of CNSL under various feeding conditions.

References

- Demeyer D.I., 1991. Quantitative aspects of microbial metabolism in the rumen and hindgut. In: Jouany JP (ed.), *Rumen Microbial Metabolism and Ruminant Digestion*, INRA, Paris, France. pp 217–237.
- FAO, 2007. *Livestock's Long Shadow. Part IV. Livestock's role in climate change and air pollution*. Available from URL: <ftp://ftp.fao.org/docrep/fao/010/a0701e/a0701e03.pdf>
- Goel G., H.P.Makkar and K.Becker, 2009. Inhibition of methanogens by bromochloromethane: effects on microbial communities and rumen fermentation using batch and continuous fermentations. *Br. J.Nutr.* 101: 1484–1492.
- Kubo I., H.Muroi and M.Himejima, 1993. Structure-antibacterial activity relationships of anacardic acids. *J. Agric. Food Chem.* 41: 1016–1019.
- McSweeney C. and R.Mackie, 2012. *Micro-organisms and ruminant digestion: State of knowledge, trends and future prospects*. Background study paper No. 61. Food and Agriculture Organization of the United Nations (FAO), Rome.
- Mitsumori M. and W.Sun, 2008. Control of rumen microbial fermentation for mitigating methane emissions from the rumen. *Asian-Aust. J. Anim. Sci.* 21: 144-154.
- Mitsumori M., T. Shinkai, A. Takenaka, O. Enishi, K. Higuchi, Y. Kobayashi, I. Nonaka, N. Asanuma, S.E. Denman and C.S.McSweeney, 2012. Responses in digestion, rumen fermentation and microbial populations to inhibition of methane formation by a halogenated methane analogue. *Br. J.Nutr.* 108: 482-491.
- Shinkai T., O. Enishi, M. Mitsumori, K. Higuchi, Y. Kobayashi, A. Takenaka, K. Nagashima, M. Mochizuki and Y.Kobayashi, 2012. Mitigation of methane production from cattle by feeding cashew nut shell liquid. *J Dairy Sci.* 95: 5308-5316.
- Wright A.D., A.J.Williams, B.Winder, C.T.Christophersen, S.L. Rodgers and K.D.Smith, 2004. Molecular diversity of rumen methanogens from sheep in Western Australia. *Appl. Environ.Microbiol.* 70: 1263–1270.

Utilization of Cassava Pulp and Corncob Fermented with *Aspergillus niger* for Animal Feed: Effect on Protein Levels

Kunthika^{*1}, V., P². Pianpumepong, & S³. Wongsutthawas

¹Department of Applied Biology, Faculty of Sciences and Liberal Arts, ²Department of Post-Harvest and Processing Engineering, Faculty of Engineering and Architecture, ³Department of Agricultural Technology and Environment, Faculty of Sciences and Liberal Arts, Rajamangala University of Technology Isan, Muang District, Nakhon Ratchasima 30000, Thailand

Abstract

Cassava pulp and corncob can be used as animal feed. However, those contain high fiber, low protein and amino acid deficiency with sulfur compounds. It would be more valuable if the waste product is fermented with microorganisms to increase protein level in animal diets. This study used a solid state fermentation with 2 experiments. In experiment I, the proportion of cassava pulp and corncob fermented with *Aspergillus niger* was determined in the laboratory scales. The sample was divided into 5 groups: cassava pulp: corncob ratio; T 1 = 100:0, T 2 = 0:100, T 3 = 50:50, T 4 = 70:30, and T 5 = 30:70. Samples were stored at room temperature and mixed 3 times/d. The fermentation of T2, T 3 and T 5 had the same protein level (4.12, 4.10, and 4.10 g / 100 g, respectively). In experiment II, cassava pulp and corncob in the ratio of 30:70 was fermented in a 200 liter fermenter at 30°C for 7 d. The protein content increased to 4.96 g / 100 g compared to those fermentation in the lab scales. In conclusion, using cassava pulp and corncob fermented with *A. niger* strain would be a good protein source for animal diets.

Keywords: cassava pulp, corncob, solid state, protein, *Aspergillus niger*

*Corresponding author: kunthikar@hotmail.com

Introduction

Cassava and maize are grown widely in Thailand. Production volume is increasing every year especially in the Northeast of Thailand. Previous study found that cassava pulp and corncob have residual nutrients (Sriroth, et.al, 2000). Which can be used as animal feed. However, since cassava pulp and corncob low protein and high fiber has 2-2.5 percent of amino acids with sulfur compounds such as methionine as well as cassava pulp acids also hydrocyanic nick toxic to animal health, which is limited in its use as animal feed, hence the use of cassava pulp and corncob to a broiler is so low. Therefore, it is a concept that has improved the nutrition of cassava pulp and corncob by microbial fermentation method to add protein to rise. It can increase the use of cassava pulp and corncob diet in broilers.

Materials and Methods

Sample preparation. The fermentation process is divided into two levels, laboratory scale is marinated in erlenmeyer flask in 250 ml and experiment scale is fermented using a 200-liter fermentation tank. The experiment in the laboratory the sample is divided into five groups: Cassava pulp : corncob ratio T1 = 100: 0, T2 = 0: 100, T3 = 50:50, T4 = 70:30 and T5 = 30:70. Add cassava pulp and corncob in 250 ml Erlenmeyer flask to autoclave at 121°C for 15 minutes and leave to cool. Then fill the *A. niger* volume of 1 ml of sterile distilled water 25 ml and mix

well. Incubated at room temperature and shaking the sample once a day, every day for seven days after the time leading to drying at 55 ° C for 2 days, the samples were analyzed for protein. The experiment scale were selected samples from laboratory scale fermentation process the highest protein content were used condition. Add cassava pulp and corncob into stainless tank 200 liter add 40 liters of distilled water and then steamed by steam at a temperature of 95 ° C for 1 hour and left to cool in the fermenter . Then fill the *A. niger* down to 330 grams and mix to combine. Incubated at 30 ° C for 7 days mixed sample 3 times a day, the samples were proximate analysis. **Sample analysis.** The nutritional composition (ash, fat and crude fiber) of the fungi fermented cassava pulp and corncob product was evaluated using the methods reported by Cordenunsi et al., 2004 and the protein content was determined using the micro-Kjeldhal method (N x 6.25).

Results and Discussion

A study in the laboratory scale. By analyzing the protein content of the ingredients two kinds of cassava pulp and dried corncob prior to fermentation correlation was 1.86 and 2.46 g / 100 g , respectively, after samples fermented with inoculum *A. niger* at room temperature . 7 days of the study. The protein content of the samples T2 T3 and T5 are similar 4.12, 4.10 and 4.10 g / 100 g, respectively. Since, the protein content of the samples T2 T3 and T5 are similar. The experiments in the fermenter actually chose T5 example, due to low prices for the raw material of corncob to be used as animal feed further. T5 samples in the batch size of 200 kg real fermentation tanks under controlled temperature at 30 ° C for 7 days and then bring the samples to be dry. The results of the analysis of the chemical were showed in table 1.

Table 1. The proximate composition of sample (T5) fermented for 7 days.

Proximate composition	Time periods (day)							
	0	1	2	3	4	5	6	7
Crude ash (g/100g)	5.47	5.17	5.41	5.63	5.89	5.34	5.75	5.94
Crude fiber(g/100g)		18.58	18.67	18.63	18.72	18.39	18.91	18.34
Gross energy (kg calories/100g)		339.76	339.24	329.59	341.50	323.52	344.50	316.95
Crude protein (g/100g)		4.19	4.46	4.36	4.54	4.61	4.44	4.96
Crude lipid (g/100g)	0.46	0.52	0.43	0.55	0.30	0.36	0.70	0.59

Conclusion

The results obtained from this experiment could have a great impact on animal feed especially using local resources-based diets. The present results indicate that fermentation of cassava pulp and corncob by fungi can improve crude protein. This method could be more useful cassava pulp and corncob with *A. niger* strain would be a good source of protein for animal diets.

Acknowledgements

The researchers acknowledge Rajamangala University of Technology Isan Nakhon Ratchasima for facilities and financially supporting this research.

References

- AOAC, 1984. Association of Official Analytical chemists. Official methods of analysis, 12th Edition, Washington DC.
- Cordenunsi BR, D. M. Wenzel E, Genovese MI, Colli C, De Souza Gonçalves A, Lajolo FM. 2004. Chemical composition and glycemic index of Brazilian pine (*Araucaria angustifolia*) seeds. *Journal of Agricultural and Food Chemistry*.52:3412-3416.
- Sriroth, K., K. Piyachomkwan, S. Wanlapatit, y C. G. Oates. 2000. Cassava Starch Technology: The Thai Experience *Starch/Stärke* 52: 439-449.

Nutrient composition and *in vitro* ruminal degradability of selected local plants used as goat feed in Malaysia

Ghani, A.A.A.¹, M.S. Shahudin¹, M. Zamri-Saad¹, A.B. Zuki¹, H. Wahid¹, A. Kasim², M.S. Salisi¹ & H.A. Hassim^{*1}.

¹Faculty of Veterinary Medicine, ²Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

Abstract

A comparative study of nutrient composition and *in vitro* ruminal degradability of selected local plants (*Macaranga* sp. and *Mallotus* sp.) and Napier grass (*Pennisetum purpureum*) used as goat feed were carried out. Napier grass was used as a control group as it is common and widely been used (e. g fibre source) in goats diet. All plants were analyzed for nutrient composition by proximate analysis (dry matter; crude protein; crude fibre; crude fat) and *in vitro* ruminal degradability was performed to determine the total gas and volatile fatty acids (acetate; propionate; butyrate) production. The result obtained from proximate analysis revealed that there were significant difference ($P < 0.05$) of all nutrient composition for each local plant species against Napier grass. Dry matter content was highest in *Macaranga* sp. (50.92%) followed by *Mallotus* sp. (45.41%) and Napier grass (13.04%). Crude protein was highest in *Macaranga* sp. (7.18%) followed by *Mallotus* sp. (6.78%) and Napier grass (3.88%). *Mallotus* sp. has the highest crude fat content (5.22%) followed by *Macaranga* sp. (4.85%) and Napier grass (2.84%). As for crude fibre content, Napier grass showed the highest content (25.38%) followed by *Mallotus* sp. (15.47%) and *Macaranga* sp. (12.00%). For the *in vitro* ruminal degradability, the highest total gas production was shown by Napier grass (31.00ml), followed by *Macaranga* sp. (28.67ml) and *Mallotus* sp. (23.33ml). Acetate production was highest in *Mallotus* sp. (733.49mM/ml) followed by Napier grass (605.61mM/ml) and *Macaranga* sp. (599.85mM/ml) whereas propionate production was highest in *Mallotus* sp. (28.61mM/ml) followed by Napier grass (24.45mM/ml) and *Macaranga* sp. (24.23mM/ml). As for butyrate production, Napier grass showed the highest value (11.26mM/ml), followed by *Macaranga* sp. (11.19mM/ml) and *Mallotus* sp. (10.36mM/ml). However, there was no significant difference shown for acetate, propionate and butyrate production between all samples. Thus, based on the nutrient composition and *in vitro* ruminal degradability findings, it shows that these local plants could be used as a good feed source for goats. In addition, a proper feeding regime using these local plants need to be considered in order to ensure a balance diet for the goats.

Keywords: *Macaranga* sp., *Mallotus* sp., napier grass, proximate analysis, ruminal degradability

*Corresponding author: haslizaabu@upm.edu.my

Introduction

Local plants had supported grazing animals even before domestication as these local plants are abundant and easily accessible in the environment. Even till now, most of small holder farmers in Malaysia are practicing feeding these local plants to goats especially at times of short supply (FFTC, 2002). Indeed, this can help to reduce the dependence on concentrates and supplements in total mix ration subsequently reducing the cost of feed and feeding. There are also farmers practicing a combination of local plants growing around the farm area together with the primary feed (e.g. concentrate; grass) offered to their livestock. In addition, the abundant availability of the local plants in Malaysia has derived the small holder goat farmers to integrate these local plants into the total feed ration of the livestock. Local plants such as *Macaranga* sp. and *Mallotus* sp. are mostly used to feed the goats but the information on their potential or benefits as goats feed is limited. Thus, this study was done to assess the potential of these local plants to be included in total mix ration for goat diet in which the nutrient composition and *in vitro* ruminal degradability were determined.

Materials and Methods

Macaranga sp. and *Mallotus* sp. leaves were harvested from bush area surrounding the farm. Meanwhile, Napier grass was harvested manually from pasture at the farm at the age of 8 weeks. Napier grass was used as a control group as it is common and widely been used in goats diet. All samples were dried at 60°C for 48 hours and ground using a laboratory blender (Waring®) to pass through 2mm sieve screens and kept in labelled air-tight plastic containers at room temperature for further laboratory analysis. The nutrient composition (dry matter; crude protein; crude fibre; crude fat) for all samples was determined by proximate analysis. The proximate analysis of the samples was done in 4 replicates using procedures of AOAC (1990). *In vitro* ruminal degradability was performed according to the method described by Hassim et. al. (2012). A total amount of 0.25g of each sample was added into 100 ml plastic syringes. A mixture of rumen fluid (10 ml) and phosphate-bicarbonate buffer (20 ml) was further added into the syringes. The syringes were incubated at 39°C for 24 hours. The volume of the gas produced at 0, 2, 4, 6, 8, 10, 12 and 24 hours of incubation period were recorded. After 24 hours, the incubation content was collected and analyzed for volatile fatty acids (acetate; propionate; butyrate) production using gas-liquid chromatography. All data were statistically analyzed by ANOVA and means were tested with Dunnett Test using SPSS version 20.

Results and Discussion

The nutrient composition of the *Macaranga* sp., *Mallotus* sp. and Napier grass (*Pennisetum purpureum*) is presented in Table 1. The result revealed that there were significant difference ($P < 0.05$) for all nutrient composition in each local plant species against Napier grass. Dry matter content was highest in *Macaranga* sp. (50.92%) followed by *Mallotus* sp. (45.41%) and Napier grass (13.04%). The crude protein content was highest in *Macaranga* sp. (7.18 %) followed by *Mallotus* sp. (6.78%) and Napier grass (3.88%) whereas *Mallotus* sp. contain the highest crude fat content (5.22%) followed by *Macaranga* sp. (4.85%) and Napier grass (2.84%). As for crude fibre, Napier grass showed the highest content (25.38%) followed by *Mallotus* sp. (15.47%) and

Macaranga sp. (12.00%). High dry matter content in *Macaranga* sp. and *Mallotus* sp. could be due to sample type (only leaves used) to mimic the feeding regime as the goats only offered with leaf parts. Meanwhile, whole part of Napier grass that used in this study contain lower dry matter as compared to the two local plants. This could be explained by a high water content of stem fraction that resulted in an increased of moisture content (Mohammed et al., 2015). Low crude protein content of the Napier grass in this study could be due to several factors including soil, species variety, maturity, pasture management and climate (Halim et al., 2013). Sampling of Napier grass was done during hot season and the pasture was poorly managed by the farmer with improper irrigation and inadequate fertilizer. In addition, crude protein and crude fat content were higher in both local plants and this could be associated with leaf: stem ratio which contain high nutritive value (Tudsri et al., 2002). The total gas and volatile fatty acids production of *in vitro* ruminal degradability is presented in Table 2. The highest total gas production was shown by Napier grass (31.00 ml), followed by *Macaranga* sp. (28.67 ml) and *Mallotus* sp. (23.33 ml). Acetate production was highest in *Mallotus* sp. (733.49 mM/ml) followed by Napier grass (605.61 mM/ml) and *Macaranga* sp. (599.85 mM/ml) whereas propionate production was highest in *Mallotus* sp. (28.61 mM/ml) followed by Napier grass (24.45 mM/ml) and *Macaranga* sp. (24.23 mM/ml). As for butyrate production, Napier grass showed the highest value (11.26 mM/ml), followed by *Macaranga* sp. (11.19 mM/ml) and *Mallotus* sp. (10.36 mM/ml). However, there was no significant difference shown for total gas and volatile fatty acids production. Thus, as there were no significant difference observed between Napier grass with these local plants, it can be concluded that these local plants have similar degradability with Napier grass.

Table 1. Nutritional composition of Napier grass (*Pennisetum purpureum*) and local plants (*Mallotus* sp. and *Macaranga* sp.).

	Napier grass	<i>Mallotus</i> sp.	<i>Macaranga</i> sp.
DM (%)	13.04±0.04 ^a	45.41±0.09 ^b	50.92±0.67 ^c
CP (% DM)	3.88±0.38 ^a	6.78±0.25 ^b	7.18±0.62 ^b
CFa (% DM)	2.84±0.03 ^a	5.22±0.09 ^b	4.85±0.01 ^c
CFi (% DM)	25.38±0.20 ^a	15.47±0.20 ^b	12.00±0.14 ^c

DM=Dry matter, CP=Crude protein, CFa=Crude fat, CFi=Crude fibre. Value expressed as mean±standard error of mean (SEM).

Table 2. Total gas and volatile fatty acids production of Napier grass (*Pennisetum purpureum*) and local plants (*Mallotus* sp. and *Macaranga* sp.).

	Napier grass	<i>Mallotus</i> sp.	<i>Macaranga</i> sp.
TG (ml)	31.00±2.65 ^a	23.33±3.29 ^a	28.67±4.56 ^a
TVFA (mM/ml)	641.31±78.80 ^a	772.46±122.07 ^a	635.27±71.00 ^a
Acetate (mM/ml)	605.61±75.77 ^a	733.49±118.86 ^a	599.85±68.26 ^a
Propionate (mM/ml)	24.45±2.61 ^a	28.61±3.80 ^a	24.23±2.05 ^a
Butyrate (mM/ml)	11.26±0.76 ^a	10.36±1.98 ^a	11.19±1.33 ^a

TG=Total gas, TVFA=Total volatile fatty acid. Value expressed as mean±standard error of mean (SEM).

Conclusion

Based on the results of nutrient composition and *in vitro* ruminal degradability, it shows that these local plants has a potential to be used as a good feed source for goats. In addition, a proper feeding regime using these local plants need to be considered in order to ensure a balance diet for the goats.

References

- AOAC, 1990. Official Methods of Analysis. 15th edition. Association of Official Analytical Chemists, Arlington, Virginia, USA.
- Ansah, T., E. L. K. Osafo and H. H. Hansen, 2010. Herbage yield and chemical composition of four varieties of Napier (*Pennisetum purpureum*) grass harvested at three different days after planting. *Agriculture and Biology Journal of North America*, 1(5): 923-929.
- Food and Fertilizer Technology Center, 2002. Feeding mixtures of tree/shrub leaves for ruminant animals. *Livestock PT2002-20*: 1-2.
- Halim, R. A., S. Shampazuraini and A. B. Idris, 2013. Yield and nutritive quality of nine Napier grass varieties in Malaysia. *Malaysian J. Anim. Sci.* 16(2): 37-44.
- Hassim, H. A., M. Lourenço, Y. M. Goh, J. J. Baars and V. Fievez, 2012. Rumen degradation of oil palm fronds is improved through pre-digestion with white rot fungi but not through supplementation with yeast or enzymes. *Canadian J. Anim. Sci.* 92(1): 79-87.
- Mohammed, I. Y., Y. A. Abakr, F. K. Kazi, S. Yusup, I. Alshareef and S. A. Chin, 2015. Comprehensive Characterization of Napier Grass as a Feedstock for Thermochemical Conversion. *Energies*, 8(5), 3403-3417.
- Tudsri, S., S. T. Jorgensen, P. Riddach and A. Pookpakdi, 2002. Effect of cutting height and dry season closing date on yield and quality of five napier grass cultivars in Thailand. *Tropical Grasslands*, 36(4), 248-252.

Effect of crude glycerin supplementation on performance of dairy heifers

Abdurrahman^{1,2}, A.M., L. Boon-ek¹, S. Buaphan¹, N. Buathong¹ & N. Uncheun¹

¹Animal science Department, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, 73140, Thailand, ²Indonesian Agencies for Agricultural Research and Development, Ministry of Agriculture 12540 Jakarta, Indonesia.

Abstract

Crude glycerin is a by-product of biodiesel production and has recently become more available as a livestock feed with the growth of the biofuel industry. The objective of this study was to investigate the effect of palm based crude glycerin as an alternative for energy feedstuff in dairy heifers. Twenty crossbred Holstein (87.5-93.75%) heifers, 211.4 ± 59.3 kg initial body weight were used in randomized complete block design, composed of 4 treatments for 60 days period. The heifers were fed concentrate diet with increasing level of crude glycerin (0, 4, 8 and 12%). Napier grass was used as a roughage source, containing 60% of total DM of the diet. Increasing crude glycerin levels had significant differences in ADG (498.2, 574.0, 621.3, 579.5 g/d, respectively). However, There were no statistically significant differences on DMI, F:G and C:G among treatments. Therefore, inclusion of crude glycerin at low level can be beneficial to dairy heifer performance.

Keywords: glycerin, heifers, performance

Introduction

Crude glycerin is a by-product of biodiesel production and has recently become more available as a livestock feed with the growth of the biofuel industry. Crude Glycerin is also called as sugar alcohol has GE content 4.3 Mcal/kg. Crude glycerin, used less than 10% in DM of the diet, also has retained energy better than dry roll corn (Hales et al., 2015). Farias et al. (2012) reported that crude glycerin inclusion up to 9.1% in DM of total diet have no change on food digestibility and heifers' behavior. Other study by Parson et al. (2009) showed that feeding less than 8% crude glycerin in steam-flaked corn diets can enhance gain and efficiency of finishing heifers. Glycerin inclusions reduce fiber digestion and bacterial population but not significantly reduce at concentration below 7% in the diet (Abo El-Nor et al., 2010). The objective of this study was to investigate the effect of palm based crude glycerin as an alternative energy feedstuff in dairy heifers.

Materials and Methods

Twenty crossbreeds Holstein heifers (87.5-93.75%) with average initial body weight 211.37 ± 59.26 kg were used in randomized complete block design for 60 days. Deworming was done before heifers got into the experimental pen. Heifers were blocked by initial body weight and randomly assigned to 4 treatments with 5 blocks as replications. Dietary treatments were used 0, 4, 8 and 12% palm based crude glycerin on the concentrate (Table 1). Fresh Napier grass was used as a roughage source, containing 60% of total DM of diets. Heifers fed twice daily at the morning (8.00 h) and afternoon (14.00 h) by *ad libitum* feeding. Feed samples were analyzed for DM, CP, ash, EE by using AOAC method (2012), and NDF, ADF by using Goering & Van Soest method (1970). Data were analyzed by using general linear model (GLM) procedure (SAS, 1995). Significant difference was declared at $P < 0.05$.

Table 1. Ingredients and chemical composition of heifer's diets.

Item	Napier Grass	Crude glycerin, %			
		0	4	8	12
Ingredients, %					
Palm based Crude glycerin		0.0	4.0	8.0	12.0
Cassava chip		36.0	32.0	28.0	24.0
Wheat bran		20.0	20.0	20.0	20.0
Palm seed meal		12.0	12.0	12.0	12.0
Soybean hull		11.7	11.5	11.3	11.1
Soybean meal-44%		4.8	5.0	5.2	5.4
Mungbean meal		14.0	14.0	14.0	14.0
Urea, 46% N		0.5	0.5	0.5	0.5
Di calcium phosphate		0.5	0.5	0.5	0.5
Mineral		0.5	0.5	0.5	0.5
Price (Baht kg ⁻¹)	0.5	8.9	9.0	9.1	9.2
Analysis content, % DM					
DM (%)	18.6	92.0	90.9	89.0	87.0
CP (%)	5.8	12.2	12.3	12.6	12.9
Ash (%)	11.6	14.0	13.9	14.4	14.6
EE (%)	1.1	2.6	2.7	2.8	2.9
NDF (%)	68.2	30.7	30.9	31.5	32.0
ADF (%)	47.3	23.9	24.0	24.3	24.3
GE (kcal kg ⁻¹)	4218.0	3838.1	3886.6	3927.0	3930.5

Roughage to concentrate ratio 60:40.

Result and Discussion

Inclusions of crude glycerin 0, 4, 8 and 12% on concentrate, with ratio of DM roughage to concentrate 60:40, had crude glycerin contents on diet 0, 1.6, 3.2 and 4.9%, respectively. These indicated that the inclusions of Crude glycerin on diet were at low level concentration that would not significantly affect of fiber digestion and bacterial population in the rumen (Abo El-Nor et al.,

2010). Increasing level of crude glycerin, as energy source at 4-12% in concentrate can reduced cassava chip of 11-33% (Table 1).

Overall means of feed intakes in terms of DMI, CPI, EEI, NDFI and ADFI were not significantly different for all dietary treatment (Table 2). However, crude glycerin inclusions at 4, 8 and 12% on concentrate had significantly increased ADG at 15.2, 24.7 and 16.3%, respectively, compared to control. Heifers fed concentrate 8% of crude glycerin has the highest ADG than others. There were no statistically significant differences on feed per gain (F:G) and cost per gain (C:G) with the control, but the numbers showed, inclusions of crude glycerin had reduced feed per gain and cost per gain. Crude glycerin inclusions at 4, 8, and 12% on concentrate reduce F:G and C:G at 19.2, 15.8, 16.9% and 17.2, 12.0, 11.2%, respectively. It means that inclusions of crude glycerin increased feed efficiency and reduced cost per gain of rearing dairy heifers ($P > 0.05$).

Facuri et al. (2014), also found that the inclusions of crude glycerin up to 15.56% had no effect on feed intake of heifers and increased the use of DM and NDF from pasture and concentrate through the better feed and rumination efficiencies. However, Parson et al. (2009) reported that crude glycerin inclusions less than 8% in the diets on finishing heifers had more ADG and feed efficiency than control, with the best ADG at 2%. Hales et al. (2015) reported that inclusions of glycerin 0-10% in total diet have an effect on increasing of metabolizable energy (ME) and Digestible energy (DE). However, the maximum retained energy was at 5% inclusions. Increasing ME and DE indicated enhancing of net energy (NE) that would effect on the better performance of animals.

Table 2. Effects of increasing of crude glycerin in concentrate diet on performance of dairy heifers.

Item	Crude glycerin in concentrate, %				SEM	P-Value
	0	4	8	12		
Initial bodyweight (kg)	211.2	205.2	229.6	207.2	5.24	0.375
Final bodyweight (kg)	238.6	237.8	267.0	241.6	5.65	0.260
DMI(kg day ⁻¹)	6.3	6.2	6.8	6.1	0.15	0.394
CPI (g day ⁻¹)	527.7	518.9	582.8	532.3	12.45	0.308
EEI (g day ⁻¹)	106.5	106.2	120.3	111.4	2.57	0.232
NDFI (g day ⁻¹)	3354.2	3288.3	3652.0	3318.4	81.31	0.399
ADFI (g day ⁻¹)	2389.1	2340.0	2597.9	2352.0	57.68	0.389
GEI (Mcal day ⁻¹)	26.0	25.4	27.9	25.1	0.60	0.394
ADG (g)	498.2 ^b	574.0 ^a	621.3 ^a	579.5 ^a	11.66	0.020
F:G (kg kg ⁻¹)	13.2	10.8	11.1	11.0	0.38	0.128
Cost (baht)	35.0	34.9	39.3	36.2	0.82	0.248
C:G (baht kg ⁻¹)	73.2	60.6	64.5	65.00	2.20	0.277

^{ab} means within a row different superscript differ ($P < 0.05$).

Conclusions

Crude glycerin can be used as alternative energy source that will be beneficial on rearing dairy heifer. The inclusions of crude glycerin up to 12% in concentrate could increase ADG and feed efficiency, with the best ADG observed at 8% in concentrate or 3.2% in DM of diet.

Acknowledgements

The authors acknowledge The Agricultural Research and Development Agency (Public organization) Grant No. PPR5705010010 for financing this research.

References

- Abo El-Nor, S., A. A. AbuGhazaleh, R. B. Potu, D. Hastings, M. S. A. Khattab, 2010. Effect of differing levels of glycerol on rumen fermentation and bacteria. *Anim. Feed Sci. Technol.* 162: 99-105
- Facuri, L. M. A. M., R. R. Silva, F. F. da Silva, G. G. P. de Carvalho, C. B. Sampaio, F. B. L. Mendes, M. M. Lisboa, D. S. Barroso, V. M. Carvalho, and M. M. S. Pereira, 2014. Ingestive behavior of heifers supplemented with glycerin in substitution of corn on *Brachiaria brizantha* pasture. *Asian Australas. J. Anim. Sci.* 27: 1584-1592
- Farias, M. S., R. R. Silva, F. Zawadzki, C. E. Eiras, B. S. Lima, and I. N. Prado, 2012. Glycerin levels for crossbred heifers supplemented in pasture: Intake behavior. *Acta Sci. Anim. Sci.* 34:63-69.
- Hales, K. E., A. P. Foote, T. M. Brown-Brandl, and H. C. Freetly, 2015. Effects of dietary glycerin inclusion at 0, 5, 10, and 15 percent of dry matter on energy metabolism and nutrient balance in finishing beef steers. *J. Anim. Sci.* 93:348–356
- Parson, G. L., M. K. Shelor, and J. S. Drouillard, 2009. Performance and carcass traits of finishing heifers fed crude glycerin. *J. Anim. Sci.* 87: 653-657

Utilization of fresh cassava with Ruzi grass fermented by microbes from Pangkhaomark as diets in swine

Pongjongmit^{*1}, T., N. Piamphon², P. Pansri¹, R. Worachinda¹,
N. Nasinprom¹ & J. Insee¹

¹Rajamagala University of Technology Isan Kalasin Campus, Kalasin 46000, Thailand

²Rajamagala University of Technology Tawan-ok, Chonburi 20110, Thailand

Abstract

Utilization of fresh cassava with ruzi grass fermented by microbes from pangkhaomark as diets in swine. The objective of this experiment was conducted on two time interval consisting of growing and finishing pigs. The objective of this experiment was to investigate the supplementation effect of fresh cassava and ruzi grass fermented by pangkhaomark on performance, production cost, digestion coefficient, carcass quality and blood urea nitrogen level of growing and finishing pigs. There were three diets in this experiments diet 1 normal pig diets diet 2 and 3 were prepared from fresh cassava and ruzi grass fermented by pangkhaomark mixing with two different herbs)fermented diet 1, fermented 2, respectively(. Eighteen cross - bred pigs were)Large white X Landrace X peatrian(raised from 24 ± 1.8 kg to 100 ± 3.8 kg of body weight were assigned in Completely Randomized Design consisting of three treatments. group 1 , group 2 and group 3 pigs were fed on normal diet, diet 1 and diet 3, respectively. The results showed that growth rate at growing stage for group 1 , group 2 and group 3 were 680, 685 and 678 and while the data of finishing stage were 731, 735 and 726, respectively. There was no significant difference on FCR and feed intakes of all groups from 25 – 100 kg of body weight. Feed intakes of pigs were 1.99, 2.07 and 2.04 kg/day, respectively. Feeding costs per pig were 3,347, 3,204 and 3,153, respectively ($P < 0.05$).

Keywords: Cassava, Microbes from Pangkhaomark , Pig

**Corresponding author: tpongjongmit@hotmail.com*

Introduction

The fact that swine feed is produced with high cost, mainly due to the elevated cost of maize, makes pig production very expensive, constituting 75% of the total cost of production. Effort to reduce the cost of production is being directed towards the use of affordable and available alternative sources of energy in the diets of pig (NNADI et al., 2010). Several studies on the utilization of cassava for animal feeds including pig feeds have been conducted for more than two decades It is evident that cassava products are good energy feed ingredients for pig. The starch in cassava is highly digestible when compare to that of maize due to the high content of amylopectin. Cassava can be used as a sole source energy feed ingredient in pig. There have been several studies of the utilization of cassava in animal ration in Thailand and the results of the studies indicate cassava is a great potential sources of energy to substitute the conventional energy feed ingredient (Uthai and Sukanya, 2005). The need to use cassava to replace swine diet is to reduce feed cost as it would constitute about 45 – 60% of energy demand in pig diet. Due to limitation of using fresh cassava as swine diet leading to study on the efficiency of fermented cassava as swine

supplement feed. Therefore, this study was designed to utilize fresh cassava with ruzi grass fermented by microbes from pang-khaomark as diets in swine.

Materials and Methods

A total of Eighteen cross - bred pigs were)Large white X Landrace X peatrian(raised from 24±1.8 kg to 100±3.8 kg of body weight were assigned in Completely Randomized Design consisting of three treatments. There were three diets in this experiments diet 1 normal pig diets diet 2 and 3 were prepared from fresh cassava and ruzi grass fermented by pangkhomark mixing with two different herbs)fermented diet 1, fermented 2, respectively(.

Results and Discussion

The study indicated that of pang khao mark-fermented cassava in growing - finishing pigs between the There were three diets in this experiments diet 1 normal pig diets diet 2 and 3 were prepared from fresh cassava and ruzi grass fermented by pangkhomark mixing with two different herbs)Table1(

Table1 The effect of Fresh Cassava with Ruzi Grass Fermented by Microbes from Pangkhaomark on performance in Swine

Item	Treatment			SEM	p-value
	1	2	3		
Number of swine	6	6	6	-	-
Initial)kg(24.2	23.7	24.5	2.42	0.65
Final) kg(99.56	99.58	100.82	3.3	0.13
Weight gain) kg(75.36	75.88	76.32	0.73	0.29
Average daily gain	703	708	712	6.4	0.30
Feed intake) kg/d(1.99 ^b	2.07 ^a	2.04 ^a	0.26	0.03
Feed conversion ratio	2.83	2.92	2.87	0.53	0.17

Different superscripts in a row indicate significant difference between the treatment means at the level of probability = $p < 0.05$

The effect of pang khao mark-fermented cassava in growing - finishing pigs was studied. Thailand is one of the world's largest exporters of cassava flour and not less than 1 million tons of cassava containing approximately by AOAC)1994(. 66.0% starch, 4.5% fiber, 3% protein, 2.7% ash, and 2.1% fat. According to the high level of starch content used as feed in animal diet (Sriroth et al., 2000). Using *Saccharomyces cerevisiae* from 4.4 to 10.9%. Boonnop et al. (2006) reported the potential of protein increasing from solid-liquid media fermentation by *S. cerevisiae* at 9.5 and 6.5 fold for cassava chip and fresh cassava root, respectively. Thongkratok et al. (2010) found that crude protein of cassava pulp could be significantly enhanced after fermentation with *A. oryzae*, *S. cerevisiae* and *Candida utilis* whereas *A. oryzae* provide highest protein and amino nitrogen up to 17.4 and 15.14%, respectively. With the addition of *S. cerevisiae* to cassava pulp, protein content was improved up to 26% in Oboh)2006(. Using of fresh cassava with ruzi grass fermented by microbes from pangkhaomark as diets in swine, therefore, had a great potential for making profit of pig producers.

Conclusion

The results showed that the swine. There were three diets in this experiments diet 1 normal pig diets diet 2 and 3. Feed intake of pigs were 1.99, 2.07 and 2.04 kg/day. Feeding costs per pig were 3,347, 3,204 and 3,153, respectively ($p < 0.05$). But there was no significant difference on FCR and feed intakes of all groups. However, replacing pang khao mark-fermented cassava resulted in a substantial reduced production costs as it reduced feed cost per 1 kilogram higher than control treatment.

References

- AOAC. 1994. Official method of analysis. 16th edn. Association of Official Analytical Chemists, Washington, DC.
- Boonnop, K., M. Wanapat, N. Nontaso and S. Wanapat. 2009. Enriching nutritive value of cassava root by yeast fermentation. *Sci. Agric. (Piracicaba, Braz.)*. 66: 629-633.
- Kaewwongsa, W., S. Triyagun, C. Yuangklang, C. Wachraprakarn, and P. Paengkoum. 2011. Protein enrichment of cassava pulp by *Saccharomyces cerevisiae*. *J. Anim. Vet. Adv.* 10: 2434-2440.
- NNADI, Pius Ajanwachukwu, OMEKE, Benjamin Chigozie and OKPE, Godwin Chidozie. 2010. Growth and reproductive performances of weaner pigs fed maize replaced cassava diet. *Animal Research International* (2010) 7(3): 1257 – 1263.
- Oboh, G., 2006. Nutrient enrichment of cassava peels using a mixed culture of *Saccharomyces cerevisiae* and *Lactobacillus sp* solid media fermentation techniques. *Electrical J. Biol.*, 9: 46-49.
- Sriroth, K., R. Chollakup, S. Chotineeranat, K. Piyachomkwan, and C. G. Oates. 2000. Processing of cassava waste for improved biomass utilization. *Bioresource Technology*. 71: 63-69.
- Uthai Kanto and Sukanya Juttupornpong. 2005. Animal Nutrition Research and Development Center, Kasetsart University,

Evaluation of urea-yeast fermented fresh cassava root as protein source by using *in vitro* gas production technique

Nitipot^{*1}, P. & C.Promkot²

¹Rajamangala University of Technology Isan Kalasin Campus, Kalasin 46000, Thailand

²Rajamangala University of Technology Isan Sakonnakorn Campus, Sakonnakorn 47160, Thailand

Abstract

The objective of this study was to evaluate pattern of fresh cassava root fermentation to develop as protein source on nutritive values. Investigated 4 methods of ground fresh cassava root fermentation; thus, solely fermented fresh cassava root (FFCR) (T1), FFCR with urea (T2), FFCR with urea - yeast in closed system (T3), and FFCR with urea and yeast in opened system (T4). The experiment was arranged following Completely Randomized Design (CRD). It found that methods of cassava root fermenting were not affected on ash ($P > 0.05$). On the other hand, CP content was increased ($P < 0.01$) when urea was mixed in fermentations. The NDF of T3 and T4 was decreased lower than of T2 ($P < 0.01$). The *in vitro* gas production technique parameters were analyzed as following. Ammonia nitrogen ($\text{NH}_3\text{-N}$) in fermented fluid of T1 was lower ($P < 0.05$) than of T2, T3 and T4 at 3, 6, and 9 hour of incubation time. At 12 and 24 hour of incubation time, the $\text{NH}_3\text{-N}$ of T3 and T4 was higher than of T1 and T2 ($P < 0.05$). The kinetics of gas production, it found that, rate of gas production (c) were not differ among treatments ($P > 0.05$). Gas volume at 0 hour of incubation time (a) of T2 was lower than of T3 and T4 ($P < 0.05$). Potential of gas production (d) and gas volume (b) of T1 was higher than the others ($P < 0.01$). It was conclude that incorporated urea with yeast in the fermentation could elevated CP level and decreased the NDF content in FFCR, furthermore, it could maintain $\text{NH}_3\text{-N}$ level in the *in vitro* ruminal fermentation.

Keywords: cassava root, in vitro gas production, urea-yeast fermented

**Corresponding author: nitipot@yahoo.com*

Introduction

Yeast supplementation for ruminants by the use of dry cassava chips and fresh cassava root fermented with pure culture of *Saccharomyces cerevisiae* had been studied and could increase its protein content from 3 % in non-fermented cassava root to 21-30 % in fermented products (Boonnop et al., 2009). However, lack of data is available for method of fermenting of fresh cassava root with pure culture of *S. cerevisiae*. Also, it needs screening test prior to using in animals where costly investigation. On the other hand, the *in vitro* gas production technique was proposed for determining fermentation kinetics of ruminant feed. The technique is a low cost, highly reproducible and easy method of obtaining a dynamic description of the nutritive value of feedstuffs, while at the same time allowing for more samples to be analyzed. Thus, the objective of this study was to evaluate

method of fresh cassava root fermentation to develop as protein source on nutritive values, NH₃-N concentration and kinetics of gas production by using *in vitro* gas production technique.

Materials and Methods

Experimental feeds and management: Fresh cassava root was ground for investigation of 4 fermenting methods; thus, solely 21 day fermented fresh cassava root in plastic bags (FFCR) (T1), FFCR with urea for 5 day additional fermentation (T2), FFCR with urea - yeast in closed system for 5 day additional fermentation (T3), and FFCR with urea and yeast in opened system without additional fermentation (T4). The last three treatments were mixed of 80g urea/kg fresh cassava root. All test feedstuff were finished by air-drying. The experiment was arranged following Completely Randomized Design (CRD) with 5 replications. Five kg each of dried samples were ground through a 1 mm screen for chemical analysis. The feedstuff samples were analyzed to determine chemical composition following standard method of AOAC (1990) and Van Soest et al. (1991).

The *in vitro* gas production technique: The experiment was arranged following Randomized Completely Block Design (RCBD) with 4 replications for kinetic of gas production and 2 replications for ammonia nitrogen (NH₃-N) analysis. The ground samples were conducted following *in vitro* gas production technique procedure (Sommart et al., 2000) where two mature Thai native cattle were used as the source of rumen inoculum. Readings of gas production were recorded from 1 to 72 h. Cumulative gas volumes were fitted using the equation $y = a + b(1 - e^{-ct})$ (Ørskov and McDonald, 1979), where y = gas production at time 't', a = the intercept, b = the fermentation of the insoluble fraction, c = rate of gas production, and $d = (|a| + b)$ = potential extent of gas production. *In vitro* ruminal fluid of sampled incubation bottles were centrifuged at 5,000 rpm for 5 minutes for ruminal fluid supernatant collection. The collected supernatants were analyzed for NH₃-Nas following AOAC (1990).

Statistical analysis: All data obtained from the trials were subjected to the analysis of variance procedure performed using the GLM procedure (SAS, 1996) (according to a CRD a RCBD). Differences for treatment mean and group-treatment mean were statistically compared using Duncan's New Multiple Range Test and Orthogonal Contrast procedure (SAS, 1996).

Result and Discussion

Chemical composition of FFCR in four methods as showed in table, it found that mixed urea in FFCR was significantly ($P < 0.01$) elevated CP level in T2, T3 and T4. The NDF content of T3 and T4, compared to T2, were decreased ($P = 0.01$) when incorporated urea with yeast in the fermentation.

The NH₃-N concentration of the *in vitro* ruminal fluid in various durations of incubation, data indicated that FFCR was significantly ($P < 0.01$) increased NH₃-N level in T2, T3 and T4 at 3, 6, and 9 h of incubation. Especially, T3 could maintain NH₃-N level longer than the others. NRC (2001) stated that the optimum NH₃-N level for microbial in the rumen is ranged from 18-100 mg%, where incorporated urea with yeast in this study (T2, T3 and T4) ranged from 15.00 – 66.20 mg%. Gas production value and kinetics of gas production, data indicated that FFCR was not effected ($P > 0.05$) on rate of gas production (c). However, T1 showed the highest ($P < 0.01$) potential of gas production (d) and gas production value (b) than the others, showed that solely urea

(T2) or urea incorporated with yeast in the fermentation (T3 and T4) had negative effected on these parameters. While in the same time, there was not differed among T3 and T4. The intercept (*a*), it found that T2 had the lowest ($P < 0.05$) value than the others, where reflected the highest soluble gas in ruminal fluid (where the negative value was not considered). It was comparable to report of Nitipot and Sommart (2003) whom studied the kinetic of gas production of cassava chip and cassava pulp.

Based on this experimental data, it was conclude that incorporated urea with yeast in the fermentation could elevated CP level and decreased the NDF content in FFCR, furthermore, it could maintain $\text{NH}_3\text{-N}$ level in the *in vitro* ruminal fermentation.

Table 1. Chemical composition of fermented fresh cassava root, $\text{NH}_3\text{-N}$ and kinetics of gas production obtained from the *in vitro* gas production technique.

Items	Treatment				P-Value			SEM
	T1	T2	T3	T4	T1 vs T2, 3, 4	T2 vs T3, 4	T3 vs T4	
Chemical composition, %								
DM	89.35	89.48	86.76	86.64	-	-	-	-
-----% of DM -----								
Ash	4.33	4.63	4.21	3.83	0.79	0.16	0.45	0.35
CP	1.29 ^b	20.04 ^a	20.03 ^a	19.21 ^a	<0.01	0.62	0.34	0.55
NDF	13.63 ^b	18.06 ^a	5.49 ^c	6.77 ^c	0.01	<0.01	0.37	0.96
ADF	4.61 ^{ab}	5.10 ^a	4.02 ^b	4.05 ^b	0.46	<0.01	0.93	0.55
Ammonia nitrogen concentration ($\text{NH}_3\text{-N}$, mg%)								
3 hr	14.85 ^b	40.35 ^{ab}	34.55 ^{ab}	44.30 ^a	0.03	0.91	0.35	4.09
6 hr	19.00 ^b	62.35 ^a	42.20 ^{ab}	46.95 ^{ab}	0.03	0.17	0.72	5.48
9 hr	9.60 ^b	34.40 ^{ab}	46.80 ^{ab}	69.15 ^a	0.04	0.18	0.26	7.58
12 hr	9.00 ^c	26.35 ^b	66.20 ^a	62.75 ^a	<.01	<.01	0.10	0.73
24 hr	4.55 ^b	15.00 ^b	60.10 ^a	37.90 ^{ab}	0.05	0.06	0.21	6.59
Kinetics of gas production								
<i>c</i> , %/hr	0.032	0.034	0.030	0.031	0.826	0.107	0.668	0.00
<i>d</i> , ml	232.50	225.68 ^a	189.78 ^b	194.53 ^b	0.003	0.002	0.602	5.56
<i>b</i> , ml	219.53 ^a	209.35 ^a	180.05 ^b	183.10 ^b	0.002	0.003	0.715	5.11
<i>a</i> , ml	-12.98 ^b	-16.33 ^c	-9.73 ^a	-11.43 ^{ab}	0.596	<0.001	0.149	0.68

DM=Dry matter, CP = Crude protein, NDF = Neutral detergent fiber, ADF = Acid detergent fiber, **d* = Potential of gas production where $d = b + |a|$ (ml/0.5 g substrate DM), *b* = Volume of gas production (ml/0.5 g substrate DM), *c*= Rate of gas production (%/hr), *a*= Gas volume at time 0 (y axis intercept, ml/ 0.5 g substrate DM)

References

- AOAC. 1990. Official Methods of Analysis. 15th edn. Association of Official Analytical Chemists, Virginia. pp684
- Boonnop, K., M. Wanapat, N. Nontaso and S. Wanapat. 2009. Enriching nutritive value of cassava root by yeast fermentation. *Sci. Agric.* 66:616–620
- Nitipot, P. and K. Sommart. 2003. Evaluation of ruminant nutritive value of cassava starch industry by products, energy feed sources and roughages using *in vitro* gas production technique. In: Proceeding of Annual Agricultural Seminar for year 2003, 27-28 January, KhonKaen University; 2003. 179-190
- NRC. 2001. Nutrient Requirements of Dairy Cattle. 7th ed. Natl. Res. Council., Natl. Acad. Press, Washington, D.C. pp408

- Ørskov, E.R. and I. McDonald. 1979. The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. *J. Agric. Sci. (Camb)* 92: 499-504
- SAS. 1996. *SAS User's Guide: Statistics*, Version 6.12th ed. SAS Institute Inc. Cary, NC.
- Sommat, K., D.S. Parker, P. Rowlinson and M. Wanapat. 2000. Fermentation characteristics and microbial protein synthesis in an *in vitro* system using cassava, rice straw and dried ruzi grass as substrates. *Asian-Australasian J. Anim. Sci.* 13(8):1084-1093
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and Non-starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74:3583-3597

The effects of soybean meal treated with green tea marc crude extract on oxidative status and milk production of dairy cows

Buaphan*, S., L. Boonek, N. Unchuen, N. Buathong, T. Hongsapak & P. Usungnern

Animal Science Department, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Nakhon Pathom 73140, Thailand

Abstract

The objective of this study was conducted to evaluate the effects of soybean meal treated with green tea marc (GTM) crude extract in diet of transition dairy cows. Ten crossbred Holstein (>93.8%) cows in late gestation were paired according to expected calving date and randomly assigned to total mixed rations (TMR) containing either untreated SBM (control) or SBM treated with 5% GTM from 1 week prepartum to 3 weeks postpartum. The TMR contained dried para grass as roughage source. There was no statistically significant difference between treatments on dry matter intake, body weight, blood thiobarbituric acid reactive substance (TBARS), non-esterified fatty acids (NEFA) and β -hydroxybutyrate (BHB) concentration. However, milk protein yield at 2 and 3 week after parturition were higher for cows fed GTM diet than control group ($P < 0.05$).

Keywords: green tea marc, oxidative status, transition dairy cows

**Corresponding author:* swksrbu@ku.ac.th

Introduction

During the transition period, cows are more susceptible to oxidative stress, which may contribute to an impaired immune function and enhanced susceptibility to periparturient diseases. Oxidative status of dairy cows was related to energy status. Cows with higher BHB and NEFA showed higher TBARS (Bernabucci et al., 2005). Moreover, cows require more high quality protein than that provided by rumen microorganisms. Soybean meal (SBM) is the most common protein supplement in the dairy feeds. However, it can be extensively degraded (≥ 57.4 to 69.6%) by ruminal microbes (NRC 2001). There are some methods for decreasing protein degradation in rumen such as using tannins. Tannins have high affinity for proteins and protect them from ruminal microbial degradation. Intake of condensed tannin at low to moderate level (2-4 % of DM) in the feed have a beneficial effect on protein metabolism (Aerts et al., 1999). Soltan (2009) pelleting SBM with 1.25% tannin plant extract reported increased milk fat percentage, fat and protein yield. Tea leaf (*Camellia sinensis*) content of tannin (condensed) and catechins (Cai et al., 2001). The catechins present in green tea in greater quantities (26.7%), which exhibit powerful antioxidant activities. Therefore, we designed this study to investigate the effects of treated SBM with GTM crude extract that contained tannin-catechins on oxidative status, blood composition and milk production of transition dairy cows.

Materials and Methods

Ten crossbred Holstein (>93.8%), average 464 kg of body weight (BW) in late gestation were paired according to expected calving date and randomly assigned to diets containing either untreated SBM (control) or treated with 5% GTM. The GTM crude extract contained 13.1% of tannin-catechins. The experiment was conducted from 1 week prepartum to 3 weeks postpartum. Before starting experimental all cows were fed the same diet for a week. Diets were fed as a TMR, in which dried para grass and concentrate (previously mixed) were weighed and mixed before feeding. Diet samples were collected and analyzed for dry matter (DM), crude protein (CP), ether extract (EE), acid detergent fiber (ADF) and neutral detergent fiber (NDF) (AOAC, 1990; Van Soest et al., 1991). On the last day of week 1, 2, and 3 postpartum, blood samples were collected before the morning feeding and analyzed for TBARS (Asakawa & Matsushita, 1980), NEFA (Johnson & Peters, 1993) and BHB (Gibbard & Watkins, 1968). Milk yield and milk samples were collected on the last day of week 1, 2, 3, postpartum and analyzed for fat, protein, lactose and somatic cell counts (AOAC, 1997). Data for all measured variables were analyzed according to the paired comparison. Treatment means were compared using *t*-test SAS, 1996(. Significance was declared at $P < 0.05$.

Table 2 .Ingredients and nutrient composition of the TMR fed to transition dairy cows.

Item	Control	GTM
Ingredient, % of DM		
Para grass	20.0	20.0
Cassava chips	30.0	30.0
Palm seed meal	16.0	16.0
Wheat bran	13.4	13.4
Soybean hull	6.5	6.5
Untreated SBM	12.0	-
Treated SBM ^A	-	12.0
Urea, 46%N	1.0	1.0
Vitamins and minerals	1.1	1.1
Analysis content, % of DM		
Crude protein	15.6	15.0
Ether extract	1.9	1.9
Neutral detergent fiber	36.7	37.1
Acid detergent fiber	25.5	25.6

^A SBM treated with GTM at 5%, GTM crude extract contained 13.1 % of tannin-catechins.

Results and Discussion

The ingredient and nutrient composition of diets are shown in Table 1. Diets were formulated to be isonitrogenous (16%), however, analyses ranged from 15.0% to 15.6%. Para grass was the main source of fiber in this study and contained 4.7% CP, 78.8% NDF, and 43.7% ADF. Treated SBM had lower in CP compared to untreated SBM (46.3% and 47.8%, respectively). Intake of tannin-catechins for cows fed GTM group averaged 5.8g/day (0.07% of diet DM). The dry matter intake

(DMI) of TMR and BW were not influenced by dietary treatments (Table 2). Aerts et al. (1999) reported that high dietary condensed tannins concentration in forage legume (6-12% of DM) can depress feed intake, digestibility and animal performance. Serum concentration of NEFA and BHB did not differ between the 2 groups (Table 2). After calving, cows showed an increase of TBARS. Oxidative status of dairy cows was related to energy status. Cows with higher BHB and NEFA showed higher TBARS and lower levels of antioxidants (Bernabucci et al., 2005). According to this study plasma concentration of TBARS at week 1 after calving had higher for both groups of cows than week 2 and week 3 ($P>0.05$). Although, no significant difference was observed in TBARS between control and GTM group, increased of TBARS immediately after calving confirms that cows during transition period are under oxidative stress conditions. O'Grady et al. (2006) has been reported the lack of an effect of dietary tea catechins on total antioxidant status of bovine plasma may indicate that tea catechins was excreted in the urine.

In present study, there were no differences in percentage of milk fat, protein and lactose and somatic cell counts between diet groups. However, cows fed GTM diet significantly improved ($P<0.05$) milk protein yield (kg/day) at week 2 (24%) and week 3 (19%) (data not shown). Theeraphaksirinont et al. (2009) also found that crossbred cows fed TMR supplemented with green tea waste at 5 or 10% of DM milk protein percentage increased ($P<0.05$). Dschaak et al. (2011) reported supplementation of quebracho condensed tannin extract at 3% of dietary DM in multiparous cow had decreased milk urea-N concentration. Therefore, in the presence of tannins, protein may be bound and protected from microbial degradation, but are released in the abomasums (Barry & McNabb, 1999). Moreover, Liu et al. (2013) reported that addition of chestnut tannins at 10g/kg could alleviate oxidative stress and inflammation of the mammary gland in lactating cows. In conclusion, dairy cow received diet contains SBM treated with 5% GTM crude extract gave higher milk protein yield, might has been improve utilization of nitrogen for milk production. Increasing level of GTM crude extract more than 5% might have both effect on reducing of protein degradation by tannins to improve milk production and giving greater sufficient levels of catechins to reduce oxidative stress in dairy cows.

Table 2. Body weight, blood composition after parturition and milk production of transition dairy cows fed TMR with untreated or treated SBM with 5%GTM.

	Control	GTM	SEM	Pr>t
BW (kg(after parturition (day 1)	410	426	11.88	0.65
BW (kg(after parturition (week 3)	400	408	11.46	0.81
DMI after parturition) %BW(2.1	2.0	0.04	0.66
NEFA (mmol/L)week 1	0.7	0.7	0.05	0.96
week3	0.4	0.3	0.02	0.67
BHB (mmol/L)week 1	0.3	0.4	0.02	0.15
week3	0.3	0.3	0.01	0.41
TBARS (nmol/mL)week 1	1.7	1.8	0.05	0.65
week3	1.6	1.3	0.06	0.18
Milk yield) kg/d(
week 1- 3	11.5	13.1	0.42	0.28
Milk protein(%)				
week 1-3	3.1	3.3	0.07	0.55
Milk fat(%)				
week 1- 3	3.4	3.7	0.07	0.54
Lactose(%)				
week 1- 3	4.7	4.7	0.06	0.32
Somatic cell counts) x10 ³ cell/mL(
week 1- 3	27.1	36.1	5.38	0.45

References

- Barry, T. and W. C. McNabb, 1999. The implications of condensed tannins on the nutritive value of temperate forages fed to ruminants. *Br. J. Nutr.* 81: 263-272.
- Bernabucci, U., B. Ronchi, N. Lacetera and A. Nardone, 2005. Influence of body condition score on the relationship between metabolic status and oxidative stress in periparturient dairy cows. *J. Dairy Sci.* 88: 2017-2026.
- Lui, H. W., D. W. Zhou and K. Li, 2013. Effects of chestnut tannins on performance and antioxidative status of transition dairy cows. *J. Dairy Sci.* 96: 5901-5907.

Utilization of *Samanea saman* pod meal as protein source in diet on voluntary feed intake and digestibility of goats

Anantasook^{1*}, N., P. Gunun², A. Cherdthong³, M. Wanapat³ & W. Kaewwongsa¹

¹Program in Animal Production Technology, Faculty of Technology, UdonThaniRajabhat University, UdonThani

²Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, SakonNakhon Campus, Phangkhon, SakonNakhon

³Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, KhonKaen University, KhonKaen

Abstract

The objective of this study was to investigate the utilization of *Samanea saman* pod meal as protein source in diet on voluntary feed intake and digestibility of goats. The experimental design were randomly assigned according to 4×4 Latin square design)LSD(and the dietary treatments were substitute of SBM by *saman* pod meal at 0%)T1(, 30%)T2(, 60%)T3(, and 100%)T4(, respectively. The results revealed that voluntary feed intake and apparent digestibility of DM, OM, CP, NDF and ADF were not significantly difference among treatment when replacement soybean meal by *saman* pod meal up to 100%. Based on this study, it could be concluded that *saman* pod meal can use as protein sources in concentrate and can replace soybean meal up to 100% without negative effect on feed intake and digestibility of goat.

Keywords: soybean meal, *samanea saman*, feed intake, digestibility

*Corresponding author: nirawan_kku@hotmail.com

Introduction

Samanea saman is local tree legume grown in tropical and sub-tropical area. Pods are black-brown, filled with a sticky, sweet, brownish pulp and falls during the dry season. Jetana et al. (2010) reported that *saman* pod contain high total sugar (10.00–17.30%) and protein (18–23%) content which potential to use as ruminant feed. Our previous study in *in vitro* have shown that substitute of soybean meal by *saman* at 60% in concentrate could improve *in vitro* gas production, degradability, microbial mass and NH₃-N concentration) Anantasook et al., 2014(. However, little information is available on the use of *saman* pod meal as a protein source in the diet for ruminant. Therefore, the objective of this study was to determine the Utilization of *saman* pod meal as protein source in diet on voluntary feed intake and digestibility of goats

Materials and methods

Four male goats with initial BW of 33±7 kg were randomly assigned according to 4×4 Latin square design (LSD(. The dietary treatments were replacement SBM by *Samanea saman* pod meal

at 0%)T1(, 30%)T2(, 60%)T3(, and 100%)T4(, respectively. Pangola grass hay)PH(was used as a roughage source. Feeds were sampled and fecal samples were collected from the total collection of each individual goats during the last 7 days of each period at the morning and

Table 1. Ingredients and chemical composition of concentrate used in the experiment.

Item	T1	T2	T3	T4
Ingredient, kg DM				
Cassava chip	65.0	62.0	59.0	56.3
Rice bran	8.0	9.9	12.0	13.0
Palm kernel meal	5.5	6.2	8.0	9.0
<i>Samaneasaman</i>	0.0	4.4	9.1	14.2
Soybean meal	14.0	10	4.4	0.0
Urea	2.5	2.5	2.5	2.5
Molasses	3.0	3.0	3.0	3.0
Salt	0.5	0.5	0.5	0.5
Sulfur	0.5	0.5	0.5	0.5
Mineral premix	1.0	1.0	1.0	1.0
Chemical composition				
Dry matter, %	87.8	88.1	88.2	87.9
	-----g/kg of dry matter-----			
Organic matter	92.9	95.0	94.7	94.4
Crude protein	14.0	13.9	13.9	13.9
Neutral detergent fiber	19.6	22.8	22.9	24.8
Acid detergent fiber	13.2	13.1	13.4	14.0

¹T1= substitute of SBM by *saman* at 0%, T2= 30%, T3= 60%, and T4=100%

afternoon feeding. Feeds, refusals and fecal samples were dried at 60 °C and ground)1 mm screen using Cyclotech Mill, Tecator, Sweden(, and analyzed for DM, ash, CP content (AOAC, 1995(. All data were analyzed as a 4x4 Latin square design using the general linear procedure in PROC GLM of Statistical Analysis System)SAS, 1998(.

Results and Discussions

Feed ingredients and chemical composition of dietary treatment was presented in Table 1. The mixture of concentrate, consisting of available local feed resources such as energy source (cassava chip, rice bran and molasses), protein sources (soybean meal, *saman* pod meal) and non-protein nitrogen (urea), had a higher quality in terms of CP (14.0 %). Feed intake and apparent digestibility of DM, OM, CP, NDF and ADF are presented in Table 2. It was found that total feed intake was not show negatively effect when replacement soybean meal by *saman* pod meal up to 100%. Moreover, apparent digestibility of DM, OM, CP, NDF and ADF were not significantly difference among treatments. The result indicated that *saman* pod meal can use as protein sources in concentrate and can replace soybean meal up to 100% without negative effect on feed intake and digestibility of goat.

Table 2. Effect of *samaneasaman* pod meal as protein sources in the diet on feed intake and digestibility

Item	Treatment ¹				SEM	P-value
	SBM	Saman 30	Saman 60	Saman 100		
DMI						
g/h/d	941.5	948.8	938.5	934.3	2.98	ns
% BW	2.8	2.8	2.8	2.6	0.07	ns
% BW ^{0.75}	66.3	66.6	66.0	62.0	1.09	ns
Apparent digestibility, %						
DM	64.9	66.5	62.0	58.8	1.62	ns
OM	70.3	71.2	67.5	64.1	1.21	ns
CP	62.6	68.2	60.8	58.5	2.21	ns
NDF	58.2	60.8	55.8	53.3	1.81	ns
ADF	41.8	43.8	40.0	39.1	2.39	ns

¹SBM = soybean meal, *Saman* 30, 60, 100 = soybean meal replacement by *Samaneasaman* pod meal 30 %, 60% and 100%, ns = non-significant

Conclusion

Based on this study it can be concluded that replacement of soybean meal by *saman* pod meal in concentrate at 100% did not show negatively effect on feed intake and apparent digestibility. These results revealed a potential use of *saman* pod meal as protein sources in concentrate for goat.

Acknowledgments

The authors would like to express their most sincere thanks to the **Research and Development Institute** for providing financial support and special thanks to the **Center of Science and Technology for Research and Community Development**, Program in Animal Production, Faculty of Technology, Udon Thani Rajabhat University for laboratory and research facilities.

References

- Anantasook, N.P. Gunun, M. Wanapat and W. Kaewwongsa, J. Jantapong, S. Sangkaew and P. Thattamlay. 2014. Effects of Different levels of *samaneasaman* pods meal in concentrate on degradability and *in vitro* gas production. Thai Journal of Animal Science. 1 SUPPL. 1:181-184.
- AOAC. 1995. Official Methods of Analysis. Association of Official Analysis Chemists, DC, U.S.A.
- Jetana, T., C. Vongpipatana, S. Thongruay, S. Usawong and S. Sophon. 2010. Apparent digestibility, nitrogen balance, ruminal microbial nitrogen production and blood metabolites in Thai Brahman cattle fed a basal diet of rice straw and supplemented with some tropical protein-rich trees. Asian-Aust. J. Anim. Sci. 23: 465-474.
- SAS Institute Inc., 1998. User's Guide: Statistic, Version 6.12th Edition. SAS Inst. Inc., Cary, NC.

Digestibility and nitrogen balance of growing goats fed different *Mimosa pigra* (L.) meal levels

Kaewwongsa, K. *, W.Tanodtaisong, N. Buddekoum, A. Sakun, P. Tayida, & S. Sonla

Program in Animal Production Technology, Faculty of Technology, Udon Thani Rajabhat University, Udon Thani, 41000, Thailand

Abstract

The present study was conducted to evaluate the effects of different levels of *Mimosa pigra* (L.) as replacement protein source of soybean meal (SBM) in growing goat diets on apparent digestibility coefficients and nitrogen balance. Growing goats were randomly assigned to four dietary treatments according to replicate 4x4 Latin square design. Dietary treatments were 4 levels of replacement of SBM to *M. pigra* (L.) at 0, 33.3, 66.7 and 100 % of protein source of SBM. The results showed that digestibility of digestible dry mater (DDM) and digestible organic mater (DOM) of goats fed with *M. pigra* (L.) replacement of SBM were not significant difference ($P>0.05$). Digestible crude protein (DCP) of goats fed with *M. pigra* (L.) replacement of SBM were significant difference ($P<0.05$). Similarly, Nitrogen balance was not significantly varied for treatment. The data suggest that *M. pigra* (L.) replace up to 100% of SBM in goat diets.

Keywords: *Mimosa pigra* (L.) meal, replacement, digestibility, Nitrogen balance

*Corresponding author: kaewwongsa@hotmail.co.th

Introduction

Mimosa, *Mimosa pigra* L., has well-documented impacts on primary industry and the environment in Australia and Asia (Miller, 1988). The chemical composition was similar to *Leuceana leucocephala*. *M. pigra* meal was weed that can be used as a feedstuff to the animals. Dried leaves of *M. pigra* can be included in the diets of quail, broiler, laying hen and pigs (Devendra, 1989). The above information obviously reflects that *M. pigra* meal are promising alternative protein source in animal diets. *M. pigra* leaves have a high protein content of 20 to 23% in DM according to Vearasilp et al. (1981a,b). Therefore, the present study evaluated productive performance of meat goats fed diets containing various CP replacement levels of *M. pigra* meal.

Materials and methods

Animals and Sampling methods

Eight growing crossbreed (Thai native x Anglo-Nubian) male meat goats of 7-8 months old were used in this study. The goats were kept in individual pens and received free choice of clean fresh water and mineral block. Individual body weight and feed consumption per cage were measured at the end of weekly to 7th week to determine the feed consumption, total feed intake, nutrient digestibility and nitrogen balance.

Experimental design and treatments

The experiment was taken according in replicated 4x4 Latin square design. Dietary treatments were four levels of replacement SBM with *M. pigra* meal at 0, 33, 67, and 100 % of crude protein (CP) in concentrates. The dietary treatments with T1: control diet with soybean meal (SBM) based, T2: replacing SBM by 33.3% of *M. pigra* meal, T3: replacing SBM by 66.7% of *M. pigra* meal and T4: replacing SBM by 100% of *M. pigra* meal. All diets were isonitrogenous and formulated to meet or exceed the NRC (1988) recommendations for all nutrients, regardless of treatment. Animal were fed concentrate 1.5% BW. The experiment was conducted for four periods, each period lasted 21 days. During the first 14 days, all animals were fed their respective dietary treatments at 1.5% BW of concentrate and fed with rice straw ad libitum.

Statistical methods

All data were subjected to analysis of variance using Proc ANOVA (SAS, 1996) and treatment means were statistically compared by Duncan's New Multiple Range Test (Steel & Torrie, 1980).

Results and discussion

The total intake of the meat goats were showed that Table 1. Total dry matter intakes expressed as g/day were increased as increasing the level of replacing soybean meal (SBM) with *M. pigra* meal ($P>0.05$) (Dawson et al., 1990); decreased ruminal lactate concentrations (Williams et al., 1991) (Table 1).

Table 1 Effects of *Mimosa pigra* (L.) meal replacement of SBM in dietary meat goats.

Items	<i>M. pigra</i> (L.) meal replacement of SBM				SEM	Contrast ¹	
	0:100	33.3:66.7	66.7:33.3	100:0		L	Q
Total dry matter intake							
g/day	511.22	516.1	523.4	525.79	11.69	ns	ns
%BW	3.82	4.05	3.81	3.61	0.09	ns	ns
gDM/kgBW ^{0.75}	72.91	76.21	73.01	70.42	1.28	ns	ns
Digestibility (%)							
DM	71.48	70.67	71.82	68.79	1.34	ns	ns
OM	76.23	75.80	76.86	74.44	1.16	ns	ns
CP	79.94 ^a	70.53 ^{ab}	70.26 ^{ab}	65.02 ^b	1.74	*	ns

¹contrast effect (L=Linear and Q=Quadratic), SEM= Standard error of means, ns=not significantly different ($P>0.05$), ^{a, b, c} Value on the same row under each main effect with different superscripts differ significantly ($P<0.05$), *Value on the same row under each main effect with different superscripts differ significantly ($P<0.05$), DM=Dry matter, OM=Organic matter, CP=Crude protein

Nitrogen intake was decreased highly significantly different ($P<0.01$) and total nitrogen excretion were increased linearly as a consequence of *M. pigra* replacement of SBM diets ($P>0.05$). Urine N excretion and N output were increased *M. pigra* replacement of SBM diets ($P<0.15$). Similarly, N absorption and N retention (g/day) were decreased at goat fed *M. pigra* replacement of SBM diets and were significantly different ($P<0.05$) in the Table 2

Table 2 Nitrogen balance of meat goats fed *M. pigra* replacement of SBM diets in growing meat goats

Items	<i>M. pigra</i> (L.) meal replacement of SBM				SEM	Contrast ¹	
	0:100	33.3:66.7	66.7:33.3	100:0		L	Q
N intake	22.08 ^a	18.78 ^b	19.39 ^b	16.74 ^c	0.59	**	ns
N Feces	3.22	4.03	4.10	4.70	0.23	ns	ns
N Urine	0.11 ^b	0.44 ^a	3.34 ^{ab}	0.45 ^a	0.07	**	ns
N output	3.33	4.46	4.40	5.18	0.25	ns	ns
N absorption	18.87 ^a	14.75 ^b	15.31 ^b	12.72 ^b	0.63	ns	**
N retention	19.22 ^a	15.80 ^{bc}	16.39 ^b	13.63 ^c	0.61	*	**
N retention (%)	87.00	83.83	83.82	81.06	0.78	ns	ns

¹contrast effect (L=Linear and Q=Quadratic), SEM= Standard error of means, ns=not significantly different (P>0.05), a, b, c Value on the same row under each main effect with different superscripts highly differ significantly (P<0.01)

Conclusion

It could be concluded that inclusion 33.33-66.67 % of *M. pigra* meal as replacement for SBM in the diet of growing meat goats were significantly affect the total feed intake, nutrient digestibility and nitrogen retention (P<0.05). It is recommended that *M. pigra* meal should be used by growing meat goats farmers to replace SBM in order to save cost and increase production.

Acknowledgements

The authors acknowledge Udon Thani Rajabhat University for facilities and financially supporting this research.

References

- Dawson, K. A., Newman, K. E., and Boling, J. A. 1990. Effects of microbial supplements containing yeast and lactobacilli on roughage-fed ruminal microbial activities. *J. Anim. Sci.* 68: 3392-3398.
- Devendra, C., 1989. Shrubs and tree fodders for farm animals. Proceedings of a workshop in Denpasar, Indonesia, 24-29 July 1989.
- Goering, H. K., and Van Soest, P. J. 1970. Forage fiber analysis. Agricultural handbook no. 379. Agricultural Research Service, US. Department of Agriculture. Washington, DC.
- Miller, I.L. 1988. Aspects of the Biology and Control of *Mimosa pigra* L. MSc. Agr. thesis, The university of Sydney, 248 pp.
- Schneider, B. H., and Flatt, W. P. 1975. The Evaluation of Feeds Through Digestibility Experiments (pp. 78-121). The University of Georgia Press, Athens. GA, USA.
- Vearasilp, T., Phuagphong, B. and Ruengpaibul, S. 1981a. A comparison of *Leucaena leucocephala* and *Mimosa pigra* L. in pig diets. *Thai Journal of Agricultural Science*, 14, 311-317.
- Vearasilp, T., Potikanond, N. and Rajja-Apai, P. 1981b. *Mimosa pigra* in sheep rations. *Thai Journal of Agricultural Science*, 14, 59-64.

Effects of banana stem supplemented on productive performance of finishing pigs

Kaewwongsa, W.*; A. Piladang, T. Woala, N. Suwunsri, & P. Suku

Program in Animal Production Technology, Faculty of Technology, Udon Thani Rajabhat University, Udon Thani, 41000, Thailand

Abstract

The study of this experiment was to investigate the effect of supplementation banana stem in diets on average daily gain (ADG), feed conversion ratio (FCR), feed intake, and backfat thickness in growing to finishing pigs. Eighteen pigs cross bred with an average 63 ± 3 kg were fed three experimental diets in a completely randomized design. The animals were randomly assigned to each sequence of feeding on the three dietary treatments. The dietary treatments were T₁ = control diet, T₂ = control diet + fresh banana stem and T₃ = control diet+ fermented banana stem with molasses. The results found that the increased ADG, FCR and body weight gain were significantly difference ($P < 0.05$) when used banana stem supplemented. Average daily feed intake and backfat thickness were not significantly difference ($P > 0.05$) when used banana stem supplemented in diets. The findings reflected that fresh banana stem supplementation can be used as fed for growing to finishing pigs.

Keywords: banana stem, finishing pigs, supplemented, ADG,

**Corresponding author: kaewwongsa@hotmail.co.th*

Introduction

Bananas (*Musa cavendishii*) and plantains (*Musa paradisiaca*) are mainly used as human food, a considerable amount of reject fruit could be fed to livestock, particularly to pigs. The vegetative part of the plant, the pseudo-stems and leaves, contains more than 60% of the dry matter of the whole plant and has been used experimentally as meal for pigs in concentrate rations (Garcia et al., 1991a,b). Banana stem is often use as animal feed especially pig by mixing with rice bran and fed directly to pig, however, banana stem is low in nutrients and high fiber which was effect on pig growth performance but nutrient was improved when banana stem incorporate silage. The aim of the present experiment is therefore to study banana stem on intake and nutrient digestibility of growing to finishing crossbred pig

Materials and methods

Animals and Sampling methods

The experiment was performed using Eighteen pigs cross bred with an average 63 ± 3 kg were divided into 3 equal groups. The animals were kept in groups in pens (6 animals each). Each pen was equipped with a self-feeder and nipple drinker to allow ad libitum access to feed and water throughout the experiment. Individual body weight and feed consumption per cage were measured at the end of weekly to 12th week to determine the feed consumption, average daily feed intake (ADFI), feed conversion ratio (FCR), feed efficiency (FE, %), backfat thickness.

Experimental design and treatments

The animals were randomly divided into three treatment groups comprising 6 replications according to Completely Randomized Design (CRD). The experimental treatments included: T1 = control diet, T2 = control diet + fresh banana stem and T3 = control diet+ fermented banana stem with molasses. All diets were provided in mash form and formulated to meet or exceed the NRC (1998) recommendations for all nutrients, regardless of treatment.

Sampling and measurement

Individual body weight and feed consumption per pen were measured at the end of weekly to 9th week to determine the average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR). At the end of the trial, all pigs were weighed and scanned with ultrasound to determine backfat thickness and loin muscle area.

Statistical methods

All data were subjected to the GLM procedures of SAS (1996) as a completely randomized design, with each pen serving as the experimental unit. Differences among all treatments were separated by Duncan's multiple range tests. Mean values and standard error (SE) were reported. Probability values less than 0.05 were considered as significant (Steel & Torrie, 1980).

Results and discussion

Final weigh were differ among all the treatments ($P<0.05$). Body weight gain and ADG were increased by T1, T2, and T3 supplementation, respectively ($P>0.05$). In addition FCR were not significantly difference ($P>0.05$) show that in Table 1. Replacing rice bran by mixture of taro foliage and banana stem silage did not showed any different in total DM intake, intake g/kg live weight, OM intake and CF intake and high crud fiber in the diets affects gut size and development, particularly the large intestine (Jorgensen et al., 1996). However, pigs are able to digest a substantial part of plant fiber pre-caecally (Lindberg and Anderson 1998).

Table 1 Main effects of banana stem supplemented on growth performance

Items	Banana stem supplemented			SEM	P-value
	T1	T2	T3		
Initial weigh (kg)	61.33	66.02	66.00	1.43	NS
Final weigh (kg)	79.17 ^b	87.33 ^a	83.16 ^{ab}	1.43	NS
Body weight gain (kg)	17.83	21.31	17.16	1.07	NS
ADG (kg/day)	0.25	0.30	0.24	0.01	NS
FCR	4.04	3.67	4.63	0.29	NS
ADFI (kg/day)	1.98 ^c	2.81 ^a	2.34 ^b	0.12	*
Feed efficiency (%)	25.1	27.4	22.6	0.01	NS
Backfat thickness (mm)	3.33 ^b	4.26 ^a	4.10 ^a	0.15	*
Meat color					
L*	41.71	40.82	43.08	1.04	NS
a*	4.50	5.25	4.78	0.04	NS
b*	12.28	13.47	12.74	0.26	NS

The dietary treatments were T₁ = control diet, T₂ = control diet + fresh banana stem and T₃ = control diet+ fermented banana stem with molasses, ^{a,b}Means in the same row with different superscripts are significantly different ($P<0.05$), SEM=standard error of mean, ADG=average daily gain, FCR=feed conversion ratio, ADFI=average daily feed intake

Conclusion

Considering the data obtained herein, the dietary banana stem supplementation had effect on finishing pigs while partially exerted beneficial effects on growth performance.

Acknowledgements

Appreciation is extended to Udon Thani Rajabhat University, Thailand for facilities and financially supporting this research. We also thank the Research Station farm crew for animal care in these experiments.

References

- AOAC 1990 Official Methods of Analysis. Association of Official Analytical Chemists. 15th edition (K Helrick editor). Arlington pp 1230.
- Bui Huy Nhu Phuc, Preston T R, Ogle B and Lindberg J E 1996 the nutritive value of sun-dried and ensiled cassava leaves for growing pigs. *Livestock Research for Rural Development* 8: (3) <http://lrrd.org/lrrd8/3/bui.htm>.
- Buntha P, Borin K, Preston T R and Ogle B 2008: Effect of Taro (*Colocasia esculenta*) leaf silage as replacement for fish meal on feed intake and growth performance of crossbred pigs. *Livestock Research for Rural Development*. Volume 20, supplement. Retrieved April 21, 2011, from <http://www.lrrd.org/lrrd20/supplement/bunt3.htm>
- García, A., Dominguez, P.L. and Ly, J. 1991a. Estudios de composición bromatológica de plantas de plátano (*Musa spp*) destinados a la alimentación porcina. Resúmenes IV Congreso ALVEC Habana. p 108.
- García, A., Ly, J. and Dominguez, P.L. 1991b. Uso de diferentes niveles de harina de residuos foliares del plátano (*Musa spp*) en piensos secos para cerdos en preceba. Resúmenes IV Congreso ALVEC La Habana p 94.
- Jorgensen H., Zhao X, Eggum B.O., and Zhao X.Q. 1996. The influence of dietary fiber and environmental temperature on development of the gastrointestinal tract, digestibility, degree of fermentation in the hind gut and energy metabolism in pigs. *British Journal of Nutrition* 75, 365-378.
- Lindberg J.E., and Andersson, C. 1998. The nutritive value of barley-based diets with forage meal inclusion for growing pig base on total tract digestibility and nitrogen utilization. *Livestock Production Science* 56, 43-52.
- Rodríguez Lylian, Preston, T.R., and Peters, K. 2009. Studies on the nutritive value for pigs of New Cocoyam (*Xanthosoma sagittifolium*); digestibility and nitrogen balance with different levels of ensiled leaves in a basal diet of sugar cane juice. *Livestock Research for Rural Development*. Volume 21, Article #27. <http://www.lrrd.org/lrrd21/2/rodr21027.htm>.

Effect of cultivation time on populations of yeast and lactic acid bacteria co-cultures in fermented milk

Polyorach^{*1} S., O. Pongchompu¹ & M. Wanapat²

¹Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, Phang Khon, Sakon Nakhon, Thailand, ²Tropical Feed Resources Research and Development Center (TROFREC), Faculty of Agriculture, Khon Kaen University, Muang, Khon Kaen, Thailand

Abstract

The aim of this study was to study effects of cultivation time on populations of yeast and lactic acid bacteria co-cultures in fermented milk. This experiment was conducted according to a Completely Randomized Design (CRD) to study growth kinetics of yeast (*Saccharomyces cerevisiae*) and lactic acid bacteria (*lacto bacillus*) from difference cultivation time at 0, 4, 8, 24, 48, 72 and 96 h post-cultivation times. The results showed that pH of fermented milk was the highest ($P<0.01$) at 0 h of cultivation (pH7.01) and the lowest ($P<0.01$) at 96 h post-cultivation (pH 3.30). For temperature was ranged from 26.0 to 31.0 °C. Yeast populations were highest ($P<0.05$) since 72 h-post (6.90 log cell/mL), while, lactic acid bacteria was highest since 48 h-post-cultivation (9.67 log cell/mL). In conclusion, co-cultures of yeast and lactic acid bacteria in fermented milk at 72 h were the highest of yeast and lactic acid bacteria populations (6.90 log cell/mL and 9.67 log cell/mL, respectively).

Keyword: yeast, fermented milk, cultivation time, lactic acid bacteria, co-cultures

*Corresponding Author: neenart324@hotmail.com

Introduction

Now a day, micro-organism are more widely used in both food and feed industry. Yeast and Lactic acid bacteria (LAB) are very interesting single cell protein strain because firstly, growth of microorganism is very much fast, secondly, a broader range of materials may be considered as suitable substrates depending on the microorganism chosen. The two chief strategies with regard to substrate to consider are: low grade waste material, or to use relatively simple carbohydrate source to produce microbial material containing very high quality of protein. Many researcher interested in using microorganism to improve feed quality (Polyorach et al., 2011; 2013)

Fermentation, the microbial degradation of organic compounds without net oxidation, is an important process in the global carbon cycle and is also exploited worldwide for the production. Fermented milk, also known as cultured milk, is a type of dairy food which is made by adding lactic acid bacteria, mold, or yeast to milk.

However, the mechanisms of possible interaction between yeasts and the lactic flora have not been widely studied. Therefore, the objective of this study was to study effects of cultivation time on population of yeast and LAB co-cultures in fermented milk

Materials and methods

Yeast and LAB cultivation

Activated yeast: by weight 20g of Baker's yeast into a flask and add with sugar 20 g and distilled water 100 mL. Then mixed and incubated at room temperature for 1 h (A). Activated LAB: by weight 50 g commercial yoghurt, 25 g molasses and 25 mL distilled water, mixed well and incubated at room temperature for 2 h (B). Mix (A) and (B) with 500 g of raw milk. Fermented milk was measure pH and took samples at 0, 4, 8, 24, 48, 72 and 96 h post-cultivation.

Determination of yeast and lactic acid bacteria growth

Yeast and LAB populations were determined by direct counting.

Analysis of data

All data were statistically analyzed using analysis of variance of a completely randomized design with CRD using Proc. GLM procedure (SAS, 1998). Treatment means were statistically compared using Duncan's New Multiple Range Test (Steel & Torrie, 1980). Differences among means with $P < 0.05$ were accepted as representing statistically significant differences.

Results and Discussion

The result from this study as showed in Table 1. The population of yeast was the highest ($P < 0.01$) (6.90 log cell/mL) started from 72 h-post cultivation, while, LAB started from 48 h-post cultivation (9.67 log cell/mL).

Under current study, optimal cultivation time was in agree with Polyorach et al. (2013) who reported that yeast fermented liquid at 66 h post-cultivation (3×10^{11} cell/mL), which could be due to different medium and an environment. While, Narvhus & Gadaga (2003) also reported that the fermentation of natural fermented milk usually takes 2–3 days and if the product is immediately consumed, or kept under refrigeration, then any effect of interaction between yeasts and LAB in the product has to be evident within this short fermentation period. The yeast counts recorded in this study were in similar range to those reported by Mathara et al. (2008) who found yeasts counts of < 1.0 – 7.4 log₁₀ cfu/mL. The presence of yeast in traditionally fermented milk products, in varying numbers, has been reported elsewhere (Njage et al., 2011). The frequent concurrence of LAB and yeasts has led to the suggestion that there could be interactions that may an influence the product characteristics and quality (Narvhus & Gadaga, 2003). The fermented milk temperature values were range from 26–31°C, while, pH was the highest ($P < 0.01$) at 0 h of cultivation (pH 7.01) and the lowest ($P < 0.01$) at 96 h post-cultivation (pH 3.4).

Table 1. Yeast and LAB populations count in fermented milk.

Time of incubation (h)	Log cell/mL		pH	Temperature
	Yeast	lactic acid bacteria		
0	3.48 ^f	3.74 ^e	7.01 ^a	26.00 ^d
4	3.80 ^e	4.37 ^d	6.30 ^b	28.00 ^c
8	4.00 ^d	4.00 ^c	5.20 ^c	28.00 ^c
24	4.85 ^b	6.77 ^b	4.51 ^d	29.00 ^{bc}
48	6.87 ^b	9.67 ^a	3.70 ^e	30.04 ^{ab}
72	6.90 ^a	9.67 ^a	3.40 ^f	31.00 ^a
96	6.91 ^a	9.66 ^a	3.30 ^f	31.00 ^a
SEM	0.006	0.042	0.080	0.387
P-Value	0.0001	0.0001	0.0001	0.0001

^{a-f} Means in the same row with different superscripts differ (P<0.01).

SEM= sum square error of the mean.

The decreasing of ruminal pH that might be due to at the beginning of milk fermentation process, lactic acid bacteria ferment the lactose in the milk to lactic acid. Wszolec et al. (2001) reported that fermented milk product utilizes a wide variety of microorganisms to produce a wide variety of products in addition to lactic acid including ethanol, free fatty acids, and acetaldehyde. When yeasts grow with LAB in milk, they need either to be able to obtain sufficient energy-giving substrates from the milk components or to be able to avail themselves of metabolites from LAB. *Saccharomyces cerevisiae* has been associated with the production of alcohols and other aroma compounds, stimulation of LAB, improvement of nutritional value, and inhibition of undesirable microorganisms (Jespersen, 2003). However, the yeasts present in this product need to be investigated further to establish their exact role in the fermentation process, including their interaction with LAB and their metabolic properties.

Conclusion

Base on this study could be concluded that co-cultures of yeast and lactic acid bacteria in fermented milk at 72 h was the highest of yeast and lactobacillus populations (6.90 log cell/mL and 9.67 log cell/mL, respectively). Moreover, the further research should be investigate the used of fermented milk to improve quality of feed resources especially in tropical area.

Acknowledgements

The authors would like to express their most sincere thanks to all who have assisted and supported the research in this study, particularly Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, Sakon Nakhon Campus, Phang Khon, Sakon Nakhon, Thailand for providing financial support of research and the use of research facilities.

References

- Gandhi, D.N. 2006. Food and industrial microbiology of fermented dairy products. Principal Scientist, Dairy Microbiology Division, National Dairy Research Institute, Karnal-132 001 (Haryana).
- Jespersen, L., 2003. Occurrence and taxonomic characteristics of strains of *Saccharomyces cerevisiae* predominant in African indigenous fermented foods and beverages. FEMS Yeast Res. 3: 191–200.
- Mathara, J.M., U. Schillinger, C. Guigas, C. Franz, P.M. Kutima and S.K. Mbugua, 2008. Functional characteristics of *Lactobacillus* spp. from traditional Maasai fermented milk products in Kenya. Int. J. Food Microbiol. 126: 57–64.
- Narvhus, J.A. and T.H. Gadaga, 2003. The role of interaction between yeasts and lactic acid bacteria in African fermented milks: a review. Int. J. Food Microbiol. 86: 51–60.
- Njage, P.M.K., S. Dolci, C. Jans, J. Wangoh, C. Lacroix and L. Meile. 2011. Characterization of yeasts associated with camel milk using phenotypic and molecular identification techniques. Res. J. Microbiol. 6: 678–692.
- Polyorach, S., M. Wanapat, C. Wachirapakorn, C. Navanukraw, S. Wanapat and N. Nontaso. 2011. Supplementation of Yeast Fermented Liquid (YFL) and coconut oil on rumen fermentation characteristics, N-balance and urinary purine derivatives in Beef cattle. J. Anim.Vet. Adv. 10: 2084-2089.
- Polyorach, S., M. Wanapat, S. Wanapat. 2013. Enrichment of protein content in cassava (*Manihot esculenta* Crantz) by supplementing with yeast for use as animal feed. Emirates Journal of Food and Agriculture. 25: 142-149.
- SAS. 1998. User's Guide: Statistic, Version 6.(12th Edition). (SAS Inst. Inc., Cary, NC.)
- Steel RGD, Torrie JH (1980) Principles and Procedures of Statistics. (McGraw Hill Book Co, New York)
- Wszolek, M., A.Y. Tamimi, D.D. Muir, M.N.I. Barclay. 2001. Properties of kefir made in Scotland and Poland using bovine, caprine, and ovine milk with different starter cultures. Lebensmittel-Wissenschaft und Technologie 34: 251-261.

Effect of dietary supplementation of bioceramic powders on production performance of broiler chickens

Mitchaothai^{*1}, J. & Ruenphet, S.²

¹Department of Veterinary Clinical Science, ²Department of Veterinary Pre-clinical Science, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Bangkok 10530, Thailand

Abstracts

This study was carried out to investigate the influence of bioceramic powders on production performance of broiler chickens. Eight, one day-old chickens with an average body weight of 46.5 g were allocated into two experimental groups (control and treatment) equally and housed individually. The broiler chickens in the control group were offered basal diet while the broiler chickens in the treatment group were offered basal diet supplemented with 0.10% bioceramic powders throughout 5 weeks of the present study. All chickens were offered experimental diets and drinking water *ad libitum* throughout the experimental duration. The body weight of the chickens and refusal feeds were weighed weekly for 5 weeks. Average daily gain (ADG) and Feed : Gain of the studied chickens were then calculated. The results have showed that the ADG of the chickens in the control group (42.93 ± 0.83 g/day) at period of week 1-3 was higher ($P \leq 0.01$) than that in the treatment group (35.73 ± 1.55 g/day). However, the Feed:Gain of the chickens in the treatment group (1.16 ± 0.05) at week 1-3 was better ($P \leq 0.01$) than that in the control group (1.37 ± 0.07). For ADG and Feed:Gain of the chickens at period of week 4-5 and overall (week 1-5), there were no significant differences ($P > 0.05$) (between both studied groups). Hence, it could be concluded that supplementation of bioceramic powders in diet at 0.10% would improve feed utilization efficiency, but lower growth rate of broiler chickens at week 1-3 of age.

Keywords: bioceramic powders, production performance, dietary supplementation, broiler chickens

**Corresponding author: jmitchaothai@yahoo.com*

Introduction

Bioceramic powders are the materials derived from chicken feces, prepared by sintering process (Thammakarn et al., 2013). The application use of the bioceramic powders is mainly for disinfectant purposes, especially for viral disease control in poultry production industry. In the present, there are still do not know the exact mode of action of the bioceramic powders. Takehara et al. (2009) reported that they possibly work by adsorbing virus particles. This proposed mode of action might be applied for balancing intestinal bacteria and enhancing growth performance. However, there is lack information for the impact of the bioceramic powder on intestinal bacteria

and also production performance. Therefore, this study was carried out to investigate the influence of bioceramic powders on production performance of broiler chickens.

Materials and Methods

A total of 8 male day-old Cobb chickens with an average body weight of 46.5 g were randomly allocated into two experimental groups (control and treatment) equally and housed individually. The broiler chickens in the control group were offered basal (corn-soybean meal) diet while the broiler chickens in the treatment group were offered basal diet supplemented with 0.10% bioceramic powders throughout 5 weeks of the present study. All chickens were offered experimental diets and drinking water *ad libitum* throughout the experimental duration. There were 2 dietary formulations for both studied chicken group; 1) the starter diet for chicken age of week 1-3 and 2) the grower diet for chicken age of week 4-5. The body weight of the chickens and refusal feeds were weighed weekly for 5 weeks. Average daily gain (ADG) and feed conversion ratio (Feed:Gain) of the studied chickens were then calculated. All data were analyzed as a completely randomized design (CRD) by independent Student's t test to detect significant difference at $P \leq 0.05$.

Results and Discussion

Average daily gain (ADG) and feed conversion ratio (Feed:Gain) of the studied chickens were demonstrated in the Table 1. At the week 1, the growth rate of the chickens offered the diet supplemented with bioceramic powders was higher ($P \leq 0.05$) than those obtained the control diet. At the week 2 and 3, the chickens offered the diet supplemented with bioceramic powders tended to lower ($P = 0.087$) and lowered ($P \leq 0.05$), respectively than those in the control group. In the meantime, the feed conversion ratio of the chickens offered the bioceramic powder diet had better than the chickens in the control group at week 2 ($P \leq 0.05$) and week 3 ($P \leq 0.01$) whereas there was no difference between both studied groups for feed conversion ratio of the studied chickens. When calculated as period of week 1-3, there were higher ($P \leq 0.01$) growth rate and feed conversion ratio ($P \leq 0.05$) of the chickens in the control group, compared with the chickens in the treatment group. However, at the week 4 and 5, the studied chickens of both groups had no difference ($P > 0.05$) for growth rate and feed conversion ratio.

Table 1. Mean±SD of average daily gain (ADG) and feed conversion ratio (Feed:Gain) at different ages of the experimental broiler chickens.

Item	Control	Treatment	P-value
<i>Average daily gain, g/day</i>			
Wk1	15.49±0.72	21.34±3.63	0.046
Wk2	31.28±2.11	28.61±1.44	0.087
Wk3	54.58±1.12	42.86±3.31	0.003
Wk4	58.14±2.09	60.89±8.40	0.566
Wk5	76.24±8.40	66.11±10.57	0.187
Wk1-3	42.93±0.83	35.73±1.55	0.001
Wk4-5	66.50±2.80	57.80±9.94	0.178
Wk1-5	57.07±1.99	48.97±6.36	0.079
<i>Feed : Gain</i>			
Wk1	1.75±0.13	1.72±0.34	0.877
Wk2	1.83±0.18	1.44±0.07	0.014
Wk3	1.70±0.11	1.12±0.08	0.000
Wk4	2.29±0.13	2.24±0.35	0.824
Wk5	2.12±0.33	2.23±0.36	0.652
Wk1-3	1.37±0.07	1.16±0.05	0.005
Wk4-5	2.21±0.22	2.47±0.42	0.322
Wk1-5	1.61±0.13	1.67±0.21	0.664

From the results of the current study, generally the bioceramic powder supplemented in diet for broiler chickens had lower growth rate (lower ADG), but improve feed utilization efficiency (lower Feed:Gain) of the chickens at the age of week 1-3 while no impact on both growth rate and feed utilization efficiency at the age of week 4-5. These results would imply that the age of the chickens is a factor affecting to the responses to dietary bioceramic powder supplementation. However, there was hardly to explain the discrepancy of the growth rate and feed utilization efficiency in the current study. The earlier study (Marcato et al., 2008), the deposition rate of protein in body of Cobb chickens remarkably increase after 21 days of age, implying slow rate of protein deposition during the first 21 days of the chickens. In the present study, the chickens received the control diet had higher growth rate, indicating lower protein deposition in body of the chickens offer the bioceramic powders. The explanation for this result might be the fact that the bioceramic powders had pH more than 10.0 (Thammakarn et al., 2015). The high pH would lower digestion of protein contents in digestive tract. Basically, the low pH of the gastric juice aids protein digestion by denaturing the tertiary structures of ingested protein to be easier for enzymatic digestion and by increasing the activation of pepsin. Thus, the chickens obtained the diet with the bioceramic powder supplementation might get less protein or meat deposition, resulting in less growth rate when compared with the chickens received the control (regular) diet. However, the feed utilization efficiency of the chickens in the treatment group was better than those in the control group. Several researchers have reported low pH in small intestine could improve nutrient utilization due to less damage of intestinal villi from pathogenic bacteria in small intestine. However, the study of Knarreborg et al. (2003) found that the high pH in the small intestine of chickens was accompanied by a significant increase in lipase activity, and coincided with a significantly lower concentration of unconjugated bile acids and a higher ratio of conjugated to unconjugated bile acids. This would lead to higher utilization efficiency of fat or energy of the chickens obtained the bioceramic powders, resulting higher feed utilization efficiency but lower

growth rate due to lower protein digestion as mentioned earlier. As data available and accessible, this study should be a first study for dietary supplement of bioceramic powders in broiler chickens, thus there still be required further studies in several aspects to elucidate the effect of the bioceramic powders.

In conclusion, the Cobb broiler chickens offered diet supplemented with 0.10% bioceramic powders throughout 5 weeks would improve feed utilization efficiency, but lower growth rate of broiler chickens at week 1-3 of age.

Acknowledgements

The authors would like to thank Mr.Chongpol Jaiwangdee, Ms.Watoo Todpetch, Ms.Prapatsorn Jaidee, Ms.Pensiri Theworksuwan and Ms.Narumol Ounyingcharean for their excellent assistant and Mahanakorn University of Technology for financial support.

References

- Knarreborg, A., S. K. Jensen and R. M. Engberg, 2003. Pancreatic lipase activity as influenced by unconjugated bile acids and pH, measured *in vitro* and *in vivo*. *J. Nutr. Biochem.* 14(5): 259-265.
- Marcato, S. M., N. K. Sakomura, D. P. Munari, J. B. K. Fernandes, I. M. Kawauchi and M. A. Bonato, 2008. Growth and body nutrient deposition of two broiler commercial genetic lines. *Brazilian J. Anim. Sci.* 10(2): 117-123.
- Takehara, K., O. Chinen, A. Jahangir, Y. Miyoshi, Y. Ueno, S. Ueda, Y. Takada, S. Ruenphet, K. I. Mutoh, M. Okamura and M. Nakamura, 2009. Ceramic powder made from chicken feces: Anti-viral effects against Avian Influenza Viruses. *Avian Dis.* 53: 34-38.
- Thammakarn, C., M. Tsujimura, K. Satoh, T. Hasegawa, M. Tamura, A. Kawamura, Y. Ishida, A. Suguro, H. Hakim, S. Ruenphet and K. Takehara, 2013. Efficacy and stability of ceramic powder to inactivate Avian Influenza Virus. *Int. J. Med. Health Biomed. Pharma. Eng.* 7(9): 257-259.
- Thammakarn, C., Y. Ishida, A. Suguro, H. Hakim, K. Nakajima, M. Kitazawa, K. Takehara, 2015. Inhibition of infectious bursal disease virus transmission using bioceramic derived from chicken feces. *Virus Res.* 204: 6-12.

Effect of cutting age and ensilage on chemical composition of Pak Chok1 and King giant napier grasses

Lukkaranukool¹, A., J. Mitchaothai^{*2}, P. Kaewhom¹ & P. Sudsarn¹

¹Department of Agriculture, Faculty of Agricultural Technology, Burapha University-Sakaeo Campus, Wattana Nakorn District, Sakaeo, 27160, Thailand

²Department of Veterinary Clinical Science, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Bangkok, 10530, Thailand

Abstracts

The current study was conducted to investigate the effect of cutting age and ensilage on chemical composition of Pak Chok1 napier and King giant napier grasses. These grasses were harvested at 30, 45 and 60 days after regrowth. The harvested grasses were made for silage, prepared and allowed to be fermented for 21 days at room temperature with 3 replications for each cutting age. Chemical compositions of each cutting age of fresh grasses and silages were quantified and physical quality (pH and % ammonia nitrogen) of silages was measured. The results have shown that there were no effects ($P>0.05$) of cutting age and grass type on the contents of EE, NDF and ADF. The DM contents in both grasses harvested at 45 and 60 days after regrowth was higher ($P\leq 0.05$) than those harvested at 30 days after regrowth. Both grasses had lowest value of CP contents at the cutting age of 45 days. The contents of OM, ash, EE, CP and NDF of both grass silages were mainly related to the nutrient composition to fresh form of the grasses. There were lowest value of pH and significant lowest of % ammonia nitrogen ($P\leq 0.05$) at the cutting age of 45 days for both grass silages. Thus, there was small difference of nutrient composition between both grasses in fresh form, but the King giant napier silage seems to be better for making silage, and suitable cutting age for making silage of both grasses seem to be at 45 days.

Keywords: cutting age, ensilage, chemical composition, Pak Chok1 napier grass, King giant napier grass

**Corresponding author: jmitchaothai@yahoo.com*

Introduction

Roughage is very crucial for ruminants as providing low cost nutrients. There were many factors affecting to nutritive values of roughage. In dry season, roughage is always shortage and then silage of forage is normally alternative way to preserve quality of roughage. Pak Chok1 napier (*Pennisetum purpureum* × *Pennisetum americanum*) and King giant napier (*Pennisetum purpureum* × *Pennisetum alopecuroides*) grasses are perennial forage crop with high growth rate,

high productivity and good nutritive value (Santos et al., 2001; Wadi et al., 2004). Cutting interval is a crucial factor affecting to their performance (Lukkananukool, 2012; Santana et al., 1989). There was scarce information for the King giant napier grown in Thailand. Therefore, this study was carried out to investigate the effect of cutting age and ensilage on chemical composition of the Pak Chok1 napier and the King giant napier grasses.

Materials and Methods

The grass species used for the current study are Pak Chok1 napier (*Pennisetum purpureum* × *Pennisetum americanum*) and King giant napier (*Pennisetum purpureum* × *Pennisetum alopecuroides*). These grasses were harvested at 30, 45 and 60 days after re-growth. Chemical composition (DM, OM, ash, EE, CP, NDF and ADF) of 3 replications for each cutting age of fresh grasses was quantified by AOAC (1990). Subsequently, the harvested grasses were made for silage, prepared and allowed to be fermented for 21 days at room temperature with 3 replications for each cutting age. Samples of the two grass silages were collected for chemical analysis by AOAC (1990) as same as the fresh form of the grasses and for physical quality (pH and % ammonia nitrogen; % NH₃-N).

Results and Discussion

The results have been shown in the Table 1. There were no effects ($P>0.05$) of cutting age and grass type on the contents of EE, NDF and ADF. The DM contents in the Pak Chok1 and the King giant napier grasses harvested at 45 and 60 days after regrowth was higher ($P\leq 0.05$) than those harvested at 30 days after regrowth. The Pak Chok1 napier at cutting age of 30 had lowest contents of OM ($P\leq 0.05$), but highest contents of ash ($P\leq 0.05$) when compared with cutting age of 45 and 60 days. In the meantime, the King giant napier at cutting age of 60 had lowest contents of OM ($P\leq 0.05$), but highest contents of ash ($P\leq 0.05$) when compared with cutting age of 30 and 45 days. The CP contents of the Pak Chok1 napier were higher ($P\leq 0.05$) than the King giant napier. Both Pak Chong1 and King giant napier grasses had lowest value of CP contents at the cutting age of 45 days. For both grass silages, there was influence of species and cutting age ($P\leq 0.05$) for DM contents by higher contents in King giant napier silage and increasing with cutting age. The contents of OM, ash, EE, CP and NDF of both grass silages were influenced by species ($P\leq 0.05$) with mainly related to the nutrient composition to fresh form of the grasses, but there was no effects ($P>0.05$) of cutting age and grass type on the contents of ADF for both grass silages. For physical quality of grass silages, there were lowest value of pH and significant lowest of % NH₃-N ($P\leq 0.05$) at the cutting age of 45 days for both grass silages.

From the results of the current study, there was small difference of nutritive values between the Pak Chong1 and the King giant napier grasses in fresh form. This would be explained by the fact that both studied grasses had closed genetic to each other. The nutrient composition of both studied fresh grasses is in agreement with earlier report (Lounglawan et al., 2014). After silage making for both studied grasses, the nutritive values of the grass silages were related to the values of fresh form in accordance with the report of Lukkananukool (2012). Although the nutritive composition of the King giant napier silage was better than the Pak Chong1 napier, these

differences were rather small, except for %NH₃-N found at quite low in the King giant napier silage.

In conclusion, it could be concluded that there was small difference of nutrient composition between the Pak Chong1 and the King giant napier grasses in fresh form, but the King giant napier silage seems to be better for making silage, and suitable cutting age for making silage of both grasses seem to be at 45 days.

Acknowledgements

The authors would like to thank Faculty of Agricultural Technology, Burapha University-Sakaeo Campus for financial support.

Table 1. Effects of species (S) and days of cutting after the re-growth (D) on chemical composition of studied fresh grasses and grass silage, and physical quality of grass silage.

Item	Pak Chong1 Napier			King Giant Napier			SEM	P-Value		
	30 D	45 D	60 D	30 D	45 D	60 D		S ^ψ	D ^Ω	S×D
<i>Fresh grass</i>										
DM (%)	10.86 ^b	24.51 ^a	26.78 ^a	6.53 ^b	28.87 ^a	27.00 ^a	0.91	ns	*	ns
OM (%)	85.75 ^c	88.85 ^a	86.95 ^b	86.99 ^b	88.08 ^{ab}	84.98 ^c	0.16	ns	*	*
Ash (%)	14.26 ^a	11.15 ^c	13.05 ^b	13.01 ^b	11.92 ^{bc}	15.02 ^a	0.16	ns	*	*
EE (%)	2.37	1.67	1.87	2.17	1.92	2.15	1.70	ns	ns	ns
CP (%)	10.93 ^a	7.24 ^{bc}	9.96 ^{ab}	8.25 ^{abc}	6.78 ^c	7.30 ^{bc}	0.36	*	*	ns
NDF (%)	71.03	63.447	69.49	66.36	67.90	70.86	0.96	ns	ns	ns
ADF (%)	51.18	48.07	50.83	49.73	47.99	54.43	0.93	ns	ns	ns
<i>Grass silage</i>										
DM (%)	14.58 ^d	16.497 ^{cd}	17.34 ^{abc}	17.05 ^{bc}	19.74 ^a	19.20 ^{ab}	0.31	*	*	ns
OM (%)	83.52 ^{ab}	77.14 ^b	86.18 ^a	84.71 ^a	79.03 ^{ab}	84.98 ^a	0.92	*	ns	ns
Ash (%)	16.47 ^{ab}	20.96 ^{ab}	13.82 ^b	15.29 ^b	22.85 ^a	15.01 ^b	0.92	*	ns	ns
EE (%)	2.93 ^a	1.67 ^b	1.68 ^b	2.65 ^a	1.73 ^b	1.49 ^b	0.04	*	ns	ns
CP (%)	7.26 ^a	5.94 ^{bc}	7.19 ^a	6.92 ^{ab}	5.26 ^c	6.66 ^{ab}	0.14	*	ns	ns
NDF (%)	53.71 ^d	65.72 ^{ab}	61.32 ^{bc}	58.18 ^{cd}	69.41 ^a	61.58 ^{bc}	0.84	*	ns	ns
ADF (%)	47.22	51.88	53.70	48.67	52.25	50.26	0.82	ns	ns	ns
<i>Physical quality of grass silage</i>										
pH	5.90 ^a	5.39 ^b	6.05 ^a	5.78 ^{ab}	5.67 ^{ab}	5.95 ^a	0.07	*	ns	ns
NH ₃ -N (%)	7.38 ^b	4.43 ^c	11.52 ^a	6.84 ^b	2.84 ^d	6.99 ^b	0.71	*	*	*

^{abc} Means in the same row with different superscripts are significantly different (* $P \leq 0.05$ and ns=not significant)

^ψ Species effect, ^Ω Days of cutting after the re-growth

References

- AOAC, 1990. Official methods of analysis association of official chemists. Washington, D.C.
- Lounglawan, P., W. Lounglawan and W. Suksombat, 2014. Effect of cutting interval and cutting height on yield and chemical composition of King Napier grass (*Pennisetum purpureum* x *Pennisetum americanum*). APCBEE Procedia. 8: 27-31
- Lukkananukool, A., 2012. Effects of forage sources and feeding regimes on rumen fermentation, nutrition digestibility and goat meat quality. Ph.D. thesis. Suranaree University of Technology. pp 195.
- Santana, J. R., J. M. Pererira and N. G. Arruda, 1989. Evaluation of cultivars of elephant grass (*Pennisetum purpureum*, Schum.) in the south of Bahia. 1. Agrosystems cacao. Rev. Bras. Zootec. 18: 273-283.
- Santos, E. A., D. S. da Silva and J. de Queiroz Filho, 2001. Productive aspects of elephant grass (*Pennisetum purpureum* Schum.) cv. Roxo in Balanga Paraibano. Rev. Bras. Zootec. 30: 31-36.
- Wadi, A., Y. Ishii and S. Idota, 2004. Effect of cutting interval and cutting height on dry matter yield and overwintering ability at the established year in *Pennisetum* species. Plant Prod. Sci. 7: 88-96.

Effects of levels of dried leucaena (*Leucaena leucocephala*) supplementation on nutritive value of milk in organic dairy cows

N. Suwanpanya^{*1} C. Kongsut² & W. Pomsema³

¹ Animal Production Technology, ² Crop Production Technology, ³ Animal Health Science Faculty of Agro-Industrial Technology, Rajamangala University of Technology Isan, Kalasin campus, Kalasin, 46000 Thailand

Abstract

The experiment was conducted to study the effects of dried leucaena (*Leucaena leucocephala*) supplementation at levels of 1.5 and 3.0 kg/h/d in organic dairy cows. The dairy cows receiving either non-urea (organic concentrate) and ureaadded (non-organic concentrate) based diets. Sixteen multiparous mid-lactating Holstein-Friesian 75% crossbred cows at an initial weight of 405±15 kg were randomly assigned to 4 treatments with 4 replications each in the randomized complete block design (RCBD). Four diets were given as follows: T1) Rice straw and organic concentrate T2) Rice straw + 1.5 kg/h/d leucaena + organic concentrate T3) Rice straw + 3 kg/h/d leucaena + organic concentrate T4) Rice straw and non-organic concentrate. The results revealed that there was significantly difference milk yield among the four treatment group (P<0.05). T2 group provided the highest milk yield at 11.46 kg/hd/d. Dairy cows in T3 group produced the highest milk compositions containing fat (4.52 %) protein (3.51%) SNF (9.60%) Omega3 (0.0065 mg/g) Omega6 (0.081 mg/g) Omega9 (1.272 mg/g). There were no differences in vitamin A (29-30 mg/100ml). In contrast dairy cows in T4 was lower on nutritive value of milk.

Keyword: leucaena, rice straw, milk, organic concentrate, non- organic concentrate, dairy cow

Corresponding email: nunthiya284@gmail.com

Introduction

The efforts of Government in the development of dairy farming in Thailand have been successful. There has been a satisfactory increase in milk production and in the number of cattle and dairy farmers, so that today Thai farmers are interested in organic farming. Because of public concerns about food safety, animal welfare and the environmental impacts of organic intensive livestock farming systems, the recent past, that there has been considerable interest in the potential health promoting properties of omega3, 6, 9, a fatty acid produced naturally in ruminant animals. The primary sources of omega3, 6, 9, are dairy fats derived from ruminant animals while vegetable fats and oil contain significantly lower levels. Omega3 was significantly lower when the grass allowance was at the lowest level (41.28 kg fresh grass/ cow/ day) compared with grazing at 0.31 and 0.58 mg/g of milk fat, respectively

(Suwanpanya et al., 2012) Rice straw was a major forage source based diets and lower % protein for organic dairy management in the dry season, the objective of this experiment was to evaluate optimum level of dried leucaena (*Leucaena leucocephala*) for organic dairy management in the dry season and study the nutritive value of milk in organic dairy cows.

Materials and Methods

Animals and experimental design

Twenty multiparous mid-lactation Holstein-Friesian 75% crossbred organic dairy cows in organic dairy farming at an initial weight of 405 ± 15 kg, were assigned to 4 treatments with 4 replications each in the randomized complete block design (RCBD). The treatments were 4 different feeding systems comprising of T1) Rice straw and organic concentrate T2) Rice straw + 1.5 kg/h/d dried leucaena + organic concentrate T3) Rice straw + 3 kg/h/d dried leucaena + organic concentrate T4) Rice straw and non-organic concentrate.

Animal management and feeding

All dairy cows in treatment 1, 2 and 3 received a non-urea (organic concentrate) diet. The diet consisted of soybean meal (35%), Soybean skin (16%), cassava chip (46%), calcium(2%), and premixed (1%) (total: 100 kg.). The dairy cows in treatment 4 received a ureaadded (non-organic concentrate) diet. The diet consisted of soybean meal (30%), Soybean skin (19%), cassava chip (46%), calcium (2%), premixed (1%) % and urea (2%) (total: 100 kg.). Concentrate was limited at 5 kg/hd/d. Animals were fed twice daily at 06.00 and 15.00 hours. Rice straw was limited at 10 kg/hd/d. Animals on treatment 1 were fed rice straw and organic concentrate. The dairy cows on treatment 2 were fed rice straw and 1.5 kg/h/d leucaena and organic concentrate. The dairy cows in treatment 3 were fed were fed rice straw and 3 kg/h/d leucaena and organic concentrate. The dairy cows in treatment 4 were fed rice straw and non-organic concentrate for 60 days experimental period.

Measurements

The concentrate was analyzed for dry matter (DM), ash, crude protein and fat (AOAC, 1990), Neutral detergent fiber (NDF) and acid detergent fiber (ADF) (Goering and Van Soest, 1970). Milk production records from cows each morning and evening of everyday of the experiment were obtained. Milk samples were collected and analyzed for components.

Results and Discussion

Organic concentrate and non-organic concentrate were limited at 5 kg/hd/d. The average chemical composition of dry matter, protein, fat, NDF and ADF, is shown in Table 1. The greatest chemical composition was found in organic concentrate was 18.88% protein. non-organic concentrate at 19.92% protein.

Table 1. Chemical composition of concentrate dietary treatment.

Chemical composition (%)	Organic concentrate	Non-organic concentrate
DM	90.41	90.61
Ash	8.57	9.41
EE	0.76	0.75
CP	18.88	19.92
NDF	17.42	13.80
ADF	10.28	11.04
NEF	44.93	43.29
CF	5.41	5.50
Ca	0.95	1.36
P	0.54	0.56

Milk yield showed significant differences among the four treatment groups ($P < 0.05$) in Table 2. Significant differences were found in milk composition between the non-urea (organic concentrate) with dried leucaena and urea added (non-organic concentrate) without dried leucaena based diets. The greatest improvement of milk yield of Treatment was by 1.5 and 3 kg/h/d dried leucaena with organic concentrate. Cows in treatment 1 and 3 group produced the highest milk compositions containing milk fat at 4.66% and 4.52%, respectively ($P < 0.05$).

Table 2. Milk yield per day and milk composition.

Items	Treatment				SEM
	1	2	3	4	
Milk yield (kg/hd/d)					
Milk yield	9.92 ^b	11.46 ^a	11.16 ^a	10.07 ^b	1.8
3.5% FCM, kg/d	11.79 ^b	12.00 ^a	13.01 ^a	10.06 ^b	1.4
Milk composition (%)					
Fat	4.66 ^a	3.79 ^b	4.52 ^a	3.49 ^b	0.06
Protein	3.31	3.42	3.51	3.37	0.05
Solid Not Fat	9.08	9.45	9.60	9.34	0.07

^{a b c} Means in the same row with different superscripts differ significantly ($P < 0.01$).

Treatment 3 was significantly higher in Omega3 (0.0065mg/ g of milk fat) as indeed reported, that increased Omega3 concentration was found in the milk of cattle fed with fiber rich diets (Dhiman, et al., 1999). The cows that received high dried leucaena of their lactation produced highest milk fat Omega 6, 9 concentration (0.081 and 1.272 mg/g) than Treatment 4 cows (0.076 and 1.173 mg/g, respectively) that received rice straw and non-organic concentrate (Table 3).

There were no differences in vitamin A (29-30 mg/100ml). Treatment 3 was significantly higher in vitamin A (30.00 pg/100ml) and vitamin E (0.05 mg/100ml) and phosphorus (848.60 mg/l) whereas T4 milk composition was significantly higher in vitamin E at 0.06 mg/100ml of milk fat (Table 4). Nutritive value of milk were lower when fed rice straw based. On the other hand, Suwanpanya et al. (2012) reported that milk composition is higher in CLA (3.67 mg/g) Omega3 (0.67 mg/g) Omega6 (0.79 mg/g) Omega9 (9.47 mg/g) Vitamin A (85.50 pg/100 ml) on grazing based.

In conclusion, dried leucaena supplementation brought out significant milk yield and produced higher fatty acid, vitamin, and mineral in milk. The data indicate that the elevation in the concentration of omega in milk was due primarily to dried leucaena the feeding regime and the treatment on organic concentrate produced milk fat higher in omega 3 and 6 than herds.

The results could be an alternative for farmers to reduce sustainable production of raising dairy cows in dry season.

Table 3. Composition of fatty acid on milk composition

Items	Treatment				MSE
	1	2	3	4	
(mg/ g of milk fat)					
Omega 3	0.0048 ^c	0.0040 ^c	0.0065 ^a	0.0042 ^b	0.002
Omega 6	0.073	0.073	0.081	0.076	0.010
Omega 9	1.254 ^b	1.254 ^b	1.272 ^a	1.173 ^c	0.150

^{a b c} Means in the same row with different superscripts differ significantly (P<0.05).

Table 4. Composition of vitamin and mineral on milk composition.

Item	Treatment				MSE
	1	2	3	4	
Vitamin A (mg/100 ml)	29.00	29.00	30.00	29.07	1.2
Vitamin E (mg/100 ml)	0.06 ^a	0.06 ^a	0.05 ^{a b}	0.04 ^b	0.1
Calcium (mg/l)	1,185 ^a	1,198 ^a	1,095.5 ^b	1,147.75 ^{ab}	7.5
Phosphorus (mg/l)	833.7 ^b	833.7 ^b	848.6 ^a	780.6 ^c	6.0

^{a b c} Means in the same row with different superscripts differ significantly (P<0.05).

Acknowledgements

The authors wish to express their gratitude to the Rajamangala University of Technology Isan (RMUTI) for financing this work and RMUTI, Kalasin Campus, Kalasin, Thailand

References

- AOAC. 1997. Official Methods of Analysis, 16th ed. Association of Official Analytical Chemists, Washington, DC
- Dhiman, T. R., Anand, G. G., Satter, L. D., Pariza, M. W. 1999. Conjugated linoleic acid content of milk from cows fed different diets, *Journal of Dairy Science*, 82, 21462156
- Goering, H. R., Van Soest. 1970. Forage fiber analysis and P. J. ARS Handbook No.379 United State Department of Agriculture, Washington, D. C.
- Suwanpanya, N., Kongsut, C., Kongchasing, Y and Mahasean, P. 2012. Cost reducing and CLA and Omega3 increasing in milk by feeding organic dairy cows with high Quality grass. Faculty of Agro-Industrial Technology, Rajamangala University of Technology Isan, Kalasin campus.

Potential of seaweed as feed to make healthy broiler meat chicken

Rahmatika Choiria¹, Riza Asti Octavira²

^{1,2} Faculty of Animal Husbandry, Padjadjaran University, Jalan Raya Jatinangor KM 22 Jatinangor Sumedang 45363 Indonesia

Abstract

Based on the survey from WHO, countries who have population like Indonesia every year there are one million people who develop heart disease, 40 % died, 60 % of them helped and saved 10% of which will also be died. One possible cause is a buildup of cholesterol in the wall coronary arteries. High cholesterol in the body is because consumption broiler chicken meat contains a lot of saturated fat. To create healthy broiler chicken meat, feed engineering is required. It is required for eliminate adverse effects on consumers. Feed engineering can done in the form of addition of seaweed in broiler feed. Approximately 4.92 million tons of seaweed Indonesian untapped much is wasted into the waste often causes pollution environment. Seaweed is rich in dietary fiber that dissolves in water and can bind fat in the digestive tract to reduce fat in the body. This study aims to determine the effect of seaweed in lowering body fat broiler chickens. Research carried out experimentally by maintenance broiler chicken divided by two group, one group with usual feed and one group with adding seaweed in the feed. Ration treatment consists of four kinds of treatment. Maintenance performed with broiler provision of commercial starter ration for 14 days and the separation to four treatment group with 2,5% seaweed, 5% seaweed, 7,5% of seaweed, and pure ransum without seaweed maintenance for 2 weeks. Measurement cholesterol levels, use Liberman Burchad method. The results of this study would be useful for seaweed product diversification for animal feed in order to function optimally. As well as reducing the risk of patient coronary heart disease through healthy chicken meat consumption in order to improve public health.

Keywords: broiler, cholesterol, seaweed.

Introduction

Based on survey do by WHO, country with population like Indonesia, annually one million people get heart attack 40% death, 60 % helped but only 90% will be alive from 60% helped and 10% again unhelped. In 2002, documented by WHO that 16,7 million life all over the world die because of cardiovascular or equal with 30% of total death in the world. One causes of heart attack is cholesterol which accumulates in the wall inside coronary arteries.

Cholesterol inside blood is because of broiler chicken industry. It is ironic, when broiler chicken becomes the source of animal proteins which consume by people but also become the source of deadly disease. Sum of broiler chicken increase from 2010 as many as 991.281.000 into 1.041.968.000 chickens in 2011. This case shows that Indonesian people in fulfilling protein need one of them is from broiler chicken. Broiler chicken meat has content many saturated fats. Percentage of abdomen fat reached 4.8% from body weight (Leeson and Summers, 1980). In order to create a healthy broiler chicken needs feed engineering thus it will not give bad impact to people consume it.

Feed engineering can be done by adding seaweed into broiler chicken. Seaweed cultivation in Indonesia in 2012 reached 5.100.000 stems that defeat Philiphine as the producer of the biggest

seaweed in the world. However seaweed industry in Indonesia only using as many as 180.000 ton, thus around 4.920.000 ton, seaweed that have not use and probably many wasted become waste that often caused environmental pollution. Seaweed is rich of soluble fiber food and can binding fat inside alimentary canal thus the fat opportunity in the food entered the body become low. In addition it content essential fat opportunity inside body become weak. Therefore feed engineering through uses of seaweed hopefully will create health and low fat of broiler chicken meat.

Materials and Methods

Chicken rearing in this research did in four weeks with two weeks give ransum of seaweed. Broiler that is rearing totally twenty with five chickens gave each treatment. Treatment is 2.5% of seaweed, 5% of seaweed, 7.5% of seaweed, and without seaweed. The procedure of determining cholesterol level did by using Liberman Burchad. One bottle of reagen 1 (buffer/enzzym/chromogent soluble by aquades 125 ml). Then the liquid mix with reagent as many as 1 ml added with H₂O 10 µL as standart, 1 ml added with 10 µL standart as cholesterol standart, 1 mL added with 10 µL blood sample for treatment test. After that read by using spectrophometer with length of phase 520 nm. The reading result is by comparing to standard.

Result and Discussion

Blood Cholesterol and Broiler Meat Chicken

Broiler meat chicken is soft and palatable but it contains high fat. At abdominal fat cock reaches 1.4 to 2.60% and females 3.2 to 4.8% (Leeson and Summers, 1980). Broiler chickens contain high cholesterol in meat is about 200 mg. (Setiawan, 2009). Basmacioglu and Ergul (2005), the normal value of broiler blood cholesterol or total cholesterol broiler are: 52-148 mg / dl, triglycerides <150 mg / dl, HDL > 22 mg / dl, LDL <130 mg / dl.

Cholesterol and The Effect to Health

According Hembing (2006), cholesterol is a fatty substance such as wax and yellowish. Most cholesterol is derived from the liver, cholesterol in the body plays an important role needed in several metabolic processes, such as the material to form the cell wall, forming hormones, neural tissue wrapping, precursor of bile acids and salts that serve as a fat emulsifier. Thus normal cholesterol levels have many benefits, but it would be a problem if the levels are excessive.

Fat intake is very important to note. Fat is a very important food ingredient, if we do not eat enough fat then your energy will decrease, but when we eat excessive fat can result in damage to blood vessels affecting mortality. However high fat can be prevented by consuming a balanced dietary fiber and a source of potential is seaweed. The main composition of seaweed which can be used as food are carbohydrates, ash, dietary fiber, and a small percentage of fat and protein. Hunninghake et al. (1994) reported the addition of dietary fiber in patients hipokolesterolemia cause a decline in total cholesterol, LDL- cholesterol and the ratio of LDL / HDL, but no significant effect on HDL-cholesterol and triglycerides. Cholesterol-lowering mechanism by fiber, which is binding bile acids in the small intestine that causes increased fecal bile acid excretion, decrease the absorption of fat and cholesterol, decrease in the rate of carbohydrate absorpsi cause decreased levels of serum insulin thereby reducing the synthesis of cholesterol and lipoprotein stimulation and inhibition of cholesterol synthesis by short chain fatty acids resulting from the fermentation of soluble fiber in the colon (Wolever et al. 1997 in Kamalia 2012). Enhancement bile acid excretion in excreta can cause a decrease in plasma cholesterol levels by about 10% - 25%.

Table 1. Influence of Seaweed to Blood Cholesterol

Treatment	Average	Significant	Decrease of Blood Cholesterol (mg/dL)
T3	11,79	a	1,82
T2	10,77	b	1,26
T1	10,53	c	1,02
T0	9,97	d	

Description: Average followed by different letters indicate a significant influence outcome ($P < 0.01$). T3 = 75g / chicken/ day of 1 kg of feed, T2 = 50 grams / chicken / day of 1 kg of feed, T1 = 25 g / chicken / day of 1 kg of feed

Duncan Test Analysis results showed that the effect of the use of seaweed on cholesterol broiler very significant effect, namely (P3) very significant effect on (P0), (P1) and (P2), (P2) very significant effect on (P0), (P1) and (P3) and (P1) very significant effect on (P0), (P2) and (P3). So that the coefficient of determination addition of seaweed affects 90% of the reduction in blood cholesterol broiler. Rahmatika (2014) found that the use of seaweed can lowering cholesterol broiler meat at 7,88mg/100. The reduction in blood cholesterol in broiler maintenance for two weeks for the provision of as much as 75 grams of seaweed can lower cholesterol as much as 1.82 mg/dL, while the provision of as much as 50 grams of seaweed can lower cholesterol as much as 1.26 mg/dL and the provision of seaweed as much as 75 grams of cholesterol as much as 1.02 mg/dL. So that potential of seaweed can lowering cholesterol is a potential to create healthy broiler meat chicken is high. The best treatment in this study was the addition of 7.5% seaweed can lower cholesterol as much as 1.82 mg/dL.

Seaweed production in Indonesia continues to increase, of total production amounted to 2.574 million tons in 2009 to 3,082 million tonnes in 2010, in 2014 the government targets seaweed production by 10 million tons. However, the absorbed in the national industry only 180,000 tonnes so still many untapped seaweed. This indicates that the potential of seaweed in Indonesia to create a healthy broiler meat chicken potentially is high.

Conclusion

Based on these study variations in the use of seaweed with a concentration of 2.5%, 5% and 7.5% in broiler chicken feed found to be the result of blood cholesterol significantly different between the treatments so that the use of seaweed can lower blood cholesterol broiler. The best use of seaweed found in treatment T3 is 75 grams / chicken / day can make lower blood cholesterol by 1.82 mg/dL.

References

- Basmacioglu, H. and M. Ergul. 2005. Research on the factor affecting cholesterol content and some other characteristics of eggs in laying hens. *Turk. J. Vet. Anim. Sci.* 29: 157-164
- Hembing. 2006. Mengendalikan Kolesterol Tinggi Dengan Herba Dan Pola Hidup Sehat. Available: <http://portal.cbn.net.id> (25 Maret 2015).
- Hunninghake DB et al. 1994. Hypocholesterolemic Effect of Dietary Fiber Supplement. *Am J Clin Nutr*: 59:1050-1054
- Kamalia. 2012. Pengaruh Penambahan Berbagai Level Daun Katuk Terhadap Kolesterol HDL,LDL, dan Trigliserida Darah Broiler. Makassar
- Leeson, S. and J. D. Summers. 1980. Production and carcass characteristic of the broiler chicken. *Poultry Science*. 59: 786-798.
- Rahmatika. 2014. Potential of Seaweed As Feed To Make An Healthy Broiler Chicken. AAAP. Jogjakarta
- Setiawan, I. 2009. Budidaya Ayam Pedaging (Broiler). [Online]. Available: <http://centralunggas.blogspot.com/2009/07/budi-daya-ayam-pedaging-broiler.html>. [25 Maret 2015].

Metabolic imprinting improves rumen development via modulation of epigenetic gene expression including histone modification

Park¹, S. K., Y. G. Oh², H. J. Lee² & M. J. Kim²

¹Sejong University, 209, Neungdong-ro, Gwangjin-gu, Seoul, 143-747, Korea, ²National Institute of Animal Science, Suwon, 441-706, Korea

Abstract

This study was conducted to investigate the effect of 2 mo. administration of high energy diet on epigenetic gene expression in Hanwoo calves. Twenty Hanwoo calves (7~10 d old) were randomly assigned into 4 different treatments (5 calves per treatment, all male) including control (cow-feeding), milk replacer + concentrate (T1), milk replacer + concentrate + forage (T2) and milk replacer + concentrate + 30% starch (T3). Concentrate diets were fed as pellets in addition to a commercial milk replacer and starch (T3) was added into pellets. Body weight and feed intake were measured and blood was sampled every 2 weeks. After an 10 weeks feeding period, the calves were euthanized and loin muscle tissue from the 10th and 11th rib junction was sampled for comparative analysis of gene expression using RNA-Seq. Differentially expressed genes (DEGs) for control versus T1, T2 and T3 were 4, 39 and 13, respectively. There were 4 DEGs between T2 and T3. DAVID functional analysis revealed muscle growth and development, glycogen metabolic process and regulation of metal ion transport as the enriched biological processes in T3 compared to control group. Interestingly, expression of ACAT1 was greatest in T2 and BPIFA1 was highest in T3 among treatment groups, suggesting that supplementation of concentrate diet or high energy diets (30% starch) induce epigenetic alterations which may substantially enhance the gene expressions related to energy metabolism and development of epithelial tissue development of rumen. In conclusion, high energy diet during early stage of development may have a positive effect on rumen development in Hanwoo calves.

Keywords: epigenetic, imprinting, hanwoo

Corresponding author: sungkwonpark@hotmail.com

Introduction

In neonate, first few weeks of life, extensive physical and metabolic adaptations occur in the rumen, including changes in its size, number and size of rumen papillae, and development of muscular tissue (Van Soest, 1994). Moreover, the early transitioning of the rumen from pre-ruminant to a ruminant state is required to ensure animal health, performance and economic benefit for producers. Previous studies reported that the effect of plane of nutrition on animal performance and rumen development, including early consumption of starter (Flatt et al., 1958; Sander et al.,

1959). The presence of volatile fatty acid (VFA) in the rumen, which induced ruminal fermentation and their end product (especially propionate and butyrate), promotes the rumen epithelium development (Stobo et al., 1966; Suarez et al., 2006a; Khan et al., 2007a).

Early morphological studies demonstrated that feeding high concentrate diets, opposed to roughage diets, induces increase in rumen volume (Warner et al., 1956; Stobo et al., 1966), surface area of rumen papillae (Zitnan et al., 2006) and density and length of papillae (Stobo et al., 1966). However, overloading energy supplementation may promote keratinization of ruminal papillae by increased papillae width (Mentschel et al., 2001). The lack of scientific studies is unable to definitively distinguish the molecular mechanisms regulating these morphological changes. The objective of this study was to assess diets differing in their potential to promote rumen papillae and to identify genes differentially expressed induced by diets in Hanwoo calves.

Materials and methods

All experimental procedures were reviewed and approved by the ethics committee on the use of animals in research, National Livestock Research Institute, South Korea. Twenty male Hanwoo calves of 7 – 10 d of age (38 ± 2 kg) were stratified by weight and age, and randomly assigned to 4 dietary treatments (5 calves per treatment): control (CON), milk replacer + concentrate (T1), milk replacer + concentrate + timothy (T2) and milk replacer + concentrate + 30% starch (T3). At 10 wk of the experiment period, calves were slaughtered, fore-stomachs and the abomasum were removed immediately and weighed, emptied and rinsed. The rumen was dissected along the dorsal line and tissue samples were collected for further morphometric analysis according to Lesmeister et al. (2004). Rumen tissue samples were saved for RNA extraction and hybridization to a custom bovine whole-genome array. The tissue samples for morphometric analysis were fixed in 10% buffered formaldehyde embedded in paraffin, stained with hematoxylin-eosin, and viewed through a microscope equipped digital camera (TDI DigiCam, Olympus, Tokyo, Japan). Total RNA was extracted from ruminal tissue samples quantitative real time PCR amplification was performed. Cuffdiff provides various output files, and using one of its outputs, “gene_exp.diff”, DEG (Differently Expressed Gene) was identified. In this paper, the main goal is to detect DEGs between Experimental groups and control groups. Final BW, average daily gain (ADG), DMI, FE and rumen tissue parameter was analyzed as complete randomized block design using the GLM procedures of SAS 9.2 (SAS Institute, Inc, Gary, NC). Quantitative PCR data were analyzed using the GLM procedures of SAS 9.2 (SAS Institute, Inc, Gary, NC).

Results

The effect of dietary treatments on starter intake, ADG, feed efficiency (FE) and final body weights are presented in Table 1. Daily starter intake was greatest in T2 group than T1 and T3 groups ($P < 0.05$). Whereas, there was no difference in the average daily gain (ADG), feed efficiency (FE) and initial and final body weight between treatments. The significant differences in CON group were not tested due to only other dietary treatments were available.

Table 1. Effects of dietary treatments on final body weight (BW), average daily gain (ADG), feed efficiency (FE) and daily dry matter intake (DMI) in Hanwoo (*Bos taurus coreanae*) calves.

Parameter	Dietary treatment				P-value
	CON	T1	T2	T3	
Final BW, kg	59.3	59.2	55.0	53.9	0.403
ADG, kg/d	0.72	0.61	0.62	0.49	0.705
Feed efficiency, kg/kg	-	0.66	0.59	0.55	0.890
Daily DMI, kg/d	-	0.91 ^b	1.07 ^a	0.87 ^c	<.0001
Starter, kg/d	-	0.91 ^a	0.88 ^{ab}	0.87 ^b	0.018
Timothy, kg/d	-	-	0.18	-	-

^{a-c} Means with different superscript letters are significantly different ($P < 0.05$)

¹Feed efficiency = kg of BW gain / kg of total DMI.

Treatments: CON = maternal milk only; T1 = milk replacer with concentrate; T2 = milk replacer with concentrate with timothy; T3 = milk replacer with 30% starch.

Rumen weight and morphometric analyses: Table 2 presents the morphometric analyses of the rumen tissues. Rumen papillae length was greatest in T1 group compared with CON and T3 groups ($P < 0.05$). As expected, calves in T3 group has higher papillae width than CON ($P < 0.05$). The thickness of mucosa and rumen wall was higher in T1 and T3 groups compared with CON group ($P < 0.01$, respectively). However, submucosa thickness was higher in T2 group followed by T3 group than in CON and T1 groups ($P < 0.05$).

Table 2. Effects of dietary treatments on morphometric variables of rumen papillae length, papillae width and thickness of mucosa, submucosa, serosa, muscles layer and rumen wall.

Parameter (μm)	Dietary treatment				P-value
	CON	T1	T2	T3	
Papillae length	878.8 ^b	1793.1 ^a	1334.0 ^{ab}	1144.4 ^b	0.013
Papillae width	285.0 ^b	342.2 ^{ab}	329.5 ^{ab}	367.9 ^a	0.023
Mucosa thickness	1495.1 ^b	3103.1 ^a	2185.5 ^{ab}	2948.8 ^a	0.002
Submucosa thickness	460.3 ^b	375.4 ^b	612.2 ^a	491.6 ^{ab}	0.021
serosa thickness	222.0	343.3	246.1	290.6	0.272
Muscles layer thickness	1867.4	2000.5	1887.2	2042.9	0.518
Rumen wall thickness	4015.4 ^b	5892.2 ^a	4874.8 ^{ab}	5828.7 ^a	0.004

^{a-c} Means with different superscript letters are significantly different ($P < 0.05$)

Treatments: CON = maternal milk only; T1 = milk replacer with concentrate; T2 = milk replacer with concentrate with timothy; T3 = milk replacer with 30% starch.

Differential gene expression: The expression of BPIFA1 gene was increased by up to 20 folds in T1 and T3 groups as compared to CON and T2 groups, while the ACAT1 gene was 3 fold greater in T2 group than T1 and T3 groups. There were no other differentially expressed genes detected in rumen epithelium tissue among treatment groups.

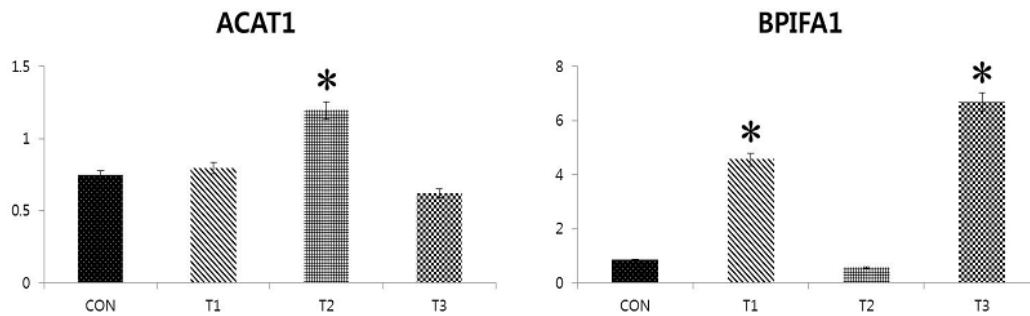


Figure 1. The gene expression in ruminant epithelium by qPCR among different dietary treatments. Treatments: CON = maternal milk only; T1 = milk replacer with concentrate; T2 = milk replacer with concentrate with timothy; T3 = milk replacer with 30% starch.

*Means with different superscript letters are significantly different ($P < 0.05$).

References

- Khan, M. A., H. J. Lee, W. S. Lee, H. S. Kim, S. B. Kim, K. S. Ki, S. J. Park, J. K. Ha and Y. J. Choi, 2007a. Starch source evaluation in calf starter: I. Feed consumption, body weight gain, structural growth, and blood metabolites in Holstein calves. *J Dairy Sci.* 90: 5259-5268.
- Mentschel, J., R. Leiser, C. Mulling, C. Pfarrer, and R. Claus. 2001. Butyric acid stimulates rumen mucosa development in the calf mainly by a reduction of apoptosis. *Arch. fur Tierernahrung.* 55: 85-102.
- Sander, E. G., H. N. Warner., H. N. Harrison., and J. K. Loosil, 1959. The stimulatory effect of sodium butyrate and sodium propionate on the development of rumen mucosa in the young calf. *J. Dairy Sci.* 42: 1600-1605.
- Stobo, I. J., J. H. Roy, and H. J. Gaston, 1966. Rumen development in the calf. 1. The effect of diets containing different proportions of concentrates to hay on rumen development. *BritJ. nutri.* 20: 171-188.
- Suarez, B. J., C. G. Van Reenen, G. Beldman, J. Van Delen, J. Dijkstra and W. J. J. Gerrits, 2006a. Effects of supplementing concentrates differing in carbohydrate composition in veal calf diets: I. Animal performance and rumen fermentation characteristics. *J. Dairy Sci.* 89: 4365-4375.
- Van Soest, P. J., 1994. *Nutritional ecology of the ruminant.* Cornell University Press.
- Warner, R. G., W. P. Flatt., and J. K. Loosli, 1956. Dietary factors influencing the development of the ruminant stomach. *J. Agr. Food Chem.* 4: 788-801.
- Zitnan, R., S. Kuhila, P. Sanftleben, A. Bilska, F. Schneider, M. Zupcanova and J. Voigt, 2006. Diet induced ruminal papillae development in neonatal calves not correlating with rumen butyrate. *Vet. Med. Czech.* 50: 472-479.

Effect of rumen content on the performance and external body measurement of sudanese desert kids

Magboul¹, Y.Y. & M.B. Elemam^{2*}

¹Al-Neelain University, Faculty of Agricultural Technology and Fish Sciences, Sudan. ²University of Kassala, Faculty of Agriculture and Natural Resources Department of Animal Production, P.O. Box 12, New Halfa, Sudan

Abstract

Twelve Sudanese desert kids (less than one year old and of initial body weight 11.4 kg) were divided into three groups of equal number to study the effect of rumen content level on the performance and external body measurements. The study was conducted at small ruminant research unit in the Faculty of Agricultural Technology and Fish Sciences, Al-Neelain University Khartoum, Sudan. Three iso-nitrogenous diets containing graded levels of rumen contents (0%, 5%, and 15%) were randomly assigned to the kids groups. Feeding was on *ad libitum* base for 35 days. External body measurements of the kids were recorded at the start and the end of the experiment. Results revealed no significant differences between groups for feed intake and feed conversion ratio but the final weight gain is significantly decreased. External body measurements were significantly influenced by inclusion of rumen content.

Key words: sudanese desert kids, rumen content, performance, body measurement

**Corresponding author: murtadaelemam@yahoo.com.*

Introduction

Goats contribute substantially to Sudanese households' well being by producing milk, meat, hair and skin (Sulieman, 1986; Mofarrah, 1995). There are four local goat breeds in the Sudan: Nubian, Desert, Tagar, and Nilotic (Sulieman, 1986). The primary aim behind the use of these by-products is to reduce the feed cost and therefore the cost of producing a unit of the product as well as to fulfill the protein demands of animals (Haapapuro et al., 1997). The dried rumen content obtained in this way not only serves as a feed nutrient, but also its recycling reduces disposal and environmental pollution problem (Swan, 1992).

Previous studies have generally indicated that dry rumen contents contained substantial amounts of crude protein and utilizable energy for ruminants (Messersmith et al., 1974; Ghosh & Dey, 1993; Salinas-chavira et al., 2007). The objective of this experiment is to study the effect of supplementation of rumen content on performance and body measurement of kids.

Materials and Methods

Rumen contents were collected from Jabel Awlia abattoir, Khartoum and dried under the sun. Proximate analysis was done as outlined by (AOAC, 1990). Based on this analysis three iso-nitrogenous and iso-caloric diets contains rumen content were formulated. Twelve Sudanese desert kids with an average live weight of 11.4 kg and age less than one year old were used in this study. A mixture containing equal proportions of experimental diets was fed to the kids. At the end of the adaptation period kids were individually weighed and divided into three groups of equal number and weight. Each group was separately kept in a pen provided with watering and feeding facilities. The diets were randomly assigned to the kids groups and offered *ad libitum* in one morning meal throughout the feeding period.

Green fodder (*Medicago Sativa*) was also offered at a rate of 1kg/ head/week as a source of vitamin A. Clean water and salt lick were available throughout the feeding period which lasted for 45 days. External body measurements were taken from the kids at the beginning and at the end of the experiment according to the procedure described by (Brown et al., 1973). Data was statistically analyzed according to the analysis of variance applicable to complete randomized design as described by (Snedecor & Cochran, 1980).

Results and Discussion

Chemical compositions of rumen content in the study were shown in (Table 1), these values were lower than those reported by Rafaelli et al. (2006). Performance characteristics of Sudanese desert kids were presented in (Table 2). Similar results were reported by Tucker et al. (1956) who found that feeding dried rumen contents did not show any evidence of pathological effects in lambs. Also, Jovanovic & Cuperlovic (1977), Gaber (1992) and Khattab et al. (1996) clearly indicated that the inclusion of dried rumen content in the rations for ruminants produced no palatability problems.

Graded percentages of rumen contents meal had no significant ($P < 0.05$) effect on the final body weight, weight gain and feed conversion ratio, but there were significant differences ($P < 0.05$) among dry matter intake, these values were differ from the findings of Abd El-Galil & Khider (2001), Abouheif et al. (1999) and Mohammed (2005), this may due to the odder of the diets. The body measurements of the kids were represented in (Table 3), however all the measurements tend to increase in the three experimental diets (control, 5 and 15 % rumen content) and these were in line with the findings of (Atta et al., 2011).

Table 1. Chemical composition (%) of rumen contents.

Item	Percentage
Dry matter	90
Crude Protein	11.5
Ether Extract	3.7
Crude fibre	15
Ash	7

Table 2. Performance characteristics of Sudanese desert kids fed experimental diets.

Item	A	B	C	S.E
Number of animals/lot	4	4	4	-
Feedlot period (days)	35	35	35	-
Initial live weight (Kg)	11.55	11.55	11.60	0.24
Final live weight (Kg/head)	15.95	16.1	15.77	0.56
Total weight gain (kg / head)	4.4	4.55	4.17	0.11
Total dry matter intake (Kg/head/day)	1.37 ^b	1.73 ^a	1.05 ^c	0.13
Feed conversion ratio (Kg DMI/Kg gain)	10.90	13.31	8.81	0.17

S.E: Standard error.

Table 3. Body measurements for experimental kids before and after the experiment.

Item	A		B		C	
	Before	After	Before	After	Before	After
Back Length	35.5	36	34.5	37	35.75	36.25
Heart Girth	52.37	53.2	55.75	56.03	55.25	56.22
Body Height	58.25	58.78	59	59.87	57	57.03
Thigh circumference	20.75	19.75	21.50	21.95	25.5	26.06

Conclusion

It could be concluded that rumen content is a safe feed without any determinant effects on kid's health. Even more it was a cheap feed and produced a reasonable growth rate.

References

- Abd El-Galil, K and R.F. Khider, 2001. Effect of dried rumen content in diets on the performance of growing Japanese quails. *J. of Agric. Sci. Mansoura Univ.*, 26 (1): 179-186.
- Abouheif, M.A., M.S. Kraidees and B.A. Al-selbood, 1999. The utilization of rumen content-barley meal in diets of growing lambs. Department of Animal Production, College of Agriculture, P.O. Box 2460 King Saud University, Riyadh 11451, Saudi Arabia.
- AOAC 1990. Official Methods of Analysis. Association of Official Analytical Chemistry (15th Ed). Washington D.C. U.S.A.
- Atta M., A. A. G. Adam and A. B. I. A. Abuzaid, 2011. Effect of two different feeding systems on body growth and measurements in Sudan Nilotic male kids. *Animal Science Journal*, 2(1): 01-06. arch Article
- Brown, J.E, Brown, C.J and Butts, T.M., 1973 Evaluating relationship among immature measurements of size, shape and performance of beef bulls .1.principal components as measures of size and shape in young Hereford and Angus bulls. *J. Anim. Sci.* 36:1010.
- Gaber, A.A., 1992. Nutritional evaluation of dried rumen and stomach contents versus berseem hay in sheep diets. *J. of Agric. Sci., Mansoura Univ.*, 17 (5): 949 – 958.
- Gosh, T.K and A. Dey, 1993. Availability of rumen contents and their utilization in diets for goats. *India J. Anim. Sci.* 63: 1194 – 1196

- Haapapuro, E.R; N.D. Barnard, and M. Simon (1997). Review. Animal waste used as livestock feed: Danger to human health. *Prev.med.* 26:599-602.
- Jovanovic, M and M. Cuperlovic, 1977. Nutritive value of rumen contents for monogastric animals. *Anim. Feed Sci. Technol.* 3: 351 – 360.
- Khattab, H.M; S.M. Abdelmawla and A.M. Singer, 1996. Nutritional evaluation of rumen contents as a slaughterhouse waste in sheep rations. *Egyptian J. Anim. Prod.* (sup) 33.
- Messersmith, T.L., W.R. Fulton and T.J. Klopfenstein, 1974. Dried pounce feed as a roughage source in beef finishing rations. *J.Anim. Sci.*39:246 (Abstr.).
- Mofarrah, M.B., 1995. Goat breeds and varieties in Sudan. In: Proceedings of Arab Center for Studies of Arid Zones and Dry lands (ACSAD). A training Course on Sheep and Goat Production, on January 1995, Khartoum, Sudan 17 – 27.
- Mohammed, H.E.H., 2005. Effect of feeding roselle (Karkadeh) seed (*Hibiscus Sabdariffal.*) on performance and carcass characteristics of Sudan desert sheep. M.Sc. Thesis, Faculty of Anim. Prod., University of Gezira.
- Rafaelli, P.M., L. Sangines –Garcin; R.F. Perez-Gil and O. Iarrosa, 2006. Documentos de trabajo. Facultad de Ciencins Agraias, PP: 1-7.
- Salinas –Chavira, J.M. Dominguez – Munoz, R. Bernal- Lorenzo, R.F.Garcia- Castillo and C. Arzola- Alvarez, 2007. Growth performance and carcass characteristics of feedlot lambs fed diet with pig manure and rumen contents. *J.Anim.vet. Adv*, 6: 505 – 508.
- Snedecor, G.W and Cochran, W.G., 1980. *Statistical Methods*, Iowa State University Press. Ames, Iowa, U.S.A.
- Suliman AH, 1986. The potential of small ruminant in eastern and southern Africa, ILCA, Nairobi, Kenya 333 – 342.
- Swan, J.E., 1992. Animal by-product processing. In: *Encyclopedia food science technology* (ed.Y.H.Hui), vol. 4, pp: 42-49.
- Tucker, J.O; M.W Glenn and G.W. Robertstad, 1956. Effects of dried preserved rumen inoculum on the rumen microorganisms of lambs. *Amer. J. vet. Res.* 17: 498 – 502.

Potassium iodate supplementation in layer drinking water for iodine enriched egg and laying performance

Chumkam, S.¹ O.Jintasataporn² and O.Jintasataporn³

¹Faculty of Agricultural Technology, Valaya Alongkorn Rajabhat, University Under the Royal Patronage, Pathum Thani, Thailand ²Department of Aquaculture, Faculty of Fisheries, Kasetsart University, Bangkok, Thailand. ³Department of Animal Science, Faculty of Agriculture at Kamphaeng Sean, Kasetsart University, Nakhonpathom, Thailand.

Abstract

This study was aimed to investigate the effects of supplementation potassium iodate in water on layer performance, egg quality, iodine content in egg. Two hundred and sixteen commercial laying hens were divided into 2 groups and subjected to the following treatments: (i) control; (ii) water supplemented with potassium iodate containing 4 ppm of iodine. Each treatment was consisted of 9 replicates with 12 laying hens per replicate. The results have shown that there were no significant difference in layer performance, haugh unit, egg shell thickness, color of egg yolk, and egg specific gravity throughout the period of experiment ($p > 0.05$). Iodine content were 80.86 $\mu\text{g}/\text{egg}$ in potassium iodate supplemented in water treatment.

Keywords: iodine, potassium iodate, layer

Introduction

Iodine is a natural element that is important for health. It is necessary for Thyroxin hormone in the Thyroid gland. These hormone acts on growth performance, regulating metabolism and development of the brain. Without iodine during the fetus and infant development, it is effect on brain development disorders. Cretinism disease is one of the resulting of brain development disorders which decrease IQ score in children. The research institute) HSRI (of Thailand report children's IQ score in 21 provinces that they survey. The score is around 91 from standard score is 90-110. This degree is in the baseline but lower than many countries. Supplementation iodine in water for hen to produce iodine's egg is another alteration to controlling and elimination iodine deficiency problem in Thailand. Robinson and Wasnidge)1981 (discovered that the iodine accumulates during production of egg by combines with calcium to accumulate in yolk granules. Charoensiriwatanta *et. al.*(2010) reported that supplement iodine in feed for one month can increase iodine content in egg from 75.96 $\mu\text{g}/100 \text{ g}$ egg for the control to 184.5 $\mu\text{g}/100 \text{ g}$ egg for the iodine enriched egg. Therefore, this experiment aim to investigate the effect of supplementation potassium iodate in water to increase iodine accumulation in egg production.

Materials and Methods

Two hundred and sixteen commercial laying hens of 32 week-old were stocked in cage. The research was assigned in completely randomized design (CRD) with two treatments: (i) control (ii) water supplemented with potassium iodate containing 4 ppm of iodine. There were 9 replicates each replicate contained twelve hens. Laying hens were fed 16 % crude protein and 2,700 kcal ME

/kg on laying period. The feed and water were allowed ad-libitum. Laying hens were maintained on a 16 hrs. light in evaporative cooling system house. The experimental period was 8 weeks in 4 subsequent interval periods each of 2 weeks. Laying hens were recorded feed intake, egg production and death. Egg quality was measured at the end of each interval period, in which 50% of egg were randomly from each treatment (6 eggs from each replicate). Egg quality including egg specific gravity, egg yolk color, albumin height, haugh unit and egg shell thickness were evaluated. One egg from each replicate was randomized to determine iodine according to Maxon & Dixon (1980). Data were analyzed as an one way ANOVA. Duncan's multiple range test was carried out to determine the differences among treatment.

Results and Discussion

The results showed that there were no significant difference ($p > 0.05$) in feed intake, feed /dozen egg, egg production rate, egg weight and survival rate (table 1). The results were similarly to Songserm *et. al.* (2006) that there were no effect of feed supplementation with either potassium iodate or potassium iodide which containing 4 ppm of iodine in layer diet on feed intake, feed /dozen egg, egg production rate, egg weight, egg mass and survival rate. However, for the other source of iodine, Jintasaporn *et. al.* (2009) founded that feed intake, feed /dozen egg, egg production rate, egg weight, weight gain and survival rate of laying hen were not difference ($p > 0.05$) on hens fed normal diet, diet supplemental potassium iodide containing 4 ppm of iodine and diet supplemental potassium iodide containing 4 ppm of iodine plus tuna oil. The performance were on the same range because level of iodine (0.30-0.40 ppm) was higher than the requirement (Cunha, 1987). Cost of feed/egg in control group was 2.08 baht that 0.48% higher than iodine group (2.07 baht).

Table 1. Laying hen performance throughout all period after drink potassium iodate for 8 weeks

Laying hen performance	Control	Iodine 4 ppm	p-value
Feed intake (g./h/d)	98.66±0.40	97.93±0.56	0.306
Feed/dozen egg (kg.)	1.36±0.03	1.35±0.01	0.777
Egg production (%)	88.16±1.63	86.72±1.13	0.480
Egg weight (g.)	57.11±0.51	57.09±0.53	0.982
Survival rate (%)	99.54±0.46	100±0.00	0.332
Cost of feed/dozen egg (baht)	24.93	24.81	-

The egg quality, haugh unit, yolk color score, egg shell thickness and egg specific gravity were not difference. The results were in agreement with Songserm *et. al.* (2006) who founded that iodine levels in 4 ppm either as potassium iodate or potassium iodide were not significant effects on egg quality because of iodine requirement in the diet was low (0.30-0.40 ppm) and supplementation 4 ppm. Iodine does not cause toxic to the hen. Lewis (2004) found that there were some effect of high concentration of iodine in layer diet on egg production by most of iodine accumulates in thyroid gland. These related to thyroid hormone which was important for bird reproduction. Thyroid is essential for the initiation and maintenance of egg production and for molting in turkey hens. Hyperthyroid state evoked by administration of thyroid hormone (T3) for few days diminishes luteinizing hormone (LH) estradiol (E2) and progesterone (P4) levels, reduces the weight of the ovary, induces atresia of preovulatory follicles and eventually causes stoppage of egg laying (Lien & Siopes, 1989; Sechman, 2013). Yaicin *et. al.* (2004) reported that egg production and egg weight decrease in iodine supplemented 12 or 24 mg iodine/kg diet group compared to the other treatment of 0, 3 and 6 mg /kg diet.

It was clearly showed that supplementation of iodine in drinking water increase egg iodine concentration. The iodine content were 9.63 and 80.82 µg./egg in control and iodine supplemented group, respectively. The results were confirmed by Abdel *et. al.* (2012) founded that increasing iodine level significantly ($p < 0.01$) increased egg iodine. Laying hen can transfer many trace

element from feed and water to egg compared to meat (Flachowsky, 2007). In addition, Kaufman *et. al.* (1998) reported that there was a significant linear correlation ($r = 0.93$) between iodine content in feed mixture up to 5 mg./kg and iodine accumulation in egg yolk.

Table 2. Egg quality and iodine content throughout all period after drink potassium iodate for 8 weeks

Laying hen performance	Control	Iodine4 ppm	p-value
Haugh unit	83.74 \pm 1.02	81.86 \pm 0.47	0.113
Yolk color score	8.60 \pm 0.10	8.63 \pm 0.90	0.812
Egg shell thickness (mm.)	0.331 \pm 0.004	0.338 \pm 0.003	0.209
Egg specific gravity	1.087 \pm 0.001	1.088 \pm 0.001	0.694
Iodine content (μ g./egg)	9.63	80.82	-

Conclusion

Water supplemented potassium iodate containing 4 ppm. of iodine enhance iodine enriched egg without any effect on laying performance and egg quality. Iodine concentration increased 8.4 time of normal egg.

Acknowledgment

The researcher would like to thank to The Valaya Alongkorn Rajabhat University under The Royal Patronage for financial support throughout the experiment.

Reference

- Abdel-Malak, N.Y., SaharM.H. Osman, A.S.A.Bahakaim, Amal S. Omar and NehadA. Ramadan. 2012. Effect of different level of iodine in layer's diet on egg iodine enrichment. *Egypt. Poult. Sci.* 32:851-864.
- Cunha, T.J. 1987. "Salt and trace mineral for livestock, poultry and other animal" Cited by S.P. Chandrakant and M.G. Venkatesh Manner. Iodine deficiency disorders and iodized salt for livestock. In S.P. Chandrakant and A.R. Rao (eds). *Iodine Deficiency Disorder in livestock.* India: Oxford University Press. p242.
- Charoensiriwatanta, W., P. Srijantr, P.Tecyapant and J. Wongvilairattana. 2010. Consuming iodine enriched egg to solve iodine deficiency endemic for remote areas in Thailand. *Nutrition J.* 9:68.
- Flachowsky, G. 2007. Iodine in animal nutrition and iodine transfer from feed in to food of animal origin. *Lohman Information.* 42:47-61.
- Kaufman, S., G. Wolfram, F. Delange and A. Rambeekw. 1998. Iodine supplementation of laying hen feed : A supplementary measure to eliminate iodine deficiency in humans. *Zeitschrift fur Ernahrungswissenschaft.* 37:288-293.
- Lien, R.J., and T.D. Siopes.1989. Effects of thyroidectomy on egg production, molt, and plasma thyroid hormone concentrations of turkey hens. *Poult. Sci.*68:1126-1132.
- Moxon, R.E.D. and E.J. Dixon. 1980. Semi-automatic method for the determination of total iodine in food. *Analyst.* 105: 344-352.
- Robinson, G.A. and D.C. Wasnidge. 1981. Comparison of the accumulation of 125-I and 144-Ce in growing oocytes of Japanese quail. *Poultry Sci.* 60:2195-2199.
- Sechman, A. 2013. The role of thyroid hormones in regulation of chicken ovarian steroidogenesis. *Gen Comp Endocrinol.* 190:68-75.
- Songserm, O., S. Chaunchom, S. Sa-nguanphan, O. Triwutanon and A. Plaiboon. 2006. Iodine supplementation in layer diet of commercial production of iodine enriched egg animal *Vet. Medicine.* 55:257-264.

Efficacy of dry powdered *Enterococcus italicus* on immune responses to *Mycoplasma hyopneumoniae* vaccination

Kasornpikul C.¹ & S. Chalorsuntisakul¹

¹Faculty of Animal Science and Agricultural Technology, Silpakorn University, Petchburi 76120, Thailand

Abstract

Enterococcus italicus is a kind of effective probiotics, which selected to be transformed into an easy dry powdered product. The objective of this study was to evaluate the safety and efficacy of the dry powdered probiotics in enhancing immune responses to *Mycoplasma hyopneumoniae* vaccination. In terms of study methods, firstly the dry powdered probiotics were prepared by mixing the probiotics with methylcellulose and steam broken-milled rice. The mixture was dehydrated at 45°C during 3 days and then mixed with swine commercial feed to get the final concentration of 10⁶ cfu/g. Subsequently, 12 – twenty-one day old - mixed sex pigs were allocated into 2 groups, with 3 replications for each group and 2 pigs per replication. A completely randomized design (CRD) was used for all experiments. The pigs were fed *ad libitum*. During a month, the pig in the first group received commercial feed, whereas the pigs in the second group were offered commercial feed supplemented with dry powdered *Enterococcus italicus*. Mortality data were collected, and all studied pigs were vaccinated against *Mycoplasma hyopneumoniae* twice at age 45 and 59 day-old. A blood test was done in order to indicate immunity level against the disease. It appeared that no studied pig was found dead and the immunity level of the pigs receiving feed supplemented with probiotics was higher ($P < 0.001$) than those receiving feed without probiotics at seven days after the second vaccination. In conclusion, the pigs receiving feed supplemented with dry powdered *Enterococcus italicus* has the highest immunity level at seven days after the second vaccination.

Keywords: *Enterococcus italicus*, immune responses, *Mycoplasma hyopneumoniae*

Introduction

Enterococcus italicus are effective probiotics (Kasornpikul & Chalorsuntisakul, 2011) (from which some products can be further developed. An easy dry powdered *Enterococcus italicus* can be useful and more accessible to farmers. However, before applying, the product should be tested in terms of safety and effectiveness on immunity enhancement in animals. The aim of the present study was to evaluate the safety and efficacy of the probiotics developed in dry powder form in enhancing immune responses to *Mycoplasma hyopneumoniae* vaccination.

Materials and Methods

To prepare dry powdered probiotics, *Enterococcus italicus* was bred in MRS broth and then spinned in a centrifuge to obtain the precipitate which was dissolved in the solution of 0.9% NaCl and then mixed with 1.25% methylcellulose. The mixture was then mixed with steam broken milled-rice in a portion of 1:2. It was then dehydrated in the decontaminated oven at 45°C during three days)modified from Suthamma & Taweechai, 2014(. The number of survival bacteria was then determined by using the pour plate method. The product obtained was mixed with commercial swine feed to get the final concentration of 10⁶cfu/g.

For experiments, 12 – twenty-one day old - mixed sex pigs were raised. Firstly, the animals were kept together for seven days prior to be allocated into 2 groups, with 3 replications for each group and 2 pigs per replication. A completely randomized design)CRD) was used for all experiments. During a month, the pigs were fed *ad libitum*; the pigs in the first group received a commercial feed, and the pigs in another group received the commercial feed supplemented with probiotics as *Enterococcus italicus* in dry powder form. Mortality data were collected, and the animals were vaccinated against *Mycoplasma hyopneumoniae* twice (Intervet Schering-Plough Animal Health’s vaccine, serial 0051908), at age 45 and 59 day-old. Then, a blood test was done in order to determine immunity level against the disease. The serum were sent to the Betagro Science Center to determine the titer using an ELISA kit (IDEXX; X3(. Results were evaluated using the sample to positive ratio (S/P ratio):

$$\text{S/P ratio} = \frac{\text{Sample mean (mean of optical absorbance)} - \text{negative control mean}}{\text{Positive control mean} - \text{negative control mean}}$$

The S/P ration was interpreted by Mean IDEXX titer=mean S/P ration; >0.4 is positive; between 0.3 and 0.4 is suspect; <0.3 is negative. All results were statistically analyzed by analysis of variance, and differences between the average values obtained from each group were evaluated using Duncan’s multiple range tests.

Results

Through the experiments, no studied pig was found dead. For the immunity levels of the pigs in both studied groups at seven days after the second *Mycoplasma hyopneumoniae* vaccination, the titers were increased. The pigs received *Enterococcus italicus* in dry powder form, had the highest level of immunity with significance (P<0.001(as showed in the Table 1.

Table 1. The level of immunity against *Mycoplama hyopneumoniae* (S/P Ratio).

Group/date	Before the vaccination	7 days after the 1 st vaccination	7days after the 2 nd vaccination
Control	0.000 ^d	0.252 ^c	1.005 ^b
Probiotic	0.002 ^d	0.285 ^c	1.236 ^a

^{a,b,c,d} The same letters in a row or column indicate no significant differences (P<0.001(.

Discussions and Conclusions

Mycoplasma hyopneumoniae is the primary pathogen of enzootic pneumonia, a chronic respiratory disease in swine and highly infection in swine producing. They cause significant economic losses

due to increased medication use and decreased performance of the swine. Moreover, *Mycoplasma hyopneumoniae* is also considered to be one of the primary agents involved in the porcine respiratory disease complex (PRDC) (Maes et al., 2008). Vaccination option is a widespread means used among pig farmers to control *Mycoplasma hyopneumoniae*. To optimize the vaccination, it needs to be used in a correct way and in consideration of the disease situation of each farm. Therefore, the program of *Mycoplasma hyopneumoniae* vaccination of each farm is different because it must be appropriate for the herd health status or disease state of the farm (Merial, 2015).

Sera from 2 Months Postvaccination were also tested for *Mycoplasma hyopneumoniae* antibodies using the IDEXX ELISA test in Rapp-Gabrielson et al. (2002). The IDEXX test detected a higher number of positive antibody titers in 21 vaccinated swine as 24% positive 14% suspect and 62% negative. Pre-challenge sera from non-vaccinated swine were negative. This is consistent with the results of the present study which show that the immunity level of the pigs receiving the probiotics and those not receiving the probiotics is higher at seven days after the second vaccination, and the former receiving the probiotics as *Enterococcus italicus* in dry powder form has the highest level with significance) $P < 0.001$.

Probiotics can enhance immune responses in the host in several ways. For example, the recruitment of enterocytes and mucosal immune cells into inflamed areas to promote the production of anti-inflammatory cytokines and reduce the production of pro-inflammatory cytokines; the probiotics (lactobacillus) improve the functions of macrophage, the number of antibody, the production of interferon, and the functions of natural killer cells)Sirichokchatchawan et al., 2008.(To control *Mycoplasma hyopneumoniae*, the vaccine given by injection (including *Mycoplasma* vaccine) cannot efficaciously enhance immune responses in mucous membrane but mainly in blood stream including helper T (Th) cell and B cell)Merial, 2015.(This is possibly because the probiotics (*Enterococcus italicus*)stimulate the functions of helper T (Th) cell and B cell and consequently enhance the efficacy of the vaccination.

References

- Kasornpikul C. and S.Chalorsuntisakul, 2011. Isolation of Probiotic Bacteria from Piglet Nostrils, Full Research Report. Faculty of Animal Sciences and Agricultural Technologypp. 41.
- Maesa, D., J. Segalesb, T. Meynsa, M. Sibilab, M. Pietersc, F. Haesebroucka, 2008. Control of *Mycoplasma hyopneumoniae* infections in swine. *Vet Microbiol* 126: 297 – 309
- Merial. 2008. Mycoplasmal Pneumonia or Enzootic Pneumonia. Available online http://www.merial.co.th/Livestock/Pages/enzootic_pneumonia.aspx (27 May 2015)
- Rapp-Gabrielson Vicki J., L. Gergen, B. Eddy, T. Wasmoen, R. Saltzman, L. Kesi, 2002. Evaluation of duration of immunity after vaccination of swine with a single dose of *Mycoplasma hyopneumoniae*bacterin (M+Pac®). Available online [http://www.thepigsite.com/focus/contents/Evaluation%20of%20M+Pac%20one%20dose.\(27](http://www.thepigsite.com/focus/contents/Evaluation%20of%20M+Pac%20one%20dose.(27) May 2015)
- SirichokchatchawanW. ,T.Chalermchaikit ,N.Prapasarakul, 2008. Roles of Probiotics in Swine Production Industry. Available online http://www.micro.vet.chula.ac.th/attachments/131_Probiotic.pdf (27 May 2008).
- Suthamma K. and N. Taweechai, 2014. Powder production of bacterial antagonistic *Bacillus subtilis* (Bs). Available online http://www.rdi.ku.ac.th/kasetresearch52/04-plant/gaigaew/plant_00.html.

Effect dietary energy concentrate on birth weight and weaning weight of Sudanese Taggar goat kid's

Bushara, I.¹, Dafalla, M. Mekki¹., M.O.Mudala¹ & A. M. Abu Nihiala²

¹ Dept. Animal Production, Faculty of Natural Resources & Environmental Studies, University of Kordofan, P.O. Box 716, Khartoum, Sudan, bushara3030@yahoo.com

² Dept. Dairy Productions, Faculty of Animal Production, University of Khartoum, Sudan

Abstracts

The objective of this study was to investigate the effect of supplementation nutrition on kid's birth weight and weaning weight of Taggar goat under traditional management in southern Kordofan state in west Sudan. Sixty three (63) kids of Taggar goats were used in this experiment. Animals were allocated to three feeding regimes in a complete random design. The results indicated that kids born to supplemented does secured higher birth weight compared with kids born to does unsupplemented (control group). Body weight at weaning was higher in kids born to supplemented does groups compared with kids born to unsupplemented does (control) group, also the daily body weight gain was higher for kids supplemented with concentrate rations as ay compared with control kids born to unsupplemented dose.

Keywords: Taggar kids, concentrate ration, birth weight, weaning, Sudan

Introduction

Goat is valuable livestock in the arid and semi-arid zones, especially adopted goat to harsh environment. The Sudanese Taggar goats, its meat breed and its production depends on the performance starting from birth weight to weaning point. Prewaning performance of kids provides stage upon which post-weaning performance is built. Generally Taggar goats, are raised by poor farmers and distressed women. Extensive production system is common practice in traditional farming system depended on communal grazing. Goats fulfill their nutrition needs mainly by eating the available vegetation. They rarely receive any supplements. The supply of nutrients from the pastures fluctuates between years and seasons. Thus, effective utilization of the available feed resources and appropriate supplementation of poor quality natural pasture and crop residue-based diets appear to be necessary steps to alleviate the nutritional constraint (Banerjee, 2000). Birth weight and the growth of kids until weaning, together with reproduction characteristics are reliable indicators of the breed efficiency in the production of meat (Husain, 1993). The present aimed to investigate the effect of supplementation nutrition on kid's birth weight and weaning weight of Taggar goat in southern Kordofan state in west Sudan.

Materials and Methods

This study were conducted in Dalanj area (longitudes 12.02° N, Latitudes 29.39°E) Southern Kordofan state. Thirty six (63) Taggar kids have been used in this experiment, were weighed and blocking according to the three groups from which they born (supplemented and unsupplemented groups), the blocked groups were assigned at random to supplement diets into three groups A (control), B and C. Kids monitored from birth to weaning age. All kids were daily allowed grazing on pasture. On their returning from pasture kids in group B and C were offered 150 g / head/day increase to 250g /day/ head of concentrate ration B and C respectively (Table 1). The data were statistically analyzed according to complete randomizes design using computer program Statistical Package for Social Sciences, software package (SPSS version 10 1999). Duncan's Multiple Range Tests (DMRT) was also used to test means significance differences

Results

Kid birth and weaning weight

Birth weight of kids data as affected by dietary energy are shown in Table (2). The supplementary rations that given to the experimental does, had highly significant ($p < 0.01$) effect on kid birth weight, weaning and daily body weight gain.

Discussion

Birth weight of animals is one of the most important factors influencing the pre-weaning growth of the young (Husain et al., 1996). The importance of supplementation during the last lap of pregnancy was confirmed in this study, does on group B and C producing the heaviest kids at parturition compared to the unsupplemented does having the lightest kids at birth, this results on line with Salim et al. (2002) and Ng'ambi et al. (2008). The significant difference in weight of kids resulting from the random effects of the dams can be attributed to the natural variation occurring in the prenatal and post birth nutrient supplied by mothers (Mavrogenis et al., 1984). The differences in birth weight between kids born could be the results of maternal nutrition (high and low nutrients available for the foetus). Therefore, a major factor contributing to the low birth rates in traditionally reared flocks is likely to be nutrition, since goats are kept almost exclusively on the natural veld. Weaning weights are crucial and indicate the milking ability of the herds as well as the growth potential of the kids. The body weight at weaning at 90 days of age was significantly heaviest for supplemented kids 8.71 and 8.37 kg compared with lower weight in unsupplemented kids 7.06 kg, and this due to high growth rate in pre weaning period in supplemented kids compared with unsupplemented kids, similar results were obtained by Berhane and Eik (2006). Generally, the availability of enough milk from the dams results to faster pre-weaning weight gain and infested the effect of birth weight on dietary treatments (Alexandre et al., 2002). The highest average daily gain values reported in this study are justifiable given the fact that, the kids were born during the wet season where, naturally grazed pasture were plenty and in additional to suckling enough milk from their mothers. The results are generally attributed to fact that these kids were more mature at birth and their physiological systems were more developed to respond better to good nutrition, also could be accounted for in terms of milk yield and composition.

Table 1. Chemical composition of the experimental feed stuffs.

Concentrate rations types	DM%	CP%	CF%	EE%	NFE%	Ash%	ME(MJ/Kg DM)
Ration B	93.2	20.4	10.3	4.5	58	6.8	12.20
Ration C	93.9	16.7	17.4	6.6	47.5	11.8	11.57

(DM= Dry matter, CP= Crude protein, CF= Crude fibre, EE= Ether Extract, NFE= Nitrogen Free Extract)

Table 2. Effect of Dietary energy level on birth, weaning weight and daily body weight gain.

Animal rations	Group and N	Birth weight	Weaning weight	daily body weight gain/g
Group A (Ration A)	20	1.84±0.07 ^{bd}	7.06±0.22 ^{bd}	58.10±2.63 ^{bd}
Group B (Ration B)	25	2.16±0.09 ^a	8.71±0.32 ^{ac}	72.78±3.35 ^{ac}
Group C (Ration C)	18	2.11±0.07 ^{ac}	8.37 ±0.38 ^a	68.03±3.06 ^a

Values in the same column followed with different letters are significant at P<0.05 and/or P<0.01.

References

- Alexandre, G., Fleury, J., Coppry, O., Archimede, H and Xande, A. 2002. Effects of mode of supplementation upon milk and growth performances of suckling Creole goats and their kids reared at pasture in Guadeloupe. *Livestock Research for Rural Development* 14 (1).
- Banerjee, S. 2000. Conserving animal genetic resources. *Animal production and Health commission for Asia and the Pacific*, Vol 16 No.3.
- Berhane, G and Eik,L.O. 2006. Effect of vetch (*Vicia sativa*) hay supplementation to Begait and Abergelle goats in northern Ethiopia II. Reproduction and growth rate. *Small Rumin. Res.* 64, 233–240
- Husain,S. S., Horst.P., and Islam, A.B.M.M. 1996.Effect of different factors on preweaning survivability of Black Bengal kids. *Small.Rum.Res.*18:1-5.
- Husain, S.S.1993.A study on reproductive performance and genetic potentials of Black Bengal goats. Ph. D thesis, Bangladesh Agric. Univ., Mymensingh.
- Mavrogenis, A. P., Constantinou, A. and Louca, A. 1984. Environmental and genetic causes of variation in production traits of Damascus goats. *Anim. Prod.* 38:91-97
- Ng'ambi, J. W., Khitsane, L., Norris, D. and Mbajiorgu, C. A. 2008. Effect of soybean meal supplementation on pre-and post-partum productivity of Angora goats in communal rangelands of Molimo-Nthuse in Lesotho. *Livestock Research for Rural Development.* ; 20 (1)
- Salim, H. M., Shahjala, M., Tareque, A. M. M. and Kabir,F.2002.Effect of concentrate supplementation on growth and reproductive performance of female sheep and goats under grazing condition. *Pakistan. J. Nutr.* 1(4): 191-193.
- S.P.S.S., 1999. *Statistical Package for Social Science* version 10.

Feeding standard for Hanwoo cattle: past, present and future

Kim, M., Y. C. Baek & Y. K. Oh

Animal Nutrient and Physiology Team, National Institute of Animal Science, RDA, Wanju-gun, Jeollabuk-do 565-851, Republic of Korea

Abstract

Since 1983, studies on feeding standard for Korean native Hanwoo cattle have been conducted. Initially, feeding standards for Hanwoo heifers and bulls were studied before 2000. Since the early 2000's, studies on feeding standard of Hanwoo steers have been concentrated due to increased demand for Hanwoo steers in Korean beef industry. To estimate energy requirements for maintenance, Hanwoo steers were fed diets with three different levels of energy. Energy requirement for maintenance of Hanwoo steers has been estimated as $124.3 \text{ kcal/BW}^{0.75}$. Also, Hanwoo steers were fed diets with three different levels of crude protein to estimate crude protein requirements for maintenance. Crude protein requirement for maintenance of Hanwoo steers has been estimated as $5.56 \text{ g/BW}^{0.75}$. Currently, we are conducting studies to update feeding standard for Hanwoo heifers. In future studies, repetitive large-scale experiments would need to be performed to determine more accurate feeding standard for Hanwoo cattle.

Keywords: energy requirements, feeding standard, Hanwoo cattle, protein requirements

Introduction

Since the first feeding standard for Hanwoo that is Korean native beef cattle (Lee et al., 2014) was announced in 1983, it continues to be updated. Because production of high-quality meat was encouraged in the late 1990's, studies on nutrient requirements for Hanwoo steers instead of Hanwoo bulls have been concentrated. Based on these studies, the revised feeding standard for Hanwoo cattle was announced in 2002. Since then, additional updates of feeding standard for Hanwoo cattle was announced in 2007 and 2012, respectively. Recently, nutrient requirements for Hanwoo heifers are actively studied because few studies on Hanwoo heifers have been conducted. In this review, we reviewed and discussed past, present and future studies on feeding standards for Hanwoo cattle. In the future, continuous studies of feeding standard for Hanwoo cattle would need to be conducted for advances in Hanwoo industry.

Past studies

Kim et al. (2004) estimated energy requirements for maintenance of Hanwoo steers as $124.3 \text{ kcal/BW}^{0.75}$ using a linear relationship between ME intake and retained energy (Figure 1A). For this experiment, 9 Hanwoo steers ($376.6 \pm 12.5 \text{ kg}$) were fed 44% rice straw and 56% concentrate with 3 different levels of energy (0.8, 1.2 and 1.6 times maintenance). On the other hand, crude protein requirements for maintenance of Hanwoo steers were estimated using 18 Hanwoo steers ($173.7 \pm 26.3 \text{ kg}$) fed 20% rice straw and 80% concentrate with 3 different levels of crude protein (Kim et al., 2006). Crude protein requirements for maintenance of Hanwoo steers were estimated as $5.56 \text{ g/BW}^{0.75}$ using a linear relationship between crude protein intake and retained crude protein (Figure 1B).

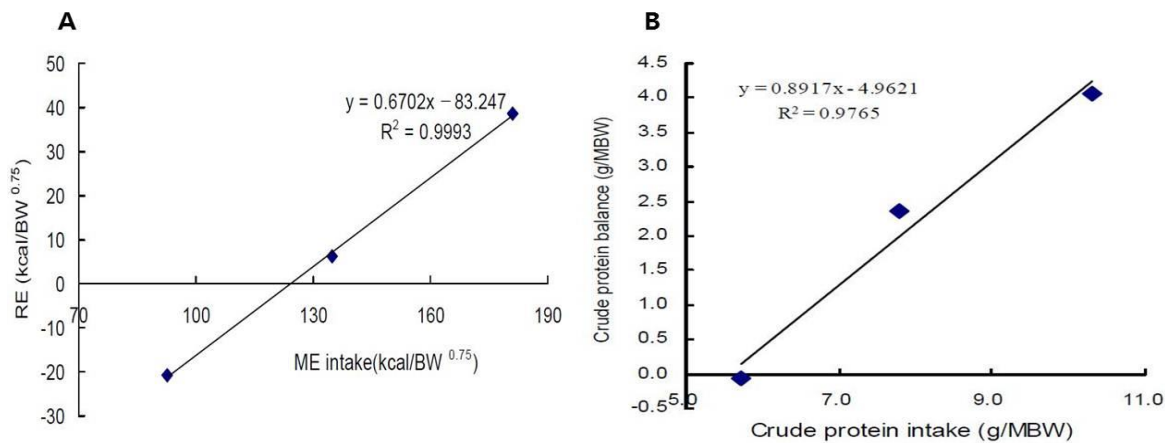


Figure 1. Estimation of energy (A) and crude protein (B) requirements for maintenance of Hanwoo steers (Kim et al., 2004 and 2006).

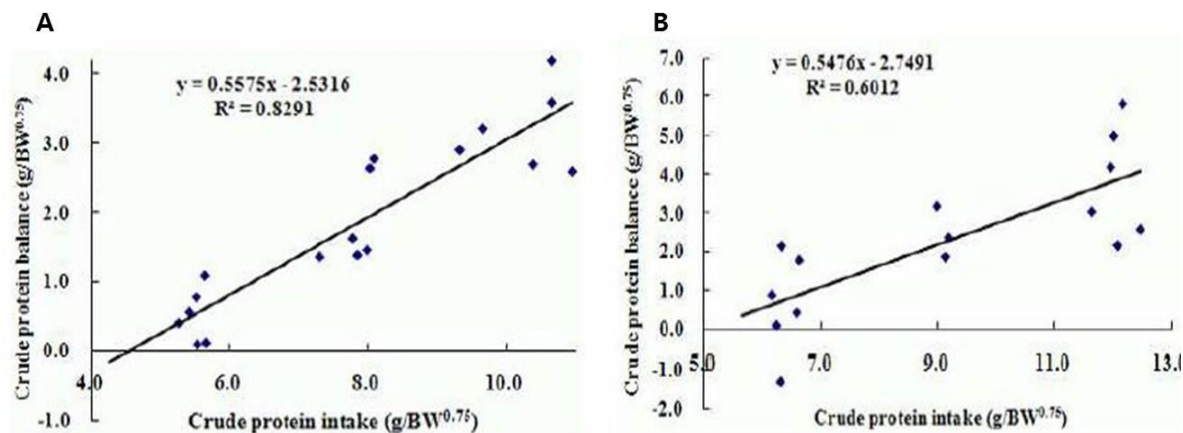


Figure 2. Estimation of crude protein requirements for maintenance of Hanwoo heifers in experiment 1 (A) and experiment 2 (B) (Kim et al., 2011).

For Hanwoo heifers, Nam et al. (2011) estimated crude protein requirements for maintenance at 2 different levels of body weight: 1) 143.2 ± 7.7 kg (experiment 1) and 2) 257.8 ± 4.2 kg (experiment 2). Linear relationships between crude protein intake and crude protein balance were $4.58 \text{ g/BW}^{0.75}$ and $5.02 \text{ g/BW}^{0.75}$ in experiment 1 and 2, respectively (Figure 2).

Present and future studies

Currently, we are conducting studies on energy and protein requirements for maintenance of Hanwoo heifer to update feeding standard for Korean native Hanwoo. In this study, heifers were fed diets with 3 different levels of energy or protein, and then energy and protein requirements for maintenance will be determined using respiration chambers. We also are conducting a study to determine nutrient requirements for Hanwoo heifer in summer and winter season, respectively.

In future studies, repetitive large-scale experiments would need to be conducted to determine more accurate nutrient requirements for both Hanwoo steers and heifers. Continuous studies of feeding standard for Hanwoo cattle will contribute to advances in Hanwoo industry.

References

- Kim, K. H., Y. G. Oh, W. Kim, S. C. Lee, K. J. Shin and B. T. Jeon, 2004. Determination of energy requirements for maintenance in Hanwoo steers. *J. Anim. Sci. & Technol. (Kor.)*. 46: 193-200
- Kim, K. H., Y. G. Oh, S. C. Lee, K. J. Shin, S. W. Kang, Y. H. Moon and M. K. Song, 2006. Crude protein requirements for maintenance of Hanwoo steers by protein balance trial. *J. Anim. Sci. & Technol. (Kor.)*. 48: 533-540
- Lee, S. H., B. H. Park, A. Sharma, C. G. Dang, S. S. Lee, T. J. Choi, Y. H. Choy, H. C. Kim, K. J. Jeon, S. D. Kim, S. H. Yeon, S. B. Park and H. S. Kang, 2014. Hanwoo cattle: origin, domestication, breeding strategies and genomic selection. *J. Anim. Sci. & Technol.* 56: 2
- Nam, I. S., Y. G. Oh, S. S. Jang, D. H. Kim, Y. J. Seol and K. H. Kim, 2011. Determination of crude protein requirements for maintenance of prepubertal Hanwoo heifers. *J. Anim. Sci. & Technol. (Kor.)*. 53: 171-176

Estimation of the TDN of spent mushroom substrates used as Hanwoo feed

Baek, Y. C., M. Kim, & Y. K. Oh

Animal Nutrient and Physiology Team, National Institute of Animal Science, RDA, Wanju-gun, Jeollabuk-do 565-851, Republic of Korea

Abstract

Recently, there has been a growing interest in the use of spent mushroom substrate as Hanwoo feed due to their low cost. Before its use as Hanwoo feed, measurement of TDN is necessary to evaluate the feed value of spent mushroom substrate. An *in vivo* experiment is commonly used to estimate TDN but needs a lot of time and cost. The objective of this study was to find an alternative method to replace a TDN estimation method based on an *in vivo* experiment. As alternative methods, both an *in situ* experiment and the NRC 2001 model were used to estimate TDN of spent mushroom substrate of *Flammulina velutipes* and spent mushroom substrate of *Pleurotus ostreatus*. TDN estimated by these two methods was comparatively assessed against TDN estimated by an *in vivo* experiment. Results indicated that the NRC2001 model may be an alternative method to estimate TDN of spent mushroom substrate instead of an *in vivo* experiment. More studies with other feed ingredients would need to be conducted to revise and verify these alternative methods for TDN estimation.

Keywords: Hanwoo, *in situ*, TDN, NRC, spent mushroom substrates

Introduction

Recently, Hanwoo cattle industry has suffered through opening of the market and increased feed cost. Therefore, there have been many studies on the use of agrifood by-products in the field of ruminant nutrition (Park et al., 2011; Chang et al., 2013). Because annual production of spent mushroom substrate (SMS), which is one of agrifood by-products, is 1.67 million tons in South Korea, there has been a growing interest in the use of SMS as Hanwoo feed. Before the use of SMS as Hanwoo feed, measurement of TDN will need to take precedence to evaluate the feed value of SMS. In the current study, we compared an *in vivo* experiment with 2 alternative methods, which are *in situ* experiment and the NRC 2001 model, to measure TDN of SMS.

Materials and Methods

Both spent mushroom substrate of *Flammulina velutipes* (SMSF) and spent mushroom substrate of *Pleurotus ostreatus* (SMSP) samples were provided by a mushroom farm in Chungcheongbuk-do Province in South Korea. Cultivation substrates for SMSF mostly consisted of corn cob and rice bran, while those of SMSP mostly consisted of sawdust and corn cob.

Standard AOAC methods (AOAC, 2000) were used to determine dry matter, crude protein, ether extract, crude fiber, nitrogen free extract, crude ash, NDF, ADF, ADL, NDICP, ADICP, NFC and gross energy (kcal/g DM).

Both an *in situ* experiment and the NRC 2001 model were used to estimate TDN of SMSF and SMSP.

Result and Discussion

Chemical compositions of SMSF and SMSP were shown in Table 1. Both SMSF and SMSP included high NDF.

Table 1. Chemical composition of SMSF and SMSP.

Nutrients	% DM	
	SMSF	SMSP
Dry matter (% As-fed)	41.6	37.8
Crude protein	10.8	13.5
Ether extract	5.1	0.4
Crude fiber	25.2	38.1
Nitrogen free extract	45.4	40.8
Crude ash	13.6	7.1
NDF	54.7	72.8
ADF	33.9	56.6
ADL	10.1	13.5
NDICP	6.5	6.3
ADICP	3.7	5.7
NFC	22.4	12.5
Gross energy (kcal/g DM)	4,437.5	4,406.2

The average TDN of SMSF or SMSP was lower in an *in situ* experiment than in an *in vivo* experiment probably due to variation in the pore size of nylon bags (Figure 1). On the other hand, the average TDN of SMSF was slightly greater in the NRC 2001 model than in an *in vivo* experiment, whereas that of SMSP was slightly lower in the NRC 2001 model than in an *in vivo* experiment.

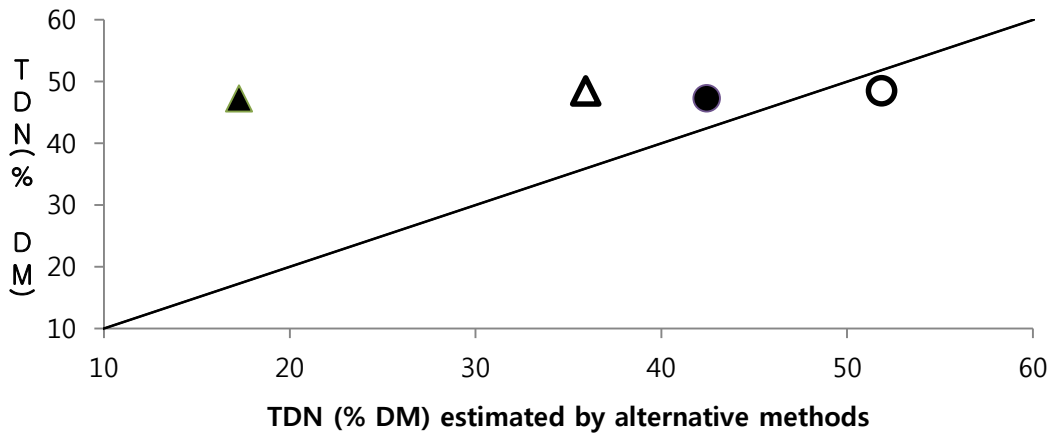


Figure 1. Comparison of *in vivo* TDN with 2 alternative methods (*in situ* method and NRC 2001 equation). *In situ*: ▲ SMSP, △ SMSF, NRC (2001) equation: ○ SMSF ● SMSP.

Nonetheless, the average TDN estimated by the NRC 2001 model was marked with a range of TDN estimated in individual animals in an *in vivo* experiment. These results indicate that the NRC2001 model may be an alternative method to estimate TDN of SMS instead of an *in vivo* experiment. More studies with other feed ingredients would need to be conducted to revise and verify these alternative methods for TDN estimation.

References

- AOAC, 2000. Official method of analysis. 17th Edition. Association of Official Analytical Chemists, Washington DC, USA
- Chang, S. S., H. J. Kwon, S. M. Lee, Y. M. Cho, K. Y. Chung, N. J. Choi and S. S. Lee, 2013. Effects of brewers grain, soybean curd and rice straw as an ingredient of TMR on growth performance, serum parameters and carcass characteristics of Hanwoo steers. *J. Anim. Sci. & Technol. (Kor.)*.55: 51-59
- Park, J. K., D. H. Lim, S. B. Kim, K. S. Ki, H. J. Lee, E. G. Kwon, W. M. Cho and C. H. Kim, 2011. Effects of partial replacement of corn grain and soybean meal with agricultural by-product feeds on *in vitro* rumen fermentation characteristics and optimum levels of mixing ratio. *J. Anim. Sci. & Technol. (Kor.)*. 53: 441-450

Feed intake, nutrient digestibility and rumen parameters in goats as affected by mao (*Antidesma thwaitesianum* Muell. Arg.) seed meal supplementation

Gunun^{*1}, P., N. Anantasook², M. Wanapat³, S. Sirilaophaisan¹ & A. Cherdthong³

¹Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, Sakon Nakhon Campus, Phangkhon, Sakon Nakhon, Thailand

²Program in Animal Production Technology, Faculty of Technology, Udon Thani Rajabhat University, Udon Thani, Thailand

³Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand

Abstract

The objective of this experiment was to evaluate the effect of mao (*Antidesma thwaitesianum* Muell. Arg.) seed meal (MSM) on feed intake, digestibility and rumen parameters in goats were fed at 0, 0.8, 1.6 and 2.4% of dry matter intake (DMI). Four female crossbred goats with initial body weight (BW) 20±2 kg were randomly assigned to a 4×4 Latin square design. Animals were fed with a roughage to concentrate ratio (R:C) of 60:40 and pangola grass hay was used as a roughage source. It was found that supplementation of mao seed had no effect on DMI and apparent digestibility when compared with the control (P>0.05). Moreover, ruminal pH and NH₃-N was not altered among all treatments (P>0.05), whereas BUN were found to be decreased (P<0.05) by MSM supplementation at 2.4% DMI. In conclusion, MSM supplementation at 2.4% DMI exhibited no negative on feed intake, digestibility of nutrients and rumen parameters in goats.

Keywords: mao seed meal, feed intake, digestibility, rumen parameters, goats

**Corresponding author: pongsatorng@hotmail.com*

Introduction

Conventional by-products and unconventional materials from the food processing industry have been frequently included in animal diets. Mao, or Makmao (*Antidesma thwaitesianum* Müll. Arg.) is favorable to be consumed and sold in the local market in Thailand because of its good color and taste (Poungpronpitag et al., 2008). Mao grows well over a variety of soil types and is naturalized in Australia, Africa, tropical Asia and islands in the Pacific Oceans (Poungpronpitag et al., 2011). Mao is used for wine and juice production industry and the major solid by-product generated is mao seed and marces. Mao seed contains large amount of underutilised condensed tannins (CT). Supplementation of wastes products from the mao into diet of ruminant could increase the efficiency of by-products utilization in the feeding systems. Our previous study (Gunun et al. 2014) reported that mao seed meal (MSM) supplementation has the potential to manipulate rumen fermentation, by decreasing protozoa and maintaining *in vitro* kinetics gas and digestibility. Therefore, the objective of this study was to investigate the effect of MSM supplementation on feed intake, digestibility and rumen parameters in goats.

Materials and methods

Four female crossbred (Thai Native × Anglo Nubian) goats with initial body weight (BW) 20±2 kg were randomly assigned to a 4 × 4 Latin square design. The dietary treatments were as follows: supplementation with mao seed meal (MSM) at 0, 0.8, 1.6 and 2.4% of DMI. All animals were fed a diet containing roughage to concentrate ratio (R:C) of 60:40 at 3.0% BW/d and pangola grass (*Digitaria eriantha* Steud., synonym *D. decumbens*) hay was used as a roughage source. The diet of R:C was offered to the animals twice per day at morning (07:00) and afternoon (16:00) feeding time. Goats were housed individually in ventilated pens with wooden slotted flooring in an open goat barn raised above the ground. Clean fresh water and feed blocks were available at all times. The experiment was conducted over four periods, each lasting for 21 days: the first 14 days were used for feed intake measurements and the remaining 7 days for total faecal collection. Rumen fluid, blood samples were collected at 0, 3 and 6 h-post feeding, and pH was measured immediately. Rumen fluid sample were then kept for analysis of NH₃-N. Blood urea nitrogen (BUN) was analysed according to the method of Crocker (1967). All data were subjected to ANOVA according to a 4 × 4 Latin square design using the General Linear Models (GLM) procedures (SAS, 1996).

Results and Discussion

The results show that feed intake and apparent nutrient digestibility were not affected by MSM supplementation ($p>0.05$) (Table 1). Supplementation of plant-containing tannins to ruminant diets usually reduces feed intake because of reduced palatability and decreased rate of digestion. The negative effect of condensed tannins on feed intake was caused by astringency of CT and short-term post-ingestive malaise. Beauchemin et al. (2008) reported that feed intake were reduced by high doses (>50 g/kgDM) of CT uptake. However, in the present study, dietary tannins sources had no effect on DMI when used suitable level (<50 g/kg DM) as a supplement. Supplementation with MSM did not affect ruminal pH and NH₃-N concentrations ($p>0.05$), whereas BUN was decreased ($p<0.05$) by MSM supplementation (Table 2). When tannins containing plant is masticated, tannins-protein complexes are formed; these are stable over the pH range 3.5 to 7.0 but dissociate in the abomasums and duodenum (Jones et al., 1994). Hence, decreasing availability of feed protein for ruminal degradation and NH₃-N release. In our study, similar ruminal NH₃-N concentrations among treatments and this could be due to the lower doses of MSM supplementation. Similarly, Animut et al. (2008) reported that rumen NH₃-N concentrations were not affected by varying source of condensed tannins in goats.

In conclusion, MSM supplementation at 2.4% DMI did not adversely affect feed intake, nutrient digestibility and rumen parameters in goats fed on pangola grass hay based diet.

Table 1 Effect of mao seed meal supplementation on feed intake and digestibility in goats.

Item	Supplementation of MSM (%)				SEM	P-value
	0	0.8	1.6	2.4		
DMI						
g/d	573.7	582.1	584.2	618.6	24.85	0.92
%BW	2.6	2.6	2.6	2.7	0.06	0.82
g/kg BW ^{0.75}	56.2	55.7	56.8	59.3	1.14	0.69
Apparent Digestibility, %DM						
DM	71.2	70.8	70.2	70.8	0.47	0.26
OM	71.9	72.2	72.0	72.3	0.41	0.36
CP	80.3	80.3	80.9	80.1	0.74	0.60
NDF	65.4	64.2	63.9	62.9	0.59	0.31
ADF	54.0	53.4	53.1	53.5	0.90	0.53

Table 2 Effect of mao seed meal supplementation on ruminal pH, NH₃-N and BUN in goats.

Item	Supplementation of MSM (%)				SEM	P-value
	0	0.8	1.6	2.4		
pH	6.5	6.6	6.7	6.7	0.07	0.18
NH ₃ -N	19.6	20.8	18.7	19.4	0.57	0.62
BUN	15.2 ^a	16.2 ^{ab}	16.8 ^b	13.9 ^c	0.35	0.03

^{a, b, c} Values on the same row with different superscripts differ (p<0.05).

Acknowledgements

The authors would like to express their most sincere gratitude and appreciation to the Program in Animal Production Technology, Faculty of Technology, Udon Thani Rajabhat University for the use of the research facilities.

References

- Animut, G., R. Puchala, A. L. Goetsch, A. K. Patra, T. Sahlu, V. H. Varel and J. Well. 2008. Methane emission by goats consuming different sources of condensed tannins. *Anim. Feed Sci. Technol.* 144:228-241.
- Beauchemin, K.A., M. Kreuzer, F. O'Mara and T.A. McAllister. 2008. Nutritional management for enteric methane abatement: a review. *Aust. J. Exp. Agr.* 48: 21–27.
- Crocker, C.L. 1967. Rapid determination of urea nitrogen in serum or plasma without deproteinization. *Am. J. Med. Technol.* 33: 361–365.
- Gunun, P., N. Anantasook, M. Wanapat, S. Sirilaophaisan, A. Cherdthong, C. Wachirapakorn and C. Yuangklang. 2014. Effect of Mao (*Antidesma thwaitesianum* Mull. Arg.) seed supplementation on *in vitro* rumen protozoal population and digestibility using a gas production technique. *Khon Kaen Agr. J.* 42) suppl. 4): 11 (abstract).
- Jones, G.A., T.A. McAllister, A.D. Muir and K.J. Cheng. 1994. Effects of sainfoin (*Onobrychis viciifolia* Scop.) condensed tannins on growth and proteolysis by four strains of ruminal bacteria. *Appl. Environ. Microbiol.* 60: 1374–1378.
- Puangpronpitag, D., P. Areejitranusorn, P. Boonsiri, M. Suttajit and P. Yongvanit. 2008. Compounds isolated from *Antidesma thwaitesianum* Müll. Arg. seeds and marcs. *J. Food Sci.* 73: 648–653.
- Puangpronpitag, D., P. Yongvanit, P. Boonsiri, M. Suttajit, P. Areejitranusorn, H.K. Na and Y.J. Surh. 2011. Molecular mechanism underlying anti-apoptotic and anti-inflammatory effects of Mamao (*Antidesma thwaitesianum* Müll. Arg.) polyphenolics in human breast epithelial cells. *Food Chem.* 127: 1450–1458.
- SAS. 1996. User's Guide: Statistic, Version 5. Edition. SAS. Inst. Cary, NC, USA.

Effects of spineless cactus feeding on milk production, milk quality and antioxidant capacity in dairy goat

Ichimura¹, H., S. Ishikawa², N. Isobe³ & Y. Hayashi^{1,2}

¹Graduate School of Agriculture, Meijo University, Kasugai, 486-0804, Japan, ²Faculty of Agriculture, Meijo University, Kasugai, 486-0804, Japan, ³Graduate School of Biosphere Science, Hiroshima University, Higashi-Hiroshima, 739-8528, Japan

Abstract

This study evaluated the effects of spineless cactus feeding on milk production, milk quality and antioxidant capacity in dairy goats. Four Japanese Saanenlactating goats (70.9 ± 7.1 kg body weight at the beginning of experiment, 106.5 ± 39.7 days of lactation and 2.3 ± 1.1 parity) were divided by two groups and fed a nutrient required feed with or without a 200 g dry matter (DM) substitution of spineless cactus for barley. One experimental term has fourteen days with ten-day adaptation period and four-day trial period. The experiment was conducted using a cross-over design. Diet was fed at 9:00 hours and 16:00 hours every day. The amount of refused feed was recorded at 9:00 hours during the trial period. Bodyweight of goats was recorded before the morning feeding on the first and the final day of each experimental term. Milk yield was measured at 16:00 hours every day in the trial period. Blood of goats was collected from jugular vein on the final day of each experimental term and plasma was collected. Feed intake, bodyweight and milk yield were not decreased by the substitution of spineless cactus. The contents of protein and antimicrobial substances (lactoferrin and S100A7) in milk, calcium (Ca) and urea nitrogen concentrations in plasma showed higher mean values with the spineless cactus substitution than without the substitution. On the other hand, the cactus feeding showed lower mean values of milk fat, plasma inorganic phosphorus (IP) and plasma total cholesterol (T-CHO) than the control feeding. The degree of oxidative stress and antioxidant capacity did not show the significant differences between the groups. The 200 g DM substitution of spineless cactus for barley did not affect to milk production and antioxidant capacity of dairy goats. However, the feeding of the cactus to goats has possibilities to promote protein metabolism and increase the contents of antibacterial factors in milk.

Keywords: antioxidant capacity, goat, milk production, milk quality, spineless cactus

Corresponding author: 153561005@c alumni.meijo-u.ac.jp

Introduction

Spineless cactus (*Opuntia* spp.) is originally bred for forage as well as vegetable, fruit and etc. However, occasions for utilizing this cactus as fodder are very few in Japan. The rate of feed self-sufficiency in Japan is around 26% in total feeds including concentrates and forage (MAFF, 2015). Thus, the supply of most feed materials, especially concentrates, is depended on the imports from the other countries and the improvement of feed self-sufficiency in Japan is required. Spineless cactus has been utilized in feeding ruminants in dry areas such as Brazil, Chile, California, Morocco, Mexico, South Africa, Texas and Tunisia due to its high biomass yield (Stintzing and Carle, 2005). The cactus feeding for dairy goats was reported to maintain the milk yield and affected to fatty acid compositions in milk (Costa et al., 2010). On the other hand, the presence of antioxidant substances (kaempferol, quercetin and etc.) has been reported in the cactus (Stintzing and Carle, 2005). Therefore, the feeding of this cactus has possibilities to promote livestock health and produce functional livestock products. Among the livestock products, goat milk has lower α_{s1} -casein content than cow milk. In addition, the specific gravity of casein micelle is lighter and the diameter of fat globule is smaller in goat milk than in cow milk (Clark and Sherbon, 2000). Thus, goat milk is remarkable for prevent milk allergy and good digestion to human consumption. If the further advantage of cactus as a feed resource for lactating goats was shown, utilization of cactus as a feed material could be increasing. Then, the cactus feeding could contribute to promote the goat health and the demand of goat milk. However, there were few reports on effects of cactus feeding on milk production, milk quality and antioxidant capacity in dairy animals. Therefore, this study was conducted for identifying the effects of substitution of spineless cactus for barley on milk production, milk quality and antioxidant capacity in dairy goats.

Material and Methods

1. Experimental location and animal management

Four Japanese Saanen lactating goats (70.9±7.1 kg bodyweight at the beginning of experiment, 106.5±39.7 days of lactation and 2.3±1.1 parity) were utilized for the experiment. The goats were raised at Experimental Farm, Faculty of Agriculture, Meijo University located in Kasugai, Japan. The does were divided by two groups (two goats each, Cactus group and Control group) and fed a nutrient required feed with or without a 200 g DM substitution of spineless cactus for barley. The diets were prepared a total ration with 9.1% of crude protein (CP), 51.0% of total digestible nutrients and 0.05% of calcium (Ca) and phosphorus (P) each according to NRC (1981), to meet the requirement for a 1.5 kg/day milk production. Each animal in Cactus group was fed 200 g of cactus, 200 g of barley, 600 g of wheat bran and 1300 g of ryegrass straw on a DM basis, twice a day at 9:00 hours and 16:00 hours. Each animal in Control group was fed 400 g of barley, 600 g of wheat bran, 1300 g of ryegrass straw and 14 g of calcium carbonate on a DM basis, twice a day at 9:00 hours and 16:00 hours. Water and mineral block were fed *ad libitum* in the both groups. The spineless cactus was offered by Goto Saboten Co., Ltd. in Kasugai, Japan. The cactus was chopped at around 3 cm square and sun-dried in advance of the experiment. The experiment was conducted using a cross-over design.

2. Sampling and measurements

One experimental term has fourteen days with ten-day adaptation period and four-day trial period. The amount of refused feed was recorded at 9:00 hours and milk yield was measured at 16:00 hours every day in the trial period. A 200 g of milk sample from each animal was collected and kept in -28°C until the analysis. Blood of goats was collected from jugular vein on the final day of each trial period and plasma was collected using a centrifuge at 3000 rpm for 10 minutes.

3. Laboratory analyses

The samples of feed supplied and refused by the goats were analyzed for DM, organic matter, CP, crude fiber (CF), ether extract (EE), crude ash (CA) and nitrogen free extract using the procedures of Association of Official Analytical Chemists (1990). Acid detergent fiber (ADF) and neutral detergent fiber contents of the samples were determined by the method of Van Soest (1973) and Van Soest et al. (1991), respectively. For analysis of Ca and P contents of feed resources, the diet samples were digested by a 1:1 mixture of 60% nitric and perchloric acids. The Ca concentration was determined by flame spectrophotometry and the P content was examined by the colorimetric method of Gomori (1942).

The milk samples were determined for specific gravity and concentration of total solids, fat, protein and ash. The specific gravity was measured by a lactometer calibrated at 15.6°C. A correction applied to a lactometer reading for other temperatures was followed by the correction table as suggested by Prasad et al. (1999). Total solids were obtained by drying in an oven at 105 °C until constant weight was reached. The Gerber method (Pearson, 1976) was used to determine the milk fat concentration. The milk protein concentration was determined through the Kjeldahl method. The analysis of ash contents was held in a muffle at 600 °C. The solids-not-fat was determined by subtracting the lipid contents from the total solids concentration. The lactose concentration was determined by deduction of protein and ash contents from the concentration of solid-not-fat. For analysis of Ca, P and potassium (K) contents of milk, the milk samples were digested by a 1:1 mixture of 60% nitric and perchloric acids. The Ca and K concentrations were determined by flame spectrophotometry and the P content was examined by the colorimetric method of Gomori (1942). Goat beta defensin, lactoferrin, S100A7 and lactoperoxidase as factors relate to antimicrobial substances in goat milk were analyzed by the enzyme immunoassay. Hematocrit values of blood were analyzed with capillary tube and total protein concentration in plasma was measured by spectrophotometer. Urea nitrogen, T-CHO, non-esterified fatty acid, glucose, Ca and inorganic P (IP) in plasma were determined by Nagahama Life Science Laboratory, Oriental Yeast Co., Ltd., Nagahama, Japan. Degree of oxidative stress and antioxidant capacity of the goats were measured by dROMs and BAP tests (Wismerll Co., Ltd., Tokyo, Japan), respectively, using blood plasma.

4. Statistical analysis

The differences in mean values of all the experimental data between Cactus group and Control group were statistically analyzed by student's t-test. All calculations were made using a commercially available computer program (Excel Statistics, SSRI Co., Ltd., Tokyo, Japan)

Results and Discussion

The chemical composition of cactus, barley, wheat bran and ryegrass straw is presented on Table 1. The CP, EE, CF, ADF, CA and Ca of cactus was higher than those of barley. In particular, the contents of EE, CA and Ca in cactus (1.8%, 17.0% and 2.4%, respectively) were around ten times higher than those concentrations in barley. The substitution of the spineless cactus did not decrease feed intake, bodyweight and milk yield ($P > 0.05$) (Table 2.). Thus, the cactus can be considered not to have adverse effects as feed for dairy goats. The chemical composition of milk did not differ significantly between the groups (Table 3). However, the content of milk protein showed a higher mean value with the cactus substitution than without the cactus feeding. The increase of milk protein with cactus feeding was reported in Tunisian local goats (Mahouachi et al., 2011). The disagreement in milk protein result in this study might have been caused by the different breed of goat. The contents of lactoferrin and S100A7 in milk showed higher mean values in Cactus group than in Control group. Lactoferrin is an iron-binding glycoprotein and is considered as a major part of non-specific disease resistance complex in the mammary gland and other epithelial tissues (Schabnacher et al., 1993). S100A7 is one of the antimicrobial peptides that exhibit antimicrobial activity against *Escherichia coli* in humans (Glaser et al., 2005). Although the reasons of this result were not clear, the cactus supplementation might induce some antimicrobial functions and increase the antimicrobial substances in milk. Further investigations are still required. On the other hand, the milk fat concentration showed lower mean value in Cactus group than in Control group. Costa et al. (2009) reported that the decrease of milk fat concentration with the cactus feeding in Alpine goats of which this report supported the results of the present study. The hematocrit value and plasma substances did not show significant differences between the groups (Table 4.). However, the urea nitrogen content in plasma tended to increase with the cactus substitution. The increase of urea nitrogen content might have been caused by accelerated proteolysis in rumen with the cactus feeding. The cactus is rich in mucilage and water soluble carbohydrates such as pectin (De Kock, 2001). The fermentation of pectin in the cactus supplies better rumen environment by increasing production of acetate, which is the main volatile fatty acid produced in the rumen by ruminal microorganisms. On the other hand, the IP concentration in plasma was tended to decrease with the cactus substitution. Since the decrease of IP in plasma may decrease vitamin D absorption in small intestine, countermeasures for preventing the decrease of plasma IP with the cactus feeding may be required. The substitution of spineless cactus for barley should be careful for the feeding rate in concentrates. The concentration of T-CHO was also tended to decrease with the cactus substitution. Stintzing and Carle (2005) reported the decline of T-CHO concentration with cactus intake in humans. This result supported the tendency of lower T-CHO concentration in plasma with the cactus substitution even in goats. The values from dROMs and BAP tests were not significant different between the groups. The 200 g DM substitution of cactus for barley might not affect to the antioxidant capacity in dairy goats.

In conclusion, the 200 g DM substitution of spineless cactus for barley did not affect to milk production and antioxidant capacity. However, the substitution of the spineless cactus for barley to dairy goats has possibilities to promote protein metabolism and antimicrobial functions in milk. Further studies on effects of the increased DM substitution of spineless cactus on milk production, milk quality and antioxidant capacity in dairy goats are necessary.

Table 1. Chemical composition of diets (%).

	Cactus	Barley	Wheat bran	Ryegrass straw
Dry matter	90.2	89.4	89.5	89.8
Organic matter	83.1	98.4	96.1	97.0
Crude protein	13.8	8.4	12.0	5.3
Ether Extract	1.8	0.2	1.8	0.5
NFE	49.6	82.3	75.2	60.5
Crude fiber	18.0	7.6	7.0	30.7
ADF	42.9	15.0	11.3	48.9
NDF	54.0	71.3	56.0	67.7
Crude ash	16.9	1.6	3.9	3.0
Calcium	2.47	0.21	0.22	0.34
Phosphorus	0.42	0.25	0.80	0.12
Potassium	0.77	0.65	0.72	0.63

On a dry matter basis.

NFE : Nitrogen free extract, ADF : Acid detergent fiber, NDF : Neutral detergent fiber

Table 2. Feed intake, body weight change and milk yield of goats.

Group	Cactus	Control
Feed intake (g DM/day/head)	1801.8±167.2	1879.4±117.4
Body weight change (kg/day/head)	48.2±71.1	176.8±75.6
Milk yield (g/day/head)	1092.6±335.0	1127.0±401.6

Table 3. Quality and antibacterial substances in goat milk.

Group	Cactus	Control
Milk quality		
Specific gravity	1.0302±0.0017	1.0297±0.0026
pH	6.56±0.11	6.54±0.13
Total solids (%)	11.35±0.71	11.54±1.76
Solids-not fat (%)	8.51±0.67	8.63±1.74
Fat (%)	2.84±0.33	2.91±0.43
Protein (%)	3.14±0.39	3.10±0.51
Lactose (%)	4.55±0.511	4.75±1.78
Ash (%)	0.818±0.073	0.784±0.069
Calcium (%)	0.076±0.091	0.081±0.066
Phosphorus (%)	0.194±0.184	0.204±0.227
Potassium (%)	0.243±0.255	0.290±0.280
Antimicrobial substances		
Goat beta defensin (ng/mL)	83.3±18.3	95.5±48.8
Lactoferrin (ug/mL)	263.0±188.1	177.8±100.3
S100A7 (ug/mL)	19.2±3.3	11.8±8.4
Lactoperoxidase (U)	6.6±1.6	7.9±3.0

Table 4. Blood components, degree of oxidative stress and antioxidant capacity of goats.

Group	Cactus	Control
Hematocrit	26.4±1.1	27.7±1.6
Blood plasma substances		
Total protein (g/dL)	7.5±0.5	7.6±0.4
Urea nitrogen (mg/dL)	15.2±3.0	12.6±2.7
Total-cholesterol (mg/dL)	78.8±18.6	81.5±12.3
Non-esterified fatty acid (µEq/L)	213.3±174.8	169.0±86.7
Glucose (mg/dL)	62.5±3.3	62.3±3.3
Calcium (mg/dL)	9.3±1.3	8.9±0.9
Inorganic phosphorus (mg/dL)	7.1±4.6	7.5±2.5
Degree of oxidative stress and antioxidant capacity		
dROMs (U.CARR)	141.25±29.84	143.50±15.25
BAP (umol/L)	2785.68±1424.72	3376.65±395.48
dROMs/BAP value	0.07±0.05	0.04±0.00

References

- Association of Official Analytical Chemists (AOAC), 1990. Official Methods of Analysis, 15th edn, AOAC, Arlington, VA
- Clark, S. and J. W. Sherbon. 2000. Genetic variants of alphas1-CN in goat milk: breed distribution and associations with milk composition and coagulation properties. *Small Rumin. Res.* 38: 135-143.
- Glaser, R., J. Harder, H. Lange, J. Bartels, E. Christophers and J. M. Schroder, 2005. Antimicrobial psoriasin (S100A7) protects human skin from *Escherichia Coli* infection. *Nat. Immunol.* 6: 57-64
- Costa, R. G., E. M. BeltrãoFilho, A. N. Medeiros, P. E. N. Givisiez, R. C. R. Queiroga and A. A. Silva Melo. 2009. Effects of increasing levels of cactus pear (*Opuntia ficus-indica* L. Miller) in the diet of dairy goats and its contribution as a source of water. *Small Rumin. Res.* 82: 62–65.
- Costa, R. G., E. M. BeltrãoFilho, R. C. R. Queiroga, M. S. Madruga, A. N. Medeiros and C. J. B. Oliveira. 2010. Chemical composition of milk from goats fed with cactus pear (*Opuntia ficus-indica* L. Miller) in substitution to corn meal. *Small Rumin. Res.* 94: 214–217.
- De Kock, G. C. 2001. The use of *Opuntia* as a fodder source in arid areas of South Africa. In: Mondragon, C. and S. Gonzalez (Eds.) *Cactus (Opuntia spp.) as Forage*. FAO Plant Production and Protection Paper, vol. 169, pp. 73-90.
- Gomori, G., 1942. A modification of colorimetric phosphorus determination for use with the photo electric colorimeter. *J. Lab. Clin. Med.* 27: 955-960.
- MAFF, 2015. Ministry of Agriculture, Forestry and Fisheries, Japan. Available from the URL http://www.maff.go.jp/j/chikusan/kikaku/lin/l_hosin/pdf/m_6.pdf
- Mahouachi, M., N. Atti and H. Hajji, 2011. Use of spineless cactus (*Opuntia ficus-indica* f. inermis) for dairy goats and growing kids: Impacts on milk production, kid's growth, and meat quality, *The Scientific World J.* Article ID: 321567.

- NRC, 1981. National Research Council. Nutrient Requirements of Goats. National Academic Press, Washington, DC.
- Pearson, D. 1976. The Chemical Analysis of Foods, 7th edn. Chemical Publishing, New York, NY.
- Prasad, J. A. K. Tyagi and Neeraj. 1999. Principles and Practices of Animal Nutrition, pp. 192. Kalyani Publishers, Ludhiana.
- Schanbacher, F. L., R. E. Goodman and R. S. Talhouk. 1993. Bovine mammary lactoferrin: implications from messenger ribonucleic acid (mRNA) sequence and regulation contrary to other milk proteins. *J. Dairy Sci.* 76: 3812-3831.
- Stintzing, F. C. and R. Carle, 2005. Cactus stems (*Opuntia* spp.): A review on their chemistry, technology, and uses. *Mol. Nutr. Food Res.* 49: 175–194.
- Van Soest, P. J., 1973. Collaborative study of acid detergent fiber and lignin. *J. AOAC.* 56: 781-784.
- Van Soest, P. J., J. B. Robertson and B. A. Lewis, 1991. Methods for dietary fiber. Neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition. *J. Dairy Sci.* 74: 3584-3597.

Effects of wet soya milk waste supplementation on feed intake and growth performance of goats fed corn stubble silage

Tatsapong, P.¹ R. Thebsawad¹ & S. Hanpok¹

¹Department of Agricultural Science, Faculty of Agriculture, Natural Resources and Environment, Naresuan University, Phitsanulok, 65000, Thailand

Abstract

This experiment was aimed to study the effects of soya milk waste on feed intake and growth rates of post weaning goat were studied. Fifteen Boer x Saanen crossbred male goats, (4-6 month of age and average live weight of 19.4 ± 1.4 kg) were used into three treatment diets under a completely randomized design (CRD) for 60 days study period. All animals were kept individually pen with free access water and mineral block. The goats fed *ad libitum* with corn stubble silage as roughage source. Three dietary treatments were commercial concentrate pellet diet (400 g), wet soya milk waste (800 g) and mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g), respectively. The results showed that feed intake, feed efficiency and body weight gain of goats were significant ($P < 0.05$) different among the diets. Dry matter (DM), organic matter (OM), crude protein (CP), gross energy (GE), neutral detergent fibre (NDF) intake of goat fed commercial concentrate pellet diet were significantly higher than those of goat fed other diet. Average daily gain of goat fed commercial diet was higher ($P < 0.05$) than those of goat fed soya milk waste diet and mixed commercial diet respectively. Feed efficiency was significantly higher in the soya milk waste diet.

Keywords: Soya milk waste, Goat, Feed intake, Saanen crossbred, boer crossbred

Introduction

In Thailand, soybean is made into various foods such as tofu, soymilk, soymilk powder, soy sauce, soy flour, dried tofu. Soya milk residue (waste), waste from soybean milk industry is high in moisture and spoil very quickly in room temperature. Soya milk waste is often considered as waste, which is mostly dumped and burned, and is a potential environmental problem because it is highly susceptible to putrefaction (Rahman et al., 2015; Li et al., 2013). Li et al. (2013) reported that soya milk waste is rich in protein, fiber, fat and trace elements, and is alternative sources of energy and protein for ruminants (Rahman et al., 2014a). The research regarding the use of soya milk waste as ruminant feed on the growth performance of goats has been limited. Although the goat is considered superior to other ruminant species in its utilization of poor quality, high fiber forages and household waste for its body maintenance and production. Therefore, this study investigated the effects of supplementing soya waste on intake, digestibility and growth rate response of growing goats.

Materials and Methods

Fifteen crossbred (Boer x Saanen) male goats, with 19.4 ± 1.4 kg initial body weight (BW), and approximately 4 to 6 months old, were used into three treatment diets under a completely randomized design (CRD) for 60 days study period. All animals were kept individually pens (1x1.2

m) with free access water and mineral block. The goats fed ad *libitum* with corn stubble silage as roughage source. Three dietary treatments were commercial concentrate pellet diet (400 g), wet soya milk waste (800 g) and mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g), respectively. Soya milk waste was supplied by a market women from a local soybean milk processing (home made) at every day, It was contained 315 g CP/kg DM and 11.3 % DM. The commercial concentrate pellet and corn stubble silage were contained 15 and 9 % CP, respectively. Feeds were offered twice a day at 0800 and 1600 h. Amounts of feed offered and refused were recorded daily to estimated feed intake. Subsamples of faeces, feed offered and residues were taken weekly for dry matter determination and dried samples were ground through 1 mm screen and then analyzed for DM, ash, CP and EE (AOAC, 1990) and fiber fraction as described by Van Soest et al. (1991). After the completion of the 60-d feeding trial, a digestibility trial was carried out for 7 d using the total faeces collection method. The body weight of the goat was recorded every 15 days. The data were analysed using the General Linear Model procedure of SPSS (version 17.0).

Results and Discussion

Nutrients intake and digestibility of goats are given in Table 1, showing the goats on the soya waste group had lower intake of all nutrients than those fed commercial pellet. It was similarly to those reported by Rahman et al. (2015; 2014a), who found that supplementation of soya waste in the diet was decreased dry matter intake of goats. This might be due to the higher content of soya waste compared with the commercial pellet diet (89 and 11.3%, respectively). High moisture content of feeds increases the bulkiness of diets, and is negatively related to the capacity of the rumen (Rahman et al., 2014b). DM digestibility was highest on the commercial pellet diet mainly because of higher values for DM intake. While the OM digestibility was highest on soya waste diet may be attributed to increase feed efficiency (Table 2) and utilization of nutrient, particularly soya protein (Rahman et al., 2015).

Table 1. Nutrients intake and digestibility by goats fed soya milk waste and commercial pellet

Parameters	Dietary treatment			SEM	<i>P-value</i>
	T1	T2	T3		
Dry matter intake (DMI)					
Roughage (g/day)	290.07	332.20	324.69	13.87	0.113
Total DMI (g/kgBW ^{0.75})	64.33 ^a	47.06 ^c	54.59 ^b	1.95	0.000
Total DMI (% BW)	2.98 ^a	2.23 ^c	2.52 ^b	0.11	0.001
Nutrients intake (g/day)					
Dry matter (DM)	644.32 ^a	443.15 ^c	549.91 ^b	11.53	0.000
Organic matter (OM)	619.60 ^a	416.02 ^c	521.34 ^b	11.47	0.000
Crude protein (CP)	80.72 ^a	66.21 ^c	75.04 ^b	0.98	0.000
Neutral detergent fiber	312.43 ^a	233.11 ^c	286.70 ^b	8.91	0.000
Acid detergent fiber	159.95 ^a	120.99 ^c	141.84 ^b	8.12	0.018
Gross energy (kcal)	2,321 ^a	1,596 ^c	1,975 ^b	47.67	0.000
Nutrients digestibility (g/day)					
Dry matter (DM)	427.88 ^a	283.31 ^c	359.04 ^b	12.77	0.000
Organic matter (OM)	135.01 ^b	237.05 ^a	224.70 ^a	12.85	0.000
Crude protein (CP)	48.46	44.16	47.01	1.66	0.28
Neutral detergent fiber	182.14	148.86	171.16	9.27	0.07
Acid detergent fiber	60.61	58.39	57.32	8.31	0.96
Digestible energy (Mcal)	1.52 ^a	0.91 ^c	1.28 ^b	0.05	0.000

T1=commercial concentrate pellet diet (400 g); T2= wet soya milk waste (800 g); T3= mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g)

The means within rows with different letters (a,b,c) differ significantly (p<0.05).

Table 2. Average daily gain and feed conversion ratio (FCR) in goat fed soya milk waste and commercial pellet

Parameters	Dietary treatment			SEM	<i>P-value</i>
	T1	T2	T3		
Initial weight (kg)	19.5	18.6	20.2	0.59	0.201
Final weight (kg)	23.9	21.1	23.7	0.89	0.084
Body weight change (kg)	4.4	2.5	3.5	0.64	0.149
Average dairy gain (g/day)	85.42 ^a	41.67 ^c	58.33 ^b	7.96	0.007
Feed efficiency	0.15 ^a	0.21 ^c	0.16 ^b	0.004	0.000
Feed conversion ratio	8.08	11.18	10.27	1.34	0.284

T1=commercial concentrate pellet diet (400 g);T2= wet soya milk waste (800 g); T3= mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g)

The means within rows with different letters (a,b,c) differ significantly ($p < 0.05$).

CP and fiber fraction digestibility were not different among dietary treatments. This might be attributed to the high nutritional characteristics of soya waste, and is positively related to the activity of rumen microbe, and lead to similar body weight change of goats (Rahman et al., 2014b). However, Average daily gain of goat was higher in commercial pellet diet ($P < 0.05$) compared with other treatments (Table 2) and this indicates greater intake of DM and CP than those of goat fed soya milk waste diet and mixed commercial diet respectively (Table 1). The goat fed soya milk waste diet showed lowest growth performance (41.6 g/day) throughout the trial period, which indicate that diets had nutrient content under the threshold level for production requirement of growing goats. This may be due to the low dry matter content (Li et al., 2013). The conclusion, supplementation of locally wet soya milk waste was decreased DM intake and BW gain of goat, and increased feed efficiency.

Acknowledgements

To the Division of Academic service and Asset Management, Naresuan University for financial support.

References

- AOAC. 1990. Official methods of analysis of the association of Official analytical chemists (AOAC). 15th ed. AOAC International, Arlington, Virginia, USA.
- Li S., D. Zhu, K. Li, Y. Yang, Z. Lei, and Z. Zhang, 2013. Soybean curd residue: Composition, utilization, and related limiting factors. ISRN Industrial Engineering, Hindawi publishing corporation. Available online at : <http://dx.doi.org/10.1155/2013/423590>.
- Rahman, M.M, M.R. Rahman, T. Nakagawa, R.B. Abdullah, W.E. Wan Khadijah, and R. Akashi, 2015. Effects of wet soya waste supplementation on the intake, growth and reproduction of goats fed Napier grass. *Anim. Feed Sci. Technol.* 199: 104-112.
- Rahman, M.M, R.B. Abdullah, W.E. Wan Khadijah, T. Nakagawa and R. Akashi, 2014a. Feed intake and growth performance of goats offered Napier grass (*Pennisetum purpureum*) supplemented with concentrate pellet and soya waste. *Sains Malaysiana.* 43: 967-971.
- Rahman, M.M, T. Nakagawa, R.B. Abdullah, W.E. Wan Khadijah and R. Akashi, 2014b. Feed intake and growth performance of goats supplemented with soy waste. *Pesq. Agropec. Bras.*, 49 : 554-558.
- Van Soest, P. J., J. B., Robertson and B. A. Lewis. 1991. **Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition.** *J. Dairy Sci.* 74: 3583-3589.

Effect of high choline levels supplementation on phosphatidylcholine concentration in egg yolk of laying hen

Janist¹, N., P. Srichana², T. Asawakarn³ & S. Kijparkorn^{1*}

¹Department of Animal Husbandry, Faculty of Veterinary Science, Chulalongkorn University, Bangkok 10330, Thailand, ²Charoen Pokphand Group, C.P. Tower, Silom Road, Bangrak, Bangkok 10500, Thailand, ³Vita Co., LTD., Sukhumvit Road, Bangkok 10110, Thailand

Abstract

Eight hundred and forty 44-week-old Isa brown layers were randomly allocated to receive 5 treatment diets with 6 replications each. Treatment diets consisted of choline supplementation at the level of 0, 1000, 1500, 2000 and 2500 mg/kg diet. Birds were raised throughout the 6 weeks of experimental period. Productive performance, egg quality and phosphatidylcholine in yolk, yolk PC, were investigated. There were no significant differences among treatment groups on productive performance and egg quality, except eggshell strength, that was improved at choline level 1000 mg/kg diet group. Yolk PC concentration in choline supplemented groups were higher than control group significantly and the highest was at 1,500 mg/kg diet. In conclusion, the highest level of choline supplementation for synthesizing highest yolk PC concentration was 1,500 mg/kg diet and without any adverse effect on productive performance and egg quality.

Keywords: choline, performance, egg quality, phosphatidylcholine

**Corresponding author: ksuwann1@chula.ac.th*

Introduction

Phosphatidylcholine (PC), lecithin, was a major phospholipid contained in egg yolk (Sourkes, 2004). The advantages of yolk PC were found to promote health benefit by reducing cholesterol absorption in rat (Jiang et al., 2001) and developed long term memory in dull mice (Moriyama et al., 1996). Moreover, PC has been generally used as an emulsifier or liposome in various industries (Palacios & Wang, 2005). Therefore, enhancement of PC concentration in egg yolk will be beneficial for consumer health and industries. Yolk PC synthesized in the liver through 2 pathways, CDP-Choline and PEMT pathway which required choline in reaction (Wurtman et al., 2007). Tsianbe et al. (1988) and Rajalekshmy (2010) showed that yolk PC can increase by supplementation choline at the level of 1,000 mg/kg diet in laying hen. The objective of present study was to find out the highest levels of choline supplementation in laying hen diet to produce highest PC concentration in yolk and productive performance and egg quality were investigated.

Materials and methods

Animals and Diets

Eight hundred and forty Isa brown layers, age 44 weeks old, were randomly allocated into 1 of 5 dietary treatments. Each treatment consisted of 6 replications which were 4 adjacent cages in each replication and 7 birds per cage. Birds were raised in evaporative cooling housing system and had free access to diet and water throughout the 6 weeks of experimental period.

The basal diet was formulated to meet or excess ISA brown requirement (A Hendrix Genetics Company, 2009-10) at the choline level 1,402 mg/kg. Treatment diets were supplemented with choline at the levels of 0, 1,000, 1,500, 2,000 and 2,500 mg/kg diet. Corn in basal diet was substituted with choline chloride (60%) corresponded to each choline supplementation group.

Data collection

Hen-day egg production, egg weight and egg mass were recorded for 1 week and analyzed to ensure the uniformity of experimental units before starting the experiment. Productive performances were recorded throughout 6 weeks. At the end of experiment, 5 eggs per replication were randomly collected to measure egg quality including Haugh units, yolk color, yolk weight, shell weight, shell thickness, shell color and shell strength and egg yolks without vitelline membrane were homogenized and lyophilized for yolk PC analysis

Sample analysis

Sample preparation for phosphatidylcholine (PC) analysis was modified from Rajaleskhmy (2010). PC analysis was performed using an Agilent 1200 series HPLC system (Agilent Technologies, United Kingdom) equipped with auto-samplers model G1329A and quaternary pump model G1311A.

Statistical Analysis

The statistical analysis was performed as a completely randomized design by using one way analysis of variances (ANOVA). Significant difference among each treatment was compared using Bonferroni t-test with using SAS statistical package (SAS Institute Inc, 2002) at significant level of 0.05.

Results

Productive performance

Choline supplementation at high levels had no adverse effect on all parameters of productive performance ($P>0.05$) as shown in Table 1.

Table 1. Effects of high choline levels on productive performance.

Parameter	Supplemented choline (mg/kg diet)					SEM	P-value
	0	1,000	1,500	2,000	2,500		
Egg production (%)	88.8	90.0	89.4	89.2	90.4	0.40	0.743
Egg weight (g)	60.6	61.3	60.6	60.6	60.6	0.21	0.846
Mortality (%)	0.00	0.00	0.60	0.00	0.00	0.12	0.426
Feed intake (g/hen/day)	105	104	106	103	106	0.77	0.707
Egg mass (g/day)	53.8	55.2	54.2	54.1	54.9	0.34	0.731
FCR (g/ g egg mass)	1.96	1.89	1.96	1.90	1.93	0.01	0.187

Egg quality

There were no significant differences in Haugh units, yolk color, yolk weight, shell weight, albumin weight, shell thickness and shell color among treatment groups ($P>0.05$) except for shell strength (Table 2). Shell strength was the highest when choline was supplemented at the level of 1,000 mg/kg diet and significantly differed from control group.

Table 2. Effects of high choline levels on egg quality.

Parameter	Supplemented choline (mg/kg diet)					SEM	P-value
	0	1,000	1,500	2,000	2,500		
Haugh units	90.9	90.1	90.2	90.4	90.3	0.46	0.989
Yolk color	9.36	9.40	9.35	9.23	9.63	0.05	0.184
Yolk weight(g)	15.6	15.3	15.2	15.3	15.2	0.10	0.704
Shell weight (g)	5.91	5.86	5.83	5.68	5.81	0.04	0.515
Albumin weight (g)	40.6	40.6	40.1	40.3	41.5	0.27	0.586
Shell thickness(mm.)	0.37	0.39	0.39	0.37	0.38	0.00	0.136
Shell color ¹	54.6	56.3	56.3	55.6	55.8	0.27	0.263
Shell strength (Kgf) ²	3.91 ^B	4.55 ^A	4.37 ^{AB}	4.09 ^{AB}	4.29 ^{AB}	0.06	0.008

¹shell color was ranked using L-value

²kgf = kilogram force

Phosphatidylcholine in yolk

The average yolk PC concentration in control group was 158.2mg/g while yolk PC concentrations of diet supplemented with choline at 1000, 1500, 2000 and 2500 mg/kg were 164.3, 169.2, 168.0 and 166.4 mg/g respectively. Yolk PC concentration of all treatment groups were higher than control group significantly ($P<0.0001$). The highest yolk PC concentration was in diet supplemented with choline at 1500mg/kg.

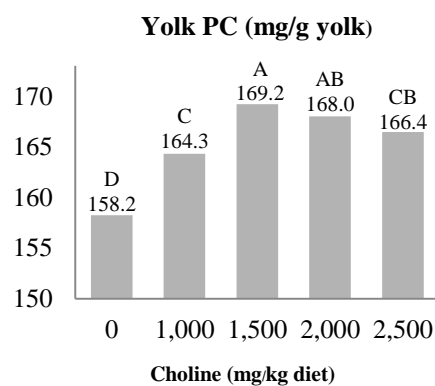


Figure 1: Effects of high choline levels on yolk phosphatidylcholine concentration.

Discussion

In general, layer can sufficiently synthesize choline to meet its metabolic requirement but larger amount will be required to accumulate in yolk in the form of phosphatidylcholine (PC) (Leeson & Summers, 2001). Crawford et al. (1968a,b) showed that 595 mg choline/kg diet was sufficient to maintain productive performance in layer and the excess choline from layer's requirement would be stored in liver as PC. Layer eliminated PC primarily through the egg due to none of choline was

reported in excreta and carcass of layer. In present study, the nutrients in all experimental diets were formulated based on breed's recommendation except choline, which was supplemented over requirement, belong to treatment groups. The high levels of choline supplementation up to 2500 mg/kg diet were not showed any adverse effect on egg production. This choline level was lower than the previous research (Zhai et al., 2013), that egg production was not interfered.

For egg quality, Yolk lipids were synthesized in the liver and transferred to the ovarian follicles by lipoproteins which mostly covered by phosphatidylcholine (Alvarenga et al., 2011). Zhai et al. (2013) who supplemented choline at the level of 0-6,800 mg in layer diet found no significant difference in Haugh units, yolk color, shell thickness and shell strength. In current study, it was interesting that supplementation of choline at 1,000 mg choline/kg diet significantly improved eggshell strength compared to control groups ($P=0.008$) while at higher levels (1,500-2,500 mg/kg diet) was not significantly different in contrast with Zhai et al. (2013). Incorporate with no previous research about the effect of choline on shell strength was documented to explain this phenomenon. Thus, further investigation is needed.

Tsiangbe et al. (1988) and Rajalekshmy (2010) indicated that choline at the level of 1,000 mg/kg diet significantly increased PC in the egg yolk. The highest level of PC concentration in yolk in current study was detected in 1,500 mg choline/kg diet. The increase of yolk PC due to the excess of choline above normal layer requirement were used to synthesize PC from CDP-choline pathway and provided methyl group to phosphatidylethanolamine (PE) until it become PC in liver and transferred to yolk (Wurtman et al., 2007). However, at the level over 1500 mg choline/kg diet did not show any beneficial effect. There were 2 possible explanations: 1) the excess PC in liver was converted to phosphatidylserine by phospholipase D enzyme (Yamane et al., 1991) or 2) PC was changed to PE by unknown mechanism in liver, which was reported by Nishijima et al. (1986). The same author reported that PE significantly increased with obtaining exogenous [^{32}P] phosphatidylcholine in cultured cells. If either or both hypothesis is true, the PE concentration in yolk should be increased. Therefore further investigation needs to be conducted and PE in yolk should be analyzes to confirm this hypothesis.

Conclusions

The highest choline level supplementation in laying hen diet to synthesize highest phosphatidylcholine concentration in yolk was at 1,500 mg/kg diet without adverse effect on productive performance and egg quality.

Acknowledgements

This study was funded by 90th Anniversary Ratchadaphiseksomphot Endowment Fund and the Faculty of Veterinary Science, Chulalongkorn University, Bangkok, Thailand. The authors would like express deep gratitude to all staffs in Feed technology Department and Development Center of Charoen Pokphand Foods PCL for their help and support all field trial and laboratory analysis facilities. The authors also wish to thanks the laboratory staffs of Vita Co., LTD. for their assistance to generate PC condition for HPLC analysis. Finally, my higher education was not possible without supporting by academic scholarship from the Graduate School, Chulalongkorn University to commemorate the 72nd anniversary of his Majesty King Bhumibala Aduladeja.

References

- A Hendrix Genetics Company, 2009-10."ISA nutrition management guide".(online). Available: <http://www.hendrix-poultry.nl>. Accessed June 2,2015.
- Alvarenga, R.R., M.G. Zangeronimo, L.J. Pereira, P.B. Rodrigues and E.M. Gomide, 2011. Lipoprotein metabolism in poultry. *World Poult. Sci. J.* 67: 431-440.
- Crawford, J.S., M. Griffith, R.A. Teekell and A.B. Watts, 1968a.Choline requirement and synthesis in laying hens.*Poult. Sci.* 620-626.
- Crawford, J.S., R.A. Teekell and A.B. Watts, 1968b.Absorption and Deposition of choline in laying hens.*Poult. Sci.* 627-633.
- Jiang, Y., S.K. Noh and S.I. Koo, 2001. Egg phosphatidylcholine decreases the lymphatic absorption of cholesterol in rats. *J.Nutr.* 131: 2358-2363.
- Leeson, S., and J.D. Summers, 2001.Nutrition of the chicken. 4th ed. M.L. Scott and Associates, Thaca, N.Y. 292-311.
- Moriyama, T., U. Kayoko, Y. Matsumoto, S.Y. Chung, E. Uezu, S. Miyaki, U. Miyoko, M. Yasunobu, K. Toyohiko, T. Tatsuo and Shigeru, 1996.Effects of dietary phosphatidylcholine on memory in memory deficient mice with low brain acetylcholine concentration. *Life Sci.* 58(6): 111-118.
- Nishijima, M., O. Kuge and Y. Akamatsu, 1986.Phosphatidylserine biosynthesis in cultured chinese hamster. *J. Biol. Chem.* 261(13): 5795-5798.
- Palacios, L.E., and T. Wang, 2005.Extraction of Egg-Yolk lecithin. *J. Am. Oil Chem. Soc.* 82(8): 565-569.
- Rajalekshmy, P.K., 2010. Effects of dietary choline, folic acid, vitamin B₁₂ on laying hen performance, egg components and egg phospholipid composition. (dissertation) University of Nebraska. pp130.
- SAS, 2002. SAS/SAT Guide for personal computers. Version 9.00 ed. SAS Inst., Inc., Carry, NC.
- Sourkes, T.L., 2004. The discovery of lecithin, the first phospholipid. *Bull Hist Chem.* 29(1): 9-15.
- Tsiangbe, V.K., M.E. Cook, A.E. Harper and M.L. Sunde, 1988. Alternations in phospholipid composition of Egg yolks from laying hens fed choline and methionine- supplemented diets. *Poult. Sci.* 67: 1717-1724.
- Wurtman, R.J., M. Cansev and I.H. Ulus, 2007.Choline and its products acetylcholine and phosphatidylcholine.Springer - Verlag Berlin Heidelberg.3-32.
- Yamane, T., L.R. Juneja, E. Taniguahi, S. Shimizu, 1991.Increasing productivity by removing choline in conversion of phosphatidylcholine to phosphatidylserine by phospholipase D. *J.Biosci. Bioeng.* 73(5): 357-361.
- Zhai, Q.H., X.F. Dong, J.M. Tong, Y.M. Guo and Y.E. Bao, 2013.Long-term effects of choline on productive performance and quality of brown-egg laying hens.*Poult. Sci.* 92: 1824-1829.

Effect of Coconut Oil Supplementation on Meal Pattern, Feed Intake, and Milk Yield in Early Lactating Crossbred Dairy Goat

Ho¹, T.K., S. Thammacharoen² & S. Chanpongsang^{*1}

¹Department of Animal Husbandry, ²Department of Physiology, Faculty of Veterinary Science, Chulalongkorn University, Bangkok 10330, Thailand

Abstract

Coconut oil is one of coconut by products found in Thailand and it contains high amount of medium chain fatty acid. Feeding coconut oil as fat supplement improved lactation performance in dairy cattle. However, there was no information of coconut oil supplementation in dairy goat. Therefore, the current study aimed at investigating the effect of coconut oil supplement on feed intake, lactation performance and meal pattern in crossbred lactating dairy goat during early lactation. Six crossbred goats were used in the current experiment. Two week before parturition, animals were randomly divided into two groups (n=3). Treatments were control diet (44% corn silage and 56% concentrate) and 2% coconut oil supplementation diet (44% corn silage and 54% concentrate). Both diets are isoenergetic and isonitrogenous, each goat was fed ad libitum twice daily as total mix ration with free access to water for 5 weeks. Body weight, dry matter intake (DMI), DMI per body weight, milk yield and feed efficiency were not significant difference between groups. However, the results from meal pattern revealed the effect of coconut oil supplement. Meal duration was longer and meal size was greater in coconut oil supplemented group than in control group. The present experiment revealed that supplementation with 2% coconut oil in dairy goats during early lactation could not influence DMI and milk yield. However, coconut oil supplementation did change the meal pattern. The latter information suggested that coconut oil supplementation may influence the palatability and change eating behavior in dairy goat.

Keywords: coconut oil, dairy goat, feed intake, milk yield, meal pattern.

**Corresponding author: somchai.c@chula.ac.th*

Introduction

Among the three periods of lactation, the early phase of lactation is the most critical and important because this period produces a significant amount of milk. An increment of milk production during this period together with feed deprivation is the main factors lead negative energy status in the dairy animal (Herdt et al., 1988; Grummer et al., 1990). To improvement lactation performance during early lactation, the researchers try to find out variable diet strategies. Dietary fat supplementation has been suggested because it was the direct way to increase both energy intake and utilization efficiency (Morand-Fehr et al., 1982; Chilliard, 1993). However, diet supplement with fat reduced DMI (Chilliard, 1993). In addition to DMI, eating behavior could also be described by meal pattern that includes meal size, duration and frequency, eating rate, intermeal interval (Abijaoude et al., 2000; Allen, 2000; Harvatine & Allen, 2006). Analysis of meal pattern is necessary to explain the regulation of food intake. According to Harvatine and Allen (2006), dairy cows fed with fatty acid decreased DMI and meal size and increased meal interval. Interestingly, these experiments revealed that fat supplementation did change to the meal pattern.

Coconut oil is one of coconut by products and has been used as fat supplement in dairy cattle (Storry et al., 1974; Lee et al., 2011; Hollmann & Beede, 2012). Coconut oil supplementation has been shown to influence DMI base on palatability or acceptability (Hollmann & Beede, 2012). In dairy cattle, supplement 2.3% coconut oil decreased DMI (Lee et al., 2011). However, Hollmann & Beede (2012) found that coconut oil supplementation at 1% apparently increased DMI, while 2 to 4% coconut oil supplementation clearly decreased DMI. Those results show that the effect of coconut oil supplementation on DMI apparently depends on the level of coconut oil in the formulation. Although, many study observed about effect of coconut oil supplement on feed intake and milk yield, but no study have observed on meal pattern. Thus, the objective of this experiment was determine effect of coconut oil supplementation on meal pattern, dry matter intake and milk yield in dairy goat during early lactation period.

Materials and methods

Six crossbred dairy goats were used in this experiment. The goats were randomly assigned to two treatments of experiment: 1) control group (without coconut oil supplement), 2) coconut oil group (with coconut oil supplement). The coconut oil was extracted by heated (hot method) processes (Mariana et al., 2009). The diet as total mix ration (TMR) offered 2 times per day and water were provided ad libitum. About two weeks before parturition six animal were randomly divided into two groups (n=3). The experimental period was last for 35 days after parturition and meal pattern would be determined in day 25 of the experiment.

The TMR was formulated according to NRC recommendation (NRC, 1981). Corn silage was used as roughage sources in this experiment. The TMR contained 44% corn silage and concentrate (56% for control and 54% for coconut oil diet). In coconut oil diet, 2% coconut oil was supplemented in concentrate. Both diets were isoenergetic and isonitrogenous. The ingredients and chemical compositions of diet in control and coconut oil group were presented in the table 1.

Table 1. Ingredients and chemical composition of experimental diets.

Diet composition	Control	Coconut oil
Ingredients, %		
Corn silage	44	44
Cassava	3.26	8.8
Soybean meal	19.62	20.24
Coconut oil	0	2
Molasses	3.69	4.97
Premix	0.5	0.5
Corn meal	25.78	7.04
Rice bran	2.25	11.25
Limestone	0.9	0.9
Chemical composition, %		
DM	35.16	35.12
CP	16.73	16.72
NE _L , Mcal/kg DM	1.66	1.67
Ca	0.52	0.54
P	0.36	0.49

Control = no added fat and 44: 56 corn silage: concentrate ratio; Coconut oil = 2% coconut oil and 44: 54 corn silage: concentrate ratio.

Two feed samples were collected once daily throughout the experiment. First sample was dried in 105°C ovens for overnight to determine dry matter. The second sample was kept at -20°C to determine nutrient compositions by proximate analysis (AOAC, 1990), neutral detergent fiber (NDF) and acid detergent fiber (ADF) by the procedure of Van Soest et al., (1991).

After parturition, milk yield was collected by hand milking and recorded twice daily at 07:30 and 15:30 and weighed.

Meal pattern was recorded continuously for 24h using digital balance equipped to data processing software (PBA 665 & Weigh Term 231G, Mettler Toledo, Zürich, Switzerland). Digital balances were fixed under the feed containers of goats. Balance with feed containers was protected by wood boxes. The actual weight of feed containers was checked and recorded automatically by a personal computer in each minute. Parameters recorded were meal size, meal duration, meal frequency and inter-meal interval (Fig. 1). Meals were defined as feed removals exceeding 5g that are separated by at least 15 minutes of non-feeding (Rossi et al., 1998).

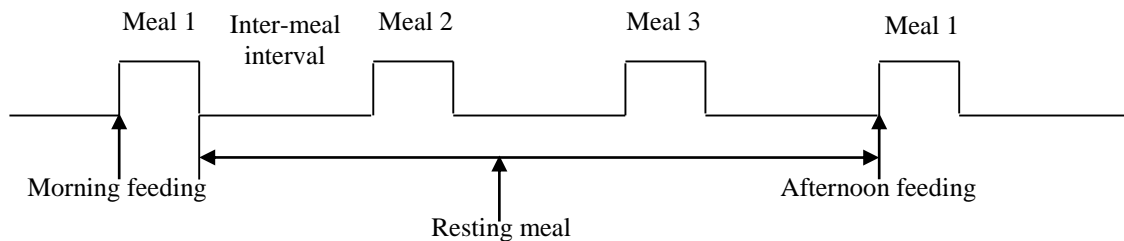


Figure 1. The diagram determined the meal patterns and resting meal.

All data were reported as the mean value \pm SEM. Statistical significant difference between groups was determined by unpaired t-test. The meal frequency between treatments was analyzed by Wilcoxon rank sum test. Significance was declared at $P < 0.05$.

Results and discussion

There were no different in bodyweight, DMI, DMI/BW, water intake and water intake/DMI (Table 2). However, depression of DMI had been proved in previous study when supplement coconut oil in TMR (Lee et al., 2011; Hollmann & Beede, 2012). Martinez et al., (2013) had mentioned that most study on dairy goat did not show the negative effect in DMI when supplement fat in diet and was similar to the result of the present study. The present study show that DMI and DMI/BW did not differ between treatments. This may be due to indifference of nutrient digestibility (unpublished data). Pantoja et al., (1994) reported that there was a linear decrease of organic matter intake in relation with a linear reduction of NDF digestibility and differ with the present study. The difference between that study and present study could be related with the level of unsaturation of fat supplement.

Table 2. Effect of coconut oil supplement on body weight, DMI, DMI/body weight, milk yield, feed efficiency, water intake and water intake/DMI (mean \pm SEM).

	Treatments		P
	Control	Coconut oil	
Body weight (kg)	33.56 \pm 0.84	32.07 \pm 3.59	0.71
Dry matter intake (kg DM/day)	1.18 \pm 0.18	1.27 \pm 0.13	0.70
Dry matter intake/body weight (g/kg BW)	35.05 \pm 3.82	38.97 \pm 3.83	0.51
Milk yield (kg/day)	1.75 \pm 0.28	1.46 \pm 0.27	0.50
Feed efficiency (kg milk yield/kg DMI)	1.43 \pm 0.01	1.11 \pm 0.15	0.10
Water intake (kg/day)	3.40 \pm 0.85	3.31 \pm 0.41	0.93
Water intake/DMI (kg water intake/kg DMI)	6.89 \pm 1.73	6.75 \pm 0.91	0.95

Control = no added fat and 44: 56 corn silage: concentrate ratio; Coconut oil = 2% coconut oil and 44: 54 corn silage: concentrate ratio.

Milk yield of the coconut oil group was lower than control group (16%), but it was not significant difference ($P > 0.05$; table 2). Martinez et al., (2013) suggested that lower milk yield when high level of fat supplement in diet could be due to negative influence on DMI and digestion. However, the present experiment unchanged either DMI or digestibility (unpublished data) and followed by unchanged milk yield. Hollmann & Beede (2012) reported that dairy cows supplement with 5% animal fat blend reduced milk yield. This differed with the result of this experiment, due to difference in degree of dietary fat.

Feed efficiency in this experiment was shown in table 2. Coconut oil reduced feed efficiency due to higher feed intake and lower milk yield, but it did not differ between treatments during experiment.

Meal size, meal frequency and meal duration in day time had higher than night time, while inter-meal interval was longer than at the night time (Table 3). This result was similar with previous study of Senn et al., (1995) when 85% of meals observed during light phase. It indicated that dairy goat ate mainly in day time and spent night time for resting.

Table 3. Effect of coconut oil on meal pattern (mean \pm SEM).

		Treatments		P
		Control	Coconut oil	
Meal size (kg/meal)	Day	0.09 \pm 0.02	0.16 \pm 0.02	0.09
	Night	0.02 \pm 0.01	0.06 \pm 0.01	0.07
	Whole day	0.06 \pm 0.01	0.11 \pm 0.01	0.04
Meal duration (min)	Day	31.00 \pm 3.46	65.00 \pm 3.51	0.01
	Night	19.33 \pm 2.19	38.00 \pm 1.00	0.01
	Whole day	50.33 \pm 3.84	103.00 \pm 3.06	0.01
Meal frequency (times)	Day	12.00 \pm 0.33	8.00 \pm 0.88	0.05
	Night	8.00 \pm 2.08	2.00 \pm 0.33	0.02
	Whole day	20.00 \pm 1.76	10.00 \pm 1.15	0.05
Inter-meal interval (min)	Day	28.67 \pm 2.40	32.67 \pm 8.76	0.68
	Night	78.33 \pm 35.89	84.00 \pm 20.60	0.90
	Whole day	53.50 \pm 18.84	58.33 \pm 7.05	0.82

Control = no added fat and 44: 56 corn silage: concentrate ratio; Coconut oil = 2% coconut oil and 44: 54 corn silage: concentrate ratio.

Coconut oil supplement in this study did not affect DMI, but it influenced meal pattern. Coconut oil group had significantly greater meal size and longer meal duration but lower meal frequency than control group (Table 3). Meal size of coconut oil diet had more weight than control diet during day time, night time and whole day, but the significant has only in whole day ($P < 0.05$). The meal duration was increase together with meal size when coconut oil supplement in dairy goat in this experiment. Moreover, coconut oil significantly increased meal duration during day, night time and whole day ($P < 0.05$). The increased meal size and meal duration may be come from the higher palatability of coconut oil supplement (Hollmann & Beede, 2012). Although, Harvatine & Allen (2006) reported that meal size had unchanged when supplement 5% saturated fatty acid in dairy cattle. In this study, coconut oil contained mainly saturated fatty acid, but the lower level of fat in diet (2% DM) may explain the higher meal size of coconut oil group than control.

Meal size of the first and resting meal was not difference between two treatments in both morning and afternoon feeding (Table 4). However, animal supplement with coconut oil spent more time for eating than control group in the first morning meal ($P < 0.05$). The eating rate in the first and resting meal was faster in control group than coconut oil group, but this difference was not significant ($P > 0.05$).

Table 4. Effect of coconut oil on first and resting meal size, meal duration and eating rate (mean \pm SEM).

		Treatments		P
		Control	Coconut oil	
Meal Size (kg/meal)	1 st Morning meal	0.32 \pm 0.15	0.44 \pm 0.08	0.51
	1 st Afternoon meal	0.29 \pm 0.08	0.32 \pm 0.05	0.76
	Resting morning meal	0.04 \pm 0.01	0.05 \pm 0.01	0.56
	Resting afternoon meal	0.07 \pm 0.01	0.21 \pm 0.06	0.09
Meal duration (min.)	1 st Morning meal	39.00 \pm 8.66	140.67 \pm 6.01	0.01
	1 st Afternoon meal	51.67 \pm 15.86	96.67 \pm 17.07	0.13
	Resting morning meal	25.67 \pm 2.60	40.00 \pm 2.65	0.02
	Resting afternoon meal	37.33 \pm 7.88	75.00 \pm 15.00	0.09
Eating rate (g/min)	1 st Morning meal	7.48 \pm 1.90	3.17 \pm 0.70	0.10
	1 st Afternoon meal	6.03 \pm 1.09	3.48 \pm 0.66	0.12
	Resting morning meal	1.72 \pm 0.21	1.27 \pm 0.16	0.16
	Resting afternoon meal	1.54 \pm 0.19	2.83 \pm 0.95	0.32

Control = no added fat and 44: 56 corn silage: concentrate ratio; Coconut oil = 2% coconut oil and 44: 54 corn silage: concentrate ratio.

Conclusion

In this experiment, 2% coconut oil had no effect on body weight, DMI, DMI/body weight, milk yield, feed efficiency and water intake, but it did increase meal size and meal duration in dairy goat during early lactation. That data suggested coconut oil can change feeding behavior in dairy goat.

Reference

- Abijaoude, J. A., P. Morand-Fehr, J. Tessier, P.H. Schmidely and D. Sauvant, 2000. Diet effect on the daily feeding behaviour, frequency and characteristics of meals in dairy goats. *Livest. Prod. Sci.* 64: 29–37.
- Allen, M.S., 2000. Effects of diet on short-term regulation of feed intake by lactating dairy cattle. *J. Dairy. Sci.* 83: 598-624.
- Association of Official Analytical Chemists (AOAC), 2000. *Official Methods of Analysis*. 17th ed. Association of Official Analytical Chemists Inc., Washington D.C, the United States.
- Chilliard, Y., 1993. Dietary fat and adipose tissue metabolism in ruminants, pigs, and rodents: a review. *J. Dairy. Sci.* 76: 3897-3931.
- Grummer, R. R., S. J. Bertics, D. W. Lacount, J. A. Snow, M. R. Dentine and R. H. Stauffacher, 1990. Estrogen induction of fatty liver in dairy cattle. *J. Dairy. Sci.* 73: 1537-1543.
- Harvatine, K.J., and M.S. Allen, 2006. Effects of fatty acid supplements on feed intake, and feeding and chewing behavior of lactating dairy cows. *J. Dairy. Sci.* 89: 1104 – 1112.
- Herd, T. H., T. Wensing, H. P. Haagsman, L. M. van Golde and H. J. Breukink, 1988. Hepatic triacylglycerol synthesis during a period of fatty liver development in sheep. *J. Anim. Sci.* 66: 1997-2013.
- Hollmann, M. and D.K. Beede, 2012. Comparison of effects of dietary coconut oil and animal fat blend on lactational performance of Holstein cows fed a high-starch diet. *J. Dairy. Sci.* 95: 1484-1499.

- Lee, C., A.N. Hristov, K.S. Heyler, T.W. Cassidy, M. Long, B.A. Corl and S.K. Karnati, 2011. Effects of dietary protein concentration and coconut oil supplementation on nitrogen utilization and production in dairy cows. *J. Dairy. Sci.* 94: 5544-5557.
- Marina, A. M., Y. B. Che Man and I. Amin, 2009. Virgin coconut oil: emerging functional food oil, *Trends in Food Science & Technology.* 20: 481-487.
- Martinez, M. L. A., H. M. Pérez, A. L. M. Perez, P. D. Carrion, S. A. I Garzon and G. C. Gomez, 2013. Fat addition in the diet of dairy ruminants and its effects on productive parameters. *Revista Colombiana de Ciencias Pecuarias.* 26: 69-78.
- Morand-Fehr, P., Y. Chilliard and D. Sauvant, 1982. Goat milk and its compounds: Secretory mechanisms and influence of nutritional factors. *Proc 3 Int Conf on Goat Production and Disease Tucson AZ.* pp 113-121.
- National Research Council (NRC), 1981. Nutrition requirements of goats. 2nd ed. National Academy Press. Washington, D.C, the United States.
- Pantoja, J., J. L. Firkins, M. L. Eastridge and B. L. Hull, 1994. Effects of fat saturation and source of fiber on site of nutrient digestion and milk production by lactating dairy cows. *J. Dairy. Sci.* 77:2341-2356.
- Rossi, R., E. Del Prete, J. Rokitzky and E. Scharrer, 1998. Effects of a high NaCl diet on eating and drinking pattern in pygmy goats. *Physiol. Behav.* 63: 601-604.
- Senn, M., B. Dürst, A. Kaufmann and W. Langhans, 1995. Feeding patterns of lactating cows of three different breeds fed hay, corn silage, and grass silage. *Physiol. Behav.* 58: 229-36.
- Storry, J.E., P.E. Brumby, A.J. Hall and V.W. Johnson, 1974. Response of the lactating cow to different methods of incorporating casein and coconut oil in the diet. *J. Dairy. Sci.* 57: 61-67.
- Van Soest, P.J., J.B. Robertson and B.A. Lewis, 1991. Symposium: carbohydrate methodology, metabolism and nutritional implication in dairy cattle: methods for dietary fiber and non starch polysaccharides in relation to animal. *J. Dairy. Sci.* 74: 3585-3597.

The effect of the use of cassava leaves silage in concentrate on goat performance

Hartutik ^{*1}, S.Chuzaemi² & Mashudi³

Animal Nutrition Department, Faculty of Animal Husbandry, Brawijaya University, Malang, East Java, Indonesia-65145

Abstract

The aim of this study was to know the optimal level of cassava leaves silage in concentrate for goat ration. The materials used were Elephant Grass (EG), Concentrate (C), cassava leaves silage with cassava waste as additive (CLS) and 15 heads goats. The experimental design used was Randomized Block Design (RBD) with 5 treatments namely R0 (EG ad libitum + 50% C), R1 (EG ad libitum + 37.5% C + 12.5% CLS), R2 (EG ad libitum + 25% C + 25% CLS), R3 (EG ad libitum + 12.5% C + 37.5% CLS), R4 (EG ad libitum + 50% CLS) and 3 replication as blocks. The ration contained 13% CP. The ratio between forage and concentrate was 50%: 50% as dry matter (DM). Variables measured were feed intake, digestibility, N retention and daily body weight gain. The results of the research showed that the use of CLS in the concentrate on goat performance did not give significantly difference ($P>0.05$) on DMI, OMI, CPI, CPD, DDMI, DOMI, DCPI, N retention and daily body weight gain, but there were significantly difference ($P<0.05$) on DMD and OMD. The highest value of daily body weight gain was R2 treatment (97.20 g/h/d). It is suggested that cassava leaves silage using cassava waste as additive could be used in concentrate up to 50% (R2) in goat ration.

Key word: cassava leaves silage, cassava waste, goat performance

Introduction

Cassava leaves, as agricultural by-products, contained high crude protein (CP) which is around 16.70% to 39.90% and total digestible nutrients around 58.85% (O'Hair, 1995). The main products of cassava, such as fresh or dried tubers are mostly used for human consumption, while the by-products e.g. cassava leaves, cassava waste and cassava peels can be used as animal feed. The utilization of cassava tubers as cattle feed is estimated 350,000 tons/year. The use of dried cassava tubers as concentrate in dairy cattle ration (85% of total concentrate) will increase milk production and milk quality (Wanapat and Petlurn, 2004). HCN content in cassava, cassava leaves as well as cassava by-products can be reduced by silage treatment. Dried cassava and cassava waste can accelerate the formation of lactic acid leading to the decrease in pH in ensilage process. Cassava leaves silage contains high protein which is expected to be used as a substitution of commercial concentrates which is generally expensive. Therefore, this study aims to determine the optimal level of use of cassava leaves silage instead of commercial concentrate in the ration on performance of goat.

Materials and Methods

The animal used in this study was 15 heads of Ettawah Crossbred goat with body weight 23-30 kg, aged 10-14 months. The feed given were Elephant Grass (*Pennisetum purpureum*, EG) (CP 10,12%), cassava leaves silage (CLS) by using cassava waste (10% of DM and 17.10% of CP) as additives, concentrate (C) with 14.91% of CP content consisting of a pollard, rice bran, coconut meal, cotton seed meal and mineral. The experimental design used was Randomized Block

Design (RBD) with 5 treatments namely R₀ (EG *ad libitum* + 50% C), R₁ (EG *ad libitum* + 37.5% C + 12.5% CLS), R₂ (EG *ad libitum* + 25% C + 25% CLS), R₃ (EG *ad libitum* + 12.5% C + 37.5% CLS), R₄ (EG *ad libitum* + 50% CLS) and 3 replication as blocks based on initial body weight. The ration contained 13% CP. The ratio between forage and concentrate was 50% : 50% as dry matter (DM). Feed given as DM base was 3% of body weight. Cassava waste is cassava by-product produced in tapioca processing. Variables measured were feed intake, digestibility, N retention and daily body weight gain. Analysis of data using analysis of covariance, if there is any difference between treatments followed by Least Significance Difference (LSD) test.

Result and Discussion

Table 1 show that the use of CLS instead of concentrate in the diet did not affect significantly ($P > 0.05$) DMI, OMI, CPI, DMD, OMD and CPD. However, the use of 0.75% CLS in the concentrate (R₃) has the highest intake, it is due to the content of CP of CLS (17.10%) was higher than the concentrate (14.91%). The higher CLS the higher CP content leading to the higher intake. Hartutik *et al* (2010) reported that the higher amount of silage substitution in the ration, the DMI, OMI, and CPI tend to increase. R₃ treatment showed that the use of silage is optimum, while the R₄ decreased the quality of ration; it is because of higher accumulation of HCN. The use of CLS to substitute concentrate in the ration showed significantly difference ($P < 0.05$) on DMD and OMD but not significant ($P > 0.05$) on CPD.

The difference of DMD and OMD, it is because of the different of DM and OM content. R₀ has the highest digestibility and consists of EG and concentrates which concentrates have greater digestibility than silage. The digestibility of R₀ was not significantly different compared R₁, R₃, and R₄. It is indicated that CLS can substitute concentrates. Hartutik *et al* (2012) reported that *in vitro* DMD and OMD of CLS were 61.91 and 76.73%, respectively and they were higher than CLS without any additives (59.96 and 70.11%). Table 1 showed that the use of CLS has no significant effect ($P > 0.05$) on the N retention. This is because the feed intake and digestion of CP (N) was not different among the treatments. In R₂, N retention was low (0.76 g / kg MBW/d) but DBWG was the highest. This indicates that this treatment give more efficient effect. Seng *et al.* (2010) reported that the BWG of goats fed cassava silage was higher than those fed urea molasses block, it were 60.4 ± 9.01 g / d and 6.70 ± 8.43 g / d respectively. Kusmartono *et al* (2010) reported that the DBWG and N retention on fatty-tailed sheep fed elephant grass and extract of cassava leaves were 62.79 g / d and 15.6 g / d respectively which are higher than the control treatment (EG and concentrates).

Table 1. Feed intake of nutrients, digestibility and digestible nutrients

Variables	Treatments					
	R ₀	R ₁	R ₂	R ₃	R ₄	
Nutrients intake (g/kg metabolic weight/d)						
- DMI (Dry Matter Intake)	113.88 ^a	95.50 ^a	113.93 ^a	117.53 ^a	98.43 ^a	ns
- OMI (Organic Matter Intake)	100.72 ^a	85.07 ^a	100.87 ^a	103.64 ^a	86.61 ^a	ns
- CPI (Crude Protein Intake)	13.78 ^a	12.51 ^a	14.13 ^a	15.16 ^a	14.02 ^a	ns
- DMI (Dry Matter Intake, %BW)	4.86	4.03	4.78	4.98	4.32	ns
Nutrients Digestibility (%)						
- DMD (Dry Matter Digestibility)	76.01 ^b	74.05 ^b	70.80 ^a	75.36 ^b	72.69 ^{ab}	s
- OMD(Organic Matter Digestibility)	78.37 ^b	76.46 ^b	73.10 ^a	77.21 ^b	74.76 ^{ab}	s
- CPD (Crude Protein Digestibility)	80.49 ^a	77.97 ^a	74.67 ^a	77.44 ^a	73.64 ^a	ns
Digested Nutrien Intake (g)						
- DMDI (DMD Intake)	84.02 ^a	76.69 ^a	80.43 ^a	86.90 ^a	78.83 ^a	ns
- OMDI (OMD Intake)	76.57 ^a	70.10 ^a	73.20 ^a	78.42 ^a	71.36 ^a	ns
- CPDI (CPD Intake)	10.70 ^a	10.13 ^a	10.45 ^a	11.46 ^a	10.85 ^a	ns
N retention (g/kg MBW/d)	0.82 ^a	0.81 ^a	0.76 ^a	0.72 ^a	0.81 ^a	ns
DBWG (Body Weight Gain, g/h/d)	66.00 ^a	71.40 ^a	97.20 ^a	73.50 ^a	63.86 ^a	ns

Conclusion

CLS can replace the concentrates in the diet to 100%, but the substitution of 50% CLS in concentrate (R₂) tends to perform the highest BWG (97.20 g / head / day). Thus, it is proposed that the use cassava leaves silage with cassava waste as additive in 50% of DM ration can replace concentrates in the diet even on 100%.

References

- Hartutik, Soebarinoto, S.Chuzaemi and Mashudi. 2012. The Effect of Additives Addition in Cassava (*Man/hot escu/enta, Crantz*) Leaves Silage on *In vitro* Digestibility and Gas Production. World Academy of Science, Engineering and Technology. Issue 66, June 2012, Paris France.Pp 1884-1887
- Kusmartono, S. Chuzaemi, Hartutik, Mashudi. 2010. Ekstraksi condensed Tannin (CT) dari Tanaman Semak dan Pohon dan Pengaruh Penambahannya dalam Pakan Konsentrat Terhadap Produksi dan Kualitas Karkas ternak Domba Ekor Gemuk. Laporan Hasil Penelitian. Hibah penelitian DP2M Desentralisasi Tahun 2010. UniversitasBarawijaya. Malang.
- Seng S, C. Phanchadcharam, M.Suy and J. Hoglund. 2010. Effects of ens led cassava (*Manihot esculenta*) foliage compared to a soybean meal supp'ement on gastrointestinal nematode infections in goats. Livestock Research for Rural Development. 22 (6) 2010.
- O'Hair, S.K. 1995. Cassava New Crop Fact Sheet Centre for New Crop and Plant product. PurdueUniversity.
- Poppi, D.P. 1996. Prediction of Food Intake in Ruminants from Analysis of Food Composition. Austr. J. Agric. Res. 47: 489-504.
- Wanapat, M. and A. Petlum. 2004. Feeding Cassava Chip-Based Rations to Lactating Dairy Cows. <http://www.mekarn.0m/procKK/wana2.htm>. Akses at 12 Juli 2004.

Effects of CNCPS fraction-enriched protein feeds on ruminal fermentation in Holstein steers Fed TMR containing low protein as a basal

Choi¹, C. W., S. Hwangbo¹, S. K. Park², D. H. Kim³ & S. H. Choi⁴

¹Daegu University, Gyeongsan, Gyeongbuk 712-714, Korea, ²Sejong University, 209 Neungdong-ro, Gwangjin-gu, Seoul 143-747, Korea, ³Korea Research Institute Bioscience and Biotechnology, Yuseong-gu, Daejeon 305-806, Korea, ⁴Chungbuk National University, Chungju, Chungbuk 362-763, Korea

Abstract

Four ruminally cannulated Holstein steers (BW 401.0± 2.22kg) fed TMR containing low protein (CP 11%) as a basal were used to investigate the effects of cornell net carbohydrates and protein system (CNCPS) fraction enriched protein feeds on rumen fermentation. The steers used in a 4×4 Latin square design were housed under the condition of temperature humidity index (THI) 71.46 (no stress, 24°C and 60%) during the entire trial. Dietary treatments were control (TMR only), AB1 (TMR + rapeseed meal), B2 (TMR + soybean meal) and B3C (TMR + perilla meal), respectively. Ruminal digesta was sampled through ruminal cannula at 1 h-interval after the afternoon feeding. The digesta was filtered using 8 layers of cheesecloth and measured for ruminal pH, ammonia-N and volatile fatty acids (VFA). Different CNCPS fraction-enriched proteins did not affect ($P > 0.05$) ruminal pH except B3C being numerically low compared with the other groups. It may be derived from relatively high B₃ and C fractionation of perilla meal. Ammonia-N and VFA were not significantly ($P > 0.05$) different among the experimental groups. Numerically low ammonia-N appeared in the steers fed rapeseed meal even though it contained high soluble N composition (A and B₁ fractions). The discrepancy is unclear; however this may be related to low protein level in the diet and/or low DM intake. Further studies on several in vivo trials including different dietary protein and investigation of blood metabolites should be required.

Keywords: cornell net carbohydrates and protein system, fraction, ruminal fermentation

Introduction

Understanding ruminal proteolysis is rather difficult because the protein metabolism includes both dietary protein degradation and microbial protein synthesis, thus an accurate method for studying rumen metabolism is required. (Wallace, 1988; Choi, 2002; Jin, 2011). As an in vitro evaluation method, the Cornell net carbohydrate and protein system (CNCPS) has been developed to fractionate protein N (Licitra et al. 1996). In the CNCPS, non protein N (NPN) is defined as A fraction whereas true protein (defined as B) is subdivided into soluble true protein N (defined as B₁), neutral detergent soluble protein N (defined as B₂) and neutral detergent insoluble, but acid detergent soluble protein N fractions (defined as B₃) based on solubility in different chemical solution. The acid detergent insoluble N (defined as C) is indigestible protein. Many studies on effects of crude protein on productivity and/or rumen metabolism (Choi, 2002) and fractionation of dietary protein by the CNCPS method published (Choi, 2002; Jin, 2011) for better understanding protein metabolism and evaluation in the rumen have been. However, few studies on effects of CNCPS fraction enriched protein (called as fraction-representative protein) on rumen metabolism. Therefore, the present study was conducted to investigate the effects of CNCPS fraction-representative protein on ruminal fermentation parameters in Holstein steers fed low protein TMR as a basal.

Materials and Methods

Four ruminally cannulated Holstein steers (BW 401.0± 2.22kg) fed low protein TMR (CP 11%) as a basal were used to investigate the effects of CNCPS fraction representative protein feeds on rumen fermentation. The steers used in a 4×4 Latin square design were housed under the condition of temperature humidity index 71.46 (no stress, 24°C and 60%) during the entire trial. Dietary treatments were control (TMR only), AB1 (TMR + rapeseed meal as A + B₁ fractions representative protein), B2 (TMR + soybean meal as B₂ fraction representative protein) and B3C (TMR + perilla meal as B₃ + C fractions representative protein), respectively. Ruminal digesta was sampled through ruminal cannula at 1 h-interval. The digesta was filtered using 8 layers of cheesecloth and measured for ruminal pH, ammonia-N and volatile fatty acids (VFA) according to Kim et al. (2009). Data were subjected to analysis of variance using the general linear models (GLM) procedure, and mean difference were examined using linear contrast (SAS, 2002). Statistical significance was defined at P < 0.05.

Results and Discussion

The highest proportion of rapeseed meal was A and B₁ fractions (35.9%), whereas that of soybean meal was B₂ fraction (50.6%) (Table 1). The C fraction of perilla meal was 57.2% as the highest fraction. With the N fractions of TMR feed, soluble N fraction (A and B₁) was similar with rapeseed meal while slowly soluble and insoluble N fractions (B₂, B₃ and C) were similar with soybean meal, respectively. Ruminal pH was not affected by different protein supplementation (p>0.05; Figure 1). Changes in ruminal pH for control, AB1 and B2 groups appeared similarly. The present pH pattern may be due to the similar N fractionation the TMR, rapeseed meal and soybean meal discussed above. Relatively lower ruminal pH for B3C than that for the other groups during the entire feeding cycle may be explained by the high proportion of B₃ and C fractions of perilla meal (see Table 1).

Table 1. Nitrogen fractionation (% of CP) of experiment feeds by CNCPS method.

Fraction [†]	TMR	Rapeseed meal	Soybean meal	Perilla meal
A fraction	24.8	24.7	10.2	22.9
B ₁ fraction	6.5	11.2	9.6	7.1
B ₂ fraction	39.7	27.1	50.6	12.8
B ₃ fraction	5.0	14.5	5.4	13.1
C fraction	24.1	22.5	24.2	44.1

[†]Fraction was analyzed according to the method of Licitra et al. (1996).

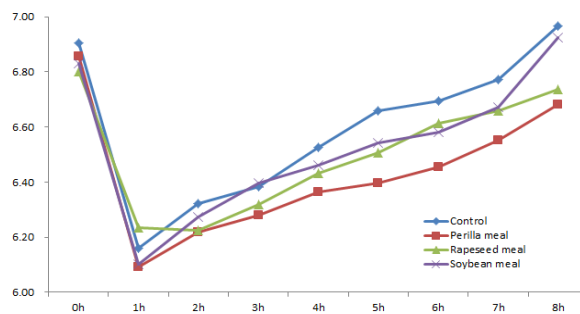


Figure 1. Changes in ruminal pH of steers fed TMR supplemented with various protein. Treatments were TMR only (◆), TMR with rapeseed meal (▲), TMR with soybean meal (X) and TMR with perilla meal (■), respectively.

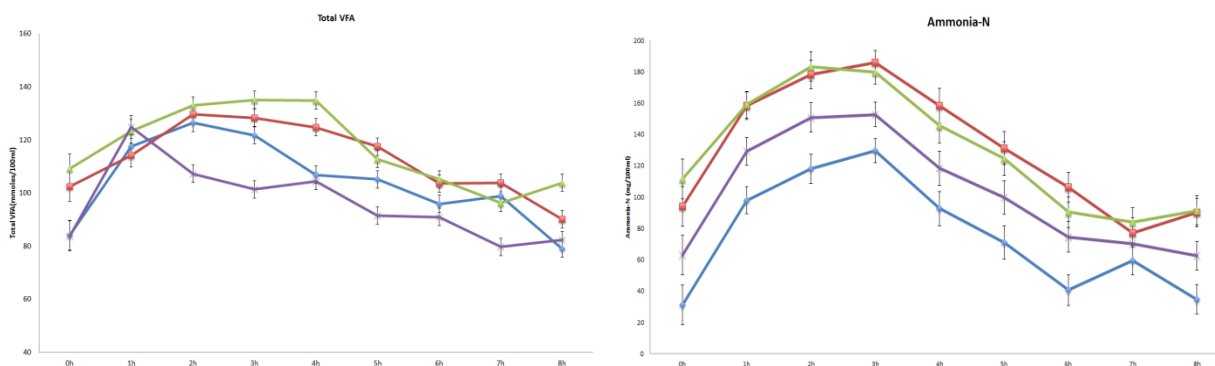


Figure 2. Changes in total VFA (a, left) and ruminal ammonia N (b, right) of steers fed TMR supplemented with various protein. Treatments were TMR only (◆), TMR with rapeseed meal (X), TMR with soybean meal (■) and TMR with perilla meal (▲), respectively.

The VFA molar proportion was not affected by different protein supplementation ($p > 0.05$; Figure 2a). In general protein supplements are not directly related to VFA (Kim et al., 2009) so that we analyzed the VFA as a fundamental data for personal database. The data may be utilized for discussing the relationship between different fractions-enriched but similar level of proteins and ruminal VFA patterns because the level of protein supplements may affect distribution and variety of rumen microbes (Wallace, 1988; Jin, 2011).

The CNCPS fraction representative protein supplements numerically affected diurnal patterns in ammonia-N concentration despite lack of statistical significances ($p > 0.05$; Figure 2b). The patterns for B2 and B3C groups appeared to be higher than the other groups, which can be explained by the feed N fractionation and the pH changes (see Table 1 and Figure 1). However, numerically low ammonia-N appeared in the steers fed rapeseed meal even though it contained high soluble N composition (A and B₁ fractions). The discrepancy is unclear; however this may be related to low protein level in the diet and/or low DM intake. Thus, further studies on different levels of protein in the basal diet supplemented with fraction-enriched protein feeds should be needed.

Acknowledgement

This work was supported by Cooperative Research Program for Agriculture Science & Technology Development (PJ009289042015), Rural Development Administration, Republic of Korea.

References

- Choi, C.W., 2003. Assessment of the flow of soluble dietary non-ammonia nitrogen escaping degradation in the rumen of dairy cows fed grass silage based diets. Doctoral dissertation. University of Helsinki. Department of Animal Science. Viikki. Finland.
- Jin, G.L., 2011. Effect of protein fractionation and buffer solubility of various feed stuffs on in vitro fermentation characteristics, degradability and gas production by rumen microbes. A dissertation for the Doctor of Philosophy. Chungbuk National University. Chungju. Korea.
- Kim, J.H., Y.K. Oh, K.H. Kim, C.W. Choi, S.K. Hong, Y.J. Seol, D.H. Kim, G.C. Ahn, M.K. Song and K.K. Park. 2009. Effects of protein supply from soyhulls and wheat bran on ruminal metabolism, nutrient digestion and ruminal and omasal concentration of soluble non-ammonia nitrogen of steers. *Asian-Aust. J. Anim. Sci.* 22: 1267-1278.
- Licitra, G., T.M. Hernandez and P.J. Van Soest, 1996. Standardization of procedures for nitrogen fractionation of ruminant feeds. *Anim. Feed Sci. Technol.* 57: 347-358.
- SAS. 2002. *Sas User's Guide. Statistics. Version 8.e.* SAS Institute. Inc. Cary, NC.
- Wallace, R.J., 1988. Ecology of rumen micro-organisms: protein use. *In Aspects of digestive physiology in ruminants.* Cornell University Press, Ithaca, USA. pp. 99-122.

Effects of dietary pomegranate by-products on performance, immunity, intestinal microbiology and odorous gas emissions from excreta in broilers

Ahmed, S. T., M. M. Islam, H. S. Mun, A. B. M. R. Bostami, K. W. Park, & C. J. Yang*

Department of Animal Science and Technology, Sunchon National University, South Korea

Abstract

The effects of dietary supplementation with pomegranate by-products (PB) on performance, immunity, intestinal microflora and odorous gas emissions from excreta in broiler chickens were investigated. A total of 240 day-old broiler chicks were fed three experimental diets (containing 0, 0.5 and 1.0% PB) until 35 days of age. The average daily gain was not significantly affected by dietary treatments, but the average daily feed intake and feed conversion ratio were reduced during the overall experimental period ($P < 0.05$). The concentration of serum IgA and IgG increased ($P < 0.05$) in response to both levels of PB. In the ileal digesta, the concentration of yeast and mold increased (0.5 and 1.0%), while decreased the *Escherichia coli* and *Salmonella* spp. ($P < 0.05$) concentration (1.0%) in response to dietary PB. In the cecal digesta, the concentration of *Bacillus* bacteria increased ($P < 0.05$) in response to both levels of dietary PB, while the concentrations of *E. coli* and *Salmonella* decreased when the diet was supplemented with 1% PB ($P < 0.05$). Dietary PB effectively reduced the emissions of ammonia and methanethiol from broiler excreta ($P < 0.05$). In conclusion, dietary PB improved immunity and the intestinal microbial ecosystem of broilers along with reduced odorous gas emissions from excreta.

Keywords: pomegranate by-products, broiler, immunity, microbial ecosystem, odorous gas

Corresponding author: tabassum_81@yahoo.com

Introduction

Since the use of antimicrobial growth promoters (AGPs) in poultry diets was banned, phytobiotics have received considerable attention as potential alternatives to AGPs owing to their antioxidant, antimicrobial and immunomodulatory activities. Pomegranate peels, are important source of several phytobiotics including hydrolysable tannins and other phenolic compounds, minerals and complex polysaccharides (Seeram et al., 2005). A number of studies have confirmed the antimicrobial and immunomodulatory activity of pomegranate peel and seed extracts against several bacteria and fungi (Gracious et al., 2001; Nuamsetti et al., 2012). In this study, we investigated whether supplementation of broiler diets with PGB impacted the performance, serum immunoglobulins, intestinal microbiota, and odorous gas emissions from excreta in broilers.

Materials and Methods

A total of 240 one-day-old Ross 308 male broiler chicks were randomly allocated into three treatment groups (PB 0%, 0.5% and 1.0%) with ten replicate pens of eight birds. The chicks were

fed a basal diet during a starter (0-21 d) and finisher (22-35 d) period. Feed intake and body weight were recorded and the average daily feed intake (ADFI), average daily gain (ADG), and FCR (feed:gain) were then calculated. Serum IgA and IgG were analyzed (after blood collection from 3 bird/replication) using chicken-specific IgA (Cat. No. E30-103) and IgG (Cat. No. E30-104) ELISA kits (Bethyl Laboratories Inc., Montgomery, TX, USA) according to the manufacturer's instructions. After blood collection the birds were slaughtered and collected the ileal and cecal digesta to measure the concentration of *Lactobacillus*, *Bacillus*, yeast and mold, *E. coli* and *Salmonella*. Fecal odorous gas emissions were measured at 0, 3, 6, 12, 24 and 48 h of incubation. Data were analyzed by SAS (2003) and level of significance was preset at $P < 0.05$. An orthogonal polynomial contrast test was performed to determine linear and quadratic effects of increasing levels of PB on each measurement.

Results and Discussion

Inclusion of PB linearly reduced the feed intake, while improved the feed efficiency of broiler without affecting the ADG (Table 1). The reduction in feed intake can be explained by reduced diet palatability due to the presence of a considerable amount of tannins in pomegranate peel. Increasing levels of PB in broiler diet increased the concentration of both IgA (Linear, $P = 0.003$; quadratic, $P = 0.006$) and IgG (linear, $P = 0.023$; quadratic, $P = 0.073$) relative to the control. In the ileal digesta, the concentration of yeast and mold increased linearly ($P = 0.003$) and quadratically ($P < 0.01$) in response to dietary PB (Table 2). Moreover, supplementation with 1% PB led to a linear decrease in *E. coli* and *Salmonella* ($P < 0.05$). In the cecal digesta, the concentration of *Bacillus* bacteria increased linearly ($P < 0.01$) in response to dietary PGB, while the concentrations of *E. coli* and *Salmonella* decreased when the diet was supplemented with 1% PGB ($P < 0.05$). A number of *in vitro* studies have confirmed the antimicrobial activity of pomegranate fruit peel against pathogenic *E. coli* and *Salmonella* (Nuamsetti et al., 2012).

Table 1. Effects of dietary pomegranate by-products (PB) on growth performance and serum immunoglobulins of broilers.

Parameter	PB (% of diet)			Probabilities		
	0	0.5	1.0	SEM	Linear	Quadratic
Growth Performance (0-35 d)						
Weight gain (g/bird/day)	55.26	55.94	54.74	0.89	0.702	0.430
Feed intake (g/bird/day)	96.08	90.53	89.45	1.80	0.033	0.353
FCR (g Feed/g Gain)	1.74	1.62	1.64	0.03	0.040	0.087
Serum immunoglobulins (mg/ml)						
IgA	0.75	0.84	0.82	0.01	0.003	0.006
IgG	1.70	1.78	1.77	0.02	0.023	0.073

^{a,b} Values with different superscripts in the same row differ significantly ($P < 0.05$).

Dietary PB effectively reduced the emissions of ammonia and methanethiol from broiler excreta ($P < 0.05$). The effectiveness of dietary PB at reducing emissions of NH_3 from broiler excreta can be attributed to a reduction in the concentration of both intestinal and excreta ureolytic bacteria including *E. coli*, together with a reduction in fecal urease activity due to the presence of high concentrations of tannins in PB. In conclusion, dietary PB improved immunity and the intestinal microbial ecosystem of broilers along with reduced odorous gas emissions from excreta.

Table 2. Effects of dietary pomegranate by-products (PB) on ileal and cecal microbiology of broiler.

Microorganism (CFU/g)	Pomegranate by-product			Probabilities		
	0%	0.5%	1.0%	SEM	Linear	Quadratic
Ileum microbiota						
<i>Lactobacillus</i> spp.	5.68	5.64	5.06	0.40	0.317	0.311
<i>Bacillus</i> spp.	5.03	4.68	5.23	0.58	0.829	0.578
Yeast and mold	3.32 ^b	5.21 ^a	4.80 ^a	0.25	0.003	0.007
<i>Escherichia coli</i>	5.97 ^a	5.26 ^{ab}	4.73 ^b	0.22	0.006	0.765
<i>Salmonella</i> spp.	1.61 ^a	1.25 ^{ab}	0.68 ^b	0.25	0.031	0.736
Cecum microbiota						
<i>Lactobacillus</i> spp.	6.65	6.50	6.78	0.22	0.708	0.489
<i>Bacillus</i> spp.	5.58 ^b	6.31 ^a	6.25 ^a	0.15	0.014	0.066
Yeast and mold	5.69	5.57	6.31	0.37	0.335	0.439
<i>Escherichia coli</i>	6.57 ^a	6.91 ^a	5.73 ^b	0.22	0.029	0.025
<i>Salmonella</i> spp.	2.16 ^a	2.39 ^a	0.91 ^b	0.40	0.002	0.009

^{a,b}Values with different superscripts in the same row differ significantly (P<0.05).

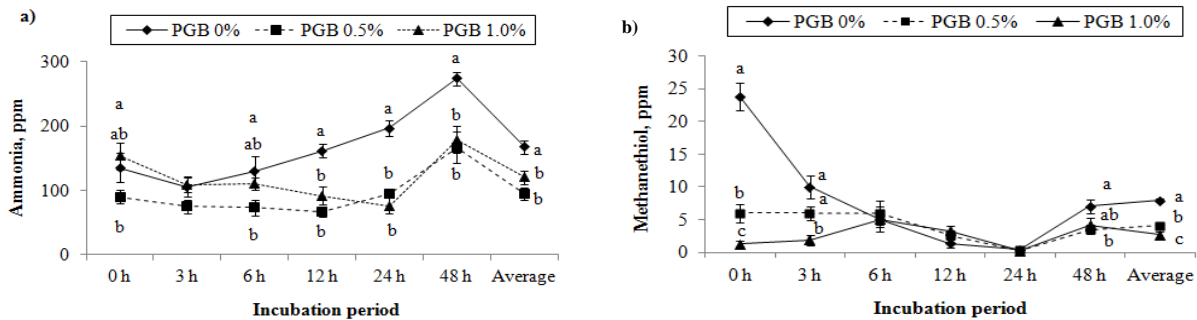


Figure 1. Effects of dietary pomegranate by-products (PB) on (a) ammonia and (b) methanethiol emissions from broiler excreta after different incubation periods.

References

- Gracious, R. R., S. Selvasubramanian, S. Jayasundar, 2001. Immunomodulatory activity of *Punica granatum* in rabbits- a preliminary study. *J. Ethnopharmacol.* 78: 85–87.
- Nuamsetti, T., P. Dechayuenyong, S. Tantipaibulvut, 2012. Antibacterial activity of pomegranate fruit peels and arils. *Science Asia* 38, 319–322.
- Seeram, N. P., L. S. Adams, S. M. Henning, Y. Niu, Y. Zhang, M. G. Nair, D. Heber, 2005. *In vitro* antiproliferative, apoptotic and antioxidant activities of punicalagin, ellagic acid and a total pomegranate tannin extract are enhanced in combination with other polyphenols as found in pomegranate juice. *J. Nutr. Biochem.* 16: 360–367.

Effects of synbiotic supplemented in broiler diet on carcass and meat quality

B. V.Raksasiri^{1,*}, S. Paengkoum², K. Poonsuk³, P. Thitisak³ and P. Paengkoum¹

¹*School of Animal Production Technology, Institute of Agricultural Technology, Suranaree University of Technology, Thailand.*

²*Program in Agriculture, Faculty of Science and Technology, Nakhonratchasima Rajabhat University, Thailand.*

³*K.M.P. BIOTECH CO., LTD., Muangchonburi, Chonburi, Thailand.*

Abstract

This study was conducted to investigate the effect of synbiotic supplementation in broiler diets on carcass and meat quality. Jerusalem artichoke and BACTOSAC-P[®] were used as the source of prebiotic and probiotic, respectively. Three hundred and twenty Ross-308 chickens with 10 days old of age were randomly allocated to four dietary treatments (T); control diet (T1), synbiotic supplemented at 0.025% of DM (T2), 0.050% of DM (T3) and 0.075% of DM (T4) in broiler diet according to a completely randomized design (CRD) with 4 replicates (20 chickens per replicate). Carcass and meat qualities were determined at the day 32 of experimental period. The results were showed that supplementation of all three supplements did not effected on carcass and meat quality. The results indicated that supplementation with synbiotic in broiler diet did not result in changes in performance of birds. However, synbiotic might provide a more beneficial immune modulation status as we are observed in the laboratory.

Keywords: Broiler, Synbiotic, Carcass quality and Meat quality

**Corresponding author: bhutharit@gmail.com*

Introduction

The utilization of antibiotic restricted in animal feed, thereby the use of prebiotic which affecting not only animal health. However, prebiotic high security and important in gastroenterology by keeping probiotic, because prebiotic be similar to antibiotic but safe than. In case use prebiotic, the probiotic will thrive and the pathogen will reduce. At present in case use prebiotic with probiotic will be safe and effective. In Jerusalem artichoke have prebiotics. In the important is inulin, inulin have fructo-oligosaccharide (FOS). Jerusalem artichoke can make Bifidobacteria and lactobacilli increase in intestinal and reduce pathogen such as clostridium and *E. coli* (Younes et al., 1995; Kaur and Gupta, 2002). In case use prebiotic with probiotic or we call synbiotic, the probiotic will thrive than add only probiotic because they are subserve together. And more probiotic pass to intestinal (Wanaporn, 2014) This research has purpose for study usage prebiotic with probiotic for increase the carcass quality and meat quality.

Materials and methods

Studied the period of three hundred and twenty Ross-308 chickens with 32 days old of age, using completely randomized design, (CRD). There were 4 groups and 4 replicates (20 chickens per replicate), and were allocated to 4 dietary treatment,

Group 1 control diet

Group 2 synbiotic supplemented at 0.025% of DM

Group 3 synbiotic supplemented at 0.050% of DM

Group 4 synbiotic supplemented at 0.075% of DM

Jerusalem artichoke and BACTOSAC-P® were used at ratio 1:9 (w/w) as the source of prebiotic and probiotic, respectively. Three hundred and twenty Ross-308 chickens with 10 to 32 days old of age, and random 5 chickens per unit, cutting for examination carcass quality (Sanchai, 2010), carcass and cutting percentage and random tender loin for examination pH, cooking loss and drip loss, (Devine,1999), chemical elements (protein, fat and moisture)AOAC (1995) shear force (Van Oeckel et al., 1999 (and color of meat (Lightness (L*),Redness (a*) and Yellowness (b*)) Sanchai, (2010).

Statistical analysis

Data was statistically analyzed according to a Completely Randomized Design (CRD) (SAS, 1996). Significant differences between treatments were determined using Duncan's News Multiple Range Test (DMRT).

Results and Discussions

A studying of consumer acceptance of synbiotic supplementation in broiler diet on carcass and meat quality. The results were showed that supplementation of all three supplements did not effected on carcass quality (breast, thigh, drumstick, wings, tender loin, visceral fat, entrails and rib) and meat quality (pH, lightness (L*), redness (a*), yellowness (b*), cooking loss, drip loss, shear force and chemical meat composition).Suparom et al (2013) found that supplementing symbiotic in broiler diet showed that the help productive performance but not effect on carcass quality. Probiotic (bactosac®) supplementation in drinking water showed that probiotic supplementation in decreased drip loss in chicken meat (p<0.05) (Nopparatmaitree et al.,2014).While Raksasiri et al.)2015 (found that prebiotic supplementation (jerusalem artichoke, curcuma white and sago palm) in broiler diet showed that , the redness value (a*) of chickens supplemented with Jerusalem artichoke and curcuma white were significantly higher than chickens supplemented with control diet (p<0.05).

Table 1. Effects of prebiotic and probiotic supplementation in diets on carcass quality in broilers

Items (%)	prebiotic and probiotic levels in rations (%)				SEM
	0	0.025	0.05	0.075	
Carcass	84.90	80.12	82.06	82.27	2.533
Breast	23.74	24.10	24.55	24.17	0.356
Thigh	16.72	18.10	18.24	18.58	0.364
Drumstick	12.01	12.35	12.32	12.52	0.316
Wings	10.71	11.67	11.86	11.73	0.191
Tender loin	4.87	4.71	5.14	4.98	0.102
Visceral fat	2.50	2.07	1.64	1.49	0.824
Entrails	20.75	18.88	19.38	19.25	6.719
Rib	41.63	43.25	44.63	44.50	1.948

SEM = Standard error of mean

Table 2. Effects of prebiotic and probiotic supplementation in diets on meat quality in broilers

Items	prebiotic and probiotic levels in rations (%)				SEM
	0	0.025	0.05	0.075	
pH (1 hr)	6.00	6.04	6.02	6.04	0.078
Lightness (L*)	50.52	49.41	49.59	49.62	0.366
Redness (a*)	1.55	1.84	1.66	1.65	0.068
Yellowness (b*)	4.62	4.60	4.74	4.20	0.300
Cooking loss (%)	28.65	27.15	28.10	27.62	1.787
Drip loss (5 day) (%)	6.97	5.63	4.17	5.57	0.205
Shear force (kg)	2.86	2.72	2.84	2.76	0.174
Protein (%)	21.42	21.76	21.63	21.71	0.813
Fat (%)	1.27	1.02	1.18	1.13	0.098
Moisture (%)	73.32	75.64	75.13	74.96	1.138

SEM = Standard error of mean

Conclusion

The studying of symbiotic supplementation in broiler diet, the affect in a better way on growth promoter and intestinal histomorphology. Even though the current study was conducted under non-challenge conditions of broiler diets did not result in changes in performance of birds, meat and carcass quality. Various possible mechanism of action probiotic have been suggested among which are the stimulation of host enzymes, production of antimicrobial substances, competition for adhesion to epithelial cells, and stimulation of the immune system of the host. Supplementation of synbiotics might provide a more beneficial immune modulation status. We are observed the immunology parameters in the laboratory.

Acknowledgements

The authors are grateful to research and researcher for industry (RRi) on the Thailand Research Fund (TRF), Suranaree university of technology, Nakhonratchasima rajabhat university and K.M.P. BIOTECH CO.,LTD.

References

- AOAC. 1995. Official Method of Analysis. 16th ed. Association of Official Analytical Chemist, Washington, DC.
- Devine, C.E. 1999. Effect of rigor temperature on muscle shortening and tenderization of restrained and unrestrained beef *m. longissimusthoracicusetlumborum*. Meat Sci. 51: 61-72.
- Kaur N. and A.K. Gupta. 2002. Applications of inulin and oligofructose in health and nutrition. J. Biosci. 27(7) : 703-714.
- Nopparatmaitree M., K. Lertchunhakiat, J. Sri-onlerd and W. Kitpipit. .2014.Effects of probiotic (Bactosac®) supplementation in drinking water on apparent nutrient digestibility, intestinal histomorphology, carcass percentage and meat quality of broiler chickens . KHON KAEN AGR. 42 (2) : 221 - 230.
- Raksasiri B. V., A. Petlum, S. Paengkoum, K. Poonsuk, P. Thitisak and P. Paengkoum. 2015.Effects of jerusalem artichoke, curcuma white and sago palm supplementation in broiler diet on production performance, carcass quality and meat quality. KHON KAEN AGR. 43 SUPPL. (1) : 202-207.
- Sanchai Jaturasitha .2010. Meat technology. Department of Animal and Aquatic Science, Faculty of Agriculture, Chiang Mai University, Thailand.
- Suparom W., S. Chumpawadee, N. Santaweesuk and J. Khajarern. 2013. Efficacy of symbiotic in broiler die. KHON KAEN AGR. J. 41 SUPPL. (1) : 381-387.
- Van Oeckel, M. J., N. Warnants and Ch. V. Boucqué. 1999. Pork tenderness estimation by taste panel, Warner-Bratzler shear force and on-line method. Meat Sci. 53: 259-267.
- Wanaporn Tapingkae. 2014. Alternatives to the Use of Antibiotics as Growth Promoters for Livestock Animals. Agri. J. 30 (2): 201 – 212.
- Younes, H., K.Garleb, S. Behr, C. Révész and C. Demigné. 1995. Fermentable fibers or oligosaccharides reduce urinary nitrogen excretion by increasing urea disposal in the rat cecum J. Nutr. 125:1010-1016.

Feed intake, digestibility, nitrogen retention and daily weight gain of steers fed on sugarcane stalk based complete diet silage

Irsyammawati, A., Chuzaemi, S., Hartutik & Hermanto

Faculty of Animal Husbandry, University of Brawijaya, Jl. Veteran, Malang, 65145, Indonesia

Abstract

This research aimed to determine the effect of feeding sugarcane stalk as a silage complete diet on feed intake, digestibility, N-retention and daily weight gain of Frisian Holstein (FH) crossbred steers and to find out the ideal proportion of sugarcane stalk in the complete diet silage which gave economically the best performance of steers.

Nine FH crossbred steers, age 10-11 months, were allocated in randomized block design (3 treatments x 3 replications). The treatments were the proportion (% as feed basis) of sugarcane stalk and concentrate in iso-protein (12 % Crude Protein/CP content) complete diet silage (CDS), i.e. CDS1 (60% sugarcane stalk + 40% concentrate + 1.23% urea); CDS2 (50% sugarcane stalk + 50% concentrate + 0.62% urea); and CDS3 (40% sugarcane stalk + 60% concentrate). Parameters of the research were intake, digestibility, N retention and daily weight gain. Data were subjected to analysis of covariance with initial body weight as covariant.

The results showed that the treatments did not show a significant effect ($P > 0.05$) on feed intake, N-retention and daily weight gain but showed significantly effect ($P < 0.05$) on organic matter digestibility (OMD), crude protein digestibility (CPD) and crude fiber digestibility (CFD). With the same effects of the treatments on crossbred steers performance, hence, CDS1 had higher economically efficiency than the other treatments, especially when the price of sugarcane stalk was less than IDR 200.00/kg (1 USD = IDR 13.000). It could be suggested to consider the particle size of CDS materials, ensilage process and storage method for the best result of CDS.

Keywords: total mixed ration, cattle, feed intake, growth, sugar cane

Introduction

Sugarcane stalk was generally harvested in the dry season when the availability of forage as animal feed is low. Rangnekar (1986) and Gomez-Vasquez et al. (2011) reported that sugarcane stalk can be potentially used as animal feed. Sugarcane stalk contains high crude fiber (CF), 28 – 36% (Rangnekar, 1988; Pate et al., 2002) and sugar (48-55%) in DM basis (Leng, 1986; Rangnekar, 1988). Sugarcane stalk also contains essential minerals for the animals. Digestibility of sugarcane stalk is relatively high at approximately 65 % (Leng, 1992). Thus, sugarcane stalk could be a potential energy source, especially for ruminants. However, crude protein (CP) content of sugarcane stalk is quite low, 2.0 – 3.6% DM (Pate et al., 2002). According to Cabral et al. (2009) whole sugarcane contains around 29 g crude protein/kg dry matter. Hence, feeding sugarcane stalk in combination with other feeds mainly richer nitrogen-protein ingredients (Gendley et al., 2009) is better for ruminants.

Sugarcane stalk also can be ensiled (Kung & Stanley, 1982; Harris et al., 1983). It was suitable with sugarcane stalk characteristics which has high soluble sugar concentration (48 – 55%). These soluble sugars are needed in ensilage to initiate the growing of *Lactobacillus* bacteria. However, these conditions also could enhance an alcoholic fermentation and deterioration in silages (De Toledo et al., 2010). It can be reduced by adding of source of fermentable nitrogen (i.e., urea), alkali (i.e., calcium propionate), or humidity absorbent (i.e., corn grain) to buffer the pH in the silo for few days in the first stage of ensilage (Ashbell et al., 1984; Lopes et al.2007). Siqueira (2005) and Junqueira (2006) reported that silage which treated with urea will reduce ethanol production and dry matter losses.

Complete diet has been popular in animal feeding methods, especially in poultry feeding and most recently for ruminant. Therefore, the aim of this study was tried to make a complete diet based on sugarcane stalk as the fiber source and concentrate as the protein source and then processed into silage.

Materials and Methods

In Vivo trial with nine FH crossbred steers, age 10-11 months, were allocated in randomized block design (3 treatments x 3 replications). The treatments were the proportion (% as feed basis) of sugarcane stalk and concentrate in iso-protein (12 %CP content) complete diet silage (CDS), i.e. CDS1 (60% sugarcane stalk + 40% concentrate + 1.23% urea); CDS2 (50% sugarcane stalk + 50% concentrate + 0.62% urea); and CDS3 (40% sugarcane stalk + 60% concentrate). Parameters of the research were intake, digestibility, N retention and daily weight gain. Data were subjected to analysis of covariance with initial body weight as covariant and analysis of variance.

Result and Discussion

Feed intake, digestibility, N retention and daily live weight gain

Average DMI on the treatments ranged from 2.49 – 2.65% of live weight. These DMI were in the range stated by Davies (1982) as well as NRC (2001) that DMI of calves should be 2- 4 % of its live weight. The data showed that intake and digestibility of nutrient (DM, OM, CP, CF) tended to increase as proportion of sugarcane stalk in the treatment diets decreased, however total nutrient intake, N-retention and body weight gain were comparable between CDS1 and CDS 3 treatments, but slightly lower on CDS2.

Table 1. Average total intake, digestibility, N retention and daily weight gain of FH crossbred steers as affected by treatments.

Parameters	CDS1	CDS2	CDS3
Intake (kg/head/day)			
DM	4.44 ± 0.07 ^a	4.01 ± 0.53 ^a	4.52 ± 0.62 ^a
OM	3.92 ± 0.06 ^a	3.56 ± 0.47 ^a	4.01 ± 0.56 ^a
CP	0.54 ± 0.01 ^a	0.49 ± 0.06 ^a	0.56 ± 0.06 ^a
CF	1.31 ± 0.04 ^a	1.19 ± 0.18 ^a	1.44 ± 0.19 ^a
Digestibility (%)			
DM	54.06 ± 0.53 ^a	53.52 ± 0.11 ^a	53.99 ± 1.02 ^a
OM	55.08 ± 0.41 ^a	55.58 ± 0.39 ^a	56.06 ± 0.45 ^b
CP	59.89 ± 3.05 ^a	66.65 ± 1.59 ^b	71.55 ± 1.55 ^b
CF	33.75 ± 2.16 ^a	35.66 ± 2.14 ^{ab}	39.37 ± 1.27 ^b
N Retention (g/head/day)	60.76 ± 6.74 ^a	52.49 ± 3.52 ^a	64.29 ± 8.38 ^a
DLWG (g/head/day)	422.62 ± 98.38 ^a	416.70 ± 180.64 ^a	625.00 ± 139.46 ^a

^{a-c} values in the same row for each parameter with different superscripts were significantly different ($p < 0.05$).

Conclusion

It could be concluded that silage complete diet based on sugarcane stalk (CDS) can be used as an alternative feed in certain conditions for ruminant. The result was as expected. By adding urea in the treatment, there was no significant effect to intake, N retention and daily weight gain except organic matter digestibility (OMD), crude protein digestibility (CPD) and crude fiber digestibility (CFD). CDS1 had higher economically efficiency than the other treatments, especially when the price of sugarcane stalk was less than IDR 200.00/kg (1 USD = IDR 13.000). It could be suggested to consider the particle size of CDS materials, ensilage process and storage method for the best result of CDS.

References

- Ashbell, G, H.H. Theuneand D. Sklan, 1984. Changes in amino acid compounds of wheat plants during ensiling and aerobic exposure: the influence of propionic acid and urea phosphatocalcium propionate, *J. Agric. Sci.* 102: 667–672.
- Cabral, A.D, A.M. Batista, A. Mustafa, F.F.R Carvalho, A. Guim, and P.S. Monteiro, 2009. Performance of dairy goats fed whole sugarcane, *Trop. Anim. Health Prod.* 41: 279–283.
- Davies, H.L.,1982. *Nutrition and Growth Manual*, Australian Universities International Development Program (AUIDP), Melbourne.
- De Toledo, P.R.A.P, R.R. Andrade, S.G. Rezende, P.R.M de Toledo, R.F. Dutra and M.R. Rodriguez, 2010. Sugarcane silage production treated with additives at different time post burning, *R. Bras. Zootec*, 39: 88–96.
- Gendley M K,P. Singh, A.K.Garg, S.P. Tiwari, K. Kumari, and G.K. Dutta, 2009. The studies on nutrient balances in crossbred cattle bulls fed chopped green sugarcane tops supplemented with some agro industrial by-products, *Trop. Anim. Health Prod.* 41: 943–949.
- Gomez-Vazquez A, J.M. Pinos-Rodriguez, J.C. Garcia-Lopez, E. de la Cruz-Lazaro, C.Luna-Palomera, R. Sanchez-Hernandez, 2011. Nutritional value of sugarcane silage enriched

- with corn grain, urea, and minerals as fed supplement on growth performance of beef steers grazing stargrass, *Trop. Anim Health Prod* 43: 215-220.
- Harris Jr B, H.H. Van Horn, K.E. Manookian, S.P. Marchall, M.J. Taylor and C.J. Wilcox, 1983. Sugarcane silage, sodium hydroxide- and steam pressure-treated sugarcane bagasse, corn silage, cottonseed hulls, sodium bicarbonate, and *Aspergillus oryzae* product in complete rations for lactating cows. *J. Dairy Sci.* 66: 1474–1485.
- Kung, Jr. L., and R.W. Stanley, 1982. Effect of stage of maturity on the nutritive value of whole-plant sugarcane preserved as silage. *J. Anim. Sci.* 54: 689–696.
- Junqueira, M. C., 2006. Aditivosquímicos e inoculantesmicrobianosemsilagens de cana-de-açúcar: perdasnaconservação, estabilidadeaeróbia e desempenho de animais, Piracicaba: USP/ESALQ, 98p. Dissertação (Mestrado).
- Leng, R.A., 1986. Principle and Practice of Feeding in Tropical Crops and by Product to Ruminants, Department of Biochemistry and Nutrition, University of New England, Australia.
- _____, 1992 Draught Feeding Strategies: Theory and Practice, R.M. Lilley, Penambul Books, Armidale.
- Lopes J., A.R. Evangelista, and G.P. Rocha, 2007. Nutritional valueof sugarcane silage enriched with urea and with additiveabsorbent of humidity. *R. Bras. Zootec.* 36: 1155–1161.
- NRC, 2001. Nutrient Requirements of Dairy Cattle, Seventh Revised Edition, National Academic Press, Washington, D.C.
- Pate, F. M. J. Alvarez, J.D. Phillips and B.R. Eiland, 2002. Sugarcane as a Cattle Feed: Production and Utilization, Bull. 844, Florida Cooperative Extension Service, Institute of Food and Agricultural Science, University of Florida, Florida.
- Rangnekar, D.V., 1986. Integration of Sugarcane and Milk Production in Western India, In : R.Sansoucy, G. Aarts and T.R. Preston (ed), Proc. FAO Expert Consultation on Sugarcane as Feed, FAO, Rome.
- _____, 1988. Availability and intensive Utilization of Sugarcane By Product, In : C. Devendra (ed), Non Conventional Feed Resources and Fibrous Agricultural Residue : Strategies for Expanded Utilization, Proc. Of A Consultation held in Hisar, India, 21-29 March, International Development Research Centre, Indian Council of Agricultural Research, pp. 76-93
- Siqueira, G. R., 2005. Cana-de-açúcar (*Saccharumofficinarum* L.) ensilada com aditivosquímicos e bacterianos, Jaboticabal: UNESP/FCAV, 91p. Dissertação (Mestrado).

Effect of nitrate addition to cassava chip on *In vitro* gas production

Wilachai K. *, J. Khotsakdee, T. Vorlaphim, B. V.Raksasiri, A. Petlum, S. Thongpea, and P. Paengkoum

School of Animal Production Technology, Institute of Agricultural Technology, Suranaree University of Technology, Thailand.

Abstract

The objective of this study was to determine the effect of nitrate addition to cassava meal and cassava chip on total gas production and mitigation of methane (CH₄) using a gas production technique. The experimental design was completely randomized design (CRD) with seven replication per treatment. The six dietary treatments consisted cassava meal (T1), cassava chip (T2), cassava meal + nitrate (1:1 vol/vol) (T3), cassava chip + nitrate (1:1 vol/vol) (T4), cassava meal + nitrate (extrusion, T5) and cassava chip + nitrate (extrusion, T6). The total gas produced were recorded at 1, 2, 3, 4, 6, 8, 12 and 24 h of incubation time and the CH₄ was collected at 24 h of incubation time. It was found that cumulative volume of gas production increased with increasing time of incubation gas produced after 24 h incubation ranged between 0.7 and 44.73 ml per 200mg of dry matter. In addition, the CH₄ was increased by T1 and T2 compared with other treatments which nitrate addition. Additional research is necessary to evaluate more effective means of reducing methane with a slow release nitrate (extrusion) in goat diets.

Keywords: nitrate, methane, slow release

*Corresponding author: wilachai_k@hotmail.com

Introduction

Nitrate effective as an electron sink in ruminal fermentation, thus lowering the amount of relatively reduced acids such as propionate and butyrate and increase in acetate. However, nitrate reduction in the rumen often causes nitrite accumulation, because nitrate reduction to nitrite is usually much faster than nitrite reduction to ammonia (Farra et al., 1971). Nevertheless, the mitigation of methane in the rumen were added of nitrate, as well as fumarate, to *in vitro* cultures of mixed ruminal microbes decreases methanogenesis (Iwamoto et al., 2002). While, Asanuma et al. (2015) suggested to the growth of nitrate- and nitrite-reducing *Selenomonas ruminantium* is stimulated by nitrate addition. Thus, *Selenomonas ruminantium* is likely to play a major role in nitrate and nitrite reduction in the rumen. Therefore, the objective of this study was determined the effect of the effect of nitrate addition to cassava meal and cassava ship on total gas production and mitigate of methane by using an *in vitro* gas production technique.

Materials and methods

Experimental design and dietary treatment

This study was conducted *in vitro* gas production technique at various incubation time intervals. The experimental using a design was completely randomized design (CRD) with seven replication per treatment. The 6 treatments were cassava meal (T1), cassava chip (T2), cassava meal+ nitrate (1:1 vol/vol) (T3), cassava chip + nitrate (1:1 vol/vol) (T4), cassava meal+ nitrate (slow release, T5) and cassava chip + nitrate (slow release, T6). Three matured rumen fistulated Saanen male goats fed with 2.5 % of body weight (% BW) DM/day containing Pangola hay and commercial concentrate (14 % of crude protein) (60:40) were used as donors of rumen fluid.

Ruminal fluid was sampled before the morning feeding from the three goats and then placed in warm (39°C) insulated flasks under anaerobic conditions. All samples were pooled in equal proportions and strained through 4 layers of cheesecloth under anaerobic conditions and then used immediately.

Artificial saliva was prepared under anaerobic conditions in a water bath at 39°C according to Menke and Steingass (1988), the strained rumen fluid was mixed in a 2:1 ratio (artificial saliva: rumen fluid) to prepare fermentation solution. Thirty-mL of buffered rumen fluid solution was dispensed into 100 ml calibrated glass syringes containing and 200 mg of sample each treatments. Syringes were incubated in water bath at 39°C for 24 h. These procedures were performed in three separated runs.

The total gas produced in the head of syringes were recorded at 1, 2, 3, 4, 6, 8, 12 and 24 h of incubation, and net gas production values were corrected by subtracting blank values from the samples (Menke and Steingass, 1988). Cumulative gas production data were fitted to the model of Ørskov and McDonald (1979) described as $Y = a + b(1 - e^{-ct})$; where Y represents the cumulative gas production at time t , a is the gas production from the immediately soluble fraction, b is the gas production from the insoluble fraction, c is the rate of gas production (/h) and $(a+b)$ is the potential gas production. At the end of 24 h incubation, the CH₄ volume was measured according to Demeyer et al. (1988) as described by Fievez et al. (2005). After the final gas volume was recorded, 4.0 mL of 10 M NaOH was introduced into the incubated contents of each syringe via the lower end of the glass syringe, which thereby avoiding gas escape. After mixed with NaOH, the CO₂ was absorbed into the content, therefore, the gas volume remaining in the syringe considered to be CH₄.

All data were analyzed as a completely randomized design using the PROC ANOVA of SAS (1998) data were analyzed using the model. The following statistical model was used $Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$, Where Y = observations, μ = overall means, τ = effect of treatment ; ε = error. Significant differences between treatments were determined using Duncan's News Multiple Range Test (DMRT) (Steel and Torrie, 1980).

Results and discussion

Cumulative gas production profiles from the *in vitro* fermentation of treatment are show in Figure 1. The cumulative volume of gas production increased with increasing time of incubation gas produced after 24 h incubation ranged between 0.7 and 44.73 ml per 200 mg of dry matter. At all incubation times cumulative gas productions (ml) of treatment were significantly ($P < 0.001$) higher than those of cassava meal (T1) and cassava chip (T2).

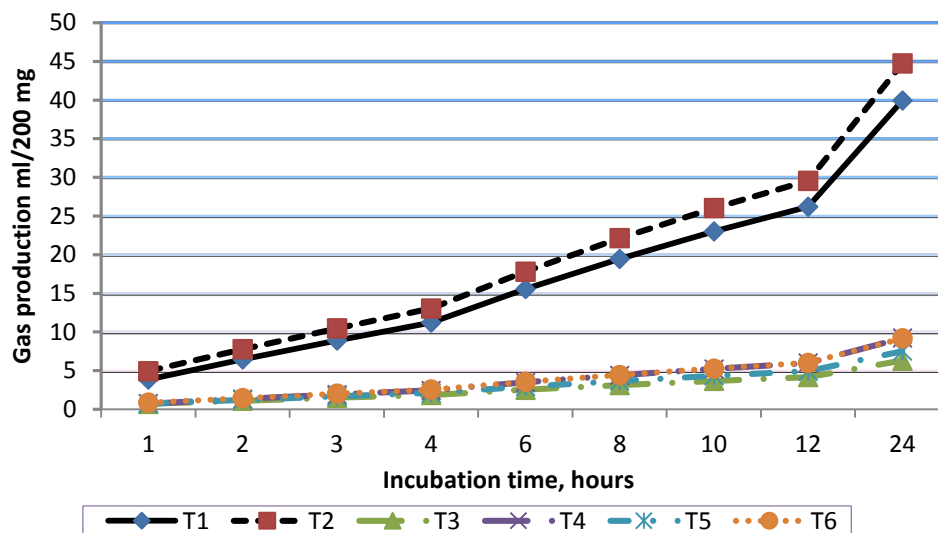


Figure 1. Gas production of treatment when incubated with rumen fluid at different incubation times, T1=cassava meal, T2=cassava chip, T3=cassava meal+ nitrate (1:1 vol/vol), T4=cassava chip + nitrate (1:1 vol/vol), T5=cassava meal+ nitrate (slow release), and T6=cassava chip + nitrate (slow release).

Therefore the estimated parameters (a , b and $a + b$) of T1 and T2 were significantly ($P < 0.01$) higher than the others, but the parameter c not affect all treatment. In addition, the effect of treatment on in vitro methane (CH_4) at 24 h founded that highly of T1 and T2 than T3, T4, T5 and T6 (Table 1.)

Gas kinetics and cumulative gas production were influenced by treatment-T1 and T2 higher than ($P < 0.01$) T3, T4, T5 and T6, these were nitrate addition. Moreover, the CH_4 was lower than group not nitrate addition. Correspondingly, Polyorach et al. (2014) reported that the gas production were influence by different R:C ratio, while gas increased concentrate level. This experiment used were 100 % of carbohydrates sources, but not increase when nitrate addition and slow release method. Nevertheless, Farra et al. (1971) and Asanuma et al. (2015) demonstrated to nitrate can be reduced CH_4 in the rumen due to it effective as an electron sink (H_2 sink) similar to this experiment.

Table 1. The estimated parameters of treatment when incubated with rumen fluid at different incubation times.

Items	Treatment*						SEM	P-value
	1	2	3	4	5	6		
c	0.05	0.05	0.05	0.05	0.05	0.05		
b	55.51 ^b	61.23 ^a	8.58 ^d	13.08 ^c	10.44 ^d	12.80 ^c	3.50	**
a	1.15 ^b	1.94 ^a	0.30 ^c	0.08 ^c	0.23 ^c	0.23 ^c	0.13	**
$a+b$	56.66 ^a	63.17 ^a	8.88 ^b	13.16 ^b	10.27 ^b	13.03 ^b	3.9	**
CH_4 , at 24 h	5.5 ^a	6.75 ^a	2.0 ^b	1.75 ^b	1.38 ^b	1.38 ^b	0.97	**

* Treatment-1) cassava meal, 2) cassava chip, 3) cassava meal + nitrate (1:1 vol/vol), 4) cassava chip + nitrate (1:1 vol/vol), 5) cassava meal + nitrate (slow release) and 6) cassava chip + nitrate (slow release). c = gas production rate, a = gas production (ml) from quickly soluble fraction, b = gas production (ml) from insoluble fraction, ($a + b$) = potential gas. ** P-Value < 0.01 .

Conclusions

Based on this study, it could be concluded that addition were nitrate and slow releases could decreased CH_4 production and then T1 and T2 were total gas production higher than other treatments with nitrate and slow release.

References

- Asanuma, N., S. Yokoyama and T. Hino. 2015. Effects of nitrate addition to a diet on fermentation and microbial populations in the rumen of goats, with special reference to *Selenomonas ruminantium* having the ability to reduce nitrate and nitrite. *Animal Science Journal*, 86(4): 378-384. doi: 10.1111/asj.12307
- Farra, P. A. and L. D. Satter. 1971. Manipulation of the Ruminant Fermentation. III. Effect of Nitrate on Ruminant Volatile Fatty Acid Production and Milk Composition. *Journal of Dairy Science*, 54(7): 1018-1024. doi: 10.3168/jds.S0022-0302(71)85965-9
- Fievez V, Babayemi O.J, and Demeyer D.2005. Estimation of direct and indirect gas production in syringes: A tool to estimate short chain fatty acid production that requires minimal laboratory facilities. *Animal Feed Science and Technology* 123-124, 197-210.
- Iwamoto, M., N. Asanuma and T. Hino. 2002. Ability of *Selenomonas ruminantium*, *Veillonella parvula*, and *Wolinella succinogenes* to Reduce Nitrate and Nitrite with Special Reference to the Suppression of Ruminant Methanogenesis. *Anaerobe*, 8(4): 209-215. doi: http://dx.doi.org/10.1006/anae.2002.0428
- Menke KH, and Steingass H. 1988. Estimation of the energetic feed value obtained by chemical analysis and in vitro gas production using rumen fluid. *Animal Research and Development* 28, 7-55.
- Ørskov ER, and I.McDonald. 1979. The estimation of protein degradability in the rumen from incubation measurements weighted according to the rate of passage. *The Journal of Agricultural Science* 92, 499-503.
- SAS. 1998. 'User's guide: Statistic.' (SAS Institute Inc.: New York).

Effects of TDN value in TMR on ruminal fermentation characteristics and effective dry matter degradability by rumen microbes

Choi¹, S. H., S. U. Lee¹, C. W. Choi², S. K. Park³ & D. H. Kim⁴

¹Chungbuk National University, Chungju, Chungbuk, 362-763, Korea, ²Daegu University, Gyeongsan, Gyeongbuk, 712-714, Korea, ³Sejong University, 209, Neungdong-ro, Gwangjin-gu, Seoul, 143-747, Korea, ⁴ Division of Applied Life Science (BK21), Graduate School of Gyeongsang National University, IALS, Jinju, Gyeongnam 660-701, Korea,

Abstract

An *in vitro* trial was conducted to examine the effects of total mixed rations (TMR) on fermentation characteristics and effective degradability (ED) by rumen microbes. Three TMR diets were growing period TMR (GR-TMR, 67% TDN), early fattening period TMR (EF-TMR, 75.4% TDN) and late fattening TMR (LF-TMR, 80% TDN). Three TMR diets (3g of TMRs in each incubation bottles) was added to the mixed culture solution of stained rumen fluid with artificial saliva (1:1, v/v) and incubated anaerobically for 48 hours at 39 °C. The pH in all incubation solutions tended to decrease up to 48h, but the opposite results were found in concentration of total gas production, ammonia-N and total VFA in all incubations. The total gas production ($P < 0.05$) in LF-TMR was highest compared with those of other diets. Also, concentration of total VFA was tended to increase in LF-TMR compared with other TMR diets in all incubations. The EDDM in both EF-TMR and LF-TMR was tended to high compared with GR-TMR ($P = 0.100$). In these *in vitro* trials, concentration of propionate in all incubation solution was not affected by increased concentration of TDN. The results of the present *in vitro* study indicate that TMR may provide more favorable condition for nutrient digestion both in the rumen.

Keywords: TMR, rumen fermentation, ruminal degradation

Introduction

The system of Total digestible nutrients (TDN) was developed almost 150 years ago because it was shown that the proximate analyses were not sufficient to characterize the nutritive value of feeds (Frik Sundtol, 1990). The term total mixed ration (TMR) or complete ration is used with complete feed, total blended ration. It is a quantitative mixture of all dietary ingredients, blended thoroughly to prevent separation and sorting, and formulated to specific nutrient content. TMR system promotes a steady state of conditions in the rumen environment (pH) and regulates ingesta flow rates (Naresh G. and Balaraman N., 2006). It is a quantitative mixture of all dietary ingredients, blended thoroughly to prevent division and sorting, and formulated to nutrient content. Optimization of forage particle size is a crucial aspect of TMR feeding strategy because it influences structural effectiveness and uniformity of the TMR digestion processes and feed intake, milk production and milk composition. Therefore, the present studies were conducted to find the Effects of TDN value in TMR on ruminal fermentation characteristics and effective dry matter degradability by rumen microbes.

Materials and methods

Rumen contents were obtained 2h after the morning feeding (09:00) from three ruminally-cannulated non-lactating Holstein cows fed 8 kg/d total diets daily (5.6 kg concentrate and 2.4 kg rice straw, as fed basis), twice (09:00 and 18:00 h) per day, in an equal volume. The rumen fluid was strained through 12 layers of cheesecloth to remove the feed particles. Carbon dioxide (CO₂) was flushed into the strained rumen fluid for 30 seconds. Culture solution was prepared by mixing 80ml strained rumen fluid with 80ml McDougall's artificial saliva (McDougall, 1948) in 250ml incubation bottle. Three grams of treatment on DM basis were placed in a nylon bag, and were placed in the flask containing the mixed solution (160ml). The flask were then sealed with rubber stoppers fitted with 3-way stopcocks and were incubated anaerobically in a shaking incubator (VS-8480 SR, VISON Science, Bucheon, Korea) at a speed of 135 rpm up to 48h an 39 °C.

Incubation was stopped by removing the bottles from the shaking incubator at 1, 3, 6, 12, 24 and 48h, and pH of culture solution was immediately measured. At the same time an aliquot of culture solution (0.8 ml, 1 ml) was collected from each bottle for ammonia and VFA analysis. Ammonia concentration was determined by the method of Fawcett and Scott (1960) using a spectrophotometer. The 0.8 ml culture solution was mixed with 0.2 ml 25% phosphoric acid and 0.2 ml pivalic acid solution as the internal standard for the VFA analysis as described by Li et al (2010). Total gas production was also measured at each incubation time through the 3-way stopcock connected to culture bottles. The nylon bag containing feed residue was washed with tap water and dried at 60 °C for 48h in the drying oven to measure dry matter (DM) degradation. Crude proteins (CP) were analyzed according to AOAC (1995). Percent disappearance of DM at each incubation time was calculated from the portion remaining after incubation in the rumen. Disappearance rate was fitted to the equation of Orskov and McDonald (1979). Non-linear parameters a, b and c was estimated by an iterative least square procedure to calculate effective degradability of DM(EDDM) according to the following equation (Orskov and McDonald, 1979)

Results and Discussion

Table 1. Chemical composition and TDN values (% , DM basis)

In vitro ruminal fermentations are widely used as a means of evaluation rumnal digestibility of feed stuffs, particularly when large numbers of samples or experimental treatments are under study (Minson and McLoed, 1972; Mould et al., 2005), or when it is desired to evaluate individual feed stuffs. Numerous studies have compared *in vitro* fermentations to *in vivo* fermentations with regard

Items	Treatments		
	GR-TMR	EF-TMR	LF-TMR
DM	82.25	66.07	74.23
EE	5.92	6.98	7.35
CP	11.21	13.70	13.1
Ash	4.73	7.58	7.48
TDN	67.00	75.40	80.00

to substrate disappearance and product formation, and the results suggest that *in vitro* methods are generally suitable for comparing digestibility of different feeds and feed components (Kaiser and Weniger, 1994, Minson andMcLoed, 1972).

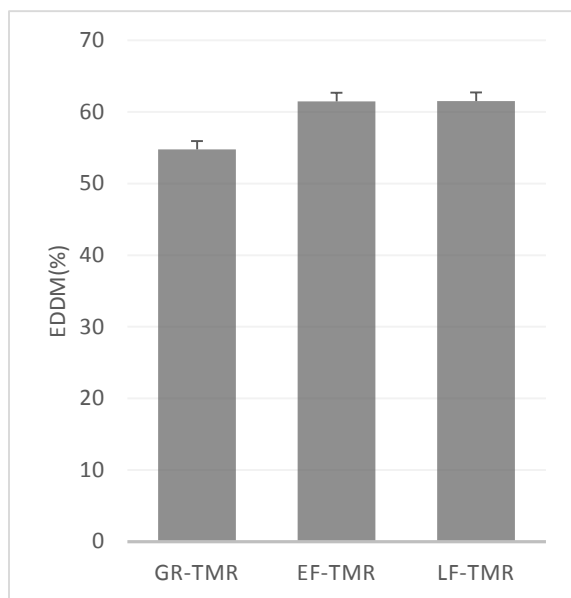


Figure 1. Effect of TDN value on EDDM by rumen microbes.

Ruminal parameters such as pH and VFA have been responded to the diet. In general, higher the starch in the diets faster in fermentation, and thus pH can be lower while total VFA concentration can be increased. It also possible that proportion of propionic acid in the rumen fluid increase as starch in the grain is degraded by the rumen microbes. It is true that great processing the grains make them be degraded rapid in the rumen. Slightly lowered pH with increased ammonia-N in the rumen fluid may indicates the highly degraded protein sources in the diet, respectively as observed from the treatments(Jin, 2009).The rate of gas production has been closely related with the rate of fermentation by ruminal microbes, and generally, grains have their own specific rate of fermentation patterns. The rate and extent of starch digestion in the rumen differed with the species of cereal. Based on the results obtained from the present study, Rumen metabolites (including gases) were increased by high energy density in TMR for fattening period of Hanwoo steers and it is concluded that TMR may provide more favorable condition for nutrient digestion both in the rumen and in the whole tract of ruminants.

References

- AOAC, 1995. Official Methods of Analysis (13th Ed.). Association of Official Analytical Chemists, Washington, DC.Frik Sundtol, 1990
- Orskov, E. R., and I. Mc Donald. 1979. The estimation of protein degradability inthe rumen from incubation measurements weighted according to the rate of passage. *J. Agric. Sci.*, 92:499-503.
- Minson, D. J., McLoed M. N., 1972. The in vitro technique; its modification for estimating digestibility of large numbers of tropical pasture samples. Division of Tropical Pastures Technical Paper No. 8. Common wealth Scientific and Industrial Research Organization, Melbourne, Australia.
- Kaiser, D., Weniger, J. H. 1994. In vivo and in vitro investigations for nutrient digestibility and heat production of ruminants under heat stress and different nutritional levels. V. Comparison of in vitro and in vivo investigations with respect to energy metabolism and energy content of rations. *Archiv.Tierzucht.*, 37;535-545.
- Mcdonald, I., 1981. A revised model for the estimation of protein degradability inthe rumen. *J. Agric. Sci. Camb.* 96, 251-252.
- Jin, G. L., 2005. Effect of dietary starch on the fermentation characteristics in the rumen and starch availability in the small intestine as influenced by dietary protein source. Unpublished doctoral dissertation, Department of Animal Science in Graduate School. Chungbuk National University, Korea.

Utilization of Paprika (*Capcicum annuum*) by product for ruminants feeding

Kim¹, B.G., A. A. Biswas¹, A. R. Son¹, S. Z. Lim¹, C.G. Avedoza¹, L. L. Mamuad¹ & S. S. Lee¹

¹Ruminant Nutrition and Anaerobe Laboratory, Department of Animal Science and Technology, Suncheon National University, Suncheon, Jeonnam, 540-742, South Korea

Abstract

The use of agricultural by product in ruminant feeding is seen to be an approach in improving animal performance as well as low cost feed production. This study was conducted to evaluate the effect of paprika (*Capcicum annuum*) by product on ruminal fermentation using *in vitro* fermentation technique. *In vitro* trial was conducted by adding paprika by product silage with the total mixed ration (TMR). There were five diets provided without paprika by product silage (control), addition of paprika silage ensiled with *L. reuteri* IMAU 70164 16.4% (T1), 33% (T2), paprika silage ensiled with *L. plantarum* KCTC 3594 16.4% (T3) and 33.0% (T4). TMR and Paprika by product silage (DM 1%) was used as the substrate for *in vitro* rumen fermentation. One g DM substrate and 100 ml buffered rumen fluid (1:3 rumen fluid-buffer ratio) were placed in serum bottles and incubated for 0, 12, 24 and 48 h, during which pH, total gas (TG), ammonia-nitrogen (NH₃-N), methane (CH₄) and volatile fatty acids (VFA) concentration, DM and OM digestibility were monitored. Results show that pH was increasing along with control, T1, T2, T3, T4 treatments, respectively and significant differences were observed at 0, 12, 24 and 48h of incubation period. Acetate concentration (44.15mM) was significantly higher (P<0.05) in T4 while butyrate concentration (29.67mM) was significantly higher in T3 after 48h of incubation. Also, higher (P<0.05) total VFA was in T1(80.62mM), T2(83.21mM), T3(81.87mM) and tended to higher total VFA was in T4(79.81mM) than control(76.45mM). Accordingly, higher total VFA suggested that paprika silage can be a potential feed source for increasing productivity of ruminant animal.

Keywords: *In vitro*, paprika, ruminant, total gas, volatile fatty acid

Introduction

Ruminant feeding systems based on agricultural by product are often a practical alternative for low cost feed production. Paprika by products such as stems and leaves are obtained after harvesting, which are mostly unusable and can cause environmental pollution. It is mostly leafy material. So we can use it as a feed for livestock especially ruminant. For its high moisture content drying is not suitable for preservation. Hence, we can preserve paprika by product by ensiling procedure and can provide as a feed for ruminant. Lactic acid bacteria (LAB) are often added to forage crops at the time of ensiling to improve the ensuing fermentation. These added organisms rapidly produce lactic acid and preserve the forage mass. LAB such as *L. plantarum* and *L. reuteri* are used with the goal of providing a faster fermentation (Weinberg & Muck, 1996). LAB inoculants in particular have been shown in various studies to improve milk yield, gain and feed efficiency (Kung et al., 2003). However, there is no available information about *L. plantarum* and *L. reuteri*

inoculants' effect on paprika by product on ensiling and ruminal fermentation. Considering the above circumstances, this study was conducted to evaluate the effect of *L. plantarum* and *L. reuteri* on paprika by product ensiling and *in vitro* ruminal fermentation parameters.

Materials and Methods

L. reuteri IMAU 70164 2.65×10^9 cfu/ml and *L. plantarum* KCTC 3594 1.08×10^9 cfu/ml were used to prepare silage. Ten percent (*L. reuteri* and *L. plantarum*) solution was mixed with paprika by product. Sixty days old silage was used for *in vitro* experiment. *In vitro* trial was conducted by adding paprika by product silage with the TMR containing 13.9 % crude protein (CP) and 79.7% total digestible nutrient (TDN). There were five diets provided without paprika by product silage (control), addition of paprika silage ensiled with *L. reuteri* IMAU 70164 16.4% (T1), 33% (T2), paprika silage ensiled with *L. plantarum* KCTC 3594 16.4% (T3) and 33.0% (T4). Ruminal contents were obtained from 48-month old rumen-cannulated Holstein Friesian cow. One hundred ml of buffered rumen fluid was anaerobically transferred under a constant flow of N₂ to serum bottles containing the one g DM substrate, sealed and incubated at 39°C for 0, 12, 24 and 48 h in a shaking incubator (100 rpm) as described by Hattori and Matsui (2008). pH, total gas (TG), ammonia nitrogen (NH₃-N), acetate, propionate, butyrate, and total volatile fatty acid (VFA) were analyzed at each incubation period.

Statistical Analysis

Differences between groups were identified by analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) using Statistical Analysis System (SAS) (version 9.1; SAS Inst. Inc., Cary, NC).

Results and Discussion

Table 1. pH, Total Gas and NH₃-N from *in vitro* rumen fermentation of paprika silage.

Parameters	Time (h)	CON	T1	T2	T3	T4	SEM	P value
pH	0	6.17 ^b	6.20 ^b	6.28 ^a	6.14 ^b	6.27 ^a	0.019	0.002
	12	5.29 ^c	5.37 ^{bc}	5.48 ^{ab}	5.36 ^{bc}	5.50 ^a	0.030	0.012
	24	5.30 ^b	5.37 ^{ab}	5.47 ^a	5.35 ^{ab}	5.44 ^a	0.030	0.039
	48	5.25 ^c	5.31 ^{bc}	5.33 ^b	5.29 ^{bc}	5.44 ^a	0.020	0.002
Total Gas (mM)	0	0.80 ^c	1.20 ^{abc}	1.00 ^{bc}	1.63 ^a	1.43 ^{ab}	0.144	0.021
	12	68.00	67.33	62.00	68.33	64.33	1.768	0.164
	24	73.33	74.33	72.67	72.00	70.33	0.970	0.197
	48	83.67	81.33	82.67	83.67	75.67	1.798	0.081
NH ₃ -N (mM)	0	8.35	6.83	7.41	7.99	8.14	0.436	0.298
	12	15.90	16.52	15.46	16.18	17.29	1.190	0.945
	24	18.98	19.51	20.45	18.20	20.11	1.140	0.792
	48	15.36	16.94	15.25	16.89	17.66	1.293	0.792

^{a,b,c}Means with difference superscript in the same row are significantly different (p<0.05).

pH level was significantly highest in T4 (5.44) followed by T2 (5.33), T1 (5.31), T3(5.29) and control (5.25) after 48 h of incubation. There was also a significant difference between control and treatment group after 0, 12, 24 and 48 h of incubation but the opposite was observed in total gas production. After 48 h of incubation, the highest total gas production was observed in control (83.67 ml) and T3 (83.67ml) followed by T2 (82.67 ml), T1 (81.33 ml) and T4 (75.67 ml). Similar

non significant total gas production was observed by Muck et al. (2007). There was no significant difference in case of NH₃-N after 0, 12, 24 and 48 h of incubation (Table 1) which is supported by Hu et al. (2009). There was significant difference in acetic acid at 0, 12, 24 and 48 h, butyric acid at 48 h and total VFA at 0, 24 and 48 h of incubations. Higher (p<0.05) acetic acid was observed in T4 with concentrations of 44.03, 46.43 and 44.15 mM after 12, 24 and 48 h of incubation, respectively (Table 2). These results differ Weinberg et al. (2007) wherein they reported non-significant effect of *L. plantarum* on wheat silage but significantly higher value on corn silage by the *L. plantarum* MTD1 (Ecosyl). Higher (p<0.05) butyric acid was observed (Table 2) in T3 (29.67mM) and total VFA was higher (Table 2) in T2 (83.21 mM), T3 (81.87 mM), T1 (80.62 mM) than the control (76.45 mM) after 48 h of incubation. Similar result was observed by Muck et al. (2007) wherein significantly higher total VFA was observed in *L. plantarum* (Biomax5) treated Alfalfa silage. There was no significant difference in propionic acid after 12, 24 and 48 h.

Table 2. Volatile fatty acid concentration from *in vitro* rumen fermentation of paprika silage.

Parameters	Time(h)	CON	T1	T2	T3	T4	SEM	P value
Acetic acid (mM)	0	24.82 ^d	26.85 ^c	28.83 ^{ab}	27.86 ^{bc}	29.88 ^a	0.388	0.000
	12	38.29 ^c	40.88 ^b	44.26 ^a	41.74 ^b	44.03 ^a	0.390	<.0001
	24	38.29 ^c	41.80 ^b	45.18 ^a	42.28 ^b	46.43 ^a	0.533	<.0001
	48	36.28 ^d	41.65 ^b	43.18 ^{ab}	39.43 ^c	44.15 ^a	0.522	<.0001
Propionic acid (mM)	0	6.06 ^b	5.98 ^b	5.74 ^b	6.45 ^a	5.97 ^b	0.097	0.016
	12	12.48	11.79	11.39	12.33	11.25	0.371	0.234
	24	11.81	11.94	11.55	11.94	11.82	0.235	0.802
	48	13.11	12.96	12.71	12.77	12.47	0.471	0.942
Butyric acid (mM)	0	10.50	10.35	9.68	10.35	9.88	0.246	0.356
	12	13.10	13.10	11.95	13.50	11.97	0.449	0.153
	24	16.55	15.43	15.19	16.97	14.46	0.876	0.381
	48	27.06 ^{ab}	26.01 ^{ab}	27.31 ^{ab}	29.67 ^a	23.19 ^b	1.174	0.051
Total VFA (mM)	0	41.39 ^b	43.19 ^{ab}	44.25 ^{ab}	44.65 ^a	45.74 ^a	0.694	0.047
	12	63.87	65.78	67.60	67.57	67.26	1.024	0.151
	24	66.65 ^c	69.18 ^{bc}	71.97 ^{ab}	71.19 ^{ab}	72.71 ^a	0.729	0.008
	48	76.45 ^b	80.62 ^a	83.21 ^a	81.87 ^a	79.81 ^{ab}	0.935	0.017
A/P value	0	4.10 ^d	4.49 ^b	5.03 ^a	4.32 ^c	5.00 ^a	0.019	<.0001
	12	3.07 ^b	3.47 ^b	3.91 ^a	3.39 ^b	3.93 ^a	0.101	0.004
	24	3.24 ^c	3.50 ^b	3.91 ^a	3.54 ^b	3.93 ^a	0.060	<.0001
	48	2.77 ^c	3.25 ^{ab}	3.40 ^{ab}	3.09 ^{bc}	3.55 ^a	0.112	0.012

^{a,b,c}Means with difference superscript in the same row are significantly different (p<0.05).

References

- Hattori, K. and H. Matsui, 2008. Diversity of fumarate reducing bacteria in the bovine rumen revealed by culture dependent and independent approaches. *Anaerobe*. 14, 87-93.
- Hu, W., R. J. Schmidt, E. E. McDonell, C. M. Klingerman and L. Kung Jr, 2009. The effect of *L. buchneri* 40788 or *L. plantarum* MTD-1 on the fermentation and aerobic stability of corn silages ensiled at two dry matter contents. *J. Dairy Sci.* 92: 3907-3914.
- Kung, L. Jr., M. R. Stokes and C. J. Lin, 2003. Silage Science and Technology. In: Silage additives (Ed. D. R. Buxton, R. E. Muck and J. H. Harrison). Am. Soc. Agron., Crop Sci. Soc. Am., Soil Sci. Soc. Am., Madison, WI. pp. 305-360.
- Muck, R., I. Filya and F. Contreras-Govea, 2007. Inoculant effects on alfalfa silage: *in vitro* gas and volatile fatty acid production. *J. Dairy Sci.* 90: 5115-5125.
- Weinberg, Z. and R. Muck, 1996. New trends and opportunities in the development and use of inoculants for silage. *FEMS Microbiol. Rev.* 19: 53-68.
- Weinberg, Z., O. Shatz, Y. Chen, E. Yosef, M. Nikbahat, D. Ben-Ghedalia and J. Miron, 2007. Effect of lactic acid bacteria inoculants on *in vitro* digestibility of wheat and corn silages. *J. Dairy Sci.* 90: 4754-4762.

Performance of Lohi Sheep and Beetal Goats fed Various Fodders

Nasrullah*¹, M. Abdullah², I.B.Marghazani¹, A.N Khosa¹, M. Nasir³, M.H kakar¹, M. Jan¹, M.Fiaz⁴ and N. Ahmad²

*¹Lasbela University of Agriculture Water and Marine Sciences Uthal, Balochistan.²Department of Livestock Production, ³Department of food and Nutrition, University of Veterinary & Animal sciences, Lahore.⁴Department of Livestock Production and Management, PirMehr Ali Shah Arid Agriculture University Murree Road Shamsabad, Rawalpindi, Pakistan

Abstract

A study was conducted to compare the performance of Lohi sheep and Beetal goats fed [Maize (*Zea mays*), Sorghum (*Sorghum bicolor*) and Millet (*Pennisetum americanum*)] fodders. A total of 90 animals [female Lohi sheep (n=45) and Beetal goats (n=45)] were randomly selected and divided equally in six groups (n= 15) animals per groups having three replicates under 2×3 factorial arrangement. Three fodders (maize, millet and sorghum) were randomly fed to the respective replicates in both species. Dry matter (DM), crude protein (CP), NDF and ADF intake was similar (P>0.05) among both species. Results Demonstrate that average daily weight gain feed efficiency and cost of production were similar (P>0.05) among both species.

Keywords: summer fodders, sheep, goats and growth performance.

Corresponding author's email: bangulzai74@yahoo.com

Introduction

Small ruminant production has the potential to become an economically viable option for small full-time farmers and growing number of part-time farmers in the country. Several factors support this assumption including increased demand, lower cost of production compared with other livestock and the ability of small ruminants to effectively utilize poor quality forage (Mc Dowell & Woodward, 1982). In Pakistan sheep and goats production system is still traditional. These animals are kept on grazing seasonal fodders and forages. i.e., summer (Kharif) and winter (Rabi). The summer fodders are Jantar (*Coriandrum sativum*), cowpeas (*vigna sinensis*), maize (*Zea mays*), guar (*cyamopsis tetragonoloba*), sorghum (*Sorghum bicolor*) and millet (*Pennisetum americanum*). The literature is lacking regarding comparative study of summer fodder and local breeds of sheep and goats. Therefore, the present study was planned to compare the voluntary feed intake and digestibility of maize, millet and sorghum fodders in sheep and goats.

Materials and Methods

Experimental procedures

Ninety animals sheep (n =45) and goats (n=45) of similar age (27 month ±15days,) and weight (32.5kg, respectively) were selected and divided under 2×3 factorial arrangement in three groups

(goats A, B and C while sheep D, E, and F,) offered Maize (DM;23, CP; 07 NDF 64, ADF 49.7), sorghum (DM; 19,CP; 08,NDF 62, ADF 47) and millet (DM; 31, CP;4.5 NDF65.5, ADF 46) fodders and left over were measured the next morning throughout the study period to calculate feed intake. The procedure of (Aregheore, 1996) for further process of samples and for proximate analysis of samples were conducted according to AOAC (2000) Collected data were analyzed statistically through two-way ANOVA technique under factorial arrangements using SAS 9.1.3 portable software. The difference among treatment means were tested through LSD test (Steel et al., 1997).

Results and Discussion

Nutrients intake

The DM and other nutrients intake of goats and sheep fed different fodders (maize, millet, and sorghum) are given in Table 1. The DM, CP, NDF, ADF intakes were higher ($P < 0.05$) in goats than sheep on all fodders are in line with the study of Gordon (2003) who reported that intake of nutrients were higher in deer than in sheep. Isaac et al. (2008) also concluded that goats had higher intakes of browse foliages than sheep are consistent to our study. In present study, goats and sheep preferred millet than maize and sorghum fodders are in consistent to Hadjigeorgiou et al. (2003) who found that goats and sheep have a similar pattern of preference for forages with a wide range of chemical characteristics.

Gain and efficiency

The weight gain of goats and sheep fed different summer fodders are given (Table 2).The average daily gain was similar in both species on all offered fodders. However, sheep performed better and registered higher weight gain than goats. The growth rates of Boer goats lower than sheep were reported by VanNiekerk & Casey (1988) is consistent to our findings. Riitta & Kangasmäki (2000) found that mean growth rate were higher for lambs than for kids raised under similar stall-feeding conditions. The efficiency among fodders was significantly different ($P < 0.05$) whereas performance of goats and sheep was similar ($P > 0.05$) on these fodders. The feed efficiency is usually different in goats and sheep. In our study, feed efficiency was similar in both species. This might be attributed to type and quality of fodders, preference of both species. The cost of production Rs/kg was similar ($P > 0.05$) between goats and sheep. The cost of production is generally higher in sheep than goats. The cost of production might be high in goats than sheep due to aggressive behavior.

References

- AOAC, 2000. Official Methods of Analysis. 17th ed. Association of Analytical Chemists, Arlington, Virginia, USA.
- Aregheore, E.M, 1996. Voluntary intake and nutrient digestibility of crop residues based ration by goats and sheep. *Small. Rumin. Res.* 22:7-12.
- Gordon, I. J, 2003. Browsing and grazing ruminants are they different beasts. *Forest Ecology and Management.* 181, 13–21.

- Hadjigeorgiou, I.E., I.J.Gordon and J.A. Milne, 2003. Comparative preference by sheep and goats for Graminaeae forages varying in chemical composition *Small. Rumi. Res.*, 49. (2): 147-156.
- Isaac Osuga, M., C.carolyne. A.Wambui., Shaukat, Abdulrazak, Toshiyoshi ichinohe and Tsutomu fujihara (2008). Evaluation of nutritive value and palatability by goats and sheep of selected browse foliages from semiarid area of Kenya. *Anim. Sci., J.* 79: 582–589.
- McDowell, R. E., A .Woodward, 1982. Concepts in animal adaptation. Comparative suitability of goats, sheep, and cattle in tropical environments. *Proc. 3rd. Int. Goat Conf. Tucson, AZ.* pp 387.
- Riitta,S.C., T. Kangasmaki ,2000. Performance of Finnish Landrace goat kids and lambs raised under stall-feeding conditions in Finland. *Small. Rumin. , Res.* 38:109-114.
- Steel, R. G. D., J. H. Torrie and D. A. Dickie, 1997.** Principles and procedures of statistics - a biometric approach. Third edition. McGraw-Hill Publishing Company. Toronto.
- Van Niekerk, W.A. and N.H. Casey, 1988. The Boer Goat II. Growth, nutrient requirements, carcass and meat quality. *Rumin. Res.* 1: 355-368.

Intake Digestibility of Summer Fodders Fed Sheep and Goats

Nasrullah^{*1}, M. Abdullah², I.B.Marghazani¹, A.N Khoso¹, M. Nasir³
M.H kakar¹ M. Jan, M.Fiaz and Nisar Ahmad

^{*1}Lasbela University of Agriculture Water and Marine Sciences Uthal, Balochistan

²Department of Livestock Production, ³Department of food and Nutrition, University of Veterinary & Animal sciences, Lahore

Department of Livestock Production and Management, PirMehr Ali Shah Arid Agriculture University Murree Road Shamsabad, Rawalpindi, Pakistan

Abstract

A study was conducted to compare the performance of sheep and goats fed on [Maize (*Zea mays*), Sorghum (*Sorghum bicolor*) and Millet (*Pennisetum americanum*)]. For this purpose a total of 90 animals [female sheep (n=45) and goats (n=45)] were randomly selected and divided equally in six groups (n= 15) animals per groups having three replicates under 2×3 factorial arrangement. Three fodders (maize, millet and sorghum) were randomly fed to the respective replicates in both species. Dry matter (DM), crude protein (CP), NDF and ADF intake was similar (P>0.05) among both species on maize millet and sorghum. NDF and digestibility was similar on sorghum in goat and sheep while different (P<0.05) on maize and millet however, ADF digestibility were similar (P>0.05) among both species.

Keywords: summer fodders, intake, sheep, goats and digestibility.

**Corresponding author: bangulzai74@yahoo.com*

Introduction

Goats and sheep production makes stable household income of full time and part time farming communities in the developing countries besides providing good quality protein to the masses of rural and urban areas. In Pakistan sheep and goats are kept on grazing fodders and forages. The availability of fodder depends mainly on the climate and soils (Bruzon, 2007). There are two seasons of fodder crops i.e., summer (Kharif) and winter (Rabi).The summer fodders are Jantar (*Coriandrum sativum*), cowpeas (*vigna sinensis*), maize (*Zea mays*), guar (*cyamopsis tetragonoloba*), sorghum (*Sorghum bicolor*) and millet (*Pennisetum americanum*). The literature is lacking in this regard. Therefore, the present study was planned to compare the feed intake and digestibility of maize, millet and sorghum fodders in sheep and goats.

Material and Methods

Experimental procedures

Ninety animals sheep (n =45) and goats (n=45) of similar age (27 month±15days,) and weight (32.5kg, respectively) were selected and divided under 2×3 factorial arrangement in three groups

(goats A, B and C while sheep D, E, and F,) offered Maize (DM;23, CP; 07 NDF 64, ADF 49.7), sorghum (DM; 19,CP; 08,NDF 62, ADF 47) and millet (DM; 31, CP;4.5 NDF65.5, ADF 46) fodders and left over were measured the next morning throughout the study period to calculate feed intake. At the end of feeding trial, the total fecal collection method was used for digestible study. The feces were collected 5 days period. The procedure of (Aregheore, 1996) for further process of samples and for proximate analysis of samples were conducted according to AOAC (2000) Collected data were analyzed statistically through two-way ANOVA technique under factorial arrangements using SAS 9.1.3 portable software. The difference among treatment means were tested through LSD test (Steel et al., 1997).

Results and Discussion

Nutrients intake

The dry matter and other nutrients intake of goats and sheep fed (maize, millet, and sorghum) are given in (Table 1). The DM, CP, NDF, ADF intakes were higher ($P<0.05$) in goats than sheep on all fodders, are consistent to work of Gordon (2003) who reported that intake of nutrients were higher in deer than in sheep. Isaac et al. (2008) also concluded that goats had higher intakes of browse foliages than sheep are consistent to our study. In present study goats and sheep preferred millet than maize and sorghum fodders are in consistent to Hadjigeorgiou et al. (2003) who found that goats and sheep have a similar pattern of preference for forages with a wide range of chemical characteristics.

Nutrients digestibility

The nutrient digestibility of different fodders fed to goats and sheep is presented in Table 2. The digestibility of DM was similar ($P>0.05$) among both species. The CP, NDF and ADF digestibility were higher ($P<0.05$) in sheep than goats. Earlier literature is also in line with our findings (Brown & Johnson, 1985; Larbi et al., 1991; Brown, 1982). However, Lamba & Rajora (2002) reported that dry matter and CP digestibility was lower in sheep (71.4%) than goats (74.0%) while, the crude fibre digestibility was higher in sheep (74.2%) than the goats (71.4%) whereas Santra et al. (1998) found that the digestibility of NDF and ADF were significantly higher in goats than in sheep.

References

- AOAC, 2000. Official Methods of Analysis. 17th ed. Association of Analytical Chemists, Arlington, Virginia, USA.
- Aregheore, E.M, 1996. Voluntary intake and nutrient digestibility of crop residues based ration by goats and sheep. *Small. Rumin. Res.* 22: 7-12.
- Brown Lynn, E, 1982. Goats and sheep: A comparison of intake, digestibility and other digestive/metabolic parameters when fed wheat straw rations. Master of Science Thesis, North Carolina State University, Raleigh. 86 pp.
- Brown Lynn, E., W. L. Johnson, 1985. Intake and Digestibility of Wheat Straw Diets by Goats and sheep. *J. Anim. Sci.* 60:1318-1323.
- Bruzon, V., 2007. Feed and fodder consultant. EC/GOP Project ALA/01/129. Strengthening of Livestock Services Project.

- Gordon, I. J., 2003. Browsing and grazing ruminants are they different beasts. *Forest Ecology and Management*. 181, 13–21.
- Hadjigeorgiou, I.E., I.J.Gordon and J.A. Milne, 2003. Comparative preference by sheep and goats for Graminaeae forages varying in chemical composition *Small. Rumin. Res.* 49: 147-156.
- Isaac Osuga, M., C.carolyne. A.Wambui, Shaukat, Abdulrazak, Toshiyoshi ichinohe and Tsutomu fujihara, 2008. Evaluation of nutritive value and palatability by goats and sheep of selected browse foliages from semiarid area of Kenya. *Anim. Sci. J.* 79: 582–589.
- Larbi, A., F.K Fianu and F.K. Akude, 1991. Voluntary intake and digestibility by sheep and goats of whole-plant, leaf and stem fractions of *Pennisetum purpureum* Schum. *Small Rumin. Res.* 6: 217-221.
- Lamba, K.S., N.K.Rajora, 2002. Comparative intake and nutrient digestibility of guar (*Cyanopsis tetragonaloba*) straw by sheep and goats. *The Indian Journal of Small Ruminants*. 8:101-103.
- Steel, R. G. D., J. H. Torrie and D. A. Dickie, 1997.** Principles and procedures of statistics - a biometric approach. Third edition. McGraw-Hill Publishing Company. Toronto.

Feed intake and nutrient digestibility in goats of silages prepared from *Stylo* legume (*Stylosanthes guianensis* CIAT184) treated with dried mao pomaces (DMP) and lactic acid bacteria

Bureenok, S.^{*}, Yuangklang C., Vasupen K., Saenmahayak B.

¹Faculty of Sciences and Liberal Arts, Rajamangala University of Technology Isan, Nakhon Ratchasima, Thailand

Abstract

The aim of this study was to investigate the effect of applying dried mao pomaces (DMP) alone and combined with LAB on the fermentative quality and nutritive value of *Thapra Stylo* legume silages. Silages were untreated (control) or prepared with DMP, or DMP plus FJLB (DMP+FJLB), or DMP plus *Lactobacillus plantarum* ST1 (DMP+Lp). DMP was applied at 100 g/kg of fresh matter (FM). FJLB and Lp were applied at log 6.03 and 5.58 cfu/g FM, respectively. Four male ruminally fistulated crossbred Boer x Saanen goats (~ 16 kg body weight) were randomly assigned to one of the four dietary treatment silages in a 4 x 4 Latin square design. The 28-d experimental period consisted of a 21-d feed intake and 7 d of sampling. All goats were received concentrate at 1.5% of body weight (BW) and *ad libitum* silage. Without any additives, the silage showed the highest pH value. The NH₃-N contents of DMP silages was lower (P<0.01) compared with the control silages, but did not appear to be significantly different from the combined silages. Silage intake tended to be higher in goats fed with DMP silages. However, no significant differences (P>0.05) of nutrient digestibility were observed in goats fed silages.

Keywords: dried mao pomaces, Thapra Stylo, silage, goat

**Corresponding author: asmerjai@hotmail.com*

Introduction

Mao luang is edible fruit tree (*Antidesma thwaitesianum*) divers in Northeast of Thailand. Mao juice and mao wine have become increasingly popular in Thailand and waste product such as mao pomaces from the process is plentiful. Several researches revealed that mao pomace contains high antioxidants, organic acid, amino acid and sugar (Samappito & Butkhup, 2008). Thus, dried mao pomaces (DMP) may have potential as silage additive by stimulating the lactic acid bacteria (LAB) growth on the ensiling process. *Thapra Stylo* is one of the most promising legume available for ruminant production in tropical area which contain high protein and grow over variety of soil types *Thapra Stylo* silage without any additives was difficult to make good quality silage, with high pH value and NH₃-N content (Liu et al., 2012). The aim of this study was to investigate the effect of applying DMP alone and combined with LAB on the fermentative quality and nutritive value of *Thapra Stylo* legume silages.

Material and methods

Fermented juice of lactic acid bacteria (FJLB) were prepared from fresh *Thapra Stylo* legumes by the method of Bureenok et al. (2011). *Thapra Stylo* legumes were chopped into 2-3 cm. length and

mixed with the silage additives. Silages were untreated (control) or prepared with DMP, or DMP plus FJLB (DMP+FJLB), or DMP plus *Lactobacillus plantarum* ST1 (DMP+Lp). DMP was applied at 100 g/kg of fresh matter (FM). FJLB and Lp were applied at log 6.03 and 5.58 cfu/g FM, respectively. Four male ruminally fistulated crossbred Boer x Saanen goats (~ 16 kg body weight) were randomly assigned to one of the four dietary treatment silages in a 4 x 4 Latin square design. The 28-d experimental period consisted of a 21-d feed intake and 7 d of sampling. All goats were received concentrate (comprising (%); cassava chips (54.5), rice bran (25.1), soybean meal (4.0) whole cottonseed (8.0), molasses (4.4), urea (1.8), salt (0.9), dicalcium phosphate (0.4), sulfur (0.1) and premix (0.4) at 1.5% of body weight (BW) and ad libitum silage. Feed and silages were sampled once a week and kept for analysing. Faeces samples were collected at the end of each period and analysed for DM, CP, EE, ash according to standard methods AOAC (1995). Neutral detergent fibre (NDF) and acid detergent fibre (ADF) were determined by the method of Van Soest et al. (1991). The data were analysed using the General Linear Model procedure by SAS. Silage profiles were compared by Duncan's multiple range test. Data of feeding trial were analysed by using the procedures of SAS for a 4 x 4 Latin square design.

Results

Dry matter content of silage was increased with addition of DMP portion. Without any additives, the silage showed the highest pH value. The NH₃-N contents of DMP silages was lower ($P < 0.01$) compared with the control silages, but did not appear to be significantly different from the combined silages (Table 1). It is generally accepted that in well preserved silages, pH values should be < 4.5 and NH₃-N < 100 g/kg total nitrogen (Umana et al., 1991). Clearly, all the treatment silages met these criteria. Silage intake tended to be higher in goats fed with DMP silages. However, no significant differences ($P > 0.05$) of nutrient digestibility were observed in goats fed silages (Table 2).

Table 1. pH value, NH₃-N concentration and chemical composition of Thapra Stylo legume silages.

Treatment	Control	DMP	FJLB +DMP	DMP+Lp	SEM	P-value
DM (g/kg)	305.81 ^d	404.59 ^a	333.27 ^c	367.28 ^b	5.09	<.0001
pH	4.49 ^a	4.01 ^c	4.31 ^b	3.98 ^c	0.03	<.0001
NH ₃ -N (g/kg TN)	89.30 ^a	52.36 ^b	80.30 ^{ab}	69.01 ^{ab}	9.85	0.1235
CP (g/kg DM)	107.12	105.79	100.55	106.44	3.70	0.6011
NDF (g/kg DM)	646.05 ^a	570.87 ^b	588.47 ^b	570.03 ^b	16.95	0.0251
ADF (g/kg DM)	365.15	337.44	340.13	328.60	11.13	0.1703

Means within the same row with different letters were significantly different ($P < 0.05$).

Table 2. Feed intake and nutrient digestibility of Thapra Stylo legume silages in goats.

Items	Control	DMP	DMP+FJLB	DMP+Lp	SEM	P-value
Silage intake						
%BW	2.5	3.1	2.4	2.6	0.11	0.314
g/kg BW ^{0.75}	51.8	64.9	50.8	55.0	2.25	0.309
Apparent digestibility (% of intake)						
DM	67.1	63.2	60.1	71.5	2.17	0.446
CP	66.9	68.6	61.6	75.6	1.77	0.224
NDF	62.0	53.7	52.0	65.4	3.12	0.511
ADF	64.8	59.6	55.7	67.7	2.46	0.446

Conclusion

Ensilage of Thapra Stylo legume with DMP showed a significant reduction in pH value and NH₃-N contents of silages. However, addition of 100 g/kg of DMP did not affect on silage intake and nutrient digestibility of goats.

Acknowledgements

Authors are greatly indebted to the Commission on Higher Education, Thailand Research Fund (TRF) and Rajamangala University of Technology Isan for funding this research.

References

- AOAC, 1995. Official Methods of Analysis, 12th ed. Association of Official Analysis Chemists. Washington, DC, USA.
- Bureenok, S., Suksombat W. and Kawamoto Y. 2011. Livest. Sci. 138, 266-271.
- Liu, Q., Chen M., Zhang J., Shi S., Cai Y., 2012. Characteristics of isolated lactic acid bacteria and their effectiveness to improve stylo (*Stylosanthes guianensis* Sw.) silage quality at various temperatures. Anim. Sci. J. 83, 128-135.
- Samappito, S., & Butkhup, L. (2008). An Analysis on organic acids contents in ripe fruits of fifteen Mao Luang (*Antidesma bunius*) cultivars, harvested from dipterocarp forest of Phupan valley in Northeast Thailand. Pakistan J. Biol. Sci., 11(7), 974-981.
- Umana, R., Staples, C.R., Bates, D.B., Wilcox, C.J., Mahanna, W.C., 1991. Effects of a microbial inoculant and (or) sugarcane molasses on the fermentation, aerobic stability, and digestibility of bermudagrass ensiled at two moisture contents. J. Anim. Sci. 69, 4588-4601.
- Van Soest, P.J., Robertson, J.B., Lewis, B.A., 1991. Methods for dietary fibre, neutral detergent fibre, and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74, 3583-3597.

Effect of combination acidifiers-garlic-*phyllanthus niruri* L. powder and encapsulated form in feed on production performance and egg quality of laying hens

Natsir¹, M.H., O. Sjoefjan¹, E.Widodo¹ & E.S. Widyastuti²

¹Department of Animal Nutrition and Feed Science, Brawijaya University, Indonesia, ²Department of Animal of Food Technology, Brawijaya University

Abstract

Effect of the combination of acidifier, garlic and *Phyllanthus niruri* L. in either powder or encapsulated form on layer performances and egg quality. One hundred ninety two laying hens were subjected to 8 different dietary groups, namely 2 forms of combination acidifier-garlic-*Phyllanthus niruri* L. (powder and encapsulated), and 4 levels usage in feed (L₀=0%, L₁=0,5%, L₂=1,0%, L₃=1,5%). Each treatment was repeated 4 times with 6 laying chickens each. Data were then analyzed by two-way Nested of Completely Randomized Design ANOVA and if there was significant effect followed by Duncan's Multiple Range Test. The results showed that combination of acidifier, garlic and *Phyllanthus niruri* L. in encapsulated form tended to decrease FCR and increase IOFC (P<0.01) and to increase (P<0.05) egg mass, but showed a no significant difference on feed consumption, HDP, eggshell thickness, color index of yolk and haught unit. Levels usage in feed of nested on forms gave significant results (P<0.05) on egg mass, but showed a no significant difference on feed consumption, HDP, feed conversion, IOFC, eggshell thickness, color index of yolk and haught unit. It is concluded that the use of combination of acidifier, garlic and *Phyllanthus niruri* L. in encapsulated form better than the powdered form. It is suggested to use 1.0% encapsulated of combination of acidifier, garlic and *Phyllanthus niruri* L. in laying hens diet.

Keywords: acidifier, garlic, phyllanthus niruri L., powder, encapsulated

Introduction

In general, problems of poultry farm are farmers use high level of antibiotics to decrease the problems of morbidity and mortality. Long term use of antibiotics may cause resistance and leave the residue in the meat and egg product that may be harmful to consumer. The combination of acidifier, garlic and *Phyllanthus niruri* L. can be used as feed additive to replace antibiotics. Acidifiers and phytobiotics however, are a natural antibiotics.

One of the efforts that can be done to go back to nature by utilizing an organic acid as a natural acidifier of fermented molasses. Garlic and *Phyllanthus niruri* L. can be used as a natural phytobiotics non antibiotics. Garlic has the ability to suppress pathogenic microbes and improve production performance, while *Phyllanthus niruri* L. give effect immunity and body resistance (immune), antibacterial and antioxidant

Materials and Methods

One hundred and ninety two 57 weeks old Isa Brown laying hens were used with uniform initial egg mass of 59.36 ± 3.08 g. They were randomly allotted to eight treatments and four replication. The experiment was designed based on Nested Completely Randomized Design with 2 main factors, namely form and level of the combination of acidifier, garlic and *Phyllanthus niruri* L. The forms comprised of powder and encapsulated forms, while levels of the combination of acidifier, garlic and *Phyllanthus niruri* L. consisted of 0, 0.5, 1.0 and 1.5% of the combination of acidifier, garlic and *Phyllanthus niruri* L. added to the basal diet. The composition of basal diet were formulated free antibiotics. Analyzed composition of basal diet are metabolizable energy 2,826 Kcal/kg, crude protein 17.43%, crude fiber 3.52%, calcium 3.73%, phosphorus 0.76%. The combination of acidifier, garlic and *Phyllanthus niruri* L. was prepared by mixing at a ratio 4: 3: 1. Then, the encapsulation process acidifier, garlic and *Phyllanthus niruri* L. mix using encapsulation method with the microwave oven (Natsir et al., 2013). The performances measured included feed consumption (FC), hen day production (HDP), egg mass, feed conversion rate (FCR), and Income Over Feed Cost (IOFC). While, the egg quality measured included eggshell thickness, haught unit (HU) and yolk colour. Data were then analyzed by two-way Nested of Completely Randomized Design and if there was significant effect followed by Duncan's Multiple Range Test.

Results and Discussion

Effect of acidifier, garlic and *Phyllanthus niruri* L. mixture form on Production Performance and Egg Quality of Laying Hens

The effects of acidifier, garlic and *Phyllanthus niruri* L. mixture in either powder or encapsulated form feed consumption, hen day production, egg mass, feed conversion rate, IOFC, eggshell thickness, HU and yolk colour were summarized in Table 1.

Table 1. Effects of powder or encapsulated acidifier, garlic and *Phyllanthus niruri* L. Mixture on production performances and egg quality of laying hens.

Variables	Treatments	
	Powder	Encapsulated
FC (g/bird/day)	119.93 ± 6.33	118.76 ± 7.03
HDP (%)	91.46 ± 6.85 ^a	94.66 ± 4.54 ^b
Egg mass (g/bird/day)	54.99 ± 4.41 ^a	57.74 ± 3.91 ^b
FCR	2.19 ± 0.12 ^B	2.06 ± 0.16 ^A
IOFC (IDR/bird)	221.11 ± 38.47 ^A	257.06 ± 42.90 ^B
Eggshell thickness (mm)	0.35 ± 0.01	0.36 ± 0.02
Color index of yolk	7.29 ± 0.62	7.41 ± 0.47
Haught unit	75.69 ± 5.91	78.55 ± 5.33

*IDR: Indonesian Rupiah (1US\$=Rp. 9.350)

The combination of acidifier, garlic and *Phyllanthus niruri* L. in encapsulated form tended to decrease FCR and increase IOFC and eggshell thickness ($P < 0.01$) and to increase ($P < 0.05$) egg mass, but showed a no significant difference on feed consumption, HDP, color index of yolk and

HU. If the comparative results between powder and encapsulated, combination of acidifier garlic and *Phyllanthus niruri* L. used in layer diet suggested that the use of encapsulated form was more effectively enhancing layer performances, respectively. El-Hakim et al. (2009) reported that the use of acidifiers, fitobiotik and mix both of them in broilers no significant effect on body weight, carcass characteristics, feed conversion, plasma proteins or organ weights of broilers aged 42 days. Furthermore, Isabel & Santos (2009) states that the addition of acidifier (a mixture of propionic acid and formic acid), essential oils from plant extracts (plant mix *Syzygium aromaticum* and *ceylanensis* Cinnamon and Cinnamon camphora (cinnamon) as well as mixed acidifier and essential oils from plant extracts did not show any significant effect on body weight broiler, but the FCR were significantly. There is a tendency of essential oils and essential oil blends-acidifier is better than the use of acidifiers. Natsir et al. (2010) reported that comparative effects of liquid and encapsulated lactic acid showed that the encapsulated form of lactic acid significantly affected body weight gain, feed conversion and IOFC, but no significant effect was observed for feed consumption and carcass percentage.

The effect of levels of acidifier, garlic and *phyllanthus niruri* L. mixture in powder and encapsulated form on production performances and egg quality of laying hens

The effects of acidifier, garlic and *Phyllanthus niruri* L. mixture addition in powder and encapsulated form on layer performances and egg quality were summarized in Table 2.

Levels usage in feed of nested on powder form gave no significant results ($P < 0.05$) on feed consumption, HDP, egg mass, feed conversion, IOFC, eggshell thickness, color index of yolk and HU. While levels usage in feed of nested on encapsulated form gave significant results ($P < 0.05$) on egg mass, but showed a no significant difference on feed consumption, HDP, feed conversion, IOFC, eggshell thickness, color index of yolk and HU. In addition, garlic powder addition did not significantly affect egg yolk index, egg shell weight and egg shell thickness, but showed gave significant results ($P < 0.05$) in egg albumen index, egg shell index, egg yolk cholesterol concentration and HU. Respectively, Narahari et al. (2009) reported that the egg size, shell thickness, albumen index, yolk index, HU and organoleptic acceptability of the eggs did no significant between zinc bacitracin and *Phyllanthus niruri* L.

Table 2. Effects of levels of acidifier, garlic and *Phyllanthus niruri* L. mixture addition in powder and encapsulated form on layer performances and egg quality.

Variables	Levels of powder/ encapsulated form in the diet (%)			
	0	0.5	1.0	1.5
Levels of powder form				
FC (g/bird/day)	121.15±4.32	117.87±9.89	120.78±1.26	122.06±1.91
HDP (%)	89.96±6.53	93.08±7.19	91.52±3.30	91.29±3.45
Egg mass (g/bird/day)	54.26±4.34	55.31±5.38	54.83±1.10	55.58±3.32
FCR	2.24±0.19	2.13±0.05	2.20±0.04	2.20±0.11
IOFC (IDR/bird)	10,176±1,123	10,189±284	10,071±1,078	11,071±834
Levels of encapsulated form				
FC (g/bird/day)	116.80±8.64	123.08±0.68	121.88±3.37	118.10±3.26
HDP (%)	89.73±6.70	94.36±0.86	96.98±1.55	97.57±1.96
Egg mass (g/bird/day)	53.38±4.51	57.27±1.56	59.32±1.59	61.00±2.80
FCR	2.19±0.10 ^b	2.15±0.07 ^b	2.06±0.08 ^a	1.94±0.10 ^a
IOFC (IDR/bird)	10,729±830	11,574±1,339	12,769±444	12,574±702

^{a,b}Means with different superscripts in the same row differ ($P < 0.05$).

Conclusion

It is concluded that the use of combination of acidifier, garlic and *Phyllanthus niruri* L. in encapsulated form better than the powdered form. It is suggested to use 1.0% encapsulated of combination of acidifier, garlic and *Phyllanthus niruri* L. in laying hens diet.

References

- El-Hakim, A.S.A., G. Cherian and M.N. Ali, 2009. Use of Organic Acid, Herbs and Their Combination to Improve the Utilization of Commercial Low Protein Broiler Diets. *Int. J. Poult. Sci.* 8 (1): 14-20.
- Isabel, B. and Y. Santos, 2009. Effects of dietary organic acids and essential oils on growth performance and carcass characteristics of broiler chickens. *J. Appl. Poult. Res.*, 18:472–476.
- Narahari, D, G.R. Manohar, S. Suba, and R. Thiruvengadam, 2009. Performance and egg value enhancing abilities of herbal feed supplements in layer diets. *Indian J. of PoultSci.* 44 (1): 55-58.
- Natsir, M.H., O. Sjojfan, K. Umam, A. Manab and E.Widodo, 2010. Effect of Liquid and Encapsulated Lactic Acid in Broiler Diets on Performances, Intestinal Characteristics and Intestinal Microflora. *J. Poult. Sci* 47: 240-243.
- Natsir, M.H., Hartutik, O. Sjojfan, and E. Widodo, 2013. Effect of Either Powder or Encapsulated Form of Garlic and *Phyllanthus niruri* L. Mixture on Broiler Performances, Intestinal Characteristics and Intestinal Microflora. *Int. J. Poult. Sci* 12 (11): 676-680.

Characteristics of fermentation kinetics and digestibility of PUFA saponification and aldehyd protected as cattle feed supplement by in-vivo

Riyanto, J* and Sudibya

Animal Husbandry Department of Agriculture Faculty of Sebelas Maret University, Surakarta, Indonesia

Abstract

This study was conducted to evaluate the effect of different saponification and aldehyde protection for poly unsaturated fatty acid (PUFA) derived soybean groats and lemuru fish oil in the diet beef cattle on the characteristics of fermentation kinetics by in vivo. Invivo treatment using three cow cattle fistulated. Oil lemuru protected saponification using salt-Ca with 2.0% level and soybean groats protected using aldehyde content of 37% with a level of 2.0%. Experimental design 3x3 latin squares. R0 (control diet), R1 (R0 + 10% PUFA saponification protected), and R2 (R0 + 10% PUFA aldehyde protected). Measurement of the average and standard deviation of the kinetics fermentability ration in the rumen on the hours to 0, 3, 6, 9, 12, and 24. Parameter research include rumen fermentation parameters are pH, VFA and microbial protein and digestibility are dry matter, organic matter and crude protein. Results of the study was the difference in treatment effect was not significant ($p > 0.05$) on dry matter, organic matter and crude protein. Protection efforts on feed with PUFA additional protection soya groats aldehyde and lemuru fish oil saponification protected gives a good fermentation characteristics in creating rumen ecology and in accordance with the needs of the rumen microbes. Conclusions are protection of saponification and aldehyde effective for PUFA protection and can be used up to the level of 10% in the composition of beef cattle rations.

Keywords: kinetics, digestibility, PUFA, saponification, aldehyde protected, in-vivo

**Corresponding author: jokoriyanto@staff.uns.ac.id*

Introduction

Improved reproductive performance of the cow can be done through the use of polyunsaturated fatty acids (PUFA). PUFA during digestion in the rumen always had biohydrogenation by rumen microbes become saturated fatty acids (Lourenc et al., 2010). It is necessary for the protection so that PUFA treatment once escaped from biohydrogenation protein called rumen bypass fat-protein to be absorbed in the small intestine. PUFA protection treatment on the feed material can be used formaldehyde to protect soybean groats (Riyanto, *et al.*, 2011) and flour cereals (Mohammadian-Tabrizi *et al.*, 2011),

Materials and Methods

In vivo treatment using three cow cattle fistulated and range of initial body weight of 350 kg of 30 head. Three ruminally fistulated Ongole crossbred cow were employed in this study as the ruminal fluid donors. Lemuru fish oil protected by saponification using salt-Ca with 2.0% level (PUFA saponification protected) and soybean groats protected using aldehyde content of 37% with a level of 2.0% (PUFA aldehyde protected) (Riyanto, *et al.*, 2011). Each treatment used rumen fluid

from three Ongole crossbred cow fistulated and repeated 3 times. In vitro test using 3 repetitions per treatment. Provided basal feed consisted of rice straw fermentation (JPF) and basal concentrate (BC) with a ratio = 40%: 60% (dry matter basis). The composition of the control feed (R0) is composed of 30% fermented rice straw, 30% elephant grass, 40% concentrate basal (6.0% of palm oil, 9.2% copra, 12.8% bran, 10.8% cassava waste, 0.8% mineral and 0.4% salts). Measurement of ration fermentability is conducted in specified time to determine the ruminal kinetics, 0, 3, 6, 9, 12, and 24 hours after feeding. Experimental design 3x3 latin squares. R0 (control diet), R1 (R0 + 10% PUFA protected saponification), and R2 (R0 + 10% PUFA protected aldehyde). Research parameters include rumen fermentation parameters are kinetics of ruminal pH, VFA (Volatile Fatty Acids) production, and microbial protein. Digestibility of dry materials, organic materials, and crude protein.

Results and Discussion

VFA kinetics, pH and microbial protein fermentation research results rumen fluid for 24 hours can be seen in Figure 1

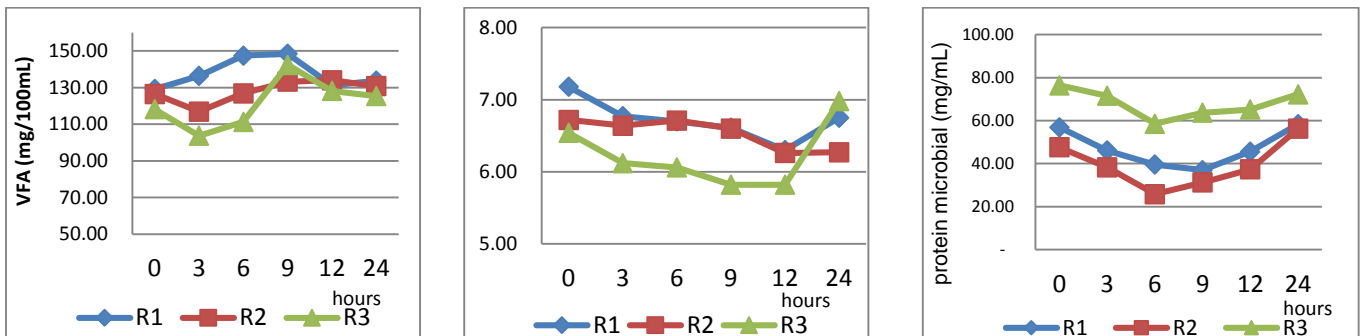


Figure 1. Kinetics of VFA, pH and microbial protein

The pattern of VFA and pH kinetics for both is almost the same while a different pattern for microbial protein kinetics of microbial protein treatment that R3 is above R1 and R2. This is related to the absorption of VFA as an energy source through the rumen wall; the lower the ruminal pH, the higher the VFA absorption (Owens and Goestch, 1988). It is caused by the synchronization of the nitrogen source provision required by the microbes in the form of NH₃ and VFA. The total range of 33.40 to 61.80 mM VFA in this research is still in the normal range. According to Mc Donald *et al.*, (2002) VFA concentration in the rumen ranges from 10-70 mM. Range pH of 6.90 to 6.98 is still within the limits of normal pH for the digestion of feed in the rumen. At these ranges is still able to maintain rumen microbial life. Protection treatment of formaldehyde on proteins can form chemical bonds with proteins that are stable at neutral pH such as rumen pH, but becomes unstable at acidic pH as the pH abomasum (Jenkins, 1993). Protein feed will be fermented into ammonia and VFA are absorbed as energy source and carbon frame together ammonia (NH₃) for microbial protein synthesis (Doreau and Chilliard, 1997). Ability limited rumen microbial cell surface to come into contact with food that is wrapped by fatty acids (Johnson, 2007). Mc Donald *et al.* (2002) microbial proteins that have undergone lysis, utilized by the host as a source of essential amino acids. Proteins that enters the duodenum is derived from feed protein that escapes from the rumen microbial degradation and microbial protein.

Digestibility of dry matter, organic matter and crude protein of the results of this research can be seen in Table 1

Table 1. Digestibility of dry matter, organic matter and crude protein of protected soybean groats and lemuru fish oil

Parameters	R1	R2	R3
Dry matter (%)	55,53±4,03 ^a	59,2±6,65 ^a	61,53±5,18 ^a
Organic matter (%)	59,71±1,09 ^a	68,86±3,73 ^b	77,23±1,20 ^c
Crude protein (%)	58,35±2,52 ^a	63,4±1,56 ^b	76,21±1,20 ^c

Footnote : P-value for main effects of parameter; means a common superscript at rows are differ significantly affect (P≤ 0.05)

In addition the average pH of the rumen fluid the third are still in the normal range so optimal rumen microbial activity and can improve the process of fermentation in the rumen whole. No negative effects of the use of formaldehyde in the ration on consumption and digestibility of dry matter (Riyanto, *et al.*, 2011). Dry matter digestibility of organic matter can affect. Organic matter digestibility is directly proportional to the dry matter digestibility because organic materials as the building blocks of dry matter. Feed with sufficient protein to provide nitrogen such as NH₃ to microorganisms and energy sources as well enough for rumen microbial digestion of organic matter will help to run normally. Stanton *et al.*, (1983) formaldehyde protected soybean meal with 0.3% levels obtained results rumen nitrogen digestibility decrease compared with the control, 0.2% and 0.6 digestibility of nutrients depends on the activity of microorganisms in the rumen fermentation process.

Conclusions

Conclusions are protection of saponification and aldehyde effective for PUFA protection and can be used up to the level of 10% in the composition of beef cattle rations.

References

- Doreau, M., and Y. Chilliard. 1997. Digestion and metabolism of dietary fat in farm animals. *Br. J. Nutr.* 78:S15–S35.
- Jenkins, T. C. 1993. Lipid metabolism in the rumen. *J. Dairy Sci.* 76: 3851 – 3863.
- Johnson, M.C. 2007. Understanding Rumen Fermentation : I. Effect o high DHA Algae oil on microbial biohydrogenation and II. Monitroing microbial shifts in response to antibiotics and oil using T-RFLP analysis. Thesis. Master o Science Nutrition and Animal Science, Raleigh, North California, USA.
- Lourenc, M., E. Ramos-Morales and R. J. Wallace- 2010. The role of microbes in rumen lipolysis and biohydrogenation and their manipulation. *Animal* (2010), 4(7):1008–1023 & *The Animal Consortium* 2010
- McDonald, P., R.A. Edward and J.F.D. Greenhalgh. 2002. *Animal Nutrition*. 4th edition. English Language Book Society, Longman, London.
- Mohammadian-Tabrizi, H. R., H. Sadeghipanah, M. Chamani, Y. Ebrahim-Nejad1 and H. Fazaeli. 2011. *In vitro* gas production of wheat grain flour coated with different fat types and levels. *African J. Biotech.* 10(39): 7710 - 7716.
- Riyanto, J., S.D. Widyawati dan W. Pratitis.. 2011. Supplementation with PUFA (poly unsaturated fatty acid) in concentrate from the local feed ingredients in Feedlot cattle business crosses of rice straw basal diet based on fermentation to produce beef low in fat and cholesterol and high in unsaturated fatty acids. Research report "National Strategy Grant" Funded by the Directorate General of Higher Education, Ministry of National Education, According to the Letter of Agreement Implementation Research Grant
- Scollan, N. D. , Nag-Jin Choi, E. Kurt, A.V. Fisher, M. Enser and J. D. Wood .2001. Manipulating the fatty acid composition of muscle and adipose tissue in beef cattle. *British Journal of Nutrition* (2001),
- Stanton T. L. , F. N. Owens and K. S. Lusby. 1983. Formaldehyde-treated soybean meal for ruminants grazing winter range grass. *J. Anim. Sci;* 56: 6 - 14.

Effect of usage probiotics powder as feed additive on the eggs quality of laying hens

O Sjoftjan¹, M. H. Natsir¹, T Ardiyati², and K Winarni³

¹Animal Nutrition Department, Animal Husbandry Faculty, Brawijaya University

²Microbiology Department, Natural Sciences Faculty, Brawijaya University, Malang Indonesia

³PT. Petrokmia Kayaku Research Center. Gresik, East Java Indonesia

Abstract

The purpose of this study was to find the effect of probiotic level in feed additive on egg weight, egg mass, specific gravity, fat and cholesterol of egg yolk. The materials used in this experiment were 400 laying hens aged one year old. Feeds were used self mix feed concentrate and probiotic powder (lactobacillus sp). Graded levels of probiotic powder were added to dietary formula treatments consisted of four treatments. Probiotic powder is added at 0% in the feed (P0), 0.2% in the feed (P1), 0.4% in the feed (P2), and 0.6% in the feed (P3). Each treatment was repeated five times. The variables measured were egg weight (g/bird), egg mass (g/bird/day), specific gravity (g/cm³), fat (%) and cholesterol of egg yolk (100mg/g). Data were analyzed by analysis of variance of the Completely Random Design (CRD), if between treatment showed significant effect were analysed by Duncan's Multiple Range Test (DMRT). The result showed that the usage of probiotic powder in feed was significant effect influenced ($P < 0,01$) by increase egg weight, egg mass and decrease fat and cholesterol content of egg yolk, but there isn't effect on specific gravity ($P > 0.05$). The addition of probiotic powder as feed additive as much as 0.4% gives best effect on egg weight, egg mass and yolk cholesterol content. It can be concluded that the addition of probiotic powder in feed can improve egg quality of laying hens.

Keywords: probiotic, powder, egg quality, laying hens

Corresponding author: osoftjan@yahoo.com

Introduction

Probiotics are microbial feed supplement, microorganisms that can improve the balance of microflora in the digestive tract of the host animal (Fuller, 1989) Probiotics can be derived from bacteria, yeast and others. Some species of probiotic bacterial origin that can be used as probiotics include *L acidophilus*, *L bulgaricus*, *L casei*, *L fermentum*, *L plantarum*, *L reuteri* and *Bifidobacterium sp*, *B subtilis*, *B cereus*, *B megaterium*, *B coagulans*, *B circulans*, and *B alvei* etc. In this study, using a mixture of probiotic cultures such as *Bacillus spp.* and *Lactobacillus sp.* Kompang (2009) states that the performance of laying hens that received *Bacillus spp.* (*Bacillus cereus*, *Bacillus subtilis* and *Bacillus licheniformis*) is better than that received antibiotics virginiamycine. Mix culture of *Bacillus spp.* can serve as a growth promoter replace egg-synthetic antibiotic growth promoter. Kompang (2000) adds the provision of *Bacillus spp.* mixed in the feed, has been reported to increase egg production and FCR. In addition it has been reported that a group of bacteria also can be used as probiotics and improve the performance of broilers and can be competitive with pathogenic bacteria such as *Salmonella* (Jin et al., 1996 and Barrow, 1992). Bacteria *Lactobacillus sp.* has the ability to remodel simple carbohydrates into lactic acid. Along

with the increase of lactic acid, the pH of the environment is low and causing pathogenic microorganisms do not grow. When it occurs on the surface of the gastrointestinal tract colonization, *Lactobacilli* prevent the growth of mold and suppress the growth of pathogenic *E coli* and gram-negative bacteria in the small intestine. *Lactobacillus sp.* can maintain the balance of the population of other bacteria in the small intestine. *Lactobacillus sp.* can colonize the surface of the digestive tract, if they get the proper environment and nutrition. After the meal, a culture of *Lactobacillus sp.* will produce lactic acid balance of microflora in the duodenum, ileum and caecum chickens within 24 hours. According Cammarota et al., (1991) *Lactobacillus sp.* has the ability to convert cholesterol into koprostanol the type of plant sterols that cannot be absorbed by the intestines of man. Koprostanol is a natural steroid that can be produced by bacteria in the lower intestine of man or animals and excreted through feces. One of the functions of lactic acid bacteria which reduce serum cholesterol. Cholesterol in the intestine will be converted into koprostanol so cannot be absorbed by the intestine and will come out with feces. The use of cholesterol reeducate enzyme produced from culture isolates of *Lactobacillus sp.* to reduce the amount of cholesterol absorbed in the intestines of animals will not degrade the quality of the resulting product, and does not cause severe side effects because the enzyme is derived from a protein in which the high temperature will be denatured. Cholesterol reductase enzyme mixes with the cytosol of *Lactobacillus sp.*; it is easy to be extracted because it dissolves in water. Barrow (1992) adds that the addition of *Lactobacillus* 2% and 4% in addition to lower feed conversion also lowers the concentration of cholesterol eggs, so as to have an attraction for farmers and consumers.

Material and methods

This study uses the period of laying hens layer 200 tails. Laying hens were placed in battery cages. Probiotic cultures used a mixture composed of two types of micro-organisms such as *Bacillus spp.* and *Lactobacillus sp.* in the form of flour content of TPC microbe that is 2.18×10^9 cfu / ml. Feed treatment used in this study are basal feed composition in the form of corn 50%, 20% bran and 30% concentrate. With a basal feed food substances: Dry matter 88.90%; ash 14.70%; crude protein 17.80%; crude fat 4.73% Crude fiber 6.72% and metabolizable energy 3750 Kcal / kg.

The method used is a field experiment with completely randomized design (CRD) using 4 treatments and each treatment was repeated 5 times. Food and drink provided ad libitum. Feed treatment is given as follows:

P0 = basal + feed probiotic powder level 0%

P1 = feed + probiotic powder basal level of 0.2%

P2 = feed + probiotic powder basal level of 0.4%

P3 = feed + probiotic powder basal level of 0.6%

Variables Research

Egg weight (Bell and Weaver, 2002). Egg Mass (Novak et al., 2006). Gravity eggs (Butcher and Miles, 1991 Egg Yolk fat (AOAC, 2005) and Egg Yolk Cholesterol (Bell. and Weaver, 2002).

Data Analysis

Data were analyzed using analysis of variance of completely randomized design (CRD) if there is a difference real effect or very real followed by Duncan's Multiple Range Test (DMRT).

Result and discussion

Table 1. Effect of Probiotic *Lactobacillus* sp Addition Form Feed Wheat In the Quality Egg Laying hens

Treatments	Variables				
	Egg Weight (g/egg)	Egg Mass (g/egg/day)	Egg Gravitation (g/cm ³)	Egg yolk Fat (%)	Egg yolk Cholesterol (mg/100 g)
P0	60.53±0.53 ^a	58.50±0.40 ^b	1.06±1.67	36.12±1,86	217.85±0.64 ^d
P1	62.15±0.22 ^b	59.77±1.25 ^b	1.06±0.89	36.35±1,21	212.93±0.21 ^c
P2	62.67±0.22 ^b	59.74±1.98 ^b	1.07±1.00	37.17±1,16	210.05±0.31 ^b
P3	60.07±0.51 ^a	52.49±1.85 ^a	1.02±2.30	36.53±0,70	20820±0.53 ^a

Description: different superscript in the same column shows the difference significantly (P <0.01).

According to table 1. The treatments provide significant influence {P <0.01} in the weight of the eggs. This is in accordance with Sjojfan (2013) which states that the addition of probiotic *Lactobacillus* sp levels in feed may increase the weight of the eggs. The addition of probiotics in the digestive tract of chicken will increase the availability of energy and protein and other nutrients needed to form a chicken egg at the time of production. Conversely, a decrease in energy and protein content in the diet tend to lose eggs. Gleaves et al., (1997) stated that in addition to the energy content in the feed which is one of the determinants of chicken egg weight, there are also environmental factors that influence egg weight that temperature.

Statistical analysis results showed that the treatment effect is highly significant (P <0.01) in to the egg mass laying hens. Egg mass is the average daily egg weight so that the percentage of egg production will affect the egg mass. Egg mass is also influenced by the production and egg weight, if one or both of the higher factor then egg mass also increased and vice versa. Standard egg mass laying hens aged 1 year according to ISA (2012) is 56.9 g / head / day. Novak et al., (2006) adds that the egg mass is also determined by protein intake during the spawn. In connection that the mass of eggs are influenced by the weight of albumin and egg yolk, which is largely composed of protein, therefore high protein intake causes high egg mass..

Analysis results showed that the treatment was not statistically significant (P > 0.05) against gravity eggs. Koelkebeck (2003) states that the factors affecting the gravity egg is an egg storage time, temperature, time of laying and the calcium content of the feed. Probiotics are used in the study contains calcium derived from skim milk are used as materials for probiotic *Lactobacillus* form of flour, but still not enough to increase the gravity of eggs. Ahmad et al., (2003) add the egg consists of four basic components, namely the yolk, egg white, eggshell and eggshell membrane. Weight of different types of each component that shell of 2.33 g / cm³, egg yolk 1.03 g / cm³, egg white 1,04g / cm³ dan eggshell membrane 1.08 g / cm³. Values higher gravity of the eggshell twice compared with egg yolk, egg white and eggshell membrane. Therefore the shell has a major influence on the gravity of the eggs intact. Gary and Richard (2003) stated the value of the eggs gravity would decrease with a decrease in the age of the chicken. This is because the size of eggs has increased from heavy shell. The difference in value of gravity on the eggs with the same weight due to the difference in eggshell thickness.

Statistical analysis results show that the addition of probiotic *Lactobacillus* sp. powder in the feed had no effect on the fat content of the yolk. Bell and Weaver (2002) states affected by the weight of the egg yolk fat because fat deposits are the largest in the yolk. Egg yolk composition is 50% water, 32% -36% fat, 16% protein and glucose 1% -2%. Fatty acids are abundant in egg yolks is linoleic, oleic and stearic. Levels of fat in an egg is influenced by the levels of fat in feed serves as a source of energy and can be manipulated via the feed, for example by adding corn or sunflower oil. The standard yellow fat content of eggs is 31.80% - 35.50%, while the observations found the egg yolk fat content of 56.12% -57.19%. The fat content is high enough this may be due to the increasing age of the chicken. In accordance with the statement of Yamamoto et al., (2007) that the fat content in the yolk increases with increasing age of the chicken.

Statistical analysis results showed that the treatment was highly significant (P, 0.01) in the content of egg yolk cholesterol. A decrease in the cholesterol content of egg yolk is caused due to the activity of the probiotic *Lactobacillus* sp. in the chicken digestive tract. This is consistent with the results of research Jernigan and Miles (1995) that the addition of *Lactobacillus* sp in the feed of laying hens capable of lowering cholesterol egg yolk. Mechanism of action of *Lactobacillus* sp. which is able to produce lactic acid in large quantities of simple carbohydrates, the excess of the basic necessities of life led to the formation of chicken fat and cholesterol in the body will decrease. A decrease in the cholesterol content of laying hen's organ will lead to a decrease in cholesterol eggs produced. Milati et al (2013) adds the use of probiotic *Lactobacillus* sp 7.5% in chicken rations Arab capable of lowering cholesterol eggs 114.58 mg / 100g lower than the control 145.88 mg / 100g. Andriani (2005) adds that the administration of probiotic microbes could help degrade cholesterol by converting cholesterol into bile acids colic and thus the concentration of cholesterol in the blood can be reduced and levels of cholesterol in the blood becomes stable. Probiotics are commonly used which are composed of microbes *Lactobacillus* sp. and *Bifidobacterium*.

Conclusion

The addition of probiotic *Lactobacillus* sp. powder in the feed can improve egg weight, egg mass and decrease egg yolk cholesterol content. The addition of 0.4% probiotics provides the best effect on the quality of egg laying hens.

Acknowledgement

I would like to thank the Ministry of Education directorate of higher education and culture which funded the research.

References

- Andriani, L. 2005. Effect of Balance bacterium *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, *Lactobacillus* and *Bifidobacterium* in Yoghurt against Blood Cholesterol Content of Mice. Faculty of Animal Husbandry. University of Malang.
- Barrow, P. A. 1992. probiotics for Chicken. In probiotics The Scientific Basis. Edited by Fuller, Chapman and Hall. London. New York. Tokyo. Melbourne. Madras.pp.225-252.
- Bell, D. and G. Weaver. 2002. Commercial Chicken Meat and Egg. Kluwer Academic Publishers, United States of America.
- Butcher, G .D. and D. R. Miles. 1991. Egg-Designing a Specific Gravity Monitoring Program. Institute of Food and Agricultural Science. Florida. www.pjbs.org.
- Fuller, R. 1992. History and development of probiotics. In: The Scientific Basis probiotics. Fuller (Ed.). Chapman & Hall. London, New York, Tokyo, Melbourne, Madras.
- Gary, D. B. and D. M. Richard. 2003. Egg-Designing A Specific Gravity Monitoring Program. Poultry Veterinarian. Poultry Nutrition. University of Florida. Gainesville. www.hammock.ifas.ufl.edu.
- Gleaves, W .e., F. B. Mather and M. M. Ahmad. 1997. Effects of dietary calcium, protein and energy on feed intake, egg shell quality, and hen performance. *Poultry Science* 56: 402-406.
- Gumilang, D. 2013. The role of Probiotic Bacteria in Production Performance and Egg Quality infected by *Salmonella enteritidis*. Faculty of Animal Husbandry. Bogor Agricultural Institute.
- ISA. 2012. Product Performance. Isa Brown Commercial Layers. www.isapoultry.com.
- Jernigan, M. A., and R. D. Miles. 1995. probiotics in Poultry nutrition. *World's Poultry Science* 2: 99- 105

- Koelkebeck, WK 2003. What Is Egg Quality and conserving It. Ilinin PoultryNet-University of Illinois. www.poultrynet.com.
- Kompiang, IP 2000. Effect of Supplementation of culture Bacillus spp. Through Feed Or Water on Performance of Laying Chicken. JITV. Vol. 5: 205-209.
- Kompiang, IP, 2009. Utilization of Probiotic Microorganisms In order to Increase Poultry Production in Indonesia. Agricultural Innovation Development 2 (3), 2009: 177-191.
- McNaught, C and J. Macfie. Probiotics 2000. In Clinical Practice: A Critical Review Of The Evidence. Nutrition Research 21 (2001) 343-353.
- Milati, A., N. Iriyanti and Mugiyono. 2013. Use of Various Types of Probiotics In rations Against Fat and Cholesterol Levels Chicken Egg Yolk Arabic. Faculty of Animal Husbandry. University General Soedirman. Purwokerto.
- NRC. 1994. Nutrient Requirements of Poultry: Ninth Revised. Ninth Revised Edition. Natural Academy Press. Washington DC.
- Novak, C., H. M. Yakout and S. E. Scheideler. 2006. The effect of dietary protein level and total sulfur amino acid: Lysine ratio parameters on egg production and egg yield in hy-line W-98 hens. Poult. Sci. 85: 2195-2206.
- Sjofjan, O. 2003. Study of Probiotic (Aspergillus niger and Bacillus sp.) As Feed rations and Implications The gut microflora and Form Laying Chicken Production. Dissertation. Padjadjaran University, Bandung.
- SNI. Chicken Eggs 1995. Consumption. The National Standards Council. Jakarta.
- Yeo, J and K. L. Kim, 1997. Effect of Feeding Diets Containing Antibiotics on, a Probiotics, Or Yucca Extract on Growth and Intestinal Urease Activity in Broiler Chicks. Poult. Sci. 76: 381-385.

Supplementation of different sources of nitrogen and its effects on rumen microbial biomass and *in vitro* feed degradability parameters

Marjuki¹, Kusmartono¹ & Daqaqa, A.L.R².

¹Faculty of Animal Husbandry, University of Brawijaya, Malang- 65145, Indonesia

²M.Sc. Student, Postgraduate Program, Faculty of Animal Husbandry, University of Brawijaya, Malang- 65145, Indonesia

Abstract

Rumen microbes have essential roles in supplying nutrients for the host animal, hence, the growth and activities of the rumen microbes affect significantly to ruminant animal performance. Non-protein nitrogen (NPN) that is easily converted to ammonia in the rumen is as main source of nitrogen for rumen microbes to grow. However, the efficiency of the ammonia utilization by the microbes must depend on some factors including the nitrogen sources. This experiment aimed to determine the effect of different sources of nitrogen on *in vitro* rumen ammonia concentration, rumen microbial biomass synthesis, apparent and true feed degradability. Completely randomized block design of six treatments and three replications were used. The treatments were the use of different form of nitrogen sources to supplement control diet (rice straw and concentrate at 1:1 ratio in DM basis). The nitrogen sources were only urea, ammonium sulphate (ZA), or NPK fertilizer, combination of urea, ZA and NPK (1:1:1 ratio N basis), and combination of urea, NPK, ZA at a total of N:P:S ratio of 12:2:1. The amount of nitrogen sources added was set to be equivalent to an addition of ammonia concentration into the rumen liquid as much as 2.5 mg NH₃-N per 50 ml rumen liquid. Supplemented diets with different sources of nitrogen showed higher feed degradability, gas production, microbial biomass synthesis, and ammonia concentration than control diet. Among the nitrogen supplemented diets, diet supplemented with mixed of urea, NPK, ZA at N:P:S ratio of 12:2:1 showed the highest total microbial protein synthesis (11.2 g microbial N), efficiency of microbial protein synthesis (57.0 g microbial N/ kg FOMR), ammonia concentration (150.7 mg NH₃-N/l), and feed degradability (61.9%) next to ZA supplemented diet. While, NPK supplemented diet showed the lowest total microbial protein synthesis (10.2 g microbial N), efficiency of microbial protein synthesis (45.6 g microbial N/ kg FOMR), ammonia concentration (111.1 mg NH₃-N/l), and feed degradability (57.6%) that were close to the control diet. Supplementation of mixed nitrogen sources (urea, ZA, and NPK at total N:P:S ratio of 12:2:1) to diets should be used to support high rumen microbial growth and increase nutrient supply for higher ruminant productivity.

Keywords: urea, ammonium sulphate, NPK, microbial protein synthesis, degradability

Corresponding author: marjuki@ub.ac.id

Introduction

Rumen microbes have very important or even essential role for ruminant. The microbes help in utilization of low quality feeds especially fibrous feeds and in conversion of non-nitrogen protein into high quality microbial protein. Both processes cannot eventually be done by the ruminant itself without any help of rumen microbes. By those two processes, hence, rumen microbes supply most of nutrients for the host animal. A major source of energy and protein for ruminant are supplied by end products of fibrous feed fermentation done by rumen microbes, and microbial rumen microbial protein flowing to the small intestine, respectively. These roles must depend mostly on rate of microbial growth in the rumen, their composition and activities. For their optimal growth, rumen microbes require optimal rumen conditions and supply of nutrients including carbohydrates and carbon skeleton, protein or nitrogen, vitamin, minerals and water. To ensure the optimum rumen microbial growth, all of the nutrients must be available in right time and in most appropriate proportion.

The unique of the rumen microbes is that they have the ability to utilize non-protein nitrogen (NPN) as their nitrogen source for their growth. Bondi (1987) reported that rumen microbes use rumen NH_3 in the range of 50 to 80% for their growth, especially in the synthesis of microbial protein. However, in the synthesis of protein, NH_3 does actually supply only amine groups in amino acids structure, hence it is required some other components to synthesize amino acids or protein, including energy, carbon skeleton, sulfur, and phosphor. Sulfur and phosphor are also essential element for rumen microbial growth, especially when the microbes utilizes NH_3 for amino acids synthesis during their growth. Sulfur is used for the formation of sulfur containing compounds in the microbial cells such as essential amino acids cysteine and methionine. Phosphor is used by rumen microbes for synthesis of DNA, RNA, ATP, membranes and protein synthesis. N:S ratio required for rumen microbial growth is 12:1 (Preston & Leng, 1987) and N:P ratio is 6:1 (Schwab *et al.*, 2005), hence based on those ratios, N:P:S ratio required for rumen microbial growth should be 12:2:1. Thus, it is important to evaluate the use of compounds which contain not only nitrogen, but also either sulfur or phosphor to supply nutrients for better rumen microbial growth and feed degradability.

Materials and Methods

The study was conducted in the Laboratory of Animal Feed and Nutrition, Faculty of Animal Husbandry, University of Brawijaya, Malang. Indonesia using *in vitro* gas techniques (Blümmel *et al.*, 1997a) and completely randomized block design of six treatments and three replications. The treatments were supplementation of three different N sources and their combination at amount to reach an additional of NH_3 -N concentration in the rumen liquid as much as 50 mg NH_3 -N/l or equals to 2.5 mg NH_3 -N/50 ml rumen liquid (Satter & Slyter, 1974). The N sources were urea, ammonium sulphate (ZA), NPK fertilizer. The treatment details were **P0** = 250 mg rice strew + 250 mg concentrate as fed basis; **P1** = P0 + urea (5.4 mg); **P2** = P0 + NPK (15.0 mg); **P3** = P0 + ZA (11.8 mg); **P4** = P0 + urea (1.8 mg) + NPK (5.0 mg) + ZA (3.9 mg) to get N supply ratio from the three sources = 1:1:1; **P5** = P0 + urea (4,2 mg) + NPK (0.2 mg) + ZA (2.7 mg) to get N:S:P supply ratio of 12:2:1. All feeds in the treatments were put in syringes separately, added with 50 ml buffered rumen liquor and incubated in incubator for 48 hours at 39°C Parameters measured were gas production and DM degradability *in vitro*, NH_3 concentration, microbial biomass

(Blümmel et al., 1997b). The data were subjected to analysis of variance followed by Duncan Multiple Range Test.

Results and Discussion

Data of in vitro gas test parameters of control and different source of nitrogen supplemented diets were presented in Table 1. It is shown in Table 3 that all supplemented diets with different sources of nitrogen showed higher feed degradability, gas production, microbial biomass synthesis, and ammonia concentration than control diet. Among the nitrogen supplemented diets, diet supplemented with mixed of urea, NPK, ZA at N:P:S ratio of 12:2:1 (P5) showed the highest total microbial protein synthesis (11.24 g microbial N), efficiency of microbial protein synthesis (57.03 g microbial N/ kg FOMR), ammonia concentration (150.73 mg NH₃-N/l), and DM degradability (61.87%) next to ZA supplemented diet. While, NPK supplemented diet showed the lowest total microbial protein synthesis (10.25 g microbial N), efficiency of microbial protein synthesis (45.60 g microbial N/ kg FOMR), ammonia concentration (111.06 mg NH₃-N/l), and feed degradability (57.64%) that were close to the control diet.

Table 1. Total and efficiency of microbial protein synthesis, ammonia concentration, in vitro true degradability, and gas production at 48 hrs incubation of control and treatment diets.

Treatment	Total MPS* (g microbial N)	Efficiency of MPS* (g microbial N/kg FOMR)	NH ₃ Conc. (mg NH ₃ -N/l)	True degrad. (%)	Gas production (ml/500 mg DM)
P0	10.34 ^{ab}	45.41	105.96 ^a	57.17 ^a	71.33 ^a
P1	11.10 ^c	46.98	119.00 ^a	59.57 ^c	81.66 ^c
P2	10.25 ^a	45.60	111.06 ^a	57.64 ^{ab}	79.55 ^{bc}
P3	11.59 ^{cd}	47.15	146.76 ^b	61.91 ^d	74.10 ^{ab}
P4	11.08 ^{bc}	47.80	148.18 ^{bc}	59.05 ^c	77.87 ^{bc}
P5	11.24 ^{cd}	57.03	150.73 ^{bc}	61.87 ^d	77.22 ^{abc}
Sign.	***	Ns	***	**	***

MPS = Microbial protein synthesis, FOMR = Fermented organic matter in the rumen, ns: not significantly different, **: significant different, ***: highly significant different.

As stated by Preston & Leng (1987), Schwab *et al.* (2005), and other researchers, it was agreed that rumen microbes require N, P, and S as a part of their substrates for their growth. Sulfur is one of important mineral to support rumen microbial growth, especially when ruminants are fed large amounts of non-protein nitrogen. Sulfur is utilized in synthesis of sulfur containing essential amino acids, methionine and cystine. In addition phosphorus is also essentially required for the growth of rumen microbes. Phosphor is mainly used as main component of cell's nucleic acid DNA and RNA as well as ATP. Hence, limited intake of sulfur may restrict rumen microbial growth (Pathak, 2008).

Conclusion

Supplementation of mixed nitrogen sources from urea, ZA, and NPK at total N:S:P ratio of 12:2:1 should be used to support higher rumen microbial growth increase nutrient supply for ruminant productivity.

References

- Bondi, A.A. 1987. Animal Nutrition. First Edition. John Wiley and Son, Ltd. New York, Chichester, Brisbane, Toronto, Singapore.
- Blümmel, M., H.P.S. Makkar, and K. Becker. 1997^a. *In vitro* gas production: a technique revisited. *J. Anim. Phys. Anim. Nut.* 77:24-34.
- Blümmel, M., H. Steinam, and K. Becker. 1997^b. The relationship between *in vitro* gas production, *in vitro* microbial biomass yield and ¹⁵N incorporation and its implications for the prediction of voluntary feed intake of roughages. *British J. Nut.* 77: 911-921.
- Pathak A. K., 2008. Various factors affecting microbial protein synthesis in the rumen. *Veterinary World*, 1 (6): 186-189.
- Preston. T. R and R. A Leng, 1987. Matching Ruminant Production System with Available Resource in the Tropics and Sub-Tropics. *Penambul Books*, Armidale, NSW. ISBN 0-9588290-12.
- Satter. L.D. and L. L. Slyter, 1974. Effect of Ammonia Concentration on Rumen Microbial Protein Production *in vitro*. *Br. J. Nutr.*, 32, 199.
- Schwab, C.G., P. Huhtanen, C.W. Hunt, and T. Hvelplund, 2005. *Nitrogen requirements of cattle*. In: Pfeffer, E. and A.N. Hristov, Editors. 2005. *Nitrogen and Phosphorus Nutrition of Cattle*. CAB International, Wallingford, UK, pp. 13–70.

Rumen adaptation for urea on feed intake, nutrient digestibility and microbial protein syntheses of swamp buffaloes

Kang^{1,2}, S., M. Wanapat² & K. Phesatcha²

¹ Agricultural Unit, Department of Education, National Institute of Education, Phnom Penh, Cambodia, ² Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

Abstract

Four, Thai – rumen fistulated swamp buffaloes male (*Bubalus bubalis*), about 5 years old with 390±18 kg liveweight, were randomly selected to investigate the rumen adaptation of urea from low to high level on feed intake and nutrient digestibility. All buffaloes were fed with rice straw *ad libitum* and supplemented with concentrate mixture containing 0% urea at 0.5% BW for a period of two weeks. Following adaptation to diet, all buffaloes were shifted to a step-up diet regimen by supplementation of concentrate mixture containing 2% and 4% urea at 0.5% BW for a period of four weeks each. The result shows that feed intake and nutrient digestibility were increased with increasing level of urea supplement. However, within two weeks of urea uptake, buffaloes could adapt well and utilized urea as N source. Increasing urea uptake for a period of two weeks increased microbial protein synthesis in swamp buffaloes especially at 4% urea. Based on this study, we could concluded that swamp buffaloes could adapt well with urea after two weeks uptake period. Moreover, urea supplement could increase feed intake, nutrient digestibility and microbial protein synthesis of swamp buffaloes. This study suggested that buffaloes could utilize urea at 4% within two weeks adaptation period.

Keywords: swamp buffaloes, urea, rice straw, rumen adaptation

Introduction

Providing supplementations with a high concentration of true protein to ruminants fed low-quality roughage stimulates roughage intake, digestion, and performance. Supplementation of natural protein to ruminants consuming low-quality forage improved forage intake, nutrient digestibility and animal performance (Khattab et al., 2013). Non-protein N (NPN) sources are usually less expensive per unit of N than natural protein sources. Moreover, research has suggested that NPN can effectively be used as a source of supplemental N to ruminants consuming low-quality forage. As reported, substituting NPN such as urea has been shown to increase voluntary feed intake (Huntingto & Archibeque, 1999), which is generally attributed to an improvement of nutrients digestibility and an increase passage from the rumen. The utilization of NPN by ruminants is often less efficient than the utilization of natural protein supplements. It has been theorized that synchronization of ruminal ammonia and energy availability will result in improved efficiency of

NPN utilization and animal performance. One strategy for using high degradable carbohydrates is to use in combination with readily available NPN sources such as urea. However, the use of NPN in N supplements can result in management concerns such as supplement palatability and refusal, urea toxicity, and decreased efficiency of N use compared with sources of natural protein. Therefore, this study was conducted to investigate on swamp buffaloes adaptation of urea as NPN in concentrate mixture on feed intake, nutrient digestibility, and microbial protein synthesis fed on rice straw based.

Materials and Methods

Four, Thai – rumen fistulated swamp buffaloes male (*Bubalus bubalis*), about 5 years old with 390±18 kg liveweight, were randomly selected and fed with rice straw *ad libitum* and supplemented with concentrate mixture without urea at 0.5% BW for a period of two weeks. Following adaptation, all buffaloes were shifted to a step-up diet regimen by supplementation of concentrate mixture containing 2% and 4% of urea at 0.5% BW for a period of four weeks each. Totally, there were ten weeks period of study and sample were collected every two weeks of period. Feeds offered and refusal were recorded daily for DMI measurement and samples of concentrate mixture and rice straw were collected daily during the last 5 days of each two weeks period. Fecal and urine samples were collected during the last 5 days of each period at 09.00 or 12.00 hours by rectal sampling and spot sampling, respectively. Acid-insoluble ash (AIA) was analyzed and used to estimate digestibility of nutrients (Van Keulen & Young, 1977). Urine samples were analyzed for allantoin by using HPLC as described by Chen et al. (1993).

Results and Discussion

Feed intake and nutrient digestibility as affected by urea containing in concentrate mixture are shown in Table 1. Rice straw and total feed intake were dramatically increased with the increasing level of urea at 4%. These results agreed with Chen et al. (2010) who indicated that DMI of sheep was improved with the increasing CP level intake and additional U (McGuire et al., 2013). It is probable that a proportion of additional U contributed to improve rumen fermentation and digestibility. Moreover, increasing level of urea from 2 to 4% in concentrate mixture increased nutrient digestibility of swamp buffaloes. Providing supplemental U to ruminants consuming low-quality forage increased intake, digestibility and performance; primarily because of it lack of amino acid N and metabolizable protein (Cappellozza et al., 2013; McGuire et al., 2013). Khattab et al. (2013) reported that there was a linear increase in digestion of DM, OM, CP and non-fibre carbohydrates with increasing U levels in the diets which could be due to the increased rate of rumen microorganisms growth as more available N in form of ammonia from the hydrolysis of U (Boucher et al., 2007). Microbial protein synthesis were increased by urea supplement. Microbial N synthesis increased linearly with increasing urea supplementation, which in reflected in increased PD in the urine (Khattab et al., 2013). The results are similar to Boucher et al. (2007), who reported the optimum ruminal ammonia N concentration required to support maximum synthesis of microbial and maximum efficiency of microbial protein synthesis. The results showed that swamp buffaloes could utilize urea efficiency for a period of two weeks especially when fed low-quality roughage.

Conclusions

Based on this study, urea supplement could increase feed intake, nutrient digestibility, and microbial protein synthesis. Moreover, swamp buffaloes could adapt to urea and utilize efficiency after uptake for a period of two weeks. It is suggested that 4% urea and a period of two weeks uptake, buffaloes could adapt and utilize efficiency when fed with low-quality roughage.

Table 1. Rumen adaptation for urea on feed intake, nutrient digestibility and urinary purine derivatives in swamp buffaloes.

Item	T0	T1	T2	T3	T4	SEM
Rice straw intake						
kg/day	6.6 ^a	6.7 ^a	6.7 ^a	7.0 ^b	7.2 ^b	0.04
g/kg BW ^{0.75}	68.3 ^a	68.4 ^a	68.8 ^a	71.6 ^b	71.9 ^a	0.73
Concentrate intake						
kg/day	1.75	1.75	1.75	1.76	1.76	0.06
g/kg BW ^{0.75}	18.86	18.86	18.88	18.91	18.95	0.45
Total intake						
kg/day	8.4 ^a	8.5 ^a	8.5 ^a	8.8 ^b	9.0 ^b	0.08
g/kg BW ^{0.75}	87.2 ^a	87.3 ^a	87.7 ^a	90.8 ^b	90.8 ^b	0.72
Nutrient digestibility, %						
Dry matter	59.6 ^a	62.6 ^b	61.9 ^b	64.6 ^c	64.6 ^c	0.16
Organic matter	61.4 ^a	63.3 ^b	63.8 ^b	63.9 ^b	64.3 ^b	0.83
Crude protein	45.2 ^a	58.1 ^b	58.1 ^b	63.2 ^c	64.4 ^c	1.25
Neutral detergent fiber	49.2 ^a	53.2 ^a	53.9	56.3 ^b	57.1 ^b	0.94
Acid detergent fiber	45.4	45.9	46.1	46.7	46.4	0.05
Urinary purine derivatives (mmol/d)						
Allantoin excretion	20.0 ^a	22.7 ^b	23.5 ^b	23.4 ^{bc}	25.8 ^c	0.73
Allantoin absorption	96.8 ^a	173.9 ^b	165.2 ^b	187.4 ^c	191.8 ^c	1.47
MNS (g/d)	70. ^a	126.4 ^b	120.1 ^b	136.2 ^c	139.4 ^c	3.26
EMNS (g/kg OMDR)	17.6 ^a	31.6 ^b	30.2 ^b	34.9 ^c	35.0 ^c	2.65

^{a, b, c} Means in the same row with different superscripts differ ($P < 0.05$); T0, Concentrate without urea; T1, Concentrate with urea 2% for a period of two weeks; T2, Concentrate with urea 2% for a period of four weeks; T3, Concentrate with urea 4% for a period of two weeks; T4, Concentrate with urea 4% for a period of four weeks.

Acknowledgements

Authors would like to express their most sincere thanks to Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand for their kind financial and facilities support for the research.

References

- Boucher, S. E., R. S. Ordway, N. L. Whitehouse, F. P. Lundy, P. J. Kononoff and C. G. Schwab, 2007. Effect of incremental urea supplementation of a conventional corn silage-based diet on ruminal ammonia concentration and synthesis of microbial protein. *J. Dairy Sci.* 90: 5619-5633.
- Cappellozza, B. I., D. W. Bohnert, C. S. Schauer, S. J. Falck, E. S. Vanzant, D. L. Harmon and R. F. Cooke, 2013. Daily and alternate day supplementation of urea or soybean meal to ruminants consuming low-quality cool-season forage: II. Effects on ruminal fermentation. *Livest. Sci.* 155: 214-222.
- Chen, S., P. Paengkoun, X. Xia and P. Na-Lumpang, 2010. Effects of dietary protein on ruminal fermentation, nitrogen utilization and crude protein maintenance in growing Thai-indigenous beef cattle fed rice straw as roughage. *J. Anim. Vet. Adv.* 9: 2396-2010.
- Chen, X. B., D. J. Kyle and E. R. Orskov, 1993. Measurement of allantoin in urine and plasma by high performance liquid chromatography with pre-column derivatization. *J. Chromatogr.* 617: 241-247.
- Huntington, G. B. and S. L. Archibeque, 1999. Practical aspects of urea and ammonia metabolism in ruminants. *Proc. of the American Soc. of Anim. Sci.* pp. 1-11.
- Khattab, I. M., A. Z. M. Salem, A. M. Abdel-Wahed and K. Z. Kewan, 2013. Effects of urea supplementation on nutrient digestibility, nitrogen utilisation and rumen fermentation in sheep fed diets containing dates. *Livest. Sci.* 155: 223-229.
- McGuire, D.L., D. W. Bohnert, C. S. Schauer, S. J. Falck and R. F. Cooke, 2013. Daily and alternate day supplementation of urea or soybean meal to ruminants consuming low-quality cool-season forage: I—Effects on efficiency of nitrogen use and nutrient digestion. *Livest. Sci.* 155: 205-213.
- Van Keulen, J. and B. A. Young, 1977. Evaluation of acid insoluble ash as a neutral marker in ruminant digestibility studies. *J. Anim. Sci.* 44: 282-287.

Study on rumen ecology of swamp buffaloes as affected by urea as protein source in concentrate mixture fed on rice straw based

Phesatcha, K.¹, M. Wanapat¹ & Kang, S^{1,2}

¹Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

²Agricultural Unit, Department of Education, National Institute of Education, Phnom Penh, Cambodia

Abstract

This study was investigated on rumen ecology adaptation of urea uptake of swamp buffaloes fed rice straw based. Four, Thai – rumen fistulated swamp buffaloes male (*Bubalus bubalis*), about 5 years old with 390±18 kg liveweight, were randomly selected and fed with rice straw *ad libitum* plus concentrate mixture containing 0% urea at 0.5% BW supplemented for a period of two weeks. Following adaptation to diet, all buffaloes were shifted to a step-up diet regimen by supplementation of concentrate mixture containing 2% and 4% urea at 0.5% BW for a period of four weeks each. The result shows that ruminal pH and temperature were not affected by urea uptake. However, feed intake were increased dramatically. Data from real-time PCR showed that total bacteria and three dominant cellulolytic bacteria (*R. albus*, *F. succinogenes*, *R. flavefaciens*) were increased with urea uptake. However, within four weeks of urea uptake, buffaloes could adapt well and utilized urea as N source and result showed clearly in the buffaloes consumed urea at 4%. Based on this study, we could concluded that swamp buffaloes could adapt well with urea after two weeks uptake period with increasing feed intake without affecting rumen ecology. Moreover, urea supplement at 4% could increase microbial growth in swamp buffaloes at 4 weeks uptake adaptation period.

Keywords: swamp buffaloes, urea, rice straw, rumen ecology

Introduction

Rice straw has been used as the main roughage source, particularly during dry season, for ruminant animal in Thailand. However, feeding rice straw alone does not provide enough nutrients for ruminants due to its low nitrogen content, poor digestibility and low intake. Low ruminal NH₃-N often limits microbial growth and ruminal fermentation (Slyter et al., 1979) in ruminants consuming low-quality forage. Consequently, provision of supplemental ruminally degradable protein (RDP) usually increases microbial CP production and enhances ruminal fermentation, thereby improving performance (Bohnert et al., 2002; Mathis et al., 1999) and reproductive efficiency. However, protein supplementation is an expensive management practice, because of the costs of supplement, labor, and equipment associated with supplement delivery. Urea is widely

used as a dietary supplement for ruminants because it is an inexpensive nitrogenous compound and has long been accepted as a replacement for some of the degradable true protein in diets (Pinos-Rodríguez et al., 2010). Previous research has indicated that providing CP supplements as infrequently to ruminants consuming low-quality forage results in performance and nutrient utilization similar to daily supplementation (Bohnert et al., 2002). However, the use of NPN in N supplements can result in management concerns such as supplement palatability and refusal, urea toxicity, and decreased efficiency of N use compared with sources of natural protein (Chalupa, 1968; Rush et al., 1976). Therefore, this study aimed to investigate on the effect of urea adaptation of swamp buffaloes on rumen ecology fed rice straw based.

Materials and Methods

Four, Thai – rumen fistulated swamp buffaloes male (*Bubalus bubalis*), about 5 years old with 390 ± 18 kg liveweight, were randomly selected and fed with rice straw *ad libitum* and supplemented with concentrate mixture without urea at 0.5% BW for a period of two weeks. Following adaptation, all buffaloes were shifted to a step-up diet regimen by supplementation of concentrate mixture containing 2% and 4% of urea at 0.5% BW for a period of four weeks each. Totally, there were ten weeks period of study and sample were collected every two weeks of period. Feeds offered and refusal were recorded daily for DMI measurement and samples of concentrate mixture and rice straw were collected daily during the last 5 days of each two weeks period. At last day of each two week period, rumen fluid of each buffalo were collected at 0, 2, 4, and 6 hour post feeding. Ruminal pH and temperature were measured immediately. Fluid samples were stored at -20 °C and analyzed for DNA extraction (Yu and Morrison, 2004).

Results and Discussion

Total feed intake were dramatically increased with the increasing level of urea at 4%. These results agreed with Chen et al. (2010) who indicated that DMI of sheep was improved with the increasing CP level intake and additional U (McGuire et al. 2013). The data on rumen ecology and microorganism affected by urea source are presented in Table 1. There were no differences on ruminal pH and temperature by urea uptake. Similarly, mean of pH was not affected by U level according to Lizarazo et al. (2014). However, U supplementation increased ruminal pH post feeding according to Van Soest (1994); since the ruminal pH is partly regulated by $\text{NH}_3\text{-N}$ concentration and the variation in pH may be explained by U entering the rumen and being hydrolyzed by microbial ureases into CO_2 and ammonia. However, microorganism (bacteria, fungi, protozoa) by direct count and total bacteria and three dominant cellulolytic bacteria (*R. albus*, *F. succinogenes*, *R. flavefaciens*) by real-time PCR were increased by supplemented with urea supplement in concentrate. It was reported that fungal zoospores, protozoa and total bacteria direct count were significantly different and had higher number in the animals fed higher protein level diet.

Table 1. Rumen adaptation for urea on ruminal pH, temperature and microbe using real-time PCR in swamp buffaloes.

Item	T0	T1	T2	T3	T4	SEM
Total intake						
kg/day	8.4 ^a	8.5 ^a	8.5 ^a	8.8 ^b	9.0 ^b	0.08
g/kg BW ^{0.75}	87.2 ^a	87.3 ^a	87.7 ^a	90.8 ^b	90.8 ^b	0.72
Rumen ecology						
Ruminal pH	6.9	6.8	6.8	6.7	6.9	0.12
Ruminal temperature	38.5	38.8	38.9	39.1	39.3	0.18
Direct count (cell/ml)						
Bacteria (x 10 ⁹)	5.2 ^a	5.8 ^b	6.3 ^b	7.7 ^c	8.6 ^d	0.43
Protozoa (x 10 ⁵)	4.3	4.6	4.2	4.7	4.5	0.09
Fungal zoospore (x 10 ⁴)	2.7 ^a	3.0 ^a	3.4 ^a	4.9 ^b	5.3 ^b	0.92
Microbe by Real-time PCR						
Total bacteria, x 10 ⁸	1.1 ^a	1.7 ^a	1.5 ^a	6.6 ^b	7.4 ^c	0.36
<i>R.albus</i> , x 10 ⁷	3.6	3.4	3.2	3.3	3.7	0.02
<i>F. Succinogenes</i> , x 10 ⁷	2.3 ^a	2.5 ^a	2.7 ^a	5.4 ^b	6.3 ^c	0.16
<i>R. flavefaciens</i> , x 10 ⁶	5.6 ^a	5.7 ^a	5.4 ^a	6.8 ^b	7.5 ^c	0.32
Methanogens, x 10 ⁴	2.2	2.3	2.3	2.4	2.6	0.02

^{a, b, c} Means in the same row with different superscripts differ ($P < 0.05$); T0, Concentrate without urea; T1, Concentrate with urea 2% for a period of two weeks; T2, Concentrate with urea 2% for a period of four weeks; T3, Concentrate with urea 4% for a period of two weeks; T4, Concentrate with urea 4% for a period of four weeks.

Conclusions

Based on this study, we could concluded that swamp buffaloes could adapt well with urea after two weeks uptake period with increasing feed intake without affecting rumen ecology. Moreover, urea supplement at 4% could increase microbial growth in swamp buffaloes at 4 weeks uptake adaptation period.

Acknowledgements

Authors would like to express their most sincere thanks to Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand for their kind financial and facilities support for the research.

References

- Bohnert, D. W., C. S. Schauer, S. J. Falck and T. DelCurto, 2002. Influence of rumen protein degradability and supplementation frequency on steers consuming low-quality forage: II. Ruminant fermentation characteristics. *J. Anim. Sci.* 80: 2978-2988.
- Chalupa, W., 1968. Problems in feeding urea to ruminants. *J. Anim. Sci.* 27: 207-219.

- Mathis, C. P., R. C. Cochran, G. L. Stokka, J. S. Heldt, B. C. Woods and K. C. Olson, 1999. Impacts of increasing amounts of supplemental soybean meal on intake and digestion by beef steers and performance by beef cows consuming low-quality tallgrass-prairie forage. *J. Anim. Sci.* 77: 3156-3162.
- Pinos-Rodríguez, J. M., L. Y. Peña, S. S. González-Muñoz, R. Bárcena and A. Salem, 2010. Effects of a slow-release coated urea product on growth performance and ruminal fermentation in beef steers. *Italian J. Anim. Sci.* 9: e4.
- Rush, I. G., R. R. Johnson and R. Totusek, 1976. Evaluation of beef cattle range supplements containing urea and biuret. *J. Anim. Sci.* 42: 1297-1308.
- Slyter, L. L., L. D. Satter and D. A. Dinius, 1979. Effect of ruminal ammonia concentration on nitrogen utilisation by steers. *J. Anim. Sci.* 48: 906-912.
- Yu, Z. and M. Morrison, 2004. Improved extraction of PCR-quality community DNA from digesta and fecal samples. *Biotechniques*. 36: 808-812.
- Chen, S., P. Paengkoun, X. Xia and P. Na-Lumpang, 2010. Effects of dietary protein on ruminal fermentation, nitrogen utilization and crude protein maintenance in growing Thai-indigenous beef cattle fed rice straw as roughage. *J. Anim. Vet. Adv.* 9: 2396-2010.
- McGuire, D.L., D. W. Bohnert, C. S. Schauer, S. J. Falck and R. F. Cooke, 2013. Daily and alternate day supplementation of urea or soybean meal to ruminants consuming low-quality cool-season forage: I—Effects on efficiency of nitrogen use and nutrient digestion. *Livest. Sci.* 155: 205-213.
- Lizarazo, A. C., G. D. Mendoza, J. Kú, L. M. Melgoza and M. Crosby, 2014. Effects of slow-release urea and molasses on ruminal metabolism of lambs fed with low-quality tropical forage. *Small Rum. Res.* 116: 28-31.
- Van Soest, P. J., 1994. *Nutritional ecology of the ruminant* (2nd Ed.) Cornell University Press, Ithaca, NY, p. 476.

Effect of *Flemingia macrophylla* (FLM) as a protein source on rumen fermentation and microbial population in dairy steers

Pesatcha¹, B., M. Wanapat¹, K. Phesatcha¹ & T. Ampapon¹

¹Tropical Feed Resources Research and Development Center (TROFREC) Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

Abstract

The objective of this research was to evaluate the effect of *Flemingia macrophylla* (FLM) as a protein source on rumen fermentation and microbial population in dairy steers. Four rumen fistulated dairy steers were randomly assigned to receive dietary treatments according to a 4x4 Latin square design. Four treatments were as follows; T1=control (no supplementation); T2=supplementation of cassava hay (CH) at 150 g/kg/d; T3=supplementation of Flemingia (FLM) at 150 g/kg/d and T4=supplementation of CH at 75 g/kg/d and FLM 75 g/kg/d)CHFLM(of total dry matter intake (DMI).The FLM contained 25.8% crude protein (CP) and 5.2% condensed tannins (CT). Rice straw, water and mineral salt block were offered *ad libitum*. The experiment was conducted for 4 periods, and each period lasted for 21 days, while the last 7 days was for sample collection. The study revealed that CH, FLM and CHFLM supplementation resulted in volatile fatty acid concentrations especially those of propionic acid were increased)P<0.05(by supplementation as compared to control group. Similarly, methane emission was reduced (P<0.05) in the CH, FLM and CHFLM as compared to control group. Based on this study, it could be concluded that FLM could be used as a protein source, while the combination of CHFLM could enhance the rumen fermentation in dairy steers

Key words: *Flemingia macrophylla*, cassava hay, rice straw, rumen fermentation, dairy steers

Introduction

Ruminants raised in the tropics largely depend on seasonal feed resources which are relatively low in quality in terms of low CP but high in crude fiber (CF); hence, the manipulation of rumen efficiency through the uses of local feed resources would be an advantage (Wanapat, 2000). *Flemingia macrophylla* (Willd.) Merrill is a drought-tolerant, perennial multipurpose shrub legume especially suited to low-input smallholder production systems in the subhumid and humid tropics. Tropical tree legumes are rich in most minerals and generally have a range of digestibilities similar to the tropical grasses (Leng et al., 1992). *Flemingia* foliage has high crude protein (CP) values ranging from 140 to 180 g/kg DM (Binh et al., 1998). The total tannin content of *Flemingia* foliage (consisting of leaves and 35–40 cm of the twigs) was 30.8 g/kg DM (Mui et al., 2002). Cassava or tapioca (*Manihotesculenta*, Crantz) is an annual tuber crop grown widely in tropical and sub-tropical areas. It thrives in sandy loam soil with low organic matter and can tolerate low rainfall and high temperatures. It is therefore a cash

crop cultivated by smallholder farmers within the existing farming systems in many countries. Cassava can also be cultivated in combined forage/root systems with two or more harvests of the foliage prior to letting the root develop to maturity (Wanapat et al., 1997).

Materials and Methods

Animals, diets, experimental design, and animal management : Four, rumen-fistulated dairy steers (180 ± 10 kg BW) were randomly assigned to receive four dietary treatments according to a 4 x 4 Latin square design. All animals were received concentrate diet at 0.5% of body weight and rice straw was fed ad libitum as roughage source. The treatments were: T1=unsupplementaion (Control), T2 = supplementation of cassava hay (CH)at 150 g/kg/d, T3 = supplementation of *flemingia*(FLM) at 150 g/kg/d, T4 = supplementation with CH and FLM at 150 g/kg/d, respectively. The experiment was conducted for four periods; each experimental period lasted for 21 days. First 14 days were used as adaptation period in which, all animals were fed with their respective diets followed by a 7-day collection period. At last day of each two week period, the first part rumen fluid of each steers were collected at 0, 2, 4, and 6 hour post feeding. The second part was immediately fixed with 10% formalin solution (1:9 v/v, rumen fluid: 10% formalin) to measure microbial populations by total direct counts of bacteria, protozoa and fungal zoospores (Galyean, 1989).

Results and Discussion

The present study has shown that propionic acid increased while acetic acid, acetic to propionic acid ratio and methane production decreased in all supplemented groups. Moreover, the TVFA concentration in ruminal fluid was not influenced by the level of protein in the diet of sheep (Merchen et al., 1986; Carro et al., 2000). Table 2 shown rumen bacteria was maximized (P<0.05) in the supplemented group compared to the control group. The general mode of action of tannins and saponins on microorganisms is their interaction with the sterol moiety, which is present in the membrane of protozoa (Ando et al., 2003).

Table 1. Effect of *Flemingia macrophylla*(FLM) as a protein source on volatile fatty acids (VFA) in dairy steers.

Items	Control	CH	FLM	CHFLM	SEM	P-value
Total VFA, mmol/l	121.4	124.3	120.5	126.7	2.35	0.49
mol/100mol total VFA						
Acetic acid) C ₂ (68.7 ^a	65.1 ^b	64.8 ^{ab}	62.2 ^b	1.35	0.04
Propionic acid)C ₃ (20.1 ^a	25.4 ^b	24.3 ^b	27.6 ^b	1.03	0.03
Butyric acid)C ₄ (8.7	8.1	8.9	8.4	0.79	0.87
C:2C ₃	3.4 ^a	2.6 ^b	2.7 ^b	2.3 ^b	0.12	0.05
CH ₄ production ^A	22.0 ^a	19.1 ^b	18.8 ^b	17.0 ^b	0.29	0.02

^{a, b, c}, Means in the same row with different superscripts differed (P<0.05)

CH, Cassava hay; FLM, *Flemingia*; CHFLM, combination of CH and FLM; SEM, standard error of the means

^A Calculated according to Moss et al. (2000) CH₄ production = 0.45 (acetate)-0.275 (propionate)+0.4 (butyrate).

Table 2. Effect of *Flemingia macrophylla* (FLM) as a protein source on rumen microbial population in dairy steers.

Items	Control	CH	FLM	CHFLM	SEM	P-value
Ruminal microbes × cell/m						
Bacteria, × 10 ⁹	2.7 ^a	3.3 ^b	3.7 ^b	4.2 ^c	0.64	0.02
Protozoa, × 10 ⁵	8.4	8.1	8.7	8.3	0.24	0.76
Fungi, × 10 ⁵	2.3	2.5	2.5	2.6	0.12	0.83

^{a, b, c}. Means in the same row with different superscripts differed (P<0.05)

CH, Cassava hay; FLM, *Flemingia*; CHFLM, combination of CH and FLM; SEM, standard error of the means

Conclusions

Based on this study, it could be concluded that FLM could be used as a protein source, while the combination of CHFLM and additionally enhanced rumen fermentation and microorganisms. The present study suggests that FLM or cassava hay supplementation could improve rumen fermentation and increased bacterial population while mitigating of dairy steers.

Acknowledgements

Authors would like to express their most sincere thanks to Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand for their kind financial and facilities support for the research.

References

- Ando, A., T. Nishida, M. Ishida, K. Hosoda and E. Bayaru (2003). Effect of peppermint feeding on the digestibility, ruminal fermentation and protozoa. *Livest. Prod. Sci.* 82: 245-248.
- Binh, D.B., Tien, N.P., Mui, N.T., 1998. Study on biomass yield and quality of *Flemingia macrophylla* and on soil fertility. In: Proceedings of Workshop of Animal Nutrition Science. Ministry of Agriculture, Vietnam, p. 137.
- Galyean, M., 1989. Laboratory Procedure in Animal Nutrition Research. Department of Animal and Range Sciences, New Mexico State University, Las Cruces, NM, USA.
- Leng, R.A., Choo, B.S., Arreaza, C., 1992. Practical technologies to optimise feed utilization by ruminants. Legume trees and other fodder trees as protein sources for livestock. *FAO Anim. Prod. Health Paper* 102, 75–93.
- Merchen, N.R., Firkins, J.L., Berger, L.L., 1986. Effect of intake and forage level on ruminal turnover rates, bacterial protein synthesis and duodenal amino acid flows in sheep. *J. Anim. Sci.* 62, 216–225.
- Mui, N.T., Ledin, I., Udén, P., Binh, D.V., 2002. The foliage of *Flemingia (Flemingia macrophylla)* or Jackfruit (*Artocarpus heterophyllus*) as a substitute for a rice bran-soya bean concentrate in the diet of lactating goats. *Asian-Aust. J. Anim. Sci.* 15, 1–10.
- Wanapat, M., Pimpa, O., Petlum, A., Boontao, U., 1997. Cassava hay: A new strategic feed for ruminants during the dry season. *Livestock Research for Rural Development* (9)2: [Http://www.cipav.org.co/lrrd/lrrd9/2/metha92.htm](http://www.cipav.org.co/lrrd/lrrd9/2/metha92.htm).
- Wanapat, M., 2000. Rumen manipulation to increase the efficient use of local feed resources and productivity of ruminants in the tropics. *Asian-Aus. J. Anim. Sci.*, 13 (Suppl.):59-67.

Effect of rice straw treatment on feed intake and nutrient digestibility in swamp buffaloes

Viennasay, B. & Wanapat, M.

Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

Abstract

The objective of this experiment was to evaluate the effect of rice straw treatment (RST) on feed intake and nutrient digestibility in swamp buffaloes. Four rumen fistulated male swamp buffaloes with 330 ± 20 kg live weight were randomly assigned according to a 4×4 Latin square design to receive four dietary treatments and were as follows; T1=untreated rice straw, T2= 1%urea treated rice straw, T3= 1%urea+1%cassava chip powder treated rice straw, and T4= 1%urea +1%cassava chip powder +0.1%yeast treated rice straw. All animals were fed concentrate mixtures at 0.2% of body weight and rice straw was fed *ad libitum* for 21 days with the first 14 days were for feed adaptation and voluntary feed intake measurement, while the last 7 days were for samples collection. The results revealed that nutritive values of rice straw were increased by urea and yeast treatment. Moreover, feed intake and nutrient digestibility of swamp buffaloes were increased in buffaloes consuming treated rice straw especially with 1%urea + 1% cassava chip powder +0.1%yeast treatment (T4). Based on this study, it could be concluded that rice straw treatment with urea plus cassava chip powder and yeast could improve nutritive value and enhanced feed intake and nutrient digestibility of swamp buffaloes.

Keywords: rice straw, intake, digestibility, swamp buffaloes

Introduction

Rice straw is the main crop-residue which farmers usually store for use as ruminant feed in tropical areas, especially in Asia. However, rice straw is low in nutritive value with low level of protein (2–5%DM), high fiber and lignin content (NDF>50%), low DM digestibility (<65%) thus resulting in low voluntary feed intake (1.5–2.0%) (Wanapat et al., 1985). Previous work discussed strategies to improve the utilization of those feeds, suggesting to provide supplements to correct the nutrient imbalances for rumen bacteria (Ørskov, 1994). It was found that using urea treatment could increase nutritive value of rice straw. Wanapat et al.(1985)reported that urea-treated rice straw could increase overall intake, digestibility, thus resulted in enhancing the performance of ruminants as compared to untreated rice straw. However, the cost of urea treatment was remarkably expensive which resulted on higher cost of production. Therefore, the objective of this experiment was to evaluate the effect of rice straw treatment (RST) on feed intake and nutrient digestibility in swamp buffaloes.

Materials and methods

Four rumen fistulated male swamp buffaloes with 330 ± 20 kg of body weight (BW) were randomly assigned to receive four dietary treated rice straw according to 4×4 Latin square design. The treatments were as follows, T1= untreated rice straw, T2= 1%urea treated rice straw, T3= 1%urea+1%cassava chip powder treated rice straw and T4= 1%urea +1%cassava chip powder +0.1%yeast treated rice straw. All buffaloes were fed rice straw *ad libitum* while additional concentrate was fed at 0.2%/kg BW. Refusals of rice straw were weighed daily prior to the morning feeding to determine daily DM intake. Feces, urine, rice straw and concentrate were collected and sampled by total collection and were analyzed chemical composition using standard method. All data were statistically analyzed using the GLM procedure of Statistical Analysis System (SAS, 1996). Differences between treatment means were determined by Duncan's New Multiple Range Test (Steel & Torrie, 1980).

Results and Discussion

Chemical composition of dietary experiment is presented in Table 1. Crude protein content of rice straw was increased by urea treatment especially by 1%urea+1%cassava ship powder+ yeast treated rice straw (T4). It was also reported by Wanapat et al. (2009) who conducted by using 2.2 + 2.2% urea-calcium hydroxide treated rice straw. As having been reported by Schiere&Ibrahim (1989), rice straw can be treated with urea, which releases ammonia after dissolving in water. Moreover, feed intake of buffaloes in the present study were significant different and the highest was in 1%urea+1%cassava chip powder treated rice straw (T3) as shown in Table 2. However, the highest nutrient digestibility of DM, OM and CP were found in 1%urea +1%cassava ship powder +0.1% yeast treated rice straw (T4) while NDF and ADF digestibility were stable. Wanapat (1992) reported that urea treated rice straw could increase digestibility of nutrient. Ammonium hydroxide formed in treated straw produces a swelling of the hemicelluloses-lignin complex in rice straw (Mapato et al., 2010). Resulting in an increased surface area available for attack by rumen microorganisms and thus increasing the rate of breakdown and the rate of passage of treated straw through the digestive tract (Ha et al., 2001). These effects may explain the action of urea treatment in improving rumen microbial degradation of rice straw by making the cellulose and hemicellulose more accessible for the rumen microbes (Shen et al., 1999).

Table 1. Chemical composition of experimental diet.

Item	Concentrate	T1	T2	T3	T4
Dry Mather	87.35	42.21	43.67	46.79	42.33
Organic Mather	87.35	38.65	38.74	42.32	37.08
Ash	8.7	3.6	4.9	4.5	5.2
Crude Protein	9.09	2.21	3.20	3.03	3.72
Neutral detergent fiber	15.24	47.73	50.46	54.49	56.77
Acid detergent fiber	70.61	71.74	71.51	80.47	78.36

Table 2. Effect of urea and yeast treated rice straw on voluntary feed intake and nutrient digestibility in swamp buffaloes.

Item	T1	T2	T3	T4	SEM
Roughage intake					
kg/day	4.8 ^a	5.2 ^a	5.9 ^b	5.1 ^b	0.14
g/kgBW ^{0.75}	67.8 ^a	64.1 ^a	75.2 ^b	63.8 ^a	3.22
Concentrate intake					
kg/day	0.6	0.7	0.68	0.68	0.03
g/kg BW ^{0.75}	8.32	8.66	8.58	8.6	0.02
Total intake					
kg/day	5.49 ^b	5.9 ^b	6.61 ^a	5.75 ^{ab}	0.24
g/kgBW ^{0.75}	1.83	1.67	1.95	1.68	0.08
Apparent digestibility(%)					
Dry matter	55.2 ^a	61.1 ^b	64.3 ^b	65.6 ^b	0.5
Organic matter	57.1 ^a	62.2 ^b	66.1 ^b	67.4 ^c	1.1
Crude protein	36.7 ^a	37.2 ^a	41.6 ^b	48.4 ^c	3
Neutral detergent fiber	57.4	61.8	57.9	56.3	4.2
Acid detergent fiber	51.4	55.1	52.7	48.7	5.4

Conclusions

Based on this study, it could be concluded that rice straw quality could be improved by treated with urea and yeast. More, Treated straw with urea and yeast could increase feed intake and nutrient digestibility of swamp buffaloes.

Acknowledgments

The authors would like to express their most sincere thanks to all who have assisted and supported the research in this study, particularly the Tropical Feed Resources Research and Development Center (TROFEC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University (KKU), Thailand for their kind facilitation and permission to use the research facilities

References

- Ha, J. K., S. S. Lee, S. W. Kim, In K. Han, K. Ushida and K. J. Cheng, 2001. Degradation of rice straw by rumen fungi and cellulolytic bacteria through mono-, co- or sequential- cultures. *Asian-Aust. J. Anim. Sci.* 14:797-802.
- Mapato, C., M. Wanapat and A. Cherdthong, 2010. Effects of urea treatment of straw and dietary level of vegetable oil on lactating dairy cows. *Trop. Anim. Health Prod.* 42:1635-1642.
- Ørskov, E.R., 1994. Recent advances in understanding of microbial transformation in ruminants. *Livest. Prod. Sci.* 39: 53–60.
- Shen, H. S., F. Sundstøl, E. R. Eng and L. O. Eik, 1999. Studies on untreated and urea-treated rice straw from three cultivation seasons: 3. Histological investigations by light and scanning electron microscopy. *Anim. Feed Sci. Technol.* 80:151-159.

- Steel, R.G. D. and J. H. Torrie, 1980. Principles and Procedures of Statistics. McGraw Hill Book Co, NY.
- Schiere, J. B. and M. N. M. Ibrahim, 1989. Feeding of urea-ammonia treated rice straw: A compilation of miscellaneous reports produced by the Straw Utilization Project (Sri Lanka). Pudoc, Wageningen.
- SAS, 1996. SAS/STAT User's Guide: Statistics, Version 6.12. Edition. SAS Inc, Cary, NC, USA.
- Wanapat, M., C. Wachirapakorn, S. Chanthai and K. Sommart, 1992. Utilization of cassava leaf (*Manihot esculenta*, Crantz) in concentrate mixtures for swamp buffaloes in Thailand. In: Proc. Feeding Strategies for Improving Ruminant Productivity in Areas of Fluctuating Nutrient Supply Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, Vienna, Austria, March 30-April 3, pp. 33-38
- Wanapat, M., Sundstol, F., Garmo, T.H., 1985. A comparison of alkali treatment methods to improve the nutritive value of straw. I. Digestibility and metabolizability. Anim. Feed Sci. Technol. 12:295-309.
- Wanapat, M., S. Polyorach, K. Boonnop, C. Mapato, A. Cherdthong, 2009. Effects of treating rice straw with urea or urea and calcium hydroxide upon intake, digestibility, rumen fermentation and milk yield of dairy cows. Livest. Sci. 125:238-243.

Efficacy of Endogenous Emulsifier in Broilers Diets on Growth Performance

Mongkoltiwattana, K., W.Jaikan, S. Wongtangtintan & J. Khajarern

*Department of Animal Science, Faculty of Agriculture Khon Kaen University,
Khon Kaen 40002, Thailand*

Abstract

The effects of endogenous emulsifier bile acid on performance of broilers were assessed in evaporating house system. The bile acid was added 0.04% to the 3 levels deduction (0.70, 1.00 and 1.30%) rice bran oil from control diet with 3 periods of feeding, starter (1-21d), grower (22-35d) and finisher (36-42d) diet was formulated to contain ME 3,121 kcal/kg and 22.52 %CP, ME 3,175 kcal/kg and 20.7 %CP and ME 3,242 kcal/kg and 18.01 %CP respectively. For the overall period (1-42d of age) of testing, birds fed with 3 graded levels deduction rice bran oil from control diet showed linear ($P < 0.05$) increasing final body weight (FBW) and body weight gain (BWG) but showed linear ($P < 0.05$) reducing feed cost/kg BWG (FCG) leading to linear ($P < 0.05$) increasing salable bird returns (SBR) and net profits return (NPR) and showed linear increasing return of investment on the 3 graded levels deduction of rice bran oil from the control diet. Feed efficiency ratio (FCR) and productive index (PI) also showed the same trends as FBW and BWG but not significant ($P > 0.05$). Addition of 0.04% bile acid in 1.30% deduction rice bran oil from control diet showed the highest in broiler performance and showed the highest return of investment (ROI).

Keywords: broilers, emulsifier bile acid, performance, return of investment

**Corresponding author: jowaman@kku.ac.th*

Introduction

The physiological limitations of the digestive system of broilers may be overcome using endogenous and/or exogenous strategies to maximize feed digestion and absorption. The addition of synthetic emulsifier to broiler diets is a recent practice as compared to other dietary supplements. The mode of action of emulsifiers is to increase the active surface of fats, allowing the action of lipase, which hydrolyze triglyceride molecules into fatty acids and monoglycerides and favor the formation of micelles consisting of lipolysis products. This is an essential step for lipid absorption, as it creates a diffusion gradient that increases absorption (Guerreiro et al., 2011). The inability of the young bird to utilize fats has been attributed to poor emulsification rather than deficiencies in lipase activity and this has led to considerable interest in the potential of exogenous bile salt preparations as a means of improving the utilization of fats in young birds (Ashraf, 2007).

Materials and Methods

Birds and Management

A total of 160 day-old mixed-sex of commercial broiler strain (Cobb 500) was randomly assigned into four treatments with 2 replications of 20 chicks per replicate. Chicks were allocated on the basis of weight so that overall weight and weight range were similar for each replicate group. Chicks were housed in fresh rice hull litter floored pens with two hanging feeders and one bell drinker located in a test facility with evaporation house at Khon-Kaen University Poultry Farm with permanent artificial illumination was applied. Chicks were vaccinated for Merek's disease, Newcastle disease and Infections bronchitis at the hatchery and at 12 d against infections bursa disease. All birds received common mash form feeds from placement 1 to 21d, 22 to 35d and 36 to 42d respectively. Feed and water were provide *adlibitum*.

Diets and parameters studied

Control starter (1-21d), grower (22-35d), and finisher (36-42d) diets was formulated to contain ME 3,121 kcal/kg and 22.52 %CP, ME 3,175 kcal/kg and 20.07%CP and ME 3,242 kcal/kg and 18.01 %CP respectively. Subsequently three other experimentdiets were formulated by deduction 0.7,1.0 and 1.3% rice bran oil and adding 0.04% bile acid to the control diet at the expense of corn Salinomysin (60 ppm) was supplemented in both starter and grower diets to control coccidiosis. Pen body weight and survival rate were recorded at 21, 35 and 42 days of age and total feed consumption during those interval was also determined. Adjustments in feed conversion were made for birds that died during the study.

Statistical Analysis

All data were subjected to analysis of variance using the GLM procedure of SAS software (SAS, 2004). Different among mean were detected by Duncan's multiple range tests. Significance was considered at $P < 0.05$ levels.

Results and Discussion

The performance of the broilers fed with bile acid for the overall period of testing (1-42d) was presented in Table 1. Broilers fed with 0.04% bile acid on the 3 levels deduction of rice bran oil from control diet showed linear ($P < 0.05$) increasing in final body weight (FBW) and body weight gain (BWG) but showed linear decreasing ($P < 0.05$) feed cost/kg BWG (FCG) leading to linear ($P < 0.05$) increasing in salable bird returns (SBR) and net profits return (NPR) and showed linear increasing return of investment on the 3 levels deduction of rice bran oil from control diet. Feeding 0.04% bile acid in the 3 levels reduction of rice bran oil from control diet also showed linear but not significant ($P > 0.05$) on feed conversion ratio (FCR) and productive index (PI) improvement on the 3 levels deduction of rice bran oil from control diet. Addition of 0.04% bile acid in 1.30% deduction rice bran oil showed the highest return of investment (ROI) and also showed improvement and enhancing digestion of nutrients especially fat and there by productive performance in broilers up to 42d of age. Aguilar et al., (2013) reported broiler fed with an

exogenous emulsifier increased the BW and decreased FCR with significant differences ($P > 0.05$) but feed intake were not affected ($P > 0.05$).

Table 1. Effect of bile acid in the diets on broilers performance for overall 42 days of testing.

Rice bran oil % reduction	Bile acid %	Survival rate %	Final weight)g(BWG)g(FI (g)	FCR g : g	PI ¹	FCG1 ² Baht/kgBW
Control	-	100.00	2,446.50 ^b	2,402.85 ^b	4,213.20	1.755	326.59	28.38 ^a
0.70	0.04	100.00	2,576.52 ^{ab}	2,532.72 ^{ab}	4,248.50	1.678	359.51	26.98 ^{ab}
1.00	0.04	97.50	2,636.22 ^a	2,592.42 ^a	4,260.50	1.643	366.25	26.20 ^b
1.30	0.04	95.50	2,640.37 ^a	2,596.67 ^a	4,256.00	1.639	358.36	26.05 ^b
<i>Pooled SEM</i>		1.77	62.49	62.60	145.16	0.044	13.19	0.73
-----Probability -----								

Contrast								
<i>L</i>		NS	*	*	NS	NS	NS	*
<i>Q</i>		NS	NS	NS	NS	NS	NS	NS
<i>C</i>		NS	NS	NS	NS	NS	NS	NS

^{a,b,c} Mean within column with no common superscript differ significant) $P \leq 0.05$ (

L = Linear Q = Quadratic and C = Cubic effect for bile acid

NS: not significant at) $P \geq 0.05$ (; *: significant) $P \leq 0.05$ (; **: significant) $P \leq 0.01$ (

¹ Productive index(PI) = $\frac{\text{BWG (kg)} \times 100 \times \text{Survival}}{\text{Age} \times \text{FCR}}$

² Feed cost per gain (FCG1) = $\frac{\text{FI} \times \text{feed cost}}{\text{BWG}}$

Table 2. Effect of bile acid in the diets on economic benefits return for overall 42 d of testing.

Rice bran oil % reduction	Bile acid %	FCG2¹ Baht/bird	SBR² Baht/bird	NPR1³ Baht/bird	ROI1⁴ Baht/bird	ROI2⁵ Baht/bird
Control	-	68.1	88.07 ^b	19.91 ^b	-	-
0.70	0.04	68.31	92.76 ^{ab}	24.45 ^{ab}	4.54	1.79
1.00	0.04	67.93	94.90 ^a	26.97 ^a	7.06	2.72
1.30	0.04	67.64	95.05 ^a	27.41 ^a	7.50	2.89
<i>Pooled SEM</i>		2.38	2.25	1.82	-	-
		----- Probability -----				
Contrast						
<i>L</i>		NS	*	*	-	-
<i>Q</i>		NS	NS	NS	-	-
<i>C</i>		NS	NS	NS	-	-

^{a,b,c} Mean within column with no common superscript differ significant)P≤0.05(

L = Linear Q = Quadratic and C = Cubic effect for bile acid

NS: not significant at)P≥0.05(; *: significant)P≤0.05(; **: significant)P≤0.01(

¹Feed cost per gain(FCG2) = FCG1 x BWG (kg)

²Salable bird return (SBR)= Price of live chicken (36 Baht) x BW

³Net profits return per bird (NPR1) = SBR-FCG2

⁴Return of investment by comparing with and the control group (ROI1) = NPR (Bile acid) - NPR (control)

⁵ROI2 = ROI1/BWG(kg)

Conclusion

The result from this experiment clearly demonstrated that bile acid showed a beneficial effects on performance, enhancing nutrient especially fat digestibility and decreasing antinutritional factors in plant protein resulting in an overall improvement of growth performance, feed efficiency, P and FCG comparable with the control groups with higher economic benefits return and increasing in ROI when comparing with the unsupplementation control group.

References

- Aguilar, Y. M., J. C. Becerra, R. R. Bertot, J. C. Peláez, G. Liu C. B. Hurtado, 2013. Growth performance, carcass traits and lipid profile of broiler chicks fed with an exogenous emulsifier and increasing levels of energy provided by palm oil. *J. Food, Agriculture and Environment (JFAE)*:ISSN: 1459-0263. 11:629-633
- Ashraf, M., 2007. Use of Emulsifiers in High Fat Level Diets of Broilers. Doctoral thesis, Department of Animal Production, Faculty of Agriculture, Al Azhar University, Cairo.
- Guerreiro, N., A. C. Pezzato, J. R. Sartori, C. Mori, V. C. Cruz, V. B. Fascina, D. F. Pinheiro, L. A. Madeira and J. C. Gonçalves, 2011. Emulsifier in broiler diets containing different fat sources. *Rev. Bras. Cienc. Avic.*13: 2.
- SAS-Statistical Analysis System. 2004. SAS User's Guide: Statistics. SAS Inst. Inc., Cary, NC; Version 9.1 for Window.

Effects of fattening length and energy levels on meat characteristics of Iranian native lamb

Karami M.

Assistance professor of meat science, Department of Animal Science, Agriculture and Natural Resources research Center, Shahrekord, 415, Iran. Karami_morteza@yahoo.com

Abstract

This study was investigating different levels of metabolisable energy (DME) (2.3 and 2.5) and fattening period (FP) (90 and 120 days) on carcass composition of Chalishtori male lambs. Rations were isonitrogenous (14% CP/DM) and used of completely random designs with factorial method (2*2). Finally 32 lambs randomly slaughtered and data analyzed by SAS. Interaction between (FP) (120 days) in (DME) for final weight, metabolic weight and carcass weight had significant ($p < 0.05$). The mean of daily weight gain was 162.37g/d, interaction between (FP) and (DME) did not influence on daily weight gain and feed intake. The mean of dressing percentage was 53.7% and interaction between (FP) (120 days) in (DME) for dressing percentage was significant ($p < 0.05$). Interaction between FP in DME for surface of loin area and back fat thickness were significant ($p < 0.05$). The mean of total carcass meat, total bone, and total subcutaneous fat and fat tail percent were 46.9, 11.7, 15.9 and 22.9 percent respectively and interaction between FP and DME on carcass compositions. However, interaction was between FP and DME influence on some traits and recommended FP 90 days with 2.3 ME, because higher daily weight gain, loser daily feed intake, better of feed conversion ratio and other wise loin area surface and carcass composition were not significant.

Keywords: fattening periods, energy, fat deposit, carcass

Introduction

Locally available breeds of livestock are important economic resources. The Chalishtory fat-tail sheep is common native breed in the south of western part of Iran and sheep are important meat producing animal in Iran. Growth in animal is defined by an increase in body cells and growth and differentiation of body cells (Bathaei & Loroy, 1996). Growth-rate and body size along with changes in body composition are of great economic importance for efficient production of meat animals. A researcher has reported that fast growing lean in animal breeds are more growing fatter breeds (Berg & Walters, 1983). Growth rate of lambs, particularly during the early stages of growth, is strongly influenced by breed (genotype), milk yield of the ewe, the environment under which the animals are maintained including levels of metabolisable energy and length of fattening periods (Al-Jassim, et al., 1996). Level of nutrition is known to influence body or carcass composition significant (Bulter-Hogg, 1984). Supporting such theory, (Aziz et al., 1992) has observed that the body fat of sheep serves as an immediate source of energy during under nutrition.

In a study reported that nutrition influences carcass yield and quality, fat deposition and composition while breed effect was observed to be greatest in influencing carcass conformation as well as carcass composition (Canton, et al., 1992). Body weight is the main determinant of body composition of animals of the same breed and sex group regardless of age or nutritional level (Turgeon et al., 1980). Therefore, the goal of this study was effects of interaction between levels of energy and fattening periods on growth and carcass composition in Chalishtory male lambs one of the Iranian native lambs.

Materials and Methods

Sixty-Four Chalishtory male lambs (initial BW 31.56 kg) were assigned to a completely randomized design with two factors were length of fattening periods (90 and 120 days) and levels of metabolisable energy (2.3and 2.5 MCal/kg DM intake) and rations were isonitrogenous (14 percent crude protein on dry matter basis). Ration of 2.3 MCal/ kgDM intake was consisted 60 percent forage (alfalfa) and 40 percent concentrate (15 percent barley, 10 percent beet pulp, 6 percent wheat bran, 8 percent cottonseed meal, 0.05 percent mineral supplement and 0.05 percent salt) and another ration 2.5 M Cal ME/kg DM intake was consisted 40 percent forage (alfalfa) and 60 percent concentrate (34 percent barley, 7 percent wheat bran, 11 percent cottonseed meal, 0.05 percent mineral supplement and 0.05 percent salt). The rations were mixed and fed *ad libitum*. The lambs were weaned at 90±5 days of age and divided randomly in to two fattening periods 90 and 120 days (2×32 lambs), and kept each two lambs in one box. At the end of every fattening period, one- half of the lambs each fattening period in two levels of energy, after 18 h fasted then they were slaughtered randomly. After slaughtering and skinning, all the abdominal and thoracic organs were removed weighed and carcass was prepared. The warm carcass were chilled at 3±2 °c for 24 h and then weighed. The method of cutting described (Farid et al., 1983). The model used to analyze included the fixed effects of fattening period by levels of energy interaction and initial weight fattening as covariate.

Results and Discussion

The average of final weight, metabolic weight and carcass weight were 49.50kg, 18.64kg and 26.53kg and interaction between fattening period (120 days) in different levels of energy for final weight, metabolic weight and carcass weight had significant ($p < 0.05$). The mean of daily weight gain was 162.37g/d, interaction between fattening periods and levels of energy did not influence on daily weight gain and feed intake, Although length of fattening period had highly significant effect on daily weight gain ($P < 0.01$). Results in this study are in agreement with reports (Al-Jassim et al., 1996) length of fattening periods have significant effect on daily weight gain (Fruzandeh et al., 2001). The mean of feed conversion ratio was 10.91 and effect of interaction between length of fattening Periods and levels of energy on feed conversion ratio in Chalishtory lambs was significant ($p < 0.05$), In a study, feed conversion Lori-Bakhtiary ram lamb that used ration with 2.64 and 2.4 M Cal ME / kg DM intake were 10.46 and 10.82 and agreement result of this study (Shadnoush, 1996). The mean of dressing percentage was 53.67% and interaction between fattening period (120 days) in different levels of energy for dressing percentage was significant ($p < 0.05$). A researcher reported dressing percentage Lori-Bakhtiary male lamb (Most Iranian native sheep breed) was 52.1 percent (Karami, 2003) and other researcher reported dressing

percentage of Naeini male lamb with 2.25 and 2.5 M Cal ME / kg DM intake were 52.3 and 55.7, respectively (Fruzandeh, et al., 2001). The means of surface loin area and back fat thickness were 15.31cm² and 8.68mm and interaction between fattening period in different levels of energy for surface of loin area and back fat thickness were significant (p<0.05). But levels of energy had not significant effect on surface lion area. The mean of total carcass meat, total bone, and total subcutaneous fat and fat-tail percent were 46.94, 11.71, 15.96 and 22.87 percent, respectively and interaction between fattening period and levels of energy on carcass compositions (total carcass meat, total bone and total subcutaneous fat percent) were not significant. Results in this study are in agreement with reports (Turgeon et al., 1980; Fruzandeh, et al., 2001; Shadnoush, 1996). However, interaction between fattening period and levels of energy influence on some traits, that we recommended fattening period 90 days with 2.3 M Cal ME/kg DM, because higher daily weight gain, loser daily feed intake, better of feed conversion ratio and otherwise loin area surface and carcass composition were not significant.

References

- Bathaei, S. S. and P.L Loroy, 1996. Growth and mature weight of Mehraban Iranian fat-tail sheep. *Small Rumin. Res.* 22:155-162
- Berg, R. T., and L. E Walters, 1983. The meat animal: Changes and challenges. *J. Anim.Sci.* 57 (suppl.2): 133-146.
- Al-Jassim, R. A. M., S. A. Hassan and A.N. Al-Ani, 1996. Metabolizable energy requirements for maintenance and growth of Awassi lambs. *Small Rumin. Res.* 20: 239-245.
- Bulter-Hogg, B. W., 1984. Growth patterns in sheep: Changes in the chemical composition of the empty body and its constituent parts during weight loss and compensatory growth. *J. Agri. Sci. (Comb.)*. 103:17-24.
- Aziz, N. N., D. M. Murray and R. O. Ball, 1992. The effect of live weight gain and live weight loss on body composition of Merino Wethers: Chemical composition of the dissected components. *J. Anim. Sci.* 70: 3413-3420.
- Canton, J. G., M. A. Velazquez and R. A. Castellanos, 1992. Body composition of pure and crossbred Black belly sheep. *Small Rumin. Res.* 7: 61-66.
- Turgeon, J. r., O. A., Brink, D. R. Barlte, S. J. Klopfenstein and C. L. Ferrell, 1980. Effects of growth rate and compensatory growth on body composition in lambs. *J. Anim. Sci.* 63:770-780.
- Farid, A., J. Izadifard, M. A. Edriss and M. Makarechian, 1983. Meat from culled old ewes of two fat-tailed Iranian breeds. II. Meat, subcutaneous fat and bone in the wholesale cuts. *Iran. J. Agri. Res.* 2:93-114.
- Fruzandeh, A. D., A. Samie and Gh. R. Ghorbani, 2001. Evaluate fattening performance of Naeini Iranian fat-tail sheep used of rations with different levels energy and protein. *J. Science and Technology of Agriculture and Natural Resources.* 4:149-160.
- Shadnoush, Gh. R, 1996. Investigation of effects different levels energy on slaughter weight and carcass characteristics in Lori-Bakhtiary Iranian fat-tail lambs. *M. Sci.Thesis. Khorasgan University.Iran.*
- Karami, M., 2003. Investigation of effects sex on feedlot performance and carcass characteristics of Lori-Bakhtiari lambs. *J. Pajouhesh-Va- Sazandegi in Animal & Fisheries Sciences.* 56: 32-37.

Comparative Efficacy of Herbal Methionine and Synthetic DL-Methionine on Performance in Laying Hen

Kaewmoonmuk, P., W. Jaikan, S. Wongtangtintharn & J. Khajarern

*Department of Animal Science, Faculty of Agriculture, Khon Kaen University,
Khon Kaen 40002, Thailand*

Abstract

A feeding trial was conducted to evaluate the bioefficacy of herbal methionine (HM) compared with synthetic methionine (DLM) in the diets of laying hens. The HM (Methiomax), synthetic methionine (DLM) and equal mixture of both were added to a control basal diet at 0.11, 0.22 and 0.33% and fed to 270 laying hens. The birds were divided into 10 treatments of 27 birds each and each treatment group was replication 3 times with 9 birds per replication. Supplementation DLM, HM and their equal mixture with three graded levels of methionine showed better improvement ($P < 0.05$) on production performance of egg production (EP), feed conversion efficiency (FCR), egg mass (EM), total number of eggs produced (TNEP) and also showed reduction in both abdominal fat content (AFC) and final body weight at termination (BWT) but showed higher feather weight at termination (FWT). In addition, there was no significant effect of methionine source or their equal mixture on all productive performance, BWT, AFC except for FWT which showed that HM and their equal mixture showed higher ($P < 0.05$) FWT more than DLM. Three graded levels of methionine added in the basal control diets exhibited a linear increase with increasing methionine levels in the diets but no significant ($P > 0.05$) influences on all productive performance, BWT, AFC and EWT or a source by level interaction were also observed. Based on this study, it was concluded that HM is an effective substitute for synthetic DLM for optimum production performance in laying hens.

Keywords: laying hen, herbal methionine, DL-methionine, performance

** Corresponding author: Jowaman@kku.ac.th*

Introduction

Since nearly three decades two synthetic methionine sources are available to balance monogastric diets. The biological value of the two main sources of methionine (DL-Methionine, DLM and hydroxyl analogue methionine, 2-hydroxy-4-methyl (thio) butanoic acid with no amino acid group in the molecule or HMTBA) has been debated around the world without reaching a definite conclusion. Methionine supplementation in poultry and swine is a common practice for various vital functions in body such as: protein synthesis, regulation of cell division, methyl donor and lipotropic agents (Aerni et al., 2005). Recent reports (Chattopadhyay et al., 2006) have shown that there are other natural herbal methionines developed to replace synthetic methionine for maintaining animal performance and well-being. Herbal methionine mixtures are phytoadditive

containing herbal ingredients that mimic the activity of methionine which has been found to replace DLM very effectively when used in broiler rations (Kalbande et al., 2009). Therefore, the present study was carried out to determine the comparative efficacy of herbal methionine and synthetic DLM on the productive performance in laying hens.

Materials and Methods

A total of 270 ISA Brown, 16 weeks of age were randomly (on the weight basis) assigned into ten treatments with 3 replications of 9 laying hens per replication. Uniform management and lighting program were provided according to the ISA Brown management manual. Feed and water provided ad libitum throughout the experimental period of 224 days (between 18 and 49 weeks of age) and was divided into pooled 8 periods of 28 days each. Feed intake, egg production and egg weight were recorded daily and data were into 28-day period. On the last three consecutive days of the 28 days periods, eggs gathered were used for egg quality determination. At the earl of the experiment, 2 hens per replicated were slaughtered for carcass grains on feather and abdominal fat weight. Control basal diet (corn-soybean-fish meal-rice bran) without methionine was formulated to contain 17.19 %CP and ME 2,800 kcal/kg. Three levels added of DLM, herbal methionine (HM) and equal mixer of both were formulated at the level 0.11, 0.22 and 0.33% in the diet.

Results and Discussion

Effects of sources and levels of methionine in laying hens on productive performance, final weight, abdominal fat and feather weight at termination were computed and analyzed for the whole lift cycle periods (18-49 wks or 224 days) are shown in Table 1. There was no significant ($P > 0.05$) effect of methionine sources or their equal mixture on productive performance, feed intake (FI), egg productive (EP), feed conversion efficiency (kg feed/kg egg or feed conversion ratio, FCR), egg weight (EW), egg mass (EM), final body weight at termination (BWT) and abdominal fat content (AFC) except for feather weight at termination (FWT) showed a significant ($P < 0.05$) effect of methionine sources. HM and equal mixture of HM and DLM showed higher ($P < 0.05$) on FWT than DLM. Dietary methionine level showed linear increased with increasing methionine level in the dies but methionine no significant ($P > 0.05$) influence on all productive performance, BWT, AFC and FWT or a source by level interaction ($P > 0.05$) were also observed. Three graded levels of methionine supplementation showed better but not significant ($P > 0.05$) on productive performance, BWT, AFC and FWT.

Table 1. The effects of dietary DL-methionine (DL-met) or Herbal methionine (HM) or their equal mixture on performance for overall (21-49 weeks).

Treatment	FI	Egg	Egg	FCR	Egg	No	Final	Abdominal	Feather	
Product (%)	(g/h/d)	production (%)	weight (g)	g egg/g feed	mass	of egg (egg/8period)	weight (%)	fat (%)	weight (%)	
Basal	0.00	109.77 ^a	86.75 ^{ab}	57.34	1.914 ^a	50.22 ^b	194.32 ^b	1.931.67	6.31 ^a	3.97 ^b
DL-met	0.11	106.03 ^{abcd}	88.90 ^{ab}	57.93	1.829 ^{ab}	52.02 ^{ab}	199.15 ^{ab}	1981.00	5.47 ^{ab}	4.11 ^{ab}
DL-met	0.22	105.93 ^{abcd}	88.99 ^{ab}	58.03	1.823 ^{ab}	52.12 ^{ab}	199.34 ^{ab}	1808.33	4.08 ^b	4.29 ^{ab}
DL-met	0.33	106.45 ^{abc}	90.48 ^{ab}	57.71	1.841 ^{ab}	52.69 ^{ab}	202.68 ^{ab}	1890.00	3.79 ^b	4.28 ^{ab}
HM	0.11	102.74 ^{bcd}	87.69 ^{ab}	57.81	1.775 ^b	51.15 ^{ab}	196.44 ^{ab}	1845.00	4.75 ^{ab}	4.76 ^{ab}
HM	0.22	101.77 ^d	88.25 ^{ab}	58.01	1.745 ^b	51.62 ^{ab}	197.67 ^{ab}	1850.33	3.95 ^b	4.75 ^{ab}
HM	0.33	107.09 ^{ab}	90.16 ^{ab}	58.15	1.839 ^{ab}	52.90 ^{ab}	201.97 ^{ab}	1975.30	3.93 ^b	5.11 ^a
DL+HM	0.11	104.43 ^{bcd}	89.96 ^{ab}	58.06	1.796 ^b	52.69 ^{ab}	201.51 ^{ab}	1778.67	3.74 ^b	5.10 ^a
DL+HM	0.22	105.19 ^{bcd}	90.81 ^a	57.85	1.831 ^{ab}	53.01 ^{ab}	203.42 ^a	1741.33	4.09 ^b	4.77 ^{ab}
DL+HM	0.33	102.41 ^{cd}	91.33 ^a	58.06	1.763 ^b	53.45 ^a	204.59 ^a	1770.70	3.54 ^b	4.37 ^{ab}
Pool SEM		2.345	1.967	1.170	0.058	1.578	4.408	163.51	1.075	0.533
Source (S)										
DL-Met		106.13	89.45	57.98	1.831	52.28	200.39	1893.11	4.14	4.22 ^B
HM		103.86	88.70	57.99	1.786	51.89	198.70	1890.22	4.21	4.28 ^A
DL+HM		104.01	90.70	57.99	1.796	53.05	203.17	1763.56	3.79	4.73 ^A
Added (A)										
0.11		104.40	88.85	57.93	1.799	51.95	199.03	1878.67	4.35	4.61
0.22		104.29	89.35	57.96	1.800	52.25	200.15	1800.00	4.03	4.60
0.33		104.32	90.66	57.97	1.814	53.01	203.08	1878.67	3.75	4.58
Significant										
Source (S)		NS	NS	NS	NS	NS	NS	NS	NS	*
Added (A)		NS	NS	NS	NS	NS	NS	NS	NS	NS
SxA		NS	NS	NS	NS	NS	NS	NS	NS	NS
Control vs other										
L1		NS	NS	NS	NS	NS	NS	NS	*	NS
Q1		NS	NS	NS	NS	NS	NS	NS	NS	NS
C1		NS	NS	NS	NS	NS	NS	NS	NS	NS
L2		NS	*	NS	*	**	*	NS	*	NS
Q2		**	NS	NS	**	NS	NS	NS	NS	NS
C2		NS	NS	NS	NS	NS	NS	NS	NS	NS
L3		NS	NS	NS	NS	NS	NS	NS	NS	NS
Q3		*	*	NS	NS	*	*	NS	NS	NS
C3		*	*	NS	*	*	*	NS	*	NS

a,b,c,d Means within column with no common superscript differ significant (P≤0.05)

A,B,C Means within column with no common superscript differ significant (P≤0.05)

L¹ = Linear for DLM., Q¹ = Quadratic for DLM., C¹ = Cubic for DLM. L² = Linear for HM., Q² = Quadratic for HM., C² = Cubic for HM. L³ = Linear for Equal mixture., Q³ = Quadratic for Equal mixture., C³ = Cubic for Equal mixture. NS : not significant at (P≥0.05) ; *: significant (P≤0.05) ; **: significant (P≤0.01)

There was significant (P < 0.05) difference in productive performance on EP, FCR, EW, EM, total number of egg produced (TNEF) and BWT but showed lower (P < 0.05) BWT and AFC which indicated that laying hen required methionine to meet the high productive performance to meet the increasing feather lesser demands associated with productive performance of methionine and protein interrelationship of both to act as lipotropic agent through its role as an amino acid (as a methyl donor) in balancing protein requirement (Chen et al., 1993). There was significant (P < 0.05) difference in performance of broiler fed methionine over that of the basal control diet

(Salome et al., 2010). Recent research reported that DLM supplementation decrease fat deposition (Jeroch & Pack, 1995) which in a agreement with the results of this study.

Conclusion

The results from the feeding trial indicated that herbal methiomax can replace DL-methionine very effectively in the commercial laying hen diets when addition at 3 levels replacement or equal mixture of both in whole eight cycle periods (18-49 wks or 224 days) can be well compared with hens supplemented with DLM. However, further trials are warranted to consolidate the present finding and cost effectiveness of using HM in layer diets.

References

- Aemi, V., M. W. G. Brinkkhof, W. H. Oester and E. Frohlich, 2005. Productivity and mortality of laying hens in aviaries: a systematic review. *World Poult. Sci. J.* 61:130-142.
- Chattopadhyay, K., M. K. Mondal and B. Roy, 2006. Comparative efficacy of DL-methionine and herbal methionine on performance of broiler chicken. *Int. J. Poult. Sci.* 5:1034-1039.
- Kalbande, V. H., K. S. Ravikanth, Maini and D. S. Rekhe., 2009. Metabolism supplementation options in poultry. *Int. J. Poult. Sci.*, 8:588-591.
- Salome, I., I. Dafwang and G. S. Bawa., 2010. Evaluation of methiorep as a substitute for methionine in broiler diets. *Int. J. Poult. Sci.*, 9:809-812.

***In Situ* evaluation of heat treated vegetable protein sources**

Marghazani, I.B.,*, M. A. Jabbar, T.N. Pasha¹, Nasrullah, M.H. Kakar, A.N. Khosa, A.H. Baloch & M. Jan

Faculty of Veterinary and Animal Sciences, Lasbela University of Agriculture, Water and Marine Sciences, Uthal, Balochistan, Pakistan, ¹Faculty of Animal Production and Technology, University of Veterinary and Animal Sciences, Lahore, Punjab, Pakistan

Abstract

Fifteen different vegetable protein sources found in Pakistan were treated at the rate of 1 hour at 15 lb per 100 g crude protein (CP) and evaluated for ruminal degradability characteristics through *in situ* procedure using rumen fistulated Nili-Ravi buffalo steer. Samples of soybean meal, corn gluten meal 60%, maize gluten feed, guar meal, sunflower meal, rapeseed meal, rapeseed cake, canola meal, cottonseed cake, cottonseed meal, coconut cake, coconut meal, palm kernel cake, almond cake and sesame cake were obtained from 10 different locations. Crude protein (CP) ruminal degradability were determined at 0, 3, 6, 12, 24 and 48 hours in triplicate and fitted to Orskov and McDonald equation to determine fractions a, b, degradation rate and effective degradability at 2, 5 and 8 percent.

Keywords: ruminal degradability, vegetable protein sources, undegradable protein,

*Corresponding author: marghazani76@yahoo.com

Introduction

The availability of feed resources in Asia and particularly in Pakistan is far less than the optimal needs of ruminant's animals. This situation is further aggravated due to inefficient management and utilization of available feeds for animal productivity (Walli, 2009). The common example is protein supplementation of ruminant's diet. However, maximizing crude protein in the diet of ruminants particularly for growing and early lactating animals is not the reasonable way out because wide-ranging breakdown of feed protein in the rumen by rumen microbes results in inefficient use of dietary protein (Castillo et al. 2001). Therefore, nutritionists are interested in controlling the rate and extent of ruminal protein degradability. For this purpose, various methods are being applied to protect degradability of high quality protein in the rumen. Of these, heat treatment remained the subject of studies at different application rates (Nakamura et al., 1994). Present study planned to determine the effectiveness of heat treatments in fifteen different vegetable protein sources used for dairy buffalo and cows in Pakistan.

Materials and Methods

Approximately 1 kg representative samples of vegetable protein sources (n=15) were collected from ten (10) different locations. All test feeds were chemically analyzed for DM, ash, CP, ether extract and crude fiber contents (AOAC, 2000). Representative sample of each vegetable protein source (500 g) was subjected to heat treatment using an automatic autoclave (HIRAYAMA Hiclave HVA-85) at the rate of 1 h at 15 lb per 100 g CP (Faran and Pasha, 2000). The ruminal protein degradability of treated vegetable protein meals was determined using the *in situ* procedure (Cottrill and Evans, 1984).

Results and Discussion

Table 1: Chemical composition (mean \pm SE) of vegetable protein sources

Feeds	DM	Ash	CP	EE	CF
Soybean meal	94.08 \pm 0.58 ^{de}	8.56 \pm 0.46 ^b	46.81 \pm 0.49 ^b	0.98 \pm 0.12 ^d	1.16 \pm 0.13 ⁱ
CGM 60 %	93.25 \pm 0.52 ^e	1.45 \pm 0.24 ^f	61.68 \pm 0.37 ^a	5.69 \pm 0.20 ^c	1.71 \pm 0.24 ⁱ
CGM 30%	94.34 \pm 0.56 ^{cde}	8.18 \pm 0.60 ^b	26.18 \pm 0.57 ^g	1.23 \pm 0.21 ^d	5.58 \pm 0.38 ^h
Guar meal	97.21 \pm 0.44 ^a	5.83 \pm 0.64 ^b	42.24 \pm 0.65 ^c	5.51 \pm 0.37 ^c	10.14 \pm 0.99 ^{dc}
Sunflower meal	94.56 \pm 0.6 ^{cde}	6.16 \pm 0.54 ^e	30.73 \pm 0.58 ^f	1.48 \pm 0.14 ^d	25.53 \pm 0.76 ^a
Rapeseed meal	96.91 \pm 0.45 ^{ab}	8.67 \pm 0.80 ^b	38.17 \pm 0.59 ^e	1.69 \pm 0.19 ^d	9.02 \pm 0.60 ^{ef}
Rapeseed cake	94.62 \pm 0.52 ^{cde}	6.14 \pm 0.61 ^e	36.74 \pm 0.45 ^e	8.50 \pm 0.55 ^a	10.1 \pm 0.65 ^{def}
Canola meal	93.89 \pm 0.58 ^e	6.46 \pm 0.54 ^d	38.17 \pm 0.54 ^e	1.05 \pm 0.11 ^d	11.4 \pm 0.51 ^{cd}
Cottonseed cake	94.79 \pm 0.41 ^{cde}	4.87 \pm 0.43 ^{ef}	23.76 \pm 0.49 ^h	8.70 \pm 0.44 ^a	21.80 \pm 0.85 ^b
Cotton seed meal	95.73 \pm 0.40 ^{abc}	5.80 \pm 0.38 ^e	40.54 \pm 0.46 ^d	1.13 \pm 0.09 ^d	8.43 \pm 0.33 ^{fg}
Coconut cake	94.00 \pm 0.52 ^{de}	6.35 \pm 0.23 ^{cd}	20.57 \pm 0.31 ^j	8.05 \pm 0.33 ^{ab}	12.61 \pm 0.38 ^c
Coconut meal	95.60 \pm 0.41 ^{bcd}	8.81 \pm 0.56 ^b	22.08 \pm 0.28 ⁱ	1.56 \pm 0.22 ^d	9.56 \pm 0.48 ^{ef}
Palm kernel cake	75.95 \pm 0.74 ^g	4.23 \pm 0.33 ^e	17.08 \pm 0.45 ^k	7.91 \pm 0.21 ^{ab}	12.64 \pm 0.52 ^c
Almond cake	93.83 \pm 0.46 ^e	21.71 \pm 0.74 ^a	41.25 \pm 0.62 ^{cd}	5.74 \pm 0.46 ^c	7.04 \pm 0.39 ^{gh}
Sesame cake	91.79 \pm 0.38 ^f	7.81 \pm 0.37 ^{bc}	37.84 \pm 0.53 ^e	7.35 \pm 0.34 ^b	3.97 \pm 0.30 ⁱ
Significance level	P<0.001	P<0.001	P<0.001	P<0.001	P<0.001

Means with different superscripts within same column are significantly different (P<0.05).

DM= dry matter; CP= crude protein; EE= ether extract; CF= crude fiber; CGM= corn gluten meal

Table 2: Mean (\pm SE) *in situ* crude protein degradation kinetics and effective degradability of heat treated vegetable protein sources at different rumen passage rates

Feeds	Crude protein degradation kinetics			Effective degradability (%)		
	a (%)	b (%)	c (h ⁻¹)	kp= 0.02	kp=0.05	kp=0.08
Soybean meal	4.23 \pm 1.06 ^g	48.56 \pm 1.47 ^{cd}	0.06 \pm 0.00	39.78 \pm 1.21 ⁱ	29.67 \pm 1.20 ^h	24.07 \pm 1.15 ^j
CGM 60%	2.85 \pm 0.38 ^g	16.54 \pm 1.62 ^h	0.09 \pm 0.00	16.18 \pm 1.37 ^k	13.22 \pm 1.08 ⁱ	11.35 \pm 0.92 ^k
CGM 30%	7.94 \pm 0.62 ^{ef}	44.97 \pm 1.62 ^{de}	0.07 \pm 0.00	42.07 \pm 1.01 ^{hi}	33.12 \pm 1.02 ^g	27.90 \pm 1.01 ⁱ
Guar meal	9.77 \pm 0.76 ^{de}	50.38 \pm 0.77 ^{bc}	0.08 \pm 0.00	49.48 \pm 0.90 ^e	39.93 \pm 0.98 ^e	34.08 \pm 0.99 ^f
Sunflower meal	11.77 \pm 0.65 ^c	62.38 \pm 1.44 ^a	0.12 \pm 0.05	59.43 \pm 0.89 ^d	46.96 \pm 0.80 ^d	39.70 \pm 0.74 ^e
Rapeseed meal	12.30 \pm 0.89 ^c	64.02 \pm 1.22 ^a	0.07 \pm 0.00	62.35 \pm 0.55 ^c	50.04 \pm 0.51 ^c	42.63 \pm 0.58 ^d
Rapeseed cake	17.01 \pm 0.61 ^b	60.93 \pm 0.80 ^a	0.08 \pm 0.00	65.63 \pm 0.40 ^b	54.37 \pm 0.56 ^b	47.36 \pm 0.62 ^c
Canola meal	7.65 \pm 0.67 ^f	46.70 \pm 1.89 ^{de}	0.08 \pm 0.00	44.63 \pm 1.21 ^{fg}	35.84 \pm 0.95 ^f	31.01 \pm 0.86 ^{gh}
Cottonseed cake	16.72 \pm 0.61 ^b	64.02 \pm 1.18 ^a	0.07 \pm 0.00	66.76 \pm 0.61 ^b	54.48 \pm 0.57 ^b	47.06 \pm 0.60 ^c
Cottonseed meal	12.91 \pm 0.73 ^c	33.50 \pm 1.16 ^g	0.09 \pm 0.00	40.02 \pm 1.11 ⁱ	34.00 \pm 0.96 ^{fg}	30.17 \pm 0.89 ^{hi}
Coconut cake	10.86 \pm 0.53 ^{cd}	45.34 \pm 0.93 ^{de}	0.08 \pm 0.00	46.85 \pm 0.74 ^f	38.60 \pm 0.76 ^e	33.17 \pm 0.74 ^{fg}
Coconut meal	8.54 \pm 0.51 ^{ef}	38.12 \pm 0.72 ^f	0.06 \pm 0.00	37.20 \pm 0.47 ^j	29.49 \pm 0.59 ^h	25.05 \pm 0.61 ^j
Palm kernel cake	8.21 \pm 0.35 ^{ef}	43.77 \pm 0.73 ^e	0.08 \pm 0.00	43.24 \pm 0.53 ^{gh}	35.21 \pm 0.42 ^{fg}	30.19 \pm 0.38 ^{hi}
Almond cake	17.96 \pm 0.71 ^b	64.53 \pm 0.84 ^a	0.09 \pm 0.00	71.06 \pm 0.40 ^a	60.00 \pm 0.70 ^a	52.72 \pm 0.82 ^b
Sesame cake	26.52 \pm 0.72 ^a	52.57 \pm 1.13 ^b	0.10 \pm 0.00	70.10 \pm 0.45 ^a	61.27 \pm 0.55 ^a	55.36 \pm 0.65 ^a
Significance level	P<0.001	P<0.001	NS	P<0.001	P<0.001	P<0.001

Means with different superscripts within same column are significantly different (P<0.05)

CGM= corn gluten meal; NS= non significant (P>0.05); a= quickly soluble fraction; b= potentially degradable fraction; c= degradation rate of fraction b; kp= rumen passage rate

The chemical composition of the test feeds is given in Table 1. The DM, ash, CP, ether extract and crude fiber contents varied significantly (P<0.001) among the test feeds. Crude protein degradation kinetics and ED at different rumen passage rates (0.02, 0.05 and 0.08) of the heat treated vegetable protein sources are given in Table 2. Fractions “a” and “b” varied significantly (P<0.001), while fraction “c” was the same (P>0.05) among the test feeds. Crude protein degradation kinetics (a, b, and c fractions) and ED of the vegetable protein sources at three rumen passage rates (0.02, 0.05 and 0.08) showed the effectiveness of heat treatment in reducing ruminal protein degradability. However, variation with the literature exist which may be due to differences in the nature of the heat treatments, such as differences in the temperature and duration of the heat

treatment (Mahala and Gomma, 2007). Exposure of a protein source to high temperature for a long period of time increases the acid detergent insoluble N content by Millard reaction (Broderick et al., 1991), damages some amino acids (Kung and Rode, 1996) and hence decreases digestibility (McNiven et al., 2002). Studying the effect of heat treatment on protein feeds, Broderick and Craig (1983) concluded that it produces two way effects by blocking reactive sites for rumen microorganisms' activity and by reducing protein solubility.

References

- AOAC. 2000. Association of official analytical chemists. Official methods of analysis, 17th ed., Washington, DC, USA.
- Broderick G.A, Craig W.M. 1983. Mechanism of protein degradation by rumen microbes. Fed. Proc. 42:532 (Abstr.).
- Broderick G.A, Wallace R.J, Orskov E.R. 1991. Control of rate and extent of protein degradation. In: T. Tsuda, Y. Sasaki and R. Kawashima (Eds.) Physiological Aspects of Digestion and Metabolism in Ruminants. Academic Press, London, pp. 541-592
- Castillo, A.R., E. Kebreab, D.E. Beever, J.H. Barbi, J.D. Sutton, H.C. Kirby and J. France. 2001. The effect of protein supplementation on nitrogen utilization in lactating dairy cows fed grass silage diets. J. Anim. Sci. 79:247-253
- Cottrill, B.R and P.J. Evans. 1984. Estimation of protein degradability: A standard method for *in sacco* measurement of nitrogen measurement of nitrogen disappearance from dacron bags suspended in the rumen. Recommendation of the Inter-departmental protein working party of Agric. Research Council, U. K.
- Kung, L. Jr., Rode, L.M. 1996. Amino acid metabolism in ruminants. Anim. Feed Sic. Technol. 59 : 167-172
- Mahala, A.G. and A. S. Gomma. 2007. Effect of heat treatment on sesame cake protein degradation. Res. J. Anim. and Vet. Sci. 2: 39-42
- McNiven, M.A., Prestlokken, E., Mydland, L.T., Mitchell, A.W. 2002. Laboratory procedure to determine protein digestibility of heat-treated feedstuffs for dairy cattle. Anim. Feed Sci. Tech. 96: 1-13
- Nakamura, T., T.J. Klopfenstein, D.J. Gibb, R.A. Britton. 1994. Growth efficiency and digestibility of heated protein feed to growing ruminants. J. Anim. Sci. 72: 774-782
- Walli, T.K. 2009. Densified TMR - an efficient way of using limited feed resources for bovine feeding in tropics. Pakistan J. Zool. Suppl. 9: 727-734

Efficacy of probiotics (Sanizyme) on performance in broiler diets

Sutthibak, N., S. Wongtangtintan & J. Khajarern

1Department of Animal Science, Faculty of Agriculture, Khon Kean University, Khon Kean 40002, Thailand

Abstract

The effects of probiotics Sanizyme on performance of broilers were assessed in evaporating house system. The probiotics Sanizyme were supplementation with 4 graded levels (0.025, 0.050, 0.075, 0.100%) in control diet with 3 periods of feeding : starter (1-21d), grower (22-35d) and finisher (36-42d) diet was formulated to contain ME 3,121 kcal/kg and 22.52% CP, ME 3,175 kcal/kg and 20.7% CP and ME 3,242 kcal/kg and 18.01% CP respectively. For the whole period of testing (1-42d) feeding with 4 graded levels of Sanizyme in control no added group showed both significant ($P < 0.05$) linear and quadratic improvement of feed conversion ratio (FCR), feed cost/kg BWG (FCG) and also showed increasing linear ($P < 0.05$) and quadratic but not significant ($P > 0.05$) on productive index (PI) with the highest improvement FCR, PI, final body weight (FBW), body weight gain (BWG) and showed the lowest on feed cost/kg BW with the highest on salable bird return (SBR) and also showed the highest on net profit return when addition Sanizyme at the level 0.075% in the broiler diet when compared with the control no added group and showed the highest return of investment (ROI). The results from this study indicated that Sanizyme can be used for probiotics assisted digestion and enhancer growth in broilers.

Keywords: broiler, probiotics, performance, economic benefits return

**Corresponding author: jowaman@kku.ac.th*

Introduction

Probiotics are defined as “living organism, which upon ingestion in certain number, exert health benefits beyond inherent basic nutrition” (FAO/WHO, 2001). Over the last 3 decades, probiotics (direct-fed microbial), including *Lactobacillus* and *Bacillus cereus* culture, have been used extensively. Probiotics have been used as nutritional bioregulators or growth promoters to replace widely used antibiotics (Fuller, 1989; Fuller, 1999; Nguyen, 1991). During the last two decade, a renewed interest has been concentrated on the role and effects of probiotics on animal production. It has been reported by many researchers that probiotics not only act as growth promoters or bioregulator and feed savers, but also as immunostimulants. The present study was, therefore, to determine the effect of feeding probiotics on growth performance in broilers.

Materials and Methods

Birds and Management

A total of 260 day-old mixed-sex of commercial broiler strain (Cobb 500) was randomly assigned into five treatments with 2 replications of 26 chicks per replicate. Chicks were allocated on the basis of weight so that the overall weight and weight range were similar for each replicate group. Chicks were housed in fresh rice hull litter floored pens with two hanging feeders and one bell drinker located in a test facility with evaporation house at Khon Kaen University Poultry Farm

with permanent artificial illumination was applied, and room temperature decreased from 34 to 25°C in the following steps: d 1 to 2: 34°C, d 3 to 4: 32°C, d 5 to 7: 30°C, d 8 to 14: 28°C, d 15 to 21: 28°C, d 22 to 42: 25°C. Chicks were vaccinated for Marek's disease, Newcastle disease and Infections bronchitis at the hatchery and at 12d against infections bursa disease. All birds received common mash from feeds from placement 1 to 21d, 22 to 35d and 36 to 42d respectively. Feed and water were provided *ad libitum*

Diets and parameters studied

Control starter (1-22d), grower (22-35d) and finisher (36-42d) diet was formulated contain ME 3,121 kcal/kg and 22.52% CP, ME 3,175 kcal/kg and 20.07% CP and ME 3,242 kcal/kg and 18.01% CP, respectively. Subsequently three other experiment diets were formulation by supplementation Sanizyme 4 levels (0.025%, 0.050%, 0.075% and 0.100%) to the control diet. Salinomycin (60 ppm) was supplementation in both starter and grower diets to control coccidiosis, Feed and water were provided *ad libitum* feed were fed in mash form. Pen body weight and survival rate were recorded at 21, 35 and 42 days of age and total feed consumption during those interval was also determined. Adjustment in feed conversion were made for birds that died during the study.

Statistical Analysis

All data were subjected to analysis of variance using the GLM procedure of SAS software (SAS, 2004). Differences among mean were detected by Duncan's multiple range tests. Significance was considered at $P < 0.05$ levels.

Results and Discussion

The performance of the broilers fed with 4 graded levels (0.025, 0.050, 0.075 and 0.100%) of Sanizyme for the whole period of testing (1-42 d) was presented in Table 1. Feeding 4 graded levels of Sanizyme in control no added group showed linear ($P < 0.05$) and quadratic ($P < 0.05$) response in improvement of feed conversion ratio (FCR) and feed cost/kg BWG and also showed increasing linear ($P < 0.05$) and Quadratic but not significant ($P > 0.05$) on productive index (PI) with the highest improvement FCR, PI, final body weight (FBW), body weight gain (BWG) and showed the lowest on feed cost/kg BWG with the highest on salable bird return (SBR) and also showed the highest on net profits return when added Sanizyme at the level 0.075% in the broiler diet. Sanizymes when added with 4 graded levels in control no added group showed improvement broilers growth performance but not significant ($P > 0.05$) by increasing both FBW and BWG. The results from this study showed that supplementation of Sanizyme maximize feed digestion and absorption. Among the Sanizyme graded 4 levels group, addition of Sanizyme 0.075% showed the highest performance on FBW, BWG, PI and the lowest FCR with the highest economic benefits return on SBR and NPR and showed the highest return of investment (ROI)

Table 1. Effects of Sanizyme on the performance in broilers for 42 d testing

Sanizyme (%)	Survival rate (%)	Final Weight (g)	BWG (g)	FI (g)	FCR (g feed : g gain)	PI ¹	FCG1 ² (Baht/ Kg BW)
Control	92.30	2,568.87	2,525.35	4,470.90	1.770 ^a	313.47 ^b	28.60 ^a
0.025	100.00	2,691.68	2,647.71	4,463.10	1.686 ^b	373.99 ^{ab}	27.47 ^b
0.050	98.10	2,696.19	2,652.57	4,354.30	1.642 ^{bc}	377.85 ^a	27.03 ^b
0.075	98.10	2,759.29	2,715.36	4,356.70	1.604 ^c	395.14 ^a	26.69 ^b
0.100	96.15	2,718.15	2,674.01	4,403.30	1.647 ^{bc}	372.38 ^{ab}	27.60 ^{ab}
<i>Pooled SEM</i>	2.96	88.42	88.42	144.96	0.02	23.57	0.39
----- Probability -----							
Contrast							
<i>L</i>	NS	NS	NS	NS	**	*	*
<i>Q</i>	NS	NS	NS	NS	**	NS	*
<i>C</i>	NS	NS	NS	NS	NS	NS	NS
<i>QU</i>	NS	NS	NS	NS	NS	NS	NS

^{a,b,c} Mean within column with no common superscript differ significant ($P \leq 0.05$)

L = Linear for Sanizyme, Q = Quadratic for Sanizyme, C=Cubic for Sanizyme, QU = Quartic for Sanizyme,

NS: not significant at ($P \geq 0.05$); *: significant ($P \leq 0.05$); **: significant ($P \leq 0.01$)

$$^1 \text{Productive index (PI)} = \frac{\text{BWG (kg)} \times 100 \times \text{Survival}}{\text{Age} \times \text{FCR}}$$

$$^2 \text{Feed cost per gain (FCG)} = \frac{\text{FI (g)} \times \text{feed cost (Baht)}}{\text{BWG (g)}}$$

Table 2. Effects of Sanizyme in diets on economic benefits return for overall 42 d of testing.

Sanizyme (%)	FCG2 ¹ Baht/bird	SBR ² Baht/bird	NPR1 ³ Baht/bird	ROI1 ⁴ Baht/bird	ROI2 ⁵ Baht/kg BW
Control	72.22	92.47	20.25 ^a	-	-
0.025	72.73	96.90	24.17 ^b	3.92	1.47
0.050	71.70	97.06	25.36 ^b	5.11	1.93
0.075	72.47	99.33	26.85 ^b	6.61	2.43
0.100	73.80	97.85	24.03 ^b	3.80	1.42
<i>Pooled SEM</i>	2.44	3.18	1.41	-	-
----- Probability -----					
Contrast					
<i>L</i>	NS	NS	*	-	-
<i>Q</i>	NS	NS	*	-	-
<i>C</i>	NS	NS	NS	-	-
<i>QU</i>	NS	NS	NS	-	-

^{a,b,c} Mean within column with no common superscript differ significant ($P \leq 0.05$)

L = Linear for Sanizyme, Q = Quadratic for Sanizyme, C = Cubic for Sanizyme, QU = Quartic for Sanizyme,

NS: not significant at ($P \geq 0.05$); *: significant ($P \leq 0.05$); **: significant ($P \leq 0.01$)

$$^1 \text{Feed cost per gain (FCG2)} = \text{FCG1} \times \text{BWG (kg)}$$

$$^2 \text{Salable bird return (SBR)} = \text{Price of live chicken (36 Baht)} \times \text{BW (g)}$$

$$^3 \text{Net profits return per bird (NPR1)} = \text{SBR} - \text{FCG2}$$

$$^4 \text{Return of investment by comparing with and the control group (ROI1)} = \text{NPR(Sanizyme)} - \text{NPR (control)}$$

$$^5 \text{ROI2} = \text{ROI1/BWG (kg)}$$

Conclusion

The result from this experiment clearly demonstrated that Sanizyme supplement in the whole period (42d) in evaporating house showed a benefits effect on performance, enhancing nutrients especially fat and protein digestion resulting in an overall improvement of growth performance, feed efficiency (FCR) and FCG with higher economic benefits return and increasing in ROI. Addition 0.075% in control diet showed the highest broiler in both performance and ROI.

References

- FAO/WHO, 2001. Evaluation of Health and Nutritional Properties of Probiotics in Food Including Powder Milk with Live Lactic Acid Bacteria. www.fao.org.
- Fuller, R. 1989. Probiotics in man and animals. *J. Appl. Bacteriol.* 66: 365–378.
- Fuller, R. 1999. Probiotics for farm animals. *J. Appl. Bacteriol.* 66: 365-378
- Nguyen T. 1991. Probiotics a nutritional bioregulator. *World Poul.* 7: 37.
- SAS-Statistical Analysis System, 2004. SAS User's Guide: Statistics. SAS Inst. Inc., Cary, NC; Version 9.1 for Window.

Efficacy of probiotics (Sanizyme) on performance and digestibility in weaning piglets diets

Patarapreecha¹, P., W. Jaikan¹, S. Wongtangtintan¹,
A. Juangsaman² & J. Khajarern¹

¹Department of Animal Science, Faculty of Agriculture, Khon Kaen University,
Khon Kaen 40002, Thailand, ²Mittraphap Feed Mill Co., Ltd 11
Soi Ramkamhaeng 81/1, Huamark, Bangkok,
Bangkok 10240, Thailand

Abstract

The experiment was evaluated the efficacy of probiotics Sanizyme with three levels (0.0125, 0.0250 and 0.0375%) in weaning piglets diets. The results from these three levels supplementation Sanizyme in weaned or nursery feed showed improvement in digestibility of carbohydrate, fat and protein in feed components and remarkable improvement in body weight, daily weight gain, better feed efficiency and uniformity of body weight at termination. linear lower feed cost per kilogram body weight gain when increasing 3 graded levels of Sanizyme in feed and also graded increasing in both net profit per head and economic benefits return over those fed the unsupplemented control group. Addition of Sanizyme at the level of 0.0375% feed appears to be the maximum response on growth and feed utilization when compared with the control unsupplemented group. The results showed that Sanizyme can be used for probiotics, assisted digestion and enhancer growth in weaning pigs.

Keywords: piglets, probiotics, growth performance, nutrients digestibility, economic benefits return

**Corresponding author: jowaman@kku.ac.th*

Introduction

Probiotics are a natural product which have been used as nutritional bioregulators or growth promoters to replace widely used antibiotics (Fuller, 1999; Nguyen, 1991). During the last two decade, a renewed interest has been concentrated on the role and effects of probiotics on animal production. It has been reported by many researchers that probiotics not only act as growth promoters or bioregulator and feed savers, but also as immunostimulants useful to animal performances and health. The effects have been reported in pigs as well as in poultry, calves, rabbit and cultured fish (Kozasa, 1978; Tournut, 1989; Nguyen, 1991; Khajarern & Khajarern, 1994; Khajarern et al., 1996; Khajarern & Ratanasethakul, 1998). Continuous usage of probiotics may reinforce the non-specific immunity system of animals and consequently anti-infectious treatments should be reduced. The purposes of this study was to assess the efficacy of probiotics Sanizyme (Quality Meat Co. LTD, Thailand) supplementation in nursery feeds.

Materials and Methods

The experiment was conducted in a well manage commercial farm (Mittraphap Farm) in Lopburi Province, Thailand. A total of 700 crossbred piglets [Duroc x (Yorkshire x Landrace)] after weaning (21 days of age with average BW 6.80 kg) with balance sexes and genetic background were divided into five treatments with 4 replications of 35 piglets each (18 for males and 17 for females). All piglets were house in an evaporation regulated nursery pen throughout the study (21-56 days of age). All piglets were given *ad libitum* access to feed and water. One eight-hole feeders and four nipple waterers were available per pen. Piglets were weighed at initial and final weight (35 days period). Feed intake and mortality of diarrhea are recorded every day until termination. Piglets uniformity was accounted as percentage units that fell into two times of standard deviation based on average live weight at termination. The five treatments consisted of the control diet was formulated to contain ME 3,245 kcal/kg and 21.43 %CP for treatment 1 and the control diet supplemented with probiotics A at the level of 1 kg/ton or 0.10% for treatment 2 and 3 levels (0.0125, 0.0250 and 0.0375%) of Sanizyme (concentration 1×10^9 spores/g) for treatment 3, 4 and 5 respectively. The diets were fed to piglets in pelleted form. Before termination 7 days, all experimental diets are added chromic oxide 0.1% for 7 days and feces are collected on d 5-7 from each pen for digestibility evaluation on dry matter, crude protein, fat or ether extract, crude fiber, Calcium, Phosphorus and Energy (AOAC, 1995).

Statistical analysis

Data for all response variable were analyzed one-way designs by analysis of variance using the General Linear Model (GLM) procedure of SAS system (SAS, 2004) Duncan's Multiple Range Test was used to determine treatment differences. All statements of significance were based on the probability level of 0.05

Results and Discussion

The results of the experiment are shown in Table 1. For the whole period of testing both probiotics in weaning piglets fed for 35 days especially three levels (0.0125, 0.0250 and 0.0375%) of Sanizyme when compared to the negative no added control showed better significant ($P < 0.05$) feed efficacy (feed conversion ratio, FCR) improved piglets performance but not significant ($P > 0.05$) by increasing body weight (BW), body weight gain (BWG), average daily gain (ADG), daily feed intake (DFI), survival rate (SR) and Uniformity of body weight at termination (UBWT). Effects of supplementation of Sanizyme on digestibility of nutrients in the diets (dry matter, DM; crude protein, CP; fat or ether extract, EE; crude fiber, CF; Calcium, Ca; Phosphorus, P and energy, EN) are shown in Table 2. Feeding weaning pigs with three levels Sanizyme showed improving in digestibility of DM, CP, EE, CF, P and EN in the diets and showed significant ($P < 0.05$) increasing DM, CP, EE, CF, P and EN when compared with the control no added Sanizyme. Linear and quadratic difference were observed ($P > 0.05$) on performance but showed difference ($P < 0.05$) on nutrients digestibility DM, CP, CF, P and EN with three levels supplementation in weaning pigs diets. Supplement of Sanizyme in weaning pigs diets showed generated benefits in both performance and nutrients digestibility especially on DM and EE. Feeding Sanizyme showed synergistic benefits in both performance and nutrients digestibility. The results from this experiment clearly showed that supplementation with three levels of Sanizyme showed improvement the digestibility of feed component which lead to give better improvement in BWG, ADG, better feed efficiency, increasing UBWT, linear decreasing feed cost per kilogram body weight gain when increasing added Sanizyme three levels in feed and also increasing net profit per head with economic benefits return per head 27.60, 32.91 and 49.96 Baht when added 0.0125, 0.0250 and 0.0375% in the weaning pigs diets.

Table 1. Effects of Sanizyme on the performance in weaning piglets during 21- 56 days of age

Parameter	T1	T2	T3	T4	T5	<i>Pooled SEM</i>
	CON	Pro A	SZ(0.0125)	SZ(0.0250)	SZ(0.0375)	
Initial no. of piglets	140	140	140	140	140	
Final no. of piglets	140	140	140	140	140	
Survival rate	100.00	100.00	100.00	100.00	100.00	
Initial weight, kg	6.82	6.83	6.82	6.82	6.82	0.161
Final weight, kg	18.67	19.05	19.18	19.12	19.53	0.692
Av. Body weight gain, kg (BWG)	11.85	12.23	12.36	12.30	12.72	0.600
<i>Improvement, %</i>		+3.21	+4.30	+3.80	+7.34	
Av. Daily gain, g (ADG)	338	349	353	351	363	17.219
<i>Improvement, %</i>		+3.25	+4.44	+3.85	+7.40	
Av. Daily feed intake, g (ADFI)	484	486	493	479	494	23.616
<i>Improvement, %</i>		+0.41	+1.86	-1.03	+2.07	
Feed : Gain (FCR) ^A	1.430 ^a	1.392 ^{ab}	1.396 ^{ab}	1.364 ^b	1.361 ^b	0.031
<i>Improvement, %</i>		+2.66	+2.38	+4.62	+4.83	
Uniformity of body weight, %	88.83	93.94	93.12	91.55	92.36	3.768
<i>Improvement, %</i>		+5.11	+4.29	+2.72	+3.53	
Feed cost/kg BWG, Baht ^A	32.60 ^a	31.94 ^{ab}	31.86 ^{ab}	31.26 ^b	31.21 ^b	0.687
Decreasing feed cost/kg BWG, Baht		-0.66	-0.74	-1.34	-1.39	
Av. Net profit/head, Baht* ^A	443.58 ^b	465.35 ^{ab}	471.18 ^{ab}	476.49 ^{ab}	493.54 ^a	27.183
Economic benefits return/head, Baht		+21.77	+27.60	+32.91	+49.96	

^{a,b} Means within row with no common superscript differ significant ($P < 0.05$)

^A Linear ($P < 0.05$) on Sanizyme inclusion

* Av. Net profit/head calculated from average overall body weight gain x sale price of pigs (70 Baht/kg)

Table 2. The effects Sanizyme on nutrients digestibility during 54-56 days of age

Item	T1	T2	T3	T4	T5	<i>Pooled SEM</i>	<i>P-values</i> ^A		
	CON	Pro A	SZ (0.0125)	SZ (0.0250)	SZ (0.0375)		<i>L</i>	<i>Q</i>	<i>C</i>
dry matter	89.45 ^c	89.65 ^c	91.76 ^a b	92.00 ^a	90.99 ^b	0.535	0.0005	<0.000 1	0.4544
crude protein	84.90 ^c	84.83 ^c	89.45 ^a	89.58 ^a	88.19 ^b	0.583	<0.000 1	<0.000 1	0.0416
fat	82.84 ^b	82.69 ^b	85.32 ^a b	86.50 ^a	86.60 ^a	1.841	0.0131	0.2380	0.9559
crude fiber	46.39 ^b	65.63 ^a	63.90 ^a	65.77 ^a	63.78 ^a	4.336	0.0003	0.0014	0.2862
calcium	58.48	57.82	59.27	61.53	58.03	4.234	0.9239	0.3249	0.4539
phosphorus	55.74 ^b	56.97 ^b	67.44 ^a	68.13 ^a	65.67 ^a	2.368	<0.000 1	<0.000 1	0.1198
energy	90.04 ^c	90.32 ^c	92.30 ^a b	92.51 ^a	91.62 ^b	0.541	0.0006	<0.000 1	0.3854

^{a,b,c} Means within row with no common superscript differ significant ($P < 0.05$)

^A *P-values* for linear (*L*), Quadratic (*Q*) and Cubic (*C*) effect for CON and Sanizyme[®]

Conclusion

The results from these experiments clearly demonstrated that supplementation of Sanizyme and probiotics A have performed remarkable improvement the digestibility of nutrients in feed components in nursery or weaned feed. Supplementation of Sanizyme with three levels in weaned feeds gives better more growth and improvement better feed efficiency and increasing net profit per head with the highest economic benefits return when supplementation at 0.0375% in feed compared with the control no added group. The results showed that Sanizyme and probiotics A can be used for stimulate appetite, assisted digestion and enhancer growth in weaning pigs diets.

References

- AOAC. 1995. Official method of analysis. 16th Edition. Association of Official Analytical Chemists, Washington, DC.
- Fuller, R., 1999. Probiotics for farm animals, Probiotics – A critical Review, Edited by G.W. Tannock, Wymondham, England: Horizontal Scientific Press.
- Khajarearn, S. and J. Khajarearn, 1994. Effects of probiotics (Toyocerin) in sow and creep feeds on resistance of diarrhea in piglets. Proceeding of 13th IPVS Congress, Bangkok, Thailand, 26-30 June 1994. p. 294.
- Khajarearn, J., S. Khajarearn and C. Ratanasethakul, 1996. The efficacy of probiotics (Toyocerin) in broiler breeder feed for inducing immunostimulant and bioregulator effect. Proceeding and Abstract of the 20th World's Poultry Congress, New Delhi, India. 2-5 September 1996. p. 26.
- Khajarearn, J. and C. Ratanasethakul, 1998. Probiotics in breeder feed. World poultry-Elsevier Volume 14 No. 7. 1998. P. 18-19.
- Kozasa, M., 1978. Feed additives of new type. Viable bacterial spores preparation Toyocerin Powder, shiryo-to-shiryo-kogno. J. Feed and Feed Industry 18(12): 45-48.
- Nguyen, T. H., 1991. Probiotics a nutritional bioregulator. PIGS-Misset Jan/Feb 1991. P. 27.
- National Research Council, 1998. Nutrient Requirement of Swine, 10th ed. National Academy Press, Washington, DC.
- SAS-Statistical Analysis System. 2004. SAS User's Guide: Statitics. SAS Inst. Inc., Cary, NC; Version 9.1 for Window.
- Tournut, J., 1989. Applications of probiotics to animal husbandry. Rev. Sci. Tech. Off. Int. Epiz. 8: 551-566.

Effects of condensed tannins of some tropical plants on ruminal gas production *in vitro*

Petlum^{1,*}, A., P. Paengkoum¹, J.B. Liang² & K. Vasupen³, K. Wilachai¹ & N. Onjai-uea¹

¹School of Animal Production Technology, Institute of Agricultural Technology, Suranaree University of Technology, NakhonRatchasima, 30000, Thailand

²Laboratory of Industrial Biotechnology, Institute of Bioscience, Universiti Putra Malaysia, 43400 UPM, Serdang, Malaysia

³Department of Agricultural Technology and Environment, Faculty of Sciences and Liberal Arts, RajamangalaUniversity of Technology Isan, NakhonRatchasima, 30000, Thailand

Abstract

The objectives of this study was to investigate the effect of condensed tannins (CTs) in some tropical plants species on *in vitro* ruminal gas production. Three plant species, include leucaena (*Leucaena leucocephala*) (LN), cassava (*Manihot esculenta*, Cranzt) (CV), and Siam neem (*Azadirachta indica* A. Juss. var. *siamensis* Valetton) (SN) (CTs contained at 2.9, 1.2 and 5.0 % of DM, respectively) were selected to use in this study. Plant species had effect on *in vitro* gas kinetic parameters and gas productions ($P < 0.01$). However, only high concentration of CTs in SN (5.0% of DM) showed the inhibitory effect on the insoluble fractions and methane production at 48 h of incubation. The results suggested that various plant species contained different concentration of CTs had different effect on *in vitro* gas and methane production, however, others factors affected on biological properties of CTs, such as molecular weight and structure should be further investigated.

Keywords: phenolic compound, secondary metabolites, rumen, polyethylene glycol.

*Corresponding author: ananpetlum@hotmail.com

Introduction

Utilization of local available plants for ruminant feeding is increasing interested, this due to limited supplied and rising costs of conventional feed stuffs (Cherdthong & Wanapat, 2010). In addition, tropical plants containing secondary compounds such as condensed tannins (CTs), have shown potential in rumen fermentation manipulation, such as increases rumen-undegradable protein (Waghorn, 2008) and especially mitigation of enteric methane emission (Kreuzer et al., 2009; Patra & Saxena, 2011). However, effects of CTs on rumen manipulation are varied and depending on their source and concentration. In ordered to find out further basically important information for efficiently using of locally available plant as ruminant feeds, therefore, the objectives of this study were to characterize the CTs content in some tropical plant species and to investigate their effects on *in vitro* gas production.

Materials and Methods

Determination of *in vitro* gas production

Leaves of 3 plant species including of leucaena (*Leucaena leucocephala*), cassava (*Manihot esculenta*, Cranzt), and Siam neem (*Azadirachta indica* A. Juss. var. *siamensis* Valetton) which contained CTs at 2.9, 1.2 and 5.0 % of DM (unpublished data) were harvested from the area of Muang District, NakhonRatchasima, Thailand, by cutting tips of about 30 cm from the youngest fully expanded leaves from several trees. All sample were dried at 50°C using a forced air oven prior to grinding through a 1.0 mm sieve.

Sample of each CT-containing plant leaves were used in the study. Additionally, control treatments (without CT effect) of each plant species were set by adding of polyethylene glycol (PEG) (CT inactivator, 160 mg/g DM of sample). This procedures were performed with 14 replicates in 2 separated runs.

Three matured rumen fistulated Saanen male goats fed with 2.5 % of body weight (% BW) DM/day containing dried-ground Pangola (*Digitaria eriantha*) and commercial concentrate (14 % of CP) (60:40) were used as donors of rumen fluid. Ruminant fluid and artificial saliva and *in vitro* gas production procedure were prepared and run according to Menke&Steingass (1988). The total gas produced were measured at 1, 2, 3, 4, 6, 8, 10, 12, 24 and 48 h of incubation, and net gas production values were corrected by subtracting blank values from the samples (Menke&Steingass, 1988). Cumulative gas production data were fitted to the model of Orskov& McDonald (1979) described as $Y = a + b(1 - e^{-ct})$; where Y represents the cumulative gas production at time t , a is the gas production from the immediately soluble fraction, b is the gas production from the insoluble fraction, c is the rate of gas production (/h) and $(a+b)$ is the potential gas production. At 4, 24 and 48 h of incubation, the methane (CH₄) volume was measured by absorbed produced CO₂ into the content by added 4.0 mL of 10 M NaOH, therefore, the gas volume remaining in the syringe considered to be CH₄ (Fievez et al., 2005).

Statistical analysis

Data of *in vitro* gas production were analyzed using the PROC GLM of SAS (1998). Multiple comparisons among treatment means were identified using Duncan's New Multiple Range Test (Steel & Torrie, 1980). Mean differences were considered significant at $P < 0.05$.

Results and Discussion

In vitro gas productions

The kinetics of gas production models and gas production at 4, 24 and 48 h of incubations of each treatments are presented in Table 1. Source of CT-containing plant species had no effect on the gas production from the immediately soluble fraction (a) ($P > 0.05$). While the gas production from the insoluble fractions (b), the gas production rate (c) and the potential extent of gas production ($a+b$) were affected by plant species ($P < 0.001$). However, there was found that, only tannins in SN leaves had effect on b value (99.4 and 125.9 in SN and SN+PEG group, respectively).

Plant species had effect on total gas (TG) and methane (CH₄) productions at 4, 24 and 48 h of incubations ($P < 0.01$) but had not effect on CH₄/TG ratio ($P > 0.05$). However, tannins in each plant species did not effected on total gas and methane productions and CH₄/TG ratio. Except for methane production at 48 h of incubation, tannins in SN had inhibitory effect on methane production at 48 h of incubation.

Different gas kinetics and gas productions among plant species probably due to different of their chemical compositions. Cassava leaves showed a highest gas productions compared to leucaena and Siam neem leaves. Inhibitory effect of CT in Siam neem leaves on the gas production from the insoluble fractions and methane production at 48 h of incubation may due to its high concentration of CT (5.0% of DM), whereas CT in cassava and leucaena leaves (2.9 and 1.2% of DM, respectively) had not effect on gas kinetics and gas productions may due to their low concentrations.

Table 1. Effects of CTs of some tropical plant leaves on *in vitro* gas and methane productions

	Treatments ¹						SEM	P-value
	CV	LN	SN	CV+PEG	LN+PEG	SN+PEG		
Gas kinetics ²								
<i>a</i>	-8.2 ^{ab}	1.2 ^a	1.2 ^a	-11.0 ^b	-4.7 ^{ab}	-8.6 ^{ab}	1.51	0.0956
<i>b</i>	158.8 ^a	118.2 ^b	99.4 ^c	173.4 ^a	133.0 ^b	125.9 ^b	2.69	0.0001
<i>c</i>	0.15 ^a	0.08 ^b	0.07 ^b	0.16 ^a	0.07 ^b	0.09 ^b	0.003	0.0001
<i>a+b</i>	150.6 ^{ab}	119.5 ^{cd}	100.7 ^d	162.4 ^a	128.3 ^{bc}	117.2 ^{cd}	3.56	0.0001
Total gas production (mL/g DM of substrate)								
4-h	60.7 ^a	34.8 ^b	27.5 ^b	72.7 ^a	38.6 ^b	34.5 ^b	2.08	0.0001
24-h	143.6 ^a	100.3 ^b	78.6 ^b	155.6 ^a	105.0 ^b	103.1 ^b	3.80	0.0001
48-h	158.8 ^{ab}	123.8 ^{bcd}	92.5 ^d	170.0 ^a	135.6 ^{abc}	106.9 ^{cd}	5.39	0.0045
Methane (CH ₄) production (mL/g DM of substrate)								
4-h	14.9 ^a	9.8 ^{ab}	6.1 ^b	14.6 ^a	5.2 ^b	4.7 ^b	0.96	0.0100
24-h	27.9 ^a	13.8 ^{bc}	9.3 ^c	27.5 ^a	15.5 ^b	11.9 ^{bc}	0.69	0.0001
48-h	32.1 ^{ab}	21.7 ^{bc}	13.5 ^c	41.5 ^a	26.0 ^{bc}	27.5 ^b	1.74	0.0052
CH ₄ /total gas ratio (v/v)								
4-h	0.28	0.43	0.85	0.22	0.15	0.18	0.09	0.2868
24-h	0.20	0.15	0.14	0.17	0.18	0.12	0.01	0.5149
48-h	0.21	0.18	0.16	0.25	0.19	0.25	0.02	0.4203

^{a,b,c,d}Within rows, values followed by different letters are significantly different ($P < 0.05$)

¹CV: Cassava leaves, LN: Leucaena leaves, SN: Siamese neem leaves, PEG: polyethylene glycol (as the tannins inactivator).

²*a*: the gas production from the immediately soluble fraction, *b*: the gas production from the insoluble fraction, *c*: the gas production rate constant, *a+b*: the potential extent of gas production.

Acknowledgements

The authors are grateful to Institute of Research and Development, Suranaree University of Technology, Thailand, for the financial support. We also thank The Center for Scientific and Technological Equipment, Suranaree University of Technology for supporting research facilities.

References

- Cherdthong A, Wanapat M (2010) Development of urea products as rumen slow-release feed on ruminant production: A review. *Australian Journal of Basic and Applied Sciences* **4**, 2232-2241.
- FAO/IAEA (2000) 'Quantification of tannins in tree foliage: A laboratory manual for the FAO/IAEA Co-ordinated Research Project on Use of Nuclear and Related Techniques to Develop Simple Tannin Assays for Predicting and Improving the Safety and Efficiency of Feeding Ruminants on Tanniniferous Tree Foliage.' (IAEA: Vienna)

- Fievez V, Babayemi OJ, Demeyer D (2005) Estimation of direct and indirect gas production in syringes: A tool to estimate short chain fatty acid production that requires minimal laboratory facilities. *Animal Feed Science and Technology***123-124**, 197-210.
- Frutos P, Hervas G, Giraldez FJ, Mantecon AR (2004) Review: Tannins and ruminant nutrition. *Spanish Journal of Agricultural Research***2**, 191–202.
- Menke KH, Steingass H (1988) Estimation of the energetic feed value obtained by chemical analysis and in vitro gas production using rumen fluid. *Animal Research and Development* **28**, 7-55.
- Ngamsaeng A, Wanapat M, Khampa S (2006) Evaluation of local tropical plants by in vitro rumen fermentation and their effects on fermentation end-products. *Pakistan Journal of Nutrition* **5**, 414-418.
- Ørskov ER, McDonald I (1979) The estimation of protein degradability in the rumen from incubation measurements weighted according to the rate of passage. *The Journal of Agricultural Science***92**, 499-503.
- Osborne NJT, McNeill DM (2001) Characterisation of *Leucaena* condensed tannins by size and protein precipitation capacity. *Journal of the Science of Food and Agriculture***81**, 1113-1119.
- Patra, A. K., and J. Saxena. 2011. Exploitation of dietary tannins to improve rumen metabolism and ruminant nutrition. *Journal of the Science of Food and Agriculture***91**, 24–37.
- SAS (1998) 'User's guide: Statistic.' (SAS Institute Inc.: New York)
- Steel RGD, Torrie JH (1980) 'Principles and procedures of statistics.' (McGraw Hill Book Co.: New York)
- Waghorn, G. C. 2008. Beneficial and detrimental effects of dietary condensed tannins for sustainable sheep and goat production – progress and challenges. *Animal Feed Science and Technology***147**, 116–139.
- Wanapat, M. 2003. Manipulation of cassava cultivation and utilization to improve protein to energy biomass for livestock feeding in the tropics. *Asian-Australasian Journal of Animal Science***16**, 463-472.

Dietary fat sources on growth performance and body composition in broiler chickens

Wongsuthavas, S¹., C. Yuangklang¹, K. Vasupen¹, W. Wongtrairat² & A.C. Beynen¹

¹Department of Agricultural Technology and Environment, Faculty of Science and Liberal Arts, Rajmangala University of Technology Isan 30000, ²Department of Electronic Engineering, Faculty of Engineering and Architecture, Rajmangala University of Technology Isan 30000

Abstract

This study was aimed dietary beef tallow, soybean oil and krabok oil on growth performance and chemical carcass composition. Growth performance was significantly improved ($P < 0.05$) by krabok oil (final body weight, average daily gain and feed conversion ratio). The diets containing soybean oil showed a significantly higher ($P < 0.01$) protein and ash content in the whole body. In contrast, fat content in whole carcass was reduced ($P < 0.01$). The results can indicated that krabok oil can be used as energy source of broiler chickens without a negative effect on growth performance.

Keywords: beef tallow, soybean oil, krabok oil, growth performance, body composition

Introduction

Composition of fatty acid in diets have been investigated that the polyunsaturated fatty acids (PUFA) supplementation to the diet at the expense of long-chain saturated fatty acids (SFA) reduces the amount of fat deposition in broiler chickens (Crespo and Esteve-Garcia, 2002a; Villaverde et al., 2005; Wongsuthavas et al., 2008). Possible mechanism could be that PUFA versus SFA are preferentially oxidized (Beynen and Katan, 1985) and yield ATP so that carbohydrates are shifted from the oxidative into the lipogenic pathway. This implies that consumption of Medium chain triglyceride (MCT) may increase fat oxidation. Consequently, the PUFA instead of SFA acids may lead to less deposition of body carcass fat. Similar to PUFA, saturated fatty acids with medium-chain length are also preferentially oxidized (Bach and Babayan, 1982). Thus, it could be presume that dietary MCT would decrease body fat deposition more than SFA diet.

Material and methods

Broiler chickens and diets

45 Arbor Acres broiler chicks at 7-day-old were used. They were randomly allocated to three groups of 15 birds each (individual cages). Feed and water were provided *ad libitum*. The experimental diets contained either tallow, soybean oil and krabok oil. The diet components consisted of (g/kg diet); tapioca starch, 460.2; soybean meal, 410.5; rice bran hulls, 40; dicalcium phosphate, 38.7; D,L-methionine, 3.0; L-lysine, 2.5; sodium chloride, 5.1; premixed, 1.0. Feed intake and body weight were measured weekly. At 28 days of age, five birds per treatment were randomly chosen for body composition (water, crude fat, crude protein and ash).

Statistical analysis

Data were subjected to analysis of variance in completely randomized design (Steel and Torrie, 1980) by using the Microsoft Excel program (Windows xp®).

Results

Growth performance and body composition

Growth performance was not significantly affected by the dietary fat sources. Moreover, ADFI and ADG was lowered by feeding soybean oil ($P < 0.01$). A decrease in ADFI in broiler chickens fed diet high in PUFA has been reported earlier (Atteh et al., 1983; Sklan and Ayal, 1989; Huang et al., 1990), but the effect does not appear to be consistent (Skrivan et al., 2000; Wongsuthavas et al., 2008). A lower feed intake when birds fed PUFA-rich could be explained by the higher digestibility of the fat component (Corino et al., 1980; Brue and Latshaw, 1985), implying a higher dietary content of metabolizable energy and less feed needed to meet the energy requirement as shown in Table 1.

Table 1 Effect of dietary fat source on growth performance, triacylglycerol and serum cholesterol concentration

Growth performance	Fat source			P-value
	Tallow	Soybean oil	Krabok oil	
Initial BW, g	150.03	150.03	149.83	0.9985
Final BW, g	802.00 ^b	750.60 ^b	839.47 ^a	0.0173
ADFI, g/b/d	59.61 ^a	49.67 ^b	58.37 ^a	0.0003
ADG, g/b/d	31.05 ^a	28.60 ^b	32.84 ^a	0.0119
FCR (g/g)	1.93	1.75	1.79	0.0799
Body composition of whole carcass, %				
Water	72.49	71.56	74.03	3.0034
Fat	14.112 ^a	12.709 ^b	14.027 ^a	0.0012
Protein	54.183 ^b	65.974 ^a	55.214 ^b	0.0001
Ash	9.014 ^b	10.838 ^a	9.0143 ^b	0.0010

^{ab} Means within the row with different superscripts differ significantly

In keeping with earlier studies (Keren-Zvi et al., 1990; Mossab et al., 2000; Wongsuthavas et al., 2008), soybean oil diet had significantly lower body fat deposition.

The composition of the whole carcass in terms of the contents of water had no significant differences ($P > 0.05$). However, the fat content of whole carcass was decreased in birds fed the diet rich in PUFA instead of SFA and MCT ($P < 0.01$). Moreover, protein, and ash contents were enhanced in PUFA diet, compared with SFA and MCT diets ($P < 0.01$) as shown in Table 1.

Conclusion

The feeding of krabok oil versus tallow did not reduce abdominal and carcass fat. The presence in dietary fat of fatty acids that are preferentially oxidized is not a determinant of abdominal fat deposition in broiler chickens. The data indicated that krabok oil can be used as energy source for broiler chickens without a negative effect on growth performance.

References

- Atteh, J.D., Leeson, S., and Julian, R.J.(1983) Effect of dietary level and types of fat on performance and mineral metabolism of broiler chicks. *Poultry Science*, 62, pp. 2043-2411
- Bach, A.C., and Babayan, V.K. (1982) Medium-chain triglycerides: an update. *The American Journal of Clinical Nutrition*, 36, pp. 950-962
- Beynen, A.C. and Katan., M.B. (1985) Why do polyunsaturated fatty acids lower serum cholesterol?. *American Journal of Clinical Nutrition* ,42, pp. 560-563
- Brue, N.R., and Latshaw, J.D. (1985) Energy utilization by the broiler chicken as affected by various fats and fat levels, *Poultry Science*, 64, pp.2119-2130
- Clarke, S.D., Romsos, D.R., and Leveille, G.A. (1976) Specific inhibition of hepatic fatty acid synthesis exerted by dietary linoleate and linolenate in essential fatty acid adequate rats, *Lipid*, 11, pp. 485-490
- Corino, C., Dellorto, V., and Pedron, O. (1980) Effects of acid composition of fats and oils on the nutritive efficiency of broiler feeds, *Review Zootechnology Veterinary*, 2,pp 94
- Crespo, N.; Esteve-Garcia, E. (2002) Dietary polyunsaturated fatty acids decrease fat deposition in separable fat depots but not in the remainder carcass, *Poultry Science*, 81,pp 512-518
- Huang, Z.B., Leibovitz, H., Lee, C.M., and Millar, R. (1990) Effects of dietary fish oil on ω 3 fatty acid level in chicken eggs and thigh flesh,*Journal of Agricultural and Food Chemistry*, 38,pp 743-747
- Keren-Zvi, S., Nir, I., Nitsan, Z., and Cahaner, A. (1990) Effect of dietary concentration of fat and energy on fat deposition in broilers divergently selected for high or low abdominal adipose tissue, *British Poultry Science*,31, pp507-516
- Mossab, A., Hallouis, J.M., and Lessire, M. (2000) Utilization of soybean oil and tallow in young turkeys compared with young chickens, *Poultry Science*, 79,pp1326-1331
- Sklan, D., Ayal, A. (1989) Effect of saturated fatty acids on growth, body fat and carcass quality in chicks, *British Poultry Science*, 30,pp 407-411
- Steel, R.G.D. and Torries, T.H. (1980) *Principle and Procedure of Statistic*. McGraw Hill Book Company, New York.
- Villaverde, C., Baucells, M.D., Cortinas, L., Hervera, M., Barroeta, A.C. (2005) Chemical composition and energy content of chickens in response to different levels of dietary polyunsaturated fatty acids,*Archives of Animal Nutrition*, 59,pp 281-292
- Wongsuthavas, S., Terapuntuwat, S., Wongsrikeaw, W., Katawatin, S., Yuangklang, C., and Beynen, A.C. (2008) Influence of amount and type of dietary fat on deposition, adipocyte count and iodine number of abdominal fat in broiler chickens, *Journal of Animal Physiology and Animal Nutrition*, 92(1), pp 92-98

Influence of rice straw treated on rumen fermentation and microbial population in swamp buffaloes

Ampapon, T. and Wanapat, M

Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

Abstract

Four rumen-fistulated swamp buffaloes with initial average liveweight 330 ± 20 kg, were used to study effect of rice straw treated on rumen fermentation and microbial protein synthesis in swamp buffaloes. Treatments were T1= treated rice straw, T2= urea 1% treated rice straw, T3= 1%urea+1%cassava ship powder treated rice straw and T4= 1%urea +1%cassava ship powder +0.1% yeast). All buffaloes were randomly allocated to receive diets according to a 4×4 Latin square design. The present results revealed that ammonia nitrogen were increased by rice straw treated with urea and yeast. Furthermore, microbial population especially proteolytic bacteria and cellulolytic bacteria were increased in buffaloes consuming treated rice straw especially with 1%urea + 1% cassava ship powder +0.1% yeast (T4). Therefore, this study could that treated rice straw with urea plus with cassava chip powder and yeast could improve rumen fermentation and changed to micorbial population of swamp buffaloes.

Keywords: rice straw, rumen fermentation, rumen ecology, swamp buffaloes

Introduction

Rice straw is an important crop residue practically used by farmers for ruminant feeding especially during the long dry season (Ørskov et al., 1999). Ruminants have the unique capacity to transform relatively low-quality dietary nitrogen (N) into high-quality animal proteins (i.e., meat and milk) (Schroeder and Titgemeyer, 2008). Relative to low nutritive value of rice straw in terms of protein (2 to 5%), fiber and lignin contents (NDF > 50%) and low digestibility (< 60%) (Wanapat et al., 1985), feeding rice straw only does not provide enough nutrients for optimum production requirement. Improving the utilization of low quality roughages could be by treatment with nitrogen sources, chemical and physical treatment (McDonald et al., 2002; Nguyen et al., 2012). However, the cost of urea treatment was remarkably expensive which resulted on higher cost of production.

Materials and methods

Four, rumen-fistulated swamp buffaloes with initial average liveweight of 330 ± 20 kg, were randomly allocated to receive diets according to a 4×4 Latin square design. The treatments were T1= treated rice straw, T2= urea 1% treated rice straw, T3= 1%urea+1%cassava ship powder treated rice straw and T4= 1%urea +1%cassava ship powder +0.1% yeast), respectively. Untreated rice straw was offered ad libitum. The experiment was conducted for four periods and each of the four periods lasted for 21 days in length. Feeds and fecal samples were collected by total collection

of each individual buffalo during the last 7 days at morning and afternoon feeding. Feeds, refusals and fecal samples were dried at 60°C and ground and analyzed using standard methods of AOAC (1995). At the last day of each sampling period, rumen fluid was collected at 0, 2, 4 and 6 h post morning feeding. Rumen fluid was immediately measured for pH and temperature using a portable pH meter. Rumen fluid samples were collected and analyzed for volatile fatty acid (VFA) by using high performance liquid chromatography (Chen et al., 1993). The last part was cultured for groups of bacteria using the roll-tube technique Hungate (1969). All data were subjected to ANOVA according to a 4 x 4 Latin square design using the General Linear Models (GLM) procedures of the Statistical Analysis System Institute (SAS, 1998).

Results and Discussion

There was no effect of urea, cassava chip powder and yeast treatment on the rumen pH and temperature. However, ruminal NH₃-N was increased by straw treatment. The average values of NH₃-N in this study were 2.4 to 6.7 mg/dl and were in the optimal range in ruminal fluid for microbial growth which was reported from 5.0 to 25.0 mg/dl and (Preston and Leng, 1987). Moreover, total ruminal VFA and propionic acid were significantly different. Ørskov et al. (1999) discussed that, when high fiber diets were offered, the VFAs in ruminal fermentation fluctuated from 65:25:10 to 70:20:10 (C2:C3:C4, in molar percentage) ratios. The results of VFAs in treated groups were similar and were higher (P<0.05) than those in the control group (untreated rice straw). Wanapat et al. (2009) also found the same result with the present study that when the animals were fed with urea or urea-lime treated rice straw would result in higher in total VFA and C3; which led to lower down the proportion of C2 to C3 ratio, if compared to untreated rice straw (Table 2) illustrates data on rumen microbes population. Especially proteolytic bacteria and cellulolytic bacteria were increased in buffaloes consuming treated rice straw especially with 1% urea + 1% cassava chip powder + 0.1% yeast (T4). According to Chen et al. (2008) reported that chemical treatments enhanced the nutritive value of rice straw through increasing the number of accessible sites of microbial attachment on the surface of the particles, increasing fibrolytic microbe quantity and hence, fibrolytic enzyme activities, and improving rumen fermentation characteristics. Therefore, this changed to studies could that treated rice straw with urea plus with cassava chip powder and yeast could improve rumen fermentation and changed to microbial population of swamp buffaloes.

Table 1. Effect of urea and yeast treated rice straw on rumen fermentation and volatile fatty acids concentration in swamp buffaloes.

Item	T1	T2	T3	T4	SEM
Rumen fermentation parameters					
pH	6.61	6.53	6.63	7.04	0.05
Temperature(°C)	38.81	38.72	38.64	38.70	0.003
NH ₃ -N ,mg/dl	2.44 ^a	5.71 ^b	6.24 ^b	6.72 ^b	0.003
Total VFA,mmol/l	64.32	92.47	86.66	114.92	22.85
Acetate (C2)	39.08	54.68	52.69	82.62	20.06
Propionate (C3)	14.46	22.62	19.37	18.72	2.84
Butyrate (C4)	10.76	14.98	14.60	13.57	1.71
C2:C3	2.85	3.07	3.51	4.88	0.82

Table 2. Effect of urea and yeast treated rice straw on microbial population in swamp buffaloes.

Item	T1	T2	T3	T4	SEM
Viable bacteria, CFU/ml by roll-tube technique					
Total viable bacteria $\times 10^9$	4.41	4.51	5.16	4.64	0.51
Amylolytic, $\times 10^7$	3.81	3.24	5.16	3.55	1.01
Proteolytic, $\times 10^8$	3.82 ^a	4.64 ^b	4.58 ^b	6.97 ^c	8.83
Cellulolytic, $\times 10^7$	5.52 ^a	6.94 ^b	7.81 ^b	9.33 ^c	3.11

Conclusions

Based on this study, it could be concluded that rice straw quality could be improved by treated with urea and yeast. More, Treated straw with urea and yeast could increase ammonia nitrogen, microbial population especially proteolytic bacteria and cellulolytic bacteria.

Acknowledgments

The authors would like to express their most sincere thanks to all who have assisted and supported the research in this study, particularly the Tropical Feed Resources Research and Development Center (TROFEC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University (KKU), Thailand for their kind facilitation and permission to use the research facilities.

References

- AOAC (1995) Official Method of Analysis, 16th ed. Animal Feeds: Association of Official Analytical Chemists, VA, USA.
- Ha, J. K., S. S. Lee, S. W. Kim, In K. Han, K. Ushida and K. -J. Cheng. 2001. Degradation of rice straw by rumen fungi and cellulolytic bacteria through mono-, co- or sequential- cultures. *Asian-Aust. J. Anim. Sci.* 14: 797-802.
- Hungate, R.E., 1969. A roll tube method for cultivation of strict anaerobes. In: Norris, J.R., Ribbons, D.W. (Eds.), *Methods in Microbiology*. Academic, New York, pp. 117– 313.
- Mapato, C, M. Wanapat and A. Cherdthong, 2010. Effects of urea treatment of straw and dietary level of vegetable oil on lactating dairy cows. *Trop. Anim. Health Prod.* 42:1635-1642.
- Ørskov, E.R., 1999. Supplement strategies for ruminants and management of feeding to maximize utilization of roughages. *Prev. Vet. Med.* 38, 179–185.
- Shen, H. S., F. Sundstøl, E. R. Eng and L. O. Eik, 1999. Studies on untreated and urea-treated rice straw from three cultivation seasons: 3. Histological investigations by light and scanning electron microscopy. *Anim. Feed Sci. Technol.* 80: 151-159.
- Steel, R. G. D. and J. H. Torrie, 1980. *Principles and Procedures of Statistics*. McGraw Hill Book Co, NY.
- Schroeder, G.F. and E.C. Titgemeyer, 2008. Interaction between protein and energy supply on protein utilization in growing cattle: a review. *Livest. Sci.* 114, 1–10
- SAS, 1996. *SAS/STAT User's Guide: Statistics, Version 6.12*. Edition. SAS Inc, Cary, NC, USA.
- Wanapat, M., C. Wachirapakorn, S. Chanthai and K. Sommart, 1992. Utilization of cassava leaf (*Manihot esculenta*, Crantz) in concentrate mixtures for swamp buffaloes in Thailand. In: *Proc. Feeding Strategies for Improving Ruminant Productivity in Areas of Fluctuating Nutrient Supply* Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, Vienna, Austria, March 30-April 3, pp. 33-38
- Wanapat, M., T.H. Sundstøl, and T.H., Garmo, 1985. A comparison of alkali treatment methods to improve the nutritive value of straw. I. Digestibility and metabolizability. *Anim. Feed Sci. Technol.* 12, 295–309
- Wanapat, M. S. Polyorach, K. Boonop, C. Mapato, and A. Cherdthong, 2009: Effects of treating rice straw with urea or urea and calcium hydroxide upon intake, digestibility, rumen fermentation and milk yield of dairy cows. *Livestock Science* 125, 238–243.

A study on nutrient intake and digestibility, rumen environment and nitrogen retention of sheep fed different levels of ensiled water hyacinth in diets

Nguyen Van Thu and Nguyen Thi Kim Dong

Can Tho University, Vietnam

Abstract

Four growing sheep (19.8 ± 0.43 kg) were allocated in a 4x4 Latin square design with 4 treatments including Para grass (EWH0), replacement of 15% Para grass by ensilaged water hyacinth (EWH15), replacement of 30% Para grass by ensilaged water hyacinth (EWH30), replacement of 45% Para grass by ensilaged water hyacinth (EWH45). This study aimed to evaluate effects of replacement of ensilaged water hyacinth (*Eichhornia crassipes*) to Para grass (*Brachiaria mutica*) in the diets (DM basis) on feed intake, rumen parameters, nutrient digestibility and nitrogen retention of growing sheep. There was a supplementation of coconut meal, soybean cake and urea to adjust the CP content of diets being 17%. Each experimental period was 14 days including 7 days for adaptation and 7 days for sample collecting. The conclusion was that EWH could be used to feed growing sheep without adverse effects on rumen parameters, and the replacement level of 30% EWH to Para grass in diet gave a better result.

Keyword: lamb, water hyacinth, supplements, diets, Para grass, replacement

Corresponding author: nvthu@ctu.edu.vn

Introduction

Water hyacinth could be used as feed resources in the humid and tropical areas where feed for ruminants is scarce in rainy season. In some countries, the water hyacinth is used as fodder for cows, goats, sheep, pig and chickens, however, it is as low economical forage because it contents high moisture, rapid deterioration and spoilage. Addition, fresh water hyacinth is unpalatable because it contents prickly crystals (Gohl, 1994). These limitations of using fresh water hyacinth for feeding ruminant may be solutions by ensiled methods. The ensiled water hyacinth was used and accepted palatable on sheep and goat feeding. The ensiled water hyacinth could rather improve on growing performance of local cattle in Mekong Delta in Vietnam when it was used to replace rice straw in diets. In recent years in Vietnam, the sheep meat demand for food consumption has been increased because it contents high nutrition, good smell and delicious. Thus, sheep has been more concerned to development in many provinces of Mekong Delta in Vietnam, where water hyacinth is available with a large biomass. Therefore an evaluation of effects of replacement of ensilaged water hyacinth (*Eichhornia crassipes*) to grass in the diets (DM basis) on feed intake, rumen parameters, nutrient digestibility and nitrogen retention of growing sheep were necessary, then recommendations could be given for farmers' practices.

Materials and methods

Four growing sheep (19.8 ± 0.43 kg) were allocated in a 4x4 Latin square design with 4 treatments including Para grass (EWH0), replacement of 15% Para grass by ensilaged water hyacinth

(EWH15), replacement of 30% Para grass by ensilaged water hyacinth (EWH30), replacement of 45% Para grass by ensilaged water hyacinth (EWH45). Para grass was fed ad-lib and supplemented with coconut meal, soybean cake and urea to adjust the CP content of diets of 17%. Each experimental period was 14 days including 7 days for adaptation and 7 days for sampling. Dry matter intake was 3.2% body live weight. Feed offered and refused, feces and urine were collected daily during sample collecting periods. Rumen fluid was taken by oesophagus gutter in order to measure N-NH₃ and volatile fatty acid at before and 3 hours after feeding. Water hyacinth was collected and eliminated the roots and then wilted under sunshine. When the DM of water hyacinth reached around 12%, it was used for making silage with molasses (3.5 kg molasses for 100 kg of fresh water hyacinth) in a plastic bag of 50kg. The silage was used for feeding sheep from day 7 to day 14. Dry matter (DM), organic matter (OM), crude protein (CP) and total ash (Ash) of samples were determined according to OAO (1990). Neutral detergent fiber (NDF) was analyzed by Van Soest *et al* methods (1991). Metabolized energy of diets was calculated described by Bruinenberg *et al.* (2002). Apparent nutrient digestibility of DM, OM, CP and NDF was determined by Mc Donald *et al* (1995) and VFAs analysis following method described by Barnett and Reid method (1957). The data were analyzed preliminary by Microsoft Excel software and analyzed of variance (ANOVA) using the General Linear Model (GLM) procedure of Minitab program (Minitab, 2000).

Results and discussion

Table 1. Chemical composition of feed ingredients used in experiment (% DM)

Ingredients	DM	OM	CP	NDF	Ash
Ensilaged water hyacinth (EWH)	19.0	84.2	11.2	54.2	15.8
Para grass (PG)	19.4	87.0	9.40	68.1	12.3
Soybean cake (SC)	87.3	83.9	42.1	23.4	16.1
Coconut meal (CM)	87.5	94.2	16.2	56.6	5.80
Urea	-	-	288	-	-

The chemical composition of feed used in the experiment was showed in Table 1. The DM, OM, CP and NDF of EWH were 19.0, 84.2, 11.2 and 54.2 %, respectively. The CP content of EWH was higher than that of Para grass, while its NDF content was lower that the Para grass. The replacement of Para grass by EWH was suitable due to the DM content was similar for both roughages. The Soybean cake, Coconut meal and urea were used to supply crude protein in the diets and to adjust the CP intake of sheep as plan proposed.

Table 2. Feed and nutrients intakes of sheep in the experiment

Items, g/day	Treatments				P	± SE
	EWH0	EWH15	EWH30	EWH45		
EWH	0 ^a	80.5 ^b	164 ^c	243 ^d	0.001	11.8
Para grass	566 ^a	500 ^b	385 ^c	310 ^d	0.001	11.8
DM	675	690	658	662	0.328	16.9
OM	576	588	560	558	0.307	16.4
CP	120	118	115	111	0.132	3.29
NDF	426 ^a	425 ^a	393 ^b	390 ^b	0.010	8.796
ME* (MJ/day)	6.05	6.10	5.83	5.89	0.854	0.370

The DM and CP intake were not significantly different ($P < 0.05$) among the treatments, however, the NDF intake was significantly higher for the EWH0 and EWH15 as compared to EWH30 and EWH45. It seemed to be the replacement of EWH at level 15% had stimulated appetite of sheep

in diet. It led to DM and OM intakes of EWH15 diet were slightly higher than the others (690 and 588g/sheep/day), respectively. In study of Vo Duy Thanh (2008) using EWH replacing Para grass at 0, 15, 30, 45% levels (DM basis) for growing cattle indicated that cattle could consume at maximum EWH level of 800 g/day.

The pH values of rumen fluid at before and after 3-hours feeding among treatments were not significantly different ($P>0.05$). However, pH value at before feeding was higher than this value at 3 hours after feeding in general. It could be explained that EWH was acidic feed. Vo Duy Thanh (2008) reported that the N-NH₃ and VFAs of rumen fluid at before feeding of cattle fed EWH were 11.6-11.9 mg/100ml and 71.7-75.2 mmole/liter, respectively. Then 3-hours after feeding, those values changed to 17.5-18.0 mg/100ml and 80.6-81.4 mmole/liter, respectively.

The nutrient digestibility (DM, OM, CP, NDF) were not significantly different ($P>0.05$) among treatments and ranged from 66.4 to 67.3%. Nitrogen retention was not significantly different ($P>0.05$) among the treatment, however the highest value was numerically for the EWH30 treatment (0.809 g/kgW^{0.75}). The daily weight gain was not significantly different ($P>0.05$) among the treatments and it was 52.5, 48.1, 57.3 and 38.1 g for the EWH0, EWH15, EWH30 and EWH45, respectively. The results in the present study indicated that EWH could be used for feeding sheep with the replacement level of 30% for the grass.

Conclusions

The conclusion was that ensilaged water hyacinth could be used to feed growing sheep without adverse effects on rumen parameters and the replacement level of 30% to Para grass in diet gave good results in term of growth performance and utilization of water hyacinth as a feed resource.

References

- Barnett A J G and Reid R L 1957 Studies on the production of volatile fatty acids from grass by rumen liquor in an artificial rumen. The volatile fatty acid production from grass. *Journal of Agricultural Science*, 48:315–321.
- Bruinenberg M H, Valk H, Korevaar H and Struik P C 2002 Factors affecting digestibility of temperate forages from seminatural grasslands. Wageningen, and Plant Research International, Wageningen, The Netherlands
- Gohl, B 1994 Tropical Feed. FAO Animal Production and Health Series
- McDonald P, Edwards R A, Greenhagh J F D and Morgan C A 1998 Digestibility. Evaluation of food. In *Animal Nutrition*. 5th Ed. Addison Wesley Longman, UK, pp. 221-237
- Van Soest PJ, Robertson J B and Lewis B A 1991 Method for dietary fiber, neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition. *Journal of Dairy Science* 74: 3583-3597
- Vo Duy Thanh, 2008 Effects of rice straw replacement by ensilage water hyacinth on rumen parameters, nutrient digestibility and nitrogen retention of local growing cattle. Bachelor thesis on Animal Science, Can Tho University

Effect of dried cassava chips in growing rabbit diets on meat performance and economic returns

Nguyen Thi Kim Dong and Nguyen Van Thu

Cantho University, Cantho City, Vietnam

Abstract

Sixty crossbred rabbits (local x New Zealand) with average initial live weight of 735 ± 4.64 g and 8 weeks of age were allocated in a completely randomized design with 5 treatments and 3 replications and were fed 5 levels of dried cassava chips (0, 10, 20, 30 and 40 g/rabbit/day) as a supplement to Para grass fed *ad libitum*. Increasing the offer level of dried cassava chips in a basal diet of Para grass fed to growing rabbits led to linear increases in total DM intake, live weight gain, coefficients of apparent digestibility and N retention. There were positive linear relationships between coefficients of apparent DM digestibility and live weight gain and N retention. It was proposed that the determinant of rabbit growth rate in forage-based diets is the overall apparent digestibility of the diet rather than the composition of the diet in terms of the relative proportions of soluble and structural carbohydrates.

Key words: carbohydrates, digestibility, N balance, Para grass

Corresponding author: ntkdong@ctu.edu.vn

Introduction

Rabbits are popularly raised in Vietnam, particularly in the Mekong delta provinces and contribute to improving the nutrition and economy of famer smallholders, both as a source of animal protein as well as a source of extra income. Rabbit meat has been also considered as good meat for high content of protein, low fat and low cholesterol levels (Owen, 1992). In the Mekong delta of Vietnam feeding of rabbits is mainly based on natural grasses and agro-industrial by products (Nguyen Thi Kim Dong & Nguyen Van Thu, 2009) so the cost for rabbit production is lower than for the other animal species ones. This study aimed to evaluate potential benefits from supplementing Para grass with dried cassava chips in diets of growing rabbits.

Materials and methods

For this feeding trial, sixty crossbred rabbits (local x improved breeds) at 8 weeks of age with average initial weight around 735g, were arranged in a completely randomized design with 5 treatments and 3 replications. Four rabbits balanced for sex were the experimental unit. The treatments were dried cassava chips (DC) supplemented to the Para grass (PG) basal diet at levels of 0 (DC0), 10 (DC10), 20 (DC20), 30 (DC30) and 40g (DC40) / day/ animal. Soybean waste was offered at 150g/day/animal and extracted soybean meal was fed at 7-11g/day/animal for adjustment of the same protein level of 11.5g/day/rabbit. The experimental period lasted 9 weeks.

Para grass was collected daily in the areas surrounding the farm. Soybean waste was bought daily from the soybean milk factory in the city; cassava chips and extracted soybean meal were bought on one occasion from an animal feed shop for using throughout the trials.

The measurements taken were intakes of dry matter (DM), organic matter (OM), ash, neutral detergent fiber (NDF), daily weight gain, feed conversion ratio, economic returns, nutrient digestibility and nitrogen retention. Chemical analyses of DM, OM, CP, EE, CF, NDF and Ash followed the procedure of AOAC (1990) and Van Soest et al. (1991). The data were analyzed using the General Linear Model (GLM) option in the ANOVA program of the Minitab software and comparison of differences between 2 treatments was done by Tukey test of Minitab (2000).

Results and discussion

Chemical composition of feeds of the experiment was presented in Table 1.

Table 1. Chemical composition of feeds (% in DM except for DM which is on fresh basis, and ME) in the feeding trial.

	DM	OM	CP	EE	CF	NDF	Ash	ME MJ/kg
Para grass	17.4	89.6	12.3	5.09	28.9	67.1	11.2	8.72
Dried cassava chips	94.3	97.1	2.70	1.59	3.39	15.6	3.09	13.4
Soya waste	12.0	95.3	21.3	15.4	3.50	35.0	4.96	13.1
Extracted soybean	87.9	90.1	42.8	3.22	3.70	27.4	11.3	12.4

Table 2. Daily intakes of feeds (g DM/animal) and fed components (g/animal) of rabbits in the feeding trial.

Item	Treatment					SEM/P
	DC0	DC10	DC20	DC30	DC40	
DM	53.4 ^a	55.2 ^a	68.2 ^b	68.5 ^b	79.9 ^c	1.43/0.001
OM	48.7 ^a	51.0 ^a	63.4 ^b	64.3 ^b	75.3 ^c	1.33/0.001
CP	11.0	10.4	11.2	10.3	10.6	0.26/0.140
EE	4.02 ^{ab}	3.92 ^a	4.47 ^{bc}	4.18 ^{abc}	4.55 ^c	0.11/0.008
NDF	28.0 ^a	25.1 ^{ab}	27.8 ^{ab}	24.7 ^b	27.7 ^{ab}	0.69/0.012
ME, MJ/animal	0.56 ^a	0.62 ^a	0.79 ^b	0.82 ^b	0.96 ^c	0.16/0.001

Intake of Para grass declined as the offer level of cassava chips was increased. Actual intake of cassava chips was slightly less than the offer level, but was strongly and positively related to total DM intake and growth rate (Table 2). Strangely, there was no relationship between cassava chip intake and feed conversion. Under the conditions in Vietnam, economic benefits were increased by almost 70% by supplementing Para grass with cassava chips (Table 3). The reason may be that the response to supplementation with a starch-rich carbohydrate is more a function of

the effect on the digestibility of the diet, than of the nature of the carbohydrate. In this respect, the digestibility of Para grass is considerably inferior to that of water spinach (Nguyen Thi Kim Dong et al., 2006).

Table 3. Mean values for changes in live weight, feed conversion and economic return.

Item	Treatment					SEM/P
	DC0	DC10	DC20	DC30	DC40	
Initial weight, g	737	735	738	727	738	5.53/0.567
Final weight, g	1755 ^a	1848 ^{ab}	2047 ^{abc}	2083 ^{bc}	2255 ^c	67.5/0.003
Daily gain, g	16.2 ^a	17.7 ^{ab}	20.8 ^{abc}	21.5 ^{bc}	24.1 ^c	1.09/0.002
FCR	3.33	3.13	3.28	3.18	3.35	0.14/0.761
Total cost, USD	2.85	2.94	3.14	3.23	3.38	-
Total income, USD	3.67	3.87	4.28	4.36	4.72	-
Net income, USD	0.82	0.93	1.14	1.13	1.34	-

Cassava chip supplementation had no effect on the proportions of total lean meat and thigh lean meat in the carcass but increased the meat: bone ratio. When corrected for final live weight, the weight and length of the caecum was decreased by cassava chip supplementation, presumably reflecting the reduction in dietary fiber as cassava chips replaced Para grass.

Conclusions

Increasing the offer level of dried cassava chips in a basal diet of Para grass fed to growing rabbits led to linear increases in total DM intake, live weight gain and coefficients of apparent digestibility. It is proposed that the determinant of rabbit growth rate in forage-based diets is the overall apparent digestibility of the diet rather than the composition of the diet in terms of the relative proportions of soluble and structural carbohydrates.

References

- Hongthong Phimmasan, Siton Kongvongxay, Chhay Ty and Preston, T. R., 2004. Water spinach (*Ipomoea aquatica*) and Stylo 184 (*Stylosanthes guianensis* CIAT 184) as basal diets for growing rabbits. *Livestock Research for Rural Development*. Vol. 16, Art. #34.
- Maertens, L., Peter, J. M., Villamide, M., Cervera, C., Gidenne, T. and Xiccato, G., 2002. Nutritive value of raw materials for rabbit. *World Rabbit Science* 10 (4), pp. 157-166.
- Nguyen Thi Kim Dong, Nguyen Van Thu, Ogle, R B and Preston, T R., 2006. Effect of supplementation level of water spinach leaves in diets based on Para grass on intake, nutrient utilization, growth rate and economic returns of crossbred rabbits in the Mekong Delta of Vietnam. from <http://www.mekarn.org/proprf/kimd2.htm>

- Nguyen Thi Kim Dong and Nguyen Van Thu 2009. The use of agro-industrial by products for raising crossbred rabbits in the Mekong delta of Vietnam. Conference on Asian Rabbit Production Development. Sep 10-11 2009. Changchun, China. pp: 156.
- Owen, J. B., 1992. Genetic aspects of appetite and food choice by animals. *J. Agric. Sci.* 119: 151-155.
- Van Soest P. J., Robertson, J. B. and Lewis, B. A., 1991. Symposium: Carbohydrate methodology, metabolism and nutritional implications in dairy cattle: methods for dietary fiber, and non starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74: 3585-3597.

Effects of wet soya milk waste supplementation on feed intake and growth performance of goats fed corn stubble silage

Tatsapong, P., R. Thebsawad & S. Hanpok

Department of Agricultural Science, Faculty of Agriculture, Natural Resources and Environment, Naresuan University, Phitsanulok, 65000, Thailand

Abstract

This experiment was aimed to study the effects of soya milk waste on feed intake and growth rates of post weaning goat were studied. Fifteen Boer x Saanen crossbred male goats, (4-6 month of age and average live weight of 19.4 ± 1.4 kg) were used into three treatment diets under a completely randomized design (CRD) for 60 days study period. All animals were kept individually pen with free access water and mineral block. The goats fed ad *libitum* with corn stubble silage as roughage source. Three dietary treatments were commercial concentrate pellet diet (400 g), wet soya milk waste (800 g) and mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g), respectively. The results showed that feed intake, feed efficiency and body weight gain of goats were significant ($P < 0.05$) different among the diets. Dry matter (DM), organic matter (OM), crude protein (CP), gross energy (GE), neutral detergent fibre (NDF) intake of goat fed commercial concentrate pellet diet were significantly higher than those of goat fed other diet. Average daily gain of goat fed commercial diet was higher ($P < 0.05$) than those of goat fed soya milk waste diet and mixed commercial diet respectively. Feed efficiency was significantly higher in the soya milk waste diet.

Keywords: Soya milk waste, Goat, Feed intake, Saanen crossbred, boer crossbred

Introduction

In Thailand, soybean is made into various foods such as tofu, soymilk, soymilk powder, soy sauce, soy flour, dried tofu. Soya milk residue (waste), waste from soybean milk industry is high in moisture and spoil very quickly in room temperature. Soya milk waste is often considered as waste, which is mostly dumped and burned, and is a potential environmental problem because it is highly susceptible to putrefaction (Rahman et al., 2015; Li et al., 2013). Li et al. (2013) reported that soya milk waste is rich in protein, fiber, fat and trace elements, and is alternative sources of energy and protein for ruminants (Rahman et al., 2014a). The research regarding the use of soya milk waste as ruminant feed on the growth performance of goats has been limited. Although the goat is considered superior to other ruminant species in its utilization of poor quality, high fiber forages and household waste for its body maintenance and production. Therefore, this study investigated the effects of supplementing soya waste on intake, digestibility and growth rate response of growing goats.

Materials and Methods

Fifteen crossbred (Boer x Saanen) male goats, with 19.4 ± 1.4 kg initial body weight (BW), and approximately 4 to 6 months old, were used into three treatment diets under a completely randomized design (CRD) for 60 days study period. All animals were kept individually pens (1x1.2 m) with free access water and mineral block. The goats fed ad *libitum* with corn stubble silage as

roughage source. Three dietary treatments were commercial concentrate pellet diet (400 g), wet soya milk waste (800 g) and mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g), respectively. Soya milk waste was supplied by a market women from a local soybean milk processing (home made) at every day, It was contained 315 g CP/kg DM and 11.3 % DM. The commercial concentrate pellet and corn stubble silage were contained 15 and 9 % CP, respectively. Feeds were offered twice a day at 0800 and 1600 h. Amounts of feed offered and refused were recorded daily to estimated feed intake. Subsamples of faeces, feed offered and residues were taken weekly for dry matter determination and dried samples were ground through 1 mm screen and then analyzed for DM, ash, CP and EE (AOAC, 1990) and fiber fraction as described by Van Soest et al. (1991). After the completion of the 60-d feeding trial, a digestibility trial was carried out for 7 d using the total faeces collection method. The body weight of the goat was recorded every 15 days. The data were analysed using the General Linear Model procedure of SPSS (version 17.0).

Results and Discussion

Nutrients intake and digestibility of goats are given in Table 1, showing the goats on the soya waste group had lower intake of all nutrients than those fed commercial pellet. It was similarly to those reported by Rahman et al. (2015; 2014a), who found that supplementation of soya waste in the diet was decreased dry matter intake of goats. This might be due to the higher content of soya waste compared with the commercial pellet diet (89 and 11.3%, respectively). High moisture content of feeds increases the bulkiness of diets, and is negatively related to the capacity of the rumen (Rahman et al., 2014b). DM digestibility was highest on the commercial pellet diet mainly because of higher values for DM intake. While the OM digestibility was highest on soya waste diet may be attributed to increase feed efficiency (Table 2) and utilization of nutrient, particularly soya protein (Rahman et al., 2015).

Table 1. Nutrients intake and digestibility by goats fed soya milk waste and commercial pellet

Parameters	Dietary treatment			SEM	<i>P</i> -value
	T1	T2	T3		
Dry matter intake (DMI)					
Roughage (g/day)	290.07	332.20	324.69	13.87	0.113
Total DMI (g/kgBW ^{0.75})	64.33 ^a	47.06 ^c	54.59 ^b	1.95	0.000
Total DMI (% BW)	2.98 ^a	2.23 ^c	2.52 ^b	0.11	0.001
Nutrients intake (g/day)					
Dry matter (DM)	644.32 ^a	443.15 ^c	549.91 ^b	11.53	0.000
Organic matter (OM)	619.60 ^a	416.02 ^c	521.34 ^b	11.47	0.000
Crude protein (CP)	80.72 ^a	66.21 ^c	75.04 ^b	0.98	0.000
Neutral detergent fiber	312.43 ^a	233.11 ^c	286.70 ^b	8.91	0.000
Acid detergent fiber	159.95 ^a	120.99 ^c	141.84 ^b	8.12	0.018
Gross energy (kcal)	2,321 ^a	1,596 ^c	1,975 ^b	47.67	0.000
Nutrients digestibility (g/day)					
Dry matter (DM)	427.88 ^a	283.31 ^c	359.04 ^b	12.77	0.000
Organic matter (OM)	135.01 ^b	237.05 ^a	224.70 ^a	12.85	0.000
Crude protein (CP)	48.46	44.16	47.01	1.66	0.28
Neutral detergent fiber	182.14	148.86	171.16	9.27	0.07
Acid detergent fiber	60.61	58.39	57.32	8.31	0.96
Digestible energy (Mcal)	1.52 ^a	0.91 ^c	1.28 ^b	0.05	0.000

T1=commercial concentrate pellet diet (400 g); T2= wet soya milk waste (800 g); T3= mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g)

The means within rows with different letters (a,b,c) differ significantly ($p < 0.05$).

Table 2. Average daily gain and feed conversion ratio (FCR) in goat fed soya milk waste and commercial pellet

Parameters	Dietary treatment			SEM	<i>P</i> -value
	T1	T2	T3		
Initial weight (kg)	19.5	18.6	20.2	0.59	0.201
Final weight (kg)	23.9	21.1	23.7	0.89	0.084
Body weight change (kg)	4.4	2.5	3.5	0.64	0.149
Average dairy gain (g/day)	85.42 ^a	41.67 ^c	58.33 ^b	7.96	0.007
Feed efficiency	0.15 ^a	0.21 ^c	0.16 ^b	0.004	0.000
Feed conversion ratio	8.08	11.18	10.27	1.34	0.284

T1=commercial concentrate pellet diet (400 g);T2= wet soya milk waste (800 g); T3= mixed commercial concentrate pellet diet and wet soya milk waste (200 g : 400 g)

The means within rows with different letters (a,b,c) differ significantly ($p < 0.05$).

CP and fiber fraction digestibility were not different among dietary treatments. This might be attributed to the high nutritional characteristics of soya waste, and is positively related to the activity of rumen microbe, and lead to similar body weight change of goats (Rahman et al., 2014b). However, Average daily gain of goat was higher in commercial pellet diet ($P < 0.05$) compared with other treatments (Table 2) and this indicates greater intake of DM and CP than those of goat fed soya milk waste diet and mixed commercial diet respectively (Table 1). The goat fed soya milk waste diet showed lowest growth performance (41.6 g/day) throughout the trial period, which indicate that diets had nutrient content under the threshold level for production requirement of growing goats. This may be due to the low dry matter content (Li et al., 2013). The conclusion, supplementation of locally wet soya milk waste was decreased DM intake and BW gain of goat, and increased feed efficiency.

Acknowledgements

To the Division of Academic service and Asset Management, Naresuan University for financial support.

References

- AOAC. 1990. Official methods of analysis of the association of Official analytical chemists (AOAC). 15th ed. AOAC International, Arlington, Virginia, USA.
- Li S., D. Zhu, K. Li, Y. Yang, Z. Lei, and Z. Zhang, 2013. Soybean curd residue: Composition, utilization, and related limiting factors. ISRN Industrial Engineering, Hindawi publishing corporation. Available online at : <http://dx.doi.org/10.1155/2013/423590>.
- Rahman, M.M, M.R. Rahman, T. Nakagawa, R.B. Abdullah, W.E. Wan Khadijah, and R. Akashi, 2015. Effects of wet soya waste supplementation on the intake, growth and reproduction of goats fed Napier grass. *Anim. Feed Sci. Technol.* 199: 104-112.
- Rahman, M.M, R.B. Abdullah, W.E. Wan Khadijah, T. Nakagawa and R. Akashi, 2014a. Feed intake and growth performance of goats offered Napier grass (*Pennisetum purpureum*) supplemented with concentrate pellet and soya waste. *Sains Malaysiana.* 43: 967-971.
- Rahman, M.M, T. Nakagawa, R.B. Abdullah, W.E. Wan Khadijah and R. Akashi, 2014b. Feed intake and growth performance of goats supplemented with soy waste. *Pesq. Agropec. Bras.*, 49 : 554-558.
- Van Soest, P. J., J. B., Robertson and B. A. Lewis. 1991. **Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition.** *J. Dairy Sci.* 74: 3583-3589.

Productive performance and production cost of different cross bred meat goats fed high levels of OPL in fermented TMR

Khamseekhiew¹, B., O. Pimpat¹ & T. Jettana²

¹Faculty of Science and Industrial Technology, Prince of Songkla University (PSU), Surat Thani Campus, P.O.Box 8, KhunThalay Post Office, T. Makhamtia, A. Maeang, Surat Thani 84100,

²Technology Project for Artificial Insemination in Dairy and Swamp Buffalo. Nuclear Research Center for Bioscience in Animal Production Technology. Faculty of Veterinary Science, Chulalongkorn University, Henry Dunant Road, Pathumwan, Bangkok 10300

Abstract

This experiment determine the productive performance and production cost of three crossbred meat goats fed fermented total mixed ration (TMR) containing high level of oil palm leaflets. Eighteen of crossbred goats (T1= native, T2= native x Anglo Nubian, and T3= native x Boer) males with a mean age of 8-9 months and average weight of 20 ± 3.8 kg were used. Six goats per treatment were allocated to the individual cages in completely randomized design. Chopped OPL was used as a sole roughage source in fermented TMR. Animals were kept individual crate fed TMR as *ad libitum* with clean water were always available. The result showed that total FI were significantly higher in T3 than those for T1 and T2. Mean N retention were lowest for T1. N excretion (g/d) of T1 were highest (8.62 g/d), resulted to the lowest N absorbed in T1. ADG was ranged from 63.6 to 86.1 g/h/d. Production cost had showed not profitable to all treatment groups. The highest loss of profit was 994.1 Baht per head for native goat. It can be concluded that chopped OPL in fermented TMR is compromising roughage source for meat goat production. Growing and production of crossbred native with Boer obtained better performance to the diets.

Keywords: oil palm leaflet, total mixed ratio, productive performance, meat goats

Introduction

Meat goat raising in the south of Thailand has high potential of development to the standard farm for domestic and exporting purposes. One vital constraint of southern part is insufficient of natural forage in the dry season with low quality reflects to its low body weight gain. Both protein and energy are main nutritive factors for growth, and production such as meat and milk. Protein level of 0.404 g/g BW gain and energy requirement of 7.25 Kcal ME/g as NRC (1981) recommended seem not to appropriate for the local meat goats. This may due to most of the crossbred goats as raised under humid environment required nutrition and feeding system differently. In Thailand, oil palm leaflets (OPL) is a huge amount produced from oil palm plantation estimated of 1.48 M metric ton annually. OPL reported low digestibility, low energy, but contain interesting amount of 11.9% CP. Reported on addition of 70% dried chopped OPL in TMR have no negative effect to the

productive performance of goats (Khamseekhiewet al., 2015). Study of the use of high levels of OPL as main roughage source in fermented TMR comparison for specific crossbred meat goats including native and exotic breeds raise in southern Thailand yet to be fully evaluated. The objective of this experiment was to determine productive performance and production cost of different crossbred meat goats fed OPL as a sole roughage source in fermented TMR.

Materials and Methods

Animals and experimental diets

Eighteen crossbred goats (T1= native, T2= native x Anglo Nubian, and T3= native x Beor) males with a mean age of 8-9 months and average weight of 20 ± 3.8 kg were used in completely randomized design. Each goat per treatment was allocated to the individual cages) 1.0x1.5x1.2m (with slatted floor, water and feeding troughs. After separating from OPF, fresh OPL were immediately chopped with a chopping machine to ground fine particles and used as sole roughage source in fermented TMR. Diet TMR was formulated to meet the predicted protein (14% CP) and energy (7.25 Kcal ME/g) requirements and kept in 120 L plastic container for 2 weeks prior fed to the goats. The ingredient detail of the diet (%DM basis) were OPL; 7, palm kernel cake; 6, dried *leucaenaleucocephala*; 10, cassava chip; 4, decanter cake; 7, urea; 0.9, molasses; 1.5, premix; 0.2, salt; 0.2, di-calcium; 0.2). One month prior the experiment, the goats were de-wormed and vaccinated. Diets were offered *ad libitum* as two equal meal at 08.00 and 15.30 h with the mineral block and the clean water was always available.

Measurement

Feed refusals were collected before the morning feeding and weighed daily during the experiment. Daily DMI were calculated as difference between feed offered and feed refusal. Rumen fluids were taken from rumen by stomach tube with a vacuum pump at 0, 2, and 4 h after morning feeding. The pH was immediately measured with a portable digital pH meter. The rumen fluid samples were strained through cheesecloth and prepared for ammonia-N (5 ml of rumen fluid was added with 0.5 ml of 0.2 N HCL) and VFA (5 ml of rumen fluid was mixed with 1 ml of 250 g/l metaphosphoric acid) analyses. Rumen subsamples were frozen at -20°C prior later analyses. Blood samples from animal were obtained from the jugular vein (10 ml into sterile tubes containing EDTA solution) for BUN analyses. Mixed sample of TMR were oven dried at 60°C then ground to pass a 2 mm screen and stored for later analyses. The live weight of each animal was measured at the start and the end of experiment.

Laboratory and Data analyses

Determination of DM, ash, CP (Kjeldahl $\times 6.25$) and EE from the samples were according to the procedure of AOAC (2005). NDF were followed the method described by Van Soest et al. (1991). The concentrates of VFA were determined by gas chromatography. Ruminant ammonia-N was analyzed by the distillation method. BUN was analyzed by the method of Crocker (1967). Data were analyzed using the mixed procedure accounting for ANOVA (SAS, 1998). The model included effect of treatment, day of sample and its interaction. Least square mean were used to detect the difference between the treatments for probability. Effect of treatment were declared significant $P < 0.05$.

Results and Discussion

Total feed intake (DMI, OMI and CPI) and digestibility of goats presented in Table 1. The results showed that T3 had significantly ($P<0.05$) higher feed intake (DM and CP) than those for the two groups. Mean values for digestibility of DM, OM and CP were 55.4, 54.4 and 57.9% respectively. N intake was significantly higher for T3 than that of T2 and T1. Mean N retention were lowest for T1. N excretion (g/d) of T1 were higher than for T2 and T3 ($P<0.05$), resulted to the lowest N absorbed in T1 (Table 1). To all treatment, the ruminal pH was recorded high in the after morning feeding and this value was dramatically decreased in the afternoon. The mean value of pH for three treatments was 6.4. Ammonia-N in the T3 was significantly higher than that of T2 and T1. However, among the tested goats, there were not significant differences for BUN and SCFA parameters. There were similar pattern of the growth of goats. The ADG showed the highest (86.1 g per day) for T3 treatment, this consecutively reflected the BW gained to 9.81 kg throughout the experimental period. Feed costs per kg gained of goats were ranged between 55.6-60.7 Baht. The highest loss of profit was 994.1 Baht per head in native meat goat (T1).

Table 1. Voluntary feed intake, digestibility, N utilization, ruminal parameters and production cost of crossbred meat goats fed fermented TMR containing high level of OPL.

Parameters	treatment			SEM
	T1	T2	T3	
Total DM intake (g/d)	411.8 x	514.8 y	550.4 z	0.91
Total OM intake (g/d)	397.8 x	497.3 y	531.6 z	0.54
Total CP intake (g/d)	56.8 x	71.0 y	75.9 z	0.78
DM digestibility (%)	54.3	55.1	56.8	1.34
OM digestibility (%)	53.6	53.4	56.1	1.22
CP digestibility (%)	56.6	57.3	59.8	0.99
Fecal N (g/d)	3.9	4.8	4.9	1.52
Urinary N (g/d)	4.72	4.24	4.87	1.20
N-absorbed (g/d)	5.14	6.56	7.22	0.66
N-retention (g/d)	0.4 x	2.3 y	2.4 y	0.84
N-retention/N-absorbed (%)	8.2	35.3	33.0	
pH	6.8	6.8	6.7	1.29
Ammonia -N (mg/L)	135.0x	137.0x	145.0y	0.77
Blood urea- N (mg/dL)	37.5	36.8	36.5	0.92
Short chain fatty acids (mmol/L)	119.5	114.1	121.6	0.44
ADG (g/d)	63.6	72.5	86.1	1.30
Feed cost per 1 kg goat gained *	55.2	60.7	54.6	
1) Cost of feed per head *	400.9	501.1	535.8	
2) Cost of management per head *	1,900	1,900	1,900	
3) Weight gain x price on sale@ *	1,296	1,486.8	1,768.8	
Profit = 3-(1+2) *	-994.1	-914.4	-670.0	

T1 = native; NT), T2 = NT x Anglo Nubian, T3 = NT x Boer

x y z = different letters in the column are significantly differed ($P<0.05$)

SEM = Standard error of the mean

@ = price of goat is 180 baht per kg referred on May 2013, * = Baht

Conclusions

Based on results in the present study, it can be concluded that fermented TMR containing high levels of chopped OPL is compromising roughage source for commercial meat goat production. Crossbred of native and Boer goats have been satisfactory performing to the experimental diets for its growing and production cost.

References

- Association of official analytical chemists 2005. Official methods for analyses of the association official agriculture chemists. 16th ed. AOAC, Washington DC.
- Crocker, C. L. 1967. Rapid determination of urea nitrogen in serum or plasma without deproteinization. American J. Medical Technol. 33: 361.
- Khamseekhiew, B., O. Pimpa, S. Rueangsuwan, & T. Jetana. 2015. Study of oil palm leaflets as roughage source in TMR for meat goats. Thai J. Anim. Sci. 1:399-403
- NRC. 1981. Nutrient requirements of goat: Angora, dairy and meat goats in temperate and tropical countries. Washington D.C., USA: National research council, National Academy Press.
- SAS. 1996. SAS User's guide: statistics, Version 6.12th Edition. SAS Institute Inc. Cary, NC.
- Van Soest, P.J., J.B. Robertson & B.A. Lewis. 1991. Methods for dietary fibre and neutral detergent fibre and non-starch polysaccharides (NSP) in relation to animal nutrition. J. Dairy Sci. 74: 3583-359

The effect of *S. rarak* microparticles on blood profile and productivity of broiler chickens raised on litter system inoculated with *E. tenella*

Pasaribu, T^{1,2}, E. Wina², Sumiati³, A Setiyono⁴ & D. A. Astuti³

¹PhD student of the Post Grad. School, Bogor Agricultural University, ²Indonesian Research Institute for Animal Production, Bogor, ³Dept. of Nutrition and Feed Technology, Fac. of Animal Science, Bogor Agricultural University, ⁴Dept. of Clinic, Reproduction and Pathology, Fac. of Veterinary Medicine, Bogor Agricultural University

Abstract

This study was conducted to evaluate the effectiveness of *S. rarak* microparticles (SRM) addition in feed on the blood profile and productivity of broiler chickens infected with *Eimeria tenella*. A total of 270 doc divided into 5 treatments with 6 replications (9 chicks per replicate) were reared in the litter system for 35 days. The treatments consisted of: R1 (without coccidiostat and SRM); R2 (coccidiostat, without SRM); R3 (SRM 1.25 g kg⁻¹ ration); R4 (SRM 2.5 g kg⁻¹ ration); R5 (SRM 5.0 g kg⁻¹ ration). The experimental design used was completely randomized design. At 14 days old, all chickens were infected with *E. tenella* by sprinkling 15,000 oocytes m⁻² on the litter. The parameters measured were blood profile, lipid profile, and body weight. The results showed that blood profile was not affected by any treatment. SRM treatment lowered cholesterol, LDL, and triglycerides, but increased HDL in the blood serum. Body weight and feed conversion in the 1.25 and 2.5 g kg⁻¹ SRM treatments were the same as those treated with coccidiostat treatment in broiler chickens infected *E. tenella*. It was concluded that the administration of *S. rarak* microparticles (SRM) 75 µm size could be used as a cholesterol reducer without affecting the productivity at up to 2.5 g kg⁻¹ dose in broiler chickens infected with *E. tenella*.

Keywords: broiler chickens, *E. tenella*, *S. rarak* microparticles, cholesterol, productivity.

Introduction

Consumers are increasingly selective in choosing meat, especially that with low levels of fat and cholesterol. Healthy broiler meat products also must be free of microbial contamination, antibiotic residues, anticoccidia, pesticides, heavy metals and other toxic materials. One of the plants that have the potency to inhibit the growth of protozoa is *Sapindus rarak* (known locally as *lerak*) because it contains a secondary compound called saponin (Wina et al, 2005). Therefore, saponin may be potential as anticoccidia and feed additive to produce healthy meat product.

The research objective was to investigate the effect of *S. rarak* microparticle on blood profile and productivity of broiler chickens on the litter system that was inoculated with *E. tenella*.

Materials and methods

The study used 270 unsexed day old chickens of Cobb strain, placed in litter pens with a capacity of 9 chicks m⁻². Prevention of ND and Gumboro was done by vaccination with ND La Sota strain and IBD. Rations were composed as isoprotein and isocalory (22% CP, 3050 kcal kg⁻¹ ME and 20% CP, 3100 kcal kg⁻¹ ME for starter and grower, respectively. *S. rarak* microparticle 75 µm (SRM) was prepared and different level was mixed with the ration. Feed and water were given *ad*

libitum. Fourteen days old chickens were naturally infected with *E. tenella* by sprinkling 15,000 *E. tenella* m⁻² over the husks, so it was expected that chickens were infected through the ingestion of *E. tenella* from the husk. At the end of the study (age 35 days), blood was sampled and analysed for blood profile, cholesterol, HDL, IgY. The study was arranged using completely randomized design (CRD) with 5 treatments, 6 replications (9 chickens/replicate). The treatment rations given were: R1 (control ration, without coccidiostat, without SRM), R2 (ration containing coccidiostat), R3 (ration contained 1.25 g kg⁻¹ SRM), R4 (ration contained 2.5 g kg⁻¹ SRM), R5 (ration contained 5.0 g kg⁻¹ SRM).

Results and discussion

Blood profile

The number of erythrocytes, hemoglobin, hematocrit, leukocytes, and albumin did not differ significantly among all treatments (Table 1). Wardiny et al. (2012) reported the same findings that saponin did not significantly affect the blood profile in quails.

Table 1. The effect of SRM on blood profile of broiler chickens infected with *E. tenella*

Variables	R1	R2	R3	R4	R5	SEM
Erythrocytes (10 ⁶ mm ⁻³)	3.91	4.24	3.68	3.91	3.82	0.24
Hemoglobin (g %)	9.78	9.25	9.54	9.21	8.85	0.34
Hematocrit (%)	29.95	27.93	28.78	28.07	28.40	0.85
Leukocyte (10 ³ mm ⁻³)	11.47	10.47	9.22	8.72	8.30	1.49
Lymphocytes (%)	45.42	45.92	51.00	57.60	46.92	4.06
Heterophiles (%)	44.67	42.33	38.50	29.90	41.08	3.31
Monocytes (%)	7.73	7.25	5.58	8.30	7.33	1.17
Eosinophils (%)	4.38	5.41	5.89	4.31	5.61	0.86
Albumin (g dL ⁻¹)	1.78	1.88	1.60	1.45	1.54	0.13
IgY (µg mL ⁻¹)	13.82	14.13	14.54	14.21	13.35	0.63

IgY level of serum tended to increase with administration of lower dose of SRM. Saponin of *Yucca shidigera* and ginseng were reported to have immune stimulating properties (Cheeke 2001, Tan and Vanitha 2004). SRM saponin improves the chicken immunity at a dose of 1.25- 2.5 g kg⁻¹ SRM.

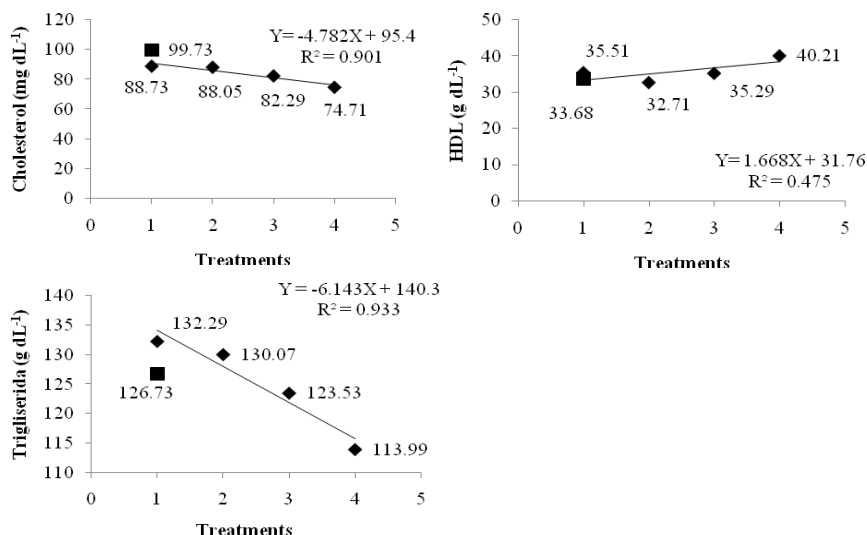


Figure 1 The influence of SRM on blood serum cholesterol, HDL and triglyceride of broiler chickens with infected *E. tenella*. 1 (◆) (R1); 1 (■) (R2); 2 (R3); 3 (R4); 4 (R5);

Blood cholesterol contents in all treatments were not significantly different, but there is a negative response of cholesterol including LDL cholesterol to SRM dose (Fig. 1). Quillaja saponin lowered the serum cholesterol level in rats (Afrose et al. 2009).

Blood triglyceride levels tended to decrease by the administration of 5.0, 2.5, and 1.25 g kg⁻¹ SRM doses (Fig. 1). The same case was reported by Pasaribu et al. (2014) in which the administration of 25-75 mg saponin extract in broiler chickens also lowered triglyceride level.

Productivity

At the age of 21 and 35 days, feed intake (FI) was not significantly different among all treatments (Table 2). On the treatment of 1, 25 - 5.0 g kg⁻¹ SRM, the chickens consumed 1.48 - 6.17 g/bird of saponin for 35 days. This indicates that saponin of 1.25-5.0 g kg⁻¹ SRM in feed had no effect on the palatability of feed by broiler chickens.

Body weights (BW) of chicken at 21 days old provided with 1.25, 2.5, and 5.0 g kg⁻¹ SRM doses were not significantly different ($P > 0.05$) from those by the coccidiostat treatment and control ($P > 0.05$) (Table 2). Chicken fed 5.0 g kg⁻¹ SRM showed lower body weights than those with 1.25 and 2.5 g kg⁻¹ SRM treatments at the age of 35.

Feed conversion ratio (FCR) of 21 days old chickens was not significantly different ($P > 0.05$) among all treatments (Table 2), but at 35 days old showed dose of 5.0 g kg⁻¹ SRM had a higher FCR. FCR is closely related to feed intake and body weight gain. This shows that the 5.0 g kg⁻¹ SRM treatment has negatively affected chicken performance.

Table 2. The effect of SRM on broiler chicken productivity that infected *E. tenella*

	R1	R2	R3	R4	R5	SEM
FI (g ^{e-1}) 21 d	1078.44	1096.89	1101.48	1098.76	1037.72	18.22
FI (g ^{e-1}) 35 d	2976.63	2919.38	2896.45	2949.16	3063.51	44.89
BW (g ^{e-1}) 21 d	618.1	654.08	648.32	645.21	615.83	14.39
BW (g ^{e-1}) 35 d	1603.13 ^a	1518.37 ^b	1573.33 ^{ab}	1550.45 ^{ab}	1404.02 ^c	25.07
FCR 21 d	1.867	1.855	1.875	1.852	1.895	0.03
FCR 35 d	1.901 ^b	1.980 ^b	1.930 ^b	1.926 ^b	2.255 ^a	0.04

Conclusion

The administration of *S. rarak* microparticles 75 µm (SRM) up to 2.5 g kg⁻¹ in broiler chickens infected with *E. tenella* had no effect on the blood profile and productivity. Concentrations of cholesterol, LDL and triglycerides were lower with increasing level of SRM. Administration of *S. rarak* microparticles 75 µm (SRM) up to 2.5 g kg⁻¹ dose in broiler chickens infected with *E. tenella* could be used as a cholesterol reducer without affecting the productivity.

References

- Afrose, S., M. S. Hossain, T. Maki and H. Tsujii. 2009. Karaya root saponin exerts a hypocholesterolemic response in rats fed a high-cholesterol diet. *Nutr Res.* 29:350-354.
- Cheeke, P. R. 2001. Actual and potential applications of *Yucca schidigera* and *Quillaja saponaria* saponins in human and animal nutrition. *Recent Adv Anim Nut Aust.* 13:115-126.
- Pasaribu, T., D. A. Astuti, E. Wina, Sumiati and A. Setiono. 2014. Pengaruh tepung *Sapindus rarak* sebagai pakan aditif terhadap performa dan profil lipida ayam broiler yang diinfeksi *Eimeria tenella*. *JITV.* 19(4):263-271.
- Wina, E., S. Muetzel, E. M. Hoffmann, H. P. S. Makkar and K. Becker. 2005. Effect of secondary compounds in forages on rumen micro-organisms quantified by 16S and 18S rRNA In: Applications of gene-based technologies for improving animal production and health in developing countries. Makkar HPS and Viljoen GJ (eds.), IAEA-FAO, Springer, Netherlands, pp. 397-410. *Anim Feed Sci and Technol.* 121: 159-174.

Effect of different Chinese herbs on antioxidant capacity and immune function in Sansui laying duck

Yang^{*1}, S.L. W. Lin¹, B. Q. Yao², M. Lu, & X. M. Li

¹Animal Science College, Guizhou University, Guiyang, 550025, Guizhou Province, China,

²The management office of Sansui duck Industry, Sansui County, 550023, Guizhou Province, China

Abstract

The aim of the experiment was to study the effect of different Chinese herbal additives on immune function, antioxidant capacity and serum biochemical indexes in Sansui laying duck. Two kinds of Chinese medicine Herbal powders Herb I and Herb II were applied in this study. 300 Sansui laying ducks at 1 day of age were randomly divided into five groups, each group consisted of three replicates and each replicate had 20 ducks. The control group of ducks were received a basal diet (BD) without any feed additive, the experimental group I was fed BD added 1% of Herb I, the experimental group II was fed BD added 1.5% of Herb I, the experimental group III was fed BD added 1% of Herb II, and the experimental group IV was fed BD with 1.5% of Herb II.

The experimental duration was 120 days and rearing environment for all groups were completely the same. The results showed that the contents of serum GsH-Px of group I, group II were significantly higher than control group ($P < 0.05$), group III, and group IV increased slightly, but the no significance ($P > 0.05$). The serum SOD levels for all groups were significantly higher than control group ($P < 0.05$). It could significantly reduce concentration of malondialdehyde (MDA) in blood while adding Chinese medicine herbs ($P < 0.05$). Compared to the control group, the serum IgA levels of group II, III, IV had significantly increased by 10.41%, 8.75%, 9.71% ($P < 0.05$), respectively. No significance was detected in group I ($P < 0.05$). The serum IgG contents for group I, II, III, IV were greater than control ($P < 0.05$), The serum IgM contents of the group II, III were significantly increased ($P < 0.05$). The serum total protein (TP) contents for group IV and group II were higher than control ($P < 0.05$). ALB levels of all groups were higher than the control group ($P < 0.05$). Group II was the highest. Suggesting that Chinese medicine herbs can improve the antioxidant capacity and immune function in Sansui laying duck.

Keywords: Chinese herbal medicine, Sansui duck, Antioxidant index, Immune parameters

**Corresponding author:* shenglinyang@126.com

Introduction

Sansui duck is one of the native laying duck breeds, which was listed in "The catalog for Chinese poultry breeds" in 1982. In recent year, with the further development of feed industry, some problems have been brought into duck industry. Particularly, some feed additives such as

hormones, antibiotics, although improve duck production, harm to human health because of its chemical residual (Ho, 2007). Alternatively, the herbal medicine additive is a good choice with its natural, versatility, low toxicity, low cost and no-residual. In this study, Chinese herbal additives were used as the substitute to antibiotics for Sansui laying ducks used as experimental animals to investigate their antioxidant capacity and immune function.

Materials and Methods

Three hundred Sansui laying ducks at 1 day of age were randomly allocated into five groups, each group consisted of three replicates and each replicate had 20 ducks. The ducks in the control group were received a basal diet (BD) without any feed additive, while the ducks in the experimental group I, II, III and IV were fed BD added 1% of Herb I, 1.5% of Herb I, 1% of Herb II and 1.5% of Herb II, respectively. The basal diets composition and nutrient levels for all groups were exactly the same.

Chinese herbal additives used in the test compatibility. Herbal feed additives were divided into two components—Formula I: *astragali aadix*, *shenqu*, *citri reticulatae pericarpium*, *glycyrrhizae radix rhizoma*, *dioscoreae rhizoma*, *schisandrae chinensis fructuse chinensis fructus*, *cinnamomi cortex*, *mume fructus*; Formula II: *foeniculi fructus*, *eucommiae cortex cortex*, *atractylodis macrocephalae rhizoma*, *hordei fructus germinatus*, *houuttyniae herbae herba*, *crataegi fructus*, *citri reticulatae pericarpium*, *zingiberis rhizoma*.

Index of measurement

Measurement of serum immunoglobulin and serum antioxidant

At 120th days of trial period, 20 Sansui duck of each group were randomly selected, and 5 ml of blood was collected from vein of wing for measuring the contents of Serum immunoglobulin G (IgG), immunoglobulin A (IgA), immunoglobulin M (IgM), superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) activity, and malondialdehyde (MDA) according to enzyme-linked assay (ELISA) using measuring kit from US biotechnology company.

Statistical analysis

The experimental data were analyzed statistically using analysis of variance (ANOVA) technique by the program SPPP 13.0 and statistical significance was recorded at the 1% significance using least significant difference test (LSD).

Results

Content of IgA、IgM、IgG

From the Table 1, the ducks in the Group II, III, IV increased by 10.41% , 8.75% , 9.71% respectively, in content of serum IgA, compared with those in the control group ($P<0.05$), The IgA of ducks in the Group I was slightly higher but no difference was detected when compared with those in the control ($P>0.05$). The IgG contents for the ducks in four groups (I, II, III and IV) increased by 9.25%, 17.98%, 9.07%, 8.99%, respectively compared with the control group ($P<0.05$). The IgM levels of ducks in Group II, III were higher than those in the control group ($P<0.05$). No differences were detected in Group I, IV compared with those in the control ($P>0.05$).

Table 1 Contents of IgA、 IgM、 IgG (g/l) in Sansui laying duck.

Groups	IgA	IgM	IgG
Control group	1.143±0.044 ^a	1.756±0.030 ^a	1.179±0.007 ^a
Group I	1.171±0.032 ^a	1.772±0.028 ^a	1.288±0.003 ^b
Group II	1.262±0.043 ^b	1.876±0.032 ^b	1.391±0.003 ^b
Group III	1.243±0.042 ^b	1.812±0.051 ^b	1.286±0.004 ^b
Group IV	1.254±0.046 ^b	1.781±0.033 ^a	1.285±0.006 ^b

^{ab} The same column data with different superscripts indicated significant differences ($P<0.05$)

Contents of GSH-PX、 SOD and MDA

The values of GSH-PX, SOD and MDA were listed in the Table 2. GsH-Px contents of ducks in group I and group II were higher than control ($P<0.05$), but no differences were observed in group III and IV when compared with those in the control group ($P>0.05$). The SOD values for the ducks in four groups (I, II, III and IV) were significantly higher than that in the control group ($P<0.05$). For MDA values, no differences were detected in all groups ($P<0.05$).

Table 2 Levels of GSH-PX、 SOD and MDA in serum (pg / ml)

Groups	GsH-Px	SOD	MDA
Control group	4.282±0.047 ^a	3.026±0.034 ^a	5.784±0.964 ^b
Group I	4.396±0.014 ^b	3.137±0.026 ^b	5.496±0.234 ^a
Group II	4.395±0.013 ^b	3.138±0.034 ^b	5.455±0.272 ^a
Group III	4.288±0.011 ^a	3.135±0.040 ^b	5.591±0.184 ^a
Group IV	4.284±0.012 ^a	3.130±0.025 ^b	5.590±0.172 ^a

^{ab} The same column data with different superscripts indicated significant differences ($P<0.05$)

Discussion

Effect of Chinese herbs on immune function in laying duck

There were a significantly increasing on the levels of serum IgG, IgA, and IgM when added 1.5% of *astragali radix*, *cinnamomi cortex*, *angelicae sinensis radix*, the results showed that the effective components of Chinese herbs can improve immune function of Sansui laying duck, this result is consistent with the report in chickens (Li, et al., 2005).

Effect of Chinese herbs on serum antioxidant index

SOD can catalyze the dismutation of superoxide anion radical generation H_2O_2 , removing-OH radicals, increased serum total antioxidant effect and reduce serum of MDA, can significantly improve the body's antioxidant function (Dong et al., 2004). Liu et al. (2007) reported that Chinese herbs can improve SOD activity in broiler when added *ligustrum*, *schisandrae chinensis fructus*, and other herbal additives (Liu et al., 2007). Yang et al. (2010) demonstrated that adding *Astragali Radix*, *Angelicae Sinensis Radix* and other herbs can significantly increase GsH-Px activity and antioxidant capacity in chicks (Yang et al. 2010). But little is known about the functions of Chinese herbs in laying duck. In current study, similar results were obtained compared to above studies.

Acknowledgements

This work were supported by the Natural Science Foundation of China (31360566) and the Agricultural Major Special Project of Guzhou province [No.(2012)6004].

References

- Ho, J., 2006. Application of Chinese herbal medicine in poultry. *J. Poul. Breed. Vet. Sci.* (6): 10-11. (in Chinese)
- Li, Q. D. and A. S. San, 2005. Effect of Chinese herbal medicine to growth and immune function in chicken. *J. Chinese Vet. Sci.* 8(41): 29-30.
- Halliwell, B. and J. Gutleridge, 1995. *Free Radicals in Biology and Medicine.* Oxford: Clarendon Press.
- Dong, X. M., C. F. Zhang and P. Wei, 2004. Effects of Chinese herbal medicine on antioxidant capacity in broiler. *China Poultry.* 26(14): 14-16.
- Jibet, A. R., J. A. Botten and D. S. Miller, 1998. Characterization of Age and Dose-related Outcomes. *Duck Hepatitis Bvirus Infection. Virology.* 244(2): 273-282.
- Chen, J., Z. Y. Pu and X. J. Liu, 2010. Effect of Chinese herbal medicine on the antioxidant index of earlier weaning pigs. *J. Sengyang Agr. Univ.* 41(4):477-480.(in Chinese)
- Liu, Y. Q. and J. L. Qui, 2007. Effect of Chinese herbal medicine on superoxide dismutase (SOD) in broiler chicken. *J. Anhui Agr. Sci.* 35(12):35-36.(in Chinese)
- Yang, F. W. and W. Yu, 2010. Influence of Chinese herbs on contents of NO and GSH-Px activity in irritated chicks. *Heilongjiang Anim.Vet. Sci.*1:143-144.(in Chinese)

Enhancing the nutritional value of soybean through supplementation with new-generation feed enzymes for poultry –Review

Mammo M. Erdaw^{1,3}, Rider A. Perez-Maldonado², M.M. Bhuiyan¹ and Paul A. Iji^{1,*}

¹School of Environmental and Rural Sciences, University of New England, Australia; ²DSM Nutritional Products, 2-Havelock Road #04-01, 059763, Singapore; ³Ethiopian Institute of Agricultural Research, Debre-Zeit Centre, Ethiopia.

Abstract

In terms of global gas emission, climate change and efficiency of feed utilization, poultry are the preferred farm animals to meet the high demand for animal protein foods. Soybean meal (SBM) remains the most important and the preferred plant protein feed source for animal feeding. However, supply of conventional SBM fluctuates while the price is on the increase due to demand, processing and transportation costs. Although there is a growing interest in the use of raw SBM for birds, the nutritive value is negatively affected by anti-nutrients. Heat treatment is advised to alleviate some of the anti-nutrients such as trypsin inhibitors and lectins, but both under- and over-processing of soybean tend to reduce the quality of the meals. Supplementing poultry diets with phytase and protease is a routine biotechnological intervention for improving the nutritional value of feed ingredients and reducing pollution. Proteases can breakdown both stored proteins and proteinaceous anti-nutrients in soybeans to improve bird performance and reduce environmental pollution. Phytase is also effective in breaking down the phytate associated with a number of nutrients, including minerals and protein. Recent *in vitro* and *in vivo* studies are showing that the use of microbial enzyme cocktails can reduce the negative effects of anti-nutrients in soybean meal for birds. This review provides information on how protease and phytase are contributing to the improvement of nutritional value and can obviate the need for pre-processing of soybean meals for poultry.

Keywords: new-generation enzymes; anti-nutritional factors; soybean meal.

*Corresponding author: pji@une.edu.au

Introduction

Poultry production is one of the most advanced industries in the livestock sector (Asyifah *et al.*, 2012) and relies on a complex feed production process. Although soybean is most widely used vegetable protein source for poultry, the raw seed meal contains a number of anti-nutritive factors (ANF) such as protease inhibitors, lectins and phytate (Chen *et al.*, 2013; Pettersson and Pontoppidan, 2013), which reduce nutrient digestibility by binding to various nutrients. To reduce the effects of ANF, soybean seeds are heated before feeding to non-ruminant animals, but some nutrients are affected by excessive heating. On the other hand, supplementation of soybean-containing diets with microbial feed enzymes reduces the negative effects of ANF, and improves nutrient availability to birds. The objective of this paper is to review findings on the benefits of supplementing soybean with new-generation feed enzymes.

Effects of antinutrients in soybean and impact of supplementation with selected microbial feed enzymes for poultry

Several ANF are found in protein-rich oilseeds/feedstuffs, which can interfere with the intake, availability or metabolism of nutrients in non-ruminant animals (Campbell and Schöne 1998; Soetan and Oyewole, 2009). As shown in Table 1, the concentration of trypsin inhibitors (TI) in raw soybean is almost double the concentration of TI in commercial SBM. The urease activities (UA) and protein solubility (KOH) are also proportionally higher in raw than in commercial SBM. Birds fed on diets containing raw soybean meals experience poor body growth, low feed intake and increased weight of pancreas due to the effects of ANF (Mogridge *et al.*, 1996; Perez-Maldonado *et al.*, 2003; Newkirk, 2010).

In addition to developing new breeds of soybean crops with low antinutrient contents, treating the raw seeds by heat or supplementation with microbial enzymes are some of the common options that are currently used to improve the nutritional value of such feeds for poultry. Protease is a protein-digesting enzyme that breaks down both stored proteins and proteinaceous anti-nutrients in feeds (Jacela *et al.*, 2013; Barletta, 2011). Various studies have shown that performance of birds is improved due to supplementation of protease enzymes to their diets (Rada *et al.*, 2014; Yadav and Sah (2005). Phytase is also effective in breaking down the phytate (phytic acid), which chelates with mineral cations, and is effective in improving N, amino acid, and energy utilization (Rezaei *et al.*, 2007).

The response of broiler chickens to diets containing soybean and supplementation with various microbial enzymes is shown in Table 2. The data generally highlight a positive response to such supplements, particularly in maize-soybean based diets.

Table 1 Some selected anti-nutrients in raw full-fat and commercial SBM

Raw full-fat SBM					Commercial SBM				References
TI (mg/g)	UA (ΔpH)	KOH (%)	Lectins, (mg/gm)	Phytate (mg/gm)	TI (mg/g)	UA (Δ pH)	KOH (%)	Phytate, (mg/gm)	
40.486	>0.20	90	3.7	-	-	0.05-0.20	85	-	Palić <i>et al.</i> (2008);Gu et al. (2010).
30-102.5	-	-	-	5-25	-	-	-	14.5	Kwanyuen & Burton (2005);Sharma <i>et al.</i> (2013).
45-60	-	-	20-200, ppm	6	1.77	.02	-	-	McNaughton <i>et al.</i> (1998); Van Eys <i>et al.</i> (2004).
26.48	-	-	-	-	1.8-2.9	-	-	-	Dragičević et al. (2010);Serrano <i>et al.</i> (2012).
48.33, TIU/mg	-	-	-	11.2	-	0.2	-	-	ILSI (2010); Crowell (2012).
23.9	-	-	7.3	-	-	-	80-85	-	Crowell (2012).
50,800, TIU/g	1.99	98	-	-	3,000,TIU/g	0.08	82	-	Ruiz <i>et al.</i> (2004).
41.5–85	-	-	-	2.3–5.6	1.50-3.45	-	77-87	-	Sharma <i>et al.</i> (2011);Ruiz, (2012).

UA= urease activity; KOH= protein solubility; TI= trypsin inhibitors; TIU trypsin inhibitor units.

Table 2 Response of birds fed on soybean-containing diets supplemented with different microbial enzymes

Enzyme types	Supplements (g/kg)	Nature of diet	Type of chickens	Response	References
Roxazyme G®	2.0	Corn-ground nut	Broilers	Improvement in dressed percentage	Omojola and Adesehinwa (2007).
Multi-carbohydrase	0.1	Corn/SBM	Broilers	No improvement in carcass yield	Vieira <i>et al.</i> (2006).
Multi-Enzymes		Corn	Broilers	Carcass yield was improved	Bangbose <i>et al.</i> (2005).
Carbohydrolase	0.05	Corn/SBM	Broilers	Breast yield was improved	Marcio <i>et al.</i> (2011).
Xylanase and β -glucanase	0.5	Corn/SBM	Broilers	Improvement in carcasses	Hajati (2010).
Protease and phytase	5,000 units/kg	Corn/SBM	Broilers	Increase in activities of enzymes	Murugesan <i>et al.</i> (2014).

Conclusion

There is a growing interest in using locally grown full-fat SBM for birds by producers. Supplementation of such diets with exogenous enzymes such as phytase and protease typically improves the nutritional value, reduces feeding cost and environmental pollution. Since proteases can breakdown the stored proteins and proteinaceous anti-nutrients in raw SBM, performance of birds can be improved through dietary supplementation with such enzymes, particularly the new-generation products.

References

- Chen, Y., Duan, W., Wang, L., Zhang, S., Zhou, Y., Ulupi, N., . . . Al-Nasser, A. (2013). Effects of thermostable phytase supplementation on the growth performance and nutrient digestibility of broilers. *Int. J. Poultry Sci.* 12(8), 441-444.
- Perez-Maldonado, R., Mannion, P., & Farrell, D. (2003). Effects of heat treatment on the nutritional value of raw soybean selected for low trypsin inhibitor activity. *Br. Poultry Sci.*, 44(2), 299-308.
- Rezaei, M., Borbor, S., Zaghari, M., & Teimouri, A. (2007). Effect of Phytase supplementation on nutrients availability and performance of broiler chicks. *Int. J. Poultry Sci.*, 6(1), 55-58.
- Rada, V., Lichovnikova, M., & Foltyn, M. (2014). The effect of serine protease on broiler growth and carcass quality. *Acta Fytotechnica et Zootechnica*, 17(3), 87-89.
- Ruiz, N., De Belalcazar, F., & Diaz, G. (2004). Quality control parameters for commercial full-fat soybeans processed by two different methods and fed to broilers. *J. Appl. Poultry Res.*, 13(3), 443-450.
- Yadav, J., & Sah, R. (2005). Supplementation of corn-soybean based broiler's diets with different levels of acid protease. *J. Int. Agric. Anim. Sci.*, 26, 65-70.

The effects of feed fermented by *Azotobacter* culture on milk production and feed efficiency of dairy cattle

Endang Setyowati¹ & Nur Cholis¹

Brawijaya University, Faculty of Animal Husbandry, Malang, East Java Indonesia

Abstract

The goal of this research was to know the effects of using feed fermented by *Azotobacter* culture on milk production and its feed efficiency in dairy cattle. Ten heads of dairy cattle were used in the study. The feed (commercial concentrate and chopped cornstalks) was fermented by 350 cc *Azotobacter* microbes culture per 100 kg feed. The animals were divided into two groups as the treatments, i.e. received fermented feed and control. Measured variables were milk production and feed consumption. The data were analyzed by t-test. Statistical analysis showed that there was no significant difference on feed consumption but on milk production ($P < 0.1$). The milk production of fermented feed was 11.16 ± 5.14 l/head/day compared to the control 10.13 ± 2.99 l/head/day. The feed efficiency of the fermented feed was 1.26 ± 0.44 . It was concluded that dairy feed fermented by *Azotobacter* microbes culture in this study could increase the milk production and its feed efficiency.

Keywords: fermented feed, microbes culture, milk production and feed efficiency

Introduction

The availability of abundant green forage during the wet/rainy season in tropical countries like Indonesia is not maximally used by the small local dairy farmers. There is fermentation feed technology by decomposer microbes such as *Azotobacter*. Microbes culture originated from roots of Alfalfas were rich with *Azotobacter* microbes (Widiasmadi, 2010). The previous research showed that animals fed fermented by *Azotobacter* originated from roots of Alfalfas culture could increase the feed nutrients and its digestibility (Setyowati & Cholis, 2013). It was hoped that by giving fermented feed to the animals could affect the feed consumption and the milk production (Adiarto, 2012, Smith & Becker, 2002). Recently, there is a lack of research studying the effect of such fermented feed on feed consumption, milk production and feed efficiency in tropical conditions.

This research goal was to know the effect of using fermented feed by *Azotobacter* culture on milk production and its efficiency in dairy cattle. The results could be used by the dairy farmers in tropical areas in applying the *Azotobacter* culture on fermented feed for the animals to increase the production.

Materials and Method

Ten dairy cattles of 3-4 parity were used as materials in this research. The animals were divided into two groups, T1 was received fermented feed while the other as control (T0), or received feed with no fermentation. The *Azotobacter* microbes was taken from noodle roots of Alfalfa (*Medicago sativa*) of 5 years age. The other microbes was come from cattle rumen fluid. There was no measuring of microbial content of fermented feed, before or after the treatment. 100 kg of chopped cornstalks or commercial concentrate were fermented by 350 ml of *Azotobacter* microbes culture, respectively. The other substances added to the composting feed were 1 litre molasses and smal amount of rice bran. After mixing they were placed in sealed plastic drum and allowed to ferment for three days. After three days the composting feed were taken out and ready to be given to the animals. There was an adaptation period of 1.5 months for the treatment cattle. Then, the feed consumption and milk production were measured daily. The collected data were analyzed by t-test.

Result and Discussion

To know the nutrient content of the fermented feed, sample was taken after the process was finished. Fermentation process occured in 3x24 hours. Proximate analysis of fermented feed is seen in Table 1.

Table 1. Proximate Analysis of FeedSource (%).

N	Source	DM	Ash	CP*	CF*	Fat*
1.	Cornstalks	40.21	14.91	15.56	31.51	2.14
2.	Commercial Consentrate	80.38	10.66	15.83	21.11	8.04

DM: Dry Matter; CP: Crude Protein; CF: Crude Fibre

Note: *based on 100% Dry Matter

The previous studies was done to know the best result of three concentration of the addition of *Azotobacter* culture of 150 ml, 250 ml and 350ml per 100 kg feed. The results showed that based on the proximate analysis, the *Azotobacter* microbes culture of 350 ml/ 100 kg feed was the best because of the decreased fibre content due to the microbes work. Nutrient content of the fermented feed of those treatment were 44.02%; 13.58%; 15.1%; 26.54%, and 3.86% of DM, ash, CP, CF and fat, repectively. The samples also run into *in sacco* rumination and the VFA content were measured. The Volatile Fatty Acid (VFA) content were 5.91 ml Mol/l, 2.39 ml Mol/l and 1.19 ml Mol/lof acetate acid, propionic acid, and butyric acid, respectively. This VFA content were higher than corn silage with high water content (Kung & Shaver, 2001, Utomo, 2012, Rao & Cooper, 1994).

Statistic analysis result showed that there was no significant differences among treatments on feed cunsumption ($P > 0.05$) but milk production ($P < 0.10$). The average dry matter consumption of the animals in this study was between 13.26 and 14.02 kg/day. Milk production in T1 and T0 were 11.16 ± 5.14 l/head/day and 10.13 ± 2.99 l/head/day. This differences was due to low crude fibres and high volatile fatty acid (VFA) in fermented feed. Thehigher the VFA content the higherthe milk production. The feed efficiency was calculated by dividing feed

consumption/head/day with milk production/head/day. The feed efficiency of T1 and T0 were 1.26 ± 0.44 and 1.35 ± 0.36 , respectively.

It could be concluded that the use *Azotobacter* microbes culture as starter of feed fermentation could increase the dairy milk production and the feed efficiency. In this study, the dose of 350 ml/100 kg dairy cattle feed gave better result than control.

References

- Adiarto. 2012. Beternak Sapi Perah Ramah Lingkungan. PT Citra Aji Pratama. Yogyakarta.
- Kung, L. and R. Shaver, 2001. Interpretation and Use of Silage Fermentation Analysis Reports. Focus On Forage-Vol 3: No. 13
- Rao, J. R. and J. E. Cooper, 1994. Rhizobia catabolize nod gene-inducing flavonoids via Cringfission mechanisms. J. Bacteriol. 176(17):5409.
- Smith, J and K. Becker. 2002. The Reproductive Status of Your Dairy Herd. Extension Dairy Specialist and 4-H Agent. New Mexico State University
- Utomo, R. 2012. Evaluasi Pakan dengan Metode Noninvasif. PT Citra Aji Parama. Yogyakarta.
- Widiasmadi, N, 2010. Multi Fungsi MA-11. Makalah presentasi di Dinas Peternakan Jawa Tengah.

Evaluation of nutrient digestibility of mixed cassava pulp and Napier Pakchong grass for use as an alternative feedstuff in laying hens

Sirisopapong M., A. Khimkem, P. Pasri, S. Chaiyasit, P. Jaiboonlue, S. Okrathok & S. Khempaka

*School of Animal Production Technology, Institute of Agricultural Technology,
Suranaree University of Technology, NakhonRatchasima 30000, Thailand*

Abstract

This study was conducted to investigate the effects of using mixed cassava pulp and Napier Pakchong grass as feed substitution in laying hen diets on nutrient digestibility and retention. A total of 40 laying hens (Isa brown), 34 weeks of age, were randomly distributed to 5 dietary treatments: one control and 4 mixed cassava pulp and Napier Pakchong grass as 80:20 ratio at levels of 5, 10, 15 and 20% through 10 days. The excreta were total collection at the last 4 days of experimental period, sprayed with 5% HCl and dried at 60 °C for 2 days. The results showed that all mixed cassava pulp and Napier Pakchong substitution levels had no negative effects on dry matter and organic matter digestibilities and nitrogen retention. In conclusion, it indicates that the mixed cassava pulp and Napier Pakchong can be used in laying hen diets up to 20% without showing negative effects on nutrient digestibility and retention.

Keywords : cassava pulp, laying hen, Napier Pakchong, nutrient digestibility

Corresponding author: khempaka@sut.ac.th

Introduction

Feed is a key economic impact on poultry production accounts for 60-70% of the total production cost. Conventional feedstuffs especially corn, fish meal or soybean meal are lacked of supply and the prices have increased continuously. Therefore, studies finding the unconventional feedstuffs would help to reduce feed cost. Nakhon Ratchasima province is the largest area of cassava production, approximately 45% of the total production is used to produce cassava starch and generated by-product approximately 2-3 tons annually. Cassava pulp contains 53.55% starch, 2.83% ash, 1.98% crude protein (CP), 13.59% crude fiber (CF) and 0.13% ether extract (EE) on a dry matter (DM) basis (Khempaka et al., 2009). Since cassava pulp still remains high in starch content, therefore the pulp has widely used as an alternative energy source in animal diets (Khempaka et al., 2009; Khempaka et al., 2014). However, the fresh pulp also contains high moisture content approximately 70-80% which can cause a problem of drying in raining season. The combination of fresh cassava pulp and dried Napier Pakchong grass may help to resolve this problem. Napier Pakchong grass is fast-growing grass and widely produced in Nakhon Ratchasima area. This grass yield 70-80 tons/rai/year and contains 10-12% protein on DM basis. Therefore, the objective of this study was to evaluate the effects of mixed cassava pulp and Napier Pakchong grass on nutrient digestibility and retention in laying hens.

Materials and methods

Preparation of mixed cassava pulp and Napier Pakchong grass

Fresh cassava pulp and Napier Pakchong grass were used in this study. Fresh cassava pulp, obtained from the Korat Flour Industry Co., Ltd, Nakhon Ratchasima, Thailand. Fresh Napier Pakchong grass aged 40-45 days after cutting was dried at 55-60 °C for 2 days. The dried Napier Pakchong grass was then ground to pass through a 1.0 mm mesh sieve. Proximate components of cassava pulp and dried Napier Pakchong were determined according to AOAC (1990). Cassava pulp and Napier Pakchong in the ratio of 75:25, 80:20, 85:15 and 90:10 were mixed and pelleted pass through a 8 mm mesh sieve and dried at 55-60 °C. The mixed pellets were used to measure nutrient composition and bulk density. It was found that the combination of cassava pulp and dried Napier Pakchong at ratio 80:20 was the most suitable to be further investigated for use as feed stuff. The nutritional composition of cassava pulp, Napier Pakchong grass and mixed cassava pulp and Napier Pakchong grass are shown in Table 1.

Table 1. Nutritional composition of fresh cassava pulp, Napier Pakchong grass and mixed fresh cassava pulp:Napier Pakchong grass at ratio 80:20 (as-fed basis).

Component	Cassava pulp	Napier Pakchong grass	Cassava pulp: Napier (80:20)
Dry matter(%)	20.00	96.52	96.95
Crud protein(%)	0.40	11.26	6.85
Crud fiber(%)	2.72	35.68	24.81
Ether extract(%)	0.03	1.69	0.56
Ash(%)	0.57	20.15	11.93

Animal and experimental design

A total of 40 ISA Brown laying hens, 34 weeks of age, were placed in individual battery cages. After a 4 days adaptation period, all hens were randomly allotted into five experimental diets (1 control and 4 mixed cassava pulp and Napier Pakchong grass diets at levels of 5, 10, 15 and 20%) to measure nutrient digestibility and retention for 10 days. All mixed cassava pulp and Napier Pakchong diets were added with fibrolytic enzymes to prevent the negative effect of high fiber content. Feed and water were provided ad libitum throughout the experimental period.

Sample collection and chemical analysis

Excreta and feed intake were collected on experimental days 7 to 10. The excreta were total collection sprayed with 5% HCl, and dried at 60 °C for 2 days. Dried excreta were pass through a 1.0 mm mesh sieve and stored at -20°C until analyses. Dry matter, organic matter, and nitrogen in diets and excreta were measured to assess their digestibilities and retention to standard methods.

Statistical Analysis

The data were analyzed by ANOVA. The experimental design was completely randomized design (CRD) and using SPSS version 16.0 (SPSS, 2004). Significant differences among treatment were assessed by Duncan's new multiple range test. A significant level of $P < 0.05$ was used.

Results and discussion

Overall, the nutrient digestibility and retention of laying hens fed diets containing cassava pulp and Napier Pakchong grass up to 20% were comparable to laying hens fed the control diet (Table 2). Although the DM, OM and ash digestibility tended to reduce as the level of mixed of cassava pulp and Napier Pachong grass increased but there were no significant differences ($P>0.05$). The combination of cassava pulp with Napier Pakchong grass can improve the CP contents compared to the regular cassava pulp (Table 1). Although the CF in mixed product was increased but it did not interfere to lower nutrient digestibility and retention. Probably due to the experimental diets were added the fibrolytic enzymes, which compose of endo-1,4- β -glucanase, endo-1,3(4)- β -glucanase and endo-1,3(4)- β -glucanase xylanase. This would help to digest CF and thus reduce the negative effect on lower nutrient digestibility (Mathlouthi et al., 2002). This indicates that the mixed cassava pulp and Napier Pakchong grass has the potential to be used as feedstuff for laying hens. In addition, it can also help to reduce the environment pollution and more efficient of utilization by-product.

Table 2. The effects of using mixed cassava pulp and Napier Pakchong grass on nutrient digestibility and retention in laying hens.

	Mixed cassava pulp and Napier Pakchong					SEM	P-value
	Control	5%	10%	15%	20%		
Digestibility (%)							
Dry matter	79.30	76.14	74.34	75.41	74.89	0.956	0.059
Ash	60.30	58.01	52.98	59.87	52.70	2.191	0.074
Organic matter	82.27	80.12	78.19	78.88	78.57	1.073	0.060
Retention (%)							
Nitrogen	59.21	58.64	57.02	58.75	59.63	2.465	0.568

References

- AOAC, 1990. Official Methods of Analysis. 15th ed. Association of Official Analytical Chemists. Washington, DC.
- Khempaka, S., W. Molee and M. Guillaume, 2009. Dried cassava pulp as an alternative feedstuffs for broilers: effect on growth performance, carcass traits, digestive organs and nutrient digestibility. *J. Appl. Poult. Res.* 18: 487-493.
- Khempaka, S., R. Thongkratok, S. Okrathok and W. Molee, 2014. An evaluation of cassava pulp feedstuff fermented with *A. oryzae*, on growth performance, nutrient digestibility and carcass quality of broilers. *J. Poult. Sci.* 50: 71-79.
- Mathlouthi, N., J.P. Lallès, P. Lepercq, C. Juste and M. Larbier, 2002. Xylanase and β -glucanase supplementation improve conjugated bile acid fraction in intestinal contents and increase villus size of small intestine wall in broiler chickens fed a rye-based diet. *J. Anim. Sci.* 80: 2773-2779.
- NRC, 1994. Nutrient Requirements of Poultry. 9th rev. ed. National Academy of Science, Washington, DC.
- SPSS, 2004. User's Guide, Version 13.0 SPSS Inc., Chicago, IL.

Effect of method of extraction of krabok oil on digestibility, milk production and milk composition in dairy cows

Panyakaew*¹ P., S. Wongsutthavas², S. Bureenok², K. Vasupen² & C. Yuangklang²

¹Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology Isan, Sakon-Nakhon Campus, Sakon Nakhon 47160 Thailand, ² Department of ²Department of Agricultural Technology and Environment, Faculty of Science and Liberal Arts, Rajamangala University of Technology-Isan, Nakhon Ratchasima, 3000 Thailand

Abstract

The objective of this experiment was to study the effect of method of oil extraction of krabok seed on nutrient digestibility, milk production and milk composition in dairy cows. Fifteen lactating Holstein-Friesian x Thai native crossbred dairy cows, with an initial milk production ranging from 10 to 15 kg/day, were used. The experiment had a randomized block design that lasted for 90 days. Cows were blocked by bloodline (HF>75% and HF<75%) and within each block cows were randomly assigned to three dietary treatments; i.e. a total mixed ration (TMR) supplemented with fresh krabok seed meal (KSM) or TMR supplemented with krabok oil (4%) obtained by means of either a cold (C-KO) or a hot (H-KO) extraction method. All cows were fed *ad libitum* and the animals had free access to water.

Dry matter intake (DMI) was significantly affected by dietary treatment and was found to be 12.3, 16.2 and 9.6 kg/day after the feeding of KSM, C-KO and H-KO, respectively. The digestibility of the ether extract was significantly different between the dietary treatments and was found to be 85, 86 and 76% of KSM, H-KO and C-KO, respectively. Statistically significant differences in milk production (kg FCM/day) were not found, but the difference in milk production between C-KO and H-KO tended to be different (P=0.090). Likewise, feed efficiency (kg FCM/kg DMI) tended (P=0.081) to be influenced by dietary treatment and was found to be 0.96, 1.00 and 0.65 after the feeding of KSM, C-KO and H-KO, respectively. Milk composition was not affected by dietary treatments and for the three dietary treatments combined values were 4.48, 2.11, 7.49 and 12.0 for the percentages of fat, protein, solids-non fat and total solids, respectively.

It can be concluded that the outcome of the current study does not provide solid evidence on the method of fat extraction in relation to milk production, but the data suggest that the cold method should be preferred.

Keywords: Dairy cows, krabok oil, fat extraction, milk production

Introduction

Krabok seeds are widely available in South-East Asian and they contain large amounts of fat and as such are of interest to be used as a fat source in animal diets by local farmers. Krabok oil is obtained from krabok seeds or barking deer's mango (*Irvingia malayana* Oliv.ex A. w. Benn) in

Thailand, which is particularly rich in C12:0 (444 g/kg) and C14:0 (437 g/kg fatty acids). Panyakaew et al. (2013a) reported krabok oil 80 and 120 mg reduced methane production in vitro. Moreover, Panyakaew et al. (2013b) found decrease rumen archaeal abundance and protozoal numbers when supplemented krabok oil 35 g/kg in feed of bulls.

In forementioned studies krabok oil obtained by means of hot extraction was used Panyakaew et al. (2013a, 2013b). Heating fat is an efficient way to alter ruminal milk fat quality when they were studied effect of the heating process of soybean oil and seed on ruminant in vitro (Troegeler-Meynadier et al., 2014). However, the extraction of oil by means of hot method is high cost for farmer. Therefore, cold method is preferred. Unfortunately, it is currently not known whether the method of oil extraction affects milk production. For obvious reasons the use of whole krabok seed is preferred under practical feeding conditions. However, to the author's knowledge effects of supplemental whole krabok seed on milk production also is not yet known. Therefore, the aim of the current experiment was to evaluate the lactational performance of dairy cows fed rations supplemented with either krabok oil extracted by means of a cold or a hot method, or whole krabok seed.

Materials and Methods

Animals and sampling

Fifteen lactating Holstein-Friesian x Thai native crossbred dairy cows, with an initial milk production ranging from 10 to 15 kg/day and similar body weights, were used. The experiment had a randomized block design that lasted for 90 days. Cows were blocked by bloodline (HF>75% and HF<75%) and within each block cows were randomly assigned to three dietary treatments; i.e. a total mixed ration (TMR) supplemented with fresh krabok seed meal (KSM) or TMR supplemented with krabok oil (4%) obtained by means of either a cold (C-KO) or hot (H-KO) extraction method. The ingredient composition of the constant components of the basal ration (TMR) was as follows (g/kg DM): cassava chips, 280; rice straw, 200; dry tomato pomace, 130; molasses, 70; rice bran, 52.0; soybean meal, 120; urea, 10; salt, 10; cotton seed, 120; di-calcium phosphate, 5; oyster meal, 5; and sulfur, 5. All cows were fed *ad libitum* and the animals had free access to water. The feed was offered twice daily so that fresh feed was available after each milking. Refusals were collected and weighed prior to the morning milking. Cows were housed individually in pens (2x4 meter per cow) and their health status was checked twice daily.

At the start and at the end of each month, milk samples were collected on Sunday evening and Monday morning and they were analyzed for their composition. Milk yield was recorded each day during the experiment.

Total collection of feces and sampling of feed were conducted at the beginning and at the end of every month of the experiment to determine the macro nutrient digestibility.

Preparation of Fats

Krabok seed was purchased from a local market. The seed was ground through pass 1-mm screen sieve of a grinding machine. Then, 1) used for krabok meal treatment. 2) The meal was extracted by the Soxhlet method using hexane (AOAC, 1975). The hexane was evaporated the residual krabok oil was used for the H-KO. 3) The krabok meal was mixed with water 50:50 and shake, applied from Hatapaet, (2002). Then, closes and leave them at the room temperature for 24 hours. The upper layer was used for C-KO.

Statistical analysis

Data were analyzed by ANOVA using a randomized block design by using Proc ANOVA data (SAS 1985). Differences between means were considered to be significant at $P < 0.05$.

Results

Feed intake

Dry matter intake was significantly affected by dietary treatment. The highest feed intake was observed when the ration was supplemented by C-KO, followed by H-KO and KSM, respectively (Table 1). Except for ether extract (EE), the macronutrient digestibility was not significantly affected by the dietary treatments. Apparent digestibility of EE was similar between KSM and H-KO but was significantly lower when C-KO was fed. (Table 1).

Milk production (kg/d) and milk composition was not affected by dietary treatments. However, 4 % fat-corrected milk production between C-KO and H-KO tended to be significant ($P = 0.090$) (Table 1).

Table 1. Effect of krabok oil extraction methods in total mixed ration on dry matter intake, digestion coefficient, milk yield and milk composition.

Items	Treatments			SEM	Bloodline	
	KSM	H-KO	C-KO		HF>75%	HF<75%
Initial weight, kg	491	493	481	15.78	0.975	0.737
Weight change, kg	15.4	16.4	10.0	11.15	0.605	0.967
Voluntary dry matter intake						
Kg/d	12.9 ^c	14.8 ^b	16.2 ^a	0.228	0.000	0.716
BW, %	2.60 ^b	2.98 ^{ab}	3.40 ^a	0.112	0.023	0.828
g/BW ^{0.75}	123 ^b	141 ^{ab}	159 ^a	4.148	0.008	0.881
Digestion coefficient, %						
DM	77.5	81.9	78.4	1.317	0.858	0.206
OM	79.5	83.5	80.3	1.198	0.874	0.200
CP	81.9	85.2	81.6	0.951	0.728	0.152
NDF	66.9	74.1	69.9	1.936	0.574	0.204
ADF	47.7	58.8	55.9	3.025	0.316	0.312
EE	85.1 ^a	86.2 ^a	76.7 ^b	0.912	0.003	0.018
Milk yield, kg/d						
Milk yield	12.5	9.39	14.8	1.332	0.479	0.167
4% FCM	12.3 ^{ab}	9.65 ^{+b}	16.2 ^{+a}	1.153	0.199	0.090
Milk composition, %						
Fat	4.34	4.32	4.76	0.179	0.341	0.586
Protein	2.08	2.06	2.20	0.036	0.278	0.260
Solids-not-fat	7.47	7.50	7.51	0.048	0.784	0.932
Total solids	11.8	11.8	12.3	0.182	0.313	0.182
Fat/Protein	2.08	2.12	2.16	0.086	0.112	0.930

^{a, b, c} Mean in the same row with different superscript differ ($P < 0.05$)

^{+a, +b, +c} Mean in the same row with tended to different superscript differ ($P < 0.10$)

Discussion

The high level of feed intake that was observed after the feeding of rations supplemented C-KO, is in line with data provided by Palmquist and Conrad. (1978) study high fat rations for dairy cows found the hydrolyzed fat 5.7 was highest feed intake but not difference on milk composition, but they found Jerseys Jerseys produced was the most fat-corrected milk per unit metabolic body size in diet hydrolyzed fat 10.8 %., which Grummer (1991) reported that milk fat alteration is dependent on the level of lipid supplementation. Therefore, the current study shows high in feed intake, in the mean time they were high on fat level intake. However, the lower EE digestibility after feeding C-KO underlying reason cannot be easily explained because all cows were except similar % fat for EE digestibility. Nevertheless, it might be related to structure of fatty acid composition in fat that will study in the future.

It can be concluded that the outcome of the current study does not provide solid evidence on the method of fat extraction in relation to milk production, but the data suggest that the cold method should be preferred.

Acknowledgements

This experiment was funded by National research council of Thailand 2011, Rajamangala University of Technology Isan, Natural Research faculty.

References

- Panyakaew, P., G. Goel, M. Lourenço, C. Yuangklang and V. Fievez, 2013a. Medium-chain Fatty acids from coconut or krabok oil inhibit in vitro rumen methanogenesis and conversion of non-conjugated dienoic biohydrogenation intermediates. *Anim. Feed Sci. Technol.* 180: 18-25.
- Panyakaew, P., G. Goel, C. Yuangklang, N. Boon, J. TH. Schonewille, W.H. Hendriks & V. Fievez, 2013b. Effect of supplementing coconut or krabok oil, rich in medium chain fatty acids on ruminal fermentation, protozoa and archaeal population of bulls. *Animal*. 1950 – 1958.
- Paliviquist, D. L. and H. R. Conrad, 1978. High Fat Rations for Dairy Cows. Effects on Feed Intake, Milk and Fat Production, and Plasma Metabolites. *J. Dairy Sci.* 890-901.
- Grummer, R.R., 1991. Effect of feed on the composition of milk fat. *J. Dairy Sci.* 74: 3244-3257.
- Trogeler-meynadier, A., S. Puaut, Y. Farizon, and F. Enjalbert, 2014. Effects of the heating process of soybean oil and seed on fatty acid biohydrogenation in vitro. *J. Dairy Sci.* 97:5657-5667.
- AOAC, Association of Official Analytical Chemists, 1975. *Official Methods of Analysis Nutrition*. Academic Press Inc., San Diego, CA.
- Hatapaet, K., 2002. Coconut oil extraction by natural way. *Agriculture Nature J.* 15-17.

Supplementation of different sources of nitrogen and its effects on rumen microbial biomass and *in vitro* feed degradability parameters

Marjuki^{1,*}, Kusmartono¹ & Daqaqa, A.L.R².

¹Faculty of Animal Husbandry, University of Brawijaya, Malang- 65145, Indonesia

²M.Sc. Student, Postgraduate Program, Faculty of Animal Husbandry, University of Brawijaya, Malang- 65145, Indonesia

Abstract

Rumen microbes have essential roles in supplying nutrients for the host animal, hence, the growth and activities of the rumen microbes affect significantly to ruminant animal performance. Non-protein nitrogen (NPN) that is easily converted to ammonia in the rumen is as main source of nitrogen for rumen microbes to grow. However, the efficiency of the ammonia utilization by the microbes must depend on some factors including the nitrogen sources. This experiment aimed to determine the effect of different sources of nitrogen on *in vitro* rumen ammonia concentration, rumen microbial biomass synthesis, apparent and true feed degradability. Completely randomized block design of six treatments and three replications were used. The treatments were the use of different form of nitrogen sources to supplement control diet (rice straw and concentrate at 1:1 ratio in DM basis). The nitrogen sources were only urea, ammonium sulphate (ZA), or NPK fertilizer, combination of urea, ZA and NPK (1:1:1 ratio N basis), and combination of urea, NPK, ZA at a total of N:P:S ratio of 12:2:1. The amount of nitrogen sources added was set to be equivalent to an addition of ammonia concentration into the rumen liquid as much as 2.5 mg NH₃-N per 50 ml rumen liquid. Supplemented diets with different sources of nitrogen showed higher feed degradability, gas production, microbial biomass synthesis, and ammonia concentration than control diet. Among the nitrogen supplemented diets, diet supplemented with mixed of urea, NPK, ZA at N:P:S ratio of 12:2:1 showed the highest total microbial protein synthesis (11.2 g microbial N), efficiency of microbial protein synthesis (57.0 g microbial N/ kg FOMR), ammonia concentration (150.7 mg NH₃-N/l), and feed degradability (61.9%) next to ZA supplemented diet. While, NPK supplemented diet showed the lowest total microbial protein synthesis (10.2 g microbial N), efficiency of microbial protein synthesis (45.6 g microbial N/ kg FOMR), ammonia concentration (111.1 mg NH₃-N/l), and feed degradability (57.6%) that were close to the control diet. Supplementation of mixed nitrogen sources (urea, ZA, and NPK at total N:P:S ratio of 12:2:1) to diets should be used to support high rumen microbial growth and increase nutrient supply for higher ruminant productivity.

Key words: urea, ammonium sulphate, NPK, microbial protein synthesis, degradability

*Corresponding author: marjuki@ub.ac.id

Introduction

Rumen microbes have very important or even essential role for ruminant. The microbes help in utilization of low quality feeds especially fibrous feeds and in conversion of non-nitrogen protein into high quality microbial protein. Both processes cannot eventually be done by the ruminant itself without any help of rumen microbes. By those two processes, hence, rumen microbes supply

most of nutrients for the host animal. A major source of energy and protein for ruminant are supplied by end products of fibrous feed fermentation done by rumen microbes, and microbial rumen microbial protein flowing to the small intestine, respectively. These roles must depend mostly on rate of microbial growth in the rumen, their composition and activities. For their optimal growth, rumen microbes require optimal rumen conditions and supply of nutrients including carbohydrates and carbon skeleton, protein or nitrogen, vitamin, minerals and water. To ensure the optimum rumen microbial growth, all of the nutrients must be available in right time and in most appropriate proportion.

The unique of the rumen microbes is that they have the ability to utilize non-protein nitrogen (NPN) as their nitrogen source for their growth. Bondi (1987) reported that rumen microbes use rumen NH_3 in the range of 50 to 80% for their growth, especially in the synthesis of microbial protein. However, in the synthesis of protein, NH_3 does actually supply only amine groups in amino acids structure, hence it is required some other components to synthesize amino acids or protein, including energy, carbon skeleton, sulfur, and phosphor. Sulfur and phosphor are also essential element for rumen microbial growth, especially when the microbes utilizes NH_3 for amino acids synthesis during their growth. Sulfur is used for the formation of sulfur containing compounds in the microbial cells such as essential amino acids cysteine and methionine. Phosphor is used by rumen microbes for synthesis of DNA, RNA, ATP, membranes and protein synthesis. N:S ratio required for rumen microbial growth is 12:1 (Preston & Leng, 1987) and N:P ratio is 6:1 (Schwab et al., 2005), hence based on those ratios, N:P:S ratio required for rumen microbial growth should be 12:2:1. Thus, it is important to evaluate the use of compounds which contain not only nitrogen, but also either sulfur or phosphor to supply nutrients for better rumen microbial growth and feed degradability.

Methodology

The study was conducted in the Laboratory of Animal Feed and Nutrition, Faculty of Animal Husbandry, University of Brawijaya, Malang. Indonesia using *in vitro* gas techniques (Blümmel et al., 1997^a) and completely randomized block design of six treatments and three replications. The treatments were supplementation of three different N sources and their combination at amount to reach an additional of NH_3 -N concentration in the rumen liquid as much as 50 mg NH_3 -N/l or equals to 2.5 mg NH_3 -N/50 ml rumen liquid (Satter & Slyter, 1974). The N sources were urea, ammonium sulphate (ZA), NPK fertilizer. The treatment details were **P0** = 250 mg rice strew + 250 mg concentrate as fed basis; **P1** = P0 + urea (5.4 mg); **P2** = P0 + NPK (15.0 mg); **P3** = P0 + ZA (11.8 mg); **P4** = P0 + urea (1.8 mg) + NPK (5.0 mg) + ZA (3.9 mg) to get N supply ratio from the three sources = 1:1:1; **P5** = P0 + urea (4.2 mg) + NPK (0.2 mg) + ZA (2.7 mg) to get N:S:P supply ratio of 12:2:1. All feeds in the treatments were put in syringes separately, added with 50 ml buffered rumen liquor and incubated in incubator for 48 hours at 39°C Parameters measured were gas production and DM degradability *in vitro*, NH_3 concentration, microbial biomass (Blümmel et al., 1997^b). The data were subjected to analysis of variance followed by Duncan Multiple Range Test.

Results and Discussion

Data of *in vitro* gas test parameters of control and different source of nitrogen supplemented diets were presented in Table 1. It is shown in Table 3 that all supplemented diets with different sources of nitrogen showed higher feed degradability, gas production, microbial biomass synthesis, and ammonia concentration than control diet. Among the nitrogen supplemented diets, diet supplemented with mixed of urea, NPK, ZA at N:P:S ratio of 12:2:1 (P5) showed the highest total microbial protein synthesis (11.24 g microbial N), efficiency of microbial protein synthesis (57.03 g microbial N/ kg FOMR), ammonia concentration (150.73 mg NH_3 -N/l), and DM degradability

(61.87%) next to ZA supplemented diet. While, NPK supplemented diet showed the lowest total microbial protein synthesis (10.25 g microbial N), efficiency of microbial protein synthesis (45.60 g microbial N/ kg FOMR), ammonia concentration (111.06 mg NH₃-N/l), and feed degradability (57.64%) that were close to the control diet.

Table 1. Total and efficiency of microbial protein synthesis, ammonia concentration, in vitro true degradability, and gas production at 48 hrs incubation of control and treatment diets

Treatment	Total MPS* (g microbial N)	Efficiency of MPS* (g microbial N/kg FOMR)	NH ₃ Conc. (mg NH ₃ -N/l)	True degrad. (%)	Gas production (ml/500 mg DM)
P0	10.34 ^{ab}	45.41	105.96 ^a	57.17 ^a	71.33 ^a
P1	11.10 ^c	46.98	119.00 ^a	59.57 ^c	81.66 ^c
P2	10.25 ^a	45.60	111.06 ^a	57.64 ^{ab}	79.55 ^{bc}
P3	11.59 ^{cd}	47.15	146.76 ^b	61.91 ^d	74.10 ^{ab}
P4	11.08 ^{bc}	47.80	148.18 ^{bc}	59.05 ^c	77.87 ^{bc}
P5	11.24 ^{cd}	57.03	150.73 ^{bc}	61.87 ^d	77.22 ^{abc}
Sign.	***	Ns	***	**	***

MPS = Microbial protein synthesis, FOMR = Fermented organic matter in the rumen, ns : not significantly different, **: significant different, ***: highly significant different

As stated by Preston & Leng (1987), Schwab et al. (2005), and other researchers, it was agreed that rumen microbes require N, P, and S as a part of their substrates for their growth. Sulfur is one of important mineral to support rumen microbial growth, especially when ruminants are fed large amounts of non-protein nitrogen. Sulfur is utilized in synthesis of sulfur containing essential amino acids, methionine and cystine. In addition phosphorus is also essentially required for the growth of rumen microbes. Phosphor is mainly used as main component of cell's nucleic acid DNA and RNA as well as ATP. Hence, limited intake of sulfur may restrict rumen microbial growth (Pathak, 2008).

Conclusion

Supplementation of mixed nitrogen sources from urea, ZA, and NPK at total N:S:P ratio of 12:2:1 should be used to support higher rumen microbial growth increase nutrient supply for ruminant productivity.

References

- Bondi, A.A. 1987. Animal Nutrition. First Edition. John Wiley and Son, Ltd. New York, Chicester, Brisbane, Toronto, Singapore.
- Blümmel, M., H.P.S. Makkar, and K. Becker. 1997^a. *In vitro* gas production: a technique revisited. J. of Anim. Phy. and Anim. Nut. 77:24-34.
- Blümmel, M., H. Steinam, and K. Becker. 1997^b. The relationship between *in vitro* gas production, *in vitro* microbial biomass yield and ¹⁵N incorporation and its implications for the prediction of voluntary feed intake of roughages. British J of Nut 77, 911-921.
- Pathak A. K., 2008. Various factors affecting microbial protein synthesis in the rumen, Veterinary World, V.1 (6): 186-189.
- Preston. T. R and R. A Leng. 1987. Matching Ruminant Production System with Available Resource in the Tropics and Sub-Tropics. Penambul Books, Armidale, NSW. ISBN 0-9588290-12.
- Satter. L.D. and L. L. Slyter. 1974. Effect of Ammonia Concentration on Rumen Microbial Protein Production *in vitro*. Br. J. Nutr., 32, 199.
- Schwab, C.G., P. Huhtanen, C.W. Hunt, and T. Hvelplund. (2005) Nitrogen requirements of cattle. In: Pfeffer, E. and A.N. Hristov, Editors. 2005. Nitrogen and Phosphorus Nutrition of Cattle. CAB International, Wallingford, UK, pp. 13–70.

Effects of lycopene on hepatic metabolic and immune-related gene expressions in chickens

Jang, I. S., Y. An & Y. S. Moon*

Gyeongnam National University of Science and Technology, Jinju 660-758, Korea

Abstract

Lycopene has been known for many years for its antioxidant properties but some studies now focus on its anti-inflammatory (protective immunity) and the modulator of lipid metabolism in mammals. There is a lack of information related the action of lycopene beyond antioxidant in chickens. The present study aimed to investigate the possible interaction of lycopene on hepatic metabolic and immune-related gene expression in laying hens. A total of 48 twenty five-week-old White Leghorn hens were randomly allocated into 4 groups consisting of 4 replicates of 3 birds. Chickens were subjected to one of following treatments: Control (BD, basal diet), T1 (BD + tomato powder-containing lycopene 10 mg/kg diet), T2 (BD + micellar of tomato powder-containing lycopene 10 mg/kg diet), T3 (BD + purified lycopene 10 mg/kg diet). Chickens were fed with a basal diet or the basal diet supplemented with lycopene *ad libitum* for 5 weeks. Total RNA was extracted from the liver for quantitative RT-PCR. PPAR γ was decreased in the liver with lycopene intake ($P < 0.05$). Lycopene intake decreased a PPAR γ target genes, fatty acid binding protein (FABP) 4 and fatty acids synthase (FASN) in T2 group ($P < 0.05$). Sterol regulatory element-binding protein (SREBP) 2 and CCAAT/enhancer binding protein (C/EBP) α also down regulated in hens fed with micellar of tomato powder-containing lycopene ($P < 0.05$). However, the gene expression of carnitinepalmitoyltransferase 1(CPT-1) was not changed by lycopene treatment. The pro-inflammatory cytokines such as tumor necrosis factor (TNF) α and interleukin (IL) 6 were inhibited by supplement with lycopene ($P < 0.05$). These data suggest that lycopene may play an important role in the modulation of lipid metabolism and immune response of chickens.

Keywords: lycopene, gene expressions, metabolism, immune, chickens

* Corresponding author: ysmoon@gntech.ac.kr

Introduction

Lycopene is a non-provitamin A carotenoid and synthesized by plants and microorganisms but not by animals. Lycopene is present in high concentrations in processed tomato products such as tomato paste (Rao, 2004). Lycopene is known to act as the most potent antioxidant among carotenoids (Sahin et al., 2008; Englmaierova et al., 2011) and this action could be beneficial to poultry because free radicals are formed under stress, fast growth, high reproduction rates and intensive metabolic conditions of poultry (Englmaierova et al., 2011). It was reported that there is a preventive effect of lycopene on oxidative stress in the Japanese quail (Sahin et al., 2006). Tomato powder supplementation increased the performance of quail including feed intake, live weight gain and feed conversion under stress conditions (Sahin et al., 2008). This molecule has been reported to display anti-inflammatory effects in several models of diseases linked to inflammation (Hung et al., 2008; Feng et al. 2009). Lycopene is able to reduce the expression of genes (IL-6, IL1 β and MCP-1) involved in the inflammation of adipose tissue in mice subjected

to a stress or diet induced obesity (Gouranton et al., 2011). Lycopene is also involved in regulation of lipid metabolism-related gene expression in mice and rabbit (Tan et al., 2014; Mao et al., 2014). The aim of the present study was to determine the effect of lycopene on metabolic and immune related gene expression in chickens.

Materials and Methods

A total of 48 twenty five-week-old White Leghorn hens were randomly assigned to 1 of 4 dietary lycopene concentrations. Dietary treatments were as follows: basal diet (BD, control), T1 (BD + tomato powder-containing lycopene 10 mg/kg diet), T2 (BD + micellar of tomato powder-containing lycopene 10 mg/kg diet) and T3 (BD + purified lycopene 10 mg/kg diet). Hens were housed in commercial cages (4 cages/diet, 3 birds /cage) with *ad libitum* access to water for 5 weeks. At the end of the experiment, liver samples were collected and stored at -80 °C until the assay for quantitative RT-PCR (qRT-PCR). First-strand cDNA was synthesized from total RNAs using ImPromII reverse transcriptase system (Promega). qRT-PCR were performed using iQTMSYGn Green supermix (Bio-Rad) in the iCycler real-time PCR detection system (Bio-Rad). The results of the qRT-PCR for gene transcripts were analyzed by the $2^{-\Delta\Delta Ct}$ calculation.

Results and Discussion

PPAR γ expression were decreased in the liver with lycopene intake ($P<0.05$). Lycopene intake decreased a PPAR γ target genes, fatty acid binding protein 4 (FABP4) and fatty acids synthase (FASN) in T2 group ($P<0.05$). SREBP2 and C/EBPa also down regulated in hens fed with micellar of tomato powder-containing lycopene ($P<0.05$). However, the gene expression of carnitinepalmitoyltransferase 1(CPT-1) was not changed by lycopene treatment. The pro-inflammatory cytokines such as tumor necrosis factor (TNF) α and interleukin (IL) 6 were inhibited by supplement with lycopene ($P<0.05$) (Table 1). Lycopene decreased pro-inflammatory cytokines and such an effect could prevent or attenuate the prevalence of pathologies. These data suggest that lycopene may play an important role in the down-regulation of lipid metabolism-related genes and may inhibit inflammation in chickens.

Table 1. Effect of lycopene on the mRNA expression levels of lipid metabolism genes in the liver using qRT-PCR.

Item	Treatment								value
	C		T1		T2		T3		
	ΔCt	$2^{-\Delta\Delta Ct}$	ΔCt	$2^{-\Delta\Delta Ct}$	ΔCt	$2^{-\Delta\Delta Ct}$	ΔCt	$2^{-\Delta\Delta Ct}$	
FASN	-0.10±0.13 ^b	1.00	-0.02±0.49 ^b	0.95	0.79±0.07 ^a	0.54	-0.32±0.44 ^b	1.17	0.0031
FABP4	7.53±0.42 ^c	1.00	7.85±0.40 ^c	0.80	9.52±0.08 ^a	0.25	8.79±0.15 ^b	0.42	<0.0001
PPAR γ	7.24±0.73 ^b	1.00	8.22±0.74 ^a	0.51	8.11±0.53 ^b	0.55	7.80±0.12 ^{ab}	0.68	0.0323
C/EBPa	6.11±0.34 ^b	1.00	6.35±0.23 ^{ab}	0.85	6.68±0.09 ^a	0.67	6.16±0.27 ^b	0.96	0.0003
SREBP1	7.37±0.81 ^a	1.00	7.82±0.17 ^a	0.73	7.89±0.24 ^a	0.70	7.36±0.18 ^a	1.01	0.2275
SREBP2	3.18±0.13 ^b	1.00	3.40±0.31 ^{ab}	0.86	3.58±0.25 ^a	0.76	3.33±0.20 ^{ab}	0.90	0.0359

The values (means, n=4) are ΔCt , which is represented as the Ct of each target gene corrected by Ct of the control gene (RPL27).

The fold difference in the relative expression of the target gene was calculated as the $2^{-\Delta\Delta Ct}$.

^{a,b,c} The different superscript letters in the same row indicate significant differences ($P<0.05$).

References

- Englmaierova, M., I. Bubancova, T. Vit and M. Skrivan, 2011. The effect of lycopene and vitamin E on growth performance, quality and oxidative stability of chicken leg meat. *Czech J. Anim. Sci.* 56: 536-543.
- Feng, D, W. H. Ling and R. D. Duan, 2010. Lycopene suppresses LPS-induced NO and IL-6 production by inhibiting the activation of ERK, p38MAPK, and NF-kappaB in macrophages. *Inflamm. Res.* 59(2): 115-121.
- Gouranton, E., C. Thabuis, C. Riollot, C. Malezet-Desmoulins, C. El Yazidi, M. J. Amiot, P. Borel and J. F. Landrier, 2011. Lycopene inhibits proinflammatory cytokine and chemokine expression in adipose tissue. *J. Nutr. Biochem.* 22(7): 642-648.
- Hung, C. F., T. F. Huang, B. H. Chen, J. M. Shieh, P. H. Wu and W. B. Wu, 2008. Lycopene inhibits TNF-alpha-induced endothelial ICAM-1 expression and monocyte-endothelial adhesion. *Eur. J. Pharmacol.* 586: 275-282.
- Mao, M., H. Lei, Q. Liu, R. He, Z. Zuo, N. Zhang and C. Zhou, 2014. Lycopene inhibits neointimal hyperplasia through regulating lipid metabolism and suppressing oxidative stress. *Mol. Med. Rep.* 10(1): 262-268.
- Rao, A.V., 2004. Processed tomato products as a source of dietary lycopene: bioavailability and antioxidant properties. *Can. J. Diet Pract. Res.* 65(4): 161-165.
- Sahin, K., M. Onderci, N. Sahin, M.F. Gursu, F. Khachik and O. Kucuk, 2006. Effects of lycopene supplementation on antioxidant status, oxidative stress, performance and carcass characteristics in heat-stressed Japanese quail. *T. Therm. Biol.* 31: 3307-3312.
- Sahin, N., C. Orhan, M. Tuzcu, K. Sahin and O. Kucuk, 2008. The effects of tomato powder supplementation on performance and lipid peroxidation in quail. *Poult. Sci.* 87(2): 276-283.
- Tan, H. L., N. E. Moran, M. J. Cichon, K. M. Riedl, S. J. Schwartz, J. W. Erdman, D. K. Jr, Pearl, J. M. Thomas-Ahner and S. K. Clinton, 2014. β -Carotene-9',10'-oxygenase status modulates the impact of dietary tomato and lycopene on hepatic nuclear receptor-, stress-, and metabolism-related gene expression in mice. *J. Nutr.* 144(4): 431-439.

The effects of different altitude to adaptability, feed consumption and weight gain's lactating ettawa's cross bred

Rachmawati, A.¹, H. Nugroho² and T. D. Putra¹

¹Animal Physiology Laboratory, Animal Production Department, Faculty of Animal Husbandry, University of Brawijaya, Jalan Veteran Malang-65144, East Java, Indonesia, Beef Cattle and Drought Laboratory, Animal Production, ²Department, Faculty of Animal Husbandry, University of Brawijaya, Jalan Veteran Malang-65144, East Java, Indonesia

Abstract

Livestock productivity is influenced by genetic and environmental factors. Genetic factors contribute 30% and environmental factors contribute 70%. Environmental factors, including: rearing management, feed, and livestock shelter (different lands or altitude). The difference in altitude could be expected to affect the productivity of livestock, especially lactating ettawa's cross bred (LEC) goat, including: adaptability, feed consumption and body weight gain. This research was conducted at the LEC Goat Ranch Paciran, Paciran Sub District, Lamongan Residence, Indonesia as lowland (2 m above sea level) and Agus Farm, Bumiaji, Batu, Indonesia as a highland (800 m asl). The purpose of this study was to compare the adaptability response (Heat Tolerance Coefficient-*HTC*), feed consumption and daily body weight gain in different land. The research materials in each land were 10 ewes with 2-3 years of age and 4-9 months of lactation. The research method used descriptive by direct observation. Data were analyzed by unpaired t-test. The results indicated that the different land did not affect the value of the *HTC*, feed consumption, and daily body weight gain.

Keywords: lowland, highland, respiratory rate, body temperature, enviromental temperature

**Corresponding author: achadiahub@gmail.com*

Introduction

Lactating ettawa's cross bred goat (LEC) is the crossing between Ettawa and a local goat well adapted to the environment in the highlands or lowlands and with a good reproduction index, ie 1.65 lamb/ewe/year (Sodiq, 2001). Female's body weight reaches 70 kg (Susilorini et al., 2008) and is capable of producing approximately 1.5 liters of milk/ewe/day (Musofie, 2000). One LEC farm center in Indonesia in East Java has a diverse topography of the area from the coastal areas of the sea, hills up to the mountains, and most of the area is at an altitude of between 0-400 m above sea level (asl) (Widada et al., 2013). This makes the region in East Java divided into two lands, the lowlands (Village Paciran Lamongan, 2 m asl) and the highland (BumiAji, Kota Batu, 800 m asl). Altitude differences affect on the ambient temperature and adaptation of livestock, so they have to maintain ideal body temperature. Monstma (1984) stated that the ambient temperature that was high or too low causes liverstock to undergo heat or cold stress. Livestock will experience heat stress, when the Heat Tolerance Coefficient (*HTC*) was more than 2 (two), while cold stress occurs in cattle with *HTC* value of less than two. Their stress caused less convenient, stress and dehydration affect to decline in feed intake, and contributes to a decrease in livestock productivity. Widada, et al. (2013) reported that the altitude affects the Limpo heifer's body temperature, but it did not affect the respiratory frequency and the value of *HTC*. Efendi et al., (2010) reported that the *HTC* value of LEC in intensive rearing was in the range of 2, while the value of semi-intensive system LEC pre and post-weaning were respectively 2.7 and

2.8. Research on the value of LEC at different altitudes was still limited so that through this research the influence of altitude on the value of HTC, feed consumption and body weight gain of LEC could be elucidated.

Materials and Methods

The material used in this study was 20 LEC with details of 10 LEC in highland and 10 animals in the lowlands. Goats were grouped according to age (2-3 years) and body weight (20-25 kg) and fed with fermented dried water spinach (lowland), dried water spinach (plateau) as a source of fiber and pollard as supporting feed. The method used was descriptive with direct observations at both locations. Observations value of HTC every two times a day at most low ambient (morning) and the highest (noon) temperature. Sampling using purposive sampling, which samples based on certain criteria (Widada et al., 2013), namely age 2-3 years old and BW 20-25 kgs.

Results and Discussion

Effect of altitude on HTC value of lactating ettawa's cross bred at different altitudes above sea level impact on the environment was positively correlated with HTC of LEC. HTC LEC grades in the lowlands was 2.13 ± 0.04 ; while the value of highlands was 2.10 ± 0.04 . The data showed that LEC in both lands experiencing heat stress (HTC's value above two). Monstma (1984) stated that livestock that experiencing heat stress will be reflected in the response body temperature and respiratory rate. Livestock have high levels of resistance to heat and if both the HTC value equal to two, meaning that animals undergo heat stress if the value of HTC was more than two, and the livestock experiences cold stress if the value of HTC is less than two. Statistical analysis showed that the different altitude did not affect the value of HTC LEC in lowland and highland, because LEC on both altitude had the same heat stress ($HTC > 2$). Ruminants were prone to heat stress caused by microbial activity in the rumen (Philips, 2001). Busono (2007) explained that all livestock homeotherm trying to maintain body temperature in a constant state by way of regulation of body temperature remained active when the temperature was hot or cold environments. Altitude influence on feed consumption and body weight gain LEC Differences in altitude were expected to affect LEC's body weight gain, especially relating to the availability of feed. LEC have adapted to the environment of each birth, including the feed given. LEC in lowland and highland fed in the form of dried water spinach as raw fiber and pollard as supporting protein needs. Based on the laboratory test results were given feed and concentrate on a different altitude had met the nutritional needs of LEC, especially fiber and crude protein (Tables 1).

Table 1. Results of laboratory tests of feed given in the lowlands and highlands

Materials*)	Content of the feed (%)				
	Dry matter	Ash	Crude Protein	Crude Fiber	Crude fat
Lowland					
Pollard	84,85	6,21	20,73	8,59	5,24
Fermented dried water spinach	62,96	17,21	13,71	30,99	3,09
Highland					
Pollard (highlands)	86,40	-	19,02	8,05	5,12
Dried water spinach	58,07	-	8,48	36,59	2,95

Measurement of LEC's body weight gain in lowland obtained was 45.97 ± 12.25 g and the highlands was 49.25 ± 12.82 g. Results showed that body weight gain on the highland was 3.28 g/head/day as compared to in the lowlands. Statistical analysis showed that the different altitude did not affect the value of LEC. This was caused by the feed given to the LEC in lowland and highland during the study using a dried water spinach and concentrate in the form of pollard, in addition to the same feed, suspected LEC in the lowlands and the highlands experiencing the

same stresses, so that the level of consumption feed and metabolic processes in the body were not much different. Proximate analysis indicated that administration of the dry matter of 5% of livestock's body weight had been meeting the LEC's needs. The feed was used by the body for the metabolism and maintain body temperature and respiratory rate steady (homeostasis). Williamson and Payne (1993) explained that weight gain will occur when the feed consumed exceed the basic necessities of life, then the excess of nutrients will be converted into sinew and fat. The indirect effect of differences in micro and macro climate was the availability of forage that old fast and cause high crude fiber, while the direct effect, for example the occurrence of heat or cold stress, so that livestock suffering from stress or feel uncomfortable livestock that decreased in feed consumption, production (weight) and reproductive (Widada et al., 2013). High content of nutrients in the feed consumed promoted growth and development of livestock, otherwise if the feed contains a low nutrient, it has increasingly inhibited the growth of livestock. Feed was a means of production which is essential for livestock, as feed serves as a growth promoter material body. The complete feed contains protein, carbohydrates, fats, water, vitamins and minerals. The number and quality of feed that will help livestock to grow, production and reproduction. The availability of adequate food both in quantity and quality as well as sustainable is one of the factors that determine the success of the business development of livestock breeding goats.

Conclusion

Altitude caused the difference in body temperature and respiratory rate, but no significant effect on the value of HTC and body weight gain. Subsequent research suggested using LEC and feed the same at a different altitude to determine accurately the adaptation.

References

- Busono, W. 2007. Keseimbangan Fisiologis untuk Optimasi Produksi Ternak. Pidato Pengukenan Guru Besar. Fakultas Peternakan. Universitas Brawijaya. Malang.
- Efendi, A. Z., Busono, W. Dan Rachmawati, A., 2010. Pengaruh Sistem Pemeliharaan Kambing Peranakan Etawah Pra dan Lepas Sapih terhadap Nilai Heat Tolerance Coefficient, Konsumsi Pakan dan Minum. Skripsi. Fakultas Peternakan. Universitas Brawijaya. Malang.
- Monstma, G., 1984. Tropical Animal Production I (Climats and Housing). T20 D Lecture Notes E400-103.
- Musofie, A., Wardhani, N.K. dan Lestari, S.B., 2000. Pengkajian Pengembangan Usaha Peternakan Kambing di Yogyakarta. Laporan Hasil Pengkajian IP2TP. Yogyakarta.
- Philips, C.J.C., 2001. Principles of Cattle Production. Head, Farm Animal Epidemiology and Informatics Unit. Departement of Clinical Veterinary Medicine. University of Cambridge. London.
- Setiawan, B., 2008. Loyalitas Pengencer sebagai Kinerja Value Drivers. Jurnal Bisnis dan Manajemen. 10 (1): 18-27.
- Sodiq, 2001. Small Ruminant Production System under Rural Area and Improving Weaning Weight. Scientific Publication of Soedirman University. Purwokerto: 27 (3): 41-52.
- Susilorini, T.E., Sawitri, M.E. dan Muharlieni, 2008. Budidaya 22 Ternak Potensial. Penebar Swadaya. Jakarta.
- Widada, A.S., Busono, W. Dan Nugroho, H., 2013. Influence of Altitude on HTC (Heat Tolerance Coefficient) Crossbreed Cattle (LIMPO) Heifer Female Before and After Concentrate Given. Mini-thesis. Faculty of Animal Husbandry. University of Brawijaya. Malang.
- Williamson, G. and W.J.A. Payne, 1993. Pengantar Peternakan di Daerah Tropis. Terjemahan oleh Darmadja, I.G.N.D. Gadjah Mada University Press. Yogyakarta.

Differential level of plasma nesfatin-1, ghrelin and leptin for onset of puberty in Murrah buffalo heifers

Gorakh Nath Prajapati¹ & Anand Laxmi²

¹Veterinary officer, U P, India ² Principal Scientist, Dairy Cattle Physiology Division NDRI, Karnal, -132001, HARYANA INDIA

Abstract

Water buffalo (*Bubalus bubalis*) is an important dairy animal, but has low reproductive efficiency when compared with the cattle species. The main problem is delay in attaining puberty. There are many factors which affect puberty. A study has to be made on some new endocrine factors like nesfatin, ghrelin and leptin which are directly or indirectly related with body weight and feed parameters, which in turn determine onset of puberty. For the present study twelve number of Murrah heifers were selected. Blood samples were collected. Hormones were estimated in the blood plasma. Animals were considered to be pubertal only when plasma progesterone level was > 1ng/ml and corpus luteum could be detected on palpation. Based on calculated dry matter intake, nutrient efficiencies were estimated. All the animals attained puberty, at the end of fourteen fortnights. Heifers of normal pubertal (NP) group attained puberty by 27 months as and those attaining puberty by 32 months of age (14 fortnights from the day of experiment) as delayed pubertal group (DP). In NP group nesfatin and progesterone concentrations were significantly higher ($P < 0.05$) and similar results were observed for nutrient efficiencies ($P < 0.001$) also, when compared with the DP group. Similarly at fifth fortnight also, all the Mean \pm SEM parameters were significantly higher ($P < 0.01$) for NP group when compared with the DP group except for ghrelin. It can be suggested that onset of puberty in buffalo heifers is also regulated by plasma nesfatin, ghrelin and leptin.

Keywords: puberty, buffalo, heifer, nesfatin, ghrelin, leptin

Introduction

In India buffaloes mostly suffer from delayed puberty, anestrus, sub-estrus, summer infertility, prolonged intercalving interval and postpartum uterine disorders (Agarwal et al., 2005). Various reports are available with regard to age at puberty in buffalo heifers (Ingole et al., 2012). Plasma nesfatin-1, ghrelin and leptin affect the activation of the hypothalamic-pituitary-gonadal axis (Garcia-Galiano et al., 2010). These hormones have been implicated in regulation of age at puberty, feed intake, body weight, energy balance and steroid hormone secretions in human and different animals; however the role of plasma nesfatin-1, ghrelin and leptin in the relation to age at which onset of puberty in Murrah buffalo heifers occurs has not been studied so far. Hence the present study has been designed to determine whether plasmanesfatin-1, ghrelin and leptin could influence age at puberty in buffalo heifers through body weight and feed parameters.

Materials and Methods

Twelve Murrah buffalo heifers within the age group ranging from 24-26 months of age with the mean body weight (BW) 290 ± 7.35 Kg were selected at random from the animal herd of National Dairy Research Institute, Karnal Haryana. None of the animals had attained puberty at the initiation of experiment. All the heifers were fed as per the standard feeding practices followed at the farm which consisted of concentrate mixture, wheat straw (40:60) and green roughages (Berseem, Maize, Oat or Jowar fodder). The period of study was from November 2013 till May 2014. Blood samples were collected and blood plasma was separated for analysis of nesfatin-1 (Raybiotech, USA) ghrelin, leptin (Cusa Biotech Co., China) and progesterone (Endocrine technologies Inc., USA) hormones using commercially available enzyme immuno assay kits. Concentration of plasma glucose (GOD-PAP Trinder's method) and nonesterified fatty acids (Shipe et al.,1980) were also estimated. Different nutrient efficiencies (Feed conversion efficiency, Crude protein conversion efficiency, Energycorrected efficiency) parameters were also estimated based on dry matter intake (DMI), body weight, crude protein and total digestibility of nutrients of feed were also estimated. The experiment continued for 14 fortnights when all the animals attained puberty and at the end of the experiment, they were divided into normal pubertal group (NP, n = 6), and delayed pubertal group (DP, n = 6) which attained puberty by 5 and 14 fortnights respectively. Heifers with an identifiable/non identifiable CL in the presence or absence of large follicles and two successive plasma samples with progesterone concentration ≥ 1 ng/ml were classified as pubertal animals. Heifers could mount and exhibit signs of heat.

Statistical analyses

The data was analyzed with unpaired t test using SAS 9.1 software package.

Results

When the animals were classified according to the age at puberty, it was observed that animals of NP group and DP group attained puberty by 5th fortnight and at the end of 14th fortnight respectively from time of initiation of experiment. Comparison of different parameters between the groups NP and DP was done accordingly for 5 and 14 fortnights and results are presented in Table 2 and 1 respectively. The significant difference between mean \pm standard error of the mean daily gain ($P < 0.01$) in body weight and concentration of plasma nesfatin-1 ($P < 0.01$), progesterone was greater ($P < 0.01$) for NP group when compared with DP group. But mean \pm sem total body weight of the DP group was higher ($P < 0.05$) and was significantly different from the NP group (Table 1). The mean concentration of plasma progesterone ($P < 0.01$) and all the nutrient efficiencies ($P < 0.001$) estimated were significantly higher at fifth and as well as at the end of fourteen fortnights ($P < 0.01$) in NP group (Table 1 and 2). Mean \pm sem Dry matter intake was however was significantly higher ($P < 0.01$) only for five fortnights tenure. Level of plasma Nesfatin, leptin was significantly greater and ghrelin was less in NP group (Table 2) at the end of five fortnights. But the estimated Mean \pm SE body weight at the end of five fortnights and at the beginning of the experiment was significantly higher ($P < 0.05$) for NP when compared with the body weight of DP group (Table 2 and 3). The mean concentration of glucose and NEFA were not significantly different at any time of study tenure. The age at puberty was recorded as 27 ± 0.49 and 32 ± 0.20 for NP and DP groups which was significantly lower ($P < 0.01$) for NP group (Table 3). The intra and inter assay CV percentage for all the hormones was $< 10\%$.

Table 1. Comparison of different parameters between NP and DP groups (after fourteen fortnights when all the animals attained puberty).

Parameters	NP group	DP group	t
Nesfatin 1(ng/ml)	1.96±0.05	1.84±0.03	2.28**
Ghrelin (pg/ml)	63.16±2.13	66±0.99	0.035
Leptin (ng/ml)	1.94±0.05	1.92±0.05	0.03
Progesterone (ng/ml)	0.85±0.09	0.66±0.04	2.09**
Body weight (kg)	346.3±8.6	363.6±8.4	1.99*
Average daily gain(kg/d)	0.61±0.02	0.48±0.02	7.28***
Glucose (mg/dl)	66.8±0.96	66.0±0.68	0.015
NEFA (umol/L)	197.58±3.67	200.4±2.20	0.026
Dry matter intake(kg/d)	8.89±0.20	8.58±0.21	0.84
FCE (%)	8.03±0.35	5.64±0.15	7.34***
CPCE (%)	65.47±2.63	46.0±1.31	7.25***
ECE (%)	13.15±0.55	9.2±0.25	6.87***

Values are expressed as Mean±SEM. **P<0.01,***P<0.001

Table 2. Comparison of different parameters when all the animals of NP group attained puberty (after five fortnights).

Parameters	NP group	DP group	t
Nesfatin 1(ng/ml)	1.96±0.05	1.75±0.01	4.24**
Ghrelin (pg/ml)	63.16±2.13	67.81±0.73	2.07*
Leptin (ng/ml)	1.94±0.05	1.75±0.02	3.43**
Progesterone (ng/ml)	0.85±0.09	0.53±0.01	3.30**
Body weight (kg)	346.3±8.6	320±1.92	2.76*
Average daily gain(kg/d)	0.61±0.02	0.47±0.01	9.99***
Glucose (mg/dl)	66.80±0.96	67.12±0.70	0.27
NEFA (umol/L)	197.58±3.67	206.93±4.04	1.71
Dry matter intake(kg/d)	8.89±0.20	7.74±0.12	5.01***
FCE (%)	8.03±0.35	6.10±0.20	4.77**
CPCE (%)	65.47±2.63	50.23±1.79	4.79**
ECE (%)	13.15±0.55	9.98±0.34	4.87**

Values are expressed as Mean±SEM. *P< 0.05, **P<0.01, ***P<0.001.

Table 3. Age and body weight of Murrah heifers.

Parameters	NP	DP	t
At the initiation of expt.			
Age (months)	26±0.47	28±0.55	0.698
Body weight (kg)	313.83±7.63	281.95±9.56	2.495*
On attaining puberty			
Age (months)	Five fortnights	Fourteen fortnights	
Age (months)	27±0.49	32±0.20	8.81**
Body weight (kg)	346.3±8.6	363.6±8.4	1.99*

Values are expressed as Mean±SEM,*P<0.05, **P<0.01. Animals were classified into respective groups at the end of fourteen fortnights of experimental period.

Discussion

The age at puberty for the normal pubertal group observed as 27 months was in confirmation with the reports of Boopathi (2013). At the end of fourteen fortnights, animals of DP group when attained puberty late at mean 32 months of age their body weights were significantly greater (P<0.05) from NP group, this suggests that higher ADG in body weight led to earlier attainment of puberty. DMI of NP group was higher at the end of five fortnights and all the nutrient

efficiencies were also significantly more. Plasma nesfatin 1 can cross blood brain barrier and can influence gonadotropin releasing hormone pathway (Patterson et al., 2011). In the DP group plasma ghrelin concentration was significantly more after 5 fortnights and decreased at fourteen fortnights, which suggests that inhibitory effect of ghrelin on gonadotropins might have been removed leading to attainment of delayed puberty for the DP group. It has the ability to modulate gonadotropin secretion and influence the time of puberty (Tena-Sempere, 2013) suggesting higher intake of feed, improved nutrient efficiencies might have influenced gonadotropin secretion which in turn might have led to earlier attainment of puberty. It is well known that ghrelin modulates feed intake, higher dry matter intake which might have reduced mean plasma ghrelin concentration at five fortnights for NP group. It can also be suggested that there can be uncoupling between feed parameters and ghrelin at the time of puberty as higher DMI and ADG is required for attainment of puberty. Effect of leptin on feed intake are central and in the present study also it was observed that with the increase in the concentration of plasma leptin, DMI, Nutrient efficiency increased and the animals consequently attained normal or delayed puberty. The higher level of plasma progesterone indicated higher luteal activity of the NP group in comparison to animals of DP group. The present study in Murrah buffalo heifers indicates differential concentration of plasma nesfatin, ghrelin and leptin in regulating onset of puberty through body weight, feed and plasma progesterone parameters.

References

- Aggarwal, S.K., Singh, S.K. and R. Rajkumar, 2005. Reproductive disorders and their management in cattle and buffalo. A review: *Ind. J. Anim. Sci.* 75:858-873.
- Boopathi, V., 2013. Studies on nutritional and hormonal interventions to improve growth and reproductive performance of Murrah buffalo heifers. Ph.D. Thesis, NDRI, deemed university, Karnal, Haryana, INDIA
- Garcia Galiano, D., V.M. Navarro, J. Roa, F. Ruiz-Pino, M.A. Sanchez-Garrido, R. Pineda, J.M. Castellano, M. Romero, E. Aguilar, F. Gaylan, C. Dieguez, L. Pinnila and M. Tena-Sempere, 2010. The anorexigenic neuropeptide Nesfatin-1 is indispensable for normal puberty onset in female rat. *J. Neurosci.* 30: 7783-92.
- Ingole, S.D., B.T. Deshmukh, A.S. Nagvekar and S.V. Bharucha, 2012. Serum profile of thyroid hormones from birth to puberty in buffao calves and heifers. *J.B.Sci.* 1: 39-49.
- Patterson, M.K. Wynne, S. Patel, K. Suzuki, J. Tadross, M. Ghatei and S. Bloom, 2011. Nesfatin stimulates the hypothalamic pituitary gonadal axis in male rats. *Endo. Abstracts* 25: pp279.
- Shipe, W.F., G.F. Senyk and K.B. Fountain, 1980. Modified copper soap solvent extraction method for measuring free fatty acids in milk. *J.D.Sci.*, 63: 193-198.
- Tena-Sempere, M., 2008. Ghrelin as a pleotropic modulator of gonadal function and reproduction. *Nature Rev. Endocrinol.* 4: 666-674.

The effect of teat seal on milk microorganism number in postpartum cows

Butprom*¹, S., P. Kupittayanant¹ & B. Likitdecharote¹

¹School of Animal Production Technology, Institute of Agricultural Technology, Suranaree University of Technology, Nakhon Ratchasima, 30000, Thailand

Abstract

The aim of the study was to examine the effect of teat seal on milk microorganism number in postpartum cows. Teat seal was inserted into teat canal during dry period for two months. During day 1- day 5 after parturition milk samples were collected for standard plate count analysis. Microorganisms were swabbed from teat orifice and stable ground floor to confirm that pathogens were present in the environment. Cows were divided into 5 groups; control (1), treated with antibiotics (2), treated with teat seal combined with 100% paraffin (3), treated with teat seal combined with 66.5% paraffin and 33.5% bismuth subnitrate (4), and treated with teat seal combined with 49.75% paraffin and 50.25% bismuth subnitrate (5). The results showed that in group 3 and 4 the number of microorganism deceased significantly when compared to group 1, but not significantly different when compared to group 2. It was also found that pathogens causing mastitis were found in both teat orifice and stable ground floor. Therefore, it can be concluded that teat seal was as effective as antibiotics and that teat seal can be used for mastitis prevention during dry period.

Keywords: teat seal, microorganism, standard plate count, dry cow.

**Corresponding author: pakanit@sut.ac.th*

Introduction

Mastitis is one of the most common diseases in dairy cows and contributes to significant economic losses in the dairy industry worldwide. The main bacteria associated with heifer mastitis are *Streptococcus uberis* and *Escherichia coli*. This is known as an environmental mastitis as the bacteria are found in mud and faces. While mastitis management programs have contributed to reduce incidence of intramammary infections and concern over the possible overuse of antibacterial increases, attention has focused on reduction of antibiotic usage and non antibiotic alternatives. A non antibiotic intramammary teat seal effectively “plugs” the teat canal preventing the pathogenic bacteria from gaining access. Previous work has shown that dry cow therapy still has a significant value in preventing new infections during the dry period and at calving (Berry and Hillerton, 2002; Hassan et al., 1999) and can reduce incidence of intramammary infections to 74 % (Parker et al., 2007). There is also an increasing interest in organic dairy production. Furthermore, it is a growing niche market that specifies that antibiotics should not be used as prophylactics. Organic milk production commands a price premium and is economically attractive. This study, therefore, used a combination of teat seal and antibiotic dry cow treatment at drying off which can reduce the number of microorganisms in milk

Materials and Methods

Animal and Teat Seal

Holstein Friesian (25 Females) of lactation 2 to lactation 5 were used in the study. Cows were divided into 5 groups; control (1), treated with antibiotics (2), treated with teat seal combined with 100% paraffin (3), treated with teat seal combined with 66.5% paraffin and 33.5% bismuth subnitrate (4), and treated with teat seal combined with 49.75% paraffin and 50.25% bismuth subnitrate (5). Teat seal was inserted into teat canal during dry period for two months.

Sample Preparation and Statistical Analysis

During day 1- day 5 after parturition milk samples were collected for standard plate count (SCP) analysis. Microorganisms were also swabbed from teat orifice and stable ground floor to confirm that pathogens were present in the environment. The data were analyzed using CRD. The data of SCP expressed as mean \pm Standard error. Analysis of variance (one-way ANOVA) was used to analyze the differences among groups. Significant differences among groups were compared using the Duncan's new multiple-range test. The results are considered statistically significant at $P < 0.05$.

Results and Discussion

Table 1 confirms that pathogens were present in the environment. Most of them were environmental microorganisms (bacteria, yeast and fungi) including *Escherichia coli*, *Streptococcus spp.* and *Staphylococcus spp.* This finding is consistent with those obtained by Eberhart (1996) showing that bacteria in the environment, such as stable housing and ground can contact to the breast causing mammary infection and that they were *Streptococcus dysgalactiae*, *Streptococcus uberis* and Coliform especially *Escherichia coli* and *Enterobacter spp.* These infections damage the lower part of the mammary ducts and spread to the tissue causing destructive mammary epithelium cells and increase the number of somatic cells in milk.

Table 1. The number of microorganisms from teat canal and environment ($\times 10^3$ colonies/ml).

Group	Selective Media	Bacterial Agents	Teat Canal	Floor
1	Nutrient Agar	Microorganisms	272	300
	EMB Agar	<i>Escherichia coli</i>	39	66
	KF Streptococcus Agar	<i>Streptococcus spp.</i>	6	26
	Staphylococci Agar	<i>Staphylococcus spp.</i>	46	86
2	Nutrient Agar	Microorganisms	192	284
	EMB Agar	<i>Escherichia coli</i>	61	102
	KF Streptococcus Agar	<i>Streptococcus spp.</i>	3	46
	Staphylococci Agar	<i>Staphylococcus spp.</i>	28	107
3	Nutrient Agar	Microorganisms	148	260
	EMB Agar	<i>Escherichia coli</i>	53	129
	KF Streptococcus Agar	<i>Streptococcus spp.</i>	16	27
	Staphylococci Agar	<i>Staphylococcus spp.</i>	36	68
4	Nutrient Agar	Microorganisms	218	284
	EMB Agar	<i>Escherichia coli</i>	65	148
	KF Streptococcus Agar	<i>Streptococcus spp.</i>	7	59
	Staphylococci Agar	<i>Staphylococcus spp.</i>	26	98
5	Nutrient Agar	Microorganisms	266	300
	EMB Agar	<i>Escherichia coli</i>	125	173
	KF Streptococcus Agar	<i>Streptococcus spp.</i>	3	56
	Staphylococci Agar	<i>Staphylococcus sp.</i>	15	87

Table 2. Effects of teat seal on the number of bacterial colonies isolated from milk.

Group	Bacteria ($\times 10^3$ colonies/ml)				
	Day1	Day2	Day3	Day4	Day5
1	12 \pm 2.98 ^a	14 \pm 1.53	10 \pm 2.43	18 \pm 6.60 ^a	20 \pm 8.04 ^a
2	10 \pm 2.00 ^{ab}	7 \pm 3.46	5 \pm 2.21	5 \pm 2.36 ^{ab}	5 \pm 2.45 ^{ab}
3	3 \pm 1.60 ^b	5 \pm 1.24	6 \pm 0.79	5 \pm 0.95 ^{ab}	3 \pm 0.67 ^b
4	7 \pm 1.61 ^{ab}	5 \pm 2.69	5 \pm 1.32	3 \pm 0.88 ^b	4 \pm 2.86 ^b
5	8 \pm 3.08 ^{ab}	7 \pm 3.61	7 \pm 3.34	7 \pm 3.34 ^{ab}	7 \pm 3.22 ^{ab}

^{a, b} Means with different superscripts in each row are significantly different at $p < 0.05$

The results showed that the number of SPC in group 3 and 4 decreased significantly ($p < 0.05$) when compared to group 1, but not significantly different when compared to group 2 (Table 2). Similarly, teat seal decreased microorganism in milk (Huxley et al., 2002). Typically, normal bacteria in raw milk should not exceed 1×10^6 CFU/ml and in retail milk were 2×10^4 CFU/ml (Boor et al., 1998). Milk is a food source for the microorganisms and the higher bacteria causing mastitis. Therefore, the bacteria multiply rapidly and when milking bacteria mixed with milk, emanating the same (Petrovski et al., 2006). From this study, teat seal may act as a physical barrier to protect teat canal from environmental microorganisms. It has also been suggested that bismuth subnitrate protected microorganism (Council on Pharmacy and Chemistry of the American Medical Association., 1914). Similarly, bismuth citrate and bismuth subcitrate could decrease *Campylobacter* in chicken intestine (Farnell et al., 2006).

Therefore, it can be concluded that teat seal was as effective as antibiotics and that teat seal can be used for mastitis prevention during dry period.

References

- Berry, E. A., J. E. Hillerton, 2002. The effect of an intramammary teat seal on new intramammary infections. *J. Dairy Sci.*, 85:2512
- Boor K.J, Brown D.P, Murphy S.C, Kozlowski S.M and Andlar D.K (1998). Microbiological and chemical quality of raw milk in New York State. *Journal of Dairy Sci.*, 81: 1743-1748. (<http://www.ncbi.nlm.nih.gov/pubmed/9684182>, 11/08/2011)
- Eberhart, J.L., and Luoma, D.L. 1996. *Truncocolumella citrina* + *Pseudotsuga menziesii*. In: Goodman, D.M., Durall, D.M., Trofymow, J.A., and Berch, S.M. (eds.). A manual of concise descriptions of North American ectomycorrhizae. Mycologue Publications, co-published by B.C. Ministry of Forests, Canadian Forest Service, Victoria B.C. Pp. CDE9.1-4.
- Farnell, M. B., A. M. Donoghue, F. Solis de los Santos, I. Reyes-Herrera, K. Cole, M. L. Dirain, P. J. Blore, K. Pandya, D. J. Donoghue. (2006). Effect of Oral Administration of Bismuth Compound on *Campylobacter* Colonization in Broilers. *Journal of Poultry Science*. 85: 2009–2011.
- Hassan, Z., R. C. Daniel, D. O'Boyle, and A. J. Frost. 1999. Effect of dry cow intramammary therapy on quarter infections in the dry period. *Veterinary record*. 145:635–639.
- Huxley, J. N., M. J. Green, L. E. Green, and A. J. Bradley. 2002. Evaluation of the efficacy of an internal teat sealer during the dry period. *J. Dairy Sci.* 85:551–561
- Parker, K. I., C. Compton, F. M. Aniss, A. Weir, C. Heuer, and S. McDougall. 2007. Subclinical and clinical mastitis in heifers following the use of a teat sealant pre-calving. *Journal of Dairy Science*. 90:207–218.
- Parker KI, Compton CWR, Aniss FM, Heuer C, McDougall S. Quarter level analysis of subclinical and clinical mastitis in primiparous heifers following the use of a teat sealant and/or an injectable antibiotic pre-calving. *J Dairy Sci*, 2008; 91:169-181.
- Petrovski K R and Emanuel S (2006). Milk composition changes during mastitis. *Internet Journal of Milk Production* 11–16
- SPSS. (2004). User's Guide. Version 13.0. SPSS Inc., Chicago, IL.
- Wilbert, M.I. (1914). Council on Pharmacy and Chemistry, American Medical Association. A Handbook of Useful Drugs. Chicago, Press of the American Medical Association. 127 pp.: 27-29.

Growth performances of PO cattle and its crossbred with European cattle (POE) maintained in different environmental conditions

Hary Nugroho & S. Suyadi

Faculty of Animal Husbandry, University of Brawijaya, Malang, Indonesia

Abstract

This study evaluated the effect of environment condition due to the different altitudes on the growth performances of PO cattle and its crossbred with European bull (POE). During August 2010 to January 2011 a survey was conducted to collect the data on birth weight, weaning weight adjusted to 105 days, average daily gain 105 days (ADG-105), yearling weight (365 days) and ADG-365 on two breeds of PO- and POE cattle farmed in two different environment condition in Nguling District, Pasuruan (5 m above sea level, 32.5°C, 59.9% RH) and Poncokusumo District, Malang (700 m above sea level, 27.6°C, 76.8% RH). The growth performances tended to be higher in POE cattle than those in PO cattle in both environment conditions of low land and high land. Similarly, cattle maintained in high land condition showed higher growth performances than those maintained in low land condition for both cattle breeds. Yearling weight of cattle maintained in low land was not significantly different for both breeds, however, in high land condition, yearling weight and ADG 365 of POE were significantly higher ($P < 0.01$) than those observed in PO cattle. There was an interaction between genetic and environmental condition, where POE cattle was more efficiently farmed in high land condition than in low land condition one.

Keywords: growth performances, PO cattle, crossbred cattle, environment condition.

Introduction

One of the famous breed of local cattle in Indonesia is Peranakan Ongole (PO) cattle. This cattle is known to have good adaptation to the tropical condition in Indonesia under high stress of temperature, humidity, poor nutrition and tick infection. Unfortunately, it showed inferior production for prepubertal growth and in adult. Farmers nowadays, tend to cross this cattle European cattle namely Limousin, that are normally reared in subtropical climate that has low adaptation to the high tropical stress condition of climate and feed resources. It is hoped that this cross can improve the growth performance of their offspring. In fact, the calf resulted from the crossing between local PO cattle and Limousin is more favourable for the farmers and market, and therefore has higher demand and higher price compared to the calf of local cattle. The crosses between female PO and Limousin bull using artificial insemination gave the LIMPO (Limousin x PO) crossbred which can presently be found in overall regions of Indonesia.

The Limpo crossbred cattle shows the combination characteristic between PO and Limousin cattle, although the phenotypic characteristics tend more towards the Limousin cattle than to PO cattle both for qualitative traits (body shape and color) and quantitative traits (body weight at weaning and body growth) (Aryogi et al., 2005). However, some factors should be considered during farming this crossbred in the tropical condition, since part of genetic of this cattle originated from temperate climate.

It is well known that East Java region consists of land with different altitudes from sea level from 0 m to 1300 m. It is considered that altitude of land on the different climate condition can influence the productive and reproductive performance of PO cattle and it crosses with temperate cattle, and it might be affected by their genetic x environment factors.

Materials and Methods

A survey study was conducted in different climate conditions of two districts i.e. Nguling, Pasuruan (± 5 m above sea level, daily temperature 32.55C, relative humidity (RH) = 60%) and Poncokusumo, Malang (>700 m above sea level, daily temperature 27 C and RH = 77%).

This study was conducted on-farm by weighing the birth weight, weaning weight corrected to 105 days, yearling body weight corrected to 365 days, and body condition score (BCS) prediction.

All animals in both locations were fed rice straws and irregularly supplemented with concentrate feed. Data obtained were analyzed using analysis of variance with unbalanced number comparison per group.

Results and Discussion

This study evaluated the effect of breed and environment climate on the growth rate of PO and Limpo cattle under farmers' rearing condition with traditional farming system. The productive traits of both breed cattle were influenced by micro-climate condition and breed of cattle (Table 1).

Table 1. Birth weight (BW), weaning weight (WW at 105 d) and growth rate of PO cattle and its crosses with Limousin (Limpo) at different altitudes.

Variable	Altitude area	PO cattle	Limpo Cattle
Birth Weight (kg)	Low land	28.437 \pm 1.780 ^a	31.857 \pm 4.450 ^b
	High land	28.800 \pm 1.303 ^a	33.631 \pm 3.612 ^b
Weaning Weight (kg) 105 days	Low land	71.773 \pm 12.123 ^a	80.897 \pm 7,124 ^b
	High land	76.769 \pm 9.308 ^c	92.17.0 \pm 4.869 ^f
Daily Gain 105 days (kg)	Low land	0.412 \pm 0.115 ^a	0.450 \pm 0.163 ^a
	High land	0.460 \pm 0.084 ^e	0.574 \pm 0.238 ^f
Body Weight 365 days (kg)	Low land	153.705 \pm 26.38 ^a	155.023 \pm 26.66 ^a
	High land	160.593 \pm 33.659 ^e	237.520 \pm 34.47 ^f
Daily Gain 365 days (kg)	Low land	0.315 \pm 0.101 ^a	0.283 \pm 0.101 ^b
	High land	0.321 \pm 0.129 ^e	0.561 \pm 0.131 ^f

The mean birth weight of calf originated from Limpo cattle was significantly higher ($P < 0.01$) than in PO cattle. In contrast, micro-climate condition and the breed x altitude interaction showed no significant difference for birth weight. The crossing program between local cattle and exotic breed is normally subjected to increase the productivity of farm animal of the offspring, although some studies showed that there were no interaction between breed of cattle and its interaction to the environment condition (Gray et al., 1978; Scott, 1979; Ebangi et al., 2002).

The average daily gain at the age of 105 (ADG-105) days of Limpo cattle was significantly higher ($P < 0.05$) than those for PO cattle. This is might be the Limpo cattle contain genetic material inherited from the large breed of Limousin cattle that has potential to express higher growth rate than those for local PO cattle. Environment condition or altitude affected significantly ($P < 0.05$) on the ADG-105 for both breeds of cattle, where the animal reared in high land showed higher ADG than those farmed in low land area. The Limpo cattle were more appropriate reared in highland area than those in lowland one.

Genetic x environment interaction was shown on the ADG-365, where in lowland area the ADG of PO cattle was higher ($P < 0.05$) than those for Limpo cattle. In contrast was for highland, the ADG of Limpo cattle was significantly higher ($P < 0.01$) than those for PO cattle. Bernabucci et al. (2010) stated that environment hot stress influence negatively to the growth

rate of cattle. Hence, Limpo inherited from temperate cattle showed higher growth rate than those for PO cattle when reared in highland area.

Conclusion

This study concluded that Limpo cattle is more appropriate reared in highlands area, since the PO cattle is in lowlands one. In farmer condition, growth rate of both cattle was significantly influenced by the interaction between breed and altitude of areas.

References

- Aryogi, Sumadi and W. Hardjosubroto, 2005. The performance of ongole grade cross cattle In low land area (a case study at kota Anyar sub district, Probolinggo district, East Java Proceeding of the national seminar on technology animal husbandry and veterinary. Bogor Indonesia.843
- Bernabucci, U., N. Lacetera., L.H. Baumgard., R.P Rhoard., B.Raonchidan and A. Nardone, 2010. Metabolic and hormonal acclimation to heat stress in domesticated ruminants animal 4:1167-1183
- Ebangi, A.L., G.J. Erasmus., D.A. Mbah., C.L. Tawahand and O. Mesine, 2002. Factors affecting growth performance in purebred gudali and two-breed synthetic Wakwa beef cattle in a tropical environment. *Revue Elev. Med. vet. Pays trop.* 55(2):149-157.
- Gray, E.F., F.A. Thrift and C.W. Absher, 1978. Heterosis expression traits under commercial beef cattle conditions. *J. Anim. Sci.* 47:370-374.
- Scott, P., 2009. Cross breeding beef cattle. Virginia cooperative extension. Virginia state university. Publication 400-805

Morphological studies of thyroid gland of Saidi rams fed mannan oligosaccharide supplemented diet

Daghash, MW.H. & M. N. Abd El-Ati

Faculty of agriculture, Animal production department, Assiut University, Assiut, Egypt

Abstract

Eighteen Saidi rams were used in this trial to study the impact of additive mannan oligosaccharide (MOS; activeMOS[®]) on morphological studies of thyroid gland. MOS are commercially available as BioMos[®], which is a nutritional supplement manufactured by MOS[®] Matrix nutrition, LLC, USA was used in this experiment. Animals were randomly divided into three equal groups. The initial average live body weight values were 24.00, 24.08 and 24.17 kg for groups 1, 2, and 3 respectively with 8 months age. The first group did not receive MOS and served as a control group, while the second and third groups were supplemented with 2 and 4 g/kg diet MOS and served as a MOS¹ and MOS² groups, respectively. At the end of the experimental period, lasted for 6 months, final average body weight values were 44.17, 48.50 and 45.83, respectively. Five animals from experimental groups were slaughtered. Thyroid glands were removed after slaughtering, cleaned carefully of extraneous tissues and prepared for histological studies. The data revealed that the overall mean of follicular diameter increased in dietary MOS1 rams compared with MOS2 and the control. Supplementation of MOS1 increased the number and size of follicular cells (height, length and width) than MOS2 and control groups. Consequently, it appears from the present study that the dietary of MOS improved the activity of thyroid gland. Moreover, MOS inclusion at 0.2% was the most effective, suggesting that MOS might be a potential type of food additive useful for the growing sheep in Upper Egypt conditions.

Keywords: morphology of thyroid gland, mannan oligosaccharides, Saidi rams

Corresponding email: wael_daghash@windowslive.com

Introduction

Oligosaccharides are promising alternatives to antibiotic growth promoters because they facilitate and support the symbiotic relationship between host and microflora (Ghosh & Mehla, 2012). Based on a large of research mannan oligosaccharide (MOS) has established itself as a one of the most important natural additives in farm animal production to provide benefits similar to antibiotic growth promoters (Sims et al., 1998). Mannan oligosaccharides are complex substances derived mainly from the cell walls of the yeast, *Saccharomyces cerevisiae* with mannose as the primary carbohydrate (Kogan & Kocher, 2007). Several benefits from the use of MOS in animals have been reported.

No available data on the effects of dietary MOS on histological studies of thyroid gland, but there is indirect effect only through the thyroid hormones on the histology of thyroid gland. In this field, Peksa et al. (2011) found that, the size and shape of the thyroid follicles structures vary in dependence on the thyroid activity (T3 and T4 hormones). Sarwar et al. (2010) found that lambs fed diets with probiotic had higher T3 and T4 levels than those fed diets without

additives. Daghash (2015) reported that rams fed MOS had elevated T3 ($P<0.05$) and T4 hormones than a control ones. Then, the present study was aimed at elucidating the effects of MOS supplemented diet on morphology of thyroid gland in Saidi rams under Upper Egypt conditions.

Materials and Methods

The experiment was conducted at the Animal Experimental Farm, Animal Production Department, Faculty of Agriculture, Assiut University, Assiut, Egypt. Eighteen Saidi rams were used in this trial to study the impact of additive mannan oligosaccharide (MOS; activeMOS[®]) on some of morphological studies of thyroid gland. MOS are commercially available as BioMos[®], which is a nutritional supplement manufactured by MOS[®] Matrix nutrition, LLC, USA was used in this experiment. Animals were randomly divided into three equal groups. The initial average body weight values were 24.00, 24.08 and 24.17 kg for groups 1, 2, and 3 respectively. The first group did not receive MOS and served as a control group, while the second and third groups were supplemented with 2 and 4 g/ kg diet MOS and served as a MOS¹ and MOS² groups, respectively. Both experimental groups were fed roughage and concentrate diets *ad libitum* during this study. At the end of the experimental period, lasted for 6 months, final average body weight values were 44.17, 48.50 and 45.83, respectively. Five animals from experimental groups were slaughtered. Thyroid glands were removed after slaughtering, cleaned carefully of extraneous tissues. Then dissected into small pieces and immediately fixed in 10% neutral formalin, and then carefully embedded in paraffin for histological examination. The samples were sectioned into slices of 5 μm thickness using rotary microtome. Sections were stained with haematoxylin and eosin (Gu & Li., 2004). The follicular diameter was measured in 50 follicles. These follicles were considered small (up to 39.9 μm) medium (40-80 μm) and large (more than 80 μm) lobules of each gland. The number of follicles was counted. The follicle size (height, length and width) was measured.

Data were statistically analyzed using general linear model (G.L.M.) procedure of SAS (2012).

Results and Discussion

The values of different quantitative parameters of the thyroid gland in control and MOS supplemented rams are showed in Tables 1 and 2. Diameter of thyroid follicles were larger in MOS1 treated rams than those of MOS2 and untreated rams in small, medium and large ($P<0.05$) ones of lobules, respectively (with high levels of thyroid hormones in MOS1 than MOS2 and control as reported by Daghash (2015). In addition, MOS1 treated rams increased significantly ($P<0.05$) number of follicular cells and the height of follicles (μm) and insignificantly the length and the width of follicular cells than MOS2 and control rams. No available data on the effects of dietary MOS on histological studies of thyroid gland, but there is indirect effect only through the thyroid hormones on the histology of thyroid gland. Peksa, et al. (2011) found that, the size and shape of the thyroid follicles structures vary in dependence on the thyroid activity (T3 and T4 hormones). Previous studies reported that, in broilers, Sohail et al. (2010) revealed that dietary supplementations of 0.5% mannan oligosaccharides increased ($P<0.05$) thyroxine (T₄) concentration compared with a control group. In addition, Sarwar et al. (2010) found that lambs fed diets with probiotic had higher T3 and T4 levels than those fed diets without additives. Daghash (2015) reported that rams fed MOS had elevated T3 ($P<0.05$) and T4 hormones than a control ones. It could be concluded from this study that MOS additives effect on the diameter and the size of the thyroid follicles indicating that the activity of thyroid gland was more active in rams fed MOS supplemented diet than unsupplemented rams.

Table 1. Diameter (μm) of thyroid glands of rams fed mannan oligosaccharide supplemented diet.

Item	Dietary treatment			SEM
	Control	MOS ¹	MOS ²	
Small	34.68	37.96	36.88	0.96
Medium	65.77	69.66	75.62	1.79
Large	90.89 ^b	110.8 ^{ab}	108.6 ^{ab}	3.89

¹MOS = Animal supplemented with 2 g /kg diet. ²MOS = Animals supplemented with 4 g /kg diet.
SEM = Standard error of means. ^{a,b} (P<0.05).

Table 2. Number of follicles cells and follicle size of the thyroid gland of rams fed mannan oligosaccharide supplemented diet.

Item	Dietary treatment			SEM
	Control	MOS ¹	MOS ²	
Number of follicular cells	22.08 ^b	25.68 ^a	23.90 ^{ab}	0.69
Height of follicles (μm)	8.20 ^b	10.69 ^a	9.68 ^{ab}	0.43
Length of follicles (μm)	110.2	122.0	118.6	13.56
Width of follicles (μm)	62.80	74.98	78.93	11.65

MOS¹ = Animal supplemented with 2 g /kg diet. MOS² = Animals supplemented with 4 g /kg diet.
SEM = Standard error of means. ^{a,b} (P<0.05)

References

- Daghash, M.W.H., 2015. The impact of using mannan oligosaccharide as growth promoter on the productive performance of male sheep in Upper Egypt. Ph.D.Thesis, Fac.Agric., Assiut University, Egypt.
- Ghosh, S. and R.K. Mehla, 2012. Influence of dietary supplementation of prebiotic (mannan oligosaccharide) on the performance of crossbred calves. *Trop. Anim. Health. Prod.* 44: 617-622.
- Gu, X. and D. Li, 2004. Effect of dietary crude protein level in villous morphology, immune status and histochemistry parameters of digestive tract in weaning piglets. *Anim. Feed Sci. Technol.* 114: 113-126.
- Kogan, G. and A.Kocher, 2007. Role of yeast cell wall polysaccharides in pig nutrition and health protection. *Livest. Sci.* 109: 161-165.
- Peksa, Z., J. Trávníček, H. Dušová, R. Konečný and Lucie Hasoňová, 2011. Morphological and histometric parameters of the thyroid gland in slaughter cattle. *J. Agrobiol.* 28(1): 79-84.
- Sarwar, M., N. Mukhtar, M. A. Shahzad and M. Nisa, 2010. Traditional versus high input feeding system: impact on nutrients intake, blood dynamics, hormonal profile, weight gain and economics in growing lambs. *Eg. J. of Sh. & G. Sci.*, 5: 127-145.
- Sohail, M.U., A. Ijaz, M.S. Yousaf, K. Ashraf, H. Zaneb, Aleem, M. and H. Rehman, 2010. Alleviation of cyclic heat stress in broilers by dietary supplementation of mannan-oligosaccharide and Lactobacillus-based probiotic: dynamics of cortisol, thyroid hormones, cholesterol, C-reactive protein, and humoral immunity. *Poult. Sci.* 89(9):1934-1938.
- SAS Institute, 2012. SAS User's Guide: Statistics. Version 9. SAS Institute Inc, Cary. North Carolina.
- Sims, M.D., P. Spring and A.E. Seften, 1998. Effect of mannan oligosaccharide on performance commercial broiler chickens. *Poult. Sci. (Suppl. 1)*: 89.

The study of extracellular hsp70 & physiological parameters, the effect of feeding improvement of the Ongole crossbred & its crossing breed

Ita Wahyu Nursita ^{*1}, Woro Busono¹, Suyadi² & Nuryadi²

^{*1}Brawijaya University, Jl. Veteran Malang 65142, East Java, Indonesia, Animal Physiology Laboratory, Faculty of Animal Husbandry; ²Brawijaya University, Jl. Veteran Malang 65142, East Java, Indonesia, Animal Reproduction Laboratory, Faculty of Animal Husbandry

Abstract

How is the expression of extracellular Hsp70 and the physiological parameters of Crossbred Ongole (CO or PO) and its crossing breed (Limousine x PO = Limpo) in low land (5m above sea level) was investigated through this study. Also, whether the improvement of feeding through giving the concentrate to the animals could reduce the expression of extracellular Hsp70 and improve the physiological performance had been identified by this study.

The conclusion of this study is that PO in low land farm yard and low feeding level had better heat tolerance than Limpo. Concentrate treatment could eliminate the effect of breed to the heat tolerance of the animals. There was a positive correlation between HTC and eHsp70 expression. There was no effect of the treatment to plasma metabolites concentration except for NEFA. After the treatment, of PO breed plasma NEFA concentration in yearling cattle were higher ($P < 0.05$) than calves (18.0 ± 8.04 vs. 15.3 ± 3.84 pg/ml). Of Limpo's, plasma NEFA concentration in calves were higher ($P < 0.05$) than yearling cattle (43.8 ± 3.39 vs. 41.9 ± 3.17 pg/ml). The range of average plasma glucose and Hsp70 concentration was 68.9-101.2 mg/dl and 2.7-3.5 pg/ml.

The suggestion of this study is that calves of PO and its crossbred could be raised in low land (± 5 m above sea level) with environment temperature of 38°C and 50 % RH until weaning period without any physiological stress. Of the yearling's, the concentrate treatment should be added with other management to increase heat dissipation from animal body such as water splashers, sprayers or sprinklers if possible.

Keywords: extracellular Hsp70, physiological parameters, ongole crossbred

**Corresponding author: iwnursita@gmail.com*

Introduction

The PO cattle faced with the threat of extinction because of the uncontrolled crossing with imported cattle such as Simmental or Limousine breed. Mostly the cattle raised traditionally by small farmers with underfed conditions, especially during the dry season. One of the strategies used for reducing the effect of hot environment to the nutrition of the animals is by increasing the energy content of the feed or simply by giving commercial concentrate. Extreme hot environment affects the animals cellularly by activating Hsf-1, increasing Hsp expression and reducing the expression and the synthesis of other proteins. The aim of this study is to assess the physiological parameters and the expression of extracellular Hsp70 of Crossbred Ongole (CO or PO) and its crossing breed (Limousine x PO = Limpo) at low land (5m above sea level) of East Java Province of Indonesia. Also to know if by giving commercial concentrate the physiological and cellular performance become better.

Materials and Methods

The study was done from 5th October till 21th November 2012. Twenty heads of PO calves and steers maintained in the small farmers in Dandang gendis Village, Nguling District, Pasuruan Regency of East Java Province were used in this study.

Methods

During the study the animals stayed in farmers' own animal housing without any management intervention except for giving commercial concentrate as much as 0,75 kg for calves and 1,5 kg for steers. The data was taken before and after treatment period (21 days). The physiological parameters data obtained were rectal temperature, respiration rate and HTC. Laboratory analysis was done in Faal Laboratory of Medical Faculty of Brawijaya University, Malang City, East Java to know plasma glucose, plasma NEFA and eHsp70 concentration using commercial kit test. Data obtained were analyzed by software Microsoft Excel and tested by anova of nested design to know if there were significant difference ($P < 0.05$).

Results and Discussion

The average ambient temperature and relative humidity before and after the treatment were 34.2-35°C and 47.2-48%; 36.0-38.0°C and 42.7-48.3%, respectively. The average initial body weights of weaned and steer were 89.4 – 133.6 kg and 142.8-162.2 kg, respectively. The main feed of the animals was untreated rice straw. The others were native grass, elephant grass (*Pennisetum purpureum*), sugar cane young leaves, young corn stalks and cornhusk.

The expression of extracellular Hsp70 was detected during the observation ranged between 2.7-3.5 pg/ml. This range was same as reported by Kristensen et al. (2004) for Friesian-Holstein cow (0.24 – 26.47 ng/ml Hsp72) but lower than of Kristensen and

Table 1. The Average Concentration of Plasma Hsp 70 (pg/ml).

Animals	Before Treatment	After Treatment
PO Calves	3.1 ± 0.42	3.5 ± 0.31
Limpo Calves	2.9 ± 0.14	3.3 ± 0.38
PO Steers	2.7 ± 0.29	3.3 ± 0.37
Limpo Steers	3.0 ± 0.13	3.5 ± 0.34

Løvendahl (2006) for heat stressed Jersey's calves age of 76.7 ± 4.5 days i.e. 0 -1.3 ng/ml of Hsp72. Statistically, there is no effect of breed or animal's age (in breed) to the concentration of Hsp70 extracellular in this experiment. Haque et al. (2012) stated that Hsp70 production started at environment temperature of 2 or 3°C higher than animal body temperature or 40°C. The average concentration of Hsp70 plasma significantly higher ($P < 0.05$) in 40, 42 dan 45°C than of 38°C of young and adult buffaloes.

Statistically, there was no plasma glucose concentration significant difference ($P > 0.05$) caused by breed or age (in breed). The average concentration was between 68.9 – 101.2 mg/dl still in normal range for ruminant (Harper, 1977).

Table 2. The Average Concentration of Plasma Glucose (mg/dl) and Plasma NEFA (pg/ml).

Animals	Glucose Before T.	Glucose After T.	NEFA Before T.	NEFA After T.
PO Calves	92.0 ± 14.9	83.0 ± 14.07	18.4 ± 5.22 ^a	15.3 ± 3.84 ^x
Limpo Calves	77.8 ± 12.26	96.6 ± 10.90	41.2 ± 4.25 ^a	43.8 ± 3.39 ^x
PO Steers	68.9 ± 34.47	101.2 ± 23.02	22.5 ± 9.43 ^b	18.0 ± 8.04 ^y
Limpo Steers	74.5 ± 11.33	79.5 ± 15.76	37.7 ± 6.83 ^b	41.3 ± 3.17 ^y

^{a,b}different superscript in the same column was significantly different ($P < 0.05$)

^{x,y}different superscript in the same column was highly significantly different ($P < 0.01$)

According to variance analysis results, there was no effect of breed but age (in breed) ($P<0.05$) to plasma NEFA concentration. Numerically, the amount of plasma NEFA concentration of Limpo's was almost twice than PO's.

There was no breed effect ($P<0.05$) to animal's rectal temperature. There was a highly significant ($P<0.01$) and significant ($P<0.05$) effect of age (in breed) to rectal temperature before and after treatment, respectively. In PO, calves had higher rectal temperature than steers. In Limpo, it was only true for before treatment.

Table 4. The average rectal temperature ($^{\circ}\text{C}$).

Animals	Rectal T Before T.	Rectal T After T.	RR Before T.	RR After T.
PO Calves	38.4 ± 0.21^a	38.7 ± 0.21^x	25.7 ± 4.19^a	28.7 ± 0.96^x
LimpoCalves	39.9 ± 0.80^a	38.8 ± 0.22^x	26.4 ± 1.52^b	28.4 ± 1.95^x
PO Steers	38.6 ± 0.22^b	39.2 ± 0.59^y	25.8 ± 2.32^a	29.5 ± 1.05^y
Limpo Steers	38.6 ± 0.15^b	39.4 ± 0.59^y	27.7 ± 2.63^b	30.9 ± 1.07^y

^{a,b} different superscript in the same column was highly significantly different ($P<0.01$)

^{x,y} different superscript in the same column was significantly different ($P<0.05$)

Statistically, there was a breed effect ($P<0.05$) to respiration rate of the animals before treatment. Crossing animals (Limpo) had higher respiration rate than PO's (26.4 ± 1.52 vs. 25.7 ± 4.19 times/ minute in calves and 27.7 ± 2.63 vs. 25.8 ± 2.32 times/ minute in steers). After treatment the difference was not exist anymore. The difference of respiration between the animals was caused by age (in breed).

According to variance analysis results, there was breed effect ($P<0.01$) to HTC values before treatment. Limpo's had higher HTC than Limpo's (2.2 ± 0.06 vs. 2.1 ± 0.18 of calves and 2.2 ± 0.13 vs. 2.1 ± 0.18 of steers). There was only age (in breed) effect ($P<0.05$) to HTC after the treatment. Steers had higher HTC than calves.

Table 7. The average HTC.

Animals	Before Treatment	After Treatment
PO Sapih	2.1 ± 0.18^a	2.2 ± 0.04^x
LimpoSapih	2.2 ± 0.06^b	2.3 ± 0.07^x
PO Setahun	2.1 ± 0.10^a	2.3 ± 0.04^y
LimpoSetahun	2.2 ± 0.13^b	2.4 ± 0.04^y

^{a,b} different superscript in the same column was highly significantly different ($P<0.01$).

^{x,y} different superscript in the same column was significantly different ($P<0.05$).

References

- Behl, Rahul, Jyotsna Behl, and B. K. Joshi, 2010. Heat tolerance mechanisms in cattle -status in zebu cattle: A review. *Indian J. Anim. Sci.* 80:9.
- Djajanegara A. and K. Diwyanto, 2002. "Development Strategies for Genetic Evaluation of Beef Production in Indonesia." *Development Strategies for Genetic Evaluation for Beef Production in Developing Countries* edited by Jack Allen and Ancharlie Na-Chiangmai ACIAR Proceedings 108.
- Wheelock, J. B., et al., 2010. Effects of heat stress on energetic metabolism in lactating Holstein cows. *J. Dairy Sci.* 93: 644-655.

Breeding Soundness Evaluation in Garut Ram

Arifiantini, I¹, D. Sudradjat² & W.M. Nalley³

¹Department of Veterinary Clinic, Reproduction and Pathology, Faculty of Veterinary Medicine, Bogor Agricultural University, Bogor 16680, Indonesia. ²Department of Animal Science, Faculty of Agricultural, University of Djuanda, Bogor Indonesia. ³Faculty of Animal Science, University of Nusa Cendana Kupang 85148, Indonesia

Abstract

Breeding soundness evaluation of the male offers predictive information on expected performance that may enhance overall herd productivity. The aim of this study was to examine breeding soundness evaluation in Garut rams. Twenty four rams with aged of 10 month to 3 years were examined at Faculty of Animals Science, Bogor Agricultural University (IPB). Clinical examination was performed with each ram. External genitalia was examined by visual assessment and palpation for testicular tone. Scrotal circumference was measured using a metal scrotal tape. Semen was collected using artificial vagina and assessed on the basis of progressive motility, sperm concentration and sperm morphology. Result demonstrated that the average body weight of the 24 rams were 29.85 ± 3.13 kg (range of 24-36 kg), scrotal circumference were 25.35 ± 2.10 cm (range of 22-28 cm). Around 33.3% (8/24) rams producing semen with a progressive motility more than 75%, 54.16% (13/24) rams ranges from 60 to 70% and less than 12.5% (3/12) demonstrated < 60% motility. Four rams demonstrated the sperm concentration > 4000×10^6 cell per ml, 13 rams ranges from 2000-4000 $\times 10^6$ cell per ml and only 1 ram demonstrate less than 2000 $\times 10^6$ cell per ml. All rams produced an excellent morphologically normal spermatozoa $> 93.82 \pm 2.03\%$.

Keyword: Breeding soundness evaluation, Garut ram

* Corresponding author: iis.arifiantinipurna@gmail.com

Introduction

Garut is one of local Indonesian germplasm sheep that is often used for competition and for general consumption. The National demand for Garut rams were quite high, thus an effort to meet that demand through increasing in population and quality should be done immediately. Improving in Garut rams quality can be done with selection of good quality and cross breeding rams. Breeding program require rams with good physic and provide healthy spermatozoa for female rams in estruses period. Thus, an evaluation for ram's performance has to be done before initiate the breeding program. One of the methods to evaluate the ram's performance is with *breeding soundness evaluation* (BSE). BSE is an evaluation method to decide the performance excellences of the male by observing the physic and reproduction performance. BSE method has been tested on several livestock such as bull (Leamaster & Duponte, 2007), buck (Bagley, 1997) and rams (Pezzanite et al., 2004). The BSE method evaluate scrotal circumference and semen quality such as motility, concentration and spermatozoa morphology (Leamaster & Duponte, 2007). The aims of this study was to examine breeding soundness evaluation in Garut rams.

Materials and Methods

Animals

Twenty four Garut rams with age ranging from 10 months to 3 years, were examined at Faculty of Animals Science, Bogor Agricultural University (IPB).

Clinical examination

Clinical examination was performed with each ram. External genitalia was examined by visual assessment and palpation for testicular tones. Scrotal circumference was measured using a metal scrotal tape.

Semen collection and evaluation

The semen was collected from the ram with the aid of an artificial vagina. Immediately after collection, the semen was evaluated for progressive motility, sperm concentration and sperm morphology. Progressive sperm motility (%) was performed by placing 10 μ L semen on a object glass and covered by cover slip pre-heated to 37 C and analyze the sample subjectively under light microscopy at 200 \times –400 \times magnification. Sperm concentration was measured using improved Neubauer hemocytometer by diluting the semen to 1:500 ratio with formal-saline on test tube and gently mix well the solution. After mixing, one drop of solution were placed in a clean and dry hemacytometer covered by cover slip and counted the sperm dispersed in the middle square and the four corner squares of the 25 squares in the grid. Number of spermatozoa was counted in 100 squares with the help of manual counter. The number of spermatozoa from 5 square covered by cover slips were multiplied by 25 x 10⁶ (Arifiantini 2012). Sperm morphology was evaluated using eosin nigrosin, 100 μ l eosin nigrosin well-mixed with 10 μ l semen, smeared at object glass by sliding with a clean another object glass. The smears were air dried and examined directly. Two smears were made from each sample. Sperm were assessed with a light microscope (Olympus CH 20) at 400x magnification. At least 200 sperms were evaluated on each smear.

Result and Discussion

Result demonstrated that the average body weight of the 24 rams was 29.85 \pm 3.13 kg (range 24-36 kg). Around 62.5% rams had body weight of 30.9 \pm 3.28 kg and this weight was reached when rams age were 1.5 years (Tabel 1). The average of scrotal circumference of garut rams were 25.35 \pm 2.10 cm (range of 22-28 cm). After rams reached one year, the scrotal circumference seems to be stable.

Table 1. Age, body weight and scrotal circumference of garut rams.

Age (years)	Number (head)	Body weight (kg)	Scrotal circumference (cm)
<1	1	28.0 \pm 0.00	22 \pm 0.00
1-1.5	15	30.9 \pm 3.28	25.50 \pm 1.92
>1.5-2	4	32.5 \pm 2.08	25.63 \pm 2.56
>2-3	4	28.5 \pm 2.65	25.38 \pm 2.5

Sperm motility is the indicator of sperm fertility. Around 33.3% (8/24) garut rams produced semen with a progressive motility more than 75%, 54.16% (13/24) rams ranges from 60 to 70% and less than 12.5% (3/12) demonstrated < 60% motility.

Table 2. Progresif sperm motility distribution of garut rams.

Sperm motility (%)	Number (head)	Percentage (%)
>75	8	33.3
>60-75	13	54.16
< 60	3	12.5
Total	24	100

Sperm concentration were defined as the number of sperm in 1 ml semen volume. The 75% (18/24) of rams demonstrated sperm concentration range from 2000 to 4000 x10⁶ cell per ml and four rams demonstrated sperm concentration of > 4000 x10⁶ cell per ml, only 1 ram demonstrate less than 2000 x10⁶ cell per ml.

Table 3. Sperm concentration distribution of Garut rams.

Sperm concentration (x10 ⁶ /ml)	Number (head)	Percentage (%)
1000-2000	1	4.16
2000-3000	13	54.16
3000-4000	5	20.8
>4000	4	16.6
Total	24	100

All rams showed an excellent normal sperm morphology. The average of normal sperm morphology was > 93.82%±2.03% range from 86.76% to 96.35%. According to Bagley (1997), ram with the age of > 14 months with the scrotal circumference <33 cm were categorized as questionable. In this study, the maximum scrotal circumference was 28 cm with the average of 25 – 35±2.10 cm. When we perform BSE according to Bagley (1997), all rams were failed to pass the test. The BSE for local ram must be adapted to the average of the rams. Garut rams more than 1 year were categorized as satisfactory if the scrotal circumference > 25 cm and categorized as questionable when less than < 25 cm. Based on semen evaluation, rams that demonstrated > 75% sperm motility were categorized as excellent, 60-75% as satisfactory and <60 % as questionable. Garut rams sperm concentration were categorized as excellent if the sperm concentration > 4000x10⁶ /ml, satisfactory if > 2000 - <4000x10⁶ /ml and questionable if < 2000x10⁶ /ml categories as questionable. Since the sperm morphology demonstrated high number of normal sperm, we were categorized garut ram with sperm morphology of > 86.76 % as satisfactory and < 86.76 % as questionable.

Based on this finding the categorization of BSE as excellent, satisfactory or questionable should be done specifically based on the breed.

References

- Arifiantini, R.I., 2012. Tehcnique of semen collection and evaluation. IPB press. Bogor Indonesia.
- Leamaster, B.R., and M.W. Duponte, 2007. Bull Power : Examination of Beef Cattle for Breeding Soundness. Cooperative Extension Service. College of tropical Agriculture and Human Resources. University of Hawai'i at Manoa.
- Bagley, C.V., 1997. Breeding soundness in rams: How to do it and how to intepret it. http://extension.usu.edu/files/publications/factsheet/AH_Sheep_13.pdf. [23 August 2011].
- Pezzanite, L., A. Bridges, M. Nearly, and T. Hutchens, 2004. Breeding soundness examinations of rams and bucks. <http://www.extension.purdue.edu/extmedia/AS/AS-599-W.pdf>. [23 August 2011].

Individual variation on the success of garut ram frozen semen production

Nalley, W.M.¹, I. Arifiantini I.², M. Nofriza² & E. Sukmawati³

¹Faculty of Animal Science, University of Nusa Cendana, Kupang 85148, Indonesia.

²Department of Veterinary Clinic, Reproduction and Pathology, Faculty of Veterinary Medicine, Bogor Agricultural University, Bogor 16680, Indonesia. ³Artificial Insemination Centre, Lembang, Bandung 40391, Indonesia

Abstract

This research aims to study the individual variation on the successful garut ram frozen semen production. The semen were obtained from four sexually mature Garut ram (Sinta, Wulung, Jabar, Batara), at Artificial Insemination Centre (AIC) Lembang, Bandung. The semen was collected using artificial vagina and evaluated macro- and microscopically. The semen was diluted with andromed, packed into 0.25 mL mini straw (Combo System, Minitube Jerman), equilibrate at cooling cabinet for 4 hours, and freeze at automatic freezing machine (Digitcool 5300 ZB 250, IMV Prancis) according to AIC standard procedures. Data were analyzed with a linear model (GLM) and Duncan 's test. The result indicated that there were differences in raw semen quality. Wulung demonstrated the highest ($P<0.05$) raw semen motility (82.50%) and Batara has the lowest raw semen motility (75.50%). There were no differences in post thawing motility (PTM) in all ram. The PTM were between 40.00 to 41.67%. The recovery rate of sperms obtained from Batara ram was significantly higher ($P<0.05$) than the others. This research concluded that there was an individual variation on the quality of Garut ram frozen semen.

Keywords: frozen semen, freezing capability, Garut ram, recovery rate

* Corresponding author: iis.arifiantinipurna@gmail.com

Introduction

Garut sheep, is one of originally small ruminant from Garut, west java. Garut sheep use as a source of meat and ram for fighting contest a cultural community of West Java. Garut sheep that have superior genetic quality is rare and very expensive, therefore some artificial insemination center (AIC) produce frozen semen of this ram for artificial insemination (AI) purposes. The successfull of AI could be influenced by many factors, including human resources such as farmer and inseminators, the female factors such as estrus cycle and age of the ewe and the male factor, in this case is the quality of the liquid or frozen semen to be used for AI.

Freezing process causes cell damage that is irreversible in the plasma membrane (Holt, 2000; Lemma, 2011), especially in the acrosomal cap (Purdy, 2006), caused by ice crystals formed during freezing (Martinez & Wallgren, 2011) as well as changes in the composition of the function of plasma membrane (Lemma 2011). Each animal has a different composition of the membrane plasma, therefore the ability of sperm to survive on the freezing process differs. Arifiantini et al. (2014) reported an effect of individuals freezing capability of goat sperm. Since they are differences in the composition of the membrane this research aims to study the influence of individuals on the success of frozen semen production in Garut Ram at Lembang Artificial Insemination Centre.

Materials and Methods

Source of Semen

Four sexually mature Garut rams age 2-3 years, body weight about 70 to 80 kg (belong to Lembang, Bandung) AIC were used as sperms sources with a total of 48 ejaculates. The Rams were kept under natural light and maintained under a uniform and constant nutrition regime, each buck fed 1 kg concentrate, 8 kg of grass, 1 kg of legume, salt lick, and water was provided *ad libitum*.

Extender Preparation

A commercially available diluent Andromed[®] (Minitube, Germany) was used in this experiment. This extender contains soybean extract with antibiotics (lincomycin, spectinomycin, tylosin, gentamycin) and glycerol (7%). One part of andromed was diluted with 4 parts of aquibidest, warmed up at 37°C water bath.

Semen collection and evaluation

The semen was collected from the ram with the aid of an artificial vagina, twice a week. Immediately after collection, the semen was evaluated macro- and microscopically including semen volume, pH, consistency, and color, mass movement, motility, and sperm concentration.

Semen Processing

Semen having volume of > 1.5 mL, sperm concentration $> 2.000 \times 10^6$ and progressive sperm motility $> 70\%$ were selected for cryopreservation. The semen was diluted with diluent to a final concentration of 200×10^6 sperm/ml. Diluted semen was loaded into 0.25 ml straws (Minitube Germany) using automatic filling and sealing machine (Combo System, Minitube Germany), equilibrated at 4°C for 3 hours and was freeze at automatic freezing machine (Digitcool 5300 ZB 250, IMV Prancis) for 9 minutes and the straws were then plunged into the liquid nitrogen and stored until thawing.

Evaluation of post-thawing quality

After 24 hours of storage the semen straws were thawed in a water bath (at 37° C for 30 second) for for futher evaluation. Sperm motility was assessed using a phase-contrast microscope (Olympus BX 53) 200 x magnification with a warm stage maintained at 37°C. A wet semen mount was made by using 5 μ L semen placed directly on a microscope slide and covered by a cover slip. Motility estimations were performed from five different microscopic fields in each sample. The post-thawing quality criterion were $< 40\%$ was bad, 40-50% was moderate, and $> 50\%$ was good.

Statistical Analyses

The study was repeated 12 times and the results were expressed as the mean \pm SEM. One way analysis of variance (ANOVA) with a subsequent Duncan test was used to compare the mean values resulting from the various individual at a significance level of $P < 0.05$. All analyses were carried out using the SPSS 18 for Windows statistical software package.

Result and Discussion

Raw semen quality

All ejaculates were collected from sexually mature ram that were of proven fertility and were undergoing regular semen collection for artificial inseminations. Therefore, it was expected that semen quality before freezing would be of a high standard. Individual variations were marked between raw semen samples used in the study; however, the qualities were in the normal range.

Table 1. Raw semen quality of Garut Ram (mean± SEM).

Parameter	Ram			
	Sinta	Wulung	Jabar	Batara
Volume (ml)	1.96±0.51 ^c	2.43±0.72 ^b	3.08±0.47 ^a	2.60±0.80 ^b
pH	6.65±0.21 ^a	6.66±0.19 ^a	6.74±0.26 ^a	6.72±0.30 ^a
Colour	Creamy white			
Consistency	tick			
Mass activity	+++	+++	+++	+++
Sperm motility (%)	79.29±4.75 ^{ab}	82.50±4.52 ^a	81.67±5.16 ^a	75.50±6.43 ^b
Velocity (1-5)	4	4	4	4
Sperm concentration (x10 ⁶)/ml	2241.43±146.23 ^{ab}	2360.00±158.40 ^{ab}	2460.00±273.64 ^a	2067.00±202.39 ^c

Mass activity scoring (0-3); 0 no mass activity, 1 poor, 2 moderate and 3 good. Different superscripts within the same row are statistically different at P<0.05

Raw semen quality demonstrated 1.96 – 2.60 ml in volume, creamy white color, tick consistency, mass activity >2, with percentage of sperm motility >70%, and sperm concentration > 2000x10⁶. There were no difference among parameters, except for sperm motility, batara ram, demonstrated the lower sperm motility as compared to others (Table 1).

Frozen semen quality of Garut Rams

Overall, the quality of post-thawing motility of sperms obtained from garut ram, demonstrated a moderate quality (41.67±0.83 to 46.25±1.09%). Sperm motilities during pre- freezing period were between 62.22±1.21 to 65.00±1.23% and no significant different among individual rams. Freeze-thawing procedure decreased the sperm motility between 32.08 to 39.17% (Table 2).

Table 2. Recovery rate of garut ram sperms after freezing (mean± SEM).

Ram name	Motility (%)			Recovery rate (%)
	Raw	Pre freezing	Post thawing	
Sinta	79.29±4.75 ^{ab}	62.14±3.23 ^a	40.36±0.34 ^{ab}	51.07±3.53 ^{ab}
Wulung	82.50±4.52 ^a	61.82±3.37 ^a	40.00±0.00 ^{ab}	48.49±2.72 ^b
Jabar	81.67±5.16 ^a	60.83±2.04 ^a	41.67±0.58 ^a	51.11±3.21 ^{ab}
Batara	75.50±6.43 ^b	61.11±2.20 ^a	40.00±0.00 ^{ab}	53.39±5.57 ^a

Different superscripts within the same columns are statistically different at P<0.05.

There was no differences was detected for post-thaw sperm motility, range from 40.00 to 41.67±0.58%. The indicator for freezing capability of sperm is not only post-thawing motility. The successful of freezing can also be seen by assessing its recovery rate (RR), by comparing the sperm motility of raw semen with post-thawing semen. According to RR value, batara showed the best freezing capability with 53.39 ±5.57% sperm recovered after freezing, even though he sperm motility of raw semen only 75.50±6.437% but the post-thawing motility was

40.00±0.00%. This value was not differ from wulung with 82.50±4.52% of raw sperm motility (Table 2). Individual differences in sperm cryo-survival were not exclusive to ram, because they have also been observed in stallion (Sieme et al., 2008), ram (D'Alessandro & Martemucci, 2003), boar (Thurston et al., 2002) and goat (Arifiantini et al., 2014). The reason for ram individual variability in cryo-survival of sperm is unknown at present, although it may have a genetic origin. Differences in specific DNA sequences have been identified between boars in which post-thaw sperm quality was classified as poor or good (Thurston et al., 2002). This research concluded that there was an individual variation on freezing capability of sperms in garut ram semen.

References

- Arifiantini, R.I., W.M.M. Nally, T. Susnawati and E. Rochmiati, 2014. Individual Variation on the Sperm Freezing Capability of Etawah Grade. The 2nd Asian-Australasian Dairy Goat Conference. P:127-130.
- D'Alessandro, A.G. and G. Martemucci, 2003. Evaluation of Seasonal Variations of Semen Freezability in Leccese Ram. *Anim. Reprod. Sci.* 79:93-102.
- Holt, W.V., 2000. Basic Aspects of Frozen Storage Semen. *J. Anim. Sci.* 62:3-22.
- Kaya, A., M. Aksoy and T. Tekeli, 2002. Influence of Ejaculation Frequency on Sperm Characteristics, Ionic Composition and Enzymatic Activity of Seminal Plasma in Rams. *Small Rum. Res.* 44:153-158.
- Lemma, A., 2011. Effect of Cryopreservation on Sperm Quality and Fertility. in: Hanafi M. Editor. *Artificial Insemination in Farm Animals*. Croatia (R): Intech.
- Martinez, H.R. and M.Wallgren, 2011. Advances in Boar Semen Cryopreservation. *Ved. Med. Int.* doi: 10.4061/2011/396181.
- Purdy, P. H., 2006. A Review on Goat Sperm Cryopreservation. *Small Rumin. Res.* 63:215-225.
- Sieme, H., R.A.P. Harrison and A.M. Petrunkina, 2008. Cryobiological Determinants of Frozen Semen Quality, with Special Reference to Stallion. *Anim. Reprod. Sci.* 107:276-292.

Efficiency of difference doses of pregnant mare's serum gonadotropin on superovulations in meat goats

Kajaysri^{1,*}, J., C. Chumchoung¹ & S. Sophon²

¹Clinic for Obstetrics and Gynecology Andrology and Artificial Insemination of Domestic Animals, ²Department of Animal Husbandry and Basic Veterinary Science, Faculty of Veterinary Medicine, Mahanakorn University of Technology, 140 Nong-chok, Bangkok 10530, Thailand

Abstract

The objective of this study was to compare the efficiency of two doses (700 IU and 1,000 IU) of pregnant mare's serum gonadotropin (PMSG) on superovulatory responses in crossbred meat goats. Twelve crossbred meat does were assigned for estrus synchronization using the intravaginal progesterone controlled internal drug release (CIDR) devices for 10 days and 125 µg prostaglandin F_{2α} injections at day 10 immediately after CIDR device removal. These twelve does were divided into two equal groups. Group 1 and Group 2 were injected with 700 IU and 1,000 IU of PMSG at 48 h before CIDR removals, respectively. After CIDR withdrawals, estrus responses were detected. In addition, on day 7 after CIDR removal, each estrus doe in both groups were examined the ovaries for superovulatory calculation. The ovulatory follicle was evaluated from the number of active corpus luteum (CL) which founded on both ovaries. The results showed 100 % of does with estrus responses in both groups. The mean estrus onsets and estrus durations of Group 1 and Group 2 were not significantly different (18.92±1.16 h and 35.42±3.06 h vs. 19.50±2.24 h and 36.08±1.46 h, respectively). The mean numbers of active CL per doe in Groups 1 and Group 2 were not significantly difference (7.17±5.46 and 9.17±3.37, respectively). On other hand, Group 1 had 50.00% superovulation rate while in Group 2 had 83.33% superovulation rate. Moreover, the does in Group 2 revealed the tendency of higher percentages of active CLs plus dominant an ovulatory follicles when compared with Group 1 (75.54±7.65% vs. 55.55±19.34%, respectively). In conclusion, the efficiency of 700 IU or 1,000 IU of PMSG on superovulatory responses obtained similar stimulations to produce number of ovulatory follicles. In contrast, the treatment with 1,000 IU of PMSG had trends of higher incidences of superovulation rate and percentage of dominant follicles than the treatment with 700 IU of PMSG in crossbred meat goats.

Keywords: CIDR, crossbred meat goat, estrus synchronization, PMSG, superovulation

*Corresponding author: jatuporn@mut.ac.th

Introduction

It widely knows that ovulation is rupture of mature follicle on ovarian surface and releasing of its contents, such as mature oocyte. The ovulation can be evaluated in the appearance of active corpus luteum on the ovary after ovulation. The ovulation rate has major influence to litter size and reproductive efficiency in goats. However, it can be increased number of ovulation and follicle for oocyte recovery by hormonal induction which is named as superovulation (Rahman et al., 2008). The superovulation protocol in goats is provided of long progesterone impregnated insertion and followed by prostaglandin F_{2α} plus gonadotropin hormone administrations at the later part of protocol. Gonadotropin hormone usually using for superovulation is pregnant mare's serum gonadotropin (PMSG) hormone (Goel and Agrawal, 2005). The PMSG is available, easily

using in several countries, low cost and practical administration like single dose hormone for superovulation (Armstrong et al., 1983). The superovulatory response to PMSG can be variable, depend on used hormone dosage (Amoah and Gelaye, 1990). The 1,000 IU of PMSG can be able to induce superovulation in goat as well (Saharrea et al., 1998).

Materials and Methods

Twelve healthy crossbred meat does with age 2 to 5 years varied in body weigh between 30 and 50 kg. They were assigned for estrus synchronization using the intravaginal progesterone controlled internal drug release (CIDR; Eazi-Breed[®], Pfizer, U.S.A.) devices for 10 days and 125 µg prostaglandin F_{2α}(Cloprostenol; Estrumate[®], Schering-Plough, U.S.A.) injections at day 10 immediately after CIDR device removals. During impregnated intravaginal CIDR, these does were allocated into two groups equally. The does in Groups 1 and 2 were injected with 700 and 1,000 IU of PMSG(Folligon[®], Intervet international, Netherlands) at 48 h before CIDR removal, respectively. After CIDR withdrawals, the does in Groups 1 and 2 were detected estrus responses with helping of fertile teaser bucks. The number of estrus does and time of estrus onset and duration in each group were recorded. In addition, on day 7 after CIDR removal, each estrus doe in both groups was examined the ovaries by laparotomy for an ovulatory follicle inspection and then superovulatory calculation. The ovulatory follicle was evaluated from the number of active corpus luteums (CLs) founded in both ovaries. The number of medium (3-5 mm), dominant anovulatory (>5 mm) follicles, active CLs and number of does that had superovulation, were recorded. The superovulatory doe was defined as the doe presenting more than 3 ovulatory follicles.

Statistical analyses

The differences between studied groups for estrus onset, estrus duration, number of 1) medium anovulatory follicles, 2) dominant anovulatory follicles, 3) active CLs and 4) active CLs plus dominant anovulatory follicles, and percentage of 1) medium anovulatory follicles, 2) dominant anovulatory follicles, 3) active CLs and 4) active CLs plus dominant anovulatory follicles were analyzed by using independent student's *t*-test while the difference of superovulation rate were detected by χ^2 test (SPSS 10.0 for Windows, SPSS Inc. Chicago, IL, U.S.A.). A probability of $P \leq 0.05$ was considered significant for all statistical analyses.

Results and Discussion

The estrus responses of does in each group were presented in Table 1. All does in groups 1 and 2 showed estrus responses. The means of estrus onsets and durations in Groups 1 and 2 were not significantly differences between both studied groups. These results were similar to earlier study (Kajaysri and Thammakarn, 2012). Ovarian responses after treatment with 700 and 1,000 IU of PMSG were presented in Table 2. The number of active CLs per doe in Group 2 was 9.17 ± 3.37 according with previous study (Saharrea et al., 1998). It was not significantly difference from Group 1 (7.17 ± 5.46). In addition, the numbers of medium, dominant anovulatory follicles and active CLs plus dominant anovulatory follicles were not difference between both experimental groups. These present results indicated that the treatment with 700 IU of PMSG could stimulate to present of ovulatory and anovulatory follicles as well as the stimulation with 1,000 IU of PMSG. According to Amoah and Gelaye (1990) explained that the PMSG can be variable to stimulate superovulatory response. Thus, it might be possible that the efficiency of difference doses between 700 and 1,000 IU of PMSG could be similar influence on superovulatory responses in crossbred meat goats. On other hand the does in Group 1 had 50% superovulation rates while 83.33% of does in group 2 had superovulation rates. Moreover, the doe in group 2 tended to present higher percentages of active CLs plus dominant anovulatory

follicles ($75.54 \pm 7.65\%$) when compared with group 1 ($55.55 \pm 19.34\%$) ($P=0.053$). These finding data showed the treatment with 1,000 IU of PMSG had the tendency of higher incidences of superovulation rate and percentage of dominant follicles than the treatment with 700 IU of PMSG in crossbred meat goats. It widely knows that human chorionic gonadotropin (hCG) hormone exhibits predominantly LH-like hormone effects and can be used for improving LH activity and increasing a number of dominant follicles to be ovulated. According to Medan et al. (2003) reported that a combination of PMSG and hCG is also worldwide used to stimulate superovulations in does. Therefore, in order to increase efficiency of PMSG with dosage 1,000 IU on superovulatory response could be able to do by using combination with hCG hormone. In conclusion, difference dosages of 700 and 1,000 IU of PMSG had similar efficiency on superovulatory responses to produce number of ovulatory follicles in crossbred meat goats. In addition, the treatment with 1,000 IU of PMSG had trends of higher incidences of superovulation rate and percentage of dominant follicles than the treatment with 700 IU of PMSG.

Table 1. Estrus responses treated with CIDR+PGF_{2α} and different doses of PMSG in goats.

Treatment group	No. of estrus does	Estrus onset (h)	Estrus duration (h)
Group 1 (700 IU, PMSG)	6/6 (100%)	18.92±1.16	35.42±3.06
Group 2 (1,000 IU, PMSG)	6/6 (100%)	19.50±2.24	36.08±1.46
P-value*		0.587	0.644

*A probability of $P \leq 0.05$ was considered significance

Table 2. Ovarian responses treated with CIDR+PGF_{2α} and different doses of PMSG in goats.

Treatment group	Ovarian responses*								
	1	2	3	4	5	6	7	8	9
Group 1	7.50 ±3.94	2.50 ±1.05	7.17 ±5.46	9.67 ±5.05	44.45 ±19.34	16.65 ±12.00	38.90 ±23.69	55.55 ±19.34	50.00% (3/6)
Group 2	4.50 ±2.07	4.17 ±3.19	9.17 ±3.37	13.33 ±1.86	24.46 ±7.65	24.74 ±22.20	50.80 ±16.17	75.54 ±7.65	83.33% (5/6)
P-value**	0.139	0.269	0.466	0.143	0.053	0.456	0.336	0.053	0.540

*1=No. of medium anovulatory follicle (3-5 mm), 2=No. of dominant anovulatory follicle (>5 mm), 3=No. active corpus luteum, 4=No. active corpus luteum plus dominant anovulatory follicle, 5=Percentage of medium anovulatory follicle, 6=Percentage of dominant anovulatory follicle, 7=Percentage of active corpus luteum, 8=Percentage of active corpus luteum plus dominant anovulatory follicle, 9=Superovulation rate

**A probability of $P \leq 0.05$ was considered significance

References

- Amoah, E.A. and S. Gelaye, 1990. Superovulation, synchronization and breeding of does. *Small Rum. Res.* 3: 63-72
- Armstrong, D.T., A.P. Pfitzner, G.M. Warnes, M.M. Ralph and R.F. Seamark, 1983. Endocrine responses of goats after induction of superovulation with PMSG and FSH. *J. Reprod. Fert.* 67: 395-401
- Goel, A.K. and K.P. Agrawal, 2005. Ovulatory response and embryo yield in Jakhrana goats following treatments with PMSG and FSH. *Trop. Anim. Health Prod.* 37: 549-558
- Kajaysri, J. and C. Thammakarn, 2012. Estrus synchronization using intravaginal medroxyprogesterone acetate (MAP), MAP plus prostaglandin F_{2α}, controlled internal drug release (CIDR) or CIDR plus prostaglandin F_{2α} in Saanen dairy goats in Thailand. *Kasetsart J. (Nat. Sci.)* 46: 71-79
- Medan, M.S., G. Watanabe, K. Sasaki, S. Sharawy and N.P. Groome, 2003. Ovarian dynamics and their associations with peripheral concentrations of gonadotropins, ovarian steroids, and inhibin during the estrus cycle in goats. *Biol. Reprod.* 69: 57-63
- Rahman, A.N.M.N., R.B. Abdullah and W.E. Wan-Khadijah, 2008. A review: Estrus synchronization and superovulation in goats. *J. Biol. Sci.* 7: 1129-1137
- Saharrea, A., J. Valencia, A. Balcazar, O. Mejia, J.L. Cerbon, V. Caballero and L. Zarco, 1998. Premature luteal regression in goats superovulated with PMSG: effect of hCG or GnRH administration during the early luteal phase. *Theriogenology.* 50: 1039-1052

Noni Effect on Goat Sperm Motility after Cooling

Isnaini, N.* & Nuryadi

Faculty of Animal Husbandry, University of Brawijaya. Jl. Veteran Malang 65145, Indonesia

Abstract

Noni (*morinda citrifolia*) is the fruit of a tropical plant with content of alkaloids (xeronin), vitamins A and C which are antioxidants that effectively prevent and neutralize free radicals. This study was conducted to evaluate Noni effect on goat sperm motility after cooling. Semen was collected from 4 crossbreed etawah bucks using artificial vagina method. Fresh semen evaluated for colour, pH, volume, concentration, mass motility, motility, life sperm and sperm abnormality. Semen was diluted with tris-egg yolk-based extender supplemented with different levels of Noni (*morinda citrifolia*) extract (0, 10, 20 and 30 %) v/v with the ratio of 1 semen : 4 diluter. Semen used had mass motility of 2+ and motility of 70%. Immediately after dilution semen was stored in 3-5°C and sperm motility percentage were observed at 0, 24 and 48 h. The obtained data were analyzed with Analysis of Variant (ANOVA) and Least Significant Difference were determined. The experiment was designed using completely random design (4 treatments and 10 replications). The results showed that the level of Noni (*morinda citrifolia*) extract had very significant effect ($P < 0.01$) on sperm motility percentage in 0, 24 and 48 h of cooling. It can be concluded that the best Noni (*morinda citrifolia*) extract level for resulting optimal sperm motility was 10%.

Keywords: AI, antioxidant, dilution, Noni extract, semen quality

Corresponding author: nurulisnaini17@yahoo.co.id

Introduction

Tris-egg yolk is the basic extender of simple semen buck diluter and it is made for the purpose of maintaining the pH and osmotic pressure and also to maintain essential inorganic ion concentrations. Noni (*morinda citrifolia*) extract is a material containing vitamin A and C which are antioxidants that effectively prevent oxidative stress (Djauhariya, 2003). However, there is very limited information could be found regarding the role of Noni (*morinda citrifolia*) extract in crossbreed etawah goat semen quality after cooling.

This study was conducted to evaluate the fortification of Noni (*morinda citrifolia*) extract in tris-egg yolk-based extender on sperm motility of crossbreed etawah goat semen after cooling.

Materials and Methods

Preparation of Noni (*morinda citrifolia*) extract

Noni (*morinda citrifolia*) extract prepared by blending: 5 ripe Noni (*morinda citrifolia*) fruit + 500 ml aquadest, and the mixture was then filtered using filter paper (0.48 µm) and deposited to form the supernatant and precipitate. Supernatant (Noni extract) was stored in a freezer temperature of -20°C.

Semen Collection and fresh semen evaluation

Semen was collected from 5 bucks of crossbreed etawah goat. Semen was collected 1 time a week by artificial vagina method. After collection, the semen was evaluated for volume, pH (with lacmus paper), concentration (calculated by using a hemocytometer), sperm mass motility (light microscope 100x magnification), motility (light microscope 400x magnification), viability and morphology (in nigrosin-eosin smears under a light microscope, 400x magnification). Only semen with mass motility of 2+-3+ and motility of sperm more than 70% was used for research material.

Semen Dilution, Storage and Evaluation

The selected semen was diluted with tris-egg yolk-based extender containing 0, 10, 20 and 30% v/v of Noni (*morinda citrifolia*) extract. Semen was diluted in ratio of 1 semen: 4 diluter (0.1 ml semen: 0.4 ml of diluter). Diluted semen was placed into 1.5 ml closed eppendorf tubes. To determine the effect of level of Noni (*morinda citrifolia*) extract on the motility of cock sperm, semen was evaluated after cooling (3-5°C) at 0, 24 and 48 h.

Data analysis

The obtained data were analyzed by Analysis of Variance (ANOVA) and continued by Least Significant Difference calculated if there was significant or very significant different between groups. The experiment was designed using a completely random design (4 treatments and 10 replications).

Results

Characteristics of fresh semen

The collected semen was examined the characteristics before treated with tris-egg yolk-based extender combined with Noni (*morinda citrifolia*) fruit extracts (Table 1).

Table 1. Characteristics of fresh semen used in the experiment (n=10).

Parameter	Mean \pm SD
Color	creamy
Consistency	less opaque
pH	7.00 \pm 0.00
Volume (ml/head)	1.01 \pm 0.21
Concentration (10 ⁷ /ml)	300.2 \pm 23.06
Mass motility (%)	2+ - 3+
Individual motility (%)	79.00 \pm 3,16
Viability (%)	95.07 \pm 1.54
Abnormal sperm (%)	4.64 \pm 0.80

Sperm Motility

Percentage of sperm motility of bucks semen after cooling diluted with tris-egg yolk-based extender containing different levels of Noni (*morinda citrifolia*) extract at different times of cooling are shown in Table 2.

Table 2. Percentage of sperm motility following treatment with different Noni (*morinda citrifolia*) extract level after cooling.

Time of cooling (h)	Noni extract levels (%)	Mean \pm SD
0	0	69.00 \pm 2.11 ^b
	10	73.00 \pm 3.50 ^a
	20	68.00 \pm 4.22 ^b
	30	67.50 \pm 4.25 ^b
24	0	62.00 \pm 4.22 ^a
	10	64.50 \pm 3.69 ^a
	20	56.00 \pm 3.94 ^b
	30	53.50 \pm 2.42 ^b
48	0	52.00 \pm 2.58 ^a
	10	54.00 \pm 3.94 ^a
	20	43.50 \pm 2.42 ^b
	30	42.00 \pm 2.58 ^b

^{a, b} highly significant different (P<0.01).

Discussion

The results showed that the level of Noni (*morinda citrifolia*) extract had very significant effect (P<0.01) on sperm motility percentage in 0, 24 and 48 h of cooling. Levels 10% Noni (*morinda citrifolia*) extract produced the highest percentage of motility of spermatozoa, followed by the level of 0%, 20%, and 30% Noni (*morinda citrifolia*) extracts. Sperm motility decreased gradually as the duration of cooling. The longer the cooling the lower the sperm motility and viability. Decrease in the percentage of sperm motility after cooling is due to fewer sperm that have sufficient energy reserves to be used to move, as long as the cooling sperm remain metabolic activity. Exogenous substrates during cooling required for mitochondrial ATP availability is limited. Secondary metabolites materials required for energy and buffer as well as antioxidants to protect sperm from damage due to the accumulation of CO₂, lactic acid and free radicals (Kaeoket et.al., 2011; Tavilani et. al., 2008). It can be concluded that the best Noni (*morinda citrifolia*) extract level for resulting optimal sperm motility was 10%.

References

- Kaeoket, K., P. Chanapai, Junchiyaphoom and P. Chanapiwat, 2011. The effect of using long term and short term extenders during cooling process on the quality of frozen boar semen. Thai Vet. Med. J. 41(3): 283 – 288.
- Tavilani H., M.T., Goodarzi, A. Vaisi-raygani, S. Salimi and T. Hassanzadeh, 2008. Activity of antioxidant enzymes in seminal plasma and their relationship with lipid peroxidation of spermatozoa. Int. Braz. Urol. J. 34(4): 485 – 491.

Productive and reproductive performance of Indigenous Lime and Parkote Buffaloes in the Western hills of Nepal

Shah¹, M. K., Y. Hayashi² & H. Kumagai³

¹Nepal Agricultural Research Council, Regional Agricultural Research Station, Lumle, Kaski Nepal, ²Faculty of Agriculture, Meijo University, Kasugai, 486-0804, Japan, ³Graduate School of Agriculture, Kyoto University, Kyoto, 606-8502, Japan

Abstract

Livestock is an integral part for the most rural livelihoods in Nepal. A very high proportion of poor and marginalized farmers depend on livestock as main or supplementary source for their income. Cattle and buffalo contribute more than 70% to the livestock sector. The productive and reproductive performances of indigenous buffalo, Lime and Parkote, were studied in Regional Agricultural Research Station (RARS), Lumle in Nepal. The compilation of 14 years (2000 to 2014) lactation records of the indigenous buffaloes maintained at RARS livestock farm was collected and analyzed to assess the production and reproduction traits. Similar surveys were also conducted in Kaski and Lamjung districts of Nepal. The buffaloes were kept on-station with the basal diet of commercial concentrate, roughages and a supplementary grazing (2-3 hours per day). The productive performance of Lime was 964.1±33 litre/lactation and Parkote was 878.5±66.3 litre/lactation, while daily milk yields of Lime was 3.16±0.10 litre/day and Parkote was 2.88±0.21 litre/day. The milk constituent of Lime was (9.03%) fat, (9.21%) solid not fat & (3.84%) protein and Parkote was (8.91%) fat, (9.81%) solid not fat, & (4.02%) protein. The maximum mating (30%) was found in October, while the minimum mating (1%) was in June. The calving time of Lime and Parkote is maximum (36%) in the month of September. The farmers of mid hill were preferred (91.5%) farmers was natural mating system while (2.1%) artificial insemination and (6.4%) farmers have both artificial insemination & natural breeding in indigenous buffaloes. The survey of Kaski and Lamjung districts was summarized that 10.4% farmers had throughout the year fodder and forage availability, whereas 14.6% farmers have nine month, 68.8% farmers have six month & 6.2% farmers have four month availability of forage and fodder. The Lime and Parkote buffaloes are highly potential milking animals in the western hills of Nepal. Furthermore, the value chain and organic production approaches would be very useful for the conservation and utilization of these indigenous buffaloes.

Keywords: buffalo, lime, parkote, production, reproduction

Introduction

Indigenous buffaloes of Nepal have the ability to adapt across different agro-ecological zone, exist in low plain of nutritional regime, efficient forage digestion ability, cold tolerance and relatively smaller body size. Therefore, they are highly suitable to thrive on narrow and steep slope of the hills and mountains of the country. The indigenous buffalo of Nepal Lime and Parkote are of reverine type having 25 pairs of chromosomes. They are mainly black in colour with certain peculiar marks or patches in the different body parts. It has been reported that within the indigenous breeds of buffaloes, there are some very high yielding individuals. However, due to the indiscriminate breeding and absence of selection, the utilization of superior genetic materials is being restricted. There are limited studies related to improvement of their production

potential. Hence, this study was carried out to evaluate the productive and reproductive performance of indigenous buffalo, Lime and Parkote, in the western hills of Nepal.

Materials and Methods

All the Lime and Parkote buffaloes and their progenies maintained at Regional Agricultural Research Station (RARS), Lumle Kaski were used for this study. The buffaloes had been maintained on the station with the basal diet of commercial feed and a supplementary grazing (3-4 hours per day). Day to day on station maintenance and management of buffaloes and their progenies have been performed with routine activities such as feeding, fodder collection, cleaning, grooming, milking, weighing, veterinary care, breeding etc. Performance records of 14 consecutive years (2000 to 2014) were used for this study. Production and reproductive performance records of the animals. i.e. age at first services, age at first calving, gestation period, lactation length, lactation yield, etc. have been recorded as per standard format and analyzed in Minitab statistical package.

Results and Discussion

Reproductive Performance

The reproductive performance of indigenous buffalo Lime and Parkote is presented in Table 1. The age at first service (days), age at first calving (days), gestation period (days), lactation length (days), dry period (days) and calving interval (days) were found as 978.1 ± 172.8 , 1333.9 ± 189.3 , 304.07 ± 6.56 , 303.65 ± 71.59 , 176.9 ± 22.1 , and 489.4 ± 66.1 respectively. The results are in agreement with the finding of Amataya et al. (2000) who reported that age of first calving of Lime and Parkote buffalo was 51.6 months. Rasali et al. (1997) reported that the age of the first calving for the indigenous buffalo was 53.8 months.

Table 1. Reproductive Performance of Indigenous buffalo Lime and Parkote from 2000 to 2014.

Parameters	Values
Age at 1 st service (days)	978.1 ± 172.8
Age at 1 st calving (days)	1333.9 ± 189.3
Gestation Period (days)	304.07 ± 6.56
Lactation length (days)	303.65 ± 71.59
Dry Period (days)	176.9 ± 22.1
Calving interval (days)	489.4 ± 66.1

Productive Performance

The productive performance of indigenous Lime and Parkote buffalo is presented in Table 2. The average daily milk yield (lit) of Lime and Parkote buffalo ranges from 1.31 to 4.30 and 1.55 to 4.39. The lactation yield (lit.) of Lime and Parkote buffalo ranges from 399.5 to 1311.50 and 473 to 1337.80 respectively.

Table 2. Productive performance of Indigenous buffalo Lime and Parkote 2000 to 2014.

Parameters	Lime			Parkote		
	Mean	Minimum	Maximum	Mean	Minimum	Maximum
Average daily milk yield (lit.)	3.16	1.31	4.30	2.88	1.55	4.39
Lactation yield (lit.)	964.10	399.5	1311.50	878.5	473.00	1337.80
Fat %	9.03	5.07	12.63	8.91	6.39	11.58
Solid- not- Fat %	9.21	6.08	11.18	9.81	8.79	11.10
Protein %	3.84	2.72	4.56	4.02	3.26	4.81

The ranges of milk constituents fat %, Solid-not-fat % and Protein % in indigenous buffalo Lime were 5.07 to 12.63 , 6.08 to 11.18 and 2.72 to 4.56 respectively, whereas in Parkote buffalo milk constituents fat %, Solid-not-fat % and Protein % were 6.39 to 11.58 , 8.79 to 11.10, 3.26 to 4.81, respectively. The lactation yield of indigenous breed of buffaloes in this study is similar to the findings of Shrestha (2003) and Amatya et al. (2000).

Mating and calving season

The mating and calving season of indigenous Lime and Parkote buffalo ranges throughout the year but peak time of mating season and calving season was August to October and June to September, respectively. The results finding are agreement with the finding of Shrestha (2003).

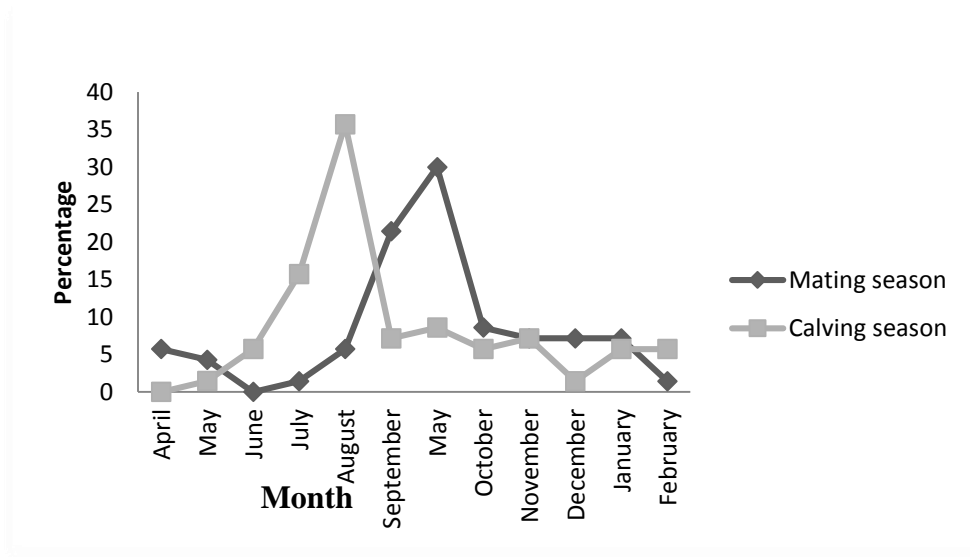


Figure 1. Mating and calving season of indigenous buffalo Lime and Parkote.

Conclusion

The population of indigenous buffalo is declining. Results obtained from this study have shown high variation in reproductive and productive performance of indigenous Lime and Parkote buffalo. Adoption of proper selection and appropriate breeding methods are essential for its genetic improvement and to increase its productivity. Relatively poor management practice for these animals also affects their production and reproduction performance to a great extent.

References

- Amatya, N., D.P.Rasali., R.S.Rana, 2000. Evaluation of phenotypic and production characteristics of indigenous buffalo type in the western hills of Nepal. Nepal Agricultural research council, ARS, Lumle, Nepal.
- Rasali, D.P., D. B. Gurung and E. R. Yadav, 1997. Performance of monsoon calver buffaloes across genotypic and non-genotypic factors under farmers management in western hills districts of Nepal, Technical Paper, ARS, Lumle.
- Shrestha, B.S. 2003. Development of suitable breeding strategy for indigenous buffaloes in the western hills of Nepal. Final Technical Report, HARP. Pp78.

The oocyte parthenogenesis stimulation by protein extract of goat spermatozoa

¹M. Nur Ihsan, ²S. Rahayu, ¹G. Ciptadi, ³E.A. Wahyuni, ⁴Mudawamah & ¹I. Novianti

¹Faculty of Animal Husbandry UB, ² Faculty of Basic Science-Biology UB, ³Trunojoyo University, Bangkalan, Madura, ⁴ Islamic University of Malang, Indonesia

Abstract

The research purpose is to analysis the potential function of proteins sperm extracts in activation of oocyte resulted from in vitro maturation (IVM). Protein sperm extract was isolated from goat semen ejaculated. This sperm extract is expected to be utilized as a candidate natural activator for gamete cell activation. Laboratory experimental of IVM is carried out in medium TCM199 + 10% FBS + 10 μ l FSH + 25 μ l LH + gentamycin in 5% CO₂ incubator for 24 hours, and then selected for second metaphase (M-II) oocytes by 1st polar body extrusion. Main Treatments were ethanol activation followed by sperm extract supplementation were: Sperm Extract Alone (Po), Ethanol 7%+ Sperm Extract (P1), ethanol 7%+ 2 mM 6-DMAP (P2). The variables measured were the percentage of cells activated base on cleavage rate. The results showed that activation of the matured oocyte by both sperm extract alone (control) and use total protein sperm extract can be used in a system proven in vitro activation of M-II parthenogenesis oocytes. It was concluded that activation of M-II oocytes by chemicals and then supplemented by sperm extract indicates the best cleavage rate of about 34.48%. It is recommended for further basic research focused on specific protein concentration of extracts sperm to oocyte activation *in vitro*.

Keywords: oocyte, parthenogenesis, activation, sperm extract, goat

Corresponding author: m_nur_ihsan@yahoo.com

Introduction

Parthenogenesis activation of oocytes M-II mammals generally done by induction of chemicals that can increase levels of intracellular Ca⁺⁺ (Ca⁺⁺ oscillation) that principally is mimicking what happens in the natural process of fertilization by sperm. Problems arise because most chemical activation treatment only resulted in increased levels of Ca⁺⁺ monotonous and its single spike, causing partial activation is less than perfect and not favorable for further development of the embryo. Extract spermatozoa have competence as biological activators replace chemicals (Ca ionophore / 6-DMAP). Extract of sperm in mice reported to have the potential to improve in vitro fertilization (IVF) and Intra cytoplasmic Sperm Injetion (ICSI), since the sperm is a highly efficient activator oocytes. Extract sperm injection into the oocyte of mammals have been shown to raise levels of intracellular calcium are similar to the conditions after the occurrence of normal fertilization by spermatozoa. Through analysis of influx Calcium (Ca⁺⁺ release) with Fluo-3, using a laser microscope confocal will be known to an increase in calcium levels as evidence for the activation of oocytes by sperm extracts.

Research Methods

Extract isolated and prepared sperm from the male ejaculate goats, refers to the method that has been used in mice (Fissore, 1998) and horses Choi et al. (2002) with a little modifications (Wahyuni, et al., 2008). Ejaculate centrifuged 900 g for 10 minutes to remove seminal plasma. Pellet plus the TALP containing 6 mg BSA and centrifuged 900 g for 10 minutes. Pellet redissolved to a concentration of sperm obtained 5×10^8 and 20×10^8 sperm / ml medium core insulation, centrifuged to eliminate TALP. In a medium containing 1 mM dithiothreitol, 100 uM leupeptin, antipain 100 um, and 100 ug / ml soybean trypsin inhibitor. The suspension obtained is frozen, thawed back and sperm created pellets with centrifuge 20,000 g for 50 minutes at a temperature of 2 ° C. Supernatant obtained by carefully drawn, aliquoted, and keep at a temperature of - 80 ° C until use.

Parthenogenesis activation method referring to Onger et al. (2001). Briefly, goat oocyte activation is as follows: (1). IVM performed up to 24 hours, and then made the release of the cumulus of the oocytes matured with 0.1% hyaluronidase for 1 minute. (2). Oocytes then washing twice in TCM 199 + 5% FBS and 1 times in TCM 199 + 10% FBS. (3). Incubation carried back for 4 hours at a temperature of 38.5o C in 5% CO2 incubator in medium TCM199 + 10% FBS with maximum humidity. (4). At the 28th hour of the beginning of the culture performed in a material exposure induced activation of oocytes, namely: extracts sperm alone (P0), 7% ethanol with sperm extract (P1) and Sperm extract with the protein kinase inhibitors (6-DMAP) (P2).

On activation of the combination treatment, then after a single treatment induced rise in intracellular calcium levels for 7 minutes, the oocytes were washed 3 times in medium TCM199 + 10% FBS and then cultured for 4 hours in a combination treatment of 7% ethanol / with 2 mM 6-DMAP.

Results and Discussion

The results of the activation parthenogenesis presented in Table 1. In general, the results showed that the sperm extract itself does not demonstrate capabilities in cell activation, so that the chemical agents remain necessary. Thus the function is more suplementatif. Figure 1 shows the pattern of non influx recurrent calcium based on their Ca^{++} intensity. Table 1 showed. Results of oocyte activation is based on the percentage of M-II oocytes that show cleavage.

Table1. Cleavage rate of M-II Oocytes.

Treatments	Total Oocyte	Numbers oocytes cleavage (%)
Control (ESC)	110	3 (2.72)
Ethanol + ESC	119	24 (20.16)
Ethanol + 6-DMAP+ESC	116	40 (34.48)

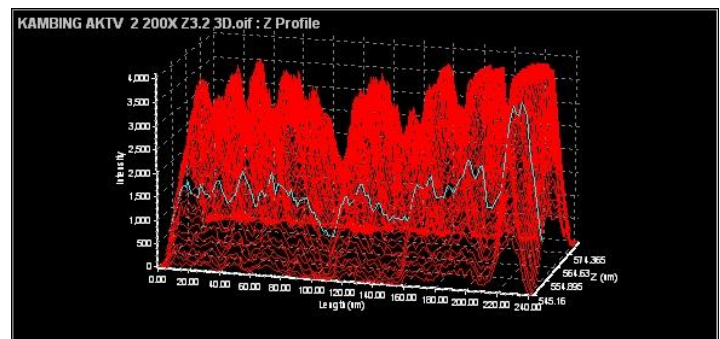


Figure 1. The intensity pattern of calcium using CLSM.

The activation success of extract sperm for oocyte because it may be supported by activating the protein content of sperm were very varieties. To obtain a functional protein, especially a protein that helps the activation of oocytes then need to do further analysis. Proteins with a molecular

weight of approximately 100 kDa have the ability to activate oocytes (Matsuura and Maeda, 2006). Results of oocyte activation induced parthenogenesis use traditional chemicals such as ethanol, Ca ionophore, cycloheximide and 6-DMAP shows the level of activation of oocytes goat relatively low at between 29-45%. (Ciptadi et al, 2004; Ciptadi 2005). Some activation treatment of artificial been proven cause an increase in the concentration of ions in intracellular calcium by release of calcium from the cytoplasm, such as strontium (Cuthbertson et al., 1981) and Ionomycin (Loi et al., 1998), while ethanol is known to induce the increase in the concentration of calcium derived of extra cellular and Ca ionophore A-23187 induces a calcium influx of extracellular and intracellular reserves (Nakada and Mizuno, 1998, Grupen et al., 2002. Effects of protein extracts of spermatozoa is in its ability to help stage parthenogenesis embryonic development Swann et al., (2004) found that the Sperm Extract contains PLC, particularly PLC

Conclusion

ES effects show the amount of cleavage activation of oocytes 2:27 - 34.48%. Extract own sperm cannot be used as activation parthenogenesis. The highest amount of cleavage after exposure to extract sperm activators supplemented in combination Ethanol and 6-DMA, yet so extract the sperm could be developed as a candidate activator that plays a role in increasing the cell cleavage parthenogenesis. More research is needed on effective use of the levels of sperm extract either alone or in combination with chemical agent cell activation.

Acknowledgements

This work was supported by Research Grant year of 2013/2014) from Direktorat General of High Education-DIKTI, Ministry of Education Republic of Indonesia.

References

- Ari Wahyuni, E. G. Ciptadi, Aulani Am and B. Siswanto. 2008. Isolation and characterization of sperm crude extract of goat and cattle. Un published. Brawijaya University.
- Ciptadi, G. 2005. Improvement of oocyte activation using sperm extract for M-II and reconstructed cells. Un published. Dissertation, Brawijaya University, Indonesia.
- Grupen, C.G., M.B. Nottle and H. Nagashima. 2002. Calcium release at fertilization: artificially mimicking the oocyte's response to sperm. *J. Reprod. Dev.* 48: 313 – 333.
- Matsuura, D and T. Maeda. 2006. Effect of Sperm Extract and its Molecular Weight Fractions on Oocyte Activation in Miniature Pig Spermatozoa. *J. Mamm. Ova Res.* 23:122-127.
- Ongeri, EM., C.L. Bormann, R.E. Butler, D. Melican, W.G. Gavin, Y. Echelard, R.L. Krisher and E. Betboodi. 2001. Development of goat embryos after in vitro fertilization and parthenogenesis activation by different method. *Theriogenology*, 55 (9): 1933 – 1945.
- Wu, H., J. Smyth, L. Veronica, F. Kiyoko, T. Tadaomi, L.B. Samuel, L.A. Nancy , and R.A. Fissore. 2001. Sperm Factor induces Intracellular free Calcium Oscillations by stimulating the Phosphoinositide Pathway. *Biol. Reprod.* 64: 1338-1349.

Supplementation of vascular endothelial growth factor (VEGF) increases the maturation of porcine COCs derived from small follicles

Bui T. T. M., A. Karata, P. Ferré, M. T. Tran, T. Wakai & H. Funahashi*

Department of Animal Science, Okayama University, Okayama 700-8530, Japan

Abstract

It is known that oocytes derived from small follicles (SF) less than 3 mm in diameter have lower competence to successfully mature *in vitro*. The aim of this study was to investigate, in prepubertal pig, whether vascular endothelial growth factor (VEGF) can stimulate the meiotic competence of oocytes derived from SF. The amount of VEGF secreted to the maturation medium was higher in cumulus-oocyte complexes (COCs) derived from middle follicles (MF) than those from SF ($P < 0.05$). Different concentrations of VEGF (0, 10, 25, 50 or 100 ng/mL) were supplemented during the first 20 h of *in vitro* maturation (IVM). Higher maturation rates were recorded ($P < 0.01$) in the presence of VEGF at 50 and 100 ng/mL (62.2% and 72.9%, respectively), while there were no significant differences among the other samples in the presence of 0, 10 and 25 ng/mL VEGF (53.8%; 55.2% and 50.7%, respectively). Our results indicate that VEGF improves the meiotic competence of oocytes derived from SF, especially at a 100 ng/mL concentration when added during the first 20 h of IVM.

Keywords: IVM, small follicle, porcine, VEGF

Corresponding author: hirofun@okayama-u.ac.jp

Introduction

Oocytes derived from follicles with less than 3 mm in diameter, which have a majority in ovaries have been known to have a lower competence to achieve metaphase II *in vitro* (Yoon et al., 2000). It has been known that oocytes from SF may lack of factors that mediate to allow them to completely mature (Romaguera et al., 2010). The amount of VEGF and its receptors are up-regulated with the folliculogenesis (Greenaway et al., 2004). In addition, the effect of VEGF supplementation in culture medium has been reported to induce follicular and oocyte growth in goat (Bruno et al., 2009). Considering the benefits obtained from VEGF protein, we investigated the effect of VEGF on the maturation of oocytes derived from SF in pre-pubertal gilts.

Material & Methods

COCs were obtained from SF (< 3 mm) and MF (3-6 mm). Groups of 40 COCs with at least 3 layers of clear and compact cumulus cells (CC) were cultured *in vitro* following a standard protocol (Funahashi et al., 1997). The amount of VEGF in the culture medium after 20 and 44 h of IVM was evaluated by Quantikine[®] ELISA, Human VEGF immunoassay (SVE00, R&D System). In a second experiment, SF oocytes were supplemented with different concentrations of VEGF (0, 10, 25, 50, 100 ng/mL, V4512-5UG, Sigma-Aldrich) during the first 20 h of IVM. After 44 hours, oocytes were fixed, stained with orcein and observed under a phase contrast microscope.

All the experiments were replicated 5 times and analyzed by one-way ANOVA (Minitab 15). Data were expressed as mean \pm SD.

Results and Discussion

VEGF protein quantification

The concentration of VEGF was demonstrated to be lowest in follicular fluid aspirated from SF and gradually increased following up to the follicle growth (Kere et al., 2014). No level of VEGF was detected in early degenerated middle size follicles (Mattioli et al., 2001). Our results show that the amount of VEGF secreted into the culture medium at 20 h or 44 h of IVM was higher in oocytes from MF when comparing with those from SF (Table 1), indicating that VEGF plays a crucial role for the development of mature and competent oocytes.

Table 1. Amount of VEGF protein secreted to the culture medium after 20 h and 44 h of IVM.

Follicle type	VEGF (pg/mL)	
	20 h	44 h
MF	115.1 ^a \pm 21.2	376.9 ^a \pm 78.9
SF	35.6 ^b \pm 4.0	67.8 ^b \pm 16.1

^{a,b} Mean do not share a letter within column are significantly different (P<0.05).

Effect of VEGF on the development of oocytes derived from small follicles

Our data shows that the maturation rate at the concentration of 50 and 100 ng/mL (62.2% and 72.9%, respectively) was significantly higher than 0, 10, 25 ng/mL (50.7%, 53.8% and 55.2%, respectively, P<0.01, Figure 1). The concentrations used in a study by Luo et al. (2002) on oocytes from bovine MF was lower than ours. This suggests that the amount of VEGF supplementation is different according to the species and the oocyte stages. It has been reported that the VEGF expressed in total follicular tissue increased significantly (and correlated) with the developmental stages of follicle growth (Shimizu et al., 2003). Together with our data, we suggest that VEGF is crucially involved in the oocyte meiotic resumption process.

In conclusion, COCs derived from MF produce higher amounts of VEGF than those from SF. The higher concentration of VEGF correlates positively with the nuclear maturation rates. Based on our data, VEGF was demonstrated as a factor affecting directly the oocyte nuclear maturation process *in vitro*. Therefore, the addition of VEGF during the first half of IVM could be used to improve maturation rates of small follicle oocytes in IVM protocols.

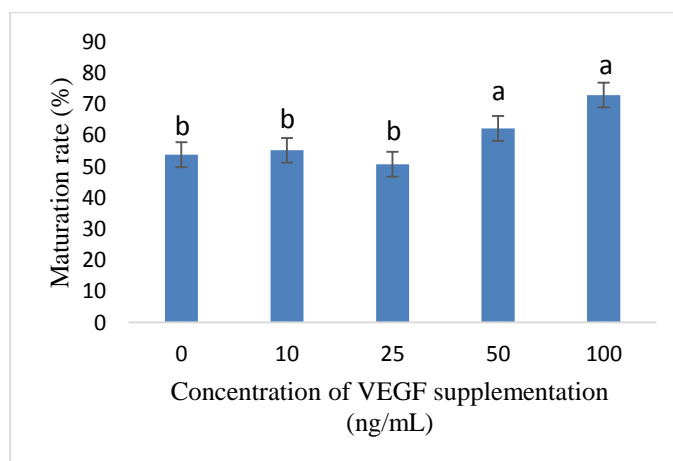


Figure 1. Effect of VEGF supplementation on maturation rate in IVM.

References

- Bruno J. B, Celestino J. J, Lima-Verde I. B, Lima L. F, Matos M. H, Araujo V. R, Saraiva M. V, Martins F. S, K. P.Name, C. C. Campello, S. N. Bao, J.R. Silva and J.R. Figueiredo, 2009. Expression of vascular endothelial growth factor (VEGF) receptor in goat ovaries and improvement of *in vitro* caprine preantral follicle survival and growth with VEGF. *Reprod. Fertil. Dev.* 21(5): 679–687.
- Funahashi, H., T. C. Cantley and B.N. Day, 1997. Synchronization of meiosis in porcine oocytes by exposure to dibutyl cyclic adenosine monophosphate improves developmental competence following *in vitro* fertilization. *Biol. Reprod.* 57:49-53.
- Greenaway, J., K. Connor, H. G. Pedersen, B. L. Coomber, J. Lamarre and Petrik J, 2004. Vascular endothelial growth factor and its receptor, Flk-1/KDR, are cytoprotective in the extravascular compartment of the ovarian follicle. *Endocrinol.* 145(6): 2896–2905.
- Kere, M., C. Siriboon, J.C. Liao, N.W. Lo, H.I. Chiang, Y.K. Fan, J. P. Kastelic and J.C. Ju, 2014. Vascular endothelial growth factor A improves quality of matured porcine oocytes and developing parthenotes. *Domest. Anim. Endocrinol* 49: 60-69.
- Luo, H., K. Kimura, M. Aoki and M. Hirako, 2002. Effect of vascular endothelial growth factor on maturation, fertilization and developmental competence of bovine oocytes. *Vet. Med. Sci.* 64(9):803-806.
- Mattioli, M., B. Barboni, M. Turriani, G. Galeati, A. Zannoni, G. Castellani, P. Berardinelli and P.A. Scapolo, 2001. Follicle activation involves vascular endothelial growth factor production and increased blood vessel extension. *Biol. Reprod.* 65: 1014–1019.
- Romaguera, R., R. Morató, A.R. Jiménez-Macedo, M. Catalá, M. Rour, M.T. Paramio, M.J. Palomo, T. Mogas, D. Izquierdo, 2010. Oocyte secreted factors improve embryo developmental competence of COCs from small follicles in prepubertal goats. *Theriogenology* 74: 1050 – 1059.
- Shimizu, T., J. Jiang, K. Iijima, K. Miyabayashi, Y. Ogawa, H. Sasada and E. Sato, 2003. Induction of Follicular Development by Direct Single Injection of Vascular Endothelial Growth Factor Gene Fragments into the Ovary of Miniature Gilts. *Biol. Reprod.* 69:1388–1393.
- Yoon, K. W., T. Y. Shin, J. I. Park, S. Roh, J. M. Lim, B. C. Lee, W. S. Hwang, E. S. Lee, 2000. Development of oocytes from preovulatory follicles of different sizes after maturation in media supplemented with follicular fluids. *Reprod. Fertil. Dev.* 12: 133-9.

Supplementation of L-Carnitine on matured goat oocyte *in Vitro*

Sri Wahjuningsih* & Nurul Isnaini

Faculty of Animal Husbandry, University of Brawijaya,
Veteran Street, Malang 65141, Indonesia

Abstract

To support the development and growth gamete cells utilize energy sources and nutrients in the culture medium *in vitro*. Gamete cells in the early phase of increased production of oxygen free radicals when cultured *in vitro*. Therefore needed antioxidants that can inhibit free radicals in the culture medium. L-Carnitine has antioxidant activity. The purpose of the study was to analyze supplementation of L-Carnitine on matured goat oocyte. Immature oocytes with compacted cumulus cell and homogenous cytoplasm performed for 26 h in basic medium TCM 199 + FCS 10 % + PMSG 10 IU + hCG 10 IU and supplemented with L-Carnitine 0, 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4 mg/mL. The study design used was a completely randomized design using 10 replicates of each treatment. Evaluation of matured oocyte *in vitro* was percentage of completely expanded cumulus cell and metaphase II. The results showed that TCM 199 + FCS 10 % + PMSG 10 IU + hCG 10 IU and supplemented with L-Carnitine 0, 0.2, 0.4, 0.6, 0.8, 1.0, 1.2 and 1.4 mg/mL yielded completely expanded cumulus cell was 62.5, 62.7, 67.8, 68.4, 70.1, 76.6, 91.3, 78.5 % and Mt - II oocytes was 59.1, 61.7, 58.2, 60.8, 66.4, 76.2, 88.7, 77.6 % respectively. There were significant differences ($P < 0.05$) on completely expanded cumulus and metaphase II oocyte. It was concluded that supplementation of L-Carnitine increase the rate of matured goat oocyte *in vitro* and the best dose of L-Carnitine for *in vitro* maturation rate was 1.2 mg/mL.

Keywords: carnitine, cumulus cell, goat, metaphase II, oocyte

*Corresponding author : yuningyuning208@yahoo.com

Introduction

Application of embryo transfer requires embryos in large quantities which can be obtained from *in vitro* fertilization. One of the factors that play a role in IVF was successful *in vitro* maturation. The process of oocyte maturation *in vitro* was useful to provide a well-developed oocytes in cumulus cells, maturation of cytoplasmic components and nucleus at metaphase II stage. Cells gametes and embryos in the early phase increased production of oxygen free radicals when cultured *in vitro* (You et al., 2012). The formation of Reactive Oxygen Species which is generated due to the plasma membrane damage causes a decrease in the ability of the fusion of sperm - oocyte. Therefore, it needs an antioxidant that can inhibit free radicals in the culture medium.

L-carnitine is a water-soluble molecule and plays an important role in the metabolism of fatty acids that have antioxidant activity to protect against DNA damage (Dunning et al., 2012). L-Carnitine has an important role in the β -oxidation via the transport of fatty acids into the mitochondria for ATP production which can improve oocyte maturation and embryo development (Yamada et al., 2006)

Materials and Methods

Ovaries were obtained from slaughter house, put into the bottle containing NaCl 0.9 % + Penicillin 0.06 g/100 ml and Streptomycin 0.01 g/ 100 ml. Aspiration medium was TCM199 (GIBCO, Cat no. 21,200,076) added with Hepes (Sigma), NaHCO₃ (Sigma) and 10% Fetal Calf Serum (GIBCO, Cat no.39N8255). Selected oocytes based on the cumulus cells surrounding oocyte and IVM process according to Wahjuningsih et al.,(2012). Only oocyte were surrounded by compact cumulus cells were used for IVM. Selected oocytes were placed in an incubator 5% CO₂, 95% humidity, and temperature of 38.5°C for 26 h.

Evaluation of oocyte maturation

Evaluation of oocyte maturation was done by completely expanded cumulus cell and Metaphase-II. Oocytes that have been matured for 26 h were observed completely expanded cumulus cell cumulus by inverted microscope with 400 X magnification. The level of metaphase II was done by staining with 1% aceto-orcein (Wahjuningsih et al., 2012)

Statistical analysis

Each experiment was replicated ten times, The data of completely of expanded cumulus cells (%) and metaphase-II (%) were transformed to arcsine. One-way analysis of variance was used to analyze significant difference between treatments ($p < 0.05$). Duncan's multiple range test was used to compare the means of significant difference of each treatment.

Results and Discussion

There were significant differences in percentage of completely expanded cumulus cells among different concentration of L-Carnitine (Table 1).

Table 1. Effect of different concentration of L-Carnitine on completely expanded cumulus cells and Mt-II oocytes

Concentration of L-Carnitine (mg/L)	Completely expanded cumulus cells (%)	Mt - II oocytes (%)
0	62.5 ^a	59.1 ^a
0.2	62.7 ^a	61.7 ^a
0.4	67.8 ^b	58.2 ^a
0.6	68.4 ^b	60.8 ^a
0.8	70.1 ^b	66.4 ^b
1.0	76.6 ^c	76.2 ^c
1.2	91.3 ^d	88.7 ^d
1.4	78.5 ^c	77.6 ^c

The different notation on the same column showed that the treatment gave significant different result ($p < 0.05$)

Supplementation of L-Carnitine in IVM medium will accelerate the maturation nucleus. L-Carnitine plays a role in the maturation of the nucleus is a factor in promoting the development of nucleus maturation. L-Carnitine is involved in the transport of long-chain fatty acids into the mitochondria for β -oxidation, therefore L-Carnitine supplementation may increase the use of fatty acids and energy as needed physiologically (Steiber et al., 2004).

Oocyte containing endogenous lipid substantially consisting mostly of triglycerides. Endogenous triglycerides play an important role during oocyte maturation in vitro, which provides ATP for protein synthesis that is necessary for meiosis and cytoplasmic maturation (Somfey et al., 2011). Therefore the acceleration of nucleus maturation can be caused by an

increase in fatty acid metabolism via supplementation of L-Carnitine. Glucose metabolism in oocyte maturation can be enhanced with lipid metabolism. As a consequence can cause lipid peroxidation, in-activation of enzymes, oxidative modification of proteins and DNA fragmentation are known to have deleterious effects, such as alteration of mitochondria, apoptosis and cell embryo block in oocytes and embryos (Wu et al., 2011) .In this study, L-Carnitine supplementation of 1.2 mg / mL provide a positive effect on the expansion of cumulus cells and oocytes reach Mt phase-II

Conclusion

Supplementation of L-Carnitine increase the rate of matured goat oocyte in vitro and the best dose of L-Carnitine for in vitro maturation rate was 1.2 mg/mL.

References

- Dunning KR, Robker, RL. 2012. Promoting lipid utilization with L-carnitine to improve oocyte quality. *Animal ReprodSci*; 134:69.
- Somfai T, Kaneda M, Akagi S, Watanabe S, Haraguchi S, Mizutani E, Dang-Nguyen TQ, Geshi M, Kikuchi K and Nagai T. 2011. Enhancement of lipid metabolism with L-carnitine during *in vitro* maturation improves nuclear maturation and cleavage ability of follicular porcine oocytes. *ReprodFertil and Dev*;23(7):912-920.
- Steiber, A, Kerner, J, Hoppel.C.L. 2004. Carnitine : a nutritional, biosynthetic and functional perspective. *Mol Aspects Med* 25:455-73
- Wahjuningsih, S. dan Djati,M.S. 2012. Study of estrus cow serum (ECS) in maturation media on in vitro maturation rate of bovine oocyte. *Journal of Agricultural Science and Technology A*..Vol.1(8) ISSN: 2161-6256
- Wu G.Q, Jia BY, Li JJ, Fu XW, Zhou GB, Hou YP, Zhu SE. 2011. L-carnitine enhances oocyte maturation and development of parthenogenetic embryos in pigs.*Theriogenology*. 15;76(5):785-93.
- Yamada, T., Imai, H., and Yamada, M. 2006.Beneficial effects of acetyl-L-carnitine treatment during IVM on post-fertilization development of bovine oocytes *in vitro*. *Reprod. Fertil. Dev.* **18**, 280–281.
- You J, Lee J, Hyun SH, Lee E. 2012. L-carnitine treatment during oocyte maturation improves in vitro development of cloned pig embryos by influencing intracellular glutathione synthesis and embryonic gene expression. *Theriogenology* 78:235-243.

The acceptability of reduced sperm concentration in frozen buffalo semen

*E.V. Venturina^{*2}, F.V. Mamuad^{**a}, R.V. Cabactulan², C.C. Ringor², & MD.B.Peralta² H.V. Venturina², F.A. Venturina²*

¹Philippine Carabao Center, Science City of Muñoz, Nueva Ecija 3120, Philippines, ²Philippine Carabao Center at Central Luzon State University, Science City of Muñoz, Nueva Ecija 3120, Philippines

Abstract

This study was conducted to determine the conception rate and calving rate of animals inseminated with frozen buffalo semen out of the semen collected from Murrah buffalo bulls. This will be the first time that we are inseminating buffalo cows with less than 100 million per ml sperm cell/ml. of frozen semen. When these become acceptable, this will cause to have more cows to be inseminated. The semen was extended with different sperm concentration of 50M, 45M, 40M, and 35M spermatozoa per dose (0.5 ml straw). There were about 37 buffalo bulls that were assigned to each concentration. The numbers of bulls depended on the number of cows to be inseminated and frozen semen doses. The number of semen donors has been identified and were given sperm doses with lower sperm concentration. Two breed types of bulls were used, the Indian Murrah and Bulgarian Murrah buffalo bulls. The number of doses produced depended on the motility and assigned to the treatment 1 (T₁), 50M/dose; T₂, 45M/dose; T₃, 40M/dose; and T₄, 35M/dose of each 0.5 ml straw. The number of ejaculates were recorded, processed and assigned to each treatment. The number of straws was 1,171 doses; 3,393 doses; 5,297 doses; and 4,454 doses for T₁, T₂, T₃, and T₄ respectively. They were used for artificial insemination by the Village Base Artificial Insemination Technicians (VBAIT) in the Provinces of Nueva Ecija, Bulacan, and Tarlac. The differences per treatment are presented. Decreasing the sperm concentration showed higher number of doses for T₁, T₂, T₃, and T₄ with 1,171 doses, 1,258 doses, 1,454 doses and 1,705 doses respectively. The conception rate and calf drop with male and female offspring showed no significant differences. The significant difference in gestation period is normal because it is within the range of 320 ± 7 days and correlation is normal.

In conclusion, trying to see the cost of producing 0.5 ml straw with different sperm concentration and the number of buffaloes to be inseminated showed that more semen could be used at 50 PhP per dose and could inseminate more animals. It is recommended that another study should be conducted by decreasing up to 15×10^6 sperm concentration/dose of 0.5 ml. straw.

Keywords: vbait-village base artificial insemination technicians

Corresponding authors: emmaventurina@yahoo.com*, fvmamuad@yahoo.com*

Rationale

The semen processing laboratory of the Philippine Carabao Center or the Philippine Carabao Research and Development Center was established last 1983. All the frozen semen produced was having 100 million sperm cells per ml or 50 million sperm concentration at a dose of 0.5 ml. straw. This was used for the last 19 to 20 years. Pregnancies and calf production was about 30 percent for estrus synchronized buffaloes and 70 to 80 percent for naturally occurring estrus buffaloes. After several years back from 2001 to 2004, there are comments that our products,

frozen buffalo semen are highly concentrated. The concentrations used were crafted from Thailand, India and Pakistan. Due to the increasing number of insemination needed in the field, the sperm concentration needs to be decreased. This would lead as to our aim to achieve high pregnancy rates with semen from genetically superior sires.

Objectives

1. To study the physical characteristics of fresh and frozen buffalo semen with different sperm concentration and their differences in the amount of straw produced.
2. To determine the conception rate/calving rate of animals inseminated with different sperm concentration.

Parameters

Individual motility, Mass movement, Sperm concentration, Abnormalities, PH (Hydrogen Ion concentration), Number of Production of Frozen Semen in 0.5ml straws, cost of production.

Review of Literature

Salisbury & Van Demark (1961) reported that it is a common perception that many of the problems with bull fertility and conception rate are due to insufficient number of sperm in AI doses from a particular bull, or all bulls in general. Dejarnette et al. (2007) reported that there is a threshold value above which adding more sperm to an AI dose does not increase fertility. The threshold value for sperm number per AI dose required to reach maximum fertility potential ranges from as low as 0.5M sperm per dose to as high as 12M per dose. Hence some bulls having more than 1-2 million sperm per AI dose does not improve fertility, where as other bulls, inclusion of 10-15M sperm per AI dose would be required to reach the upper limit value.

Abbas et al. (2001), says that there are numerous efforts that have been made to find the optimal sperm concentration per insemination dose without compromising the bull fertilities. Tahir et al.(1981)found that in buffaloes, different insemination doses of <20,20-30, 30-40, 40-50, 50-60 and >60 million frozen thawed spermatozoa resulted in similar fertility rate. Andrabi et al. (2006) states and concluded that reduction of sperm number from 30×10^6 to 15×10^6 spermatozoa in 0.5ml straw on dose of insemination did not affect fertility of cryopreserved buffalo bull semen investigation through pregnancy rate under field condition.

Methodology

The usual protocol in semen processing of buffalo semen is to follow the procedure written in the Semen Processing Manual. The sperm concentration was written to be 50 million sperm cells in 0.5 ml straw and not less than 30 percent individual sperm motility. They were news or statements from other countries who visited the laboratory, that the semen that was produced here are above the normal sperm concentration and suggested that we could lower it down. To make the necessary decreases the following sperm concentration will be followed. This can be tested to inseminate animals and will be re inseminated when they will be in heat after 21-42 days and will be pregnancy diagnose 45-65 days after. Final calf drop will be made during their term.

Experimental Procedure

A. Sperm Concentration: T.1 100M/ml -50M/dose, T.2 90M/ml-45M/dose, T.3 80M/ml-40M/dose, T.4 70M/ml - 35M/dose

B. Percent Motility: All semen that has 60% motility and above shall be processed and classified under the four different sperm concentration in letter A.

C. Other parameters: All semen ejaculate that pass the standard shall pass thru the evaluation and given to the four different sperm concentration.

D. Determination of fertilizing Ability by Artificial Insemination and Calf Drop: The AI technician shall be given from semen with four different sperm concentration. The semen doses shall be given some ID or mark to show then sperm concentration during their recording system in the forms. The pregnancy rate and calving will be recorded and will form part of evaluation. A total number of AI technician will be 10 to 15, they will be given 100/doses of each of the four groups “T.1; T.2; T.3 and T.4”. The kind of semen that will be use shall be T.1; T.2; T.3 and T.4 and repeated again to minimize error. The pregnancy rate, calving record, body condition score of carabaos, date, time of insemination, place of semen deposition, normally cycling carabaos or cross breeds/pure breeds will be recorded.

The production of different semen doses under each of the four categories will be given consideration. The cost of production will be made and will be given some projection in targeting the production for one year.

E. Experimental Design and Analysis. The experiment was arranged and analyzed by Randomized Complete Block Design, (RCBD) and analysis of variance was differentiated with Duncan’s Multiple Rage Test, (DMRT). Descriptive analysis was made with different sperm concentration level made. Correlations were made to PD and CD for different sperm concentration.

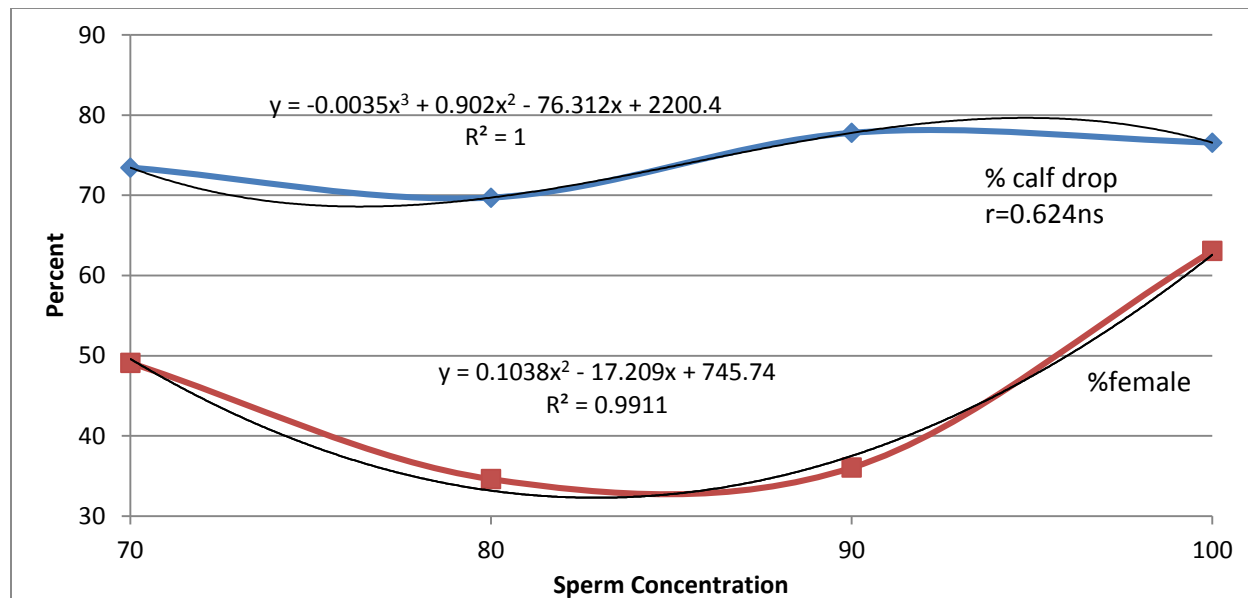
Table 1. The following bulls and village base AI technicians were available and used to get the results.

Treatment	Number of Bulls per Sperm Concentration	Number of AI Technicians
T1 100M	8 Buffalo Bulls	10 VBAIT
T2 90M	9 Buffalo Bulls	12 VBAIT
T3 80M	11 Buffalo Bulls	15 VBAIT
T4 70M	9 Buffalo Bulls	11 VBAIT

Table 2. The Gestation Period of animals monitored for Pregnancies and Calf drop and sex of crossbreds.

Sperm Concentration (M/ml.)	GP (days)	AI'd Animals	PD*	CD (%)	Male (%)	Female (%)
T1-100	314.00	111	85	76.58	36.93	63.07
T2-90	315.00	108	84	77.77	63.95	36.05
T3-80	317.00	208	145	69.71	65.37	34.64
T4-70	318.00	113	83	73.45	50.88	49.12

*animal pregnant



Summary, Conclusion, and Recommendation

The acceptability of frozen buffalo semen with different sperm concentration was conducted to determine the conception rate, calving rate of animals inseminated with the normal frozen semen with 50M sperm cells in 0.5 ml. straw and decreases to 45M, 40M, and 35M sperm cells in 0.5ml.straw. The physical characteristics of the neat semen should pass the requirement and determine the number of frozen semen produce when the sperm concentration is reduced. The number of doses and value and the number of carabaos inseminated were analyzed.

The study also looks into the calf drop of animals' impregnated. There was no significant difference between the 50M, 45M, 40M and 35M/dose. When the cost of production was made, there was a significant increase based on number of straws produce from 50 PhP per dose. When the number of cost was translated to number of carabaos inseminated, more animals will be inseminated from 585 to 1,111, 2,063 and 1,641 animals for T₁ to T₄ respectively.

Recommendation

The result of the study is very promising because we could increase the number of frozen semen doses that will be produce without hurting the calf drop of the animals. This could be attained by using CASA and phase contrast microscopy. However, since there were no significant differences, we could bring down the number of semen up to 30M/dose, 25M/dose 20M/dose and 15M/dose of 0.5ml/straw. This may be made base on the recommendation of Andrabi et al. (2006) where it has reported 15×10^6 spermatozoa/0.5ml.dose for insemination did not affect fertility of cryopreserved buffalo bull semen. In here, we will do two batches of artificial insemination to cover all months of the year.

References

- Andrabi, S.M.H, M.Siddique, N.Uledh and L.A. Khan, 2006. Effect of Reducing Sperm numbers per inseminations dose in fertility of cryopreserved buffalo bull semen, Pakistan Vet. 5, 2006, 26(r): 17-19.
- Dejarnette, J.M., R.L. Nebel, C.E. Marshall, 2007. Understanding Estimates of AI Sire fertility, SELECT SIRE (Lecture)
- Hafez, E.S.E. 1987. Reproduction in Farm Animals 5th Ed. Lea and Febigu. Philadechphia
- Mamuad, F.V, H.V. Venturina, E.V. Venturina, R.T. Morcoso, E.C. Atabay and Kazuhiro Kudo, 2005. Artificial Insemination Manual for Water Buffaloes, WBBCIP 2000-2005. Under JICA, PCC and BAI.
- Marphy, C.,A.G. Fahey, A. Shafat and S. Fair, 2013. Reducing sperm concentration is critical to limiting the oxidative stress challenge in liquid bull semen. ADSA. Elsevier Inc.

Reproductive performance of cattle and buffaloes treated with prostaglandin 2_{α} and gonadotropin releasing hormone in Thailand and Philippines

*F.V. Mamuad**¹, K.Dechma³, V. Jamsawat*², C. Saetung², V.L. Mamuad¹, E. V. Venturina¹, E.C. Encarnacion¹, & R.M. Labindao¹*

¹Philippine Carabao Center, Science City of Muñoz, Nueva Ecija 3120, Philippines,²RajamangalaUniversity of Technology Tawan-ok Chonburi, Thailand, ³Sisaket College of Agriculture & Technology, Sisaket, Thailand

Abstract

The use of hormones on the effect on reproductive performance of dairy cattle in Thailand and on beef cattle and buffaloes in the Philippines was made. Cloprostenol (PGF 2_{α}) are used in both countries. The reproduction in cows and heifer in Thailand and buffaloes and Beef Cattle in the Philippines were observed. The estrous signs, conception rate, pregnancy rates and costs were determined. The first study was in Thailand with of 392 heads were used. There are 82 cows and 62 heads of cattle in Thailand and 160 buffaloes and 36 cattle with lulatory and 45 buffaloes and 5 cattle in the Philippines for cloprostenol.

Chi-square and T-test in the Philippines and Chi square test analysis for factorial in CRD in Thailand. DMRT are used to determine for significant differences.

Results show that, dairy cows and heifers responded positively to the GnRH and PGF 2_{α} injection, resulting in higher incidence of estrus. Synchronization of estrus and ovulation by ovsynch protocols could improve percent non-return to estrus and conception rate of dairy cattle. The pregnancy rate of dairy cows and heifer was increased by synchronization of estrus and ovulation.

The result shows no significant change in the reproductive performance of buffaloes using lulatory or cloprostenol, similarly, dairy cows and heifer in Thailand is insignificant. The use of PGF 2_{α} and GnRH in Thailand are almost the same. Environmental condition does not affect their performance. The cost is likely to be used by the AI technicians in the fields. Both are the same in Thailand and Philippines.

It is recommended to use the hormone using the PGF 2_{α} . It is noteworthy to know that the price is cheaper and effective. It is easier to apply under field conditions; however, the technical knowledge of the AI technician in ovarian palpation is necessary.

Keywords: pgf 2_{α} -prostaglandin f 2_{α} ,gnrh-gonadotrophin releasing hormone, dmrt-duncanmultifactor range test

*Corresponding author: fvmamuad@yahoo.com**, virapolj@yahoo.com**

Introduction

Various combinations of synthetic hormones have been used in inducing ovarian activity, postpartum and increasing conception rates. The responses were variable, indicating that physiological effects of hormones may be influenced by age, nutritional status of the cow, body condition and seasonal variations.

Scope and Limitation of the Study

In Thailand it is focused mainly on the effects of GnRH and PGF_{2α} on the reproductive performance of dairy cattle raised in the northeastern part of Thailand which aimed to improve the reproductive performance by the use of hormones: for estrus synchronization, induction of ovulation and conception rate. The number of dairy cattle used in this study was 82 lactating dairy cows and 62 dairy heifers. In the Philippines, treated of FGF_{2α} is normally used in buffaloes and cattle. The use of cloprostenol is a new hormone that is introduced for estrus in buffalo and cattle. This will be the emphasis of this study.

Objectives of the Study

The primary objective of this research is to improve the present level of reproductive performance of crossbred dairy cows (cattle and buffaloes) and heifers raised with the use of hormones.

Materials and Methods

Selected artificial insemination technicians who are conducting estrus synchronization in the villages were given PGF_{2α} for injection to selected buffaloes. Each animal is given PGF_{2α} (cloprostenol and Lutalyze). After 45 to 60 days, the animals are diagnosed for pregnancies and wait to the animal to give calves. Each result was analyzed for T test.

In the Philippines a total of 392 heads were used. The first group of 50 (45 buffaloes and 5 cattle) given estrumate (cloprostenol), the 196 heads of 160 buffaloes and 36 cattle were given Lutalyze. In Thailand 82 heads of lactating dairy herd were given estrumate (Cloprostenol) and GnRH (Buserelin, 0.01 mg) and 62 heads in Thailand for PGF_{2A} and GnRH.

Statistical Design and Analysis

Data collected in the Philippines were analyzed by T-test and in Thailand by Chi-square test analysis for factorial in CRD. Treatment means were compared using Duncan Multiple Range Test (DMRT).

Results and Discussion

The response of the animals, cattle and carabaos, after the injection of cloprostenol, to effect estrus is controlled by hormone after the 72-96 hours interval and has different time of estruses. In cattle, estrus was first detected at 72 hours after injection. The animals showed different signs of estrus, like swelling of the vulva, frequent mucus discharge, urination, bellowing, switching of tail, mounting and allowing to be mounted. The said duration was observed 14-18 hours. The ovaries have already ovulated after 92 hours.

In buffaloes, the first sign was detected after 72 hours after injection. Ovaries were showing a size of 10 nanometer. Only 30% of injected animals showed estrus (pro-estrus), mucus discharged was clear and watery. But on the 92-96 hours, the animal shows standing heat. The animal shows all the clinical symptoms of estrus like reddening of the vulva with good mucus discharge and mounting of other animals. The ovaries showed a 15-20 mm of Graafian follicle.

Table 1. Performance of Animals treated with different Hormones, PGF2.

Hormone /PGF2 α	N	Breed	Age	Return Estrus After 21 days	Response to Monitoring after Delivery Calf Drop/%
Synchoromate (Cloprostenol)	45	Philippine Carabao	5-17	15	18 (40%)
	5	Cattle	3-15	0	5 (100%)
Lutalyze	160	Philippine Carabao	3-12	22	5 (100%)
	36	Cattle	3-15	26	10 (28%)

Table 2. Cost of Hormones/head of cows and heifers.

Hormones	COST Baht/Pesos
A1 = PGF2 α and times AI	= 300/450
A2 = GnRH + PGF2 α ; 2 nd doze GnRH+AI	= 675/1012.5
A3 = GnRH + PGF2 α ; time/AI+ GnRH+AI	= 675/1012.5

Based on the results of this study

Hormone treatment protocols may be applied to lactating dairy cows and heifers without adverse effects on their reproductive performance, particularly non-return to estrus, conception or pregnancy rates. A distinct advantage of hormone treatments 1 or 2 is that rigid observation of estrus signs under field or confinement conditions can be dispensed with by the animal breeder or AI technician. Dairy cows and heifers responded positively to the GnRH and PGF2 α injection resulting in higher incidence of estrus. Synchronization of estrus and ovulation by Ovsynch protocols could improve percent non-return to estrus and conception rate of dairy cattle. There is follicular development or the growth of a new follicular wave in ovaries of cows and heifers after hormone treatment protocols.

The results indicate comparable effects of the three hormone treatments in terms of percent conception rate. For AI, it is thus recommended to apply the hormone used in Treatment 1, which used only PGF2 α . It is noteworthy to know that the price of this hormone is cheaper than the hormones used in Treatments 2 and 3. Also, administration of one hormone is an easier procedure to apply under field conditions; however, the technical knowledge of the AI technician in ovarian palpation is necessary.

Conclusion and Recommendations

The result shows that there was no significant change in the reproductive performance of buffaloes using with lutalyze or cloprostenol. Similarly, dairy cows and heifer in Thailand do not show significant differences in their estrus and conception rate. The use of PGF2 α and GnRH in Thailand are almost the same. PGF2 α alone is enough to get reproductive performance of cattle and buffaloes in the Philippines and in Thailand. Environmental condition does not affect their performance. The cost is likely to be used by the AI technicians in the field.

References

- Abeyguna, Wardena, H. and J. A. De S.Siriwardene, 1999. Cattle and Buffalo Farming: Handbook for Veterinarians. National Science Foundation Press, Colombo, Sri Lanka; 382 pp.
- Bondoc, O. L., M. P. G. Ripoll., N. D. Molina., R. A. Garcia., J. M. Malabanan and N. C. Vera Cruz, 1997. Local heat response parameters and conception rate in estrus-synchronized cattle. Proceeding of the PSAS 34th Annual Convention, 23-24 October 1997. Metro Manila Philippines; pp. 125-132.
- Hafez, E. S. E., 1987. Reproduction in Farm Animals. (5th Edition) Lea &Febiger, Philadelphia; 649 pp.
- Hafez, E. S. E., 1993. Reproduction in Farm Animals. (6th Edition) Lea &Febiger, Philadelphia; 573 pp.

Morphometric dimensions of the spermatozoa in Thai native boar depend on the ejaculates

N. Nasinprom^{*1}, P.i Wongsreesom²,. Kuha², P. Termsombatthaworn², J. Nutthakornku¹ T. Pongjongmit¹ and J. Insree¹

¹ Department of Animal Production Technology, Faculty of Agro-Industrial Technology, Rajamangala University of Technology Isan Kalasin Campus, Kalasin 46000, ² Department of Animal and Fisheries Science, Faculty of Science and Agriculture Technology, Rajamangala University of Technology Lanna Nan, Nan 55000

Abstract

Size and shape of spermatozoa are known to be associated with the fertility. The objectives of this study were 1) to determine the sperm morphometric of Thai native boar and 2) to investigate the effect of ejaculates on morphometric traits of spermatozoa. Semen sample were collected for 5 consecutive weeks from 5 Thai native boars. Spermatozoa were fixed with formaldehyde after stained with Nigrosin-Eosin then smeared on replicated slides. Normal sperm morphometry characteristics were measured on computer screen by used microscope software. The sperm head dimensions by average were 8.67 μm in length, 4.66 μm in width, 0.30 μm in elongation and 1.86 μm in ellipticity. The tail lengths by average were 11.70 μm in midpiece, 33.24 μm in principal piece and 2.64 μm in end piece. The sperm head dimensions including head length, head width, elongation and ellipticity were significantly differences between ejaculates. However, there were not significantly differences in tail lengths between ejaculates. The data suggest that differences in the ejaculates affect on sperm head dimensions in Thai native boar.

Keywords: Ejaculates, Morphometric dimensions, Thai native boar

**Corresponding author: krukeaw@yahoo.com*

Introduction

Meat production from pigs is growing rapidly. Thai native boar has played an important role in smallholder farms and local populations for a long time. They have been raised using low input but they still generate their products and by-products to meet household needs. Moreover, in relation to biodiversity, Thai native boar seems to be a reservoir of genes that could be an asset for future use. One of the key factors for pig production is the reproductive system. Animal reproductive systems can be divided into male and female reproduction. Male fertility depends on the production of normal sperm. Mammalian spermatozoa share the same basic sperm structure consists of a head and a tail. The principal function of a sperm head is to deliver a haploid set of chromosomes to the oocyte. The function of the flagellum is to provide cell motility to allow a spermatozoon to penetrate the boundaries of the female reproductive tract and the zona pellucida. The structural features of the head and tail of a spermatozoon reflect these functional roles (Mortimer, 1997; Pesch and Bergmann, 2006)

Sperm head size and shape have been associated with fertility in human (Chan et al., 1999) and horse (Casey et al., 1997). The tail is the longest part of sperm and consists of midpiece, principle piece and end piece. The midpiece is characterized by mitochondria sheath which provide the energy (ATP) that is mostly used for motility (Pesch and Bergmann, 2006). The

principle piece is the longest segment of the tail. It is most effective for movement. The data indicated that sperm morphometric characteristics associated with semen quality, sperm function and fertility. Thus the study aims to 1) determine the sperm morphometric characteristics and 2) investigate the effect of ejaculates on morphometric traits of Thai native boar spermatozoa.

Materials and methods

Semen Collection

Semen was collected from 5 Thai native boars (3-5 years of age) at Rajamangala University of Technology Lanna Nan. It was collected for 5 consecutive weeks. The ejaculates were taken by means of the gloved-hand technique.

Preparation of Nigrosin-Eosin (NE) Stain

One g of Eosin Y (230251, Sigma-Aldrich, USA), 5 g of Nigrosin (198285, Aldrich, USA) and 3 g of sodium citrate (468500, Unilab, Australia) were mixed. Then, 100 mL of distilled water was added and allowed to mix with the stain components for 20 min in a water bath at 100 °C. The whole content was then filtered with filter paper (1005090, Whatman, England). The stain was placed in a dark glass container and kept at 4 °C until further use.

Sperm Staining

For staining, 100 µL of a semen sample was placed in a 1.5 mL centrifuge tube and fixed with 100 µL formaldehyde for 5 min in a water bath at 37°C. Then 100 µL of NE stain was added and left for 5 min at 37 °C.

Measurement of head dimensions and flagellum length

For each ejaculate 4 slides were prepared for the measurement and counting at 100x magnification, using an oil immersion lens under a light microscope (Leica, DM500, Switzerland) equipped with camera (Leica ICC50 HD, Switzerland) connected to a personal computer. On each slide, around 50 spermatozoa (out of the total number of around 3,000) were measured for their head length, head width and flagellum length by the microscope software (Leica LAS EZ, version 2.0.0, Switzerland). The distances between a-b was accepted as head width (HW); c-d was accepted as head length (HL); d-f was accepted as midpiece length (ML); f-g was accepted as principal piece length (PPL); g-e was accepted as end piece length (EPL) and d-e was accepted as flagellum length (TTL). According to these definitions, ellipticity (HL/HW) and elongation $[(HL-HW)/(HL+HW)]$ could also be calculated (Fig 1)



Fig 1: A typical of boar spermatozoa.

Results and discussion

Parameters for the morphometric characteristics of normal sperm in Thai native boar are presented in Table 1. The head width (4.66 µm) of sperm in this study was similar to the value for Pietrain (4.65 µm) but smaller than the value for Duroc (4.80 µm) reported by Kondracki et

al. (2012a) and larger than the value for Iberian boars (4.07 μm) reported by Gil et al., (2009). In addition, the sperm head length (8.66 μm) of Thai native boar in this study was shorter than the value for Duroc (9.41 μm) and Pietrain (9.20 μm) reported by Kondracki et al. (2012a) but longer than the value for Iberian boars (8.11 μm) reported by Gil et al., (2009). Regarding the tail length, the value for sperm of Thai native boar (47.58 μm) was longer than the value for Duroc (44.72 μm) and Pietrain (44.52 μm) reported by Kondracki et al. (2012a). Taking into account the above data, it is indicated that Thai native boar spermatozoa had lower elongated head shape but longer tail length compared to the sperm from Iberian boars, Duroc and Pietrain.

Table 1. Morphometric characterization of Thai native boar spermatozoa

Sperm morphometric traits	Mean	Minimum	Maximum	SD	CV
Head width (μm)	4.66	4.25	4.95	0.15	3.23
length Head(μm)	8.66	8.25	8.99	0.17	2.00
Ellipticity	1.86	1.68	2.08	0.07	3.87
Elongation	0.30	0.25	0.35	0.01	5.50
length Midpiece(μm)	11.70	11.35	11.99	0.17	1.46
length piece Principle(μm)	33.23	32.51	33.99	0.40	1.20
length piece End(μm)	2.64	2.21	2.95	0.18	6.96
Total tail length (μm)	47.58	46.27	48.70	0.46	0.95
Total sperm length (μm)	56.25	54.91	57.62	0.50	0.86

The morphometric characteristics of Thai native boar spermatozoa according to semen ejaculated are presented in Table 2. The head size and shape were influenced by the ejaculates ($P < 0.01$). The second ejaculates have significant more elongation and ellipticity head shape than other ejaculates ($P < 0.01$). Accordingly, the second ejaculates had the lowest head width and slightly longer head length ($P < 0.01$). However, no significant differences were found in tail morphometric traits between ejaculates ($P > 0.05$), except the was length piece end influenced by the semen ejaculated ($P < 0.01$).

Table 2. The morphometric characteristics of Thai native boar spermatozoa according to ejaculated.

Ejaculates	HW (μm)	HL (μm)	Ellipticity	Elongation	MPL (μm)	PPL (μm)	EPL (μm)	TTL (μm)	TSL (μm)
1	4.67 ^a	8.63 ^c	1.85 ^b	0.29 ^b	11.70	33.27	2.66 ^a	47.64	56.28
2	4.61 ^b	8.67 ^a	1.88 ^a	0.31 ^a	11.70	33.20	2.66 ^a	47.56	56.23
3	4.66 ^a	8.69 ^a	1.86 ^b	0.30 ^a	11.69	33.24	2.61 ^b	47.53	55.22
4	4.67 ^a	8.68 ^a	1.85 ^b	0.30 ^a	11.71	33.24	2.63 ^b	47.57	56.25
5	4.68 ^a	8.65 ^b	1.84 ^b	0.29 ^b	11.71	33.24	2.62 ^b	47.58	56.23

HW, head width; HL, head length; ML, midpiece length; PPL, principal piece length; EPL, end piece length; TTL, flagellum length; TSL, total sperm length

The data presented in this work demonstrated that the ejaculates affect the morphometric characteristics of Thai native boar spermatozoa. This may be because the sperm morphometric can depend on sperm concentration. Banaszewska et al., (2009) observed that there are correlations between the sperm morphometric characteristics and sperm concentrations in an ejaculate. In their study, the semen with the smallest sperm concentration was characterized by spermatozoa with slightly longer head length, higher head width, and longer tail length than the semen with high sperm concentration. Similarly to what was reported in Duroc, it was found that the boar ejaculates with low sperm concentrations are characterized by larger spermatozoa as compared to ejaculates with high sperm concentrations (Kondracki et al. 2011). In addition, the recent studies in bull spermatozoa reported by Kondracki et al. (2012b) show that the less concentrated ejaculates contained spermatozoa with a slightly larger head circumference and a

more elongated head shape in comparison with the spermatozoa in the more concentrated ejaculates.

Conclusions

The sperm head dimensions by average of Thai native boar were 8.66 μm in length, 4.66 μm in width, 0.30 μm in elongation and 1.86 μm in ellipticity. The tail lengths by average were 11.70 μm in midpiece, 33.23 μm in principal piece and 2.64 μm in end piece. The sperm morphometric traits including head length, head width, elongation, ellipticity and end piece length were significantly different between ejaculates. However, there were no significant differences in lengths of midpiece and principal piece between ejaculates. The data suggest that differences in the ejaculates affect sperm morphometric traits in Thai native boar.

Acknowledgements

The authors express their appreciation to the Rajamangala University of Technology Lanna Nan for support funds and facilities.

References

- Banaszewska, D., S. Kondracki and A. Wysokinska. 2009. Effect of sperm concentration on ejaculate for morphometric traits of spermatozoas of the pietrain breed boars. *J. cent. Eur. Agric.* 10: 383-396.
- Casey, P.J., C.G. Gravance, R.O. Davis, D.D. Chabot and I.K.M. Liu. 1997. Morphometric differences in sperm head dimensions of fertile and subfertile stallions. *Theriogenology* 47: 579-589.
- Chan, P.J., H.C.L.D. Johannah U. Corrselli, J.D. Jacobon. W.C. Patton and A. King. 1999. Spermac stain analysis of human sperm acrosomes. *Fert. Steril* 72: 124-128.
- Gil, M.C., M. Garcia-Herreros, F.J. Baron, I.M. Aparicio, A.J. Santos and L.J. Garcia-Marin. 2009. Morphometry of porcine spermatozoa and its functional significance in relation with the motility parameters in fresh semen. *Theriogenology* 71: 254-263.
- Kondracki, S., A. Wysokińska, M. Iwanina, D. Banaszewska, and D. Sitarz. 2011. Effect of sperm concentration in an ejaculate on morphometric traits of spermatozoa in Duroc boars. *Pol. J. Vet. Sci.* 14: 35-40.
- Kondracki, S., M. Iwanina, A. Wysokińska, and M. Huszno. 2012a. Comparative analysis of Duroc and Pietrain boar sperm morphology. *Acta Vet. Brno* 81: 195–199.
- Kondracki, S., D. Banaszewska, A. Wysokińska and M. Iwanina. 2012b. The effect of sperm concentration in the ejaculate on morphological traits of bull spermatozoa. *Folia Biol (Krakow)* 60: 85-91.
- Mortimer, S.T. 1997. A critical review of the physiological importance and analysis of sperm movement in mammals. *Hum. Reprod.* 3: 403-439.
- Pesch, S. and M. Bergmann. 2006. Structure of mammalian spermatozoa in respect to viability, fertility and cryopreservation. *Micron* 37: 597-612.
- Vongpralab, T. 1999. *Reproduction in Domestic Animal*. Department of Animal Science, Faculty of Agriculture, Khon Kaen University (in Thai).

Effects of addition juice date palm to the extender on the percentage of live and motility of frozen thawed bull spermatozoa

Abd. Malik^{1*}, Yayan², IrwanZakir M.¹ & Syarif Djaya, M.¹

¹Department of Animal Science, Faculty of Agriculture. Islamic University of Kalimantan. Banjarmasin- Indonesia. ²Officer Animal husbandry, District of Tanah Bumbu, South Kalimantan. Indonesia

Abstract

The objective of the present study was to investigate the effects of supplementation of juice date palm to the extender on post-thaw percentage of live and motility in bull semen. A total of four Bali bull cattle were used to the study. Juice date palm was added at the concentration of 0.1%, 0.2%, 0.3% and 0.4% to bovine semen cryoprotective medium. The cryoprotective extender for the control group was the same as that for the treatment groups except that it was not supplemented with juice date palm. The results indicated that percentage of live sperm and motility on fresh sperm was no significant different ($P > 0.05$) between control and all treatments. Whereas, sperm motility of frozen thawed was significantly different ($P < 0.05$) between control and all treatments. Furthermore, percentage of live sperm was no significant different ($P > 0.05$).

Keywords: *Juice date palm, spermatozoa, frozen thawed, percentage of live and motility*

Correspondent author: *sidol_99@yahoo.com*

Introduction

Date palm is one of the oldest fruit crops grown in the Arabian Peninsula, North Africa, and the Middle East. Fruits of date palm (*Phoenix dactylifera* L.) is rich in mineral salts and vitamins (Booijet al., 1992). Dates contain small amounts of vitamins C, B1 thiamine, B2 riboflavin and nicotinic acid (Al Shahibet al., 2002), and studies have shown that dates have strong antioxidant (Al Farsi et al., 2005). Date palm fruits consist of 3 main parts: date flesh, date pit, and skin. The glucose, fructose and sucrose are the main sugars of date flesh. The palm date fruit has a high content of sucrose at early stages of maturing, but during the maturation process it is converted to glucose and fructose. Proteins appear in date fruits as 1–3% of dry matter, while its fat content was reported to be 0.52–3.25% (Myharaet al., 1999). Existing sugar palm juice is an important ingredient for sperm. Thus, the objective of this study was to investigate the effects of addition juice date palm to the extender on the percentage of live and motility of frozen thawed bull spermatozoa.

Materials and Methods

This study was conducted at the artificial insemination center Banjarbaru south Kalimantan Indonesia. A total of four Bali bull cattle were used to the study. The cryoprotective extender for the treatments follows: skim milk, glucose, egg yolk and glycerol according to Bustamante et al. (2009), and was supplemented with different concentrations of juice date palm from Aljazirah

Ltd with 0.1%, 0.2%, 0.3%, and 0.4 %. The cryoprotective extender for the control group was the same as that for the treatment groups except that it was not supplemented with juice date palm. The semen was collected from the Bali cattle bulls as long as two months with the aid of an artificial vagina. Immediately, after collection, the semen was kept in a water bath (37°C), and semen parameters were assessed, including volume, pH, consistency, color and concentration of the semen. The percentage of live sperm was evaluated using eosin-nigrosin stain according to Dott & Foster (1972) whereas, the analysis motility of sperm was adopted from Hanget al. (2009).

Results and discussion

The effect of addition juice date palm to the extender on the percentage of live and motility of frozen thawed Bali bull spermatozoa has been presented in table 1 and 2. Based on the evaluation of fresh semen are shown in table 1, overall parameters of sperm characteristic were considered as standard. The sperm motility of frozen thawed are shown in the table 2.

Table 1. Percentages of fresh sperm including percentage of live sperm and motility.

Item	Control	Concentration of juice date palm (%)			
		0.1	0.2	0.3	0.4
Percentage of live sperm (%)	81.25±5.56	80.50±3.11	79.62±5.94	80.15±4.75	79.10±4.40
Motility (%)	73±2.43	75±1.05	74±4.12	72±2.57	72±1.90

Table 2. Percentages of sperm of frozen thawed including percentage of live and motility.

Item	Control	Concentration of juice date palm (%)			
		0.1	0.2	0.3	0.4
Percentage of live sperm (%)	61.75±4.06	68.50±1.02	67.50±1.96	61.75±1.78	65.25±3.25
Motility (%)	43.50±1.00 ^a	41.25±1.26 ^{ab}	39.75±1.26 ^b	36.00±1.41 ^c	31.00±2.94 ^d

^{a,b,c}Values in the same row with different superscripts indicate significant difference (P<0.05).

The assessment of percentage of live sperm and motility is one of the most often used parameters for semen evaluation. The results of this study indicate that percentage of live sperm and motility shown decreased between fresh semen and post thawed. Cryopreservation is a major cause of damage to the sperm thawed (Hauganaet al., 2007; Hong *et al.*, 2009). These were probably due to the induction of reactive oxygen species (ROS) produced from cryopreservation can also induce damage to sperm thawed (Mammotoet al., 1996; Alemayehu, 2011). On the other hand mitochondria are the source of sperm energy, and damage to their structure during the cryopreservation process is associated with reduced post-thaw including sperm viability and motility (Ortega et al., 2008).

References

- Alemayehu, L., 2011. Artificial Insemination in Farm. Chapter 12. Effect of cryopreservation on sperm quality and fertility. Published CC.BY-NC-SA 3.0. Doi.10.5772/16563.
- Al-Farsi, C. Alasalvar, A. Morris, M. Baron and F. Shahidi, 2005. Comparison of antioxidant activity, anthocyanins, carotenoids, and phenolics of three native fresh and sun-dried date (*Phoenix dactylifera* L.) varieties grown in Oman. *J. Agric. Food Chem.* 53: 7592–7599.
- Al-Shahib, J.R. Marshall, 2002. Dietary fibre content of dates from 13 varieties of date palm *Phoenix dactylifera*. *Int. J. Food Sci. Technol.* 37 (6): 719–722.

- Booij, G. Piombo, J.M. Risterucci, M. Coupe, D. Thomas and M.Ferry, 1992. Study of the chemical composition of dates at various stages of maturity for varietal characterization of various of date palm cultivars (*Phoenix dactylifera* L.), *Fruit Paris* 47: 667–677.
- Bustamante F, IC, C.D. Pederzoli, A.M. Sgaravatti, R.M. Gregory, C.S. Dutra Filho M.I.M. Jobim, R.C. Mattos. 2009. Skim milk-egg yolk based semen extender compensates for non-enzymatic antioxidant activity loss during equine semen cryopreservation. *Anim. Reprod.* 6 (2): 392-399,
- Chatterjee, S., Gagnon, C. 2001. Production of reactive oxygen species by spermatozoa undergoing cooling, freezing, and thawing. *Mol. Reprod. Dev.* 59: 451–458
- Dott HM, Foster GC. 1972. A technique for studying the morphology of mammalian spermatozoa which are eosinophilic in a differential 'live/dead' stain. *J.ReprodFertil.* 29: 443-445.
- Hong, J.H.U., Wang, Q.L.I., Chen, Y.L., Jlang, Z.L., Jia, Y.H., Wang, L.Q., and Ou, B.B. 2009. Effects of addition of vitamin B12 to the extender on post-thaw motility, acrosome morphology, and plasma membrane integrity in bull semen. *Turk. J. Vet. Anim. Sci.* 35: 379 – 384
- Haugana, T, Y.T. Gröhn, E. Kommissrud, E. Ropstad, O. Reksen. 2007. Effects of sperm concentration at semen collection and storage period of frozen semen on dairy cow conception. *Anim. Reprod. Sci.* 97: 1-11.
- Mammoto, A., Masumoto, N., Tahara, M., Ikebuchi, Y., Ohmichi, M., Tasaka, K., and Miyake, A. 1996. Reactive oxygen species block sperm-egg fusion via oxidation of sperm sulfhydryl proteins in mice. *Biol. Reprod.* 55: 1063-1068.
- Myhara, R. M., Karkalas, J. & Taylor, M. S. 1999. The composition of maturing Omani dates. *J. Sci. Food Agric.* 79:1345-1350.
- Ortega Ferrusola, C., Sotillo-Galán, Y., Varela-Fernández, E., Gallardo-Bolaños, J.M., González Fernández, L., Tapia, J.A., Peña, F.J. 2008. Detection of apoptosis like changes during the cryopreservation process in equine sperm. *J. Androl.* 29: 213–221.

Antibacterial activity of wood vinegar against *Salmonella* Enteritidis and *Salmonella* Typhimurium

Sunisa Sirimongkolvorakul^{1*}, Kornkamon Paditporn² & Rachakris Lertpatarakomol³

¹ Department of Pre-Clinical Veterinary Science, ²Department of Clinical Veterinary Science, ³Department of Animal Science and Veterinary Basic Science, Faculty of Veterinary Medicine, Mahanakorn University of Technology, 140 Cheum-Sampan Rd., Nong Chock, Bangkok 10530 Thailand.

Abstract

The *in vitro* antibacterial activities of wood vinegar were investigated against *Salmonella* Enteritidis and *Salmonella* Typhimurium. The antibacterial effect was tested by minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC) methods. Wood vinegar's pH was also evaluated. The broth-determined MIC was found at 10% (v/v). The agar-determined MBC was noticed at 1% (v/v) in both those bacteria strains. Results indicated that wood vinegar showed antibacterial activities against *Salmonella* Enteritidis and *Salmonella* Typhimurium. Moreover, we found that wood vinegar at corresponded concentration had pH at 4.15-4.59. These suggested that the efficacy of wood vinegar may be associated with their pH and its constituents. The current study might be applied wood vinegar for an alternative antibacterial agent towards the *Salmonella* Enteritidis and *Salmonella* Typhimurium treatment.

Keywords: wood vinegar, antibacterial activity, *Salmonella* Enteritidis, *Salmonella* Typhimurium

*Corresponding author: sirikul_sunii@hotmail.com

Introduction

Many diseases are known to be treated with herbal medicines as well as some natural products throughout the history of mankind. Amongst those reports, wood vinegar was reported to treat skin infection and also used as insect repellent, antibacterial agents, alkali bath and food additive. It is natural product obtaining in the process of making charcoal. It has a number of chemicals components mainly include acetic acid, phenol, methanol, acetone, and tar compounds (Cerezo et al., 2010). Constituents of wood vinegar showed variability depended on wood species and process conditions.

Salmonellosis is a bacterial food-borne pathogen worldwide (Wang et al., 2015, Rivoal et al., 2009, Tauxe, 2002). *S. Enteritidis* and *S. Typhimurium* are the most common serovar in human and animals. Due to *Salmonella* spp. are transmitted through the human/animal reservoir and have also been isolated from Thai seafood and chicken products (Bernbom et al., 2009). Therefore, it has been increasing interest in the improvement of new agents of which effective and nontoxic antimicrobial compounds. Moreover, some natural products and their constituents are recognized to be safe due to their traditional use. However, the reports of those antimicrobial agents are not well document.

Since wood vinegar is naturally occurring therefore, it is suitable for using as an alternative antibacterial agent. Thus, the aim of this study was to investigate the *in vitro*

antibacterial activities of wood vinegar against *Salmonella* Enteritidis and *Salmonella* Typhimurium.

Materials and Methods

Salmonella Enteritidis (DMST 15676) and *Salmonella* Typhimurium (DMST 17242) were used in this test. Equal volumes of each bacterial strain culture, containing approximately 1×10^5 CFU/mL, were applied into Mueller Hinton broth (MHB, Difco, USA) with wood vinegar at concentrations ranging from 0.001 to 100% (v/v) in tubes. These serially diluted cultures were then incubated at 37°C for 24 h. Turbidity indicates growth of the microorganism and MIC was defined as the lowest concentration where no growth is visually observed. Subsequently, 100 μ l of MIC tubes was placed on Mueller Hinton agar (MHA, Difco, USA) and incubated at 37 °C for 24 h. To determine the MBC, the dilution representing the MIC and at least two of the more concentrated dilutions are plated and to determine viable CFU/ml.

Results

Results obtained in this study revealed that wood vinegar posses potential antibacterial activity against *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242). After incubation, the tubes were visually investigated to determine whether *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242) grew. The 10% (v/v) of wood vinegar inhibited the visible growth of both *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242) (no turbidity) was defined as MIC (Fig 1A and Fig 1B). For further MBC determination, 10 μ L of bacteria suspension from clearly visible tubes was grew on MHA and cultured for 24 hr. The MBC was noticed at 1% (v/v) in both those bacteria strains. Moreover, we found that wood vinegar at corresponded concentration had pH at 4.15-4.59.

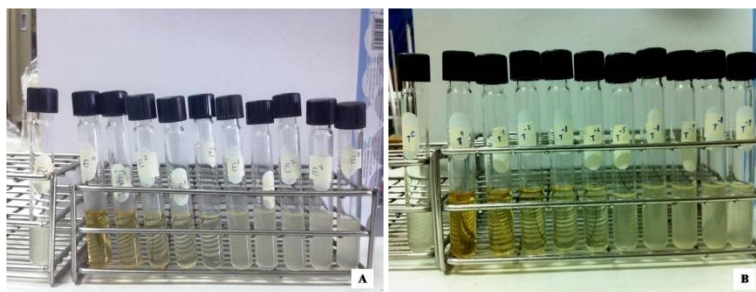


Fig 1. Wood vinegar against *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242). The inhibition of growth (MIC) was observed in both tested organisms at 10% (v/v) of wood vinegar.

Discussion and Conclusion

Natural products are interesting source of potential new antibacterial agents. The first step towards this goal is the *in vitro* antibacterial activity test. However, not many studies are available to identifying on the benefit of those products. In this study, wood vinegar showed the activity against *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242). Though, a variety of natural products showing an antimicrobial activity have also been reported (Lee et al., 2006; Zweifel & Stephan, 2012), variation of MIC still exist. The susceptibility of *S. Enteritidis* (DMST 15676) and *S. Typhimurium* (DMST 17242) to wood vinegar at different concentrations were examined and screened with broth dilution method. The 10% (v/v) of wood vinegar had strongest anti-*S. Enteritidis* (DMST 15676) and anti-*S. Typhimurium* (DMST 17242) activities, whereas MBC was noticed at 1% (v/v) in both those bacteria strains. The results indicate that

wood vinegar can be used as useful source for novel product against *S. Enteritidis* and *S. Typhimurium*. However, potential efficacy of wood vinegar remains to be elucidated into mechanisms of activity against *S. Enteritidis* and *S. Typhimurium*.

References

- Bernbom, N., Y. Y. Ng, C. Paludan-Müller and L. Gram, 2009. Survival and growth of *Salmonella* and *Vibrio* in *som-fak*, a Thai low-salt garlic containing fermented fish product. *Int. J. Food Microbiol.* 134: 223-229.
- Cerezo, A. B., W. Tesfaye, M. E. Soria-Díaz, M. J. Torija, E. Mateo, M. C. Garcia-Parrilla and A. M. Troncoso, 2010. Effect of wood on the phenolic profile and sensory properties of wine vinegars during ageing. *J. Food Comp. Anal.* 23: 175–184.
- Lee, M. H., H. A. Kwan, D. Y. Kwan, H. Park, D. H. Sohn, Y. C. Kim, S. K. Eo, H. Y. Kwan, S. W. Kim and J. H. Lee, 2006. Antibacterial activity of medicinal herb extracts against *Salmonella*. *Int. J. Food Microbiol.* 111: 270-275.
- Rivoal, K., J. Protais, S. Queguiner, E. Boscher, B. Chidaine, V. Rose, M. Gautier, F. Baron, N. Grosset, G. Ermel and G. Salvat, 2009. Use of pulsed-field gel electrophoresis to characterize the heterogeneity and clonality of *Salmonella* serotype Enteritidis, Typhimurium and Infantis isolates obtained from whole liquid eggs. *Int. J. Food Microbiol.* 129: 180-186.
- Tauxe, R.V., 2002. Emerging food borne pathogens. *Int. J. Food Microbiol.* 78: 31-41.
- Wang, Y., B. Yang, Y. Wu, Z. Zhang, X. Meng, M. Xi, X. Wang, X. Xia, X. Shi, D. Wang and J. Meng, 2015. Molecular characterization of *Salmonella enterica* serovar Enteritidis on retail raw poultry in six provinces and two National cities in China. *Food Microbiol.* 46: 74-80.
- Zweifel, C. and R. Stephan, 2012. Spices and herbs as source of *Salmonella*-related foodborne diseases. *Food Rev. Int.* 45: 765-769.

Virucidal efficacy of *Clinacanthus nutans* and *Houttuynia cordata* extract against virulent Newcastle Disease virus

Punyadarsaniya¹, D., S. Sirimongkolvorakul¹, J. Mitchaothai² & S. Ruenphet^{1*}

¹Pre-clinical Veterinary Science Department, ²Clinical Veterinary Science Department, Faculty of Veterinary Medicine, Mahanakorn University of Technology, 140 Cheum-Sampan Rd., Nong Chock, Bangkok 10530 Thailand

Abstract

Determining the virucidal efficacy of *Clinacanthus nutans* (*C. nutans*) and *Houttuynia cordata* (*H. cordata*) extract against virulent Newcastle disease viruses (vNDV) was the aim of this study. Both candidate agents were extracted using 50% and 70% ethanol and then were stock at 200 mg/ml of concentration until test. Concentration as 12.5, 50, 100 and 200 mg/ml of both candidate agents were tested for virucidal efficacy by inoculating vNDV into the cell culture of chicken embryo fibroblasts (CEFs). The neutralizing index (NI) to vNDV was determined to evaluate virucidal efficacy for the present study. The results have illustrated that minimal concentration of *H. cordata* using 50% and 70% ethanol extracted, could inactivate vNDV at concentration of 12.5 and 100 mg/ml, respectively. In the meantime, *C. nutans* extracted by 50% ethanol could inactivate vNDV at only concentration of 200 mg/ml. Therefore, the present study could be concluded that the ethanol extracts of *C. nutans* and *H. cordata* have a virucidal property against vNDV at different concentrations and they might be used as an alternative disinfectant for bio-security purpose, particularly in poultry farms.

Keywords: virucidal efficacy, *Clinacanthus nutans*, *Houttuynia cordata*, Newcastle disease virus

* Corresponding author: rsakchai@hotmail.com

Introduction

Herbal medicine have been long history especially Chinese have utilized herbs and plants to treat various diseases for more than 8,000 years (Drasar & Moravcova, 2004). Since 1980s, the World Health Organization estimated that about 80% of the world's population relied on traditional medicines including herb medicines for their primary health care needs (Farnsworth et al., 1985).

In Thailand, the leaves of *Clinacanthus nutans* (*C. nutans*) have long been traditionally used such as an anti-inflammatory drug for the treatment of insect bites (Sakdarat et al., 2008). Satayavivad, et al. (1996) reported that the leaves extract possess analgesic and anti-inflammatory activities, and several researcher described antiviral activities such as varicella-zoster virus and herpes simplex virus type-2 (Thawaranantha, et al., 1992; Jayavas, et al., 1992a; Jayavas, et al., 1992b; Sangkitporn, et al., 1995)

Houttuynia cordata (*H. cordata*), called in Thai “*Khao-tong* or *Plu-khao*” is well known traditionally used as a medicinal material in the indigenous medicine systems of Southeast Asia, especially Thailand. The functions of *H. cordata* are relieve fever, resolving toxin, reducing swelling, draining pus and promoting urination (Zheng et al., 1998). Several researchers reported *H. cordata* was virucidal, such as Severe Acute Respiratory Syndrome (SARS) (Lau et al., 2008).

The objective of this study is to evaluate the efficacy of *C. nutans* and *H. cordata* extract against virulent Newcastle disease that is important viral diseases of birds, especially commercial chickens. They could potentially be used as an alternative disinfectant for enhancement of bio-security at poultry farms.

Materials and Methods

Virus and cell: Virulent Newcastle disease viruses (vNDV), namely NDV-G7 strain, that have titer as $10^{7.5}$ ELD₅₀, and chicken embryo fibroblast (CEF) cells, were used for the present study.

Extraction: The air-dried leaves of *C. nutans* and *H. cordata* leaves were sequentially extracted using 50% and 70% ethanol. Briefly, 50 gram of air-dried leaves of *C. nutans* and *H. cordata* were pre-extracted using 500 ml of 50% and 70% ethanol at room temperature for 24 hr. Each extract was filtered through filter cloth and a 0.45 µm filter (Nalgene®, Thermo scientific, USA) with vacuum pump. *C. nutans* filtrate was evaporated using a rotary evaporator below 50°C and then evaporated on 60°C water bath for 5 hr while *H. cordata* filtrate was only evaporated on 60°C water bath for 8 hr. Then crude extracts were dissolved using maintenance medium (MM) containing 1% dimethyl sulfoxide (DMSO) as 1/50 of weight/volume (concentrated 200 mg/ml) for stocking solution. Each diluted extract was aliquot as 500 µl/tube and keep at -30°C until test.

Viral inactivation procedure: The each 50% and 70% ethanol stock solution of *C. nutans* and *H. cordata* was diluted as 200, 100, 50, 25, and 12.5 mg/ml for virucidal testing. Four hundred and fifty microliter (µl) of each treatment was mixed with 50 µl of vNDV. The mixer was diluted in serial 10-fold dilutions for virus titration using CEF cells into 96-well cell-culture plates for 4 wells per dilution. The cytopathic effect (CPE) was observed twice a day and confirmed by hemagglutination assay (HA) at 3 days post inoculation. The virus titer was calculated by the Behrens-Kaerbel method with HA results using 50% tissue culture infectious dose (TCID₅₀).

Viral inactivation index: Neutralizing index (NI) was calculated using the following equation: $NI = t_{pc} - t_a$ (Belsham et al., 2011), where “ t_{pc} ” is the converted titer of the positive control (no sample-treated), and “ t_a ” is the titer converted into an index in log₁₀ of the virus recovered from the sample-treated tube. Inactivation of viruses was considered effective when the NI > 3.0 (Lombardi et al., 2008.; Takehara et al., 2010).

Results

Table 1 shows the neutralizing index (NI) to virulent Newcastle disease virus (vNDV) of *H. cordata* and *C. nutans* extracted by 50% and 70% ethanol using concentration as 200, 100, 50, 25 and 12.5 mg/ml. Minimal concentration of *H. cordata* using 50% and 70% ethanol extraction, could be inactivated vNDV as 12.5 and 100 mg/ml, respectively while *C. nutans* extracted by 50% ethanol could only inactivated as 200 mg/ml.

Table 1. Neutralizing index (NI) to virulent Newcastle disease virus (vNDV) of *H. cordata* and *C. nutans* extracted by 50% and 70% ethanol using concentration as 200, 100, 50, 25 and 12.5 mg/ml.

Item	Neutralizing index (Log ₁₀ TCID ₅₀ /ml)				
	200 (mg/ml)	100 (mg/ml)	50 (mg/ml)	25 (mg/ml)	12.5 (mg/ml)
<i>H. cordata</i> at 50%	-	6.00±0.00	4.88±0.53	4.50±0.00	3.00±0.00
<i>H. cordata</i> at 70%	5.75±0.71	4.58±1.63	2.25±0.00	-	-
<i>C. nutans</i> at 50%	4.63±0.18	1.33±0.14	-	-	-
<i>C. nutans</i> at 70%	1.25±0.00	0.63±0.18	-	-	-

- stands for “not test”

Summary and discussion

Usually, mostly viruses especially lipid envelope viruses such as NDV, are easily inactivated by organic solvents, detergents and chemicals (Swayne & Halvorson 2008). The present study was showed the virucidal efficacy of *H. cordata* and *C. nutans* extracted from 50% and 70% ethanol. These results indicated that both extracts could be inactivated vNDV however efficacy of *H. cordata* is better than *C. nutans* (Table 1).

Bauer et al. (1996) described that *H. cordata* contains groups of such chemical components as flavones, essential oil and alkaloids. During the period of the Severe Acute Respiratory Syndrome (SARS) outbreak, it was one of the ingredients in the SARS prevention formulas recognized by the Health Ministry of China (Lau et al., 2008).

Sakdarat et al. (2008) described that *C. nutans* is consisted 8 compounds related to chlorophyll a and chlorophyll b. *C. nutans* extracts from the leaves were reported to possess analgesic and anti-inflammatory activities (Satayavivad et al., 1996), antiviral activities against varicella-zoster virus (Thawaranantha et al., 1992) and herpes simplex virus type-2 (Jayavasu et al., 1992a).

We believe that the isolation of new active principles for drug discovery from individual perspective, and establishment of detail chemical profiles for standardized extracts from holistic perspective would be of great scientific merit. In addition, they might be used as an alternative disinfectant for bio-security purpose, particularly in poultry farms.

Acknowledgements

We thank Pakawadee Kumpolngam, Yaowalak laksub, Theerasak Kunwandee, Nattapong Seungsui, Preeyanuch Sudphawa and Pattaradanai Musksing for their excellent assistance. This study was supported by Mahanakorn University of Technology, Thailand.

References

- Bauer, R, A. Proebstle and H. Lotter, 1996. Cyclooxygenase inhibitory constituents from *Houttuynia cordata*. *Phytomedicine*. 2(4): 305-308
- Belsham, G. J., S. M. Jamal, K. Tjornehoj and A. Botner, 2011. Rescue of foot-and-mouth disease viruses that are pathogenic for cattle from preserved viral RNA samples. *Plos One*. 6(1): e14621
- Drasar, P, and J. Moravcova, 2004. Recent advances in analysis of Chinese medicinal plants and traditional medicines. *J. Chromatography B*. 812(1): 3-21
- Farnsworth, N. R., O. Akerele, A. S. Bingel, D. D. Soe-jarto and Z. Guo, 1985. Medicinal plants in therapy. *Bulletin of the World Health Organization*. 63(6): 965-981
- Jayavasu, C., K. Balachandra and S. Sangkitjaporn, 1992a. Clinical trial in the treatment of genital herpes patients with *Clinacanthus nutans* extract. *Com. Dis. J*. 18: 152-161
- Jayavasu, C., T. Dechatiwongse and K. Balachandra, 1992b. The virucidal activity of *Clinacanthus nutans* Lindau extract against *Herpes simplex* virus type-2: an in vitro study. *Bull. Dept. Med. Sci*. 34: 153-158
- Lau, K. M., K. M. Lee, C. M. Koon, C. S. F. Cheung, C. P. Lau, H. M. Ho, M. Y. H. Lee, S. W. N. Au, C. H. K. Cheng and C. B. S. Lau CBS, 2008. Immunomodulatory and anti-SARS activities of *Houttuynia cordata*. *J. Ethnopharma*. 118(1): 79-85
- Lombardi, M. E., B. S. Ladman, R. L. Alphin and E. R. Benson, 2008. Inactivation of avian influenza virus using common detergents and chemicals. *Avian Dis*. 52: 118-123
- Sangkitjaporn, S., K. Balachandra, M. Bunjob, S. Chaiwat, T. D. Na-Ayudhya, C. Jayavasa, 1995. Treatment of *Herpes zoster* patients with *Clinacanthus nutans* (Bi Phaya Yaw). *J. Med. Assoc. Thailand*. 78: 624-627

- Sakdarat, S., A. Shuyprom, T. D. Na-Ayudhya, P. G. Waterman and G. Karagianis, 2008. Chemical composition investigation of the *Clinacanthus nutans* Lindau leaves. Thai J. Phytopharm. 13(2): 13-24
- Satayavivad, J., N. Bunyaoraphatsara, S. Kitisiripornkul and W. Tanasomwang, 1996. Analgesic and anti-inflammatory activities of extract of *Clinacanthus nutans* Lindau. Thai J. Phytopharm. 3: 7-17
- Swayne, D. E. and D. A. Halvorson, 2008. Disease of Poultry, 12th ed. Y. M. Saif, H. J. Barnes, A. M. Fadly, J. R. Glisson, L. R. McDougald, and D. E. Swayne, eds. Blackwell Science, Ames.
- Takehara, K., K. Yamazaki, M. Miyazaki, Y. Yamada, S. Ruenphet, A. Jahangir, D. Shoham, M. Okamura and M. Nakamura, 2010. Inactivation of avian influenza virus H1N1 by photocatalyst under visible light irradiation. Virus Res. 151: 102-103
- Thawaranantha, D., K. Balachandra, S. Jongtrakulsiri, P. Chavalittumrong, J. Bhumiswasdi and C. Janyavasud, 1992. *In vitro* antiviral activity of *Clinacanthus nutans* on varicellazoster virus. Siriraj Hosp. Gaz. 44: 285-291
- Zheng, H., Z. Dong and J. She, 1998. Modern study of traditional Chinese medicine. Xue Yuan Press, Beijing

Ciliary activity and life span of swine precision-cut lung slices: Comparison between changed and unchanged medium

S. Rattanachan¹, D. Punyadarsaniya^{2*}, S. Ruenphet², B. Chuppava³, & G. Herrler⁴

¹Aquatic animal clinic, Faculty of Veterinary Medicine, Mahanakorn University of technology, Bangkok, Thailand, ²Virology and Immunology Department, Faculty of Veterinary Medicine, Mahanakorn University of technology, Bangkok, Thailand, ³Student of Veterinary Medicine faculty, Mahanakorn University of technology, Bangkok, Thailand, ⁴Institute of Virology, University of Veterinary Medicine, Hannover, Germany

Abstract

Precision-cut lung slices (PCLS) offer the possibility of investigating thin tissue culture material under various changing conditions. In PCLS, ciliary function is pertained, and the interactions of cilia with infectious agents and its functional aspects can be investigated. PCLS is possible to evaluate morphologic changes over a longer period of time using only a small group of animals. To investigate the life span of cilia in changed and unchanged medium groups, PCLS were prepared from lungs of three months old crossbred pigs and shown fully retained of the epithelial cells viability. One of the groups was changed medium daily and another was not changed until the end of the experiment. The result revealed that the changed medium group retained full ciliary activity for 10 days, longer than the unchanged medium group for 4 days. This information could help for design the experiment for this culture system in the future.

Keywords: precision-cut lung slices, ciliary activity, tissue culture

**Corresponding author: darsaniya_p@yahoo.de*

Introduction

Culture system for differentiated respiratory epithelial cells is precision-cut lung slices (PCLS), where the epithelial cells are maintained in their original setting. In addition to ciliary activity and bronchoconstriction, this culture system provides another characteristic feature of the airway (Ebsen et al., 2002). Prior to the first studies using agar-filled precision-cut lung slices published in 1992 (Stefaniak & Brendel, 1992) lung slices were used as an *in vitro* system for many studies which is proved to be an efficient method to do an experiment for virological (Goris et al., 2009; Punyadarsaniya et al., 2011; Meng et al., 2013), toxicological (Henjakovic et al., 2008), pharmacological (Henjakovic et al., 2008) and physiological (Morin et al., 2013) approaches. PCLS are efficient *ex vivo* maintaining functions one of the most similar to *in vivo*. To determine the viability of the PCLS, the results of the study are important in various fields such as the viruses which require only living cells to proliferate. The ciliary activity represents cell viability of respiratory epithelial cells (Punyadarsaniya et al., 2011). Determination of the viability of cells in different ages, leads to know the appropriate time to use the PCLS in the experiment. The aim of this study was to investigate ciliary activity life span of PCLS to compare them with changed and unchanged medium.

Medium is a mixture of inorganic salts and other nutrients capable of sustaining cell survival *in vitro* for 24 hours. For the meaning of maintenance medium this will retain cell survival without growth. Changing the medium or Feeding a culture can keep the cells healthy

by providing fresh nutrients, while cell passage or splitting is required to maintain cells in exponential growth. Given the necessary care and attention, most cell lines are easy to maintain and grow (Freshney, 1987). More recent paper detailed the long-term maintenance of lung slices cultured in defined media (Siminski et al., 1992). In addition, precision-cut lung slices have excellent preservation of the ciliated layer on bronchial epithelium while connective tissue mast cells remained granulated throughout the culture period (Parrish et al., 1995).

Materials and Methods

Ethics statement

Pigs used for these experiments were kept in the Clinic for Swine and Small Ruminants for demonstration and student veterinary training (approval number 33.9-42502-05-09A627). All studies were carried out in strict accordance with the recommendations of the European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes (European Treaty Series, nos. 123 [<http://conventions.coe.int/Treaty/en/Treaties/Html/123.htm>] and 170 [<http://conventions.coe.int/Treaty/en/Treaties/Html/170.htm>]). The protocol was approved by the national permitting authorities (animal welfare officer of the University of Veterinary Medicine, Lower Saxony State Office for Consumer Protection and Food Safety). All measures were in accordance with the requirements of the national animal welfare law. Killing and tissue sampling were performed under sodium pentobarbital anesthesia, and all efforts were made to minimize suffering.

Precision-cut lung slices (PCLS)

PCLS were prepared from lungs of three months old crossbred pigs originated from conventional farms and housed in the Clinics for Swine and Small Ruminants and Forensic Medicine at the University of Veterinary Medicine, Hannover. Pigs showed no clinical symptoms of respiratory or systemic disease. Immediately after euthanasia with pentobarbital, lungs were carefully removed and the cranial, middle, and intermediate lobes were filled with 37°C warm low-melting agarose (agarose LM GQT; GERBU, Gaiberg, Germany) followed by solidification on ice. Tissue was stamped out as cylindrical portions (8-mm tissue coring tool) and approx. 250 µm thick slices were prepared by using the Krumdieck tissue slicer (TSE systems, model MD4000-01) with a cycle speed of 60 slices/min. PCLS were incubated in 1 ml of RPMI 1640 medium (Invitrogen/Gibco, Germany) containing antibiotics and antimycotics (Amphotericin B, Clotrimazole, Enrofloxacin, Kanamycin, Penicillin/ Streptomycin) per slice in a 24-well plate at 37°C and 5% CO₂. The medium was changed every hour during the first four hours and once after 24 hours in propose to remove the low-melting agarose.

The changed medium group was continued changed medium daily and unchanged medium group was incubated without any treatment. All the experiments were performed at least three times with 6 slices per group.

Ciliary activity assay

PCLS viability were analyzed by observing the ciliary activity under the light microscope (Zeiss Axiovert 35) equipped with an ORCA C4742-80 digital camera (Hamamatsu) and SIMPLE-PCI analysis software (Compix Imaging Systems). To dedicate the ciliary activity score of bronchus, each bronchus was virtually divided into ten segments is shown in Figure1. Each of which was monitored for the presence or absence of ciliary activity. Slices were selected that showed 100% ciliary activity at the beginning of the experiment.

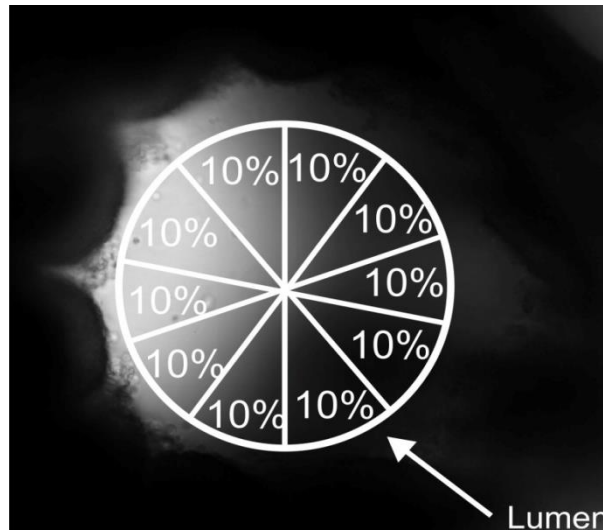


Figure 1. PCLS viability was analyzed by observing the ciliary activity under the light microscope. Each bronchus was virtually divided into ten segments and monitored for the presence or absence of ciliary activity.

Results

The effect of changed and unchanged medium on ciliary activity

The response of ciliary activity to medium changes is shown in Figure 2. On the first day after preparation which presence the 100% ciliary activity and gradually decreased after the ninth day after preparation. However, when the medium was changed then viably occurred. After the ninth day medium changes the ciliary activity was decreased to 90 % and constantly until fourteenth day. In contrast, the result of ciliary activity of PCLS without medium change which presence 100% ciliary activity up to the fifth day after preparation and suddenly decreased to 84% and gradually changed to 75% on the fourteenth day. To compare ciliary activity with changed and unchanged medium were found any relation with ciliary activity of the swine respiratory epithelial cells from this experiment. The slopes of the curves for both of results are similar up to fifth day after preparation but the changed medium are remain 100% ciliary activity up to ninth day, presumably because PCLS was carried out in plates which medium change every 24 hours.

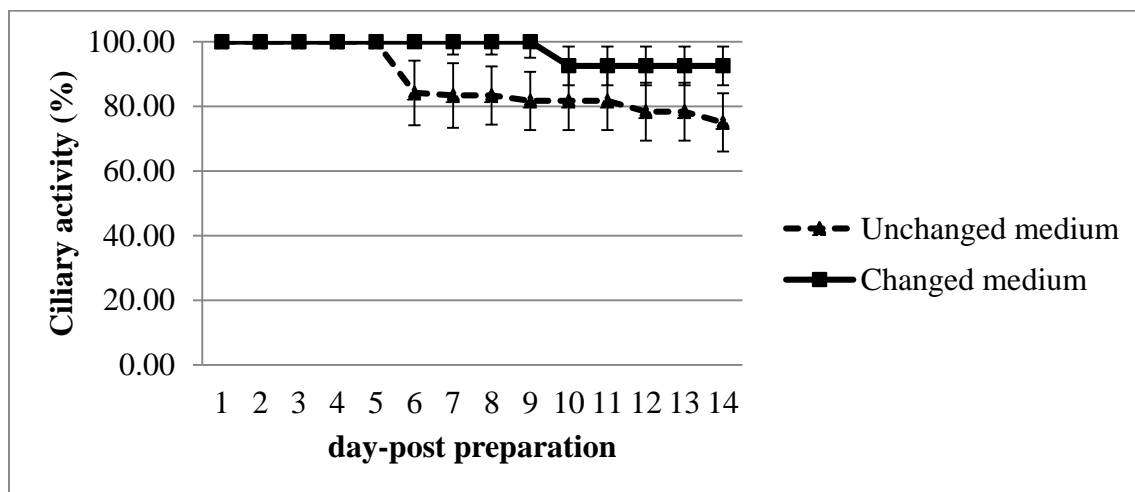


Figure 2. Comparison of ciliary activity of swine PCLS between changed and unchanged medium within fourteen days life span. PCLS were analysed for ciliary activity at daily intervals.

Discussion

In this study, Precision-cut lung slices are an interesting alternative culture system as their preparation is comparably easy providing a large number of slices. Furthermore, the epithelial cells remain differentiated as the original setting lining of the respiratory track. Ciliary activities of epithelial cells were proved to refer as viability of the epithelial cells from PCLS (Punyadarsaniya et al., 2011). This paper reports the effects of a medium change on ciliary activity from the longer life span for changed at frequent intervals group. A direct relationship was found between ciliary activity and PCLS life span of medium changes given to a culture. This was an expected result because normally when a culture is prolonged by changed medium the viability of cell remaining. Medium changes had a profound effect on cellular metabolism (Griffiths, 1970). After a medium change there was a sequential stimulation of cell metabolism in the same order as they were inhibited. The inhibitory mechanism that is affected by cell crowding is obviously reversed by a medium change. Surprisingly that by non-treatment group could found fully retain of ciliary activity until 5 days. The results presented in this paper suggest that the cell is able to take up a sufficient supply of nutrients in a culture depend on the 2 factors that could be involved are, first, a reduced nutrient uptake capacity due to the structure of cell membranes and secondly, the large reduction in cell membrane area exposed to the environment in a confluent culture (Kruse & Miedema, 1965). With the previous reasons also leads to reduction of ciliary activity after 9 days.

In conclusion, this is the study on precision-cut lung slices from pig lungs. The slices are viable for at least 5 days without medium changes. This can apply for the experiment with long treatment period such as viral infection. The changed medium daily method would perfectly use for prolong PCLS viability for a long time. Although the precision-cut slice system is continually evolving with development and modification of new slicing and preservation buffers as well as application of molecular biology techniques to slices, slices are currently a well-established and a powerful *in vitro* model.

References

- Ebsen, M., G. Mogilevski, O. Anhenn, V. Maiworm, D. Theegarten, J. Schwarze and M. Morgenroth, 2002. Infection of murine precision cut lung slices (PCLS) with respiratory syncytial virus (RSV) and chlamydomphila pneumoniae using the Krumdieck technique. *Pathol. Res. Pract.* 198: 747-753.
- Freshney, R. I., 1987. *Culture of animal cells: a manual of basic technique.* Edition 2nd. A.R. Liss, New York, pp 397
- Goris, K., S. Uhlenbruck, C. Schwegmann-Wessels, W. Köhl, F. Niedorf, M. Stern, M. Hewicker-Trautwein, R. Bals, G. Taylor, A. Braun, G. Bicker, M. Kietzmann and G. Herrler, 2009. Differential Sensitivity of Differentiated Epithelial Cells to Respiratory Viruses Reveals Different Viral Strategies of Host Infection. *J Virol.* 83: 1962-1968.
- Griffiths, J.B., 1970. The effect of medium changes on the growth and metabolism of the human diploid cell, WI-38 *J. Cell Sci.* 8: 43-52.
- Henjakovic, M., C. Martin, H.G. Hoymann, K. Sewald, A.R. Ressmeyer, C. Dassow, G. Pohlmann, N. Krug, S. Uhlig and A. Braun, 2008. Ex vivo lung function measurements in precision-cut lung slices (PCLS) from chemical allergen-sensitized mice represent a suitable alternative to in vivo studies. *Toxicol Sci.* 106: 444-453.
- Kruse, P.F. and E. Miedema, 1965. Production and characterization of multiple-layered population of animal cells. *J. Cell Biol.* 27: 273-279.
- Meng, F., D. Punyadarsaniya, S. Uhlenbruck, I. Hennig-Pauka, C. Schwegmann-Wessels, X. Ren, R. Dürrwald and G. Herrler, 2013. Replication characteristics of swine influenza

- viruses in precision-cut lung slices reflect the virulence properties of the viruses. *Vet Res.* 44: 110.
- Morin, J.P., J.M. Baste, A. Gay, C. Crochemore, C. Corbiere and C. Monteil, 2013. Precision cut lung slices as an efficient tool for *in vitro* lung physio-pharmacotoxicology studies. *Xenobiotica.* 43:63–72.
- Parrish, A.R., A.J. Gandolfi and K. Brendel, 1995. Minireview precision-cut tissue slices: application in pharmacology and toxicology. *Life Sci.* 57(21): 1887-1901.
- Punyadarsaniya, D., C.H. Liang, C. Winter, H. Petersen, S. Rautenschlein, I. Hennig- Pauka, C. Schwegmann-Wessels, C.Y. Wu, C.H. Wong and G. Herrler, 2011. Infection of differentiated porcine airway epithelial cells by influenza virus: differential susceptibility to infection by porcine and avian viruses. *PLoS. ONE.* 6: e28429.
- Siminski, J.T., T.J. Kavanagh, E. Chi and G. Raghu, 1992. Long-term maintenance of mature pulmonary parenchyma cultured in serum-free conditions. *Am. J. Physiol.* 262: 105-110.
- Stefaniak, M.S. and K. Brendel, 1992. Biochemical and histological characterization of agar-filled precision-cut rat lung slices in dynamic organ culture as an *in vitro* tool. *In Vitro Toxicol.* 5: 7–19.

Factors influencing the incidence of Babesiosis in sheep of district Toba Tek Singh, Pakistan

Babar*¹, W., T. Rehman¹, RZ. Abbas¹, R. Hussain¹, F. Shahzad¹, A. Sikandar² & MA. Zaman²

¹Department of Parasitology, UCV&AS, The Islamia University of Bahawalpur, Pakistan

²College of Veterinary and Animal Sciences, Jhang

Abstract

The prevalence of Babesia was studied in sheep in district Toba Tek Singh, Punjab, Pakistan from 2009-2010. A cross-sectional survey was performed in order to determine the prevalence of Babesia and effect of various risk factors on its prevalence in sheep of district Toba Tek Singh. A total 43 (9.29%) out of 463 sheep showed the Babesia infection. Month wise peak prevalence was observed in July of 17.95%, while no positive case was registered in Dec-2009 and Jan-2010. The prevalence of Babesia was found to be significantly ($P < 0.05$) dependent to age and sex. The prevalence of Babesia was found 14.18% and 13.67% higher adults and female verses young and male sheep. No correlation was observed between body condition and Babesia prevalence.

Keywords: Sheep, Babesia, factors influencing, management

**Corresponding author: wasim_babar11@hotmail.com*

Introduction

Sheep babesiosis is of considerable economic importance. Babesia is transmitted by hard ticks and causes fever, anemia, haemoglobinuria and icterus in small ruminants. Babesia ovis and Babesia motasi are known to be pathogenic in sheep and goats. The effects of B. ovis are usually less severe than B. motasi (Razmi et al., 2003). Studies on the determination of associated risk factors of protozoan infections have been conducted by various workers in Iran (Razmi et al., 2003a), Greece (Theodoropoulos et al., 2006) and Uganda (Magona et al., 2008). Iqbal et al. (2011) studied prevalence of babesiosis in small ruminants from Southern Punjab, Pakistan. Bashir et al. (2009) performed vector identification studies on canine babesiosis and Rashid et al. (2010) studied prevalence and chemotherapy of babesiosis among lohi sheep in the livestock experiment station, Sahiwal district, Pakistan. There is lack of information on the occurrence and determination of associated risk factors with babesiosis in sheep in central Punjab, Pakistan. Therefore, present study was designed to evaluate the prevalence of Babesia as well as associated risk factors in sheep from district Toba Tek Singh (T.T. Singh), Punjab, Pakistan.

Materials and Methods

Development of questionnaire

The closed ended (dichotomous and multiple choice) questionnaire was developed for collecting necessary information from farmers regarding associated risk factors using according the prescribed method (Thrusfield 2008). Information regarding the following determinants was collected through questionnaire.

1. Age: Animals were divided into two age categories; adults (>6 months) and young-stock (<6 months).
2. Sex: Both sexes were sampled during study.
3. Climate: Season-wise prevalence was noted separately as; (a) Cold season (December-March), (b) Hot season (April-June), (c) Rainy season (July-September) and (d) Post-rainy season (October-November).

Collection of samples and parasitological examination

A total of 463 blood samples were collected from study area. 5-10 ml blood was collected from jugular vein using a sterile syringe in screw capped bottles containing 10mg EDTA as preservative (Iqbal et al., 2006). Thin and thick smears were prepared for identification of blood protozoa according to method given by Iqbal et al (2006). Haemoprotozoa were identified as described by Soulsby, (2006) and Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (2004). The percentage prevalence of Babesia in goats was recorded using following formula.

Statistical Analyses

Logistic analysis was performed using logit model including all variables in the model with backward elimination procedure. Factors with paired characteristics were analyzed using Odds Ratio (OR) and Mantel–Haenszel (MH) chi-square. Hosmer– Lemeshow goodness-of-fit test indicated that model fits well. All the analyses were carried out using SAS software package (1998) at 95% confidence level (SAS 1998).

Results

The prevalence of Babesia among sheep was found to be dependent on various factors. Of the total 463 fecal samples of sheep examined, 43 (9.29%) showed presence of Babesia. Mean OPG was recorded highest in July (2009) and lowest in December-2009 and January-2010. Mean OPG was recorded highest in August (2007) which lies in rainy season in study area. Generally, calves revealed higher mean OPG than adults. The month wise Babesia prevalence is given in fig. 1 which indicates that rain fall has a significant role in Babesia prevalence in sheep. The prevalence of Babesia in sheep from central Punjab, Pakistan was investigated in association of various risk factors. Of the total 463 fecal samples of sheep examined, 43 (9.29%) showed Babesia infection. Month wise results of Babesia prevalence in sheep are shown in Fig. 1. In Apr-2009 the prevalence was observed 7.5 % in 463 examined sheep. The prevalence of Babesia was increased constantly until July-2009. In the month of July-2009 17.95 % samples revealed the presence of Babesia out of total 463 and then there was a decreasing trend and reached 0 % in the month of Dec-2009 and Jan-2010 and again increasing trend was observed, reached 7.5 % in Mar-2010. Weather conditions also seemed to affect the Babesia prevalence in sheep and results obtained are shown in Fig. 1. The highest prevalence of Babesia in sheep was recorded in the months of July and August of 17.95 % and 15.38 %, respectively. The data obtained regarding Babesia infection showed that the rain fall was more supportive as compared to temperature and relative humidity. The prevalence of Babesia was recorded 0% in the months of Dec-2009 and Jan-2010 and in the meantime the rain fall was recorded minimum which indicates the dominant relation of Babesia prevalence with rain fall as compared to temperature and relative humidity. The detailed relationship between rain fall; temperature; relative humidity with Babesia prevalence can be seen in Fig. 2.

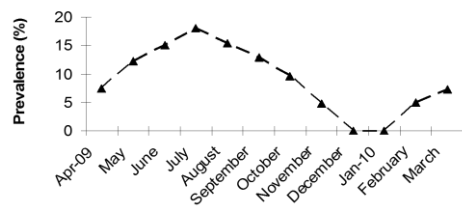


Fig. 1: Month wise prevalence of Babesia in sheep from T. T. Singh District, Punjab, Pakistan

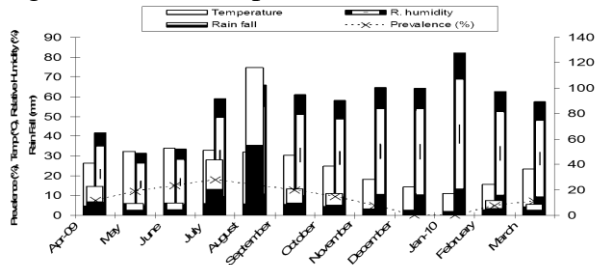


Fig. 2: Association of weather condition with risk of Babesia in sheep from T. T. Singh district, Pakistan

Associated risk factors

The prevalence of Babesia in sheep from T. T. Singh District, Punjab, Pakistan was evaluated with respect to associated risk factor and the result of Babesia relation with age and sex are shown in Fig. 3. Analysis of the hypothesized associated risk factors by stepwise multivariate logistic regression model of Mantel-Haenszel Chi-Square and Hosmer-Lemeshow goodness-of-fit analysis of goat age, sex and breed are shown in table 1 and 2. Statistical analyses revealed that the risk of Babesia infection was strongly predisposed ($P < 0.05$) by age of animal (between young and adults sheep). Higher prevalence was observed in adults sheep (17.52%; 34/194; $\chi^2 = 26.843$; $OR = 4.742$) as compared to young sheep (3.34%; 9/269) (Fig. 3). Sex was found a strongly associated risk factor ($P < 0.05$) influencing the prevalence of Babesia in sheep. Generally higher prevalence was observed in female animals versus male animals. The Babesia prevalence in female and male sheep was found to be 17.08% (34/199; $\chi^2 = 25.137$; $OR = 0.381$) and 3.41% (9/264), respectively. The effect of body condition (poor and good condition) was found non-significant ($P > 0.05$). However, a marginally high prevalence was observed in poor body condition animals than animals with good body condition.

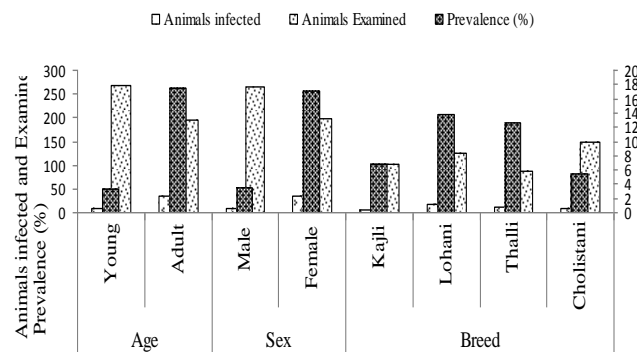


Fig 3: Association of associated risk factor (age, sex and breed) with risk of Babesia in sheep

Table 1: Multivariate logistic regression analysis of associated factors with risk of babesia infection in sheep

Term	Odds ratio	95% C. I.	P-value
Sex	0.381	0.153-0.950	0.038
Age	4.742	1.892-11.885	0.001

HOSMER-LEMESHOW GOODNESS-OF-FIT TEST: P = 0.043

Table 2: Mantel-haenszel chi-square analysis of all hypothesized risk factors with babesia infection

Associated determinants	Variables	Prevalence	Mantel-Haenszel Chi-Square
Age	Young	3.34% (9/269)	26.8432
	Adult	17.52% (34/194)	(<.0001)
Sex	Male	3.41% (9/264)	25.1374
	Female	17.08% (34/199)	(<.0001)
Body condition	Poor	11.44% (23/201)	3.1759
	Good	7.63% (20/262)	(0.0510)

Discussion

During the study, a total of 363 sheep of four breed, including 194 adults and 269 young lambs and of both sex, were examined. The overall prevalence of Babesia was 9.29 % including all risk factor and management practices. The Babesia presence has been reported from Lahore, Sahiwal, lower Punjab, Layyah and Muzafaargarh, Pakistan, but the prevalence as well as its correlation with various risk factors in sheep from central Punjab, Pakistan has not been previously reported. Similar results have also been reported by various other researchers from Pakistan in different animals such as Ahmad et al. (2007 and 2011) reported the prevalence of Babesia prevalence in dogs and cats, respectively from Lahore, Pakistan and its association with various risk and management factors and Rashid et al (2010) who reported the prevalence of babesiosis among Lohi sheep in the Livestock Experiment Station, Qadirabad, District Sahiwal, Pakistan and in environs. Furthermore, the Babesia prevalence from District T. T. Singh, Punjab, Pakistan has also been correlated with reports from other countries such as Egypt (Mazyad et al., 2002), Sri Lanka (Jorgense et al., 1992), Gambia (Mattioli et al., 1997) and Sudan (Salih et al., 2008) ranging from 1-100% Babesia prevalence. However, our findings are somewhat different from Papadopoulos et al. (1996) and Hosein et al. (2007), who reported 52.1% and 50.92% ovine babesiosis, respectively versus our study (9.23%). This difference might be attributed to the application of highly sensitive tests like indirect fluorescent antibody technique for diagnostic purpose and Mazyad and Khalaf (2002), who reported very low prevalence, i.e. only 2.73% among 475 sheep, which may be due to low existence of vector parasites that spread the disease in area under study. In present study, the adult and female sheep were found more susceptible to Babesia infection. The significant difference in Babesia prevalence in different ages and sex group of animals has also been reported previously by Zulfiqar et al. (2012) and Iqbal et al. (2011). The prevalence of Babesia in goat from T. T. Singh, Punjab, Pakistan was also found to be correlated with the weather conditions such as rain fall, temperature and relative humidity. In Pakistan, the weather condition varies whole the year including hot dry to humid rainy season. So, the Babesia prevalence is expected different. In rainy season, the Babesia prevalence was found highest (17.95%, July), while lowest (0 %, Dec-2009 and Jan-2010). In rainy season the higher Babesia infection may attributed with humid climate because minimum prevalence was observed in dry weather condition and result are in accordance with Ahmad et al. (2011) who

have reported a significant variation of Babesia prevalence among seasons and peak infection was observed during summer and autumn. Similar results have also been documented by other workers (Birkenheuer et al., 1999, Kar et. al., 2008 and Pavlidou et al., 2008) and attributed the higher Babesia prevalence with high risk of vectors during peak infection periods.

References

- Ahmad, S. S.; M. S. Khan; M. A. Khan; and N. Ahmad (2011). Prevalence of Babesiosis in cats in Lahore, Pakistan. *The J. Animal Plant Sci.* 21: 354-357
- Bashir, I.N.; Z.I. Chaudhry; S. Ahmed and M.A. Saeed (2009). Epidemiological and vector identification studies on canine babesiosis. *Pakistan Vet. J.* 2009, 29: 51-54.
- Hosein, HI.; S.A. Ahmed; F.A. Ibrahim; T.R. Abou-Elnaga; M.A. Gebely; M.A. Mahmoud (2007). Seroprevalence of Babesia ovis in small ruminants in Siwa Oasis, Egypt. *Bs Vet Med J.* 17: 19-24
- Iqbal, Z.; M.S. Sajid; A. Jabbar; Z.A. Rao and M.N. Khan (2006). Techniques in Parasitology. pp. 52-54. D.G. Administration, Higher Education Commission, Islamabad-Pakistan.
- Iqbal, F.; M. Fatima; S. Shahnawaz; M. Naeem; R. Shaikh; M. Ali; A. Shaikh; M. Aktas; M. Ali (2011). A study on the determination of risk factors associated with babesiosis and prevalence of Babesia sp., by PCR amplification, in small ruminants from Southern Punjab (Pakistan). *Parasite.* 18(3):229-234.
- Jorgensen, W.K.; D. J. Weilgama; M. Navaratne and R. J. Dalgliesh (1992). Prevalence of Babesia ovis and Anaplasma marginale at selected localities in Sri Lanka. *Tropical Animal Health and Production.* 24: 9-14.
- Magona, J.W.; J. Walubengo; W. Oluho-Mukani; N.N. Jonsson; S.C. Welburn; M.C. Eisler (2008). Clinical features associated with seroconversion to Anaplasma marginale, Babesia bigemina and Theileria parva infections in African cattle under natural tick challenge. *Veterinary Parasitology.* 155: 273–280.
- Mattioli, R.C.; L. Janneh; N. Corr; J.A. Faye; V.S. Pandey and A. Verhulst (1997). Seasonal prevalence of ticks and tick transmitted haemoparasites in traditionally managed N'Dama cattle with reference to strategic tick control in the Gambia. *Medical and Veterinary Entomology.* 11: 342-348
- Mazyad, S. A.; and S. A. Khalaf (2002). Studies on Theileria and Babesia infecting live and slaughtered animals in Al Arish and El Hasanah, North Sinai Governorate, Egypt. *J. Egypt. Soc. Parasitol.* 32:601–610.
- Papadopoulos, B.; M. Brossard and N.M. Perie (1996). Piroplasms of domestic animals in the Macedonia region of Greece 3. Piroplasms of small ruminants. *Veterinary Parasitology.* 63: 67-74.
- Rashid, A.; J.A. Khan; M.S. Khan; K. Rasheed; A. Maqbool and J Iqbal (2010). Prevalence and chemotherapy of babesiosis among Lohi sheep in the Livestock Experiment Station, Qadirabad, Pakistan, and environs. *The Journal of Venomous Animals and Toxins including Tropical Diseases.* 16: 587-591.
- Soulsby, E.J.L (2006). Helminths, arthropods and protozoa of domesticated animals, pp. 720. Baillier Tindall, UK.
- Theodoropoulos, G.; M. Gazouli; J.A. Ikonomopoulos; V. Kantzoura and A. Kominakis (2006). Determination of prevalence and risk factors of infection with Babesia in small ruminants from Greece by polymerase chain reaction amplification. *Veterinary Parasitology.* 135: 99–104
- Zulfiqar, S.; S. Shahnawaz; M. Ali, A.M. Bhutta; S. Iqbal; S. Hayat; S. Qadir; M. Latif; N. Kiran; A. Saeed; M. Ali; F. Iqbal (2012). Detection of Babesia ovis in blood samples and its effect on the hematological and serum biochemical profile in large ruminants from Southern Punjab. *Asian Pacific Journal of Tropical Biomedicine.* 104-108

The *in vitro* antibacterial activity of *Muntingia calabura* against *Staphylococcus aureus* and *Streptococcus agalactiae*

Surjowardojo, P*, Sarwiyono, I. Thohari & A. Ridhowi

Faculty of Animal Husbandry, University of Brawijaya Jl. Veteran, Malang 65145, East Java, Indonesia

Abstract

The present study investigated the possible antibacterial activity of methanol, ethanol and aqueous extracts of *Muntingia calabura* using *in vitro* discs diffusion method. The sterilized blank discs (5mm diameter) was impregnated with 10 µL of the respective extract (the concentration: 10%, 20%, 30%, 40%, 50%, control (iodip) and tested against *Staphylococcus aureus* and *Streptococcus agalactiae* were obtained from fresh milk CMT score 3. Statistical analysis showed that extract from methanol, ethanol and aqueous were effective in inhibiting growth of *Staphylococcus aureus* bacteria as control treatment at concentration 30% (6.85 ± 0.35 ; 6.43 ± 0.65 ; 6.62 ± 0.19 vs. 6.34 ± 0.07 mm) respectively. Inhibiting zone for *Streptococcus agalactiae* from methanol and aqueous was effective as control at concentration 30% (7.42 ± 0.62 ; 7.77 ± 0.37) while from ethanol it's effective as control treatment at concentration 30%, 7.77 ± 0.37 mm. Inhibiting zone against *Streptococcus agalactiae* slightly bigger compare to *Staphylococcus aureus*. The increasing concentrations of extract will higher inhibiting zone area. We concluded that *Muntingia calabura* has potency to be used as an antimicrobial activity against the *Staphylococcus aureus* and *Streptococcus agalactiae*.

Keywords: antibacterial activity, *muntingia calabura*, *Staphylococcus aureus*, *Streptococcus agalactiae*.

*Corresponding author: puguh.surjowardojo@yahoo.co.id

Introduction

Staphylococcus aureus and *Streptococcus agalactiae* have been reported as the most common bacterial agent of mastitis (Khan et al., 2003). Accumulation of bacteria waste products intensifies the inflammatory response resulting in destruction of milk producing tissues and reduced milk yield (Myllys & Rautala, 1995).

The use of herbal plants are widely known to be used in the treatment of various infectious diseases throughout the history of mankind. Plant materials continue to provide the major source of natural therapeutic remedies and play an important role in health care in many developing country (Ody, 1993). It is believed that chemical compounds of plants have an important role. They can work as pollinator attractants and as chemical defenses against insects, herbivores and microorganisms (Harbone, 1998). These antimicrobial compounds produced by plants are active against plant and human pathogenic microorganisms (Sarac & Ugur, 2007). There are several reports in the literature regarding the antimicrobial activity of plant crude extracts and the bioassay-guided fractionation of those extracts that yielded active principles (Portillo et al., 2001)

Muntingia calabura have been reported to contain some varieties of chemical substances and biological properties. Therefore, the main objective of this study is investigate the possible antibacterial activity of methanol, ethanol and aqueous extracts of *Muntingia calabura* against *Staphylococcus aureus* and *Streptococcus agalactiae* bacteria using *in vitro* discs diffusion method.

Material and Method

2.1 Collection of plant and Extraction

Fresh plant leaves of *Muntingia calabura* were collected from Sawiran Village, Tutar Sub District, Pasuruan Regency, East Java Province, Indonesia. The leaves were cleaned and dried in an oven at 60°C for 24 hrs. Leaves were crushed to powder using grinding machine. All extraction was carried out in the ratio 1:5 (v/w) for 24 hrs by using maseration method extraction. The residues of extraction from various solvent (methanol, ethanol, aqueous) were further evaporated by using rotary evaporator at 60°C. The concentration of extraction to determine inhibiting zone in each solvent were 10%, 20%, 30%, 40%, 50%.

2.3 Preparation of Microorganism Culture

The bacteria were taken from fresh milk with CMT score 3. *Staphylococcus aureus* and *Streptococcus agalactiae* bacteria were incubated at 37°C for 24 hrs after injection into nutrient broth, MSA and MRSA, sterilized in a flask and cooled to 40-50°C was poured, in the volume of fiftymicroliter, into sterilized petri dishes (diameter of 5 mm) and allowed to harden under room temperature. This is followed by homogenous distribution of 0.1 mL bacteria culture (10^7 bacteria per mL) onto medium of petridishes. The inhibiting zones (mm) were performed from fourfold replication in each treatment, which were calculated as mean \pm standard deviation. All data were analyzed using complete randomize design.

Result and Discussion

The results of this study were expressed as mean \pm standard deviation (Table 1 and 2). Statistical analysis showed that extract from methanol, ethanol were effective ($P < 0.01$) in inhibiting growth of *Staphylococcus aureus* bacteria as control treatment at concentration 30% (6.85 ± 0.35 ; 6.43 ± 0.65 vs. 6.34 ± 0.07 mm) respectively, while extract from aqueous was effective ability as control treatment at concentration 30% (6.62 ± 0.19 vs. 6.34 ± 0.07 mm)

Table 1. The antibacterial activity of methanol, ethanol and aqueous extracts of *Muntingia calabura* against *Staphylococcus aureus* bacteria.

Treatment	<i>Staphylococcus aureus</i>		
	Methanol	Ethanol	Aqueous
P0. Iodip	6.34 ± 0.07^a	6.34 ± 0.07^a	6.34 ± 0.07^c
P1. 10%	6.36 ± 1.08^{ab}	6.10 ± 0.33^a	5.58 ± 0.22^a
P2. 20%	6.57 ± 0.33^{ab}	6.21 ± 0.60^a	5.89 ± 0.26^b
P3. 30%	6.85 ± 0.35^{abc}	6.43 ± 0.65^{ab}	6.62 ± 0.19^c
P4. 40%	7.21 ± 0.69^{bc}	7.02 ± 0.11^{bc}	7.45 ± 0.18^d
P5. 50%	8.18 ± 0.40^c	7.51 ± 0.28^c	8.26 ± 0.16^e

a,b,c,d Superscript letters within the same column indicate significant ($P < 0.05$)

* Value are presented as mean \pm SE, of fourfold experiments

* Diameter of inhibition zone not including diameter of disc 5

Statistical analysis showed that extract from methanol and aqueous were effective ($P < 0.05$) in inhibiting growth of *Streptococcus agalactiae* bacteria as control treatment at concentration 30% (7.63 ± 0.96 ; 7.77 ± 0.37 vs. 7.62 ± 0.25 mm) respectively, while extract from ethanol was effective ability as control treatment at concentration 40% (7.02 ± 0.76 vs. 7.62 ± 0.25 mm). The antimicrobial activity increased significantly with the increasing concentration. Believed that the bioactive constituent of *Muntingia calabura* leaves i.e. saponin, flavonoid and tannin has a role in inhibiting zone area.

Table 2. The antibacterial activity of methanol, ethanol and aqueous extracts of *Muntingia calabura* against *Streptococcus agalactiae*

Treatment	<i>Streptococcus agalactiae</i>		
	Methanol	Ethanol	Aqueous
P0. Iodip	7.62 ± 0.25 ^{bc}	7.62 ± 0.25 ^c	7.62±0.25 ^b
P1. 10%	6.34 ± 1.01 ^{ab}	5.85 ± 0.53 ^a	6.37±0.28 ^a
P2. 20%	6.73 ± 0.90 ^{ab}	5.91 ± 0.42 ^a	7.42±0.50 ^b
P3. 30%	7.42 ± 0.62 ^{bc}	6.61 ± 0.59 ^{ab}	7.77±0.37 ^{bc}
P4. 40%	7.63 ± 0.96 ^{bc}	7.02 ± 0.76 ^{bc}	8.27±0.35 ^{cd}
P5. 50%	8.17 ± 0.30 ^c	8.02 ± 0.46 ^d	8.42±0.34 ^d

^{a,b,c,d}Superscript letters within the same column indicate significant ($P < 0.05$)

* Value are presented as mean ± SE, of fourfold experiments

* Diameter of inhibition zone not including diameter of disc 5

Muntingia calabura extracted by methanol and ethanol had inhibiting zone value less than aqueous. Maybe this is due to, polarity indexed of aqueous solvent. Kaneda et al. (2011) reported that successful determination of biologically active compounds from plant material is largely dependent on the type of solvent used in the extraction procedure. The lower value obtained from *Staphylococcus aureus* bacteria compare with *Streptococcus agalactiae* indicated that *Streptococcus agalactiae* more sensitive to antimicrobial activity of *Muntingia calabura*. This means that the type of bacteria has a role in effectiveness of antimicrobial activity. This opinion is in line with Zakaria et al. (2006) which reported that *Muntingia calabura* and *Corchorus olitorius* has different effectiveness for diameter of inhibitory zone.

Conclusion

The extract of leaves of *Muntingia calabura* provides evidence that *Muntingia calabura* contains has important bioactive compounds and their potential as a source of the use of plant species as antimicrobial activity in mastitis disease.

References

- Harbone, B., 1998. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. Chapman and Hall Co. New York.
- Kaneda N., Pezzuto, J. M., Soejarto, D.D., Kinghorn, A.D., Farnworth, N.R., Santisuk, T., Tuchinda, P., Udachon, J & Reutrakul, V, 1991. Plant anticancer agents, XLVII. new cytotoxic flavanoids from *Muntingia calabura* roots. J. Nat. Prod. 54: 196-206.
- Khan, I.U., A.A. Hassan, A. Abdulmawjood. C. Lanimler, W. Wolter, and M. Zschock. (2003); Identification and epidemiological characterization of *Streptococcus uberis* isolated from bovine mastitis using conventional methods. J. Vet. Sci. 4:213-223.
- Myllys, V. and Rautala, H, 1995. Characterization of clinical mastitis in primiparous heifers. J. dairy Sci. 78: 538-545.
- Ody, P., 1993. The complete medical herbal. New York, Dorling Kindersley Limited, pp: 132-171.
- Portillo, A. R., R. Vila, B. Freixa, T. Adzet, and S. Cañigueral, 2001. Antifungal Activity of Paraguayan Plants Used in Traditional Medicine. J. Ethnopharmacol. 76: 93-98.
- Sarac, N. and A. Ugur, 2007. Antimicrobial activities and usage in Folkloric Medicine of some lamiaceae species growing in Mugla, Turkey. Eur. Asian J. Bio. Sci. 1(4): 28-34.
- Zakaria, Z. A., H. Zaiton, E.F.P. Henie, A.M. Mat Jais, D. Kasthuri, M. Thenamutha, F. W. Othman, R. Nazaratulmawarina and C. A. Fatimah. 2006, Journal of Pharmacol.Toxiol. 8: 480-486.

Study of the optimal condition of RNA in situ hybridization for *HER-2/neu* in canine mammary gland

Pavasutthipaisit^{1,2}, S., T. Mamom^{2,3} & P. Sotthibandhu^{*2,3}

¹Student of Master of Science in Animal Biotechnology, ²Department of Preclinical Veterinary Science, ³Histopathology section of MVDC, Faculty of Veterinary Medicine, Mahanakorn University of Technology, 140 Cheum-Sampan Rd., Bangkok 10530 Thailand

Abstract

The mammary gland tumors are the second most common occurring canine neoplasm. The investigation of *HER-2/neu* gene expression by chromogenic in situ hybridization was more valuable prognostic marker than protein expression by immunohistochemistry. Study of canine mammary gland tumor with chromogenic in situ hybridization (CISH) is rarely reported. The purpose of this study was to investigate the optimal condition of *HER-2/neu* gene expression by RNA CISH in canine mammary tumors. The canine mammary tissue samples were diagnosed with H&E stain then CISH for *HER-2/neu* was performed with RNA probe. The hybridization conditions were divided into 2 durations, 12 and 16 hours, each duration was divided into 6 groups with different concentration of antisense-sense probe and hybridization temperature as follows: 1:100-45°C, 1:100-55°C, 1:200-45°C, 1:200-50°C, 1:200-55°C and 1:200-60°C. The *HER-2/neu* mRNA signal visualization scored as follows: 0 = no signal; 1 = weak signal; 2 = moderate signal; 3 = strong signal. The best condition was 1:200-60°C at 16 hybridized hours. The signals of benign tumor scored 1, tubulopapillary mammary carcinoma scored 2 and anaplastic carcinoma scored 3. The present study revealed the optimal condition of chromogenic in situ hybridization for detection of *HER-2/neu* mRNA by adjust 1) Duration of hybridization 2) Temperature of hybridization and 3) probe concentration. The *HER-2/neu* RNA CISH may play an important role as prognostic biomarker for the risk of developing metastasis in canine mammary tumor.

Keywords: HER-2/neu, CISH, canine mammary gland tumor

**Corresponding author: email address: pongsiwa@mut.ac.th*

Introduction

The proto-oncogene *HER-2/neu*, one of a four member family of the human epidermal growth factor receptor also known as *erbB-2* encoding is a transmembrane tyrosine kinase growth factor receptor. In canine, *HER-2/neu* gene has been mapped to chromosome 1q13.1 which cytogenetic studies revealed that this regions frequently clonal chromosome aberrations in canine mammary tumor (Murua Escobar et al., 2001). Several studies showed a good beneficial immunohistochemistry result of *HER-2/neu* overexpression in canine mammary carcinoma (Antuofermo et al., 2007). In earlier reports indicated that *HER-2/neu* plays an important role in canine mammary neoplasia and discussed as a valuable prognostic biomarker to be a suitable natural model of human breast cancers (Hsu et al., 2009). Many others publish reports indicated that the chromogenic in situ hybridization (CISH) has a good correlation with fluorescent in situ hybridization (FISH) which was the gold standard methodology of *HER-2/neu* gene evaluation and given more informative diagnostic evaluation greater than immunohistochemistry in breast

cancers (Kostopoulou et al., 2007; Todorović-Raković et al., 2007) whereas *HER-2/neu* gene expression by CISH was rarely reported in canine mammary neoplasm. The goal of present study was to determine the optimal condition for detection of *HER-2/neu* mRNA on canine mammary gland tumor by CISH.

Materials and Methods

Tumor specimens and histological examination: Three samples of mammary tumors were surgically removed from female dogs, then fixed in 4% paraformaldehyde in phosphate buffer (pH 7.2) and processed for paraffin sections in RNase free-condition. Four micron-thickness was stained with hematoxylin and eosin (H&E) for diagnosis according to the WHO Histological classification of Mammary Tumors of the Dogs and Cats (Misdrop et al., 1999). Serial sections on Superforst/Plus microscope slide was prepared for CISH.

Preparation of cRNA probes: The cRNA probes of canine erbB-2 DNA were designed according to GenBank accession no. AB008451, size 404 bp as follows:

```
GAGAGTATGTGAAGGACAGGTAAGTGTCTACCGTGCCACTCAGAGTGTGTCAGCCCCAG
AATGGCTCAGTGACCTGTTTCGGATCGGAGGCTGACCAGTGTGTGGCCTGCGCCCAC
TACAAGGACCCTCCCTTCTGTGTGGCTCGCTGCCCCAGTGGTGTGAAACCTGACCTG
TCCTTCATGCCATCTGGAAGTTCGCAGATGAGGAGGGCACTTGCCAGCCGTGCCCC
ATCAACTGCACCCACTCCTGTGCGGACCTGGACGAGAAGGGCTGTCCC GCCGAGCA
GAGAGCCAGCCCTGTGACATCCATCATTGCCGCTGTGGTGGGCATTCTGCTGGCTGT
GGTCGTGGGGCTGGTCCTCGGCATCCTGATCAAGCGAAGGCGGCAGAAGATCCGGA
AGTACACT (Martin de las Mulas, et al. 2003)
```

This was synthesized by GenScript USA with pCRTM4-TOPO[®] TA vector. The plasmid DNA was digested in 37 °C for an hour by using SpeI (Promega[®]) for linear DNA of anti-sense cRNA probe and NotI (Promega[®]) for linear DNA of sense cRNA probe. After the digested plasmid DNA was purified by ethanol precipitation, the cRNA probe was prepared by DIG RNA Labeling Kit (Roche[®]) and precipitated in 7.5 M lithium chloride solution.

CISH: The CISH was performed according to Sothibandhu (2009). The hybridization conditions were divided into 2 durations, 12 and 16 hours, each duration was divided into 6 groups with different concentration of antisense-sense probe and hybridization temperature as follows: 1:100-45°C, 1:100-55°C, 1:200-45°C, 1:200-50°C, 1:200-55 °C and 1:200-60 °C. The sections were deparaffinized, dehydrated in RNase free-condition. Antigen retrieval was carried out by autoclave treatment in citrate buffer (pH 6), 121°C, 5 min. Then blocking endogenous alkaline phosphatase was done with 0.2 N HCl, 10 min and post-fixation with 4% paraformaldehyde in phosphate buffered saline at room temperature, 5 min. Denaturation was carried out at 95 °C, 5 min and hybridized. After stringency washes, the sections were blocking for a nonspecific background staining with blocking reagent (DIG nucleic acid detection kit, Roche[®]) at room temperature, 5 min then applied with detection buffer (DIG nucleic acid detection kit, Roche[®]) at room temperature, 10 min and performed a signal detection with detection solution 1x NBT/BCIP (NBT/BCIP stock solution, Roche[®]). The *HER-2/neu* mRNA signal was visualized and captured by AxioCam[®] ERc5S.

Results

Observation on H&E stained slide revealed that the mammary tumors were classified as benign mixed mammary gland, tubulopapillary carcinoma and anaplastic carcinoma.

CISH: The best condition was 1:200-60 °C at 12 hybridized hours. The signal of antisense was found in cytoplasm of glandular epithelium at condition (Fig.1 and 2). The signal of benign tumor scored 1, tubulopapillary carcinoma scored 2 and anaplastic carcinoma scored 3. Other conditions showed no signal or very weak signal. Sense probe showed no signal (Fig3.). The results showing that score of other condition was 0 or 1 (Table 1).

Table 1. Chromogenic in situ hybridization results.

Mammary tumor	12 hybridized hours						16 hybridized hours					
	1:100 45°C	1:100 55°C	1:200 45°C	1:200 50°C	1:200 55°C	1:200 60°C	1:100 45°C	1:100 55°C	1:200 45°C	1:200 50°C	1:200 55°C	1:200 60°C
Benign tumor	0	0	0	0	0	1	0	0	0	0	0	0
Tubulopapillary carcinoma	0	0	0	0	0	2	0	0	0	0	0	0
Anaplastic carcinoma	0	0	0	0	0	3	0	0	0	0	0	0

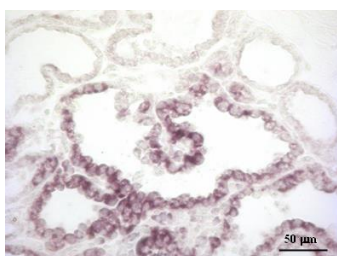


Figure 1. Mammary anaplastic carcinoma 1:200-60 °C. Antisense

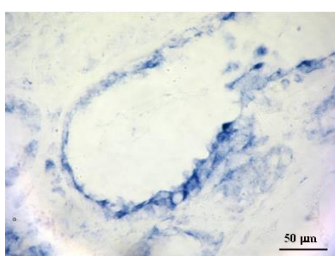


Figure 2. Benign mixed mammary tumor 1:200-60 °C Antisense (rinse with 90-95% ethanol).



Figure 3. Benign mixed mammary tumor 1:200-60 °C. Sense

Discussion and Conclusion

In previous reports of canine mammary tumor could not observed gene amplification by chromogenic in situ hybridization, however our finding of marked *HER-2/neu* mRNA in anaplastic carcinoma seem to be similar to several reports which suggested that marked *HER-2* overexpression were impact in malignancy especially in invasive carcinoma (Dutra et al., 2004) or high-grade ductal carcinoma (Antuofermo et al., 2007). The number of *HER-2/neu* mRNA and protein overexpression may associated with the tumor aggressiveness. This present study revealed the optimal condition of RNA in situ hybridization for *HER-2/neu* in canine mammary tumor. We suggest that the preservative agent, the duration and temperature of hybridization and the concentration of RNA probe are the important points. Small number of target gene might need longer duration and higher temperature of hybridization. According to CISH in human breast cancer was more accurate detection of *HER-2/neu* status than immunohistochemistry method. Hence, these findings of revealed condition will be further processed for study of prognostic biomarker for the risk of developing metastasis in canine mammary tumor.

Acknowledgement

The present study was funded by Mahanakorn University of Technology.

References

- Antuofermo, E., M. A. Miller, S. Pirino, J. Xie, S. Badve and S. I. Mohammed, 2007. Spontaneous Mammary Intraepithelial Lesions in Dogs – A model of Breast Cancer. *Cancer Epidermiol. Biomarkers Prev.* 16: 2247-2256.
- Dutra, A.P., N.V.M. Granja, F.C. Schmitt and G.D. Cassali. 2004. c-erbB-2 expression and nuclear pleomorphism in canine mammary tumors. *Braz. J. Med. Biol. Res.* 37: 1673-1681.
- Hsu, W. -L., H. -M. Huang, J. -W. Liao, M. -L. Wong and S. -C. Chang, 2009. Increased survival in dogs with malignant mammary tumours overexpressing *HER-2* protein and detection of a silent single nucleotide polymorphism in the canine *HER-2* gene. *Vet. J.* 180: 116-123.
- Kostopoulou, E., D. Vageli, D. Kaisaridou, M. Nakou, M. Netsika, N. Vladica, A. Daponate, G. Koukoulis, 2007. Comparative evaluation of non-informative *HER-2* immunoreactions (2^+) in breast carcinomas with FISH, CISH and QRT-PCR. *The Breast.* 16: 615-624.
- Martin de las Mulas, J., J. Ordás, Y. Millán, V. Fernández-Soria, S. Ramón y Cajal, 2003. Oncogene *HER-2* in canine mammary gland carcinomas. *J. Breast Canc. Res.Treat.* 80: 363-367.
- Misdrop W, R. W. Else, E. Hellmé, T. P. Lipscomb, 1999. Histological classification of mammary tumors of the dog and the cat, 2nd series, Vol. VII, Armed Forces Institute of Pathology, American Registry of Pathology, Washington, DC and the World Health Organization Collaborating Center for Worldwide Reference on Comparative Oncology, pp59.
- Murua Escobar, H., K. Becker and J. Bullerdiek, 2001. The canine ERBB2 gene maps to a chromosome region frequently affected by aberrations in tumors of the dg (*Cans familiaris*). *Cytogenet. Cell Genet.* 94: 194-195.
- Sotthibandhu, P. 2009. Morphology studies of mammalian taste chemosensory cells. PhD degree Thesis. Laboratory of Veterinary Anatomy, Kitasato University.
- Todorović-Raković, N., D. Jovanović, Z. Nešković-Konstantinović and D. Nikolić-Vukosavljević. 2007. *Exp. Mol. Pathol.* 82: 262-268.

Process development of cooking wine from whey of buffalo milk

Mamuad*¹, L. L. & F. V. Mamuad²

¹*Ruminant Nutrition and Anaerobe Laboratory, Department of Animal Science and Technology, College of Bio-industry Science, Sunchon National University, Suncheon, Republic of Korea 540-742*

²*Philippine Carabao Center, Science City of Munoz, Nueva Ecija Philippines 3120*

Abstract

Development of process in the production of cooking wine from whey of buffalo milk was undertaken. Whey from buffalo milk was treated with different inocula, sugar concentrations and fermented in different types of container. Data gathered on the physico-chemical analysis and sensory qualities of wine were tabulated and statistically analyzed using the split-split plot design in Completely Randomized Design. The relationships of the physico-chemical data to sensory evaluation scores were also studied. Results of the study revealed that *Kluyveromyces marxianus* is a better source of inoculum for wine fermentation of whey because it has high percent Total Titratable Acidity (TTA), alcohol taste, good clarity, sourness, sweetness, and general acceptability but low on off-odor, off-flavor, aroma, after-taste and saltiness. Wine developed from 24°Brix has high Total Soluble Solids (TSS), percent alcohol, alcohol taste and clarity than 20°Brix. On the other hand, 20°Brix has also higher percent TTA, sourness, sweetness and general acceptability than 24°Brix. Both have low values on off-odor, off-flavor, aroma and saltiness. Thus, 20 and 24°Brix are the best sugar concentrations that could be used for whey fermentation. On the other hand, the use of earthen jar converted and produced more alcohol than the other containers use. It also has the highest percent TTA, TSS, clarity, aroma, alcohol taste, sourness, and general acceptability. Furthermore, it has low sensory scores on saltiness, off-odor and off-flavor. Earthen jar is the best type of container that could be used for the fermentation of whey for wine production.

Keywords: buffalo milk, cCooking wine, process development

**Corresponding author: loveliamamuad2306@gmail.com*

Introduction

Milk is a white liquid secreted by the mammary glands of female carabaos, cows, goats, sheep and other mammals. It is regarded as a highly nutritious substance because of its constituents such as protein, fat, sugar, minerals, vitamins, and other minor components, which are known to be essential for human nutrition. It is also widely consumed both in its natural form and in dairy products.

The Philippine Carabao Center (PCC) produces several milk products to preserve milk in other forms that are acceptable to consumers. Two of these milk products are the white soft cheese and mozzarella cheese. However, in cheese making process, about 50-60 percent of it is the whey in buffalo milk, which is commonly removed and discarded by cheese makers.

Whey is a by-product of cheese making which contains valuable food components such as protein, vitamins, minerals and lactose. It is formed when the curds separate from the milk or cream. After the cheese curds are formed, the remaining liquid is called whey. Whey proteins

mainly consist of α -lactalbumin and β -lactoglobulin. Depending on the method of manufacture whey may also contains glycomacropeptides (GMP) (Yares, 2007). It also contains lactic acid, citric acid and non-protein nitrogen (FAO, 1977). To be able to utilize this whey, the process of producing wine out of whey from cheese manufacturing process was studied.

Materials and Methods

Whey from curd making was collected from the National Gene Pool Dairy Processing Plant, Philippine Carabao Center, Science City of Muñoz, Nueva Ecija using sanitized containers. Then, the collected whey was brought to the laboratory.

Productions of wine from whey from buffalo milk as influenced by different factors like the inoculum (*Kluyveromyces marxianus* (E.C. Hansen) Van der Walt and *Saccharomyces cerevisiae* Ludwigii), sugar concentrations (11, 16, 20, and 24 °Brix, respectively) and types of container (earthen jar, transparent glass jar and amber glass jar) were undertaken. Determination and comparison of the physico-chemical properties of cooking wine as affected by different factors in terms of pH, total soluble solid, total titratable acidity and alcohol content were undertaken. Likewise, evaluation and comparison of the sensory qualities of wine produced from whey was done. Also, determination of the relationship between and among the physico-chemical analysis and the sensory evaluation scores of cooking wine and computation for the economic cost of producing wine was done. Results of physico-chemical analysis and sensory evaluation of wine gathered were tabulated and statistically analyzed using split-split plot design using CRD. Moreover, correlation of the physico-chemical analysis with the sensory evaluation scores was also analyzed.

Results and Discussion

Based on the results gathered, different nutritional and physical factors such as inoculum (*Kluyveromyces marxianus* and *Saccharomyces cerevisiae*), sugar concentrations (11, 16, 20 and 24 Brix, respectively) and types of container (amber glass jar, transparent glass jar and earthen jar) affect the production of wine (Table 1).

Table 1. Mean total soluble solids of wine as affected by inoculum, sugar concentrations and types of container.

INOCULUM	SUGAR CONCENTRATION	TYPE OF CONTAINER			MEAN
		AMBER	TRANSPARENT	EARTHEN	
<i>Kluyveromyces marxianus</i>	11 °Brix	3.48	3.34	-	3.41
	16°Brix	6.17	5.54	5.00	5.57
	20 °Brix	8.50	6.34	7.63	7.49
	24 °Brix	8.58	8.40	10.90	9.29
	Mean	6.68	5.91	7.84	6.72 ^a
<i>Saccharomyces cerevisiae</i>	11 °Brix	0.00	0.00	-	0.00
	16°Brix	5.80	5.32	6.00	5.71
	20 °Brix	8.44	8.58	9.25	8.76
	24 °Brix	11.44	11.70	11.90	11.68
	Mean	6.42	6.40	9.05	7.13 ^a

Means followed by the same letter superscript in a column (a, b, c, d) or in a row (x, y, z) are not significantly different at 5% level by DMRT

Kluyveromycesmarxianus is a better source of inoculum for wine fermentation of whey because it has high percent TTA, alcohol taste, good clarity, sourness, sweetness, and general acceptability but low on off-odor, off-flavor, aroma, after-taste and saltiness. Wine developed from 24°Brix has high TSS, percent alcohol, alcohol taste and clarity than 20 °Brix. On the other hand, 20 °Brix has also higher percent TTA, sourness, sweetness and general accept ability than

24°Brix. Both have low values on off-odor, off-flavor, aroma and saltiness. Thus, 20 and 24°Brix are the best sugar concentrations that can be used for whey fermentation. On the other hand, the use of earthen jar converted and produced more alcohol than the other containers used. It also has the highest percent TTA, TSS, clarity, aroma, alcohol taste, sourness, and general acceptability. Furthermore, it has low sensory scores on saltiness, off-odor and off-flavor. Therefore, it is concluded that earthen jar is the best type of container that can be used for the fermentation of whey for wine production.

Table 2. Mean percent alcohol of wine as affected by inoculum, sugar concentrations and types of container.

INOCULUM	SUGAR CONCENTRATION	TYPE OF CONTAINER			MEAN
		AMBER	TRANSPARENT	EARTHEN	
<i>Kluyveromyces marxianus</i>	11 °Brix	2.42	2.12	-	2.27
	16°Brix	3.67	2.96	2.90	3.18
	20 °Brix	5.36	2.70	1.93	3.33
	24 °Brix	5.69	4.38	6.19	5.42
	Mean	4.28	3.04	3.67	3.66 ^a
<i>Saccharomyces cerevisiae</i>	11 °Brix	0.24	0.00	-	0.12
	16°Brix	2.69	2.92	2.14	2.58
	20 °Brix	4.50	3.95	4.60	4.35
	24 °Brix	8.08	6.79	5.55	6.80
	Mean	3.88	3.41	4.09	3.77 ^a

Means followed by the same letter superscript in a column (a, b, c, d) or in a row (x, y, z) are not significantly different at 5% level by DMRT

Table 3. Mean general acceptability of wine as affected by inoculum, sugar concentrations and types of container.

INOCULUM	SUGAR CONCENTRATION	TYPE OF CONTAINER			MEAN
		AMBER	TRANSPARENT	EARTHEN	
<i>Kluyveromyces marxianus</i>	11 °Brix	1.50	1.40	-	1.45
	16°Brix	1.90	3.30	1.80	2.33
	20 °Brix	4.20	3.20	4.80	4.07
	24 °Brix	2.60	3.60	3.50	3.23
	Mean	2.55	2.88	3.37	2.89 ^a
<i>Saccharomyces cerevisiae</i>	11 °Brix	1.30	1.00	-	1.15
	16°Brix	2.70	2.40	1.40	2.17
	20 °Brix	2.80	4.00	3.00	3.27
	24 °Brix	3.30	3.60	2.60	3.17
	Mean	2.53	2.75	2.33	2.55 ^b

Means followed by the same letter superscript in a column (a, b, c, d) or in a row (x, y, z) are not significantly different at 5% level by DMRT

References

- Amerine, M. A. and E. B. Roessler. 1976. Wine. Their Sensory Evaluation. USA: W. H. Freeman and Company.
- de Carvalho Lins, A. and M. Rocha Leão. 2002. Removal of skim milk lactose by fermentation using free and immobilized *Kluyveromyces marxianus* cells. World J. Microbiol. Biotechnol.18:187-192.
- Mamuad, F. L.2001. Physico chemical characteristics of soft cheese produced from buffalo milk and abomasal extract of suckling buffalo and its whey. Undergraduate thesis. College of Arts and Sciences. Department of Chemistry. Central Luzon State University.
- Pessoa Jr, A. and M. Vitolo. 1999. Inulinase from *Kluyveromyces marxianus*: culture medium composition and enzyme extraction. Braz. J. Chem. Eng. 16:237-245.

Study of differential protein composition of raw milk and processed milk by using SDS-PAGE and Native-PAGE

Janmanee¹, O., W. Junlapho^{*1,2}, S. Naktong¹ & J. Kanjanapruthipong¹

¹Department of Animal Sciences, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, 73140, Thailand, ²Center of Excellence on Agricultural Biotechnology: (AG-BIO/PERDO-CHE), Bangkok, 10900, Thailand

Abstract

This study was investigation the impact of the heat treatment of processed milk conducted at different processing temperatures, and the adding of demineralized whey on the protein solubility, soluble protein composition and interactions involved between proteins in a chemical complex. Commercial powder milk has been reconstituted and the soluble protein composition has been determined by the polyacrylamide gel electrophoresis with SDS-PAGE and Native-PAGE. Gel scans from SDS-PAGE and Native-PAGE patterns of powder milk samples were not showed low molecular weight complex as β -lactoglobulin (19 kDa) and α -lactalbumin (13.6 kDa) while compared to raw milk. Protein bands of high molecular weight complex as immunoglobulins (75 kDa), lactoferrin (69 kDa) and serum albumin (60.6 kDa) in powder milk were absent except casein (~33 kDa) was slightly present in SDS-PAGE. Due to the different changes occurred heat and time during processing treatments. The disulfide interactions between denatured molecules of these proteins are mostly responsible for the formation.

Keywords: cow milk, heat treatment, whey proteins, processed milk, native, sodium dodecyl sulfate polyacrylamide gel electrophoresis

*Corresponding author: fagrwn@ku.ac.th

Introduction

Heat treatments are usually used to obtain bacteriological safe milk and to ensure a long shelf life. However, as a lot of literature pointed out, intensive thermal treatment would compromise milk nutrition and flavor (Lacroix et al., 2006), thus consumers prefer pasteurized milk to reconstituted milk and were concerned at possible economic fraud by labeling reconstituted milk as pasteurized milk. A few analysis techniques such as HPLC (Resmini et al., 1992), ELSD (Guyomarch et al., 2000) have been applied in differentially pasteurized milk and reconstituted milk to quantified individual protein or sugar ingredient such as furosine, lactoglobuline or HMF (hydroxymethylfurfural), which demands complicated pre-processing of milk sample. Proteomics have the potential to facilitate further advances in our knowledge of milk proteins. (Ramona et al., 2010), we presented the application of SDS-PAGE in an overall analysis of protein profile change related to milk thermal procession, revealing a significant alteration of protein component between raw milk, processed milk and reconstituted milk.

Materials and Methods

Sample preparation and protein content

Raw bovine milk and processed milk samples from different types [two pasteurized milks, UHT milks and sterilized milk] were purchased from a local market and dry milk powder was reconstituted by diluted 0.15 g of powder milk in 1 ml water. 100 μ l of each milks sample was mixed with 100 μ l of chloroform, vortex, and centrifuge for 5 minute at 4,500 rpm and uses a large aqueous layer on top to measure protein content by Bradford (1976).

SDS-PAGE and Native-PAGE

The SDS-PAGE and Native-PAGE were run under reducing and non-reducing condition, respectively by using a gel electrophoresis apparatus with a 4% acrylamide stacking gel and a 15% separating gel. Marker protein ladder with 14.4 to 97 kDa proteins was used as a molecular weight standard. Current was applied at a constant voltage of 70 V until samples entered the separating gel, and voltage was increased to 120 V. Gels were stained with 0.1% coomassie brilliant blue R-250 in 10% acetic acid and 40% methanol, and destained overnight in 10% acetic acid and 10% methanol. Images were captured on scanner in transmission mode.

Results and Discussion

All milks samples were run in SDS-PAGE with 250 mg/ml of protein concentration and result of proteins profile has shown in Figure 1. Raw milk and pasteurized milk (lane 2 and 3) were showed proteins profile containing with eight bands, those bands were identified as immunoglobulins (~75 kDa), lactoferrin (~69 kDa), serum albumin (~60.6 kDa), three subunits of casein (~38, 33.4 and 30 kDa), β -lactoglobulin (~19 kDa) and α -lactalbumin (~13.6 kDa), respectively.

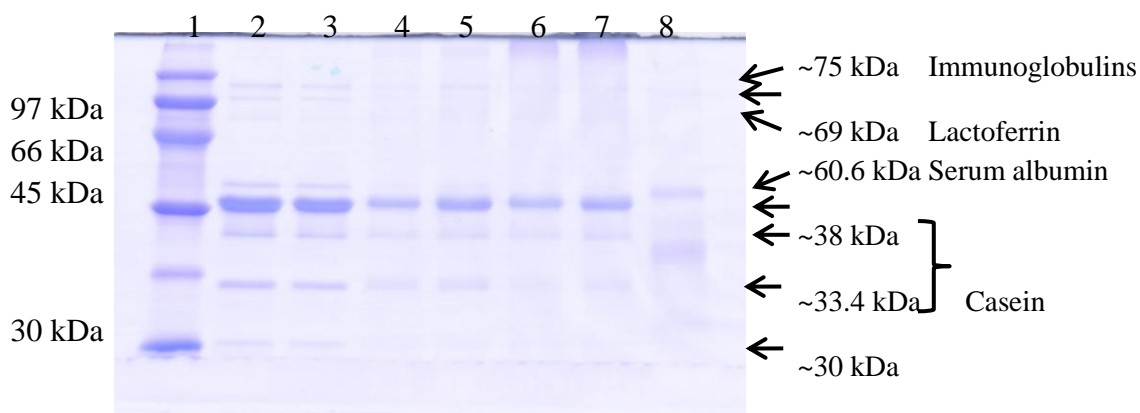


Figure 1. SDS-PAGE profiles of milk samples: Lane 1) Protein marker, 2) Raw milk, 3) Pasteurized milk, 4) UHT milk, 5) Low fat UHT milk, 6) Sterilized milk, 7) Low fat sterilized milk and 8) Powder milk.

In processed milk samples, the proteins bands of immunoglobulins, lactoferrin, serum albumin, casein (~38 and 30 kDa), β -lactoglobulin and α -lactalbumin in pasteurized, UHT, sterilized milk and powder milk samples were decreased while compared to raw milk except casein (~33.4 kDa) was showed clear bands in pasteurized, UHT and sterilized milk but these protein band was present slightly band in powder milk sample in lane 8. The result from Native-

PAGE in Figure 2 has shown the high molecular weight protein complex of powder milk as immunoglobulins, lactoferrin and serum albumin were decreased bands while compared to raw milk and the low molecular weight protein as β - and α -lactalbumin were absent. However, protein band of casein in powder milk has shown clearly band.

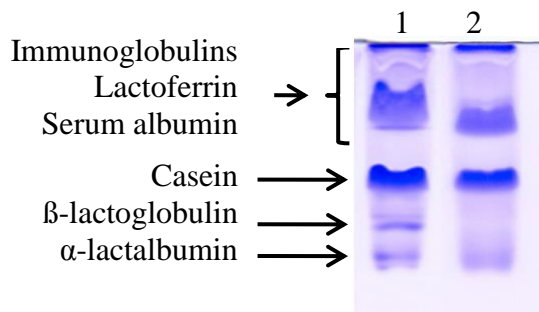


Figure 2. Native-PAGE: Lane 1 = Raw milk, Lane 2 = Powder milk.

The proteins profile of processed milk differentially affected by the privatization process method used to sterilize heat treatment and extend the shelf life of the milk product by using different heat treatment levels (Morales et al., 2000). The heat treatment height and long duration it will effect to denaturation of protein increased. The results corresponding with the reports of Lin et al. (2010) is study about denaturation of whey protein in bovine milk product by using different heat treatment levels by using Native-PAGE techniques.

References

- Bradford, M.M. (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.* 72: 248-254.
- Guyomarch, F., F. Warin, D. Muir and J. Leave, 2000. Lactosylation of milk proteins during the manufacture and storage of skim milk powders, *International Dairy Journal.* 10: 863-872.
- Lacroix, M., J. Leonil, C. Bos, G. Henry, G. Airinei, J. Fau- quant, D. Tome and C. Gaudichon, 2006. Heat markers and quality indexes of industrially heat-treated [15N] milk protein measured in rats, *J. Agric. Food Chem.* 54: 1508-1517.
- Lin,S., J. Son, D. Cao, J. Cao and W. Jiang, 2010. Distinction of different heat-treated bovine milks by native-PAGE fingerprinting of their whey proteins, *Food Chemistry.* 121: 803-808.
- Morales, F. J., C. Romero and S. Jimenez-Perez, 2000. Characterization of industrial processed milk by analysis of heat-induced changes. *International Journal of Food Science and Technology.* 35:193-200.
- Ramona S., C.I. Bartalici, R. Petrut, A. C. Semeniuc and M. A. Rotar, 2010. Two-Dimensional gel electrophoresis of peptides as lap in bovine milk. *Journal of Agroalimentary Processes and Technologies.* 16 (2): 169-176.
- Resmini, P., L. Pellegrino, F. Masott, A. Tirelli and F. Prati, 1992. Detection of reconstituted milk powder in raw and in pasteurized milk by HPLC of furosine, *Scienze e Tecnica Lattiero Casearia.* 3(3): 169-186.

Development and production of Shrikhand by utilization of pomegranate fruit

Gaikwad, S.M.^{1*}, Mundhe S.J.², Chate B.N.³, Landge B.D.¹, & Jangewad N.M.¹

^{1*}Corresponding author, Department of Dairy Science, Sanjeevani College, Chapoli, Tq. Chakur, Dist. Latur (M.S.) INDIA gaikwadsunil39@yahoo.com, ²Department of Microbiology, Sanjeevani College, Chapoli, Tq. Chakur, Dist. Latur (M.S.) INDIA ³Department of Chemistry, Sanjeevani College, Chapoli, Tq. Chakur, Dist. Latur (M.S.) INDIA

Abstract

Shrikhand is an Indian sweet dish made of strained dahi. It is one of the main desserts in Kerala cuisine, Gujarati cuisine and Maharashtrian cuisine. Several attempts have been made to incorporate different additives into shrikhand to address the growing interest in the diversification of food products to attract a wider range of consumers. However, nobody tried to incorporate pomegranate in shrikhand. Therefore, present investigation was carried out to study the acceptability of pomegranate shrikhand. It can be concluded that the proportion of pomegranate pulp may be taken as 20 gm pulp and 80 gm of chakka for the preparation of pomegranate shrikhand which was superior over all combination for its organoleptic quality. It is also recommended that pomegranate can be effectively used for the production of this newly invented product.

Keywords: Shrikhand, pomegranate, fermented milk product, traditional dairy product

Introduction

Shrikhand is an Indian sweet dish made of strained dahi. It is one of the main desserts in Kerala cuisine, Gujarati cuisine and Maharashtrian cuisine. To prepare shrikhand, dahi is tied in a cotton cloth and left under pressure to drain. The strained dahi, referred to as "Chakka", and sugar are mixed thoroughly in a deep bowl. Cardamom, saffron, and any other flavors are then added and mixed. It is then left in the refrigerator for the sugar to dissolve. The dish is served chilled.

Shrikhand is an extremely popular Indian dessert where plain yogurt is transformed into a melt-in-the-mouth delicacy. Essentially a combination of yogurt with fruits and sugar, this dish has a very sweet taste. In North India, it's commonly served for breakfast along with hot pooris, a fried type of bread. In South India, this dish is served as a dessert after the end of the meal. Made from fermented yogurt, shrikand has a creamy, semisoft consistency and is slightly yellowish in color.

One of India's ancient traditional desserts found widely throughout the country. There are many local variations found in different states. For instance, amrakhand is shrikhand containing mango pulp and is found in some parts of Maharashtra.

Pomegranate fruit is one of the most popular, nutritionally rich fruit with unique flavor, taste, and health promoting characteristics. The fruit is moderate in calories; 100 g provides 83 calories, slightly more than that in the apples. It contains no cholesterol or saturated fats. It is a good source of soluble and insoluble dietary fibers, providing about 4 g per 100 g (about 12% of RDA), which aid in smooth digestion and bowel movements.

Several attempts have been made to incorporate different additives into shrikhand to address the growing interest in the diversification of food products to attract a wider range of consumers. The pulp of fruits such as apple, mango, papaya, banana, guava and sapota (Bardale

et al., 1986; Dadarwal et al., 2005), cocoa powder with and without papaya pulp (Vagdalkar et al., 2002) and incorporation of probiotic organisms (Geetha et al., 2003) have been tried in shrikhand. However, nobody tried to incorporate such important fruit i.e. Pomegranate in shrikhand. Therefore, present investigation was carried out to achieve the following objectives.

Objectives

The present investigation was carried out to study the effect of different level of pomegranate juice to judge the acceptability of pomegranate shrikhand. The present investigation was also carried out to study the standardization procedure to prepare pomegranate shrikhand.

Materials and Methods

This study was conducted in the Department of Dairy science, Sanjeevane College, Chapoli. The present investigation was carried out to standardize the pomegranate shrikhand. Hence details regarding the materials used and the method adopted for the present investigation are given here.

Considering the initial investigation the effect of three different levels of pomegranate juice were studied on the sensory quality of this product. The levels of these variables that resulted in most of liked product on the basis of sensory evaluation were selected. Good quality food grade cane sugar was purchased from the local market. This sugar was used at 40% of chakka. Chakka was prepared by using dahi.

Pomegranate pulp

Arils of pomegranate fruits were manually separated from the peels and piths and their juice was extracted using an electric juicer. Fresh juices thus extracted from the fruits were pooled separately, and stored in a refrigerator at 4 °C for subsequent product preparation.

Pomegranate was obtained from local market, peeled properly, ground in a mixer to make paste. The paste kept overnight at refrigerator temperature. The pulp was thereafter incorporated at 0, 5, 10 and 15% replacing chakka in the formulation.

Process combination

For the present investigation following parameters were used to standardize the product. Following parameters were used for the both milk i.e. cow and buffalo milk.

Table 1. Process combinations.

Sr. No.	Process combination	Level of pulp (%)
1	T0	0
2	T1	5
3	T2	10
4	T3	15
5	T4	20

Details of Manufacture

Fresh milk was procured from the local market of Chapoli with 3.5% fat and 8.5% Solid not fat (SNF). Dahi was procured from local market and used as culture. Milk was boiled and then cooled down at 28 to 30°C and inoculated with dahi at the rate of 1.5% and incubated at 30 to

32°C for 10 to 12 h until a firm coagulum was formed. Coagulum was then crushed and transferred to a double muslin cloth and hung for expulsion of whey for 8 to 10 h in refrigerated conditions ($4 \pm 1^\circ\text{C}$). The semi solid chakka obtained after drainage of whey was used as the base for shrikhand. The level of sugar was adjusted at 40%. The sugar was powdered and kneaded uniformly with the chakka. Shrikhand was prepared by supplementing different levels of pomegranate pulp viz. 5, 10, 15 and 20% to chakka. Shrikhand prepared without pomegranate pulp served as control and was compared with the treatments. The sensory evaluation of the product was carried for attributes, namely colour and appearance, flavour, body and texture, sweetness and the overall acceptability of fresh shrikhand.

The scores for qualitative data such as colour and appearance, flavour, body and texture, sweetness, and the overall acceptability given by different judges were tabulated. The data thus obtained was analyzed as per one way ANOVA by Snedecor and Cochran (1994).

Sensory Evaluation

Sensory evaluation of fresh samples was done by a panel of six semi trained members, based on a 9-point hedonic scale, wherein 9 denoted extremely desirable and 1 denoted extremely undesirable. Water was provided for oral rinsing between the samples.

Result and Discussion

Table 2. Effect of level of pomegranate pulp on the sensory quality of shrikhand.

Treatment	Attributes			
	Smell	Taste	Appearance	Overall acceptability
T0	7.66±1.63	8.16±1.16	8.5±0.83	8.10±0.42
T1	7.33±1.75	8.16±1.16	8±0.89	7.83±0.44
T2	7.5±0.83	8±0.63	8±0.63	7.83±0.28
T3	7.33±1.03	7.66±1.21	7±1.09	7.33±0.33
T4	7.66±0.81	8±1.09	7.83±0.75	7.83±0.17

Effect of level of pomegranate pulp on the sensory quality of Shrikhand

The mean sensory score given by the panelists to the pomegranate shrikhand sample prepared at different treatment given in Table 2 the maximum mean sensory score was obtained by the product sample of treatment T0 and 4 combinations for the smell, taste, appearance and overall acceptability. Out of all these combinations for the taste sample T1 got highest (8.16) score as compared to the other combinations however sample T3 got lowest (7.66) score as compared to other combinations. From present investigation it is reported that for the overall acceptability as level of fruit pulp increased the acceptability also increased but with increase in level of pulp the smell score reduced however taste score of the product increased. The present investigation corroborates with that of Nigamet al. (2009) who also reported that as papaya pulp increased the fat, protein, lactose, sucrose, ash and total solid contents and sensory score significantly decreased with increase in the level of papaya pulp. Hossain et al. (2012) reported that the smell and taste, body and consistency and color and texture of the fruit yoghurts were equally acceptable. 10% and 15% strawberry fruit yoghurt contain more acid and its texture was cracked down in refrigeration temperature. Statistical analysis showed that yoghurt with 10% orange juice was more acceptable than others comparing all quality characteristics. Rita & Jyothi (2013) reported that the incorporating banana pulp at 10% (T1), 20% (T2) and 30% (T3) sensory analysis showed a significant difference in different sensory attributes of T2 sample with the rest of the treatments. Parveez et al. (2014) were undertaken a study to evaluate the effect of orange pulp and chiku pulp in combination (1:1) on the quality attributes of shrikhand. The pulp

combination was incorporated at 0%, 7%, 14% and 21% level (replacing chakka) into the formulation of shrikhand. On the basis of various sensory parameters, shrikhand containing 14% pulp combination was selected as optimum. The pulp combination had a significant ($p < 0.05$) influence on colour and appearance, flavour, body and texture, sweetness and overall acceptability of the Shrikhand.

Conclusion

It can be concluded that the pomegranate shrikhand could be prepared using the pomegranate pulp. The proportion of pomegranate pulp may be taken as 20 gm pulp and 80 gm of chakka for the preparation of pomegranate shrikhand which was superior over all combination for its organoleptic quality. It is also recommended that pomegranate can be effectively used for the production of this newly invented product.

References

- Arjmand, A. 2011. Antioxidant activity of pomegranate (*Punica granatum L.*) polyphenols and their stability in probiotic yoghurt, A M.Sc. Thesis submitted to School of Applied Sciences College of Science, Engineering and Health RMIT University.
- Bardale, P. S., Waghmare, P. S., Zanjad, P. N. and Khedkar, D. M. 1986. The preparation of Shrikhand-like product from skimmed milk chakka by fortifying with fruit pulps. *Indian J. Dairy Sci.* 39(4): 480- 483
- Dadarwal, R., Beniwal, B. S. and Singh, R. 2005. Process standardization for preparation of fruit flavored Shrikhand. *J. Food Sci. Technol.* 42(1): 22-26.
- Geetha, V. V., Sarma, K. S., Reddy, V. P., Reddy, Y. K., Moorthy, P. R. S. and Kumar, S. 2003. Physicochemical properties and sensory attributes of probiotic shrikhand. *Indian J. Dairy Biosci.* 14(1): 58 - 60.
- Hossain M.N., Fakruddin M., Islam M.N. 2012. Quality Comparison and Acceptability of Yoghurt with Different Fruit Juices. *J. Food Process. Technol.* 3:171. doi:10.4172/2157-7110.1000171
- Kumar Sunil, Z.F. Bhat and Pavan Kumar. 2011. Effect of Apple Pulp and *Celosia argentea* on the Quality Characteristics of Shrikhand. *American J. Technol.* 6: 817-826.
- Mehrotra Rolly, Dheer Singh and Svinash Tiwari. 2014. Effect of sugar replacement on chemical composition and organoleptic properties of shrikhand, *Innovar J. Food Sci.* 2(1):22-25
- Nigam Navita; Rashmi Singh; Upadhayay, P. K. 2009. Incorporation of Chakka by papaya pulp in the manufacture of shrikhand, *J. Dairyng, Foods Home Sci.* 28(2):115-118
- Parveez Ahmad Para, Raheeqa Razvi and Nisar Ahmad Nisar. 2014. Effect of Orange Pulp and Chiku Pulp in Combination (1:1) on the Quality Characteristics of Shrikhand, *World J. Dairy Food Sci.* 9 (2): 135-137.
- Rita Narayanan and Jyothi Lingam. 2013. Sensory analysis of banana blended shrikhand, *African J. Agri. Res.* 8(44):5518-5521
- Singh S, Immanuel G. 2014. Extraction of Antioxidants from Fruit Peels and its Utilization in Paneer. *J. Food Process. Technol.* 5:349. doi:10.4172/2157-7110.1000349
- Thakur S.N., Rajni Kant and Ramesh Chandra. 2014. Preparation of Shrikhand by using mango pulp. *Bioved.* 25(1): 79-82.
- Vagdalkar, A. A., Chavan, B. R., Mokile, V. M., Thalkari, B. T. and Landage, S. N. 2002. A study on preparation of shrikhand by using cocoa powder and papaya pulp. *Indian Dairyman.* 54(4): 49-51.
- Varghese Kochupurakkal Shiby, Kolpe Radhakrishna and Amarinder Singh Bawa. 2013. Development of whey-fruit-based energy drink mixes using D-optimal mixture design. *Int. J. Food Sci. Technol.* 48 (4):742-748.

Physico-chemical quality of kefir of etawah crossbred goat milk

Imam Thohari, Hari Purnomo, & Lilik Eka Radiati

Faculty of Animal Husbandry, University of Brawijaya, Malang, Indonesia-65145

Abstract

A study was conducted to determine the effect of incubation time on pH, acidity, protein, fat and alcohol content of etawah crossbred goat milk kefir. The kefir was prepared using etawah crossbred goat milk and 4 % (w/v) kefir grain as starter, and incubated in five different time of incubations as treatments, i.e. 0, 6, 12, 18, and 24 hours. All treatments were repeated four times. Data collected were subjected to analysis of variance of randomized block experiment design. The results showed that time of incubation significantly affect ($p < 0.05$) on pH, acidity, protein, and the ethanol content, while on fat content didn't significantly effect. Incubations time could reduce pH and fat content, while the acid content, protein, and alcohol has increased. It is concluded that 18 hours incubation resulted the best quality of etawah crossbred goat milk kefir.

Keywords: incubation, kefir, etawah crossbred goat milk

Corresponding author: itohfptub@gmail.com

Introduction

Kefir is a fermented milk originating from the Caucasus and has produced hundreds of years ago (Sady et al., 2007). Fermentation on kefir as a result of the work of bacteria and yeasts contained in kefir grain (Farnworth, 2006). Fermented milk kefir-making process will be changes in the composition of nutrients and other elements.

The chemical composition of kefir according to the Codex Standard For Fermented Milks Codex Stan 243-2003 issued by the FAO / WHO (2001) are: protein of at least 2.8% (w/w), milk fat <10% (m/m), acidity expressed as % lactic acid at least 0.6% (m/m), which is the number of specific microorganisms minimum 10^7 starter culture (cfu / g) and a minimum of 10^4 yeast (cfu / g). According Libudzisz & Piatkikiewicz (1990), the dry mass of fresh kefir grains is 10-16 g / 100 g, consisting of 30 g / 100 protein and 25-50 g / 100 carbohydrates. While the chemical composition of kefir grain from Russia, Yugoslavia and Bulgaria according Ottogalli et al., (1973) contains a water content of 90%, 3.2% protein, 0.3% fat, non-protein nitrogen dissolved 5.8% and 0.7% ash.

Kefir chemical content will depend on the type of milk, kefir microflora in the grain, incubation temperature and others. Research by Dimitrelia & Antoniou (2011) on bovin milk kefir with incubation time different would produce a different chemical quality.

Materials and Methods

Kefir made from Etawah crossbred goat milk in Lawang Subdistrict Malang East Java. Variables were measured by pH, acidity, protein content Kyeldahl method, Fat content Gerber method and ethanol content High-performance liquid chromatography (HPLC) method (AOAC, 2000). Experimental design Randomized block design (Steel and Torrie, 1980) and replication 4 times.

The treatments were 0 (control), 6, 12, 18, and 24 hours. The use of kefir grain in each treatment as much as 4%. The incubation at room temperature.

Results and Discussion

Results of the study were pH, acidity, protein, fat content, and the concentration of ethanol as shown in Table 1. Incubation time 0, 6, 12, 18 and 24 hours had a significantly effect ($p < 0.05$) on pH, acidity, protein content, and ethanol content, while the time incubation did not significantly affect on fat content.

Table 1. Effect of incubation on pH, acidity, protein content, fat content, and the ethanol content of goat milk kefir.

Incubation (hours)	pH	Acidity (%)	Protein (%)	Fat (%)	Ethanol (%)
0	6.70 ^a	0.33 ^a	4.011 ^a	5.20	0.31 ^a
6	4.27 ^b	2.01 ^b	4.343 ^b	5.10	0.58 ^b
12	3.74 ^b	2.24 ^b	4.357 ^b	5.05	0.61 ^b
18	3.72 ^b	2.29 ^b	4.530 ^b	4.73	0.62 ^b
24	3.71 ^b	2.31 ^b	4.340 ^b	4.70	0.61 ^b

The highest pH was obtained at 0 hour incubation was 6.70 and the lowest at 24 hours incubation pH of kefir was 3.71. The pH decrease due to lactic acid bacteria and yeasts activity derived from kefir grain. Kefir grain change lactose milk into lactic acid, with the length of incubation, lactic acid was formed more and more, causing the pH to fall.

Incubation 6 hours obtained pH 4.27. Fermentation of kefir begins after inoculation kefir grain, then lactic acid is formed as activity of streptococci that grow faster and effects to decrease of pH, so that lactobacilli can grow (Koroleva, 1988). During the fermentation process of the growth of lactic acid bacteria beneficial to the development of yeast and acetic acid bacteria. Koroleva (1988) states that the microorganisms responsible for acidification of milk in the process of making kefir was streptococci.

The highest acidity was obtained at 24 hours incubation with acidity 2.31% and the lowest at 0 hour of incubation with the acidity 0.33%. The average acidity goat milk kefir was 1.83 (Table 1). Treatment incubation 6, 12, 18, and 24 hours established by the FAO / WHO (2001). Acidity expressed as lactic acid levels in goat milk kefir depending on the levels of lactose fermented by lactic acid bacteria. The fermentation process in the milk after inoculated with kefir grains occur for 24 hours. The lactic acid as a result of activity of streptococci grow faster then lower the pH, so that lactobacilli can grow. This causes the number of streptococci decreased. Yeast in grain kefir together with lactic acid bacteria to ferment at a temperature of 21-23°C. During the fermentation process of the growth of lactic acid bacteria beneficial to the development of yeast and acetic acid bacteria (Koroleva, 1982).

The highest protein content was obtained at 18 hours incubation was 4.340% and the lowest at 0 hour incubation with protein content 4.011%. Time incubation will affect the protein content of kefir. An increase in the amount of protein because of the additional bacterial cells and yeasts. However, the results of research conducted by Belakaaloul et al. (2010) by using SDS-PAGE analysis of the milk fermented with the bacteria found during fermentation will occur hydrolysis of protein by proteolytic enzyme activity.

The average fat content of goat milk kefir in this study was 4.70%. According to Livia (1982), lipolytic enzyme activity will be low in acidic conditions, so that changes that occur in a small fat composition. Regula (2007) said that fermented beverages such as yogurt, bioyogurt, sour milk and kefir made from sheep's milk, the amount of free fatty acids will be influenced by the type of culture stater used. Yogurt has more fatty acids lauric, myristic, palmitic, stearic, and oleic acid compared with kefir, it is inversely proportional to the concentration of acetic acid.

The highest ethanol content obtained at 18 hour incubation was 0.62% and the lowest at 0 hour incubation with ethanol content was 0.31%. Kefir was a dairy beverage that was acid-alcohol (Bosch et al., 2006). Specific yeasts during fermentation to produce ethanol about 0.7 to 2.5% and CO₂ and made kefir has special characteristics (Simova et al., 2002). Ethanol together with other flavor components form the aroma and flavor on kefir ((Beshkova et al., 2003). The factors that influence the production of ethanol was the pH of the culture medium, inoculum concentration, substrate composition, kefir washed or not, type of milk, and no less important variable was the type of milk (Sánchez et al., 2004).

Conclusions

The results showed that the incubation time had significantly effect on pH, acidity, protein content, and the content of ethanol, whereas no effect on lipid levels. Time incubation ranging from 6 hours to 24 hours appropriate by FAO / WHO (2001), but the majority acquisition of kefir grains obtained in 18 hours of incubation.

References

- AOAC., 2000. Official Methods of Analysis of AOAC International. Horwitz, W. ed., 17th ed. Gaithersburg, Maryland.
- Belkaaloul, K., Chekroun, A., Abdessalam, A., Saidi, A. D. and Kheroua, O. 2010. Growth, Acidification and Proteolysis Performance of Two Co-Cultures (*Lactobacillus Plantarum* *Bifidobacterium Longum* and *Streptococcus Thermophilus Bifidobacterium Longum*). Afr. J. Biotech., 9(10): 1463-1469.
- Beshkova D.M., Simova E.D., Frengova G.I., Simov Z.I. and Dimitrov Zh.P., 2003, Production of volatile aroma compounds by kefir starter culture, Int. Dairy J. 13, 529-535.
- Bosch A., Golowczyc M.A., Abraham A.G., Garrote G.L., De Antoni G.L. and Yantorno O., 2006, Rapid discrimination of lactobacilli isolated from kefir grains by FT-IR spectroscopy, Int. J. Food Microbiol. 111, 280-287.
- Buffler. 2008. Compilation of various methodes for protein determination in milk, best on the classic determination by Kjeldahl. BÜCHI Labortechnik AG, Flawil.
- FAO/WHO. 2001. CODEX Standard for Fermented Milks. <http://www.codexalimentarius.net>. Akses 15-06-2009.
- Dimitrelia, G. And Antonioub, K.D., 2011. Effect of incubation temperature and caseinates on the rheological behaviour of Kefir, Procedia Food Science 1 (2011) 583 – 588.
- Koroleva, N.S. 1988. Technology of Kefir and Kumys. IDF. Bull., 227: 96-100.
- Regula, A., 2007. Free Fatty Acid Profiles of Fermented Beverages Made from Ewe's Milk. INRA, EDP Sci., 87: 71-77.
- Simova E., Beshkova D., Angelov A., Hristozova T., Frengova G. and Spasov Z., 2002, Lactic acid bacteria and yeasts in kefir grains and kefir made from them, J. Ind. Microbiol. Biotechnol. 28, 1-6.
- Steel, R.G.D. and Torrie, J.H. (1980). Principles and Procedures of Statistics. A biometrical approach. 2nd edition. McGraw-Hill, New York.
- Livia. A. 1982. Fermentation on Milk Fat of Swedish Fermented Milk Products, 65: J Dairy Sci 65:521-530

Histological Liver of Mice (*Mus musculus*) Consumption Collagen Extract of Broiler's Bone in South Sulawesi, Indonesia

Said, M. I¹., E. Abustam², F.N. Yuliati³ & B. Utomo⁴

¹Lab. By Product Processing Technology, Faculty of Animal Science, Hasanuddin University, Jl. Perintis Kemerdekaan Km.10 Makassar, Indonesia 90245, ²Lab. Meat and Egg Processing, Faculty of Animal Science, Hasanuddin University, Jl. Perintis Kemerdekaan Km.10 Makassar, Indonesia 90245, ³Lab. Microbiology and Animal Health, Faculty of Animal Science, Hasanuddin University, Jl. Perintis Kemerdekaan Km.10 Makassar, Indonesia 90245, Graduate of Animal Science Study Program, Faculty of Animal Science, Hasanuddin University, Jl. Perintis Kemerdekaan Km.10 Makassar, Indonesia 90245,

Abstract

The purpose of this research was to study the effect of the broiler's bone collagen extract consumption in different doses to the liver histology in mice (*Mus musculus*). A total of 30 male mice were used for the experiments and were randomly allocated into five groups (n=6 each group). Level doses of collagen were: Group A as control (0 mg/kg of Body Weight (BW)), dose of group B (5 mg/kg of BW), dose of group C (50 mg/kg of BW), dose of group D (500 mg/kg of BW) and dose of group E (5,000 mg/kg of BW). Microscopic observation in the liver of mice was done after the consumption of collagen extracts. The study of histological liver of mice that were given consumption of collagen extract from broiler's bone with level doses respectively: 0 ; 5 ; 50 and 500 mg/kg of BW indicated that the observation the level of damage to liver tissue <30%, but to level dose of 5,000 mg/kg of BW has shown the extent of damage >30% and has been characterized by inflammatory cell infiltration process. The results of this research showed that the dose limits extract collagen that is still acceptable of mice (*Mus musculus*) is 500 mg/BW.

Keywords: liver, mice, collagen extract, broiler, bone

Introduction

Broiler's bone riched in collagen compounds is a kind of waste produced by the chicken slaughter. Collagen, a hydrocolloid product resulted from partial hydrolysis of animal tissue, is currently used widely in food and pharmaceutical fields. Structurally, bone is rich in bioactive compounds particularly collagen protein (Zeugolis et al., 2008, Nagai et al., 2008, Muyonga et al., 2003). Collagen extracts widely used by humans as a food supplement, especially for the elderly. Collagen extract is generally produced using chemical processes such as acids and bases (Kolodziejka et al., 2003; Wang et al. 2008).

Chemical compounds such as strong acids and a strong bases type are now widely used as a main ingredient in the production process of collagen extract. This material was aims to increase of the yields and accelerate in the production process. The collagen extract is a non-toxic compound, but the use in large doses can be cause pseudo-agglutinations and fixation of proteins in tissues. These effects are reversible which continually be eliminated from the body through the elimination process (Rachmawati et al., 2011). However, some of the chemical compounds that are used in the production process of collagen extract such as HCl, H₂SO₄ and NaOH are toxic compounds.

Chemical compounds such as weak bases can be also using in the production process of collagen extract from bovine's bones. Such as research of Said et al., (2011), that the production process of collagen extracts especially for curing stage, chemical compound $(\text{Ca}(\text{OH})_2)$ 100 g/L can be used to increase the yields of collagen.

Use of chemical compounds strong acids and strong bases in the production process of food for human thought to have an impact on human health. Liver is the very sensitive organ to chemical contamination in humans. The liver is an organ that plays an important role in detoxifying various toxic substances that get into the digestive tract. Toxic substances that enter and absorbed in the gastrointestinal tract and then are carried by the portal vein to the liver (Lu, 1995).

Preliminary studies have been conducted by the authors of the lethal dose (LD_{50}) in mice given consumption of the collagen extract from bovine's bone. The study showed that the extract collagen from bovine's bone with doses orally to 15,000 mg/kg of body weight in mice (*Mus musculus*) results LD_{50} is not achieved (the mortality rate is only 30%), which indicates the category of practically non-toxic by Lu (1995) (Said, 2013).

In this study will be assessed, how to effect of the consumption of the collagen extract from the broiler's bone that using a chemical compound (H_2SO_4) in their production process (bone demineralization process). The purpose of this research was to study the effect of the consumption of the collagen extract from broiler's bone at different dose levels to the liver histology in mice (*Mus musculus*).

Materials and Methods

Materials

A total of 30 male mice (*Mus musculus*), Balb/C strain, 2-3 months age, body weight of 20 to 25 grams was used as animal experiment and extract collagen from broiler's bone, distilled water and commercial feed was used as research material. Trial cages, feeding and drinking tools, digital scales, water bath, and oral needles no. 14 was used as research tools.

Methods

Research design

Complete Randomized Design (CRD) with five group treatments and three replications was used as the research design. There are five group levels doses of collagen extract that have been applied in this study (n=6 each group), namely: group A (0 mg/kg of BW (as control); group B (5 mg/kg of BW); group C (50 mg/kg of BW); group D (500 mg/kg of BW) and group E (5,000 mg/kg of BW).

Preparation of experimental animals

A total of 30 male mice (*Mus musculus*), 2-3 months age, BW of 20 to 25 grams were placed in the main cage (100 cm x 200 cm x 100 cm size) made of wire. It was placed in room with a good air circulation and natural lighting systems (24 hours). A total of 15 units of small cages (15 cm x 20 cm x 15 cm size) from PVC (*polyvinylchloride*) plastic were placed inside at the main cage. Each small cages were placed 2 male mice as experimental animals. In the small cages were placed each one unit of feeding and one unit water container from PVC plastic. The lighting system using a natural lighting for 24 hours. Feeds and water were provided *ad-libitum*, but previously dosed to determine the amount of feed and water intake.

Production process of collagen extract

In this study was referred to the collagen extract production process from broiler's bone according to Ockerman and Hansen (2000) which had been modified. A total of 300 g broiler's

bone composite was inserted into the glass beaker containing ethanol 70% as degreasing solution. Degreasing process was carried out for 24 hours at a room temperature. The bone demineralized by H_2SO_4 , was called *ossein*. Then, it was neutralized with alkaline solution ($Ca(OH)_2$ 20% w/v) for 24 hours. The first extraction process was carried out in a water bath under temperature of 70°C (fraction 1) for 24 hours followed by a second extraction at a temperature of 75°C (fraction 2) for 24 hours. The result of the extraction was collagen extract.

Preparation of extract collagen solution

The collagen extract were dissolved in the 100 ml of distilled water as temperature 80°C with several doses level, namely: 0 mg/100 ml (0 mg/kg of bw) (group A); 0.013 mg/100 ml (5 mg/kg of bw) (group B); 0.13 mg/100 ml (50 mg/kg of bw) (group C); 1.3 mg/100 ml (500 mg/kg of bw) (group D) and 13 mg/100 ml (5,000 mg/kg of bw) (group E).

Treatment in experimental animals

Treatment was given for 30 days in experimental animals. Overall experimental animals were given a standard diet in the form of commercial pelleted feed and sterile water. Before being granted a suspension of collagen, the mice were fasted for 2-3 hours. Giving oral suspension was done by using the syringe and given one time a week for 30 days (four times for one month). The doses level was applied in animal experiments as follows: group A: 0 mg/kg of BW as control; group B: 5 mg/kg of BW; group C: 50 mg/kg of BW; group D: 500 mg/kg of BW and group E: 5,000 mg/kg of BW. On the 31th day, as many as two male mice were randomly selected from each group to be killed as an object of observation. The liver was taken and then washed with physiological solution as well as in the fixation with formalin solution.

Techniques of data collection

Making preparations for histology and staining was performed in the Center for Veterinary in Maros regency, South Sulawesi. Making preparations have been carried out by fixation technique using formalin, and then soaked in 70% alcohol. The next stage was dehydrated in alcohol 80%. Liver tissue purification process has been carried out in the next stage of xylol and paraffin embedding. Tissue blocks were cut using a microtome (5 μ m) and a piece of tissue attached to the glass object.

Result and Discussion

In generally, collagen extract are protein that can be extracted both from bones and skin of the animal. Collagen is a product of the partial hydrolysis portion of the tissue of animal. Based on the type of material, collagen extracts are considered safe for consumption because it is natural, but in the production process is often questionable. This is because the process is still using some kind of chemicals. Presence of the chemical compounds contained in human food can certainly have an impact on body organs, one of which is a very sensitive organ is the liver.

The liver is the organ absorption of nutrients absorbed from the gastrointestinal tract, processed and stored for other organs. The bloods from the abdominal cavity and intestines will flow in to the first into the portal vein. Blood contains many important substances such as carbohydrates, fats and vitamins that can be absorbed and stored in the liver as long as needed (Stine and Brown, 1996). Histological liver of male mice (*Mus musculus*) that had been given collagen extract consumption at various doses for 30 days presented in Figure 1-5.

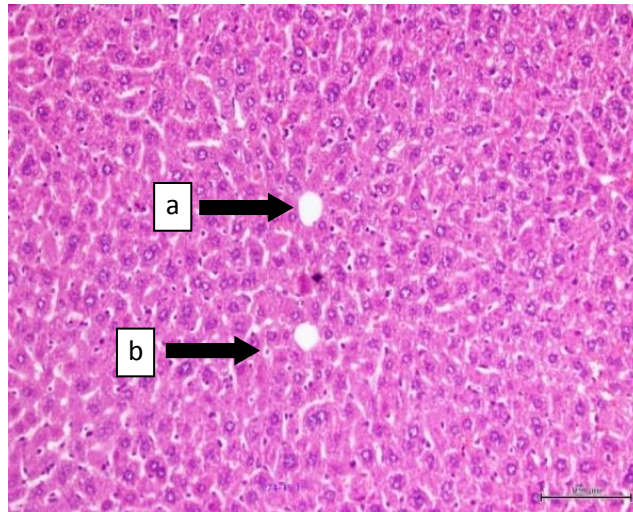


Figure 1. Histological appearance of the liver of mice after consumption of collagen extract (0 mg/kg of BW dosage) (control) for 30 days (magnification of 100x)
 (Note : (a) venous centralis normal/no damage ; (b) hepatocit cells normal bw = body weight of mice (*Mus musculus*))

Fig.1 shows the structure of the liver tissue of mice that were not given collagen extract consumption during the study (0 mg/kg of bw) (group A) (controls). The results showed the rats' network structure unchanged (normal). The centralis venous and hepatocytes showed normal shape. Hepatocytes or liver cells are the biggest part of the liver. These cells are responsible for the metabolism of the liver. These cells are located between the sinusoids containing blood and bile ducts. Sinusoid is coming from the edge of the lobules in the venules in the branches of the portal vein and hepatic artery towards the center and empties into the central vein (Sri-Gandono, 1992). A toxicant can be absorbed through various channels after absorbed, toxicant is absorbed into the various parts of the body, including organs of excretion and then excreted from the body. Many chemicals undergo biotransformation in the body. The most important place is the liver, this process also takes place in the lungs, kidneys and stomach.

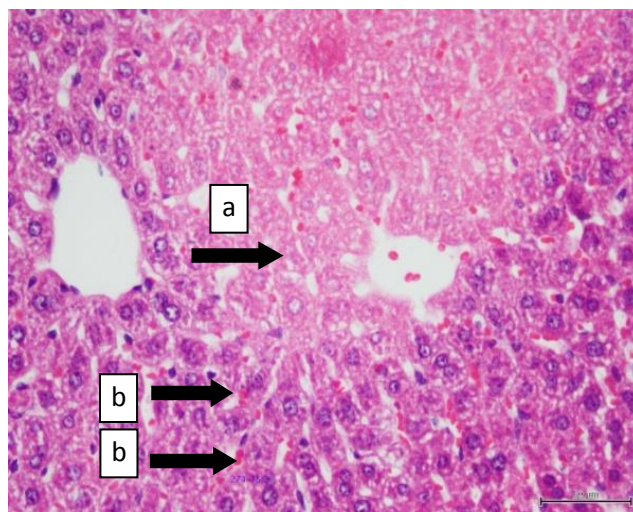


Figure 2. Histological appearance of the liver of mice after consumption of collagen extract (5 mg/kg of bw dosage) for 30 days (magnification 200x)
 (Note : a = necrotic hepatocytes around the vena centralis ; b = occurred hemorrhage ; bw = body weight of mice (*Mus musculus*))

Consumption of collagen extract on male mice (*Mus musculus*) (5 mg/kg of bw) (group B) for 30 days showed changes in liver histology (Fig.2). Changes that occur were the presence of necrosis of the hepatocytes around the central vein and hemorrhage, but did not show significant damage. Consumption of collagen extract is closely related to the digestive process in the body. According to Ganiswarna (1995), that in the pharmacokinetics of each drug in your body will be undergo a process of absorption, distribution, metabolism and excretion. Similarly, the extract of collagen will be absorbed by the intestine and then metabolized in the liver. The liver is the first organ to be achieved by drugs and other substances absorbed from the intestine via the portal vein, so it is mentioned that the liver is the main place of drug metabolism and detoxification. The buildup of toxic substances in the liver parenchymal cells can injure hepatocytes and causes varying histopathological changes (Himawan, 1992).

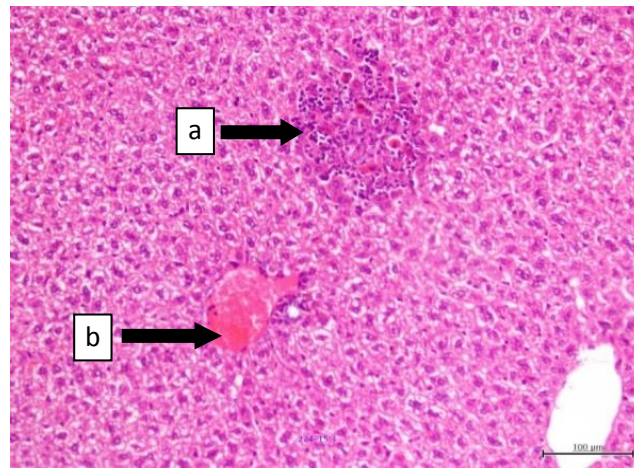


Figure 3. Histological appearance of the liver of mice after consumption of collagen extract (50 mg/kg of bw dosage) for 30 days (magnification 100x)

(Note : a = fibrin ; b = makrofag on the focal cell ; bw = body weigh of mice (*Mus musculus*))

The appearance of liver histology of male mice given bone collagen extract consumption of broiler (50 mg/kg of bw) for 30 days (Fig.3), shows that there has been a change in the form dilatation of the blood vessels and filled with fibrin, as well as macrophages occurs in focal cells. Damage of the liver due to toxic substances are influenced by several factors, such as the type of chemicals, the dose given and the duration of exposure to substances such as acute or chronic subchronic. Higher of the concentration of a given compound the toxic response caused even greater. Damage of the liver can occur immediately or after a few weeks to several months. Damage can be shaped necrosis of hepatocytes, cholestasis, hepatic dysfunction or onset slowly (Amalina, 2009). Damage of the liver is closely related to bleeding and an array of smaller units that acini of the liver, which is the latest concept from the smallest functional unit of the liver. Hepatocyte is a cell with polyhedral shape which has a surface of six or more, with a clear cell membrane and spherical nucleus in the middle (Bhara, 2004).

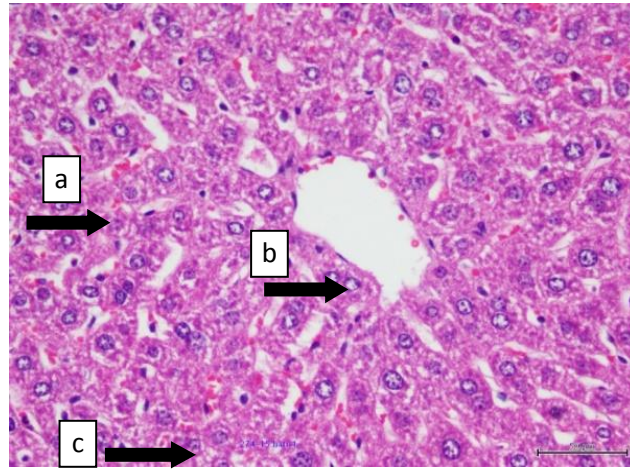


Figure 4. Histological appearance of the liver of mice after consumption of collagen extract (500 mg/kg of BW dosage) for 30 days (magnification 200x)
 (Note : a=hemorrhage, b=necrotic, c=venous centralis; bw=body weight of mice (*Mus musculus*))

Damage to each treatment has shown tend to be similar, namely necrotic, hemorrhage and there macrophages on the consumption collagen extract (500 mg/kg of bw) (group D) (Fig.4). It has started to look for damage to be nekrosa on hepatocyte cells and hemorrhage. Hepatic necrosis has occurred spontaneously in test animals after the administration of high doses of therapeutic agent. This is due to the direct influence of toxic agents such as chemicals or toxins germs. Necrosis is the death of cells or tissue in a living organism. The core of dead cells look smaller, chromatin fibers and reticular be multiplied and will become more dense nucleus and eosinophilic (kariolisis) (Kasno, 2003).

The changes are visible in the microscopic observation such as: inflammation, fibrosis, degeneration, and necrosis. According to Robins and Kumar (1992), that the liver damage due to chemical compounds characterized by biochemical lesion gives a series of changes in the function and structure. Some changes in the structure of the liver due to a chemical compound that can be seen in such microscopic observation, inflammation, fibrosis, degeneration, and necrosis. Necrosis is the death of cells or tissue in organism life.

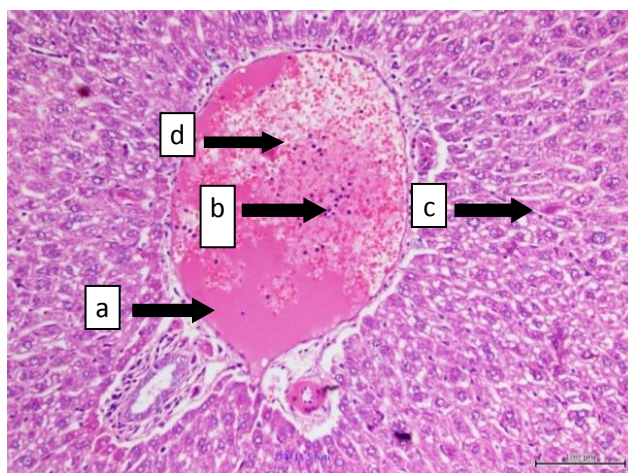


Figure 5. Histological appearance of the liver of mice after consumption of collagen extract (5,000 mg/kg of bw dosage) for 30 days (magnification 400x)
 (Note : a = fibrin ; b = Inflammatory cell infiltration ; c = hydropic degeneration ; d = macrophages ; bw = body weight of mice (*Mus musculus*))

Fig.5 shows the histological liver of male mice giving of collagen extract consumption with a dose of 5,000 mg/kg of bw for 30 days. The analysis showed a highly significant structural change. Hepatotoxicity chemical compound is a potential complication that is almost always present in any given chemical compound. The liver is the central metabolic disposition of all drugs and foreign substances that enter including collagen extract. The liver cell damage caused by a substance directly, but often by the toxic metabolites of the substance was in question. The process of metabolism is not running normally, it will cause various diseases, one of which is a disease that occurs in the liver. The cells contained in the liver will be deposited so that it will undergo a change (Jayanti, 2011). Results of this study showed that the higher dose of the extract is applied, and then the change liver damage caused greater. Dose is a factor that determines whether a chemical substance is toxic or not (Rasyid et al., 2011).

Conclusion

The higher the dose of extract collagen consumed by the mice, it is more visible changes in liver histology of mice (*Mus musculus*). The use dose of collagen extract of broiler's bone until the level of 500 mg/kg of BW still showed normal liver tissue structure changes in mice (*Mus musculus*).

Acknowledgement

The authors sincerely thanks to the Ministry of Technology Research and Higher Education on funding research grants of National Strategic on the second year with the contracts number: 1764/UN4.20/ PL.09/2015 on March 31, 2015 and then my students Selfin Tala, S.Pt as colleagues in the laboratory

References

- Amalina, N., 2009. Uji toksisitas akut ekstrak valerian (*Valerina officinalis*) terhadap hepar mencit balb/c. Karya Tulis Ilmiah. Fakultas Kedokteran Universitas Diponegoro, Semarang.
- Bhara, M., 2004. Pengaruh Pemberian Kopi Dosis Bertingkat Per Oral 30 Hari Terhadap Gambaran Histologi Hepar Tikus Wistar. Laporan Akhir Karya Tulis Ilmiah. Fakultas Kedokteran Universitas Diponegoro, Semarang. Hal22-28.
- Ganiswarna., 1995. Farmakologi dan Terapi. Edisi IV. Badan Penelitian dan Pengembangan Kesehatan. Departemen Kesehatan, Jakarta. Hal12.
- Himawan, S., 1992. Patologi. Penerbit Buku Kedokteran, Jakarta. Hal86.
- Jayanti, D. P., 2011. Pengaruh perbedaan lama pemberian diet kolesterol terhadap perlemakan hati pada Tikus Putih (*Rattus norvegicus*). Skripsi. FST Universitas Islam Negeri Maulana Malik Ibrahim, Malang. Hal 38-39.
- Kasno, P. A., 2008. Patologi Hati dan Saluran Empedu Ekstra Hapatik. Balai Penerbit Universitas Diponegoro, Semarang. Hal 86.
- Kolodziejaska, I., K. Kaczorowski, B. Piotrowska and M. Sadowska, 2003. Modification of the properties of gelatin from skins of Baltic cod (*Gadus morhua*) with transglutaminase. J. Food Chem. 86: 203-209.
- Lu, F. C., 1995. Toksikologi Dasar. Edisi Kedua. Universitas Indonesia Press, Jakarta hal 206-214.
- Muyonga,J.H., C. G. B. Cole and K. G. Duodu, 2004. Extraction and physico-chemical characterization of Nile perch (*Lates niloticus*) skin and bone gelatin. J. Food Hydrocolloids. 18: 581-592.

- Nagai, T., N. Suzuki and T. Nagashima, 2008. Collagen from common minke whale (*Balaenoptera acutorostrata*) unesu. *J. Food Chem.* 111: 296-301.
- Ockerman, H. W and C. L. Hansen, 2000. *Animal by Product Processing and Utilization*. CRC Press, USA. pp183.
- Rachmawati, N., R. Triwibowo dan A. K. Putri, 2011. Toksisitas subkronik gelatin kulit ikan patin Siam (*Pangasius hypophthalmus*) terhadap mencit. *Jurnal Pascapanen dan Bioteknologi Kelautan dan Perikanan*. 6: 81-90.
- Said, M.I., S. Triatmojo, Y. Erwanto dan A. Fudholi, 2011. Gelatin properties of goat skin produced by calcium hydroxide as curing material. *Jurnal Media Peternakan*. 34(3): 184-189.
- Said, M. I., S. Triatmojo, Y. Erwanto dan A. Fudholi, 2013. Profil histologis serabut kolagen pada kambing bligon yang direndam dalam larutan asam dan basa lemah pada konsentrasi berbeda. *Jurnal Ilmu dan Teknologi Hasil Ternak (JITEK)*. 8(1): 19-24.
- Sri-Gandono., 1996. *Anatomi Fisiologi Ternak*. Gadjah Mada University Press, Yogyakarta. Hal15.
- Stine, K.E and T. M. Brown, 1996. *Principles of Toxicology*. USA: CRC. Press Inc. Lewiss Publishers. pp123.
- Rasyid, M., Usmar dan Subehan, 2011. Uji toksisitas akut ekstrak etanol lempuyang wangi (*Zingiber aromaticum* Val.) pada mencit. *Fakultas Farmasi Universitas Hasanuddin, Makassar*. Hal 13-19.
- Robbins, S. L dan V. Kumar, 1992. *Buku Ajar Patologi 1*. Penerbit Buku Kedokteran EGC, Surabaya. Hal 14-17.
- Wang, L., B. Yang, R. Wang and X. Du, 2008. Extraction of pepsin-soluble collagen from grass carp (*Ctenopharyngodon idella*) skin using an artificial neural network. *J. Food Chem.* 111: 683-686.
- Zeugolis, D. I., S. T. Khew, E. S. Y. Yew, A. K. Ekaputra, Y. W.Tong, L. L. Yung, D. W. Hutmacher, C. Sheppard and Michael, 2008. Electro-spinning of pure collagen nanofibres-Just an expensive way to make gelatin. *J. Biomaterials*. 15: 2293-2305.

Effect of jelly addition on Kefir quality

Amertaningtyas D.¹, Arin M.R.², Rimayanti I.P.², Imtichanul F.² & Valentine F.S.²

¹Lecture of Animal Product Technology of Animal Husbandry Brawijaya University Indonesia,

²Student of Animal Husbandry Brawijaya University Indonesia

Abstract

This study aims to the effect of addition of jelly on physico-chemical and organoleptic quality of kefir. The research material is goat milk kefir and commercial jelly. The research method was an experiment with completely randomized design (CRD), 4 treatments, the addition of jelly concentration of 1%, 1.5%, 2% and 2.5%, 4 replications, followed by Least Significant Difference Test (LSD) if there different and to determine the best treatment. The results showed that the kefir added various concentrations of jelly showed different significant ($P < 0.01$) in viscosity (4474.75 cP), fat content (3.545%), moisture content (68.76%), pH (3.99), but not significance ($P > 0.05$) in fiber content (0.297%) and protein content (2.446%). Organoleptic qualities provide different significant ($P < 0.01$) such as colour (3.24), flavour (3.26), texture (2.99) and taste (2.97). The conclusion is the addition of jelly on kefir is a different significant on the viscosity, fat content, moisture content, pH and organoleptic quality but not significance on the fiber content and protein content. Addition of the best jelly on goat milk kefir is 1.5%.

Keywords: goat milk kefir, jelly, physico-chemical, organoleptic

Introduction

Kefir is a dairy products through the fermentation process. Belkaaloul et al. (2010), milks can be fermented into a new product which is fermented milk and contains ethanol, the Bulgarian specifications (high acid), acidophilus and yoghurt (acidic medium), cultured buttermilk and cultured cream (low acid). Otles & Oslem (2003) and Sady et al. (2007), kefir is a native drink the Caucasus mountains. These characteristic are flavor, colour and consistency kefir like yoghurt but kefir has a distinctive yeasty aroma. Kefir is fermented with kefir grains, namely white granules from a collection of bacteria and yeasts. Hertzler & Clancy (2003), kefir commercially in the United States using a mixture of cultures, among others *Streptococcus lactis*, *Streptococcus cremoris*, *Streptococcus diacetylactis*, *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, *Leuconostoc cremories* and *Saccharomyces florentinus*. Kefir is a probiotic drink or functional foods, its means foods containing compounds, works for consumers' health, reduce the risk and protection of diseases such as hypertension, diabetes, cancer, osteoporosis and liver disease. For Fermented Milks Codex Standards Codex Stan 243-2003 issued by the FAO/WHO (2001), the composition of kefir is a protein of at least 2.8% (w/w), milk fat <10% (m/m), minimum acidity of lactic acid 0.6% (m/m), the number of specific microorganisms which is the starter culture of at least 10^7 (cfu/g) and a minimum of 10^4 yeast (cfu/g).

Materials and process of making kefir affects the composition of kefir. Sheep's milk kefir and treatment of different long incubation significant effect on the physico-chemical kefir (Motaghi et al., 1997). Karagozlu & Kavas (2000), the process of making traditional kefir milk is heated, cooled to a temperature of 20-25°C and inoculated 2-10% kefir grains, fermented 18-24 hours 20-25°C temperature, filtering kefir grain, stored cold temperatures. The study was

developed to improve the kefir quality and consumer preferences, with the addition of honey (Jaya et al., 2014), modified starch canna (*Canna edulis, Ker*) (Thohari et al., 2013). It should be research on the addition of jelly on the quality of kefir. Jelly. The main content in the form of carrageenan jelly extracted from red algae (*Rhodophyceae*),

Materials and Methods

Materials: goat milk kefir “Peranakan Etawa” commercial, jelly powder commercial, commercial sugar and water. Viscosity test equipment is a Brookfield viscometer DV type-II + Pro, test equipment fat content (Gerber), protein assay (Buchi Auto Kjeldahl Unit K-370. Methods: experiment with completely randomized design (CRD), which is the percentage jelly 4 treatments: 1%, 1.5%, 2% and 2.5%, 4 replications, assessment of the 30 panelists (non-trained testers) for organoleptic test, based on a 1-5 scale preference. Treatment of jelly addition (1%, 1.5 %, 2% and 2.5%) of the weight of kefir, add sugar 1%, dissolved warm water (300 ml), stirred, put into kefir (700 ml), mixed, heated 30°C for 5 minutes, be cooled. Data test tested Least Significant Difference (LSD). the variables measured were: Brookfield viscosity methods (2005), fat contents AOAC methods (2000), fiber contents AOAC methods (2000), protein contents AOAC methods (2000) and organoleptic test is colour, flavour , texture and taste.

Results and discussion

Physicochemical quality and organoleptic quality of kefir jelly are shown in Table 1.

Table 1. Data of different concentration jelly effect on kefir quality.

Treatments	Viscosity	Fat content	Protein content	Fiber content	Moisture content	pH	Colour	Flavour	Texture	Taste
1%	116.00 ^a	2.385 ^a	2.245	0.170	73.13 ^e	3.95 ^a	3.45 ^c	3.06 ^a	3.36 ^{bc}	2.67 ^a
1,5%	5061.75 ^b	4.823 ^b	2.635	0.368	63.92 ^a	3.98 ^{ab}	3.18 ^{ab}	3.40 ^b	3.00 ^b	3.06 ^b
2%	4172.25 ^b	4.228 ^c	2.495	0.228	71.67 ^d	3.94 ^a	3.17 ^a	3.32 ^b	3.03 ^b	3.03 ^b
2,5%	8549.00 ^c	2.745 ^d	2.410	0.423	66.23 ^b	4.08 ^b	3.17 ^a	3.29 ^b	2.56 ^a	3.11 ^{bc}
average	4474.75	3.545	2.446	0.296	68.74 ^c	3.99 ^{ab}	3.24	3.27	2.99	2.97

Description: Notation superscript letters in the same column indicates different significant (P<0.01).

The average on viscosity(4474.75 cP), fat content (3.545%), protein content (2.446%), fiber content (0.296%), moisture content (68.74%) and pH (3.99), organoleptic quality are 3.24colour, 3.26 aroma, 2.99 texture and 2.97taste. The addition of jelly effect on the viscosity of kefir for making matrix viscosity increase which affects the viscosity kefir because of jelly serves as a stabilizer. Belizt & Grosch (1999), the viscosity is affected by the concentration and molecular weight stabilizer. The higher the value of molecular weight and the concentration of the stabilizer, the viscosity increased. Jelly also affect the levels of fat kefir because microorganism survive and kefir has the prebiotics. The microorganism produces lipase and hydrolyzes fatty acids. Livia (1982), lipolytic enzyme activity will be low in acid condition so that changes that occur in a small fat composition. Motaghi et al. (1997), the kefir grain microorganisms will metabolize fermentation by converting proteins, fats and carbohydrates for survival. Coskun & Ondul (2004), *lactic acid bacteria* (LAB) *L. Acidophilus*, *L. Brevis*, *L. fermentum*, *L. Plantarum*, *S.thermophilus*, and *S. bulgaricus* is a bacteria that play a role in lipolysis contribution and increase of fatty acids in the product milk. The addition of jelly does not affect the levels of protein and fiber content kefir because jelly is added a little protein so it does not give effect to levels of protein and fiber content. The addition jelly very significant effect on the organoleptic quality. Panelists like kefir mixed with jelly because jelly can be change the colour, aroma, texture and taste of kefir. Plessas et al. (2004), kefir in bread dough to improve the quality of bread is better to loaf volume, flavour, texture and self life compared

dough bread using baker's yeast, firmer texture. Jaya et al. (2014), the addition of 5% rambutan honey on kefir during 36 hours fermentation provide the best antioxidant (86.53%). Thohari et al. (2013), the addition of 1% starch modification canna (*Canna edulis*, Ker) improve the quality of kefir in terms of pH, water content, viscosity and protein. The conclusion was that the addition of 1-2.5% jelly can improve the quality of goat milk kefir physicochemical quality and organoleptic quality. Jelly additional treatment of 1.5% is the best jelly additions.

References

- AOAC. 2000. Official Methods of Analysis of AOAC International. Horwitz, W. Ed., 17th Ed. Gaithersburg, Maryland.
- Belkaaloul, K., A. Chekroun., A. Abdessalam., A.D. Saidi., and O. Kheroua, 2010. Growth, Acidification and proteolysis Performance of Two Co-Cultures (*Lactobacillus plantarum*, *Bifidobacterium Longum* and *Streptococcus Thermophilus*, *Bifidobacterium Longum*). Afr. J. Biotech. 9(10):1463-1469.
- FAO/WHO. 2001. Codex Standard for Fermented Milks. <http://www.codexalimentarius.net>. Akses 15-06-2009.
- Hertzler, S.R, and Clancy, S.M. 2003. Kefir Improves Lactose Digestion and Tolerance in Adults With Lactose Maldigestion. The American Dietetic Ass. J. 103:582-587.
- Immerson, A.P. 2000. *Carrageenan*. Citation on Philips, G.O. and P.A. Williams, Editor. Handbook of Hydrocolloids. Florida: CRC Press.
- Karagozlu, C, and G. Kavas. 2000. Alkllu Fermented SutIcecekleri: Kefir ve Kimizin Ozelliklerive Insan Beslen Mesindeki Onemi. Gida. 6:86-93.
- Motaghi, M. Mazaheri, N. Maozami, A. Farkhondeh, Fooladi, M.H and Goltapeh, E.M. 1997. Kefir Production in Iran. Word Microb. And Biotechnol.J. 13:579-581.
- Otles, S. and Ozlem, C, 2003. Kefir: A Probiotic Dairy-Composition, Nutritional and Therapeutic Aspects. Pakistan. J. of Nut. 2(2):54-59
- Plessas, S., L. Pherson., A. Bakatarou., P. Nigam, and A.A. Koutinas. 2004. Bread Making Using Kefir Grains as Baker's Yeast. Foodchem. J.10:1-5.
- Thohari, I., D. Amertaningtyas, D. Rosyidi, Purwadi, L.E.Radiati, and F. Jaya, J. 2013. Effect of Canna Starch (*Canna edulis*, Ker) Modification on Kefir Quality. Departement of Animal Products and Technology. Animal Husbandry. Brawijaya University. Indonesia.pp 17-23
- Sady, J., Domagala, T., T. Grega and Najgebauer-Lejko, D. 2007. Sensory and PhysicoChemical Properties of Commercially Available Kefir. Biotech. J. in Animal Husbandry. 23:199-206.
- Jaya,F., D. Amertaningtyas., D. Rosyidi., M. E.Sawitri., and E. S.Widyastuti.2014. Natural Antioxidant Properties and Physico-Chemicals of Kefir Prepared by Combination of Local Honey and the Time of Fermentation of Goats Kefir. Sustainable Livestock Production in the Perspective of Food Security, Policy, Genetic Resource, and Climate Change. The 16th AAAP Congress. Proceedings Vol. II. Yogyakarta. Indonesia.pp 332

Crude triterpenoid, phenolic compounds and enzyme activities of fermented soybean hull by *Antrodia cinnamomea*

Lee*, T. T., H. C. Lin & B. Yu

Department of Animal Science, National Chung Hsing University, Taichung, Taiwan

Abstract

One of brown-rot fungus, *Antrodia cinnamomea*, also known as Niu-Chang-Chih, was a unique species which only parasitizes in decayed heart woods of stout camphor (*Cinnamomum kanehirae*) in Taiwan. The study was demonstrated that using solid-fermented soybean hulls by *A. cinnamomea* and evaluated its functional components and enzyme activities to evaluate as a functional fermented feed. The results showed that soybean hull inoculum contained 60% of fungal mass after 21 days fermented which contained 16.80 mg/g DW and 4.50 mg gallic acid equivalent (GAE)/g DW of crude triterpenoid and total phenolic contents, respectively. Hence, the carbohydrates enzymes were produced, such as mannanase (5.40 U/g DW), xylanase (21.30 U/g DW) and cellulases (3.0 U/g DW), respectively. In conclusion, solid-fermented soybean hulls by *A. cinnamomea* could increase functional metabolites and carbohydrates enzymes activity; therefore, the fermented feed by *A. cinnamomea* could be potential functional feed.

Keywords: *Aureobasidium pullulans*, solid-state fermentation, functional compounds.

Introduction

Biological pretreatments, wood destroying microorganisms that attack wood naturally, degrading lignin and cellulose, are allowed to grow on the biomass, producing holocellulose–lignin complex breaking. Brown-rot fungi are the most destructive type of wood decay fungus. Brown-rot fungi degrade preferentially wood polysaccharides and partially oxidize lignin substrate (Damisa et al., 2008). They degrade holocellulose causing a rapid decrease in degree of polymerization with a low mass loss as well as its degradation products are produced faster than they are utilized. The fungi could naturally break down the highly ordered cellulose crystalline structure (Green, 1997).

Antrodia cinnamomea, also known as *Antrodia camphorata* or Niu-Chang-Chih, is a fungus indigenous to Taiwan which grows on decayed *Cinnamomum kanehirae*. Currently, the use of soybean hulls in the food and feed industry has increased distinctly and visibly over the last decade. The purpose of the study was assay the crude triterpenoid, phenolic compounds and enzyme activities of fermented soybean hull by *Antrodia cinnamomea* and thus evaluate as a functional feed.

Materials and Methods

***Antrodia cinnamomea* packing bags cultivation and treatment**

Soybean hulls were autoclaved at 121 °C for 15 min. The soybean hulls were inoculated with *Antrodia cinnamomea* packing bags containing 60% of fungal mass.

Bioactive components assay

Trichloroacetic acid, chloroform-sulfuric acid and glacial acetic acid-acetyl chloride were used for the qualitative detection of crude triterpenoids according to the procedures described previously (Wu, 2004). The Folin-Ciocalteu reagent was mixed evenly with CMWM extracts before adding the Na₂CO₃ solution and thus measured with a spectrophotometer at 730nm. Then, with the contents of the phenolic compounds of the extracts, a microgram of the gallic acid equivalent (GAE) was determined using the equation that was obtained from the standard gallic acid graph (Lee et al., 2003).

Enzyme activities

The contents of fermented soybean hulls were then filtered through a microstrainer followed by vacuum filtration using a Buchner funnel lined with Whatman No.1 filter paper. Enzyme activities were determined using a microprocessor-based double beam UV–vis spectrophotometer at 540 nm. One International unit (IU) of both hemicellulase activities was defined as the quantity of enzyme required to liberate 1 μm of reducing sugar (mannose/xylose/cellulose) of crude filtrate per minute under standard assay conditions.

Results

Bioactive components assay

The results showed that soybean hulls inoculum contained 60% of fungal mass after 21 days fermentation contained 16.80 mg/g DW and 4.50 mg gallic acid equivalent (GAE)/g DW of crude triterpenoid and total phenolic contents, respectively.

Enzyme activities

The carbohydrates enzymes were produced such as mannanase (5.40 U/g DW), xylanase (21.30 U/g DW) and cellulases (3.0 U/g DW), respectively.

Conclusion

Solid-fermented soybean hulls by *A. cinnamomea* could increase functional metabolites and carbohydrates enzymes activity; therefore, the fermented feed by *A. cinnamomea* has potential to be a functional feed.

Acknowledgments

The authors thank the National Science Council (MOST 103-2628-B-005-001-MY3) for financially supporting this study.

References

- Damisa D, Ameh J. B. and Umoh V. J. 2008. Effect of chemical pretreatment of some lignocellulosic wastes on the recovery of cellulase from *Aspergillus niger* AH3 mutant. *Afr. J. Biotechnol.* 7:2444–2450.
- Green F, Highley T. L. 1997. Mechanism of brown-rot decay: paradigm or paradox. *Int. Biodeterior. Biodegrad.* 39:113–124.
- Lee TT., Ciou J. Y., Chen C. L. and Yu B. 2013. Effect of *Echinacea purpurea* L. on oxidative status and meat quality in Arbor Acres broilers. *J. Sci. Food Agric.* 93:166-172.
- Wu L.J. 2004. *Natural Pharmaceutical Chemistry*. People's Medical Publishing House, Beijing (in Chinese) pp. 376-377.

Effect of performance and intestinal characteristics on supplemented with protease in the broiler diet

Yu*, B., Y. J. Wang & T. T. Lee

Department of Animal Science, National Chung Hsing University, Taichung, Taiwan

Abstract

This study was demonstrated that supplementation of protease in the diets to evaluate the effect on performance and intestinal characteristics of broilers. One thousand four hundred 0-day old broilers were divided to two batches. There were four replications in every batch, two males and females respectively. Broilers were randomly allocated into following treatments: T1 (maize-soybean meal diet); T2 (T1 diet decreasing 5% CP); T3 (non-conventional diet including DDGS, meat bone meal and feather meal); T4 (T3 diet decreasing 5% CP), and 0.05% protease was added in the T2, T3 and T4 to be the enzyme supplement groups. The results showed that the based on different protein sources, T1 and T2 had the better weight gain and FCR than T3 and T4 in starter (0-10d) and grower (11-20d) period ($P<0.05$), but not occurred in finisher (21-35d) period. There was a significantly better FCR in normal protein diet than that of the low-protein diet ($P<0.05$) in finisher period. Both decreasing the protein level and protease inclusion significantly reduced concentration of ammonia in excreta ($P<0.05$). Supplementation of protease significantly decreased the total VFA concentration in ceca ($P<0.05$). The availability of protein was improved in low-protein diet treatments by adding protease ($P<0.001$). In conclusion, protease inclusion could improve the utilization of protein in low-protein diet, and reduce ammonia emission in broilers.

Keywords: protease, broilers, performance.

**Corresponding author: byu@dragon.nchu.edu.tw*

Introduction

Protein and amino acids are the most important but expensive in broiler diet. For the requirement of protein and amino acids, higher energy raw material (ex. plant oil) would be included in the formula therefore increased the cost. After protein digestion and absorption, the residue protein arrives in hindgut and turns it into the medium for microorganism. It would enhance the growth of pathogen bacteria which produce some harmful metabolites affecting the animal health. Ammonia is one of the harmful metabolite produced by bacteria in the gut, and the study had showed that if a lot of ammonia exists in the chicken house, it would increase the incidence of foot pad dermatitis and injure the lung on broilers (Oyetunde et al., 1978; Kjaer et al., 2006). Due to the lack of protease in young animal (Nitsan et al., 1991), protease supplement is needed. Supplementation of protease aims to hydrolyze the indigestible proteins and anti-nutritional factors, therefore improving the amino acids availability on broilers (Angel et al., 2011) and decreasing the ammonia nitrogen concentrations on weaned piglets (Wang et al., 2011). The purpose of this study was based on the low-protein and non-conventional diets by adding commercial protease to estimate the effect on growth performance and intestinal characterizes of broilers.

Materials and Methods

Broilers, management and sample collection

A total of 1,400 0-day old broilers (Ross 308) were divided to two batches. There were four replications in every batch, two males and females respectively. Broilers were randomly allocated into following treatments: T1 (maize-soybean meal diet); T2 (T1 diet decreasing 5% CP); T3 (non-conventional diet including DDGS, meat bone meal and feather meal); T4 (T3 diet decreasing 5% CP), and 0.05% protease was added in the T2, T3 and T4 to be the enzyme supplement groups. Feed consumption, feed conversion ratio (FCR), and body weight gain were recorded at day 10, 20 and 35. At day 35 after weighed, 12 chicks from each treatment were randomly selected and analyze the intestinal traits (concentration of ammonia and total volatile fatty acids (VFA)).

Apparent metabolizability trial

Six broilers (three males and three females) at 35 days were selected. After a 3-day adjustment period and fasted 1 day, faeces were collected separately during a period of 4 consecutive days. Diets and faeces were analyzed to calculate the metabolizability of dry matter, ash, crude protein and energy.

Results

Growth performance

The results showed that the based on different protein sources, T1 and T2 had the better weight gain and FCR than T3 and T4 in starter (0-10d) and grower (11-20d) period ($P<0.05$), but not occurred in finisher (21-35d) period. There was a significantly better FCR in normal protein diet than that of the low-protein diet ($P<0.05$) in finisher period.

Intestinal traits

The results of Dietary supplementation of protease on the concentration of ammonia and total VFA shows that both decreasing the protein level and protease inclusion significantly reduced concentration of ammonia in excreta ($P<0.05$). Supplementation of protease significantly decreased the total VFA concentration in ceca ($P<0.05$).

Apparent metabolizability

The high-protein diet treatments had higher ash, protein and energy metabolizability than low-protein diet ($P<0.05$). The availability of protein was improved in low-protein diet treatments by adding protease ($P<0.001$).

Conclusion

Protease inclusion could improve the utilization of protein in low-protein diet, and reduce ammonia emission in broilers.

References

- Angel, C. R., W. Saylor, S. L. Vieira and N. Ward, 2011. Effects of a monocomponent protease on performance and protein utilization in 7- to 22-day-old broiler chickens. *Poult. Sci.* 90: 2281-2286
- Dong, G., A. Zhou, F. Yang, K. Chen, K. Wang and D. Dao, 1996. Effect of dietary protein levels on the bacterial breakdown of protein in the large intestine, and diarrhoea in early weaned piglets. *Acta. Vet. Zootec. Sin.* 27: 293-302
- Kjaer, J. B., G. Su, B. L. Nielsen and P. Sørensen, 2006. Foot pad dermatitis and hock burn in broiler chickens and degree of inheritance. *Poult. Sci.* 85: 1342-1348
- Nitsan, Z., G. Ben-Avraham, Z. Zoref and I. Nir, 1991. Growth and development of the digestive organs and some enzymes in broiler chicks after hatching. *Br. Poult. Sci.* 32: 515-523
- Oyetunde, O. O., R. G. Thomson and H. C. Carlson, 1978. Aerosol exposure of ammonia, dust and *Escherichia coli* in broiler chickens. *Can. Vet. J.* 19: 187-193
- Wang D., X. S. Piao, Z. K. Zeng, T. Lu, Q. Zhang, P. F. Li, L. F. Xue and S. W. Kim, 2011. Effects of keratinase on performance, nutrient utilization, intestinal morphology, intestinal ecology and inflammatory response of weaned piglets fed diets with different levels of crude protein. *Asian-Aust. J. Anim. Sci.* 12: 1718-1728

The levels of TBARs in pellet fish feed mixed with different astaxanthin levels and packaging methods

Simawan¹, J., N. Senasri¹, N. Chumnanka¹ & T. Sreelawong¹

¹Department of Fisheries Science, Faculty of Natural Resources, Rajamangala University of Technology Isan, SakonNakhon Campus, SakonNakhon Province, 47160, Thailand.

Abstract

The objective of this experiment was to study the levels of TBARs in pellet fish feed mixed with different astaxanthin and packaging methods. This study was conducted using a 3x4 factorial experiments in completely randomized design with 3 replications, treatment contained 2 factors, factor A levels of astaxanthin were 0, 250 and 500 mg/kg of feed, factor B type of packaging were polyethylene bag, polyethylene vacuum packing bag, aluminium foil bag and aluminium foil vacuum packing bag, 100 g/bag samples were collected and storage at amphibian temperature. TBARs were measure after 3 months, the results showed that the level of TBARs was lowest at 0.0660 ± 0.0006 mg MDA/kg of feed ($P < 0.05$) when applying astaxanthin at 500 mg/kg of feed with aluminium foil vacuum packing bag. And the highest ($P < 0.05$) when using astaxanthin at 0 mg/kg of feed with polyethylene vacuum bag (0.0713 ± 0.0019 mg MDA/kg of feed). Therefore, based on this study, it could be concluded that supplementation of astaxanthin in pellet fish feed at 500 mg/kg of feed with aluminium foil vacuum packing bag could preserve the quality of ornamental pellet fish feed.

Keywords: Oxidation, astaxanthin, packaging, fish feed, storage

Corresponding author: jitra203@hotmail.com

Introduction

The deterioration of pellet fish feed might cause by several factors such as the package has been opened; food was exposed to oxygen or light, or the property of food packaging. The deterioration caused by the fat oxidation reaction in pellet fish feed results rancid, discoloration and loss of nutrients (Tongsiri, 2010). The rancidity of fat was driven by the reaction of polyunsaturated fatty acids and oxygen in the air which caused the rancidity in food (Crawley, 1993). The substances from food rancidity were peroxide, aldehyde and ketone (Earpin and Chamchumroon, 2007). Which were toxic to fish and thus resulting in uncontrollable body and hinder amount of antioxidant intake to normal level which lead to aquatic chronic disease, effecting to low growth rate and pale in aquatic animals (Supis, 1992).

Antioxidants or a substance inhibitor of oxidative obtained from synthetic were named as Butylated Hydroxyl Anisole (BHA), Butylated Hydroxyl Toluene (BHT), Tertiary Butyl Hydroquinone (TBHQ) and Propyl Gallate (PG) Earpin and Chamchumroon. (2009). These antioxidants were chemical substances used to be added in fish feed with fat added or high level of fat added to slow down fat oxidation Hernandez et al., 2014). Nevertheless, the over dose used of legal limit of antioxidant level may result in toxic in human and in animal (Siwaporn, 2003).

Astaxanthin, a substance in xanthophyll group and carotenoid family, can be found in both plants and animal, such as salmon, caviar, prawn shell and some types of microalgae

(*Haematococcus pluvialis*) EFSA Panel on Additives and Products or Substances used in Animal Feed, 2014.) From the study of the capacity of antioxidant in *Haematococcus pluvialis* found that this algae contains astaxanthin (Kobayashi, 1997). Astaxanthin and beta-carotene contains 10 times antioxidant more than other carotenoid (Miki, 1991). Tungsiri (2010) has applied the lycopene extracted from tomato with different intensity in fish feed and packed in different packaging and stored in temperature room with 25 ± 3 °C for 24 weeks. The result showed that the fish feed with lycopene 100 mg/kg stored in aluminium packaging contains TBARS minimum 0.33 ± 0.06 mg MDA/kg of feed. Lycopene from tomato is categorized in the same Carotenoid group as Astaxanthin. To store the Ornamental fish feed in the appropriate storage could reduce the antioxidant in fish feed (Jintasataporn et al., 2000).

Francis and Clydesdale (1975) reported that the cause of reduction of carotenoid in food was the oxidation. Therefore, the rate of oxidation depended on the exposed to oxygen, light, heat and the catalyst such as chitin and chitosan (Makkhun, 2003). Astaxanthin is a red, pink or orange carotenoid with the best qualification of antioxidant comparing to other type of carotenoid (Terao, 1989).

Materials and Methods

Ornamental fish feed contained astaxanthin as required (Plus Value Enterprise Co., Ltd) and packed in different packaging 100g/package. The experiment was conducted 3x4 factorial experiments in Completely Randomized Design: CRD with 3 repetitions. There were 2 factors in this study: 1. Ornamental fish feed mixed with 3 levels of astaxanthin (0, 250 and 500 mg/kg) 2. Packaging and 4 types of packaging method were polyethylene bag, polyethylene vacuum packing bag, aluminium foil bag and aluminium foil vacuum packing bag and stored in the room temperature for 3 months.

This experiment studied the changes of fish feed rancidity by analyzing the Thiaobarbituric acid reactive substance (TBARS) after the experiment by using the method A.O.A.C. (2000). This method measured the level of oxidation in food by using the absorption of samples of fish feed by using the TBARS. The TBARS substituted linear equation in Malondialdehyde: MDA shown in mg MDA/kg of feed. Then, The data gained used to analyze the Analysis of Variance and by using 3x4 factorial experiment in CRD to compare the different of mean of the experiment by Duncan's new multiple range test (DMRT) with reliability 95 % ($P < 0.05$). The data was analyzed by using SPSS statistics.

Result and Discussion

The results revealed that the average TBARS in ornamental fish feed in different level of astaxanthin presented in 3 levels ($P < 0.05$). The ornamental fish feed with astaxanthin 500 g/kg feed was found existing the lowest TBARS (0.067 ± 0.0010 mg MDA/kg of feed) and the ornamental fish feed with astaxanthin 250 and 0 mg/kg of feed showed that the TBARS was lower as can be seen in Table 1. Similar to Makkhun (2003) study the use of astaxanthin from prawn shell oxidation reaction frozen Fish Tilapia. The study found that astaxanthin from prawn shell 0.5-1.0 % (w/v) effected of slowing oxidation of frozen fish in good level and astaxanthin extract 0.5, 0.7 and 1.0% respectively TBARS of skin and flesh fillet increase in the lowest level.

Table 1. Effects of astaxanthin level different on the concentration of TBARS in fish feed.

Level of astaxanthin (mg/kg of feed)	TBARS value (mgMDA/kg of feed)	Cost of feed (Baht/kg)
0	0.0693±0.0023 ^a	75
250	0.0678±0.0011 ^b	100
500	0.0671±0.0010 ^c	125
P-value	0.000	

^{a-c} Means with different letters within a column of each group are significantly different at P<0.05

Comparing TBARS in ornamental fish feed which stored in different packaging and different storage methods found that the TBARS were different. The TBARS of ornamental fish feed kept in polyethylene bag and polyethylene vacuum packing bag were different. The polyethylene vacuum packing bag found the TBARS lower than (P<0.05) the polyethylene bag (Table 2) related Earpinand Chamchumroon (2009) reported that the more food surface is exposed to oxygen the quicker fat oxidation reaction would occur to the fish feed so that the TBARS in fish feed in polyethylene vacuum packing bag were lower than the TBARS in fish feed kept in polyethylene bag. Removal oxygen from the packaging was the way to reduce the spoilage of fish feed. On the other hand, there were several ways to reduce the spoilage of fish feed such as vacuum packaging (Morais et al., 2001), modified atmosphere packaging and oxygen scavenger (Bauernfeind, 1981).

Table 2. Effects of packaging and storage methods different on the concentration of TBARS in fish feed.

Type of package	TBARS value (mg MDA/kg of feed)
Polyethylene bag	0.0693±0.002 ^a
Polyethylene vacuum packing bag	0.0668±0.0005 ^b
Aluminium foil bag	0.0691±0.0015 ^a
Aluminium foil vacuum packing bag	0.0665±0.0006 ^b
P-value	0.000

^{a-b} Means with different letters within a column of each group are significantly different at P<0.05

From the studies of using astaxanthin, packaging, storage methods with different method TBARS in ornamental fish feed kept on room temperature for 3 months with 12 experiments (Table 3) found that Experiment 12 established the lowest TBARS which was significantly different from Experiment 1, 2, 3, 7 and 8 (P<0.05). This finding of this experiment is similar to the studies of Tongsir (2010) which found that the fish feed mixed lycopene 100 mg/kg and stored in aluminium foil bag in room temperature (25 – 30 °C) had the TBARS average minimum 0.33±0.06 mgMDA/kg of feed. On the other hand, the ornamental fish feed with no lycopene from tomato kept in aluminium foil vacuum packing bag has TBARS average maximum 1.47±0.00 mgMDA/kg of feed. Similarly, a study of Hernandez et al. (2014) reported that antioxidant extracted from natural and synthetic substances in fish feed 5 formula as follows; BD (fish feed with no antioxidant added), BHT (fish feed mixed Butylhydroxytoluene), RO (fish feed mixed Rosemary extract), TC (fish feed mixed essential oil of *Thymbracapitata*) and TZ (fish feed mixed essential oil of *Thymus zygis*). All 5 formula were kept in room temperature (20 – 28 °C) found that the fish feed with RO and BHT were able to inhibit antioxidant reaction in week 12 (TBARS = 9.9 and 8.1 mgMDA/kg of feed respectively) (P>0.05). This can be concluded that the storage of fish feed mixed rosemary extract can restrain the antioxidant reaction as well as the synthetic BHT. From this study revealed that the packaging and storage effected the antioxidant reaction to TBARS in fish feed as the vacuum packing was able to reduce the reaction between fish feed and oxygen (Bauernfeind, 1981). Moreover, storage fish feed in the opaque packaging was reduction of antioxidant reaction better translucent packaging (Morais et al., 2001).

Table 3. Effects of astaxanthin level, packaging and storage methods different on the concentration of TBARS in fish feed.

Experiment	Type of package and storage method	Level of astaxanthin (mg/kg of feed)	TBARS value (mgMDA/kg of feed)
1	Polyethylene bag	0	0.0713±0.0019 ^a
2		250	0.0687±0.0002 ^b
3		500	0.0677±0.0009 ^{bc}
4	Polyethylene vacuum packing bag	0	0.0671±0.0005 ^{cd}
5		250	0.0669±0.0008 ^{cd}
6		500	0.0665±0.0003 ^{cd}
7	Aluminium foil bag	0	0.0705±0.0004 ^a
8		250	0.0687±0.0004 ^b
9		500	0.0675±0.0012 ^{bcd}
10	Aluminium foil vacuum packing bag	0	0.0668±0.0006 ^{cd}
11		250	0.0667±0.0005 ^{cd}
12		500	0.0660±0.0006 ^d
P-value			0.008

^{a-d} Means with different letters within a column of each group are significantly different at P<0.05

Hence, ornamental fish feed with synthetic astaxanthin from 250 – 500 mg/kg of feed was able to detain antioxidant reaction in fish feed and the storage method of ornamental fish feed in aluminium foil vacuum packing bag and polyethylene vacuum packing bag were able to inhibit the rancidity especially ornamental fish feed with astaxanthin 500 mg/kg of feed packaged in aluminium foil vacuum packing bag which found the best way to inhibit oxidation reaction with cost 125 Baht/kg.

Conclusion and Suggestion

Ornamental fish feed with astaxanthin 500 mg/kg of feed presented the lowest level of TBARS (0.0671mgMDA/kg of feed) and packaged in aluminium foil vacuum packaging bag revealed that the TBARS was in the lowest level (0.0665mgMDA/kg of feed). The TBARS increased when the ornamental fish feed was packed in the Polyethylene bag and mixed astaxanthin in the small amount. The ornamental fish feed with astaxanthin extract 500 mg/kg of feed packed in aluminium foil vacuum packing bag found the TBARS was downward to lowest level (0.0660mgMDA/kg of feed). So that the mix of astaxanthin in ornament fish feed at amount 500 mg/kg of feed with packed in vacuum packaging was able to slow down the antioxidant reaction. However, this experiment studied the astaxanthin effected to the TBARS in ornamental fish feed. The next experiment should have studied the astaxanthin in higher level to reveal the clearer result.

Acknowledgement

This research was supported by grants from the national research council of Thailand, project of “Utilization of fairy shrimp as protein and carotenoid supplementary in dietary of Gold fish”.

Reference

- A.O.A.C. 1995. Official Methods of Analysis of the Association of the Official Analysis Chemists. Association of Official Analytical Chemists Arlington: USA.
- Bauernfeind, J.C. 1981. Carotenoids as colorants and vitamin A precursors. Academic Press. New York.19-41.

- Crawley, H. 1993. Natural occurrence of vitamins in food. The technology of vitamins in food. Blackie Academic and professional. Glasgow. England.
- Earpin C. and P. Chamchumroon. 2009. Effect of temperature and time on antioxidant activity of dried onion. *Agricultural Sci. J.* 38(6) : 139-142. [Online] Available from http://www.crdc.kmutt.ac.th/agr/agr_1_99-150.html. (in Thai)
- EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP). 2014. Scientific Opinion on the safety and efficacy of synthetic astaxanthin as feed additive for salmon and trout, other fish, ornamental fish, crustaceans and ornamental birds. European Food Safety Authority. 12(6):3724.
- Francis, F.J. and Clydesdale F.M. 1975. Food Colorimetry: Theory and applications. Westport Conn, AVI.
- Hernández, A. GarcíaGarcía, B. Jordán, M.J. and Hernández, M.D. 2014. Natural antioxidants in extruded fish feed: Protection at different storage temperatures. *Journal of Animal Feed Science and Technology*. [Online] Available from <http://www.sciencedirect.com/>[2014, August 1]
- Jintarataporn O., S. Chumkam and O. Triwatanon. 2000. Shelf Life of Ornamental Fish Feed. Proceedings of the 38th Kasetsart University Annual Conference: Fisheries and Science. Department of Aquaculture, Kasetsart University. [Online] Available from http://kukr.lib.ku.ac.th/ku_proceed/KUCON/search_detail/result/7_3_4_1/ [2014, August 1] (in Thai)
- Kobayashi, M. Kurimura, Y. Sakamoto Y. and Tsuji, Y. 1997. Selective extraction of astaxanthin and chlorophyll from the green alga *Haematococcus pluvialis*. *Biotechnology Techniques*. 11(9): 657-660.
- Makkhun S. 2003. Extraction and effect of astaxanthin from shrimp shell on the color and TBA values changing of chilled tambora fish (*Oreochromis* sp.). Master's Thesis. Naresuan University, Phayao. [Online] Available from <http://www.lib.ku.ac.th/KUthesis/SakunkhunMAK/SakunkhunMAKAll.pdf> (in Thai)
- Miki, Y. 1991. Biological functions and activities of animal carotenoids. *Pure Applied Chemistry* 63: 141-146.
- Morais, F., Kuhnk, K., Steward D.H., Barber, J., Brudvig, G.W. and Nixon, P.J. 2001. Photosynthetic water oxidation in cytochrome b₅₅₉ mutants containing a disrupted heme-binding pocket. *Journal of Biological Chemistry*. 276: 31986-31993.
- Tongsiri, K. 2010. Utilization of lycopene from tomato (*Lycopersicon esculentum* Mill) as natural antioxidant in fancy carp (*Cyprinus carpio* Linn.) feed. Master's Thesis. King Mongkut's Institute of Technology Ladkrabang. Bangkok. 89. (in Thai)
- Terao J. 1989. Antioxidant activity of β -carotene-related carotenoids in solution *Lipid*. 24: 659-661.
- Siwaporn, S. 2003. Food additive. National Agricultural Extension and Training Center. Kasetsart University Kamphaeng Sean Campus. Nakhon Pathom. 380. (in Thai)
- Supis, T. 1992. Importance of fat in aquatic animal diet. *Journal of Thai Fisheries Gazette*. 45 (4):943- 950. (in Thai)

Standardized total tract digestibility of phosphorus of various meal diets as protein source in growing-finishing pigs

Park¹, S. K. & M. J. Kim²

¹Sejong University, 209, Neungdong-ro, Gwangjin-gu, Seoul, 143-747, Korea, ²National Institute of Animal Science, Suwon, 441-706, Korea

Abstract

This study was conducted to estimate apparent total tract digestibility (ATTD), standardized total tract digestibility (STTD) of phosphorus (P), ATTD of crude protein (CP) and dry matter (DM) in various meals fed to growing-finishing pigs. Twelve barrows (initial BW, 71.7±17.0 kg) were allocated in metabolism cages. The experimental design was a 12×8 Latin square with 12 dietary treatments and 8 replication periods. The diets were based on sesame meal (SM), dehulled soybean meal from Korea (KD-SBM), dehulled soybean meal from India (ID-SBM), soybean meal from Korea (K-SBM), corn high protein distillers dried grains (HPDDG), perilla meal (PEM), canola meal (CAM), copra meal (COM), corn germ meal (CGM), palm kernel meal (PKM), cassava root distillers dried grains (CRDDG), respectively, and P-free diet for measuring basal endogenous losses of P. ATTD of DM was higher ($P<0.001$) in the K-, KD- and ID-SBM compared with SM, PEM, PKM and CRDDG. ATTD of CP in the K-, KD- and ID-SBM was greater ($P<0.001$) than that in SM, PEM, COM and CRDDG. Nitrogen retention was increased ($P<0.001$) in the K-, KD- and ID-SBM than the SM, PEM and HPDDG. ATTD of P was higher ($P<0.001$) in the CGM than the SM, PEM, CRDDG and KD-SBM. STTD of P was greater ($P<0.001$) in the HPDDG and CGM compared with the SM, KD-SBM, PEM and CAM. In conclusion, data from our current study can be utilized as a platform for formulating the diets with precise P utilization in various meals for pigs.

Keywords: *various meals, standardized total tract digestibility, phosphorus, crude protein, pigs*

Corresponding author: sungkwonpark@sejong.ac.kr

Introduction

Growth performance is closely related to the absorption of nutrients and their digestibility in animals. Providing adequate nutrient which meet animals requirement accounts for desired or expected performance and health of animals. Feeding diet is the only nutrient source for farm animals. Therefore, precise estimation of nutrition value and digestibility of feed ingredients is very important for animal livestock. Feed ingredients such as meals have been utilized as a protein source for swine diet. Among them, soybean meal accounts for about two-thirds of the world's high protein animal feed, followed by cottonseed and rapeseed meal, which together account for less than 20%. Price of soybean meal has been moving to record highs in recent years due to its increased demands with decreased production is a representative protein feed ingredient showing a better digestibility than other feed stuffs (NRC, 2012). However, price of soybean meal is recently raised due to increased demand and decreased supply. Studies have evaluated the feed value of canola, rapeseed, cottonseed, perilla, sesame, palm kernel and copra meal as a replacement of soybean meal (Kim & Kwon, 1994; Li et al., 2000; Kim et al., 2001; Babiker, 2012). However, there was less data available for digestibility including apparent total

tract digestibility (ATTD) and standardized total tract digestibility (STTD) of crude protein (CP) and phosphorus (P) for these feed ingredients in NRC (2012). Therefore, the purpose of our current study was to evaluate the ATTD of CP and STTD of P of various feed ingredients including sesame meal, soybean meal, dehulled soybean meal, canola meal, corn high protein distillers dried grains, perilla meal, copra meal, corn germ meal, palm kernel meal, cassava root distillers dried grains in growing-finishing pigs

Materials and methods

Diets, Animals, and Experimental Design: The experimental protocols describing the management and care of animals were reviewed and approved by the Animal Care and Use National Institute of Animal Science. Twelve barrows with initial of BW of 71.7 ± 17.0 kg were prepared from local farm and allocated in individual metabolism cage. Experimental design was a 12×8 Latin squares with 12 diet treatments and 8 periods. Nutrient compositions:

Table 1. Nutrient composition of coproduct ingredients.

Composition ²	SM	KD-	ID-	K-	HP	PEM	CAM	COM	CGM	PKM	CR
		SBM	SBM	SBM	DDG						DDG
Moisture, %	4.37	11.9	11.2	11.5	9.58	9.59	9.41	10.7	6.23	11.4	7.87
GE, kcal/kg	4,973	4,520	4,494	4,575	5,285	4,601	4,329	4,169	4,888	4,574	4,049
CP, %	48.6	46.7	39.6	47.1	38.3	44.5	36.8	21.7	21.9	15.7	17.8
EE, %	3.69	2.03	1.96	1.99	4.11	0.84	1.66	1.50	8.42	6.52	2.67
CF, %	4.32	5.13	5.40	5.95	9.84	21.1	9.70	25.4	10.3	25.6	26.1
Ash, %	5.84	6.57	6.55	6.03	1.38	9.01	9.92	6.58	2.31	4.62	15.5
NDF, %	22.8	8.70	11.4	10.4	39.6	43.2	22.4	54.1	49.3	56.1	58.7
ADF, %	11.8	4.54	5.32	5.70	12.1	24.7	14.1	35.4	12.3	36.1	36.0
Ca, %	0.34	0.56	0.61	0.39	0.02	1.39	1.07	0.21	0.12	0.39	0.75
P, %	0.63	0.57	0.54	0.63	0.24	1.28	0.99	0.56	0.53	0.54	0.23

Feeding and Sample Collection: The daily amount of feed provided to the pigs was calculated as 2.5 times the estimated requirement for maintenance energy (i.e., 106 kcal ME per $\text{kg}^{0.75}$; NRC, 1998) and divided into 2 equal meals at 09:00 and 17:00. Feces were collected according to the marker-to-marker approach (Adeola, 2001). Urine was collected daily in buckets to contain 50 mL of 4N sulfuric acid and during collection periods.

Sample Analyses and Data Processing: Fecal samples were dried at 60 °C in a forced-air drying oven and ground. All samples, which are ingredients, diets, feces and urine, were dried in a forced-air drying oven at 135 °C for 2 h (method 930.15; AOAC, 2005) and were analyzed for crude protein (method 990.03; AOAC, 2005). The ingredients, experimental diets were analyzed for gross energy (Model C2000, IKA, Germany), ether extract (method 920.39; AOAC, 2005), crude fiber (method 978.10; AOAC, 2005), ash (method 942.05; AOAC, 2005), acid detergent fiber (method 973.18; AOAC, 2005) and neutral detergent fiber (Holst, 1973). The Ca concentrations in the ingredients, diets, and feces were analyzed using an atomic absorption

spectrophotometer (method 978.02; AOAC, 2005; Perkin Elmer 3300, Perkin Elmer, USA). The P concentrations in the ingredients, diets, and feces were analyzed using a spectrophotometer (method 946.06; AOAC, 2005; Optizen 2120UV, Mecasys, Republic of Korea). The apparent total tract digestibility (ATTD) and standardized total tract digestibility (STTD) of P and the endogenous P loss in each diet were calculated as previously described (Almeida & Stein, 2010). Values for ATTD of CP and DM, expressed as a percentage of their intake, were calculated by subtracting DP and DM excreted in the feces from intake, dividing by intake, and multiplying the result by 100. All data were analyzed using the MIXED procedure (SAS Inst. Inc., Cary, NC). Homogeneity of the variances among treatments was confirmed using the UNIVARIATE procedure and this procedure was also used to test for outliers, but no outliers were identified. The model included diet as the fixed effect and replicate as a random effect. Least squares means (LSMEANS) were calculated and the means were separated using the PDIFF option with the Tukey's adjustment. The pig was the experimental unit for all analyses and an alpha level of 0.05 was used to assess significance among means.

Results

ATTD of CP in various co-products fed to pigs was shown in Table 5. Difference of DM intake among ingredients was not observed, whereas intake of N in KD- and K-SBM was greater ($P < 0.001$) than that of ID-SBM, PKM and HPDDG. The output of feces in KD-, ID- and K-SBM (SBM groups) compared with SM, PEM, COM and CRDDG was reduced ($P < 0.001$). Fecal DM output was lower ($P < 0.001$) in KD-, ID- and K-SBM (SBM groups) compared with SM, PEM, COM, PKM and CRDDG. Excretion of N in SBM groups, HPDDG and CAM was decreased ($P < 0.001$) than that of SM, PEM and CRDDG. ATTD of DM from SBM groups was higher ($P < 0.001$) than that of SM, PEM, PKM and CRDDG. ATTD of CP was greater ($P < 0.001$) in SBM groups compared with SM, PEM, COM and CRDDG and also was higher ($P < 0.001$) in HPDDG and CAM than that of SM, PEM and CRDDG. Significance among ingredients of the total urine output was exited ($P = 0.031$), but, by Tukey's adjustment, was not presented among them. Difference of N output in urine was not observed among ingredients, however, N retention of SBM groups was greater ($P < 0.001$) than that of SM, HPDDG and PEM.

The effects of various feed ingredients on STTP and ATTD of P in growing finishing pigs were presented in Table 6. Intake of Ca among ingredients was as follows that, in order of high value, SM, PEM, CAM, KD-SBM, K-SBM, CRDDG, ID- SBM, COM, CGM, PKM and HPDDG. Intake of P of experimental diet fed to pig was showed by serially high value that SM, PEM, CAM, KD-SBM, K-SBM, CGM, ID-SBM, COM, PKM, CRDDG and HPDDG. Intake of Ca and P in SM, PEM and CAM was greater ($P < 0.001$) than those of other ingredients, whereas, in numerical aspect, HPDDG compared with other ingredients was lowest in both. However, excretion of Ca and P in feces from pigs fed experiment diets with ingredients was greater ($P < 0.001$) in SM and PEM compared with other ingredients. In contrast, the output of fecal Ca and P in HPDDG tended decreased than those of other ingredient. ATTD of Ca in SM compared with other ingredients was lowest ($P < 0.001$), and the value of PEM was lower ($P < 0.001$) than that of SBM groups, HPDDG, COM, CGM and CRDDG. ATTD of P was higher ($P < 0.001$) in CGM compared with SM, KD-SBM, PEM and CRDDG. STTD of P in HPDDG and CGM was greater ($P < 0.001$) than that of SM, KD-SBM, PEM and CAM, and the value of K-SBM and COM was higher ($P < 0.001$) than that of PEM. STTD-P was higher ($P < 0.001$) in SM and PEM compared with other ingredients except for CAM, whereas the value of HPDDG and CRDDG was lower ($P < 0.001$) than that of other ingredients.

Table 2. Apparent total tract digestibility (ATTD) of Ca and standardized total tract digestibility (STTD) of P in experimental diets.

Item	SM	KD-SBM	ID-SBM	K-SBM	HP DDG	PEM	CAM	COM	CGM	PKM	CR DDG	SEM ⁴	P-value
Diet intake, kg	6.70	6.77	6.56	6.70	6.14	6.65	6.59	6.86	7.18	6.53	6.44	0.31	0.237
Diet DM ⁵ , %	92.4	90.6	90.8	90.7	91.6	91.1	91.9	91.5	92.1	90.9	91.9	-	-
Diet N, %	2.90	3.01	2.51	3.16	2.36	2.84	2.67	2.58	2.57	2.57	2.88	-	-
DM intake, kg	6.20	6.13	5.95	6.08	5.63	6.06	6.05	6.28	6.61	5.93	5.91	0.29	0.188
N intake, g	195 ^{abc}	204 ^{ab}	165 ^{de}	212 ^a	145 ^e	189 ^{abcd}	175 ^{bcd}	177 ^{bcd}	185 ^{abcd}	168 ^{cde}	185 ^{abcd}	9	< 0.001
Fecal output, kg	0.74 ^{ab}	0.25 ^{ef}	0.26 ^{def}	0.18 ^f	0.45 ^{cdef}	0.90 ^a	0.47 ^{bcd}	0.55 ^{bc}	0.45 ^{cdef}	0.54 ^{bcd}	0.90 ^a	0.07	< 0.001
Fecal DM, %	93.1 ^{cd}	91.4 ^{fg}	92.6 ^{de}	91.3 ^g	94.4 ^{ab}	93.6 ^{bc}	92.1 ^{ef}	93.4 ^{cd}	93.9 ^{abc}	93.5 ^{bc}	94.7 ^a	0.3	< 0.001
Fecal N, %	7.54 ^a	4.30 ^d	3.91 ^{ef}	4.24 ^{de}	3.03 ^g	5.34 ^b	3.54 ^f	5.20 ^{bc}	4.86 ^c	3.69 ^f	3.82 ^f	0.10	< 0.001
Fecal DM output, kg	0.69 ^{ab}	0.23 ^{de}	0.24 ^{de}	0.17 ^e	0.43 ^{bcd}	0.85 ^a	0.43 ^{bcd}	0.51 ^{bc}	0.42 ^{cde}	0.51 ^{bc}	0.85 ^a	0.06	< 0.001
Fecal N output, g	56.4 ^a	10.8 ^d	10.1 ^d	8.00 ^d	13.2 ^d	48.6 ^a	16.9 ^{cd}	28.7 ^{bc}	21.7 ^{bcd}	20.2 ^{bcd}	33.8 ^b	3.28	< 0.001
ATTD of DM, %	89.0 ^{ef}	96.2 ^{ab}	95.9 ^{abc}	97.3 ^a	92.2 ^{bcd}	86.0 ^f	92.8 ^{bcd}	92.0 ^{cde}	93.5 ^{abcd}	91.8 ^{de}	85.4 ^f	0.9	< 0.001
ATTD of CP ⁶ , %	71.6 ^e	94.6 ^{ab}	93.8 ^{ab}	96.3 ^a	90.4 ^{abc}	74.2 ^e	90.4 ^{abc}	84.0 ^{cd}	88.0 ^{bcd}	88.5 ^{bc}	81.4 ^d	1.6	< 0.001
Urine output, kg	10.49 ^a	21.72 ^a	19.85 ^a	20.11 ^a	20.42 ^a	16.44 ^a	16.93 ^a	21.33 ^a	17.03 ^a	11.58 ^a	8.19 ^a	3.54	0.031
Urinary N, %	0.87 ^{ab}	0.42 ^b	0.35 ^b	0.51 ^b	0.53 ^b	0.67 ^b	0.53 ^b	0.52 ^b	0.59 ^b	0.92 ^{ab}	1.45 ^a	0.14	< 0.001
Urinary N output, g	69.7	72.5	51.4	76.5	76.0	73.9	59.2	63.1	80.5	66.5	66.4	7.2	0.102
N retention ⁷ , %	36.4 ^c	58.7 ^a	61.8 ^a	60.1 ^a	37.1 ^{bc}	34.8 ^c	56.3 ^{ab}	48.0 ^{abc}	44.3 ^{abc}	50.2 ^{abc}	47.8 ^{abc}	4.3	< 0.001

^{a,b,c,d,e,f,g}Means within a row without a common superscript letter differ ($P < 0.05$).

References

- Adeola, O. 2001. Digestion and balance techniques in pigs. In: Swine Nutrition. A. J. Lewis and L. L. Southern, ed. CRC press, Washington, DC, p. 903-916.
- Almeida, F. N., and H. H. Stein. 2010. Performance and phosphorus balance of pigs fed diets formulated on the basis of values for standardized total tract digestibility of phosphorus. *J. Anim.Sci.* 88: 2968-2977.
- Holst, D. 1973. Holst filtration apparatus for Van Soest detergent fiber analysis. *J. AOAC* 56: 1352-1356.
- Kim, B. G. et al. 2001. Effect of partial replacement of soybean meal with palm kernel meal and copra meal on growth performance, nutrient digestibility and carcass characteristics of finishing pigs. *Asian-Aust.J. Anim. Sci.*14: 821-830.
- Kim, H. M. J., and S. K. C. Kwon. 1994. Growth performance and amino acid digestibilities affected by various protein sources in growing-finishing pigs. *Asian-Aust.J. Anim. Sci. (AJAS)* 7: 537-546.
- Li, D. et al. 2000. Performance of growing-finishing pigs fed sesame meal supplemented diets formulated amino acid digestibilities determined by the regression technique. *Asian-Aust.J. Anim. Sci.(AJAS)* 13: 213-219.
- NRC. 2012. Nutrient Requirements of Swine. 11th rev.ed. ed. Natl. Acad. Press, Washington, DC.

Application of feed technology of quail (*Coturnix-coturnix Japonica*) using waste of Skipjack (*Katsuwonus pelamis*)

Najoan¹, M., A. Makalew², F.N. Sompie¹, M.A.V. Manese², B. Bagau¹ & M.T. Massie²

¹Department of Animal Nutrition, Faculty of Animal Husbandry, University of Sam Ratulangi, Jl. Campus Bahu Kleak Manado 95115, North Sulawesi, Indonesia

²Department of Social Economics, Faculty of Animal Husbandry, University of Sam Ratulangi, Jl. Campus Bahu Kleak Manado 95115, North Sulawesi, Indonesia

Abstract

Livestock of quail (*Coturnix-coturnix Japonica*) can be cultivated with not require a large area. Quail livestock enterprises have been introduced to the housewife, who joined the group of “dasa wisma”, with the aim to improve their welfare. Quail livestock enterprises will succeed one of which depends on the feed. The problem of knowledge housewife group "dasa wisma" is low, on the use of local resources in quail feed. Based on these problems, it has made the application of technology with the goal for utilization of skipjack waste in quail feed. Method of application is through extension and training. Housewives as a member “dasa wisma”, had trained preparing rations by utilizing local resources. One of the feed material used is meal from waste of skipjack. Fish waste is cheaper, than the meal of fish that must exist in the preparation of rations quail. Based on the results of the activities that have been carried out can be concluded that the application of technology by adding waste skipjack in rations can reduce feed costs. Suggestions submitted are necessary assistance both from local government and universities for the development of livestock quail by utilizing local resources.

Keywords: quail, technology, feed, waste of skipjack

Introduction

Quail (*Coturnix-coturnix Japonica*) are cattle that can be cultivated without requiring large land. Quail farms can be used as permanent or part-time businesses, which provide additional income for businesses (Anugrah, 2009). Enterprises of quail can be the people's choice in order to increase their income (Amin, 2011). Cultivation quail enough potential as one of the business to rural communities.

Constraints in the development of quail, is the unavailability of seeds of quail, commercially as well as livestock chicken (Kaharuddin et al., 2008). Quail livestock enterprises have been introduced to the housewives who are members of the group "dasa wisma", with the aim to improve their welfare. The main obstacle, the efforts are quail feed costs tend to increase. These conditions indicate the need for a solution, so that livestock quail can be developed and profitable for entrepreneurs. Quail cattle business will be successful, one of which depends on the feed. However, the use of feed for livestock quail manufacturer results, which are expensive, may cause losses for farmers. The problem of knowledge housewives group "dasa wisma" is still low on the use of local resources in feed quail. Based on these problems it has been done the application of technology, with the aim of utilization of waste skipjack in the quail rations.

Materials and Methods

Methods of research that has been done is a survey method to housewives who are members of the group "dasa homestead". Introduction of technology has done for business quail, belonging to members of the group "dasa wisma". Methods of application of the technology that has been done is a method of extension and training. Housewives member "dasa wisma" have been trained arrange ration, utilizing local resources. One of the feed materials used is a waste meal of skipjack.

Results and Discussion

Livestock raising which quickly and economically to produce meat and eggs are livestock quail (Djaya, 2010). Housewives group "dasa wisma" have been trained to mix ration of livestock quail by utilizing local resources (meal of skipjack fish waste). The purpose of this activity is that the cost of livestock feed quail cheaper than if housewives buying finished feed, in food shops of Livestock. Waste of skipjack that production is very much in North Sulawesi, can be used as fish meal. The waters of North Sulawesi and Irian Jaya are an area of considerable potential for the production of skipjack (*Kahsuwonus pelaris*, L) including fish "tropical tuna" (Blackborn, 1985, quoted from Kaseger, 1986). After passing through the processing, skipjack meat, and the rest is waste. Waste of skipjack consists of: head, gills, tail, fins and innards. These sections coupled with the heart and the air bubbles non edible portion (Winarno et al., 1989), but can be utilized as an adjunct in animal feed.

Waste of skip jack contains nutrients such as 57.50% crude protein, 10-20% fat, 1.89% crude fiber, calcium 6.40%, 2.84% phosphorus and the metabolic energy 2779 Kcal/kg (Najoan, 1991). These data indicate that the skipjack waste has nutritional value is quite good, and can be used as a mixture in the ration. The results of the Najoan (1991) showed that the meal of skipjack fish waste to 15% level in the ration, not to move the odor in egg sand meat. Utilization of waste is carried out as an effort to reduce the cost of feed, to increase the income housewives group members. Fish waste is cheaper than fish meal should be in the preparation of rations quail.

Quail rations that had been developed after the adoption of technology, consisting of: corn (52%), fine bran (13%), coconut cake (15%) and flour from of skipjack waste (20%). The price of fish meal is currently about Rp 11,000 per kg, while the fish meal is derived from skipjack waste, costs only around Rp 5,000/kg. The price of corn Rp 5,000/kg, fine bran Rp 3,000/kg, coconut cake Rp 11,000/kg. Based on this ration composition, then the use of fish meal derived from fish waste skipjack caused feed cost can be reduced up to 17.54 percent. These conditions have benefited housewives group "dasa wisma".

Quail live stock productivity is determined by several factors, including rations. Feed is one of the factors that should be a major concern, given the cost of feed is 60-70% of the total cost of production. Even the cost of feed in the poultry business ranges between 70-80 percent (Sumekar et al., 2012). According to Anugrah (2009), nationally the percentage of ration needs, in units of the cycle of livestock business, reaching an average of 79.84%. Ration is a mixture of several ingredients of feed given to of livestock to meet the needs for one day (Supratman & Setiyatwan, 2012). According Achman et al. (2011), the feed given, said to be efficient, if such feed can be optimally consumed by of livestock quail. Although, the highest feed consumption by of livestock quail indicated that the highest weight (Kaharuddin & Kusdiyah, 2006).

Conclusions and Suggestions

Based on the results of the activities that have been carried out can be concluded that the application of technology by adding waste skipjack in rations can reduce feed costs. Suggestions submitted are necessary assistance both from local government and universities for the development of livestock quail by utilizing local resources.

References

- Achmanu., Muharlieni., and Salaby, 2011. Influence of cage floor (meetings and tenuous) and the male-female balance on feed consumption, egg weight, feed conversion and egg shell thickness in quail. *J. Livestock Tropika*, 12: 1-14.
- Amin, L, 2011. The effect of cumin (*Cuminum cyminum*) in feed for quail egg production. *Journal Agrisains*, 2: 29-39.
- Anugrah, I.S., I. Sadikin, and W.K. Sejati, 2009. Institutional policy of the traditional poultry business as a source of rural household economy the case of quail farmers Yogyakarta. *J. Agricultural Policy Analysis*, 7: 249-267.
- Djaya, M.S, 2010. The influence of the use of blood meal in the ration on performance of quail. *Media Science*, 2: 142-145.
- Kaharuddin, D., and Kususiya, 2006. Performance of qualitative and quantitative properties of quail from Yogyakarta, Padang and Bengkulu. *Journal Sain Animal Husbandry Indonesia*, 1: 21-25.
- Kaharuddin, D., Kususiya and Deva, 2008. Performance of growing quail (*Coturnix-Coturnix japonica*) white and brown. *Journal of Animal Husbandry Science Indonesia*, 3: 1-4.
- Kaseger, B.E. 1986., Silage of Skipjack (*Katsuwonus pelamis* L). Thesis. Faculty of Graduate Studies. Bogor Agricultural Institute, Bogor.
- Najoan, M. 1991., The effect of granting for Various Level Meal of Waste Skipjack (*Katsuwonus pelamis* L) for Performance of quail (*Coturnix Coturnix japonica*) During the First Five Months. Dissertation. Faculty of Graduate Studies UNPAD, Bandung.
- Sumekar, W., Isbandi, U. Atmomarsono and I. Susilowati. 2012. Level of empowerment Farmers Ducks for Environmental Access Businesses in Brebes. Proceedings of the National Seminar on Sustainable Animal Husbandry 4, Farmers Agribusiness Innovation for Food Security, Faculty of Animal Husbandry, Padjadjaran University. Bandung. pp 654
- Supratman, H and H. Setiyatwan. 2012. The Effect of Energy and Protein Balance Rations, for Performance of Broiler Chickens. Proceedings of the National Seminar on Sustainable Animal Husbandry 4, Farmers Agribusiness Innovation for Food Security. Faculty of Animal Husbandry, Padjadjaran University. Bandung. pp 654
- Winarno, F.G. A.F.S. Boediman, T. Silitonga and B. Soewardi. 1989., Waste of Agricultural Products. Monograph Office Men Mud Increased Food Production, Jakarta.

Quality of protein concentrate from *Jatropha curcas* seed cake produced by chemical and biological processing

Titin Widiyastuti*, Mohandas Indrajai & Tri Rahardjo Sutardi

Feedstuff Laboratory, Faculty of Animal Science
Jenderal Soedirman University, Purwokerto, 62-53122 Indonesia

Abstract

The objectives of this study were to optimize the quality of protein concentrate from *Jatropha curcas* seed cake (JSC) that obtained by chemical precipitation and fermentation using lactic acid bacteria. This experimental research used Completely Randomized Design. The treatments were applied to *Jatropha curcas* seed cake with three time repetition. Six treatments were performed, which consist of: R0 (raw *Jatropha curcas* seed cake without treatment), R1 (Protein concentrate precipitation without fermentation), R2 (fermentation mediated by *Lactobacillus acidophilus*), R3 (fermentation mediated by *Bifidobacterium spp*, R4 (*Jatropha curcas* seed cake precipitation/R1 continued by *Lactobacillus acidophilus* fermentation), R5 (*Jatropha curcas* seed cake precipitation/R1 continued by *Bifidobacterium spp* fermentation). The observed variables were: crude protein, protein solubility, antinutritive compound (lectins, phorbol ester, and antitrypsin). The result showed that precipitation and fermentation increase protein content of *Jatropha curcas* with 32.79% to 48.21% for the control and fermentation, respectively. Fermented protein concentrate of JSC (R4, R5) showed a higher solubility than protein concentrate without fermentation (R1). Protein concentrate produced by precipitation and fermentation using *Lactobacillus acidophilus* (R4) showed the lowest antinutrient content (lectins, antitrypsin and phorbol ester). Based on the result, the conclusion is protein concentrate produced by precipitation and fermentation using *Lactobacillus acidophilus* (R4) showed the best quality in terms of nutrient and antinutritive content.

Keywords: *Jatropha curcas*, *L. acidophilus*, *Bifidobacterium spp*, protein concentrate

*Corresponding author: dyast72@yahoo.com

Introduction

The availability of high quality of feed is an important factors and become a major problem in Indonesia's poultry industry, especially because the availability of feed ingredient as a protein source depending on import. These problems require concrete solutions in order to support the sustainability of poultry industry. One of the solution is exploring the local feed ingredients. An alternative protein resources in feed is agro-industry wastes, such as *Jatropha* seed cake. *Jatropha* seed cake has not been use as component of animal feed whereas its potential in term nutrient composition. Toxic varieties of *Jatropha* bean (based on dry weight) contain 56.4% of crude protein whereas non-toxic varieties has 63.8% of crude protein. *Jatropha* seed cake is potential to be used as feed compounds, due to their high nutrient composition and amino acid content. But there is an obstacles in *Jatropha* seed cake utilization, since it contain anti-nutritive factors i.e. phorbol esters, trypsin inhibitors, lectins and phytate (Makkar *et al.*, 2008). Phorbol esters (PEs) are the major impediment to the wide commercial use of *Jatropha* meal as a feedstock. During extraction of oil from *Jatropha* seeds, 70 - 75% of PEs goes with the oil, and

25 - 30% remains strongly bounded to the matrix of seed meal. Due to the high toxicity of PEs, seed cake cannot be used as animal feeds without detoxification Gogoi *et al.* (2014). We have to reduce or eliminate antinutritive factors inside jatropha seed cake before it can be used as animal feed. Various detoxification methods were widely used to eliminate antinutritive factors of seed cake i.e. addition of sodium butyrate on feed (Arnouts and Vandendriessche, 2007), heat and chemical treatments (Herera *et al.*, 2007; Chivandi, 2006) alkaline precipitation (Makkar *et al.*, 2008), lactic acid bacteria mediated fermentation (*Lactobacillus* spp and *Bifidobacterium* spp) and addition of various saccharides (Widiyastuti *et al.*, 2013) More over, Widiyastuti *et al.* (2014) reported the added value of Jatropha cultivation can be increased by optimizing Jatropha seed cake as complete feed. Addition of 12% of fermented jatropha seed cake in rabbit feed, has no effect on blood profile of post weaned Rex rabbits. Previous studies proved that jatropha seed cake, could not increase farm productivity. This indicates that high protein content in seed cake could not be fully utilized. This provide opportunity to develop an appropriate detoxification technology. Chemical and physical processes leaved antinutritional factors, however combination of chemical and biological techniques is expected to rise protein content and reduce antinutritive factors in seed oil cake. The objective of this study is to improve the quality of protein concentrate from jatropha seed cake as feed component through precipitation and fermentation using lactic acid bacteria (*Lactobacillus* spp and *Bifidobacterium* spp).

Materials and methods

Precipitation process

Precipitation process according to Makkar *et al.* (2008) modified by Widiyastuti *et al.* (2012).

Fermentation process

Fermentation for R2 and R3: 500 g jatropha seed cake, mixed with 5 % molasses as a carbohydrate source and 500 ml of water and then sterilized by autoclaving at 121°C and a pressure of 1 atm for 15 minutes. Substrate then inoculated with *Lactobacillus acidophilus* (R2) and *Bifidobacterium* spp (R3) and incubated at 37°C for 3 x 24 hours. Fermentation for R4 and R5: 200 grams of protein concentrate from jatropha seed cake, mixed with 5 % molasses then sterilized by autoclaving at 121°C and a pressure of 1 atm for 15 minutes. Substrate then inoculated with *Lactobacillus acidophilus* (R4) and *Bifidobacterium* spp (R5) and incubated at 37°C for 3 x 24 hours. Post fermented, sampel were drying at 50-60°C and taken for analysis of nutrient content (proximat analysis), protein solubility, and antinutritive content (lectins, phorbol ester and antitrypsin).

Nutrient assay

Crude protein was measured according to AOAC (1990), protein solubility was measured before and after incubation in pepsin solution (pepsin solution concentration was 0.2% (activity 1:10,000).

Antinutrients assay

Trypsin inhibitor activity was determined according to Smith *et al.* (1980). Lectin was determined by using hemagglutination assay as described by Makkar *et al.* (1999), phorbol ester assay according to Makkar *et al.* (2007).

Statistical methods

The study design used was completely randomized design (CRD) and continued by contrast orthogonal. Treatments consist of : R0 (raw jatropha seed cake without treatment), R1 (Protein concentrate precipitation without fermentation), R2 (fermentation mediated by *Lactobacillus acidophilus*), R3 (fermentation mediated by *Bifidobacterium* spp, R4 (jatropha seed cake precipitation and *Lactobacillus acidophilus* fermentation), R5 (jatropha seed cake precipitation and *Bifidobacterium* spp-mediated fermentation). Each treatment was repeated for 3 times. The

variables measured were crude protein, protein solubility, anti-nutritive content (lectins, phorbol ester and anti-trypsin). Data were analysed by analysis of variance using Microsoft Excel ver. 2007.

Results and discussion

Protein content and protein solubility- of concentrate protein of Jatropha seed cake

Nutrient content of seed oilcake control (without treatments) were highly significant ($P < 0.01$) compared to treated seed oilcake (R1 through R5, except R2). Optimizing protein seed cake by precipitation and fermentation methods showed significant results obtained high levels of a protein that is from 32.79% (R0) to 48.21% (R1) or its increased as many as 15.42%. But then a decline after seed oilcake and protein concentrate fermented by *Lactobacillus acidophilus* and *Bifidobacterium bifidum*. Decreasing in protein content was greater in protein concentrate that fermented using *Bifidobacterium bifidum*. These results indicated that *Bifidobacterium bifidum* greedier use some nutrients that exist on the seed oilcake or protein concentrates for growth. The results showed that the protein solubility varies from 48.97% to 60.98% (Tabel 3). The control treatment (R0) had lowest protein solubility, this suggested that the fermentation process increased the proteins solubility of oilseed cake and concentrate protein. Analysis of variance informed that treatments were highly significantly effect to pepsin solubility. Orthogonal contrast test showed that control (without treatment) significantly differed with all other treatments. Pepsin solubility of fermented protein concentrate (R4, R5) showed a higher solubility than protein concentrate without fermentation (R1). While the pepsin solubility of protein concentrate fermented by *Bifidobacterium bifidum* (R4) was significantly higher than *Lactobacillus acidophilus* (R5).

Antinutritive Compound

The result showed that phorbol ester content of protein concentrates (R1) were $0.410 \text{ mg.g}^{-1} \pm 116.54 \text{ mg.g}^{-1}$, as well as fermented protein concentrates (R4 and R5) were $0.259 \pm 72.23 \text{ mg.g}^{-1}$ and $0.343 \pm 139.44 \text{ mg.g}^{-1}$. Fermentation process using *Lactobacillus acidophilus* and *Bifidobacterium bifidum* to protein concentrates were effectively reduce phorbol ester and antitrypsin. Based on orthogonal contrast test, Antitrypsin and phorbol ester were significantly decreased in protein concentrate fermented by *Lactobacillus acidophilus* (R4) compared to *Bifidobacterium bifidum* (R5) ($P < 0.01$). On the other hand, there is no differences among other treatments. it can be stated that the process of making protein concentrates from jatropha seed cake, followed by fermentation process is the right step to reduce the content of anti-trypsin and phorbol ester. a decline of 59.20 % phorbol ester from 634 ppm (R0 /raw jatropha seed cake) to 258.67 (R4). Otherwise Makkar *et al.* (2008) reported protein concentrate from *Jatropha curcas* was comprise a large number of phorbol ester ($0.86\text{-}1.48 \text{ mg g}^{-1}$). The research showed that fermentation led to decrease phorbol ester. This is presumably due to fermentation using lactic acid bacteria produce acids that can dissolve fat, which in turn will lower phorbol ester. Goel *et al.* (2007) stated that the phorbol ester compound was defined as "polycyclic" in which the two hydroxyl groups on the carbon atom nearest is esterified with fatty acids. Ahmed and Salimon (2009) informed that there are significantly variations in oil content and the levels of phorbol ester in the seeds that are originated from three countries, phorbol ester found in Malaysian oil seeds is 0.23%, while from Indonesian and Indian are 1.58% and 0.58% respectively. The content of trypsin inhibitor, lectins and phorbol ester in R1 until R5 were approximately ten-fold lower than non treated oilseed cake (R0). Lectins were in high levels, but their rates were lower than the seed cake. Lectins content of post fermented oilseed cake was unidentified. This indicated that fermented jatropha seed cake on protein concentrates and seed expeller processing was capable to eliminate the lectin content. Fermentation processing could increased the nutrient composition of Fermented *Jatropha curcas* seed cake and concentrate protein compared to oil seedcake without treatment (Table 1).

Table 1. Crude Protein, Protein Solubility, and Antinutrient Content of Protein Concentrate of *Jatropha curcas* Seed Cake Produced by Different Processing

Treatments	Crude Proteins (%)	Protein Solubility (%)	Lectins (Unit)	Antitrypsin (TIU)	Phorbol ester (ppm)
R0	32.79 ± 0.05	48.97 ± 2.17 ^a	48.3	24270.05 ± 765.3857	634.00 ± 92.92 ^a
R1	48.21 ± 0.28	51.85 ± 0.41 ^{ab}	NID	24077.97 ± 17208.55	410.03 ± 116.54 ^a
R2	32.45 ± 0.26	49.71 ± 1.98 ^a	NID	11907.76 ± 9707.513	455.67 ± 183.09 ^a
R3	31.36 ± 0.29	53.64 ± 2.43 ^{ab}	NID	20439.98 ± 2615.456	388.00 ± 65.83 ^a
R4	44.42 ± 0.19	57.88 ± 0.64 ^b	NID	1050.532 ± 575.1024	258.67 ± 72.23 ^b
R5	42.78 ± 0.23	60.98 ± 6.32 ^b	NID	8362.785 ± 1274.059	343.00 ± 139.44 ^c

Note : R0: *Jatropha curcas* seed cake (control), R1: protein concentrate produced by acid, R2: *Jatropha curcas* seed cake fermented by *Lactobacillus acidophilus*, R3: *Jatropha curcas* seed cake fermented by *Bifidobacterium spp*, R4: protein concentrate produced by precipitation and fermentation using *Lactobacillus acidophilus*, R5: protein concentrate produced by precipitation and fermentation using *Bifidobacterium spp*. NID (Not Identified), TIU (Trypsin Inhibitor Unit)

Conclusions

Precipitation processes followed by fermentation using lactic acid bacteria improve the quality of *Jatropha curcas* seed cake. Protein concentrate fermented by *Lactobacillus acidophilus* is the best performance in term of nutrient content and antinutritive compounds. Protein concentrate *Jatropha curcas* seed cake produce by precipitation and fermentation method potentially be used as a feed of rabbits, poultry and ruminants.

Acknowledgement

The author wish to thanks to Jenderal Soedirman University and PT. Jatropha Green Energy Kudus Central Java Indonesia. This work was supported in a part by grant from Indonesia Directorate General of Higher Education for Featured research.

References

- Ahmed, W.A and J. Salimon. 2009. Phorbol Ester as Toxic Constituents of Tropical *Jatropha Curcas* Seed Oil. © *EuroJournals Publishing, Inc.* ISSN 1450-216X Vol.31 No.3 , pp.429-436..
- AOAC. 1990. *Official Methods of Analysis of the Association of Official Analytical Chemist.* Editor by W. Harwitz. Benjamin Franklin Station. Washington.
- Arnouts, S. and Vandendriessche, J. 2007. The Effect of lectin in Combination with Sodium Butyrate on The Performance of Broilers. *INVE Technologies. Hoogveld 93.* 9200 Dendermonde. Belgium.
- Chivandi, E. K.H. Erlwanger, S.M. Makuza, J.S.Read and J.P. Mtimuni. 2006. Effects of Dietary *Jatropha curcas* Seed cake on Percent Packet Cell Volume, Serum Glucose, Cholesterol and Triglyceride Concentration and Alpha- amylase Activity of Weaned Fattening Pigs. *Research Journal of Anim. Vet. Sci.* 1 (1): 18- 24.
- Goel, G. H P. S. Makkar, G. Francis, and K. Becker. 2007. Phorbol Esters: Structure, Biological Activity, and Toxicity in Animals. *International Journal of Toxicology.* 26:279–288. DOI: 10.1080/10915810701464641.
- Gogoi R., U.K.Niyogi and A.K. Tyagi. 2014. Reduction of phorbol ester content in jatropha cake using high energy gamma radiation. *Journal of Radiation Research and Applied Sciences.* Vol 7. P: 305 -309

- Herera, M.J., P.Siddhuraju, G.Francis, G. D. Ortiz and K. Becker. 2006. Chemical composition, toxic/antimetabolic constituents, and effects of different treatments on their levels, in four provenances of *Jatropha curcas* L. From Mexico. *Food Chem.* 96 p : 80-89.
- Makkar, HPS, G. Francis, and K. Beckers. 2008. Protein concentrate from *Jatropha curcas* screw-pressed seed cake and toxic and antinutritional factors in protein concentrate. *Journal of the Science of Food and Agriculture*, Volume 88, Number 9, July 2008, pp. 1542-1548 (7).
- Makkar, HPS, J. Maes, W. De Greyt and K. Beckers. 2007. A Laboratory Manual on Quantification of Plant Secondary Metabolites. Humana Press. New Jersey. P:101 – 105
- Makkar, HPS and K. Becker. 1999. Nutritional Studies on rats and fish (carp *Cyprinus carpio*) fed diets containing unheated and heated *Jatropha curcas* meal of a non-toxic provenance. *Plant food for human nutrition.* 53 : 183 – 192.
- Smith, C., W.V. Megen, L. Twalfhoven and C. Hitchcock. 1980. The determination of trypsin inhibitor levels in foodstuffs. *Journal of the Science of Food and Agriculture.* Vol. 31. P: 341 – 350.
- Widiyastuti, T., C.H. Prayitno, and N. Iriyanti. 2013. Digestibility and Blood Metabolite Profiles of Chicken Fed Fermented *Jatropha* Seed Meal.. *Animal Production* 15(2). P:98-105
- Widiyastuti, T., M. Indrajati, A. Wibowo and R. Hendroko. 2014. Biorefinery of *Jatropha* Seed Cake by Lactid Acid Bacteria and The Effects on Hematological Profile of Rex Rabbit. *J. Egypro* Vol 47 p: 290 – 294. DOI : 10.1016/j.egypro.2014.01.227.

Serum testosterone, testes size and semen quality of rams fed sugarcane bagasse treated with urea or pronifer

Kobeisy, M. *¹, G. Abd El-Hafez¹, S. Mousa¹ & A. Kabak²

¹Animal Production Department, Faculty of Agriculture, Assiut University, Assiut, Egypt

²Animal Production Department, Faculty of Agriculture, Al-Azhar University, Assiut, Egypt

Abstract

This study was to investigate the influence of sugarcane bagasse silage treated with urea, live yeast and pronifer on serum testosterone concentration, testes size and semen quality of rams. Sixteen Rahmani rams were used in four feeding groups, Group 1, untreated bagasse, Group 2, 3% urea treated bagasse; Group 3, urea and yeast (10 g per animal daily) treated bagasse and Group 4, urea and pronifer (2 g per animal daily) treated bagasse. Pronifer is feed additive made by specific lactic acid fermentation of heat-treated soy bean meal and malt. Blood samples were collected for serum testosterone level determination. Testes size was measured and semen quality was tested. The experiment lasted for six months. All results were analyzed using GLM procedure of SAS. Testosterone level in blood of animals fed urea treated silage with live yeast or pronifer have higher ($P < 0.05$) values than those of control or urea group. No significant effects of urea supplementation on testosterone level, testes size and semen quality. No differences among treatments for testes size but semen quality showed that mass activity, volume and semen concentration for yeast and pronifer groups were higher ($P < 0.05$) than those of control or urea treated groups. Pronifer supplement improve semen quality by decreasing total abnormalities and increasing mass activity ($P < 0.05$). In conclusion, urea treated bagasse have no deleterious effects on semen quality, however, live yeast and pronifer can enhance ($P < 0.05$) semen quality in rams fed urea treated bagasse.

Keywords: testosterone, semen quality, urea, pronifer, Sugarcane bagasse.

Corresponding author: mostafa_kobeisy@yahoo.com

Introduction

Most of sugarcane bagasse which is produced during sugar production is used in producing fire and consequently increased air pollution. The low nitrogen and high fiber content formed a problem for using it for feeding animals (Khan et al., 1992), so some processing could be helpful for using it as low cost roughage. These processing such as preserving it as a silage and addition of urea (Singh et al., 1999) and yeast (Harrison et al., 1988) or even feed additives like pronifer which is a probiotics made by specific lactic acid fermentation of heat – treated soybean meal and malt. The objective of the recent study is to evaluate the effect of sugarcane bagasse silage treated with urea, live yeast or pronifer on Serum testosterone, testes size and semen quality of rams.

Materials and Methods

The study was carried out on 16 *Rahmani* lambs over five months old with 29.06 ± 0.54 kg an average body weight. Sugarcane bagasse was collected and sun-dried for two months then

ensiled with 5% molasses for two months in two treatments, untreated and treated silage, with 3% urea calculated as dry matter basis. Animal were assigned to 4 groups and receiving 600 g concentrate diet and one of the following treatment roughage diet (ad libitum) for 6 months: Group1, untreated sugarcane bagasse silage, UBS (control). Group 2, Urea treated (3%) SBS (Urea). Group 3; diet No 2 with live yeast culture, 10 mg/animal/day (Yeast). Group 4; diet No 2 with pronifer, 2 mg/animal/day. Pronifer is a probiotic made by specific lactic acid fermentation of heat-treated soybean meal and malt, from P.G.E. (EGGER), GmbH, Mitterlabill, Austria). Animal were fed once daily and water was offered two times daily. Blood samples were collected and serum was separated and analyzed for serum testosterone using ELISA kits (Biosource, Belgium). The size of testis (both testes and scrotum sizes) for all studied groups were measured monthly by volume subtracted method. Semen samples were collected weekly at the last 3 weeks of feeding trial, in the morning at 9:00 am, by using artificial vagina. Semen quality of these lambs (approximately one year old) were examined for volume, mass activity, live sperm %, total abnormalities % and sperm concentration according to Zemjanis (1970) Procedures. All data were analyzed by ANOVA using General Linear Models Procedure of SAS (1985).

Results and Discussion

Serum testosterone concentration level

Serum testosterone concentration level in live yeast and pronifer groups were higher ($P < 0.05$) than those of control and urea groups (Table 1). Pronifer and live yeast supplementation improved serum testosterone level; such effect may be due to their effects on reproduction and NPN assimilation in sheep. However, Urea supplement had no enhanced effect on serum testosterone. Different result was found by Megahed et al. (2000) and their results may be due to the difference in urea level intake, feeding period and/or crude protein of the basal diet.

Table 1. Effect of feeding sugarcane bagasse silage treated with urea, live yeast and pronifer on serum testosterone (ng/ml) in sheep.

Experimental Period, month.	Treatment*				
	Control	Urea	Yeast	Pronifer	SE
1	1.34	1.33	1.51	1.66	0.15
2	1.79	1.80	1.88	2.12	0.15
3	2.13	2.13	2.47	2.91	0.15
4	2.20	2.29	2.85	3.11	0.15
5	2.27	2.34	3.15	3.35	0.15
6	2.48	2.45	3.37	3.74	0.15
Mean	2.03 ^c	2.06 ^c	2.54 ^b	2.82 ^b	-

*Values are least square means (LSM) and SE is a standard error of LSM. Control, sugarcane bagasse silage (SBS); Urea, SBS+3% urea; Yeast, SBS+3%urea+10 gm yeast /animal and day and Pronifer, SBS + 3% urea + 2 gm pronifer /animal and day. Means within rows differ ($P < 0.05$) when superscripts differ.

Testes size

By observation and measurement of testis size founded the both testes of all experimental lambs were quit balancing sizes” in this results table (2) showed that, no significant differences among treatments for testes size indicating no significant effect of urea, yeast or pronifer on rams’ testes size. In contrast, Megahed et al. (2000) found higher testicular growth for sheep fed sugarcane bagasse or tops treated with 1 % urea than control. Such differences may be due to differences in urea intake, crude protein of basal ration, feeding period, levels and source of nitrogen (Murray et al., 1991; Thwaites, 1994) and/or association effect of feeds (Mehrez,1989).

Table 2. Effect of feeding sugarcane bagasse silage treated with urea, live yeast and pronifer on testes size (ml) in sheep.

Experimental Period, month.	Treatment*				
	Control	Urea	Yeast	Pronifer	SE
1	255	275	290	263	10.45
2	313	327	343	310	10.45
3	388	378	390	355	10.45
4	445	433	463	398	10.45
5	500	528	520	480	10.45
6	552	598	548	568	10.45

*Values are least square means (LSM) and SE is a standard error of LSM. Control, sugarcane bagasse silage (SBS); Urea, SBS+3% urea; Yeast, SBS+3%urea+10 gm yeast /animal and day and Pronifer, SBS + 3% urea + 2 gm pronifer /animal and day.

Semen quality

No differences were found between control and urea treatment in sperm concentration, total abnormalities, semen volume, mass activity and alive sperm (Table 3). Similarly no beneficial effect of urea on semen quality was found by (Al-Haboby et al., 1999). In contrast Ferguson & Chalupa (1989) reported harmful effect of urea treated diets on spermatozoa and semen quality. However, yeast and pronifer improved ($P < 0.05$) sperm concentration and semen volume. Also, pronifer group had lower ($P < 0.05$) total abnormalities and higher ($P < 0.05$) mass activity than those of control and urea groups. Similar result was found by Abd- El-Aziz (2001), they found higher ($P < 0.05$) semen volume in rams fed sugarcane tops silage treated with 1% urea and yeast (5 kg/ton) than those fed the same diet without yeast. The enhancement of semen quality in rams fed yeast or pronifer in the present study may be attributed to the increase in serum testosterone (Table 1) which activates spermatogenesis and the secretary function of the accessory glands (Salisbury, 1987). Spermatogenesis is dependent on endocrine support (Johnson, 2007). In conclusion: Dietary yeast or pronifer may be useful for improvement of serum testosterone level, testes size and semen quality of rams fed urea treated sugarcane bagasse silage.

Table 3. Effect of feeding sugarcane bagasse silage treated with urea, live yeast and pronifer on semen quality in sheep.

Item	Treatment*				
	Control	Urea	Yeast	Pronifer	SE
Sperm concentration, $\times 10^6/\text{mm}^3$	2.66 ^b	2.77 ^b	3.73 ^a	3.88 ^a	0.21
Total abnormalities, %	15.64 ^a	15.57 ^a	14.46 ^{ab}	14.05 ^b	0.39
Volume, ml	0.97 ^b	1.00 ^b	1.77 ^a	1.87 ^a	0.09
Mass activity	4.00 ^b	4.33 ^{ab}	4.67 ^{ab}	5.00 ^a	0.24
Alive sperm, %	82.31	83.74	85.04	86.67	1.64

*Values are least square means (LSM) and SE is a standard error of LSM. Control, sugarcane bagasse silage (SBS); Urea, SBS+3% urea; Yeast, SBS+3%urea+10 gm yeast /animal and day and Pronifer, SBS + 3% urea + 2 gm pronifer /animal and day. Means within rows differ ($P < 0.05$) when superscripts differ.

References

- Abd-El-Aziz, M. M.F., 2001. Utilization of sugarcane tops in sheep feeding. M. Sc. Thesis, Fac. Agric., Assiut Univ., Egypt
- Al-Haboby, A. H., A. D. Salman and T. A. Abdul-Kareem, 1999. Influence of protein supplementation on reproductive traits of Awassi sheep grazing cereal stubble. Small Rumin. Res. 34:33-40.

- Ferguson, J. D. and W. Chalupa, 1989. Impact of protein nutrition on reproduction in dairy cows. *J. Dairy Sci.* 72: 746-766.
- Johnson, M.H., 2007. Testicular function in the adult, in: *Essential Reproduction* (6th Ed.), Blackwell Publishing. pp:69.
- Khan, M. E., A.Ali and Z.O. Muller, 1992. Nutritional evaluation of sugarcane bagasse based rations treated with urea and cattle manure. *Anim. Feed Sci. Technol.*, 38:135-141.
- Harrison, G. A., R. W. Hemken, K. A. Dawson, R. J. Harmon and K. B. Barker, 1988. Influence of addition of yeast culture supplement to diets of lactating cows on ruminal fermentation and microbial population. *J. Dairy Sci.* 71: 2967-2975.
- Megahed, G.A., H. A. Daghsh and M. M. Anwar, 2000. Reproductive performance in Ossimi rams as influenced by crude protein. *Egypt. Soc. Anim. Reprod. Fert.* 12th Annual Congr. Giza, 16-18 January, pp.67-77.
- Mehrez, A. Z., 1989. Associative effect of feeds. *Proc. First scientific symposium on animal and poultry nutrition.* Assiut University, 15-16 Nov., Assiut, Egypt.
- Murray, P.J., J.B. Rowe and D.W. Pethick, 1991. Effect of season and nutrition on scrotal circumference of Merino rams. *Aust. J. Exper. Agric.* 31:753-758.
- Salisbury, G.W., N.L. Vandemark and J. R. Lodge, 1987. *Physiology of reproduction and A.I.* 2nd Ed. W.H. Freeman and Co., Sanfrancisco, USA, pp:199.
- SAS, 1985. *SAS User's Guide: Statistics.* SAS Int., Cary, NC
- Singh, A. P., V.S. Upadhyay, A. K. M.isra and A. K.Samanta, 1999. Influence of urea and cow pea addition on the silage quality of sugarcane bagasse. *Ind. J. Dairy Sci.* 52:337-343.
- Thwaites, C. J., 1994. The effects of feeding supplements containing different amounts and sources of nitrogen on live weight and the testes of rams during mating. *Anim. Feed Sci. and Technol.* 48:177-184.
- Zemjanis, R., 1970. *Diagnostic and Therapeutic Technique in Animals Reproduction.* 2nd ED. Williams and Wilkines Company, Baltimore. USA.

Effects of storage on pathogenic bacteria content of layer manure extract

Usungnoen¹, P., W. Jintanawich¹ & S. Juttupornpong¹

¹Department of Animal Science, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, 73140, Thailand.

Abstract

The effects of storage of layer manure extract (LME) produced from 1) newly dried manure and 2) and 3) dried layer manure stored for 1 and 2 month, respectively, on the content of pathogenic bacteria including coliform bacteria, *E.coli* and *Salmonella* spp. were studied. LME was produced by steeping of dried layer manure in water at a ratio 1:10 w/v for 24 hr. and then remove out the solid. The LME produced were stored in 200 liter plastic drum covered with plastic lid. Samples of the LME were taken at Day 0, 7, 14, 21, 28, 35, 42, 49 and 56 of storage and analyzed for population of coliform bacteria, *E. coli*, *Salmonella* spp., pH and total N content. Results of the study showed that storage of dried layer manure could reduced ($P < 0.001$) the initial loads of the pathogenic bacteria in LME produced from the layer manure. The storage of LME significantly reduced ($P < 0.05$) population of coliform bacteria, *E. coli* and *Salmonella* spp. to nil at Day 35, 28 and 14, respectively. LME produced from stored dried layer manure has more rapidly decreasing of the pathogenic bacteria content than those produced from the newly dried manure. There were apparently no change in total N content, but there was a significantly increase ($P < 0.001$) of pH in LME during the storage. The research data has indicated the safe use of LME for food crop production after the storage of LME for 14 to 35 days. In additional, the uses of stored layer manure for 1 month will reduce the risk of pathogenic bacteria contamination in the LME.

Keywords: layer manure extract, pathogenic bacteria, coliform bacteria, E.coli, Salmonella

*Corresponding author: patima2518@gmail.com, swkpmu@ku.ac.th

Introduction

Expansion of commercial animal production in Thailand has created a great problem in animal waste pollution to the surrounding environment. The department of Animal Science, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University (KU), Kamphaeng Saen Campus, Nakhon Pathom, Thailand has studied the utilization of animal manure including the manure of pig, quail, chicken and dairy for production of the animal manure extract as plant nutrient sources and it can be used as organic fertilizer for both soil and foliar application (Juttupornpong et al., 2012). Results of the study showed that chemical composition and physical property of layer manure extract (LME) are as followed: N 0.15%, P 0.03%, K 0.27%, Ca 400 ppm, Mg 100 ppm, Cu 1.58 ppm, Fe 8.65 ppm, Mn 1.91 ppm, Zn 2.18 ppm, B 1.94 ppm, Na 0.03 ppm, pH 7.0 (Juttupornpong & Usungnoen, 2013). However, contamination of pathogenic bacteria found in the gastrointestinal tract of animals including coliform bacteria, *Escherichia coli* and *Salmonella* spp. are the prime concern for the utilization of animal farm waste as plant nutrient sources. The high contamination of the pathogenic bacteria in food will be harmful to the consumer health (Maier et al., 2000). Objectives of the study to evaluate the effects of storage LME at different

time interval on pathogenic bacteria content in LME, which will determine the appropriate storage time of LME to be used as a safe foliar fertilizer for vegetable production.

Materials and Methods

The study was conducted at Animal Nutrition Research and Development Center, KU, Kamphaeng Saen Campus, Thailand. Three types of dried layer manure; the newly dried manure (LM0) and the dried manure stored for 1 (LM1) and 2 (LM2) months were steeped in the water at a ratio of dried manure: water = 1:10 w/v for 24 hr. Then, the remaining solid of the manure was filter out, and the liquid part was classified as layer manure extract (LME). Store the LME in plastic drums covered with plastic lid. Sampled the LME from each drum at Day0, 7, 14, 21, 28, 35, 42, 49 and 56 of storage and analyzed for the content of coliform bacteria, *E. coli*, by pour plate method where the coliform agar (Merk Cat.No.1.10426.0500) was incubated at 37 °C for 24 hr, and the content of *Salmonella* spp. by the method adapted from Arun et al. (1994). The samples of LME stored at 0, 7, 28 and 56 days were analyzed for total N content by Kjeldahl method (AOAC, 1990) and the pH by pH meter. The experiment was designed as 3×9 Factorial in RCB with 3 replications. The data were subjected for analysis of variance (ANOVA) and the means of treatments were compared by Duncan's Multiple Range Test (DMRT) (P<0.05) using SAS software version 9.0 (SAS Institute, Cary, NC, USA)

Results

Coliform bacteria content

LME prepared from LM0 had significantly higher (P<0.001) initial loads of coliform bacteria than those prepared from LM1 and LM2. However, storage of LME significantly reduced (P<0.001) the contents of coliform bacteria in LME produced from LM0, LM1 and LM2. The coliform bacteria contents in LME was highest in Day 0 and drastically reduced (P<0.001) to nil at Day 35 onward as shown in Table 1. There was a significantly interaction between the type of layer manure and the storage time of LME (P<0.001) (Table 1).

Escherichia coli content

The storage of LME significantly reduced (P<0.05) the *E. coli* content in LME produced from LM0, LM1 and LM2. The *E. coli* content in LME was highest in Day 0 and drastically reduced (P<0.05) to nil at Day 28 onward (Table 1).

Salmonella spp.

It is clearly demonstrated that layer manure type affects the contents of *Salmonella* spp. count in the LME product. LME product from LM0 has *Salmonella* spp. content until Day 7 of storage. LME produced from LM0 can detect the bacteria content only until Day 14, mean while LME produced from LM1 and LM2 cannot be detected the bacterial content at Day 0 of storage.

Chemical composition and pH value

Total N content in LME was ranging from 0.05 to 0.09% and there were apparently no changing of total N content during the storage from 0 to 56 days. The average pH of newly produced LME was 7.21, and it was significantly increased (P<0.001) to 7.72, 8.75 and 8.86 at Day 7, 28 and 56 of storage, respectively (Table 2).

Discussion

Results of the study indicated that there was a content of pathogenic bacteria in the layer manure and LME. However, there were evidences that the storage of layer manure can reduce the pathogenic bacteria population in LME produced from the manure. Regardless of the initial loads of the pathogenic bacteria population in the LME (Day 0 storage), storage of LME significantly reduced the content of coliform bacteria. During the storage, the bacteria will use-up all nutrients available in the medium (carbon and nitrogen sources) for growth until exhausting as well as the unfavorable condition for living i.e. the increasing of pH, and then become the death of the bacteria (Bussakorn, 2009). There was no significantly changing of total N content in LME during the storage. Although the bacteria requires N in the medium to be in cooperated into amino acids, proteins, nucleotides, peptides and some vitamins in the bacterial cells (Kuivakanon et al., 1984). Death of the bacterial cells will release the N back into the medium and caused no change of total N content in the LME. However, metabolism of the bacteria produced metabolite including organic acids, gasses, CO₂ and H₂ (Bussakorn, 2009) which may have caused the increasing of pH of the LME and the unfavorable condition for bacterial living.

Table 1. The effect of storage of dried layer manure and LME on coliform bacteria and *E. coli* population in the LME. (cfu/ml).

Storage of LME (days)	Coliform population (cfu/ml)				<i>E. coli</i> population (cfu/ml)				
	Storage of dried layer manure			Mean	Storage of dried layer manure			Mean	
	LM0	LM1	LM2		LM0	LM1	LM2		
0	7,206,250	1,042,500	3,840,625	4,029,792 ^a	1,250,000	398,750	310,000	652,917 ^a	
7	2,714,125	349,791	380,750	1,148,222 ^b	1,280,063	298,975	135,662	571,567 ^a	
14	229,293	87	10,382	79,921 ^c	22,131	282	7,414	9942 ^b	
21	0	2	398	133 ^c	0	5	271	92 ^b	
28	0	4	2	2 ^c	0	0	1	0 ^b	
35	0	0	0	0 ^c	0	0	1	0 ^b	
42	0	0	0	0 ^c	0	0	0	0 ^b	
49	0	0	0	0 ^c	0	0	0	0 ^b	
56	0	0	0	0 ^c	0	0	0	0 ^b	
Mean	1,127,741 ^a	154,709 ^b	470,240 ^b		283,577 ^a	77,557 ^a	50,372 ^a		
C.V. = 174 Grand mean = 584,230				C.V. = 332 Grand mean = 154,315					

Table 2. The effect of storage of dried layer manure and LME on total nitrogen content and pH of the LME.

Storage Of LME (days)	Total Nitrogen (%)				pH				
	Storage of dried layer manure			Mean	Storage of dried layer manure			Mean	
	LM0	LM1	LM2		LM0	LM1	LM2		
0	0.07	0.07	0.06	0.07 ^a	7.19	7.22	7.22	7.21 ^c	
7	0.07	0.09	0.08	0.08 ^a	7.73	7.72	7.72	7.72 ^b	
28	0.07	0.05	0.08	0.07 ^a	8.74	8.75	8.75	8.75 ^a	
56	0.07	0.05	0.07	0.06 ^a	8.85	8.87	8.87	8.86 ^a	
Mean	0.07 ^a	0.07 ^a	0.07 ^a		8.12 ^a	8.14 ^a	8.14 ^b		
C.V. = 31 Grand mean = 0.07				C.V. = 1.68 Grand mean = 8.14					

Note: Means with the same letter in the column do not differ significantly at (P≥0.05).

LME = Layer manure extract

LM0 = New manure, LM1 = Layer manure stored for 1 month, LM2 = Layer manure stored for 2 month

C.V. = Coefficient of Variation

Conclusion

Storage of dried layer manure can reduce ($P < 0.001$) the initial loads of pathogenic bacteria in LME produced from the layer manure. Regardless of the initial loads of the pathogenic bacteria in LME, storage of LME greatly reduced ($P < 0.05$) population of coliform bacteria, *E. coli* and *Salmonella* spp. to nil at Day 35, 28 and 14, respectively. LME produced from stored dried layer manure had a more rapid decreasing of the pathogenic bacteria content than those produced from the newly dried manure. There were apparently no change in total N content, but a significantly increase of pH in LME during the storage. The research data clearly indicated that LME after 14 to 35 days storage is a safe plant nutrient source for food crop production. In additional, the used of stored layer manure for 1 month will reduce the risk of pathogenic bacteria contamination in the LME.

References

- Arun, B., 1994. Laboratory Diagnosis of Pathogenic Bacteria in the Digestive Tract. Division of Clinical Pathology, Department of Medical Sciences, Ministry of Public Health, Bangkok, Thailand.
- AOAC, 1990. Method of the Association of Official Analytical Chemists. Benjamin Franklin Station, Washington DC.
- Bussakorn U., 2009. Food Microbiology. Department of Biology, Faculty of Science, Thaksin University. 457 p.
- Juttupornpong, S., P. Usungnoen and U. Kanto, 2012. Utilization of Pig Manure and Effluence from Pig Farm as Organic Fertilizer for Economic Crop. 1th Edition. SARDI, Kasetsart University, Kampaeng Saen Campus, NakhonPathom, Thailand. pp 3.
- Juttupornpong S. and P. Usungnoen, 2013. Utilization of Chicken Manure as Organic Fertilizer for Plant. 1th Edition. SARDI, Kasetsart University, Kampaeng Saen Campus, NakhonPathom, Thailand. pp 9.
- Kuiyakanon T., U. Tattawasart and W. Lulitanon, 1984. Bacteriology. Faculty of Medicinal, KhonKaen University, Khon Kaen, Thailand. 147 p.
- Maier, M.R., L.I. Pepper and P.C. Gerba, 2000. Environmental Microbiology. Academic Press, Canada. 585 p.

Effect of heat stress environment on the blood parameters and behavior pattern in Korean native calves

Won-Seob Kim, Jae-Sung Lee, Min-Jeong Kim, U-Suk Jung, Seung-Woo Jeon, Dong-Quio Peng, Young-Sin Kim, Mun-Hee Bae¹, Sang-Rak Lee & Hong-Gu Lee

Department of Animal Science and Technology, College of Animal Bioscience and Technology, Konkuk University, 120 Neungdong-ro, Gwangjin-gu, Seoul 143-701, Korea

Abstract

Heat stress in hot climates is a major factor that strongly affects Korean beef industry in negative ways, leading to immeasurable economic loss. For this reason, we investigated the effects of a heat stress environment on the blood parameters and behavior in Korean native calves (6~7 months, male). The temperature-humidity index (THI) is commonly used to indicate the degree of heat stress on cattle. The study used 6~7 month-old Korean native calves (204.5 ± 6.79 kg) and was conducted in two designated THI periods: low THI (LTHI; $\text{THI} = 70.01 \pm 0.81$) and high THI (HTHI; $\text{THI} = 87.73 \pm 0.92$). According to the temperature and humidity change (THI 70-87), the blood metabolites and hormone as stress-related indicators were analyzed by biochemical analyzer-TBA-40FR, and CBC (Complete blood Count) test was conducted to determine the change in blood cells. Through a video monitoring system, we also analyzed the behavior pattern, feed intake, water intake, heart rate and rectal temperature. The results showed that there was a decrease in NEFA and an increase in GOT and GPT in the serum of calves under hot conditions (HTHI) ($P < 0.05$), and that these calves have lowered WBC and platelet counts, but an enhanced rectal temperature and heart rate ($P < 0.05$). The level of serum cortisol as indicator of stress steroid hormone was significantly increased in the HTHI period compared with the LTHI period ($P < 0.05$). In addition, calves in the HTHI period had an increased water intake but a decreased feed intake compared with calves in the LTHI period. This means that higher THI is related to high stress in Korean native calves. In conclusion, the data indicate that determining the basis for altered blood parameter and behavioral pattern during heat stress will lead to opportunities for improved animal performance via altered nutritional management in Korean native cattle.

Keywords: heat stress, THI, behavior pattern, blood parameter, Korean native calves

Corresponding author: kws9285@naver.com

Introduction

Heat stress in hot climates is a major factor that strongly affects Korean beef industry in negative ways, leading to immeasurable economic loss. Such heat stress also has been clearly linked to decreased dry matter intake and other measures of performance including milk yield, average daily gain (Meyer et al., 2010). The reduced weight gain and feed efficiency (Ko et al., 2006) is a result of increased body temperature that will induced the decline in feed intake as well as alterations in endocrine profiles, energy metabolism (Baumgard & Rhoads, 2007) and other unidentified factors (Collier et al., 2008). Heat stress in cows commonly is thought to be a more relevant concern to the industry than that in calves because the calves produce less metabolic

heat and have a greater surface area to body mass ratio with which to dissipate heat (Meyer et al., 2010). Environmental factors along with metabolic heat production prevent the animal from maintaining thermal balance or homeostasis, thus body temperature rise and the animal must compensate and adapt in order to re-establish homeostasis (West et al., 1999).

Commonly, heat stress is being quantified by calculating THI (Temperature Humidity Index) and it was already developed in lactating dairy cows (Collier et al., 2012). THI is a value associated with different levels of thermal stress and combines the effects of humidity and temperature (Bohmanova et al., 2007). The current THI was originally developed by Thom (1958) and extended to cattle by Berry et al (1964). It is currently used to estimate cooling requirements of dairy cattle in order to improve the efficiency of management strategies to alleviate heat stress (Zimbelman et al., 2008). Currently it is widely used in computerized cooling systems to estimate cooling requirements of dairy cattle. However, there was no studies about beef cattle or calve THI chart, especially THI for Korean native calves. Therefore, we attempted to establish THI chart for Korean native calves under heat stress condition.

Materials and Methods

Animals

The data analyzed in this study was obtained from 8 different studies during the course. Four Korean native calves (6-7 months, 204.5 ± 6.79 kg) were housed in individual tie stalls in one of the environmentally controlled chamber. All experimental procedures were in accordance with the "Guidelines for Care and Use of Experimental Animals of Konkuk University (KUB151043)"

Experimental design

Eight different studies were designed involving four levels of dry bulb temperature (22-34°C) and two levels of humidity (60-80%). These parameters were then combined to produce eight experimental environments. Animals entering the facility were provided three days to acclimatize to the chambers in a thermal neutral environment. After adaptation, calves experienced the experimental environment for four days.

Physiological parameters under heat stress

Calves were fed and given water four times daily with orts measured once a day (0800 h). During adjustment periods and experiment periods, water intake and feed intake were measured daily. Entire behavior was recorded by video system (0900-1900 h). Heart rate was determined by measuring the beat per minute of the heart using stethoscope (1100 h). Rectal temperature was measured at the 2nd and last day (1100 h) during the experimental period.

Blood parameter under heat stress

Blood samples were obtained from jugular vein of each calve (1100 h), then centrifuged (3,000 rpm, 4°C, 15 min), serum and plasma were collected and kept in -80°C until analysis. Hematological traits, parameters of metabolism (NEFA, GOT, GPT), and cortisol were analyzed by VetScan HM2 (Diamond Diagnostics, Abaxis Inc., MA, USA), Biochemical analyzer (Diamond Diagnostics) and Bovine Cortisol ELISA test kit (Endocrine technology Co., Ltd, city, country), respectively. Behavior pattern were recorded and marked the frequency of ruminant's lying and standing on the 3rd day (0900-1900 h).

Statistical analysis

All results are expressed as the means with standard errors. Comparison between two groups or multiple groups were analyzed using Student's *t*-test or TURKEY Kramer's multiple comparison test, respectively. Differences were considered statistically significant if the probability less than 0.05. All statistical analyses were performed using the JMP 5.0 (SAS Institute Inc., Cary, NC, USA).

Results and Discussion

The study was conducted in two designated THI periods: low THI (LTHI; THI = 70.01 ± 0.81) and high THI (HTHI; THI = 87.73 ± 0.92). Temperature humidity index was calculated using dry bulb temperature (Tdb, °F) and relative humidity (RH), $(Tdb - (0.55 - (0.55 * RH / 100)) * (Tdb - 58))$; Buffington et al., 1977). The results showed that there was a decrease in NEFA and an increase in GOT (Glutamic oxaloacetic transaminase) and GPT (Glutamic-pyruvic transaminase) in the serum of calves under hot conditions (HTHI) ($P < 0.05$), and that these calves have lowered WBC (White blood cell) and platelet counts, but an enhanced rectal temperature and heart rate ($P < 0.05$). The level of serum cortisol as indicator of stress steroid hormone was significantly increased in the HTHI period compared with the LTHI period ($P < 0.05$). In addition, calves in the HTHI period had an increased water intake but a decreased feed intake compared with calves in the LTHI period. This means that higher THI is related to high stress in Korean native calves. In conclusion, blood parameter and behavior pattern of heat stress were shown to be correlated with THI and therefore are measurements that can be obtained to evaluate the degree of heat stress in the Korean native calves.

References

- Anderson, A. D, B. J. Bradford, J. P. Harner, C. B. Tucker, C. Y. Choi, J. D. Allen, L. W. Hall, S. Rungruang, R. J. Collier and J. F. Smith, 2013. Effects of adjustable and stationary fans with misters on core body temperature and lying behavior of lactating dairy cows in a semiarid climate. *J. Dairy Sci.* 96:4738-4750.
- Allen, J. D., L. W. Hall, R. J. Collier and J. F. Smith, 2015. Effect of core body temperature, time of day, and climate conditions on behavioral patterns of lactating dairy cows experiencing mild to moderate heat stress. *J. Dairy Sci.* 98:118-127.
- Baumgard, L.H. and R.P. Rhoads, 2007. The effects of heat stress on production and its nutritional implications. Penn State Dairy Cattle Nutrition Conference. pp 29-38
- Bohmanova, J., I. Misztal and J. B. Cole, 2007. Temperature-humidity indices as indicators of milk production losses due to heat stress. *J. Dairy Sci.* 90: 1947-1956.
- Brien, M. D. O R. P. Rhoads, S. R. Sanders, G. C. Duff, L. H. Baumgard, 2009. Metabolic adaptation to heat stress in growing cattle, *Domest. Anim. Endocrinol.* 38:86-94.
- Cassandra B. Tucker, Andrea R. Rogers, Karin E. Schutz, 2008. Effect of solar radiation on dairy cattle behavior, use of shade and body temperature in a pasture-based system, *Appl. Anim. Behav. Sci.* 109:141-154
- Collier, R. J., J. L. Collier, R. P. Rhoads, and L. H. Baumgard. 2008. *Invited Review: Genes involved in the bovine heat stress response.* *J. Dairy Sci.* 2008 91: 445-454.
- Ko, Y. D, H. S. Kang, S. J. Kwack, D. H. Kim, D. J. Kim, S. C. Kim, Y. M. Kim, Y. H. Kim, J. Y. Kim, J. T. Jeon and M. H. Jeong, 2006. Hanwoo Science, Sunjin Press, Seoul, Korea.
- Mader, T. L., 2003. Environmental stress in confined beef cattle. *J. Anim. Sci.* 81:E110-E119
- Meyer, D.M., M. J. Brouk and L. C. Hollis, 2010. Effects of Heat stress on dairy calves, The Dairy Site, <http://www.thedairysite.com/articles/2370/effects-of-heat-stress-on-dairy-calves>.

- Robert J. Collier, Laun W. Hall, Sunthorn Rungruang and Rosemarie B. Zimbleman. 2012. Quantifying Heat Stress and Its Impact on Metabolism and Performance, The University of Arizona
- Rhoads. M. L., R. P. Rhoads, M. J. VanBaale, R. J. Collier, S. R. Sanders, W. J. Weber, B. A. Crooker and L. H. Baumgard, 2009. Effect of heat stress and plane of nutrition on lactating Holstein cows: I. Production, metabolism, and aspects of circulating somatotropin. *J. Dairy Sci.* 92:1986-1997.
- West, J. W, G. M. Hill, J. M. Fernandez, P. Mandebvu and B. G. Mullinix, 1999. Effects of dietary fiber on intake, milk yield, and digestion by lactating dairy cows during cool or hot, humid weather. *J. Dairy Sci.*82:2455-2465
- Zimbelman,R. B., 2008. Management strategies to reduce effects of thermal stress on lactating dairy cattle, The University of Arizona.

Study of local feeds potency for pig farming development in Manokwari, West Papua, Indonesia

Lekitoo, M.N., S. Hartini, S. Lumatauw, D.D. Rahardjo, S.Y. Randa & T.W. Widayati

University of Papua, Gunung Salju St. Amban, 98314, Manokwari Indonesia

Abstract

A research was carried out to study the potential of local feeds for pig farming development in Manokwari. Descriptive method with technical survey was used to identify general characteristic of each farm and to identify the common local feeds using by the farmers in their pig rations, measure their quantity and quality and formulate appropriate rations for each growth stage by using available local feeds.

All identified local feeds were analyzed for protein (%) and ME (kcal/kg) content to evaluate the feed status at each farm. Results showed that majority of the farmers kept less than 50 pigs in a flock with simple permanent housing. All of the farmers used local feeds in pig rations without formulating them properly. In fact these local feeds were found potential with daily production as follow: Fish waste 1000 kg, tofu waste: 2400 kg, soybean hull 55.5 kg, mung-bean hull 83.4 kg, rice bran 11,586.6 kg, banana peel 127.5 kg, taro peel 11.4 kg, market waste (vegetable stalks) 546 kg, and restaurant waste 2056.6 kg. Only two farmers provided ration for their animals with enough protein and ME as required.

Keywords: local feeds, pig farming, pig ration, by-product

Introduction

Population, family income and economic status of Manokwari people increase. This results in increase of animal products demand. Data from the Department of Statistics of West Papua Province (2012) showed that Animal food consumption had been increased for the past four years. The rate of increase were 56.60% for beef, 59.15% for pork, 33.75% for goat meat, 43.33% for native chicken, 9.58% for broiler and 36.75% for ducks. Majority of these animal products had been imported from outside of Papua due to small population available in the region. Consequently, animal products were not only limited but also found high price in the market especially pig products.

Raising pigs is not new to local people but the pig farming practices is still subsistence (Iyai, 2008). This is due to limited animal feeds availability. The commercial feed for pigs was uncommon in the market and considered high price. Therefore, more farmers were using available local feeds such as taro peel, tofu by-product, coconut pulp, soybean hull (tempe by-product), mung-bean hull, banana peel, fish waste, market waste (vegetable stalks) and restaurant waste as mixture in pig rations. In using these local feeds, most farmers failed to consider the nutrition needed by the pigs. Consequently, the pig production is found not optimal.

This study was conducted to identify the common local feeds using by the farmers in their pig rations, measure their quantity and quality and formulate appropriate rations for each growth stage by using available local feeds.

Materials and Methods

A survey was applied in seven pig farms which were under a supervision of The Department of Animal Husbandry and Food Security of West Papua Province. The study was initiated by

observing the general status of each farm included: number of pigs raised, animal and housing condition, kinds of feeds used, farming system, and other management practices applied in their farms.

The focus of the study was to determine kinds and amount of local feeds used by farmers. Each local feed observed in each farm was recorded and weight to get the total (kg) of daily fresh weight used by the farmer. About 1 kg fresh weight of each local feed observed was brought to the Laboratory of Animal Nutrition at the Faculty of Animal Science, Papua University for water content determination. The dry form of each local feed then was sent to the Integrated Laboratory of Gadjah Mada University at Yogyakarta for nutrient analysis (Protein and ME content). The protein and ME content of each ration at each farm then was computed, and from this computation showed whether each farm gave ration to their pigs as required or not. Another focused in this study was to formulate and recommend rations for each stage of pig growth based on the available local feeds that commonly used by the farmers based on the nutrient requirement by pigs (NRC, 1998). Thus the amount of each local feed recommended at each ration was computed to meet the protein and ME requirements. In other words, the recommended ration in this study had the same protein (%) and ME (kcal/kg) content with the requirement according to NRC (1998).

Results and Discussion

General characterization

Number of pigs raised by the farmers in Manokwari were varied from 6 to 51 heads, and 30% of them were growers. Half brick open-sided housings were found to be the greater proportion (75%). Housing system was relatively simple with cemented floor and wooden wall. Housing size requirements were met due to small pig number being raised.

Local feeds commonly used by the farmers were: tofu by-product, fish waste, soybean hull, mung-bean hull as protein sources, and rice bran, banana peel, taro peel, market waste (vegetable stalks), restaurant wastes as carbohydrate sources. Table 1 shows kinds and potency of each local feeds in Manokwari.

Table 1. Kind and potency of local feeds in Manokwari.

No.	Commodity	Potency (kg/d)**	By-product	Total Estimation (kg/d)***
1.	Soybean	1,519.0	Tofu by-product	2,400.0
			Soybean hull	55.5 kg
2.	Fish	2,791.0	Fish waste	1,000.0
3.	Mung-bean	189.0	Mung-bean hull	83.4
4.	Paddy	115,865.8	Rice bran	11,586.6
5.	Banana	9,441.1	Banana peel (king banana)	127.5
6.	Taro	870.1	Taro peel	11.4
7.	Vegetables	4,345.2	Vegetable waste	546.0
8	Restaurant	152.0*	Restaurant waste	2,056.0

*Yafur (2012) **Department of Statistics of West Papua Province (2012) *** as computed

Quantity and quality of feed ration at each farm

Table 2 presents the information on nutrient evaluation of ration at each farm based on the total ration, crude protein and metabolizable energy (ME) given and required. It was found that the farmers did not provide ration based on the age and pig production phase. All pigs in the farms were given the same ration. Table 2 indicates that some farmers failed to meet total feed requirements, but some of them exceed. Feed quality given by the farmers was varied from one farm to another. All rations formulated by the farmers did not meet the requirements. In order to get an optimal growth, starter pigs require as much as 23.7 to 26.0 % protein, growers require 18 to 20 % protein and finishers require 13.2 to 15.5 % (NRC, 1998).

Table 2. Feed given, nutrient content and required daily feed on each farm.

Farm	Population (head)	Given feed weight (kg/d)		CP* (kg)	ME** (kcal/kg)	Required CP* (kg)	Required ME (kcal/kg) **	Evaluation
		Fresh	Dry					
1	18	217.8	33.22	7.63	126841.38	4.59	80260	CP (+) ME (+)
2	39	94.5	34.49	4.38	112740.43	6.61	126080	CP (-) ME (-)
3	46	43.5	29.96	4.13	96369.82	11.57	214670	CP (-) ME (-)
4	26	137	35.75	3.99	116534.45	5.44	107130	CP (-) ME (+)
5	51	94.5	22.99	3.72	73138.47	11.23	211625	CP (-) ME (-)
6	6	164	35.96	3.14	119854.28	1.52	38505	CP (+) ME (+)
7	39	103	39.48	3.24	127021.14	9.30	191020	CP (-) ME (-)

Recommended feed formulation

Normally animal ration is formulated based on dry weight feeds but in Manokwari, the pig raisers formulated their rations based on fresh feeds. Therefore, the pig rations recommended here was formulated by using commonly available local feeds for daily requirement in dry matter bases but had converted in the form of fresh feeds. The amount of each local feed was computed to meet the protein and ME requirements. Therefore, the recommended rations have the same protein and ME content as required by NRC (1998) in Table 3.

Table 3. Recommended local feed formulation for pig at each growth stage.

No.	Local feed (kg)	Growth stage					
		Pre starter	Starter	grower	Boar/Sow (Non-lactation)	Gestation	Lactation
1.	Fish waste	0.19	0.14	9.28	-	-	-
2.	Tofu by product	1.07	1.95	2.45	1.43	1.79	0.86
3.	Broiler ration CP II	0.28	0.42	0.38	-	-	-
4.	Taro peel	-	0.13	0.37	1.38	21.64	2.29
5.	Vegetable stalks	-	0.18	1.29	1.96	0.93	2.59
6.	Restaurant waste	-	0.18	1.29	1.96	0.93	2.59
7.	Tempe by product	-	-	0.64	1.82	2.73	0.29
Daily requirements (kg fresh weight)		1.53	4.03	8.09	9.25	10.45	12.98
Required Protein (%)		23.7	20.9	18.0	13.0	12.9	16.3
Required ME (kcal/kg)		3265	3265	3265	3265	3265	3265

Summary

Local feeds commonly used by the farmers were: tofu by-product, fish waste, soybean hull, mung-bean hull, rice bran, banana peel, taro peel, market waste (vegetable stalks) and restaurant wastes. These feeds were potential in supporting pig farming development in Manokwari when properly formulated.

References

- BPS Papua Barat, 2012. Papua Barat dalam Angka (West Papua in Number). Department of Statistics of West Papua. Manokwari.
- Iyai, D.A., 2008. Innovation of Pig Keeping System in Pig Farmers at Manokwari, West Papua Province, Indonesia. Tesis. Wageningen University. The Netherlands.
- NRC, 1998. Nutrient Requirements of Swine. 10th Revised Edition. National Academic Press. Washington, D.C.
- Yafur, F. N., 2008. Tingkat Pemanfaatan Limbah Warung Makan Nasi Campur dan Bakso Sebagai Pakan Ternak Babi di Kota Manokwari. Skripsi Sarjana Peternakan Fakultas Peternakan Perikanan dan Ilmu Kelautan Universitas Negeri Papua, Manokwari.

A response of *in vitro* and *in vivo* methane production, nutrient digestibility and rumen parameters of sheep by Cat fish oil (CFO) supplementation

Nguyen, V. T. & T.K.D. Nguyen

Dept. of Animal Sciences, College of Agric & Appl. Biology, Can Tho University, Vietnam

Abstract

Two experiments (Exp) were carried out. The first one was the *in vitro* gas production experiment using 50 ml-syringes in a complete randomized design with 6 treatments and 3 replications. The treatments were 0, 1, 2, 3, 4 and 5 % of Cat fish oil (CFO) in the total substrate dry matter (DM). The substrate used in the experiment was Para grass. The second experiment was a 4 x 4 Latin square design with the growing sheep (19.7 ± 1.43 kg). The treatments were the supplementation levels of Cat fish oil of 0, 1, 2 and 3 % (DM basis) corresponding to the CFO0, CFO1, CFO2 and CFO3 treatment. Each experimental period was for two weeks including one week for dietary adaptation and another one for the sample collection. Methane (CH₄) production was measured over a 24 h period while the sheep were in respiration chambers. The results showed that the *in vitro* CH₄ and CO₂ production from 0-96 h were significantly different ($P < 0.05$) among the treatments (Exp 1), particularly there was a significantly gradual reduction of CH₄ ($P < 0.05$) when increasing CFO levels from the CFO0 to the CFO5 treatment. In Exp 2, methane production was significantly higher ($P < 0.05$) for the CFO2 and CFO3 treatments, while the daily weight gains were significantly different ($P < 0.05$) among the treatments with the highest value for the CFO2. A gradual reduction of CH₄ by increasing the CFO in diets with a linear relationship of $R^2 = 0.89$ was found.

Keywords: ruminant, green house gas, fat supplementation, digestion, rumen environment

Introduction

The World demand for ruminant products is expected to increase due to the increase in the size of the human population and its increasing wealth. The production of ruminant meat and milk is associated with a relatively large environmental impact when compared to other animal products. This is, for a large part, caused by the fact that ruminants produce enteric methane, a greenhouse gas, during the digestion of their feed. Many dietary strategies have been proposed to lower methane production in ruminants, although most of these have only been tested in *in vitro* experiments. In the Mekong delta the production of Tra fish (*Pangasius hypophthalmus*) was about 5,400 ha and 645,000 ton in 2010 (HBS, 2011). As a result the byproduct as the fish oil was also at a big amount. The supplement of fish oil in the animal diets improved the energy feed source and production (Mon Seng, 2001). Some studies stated that the supplementation of fish oil in the ruminant diets reduced methane production. Therefore this study aimed to evaluate methane production, nutrient intake and digestibility and rumen parameters of sheep by fish oil supplementation for the further research on using it as the energy source in the diet and reduction the green house gas emissions.

Materials and Methods

The first experiment was an *in vitro* gas experiment. It was a complete randomized design with 6 treatments and 3 replications. They were 0, 1, 2, 3, 4 and 5 % of CFO in the total substrate (DM basis) by using the syringe system. The substrate used in the experiment was the Para grass. Representative samples were put into the incubation 50 ml- syringes following the method described by Menke et al. (1979). The second one was a 4x4 Latin square design with growing sheep (19.7 ± 1.43 kg). The treatments were the supplementation levels of Cat fish oil of 0, 1, 2 and 3 % (DM basis) corresponding to the CFO0, CFO1, CFO2 and CFO3 treatment. Each experimental period was for two weeks including one week for dietary adaptation and another one for the sample collection. CH₄ production was measured over a 24 h period while the sheep were in respiration chambers for 2 days following the method developed by Bird et al. (2008). Concentrations of CH₄ in chamber were automatically recorded during the measurement period by using Fast Greenhouse Gas Analyzer, USA.

Results and Discussion

The total gas, CH₄ and CO₂ production from 0-96 h were significantly different ($P < 0.05$) among the treatments (Table 1). Particularly there was significantly gradual reduction of CH₄ when increasing CFO levels from the CFO0 to the CFO5 treatment ($y = -2.37x + 55.6$, $R^2 = 0.949$, $p = 0.001$, $SE = 1.15$). The OMD values significantly increased ($P < 0.05$) for the supplemented CFO treatments.

Table 1. *In vitro* total gas, CH₄, CO₂ production (ml/gOM) and organic matter digestibility (OMD) at 96 h supplemented Cat fish oil (CFO) in Exp 1.

Item	Treatment						±SE	p
	CFO0	CFO1	CFO2	CFO3	CFO4	CFO5		
Total gas	351 ^a	316 ^{ab}	288 ^{bc}	393 ^{ac}	298 ^{ac}	270 ^{bc}	11.7	0.007
CH ₄	57.0 ^a	52.3 ^{ab}	49.4 ^{ab}	48.8 ^a	46.4 ^b	44.1 ^b	2.23	0.019
CO ₂	287 ^a	265 ^{ab}	230 ^{ab}	248 ^{ab}	230 ^{ab}	212 ^b	13.9	0.028
OMD, %	68.9 ^a	72.9 ^{ab}	78.9 ^b	76.4 ^b	75 ^{ab}	77.6 ^b	1.31	0.002

^{a,b,c}: Means with different letters within the same rows are different at $p < 0.05$

Table 2. Rumen parameters at 3 hrs after feeding, methane production and daily weight gain of sheep supplemented different levels of CFO in Exp. 2.

Item	Treatment				p	± SE
	CFO0	CFO1	CFO2	CFO3		
pH	6.74	6.80	6.74	6.59	0.28	0.06
NH ₃ -N, mg/100ml	59.5	66.7	61.5	67.9	0.87	8.29
VFAs, μmol/ml	91.2	97.0	97.3	96.4	0.39	2.65
CH ₄ production, g/kgDMI	13.3 ^a	10.2 ^{ab}	9.07 ^b	8.04 ^b	0.021	0.991
Weight gain, g	50.0 ^a	52.5 ^{ab}	60.0 ^b	55.0 ^{ab}	0.032	0.757

^{a,b} Means with different letters within the same rows are different at $p < 0.05$

Daily nutrient intake and digestibilities, and the rumen parameters of sheep were similar among the treatments. Methane production was significantly lower ($P < 0.05$) for the CFO2 and CFO3 treatments, while the daily weight gains were significantly different among the treatments with the highest value for the CFO2. There was also a gradual reduction of CH₄ by increasing the CFO supplementation with a linear relationship of $R^2 = 0.89$ ($y = 12.58x + 12.6$, $p = 0.05$, $SE = 0.89$). Roger (2010) also reported that including 2% fish oil in the diet of cattle reduced

flatulence, apparently due to the omega 3 fatty acids in the oil, and the technique cut methane output of three cows by 21%.

Conclusion and Implications

There was a consistent reduction of CH₄ production investigated between *in vitro* and *in vivo* experiments when supplementing the CFO in the substrates and diets, respectively. No significant difference of nutrient intakes and digestibilities, and rumen parameters of sheep were found in the present study.

References

- Bird, S.H., Hegarty, R.S. and Woodgate, R., 2008. Persistence of defaunation effects on digestion and methane production in ewes. *Aust. J. Exp. Agric.* 48: 152–155.
- HBS, 2011. The analysis of Tra fish production. Documents%20and%20Settings/Admin/My%20Documents/Downloads/HBS_PhanTichNhomCATRA_-_20110729%20(1).pdf
- Gutierrez, D. , 2009. Feeding Fish Oils to Cows Reduces Greenhouse Gas Emissions from Bovine Farts. http://www.naturalnews.com/026708_emissions_fish_methane.html.
- Rogers, S., 2010. Fish Oil Could Control Methane-Packed Cow Farts. *Earth first*. <http://earthfirst.com/fish-oil-could-control-methane-packed-cow-farts/>
- Menke, K. H., Raab, L., Salewski, A., Steingass, H., Fritz, D. and Schneider, W. (1979). The estimation of the digestibility and metabolizable energy content of ruminant feeding stuffs from the gas production when they are incubated with rumen liquor *in vitro*. *J. Agric. Sci.* 93: 217-222.

Water footprint of milk production in Thailand

Boonyanuwat, K.¹ & W. Sirikul²

¹Bureau of Animal Husbandry and Genetic Improvement, Department of Livestock Development, Ratchathevi, Bangkok, 10400, Thailand. ²Pakchong Livestock Research Center. ¹Bureau of Animal Husbandry and Genetic Improvement, Department of Livestock Development, Pakchong, Nakornratchasrima Province, 30130, Thailand

Abstract

The green, blue and grey water demand of a dairy farm plays a pivotal role in the regional water balance. Considering already existing and forthcoming climate change effects there is a need to determine the water cycle in the field and in housing for process chain optimization for the adaptation to an expected increasing water scarcity. Resulting investments to boost water productivity and to improve water use efficiency in milk production are two pathways to adapt to climate change effects. In this paper the calculation of blue water demand for dairy farming in Thailand in 2013 is presented. The water used for feeding, milk processing, and servicing of cows in 1 year was assessed in this study. The results of the calculation of the green, blue, and gray water footprint showed as 93.39, 6.16, and 0.46 % respectively. The total water for 1 kg milk production at farm gate was 366.22 kg. The water footprints of milk processing were 88.05 and 78.03 kg per pack of 200 cc UHT and pasteurized milk respectively. The major part of water footprint in milk production was green water footprint from rain using by forage crop production.

Keywords: milk, water footprint.

**Corresponding author: kalayabo@gmail.com*

Introduction

The overall goal of the strategy plan of climate change in agricultural sector of Ministry of Agriculture and Cooperatives, Thailand for adaptation strategy is to assess the climate change impacts on agricultural farming systems and to identify adaptation measures, investigated on the process chain level in farms. Investigations concerning the water consumption of dairy farms in each dairy cooperatives. Due to climate change and population growth, pathways for reducing the water footprint of food production chains are increasingly sought, but poorly understood. Agricultural production accounted for about 90% of global freshwater consumption during the past century (Shiklomanov, 2000). Even without negative climate change effects, the water consumption for food production will increase to meet demands of a 50% larger global population (UNDP, 2006). Observational evidence from all continents and most oceans shows that many natural systems are being affected by regional climate changes, particularly through temperature increases. Globally the negative impacts of climate change such as more frequent floods, endangered ecosystems and increasing ground instability, are likely to outweigh their benefits (IPPC, 2007). For regional agriculture, more detailed scenarios of the climate development were necessary. The resulting effects depend on current climatic and soil conditions, the direction of change and the availability of resources and infrastructure to cope with change. One adaptation strategy can be the modification of water use efficiency. The efficiency of a specific crop or a specific housing system varies with climate and agronomic

practice (Allen et al., 1998). Regarding crops, the sowing date and plant density, supplemental irrigation, and humus conservation management can be factors to raise water efficiency (Drastig et al., 2010). The water footprint concept can be used as an indicator of water use that looks at both direct and indirect water footprint of the feed crop cultivation, the livestock farming, a food processor, a retailer or a consumer.

Materials and Methods

This assessment is based on the methodology for LCA, Life Cycle Assessment. The assessment encompasses the entire production chain of cow milk, from feed production through to the final processing of milk, including transport to the retail sector,

- 1) cradle to farm-gate and 2) farm-gate to retail.

Water footprint analysis for the assessment of processes in dairy farms and milk processing (Figure 1) are:

Green water use is assumed to be equal to the precipitation and the soil water absorbed by the crop. The cropevapotranspiration requirement will be calculated using different complex approaches.

Blue water use depends mainly on process water and drinking water consumption.

Grey water use depends on process water that causes pollution.

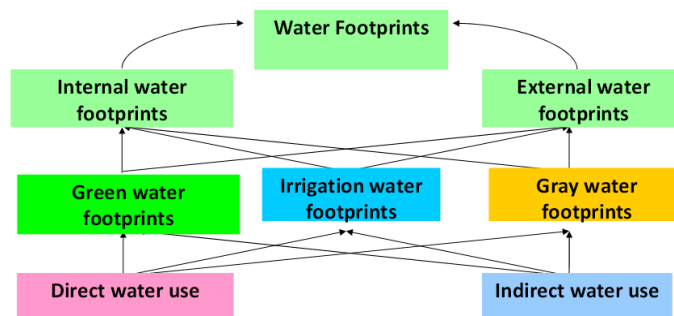


Figure 1. Water Footprint in milk production.

Results and Discussion

Water footprint for milk production

This part of water footprint consisted of forage crop, feed production, housing management, milking, and electricity use. The amount of milk produced in 2013 was about 1,138,661 tons (DLD, 2013). The water footprint from the dairy herd, including from milk production before farm gate are 366.22 kg/kg milk (Table 1).

Table 1 Water Footprint for milk production from farm to gate.

	Water Footprint (ton/year)			
	Green	Blue	Grey	Total
Water Footprint (ton)	386,984,956.50	25,510,262.27	1,887,989.73	414,383,208.50
Milk yield (ton/year)				1,131,500.00
Water Footprint (kg/kg milk)	93.39%	6.16%	0.46%	<u>366.22</u>

The mean blue water consumption for the production of 1 kg milk was 6.16%. We found that the highest blue water consuming process was the supply of the cows with drinking water.

Beside the drinking water consumption of cows, losses from leaky tubes and watering places may cause a huge loss of drinking water.

The results of the presented water consumption used per kg of produced milk show in relation to the literature comparable results. The main part of the consumed water seems to stem from the indirect used green water for the production of the feed for the cows. Hoekstra (2008) calculated for the beef production a major share of 99% used water for the production of the feed. Chapagain & Hoekstra (2004) calculated a world average of 990m³ water per ton of produced milk. If the blue water footprint accounts to only 0.4% of the whole water footprint, the measures to raise water efficiency in milk production will not be focused in this area. In this study the blue water footprint is 6.16% so it should be focused in this area and it can be developed.

Water footprint for milk processing

There are 2 milk products in this study, 200 cc packing of UHT and pasteurized milk for school milk. The water footprint of milk processing were 88.05 and 78.03 kg. per pack of 200 cc UHT and pasteurized milk respectively (Table 2). Water footprints in milk products, they carried water footprint from fresh milk. The sources of water footprint in milk products depended on pollution from processing and transportation. This water footprints could be controlled by production standards. The results in Table 2, the major water footprints were carried by raw milk, and the second were processing. So in milk production chain, it should be focused at processing and dairy farming to control water footprints.

Table 2. Water Footprint for milk processing.

200 cc/pack	Water Footprint (%)			Water Footprint (kg/Pack)
	raw milk	transportation	processing	
UHT	97.87	0.03	2.10	88.05
Pasteurized	98.43	0.04	1.54	78.03

References

- Allen, R.G., L.S. Pereira, D. Raes and M. Smith. 1998. Crop evapotranspiration: Guidelines for computing crop water requirements. FAO Irrigation and Drainage Paper. 56 Rome. Italy. Food and Agriculture Organization of the United Nations.
- Chapagain, A.K. and A.Y. Hoekstra. 2004. Water footprint of nations. UNESCO-IHE.
- DLD, 2013. Livestock Statistic. Available: from: <http://ict.dld.go.th/th2/index.php/th/report>, May 1, 2014.
- Drastig, K., A. Prochnow and R. Brunsch, 2010. Wasser management in der Landwirtschaft. Berlin Brandenburg Academy of Sciences and Humanities. Discussion Paper. 2010 (in German).
- Hoekstra, A.Y., 2008. The water footprint of food in: Water for food. Edited by: Forare, J., The Swedish Research Council Formas.
- IPCC. 2007. Fourth Assessment Report (AR4). in: Working Group II Report "Impacts, Adaptation and Vulnerability". Summary for Policy makers. Inter governmental Panel on Climate Change (IPCC). 939pp.
- UNDP. 2006. Human Development Report 2006. Beyond Scarcity: Power. Poverty and the Global Water Crisis. New York. United Nations Development Programme. 422pp.
- Shiklomanov, I. 2000. Appraisal and assessment of world water resources. Water Int. 25: 11-32.

Biogas unit, an alternative solutions for reducing green house gas effect of animal waste

Mochammad Junus

Universitas Brawijaya, Faculty of Husbandry

Abstract

Animal waste is not only a major source of CO₂, a green house gas (GHGs), but also a good source of manure and in 2014 a national survey in Indonesia estimated that its animal husbandry produced 180,000,000 tons of waste and 187,000,000 m³ of CO₂ annually. It is imperative to find ways to reduce this and one way is to increase manure production and thus crop growth to take up as much CO₂ as possible. Therefore, the aim of study was to find ways of improving and speeding up manure production. They study was enacted in Bantur district, Malang Indonesia between June and November 2014. A randomized full design was employed with 5 treatments and 4 replications. Sludge was mixed with water to speed up anaerobic fermentation. Straw was added as fellows 1:1, 1:2, 1:3, 1:4 and 1:5, respectively. In conclusions biogas units can reduce GHGs by managing CO₂ production through anaerobic metabolism and the reabsorbs ion of CO₂ through encouraging more flourishing crop growth.

Keyword: solid, sludge, organic, livestock, fish

Corresponding author: yunusbrawijaya@ub.ac.id

Background

Animal waste (manure) is organic matter mostly used only for organic fertilizer. Composing process of organic fertilizer for plant is taking quite long time, approximately three months. The highly needs of organic fertilizer for plant per area is caused by the small amount of nitrogen contained in it. However, the existence of synthetic fertilizers to replace organic fertilizer decrease the using of manure as fertilizer.

The use of synthetic fertilizers will provide nutrient which is quickly absorbed by plants. However, there are more nutrients inside organic fertilizer made from manure or other organic waste compared with synthetic fertilizer. Frequent use of synthetic fertilizer will cause the plants lack certain nutrient during the growth of animal or human. As the result, the balance of nutrient inside fodder or food becomes imbalance. The imbalance amount of nutrient causes many metabolism diseases and increase protracted lack of fodder or food.

Moreover, the existence of wasted animal waste on the land without water involvement will create kinds of GHGs such as CO₂, N₂O, NO₂, NO_x and CH₄. Then, animal waste which is thrown away into the water will create GHGs such as CO₂ and CH₄. It shows that the existence of animal waste both in the water and on the land creates various kinds of harmful GHGs. Based on that fact, FAO summarized that the animal waste creates GHGs with 51% (Mayer, 2006).

CO₂ and CH₄ from biogas unit can be burnt in stove, lamp etc. The combustion result of CO₂ still produces CO₂ while CH₄ produces CO₂ and water. Solid sludge biogas unit (SSBGU) in the form of solid contains many nutrition, while the liquid sludge biogas unit (LSBGU) contains many nutrients. The right use of SSBGU as fodder will minimize the production of NO₂, N₂O or NO_x, while the LSBGU will not produce CO₂ or CH₄. Furthermore, the use of

LSBGU will improve plant production between 100 – 400%. The ability of LSBGU in improving the plant production will also improve CO₂ absorption so GHG's can be completely absorbed in order to create environment balance.

The Potential of Animal Waste (Manure) in Indonesia 2014 in Producing GHGs

People in Indonesia who work both as farmers and breeders always use their animal waste as organic fertilizer. Meanwhile, people who only work as breeders tend to ignore the animal waste though they know that animal waste can be used as organic fertilizer for plants. There is no much animal waste available in society, in 2014 there was only 494,861.00 tons/day or 180,558,565.00 tons/year. It can produce 1,577,029.00 tons/day or 575,615,585.00 tons/year organic sludge using biogas unit (Junus, 2013; Junus, 2015).

The Potential of Animal Waste (Manure) in Producing Livestock/Fish Fodder and Reducing GHGs

Based on the research in organic sludge biogas unit (OSBGU) taking, after 80 kg of sludge were dripped, there were 35kg of wet solid (43%) and 50 liters of LSBGU (62%). Then, after it was drained there were 7 kg of SSBGU (8.75%). Therefore, the estimation result from each dry solid and liquid fertilizer were 50,366,363.69 ton/year and 356,881,662.7 ton/year.

The Ability of Organic Sludge Biogas Unit (OSBGU) as Organic Matter Decomposer

Organic sludge biogas unit is organic matter from various microorganisms remains. It makes the sludge contains much nutrition and nutrient. Furthermore, the others organic matters also contain many inactive microorganisms. If organic matters such as straws and agricultural waste are mixed with organic sludge, the remains microorganism will actively decay organic sludge and the straws or agricultural waste. As the result, the decomposition process of organic matters will happen faster.

Compost Ingredients

Compost change from straws mixed with organic sludge biogas unit decomposer (LOUGB) physically showed the pile extent change, biological changes included temperature inside the compost and straws decay, while chemical changes included the increase of PH, organic carbon, total of nitrogen, C/N, organic source, phosphor, potassium and magnesium.

Conclusion and Suggestion

Conclusions

The conclusions are summed as follows:

1. GHGs production from animal waste (manure) in Indonesia using biogas unit is quite high, in the range of 74,830,037 m³ up to 112,245,055.5 m³/year CH₄
2. Organic sludge biogas unit is able to fasten the process of compost making that can be used as plant fertilizer to absorb CO₂ from biogas unit or combustion result of CH₄, in the range of 300%-500% or 400%.

Suggestions

1. Utilizing organic sludge from biogas unit is needed to make livestock/fish fodder in order to control the production of GHGs from animal waste.

2. Biogas unit technology making by optimizing its production is needed to fasten the absorption of CO₂ from animal waste.

Acknowledgements

The researcher would express his profound sense of reverence to the Head of LPPM University of Brawijaya and the Dean of Faculty of Animal Husbandry for entrusting the researcher to do a research and seminar.

References

- Agustina, Junus, M. and Rustidja, 2007. Fish Livestock Production Systems Organic Vegetable based on organic waste (waste bio gas unit). State Ministry for Research and Technology. Faperta-diffusion of Research and Technology.
- Idris, S. Junus, M, and K. Sukesi, 2007. Role of Women Farmers of the energy supply of dairy cattle waste in Pujon Malang. State Ministry for Research and Technology. PSW-diffusion of Research and Technology.
- Junus, M., 2010. Practical Guidelines. The Making and Utilization of Integrated Biogas Unit. Biogas Unit Development Programme UNDP-PT. Earth Harmony Indoguna, Indonesia.
- Junus, M., 2011. Development Acceleration of Biogas Unit for Livestock Development in Indonesia. Inauguration Speech Professor Position in the Field Production Science Arts Faculty of Animal Husbandry At UB.
- Junus, M. and S. Minarti, 2012. Existence of biogas units in the Community in supporting roadmap of Desa Mandiri Energy based on livestock biogas units Coordinating Ministry for Economic Affairs-Faculty of Animal Husbandry, UB.
- Junus, M. Agung, W. Wahyono and Zamrudy, 2014. Management and Use of animal waste into organic products. Management and Use of animal waste into organic products.
- Junus, M., 2013. Bioenergy sources from livestock waste by using biogas system in small scale farms. In data inventory and mitigation on carbon emission and nitrogen recycling from livestock in Indonesia. Jakarta IAARD Press.
- Junus, M., 2013. Reduction of green house gas using biogas integrated livestock unit (BGU). The 2nd Animal Production International Seminar. Sustainable livestock production based on local resources in the era of global climate changes: Prospects and chalanges. 2nd APIS-2013. University of Brawijaya. Malang.
- Junus, M. and S. Minarti, 2013. The appearance and the conversion of plastic biogas units in Indonesia. The 2nd Animal Production International Seminar. Sustainable livestock production based on local resources in the era of global climate changes: Prospects and chalanges. 2nd APIS-2013. University of Brawijaya. Malang.
- Kawashima, T. Invenory the data and mitigation activities of carbon gas and nitrogen cycling from livestock in developing countries: Asia case as an example. In data inventory and mitigation on carbon emission and nitrogen recycling from livestock in Indonesia. Jakarta IAARD Press.
- Kristanto, A. 2011. Analysis in burning process of natural gas <http://aqunqkristanto.wordpress.com/2011/01/24lanalisis-proses-pembakaran-bahan-bakar-qas-alamnatural-qas-2/> accessed on 7/4/2013
- Meyer. L., 2006. The IPCC Special Report on Carbon dioxide Capture and Storage EU-OPEC Roundtable on Carbon Dioxide Capture and Storage Riyadh, Saudi Arabia (CCS) IPCC Working Group III
- Werner, U., U. Stohr and N. Hees, 1989. Biogas in Animal Husbandry. Friedr. Vieweg & Sohn Braunschweig / Wiesbaden.

Development of Farmer Champions and their Role in Progressing Smallholder Beef Production in Vietnam

Lydia Turner*¹, Jeffrey Corfield², Nguyen Thi Da Thao³ and Lawrence Bonney⁴

¹Tasmanian Institute of Agriculture, University of Tasmania, Burnie 7320, Tasmania, Australia, ²Corfield Consultants, Townsville 4810, Queensland, Australia, ³Hue University of Agricultural and Forestry, Hue City 47000, Vietnam, ⁴Tasmanian Institute of Agriculture, University of Tasmania, Hobart 7001, Tasmania, Australia

Abstract

‘Farmer Champions’ have an important role in promoting adoption of best practice recommendations in the communities that participate in international research for development. This paper draws on data from in-depth interviews with three Farmer Champions. It describes how the role of these Farmer Champions developed and how they have progressed beef smallholder production in Cat Trinh, a commune of the Vietnamese South Central Coastal province, Binh Dinh. Farmer Champions firstly adopt or adapt study recommendations to improve their own farming systems. They are subsequently recognised as valuable sources of knowledge and resources by surrounding farmers. Farmer Champions then facilitate significant spread (or ‘scale out’) of improved practices to other farmers. Within their community this was found to occur primarily through informal knowledge transfer pathways, while their involvement in formal pathways extended benefits to farmers outside their community. Understanding the development of Farmer Champions and how they influence scale out provides information to guide knowledge transfer methods used in future research for development, and further improve adoption of best practice recommendations.

Keywords: adoption, beef production, knowledge transfer, smallholder farmer, Vietnam

**Corresponding author: Lydia.Turner@utas.edu.au*

Introduction

In response to the growing domestic demand for beef in Vietnam, and the constraints on smallholder farmers to meet this demand (Ba *et al.* 2013), a recent ACIAR study (2009-13) focused on improving the productivity of beef smallholders in Cat Trinh, a commune of the Vietnamese South Central Coastal province, Binh Dinh. In this study, groups of 15 farmers worked alongside researchers in a participatory approach that is described in Lisson *et al.* (2010). This approach builds on the farmers’ existing knowledge and practices, and is an effective way to facilitate on-farm practice change and increase adoption of best practice recommendations. Farmers that adopted many of the forage and cattle management recommendations and then championed the importance of their new knowledge and practices became known as Farmer Champions (FCs). Collection of quantitative data within the previous study demonstrated the importance of these early adopters in the subsequent scale out of recommendations to other farmers (Khanh *et al.* 2014). The participatory approach to research for development continues on the South Central Coast of Vietnam through a current ACIAR study (2014-17). This study aims to further improve the efficiency and profitability of beef smallholders and requires an understanding of local knowledge transfer pathways to maximise impact. Understanding how the role of FCs developed and how they have progressed beef cattle smallholder production in Cat Trinh provides information to guide future extension methods, with a goal of further improving adoption of best practice recommendations.

Methods

The development of FCs and their role in progressing smallholder beef production was explored using a Case Study approach. Semi-structured interviews were carried out with three FCs in Phu Kim village of Cat Trinh commune in April 2015. Farmer Champions were selected on the basis of their involvement in the previous study and their names have been abbreviated in this paper to maintain anonymity. The interviews focused on changes to their farming systems since their initial involvement in the previous study, the extent and patterns of scale out they have facilitated, and the methods of knowledge transfer they have been engaged in. Themes that occurred across the interview data were manually coded; they focused on the FCs developing through the following stages as: 1) Adopters; 2) Sources of Knowledge and Resources; and 3) Facilitators of Knowledge Transfer.

Findings

Farmer Champions as Adopters

Some of the key practices that the FCs have implemented as a result of the knowledge gained through the participatory process are summarised in Table 1. Utilising new forage species with improved quality, productivity and persistence, and increasing the quantity of cultivated forages for cut and carry, have reduced the requirement for cattle to graze marginal common land. Mr T emphasised the importance of his new knowledge and increased reliability of the feed source for his cattle, “now Mulato is always available in the garden...I can control the feed source in wet and dry seasons when native grass, rice straw and other crop residues may be scarce.” Although cattle numbers have not changed, there has been a change from mainly cattle keeping (involving opportunistic sales when money is needed) to more efficient systems that involve regular sales. The three FCs indicated how important cattle production has become to their overall farming system by ranking it first of a number of farm activities, in terms of contributing to income and planning for future expansion.

Table 1. Changes implemented by Farmer Champions between 2010 and 2015.

Change in Practices	Mr T	Mr X	Mr P
<i>Grazing in 2010 (hrs/month)</i>	200 hours/month	240 hours/month	240 hours/month
<i>Grazing in 2015 (hrs/month)</i>	120 hours/month	120 hours/month	160 hours/month
<i>Cultivated forages in 2010</i>	250m ² local King grass	‘Some’ local King grass	500m ² local King grass
<i>Cultivated forages in 2015</i>	>100m ² VA06 King grass 1000m ² Mulato	500m ² Mulato 200m ² Panicum 150m ² Paspalum 30 Leucaena trees	400m ² Mulato 200m ² Panicum

Farmer Champions as Sources of Knowledge and Resources

The benefits that the FCs experienced through adopting the study recommendations led to them becoming known as valuable sources of knowledge and resources. The benefits observed by neighbours, relatives, friends, acquaintances and service providers included:

- Increased confidence about feeding and managing cattle due to new knowledge;
- Increased availability of labour due to decreased requirement for grazing cattle;
- Decreased costs due to decreased requirement to buy crop-residues for feeding cattle;
- Increased convenience and reliability of cattle feed supply due to cultivating new forages close to the home;
- Improved cattle condition due to improved management of their feeding;
- Increased financial security due to more regular income from cattle sales.

Farmer Champions as Facilitators of Knowledge Transfer

The three FCs have collectively provided the knowledge and resources to increase the efficiency and profitability of an estimated 60 Scale Out Farmers (SOFs) since 2011, through informal and formal knowledge transfer pathways. A key to becoming effective facilitators of knowledge transfer was their willingness to respond to requests and initiate helping other farmers. It was common for there to be multiple visits between farms as SOFs developed questions around the next stage of adoption. The three FCs estimated that 90% of their knowledge transfer occurred informally through social interaction with other farmers and visits between the smallholder farms. An example of this facilitation flowed on from a conversation between Mr T and Mr K at a commune event, about difficulties Mr K was experiencing feeding cattle during the wet season. After the event Mr T immediately took Mr K to his house to provide him with 10kg of forage cuttings and key advice about how to manage them to ensure a reliable feed supply. Similarly, Mr X described how a visit to his farm changed the life of Mrs M, who was running a small market business and did not have enough time at home to care for her three children. After observing Mr X's successful cattle production, Mrs M sought from him the knowledge and free forage resources needed to raise cattle and improve the wellbeing of her household. The FCs demonstrated a common desire to help improve livelihoods in their communities by providing forage resources and time to share advice. Their reputation as experts led to formal requests to be involved in other cattle-related projects, developing commune policy around forage and cattle management, and organising the collection of large quantities of forage resource for other communities.

Conclusion

Farmer champions naturally emerge by confidently applying new knowledge, being seen to experience the benefits of continually improving their practices, and generously sharing their knowledge and forage resources with other farmers. This case study emphasised the importance of smallholder farmers observing the benefits from implementing recommended practices before initiating system changes, and their need for continued access to advice throughout the adoption process. These features can be lacking in traditional extension activities where pre-determined recommendations are delivered in one-off events. The FCs have progressed the development of smallholder production in their communities primarily through informal knowledge transfer pathways, with their ongoing availability ensuring the quality of scale out. Millar and Connell (2010) suggest that both informal and formal knowledge transfer pathways are required to maximize impact. Indeed, the role of FCs in formal knowledge transfer pathways (which followed their natural establishment as successful facilitators), increased the quantity of scale out by extending their sphere of influence outside the local community. Further research should now assess how to maintain the quality of scale out from FCs involved in these formal extension methods.

Acknowledgements

We wish to acknowledge ACIAR research funding through project LPS/2012/062.

References

- Ba, N. X., Lane, P., Parsons, D., Van, N. H., Khanh, H. L. P., Corfield, J. & Tuan, D. T. 2013. Forages improve livelihoods of smallholder farmers with beef cattle in South Central Coast Vietnam. *Tropical Grasslands Forrajes Tropicales*, 1:225-229.
- Lisson, S., MacLeod, N., McDonald, C., Corfield, J., Pengelly, B., Wirajaswadi, L., Rahman, R., Bahar, S., Padjung, R., Razak, N., Puspadi, K., & Dahlanu. 2010. A participatory, farming systems approach to improving Bali cattle production in the smallholder crop–livestock systems of Eastern Indonesia. *Agricultural Systems*, 103:486-497.

- Khanh, H., Ba, N. X., Van, N. H., Corfield, J., Parsons, D., Tung, H. V., Vy, L. V., Nghi, N. V. & Tuan, D. T. 2014. Influence of Labour Saving in Uptake of Improved Forage Technologies by Smallholder Farmers in South Central Vietnam. Proceedings, 16th AAAP Congress, Jogjakarta, Indonesia, November 2014.
- Millar, J. & Connell, J. 2010. Strategies for scaling out impacts from agricultural systems change: the case of forages and livestock production in Laos. *Agriculture and Human Values*, 27:213–225.

The assessment of cattle and palm oil plant integration system in West Sumatera, Indonesia

*Abdullah Bamualim**, *Hardiyanto and Ratna A. Dewi*

Assessment Institute for Agricultural Technology of West Sumatera (BPTP Sumatera Barat), Indonesia.

Abstract

A study has been undertaken in 2014 to assess the local cattle production using palm oil plant by products in a integration system in West Sumatera, Indonesia. The expected output was the utilization of feed technology and the utilizing of the organic fertilizer to increase production of oil palm plantation by based on oil palm plantation-cattle integration system. The study used 40 head of Pesisir cattle with the age of 2-3 years old, consisted of 36 females and 4 males. The pen using the group pen based on the Grati model. The feeds given to the animals consisted of 60% oil palm frond sillage, 25% palm kernel cake, 10% molasses, plus 1 kg of fermented rice straw and 2,5 kg/head/day of fresh grass. Cattle manure was used as organic fertilizer for oil palm plants. The parameters observed include: feed consumption, animal growth, animal reproduction, animal manure produced, and palm oil fruit production. The results indicated that there was a potential to utilize oil palm plantation by-products for local cattle feed resources. The body weight gains of both heifers (0.19 kg/hd/day) and young bulls (0.085 kg/hd/day) were relatively small as the animals of small breed. Out of 36 cows, 26 head were pregnant (72%). The results from organic ferlizer applicaton indicated that the oil palm fruit production was increased by 50%. It is hoped that the research will accelerate the integration system of local beef cattle raise under the oil palm plantation in West Sumatera.

Keywords: Local beef cattle production, Feeds, Oil palm plant by-products, Organic fertilizer, Oil palm fruit production

**Corresponding author: abamualim@yahoo.com*

Introduction

In general, feed supply is the main constraint due to the low quality of forages and lack of manpower to provide adequate feeds to the animals. The nutritional limitation is more obvious in lowering animals condition and growth rate of young animals, increasing mortality rates, reduce the reproductive rates, and hence reduce the cattle population.

One of the solutions to provide high quality feeding is the use of agricultural by products. This option is the best choice to anticipate the shortage of feed as native grasses supply. The oil palm plantation, with its diverse products and by-products, offers opportunities to increase animal production. Oil palm cultivation is rapidly expanding in Indonesia and become the leading producer of oil palm in the world with the plantation of more than 10 million hectares. Within the Indonesia region, the largest plantation is found in Sumatera island, whereas West Sumatera alone contributed some 457.000 ha.

One of the by products is oil palm fronds, accounted for more than 1.07 million tons which could support to feed 595,000 head of cattle in West Sumatera. Palm kernel cake (PKC)

was available at 25,000 tons and potentially to feed 25,000 head of cattle (Buharman, 2011). In addition, organic fertilizer from cattle manure can be applied to increase oil palm production.

A study was conducted in West Sumatera used the Pesisir cattle, a small local cattle breed, which dominated along the coastal areas. The adult body weight of only 150-200 kg/head, however its population contributed some 20% of beef cattle population in West Sumatera (Bamualim *et al.*, 2006). The research was aimed to accelerate the adoption of crop-livestock integration system using oil palm by-products to support beef cattle production.

Materials and methods

The study was conducted in 2014 at West Sumatera Province, Indonesia. The cattle used in this study were 40 head of Pesisir cattle of 2-3 years old. The cattle consisted of 36 heifers and four young bulls, however only two bulls were used to mate the heifers in two communal pens. The initial body weights were 113 kg/head for heifers and 140 kg/head for young bulls.

The pen consisted of two separated group pens using the Grati model, where the animal's dung was left on the floor for three months before taken out. Each group pen accommodated 20 head of cattle and the animals were weighed every month.

The feed used was oil palm fronds silage consisted of 60% oil palm fronds, 25% PKC, 10% molasses and 5% rice bran mixed evenly then fermented in plastic bags for two weeks before giving to the animals. Fresh grasses and fermented rice straw were also given daily. The amount of feed given: 5 kg of fronds silage, 2.5 kg of fresh grass and 1 kg fermented rice straw per head daily. The parameters measured include bodyweight changes and pregnancy rates.

To evaluate the plant production, cattle manure was applied on oil palm plant at the rate of 50 kg/tree/year. The animal manure produced and oil palm production were also measured.

Results and discussion

1. Animal Production

The results of body weight changes and the average body weight gains from May to November, 2014, is shown in Table 1. The body weight changes were relatively small as the animals used were small breed, as reflected by very small body weight gains of both heifers and young bulls.

Tabel 1. Monthly body weight changes and the average body weight gain from May to November 2014.

Items	Monthly bodyweight (kg/head)						Average bodyweight gain (g/hd/day)
	I	II	III	IV	V	VI	
Heifers (36 head)	113,1	117,1	117,9	127,7	135,4	135,8	190
Young bulls (4 head)	139,9	141,4	142,4	144,3	154,5	151,0	85

The body weight gains were the indicators that the feeds derived from oil palm by products contained adequate nutritional derivatives to meet animal requirements. The PKC supplementation was considered to be the main affect to the animal performances. The proximate analyses of PKC showed that its protein content is 15-17% and the metabolize energy (ME) for ruminants is 10.5-11.0 MJ/kg which is considered to be suitable for most ruminants (Bamualim and R.A, Dewi, 2014). It appears that the PKC supplementation in this research tended to improve the body weight gains. Much of the response to be influenced by the increased in dry matter intake which may be stimulated by increased supply of protein. Based on the nutrient requirements of ruminants in developing countries (Kearl, 1982), the estimated intake of protein (7.7%) and TDN (63.2%) was adequate to support a positive growth rates for the animals.

The pregnancy rate was 72%, as 26 head out of 36 head of heifers were pregnant. The reproduction rate was relatively high compared to other breeds in Indonesia (Bamualim *et al.*, 2006).

2. The Affect U of Manure for Oil Palm Production

One of the benefits derived from the crop livestock integration system was the utilization of manure as a source of fertilizer. The amount of cattle manure from 40 cattle measured in two collection periods, in three months interval, yielded 10 and 16 t of dry manure respectively.

In this study, the manure was used to fertilize the oil palm trees. The results indicated that providing organic fertilizer and anorganic fertilizer increased palm fruits production by 50% as the production increased from 1,200 to 1,800 kg/harvest/ha. In general, there was a positive result of providing organic fertilizer in order to increase the oil palm fruit production. Similar research at other locations also showed the increasing trend of oil palm fruit production by organic fertilizer application (Bamualim *et al*, 2015, in press).

Conclusions

There was a positive effect of providing feed based on the oil palm by products to increase cattle production and reproductive in West Sumatera. In addition, the production of oil palm fruits was increased by 50% with the implementation of organic fertilizer above the chemical fertilizer.

It is recommended that utilization of oil palm by products as a potential feed for beef cattle should be always disseminated to stakeholders of oil palm plantation in West Sumatera.

Acknowledgments

The authors wish to thanks the Agency of Agricultural Research and Development, Indonesian Agriculture Ministry for the financial support of this study. The help of the technicians during the trial is also acknowledged.

References

- Bamualim, A., Wirdahayati, dan Marak Ali. 2006. Profil Peternakan Sapi dan Kerbau di Sumatera Barat. Balai Pengkajian Teknologi Pertanian (BPTP) Sumatera Barat.
- Bamualim, A. and Ratna A. Dewi. 2014. The effect of palm kernel cake supplementation on the body weight gain of local beef cattle fed grass, rice straw and oil palm frond basal diets. Proceedings of the 16th AAAP Animal Science Congress Vol. II 10-14 November 2014, Gadjah Mada University, Yogyakarta, Indonesia. pp. 1754-1757.
- Bamualim, A., F. Madarisa, Y. Pendra, E. Mawardi and Asmak. 2015. The crop-livestock integration innovation using palm oil by-products technology to support beef cattle production in West Sumatera. Journal of Anim. Sci. published by Animal Husbandry Faculty, the Andalas University in Padang, Indonesia (*in press*).
- Buharman, B. 2011. Pemanfaatan teknologi pakan berbahan baku lokal mendukung pengembangan sapi potong di Provinsi Sumatera Barat. Wartazoa Vol 21, No.3:133-144.
- Kearl, L.C. 1982. Nutrient Requirements of Ruminants in Developing Countries. International Feedstuffs Institute, Utah state University, Logan, Utah, USA.

Seasonal changes in pasture and animal productivity of Korean native goats grazed at different pasture type

S.Y. Jang¹, B.T. Jeon¹, J.H. Park¹, M.R. Oh¹, S.W. Kim² & S.H. Moon¹

¹Division of Food Bio Science, Konkuk University, Chungju, 380-701, Republic of Korea

²National Institute of Animal Science, RDA, Namwon, 331-801, Republic of Korea

Abstract

The objective of this study was to investigate seasonal changes in forage and livestock productivity of Korean native goats grazed at different pasture types. Four farms which have a certain size of animals (above 200 heads) and pasture (above 10 ha) were selected as experimental farms. Dry matter yield of pasture and average daily gain of goats were seasonally evaluated. Growing goats of farm A, C, D were grazed at rangeland and those of farm B were grazed at pasture. Forage productivity was high at farm B in spring and at farm A, C, and D in summer. Nutritive value of pasture was higher at farm B than at farm A, C, and D. Goats of all farms increased consistently body weight and daily gain of goats was high at farm A and C and was low at farm D. These differences were mainly attributed by differentiation of seasonal forage productivity and nutritive value between pastures. Therefore, it is greatly demanded that rangeland would be changed or improved to pasture type for effective grazing system of goats.

Keywords: goat, grazing, pasture, seasonal changes

Introduction

Korea is a mountainous country with 64 percent of its terrain consisting of hills and peaks (Kim, 2002). Goats differ from other ruminants their feeding habits, and they appear to have more efficient in digestion of crude fiber and being better utilizers of poor roughages (Gihad et al., 1980). Hence, goats are highly adaptable with good production potential in the Korea. Also, the weather in Korea is unique with four distinct seasons. However, little information is available on the seasonal changes in forage and livestock productivity at different pasture types. Consequently, the objective of this study was to investigate seasonal pasture and animal productivity in Korean native goats grazed at different mountainous pastures.

Materials and Methods

Animal productivity in mountainous pasture

Field studies were conducted from May to October 2014 to determine the seasonal pasture and animal productivity. Four farms which have a certain size of animals (above 200 heads) and pasture (above 10 ha) were selected as experimental farms and dry matter yield of pasture and average daily gain of goats were seasonally evaluated. Growing goats of farm A, C, D were grazed at rangeland and those of farm B were grazed at pasture.

Chemical analysis

Samples were analysed for crude protein (CP), ether extract (EE), crude fiber (CF) and ash according to the methods of the Association of Official Analytical Chemists (AOAC, 1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed by the procedures of Van Soest et al. (1970).

Statistical analysis

Data were expressed as means and standard errors and were statistically analysed with Duncan's multiple range tests using the SAS package (2008) general linear models (GLM) procedure.

Results

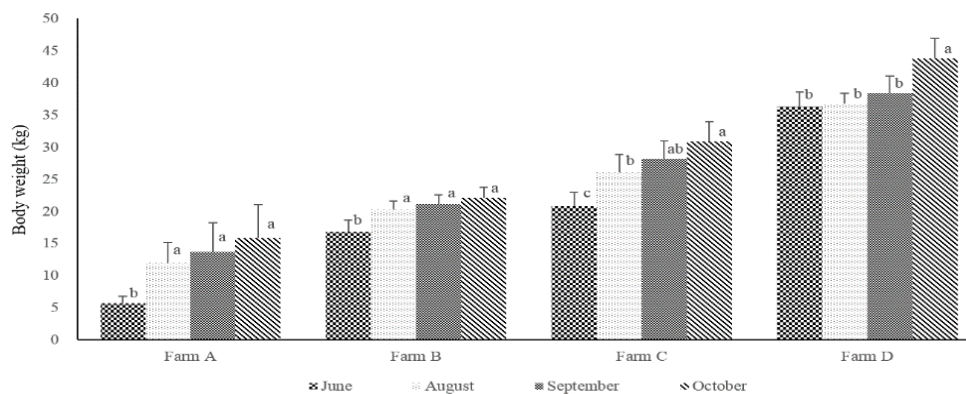
Nutrient composition at pasture

The seasonal chemical composition of the samples produced at experimental pastures is shown in Table 1. The dry matter content was not seasonally different at pasture, but it was changeable from spring to autumn at rangeland. The crude protein content increased in autumn at pasture ($P<0.05$), and decreased in summer and autumn compared with spring at rangeland. The crude fiber content was lower and the ether extract was higher at pasture compared with range land.

Table 1. Seasonal changes in chemical compositions at different pastures grazed by Korea native goats.

Season	Farm	DM	CP	CF	EE	NDF	ADF	Ash
		%				% in DM		
Spring	Farm A	17.59±0.58 ^c	12.47±0.43 ^a	20.61±0.61 ^b	3.50±0.06 ^c	44.22±0.35 ^c	47.65±0.04 ^a	7.23±0.08 ^a
	Farm B	21.93±1.35 ^b	11.71±0.30 ^b	20.84±0.17 ^{ab}	4.85±0.26 ^a	49.68±1.61 ^b	29.45±0.65 ^d	4.43±0.35 ^c
	Farm C	20.64±0.39 ^b	11.37±0.10 ^b	16.48±0.48 ^c	4.34±0.20 ^b	40.08±0.60 ^d	31.36±0.46 ^c	6.01±0.27 ^b
	Farm D	26.81±0.62 ^a	11.05±0.45 ^b	21.75±0.78 ^a	4.24±0.40 ^b	54.57±0.38 ^a	41.92±0.48 ^b	5.59±0.32 ^b
Summer	Farm A	16.42±0.72 ^d	10.75±0.39 ^a	20.73±0.88 ^b	4.50±0.52 ^{bc}	52.04±1.01 ^a	42.12±2.05 ^b	7.55±0.07 ^a
	Farm B	21.45±2.08 ^c	10.70±0.19 ^a	22.35±1.08 ^b	5.14±0.21 ^a	54.80±0.83 ^b	39.46±0.41 ^{bc}	5.23±0.19 ^c
	Farm C	26.97±2.94 ^b	11.14±0.59 ^a	21.04±2.57 ^b	4.58±0.16 ^{ab}	51.39±1.15 ^c	37.40±0.60 ^c	6.16±0.17 ^b
	Farm D	39.03±3.41 ^a	6.64±0.14 ^b	26.32±1.37 ^a	3.95±0.24 ^c	70.30±0.92 ^c	52.89±1.82 ^a	3.49±0.14 ^d
Autumn	Farm A	20.06±4.33 ^c	8.83±0.30 ^b	29.16±0.81 ^c	4.21±0.71 ^b	51.55±0.46 ^c	46.34±0.36 ^b	8.00±0.18 ^a
	Farm B	21.48±1.37 ^c	14.03±0.16 ^a	26.98±0.97 ^d	5.19±0.12 ^a	54.89±0.26 ^b	37.34±0.61 ^c	6.93±0.16 ^b
	Farm C	35.03±6.41 ^b	8.92±0.53 ^b	31.36±0.52 ^b	4.03±0.23 ^{bc}	55.86±0.88 ^b	45.76±0.69 ^b	6.86±0.16 ^b
	Farm D	45.37±3.26 ^a	5.80±0.80 ^c	34.53±0.20 ^a	3.37±0.09 ^c	72.03±0.55 ^a	60.52±2.39 ^a	4.39±0.08 ^c

^{a-d} Means with different superscripts in the same column are different ($p<0.05$).

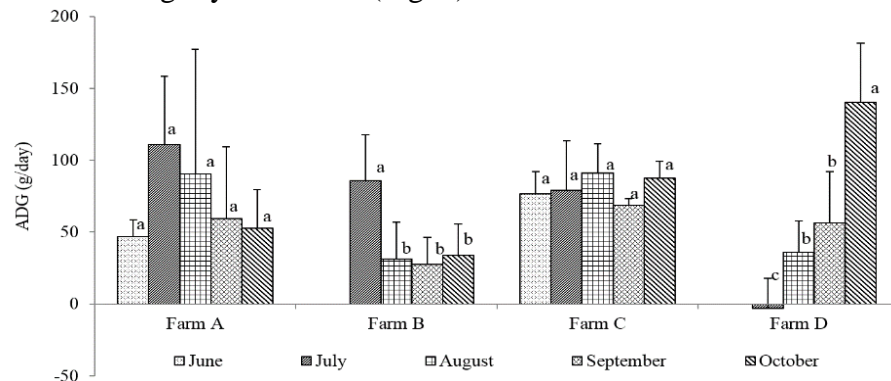


^{a-c} Means with different superscript in the same vertical bars are different ($P<0.05$)

Figure 1. Seasonal changes in body weight of Korean native goats grazed at different type.

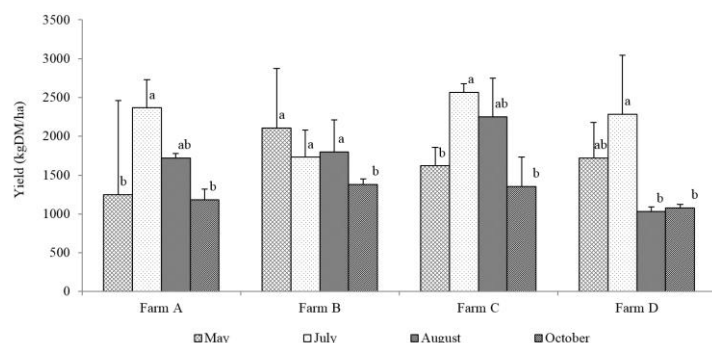
Body weight, ADG and forage productivity

Goats of all farms increased consistently body weight and daily gain of goats was high at farm A and C and was low at farm D (Fig. 1). Average daily gain of goat was different depending on forage productivity and pasture type (Fig. 2). The forage productivity of rangeland was the highest ($P<0.05$) in summer and decreased in autumn; but that of pasture was the largest ($P<0.05$) in spring and had relatively stable productivity with minor seasonal differences, although it decreased slightly in autumn (Fig. 3).



^{a-c} Means with different superscript in the same vertical bars are different ($p<0.05$)

Figure 2. Seasonal changes in average daily gain of Korean goats grazed on different pastures.



^{a-c} Means with different superscript in the same vertical bars are different ($p<0.05$)

Figure 3. Seasonal changes in forage productivity of different pastures grazed by Korean goats.

Conclusion

Goat farming has very different characteristics according to pasture type and seasons. Seasonal goat production was different depending on forage productivity and pasture types. Consequently, the lowered forage productivity at rangeland compared with pasture means that rangeland needs to be changed or improved to a pasture type that provides a more effective grazing system for goats.

References

- A.O.A.C., 1990. Official methods analysis. Association of official analytical chemists. 15th edition. Washington, D.C.
- Gihad, E. A., T. M. El-Bedawy and A. Z. Mehrez., 1980. Fiber digestibility by goats and sheep. *J. Dairy Sci.* 63: 1701.
- Kim, T.H., Sung, K.I., Kim, B.W., 2002. Sustainable livestock production in hill an woodland. Forage production and environment in 21C. pp. 131-156.
- SAS. 2008. SAS/STAT Software for PC. Release 9.2, SAS Institute Inc. Cary. NC. USA.
- Van Soest, P.J, Wine, R.H, 1967. Use of detergents in the analysis of fibrous feeds. Determination of plant cell-wall constituents. *J. Assoc. Official Anal. Chem.* 50:50.

Characterization of Native Pig raisers and their current production systems in the integrated sweet potato –native pig production system in Baliem Valley, Jayawijaya Regency, Papua Province, Indonesia

Hanike Monim¹, Enrico P. Supangco², Cesar C. Sevilla², Virginia L. Barraquio² & Virginia R. Cardena³

¹Faculty at College of Animal Husbandry, UNIPA-Indonesia; ²Faculties at College of Agriculture, UPLB-Philippines, ³ Faculty at College of Public Affair, UPLB-Philippines

Abstract

Participatory Rural Appraisal approach was utilized to characterize and describe pig raisers and their families as well as their current management and production system. Result shows the characteristics of pig raisers in Baliem Valley as: 1) 46 to 55 years old, 2) having one big family called *Sili* with more than 2 household heads, 3) married with one wife but mostly having at least two wives, 4) mostly uneducated, 5) *Sili* size ranged from 8 to 28 members, and 6) *Sili* members, especially the women, serve as farming labor.

Low education, lack of technical knowledge and skills, as well as very little effort brought about poor management and production performance of native pig in all levels (family-*Sili*-, and community-level) in the village. The main sources of *Sili* income were cultivating multi-crops (daily cash) and raising pigs (big amount of cash when need arises). Sweet potato tubers and vines, given uncooked and/or cooked at a frequency of twice a day, were the native pig's primary source of energy. Extensive rearing system was applied to satisfy its need for other source of nutrients especially mineral and protein (earthworm). Low technical knowledge and skill contributed to the inability of pig raisers to prevent and cure diseases, especially Hog Cholera (HC), that was associated with high mortality and culling rates. After 5 years of HC outbreak, the role of native pig used mainly for social capital declined by 42.67% while it increased by 19.86% and 22.81% for financial capital and consumption, respectively. In addition, the lack of effort of the pig raisers to supply enough quantity and quality of feed was also a major factor for the low reproduction efficiency. Mortality rate for pre-weaning, post-weaning and adult pigs in this study were 26.30 %, 11.40% and 5.98%, respectively; consumption rate for pre-weaning, post-weaning and adults pigs were 9.36%, 14.87% and 6.05%, respectively; social activity rate for pre-weaning, post-weaning and adult were 2.45%, 17.15% and 25.57%, respectively; and selling rate for pre-weaning, post-weaning and adult were 17.78%, 24.66% and 9.86%, respectively.

Introduction

The integrated organic sweet potato-pig system has been traditionally practiced by people in the Baliem Valley for many centuries. Sweet potato serves as the main source of food for both people and pigs in this site, while the animal's manure is used as fertilizer. However, the performance of this production system is very low as reported by Cargill (2009) and the existing organic sweet potato-pig system has several problems, notably the low fertility rate and slow growth of pigs, which may be caused by unbalanced and erratic feeding regimes, health problems, genetics, management issues and other factors. These problems contribute to the system's inefficiency and farm income that should be addressed to increase production efficiency.

Sweet potato varieties grown for human and pigs consumption are being sold in the local markets, while pork is a significant protein source for the people and is often a widely traded commodity. With the need to have cash, pig production offers a good opportunity for higher income and is considered as the most important key to reducing poverty levels. However, its productivity has remained low and inefficient. One of the major cause of these dwindling pig population was the Hog Cholera outbreak which resulted in sharp increase in the market price of pork (by 300% more than before). Considerable studies should be done to enable producers to respond to this increased demand based on the current type of production systems. The management intervention proposed by ACIAR as an internal stakeholder for almost 10 years brought about considerable improvement in the economic, social and environmental aspects of the community where the project was implemented (Mahalaya, 2010). However, in early 2009, it was found that the practices during the implementation of project can no longer be sustained since there were no more technical and financial support from any local institution especially for feeding the animals. This condition brought the farmers back to the previous (traditional) management practices and created new problems. With this situation, it is important to explore the farmers' characteristics in terms of family attitude and decision making, and the role of each family member. There is a need to analyze the current practices related to the technologies developed in order to determine the constraints faced by the farmers during the implementation of said technologies. Information concerning the role of each family member in farm activities is also important and relevant and should be given priority since the quantity and quality of time and energy spent for farm activities are crucial. This study aimed to describe and characterize the native pig raisers and their current production systems in an integrated sweet potato – native pig production system.

Material and Methods

A survey was done to assess and characterize the native pig raisers and their major production system practices in an integrated sweet potato – pig production system. Four districts (Wamena, Kurulu, Walelagama and Hubikosi) with the highest pig population were selected using Judgmental Sampling. Four villages (Kama, Jiwika, Laytopo and Hubikosi, respectively) were chosen from the selected districts by their leaders and 10 *Sili* per village were randomly selected based on their willingness to be interviewed. *Sili*, therefore, is the unit of analysis in this research. Primary data were collected from the respondents in every *Sili* in two phases. Phase 1 involved PRA approach in which 15 people of different groups and gender per village participated in the meeting held in each village office. PRA techniques were explained by using mapping, group discussion, triangulation or cross-checking, direct observation and/or participant observation. Phase 2 involved the use of structured questionnaires and semi-structured interviews for household and key informants, respectively. There were 109 households from 40 *Sili* of the four villages interviewed. Quantitative data collected were the body weight of the pigs, nutrient content of forages and earthworm, and number of healthy and sick pigs sold out in the local market as well as their prices. The different range areas of pigs in Jiwika were explored at different times and tracked down during their range of periods. Samples of forages, grasses, and earthworms eaten by the pigs were taken to the Biodiversity Research Centre of the State University of Papua (UNIPA) and University of Gadjah Mada (UGM) for proper identification and nutrient analysis, respectively. Secondary data were also collected from the Bureau of Livestock, Bureau of Agriculture, Statistic Office and each District and Village Office. All data gathered were tabulated and analyzed using simple statistical tools such as summation, average, percentage, and frequencies.

Results and Discussion

The average number of households per *Sili* is three (3) households. A *Sili* could be headed by males only or mixed, some males and some females. Almost all households in the *Sili* (90.00%) were married and having more than one wife. The average number of wife/wives per *Sili* was 4. The number of wives was closely related to the number of pigs raised and the farm size of each *Sili*. *Sili* size for each *Sili* ranged from 8 to 28 members.

Reports on Jayawijaya in Figures year 2010 have shown that sweet potato contributes 94.67% to food crop production and occupies 71.97% of land for cropping in this regency. Planting sweet potato and raising native pigs are the major on-farm activities. Every *Sili* planted at least 4 kinds of crops. Beside pigs, native chicken, cattle and rabbit were the 3 species of livestock raised by the farmers. The average number of piglets raised by farmers per *Sili* was 9 heads. Sows were the second largest group of pigs raised by farmers since they were kept as stock animal in the herd and were never culled unless when in an emergency state. Every wife was responsible for keeping the sows for the sake of the whole family's needs. The traditional management practices of this tribe is unique called

Communal Management Practices.

The management system for pig production in Baliem Valley is extensive where pigs are let out in the morning and penned at night. The foremost effort done by pigs was to find and feed on earthworms (*omali*) which are their main source of protein.

The average litter size for gilt was 4 and for sow was 7 with the average weaning age of 5 months old. Disease was the main cause of mortality of native pigs in the whole Central Mountain Range area. In 2006, the new viral disease is known as Swine Fever or Hog Cholera killed 65,000 pigs in Baliem Valley.

There are five (5) main social activities that utilize the pigs and these are as follows: 1). Traditional mass wedding fiesta happens every 5 years (*Maue*); 2). Celebration for traditional house (*Honai*) building or renovation; 3). Celebration for land and animal fertility; 4). Death of the Chieftain; and 5). Payment of debt to those who donated when a family member passed away. Adult pigs are the most utilized (25.57%) compared to grower (17.15%) while piglet utilization was only 2.45% in customary and social events in the area. Mahalaya (2010) reported that pigs contributed 73% to family income, 26% for social activity and 1% for consumption. The same report also stated that before the implementation of the project or without any intervention, utilization for pigs in this area for social activity was up to 76%.

The data on average body weight were taken from several pigs and from several farmers without any given treatment shows that BW of 5 month was 17.2 kg. At the same age, this is lower than that of Papua New Guinea pigs which was 20.5 kg (Malynicz, 1971).

Conclusion

Native pig production in integrated sweet potato – native pig system in Baliem Valley depended mainly on the characteristics of its community and the management practices applied. The more number of pigs owned, the more powerful the owner is in his social, political and recently, in his economic life. The characteristics of pig raisers in Baliem Valley can be described as follows: 1) 46 to 55 years old, 2) having one big family called *Sili* with more than 2 household heads, 3) married with mostly having at least two wives, 4) mostly uneducated, 4) *Sili* size ranged from 8 to 28 members, and 5) *Sili* members especially women serve as farming labor.

High mortality rate, high culling rate, and low reproductive efficiency that brought about the slow population growth and the ultimate result of low *Sili* cash income were the major problems faced by pig raisers in this study area. This reality has been occurring from time to time

like a cycle without much improvement. The internal factors that affected this situation were identified as lack of technical knowledge and skills, as well as the attitude and cultural constraints.

Production performance of native pig in integrated sweet potato – native pig system was generally poor in all levels: family, *Sili*, and community levels. Communal management system in the study area was influenced by their own knowledge and norm inherited for generations including division of family labor. Although awareness of the benefits gained from native pig was high, it was not translated into good management practices. Lack of budget and effort as well lack of technical knowledge and skills to protect the animals from various diseases especially Hog Cholera were the biggest disasters for pig raisers in the study area. Before the outbreak of HC, it was reported that 76% of the animals were used for social activities, 23% for selling and 1% for consumption. In this study (after 5 years of HC outbreak), 33.33% were used for social activities, 42.86% for selling and 23.81% for consumption. The role of native pig for social capital declined by 42.67%, while it increased by 19.86% and 22.81% for financial capital and consumption, respectively.

Lack of technical knowledge and skills was also the main reason of poor management practices that gave rise to the slow population growth. Long lactation (weaning age) of 5 months and dry period of 2 months led to long interval between weaning and conception of 30 - 60 days and finally accounted for inefficient farrowing index of 1.24 – 1.38.

References

- Cargill, C. F., 2009. "Poverty alleviation and food security through improving the sweet potato-pig systems in Papua, Indonesia", Final Report, Australian Centre for International Agricultural Research, Canberra.
- Mahalaya, S., 2010. Impact Evaluation of Agricultural Research in Papua, Indonesia Using the Sustainable Livelihood Framework. Dissertation. ANU, Canberra, Australia.
- Malynicz, G. L., 1971. Use of raw sweet potato, raw peanuts and protein concentrate in rations for growing pigs. Papua New Guinea Agricultural Journal. 22(3):165-166.

Smallholder identified constraints to adoption of new forage options in South Central Coast Vietnam.

Nam Van Le^{1*}, Nguyen Thi Da Thao¹, Lydia Turner², Jeff Corfield^{1, 3}

¹Hue University of Agricultural and Forestry, Hue City 47000, Vietnam, ²Tasmanian Institute of Agriculture, University of Tasmania, Burnie 7320, Tasmania, Australia, ³Corfield Consultants, Townsville, Australia, 4811

Abstract

Adoption of new farming practices by smallholders is a complex process and service providers need to understand reasons for adoption and non-adoption to ensure that extension efforts can be targeted for maximum impact. This paper presents the adoption patterns of the tree legume *Leucaena leucocephala* cv. Taramba by 41 smallholder farmers in South Central Coastal Vietnam, where it was introduced in 2010-11 as an option to improve cattle nutrition. Smallholder farmers were interviewed in 2015 about their experiences with the establishment, management and use of Taramba *Leucaena* and how these experiences influenced their subsequent adoption decisions. Lack of labour and knowledge to manage seedlings, natural causes, perceived low feed provision and lack of land to manage mature stands were the main reasons for non-adoption. Successful adopters protected their seedlings from grazing and weed competition, carried out good cutting management, and were able to establish larger stands of mature trees. Future extension efforts should focus on how the long term introduction of Taramba *Leucaena* interacts with land, labour, and competing farm activities, in order to achieve more integrated and sustainable smallholder adoption outcomes.

Keywords: adoption constraints, new forage technologies, Leucaena, smallholders

*Corresponding author: levannam@huaf.edu.vn

Introduction

The extent of forage adoption by smallholder farmers in SE Asia has been variable (Paris, 2002), despite extensive research and development programs over many decades (Stür *et al.*, 2000). Adoption is a complex and continuing step-wise process that includes non-adoption, partial adoption, gradual adoption, adaptation, integration, innovation, and sometimes dis-adoption (Wilkinson, 2011). Smallholders often have rational reasons for non-adoption, which are not always immediately apparent to researchers and extensionists (Vanclay, 1992). They need to understand the reasons for adoption / non-adoption decisions to ensure that perceptions of smallholder motivations are accurate. By doing so, extension efforts can be better targeted for improved adoption outcomes. While *Leucaena* is widely used to feed livestock in Indonesian smallholder systems (Panjaitan *et al.*, 2013) it has limited use in Vietnam, and smallholders have little experience in growing, managing and feeding it to cattle. This paper draws on experiences from a recent study in South Central Coastal (SCC) Vietnam to explore the reasons for adoption and non-adoption of the tree legume *Leucaena leucocephala* cv. Taramba. Taramba *Leucaena* was introduced along with a range of new forage grasses in 2010-11 to three study communes (Cat Trinh, Binh Dinh province; An Chan, Phu Yen province and Phuoc Dinh, Ninh Thuan province) in SCC Vietnam. The study used participatory research methods to introduce a range of forage, cattle feeding and husbandry practices to 15 smallholders in each commune. Details of study design and site descriptions can be found in Ba *et al.* (2013). Forty one farmers participated

in workshops and monitoring visits during the study; they received Taramba seedlings, and were advised about how to plant and protect seedlings, and how to grow, manage and feed mature tree forage to cattle. This paper explores the experiences of these farmers with Taramba since its introduction, examines how these experiences shaped their adoption decisions and suggests how lessons learned from this study can inform future extension activities.

Materials and Methods

In April-May 2015, 41 former study farmers in the three communes were re-visited to determine the current adoption status of Taramba. Farmers were interviewed about the status of the original Taramba stands, current patterns of use and their reasons for continuing use or non-use. Short narratives were also collected to contextualise the reasons for adoption / non-adoption decisions. Interview data was classified into themed categories and simple descriptive statistics derived to help explain response patterns.

Results

Figure 1 illustrates the patterns of Taramba *Leucaena* introduction and adoption between 2010/11 and 2015. Of 41 farmers who received seedlings, 18 (44%) failed to have them mature to adult stage due to uncontrolled grazing by small livestock (chickens, goats) or severe weed/light competition. Of the 24 farmers (59%) whose plantings reached adult stage, 4 had stands later destroyed by flood or drought and did not replant.

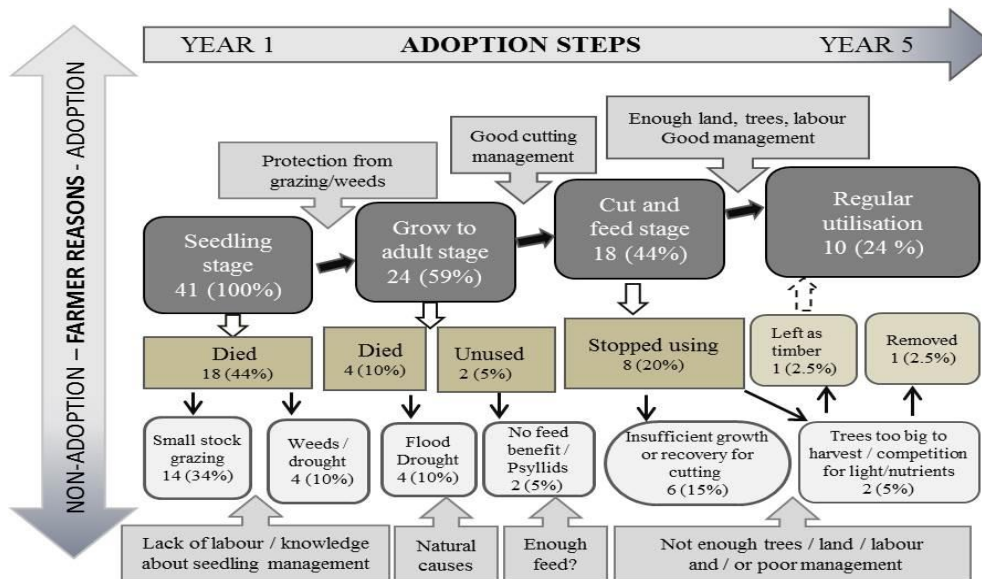


Figure 1. Schema of Taramba *Leucaena* adoption steps in 3 communes in SCC Vietnam, showing stepwise outcomes, farmer-identified reasons for adoption success / failure and contributing factors. Note: Perforated arrow indicates farmer may resume harvesting Taramba forage in the future.

Two farmers (5%) with adult stands did not attempt to use them for feed, due to perceived lack of benefit or *Psyllid* insect attack issues. Of the 44% who initially cut and fed Taramba, 20% later discontinued use due to either perceived slow recovery when cut too regularly, or inability to harvest forage if trees were left to grow too tall. Of the 24% still actively feeding Taramba to stock by April 2015, 12% cut and fed regularly, maintaining good cutting management for optimum feed quality, while the remainder harvested opportunistically. Quantity and frequency of forage harvested varied with number and size of trees available. All those still feeding Taramba reported benefits for cattle condition and productivity. There were differences between communes, with 21% of An Chan farmers failing to establish Taramba beyond seedling stage compared to 53% and 58% in Cat Trinh and Phuoc Dinh respectively. By contrast 50% of An

Chan farmers never used or stopped using well established Taramba stands compared to 7% in Cat Trinh and 15% in Phuoc Dinh, where half the active Taramba adopters were feeding it to goats, solely or in addition to cattle.

Discussion and conclusion

In Cat Trinh and Phuoc Dinh, the most significant barrier to initial adoption was the difficulty farmers had in applying recommended seedling management - a critical step for successful *Leucaena* establishment. Reasons offered included poor understanding of management needs or lack of resources or ability to protect seedlings, especially from uncontrolled grazing. Farmers who successfully established Taramba there had livestock effectively excluded. In An Chan, land constraints were the prime reason why farmers who initially grew and fed Taramba stopped doing so. Farmers with limited land, who could only grow a small number of Taramba trees either harvested them too frequently, producing only small quantities of forage and slower recovery times, or harvested infrequently to gain more biomass, which risked trees growing too tall to harvest, causing shading and perceived water and nutrient competition problems. Both outcomes invoked relative 'cost/benefit', or 'reward for effort' issues for these farmers, leading at least one An Chan farmer to remove Taramba trees from his backyard area. Farmers who successfully incorporated Taramba into their production system all had sufficient land to grow enough Taramba trees for useful forage contribution, without interfering with other farming activities – a key adoption precursor. The next constraints were labour and management, especially for some An Chan and Phuoc Dinh farmers who used Taramba opportunistically and sometimes let it grow too tall for easy harvesting in the rainy season. By contrast, one Phuoc Dinh farmer with 200 m² of well managed Taramba says it takes only one hour per day to cut 100 kg of fresh forage to feed her cattle and goats. Establishing and using tree legumes like Taramba for forage involves a much longer term commitment of land and labour than does forage grasses, and so different risk-reward considerations for smallholders. Taramba has proved a highly valuable dry season forage asset for the successful adopters in this study. Future extension efforts should focus on how the long term introduction of Taramba interacts with available resources - especially land, labour and competing farm activities - in order to achieve more integrated and sustainable smallholder adoption outcomes.

References

- Nguyen Xuan Ba, Peter A. Lane, David Parsons, Nguyen Huu Van, Ho Li Phi Khanh, Jeff P. Corfield, Duong Tri Tuan. 2013. Forages improve livelihoods of smallholder farmers with beef cattle in South Central Coast Vietnam. *Tropical Grasslands Forrajes Tropicales* 1:225-229.
- Panjaitan, T., and M. Fauzan. "Dahlanuddin; Halliday MJ; Shelton HM. 2013. Growth of Bali bulls fattened with forage tree legumes in Eastern Indonesia: *Leucaena leucocephala* in Sumbawa. *Proceedings of the 22nd International RC; Shelton HM, eds. Forage Tree Legumes in Tropical Agriculture. CAB International, Wallingford, UK* (2013).
- Paris, T. R. 2002. Crop–animal systems in Asia: socio-economic benefits and impacts on rural livelihoods. *Agricultural Systems*, 71, 147-168.
- Stür, W.W., Horne, P. M., Hacker, J. B. and Kerridge, P.C. 2000. Working with Farmers: The key to adoption of forage technologies. ACIAR Proceedings No. 95, Australian Centre for International Agricultural Research, Canberra.
- Vanclay, F. 1992. The barriers to adoption often have a rational basis. In 'Proceedings of the 7th International Soil Conservation Organisation Conference, Sydney, September 1992, People protecting their land, Vol. 2'. (Eds PG Haskins, BM Murphy) pp. 452–458. (International Soil Conservation Organisation: Sydney).
- Wilkinson, R.L. 2011. The many meanings of adoption. In Pannell, D.J and Vanclay, F.M. (eds), *Changing land management: adoption of new practices by rural landholders*. CSIRO Publishing, Collingwood, Victoria. pp 39–49.

Studies on socio-economical profile of the dairy farmers in Latur district of Maharashtra state

Dande, K.G.

Corresponding author, Head Department of Dairy Science, Mahatma Basweshwar College, Latur (M.S.) India

Abstract

Present investigation was carried out to know personal and socio-economic characteristics of dairy farmers. To achieve the objective of the present investigation data from 3000 dairy farmers were collected. It is observed from the present investigation that only 7% respondents observed below the age of 25. The highest number of respondents observed in above age level 46. However between age 26-35 only 26% were observed. About 53% of respondents fall below 5000 rupees per month income group. After this the income between the range of 5001-10000 rupees only 23 respondents were observed however between the income range 10001-15000 and above only 10 and 14 respondents were observed respectively. 81 percent of the respondents were educated up to primary level, whereas around 16% of the respondents had graduate level education and illiterate (3%). Regarding family structure, 1% was from the joint family and rest 99 % belonged to the nuclear family.

Keywords: dairy farmer, respondents, illiterate, socioeconomic

Corresponding author: dande.kiran@yahoo.com

Introduction

The importance of dairying in our country hardly needs emphasizing. India has more than 50 percent of the world's buffalos and 20 percent of its cattle which play an important role in the national economy as well as in the socio-economic development of millions of rural households. The operation flood programme, organizing dairy farmers' cooperatives in rural areas created a strong network for procurement, processing, and distribution of milk. Livestock sector plays pivotal role in providing nutritive food rich, in animal protein and also helps in supplementing family incomes and generating gainful employment in the rural sector. Presently, India has a huge population of 485 million livestock and 489 million poultry population, holding the second highest position in cattle strength. It possesses the highest strength of buffaloes, third highest number of sheep, holds second highest position in goat population, fifth highest number of chicken (Basic Animal Husbandry Statistics, 2003) and accounts for 4.5 % of GDP. Though India possesses the richest animal wealth in the world, the productivity in the livestock sector is less than its optimum (Sathyanarayan et al., 2010)

Despite of vital role of dairy farmers in the socio economic development, with this broad object in view the present investigation was undertaken to know the personal and socio-economic characteristics of dairy farmers in Latur district.

Materials and Methods

For the present investigation sample population was selected from the Latur districts of Maharashtra state India. To achieve the objective of the present investigation ten tehsils, from each tehsil 10 villages and from each village 30 farmers were selected. In this way by conducting personal interview data from 3000 dairy farmers were collected. The survey research design was used for data collection. The interview schedule constructed for the research was used to collect the information regarding age, education, land holding, family size, type of family, social participation, experience etc. from these respondents. The collected data tabulated and analyzed accordingly.

Results and Discussion

Socioeconomic profile (age in years) of the respondents in Latur district

The investigation shows socioeconomic profile of the respondents in Latur district. The present investigation showed that only 7% respondents observed below the age of 25 the reason. The highest number of respondents observed in above age level 46. However, between age 26-35 only 26% were observed.

Monthly income of dairy women farmers in Latur district

Monthly income from sale of milk was used as an indicator for estimating the income generation. Monthly income refers to the gross income earned by a family from sale of milk in one month. Monthly income is totally depends on how many milch animals is to be reared. The investigation shows that the maximum (53%) respondents fall in the range of below 5000 rupees per month. The reason might be due to unavailability of green fodder round the year, non availability of water for performing various activities on the farm etc. Due to non availability of green fodder, water, low milk producing cattle's and buffaloes, marketing facility it is becoming very difficult to produce high milk quantity. Therefore due these causes about 53% of respondents fall below 5000 rupees per month income group. after this the income between the range of 5001-10000 rupees only 23 respondents were observed however between the income range 10001-15000 and above only 10 and 14 respondents were observed respectively.

Educational qualification of the dairy women farmers in Latur district

Investigation shows the educational qualification of dairy farmers in Latur district. It is reported that education is one of the most important determinants of a person's social status. Investigation indicates that 81 percent of the respondents were educated up to primary level, whereas around 16% of the respondents had graduate level education and illiterate (3%). Regarding family structure, 1% was from the joint family and rest 99 percent belonged to the nuclear family.

Conclusion

It is observed in the studies that, majority of respondents were satisfied with family support and majority of the respondents were nuclear family. Most of the respondents were allocated business profits for business development and family members were giving good guidance to the women entrepreneurs. The study was conducted following exploratory research design to ascertain the profile characteristics of livestock farmers. Findings of the present investigation indicated that majority of the dairy farmers had low to medium profile. Hence efforts should be undertaken by the Government in providing information on livestock farming practices so that they could bring about change in their living and improve the socio-economic status of livestock farmers.

References

- Basic Animal Husbandry Statistics, 2006. Government of India, Ministry of Agriculture, Department of Animal Husbandry, Dairying and Fisheries, New Delhi.
- Jain P. and A. Singhal, 2012. Participation of rural women in livestock management activities in Bhilwara district, *Rajasthan Journal of Extension Education*, 20: 190-193.
- Kishtwaria J., A.R. Kishtwaria, and S. Sood, 2009. Work Pattern of Hill Farm Women -A Study of Himachal Pradesh, *Stud. Home Comm. Sci.*, 3(1): 67-70.
- Manimekalai, P. and B.R.Subramaniya, 2013. A study on satisfaction level of women entrepreneurs in dairy sector in salem district of Tamilnadu, *Asia Pacific Journal of Research*, 1(4):43-53.
- Rathod, P.K., T.R. Nikam, S. Landge, S. Vajreshwari and A. Hatey, 2011. Participation of Rural Women in Dairy Farming in Karnataka Veterinary College, Parel, Mumbai. *Indian Research Journal Extension Education*, 11 (2).45-50.
- Sathanarayan K., V. Jagadeeswary, V. Chandrashekhar Murthy, S. Wilfred Ruban and G. Sudha. Socio-economic Status of Livestock farmers of Narasapura Village - A Benchmark Analysis, *Veterinary World*, 3(5):215-218.
- Tayde Vidya V. and R.R. Chole, 2010. Empowerment Appraisal of Rural Women in Marathwada Region of Maharashtra State, *Indian Research Journal of Extension Education*, 10(1):33-36.
- Tiwari N and R. Upadhyay, 2012. Technological needs of rural women in selected drudgery reducing technologies related to animal husbandry, *Rajasthan Journal of Extension Education*, 20: 10-14.
- Upadhyay R. and D. Solanki, 2009. Income generation among women through dairy cooperatives and constraints thereof, *Rajasthan Journal of Extension Education*, 17: 74-78.
- Upadhyay R. and D. Solanki, 2010. Income generation among women through dairy cooperatives and constraints thereof, *Rajasthan Journal of Extension Education*, 18: 74-78.
- Vallapureddy M., 2013. Performance of Women Cooperative Dairy A Study on Mulkanoor Women Cooperative Dairy in Andhra Pradesh, *International Journal of Contemporary Business Studies*, 4(3):49-62.

Economic impact of spatial development on goat farming in Banjarnegara district Indonesia

Moch. Sugiarto and Syarifuddin Nur

University of Jenderal Soedirman

Abstract

Goats in Banjarnegara District, Indonesia are kept under varying ecological conditions consisting of the high land (>1000m), middle land (500-1000 m) and low land (<500 m). These conditions can lead to differences in the availability of feed, climate and culture that will affect profit ability and economic efficiency. The farmers in middle land has better profile (age, experience and education) compared to farmers in other ecological zones. The spatial differences in the development of goat farming would encourage income disparities of farmers and significant difference in the economic efficiency ($p < 0.01$). Goat farmers in the middle land generate the higher income and economic efficiency compared to other spatial zones ($p < 0.01$). Based on this analysis it can be concluded that spatial development of goat farming has significant economic impact. Spatially, goat farming would be more efficient and profitably performed at middle land. Strengthening human resources of goat farmers and their social resources must be conducted intensively and continuously to strengthen goat farm management.

Keywords: spatial development, goat management, income, economic efficiency

Corresponding author: zoegic@yahoo.com

Introduction

Banjarnegara district has a suitability level for goat development, both in terms of the suitability of vegetation, topography, climatic, or even from a social-cultural local area. Nevertheless goat farming has major challenges in livestock production systems include the availability of food and land, whereas the main factor in determining the productivity of livestock is the assured availability of forage. Animal agriculture systems have been categorised on the basis of agro-ecological opportunities. In general, these systems are shaped by prevailing biophysical and socio-cultural environments. In many of these systems, the livestock element is interwoven with crop production (Steinfeld et al., 2006). An increase in population, industrialization, and declining soil fertility are an important issue in locating the goat farming. Changing conditions and potential of the region would encourage changes in the direction of development of goats farming.

Income of farmers, population and production are important indicators of goat farming development. Different socioeconomic, market and agro ecological factors are significant in explaining the richness of farmers (Gauchan et al., 2005). The increase in farmers' income is driven by the number of goats owned by farmers. Nemeth et al. (2004) stated that income is increased by the number of productive animals which ensures the covering of expenses. Greater

ownership of goats related to the availability of natural resources as inputs (forage) and human resources. In addition, the development of goat farming is driven by the development of the area which includes the potential of natural resources, human resources, institutional and capital resources / capital and infrastructure simultaneously. Therefore, this study was purposed to identify and compare the income of the goat farmers in three different agro ecological zones in Banjarnegara district.

Materials and Methods

Studies on economic impact of spatial development on goat farming in Banjarnegara District were conducted by using a questionnaire survey through interviews and observations of goat farmers in Banjarnegara.

179 goat farmers selected as respondents using multistage sampling method. First, the area sample was selected by stratified random sampling based on the agro ecological zones (high, medium and low land). The sub districts were selected randomly as much as 20 percent of each stratum. Second, respondents (farmers) were selected by random sampling method as much as 20 percent in each selected sub districts.

Once the data is obtained and the results are analyzed using one-way ANOVA to determine differences in the income of goat farmers in three agro-ecological regions (lowland: <500 m, middle land: 500-1000 m, and highland:> 1000 m).

Results and Discussion

Socioeconomic characteristics of goat farmers

Goat farmers in Banjarnegara mostly categorized as productive age with average age of 47.7 years. Goat farmers in middle land have younger age compared to farmers in high and low land. In average, goat farmers have been graduated from elementary school. Goat farmers in middle land are more educated (7.4 years) than those in low and high land. Manzi et al. (2013) stated that high illiteracy rates tend to hinder adoption of suitable technology and do not make it easy to communicate to such producers' messages of technical nature. Meanwhile Meretiwon (1981) stated that education is significant and important to the increased of agricultural production. Most of the farmers have adequate experience in goat farming activities (9.13 years). Goat farmers in middle land have longer experience (10.8 years) than farmers in low and high land. Different agro ecological zone is significantly resulted different number of goats. Middle land of agro ecological zone generates number of goats than other area of Banjarnegara. The number of goats is categorized in animal unit (AU). Sodik (2004) stated that Small Ruminant Unit (SRU): livestock unit for small ruminant, doe (more than 12 months) =1 SRU, young (4-12 months) = 0.7 SRU, kid (less than 4months) =0.25 SRU. Socio economic characteristics of farmers are presented in Table 1.

Table 1. Socioeconomic characteristics of goat farmers in Banjarnegara district.

Parameter	Highland	Middle land	Lowland
Age (year)	46	45.1	50.5
Education (year)	6.5	7.4	7.1
Farming Experience (year)	10.3	10.9	7.3
Number of family members (persons)	3.4	4.2	3.9
Number of goat (AU)	0.55	0.66	0.32

Cost and return of goat farming

The revenue of goat farming mostly comes from selling goats and feces in one year period. The total revenue from goat farming in Banjarnegara is an average of Rp 4,255,586.59 per year. Revenue of goat farmers in middle land is significantly higher than that of low and high land farmers ($p < 0.05$) because of their ability to sell more number of goats. Nemeth et al. (2004) stated that a minimum herd size is needed for covering fixed and variable expenses. Income is increased by the number of productive animals which ensures the covering of expenses, but only to a certain herd size.

Variable costs such as concentrates, transportation, medicine and forage cost contributed 92.5 percent to total cost. The variable cost is significantly different among the agro ecological zones and variable cost of goat farming in middle land is higher than other zones. Goat farming in Banjarnegara needed total cost of Rp 1,904,427.53/year for 0.51 Animal Unit (AU) which equivalent to 4 heads of goat. The cost and return to goat farming is Rp1,904,427.53 and Rp 4,255,586.59 respectively. The indicated revenue, cost and income is described in Table 2.

Table 2. Cost and return of goat farming.

Parameter	High Land	Middle Land	Low Land
Total Revenue	3,704,878.05	5,992,241.38	3,278,750.00
Variable Cost	1,983,292.68	2,245,862.07	1,297,967.50
Total Cost	1,820,780.49	2,270,484.48	1,679,088.61

Profitability of goat farming based on spatial development

Goat farming in Banjarnegara district generally was driven by variable cost which contributed more than 90 percent of total cost. Goat farming in middle land spend more variable cost than other zones ($P < 0.01$). Revenue of goat farming is contributed mostly by selling goat and faces. Alibejov et al. (2001) stated that different in farm size between agro ecological zone were statistically significant. Farm size becoming an important factor to increase number of animal sold. In a year goat farmers in Banjarnegara have generated average revenue of Rp4, 255, 586.59. Farmers in middle land has capability to generate more income significantly different ($P < 0.01$) to the farmers in low and high land. Table 3 indicated that net income of goat farmers in middle land is significantly higher ($P < 0.01$) than low and high land farmers.

Table 3. Profitability and Economic Efficiency of goat farming.

Parameter	High Land	Middle Land	Low Land
Net Income (Rp)	1,163,453.25	3,822,344.82	1,689,107.59
Economic Efficiency	1.64	4.03	1.99

The effort of farmers in middle land to generate more income also resulted from efficiency in using local feed for goat. Steinfeld (2006) mentioned that in extensive system livestock production grows depends more on locally available feed resources. More availability of grass in middle land has generated low cost to get feed for goat. Availability of grass in middle land becomes an important factor to maintain more goat production. Ebrahim and Hailemichael (2012) mentioned that major constraint for goat farmers is shortage of grass/feed. Availability of land for producing grass and suitable climate in middle land has produced more efficient goat farming. Small scale goat farming rely more on grass production in their area. Based on Table 3, goat farmers in middle land tend to produce more efficient farming than other agro ecological zones ($P < 0.01$). Hosu et al. (2013) mentioned that smallholder farmers when subjected to different levels of land, labor, soil status and climate variability, the general observation is that they can produce relatively efficient under climate variability condition compared to other scenarios.

Conclusion

Spatial development in Banjarnegara has driven some changes in different agro ecological zones. Human resources of farmers in middle land have more benefit in terms of age, education and experience compared to other area. Middle land has contributed more supports to goat farming development especially in providing grass and other feed. Spatial development has produced different income of farmers in each agro ecological zone. Farmers in middle land have larger farms size to generate more revenue and make the goat farm more efficient. Spatial development of Banjarnegara District has resulted in different number of income of goat farmers. Goat farmers in middle land have generated more income than other area and therefore goat development must be developed more efficiently in middle land of agro ecological zone.

References

- Alibejov, N., S. Krusche, J. Wesseler, 2001. Agro-Ecological Zones and their Impact on Farm Production and Farm Organization after Privatization in Azerbaijan. Paper presented at the Workshop Approaching Agricultural Technology and Economic Development of Central and Eastern Europe. IAMO (Institut für Agrarentwicklung in Mittel- und Osteuropa), Halle, Germany
- Ebrahim, A and A. Hailemichael, 2012. Sheep And Goat Production And Utilization In Different Agro-Ecological Zones in Tigray, Ethiopia. *Livestock Research for Rural Development* 24 (1) 2012.
- Gauchan, D., M. Smale, N. Maxted, M. Cole, B.R Sthapit, D. Jarvis and M. P Upadhyay. 2005. Socioeconomic and Agroecological Determinants of Conserving Diversity On-farm: The Case of Rice Genetic Resources in Nepal. *Nepal Agric. Res. J.* Vol. 6, 2005
- Hosu, S., M. Sibanda, and A. Mushunje, 2013. Scenario Simulation of Small Farms' Production Efficiencies In The Eastern Cape Province, South Africa. Paper presented at the 4th International Conference of the African Association of Agricultural Economists, September 22-25, 2013, Hammamet, Tunisia
- Manzi, M., J Mutabazi, C D Hirwa and D R Kugonza, 2013. Socio-economic assessment of indigenous goat production system in rural areas of Bugesera District in Rwanda. *Livestock Research for Rural Development* 25 (11) 2013.

- Meretiwon, B. 1981. Factors Associated With Maize Storage Techniques At Farmers Level in Oyo State of Nigeria. Stored Product Research Institute Technical Report No. 8. Ibadan
- Németh, T. L. Branduse, M. Ábrahám and S. Kukovics, 2004. Factors Affecting The Profitability Of Different Goat Farm Sizes In Hungary. South African Journal of Animal Science 2004, 34. Oyo state of Nigeria.
- Sodik, A., 2004. Doe productivity of Kacang and Peranakan Etawah Goats and Factors Affecting Them In Indonesia. Journal of Agriculture and Rural Development in the Tropics and Subtropics. Supplement 78. Kassel University.
- Steinfeld, H., T. Wassenaar and S. Jutzi, 2006. Livestock production systems in developing countries: status, drivers, trends. Rev. sci. tech. Off. int. Epiz., 2006, 25 (2)

LF-349

Pasture management and supplemented feed enhanced the performance of farmed buffaloes in Sabah, Malaysia

Zamri-Saad, M^{1*}, K. Azhar², A.B. Zuki³ & A. Punimin⁴

¹Research Centre for Ruminant Diseases, Faculty of Veterinary Medicine, Universiti Putra Malaysia, 43400 Serdang, Malaysia. ²Department of Animal Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Malaysia, ³Department of Veterinary Preclinical Sciences, Universiti Putra Malaysia, 43400 Serdang, Malaysia, ⁴Department of Veterinary Services and Animal Industry Sabah, Kota Kinabalu, Malaysia

Abstract

A buffalo-breeding farm that practices extensive grazing system without supplementation was selected for this study. Farm records between 2004 and 2011 were analyzed for selected parameters, particularly the calving rate, calving interval, daily weight gain of calves and calf mortality. Following the analysis, interventions were implemented in January 2012, which included the increased in pasture area from 399 to 441 acres followed by application of fertilizer. The selected breeder buffaloes were prepared for breeding by supplementing palm kernel cake-based feed at the rate of 1.5kg/animal/day for 2 weeks before breeder males were introduced at the rate of 1 male to 20 females. Prior to the intervention, proximate analysis of pasture revealed 7.6% protein content, 79% of breeder buffaloes were with body score of ≥ 3 , the average calving rate was 22%, the calving interval was 24 ± 11.2 months, average daily weight gain of calves was 0.89 ± 0.21 kg, the average birth weight was 28.31 ± 3.26 kg and calf mortality was $26.8 \pm 7.0\%$. Following intervention, proximate analysis of grass revealed 12% crude protein. With feed supplementation, the percentage of breeder female with body score of ≥ 3 increased to 95% leading to an average annual calving rate of 50%. Average birth weight was significantly ($p < 0.05$) improved to 35.15 ± 5.39 kg while the average daily weight gain was 0.95 ± 0.32 kg. Subsequently the average calving interval was reduced to 15.2 ± 9.2 months. Similarly, the calf mortality rate was significantly ($p < 0.05$) reduced to $17.6 \pm 4.7\%$. In conclusion, feeding intervention significantly enhances the performance of farmed buffaloes.

Keywords: performance, farmed buffaloes, Malaysia

**Corresponding author: mzamri@upm.edu.my*

Introduction

Buffalo remains as high economic importance for farmers in many developing countries, particularly in Asia (Cruz, 2010). They are kept mostly on small farms and are provided with relatively poor digestible feeds (Yindee, 2011). They graze in the harvested paddy fields, along roadsides and on the edges of cultivated plots during daytime and are kept within the village, usually under the house at night (Khajareern and Khajareern, 1989). Therefore, their weight gain is low leading to poor reproductive performance (Nanda et al., 2003). This paper describes the attempt to enhance the selected performance parameters of buffaloes kept at a breeding farm in Sabah, Malaysia through improved feed and feeding.

Materials and Methods

This study involved a buffalo-breeding farm located at Telupid, Sabah, Malaysia (5° 30' N, 117° 7' E). At the start of the study, the farm had a total of 335 head of buffaloes. Of these, 180 were breeder females, 78 breeder heifers, 7 breeder males and 70 calves, of which 51 were females and 19 were males (Othman et al. 2014). The 399 acres of pastureland were divided into paddocks with established pasture (*Brachiaria decumbens*) and wallowing sites. Although the pasture was generally poorly maintained, the farm practiced an extensive 30-day rotational grazing system without feed supplementation (Othman et al., 2014).

At the start of the study, 8-year farm records between 2004 and 2011 were selected and analyzed retrospectively for selected parameters associated with feed and feeding. Among the parameters analysed included calving rate, calving interval, birth weight, average daily weight gain of calf and calf mortality. Pasture samples were collected from four representative paddocks, each paddock at 6 sites of 1m² area for proximate analysis of the nutrient contents (Galyean, 2010).

The interventions were implemented in January 2012. These included the use of organic fertilizer at the rate of 20 tonnes of organic fertilizer supplemented with 200kg of urea/ha/year to improve the pasture. After 6 months, pasture samples were collected from the four paddocks for re-analysis of the proximate nutrient contents. A total of 150 breeder females of more than 350kg body weight were selected, re-grouped into 20-head per group and were allowed to graze in paddocks. Breeders were provided with supplemented feed (1.5kg/animal/day) for 14 days before breeder males were introduced in January 2012 at the rate of 1 male to 20 females. Pregnant buffaloes were provided supplemented feed at 1kg/animal/day.

Following intervention, weaning age was reduced from 6 months to 3 months old. Birth weights were measured and recorded within 3 days of birth while body weights were recorded every 3 months and the daily weight gain was calculated based on the body weight of the first 3 months.

Results

Prior to the intervention, proximate analysis of the pasture revealed 7.6% protein content while 79% of breeder buffaloes had body score of ≥ 3 . The average annual calving rate was $22.1 \pm 6.4\%$ and the average calving interval was 24 ± 11.2 months. During the 8-year period, 73 (29%) breeders calved twice and 35 (14%) calved 3 times. The average birth weight was 28.31 ± 3.26 kg, the daily weight gain was 0.89 ± 0.21 kg (Table 1) and the calf mortality was $26.8 \pm 7.0\%$ (Table 2).

Following intervention, proximate analysis revealed an average of 12% crude protein. The breeder females with body score of ≥ 3 increased significantly ($p < 0.05$) to 95% leading to a significant ($p < 0.05$) increased annual calving rate to $50.2 \pm 16.1\%$. A total of 69 (46%) had calved twice ($p < 0.05$) and 8 (5%) calved 3 times, significantly ($p < 0.05$) less than the pre-intervention period. The average birth weight had significantly ($p < 0.05$) improved to 35.15 ± 5.39 kg and the average daily weight gain was improved to 0.95 ± 0.32 kg (Table 1). Subsequently the calving interval was significantly ($p < 0.05$) reduced to 15.2 ± 9.2 months. Similarly, the calf mortality rate was significantly ($p < 0.05$) reduced to $17.6 \pm 4.7\%$ (Table 2).

Table 1. Average birth weight and daily weight gain of buffalo calves before and after interventions

Year	Birth weight (kg) (Mean±SD)	Daily weight gain (kg) (Mean±SD)
<i>Pre-intervention</i>		
2009	28.47±1.36	0.96±0.16
2010	27.15±1.04	0.97±0.16
2011	28.74±2.08	0.73±0.21
Average	28.31±3.26 ^a	0.89±0.21 ^c
<i>Post-intervention</i>		
2012	30.53±3.43	0.85±0.24
2013	32.45±5.27	0.89±0.25
2014	37.05±4.76	1.16±0.35
Average	35.35±5.39 ^b	0.95±0.32 ^d

^{a,b}Different superscripts indicate significant difference (p<0.05)

Table 2. Calf mortality before (2004 to 2011) and after interventions (2012 to 2014)

Year	No. of calves		% calving	% mortality
	Birth	Death		
<i>Pre-intervention</i>				
2004	60	13	24.0	21.6
2005	73	12	29.2	16.4
2006	43	15	17.2	34.8
2007	78	29	31.2	37.2
2008	42	13	16.8	31.0
2009	37	9	14.8	24.3
2010	66	17	26.4	25.8
2011	43	10	17.2	23.3
Average	55.3±15.9 ^a	14.8±6.3 ^c	22.1±6.4 ^e	26.8±7.0 ^f
<i>Post-intervention</i>				
2012	53	8	35.3	15.9
2013	101	14	67.3	13.8
2014	72	9	48.0	12.5
Average	73.3±24.2 ^b	10.3±3.2 ^b	50.2±16.1 ^d	14.1±1.7 ^g

^{a,b}Different superscripts indicate significant difference (p<0.05)

Discussion

Diet is the main factor that affects body weight and body condition of livestock (Zerbini and Wold, 1999). Nevertheless, Frisch and Vercoe (1984) found low potential for buffalo to increase milk yield through improved feeding. This explains the slight increased in the post-intervention average daily weight gain in this study. However, feeding and management can enhance other parameters such as the birth weight (Khajarearn and Khajarearn, 1989). This is because the nitrogen requirements for maintenance of growing swamp buffalo can be fulfilled by grass of higher crude protein value as observed in this study (Tatsapong et al., 2010). Similarly, buffaloes are shown to gain weight with feed supplementation, which is less pronounced when the basal diet is only grass (Van Thu and Preston, 1999). Therefore, feed supplementation helps in maintaining the body condition of breeder buffaloes by reducing the weight loss (Jabbar et al., 2013).

Body condition score (Anitha et al., 2011) has been used as an indicator of energy status that is provided via feed supplementation (Qureshi, 2009). Animals receiving ME above the requirements during prepartum period are able to maintain a relatively good BCS (Qureshi, 2009), a phenomenon observed in this study when the percentage of breeder buffalo with good body score increased to 95% following feed supplementation. Furthermore, feed supplementation improves reproduction as evident from the shortest postpartum ovulation interval and lowest incidence of silent ovulations (Qureshi, 2009). Therefore, high energy supplemented feed such as the palm kernel cake-based diet implemented in this study helps to improve the reproductive performance of buffaloes.

Early weaning at 45 days old has been recognized as a major cause of mortality among buffalo calves (Parera, 1999). Nevertheless, rainy season (Othman et al., 2014) and pre-weaning nutrition level also play important role in calf mortality. However, better growth rate leads to better survivality (Thevarnanoharan et al., 2001) and the improved birth weight and weight gain observed in this study were influenced calf mortality.

Acknowledgements

This project was financially supported by the research grant (I-gt/15 (UPM) from the Ministry of Education (MoE) Malaysia and the Economic Planning Unit (EPU) Malaysia. The technical support from Mr. Jafred Tudok, Manager of the farm and official approval by the Director, Department of Veterinary Services Sabah are greatly appreciated.

References

- Anitha, A., K. Sarjan Rao, J. Suresh, P.R. Srinivasa Moorthy and Y. Kotilinga Reddy, 2011. A body condition score (BCS) system in murrha buffaloes. *Buffalo Bull.* 30(1): 79-99
- Cruz, L.C., 2010. Recent developments in the buffalo industry of Asia. *Proc. 9th World Buffalo Cong.*, Buenos Aires, Argentina, 7-19
- Frisch, J.E. and J.E. Vercoe, 1984. Improving the swamp buffalo of South-east Asia. Evaluation of large ruminants for the tropics. Copland, J.W. (ed.). *ACIAR Proc. No. 5*: 37-43.
- Galyean, M.M., 2010. Laboratory procedures in animal nutrition research. Department of Animal and Food Science, Texas Tech University, Lubbock
- Jabbar, M.A, I.B. Marghazani, T.N. Pasha, A. Khalique and M. Abdullah, 2013. Effect of protein supplements of varying ruminal degradability on nutrients intake, digestibility, nitrogen balance and body condition score in early lactating Nili-Ravi buffaloes. *J. Anim. Plant Sci.* 23(Suppl.): 108-112

- Khajareern, S. and J.M. Khajareern, 1989. Feeding swamp buffalo for milk production. In: Feeding Dairy Cows in the Tropics. Speedy, A., Sansoucy, R. (eds). Proceedings of the FAO Expert Consultation, Bangkok. FAO
- Nanda, A.S., P.S. Barr and S. Prabhakar, 2003. Enhancing reproductive performance in dairy buffalo: major constraints and achievements. *Reprod. (Suppl.)*: 61, 27-36
- Othman, R., M.Z.A. Bakar, A. Kasim and M. Zamri-Saad, 2014. Improving the reproductive performance of buffaloes in Sabah, Malaysia. *J. Anim. Health Prod.* 2(1): 1-4
- Parera, O., 1999. Management of reproduction. In; *Smallholder Dairying in the Tropics*. Zerbini, E., Wold, A.G., Falvey, L., Chantalakhana, C. (eds.). ILRI (International Livestock Research Institute), Nairobi, Kenya. 462pp
- Qureshi, M.S., 2009. Nutritional and management support to reproduction in dairy buffaloes under tropical conditions. *Pak. J. Zool.* 9 (Suppl.): 895-909
- Tatsapong, P., P. Peangkoum, O. Pimpa and M.D. Hare, 2010. Effects of dietary protein on nitrogen metabolism and protein requirements for maintenance of growing Thai swamp buffalo (*Bubalus bubalis*) calves. *J. Anim. Vet. Adv.* 9: 1216-1222.
- Thenarnanoharan, K., W. Vandepitte, G. Mohiuddin and C. Chantalakhana, 2001. Environmental factors affecting various growth traits of swamp buffalo calves. *Pak. J. Agric. Sci.* 38(3-4): 5-10
- Van Thu, N. and T.R. Preston, 1999. Rumen environment and feed degradability in swamp buffaloes fed different supplements. *Livest. Res. Rural Dev.* 11(3): Paper No. 25. <http://www.lrrd.org/lrrd11/3/thu113.htm>
- Yindee, M., 2011. Reproduction and genetic diversity of the swamp buffalo. www.vetscite.org/publish/articles/000096/index.html [May 2014]
- Zerbini, E. and A.G. Wold, 1999. Feeding dairy cows for draught. In; *Smallholder Dairying in the Tropics*. Zerbini, E., Wold, A.G., Falvey, L., Chantalakhana, C. (eds.). ILRI (International Livestock Research Institute), Nairobi, Kenya. 462pp

Developing technology and husbandry skills required for efficient animal production in villages in the highlands of Papua Indonesia

S. Mahalaya^{*1} & C.F. Cargill²

1Dinas Perikanan dan Peternakan, Kabupaten Jayawijaya, Papua Province, Indonesia, 2South Australian Research and Development Institute, Roseworthy Campus University of Adelaide, Roseworthy 5371, Australia

Abstract

A participatory approach, using a multidisciplinary project team, was applied to improve the technology of pig production and pig husbandry skills of a group of farmers in the Baliem Valley of Papua Province in Indonesia. The multidisciplinary team included social scientists, agronomists, extensionists and specialists in animal production, health and welfare. A series of on-farm experiments and demonstration trials was used to develop the technology and husbandry skills required to produce pork efficiently and cost effectively. Once a sustainable confinement system had been developed and validated for pig production, the multidisciplinary team worked with farmers to diversify cropping from a monoculture of sweet potato production, to enable crop diversification to provide more balanced diets for humans, pigs and other monogastrics. When new crops had been established and farmers were competent at pig production, two other monogastric species, rabbits and village chickens, were introduced. Again a participatory approach was used to introduce the technology required and transfer husbandry skills acquired for pig production to rabbit and poultry production, including egg production. The experience from the field trials and demonstrations, together with the farmers' comments and assessments, were used to write a series of extension materials as a basis for training other farmers. The extension material consisted of two levels of information transfer. The first was a series of single one page documents with minimal descriptions, but suitably illustrated, to enable a basic understanding of how to establish and operate each facet of the production system, and published in a manual format with replaceable pages. The second was a book containing more detailed information including the concepts involved in each facet of production, as well as how to build, develop, operate and manage each system. A group of successful farmers was selected, and provided with the knowledge and skills required to train other farmers. Finally a series of training workshops was offered in each production system: pig production, village poultry production, rabbit production and various crop production systems. During a period of 5 months, training sessions (2 sessions for each crop and livestock system, 6 days/session, 3 days/week) were provided to farmers and their families and 309 individuals from 18 villages received training. Some chose only strawberries and pigs, while others chose multiple crops, as well as rabbits and/or poultry. The training sessions were divided each week between a day of class meetings, a full day practical session, and a full day field visit to a local farm. The model described proved effective in enabling farmers to move from subsistence to small commercial production systems.

Keywords: farmer-training, multidisciplinary, pigs, poultry, rabbit

**Corresponding author: s.mahalaya@gmail.com*

Introduction

The development of technology and husbandry skills is required for efficient animal production particularly in many remote villages in the developing countries of Asia, including the highlands of Papua, Indonesia. Developed technology and improved husbandry skills through a series of participatory on-farm experiments, demonstration trials and farmers training sessions have led farmers in the Baliem Valley of Papua to move from subsistence agriculture to smallholder commercial enterprises (Cargill & Mahalaya, 2015). This paper aims to demonstrate how a participatory approach, using a multidisciplinary project team, was applied to improve the technology of animal production and husbandry skills of a group of farmers in the Baliem Valley of Papua.

Methods

A participatory approach, using a multidisciplinary project team, was applied to firstly improve the technology of pig production and pig husbandry skills of a group of farmers in the Baliem Valley of Papua. The multidisciplinary team included social scientists, agronomists, extensionists and specialists in animal production, health and welfare. A series of on-farm experiments and demonstration trials was used to develop the technology and husbandry skills required to produce pork efficiently and cost effectively. Once a sustainable confinement system had been developed and validated for pig production, the multidisciplinary team worked with farmers to diversify cropping from a monoculture of sweetpotato production, to enable crop diversification to provide more balanced diets for humans, pigs and other monogastrics. When new crops, i.e. soybean, red bean, pigeon pea and strawberry had been established and farmers were competent at pig production, two other monogastric species, rabbits and village chickens, were introduced. Again a participatory approach was used to introduce the technology required and transfer husbandry skills acquired for pig production to rabbit and poultry production, including egg production.

Results & Discussion

Extension materials

The experience from the field trials and demonstrations, together with the farmers' comments and assessments, were used to write a series of extension materials as a basis for training other farmers. The extension material consisted of two levels of information transfer. The first was a series of single one page documents with minimal descriptions, but suitably illustrated, to enable a basic understanding of how to establish and operate each facet of the production system, and published in a manual format with replaceable pages (Appendix 1 – To be shown during presentation). The second was a book containing more detailed information including the concepts involved in each facet of production, as well as how to build, develop, operate and manage each system (Appendix 2 – To be shown during presentation). Every farmer opinion was counted. The multidisciplinary team worked together with the participating farmers from planning, doing experimentations or demonstrations, to monitoring and evaluation. This participatory model included the development of extension materials. It makes sure that the developed technology meets the need of farmers. There was evidence that a participating farmer from the village of Napua using the extension materials as a guide was able to adapt the technology by running an extra experiment after the project has no longer provided any supports.

Training sessions: Training of trainers and farmers-to-farmers extension

A group of successful farmers was selected, and provided with the knowledge and skills required to train other farmers, i.e. Training of Trainers (ToT). Finally a series of training workshops, i.e.

Farmers-to-Farmers extension (FtF) was offered in each production system: pig production, village poultry production, rabbit production and various crop production systems. During a period of 5 months, training sessions (2 sessions for each crop and livestock system, 6 days/session, 3 days/week) were provided to farmers and their families and 309 individuals from 18 villages received training (Table 1). Some chose only strawberries and pigs, while others chose multiple crops, as well as rabbits and/or poultry. The training sessions were divided each week between a day of class meetings, a full day practical session, and a full day field visit to a local farm.

Table 1. Training sessions conducted in the Baliem Valley of Papua: August – December 2014

Session	Date	Host	Participant	Subject
I	20 – 22 Aug	Wamena	26 Farmers (16 villages)	Pig
	27 – 28 Aug	Wamena	26 Farmers (16 villages)	Chicken and Rabbit
II	15 – 17 Sep	Asologaima	20 Farmers (3 villages)	Pig
	22 – 23 Sep	Asologaima	20 Farmers (3 villages)	Chicken and Rabbit
III	20 – 22 Oct	Wamena	24* Farmers (16 villages)	Sweetpotato, Strawberry, Soybean, Red bean, Pigeon pea
	27 Oct	Wamena	26* Farmers (16 villages)	Crop processed products
	28 Oct	Wamena	37 WVI students (5 districts) and staff, and Churches' woman members	Crop processed products
	29 Oct	Wamena	16 Local government staff	Crop processed products
IV	10 Dec	Wamena	80 Farmers (18 villages)	Crop processed products – Competition
	11 Dec	Wamena	80 Churches' woman groups and local government staff	Crop processed products – Competition
Total	Aug – Dec 2014		309 participants (All the groups)	

*The 24 and 26 farmers who studied crops in the session III were part of the 46 farmers who studied livestock during sessions I and II

The ToT provided the selected farmers with the knowledge and skills required to train other farmers. When they led the FtF, they gained more attention and respect than outsiders who come to train. One of the advantages was that they spoke with their local languages in which everyone is familiar with any terms used during the training. Moreover, the FtF was an efficient method to extend the transfer of technology. When it combined with a competition as it was applied by the project for which an alumnus of the FtF brought four other farmers, this strategy has delivered the technology to reach even more farmers. As a conclusion, the model described proved effective in enabling farmers to move from subsistence to small commercial production systems.

References

Cargill, C. and S. Mahalaya. 2015. Improvement and Sustainability of Sweetpotato-Pig Production Systems to Support Livelihoods in Highland Papua and West Papua, Indonesia. Final Report. Australian Centre for International Agricultural Research (ACIAR), Canberra. pp40.

Acknowledgment

The project was funded by the Australian Centre for International Agricultural Research (ACIAR).

Partnership in broiler farm closed house system (case study at Tuban, East Java, Indonesia)

*Utami H.D. *, A.P. Seruni & G.Maliton*

Social Economic Department of Animal Husbandry Faculty, Brawijaya University, Veteran street, Malang, East Java 65145 Indonesia.

Abstract

Study was conducted in the broiler closed house partnership program with “Semesta Mitra Sejahtera” (SMS) company at Tuban Regency, East Java Province, Indonesia. Research was aimed to examine financial performance of seven broiler production periods (BPP) during one year. Purposive sampling method was employed to select one plasma farmer (owned about 10,000 birds). Data collections were held during a month from 21st August to 21st September 2014. The data were analysed by descriptive analysis with applying economic equation involving production costs, revenue, profit, Break Even Point (BEP), and R/C ratio. Results discovered that producing one kilogram live weight broiler required the lower production costs of IDR 13,181 (US\$1.08) in BPP-1, with more revenue (IDR 17,767.79 or US\$ 1.46) of BPP-7, and the higher profit (IDR 2,160.39/Kg or US\$ 0.18) come from the sixth broiler production period. The BPP-1 appeared more efficient in operating this poultry farm with IDR 13,181 of BEP per Kg live weight broiler and 1.141 of R/C ratio.

Keywords: production costs, revenue, profit, R/C ratio

Introduction

Meat Chicken industry contribution accounts for up to 49.95% of total meat production (Directorate General of Animal Husbandry, 2011). Indonesia’s “Poultry Partnership Farming” system can promote promising broiler farming benefit for both the core company and the plasma farmers. This partnership plays an important role in generating revenue and income and therefore, it can improve farmer’s welfare (Wainaina, 2012), as means in reducing poverty for most rural poor (Jensen & Dolberg, 2003; McDermott, 2010). However, some divergence implementation can emerge the conflict of interest because of the unbalance in profit sharing between core Poultry Company and plasma farmers (Sumartini, 2004). The study therefore, was aimed to examine the broiler partnership farming program and its role in plasma farmer’s income at Malang, East Java Province, Indonesia.

Methodology

The case study was held at broiler closed house scheme (controlled 10,000 birds) partnership with “Semesta Mitra Sejahtera” (SMS) company at Tuban Regency, East Java Province, Indonesia. Purposive sampling method was employed to select one plasma farmer with applying seven broiler production periods (BPP) during one year. Data collections were held during a month from 21st August to 21st September 2014. Survey method using structured questionnaire were applied to interview farmers for gathering primary data, while secondary data were obtained from the “SMS” company and related sources. The data were analysed by descriptive analysis with applying economic equations (i.e. production costs, revenue, and profit, Break Even Point (BEP), and R/C ratio).

Results and Discussions

Producing one Kilogram live weight the broiler provide profit about IDR 1,687 on average, with less (IDR396) and higher (IDR 2,160) income were structured by BPP-7 and BPP-6, respectively. The broiler plasma farm represents efficient in operating this farming as seen in IDR 15,101 of BEP per Kilogram of broiler and 1.11 of R/C ratio.

Revenue of one Kilogram broiler was IDR 17,227 on average, with the maximum revenue of IDR 17,768 come from BPP-7, whereas BPP minimum one was IDR 15,041 from the first broiler production period (BPP-1). Figure 1 describes the dominant selling live broiler (98.10%) in the fourth production period. However, Figure 2 discovered the BPP-1 has maximum bonus and incentive (2.93 %) as well as selling manure and sack feed (0.74%).

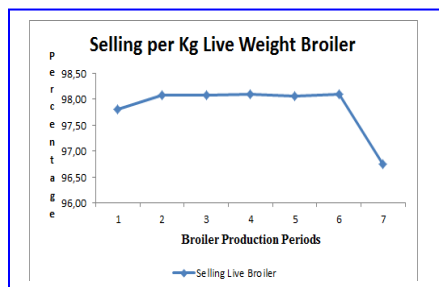


Figure 1. Selling Broiler and Its

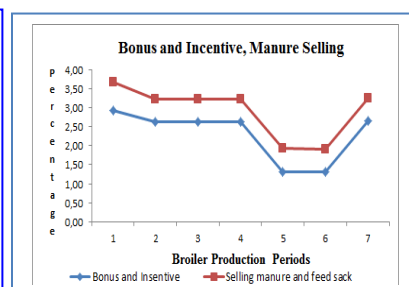


Figure 2. Bonus, Incentive,

Production costs per Kilogram broiler live weight, it was IDR 17,372 on average with BPP-1 .represented efficient (IDR 13,181) and he maximum one indicated for BPP-7 (IDR 17,372). The efficient expenditure was DOC (16.86%) of BPP-3, feed cost (72.21%) of BPP-7 (Figure 4), and medicine (0.99%) and rise husk (0.99%) in BPP-2 (Figure 3).

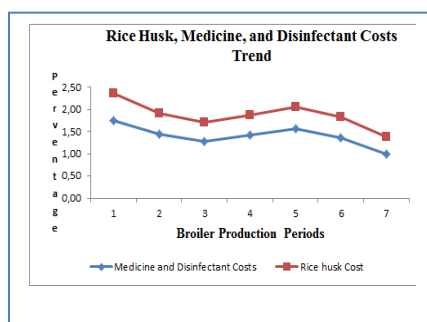


Figure 3. Rice Husk, Medicine, Desinfectant Trend.

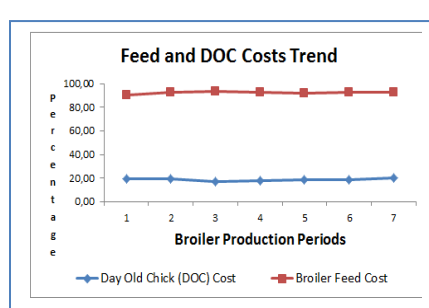


Figure 4. Feed and DOC Costs. Trend

The first broiler production period appeared more efficient in operating this poultry farm with IDR13,181 of BEP per Kg broiler live weight and 1.141 of R/C ratio. In contrast, BPP-7 was inefficient based on per Kg BEP (IDR 17,372) and R/C ratio (1.03). Therefore, the BPP-1 indicated efficient since they implemented a good management in raising their farms. Also, more cash bonus were provided by core poultry industry and it can encourage farmers to join in promising prospect of broiler farming in poultry partnership scheme.

Conclusions

1a. BPP-1 required the lower production costs of IDR 13,181 (US\$1.08) than IDR17,372.03 (US\$1.43) of BPP-7 in producing one kilogram live weight broiler.

1b. BPP-7 can gain more revenue (IDR17,767.79 or US\$ 1.46) than those of BPP-1 (IDR 15,048.58/ or US\$1.24).

1c. BPP-6 obtained the higher Profit (IDR2,160.39 or US\$ 0.18)

2. The first production period appeared more efficient in operating this poultry farm with IDR13,181 (US\$1.08) of BEP per Kg broiler live weight and 1.141 of R/C ratio. In contrast, BPP-7 was inefficient based on BEP (IDR17,372.03) and R/C ratio (1.03).

References

- Jensen, H.A., and F. Dolberg, 2003. A conceptual framework for using poultry as a tool in poverty alleviation (1). *Livestock Research for Rural Development* 15 (5)2003.
- Sumartini. (2004). Partnership in agribusiness and its effects on farmers' income: A study at Broiler industry in Bandung Regency, Indonesia. Unpublished Thesis. Agricultural University, Bogor, Indonesia.

Moving families from subsistence animal production to small commercial production using a participatory approach with a multidisciplinary team

Cargill¹, C. F., & S. Mahalaya²

¹South Australian Research and Development Institute, Roseworthy Campus University of Adelaide, Roseworthy 5371, Australia, ²Dinas Perikanan dan Peternakan, Kabupaten Jayawijaya, Papua Province, Indonesia

Abstract

Pigs and sweetpotato (SP) production are the dominant agriculture practiced in the highlands of Papua Indonesia. SP is the principal food for humans and pigs, and pigs providing the majority of social and economic capital for families. The dominance of the SP-pig system and the lack of agricultural diversity are key factors in childhood malnutrition. Without improved agricultural and husbandry technology, families are trapped in subsistence agriculture and lack the skills to move to smallholder commercial enterprises. Zoonotic parasite infections in pigs are also a major health risk for humans. A participatory approach, with a multidisciplinary team, was taken to improve husbandry skills and diversify animal production. Pig production technology and husbandry skills were improved and transferred to other monogastric species. Nutritional practices were improved through increasing dietary knowledge and developing more balanced diets through crop diversification. Pig health problems, including zoonotic parasites, were reduced by developing a sustainable pig confinement system (PCS). Pigs were housed in pens overnight and given access to high protein pastures during the day. Growth rates in PCS pigs fed diets based on silaged SP and supplemented with golden snails ranged from 230 ± 23.4 to 280 ± 32.3 g/day compared with 110 ± 26.5 g/day to 150 ± 51.4 g/day in pigs fed traditional uncooked diets. The prevalence of parasites was reduced in PCS pigs over an 18 month period to below 15% (0-15% depending on species) in PCS pigs, but increased (20-200%) in scavenger (non-PCS) pigs. More importantly, the prevalence of antibodies to *C. Cellulosae* was reduced by 22% in PCS pigs but increased by 28% in non-PCS pigs. Once farmers were proficient at pig production, their new husbandry skills were transferred to rabbit and poultry production, which have similar dietary requirements to humans and pigs and need minimal land area. Based on current prices a household with 2 sows will recover cost of developing a PCS within 3 years. Households who adopted rabbit production produced up to 40 rabbits for consumption or sale/year and sold 3 month old kittens for around US\$ 30. Households who diversified into poultry production consumed eggs and sold the excess for US\$ 0.43.

Keywords: diversification, pigs, monogastrics, malnutrition, health

Corresponding author: snout-n-about@bigpond.com

Introduction

High poverty levels, together with a high prevalence of Cysticercosis in humans and pigs were the major reasons for investigating the pig - sweetpotato (SP) production systems in the highlands of Papua Province Indonesia. Pigs husbanded in the existing free range scavenger system share the family compound with dogs and children, creating a high risk for cross infection with zoonotic parasites. The dominance of pigs and SP was also a major factor in

childhood malnutrition and preventing families from moving from subsistence to small scale commercial agriculture (Cargill et al., 2009). Verma (2007) demonstrated that farm incomes can be increased through agricultural diversification with the introduction of high value crops and livestock. This report describes a methodology for moving families from subsistence to small scale commercial farming by improving pig husbandry skills, and then transferring those skills to other livestock production system to diversify income streams and improve human health and childhood nutrition.

Materials and Methods

A participatory approach, with a multidisciplinary team, was taken to improve husbandry skills and diversify animal production. Participating families, technicians, and scientists were included in all planning and review sessions and technical support provided to families regularly. A diet based on cooked SP roots and vines was designed and compared under experimental conditions with the traditional feeding methods. This diet provided a base-line for assessing other diets using local ingredients and ensilaged materials. Experimental pigs were fed approximately 10% of their body weight daily and feed intake recorded. Pigs were weighed monthly and feed intake adjusted. Experimental pigs were medicated monthly to eliminate parasites. Once diets had been validated, a pig confinement system (PCS) was designed to provide pigs with suitable housing at night and access to high protein pasture, shade and water during the day. The cost of construction and operation was recorded and once production in the PCS had been validated, the health of pigs was investigated. Ten PCS households and 10 non-PCS households were selected for study. Pigs were treated with a single dose of Oxfendazole at 30 mg/kg prior to the study. Individual fecal samples were collected from 5 pigs at each household at 3 monthly intervals over an 18 month period, preserved in sodium acetic formaldehyde (SAF) and qualitatively examined for intestinal parasite stages by the SAF concentration technique (Marti and Escher, 1990). Blood samples were collected and serum stored at -20⁰C until qualitatively screened for IgG antibodies to *C. cellulosae* using an ELISA. A list of crops suitable for climate, altitude and soil types was used to determine farmer and community preferences. High protein crops that grew at higher altitudes were preferred. Animal species that could be introduced into the region were ranked according to land use, nutritional requirements, and social and religious limitations.

Results and Discussion

Pig Diets

The basic cooked diet developed consisted of 56% cooked SP vines, 33% cooked SP roots, 11% cooked banana trunk, and 0.5% salt. The basic ensilaged diet consisted of 33% cooked SP vines, 22% cooked SP roots, 34% ensilaged SP tubers and vines (85% roots + 15% vines), 11% cooked banana trunk and 0.5% salt. Both diets were later supplemented with either 20% fish offal or 5% golden snails (*Pila spp*) as a source of animal protein. Snails proved more sustainable than fish offal and were produced in small ponds and easily harvested. Adding papaya fruit or betel nut to diets was found to reduce endoparasite burdens further.

Pig confinement system (PCS)

Pigs were confined in houses overnight and given access to high protein pastures, shade and water during the day. Up to 8 small pasture paddocks were provided and pigs were moved to fresh pasture when 50% of the leaf material had been consumed. Healthy village pigs husbanded in this system and fed ensilaged and cooked diets grew up to 10 times faster (250 to 300 g/day) than scavenger pigs fed diets of raw SP leaves roots and vines. Families using PCS models produced an average of 12 pigs/sow/year, compared with 5 in traditional systems, and recovered the cost of new facilities within 3 years. Significant health benefits for both pigs and humans were also recorded. The prevalence of internal parasites decreased in PCS pigs to below 15% (0-

15% depending on species) but increased in non-PCS pigs by 20 to 200%. The prevalence of antibodies against *C. cellulosae* also decreased by 22% in PCS pigs but increased by 28% in non-PCS pigs, thus reducing the risk of zoonotic parasites in pork.

Livestock Diversification

Rabbits: Each farmer was given 2 does and 1 buck and assisted to build rabbit hutches. A standard FAO model with three cages (30 x 50 cm/cage), set one meter above ground, with wooden floors and walls, and a thatched roof was used. The cages were covered by a larger thatched or zinc roof (3 x 0.5 m to 4 x 0.5 m). Simple diets using combinations of SP roots and carrots supplemented with green leaf material (SP vines, cabbage leaves, high protein forage pasture and fodder trees) were formulated. Water and fresh green leaf material was provided twice daily and SP roots and carrots three times weekly. Whole corn cobs were provided 2-3 times/month. Families produced up to 40 rabbits per year, which they either consumed or sold as live rabbits in local markets. Full grown rabbits sell for between US\$ 43.50 to US\$ 70 and 3 month old kittens sell for around US\$ 30. However, prices are expected to drop as production increases.

Village poultry: Fifteen 4 month old KUB village chickens bred by the Indonesian Institute of Animal Production (13 females and 2 males) were distributed to each family. Birds were kept in houses (12.5 m² - 2.5 x 5 m) constructed with wood and a metal roof, with a secure outside area. Nest boxes (50 x 30 cm) were located one meter above the ground and filled with dry grass. Feeders and drinkers were provided. Birds commenced to lay at 7 to 8 months and continued to lay for around 9 months. The average egg weighed 30g and they were consumed by families or sold locally (US\$ 0.43/egg). As meat from hybrid village chickens is preferred to imported poultry meat, the current price for a full grown bird ranges from US\$ 15.00 to 18.00, compared with imported chicken meat which sells for US\$ 3.40 to US\$ 3.90. It is assumed that prices will decrease as supply increases.

Conclusion

A participatory approach with a multidisciplinary team, which included farmers and their families in all stages of planning and review, proved to be a successful methodology for improving pig production and establishing new crops and livestock production systems. Cash flow, family income, and human and pig health were significantly improved by developing a confinement model for pig production. New crops were introduced to improve animal and human nutrition to enable diversification of livestock production. Increased animal husbandry skills were used to successfully introduce rabbit and chicken meat and egg production. Farm incomes were significantly increased, human health improved, and childhood malnutrition reduced.

References

- Cargill, C. F., S. Mahalaya, Tjintokohadi, and T. A. Syahputra. 2009. Poverty alleviation and food security through improving the sweetpotato-pig systems in Papua Indonesia (AS1/1998/054). Australian Centre for International Agricultural Research, GPO Box 1571, Canberra ACT 2601, Australia. Final Report FR2009-39
- Verma, M. R., K. K. Datta, S. Mandal and A. K. Tripathi. 2007. Diversification of Food Production and Consumption Patterns in India. *J Agric & Food Information* 8: 87-100.

Acknowledgement

The project was funded by the Australian Centre for International Agricultural Research (ACIAR), GPO Box 1571, Canberra ACT 2601, Australia.

Development of local cattle with sustainable in North Sulawesi

Rundengan, M. L & T.F.D. Lumy

¹*Department of Social Economics, Faculty of Animal Husbandry,
University of Sam Ratulangi, Jl. Campus BahuKleak Manado 95115, North Sulawesi, Indonesia*

Abstract

Local cattle is a source of income for people in North Sulawesi. The problem is local cattle was developed traditionally. Cattle in certain areas, developed under the coconut trees, where the cattle consume grass that grows wild. Cattle, in other areas of grazing farms, and cattle consuming agricultural wastes, such as corn waste. Development in other areas, cattle are given rice straw as feed to meet the needs of cattle. The purpose of this study was to determine the pattern of development of local cattle with sustainable. Sample location is South Minahasa and Bolaang Mongondow, which has been determined by purposive sampling, because it has the largest cattle population. The results showed that farmers' income is affected by several factors. Earned income of farmers is higher when cattle were developed with an integrated pattern of cattle-coconut and cattle-food crops. In conclusion, the development model of local cattle in North Sulawesi should be integrated and sustainable. Introductions feed technology can be done to improve the productivity of local cattle. Development of local cattle with sustainable, will be maximal with the intervention of the government.

Keywords: development, local cattle, sustainable

Introduction

North Sulawesi is one of the provinces that have the prospect of the development of local cattle as sources of rural income. Local cattle can be relied upon by farming communities to support their needs. Local cattle farming was successful when contributed income to meet the needs of farmers' lives (Hoddi et al., 2011). Local cattle, is also used as labor in managing agricultural land, for plowing and transporting agricultural products. Waste from cattle has been used as raw material compost, and even thrive as alternative energy sources (biogas).

The problem, local cattle still cultivated sideline with traditional maintenance system. Local cattle, in certain districts in North Sulawesi, developed by utilizing the land under coconut trees. Local cattle, in this case eating wild grass that grows under the coconut trees. Local cattle, in other districts, are left in agricultural land which then consume agricultural waste such as corn. Development in other areas, local cattle are given rice straw as feed in meeting the needs of the animal. The question is, to what extent, the pattern of development of local cattle, in North Sulawesi. Study that has been done is aimed to determine the pattern of development of sustainable local cattle.

Materials and Methods

This research has been carried out in North Sulawesi by using the survey method. The district has been determined by purposive sampling the district has the largest cattle population is South Minahasa and Bolaang Mongondow. Respondents in this study were 150 farmers for the South Minahasa District and 65 respondents to Bolaang Mongondow. The data collected is primary data and secondary data. Analysis of the data used is descriptive analysis and multiple regression analysis.

Results and Discussion

Cattle farmers in the research area has largely been developing cattle integrated with coconut plantations. System integration cattle and crops are often considered as a step forward. Land held by farmers in the area of research, ranging from 0.5-3 ha. Most of the land owned by the farmers according to the results of research conducted Rundengan (2013), ranging between 1-1.5 ha. Land tenure is very supportive of efforts for the development of local cattle, because according to Hermawan & Utomo (2012) of local cattle is a strategic commodity with multiple functions for dry land farmers. The development of beef cattle cannot be separated from the development of agriculture and plantation (Hartono, 2012). Results of regression analysis, income from cattle farming, which is integrated with coconut and corn accordance Rundengan (2013) can be seen in the following equation:

$$Y = 779,524 + 7,295,119LH - 3.24PKN - 1.75PO - 1.16PS - 0.14TKS.....(1)$$

Income from the cattle farming is affected by the plantation area. Increasingly plantation area (LH), then the income tends to increase (Equation 1). Coconut is a commodity that is inseparable from public life. Coconut dubbed the tree of life, because of coconut has arolet people's lives, from the fulfillment of social needs, culture until the economic interests.

Equation (1) shows the cost to feed (PKN) the higher cause income tend to decline. The indication, with an integrated cattle farming, the waste from the farm under the coconut trees can be used as feed. Local cattle farming, managed by farmers without the cost of feed (zero cost). Advantages of application integration pattern can be obtained because of the synergy between activities, which in turn almost no resource is wasted (zero waste). Farmers for crops has not optimally utilize agricultural waste even some farmers burning the waste which can affect the loss of main nutrients, such as N and P, kill organisms in the soil and produces CO₂ gas that damage the environment.

Increase in the cost of organic fertilizers (PO) cause income tend to decline (Equation 1). According to Haryono (2013), the use of compost as organic fertilizer, a choice in favor of an increase in the productivity of food crops. Development of local cattle, will have a negative impact on environment, due to the waste generated from the cattle farming. According Harlia et al. (2012), waste from cattle is increasing, causing nature is not able to decipher, absorb and neutralize the waste. Local cattle, for farmers, in this case, serves as a producer of manure (Roehani et al., 2005), as an organic fertilizer raw materials. Baba et al. (2012) suggested that the integration of cattle corn, provide many advantages for both farmers (increase income and food security), local cattle (sustainability feed), and land (conservation land). Land degradation at this time, a problem faced by many countries (Herrick et al., 2010), including our country. Waste utilization of local cattle, as organic fertilizer, which is useful to the plant, as well as improve the

physical structure and chemical soil on degraded land.

Labor costs of rent (TKS), which is the higher resulting in income tends to decline (Equation 1). Local cattle farm in the research area are traditionally cultivated so that farmers use family labor. The number of cattle owned by farmers increasingly means requiring more labor. But the development of local cattle farming with an integrated system, resulting in the use of labor becomes more efficient because its activity can synergize. According Utomo et al. (2005), there are several components of the activities implemented with an integrated approach, first: technology cultivation of food crops, namely the test of adaptation of improved varieties. Second: cattle farming technology, which includes management of the enclosure, the technology of straw fermentation processing, and processing of compost. Technological innovation is critical to the development and enhancement of the added value of local cattle farming (Mariyono, 2009).

Conclusions and Suggestions

Based on the research that has been done can be concluded, that the model of development of local cattle, in North Sulawesi, should be done in an integrated, and sustainable. Introductions feed technology can be done to improve the productivity of local cattle. Development of sustainable local cattle, will be maximized if no intervention from the government.

References

- Baba, S., B. Ibrahim., A. Muktiani., and M. Yasin, 2012. Farmers' Problem in Carrying Out Corn and Cattle Integration in South Sulawesi. A Case Study in Bontonompo Gowa South and Mallawa Maros Subdistrict. Proceedings of the National Seminar on Sustainable Livestock 4, Farmer Agribusiness Innovation for Food Security. Animal Husbandry Faculty of Padjadjaran University. Bandung. pp: 654
- Harlia, E., E.T. Marlina., A. Firman and A. Fitriani, 2012. Dairy Farm Waste Utilization, and Beef Cattle As Alternative Energy Sources Biogas in Sumedang. Proceedings of the National Seminar on Sustainable Livestock Breeders 4. Innovation Sustainability for Food Security. Faculty of Animal Husbandry, Padjadjaran University. Bandung. pp: 654
- Hartono, B., 2012, Role of the carrying capacity of the region, to the development of Madura cattle farming. Journal of Economic Development. Vol. 13. No2, Des 2012. pp: 316-326.
- Haryono, 2013. Policy Strategy of the Ministry of Agriculture in Optimization of Land Suboptimal, National Food Security Support. Proceedings of the National Seminar on Land Suboptimal: "Intensification of Suboptimal Land Management in the Context of National Food Self-Reliance Support. Palembang, 20-21 Sept 2013. pp:15-22.
- Hermawan, A and B. Utomo, 2012. Role of Ruminant in the Development of Conservation Farming Systems in Dry land watershed upstream section. Proceedings of the National Seminar on Sustainable Livestock 4, Farmers Agribusiness Innovation for Food Security. Faculty of Animal Husbandry, Padjadjaran University. Bandung. pp: 654
- Herrick, J.E. V. C. Lessard, K. E. Spaeth, P. L. Shaver, R. S. Dayton, D. A. Pyke, L. Jolley, and J. J. Goebel, 2010. National ecosystem assessments supported by scientific and local knowledge. J. Frontiers in Ecology and the Environment. Volume 8, Issue 8 (October 2010). pp: 403–408. 12/12/2011.

- Hoddi, A.H., M.B. Rombe and Fahrul, 2011. Income analysis of beef cattle breeders in Tanete Rilau Subdistrict, Barru Regency. *Journal of Agribusiness*, Vol X (3) Sept. 2011: 1-11.
- Mariyono., 2009. Beef Cattle Research, to Support Agribusiness Farmer. National Seminar on Technology Farmer and Veterinary.
- Roehani, E.S., A. Subhan., N. Amali., Sumato and A. Darmawan., 2005. Income contribution from Maintenance of Cattle, in System Integration Corn and Cattle in Dryland. National Seminar on Animal Husbandry and Veterinary Technology.
- Rundengan, M.L., 2013. Optimization of Integrated Farming Between Beef Cattle With Coconut Plantation in South Minahasa District of North Sulawesi Province. Dissertation. Doctoral Program Livestock Agribusiness Interests. Faculty of Animal Husbandry, UB, Malang. pp: 256
- Utomo, B.N., A. Krismawati and E. Widjaja, 2005. The study for Integration Cow-Rice /Corn in Dryland Central Kalimantan. BPTP of Central Kalimantan. National Seminar on Crop Livestock Integration System.

Analysis of the resource potential of the coconut crop-cattle in the District of East Likupang

Salendu, A.H.S., F.H. Elly, F.S.G. Oley & A. Makalew

Department of Social Economics, Faculty of Animal Husbandry, University of Sam Ratulangi, Jl. Campus Bahu Kleak Manado 95115, North Sulawesi, Indonesia

Abstract

Coconut land in District of East Likupang, an area of 2,942 ha (8.3% of the area of North Minahasa Regency), mostly used for cattle development. The problem is, whether the coconut land has the potential to support the development of cattle in this area. The purpose of research that has been done is to analyze the potential of carrying coconut-cattle. The research method that has been used is a survey method. East Likupang districts have been determined by purposive, because it has the largest cattle population. Analysis of the data that has been used is the analysis of the Effective Potential of Livestock Development. The results showed that the maximum potential of land resources (PMSL), is equal to 2364.172 UT. That is, based on land resources, can still accommodate cattle of value PMSL. The maximum potential by farmers households (PMKK), is at 6627 UT. That is, based on the availability of labor, livestock population can be increased up to the amount of the PMKK. In conclusion, the land under a palm tree in the district of East Likupang, has the potential for development of cattle. It is, seen from the potential of land as a source of forage, and resource potential farmers. The role of government, is needed in order to increase the potential carrying capacity of land, environmentally friendly in this area.

Keywords: potential, carrying capacity, coconut, cattle

Introduction

Environmentally sustainable agricultural approach starting with the ecosystem approach. Ecosystem is a unit arrangement completely and thoroughly between elements of the environment which interplay. Agroecosistem, by experts, distinguished on the paddy field, dry land, and the coast. Dry land has the potential for agricultural development, such as plantation crops. Coconut is one of the plantation crops, as a source of income of the people in the district of East Likupang. Coconut plant according Rusdiana & Adawiyah (2013), is a commodity that is most widely spread in the archipelago including in the district of East Likupang. Cattle in this area as the entry point is very beneficial for dry land agro ecosystem. Coconut plantations, has the opportunity to be developed into are gional cattle farming, especially farming from the cow calf operation (CCO) with CLS integration patterns in-situ (Rusdiana & Adawiyah, 2013).

Land of coconut plantations in this area, covering an area of 2942 hectares, which is about 8.3% of the North Minahasa Regency. The land, mostly used for cattle development. However, cattle are still traditionally developed, by way tied under the coconut trees, and

consume the grasses that grow on the land. In fact, cattle require a consistent feed intake, both in quantity and quality.

The problem is, whether the palm plantation in the district of East Likupang has the potential to support the development of cattle. The extent of potential oil plantation in this area in supporting the development of cattle. This research was conducted aiming to analyze the potential of coconut-carrying cattle, in District Likupang East, North Minahasa regency.

Materials and Methods

This research has been conducted in the district of East Likupang, using survey methods. District of East Likupang has been determined by purposive sampling, namely one in the District of North Minahasa Regency which has the largest cattle population. The data source that has been used, is the North Sulawesi BPS data (2013) and data from BPS District of East Likupang (2013), which has been used as the primary data. The data analysis was conducted, was using Effective Livestock Development Potential Analysis.

Results and Discussion

The dry land today, faced with the challenge of highland degradation, which causes degraded land sorun productive. Land area, is the determining factor of survival of the agricultural sector. Coconut is an important commodity for the economy of East Likupang districts, and as the brand image of this area. Cattle development can encourage farmers to apply the principles of land conservation, sustainable manner. Dry land management, sustainable, requires professional handling and follows the rules of the environment.

The amount of carrying capacity and sustainability of land productivity is determined by the interaction between the ways humans manage the resource itself with biophysical environmental factors (Salendu et al., 2012). Results of research have been done on the potential development of livestock effective, in the district of East Likupangsummarizedin Table1.

Table 1. Effective Livestock Development Potential in District East Likupang.

Coefficient/Variable	Development Potential Value
PMSL	2364.17
KPPTR (SL)	666.17
PMKK	6627.00
KPPTR (KK)	4929.00

The maximum potential of land resources (PMSL) under the coconut trees according to analysis doneby2364.17UT. That is, based on coconut plantation land resources, can still accommodate cattle with a population of value PMSL. Results of the analysis has been done, the capacity increase in the cattle population by land resources (KPPTR (SL) coconut plantations amounted to666.17UT. That is, to meet the maximum potential of land resources, the cattle population can still be increased by666.17UT. The indication, land in coconut plantations in this area can be optimized through the introduction of forage. Feed for cattle is a problem faced by cattle farmers (Alfian et al., 2012; Nugrahaetal., 2013; Rusdiana & Adawiyah, 2013; Rahmansyah et al., 2013; Salendu & Elly, 2013).

Adequacy of forage, in the sense of quantity and quality is a major requirement in

breeding as well as an increase in the cattle population (Rusdiana & Adawiyah, 2013). Planting forage is a central point for the development of cattle so it is necessary efforts to optimize land under coconut. Availability of feed during the dry season is reduced, but abundant rainy season (Riswandi et al., 2012; Rahmansyah et al., 2013), so the cattle development in the future very promising (Nugraha et al., 2013). Cattle population is difficult to achieve, due to the low productivity of the cattle (Noferdiman & Afzalani, 2013). Cattle development is done integrated with coconut plantations, is the best farm system (Ahmed et al., 2011). This is done to increase the productivity of cattle and land.

Analysis has been done to produce that, the maximum potential based head of family farmers (PMKK) amounted to 6627.00 UT. That is, based on the availability of labor, the cattle population may be increased to 6627.00 UT. Results of the analysis of the cattle population by KK farmers (KPPTR (KK) can be increased up to 4929.00 UT. In the agricultural system, which is based populist, labor generally come from the farm household itself (Abdullah et al., 2012).

Conclusions and Suggestions

Based on the results of research conducted, shows that the land under of coconut trees in the District Likupang East, has the potential for development of cattle, seen from the potential of land as a source of animal feed, and resource potential farmers. The role of government is needed in efforts to increase the potential carrying capacity of land environmental friendly in this area.

Acknowledgement

Thanks submitted to the Rector of the University of Sam Ratulangi which has given opportunity to the author to obtain research funding through seed grants UNSRAT2015.

References

- Abdullah, A., M.A. Asja and Zulkarnaen, 2012. Potential analysis capabilities livestock breeders in (KPT) cattle based integration on regional center for beef cattle South Sulawesi. Proceedings of the Seminar's National animal husbandry, sustainable livestock systems revitalization based integration with plantations to achieve food security. Medan, Aula Soeratmat FP-USU. pp 34-39.
- Ahmed, N., K. K. Zander and S. T. Garnett, 2011. Socioeconomic aspects of rice-fish farming in Bangladesh: opportunities, challenges and production efficiency. *Australian J. Agric and Resour Ec.* 55 (2011), 2 (April): 199–219.
- Alfian, Y., F.I. Hermansyah., E. Handayanto., Lutojo and W.P.S. Suprayogi, 2012. Analysis of the capacity of ruminants in the dry season in dry land farming areas in Semin Gunung Kidul Regency. *Tropical Animal Husbandry*, Vol. 1 (1). Okt. 2012: 33-42.
- BPS North Minahasa, 2013. North Minahasa in figures. Statistics North Minahasa.
- BPS Likupang Eastern District., 2013. In the district of East Likupang figure. Statistics Likupang Eastern District.
- Noferdiman, A.Y and Afzahani, 2013. Conversion of organic waste, into a complete feed silage, with the use of fermentation technology and probiotics supplementation on the growth of Bali cattle. *Journal of Research Univ. Jambi Science Series*. Vol. 15. No. 2: 51-56.

- Nugraha, B.D., E. Handayanta and E.T. Rahayu, 2013. Analysis of the capacity of ruminants in the rainy season in dry land farming areas in the district of Semin, Gunungkidul Regency. *Tropical Animal Husbandry*, Vol. 2 (1). Jan. 2013: 34-40.
- Rahmansyah, M., A. Sugiharto., A. Kanti and I.M. Suidiana, 2013. Kesiagaan pakan pada ternak sapi skala kecil sebagai strategi adaptasi terhadap perubahan iklim melalui pemanfaatan biodiversitas flora local. *Buletin Peternakan*, Vol 37 (2): 95-106.
- Riswandi., S. Sandi., M.L. Sari., Muhakha and A.I.M. Ali, 2012. PIncreased production of cattle, with ammoniation fermentation technology (amofer) rice straw in the village of Tanjung Pering District of North Indrabaya Oguilirregency of South Sumatera. *SriwijayaServiceJournal*.pp73-79.
- Rusdiana, S and C.R. Adawiyah, 2013. Economic analysis and business prospects crops and cattle, in the coconut plantations. *SEPA*. Vol. 10. No. 1. Sept 2013: 118-131.
- Salendu, A.H.S. and F.H.Elly, 2012. Land use under a coconut tree forage for cattle in North Sulawesi. *J. Pastura* Vol. 2 No. 1: 21-25.
- Salendu, A.H.S. and F.H. Elly, 2013. Agro ecosystem of coconut-cattle and carrying capacity analysis in Lolayan Subdistrict of Bolaang Mongondow Regency. *European Journal of Social Science*. Vol. 40. No. 4. Nov. 2013: 549-555.
- Salendu, A.H.S., Maryunani, Soemarsono and B. Polii, 2012. Integration of cattle-coconut in South Minahasa Regency. *Proceeding: The 2nd International Seminar on Animal Industry 2012 (ISAI) Faculty of Animal Science Bogor Agricultural University*.
- Salendu, A.H.S., 2012. Agro ecosystem Management perspective Coconut-Cattle in South Minahasa. *Doctoral dissertation. Graduate Program UB Faculty of Agriculture, Malang*. pp 233

Utilization of cattle waste as compost fertilizer

Elly¹, F.H., Syarifuddin², D. Polakitan³ & F.N.S. Oroh¹

¹Department of Social Economics, Faculty of Animal Husbandry, University of Sam Ratulangi, Jl. Campus BahuKleak Manado 95115, North Sulawesi, Indonesia, ²BP3K District of Bintauna, Regency of North Mongondow Bolaang, North Sulawesi, Indonesia, ³BPTP Kalasey, North Sulawesi, Indonesia

Abstract

The government of North Mongondow Bolaang, continuously motivate farmers to develop farming of cattle, as an effort to increase their income. The problem, livestock revolution, can exacerbate environmental problems. Waste of cattle can cause pollution to the environment. The purpose of this research has been done is to analyze the benefits of cattle waste is used as compost. Methods of research that has been done is a survey method and direct observation. The location of this research sample, is the District Bintauna, with respondents who have been determined, is a member of the group Mototavia. The results showed that the cattle population in the District Bintauna, as many as 1579 tails, can produce wet feces as much as 6.82128 million kg / year or 3.97908 million kg / year dry feces. Cattle waste has been made into compost by members of the group, and is sold at a price of Rp 1500 per kg. In conclusion, cattle waste which was created as compost, beneficial for the improvement of soil fertility, and farmers can minimize expenditure for the purchase of inorganic fertilizers. Suggestions, need further research to demonstration plots crops by using compost from cattle waste.

Keywords: waste, cattle, fertilizer of compost

Introduction

Cattle are a source of income for farmers in the North Mongondow Bolaang Regency. The Government of the Regency of North Mongondow Bolaang continues to encourage farmers in developing cattle business in an effort to increase their income. The problem, livestock revolution can exacerbate environmental problems. Waste from cattle can cause pollution to the environment. Cattle farm according Salendu & Elly (2013) is regarded as one of the causes of CO₂ emissions that can lead to increased global warming.

According Kuruseng & Alianti (2011), waste in the form of animal manure, an excellent basic ingredient in making organic fertilizer. Cattle are the largest producers of feces when compared to other livestock. In this case, the feces of cattle can be used as compost raw material is fertilizer. Fertilizer is cattle waste has been fermented properly, and contains a complete nutrient, required by plants for growth. Introduction of technology utilization of waste as compost in North Sulawesi has been undertaken for community service (Elly et al., 2010; Elly et al., 2011; Elly et al., 2012).

Based on the above problems, the research has been done on the use of cattle waste as compost. The purpose of this study was to analyze the benefits of cattle waste as compost.

Materials and Methods

Methods of research that has been done is to survey and direct observation method. The data have been collected primary data and secondary data. The location is the District Bintauna samples which have been determined by purposive sampling. Respondents have been determined in this research is purposive sampling is a member of the group Mototavia with consideration of this group has been selling compost. The number of cattle as many as 24 tails. Data analysis is descriptive analysis.

Results and Discussion

Results of research conducted show that the population of cattle, in District Bintauna, as many as 1579 animals, produce wet dirt as much as 6.82128 million kg / year, or 3.97908 million kg / year of dry feces. Dry dirt in this case as fertilizer could substitute inorganic fertilizer as much as 796 hectares paddy fields. The results of the Suwandi (2005), showed that the use of input from fertilizer by 10%, assuming other variables constant (*ceteris paribus*) can increase production by 1.25%. The results of the Nurshanti (2009) shows the mustard plant requires 10 tonnes per ha fertilizer. According Dahono et al. (2011), that the economic benefits arising from the use of a combination of NPK fertilizer and fertilizer from cattle waste, higher than that of NPK fertilizer without fertilizer from cattle waste. This shows the fertilizer from cattle waste can be considered in the cultivation of plants.

Compost an organic fertilizer derived from crop residues and cattle waste that has undergone a process of decomposition or weathering. Cattle belonging to the group members Mototavia as many as 24 tails, with the maintenance system grounded. Cattle waste can be directly collected by members of the group. Cattle waste amount of 10 kg per day can produce 3 kg of compost. Thus, members of the group can produce compost as much as 80 kg per day. Cattle waste compost have been made by members of the group and is sold at a price of Rp 1500 per kg. Mean, group members earn revenues from compost Rp 120,000 per day or Rp 43.8 million / year.

The compost is used as fertilizer for crops, which can improve soil texture, increases the cation exchange capacity, improve the ability to retain water, increasing the biological activity of the soil, improve soil pH, and others. According Riyanto et al (2012), the economic value of waste feces and urine increases as well as a new source of revenue so that the welfare of farmers has also increased. Applications of organic matter according to (Riley et al., 2008), in addition to improving soil structure, can increase the water holding capacity, also increase soil biological life (Dinesh et al., 2010). The use of compost according to Haryono (2013), a choice in favor of an increase in the productivity of upland rice on dry land.

Conclusions and Suggestions

Based on the research that has been done can be concluded that cattle waste which was created as compost, beneficial for the improvement of soil fertility, and farmers can minimize expenditure for the purchase of inorganic fertilizers. Suggestions, need further research to demonstration plots crops by using compost from cattle waste.

References

- Dahono., M. Ghulamahdi., S.A. AzizandAdiwirman, 2011. Combination of NPK fertilizers and fertilizers from cattle waste in enhancing the growth and production of plants *Centella asiatica*. *Journal Littri*, 17 (2), Juni 2011. pp 51-59.
- Dinesh, R., V. Srinivasan., S. Hamza and A. Manjusha, 2010. Short-term incorporation of organic manures and biofertilizers influences biochemical and microbial characteristics of soils under an annual crop turmeric (*Curcuma longa* L.). *Bioresource Technol.* 101. pp 4697-4702.
- Elly, F.H., M.A.V. Manese., and F.S.G. Oley, 2010., IBM Cattle Farmers Group Saleo Village, District Bolangitang East, North Mongondow Bolaang. Report of Activities of Science and Technology for People (IBM). Funded by the Directorate General of Higher Education, Ministry of National Education In accordance with the letter of the Implementation Agreement Grant Program Community Service No: 152/SP2H/PPM/DP2M/ VIII/2010, August 24, 2010. pp 34
- Elly, F.H., M.A.V. Manese and T.F.D. Lumy, 2011. IBM of Livestock Farmers Tonsewer Pelita Village, District Tompasso, Minahas district. Report of Activities of Science and Technology for People (IBM). Financed from the Fund Budget Implementation List (DIPA) No. 9748 / 023-04.2.01 / 27/2011, dated December 20, 2011, Unit UNSRAT, Ministry of National Education. pp 36
- Elly, F.H., A.H.S. Salendu and M.A.V. Manese, 2012. IBM of Cattle Farmers Group Ongkaw village. Report of Activities of Science and Technology for People (IBM). Financed from DIPA No: 012 / SP2H / KPM / DIT. Litabmas / III / 2012, dated March 6, 2012 Unit University of Sam Ratulangi Ministry of Education and Culture. pp 34
- Haryono, 2013. Policy Strategy Ministry of Agriculture in the Land Optimization of Suboptimal support of National Food Security. Proceedings of the National Seminar on Land Suboptimal: "Intensification of Suboptimal Land Management in the Context of National Food Support Independence. Palembang Sept 20 to 21 2013.
- Kuruseng, M.A and Alianti, 2011. Counseling evaluation and analysis of fine compost farm manure on the growth and production of mustard. *Journal Agrisistem*, Juni 2011. Vol. 7 No. 1. pp 60-65
- Nurshanti, D.F., 2009. Pengaruh pemberian pupuk organik terhadap pertumbuhan dan hasil tanaman sawi caisin (*Brassica juncea* L.). *Agronobis*, Vol. 1, No. 1, Maret 2009. Pp 89-98.
- Riyanto, J., Lutujo and Sunarto, 2012. Production of Organic Fertilizer Solids and Liquids as New Revenue Source For Cattle Farmers. Proceedings of the National Seminar on Sustainable Livestock. Innovation agribusiness Animal Husbandry for Food Security. Faculty of Animal Husbandry, University of Padjadjaran, Bandung. pp 654
- Salendu, A.H.S and F.H. Elly, 2013. Agroecosystem of Coconut-Cattle and Carrying Capacity Analysis in Lolayan Subdistrict of Bolaang Mongondow Regency. *European Journal of Social Sciences* Vol. 40(4) November 2013, pp 549-555.
- Suwandi, 2005. Sustainability integrated farming, pattern Beef and Rice Integrated, In Sragen: Approach RAP-CLS. Doctoral dissertation. Bogor Agricultural University Graduate Program, Bogor. pp 134

The study of nutritive value of plant for goat in Pattani Province of Thailand

T. Kraiprom* & N. Kueasiti

Department of Technology and Industried, Faculty of Science and Technology, Prince of Songkla University, 94000, Thailand

Abstract

The purpose of this research was to study the nutritive value of plant that most famer used for raising goat in Pattani Province. The sample of 379 farmers was randomly selected from the population of 7,157 farmers in twelve districts of Pattani province. The research tool was a structural questionnaire and collected by interview. The statistic employ were frequency, distribution and percentage. The results revealed that most of the interviewed farmers were male, and 36-45 years of age. They were graduated in Primary education. The farmer raised their goats in barn, free ranch and tether in nature pasture. They also used leaf plant as 11 species for goat fed such as Leucaena (*Leucaena leucocephala*), Siamese rough bush leaf (*Streblus asper* Lour.), Jackfruit leaf (*Artocarpus heterophyllus* Lam), Coconut leaf (*Cocos nucifera* L. var. *nucifera*), Guava leaf (*Psidium guajava* L.), Napier Pak Chong 1 grass (*Pennisetum purpureum* x *Pennisetum americanum*), Manila tamarind (*Pithecellobium dulce* (Roxb.) Benth.), Mango leaf (*Mangifera indica* Linn.), Ubon Paspalum (*Paspalu matratum* cv.Ubon), Ruzi grass (*Brachiaria ruziziensis*) and Para grass (*Brachiaria mutica*). The range of dry matter, crude protein, neutral detergent fiber, acid detergent fiber and ash were 17.67- 45.74, 5.45-25.84, 20.38-55.85, 13.11-35.20 and 5.70-20.38% respectively.

Keywords: nutritive value, plant, goat

**Corresponding author: thaintip@gmail.com*

Introduction

Goats have been raised in Thailand for their meat and milk. The number of goat population in 2013 was 440,277 heads (420,264 and 20,013 heads for meat and milk goats, respectively). The population of goat in southern of Thailand was 34,403 heads (47.8 %). Thus, the second range of Thailand that farmer most raised goat (Department of Livestock Development, 2013). The local demand of goat meat and milk products was increasing in south of Thailand especially Thai-Islamic. Thai farmers utilized native pasture or brush for raising their goats (Davendra, 1980). Ruminants in the tropics are fed on low quality roughages, especially agricultural crop-residues and by-products from agro-industry (Wanapat, 1999). Thai farmers owned on average area for such farming is only 0.40-4.80 ha per farm. This amount of area cannot support sufficient feed for ruminant (Tudsri & Swasdiphanich, 1993). Thus, this study was aimed to survey the type and nutritive value of plant that most famer used for raising goat in Pattani Province.

Material and methods

A total of 379 samples were collected from 7,157 farms in twelve districts of Pattani Province was randomly selected by Yamane method (Yamane, 1973). The research tool was a structural questionnaire and collected by interview. The statistic employ were frequency, distribution and percentage. In addition, collecting and sampling of forage that farmer most raised goat. The 3

samples of each forages were evaluated the dry matter (DM), crude protein and ash (AOAC, 1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were evaluated using the methods of Goering & Van Soest (1970).

Results and discussion

The most of farmers raised goat in Pattani Province were male, Muslim and 36-45 years of age. They were graduated in Primary school education. The farmer raised their goats 1-5 heads per family (36.62 %). They raised goats in barn (52.11%), free ranch (38.03%) and tether in nature pasture (9.86%). The most of breed is native goat (35.21%). Department of Livestock Development (2013) reported that the majority of Thai farmers of goats were raised by smallholders in approximately 36,753 households. In Thailand, goat farms belong to small holders who raise livestock in pararubber plantations (Thongchumroon & Anothaisinthawee, 1996). They also used leaf plant as 11 species for goat fed such as Leucaena (*Leucaena leucocephala*), Siamese rough bush leaf (*Streblus asper* Lour.), Jackfruit leaf (*Artocarpus heterophyllus*), Coconut leaf (*Cocos nucifera*), Guava leaf (*Psidium guajava* L.), Napier Pak Chong 1 grass (*Pennisetum purpureum* x *Pennisetum americanum*), Manila tamarind (*Pithecellobium dulce*), Mango leaf (*Mangifera indica* L), Ubon paspalum (*Paspalum atratum* cv. Ubon), Ruzi grass (*Brachiaria ruziziensis*) and Para grass (*Brachiaria mutica*). The range of dry matter, crude protein, neutral detergent fiber, acid detergent fiber and ash were 17.67 - 45.74, 5.45 - 25.84, 20.38 - 55.85, 13.11 - 35.20 and 5.70 - 20.38% respectively. Supaneeet al. (2013) was to study the information on goat farming status in the Northern Central Region of Thailand. Most commonly used feedstuffs for dairy goat farming were fresh pangola and paragrass, corn stover and husk either fresh or silage as roughage. In addition, other feedstuffs such as dry pangola, rice straw, pressed pineapple cake, and fresh Leucaenamay be fed or allowed free grazing on fresh forages depending on their availability.

Conclusion

It was concluded in this study that the most of farmers in Pattani province were male, graduated in Primary school education. The farmer raised their goat 1-5 heads per family. They also used leaf plant for raised goat such as Leucaena, Siamese rough bush leaf, Jackfruit leaf, Coconut leaf, Guava leaf, Manila tamarind, Mango leaf, Ubon paspalum, Napier Pak Chong 1 grass, Ruzi grass and Para grass.

Table 1. The chemical composition of forage source for most farmer raising goat.

Chemical composition	Dry Matter	Crude Protein	Neutral Detergent Fiber	Acid Detergent Fiber	Ash
Leucaena	24.00	25.84	35.96	13.11	7.47
Siamese rough bush leaf	35.69	12.44	30.83	22.91	20.38
Jackfruit leaf	39.18	10.59	20.38	18.81	11.75
Coconut leaf	45.74	10.23	27.03	27.03	5.74
Guava leaf	31.37	21.10	25.09	22.25	9.35
Manila tamarind	38.05	8.67	34.12	17.58	10.44
Mango leaf	38.06	7.39	33.75	22.89	8.88
Ubon paspalum	27.10	5.45	55.06	31.61	10.89
Napier Pak Chong 1 grass	17.67	7.26	55.85	33.60	12.00
Ruzi grass	21.39	6.97	42.53	42.53	11.76
Para grass	31.95	8.80	54.82	28.02	5.70

References

- AOAC, 1990. Official Method of Analysis of the Association of Official Chemists. 14th ed., Association of Official Agricultural Chemists, Washington, D.C. 1141p.
- Department of Livestock Development, 2013. Goat farming statistic in Thailand http://www.dld.go.th/ict/stat_web/yearly/yearly52/. Accessed on January 31, 2015.
- Davendra C., 1980. Potential of sheep and goats in less developed countries. *J. Anim. Sci.* 51:461-534.
- Goering, H. K. and P. J. Van Soest, 1970. Forage Fiber Analyses (apparatus, reagents, Procedures and some applications). *Agric. Handbook No. 379*. Washington, D.C. ARS, USDA.
- Thongchumroon, T. and S. Anothaisinthawee, 1996. Milk Production in Anglo-Nubian Crossbred Goats at Yala Livestock Breeding and Research Center. Annual Report on Animal Production Research. Department of Livestock Development. Ministry of Agriculture and Co-operation. p. 288-297.
- Tudsri, S. and S. Sawasdipanit, 1993. Managerial approach to pasture production in Thailand. 3rd Proc. Forage RWG of S.E. Asia. Khonkaen, Thailand. pp. 111-125.
- Wanapat, M., 1999. Feeding of ruminants in the tropics based on local feed resources. KhonKaen Publishing Company Ltd., KhonKaen, Thailand. pp. 236.
- Yamane, T., 1973. *Statistics: An Introductory Analysis*. (3rd ed.). New York: Harper and Row Publication.

Production performance of laying hen housing on litter system with different temperature

Ulupi, N^{*}, R. Afnan & T. Setiawati

Department of Animal Production and Technology, Faculty of Animal Science, Bogor Agricultural University, Bogor, 16680, Indonesia

Abstract

High ambient temperature in the cage system during rearing time of laying hens are causing high level of stress and low productivity. So the purpose of this study was to evaluate the response of laying hens with different rearing temperature in the litter system of housing toward the production performance and egg quality. A number of 36 laying hens with 30 weeks old were used. They were placed at 2 pens in small closed house (18°C and 30°C). This study used completely randomized design. Data of production performance was analyzed by t-test, whereas data of egg quality was analyzed descriptively. Performance of production included henday production, egg weight, egg mass and feed conversion ratio (FCR) of laying hens that reared in 18°C and 30°C were not significant different. The 18°C of rearing temperature produced income over feed cost (IOFC) of Rp 376/chicken/day, and it was only Rp 5 higher than 30°C. All of egg had the same value of Haugh unit (HU), thick and egg shell percentage and there was no broken or cracked egg. At 18°C rearing temperature, there was no dirty egg, but at 30°C there were 16.67% of dirty eggs. It can be concluded that the negative impact of high environmental temperature on the rearing of laying hen toward the production performance and eggs quality (except of dirty eggs) can be overcome by using the litter system of housing.

Keywords : egg quality, income over feed cost, litter system, production performance

**Corresponding author: niken.ulupi@gmail.com*

Introduction

Badan Pusat Statistik (2014), stated that the needs of people of Indonesia to eggs in 2013 was 1,159,549 tons, and it increased by 5.53% from the previous year. To meet this need, laying chicken population in 2013 was amounted to 147.2 million chickens, and it increased by 6.17% from 2012. Therefore the commercial laying hens were one kind of the poultry that very potential in Indonesia.

Environmental factors which have great impact on the productivity of the chicken is the temperature of rearing. The comfortable temperature (thermoneutral zone) for chicken is 20-24°C (Bell and Weaver, 2002). In this temperature range, chicken is not much to produce body heat, so the use of energy becomes more efficient. The temperature change will be responded quickly by chickens.

The environmental temperature in Indonesia, especially during the daytime (30-34°C), is above the range of comfortable temperatures for chicken. This is a major constraint in rearing of laying hens. Moreover, the majority of laying hens in Indonesia were reared in cage system. These fact make the laying hens more stress. At high temperatures, the chicken release the body heat through panting. Respiratory rate of chicken can increase by up to 200 times/min (Cunningham and Klein, 2007). The impact of it is a decrease in body resistance, production and

quality of eggs produced. Even at extreme temperature (> 34°C), can lead to death until 31.7% (Mashaly *et al.*, 2004).

Efforts that could be made to suppress the negative effects of high temperature on productivity of laying hens, among others, is the use housing of litter system with control the rearing temperature. Therefore, the purpose of this study was to find out the response of laying hens toward the difference of rearing temperature in the litter system on production performance and quality of eggs produced.

Materials and methods

Animal Experiments and Rearing

The study was conducted in the Poultry Laboratory, Departement of IPTP, Faculty of Animal Science, IPB. 36 commercial laying hens from Lohmann strains, 30 weeks old were used. The small closed house that be equipped temperature controller was used in this study. It consists of 2 pens, each measuring 2 x 2 m².

Each of pen was filled with 18 chickens. Temperature in the first pen was set at 18°C, and 30°C in the second pen. The feed and water were placed in the pen. Every pen was equipped with light bulb (75 Watt). The feed was commercial feed that contain 14-17% of crude protein, and 2850 kcal/kg of metabolizable energy. Feed was given 120 g/chicken/day, but the water was given *ad libitum*.

Rearing was carried out for 6 weeks. Every day was done recording of egg production, then they were weighed. Feed were weighed every week for calculate FCR. Every weekend, all of the eggs produced were analyzed their quality (HU, shell thickness, shell weight, shell integrity and level of dirtiness eggshell).

Data analysis

Data were analyzed with t-test using completely randomized design. The temperature of rearing (18°C and 30°C) were as treatment and observation data were as response. Statistical model was used $Y_{ij} = \mu + P_i + \varepsilon_{ij}$ (Mattjik and Sumertajaya, 2002). Data on egg quality were analyzed descriptively.

Results and discussion

Performance of Production

Production performance of laying hens that reared in housing of litter system with different temperature were presented in Table 1.

Table 1. Henday production, egg weight, egg mass and feed conversion ratio of laying hens for 6 weeks of rearing at different temperature

Performace of production	18°C	30°C
Henday production (%)	89.20 ± 10.50	86.10 ± 14.00
Egg weight (g/egg)	61.40 ± 3.98	63.38 ± 3.14
Egg mass (kg/6 weeks)	41.43 ± 0.75	41.25 ± 0.74
Feed conversion ratio	2.20 ± 0.16	2.21 ± 0.15
<i>Income over feed cost</i> (Rp/chicken/day) ^{*)}	376.00	371.00

^{*)} Not analyzed statistically

Production performances (henday production, egg weight, egg mass, and FCR) of laying hens that were reared at temperature of 18°C and 30°C were not statistically different. This is because in this study using the litter system, so the heat stress due to high temperature (30°C) still be overcome. This will be very different if the chickens were reared in individual cages. The chickens will suffer a double stress (from the cage system and high temperature of rearing). This condition significant reduced the performance of laying hens (Yousev, 1985).

These results were consistent with the IOFC value that obtained. Chickens that were reared at 18°C in the litter system, generating value of IOFC Rp 376/chicken/day, and only Rp 5/chicken/day higher than the chickens were reared at high temperature (30°C).

Egg Quality

The quality of eggs produced by chickens that were reared in housing of litter system with different temperature were presented in Table 2.

Table 2. Quality of egg that produced during the 6 weeks at different rearing temperature

Egg quality	18°C	30°C
Haugh Unit	74.20 ± 6.62	73.90 ± 8.30
Thick of eggshell (mm)	0.42 ± 0.04	0.42 ± 0.04
Weight of eggshell (%)	11.25 ± 0.65	11.97 ± 0.80
Shell integrity (%)	100.00 ± 0.00	100.00 ± 0.00
Level of dirtiness eggshell (%)	0.00 ± 0.00	16.67 ± 0.40

Haugh Unit value is reflecting the level of egg white thickness. The higher of the rearing temperature caused the egg whites produced more dilute, and the quality is declining. The chickens that were reared at 18°C, generate eggs with the value of HU slightly higher than at 30°C, but based on the USDA (1964), the quality of them is the same, AA (HU ≥ 72).

All of egg that were produced (at 18°C and 30°C), have ideal eggshell thickness (≥ 0.33 mm). The percentage of eggshell weight that produced were also in the normal range, namely 11% (Stadelman dan Cotterill, 1995).

Although all chickens were reared in litter system, but the egg of cracked or broken was not found. At 18°C was also not found the dirty eggs, but at 30°C was found as much as 16.67%. This is due to the rearing at high temperature (30°C), the metabolic interference was occurs, which in turn produces the excreta that more dilute (Daghir, 2008), and cause the percentage of the dirty eggs increased.

Conclusion

Rearing of laying hens in litter system with different temperatures (18°C and 30°C) resulted in the production performances, value of income over feed cost, and eggs quality were almost the same. At 18°C temperature rearing, was not found the dirty eggs but there was as much as 16.67% at 30°C.

References

- Badan Pusat Statistik. 2014. Statistik Indonesia 2004. Jakarta. Indonesia.
- Bell, D.D., W.D. Weaver. 2002. *Commercial Chicken Meat and Egg Production*. 5th Ed. New York. USA. Springer Science and Business Media Inc.
- Cunningham, J.G., B.G. Klein. 2007. *Textbook of Veterinary Physiology*. 4th Ed. St. Louis Missouri. WB Saunders Company.
- Daghir, N.J. 2008. *Poultry Production in Hot Climates*. 2th Ed. CABI Publishing. London, UK.
- Mashaly, M.M., G.L. Hendricks, M.A. Kalama, A.E. Gehad, A.O. Abbas, P.H. Patterson. 2004. Effect of heat stress on production parameters and immune responses of commercial laying hens. *J Poult Sci*. 83: 889-894.
- Mattjik, A.A., M. Sumertajaya. 2013. *Rancangan Percobaan dengan Aplikasi SAS dan Minitab*. Jilid 1. IPB Press. Bogor.
- Stadelmen, W.J., O.J. Cotterill. 1995. *Egg Science and Technology*. 4th Ed. New York. Food Products Press.
- USDA. 1964. Egg Grading Manual. Washington DC (US) : Federal Crop Insurance Corporation.
- Yousef, M.K. 1985. *Stress Physiology in Livestock*. Vol 3 Poultry. Florida, USA. CRC Press, Inc.

Use of agricultural by-product in pig ration to reduce feed cost in Manokwari Regency, West Papua Province, Indonesia

Widayati T.W., M.N. Lekitoo, D.J.Rahardjo, S.Y. Randa, S. Lumatauw, S.Hartini & B.Santoso

Department of Animal Science, University of Papua, Manokwari, Indonesia

Abstract

The paradigm of modern animal husbandry is based on zero waste concept. The use of agricultural and food industry by-products into valuable materials is an important issue that needs to be done. Pigs are the favorite animals for the Papuan because they are valuable in social, cultural and economical aspects. However, pig farm in Papua is constrained by providing concentrate ration because there is competition between pig ration and human food. The aim of this study was to know the potential of agricultural and food industry by-products as constituents of pig ration; and its possibility to reduce feed cost. This study was conducted at Manokwari regency, West Papua Province, Indonesia. The agricultural and food industry by-products used as pig ration constituents were collected from 2 traditional market, 5 restaurant and 15 small-scale food industries. The ingredients of ration comprised fish waste, soybean curd, taro skin, soybean skin, restaurant waste and commercial broiler ration. All materials used as ration were proximate analyzed to determine nutrition content. Feed cost was estimated using local market prices. Tabulation was used to analyse the data. Results of this study showed that crude protein and gross energy contents of agricultural and food industry by-products varied 4.26 to 31.21% and 3432.94 to 4950.57 kcal/kg, respectively. Use of agricultural and food industry by-products in pig ration reduced ration cost for phases of pre-starter, starter, grower, non lactation pig, gestation pig and lactation pig by 36.65, 38.58, 46.92, 55.00, 40.59 and 65.52%, respectively. It was concluded that agricultural and food industry by-products could be used as an alternative ration in order to reduce cost of ration in Manokwari Regency, West Papua.

Keywords: agricultural by-product, pig, concentrate ration, ration cost, protein, gross energy

Corresponding author: trieswd4@gmail.com

Introduction

Pigs are the favorite animals for the Papuan because they are valuable in social, cultural and economical aspects of their lives. The market demand of this commodity is quite high and become a primary saving for the households. Selling price of this animal is sufficiently high, weaning period price ranged from IDR 1.000.000 to 1.500.000 and that of at the age of cut (8-12 months) varied from IDR 3.000.000 to 5.000.000. So far, pig in Papua has not been intensively raised. The animal are mostly taken as the home consumption, so that aspects of feeding, reproduction and health are not cared properly (Randa, 1994). In general, farmers fed their animals as single feed such as tubers which is of low quality. The minimum amount of feed with low quality are factors that affect the growth of pig to be slow and easy to be infected by diseases (Iyai, 2008). Pig is a monogastric animal, favors concentrate ration and hence compete with human food. This condition caused a problem in feed availability during intensive pig raising. In addition, the commercial feed is not always available and its price is expensive. Based on BPS Papua Barat Province (2005) that West Papua has abundant forages and agricultural by-product and

potentially for the development of animal husbandry that is 42.442.750 tons produced from the area of 4.244.274 ha.

A good livestock development is adjusted to the availability of feed, socio-cultural conditions and local climate. In fact, the potency market of pig is high, but on the other hand there is still problems in the continuous feed availability, quality and economic, thus it is necessary to use ingredients from agriculture and food industry by-products as an alternative of pig ration. Based on above reasons, a study was conducted to evaluate potency of agriculture and food industry by products in Manokwari Regency as nutrient source to pig and its ability to substitute commercial ration in order to reduce ration cost.

Materials and Methods

This study was conducted at Manokwari regency, West Papua Province, Indonesia. The agricultural and food industry by-products used as pig ration constituents were collected from 2 traditional markets, 5 restaurants and 15 food industries. Ingredients used as pig ration were fish waste, soybean curd, taro skin, soybean skin, vegetables waste, waste of restaurant and broiler commercial ration. All ingredients used as ration were proximate analyzed to determine nutritional content. Ration cost was estimated by local market prices. Tabulation was used to analyze the data.

Results and Discussion

Proximate analysis of agricultural and food industry-by products as pig ration constituent presented in Table 1.

Table 1. The Potency and nutrients content of ingredients in pig ration.

No.	Ingredients	Potency (kg/day)	Nutrients Content			
			DM (%)	CP (%)*	GE (kcal/kg)*	ME (kcal/kg)*
1.	Fish waste	1000.00	29.41	31.21	3432.94	2709
2.	Soybean curd	2400.00	14.31	23.85	4950.57	3906
3.	Soybean skin	55.50	15.96	15.1	4022.23	3174
4.	Taro skin	11.40	26.45	4.26	3648.96	2879
5.	Vegetables waste	546.00	9.84	15.8	3683.99	2907
6.	Waste of restaurant	2056.56	35.84	13.72	4202	3315
7.	Commercial broiler ration (CP 11)		87	19.5	-	3100

*Dry matter basis

Two kind of ingredients such as fish waste and soybean curd used in this study were included as protein sources. Skin taro had the lowest CP content (4.6%), whereas the highest CP content obtained in fish waste (31.21%). The agricultural and food industry by-product are abundantly available in Manokwari Regency, however those by-products have not been used efficiently. Verkan (2011) stated that the lost of economic value of food such as vegetables, fish, legumes which caused by inefficiency in retail and consumer in America as much as US\$ 197.68 billion per year. Moreover, Kariasa&Suryana (2012) revealed that food availability could be increased by preventing food wastes.

Table 2 shows that the use of agricultural and food industry by-product as pig ration at different stage of production could meet nutrients requirement of pig and provides economical benefit by reducing the cost of the ration.

Table2. Formulation of pig ration using agriculture and food industry by-products at different stages of production.

No	Items	Stages of Production					
		Prestarter	Starter	Grower	Boar and Non Lactation	Gestation	Lactation
1	CP requirement (%)	23.7	20.9	18	13	12.9	16.3
2	ME requirement (Kcal/kg)	3265	3265	3265	3265	3265	3265
3	Dry matter requirement (kg)	0.5	1	1.86	2	1.96	4.31
4	Ration formulation (kg fresh weight)						
	a Soybean curd	1.07	1.95	2.45	1.43	1.79	0.86
	b Soybean skin			0.64	1.82	2.73	0.29
	c Taro skin		0.13	0.37	1.38	1.64	2.29
	d Fish waste	0.19	0.14	0.28			0.13
	e Vegetables waste		1.22	2.69	2.66	3.35	6.26
	f Restaurant waste		0.18	1.29	1.96	0.93	2.59
	g Broiler ration (CP 551)	0.28	0.42	0.38			0.56
5	Total ration as fed (kg)	1.53	4.03	8.09	9.25	10.45	12.98
6	Cost of commercial ration (IDR)	6000	12000	22260	24000	23520	51720
7	Ration cost using agricultural by-product (IDR)	3801.18	7370.1	11816.49	10798.29	13972.62	17831.94
8	Reduction of ration cost (%)	36.65	38.58	46.92	55.01	40.59	65.52

The lowest economic profit was obtained in the use of agricultural and industry by-product as pig ration at prestarter phase where the ration cost was reduced by 36%. Meanwhile, the highest profit was obtained at lactation phase where ration cost was reduced by 65.52% as compared to the use of commercial ration.

Conclusion

Agricultural and food industry by-products could be used as the alternative feeds which are not only favorable in nutrition quality but also help to reduce feed cost of pig in Manokwari Regency, West Papua.

References

- BPS Papua Barat, 2005. Papua Barat dalam Angka. BadanPusatStatistik, Provinsi Papua Barat.
- Iyai, D.A., 2008. Innovation of pig keeping system in pig farmers at Manokwari, West Papua Province, Indonesia. Thesis. Wageningen University, The Netherlands.
- Randa, S.Y., 1994. Performance of indigenous pigs of upland and lowland Irian Jaya. MSc Thesis. Department of Animal Science. Los Baños, The Philippines.
- Venkat, K., 2011. The Climate Change and Economic Impact of Food Waste in The United States. Kvenkat@celanmetrics.com.

Evaluation of goat milk quality to support dairy goat development

Cyrilla, L.¹, B.P. Purwanto², A. Atabany¹, D.A. Astuti³ & A. Sukmawati⁴

¹Department of Animal Production and Technology, Faculty of Animal Science, Bogor Agricultural University, Indonesia, ²Diploma Program Bogor Agricultural University, Indonesia, ³Department of Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agricultural University, Indonesia, ⁴Department of Management, Faculty of Economics and Management, Bogor Agricultural University, Indonesia

Abstract

The present study was to evaluate factors that affect goat milk quality, goat milk consumer' satisfaction, and technical responses associated with goat milk quality. Three farms having more than 100 dairy goats were purposively selected for the study. Thirty samples were determined using judgment sampling techniques to assess goat milk consumer' satisfaction. Data were analyzed using House of Quality matrix. The study revealed that dairy goat farms development in Bogor Regency is feasible with the support of dairy goat farms that have been there all along. The goat milk produced on farms studied already met the standards of goat milk quality, and could be classified into the premium category. Consumers are satisfied with the important attributes of goat' milk. Goat milk attribute that has been able to achieve customer' satisfaction targets are nutritional content, packaging size and goat milk color. However, to further ensure the fulfillment of the expectations of consumers about the quality of goat milk farmers still need to make improvements in the quality of the livestock' health conditions.

Keywords: dairy goat, milk quality, consumer' satisfaction, technical response, house of quality

Introduction

Milk is one of the protein sources for humans. People consider that milk especially goat milk provide health benefit for them, and it is greatly improves the diet of many rural families. Yet market demand for goat milk began to increase in the last few years, but they have not been met as the goat milk production is still limited. It is attributed to the low productivity of the existed dairy goats, and the lower population as well. One of the biggest problems facing someone getting into the dairy goat business is recognizing that it is not a quick easy business to get into and operate. Benefits of goat milk are more potent and better when compared to other milks on the market. Goat's milk benefits are superior to cow' milk, as it is richer than cow' milk in some important nutrients: vitamin A, niacin, choline, and inositol; it is poorer in folic acid (Park, 2009). Therefore, the selling price of fresh goat milk is still quite high, namely between IDR 25000-60000/l. Goat milk farmers are aware that maintaining the good quality of goat milk is important in order to maintain the consumer's confidence and the sustainability of their businesses. How farmers maintaining their products to meet standardized milk quality is still questionable. Therefore, it is deemed necessary to implement research program related to the topic needs to reponds the questions.

The objectives of the study were to evaluate the quality of the goat milk produced by dairy goat farms, as well as to evaluate customer requirements to goat milk. It is expected that the study will be beneficial to all parties associated with the development of dairy goat farms, especially in the city and district of Bogor.

Methods

The study was carried out in three dairy goat farms in the city and district of Bogor, West Java. Samples were collected purposively from farmers who own more than 100 goats. Sample of 30 consumers were determined using judgment sampling techniques to assess their satisfaction on goat milk. The data was collected using questionnaire and checklist forms which contains the characteristics of dairy goats, goat farms and its farmers characteristics, consumers attitudes toward goat milk, consumer' assessment for goat milk attributes (Ozawa et al., 2009).

The analytical method was based on House of Quality Matrix (Gaspersz, 2007). The House of Quality (HoQ) is the first matrix that a product development team uses to initiate a Quality Function Deployment process. This matrix is especially powerful because of the amount of information that can be documented and analyzed. This matrix consists of two main parts. The horizontal matrix containing information about customer needs, it is called the *customer table*, whereas the vertical matrix containing information how the organization will meet the challenges of providing products that delight the customer, it is called *technical table*.

Results and Discussion

Goat Milk Quality

Good quality goat milk is indicated by its color, smell, taste, cooking test, a test screening (cleanliness), specific gravity, fat content, total solid-fat and protein content (Park, 2009). Fat is one of the most important components in goat's milk relate to prices, nutrition and physical and sensory characteristics that affect the goat milk products. The higher amounts of shorter-chain fatty acids in goat milk fat significant difference from cow milk. Currently Indonesia has no specific standard for goat milk. The current reference standard applicable for goat milk quality in Indonesia is the Thai *Agricultural Standard for Raw Goat Milk* issued by the *National Bureau of Agricultural Commodity and Food Standards Ministry of Agriculture and Cooperatives of Thailand* (Table 1). Based on the goat milk quality analysis (Table 2) it was revealed that goat milk produced by dairy goat farms have met Indonesia fresh milk standard (SNI 01-3141-1998) and Thai Agricultural Standard of Raw Goat Milk (TAS, 2008) as well, particularly in the basic components, i.e. specific gravity, total solid, fat, protein and total solid non-fat. Moreover, further analysis of its fat and protein content, goat milk from those farms could be classified into the premium category.

Table 1. Fresh milk quality standard

No.	Parameter	Thai Agricultural Standard 6006*		
		Premium	Good	Standard
1	Specific gravity (at 27°C)	-		
2	Fat	>4	>3.5 to 4	3.25 to 3.5
3	Total solid non-fat	-	-	8.25
4	Protein	>3.7	>3.4 to 3.7	3.1 to 3.4

Note: *TAS (2008)

Table 2. Goat milk quality of the analyzed farms

Description	Farm A	Farm B	Farm C
Dairy goat' breed	Saanen	PE & Saanen	Etawah & PE
Population of lactation does (head)	90	22	44
Milk production (liter/day)	110 ± 30	22.2 ± 2.5	40.5 ± 7.5
Average milk production (liter/head/day)	1.2 ± 0.3	0.9 ± 0.25	0.85 ± 0.2
Quality parameter*			
Specific gravity	1.0295 ± 0.0003	1.0300 ± 0.0002	1.0300 ± 0.0002
Total solid (%)	15.88 ± 0.02	14.78 ± 0.01	14.40 ± 10.02
Fat (%)	6.6 ± 0.4	5.6 ± 0.5	5.3 ± 0.5
Protein (%)	3.70 ± 0.06	4.09 ± 0.03	3.70 ± 0.04
Total solid non-fat (%)	9.28 ± 0.04	9.18 ± 0.03	9.11 ± 0.04

Note: *Analyzed in Dairy Production Laboratory, Department of Animal Production and Technology Faculty of Animal Science Bogor Agricultural University (2014)

Customer Satisfaction on Goat Milk

Trend of research results on the level of consumer interests to goat milk show that attributes for nutrient content of goat milk is very important for consumers. Consumers need important information on the nutrient content and expiration dates of the goat milk, they must be printed clearly on its packages. It is very important to ensure the consumers that the products they consume safe and healthy. For consumers relatively expensive price of goat milk if compared to cow milk won't be problem for them as long as they are convinced that consuming goat milk is beneficial for them. Consumers expect that goat milk farmers guarantee the availability of goat milk at any time.

This study reveals that consumers satisfied with the attributes of nutritional content, flavor, aroma, color, packaging design, package size and price of goat milk produced by dairy goat farm A. Similar assessments were also given by consumers for goat milk of farm C. While for goat milk produced by farm B, the study revealed that consumers were satisfied with similar attribute own by farm A and B, except on its practicality consumption.

Goat milk prices at farm B was relatively lower than the other two farms. This was due to that farm B did not put labels in its packaging, whereas the other two farms have implemented this. Goat milk products of farm B are sold in 200 cc size of plastic packs. In addition, farm B does not only perform direct selling for individual consumers but also for retailers or distributors that resell the milk using their own packages. There are opportunities for farm B to sell its milk production at the same price of other two farms or even higher if the farm improve the design of its packages to suit its customer' expectations.

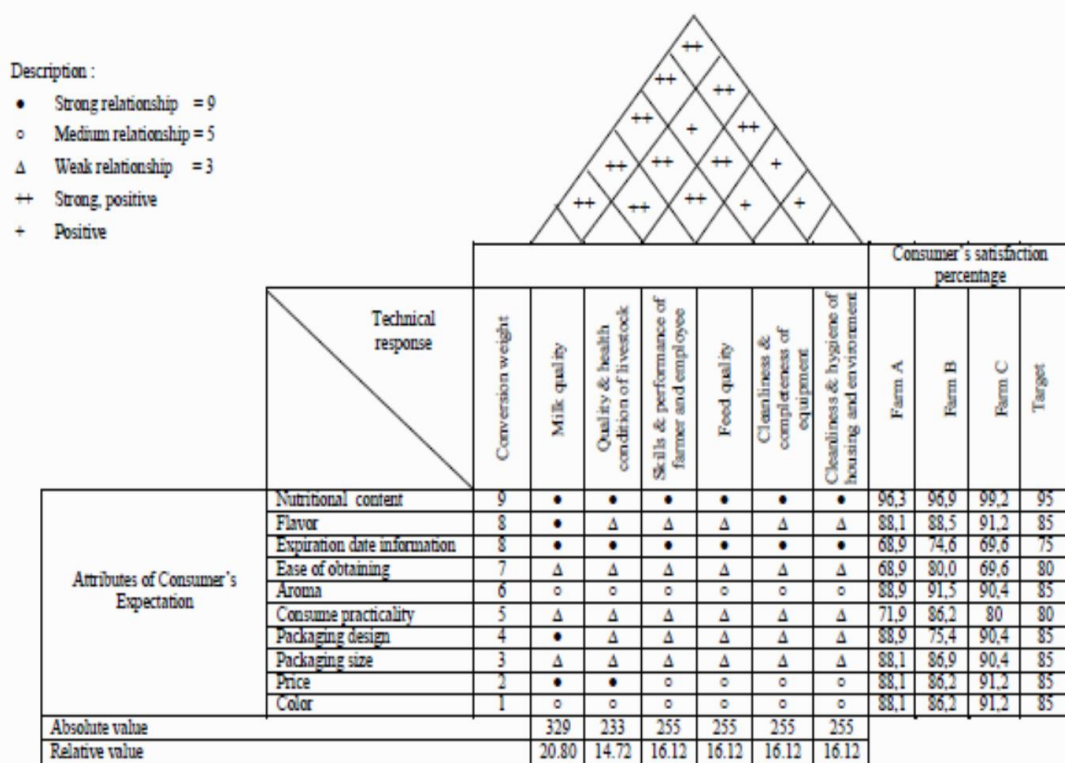


Figure 1. Goat milk house of quality

Goat Milk House of Quality

House of Quality (HoQ) is one of Quality Function Deployment (QFD) Matrix to explain the expectations of consumers and how to meet these expectations (Al-Marsumi, 2009, Gaspersz, 2007; Suyraningrat et al., 2010). The producer's ability to meet consumer expectations will be reflected in the ratio between the company target and level of customer satisfaction. Expectations reflect both past and current product evaluation and use experiences (Midauet al., 2010). Goat

Milk House of Quality (Figure 1) shows the highest customer satisfaction scores for goat milk. The highest scores were given to the nutrient content's attributes, it means that farmers had been able to achieve the targeted expectation for consumer satisfaction (the ratio is greater than one). Other attributes that reached the targets were the packaging size and color of goat milk. Efforts must be made by goat milk farmers to achieve other customer satisfaction attributes by improving their products. The relationship between technical responses to consumer expectations. Based on the data analyzed it was revealed that the technical response for the quality and health condition of goats was the first priority for improvement, as its relative value was rated 14.72 percent, the lowest. Based on their experiences dairy goat farmers are aware of and understand the importance of keeping their goat always in good health to ensure the quality of milk produced to sustain their businesses.

Conclusion

Dairy goat farms development in Bogor Regency is feasible with the support of dairy goat farms that have been there all along. The goat milk produced on farms studied already met the standards of goat milk quality, and could be classified into the premium category. Consumers are satisfied with the important attributes of goat' milk. However, to further ensure the fulfillment of the expectations of consumers about the quality of goat milk farmers still need to make improvements in the quality of the livestock' health conditions.

References

- Al-Marsumi, M. 2009. Total Quality Management in the top rank of the dairy industry in Jordan. *JJMIE* 3(1): 47- 58
- Gaspersz, V. 2007. *Lean Six Sigma for Manufacturing and Service Industry*. Gramedia Pustaka Utama, Jakarta.
- Hibma, J. 2015. Marketing "Value Added" Dairy Goat Products : Find a Niche and Fill It. http://www.dairygoatjournal.com/85-4/john_hibma/
- Midau, A., A.Kibon, M. S.Morrupa and C. Augustine. 2010. Acceptability and consumption of goat milk in Adamawa State, Nigeria: a case study of Mubi North and Mubi South local government areas. *J. Agric. Soc. Sci.*, 6: 11–13
- Ozawa, T., K. Mukuda, M. Fujitaand J. Nishitani. 2009. Goat milk acceptance and promotion methods in Japan: The questionnaire survey to middle class households. *J AnimSci*80 (2) :212-219. doi: 10.1111/j.1740-0929.2008.00611.x.
- Park, Y. W. 2009. *Bioactive Components in Milk and Dairy Products*. Ed Ke-1. Blackwell, Iowa.
- Suryaningrat, I.B., Djumarti, E. Rurianiand I. Kurniawati. 2010. Aplikasimetode quality function deployment (QFD) untukpeningkatankualitasmiejagung. *AGROTEK* 4 (1) : 8-17.
- [TAS] Thai Agricultural Standard 6006.Standard for Raw Goat Milk. 2008. National Bureau of Agricultural Commodity and Food Standards Ministry of Agriculture and Cooperatives Thailand.
- Ulya, M. 2012. Identifikasipersyaratanpelangganterhadapproduktahu. *AGROINTEK* 6 (2): 72-78

Insight into broiler development in East Java

Eko Nugroho, Priyo Sugeng Winarto, Rizki Prafitri, Anie Eka Kusumastuti & Bambang Ali Nugroho*

Department of Socio-Economic of Animal Husbandry, Faculty of Animal Husbandry, Brawijaya University, Jalan Veteran, Malang, East Java, 65145 Indonesia

Abstract

The demand of chicken meat has been predicted to increase in the future in line to the increase of human population, income per capita and awareness of nutrient sufficiency. However, with the population of 250 million, Indonesia is still classified as a low chicken meat consumer as compared to other countries. The province of East Java is one of the largest suppliers for the national chicken meat demand in Indonesia. This paper, therefore, tends to discuss broiler development in East Java including the production, consumption, constraints, and the alternative solution to the problems. The primary data were collected from broiler farmers who were interviewed using a questionnaire. Meanwhile secondary data were compiled from various references and related institutions. The results showed that broiler farming practices were formed in nucleus-client partnerships and self-financing business. Several regencies such as Blitar, Ponorogo, Jombang, Malang, Lumajang, Lamongan, Nganjuk, Pamekasan, Bojonegoro and Mojokerto were the largest broiler suppliers in East Java. Another finding showed that broiler meat production in East Java reached 168,306 tons in 2013 which is 1.33% increase for the last four years. Nevertheless, the percentage of meat consumption in East Java had average growth of 0.55% during 2010-2014. Farmers who raised broiler through nucleus-client partnership sold their products to the nucleus company such as poultry breeders, feed industries and poultry shops. Although the partnerships was able to minimize the business risks for small farmers, the partnership agreement however was still considered unfair because the price of feed, Day Old Chick (DOC) and selling prices were set unilaterally by the nucleus company. It, therefore, needs a policy which protects broiler partnership scheme based on fairness principle for both nucleus and clients, strengthens each other, and provides benefit to each other.

Keywords: poultry, production, consumption, East Java, nucleus company, partnership

**Corresponding author: eko_nug@ub.ac.id*

Introduction

One of the major developmental challenges facing most developing countries is their inability to adequately feed their ever-increasing population with the right proportion of calories and protein (Apantaku, 2006). Poultry are important providers of eggs and meat as well as being valued in religious and cultural life (Teklewold et al., 2006). The national development of poultry production showed a large increase in past few decades. Nevertheless, Indonesia with human population 250 million people is still considered a country with low consumption of chicken meat compared to nutritional requirement and consumption of other countries. Based on these conditions, broiler farmers have priority in the economic development especially small medium enterprises (SMEs) and cooperatives.

Broiler production has relatively bright prospects particularly in East Java province for several reasons, such as (a) 37 million people in East Java is a potential market and East Java is also the

important national supplier of poultry and poultry products, (b) market globalization (AEC, WTO, AFTA and APEC) become tremendous opportunity, (c) the development of upstream agroindustry needs raw material livestock product such as chicken meat processing industry, and (d) livestock product is perishable that needs further processing. The study therefore tends to review the development of broiler farm in East Java.

Material and method

The study was conducted in Banyuwangi regency and the parameters were based on indicators found by Chilonda & Otte (2006) such as population, production, consumption, and marketing. Primary data were collected from in-depth interview to several key persons guided with questionnaire. Secondary data obtained directly from relevant technical agencies such as Indonesia Statistical Agency, Department of Livestock and Animal Health and Department of Livestock Service. The obtained data were then analyzed descriptively.

Results and discussion

Broiler population in East Java

East Java province has been known to be one of the national broiler producers. However, the data from Department of Livestock and Animal Health (2013) showed the growth of broiler population in East Java decreased 2.13% during 2010- 2013 as compared to the growth of broiler population in West Java (8.13%) and the national growth (8.25%). Broiler farming practices could be found in almost all of East Java region although their population varied among regencies. Blitar, Jombang, Ponorogo, Malang, Lumajang, Lamongan, Nganjuk, Pamekasan, Bojonegoro, and Mojokerto districts were 10 broiler producers in East Java. Data issued by BPS East Java (2014) showed that Lamongan, Blitar, and Jombang districts were areas which had the largest broiler population.

Broiler meat production and consumption in East Java

Total production of broiler meat at 2013 in East Java were more than 168.306 tons, and Surabaya manage almost all of production and the rest were Malang, Blitar, Jombang, Lumajang, Ponorogo, Nganjuk, Mojokerto, Bojonegoro and Gresik. The data from Department of Livestock and Animal Health (2013) showed that broiler production in East Java during 2010-2014 growth with lower average 1.33% compared to the average national broiler production 5.07%.

In relation to broiler meat consumption, data produced by the Department of Livestock and Animal Health (2013) shows the average growth in the broiler price in East Java increased by 0.55% during 2010-2014. Nevertheless, this growth was lower than average national broiler consumption (0.73%) in the same period. In the case of avian influenza outbreak at 2005, the population of broiler decreased to 94 million especially in five areas including East Java, Banten, Bali, West Java and South Sulawesi (Yusdja, 2007).

Distribution of broiler meat product in East Java

Broiler product in East Java are distributed by farmers in several ways. Firstly, big farmers sell the product to commission agent and then continued sell to big scale traders. Secondly, farmers sell the product to big scale traders directly and forwarded to bigger traders who supplied markets in certain areas. Farmers with large broiler population had capability to distribute the product to the certain market directly in certain province.

Problems of broiler farm in East Java

The basic government regulation through Undang-Undang Peternakan Tahun 1967 in principle was to develop livestock sector as rural business for employment and income resources. At preliminary stages of broiler industry development, rural business had growed rapidly until 1980. Rural business at that time

run individual broiler farming practices and they made their own feed and breeder. Even the growing scale of rural business at that time was relatively small (less than 2,000 birds), the characteristic of rural business was in fair competition which was developed later as industrial broiler in developed countries. Nevertheless, the development of national poultry sector was not always ideal as mentioned previously. Unlike layer business, broiler farming in the study area almost could be expressed entirely in the form of nucleus-client scheme. The nucleus company provided DOC, feed, medicine for the clients (small farmers) as loans that must be paid at harvest time. In addition, the client must sell the whole chickens to the nucleus companies. Partnership pattern in the early days of development found a partial cooperation which put clients sometimes in a disadvantaged position and it began to evolve naturally into better shape. The partnership pattern in Banyuwangi guaranteed a fixed income for plasma which ranged from IDR. 900 to IDR.1000 per bird. However, the total cost for this business wasn't small (at least 3.000 chickens). It means that not all rural residents could access this broiler partnership pattern. In addition, although the partnerships was able to minimize the business risks for small farmers, the partnership agreement however was still considered unfair because the price of feed, DOC and selling prices were set unilaterally by the nucleus company.

Alternative solution and business opportunity of broiler farm in East Java

The alternative solution would be a horizontal integrated system developed by broiler small and medium enterprises. Even if the farmers are not able to make their own decision, this partnership scheme is however helpful for the growth of rural economic because it offers business opportunity such as broiler housing-made service, farm labor, security service and other opportunity to earn income. Thus, it needs a policy which protects broiler partnership scheme based on fairness principle for both nucleus and clients, strengthen each other, and provide benefit each other.

References

- Apantaku, S. O. 2006. Analysis of participation of farmers in participatory poultry production research in Lagos State, Nigeria. *Livestock Research for Rural Development* 18 (7) 2006.
- BPS East Java. 2014. Jawa Timur dalam angka 2014. BPS Provinsi Jawa Timur. Surabaya. (in Bahasa Indonesia).
- Chilonda, P. and Otte, M. J. 2006. Indicators to monitor trends in livestock production at national, regional and international levels. *Livestock Research for Rural Development* 18 (8) 2006.
- Department of Livestock and Animal Health. 2013. Statistik peternakan dan kesehatan hewan 2013. Jakarta. (in Bahasa Indonesia).
- Teklewold, H., Dadi, L., Yami, A. and Dana, N. 2006. Determinants of adoption of poultry technology: a double-hurdle approach. *Livestock Research for Rural Development* 18 (3) 2006.
- Yusdja, Y. 2007. Pembangunan peternakan: Pencapaian dan Prospek. Dalam *Prosiding Kinerja dan prospek pembangunan Pertanian Indonesia*. Editor: Kedi Suradisastra, Yusmichad Yusdja and Prajogo U. Hadi. Pusat Analisis Sosial Ekonomi dan Kebijakan Pertanian. Badan Penelitian dan Pengembangan Pertanian Departemen Pertanian. (in Bahasa Indonesia).

Association among fat, protein, lactose and total solid of milk produced by farmers in central part of Thailand

Lasa-ard, T., T. Suwanasopee* & S. Koonawootrittriron

Department of Animal Science, Faculty of Agriculture, Kasetsart University, Bangkok 10900, Thailand

Abstract

Composition of milk produced by farmers is used for price determination in dairy industry, and it is also considered as an indicator for management quality and health status of the dairy cattle. The objective of this study was to investigate the association among fat (F%), protein (P%), lactose (L%), and total solid (TS %) of milk produced by the farmers. Milk composition that evaluated from 40,511 milk samples was used in the study. These samples were taken from 432 dairy farms in Central part of Thailand during January 2012 and December 2014. Size of farms was classified into small (1 to 9 milking cows), medium (10 to 19 milking cows), and large size (20 to 90 milking cows). The mixed model considered milking year-season and farm size as fixed effects, and farm and residual as random effects. Least squares means were estimated for each factor and then they were compared using t-test. Association among F, P, L and TS were considered using correlation coefficients. Year-season and farm size had effect on all traits ($P < 0.001$). The differences among individual farms associated with the variation of milk composition 30% for F, 25% for P, 34% for L and 27% for TS. A unit of TS composed 31% of F, 25% of P, 39% of L and 6% of the others. The correlation estimate was 0.88 for TS and F, 0.59 for TS and P, 0.31 for F and P, and 0.24 for TS and L ($P < 0.001$) the others were close to zero. These results imply possibility to improve TS and others related milk component of the farmers for their sustainability.

Keywords: dairy, raw milk, tropics

* Corresponding author: agrtts@ku.ac.th

Introduction

Milk composition is widely used as a key factor to determine purchasing price of raw milk which produced by dairy farmers. The current standard of milk compositions in Thailand were 3.2% for fat, 3.0% for protein, 4.5% for lactose and 12.3% for total solid (The Agricultural Commodity and Food Standard, 2008). Induction and deduction of purchasing price of raw milk were issued by individual dairy cooperatives or private milk collecting centers. The farmers who could control the quality of raw milk to be over the standard, they would get more incomes. Under tropical conditions, not only genetic but also non-genetic factors (e.g., farm size, farm location, quality of farm management, climates, and health status of dairy cows) were important for milk composition at farm level (Rhone et al., 2008; Mirzah et al., 2010; Yeamkong et al., 2010a). Small size farms in Central Thailand could produce higher fat percentage in raw milk than medium and large farms (Rhone et al., 2008). Large size farms had higher potential for milk quality and milk revenues than small and medium farms (Yeamkong et al., 2010a). Fluctuations of farm management across year also affect milk composition. However, the consequence of changes in any solid matters of raw milk under farm management in tropical country is unclear. Therefore, the objective of this study was to

investigate the association among fat, protein, lactose, and total solid of raw milk produced by farmers in Central part of Thailand. The understanding of relationship among milk composition may lead to suitable cares for dairy cows and the sustainability of dairy farming in this region.

Materials and Methods

The 40,511 records of bulk tank milk composition including percentage of fat (F; %), protein (P; %), lactose (L; %) and total solid (TS; %) was collected from 432 dairy farms located in Central part of Thailand. Milk composition records of each farm were determined three times a month during January 2012 and December 2014. The proportion of F, P, L, and others in TS was preliminary calculated and described as shown in Table 1. The determination of factors affecting milk composition was done by considering milking year-season (YS) and farm size (FS) as fixed factors and farm and residual as random factors. Season was classified as winter (November to February), summer (March to June) and rainy season (July to October). Farm sizes were classified by the number of milking cow presented within the farm as small (1 to 9 milking cows), medium (10 to 19 milking cows), and large size (20 to 90 milking cows). Least squares means (LSM) were estimated for the significant factors and then they were compared using t-test ($P=0.05$). Association among milk compositions were estimated using Pearson correlation.

Table 1. Descriptive statistics for milk composition traits.

Traits	Records	Mean	Standard deviation	Minimum	Maximum
Fat (%)	39,627	3.77	0.45	2.54	5.01
Protein (%)	39,640	3.03	0.19	2.33	3.54
Lactose (%)	39,257	4.69	0.14	4.19	5.05
Total solid (%)	39,624	12.17	0.59	10.03	13.75

Results and Discussion

Milking year-season had significant influence on F, P, L and TS ($P<0.001$). The highest components of F ($3.89 \pm 0.01\%$) and TS ($12.34 \pm 0.02\%$) in raw milk were found in rainy season, while the lowest of F ($3.64 \pm 0.01\%$) and TS ($11.95 \pm 0.02\%$) were presented during summer. The variation of these milk compositions might be due to abundant supply of green and fresh grasses in rainy season, while, high temperature during summer had influenced on insufficient of quality roughage and low feed intake of dairy cows (Rajcevic et al., 2003).

Variation of farm size affected all milk composition traits in this study ($p<0.001$; Table 2). Small farms produced raw milk with the lowest F, P and TS, while the highest values of these traits, except for L, were found in medium farms. The LSM for milk compositions of large farms in this studied were ranged between the values of small and medium farms. Moreover, LSM of the studied traits were in the range of previous studies (Yeamkong et al., 2010b; Mirzah et al., 2010). The high ability of farmers in medium farms found in this study was possible due to their more experience in dairy production and better understanding in farm management under specific condition.

The variance ratios of farm and total variance are shown in Table 2 revealed that variability among farms accounted for 25% to 34%. The different between farms had fairly affected the variation of milk composition and it could be varied by farmer themselves (education, experience and training) and their farm condition (Yeamkong et al., 2010a).

The average proportion of F, P, L and others in TS were 31%, 25%, 39% and 6%, respectively. This revealed that TS of raw milk produced by farmers in this study was mainly composed of L. The result was similar to the study of Mirzadeh et al. (2000) in dairy cows raised in Lordegan region of Iran. This high proportion of L in raw milk is caused by overfeeding of concentrate diets and this unbalance nutrient will consequently increase the synthesis of lactose (Chalupa & Sniffen, 2000).

Table 2. Least squares means and standard deviations for milk composition traits.

Traits	Farm size			p-value	Ratio of farm variance to total variance
	Small	Medium	Large		
Fat (%)	3.76 ± 0.48 ^b	3.79 ± 0.44 ^a	3.77 ± 0.41 ^b	<0.001	0.30
Protein (%)	3.02 ± 0.21 ^b	3.04 ± 0.19 ^a	3.03 ± 0.18 ^a	<0.001	0.25
Lactose (%)	4.69 ± 0.15 ^a	4.68 ± 0.15 ^b	4.69 ± 0.13 ^a	<0.001	0.34
Total solid (%)	12.16 ± 0.65 ^b	12.20 ± 0.59 ^a	12.18 ± 0.53 ^{ab}	<0.001	0.27

^{a, b} Least squares means within the same row with different superscripts differ (p < 0.01)

The correlations among milk compositions ranged from -0.02 to 0.88 (P < 0.001). The highest estimate were found between TS and F (r = 0.88). The other correlations among milk composition were 0.59 for TS and P, 0.31 for F and P and 0.24 for TS and L, while the others were close to zero. This result indicated that the significant change of F content in raw milk could positively improve TS. Consequently, dairy farmers would get higher purchasing price to sustain their business.

Conclusions

Milking year-season and farm size had significantly influence on F, P, L and TS. Variation among individual farm accounted for 25% to 34% of the total variance for each trait. The highest correlation among milk composition was found between TS and F.

Acknowledgements

The authors would like to thank the Center for Advanced Studies for Agriculture and Food, Institute for Advanced Studies, Kasetsart University under the Higher Education Research Promotion and National Research University Project of Thailand, for financial support. We also thank the Muaklek Dairy Cooperative Limited for providing the milk quality records.

References

- Chalupa, W. and C. J. Sniffen, 2000. Balancing ratio for milk components. *Asian-Aust. J. Anim. Sci.* 13(Suppl): 388-396.
- Mirzah, Kh., A. Masoudi, M. Chaji and M. Bojarpour, 2010. The composition of raw milk produced by some dairy farms in Lordegan region of Iran. *J. Anim. Vet. Adv.* 9(11): 1582-1583.
- Rajcevic, M., K. Potocnik and J. Levstek, 2003. Correlations between somatic cells count and milk composition with regard to season. *ACS.* 63(3): 221-226.
- Rhone, J. A., S. Koonawootrittriron and M. A. Elzo, 2008. Factors affecting milk yield, milk fat, bacterial score, and bulk tank aromatic cell count of dairy farms in the central region of Thailand. *Trop. Anim. Health Prod.* 40:147-153.
- Thai Agriculture Commodities and Food Standard, 2005. Raw milk, National Bureau of Agricultural commodities and Food Standards, Thailand. pp 9.
- Yeamkong, S., S. Koonawootrittriron, M. A. Elzo and T. Suwanasopee, 2010a. Milk quantity, quality and revenue in dairy farms supported by a private organization in Central Thailand. *Livest. Res. Dev.* 22(2).
- Yeamkong, S., S. Koonawootrittriron, M. A. Elzo and T. Suwanasopee, 2010b. Effect of experience, education, record keeping, labor and decision making on monthly milk yield and revenue of dairy farms supported by a Private Organization in Central Thailand. *Asian-Aust. J. Anim. Sci.* 23(devil): 814-824.

Conventional and deep - litter pig production system: income over feed cost of three breed cross fattening pigs

Phoowadon Prapruetdee

King Mongkut's Institute of Technology Ladkrabang, Prince of Chumphon campus, Chumkoh, Pathiu, 86160, Thailand

Abstract

A previous study to investigate the effects of conventional and deep - litter pig production systems on growth performance showed no significant difference between these 2 systems, although a conventional group received 100% commercial diet while a deep - litter pig group received only 50% commercial diet and 50% grasses/native vegetables. The objective of this study was to calculate income over feed cost of finishing pigs from the conventional and the deep - litter pig production systems. A total of 16 crossbred (Largewhite-Landrace x Duroc), mixed sex (8 gilts and 8 barrows) pigs at 15.75 kg live weight were randomly allocated into 2 treatments (8 pigs/treatment): a traditional concrete - based house (TCBH) received only commercial diet. The deep - litter house (DLH) treatment group received the commercial diet and grasses/ native vegetables (50/50), bedding materials with 10 parts of rice hulls and 1 part of soil, then topping by salts about 500 g and addition of a mixed culture as 30 cm height layer and repeated 2 times to completed 90 cm. height. Results indicated that the average feed cost was higher in the TCBH group due to 100% commercial diet consumption (54.08 versus 29.24 Baht/head/day) as compared to the DLH group, leading to a greater feed conversion ratio (2.11 versus 1.72); followed by slightly higher average weight gain (1 versus 0.96 kg/day) in the TCBH group. The TCBH group only received 4,990.20 Baht/head income from pig sold while the DLH group received 5,297 Baht/head income from pig and litter sold (a live weight pig sold price at farm was 60Baht/kg), so the income per day of the TCBH group was less (57.36 Baht/head/day) than DLH group (60.19 Baht/head/day). Income over feed cost (IOFC) in the DLH group was higher than TCBH group (30.95 versus 3.28 Baht/head/day) according to the existing feed cost and income data. Therefore, the deep - litter pig production system could be more beneficial for fattening three breed cross pigs when focusing on Income over feed cost.

Keywords: deep – litter pig production, income over feed cost, three breed cross pig

Introduction

Pig production involves very high feed cost, about 60 - 70 % of total production cost, therefore the sustainable various feed resources with more “natural low cost” have been practiced in tropical countries (Suryatni & Sutan, 2014). One of the low cost pig production systems is a deep – litter pig housing and has been saved up to 40 % of construction cost (Margeta et al., 2010). Prapruetdee (2013) reported no significant difference on some of growth performance between conventional and the deep - litter pig production system, even the deep - litter group were fed only half of conventional and replaced the rest with grasses/native vegetables and had inferred that the deep - litter pig production is a suitable way of pig production for small holder farms in Thailand.

The objective of this study was to evaluate the effect of conventional and deep - litter pig production system on income over feed cost (IOFC) of three breed cross fattening pigs.

Materials and Methods

A total of 16 crossbred mixed sex (8 barrows and 8 gilts) piglets were randomly placed into 2 treatments (8 pigs/treatment): a traditional concrete - based house (TCBH) received 100 % commercial diet. The deep - litter house (DLH) treatment group received the commercial diet and grasses or native vegetables (50/50), bedding materials with 10 parts of rice hulls and 1 part of soil, topping by salts about 500 g and addition of a mixed culture. Body weights were measured in every 21 days and feed intakes were observed daily throughout the study. At the end of the experiment (similar weight at 101.55 ± 1.65 kg) all pigs were sold by the live weight at farm - price. Income over feed cost (IOFC) using total average feed cost (Bath/head/day) of concentrate and roughage (in DLH), average finishing weight (kg/head), days of finishing, live weight pig sold price at farm (Bath/kg), litter sold (Bath/head) in the DLH group, average prices of concentrate and roughage during the experiment and income per day (Bath/head/day). IOFC of the TCBH group and the DLH group were calculated using the formula as described by Suryatni & Sutan (2014): [price of pigs sold (Bath)] + litter sold in DLH (Bath)] – [feed cost (concentrate and roughage in DLH), + (piglets price)].

Results and discussions

Results indicated that the average feed cost was higher in the TCBH group due to 100% commercial diet consumption as compared to the DLH group (54.08 versus 29.24 Baht/head/day), leading to a greater feed conversion ratio (2.11 versus 1.72); followed by slightly higher average weight gain (1 versus 0.96 kg/day) in the TCBH group.

Table 1. The effect of treatments on IOFC of three breed cross fattening pigs.

Treatment	Way of fattening	
	TCBH	DLH
Average feed cost (Bath/head/day).	54.08	29.24
Total income (Bath/head)	4,990.20	5,297
Income per day (Bath/head/day)	57.36	60.19
IOFC (Bath/head/day)	3.28	30.95

The TCBH group only received 4,990.20 Baht/head income from pig sold while the DLH group received 5,297 Baht/head income from pig and litter sold (a live weight pig sold price at farm was 60 Baht/kg). So, the income per day of the TCBH group was less (57.36 Baht/head/day) than DLH group (60.19 Baht/head/day). Income over feed cost (minus the average piglets cost at the beginning for 1,200 Baht/ head) in the DLH group was higher than TCBH group (30.95 versus 3.28 Baht/head/day).

Conclusion

Deep – litter pig production system could be beneficial for fattening three breed cross pigs when focusing on the income over feed cost. Obviously, for Thai small farm holders, achieved by its lower feed cost, by replacing half of concentrate with very cheap grasses or native vegetables. Moreover the deep – litter pig production system could make more income from selling decomposed bedding material after fattening pigs.

References

- Margeta, V., G. Kralik, D. Hanzek and P. Margeta, 2010. Deep – litter pig keeping (A review). *Acta Agraria Kaposvariensis*. 14(2): 209 – 213.
- Prapruetdee, P., 2013. Conventional and deep-litter pig production system: the effects on growth performance. Proceedings of the 4th international conference on sustainable animal agriculture for developing countries. 27 - 31 July, 2013, Lanzhou, China. 396 – 397.
- Suryanti, N.P.E and Y.F.G. Dillak, 2014. Ration efficiency and income over feed cost of native chickens fed fermented local feeds. Proceeding of the 16th AAAP Congress. 10-14 November, 2014. Yogyakarta, Indonesia. 2355 - 2358.

Application of PCR technique for detection *Staphylococcus aureus* cause of mastitis in dairy cows

Junlapho^{*1,2}, W. , T. Puangmalee² & J. Kanjanapruthipong¹

¹Department of Animal Sciences, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, 73140, Thailand,

²Center of Excellence on Agricultural Biotechnology: (AG-BIO/PERDO-CHE), Bangkok, 10900, Thailand

Abstract

Staphylococcus aureus causes one of the most common types of chronic mastitis and difficult to control by treatment alone. Spread of infection can occur through milkers' hands, washcloths and teat cup liners during milking. Successful control is gained only through prevention of new infections and culling of infected animals by reduce bacterial numbers in milking process is recommended. Polymerase Chain Reaction has made bacterial detection possible without the need for bacteria isolation, fast, highly sensitive and specific assay than conventional culture. Generate PCR primers derived from staphylococcal species coagulase gene were used to amplify a target region of *Coa1 Enterotoxin* product size 821bp (Forward-ACC ACA AGG TAC TGA ATC AAC G and Reverse-TGC TTT CGA TTG TTC GAT GC), annealing temperature at 58°C and *Coa2 non Enterotoxin* (Forward-TGC AGAT GGT ACT GCG ACA T and Reverse-CTT CCG ATT GTT CGA TGC TT) annealing temperature at 56°C and PCR product 158 bp. The samples were swabbed from 21 risk points during milking process. The result of bacterial detection from plate count on egg-yolk mannitol salt agar were found colony forming units of 7 samples were similar with result from amplify by PCR by *Coa2 non Enterotoxin*, showing the specificity and high sensitivity of the assay.

Keywords: polymerase chain reaction, *Staphylococcus aureus*, mastitis, dairy cows, coagulase gene

*Corresponding author: fagrwnn@ku.ac.th

Introduction

Mastitis is the most common infection that affects cows and still is the most important economic impact on the quantity and quality of raw milk. (Bhushan, 2000) The major cause of bovine mastitis is the infection of the udder by pathogenic bacteria. A wide variety of bacteria can be involved, but the most common mastitis pathogens are *S. aureus* (William et al., 2002). The treatment of mastitis was ineffective due to the emergence of drug resistance in bacteria and collection sample of bacterial pathogens were identification to help determine the effectiveness of antibiotics should also be used to treat a type of bacteria that cause mastitis, which is good for planning and controlling mastitis in the farm. (Phuektesat al., 2001). Identification of bacterial pathogens from mastitis of dairy cows is the detected with mannitol salt egg yolk agar takes longer to detect. Therefore, application method is fast and precision that is the PCR technique. The samples were collected from The critical control point is likely to mastitis including the milker's hands, the floor of stall, washcloths and teat cup liners during milking. Coagulase is an enzyme important in diagnosis due to the properties of the formation and separate out the different types of *S. aureus* from *Staphylococcus*. (Bryan, 1976) Therefore, coagulase gene sequencing was used to design primer, to be used to determine *S. aureus* in PCR reaction.

Materials and Methods

Sample collection and selective media test

The Samples were collected from the infected farm 7 critical control points with cotton swab technique from the critical control point is likely to mastitis including the milker's hands, the floor of stall, washcloths and teat cup liners during milking. Store the sample at temperatures not exceeding 4°C. Samples were cultured in mannitol salt egg yolk agar and using 100µl of the appropriate dilution into a petri dish containing agar with 2 replication per dilution. Each dilution was spread across the agar surface until dry and incubated at 37°C for 48 h in incubation cabinet.

Sample preparation for PCR

Each sample in 1 µl the solution peptone 0.1% was centrifuged at 10,000 rpm for 10 minutes to absorb all the broth left. Add 1 ml of water to washed sediment cells at the bottom of the tube and centrifuged at 9,000 rpm for 10 minutes. Remove sterilized distilled water mixed with sediments microliter volume of cells at the bottom of the tube will have samples that can be detect by PCR technique.

PCR test with primer *Coa1* and *Coa2*

Samples prepared to infection detected by PCR using primer *Coa1* detection of *S. aureus* to produce enterotoxin, PCR product size 821 bp, (Forward-ACC ACA AGG TAC TGA ATC AAC G and Reverse-TGC TTT CGA TTG TTC GAT GC), annealing temperature at 58°C, 40 cycles. Primer *Coa2* detection of *S. aureus* to non-produce enterotoxin, there are PCR product size 158bp, (Forward-TGC AGAT GGT ACT GCG ACA T and Reverse-CTT CCG ATT GTT CGA TGC TT) annealing temperature at 56°C, 40 cycles. After completing the PCR reactions Check PCR product with a 2% agarose gel electrophoresis. And find them under ultraviolet.

Results and Discussion

The samples from the 7 critical control points for PCR using Primer *Coa1* to detected annealing temperature at 58°C and the results were examined by agarose gel electrophoresis.

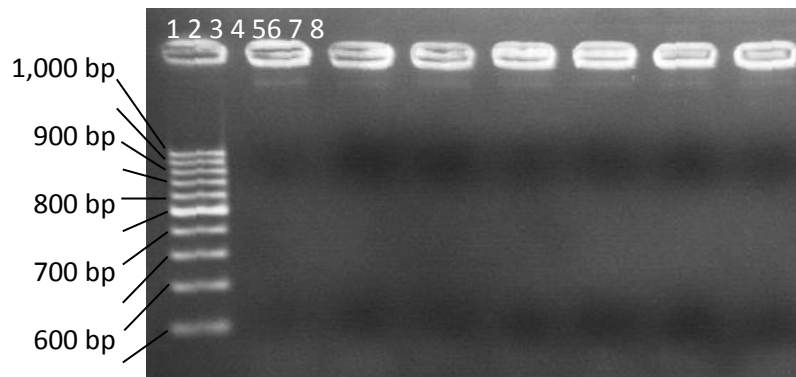


Figure 1 PCR product from primer *Coa1* : Lane 1) DNA marker ladder, 2) The teats of cows after clean, 3) Washcloths after used, 4) Milker's hands before milking, 5) The floor of waiting area, 6) The floor of bedding, 7) The floor of stall and 8) The floor of entrance milking stall

The Primer *Coa1* hasn't shown any PCR product (Figure. 1). PCR product from primer *Coa2* was detected all samples from the 7 critical control points by using annealing temperature at 56 °C for 40 cycles and the results were examined by agarose gel electrophoresis. The results was showed that all samples from the 7 critical control point and *S.aureus* purified can be detected and showed 158 bp DNA size in gels (Figure 2).

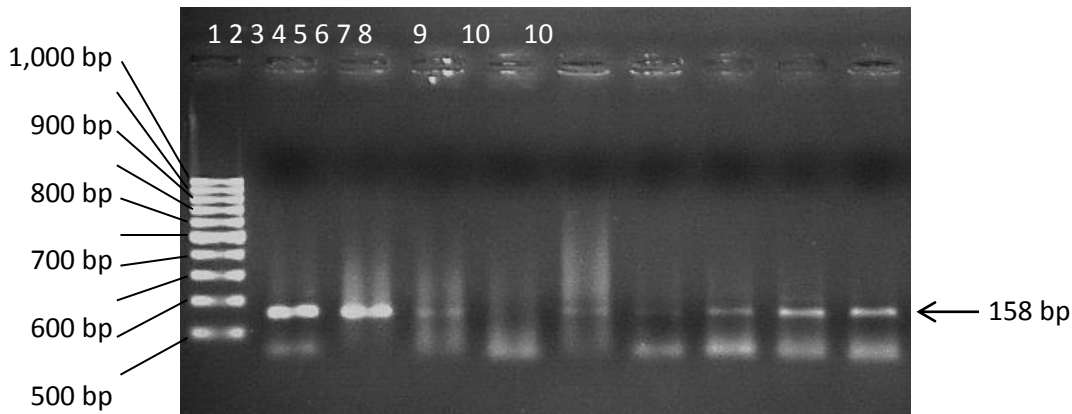


Figure 2 PCR product by primer *Coa 2*: Lane1) DNA markerladder, 2) *S. aureus* purified, 3) *S. aureus* purified in dilution 10^{-1} , 4) The teats of cows after clean,5) Washcloths after use, 6) Milkers' hands before milking, 7) The floor of waiting area, 8) The floor of bedding, 9) The floor of stall and 10) The floor of entrance milking stall.

The critical control in the milking process as 7 points to detect by PCR techniques using first PCR primer is primer *Coa1* which cannot be detected *S. aureus* in the samples because the primer can detected the only the *S. aureus* produce enterotoxin.(Borelli et al.,2011).The primer *Coa2* has designed and can detect the *S. aureus* from the sample were collected from the infected farm. Primer *Coa2* concluded that it can be used to determine the area of *S. aureus* mastitis in dairy cows and milking equipment used in the process and can also indicate that the samples collected seven samples were infected with *S. aureus* species does not create enterotoxin.

References

- Bhushan, J. 2000. Bovine Mastitis. Department of Veterinary Science. Pennsylvania State University.
- Borelli, B.M., I.C.A. Lacerda, L.R. Brandão, C.R. Vianna, M.C. Ferreira, F.C.O. Gomes, L.S. Carmo, L.G.D. Heneine and C.A. Rosa. 2011. Identification of *Staphylococcus* spp. isolated during the ripening process of a traditional Minas cheese. *Arq.Bras. Med. Vet. Zootec.*, v.63, n.2, p.481-487.
- Bryan, F.L. 1976. *Staphylococcus aureus*. pp. 12-105. Cited M.P. Defigueiredo and D.F. Splittstoesser, eds. *Food Microbiology: Public Health and Spoilage Aspects*. AVI Publ. Co. Inc., Westport, Connecticut.
- Phuektes,P., P. D. Mansell, and G. F. Browning. 2001. Multiplex Polymerase Chain Reaction Assay for Simultaneous Detection of *Staphylococcus aureus* and Streptococcal Causes of Bovine Mastitis.*J. Dairy Sci.* 84:1140–1148
- William, L., Robert, C. J., Harmon, O., Leary, J. and Jack, M A. 2002. *Mastitis and Its Control*. Kentucky University

Enhancing goat farm performance thru the farmer livestock school –goat enterprise management modality

Barcelo¹, P.M., J. Castillo², N. del Rosario³, R. Hipe⁴, R. Intong⁵, J. Pepito⁵, N. Naanep⁶, E.C. Villar⁷ & A.M. Alo⁷

¹Don Mariano Marcos Memorial State University, Bacnotan, La Union, Philippines,

²Department of Agriculture-Regional Field Office 02, Upi, Gamo, Isabela, Philippines, ³Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines, ⁴Department of Agriculture-Region 08, Tacloban City, Philippines,

⁵Central Mindanao University, Musuan, Bukidnon, Philippines,

⁶Sultan Kudarat State University, Tacurong, Sultan Kudarat, Philippines

⁷Philippine Council for Agriculture, Aquatic, Forestry and Natural Resources Research Council and Development, Los Banos, Laguna, Philippines

Abstract

The Farmer Livestock School-Goat Enterprise Management (FLS-GEM) Modality is an extension modality in which the farmers were trained the goat management interventions by attending barangay classes at their most convenient time. Initially, a curriculum was developed by a group of experts in the Philippines funded by Department of Science and Technology-Philippine Council for Agriculture Aquatic and Natural Resources Research and Development Council (DOST-PCAARRD). This curriculum served as the material in training the 12 national facilitators coming from 6 regions in the Philippines. After the national training, each set of regional trainer trained 25 regional facilitators the way they were trained during the national training. These regional trainers in turn trained at least 25 farmers on a scheduled basis based on the most convenient time of the farmers, preferably once a week for 4 hours. To assess the effects of the extension modality, 10% of the trained farmers served as cooperators in which monthly monitoring of their farm performance was done. The monitoring data included the inventory, dam performance, health, inflow and outflow.

Based from the results of the monitoring of farms, inventory of goats increased from an average of 5 does to 8 does at the backyard level, conception rate increased from 72% to 85% for the naturally-bred does, pre-weaning mortality was significantly reduced from 25% to 4.49% (t-test, .01), birth weight of goats increased from 1.50 kg to 2.25 kg, slaughter weight increased from an average of 18 kg to 22.50 kg.

This proves that the FLS-GEM extension modality is an efficient method of training farmers in the Philippines.

Keywords: farmer livestock school-goat enterprise management, enhancing goat farm performance, curriculum

Introduction

Goat production in the Philippines is an industry that is slowly gaining favor with investors. Demand as evidenced by slaughter rate is higher than production. In fact in 2010, goats slaughtered was 108% higher than kids born alive (BAS, 2012). This means that production cannot meet demand. Hence, population growth rate from 2006 to 2010 was low at 0.97% and production, 1.2%. In 2010, goat population in the Philippines was placed at 3.88 million heads,

with 98.5% in the backyard farms and 1.8% in commercial farms. Goat inventory however decreased in 2011 (BAS, 2012). Although the industry is still predominantly backyard, the share of the commercial sector continued to increase with 1.9% share in 2011.

From the 3-year data analysis from the study entitled “*National Goat Farm Performance*”, funded by the Department of Science and Technology-Philippine Council in Agriculture, Aquatic and Natural Resources Research and Development Council (DOST-PCAARRD) in 2008, it was seen that across regions, productivity was very low, characterized by a kidding interval of 8.37 and 8.67 months for the backyard and commercial farms, respectively, slow growth of kids and relatively high preweaning mortality of 18.2% and 14.7% for backyard form (BF) and commercial farm (CF), respectively (Cruz et al., 2010).

Kids that survived weighed on average 16kg and 20kg at 8 months for BF and CF, respectively. Those reached by project interventions of another PCAARRD-funded project, *Upscaling Rural Enterprise Development thru Innovative Goat Production Systems (URED)*, performed better in just two years (Cerbito et al., 2010). URED preweaning mortality was on average 10.7% while slaughter weight was 24.6kg. URED kidding interval was 237 days and conception rate, 91%. In this study, proper technological interventions and training reached the goat raisers through the Farmer Livestock School-Goat Enterprise Management (FLS-GEM) modality.

The FLS-GEM is a technology transfer that was developed by PCAARRD for teaching adults with goat as subject for training (Alo et al., 2014). The curriculum consists of the following topics: (1) Production management and housing, (2) Feeding management, (3) Breeding management, (4) Health management, (5) Ecological and nature management and (6) Enterprise management. This extension modality acknowledges the farmers’ ability to mix-match options suited to the farmers’ endowments.

The ultimate goal of this project, was to enhance goat production in the six participating regions by accelerating the delivery of alternative technology options to a bigger number of stakeholders. It also aimed to improve goat production among trained goat raisers by adopting the technology interventions as taught through the extension modality.

Materials and Methods

Preliminary Activities

The six regions that have identified goat as their top three regional commodity namely Regions 1, 2, 3, 8, 10 and 12 were included in this study. The generated technologies in goats by previous research studies were transposed into training materials through the production of training manuals. After which the national and regional facilitators were trained.

A two-stage purposive sampling method was followed in selecting the local government units to be the sites of the FLS-GEM training like, it should be the top three goat producing province and within the province, the municipality should be the top three producing within the province. Other criteria were also set as accessibility and without peace and order problem.

In the selection of farmer trainees, criteria were set as: (a) at least 5 does, (b) willingness to attend the scheduled trainings regularly, (b) willingness to do record keeping then share the recorded data, and (d) willingness to perform the selected technology interventions. When all the technical sessions were completed, each farmer was given 8 weeks to mix and match the options he likes and see for himself the effects of the tech-mix on his animals and his family. All data related to his on-farm trials were recorded.

Farm recording and monitoring

At least 5 and 10 does for each backyard and commercial farms, respectively, were tagged as the sources of data for dam performance. The monthly inventory, health, inflow and outflow were monitored on a monthly basis.

Results and Discussion

There were 23 national and total of 305 regional trainer strained. These trainers were responsible in training the goat farmers. This methodology ensured a more widespread dissemination of technologies at a faster and wider scope using the same modality. Within the two years implementation of the project, there were 2003 graduates of the FLS-GEM.

Improvement of the Goat Industry with S and T interventions Disseminated through the FLS-GEM

As shown in Table 1, through the FLS-GEM, the conception rate increased from 72% to 82%. As such, with the existing 6,200 does at the backyard level, there will be 49,055 kids produced with the practices of the S and T interventions over the traditional method of raising goats.

Table 1. Comparison between the traditional versus the with S and T interventions thru the FLS-GEM modality.

Science and Technology Interventions	Traditional Method of Raising	With Interventions Taught thru the FLS-GEM
Feeding and Breeding (Conception Rate from 71% to 81%)	Does to conceive 4,650 does	Does to conceive 5,022 does
Management (Kidding interval from 9 to 8 months)	Kids produced 32,552 kids	Kids produced 60,264 kids
Health (Pre weaning mortality from 25% to 18.6%)	Kids that will survive 24,412 heads	Kids that will survive 49,055 heads
Upgrading/breeding (Increased slaughter weight from 15 kg to 24 kg)	Volume of production at market age 366,187 kg	Volume of production at market age 1,177,320 kg
Effect of market weight on farm gate price (P100.00 to 110.00)	Price (P100.00/kg LW) P36,618,750.00	Price(P110.00/kgLW) P129,505,200.00

Assumption: 1,242 farmers with an average of 5 does= 6,200 does.

Monitored Data

Based on the performance of monitored goat farms as shown in Table 2, the conception rate increased from 72% to 82%. There was a significant decrease in the pre-weaning mortality of the goat farms from a baseline data of 25% to 4.49% ($t < .01$). This could be attributed to the regular deworming of the goats as prescribed in the FLS-GEM curriculum. Due to the infusion of the bloodlines of the improved breeds, the birth weights increased from 1.50 kg to 2.25 kg. The slaughter weight of the goats also increased from 15 kg to 22.50 kg. The results of the analyses of the data monitored proves that with proper technological interventions, there will be an improvement in the performance of the animals. The findings support the findings of Cerbito et al. (2010).

Table 2. Monitored performance of goat farms before and during the conduct of the FLS-GEM (As of December 2014).

PERFORMANCE INDICATOR	BEFORE	DURING THE CONDUCT
Conception rate (%)	72	82
Pre-weaning mortality (%)*	25 ^a	4.49 ^b
Birth weight (kg)	1.50	2.25
Slaughter weight (kg)	15.00	22.50

*-Significant, t-test

Conclusions and Recommendation

Based from the results of the study, the following can be concluded:

1. The Farmer Livestock-Goat Enterprise Modality is a fast method of disseminating technologies on goat production because trainings can be done simultaneously among the regions, provinces, municipalities and local government units.
2. With the adoption of the technologies that are included in the FLS-GEM curriculum, performance of animals in the farm improved particularly the reduction of mortality among the pre weaners.
3. The birth weight, weaning weight and weight at 8 months increased with the adoption of the technologies on upgrading and artificial insemination.

Recommendations:

1. The conduct of the FLS-GEM could be improved with an institutionalized support from the local government units like a policy to be promulgated at the barangay or municipality to undergo trainings before a farmer can avail of funded projects on goats.

References

- Cerbito, W., J. O. Datuin, J. N. Nayga, C. E. O. Orden, A.M. Alo and E.C.Villar, 2010. Rural Enterprise Development thru Innovative Goat Production Systems. PCAARRD Unpublished Terminal Report.
- Cruz, E.M., P.M. Barcelo, A. Asuncion, M.E. Orden, R. Hipe, J. Pepito, R. Intong, A.M. Alo and E.C. Villar, 2010. National Goat Farm Performance. PCAARRD Unpublished Terminal Report.

Constraints to improved productivity of smallholder cow-calf systems in South Central Coast Vietnam – insights from recent surveys

Dinh Van Dung¹, Nguyen Thi Mui², Nguyen Xuan Ba², Nguyen Huu Van², Rowan Smith³, David Parsons³ & Jeff Corfield^{2, 4*}

¹Hue University of Education, Hue 47000, Vietnam, ²Hue University of Agricultural and Forestry, Hue 47000, Vietnam, ³Tasmanian Institute of Agriculture, University of Tasmania, Hobart 7001, Tasmania, Australia, ⁴Corfield Consultants, Townsville, Australia, 4811

Abstract

Growth in demand for beef in Vietnam has resulted in historically high prices for cattle, especially crossbred stock, across most age/size classes. Despite increasing live cattle imports, a significant proportion of cattle are still sourced from smallholders, especially for regional markets. Smallholder cattle producers have benefited from this rising demand for cattle. However, they face many challenges in order to meet the continuity of supply and product consistency and quality increasingly demanded by the market. In South Central Coastal (SCC) Vietnam, smallholder cattle production has traditionally relied on extensive grazing supplemented by crop by-products like rice straw. Decreasing access to common grazing land has exacerbated existing cattle nutrition and husbandry issues, contributing to long calving to conception intervals (CCI) and irregular calving, making it difficult for smallholders to coordinate calving with optimum seasonal feed supply. This paper examines results from two recently surveyed South Central Coastal Vietnam smallholder cow-calf producer communities which exhibited significantly different ($P < 0.001$) CCIs. The survey found that the main contributor to CCI length was the interval between calving next heat detection (CNHDI) rather than number of inseminations before conception. Preliminary survey results indicated no significant correlation between CCI and key resource and production variables associated with cow-calf systems examined, despite there being significant ($P < 0.005$) between-commune differences for many of these. The paper also considers the implications of these findings for future research into CCI related issues.

Keywords: body condition, calving interval, cow-calf production, nutrition, smallholder

*Corresponding author: jeffcorfield@yahoo.com.au

Introduction

Cattle production is an integral part of smallholder farming systems in Vietnam, with the South Central Coastal (SCC) region accounting for 22.7% of national cattle production in 2014 (MARD, 2014). Farming in the SCC region is constrained by low fertility sandy soils, and harsh climatic conditions characterized by long, hot, dry seasons and shorter rainy seasons (Parsons et al., 2013). Smallholder cattle production has traditionally relied on extensive grazing, supplemented by use of crop residues, especially rice straw. Poor cattle nutrition and husbandry contribute to long and variable calving to conception intervals (CCI). This results in irregular calving making it difficult for smallholders to coordinate calving with optimum seasonal feed supply (Parsons et al., 2013). Previous research (Montiel & Ahuja, 2005; Phung, 2009) suggest that long CCIs may be linked with post-partum anoestrus (PPA) in tropical cattle, which may be more associated with factors such as nutrition and husbandry, than genetic differences. This paper compares CCI results from recent surveys conducted in two smallholder cattle producer communities in SCC Vietnam. It also compares these with key resource and production system

factors within and between communes to explore possible links to long CCI and by inference, PPA.

Materials and Methods

The study was conducted in two communes: An Chan in Phu Yen province and Tay Giang in Binh Dinh province, SCC Vietnam. Both communes have similar mixed crop-livestock smallholder communities, but differ significantly in availability of land and other resources used for cattle production and to some extent seasonal conditions. Surveys of 61 households in An Chan and 66 households in Tay Giang (with a total of 128 and 149 cows respectively) were conducted between January and August 2014, to record data on farm resources; cattle production system and cow performance, including body condition score (BCS) (Gaden et al., 2005) and reproductive history, specifically CI, CCI, number of inseminations per conception and calving to next heat detection interval (CNHDI). Data were subjected to ANOVA and Pearson correlation analyses using *Statistix* 10 (2010) to compare results between communes and explore associations between CI, CCI, and various production variables.

Results and Discussion

While both communes are similar mixed crop-livestock smallholder communities, Tay Giang had 61% more total land area per household (5241 m² vs. 3367 m² (P<0.005) and 3 times the arable cropland (4899 m² vs. 1653 m², P<0.0005) per household than An Chan. However, An Chan had twice the area of planted forages per household compared to Tay Giang (1401 m² vs. 615 m², P<0.0005). While both communes had similar cow numbers per household (mean 2.1, 2.2, 0.73) Tay Giang had (92%) crossbred cows 92% crossbred cows compared to 69% for An Chan (P<0.0001). Farmers in both communes spent similar time grazing cattle in the dry season (693 hours vs. 666 hours, P=0.8310 but An Chan farmers spent 3 times longer grazing during the rainy season (394 hours, vs. 137 hours, P<0.0005). Farmers in both communes fed similar amounts of crop by-products as pre-partum supplements (0.6 kg/d vs. 0.73 kg/d) but Tay Giang farmers fed nearly 50% more (0.63 kg/d to 0.93 kg/d) to cows post-partum (P<0.005). There was a significant difference (P<0.0005) in mean CCI (197 vs. 118 days) between Tay Giang and An Chan, with a much wider spread of longer CCIs in Tay Giang than An Chan (figure 1a). Forty five percent of An Chan CCIs were <90 days, compared to 18% in Tay Giang, while CCIs <120 days were 64% and 31% respectively (figure 1b).

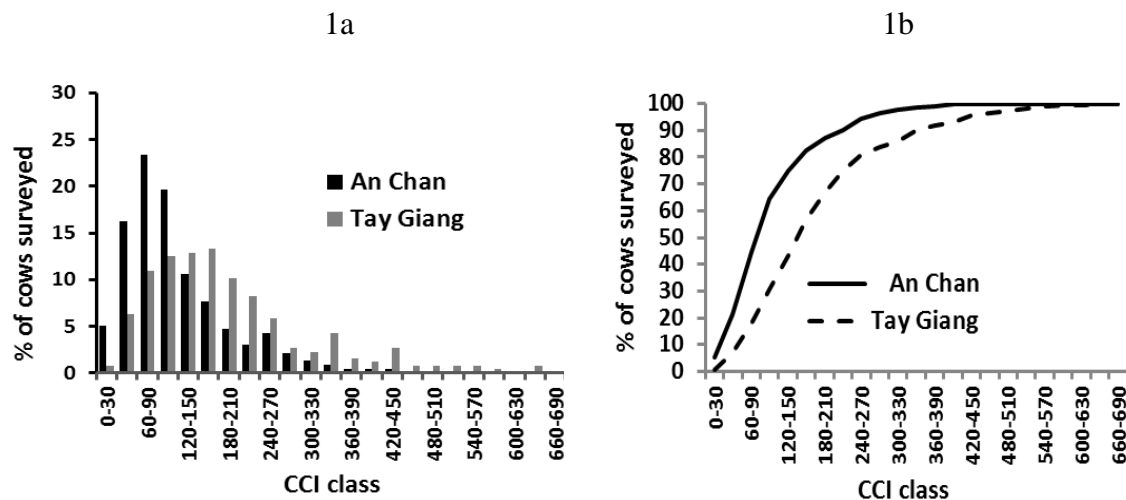


Figure 1. CCI frequency data comparisons for cows from households surveyed in An Chan and Tay Giang communes, 2014. 1a is cumulative frequency and 1b is frequency distribution.

With a high percentage of conceptions occurring on first insemination (80% for Tay Giang and 95% for An Chan) irrespective of insemination method (bull or AI) or breed, there was a strongly significant correlation ($R=0.98$, $P<0.0001$) between CCI and CNHDI. Long CHNHDI may point to problems with oestrus detection by farmers or genuine post-partum anoestrus, which may be linked to resource or production variables. However, we found no statistically significant ($P>0.05$) correlations between CCI or CNHDI and individual resource variables such as area of forage grown, pre- or post-partum feeding of crop by-products or time spent grazing. Likewise there was no significant correlation between CCI and performance indicators such as breed, calving time, number of calves produced / cow and BCS, within in or across communes. This is despite a significant difference ($P<0.0005$) between mean BCS for Tay Giang (2.96) and An Chan (2.76). Variability within and between cows from surveyed households contributed to this. Survey results show that average CCI was nearly 80 days longer in Tay Giang than in An Chan - equivalent to 2.5 months or nearly 4 oestrus cycles. Most of this difference can be attributed to longer and more variable CNHDI in surveyed Tay Giang cows, some 25% of which exceeded 200 days (figure 1b). While failure to detect oestrus may be a factor, only 4% of surveyed farmers in An Chan and Tay Giang whose cows had long CCIs felt this was an issue. By contrast, 60% in An Chan and 45% in Tay Giang felt poor nutrition due to lack of feed contributed to long CCIs, while 32% and 38% respectively nominated poor body condition and 40% and 27% suggested length of suckling time as a possible cause. Montiel and Ahuja (2005) suggested suckling time and BCS as possible contributors to post-partum anoestrus, but the surveys here indicated no significant correlation between BCS and CCI. Data on suckling length was not collected.

Conclusion

Survey data used here was collected to profile smallholder cattle production systems and their supporting resource base, rather than identify specific factors contributing to CCI length or indeed post-partum anoestrus. While the surveys identified significant issues with long CIs and CCIs, in Tay Giang commune especially, as well as differences between the resource base and associated cattle production systems of both communes, it was not possible to identify specific cause and effect associations between them and CCI here. However, the results presented point to the need for further targeted research into CCI length, which is a major constraint to improving productivity in smallholder cattle production in this region.

Acknowledgements

We wish to acknowledge the research funding contribution by the Australian Centre for International Agricultural Research (ACIAR) through project LPS/2012/062.

References

- MARD, 2014. Ministry of Agriculture and Rural Development. Livestock statistic up to 01/10/2014. Updated 01/01/2015.
- Parsons, D., P. A. Lane, L. D. Ngoan, N. X. Ba, D. T. Tuan, N. H. Van, D. V. Dung, and L. D. Phung. "Systems of cattle production in South Central Coastal Vietnam." *Livestock Research for Rural Development* 25, no. 2 (2013): 1-8.
- Gaden, R., G. Duddy, and J. Irwin . "Identifying live animal condition scoring system for the Australian livestock export industry". MLA, North Sydney NSW (2005).
- Montiel, F., and C. Ahuja. "Body condition and suckling as factors influencing the duration of postpartum anestrus in cattle: a review." *Animal Reproduction Science* 85, no. 1 (2005): 1-26.

Phung, L. D. "Genotype by environment interaction: a case study of productive and reproductive performance of Yellow local and F1 (Red Sindhi x Yellow local) cattle in two production zones in Quang Ngai, Vietnam." *Livestock Research for Rural Development* 21, no. 2 (2009).

Statistix 10 for Windows. 1985-2013. Analytical Software, PO Box 12185, Tallahassee, USA

Efforts to increase production of cow's milk through the cooperation empowerment in Sinjai regency

Rohani, S¹., R.Malaka², & M.Darwis³

*Faculty of Animal Science, Hasanuddin University, Indonesia
Jl. Perintis Kemerdekaan Km.10, Makassar, 90245, Indonesia*

Abstract

People needs of milk are not fulfilled because of low development of dairy cattle production. Therefore, it is necessary to be developed so the milk production meets the people needs of milk. The causing factors why milk production cannot fulfill demand for milk are small scale ownership of dairy cattle, low milk production ability, unprofitable milk price and high production cost. Dairy cattle business cannot be separated from cooperation existence. To improve the development of dairy cattle production needs the cooperation empowerment to increase business scale, improve milk production ability and reduce production cost. The cooperation empowerment are done by providing female dairy cows, quality feed and concentrate at affordable price, and cooperation business network development.

Keywords: Milk production, empowerment, cooperation

Email : ¹nanirohani24@yahoo.co.id ²malaka_ag39@yahoo.co.id ³Darwism24@gmail.com

Introduction

Dairy cattle agribusiness succeeds developing business organization become a community farm business, until now fresh milk business start from headwaters till downstream are owned by dairy farmers through cooperation as primary and secondary cooperation. However, the farmers and their cooperation do not escape from any problems. Apart from income tax for milk commodity, levies exist, 'protection against dairy commodity revoked'. As a result, cooperation and farmers need to work harder to fulfill dairy processing industry standard and find another marketing alternative for products preferred by consumers. It is the biggest challenge that needs to succeed in order to win consumers' trust providing quality product, various and affordable product, dairy cooperation will become "agribusiness cooperation" (Yunasaf, 2008).

Presence of dairy cow group, including their dynamism cannot be separated from external influences; from cooperation and a counselor who assigned to the local farm. Thus, group dynamism of dairy farmers will be influenced by cooperation in running its functions and counseling as reformer agent. The element can be the existence of effective leadership, clear goal, complete group structure, well-organized group function, good development and group maintenance, teamwork and group efficiency.

The presence of livestock cooperation as one supporting infrastructure development livestock sub sector has an important role in improving member's welfare that consist of farmers. In South Sulawesi, livestock cooperation is only in Sinjai and Enrakang Regency which are dairy development area and milk production supported by government and climate or location which is good for dairy development.

Dairy cooperation Sintari existed since 2005 because Sinjai government has seen community seriousness in gunung perak village in raising dairy cattle which increase their

income even their primary income is farming. Sinjai government provide development funds to dairy cooperation “Sintari” because in 4 years, government looked the development of dairy cattle raising in that area, whether they can produce or not.

Dairy cooperation “Sintari” capital in the beginning was supported by government fund, especially funding milk processing machine and the rest was from members who paid principal and mandatory savings. However, the problem now is lack of member’s awareness and responsibility – they have not completed the administrative requirements, signed the membership book and have not paid whole principal savings include mandatory savings and others which was set at bylaw for candidate member.

According to the issues, dairy cooperation empowerment is necessary due to its strategic role in the dairy agribusiness development. This study aimed to discuss the empowerment aspect which can be done by dairy cooperation to improve the dairy cattle agribusiness, so it can give a positive impact to the dairy production improvement.

Materials and Methods

Research method used in the study is descriptive method which describe or details about a situation clearly – capital in dairy cooperation “Sintari” in Gunung Perak village, West Sinjai, Sinjai.

The used data is (a) qualitative data – sentence, statement given by leaders and members of dairy milk “Sintari” in Gunung perak village, West Sinjai, Sinjai, (b) quantitative data that comes from dairy cooperation “Sintari”.

Data source which is used in the research is: (a) Primary data – the data obtained directly through interview and observation with members of dairy cooperation Sintari, (b) secondary data – data obtained by related parties. Obtained data is analyzed using descriptive statistic. (Sugiyono, 2004).

Result and Discussion

Obstacles and solution of dairy cow development

Several obstacles faced in the dairy cow development are the farmer powerlessness to develop business because of low income. Income that they earn is used to pay the family needs so they cannot develop the dairy business. According to needs or demand is bigger than the milk availability. Based on the condition, dairy business to produce fresh milk is very prospective to be developed in Indonesia, especially in South Sulawesi (Sirajuddin, 2009).

Business scale improvement

Dairy cow business scale is defined as number of cow raised, both lactating cows and not-lactating cows. The cow which is raised by dairy cooperation Sintari categorized small business scale, with ownership scale 3 – 5 cows, and producing ability 10 – 12 liter/cow/day. Not all cow produce milk in a year because they are not lactating. Muatip (2008) stated that dairy cooperation is a livestock business to increase member’s welfare by providing business field – dairy cattle. Therefore, dairy cattle cooperation in west java form a *top-down* policy which gain fund in a procurement of imported dairy cow give to the cooperation members as a loan. Farmers return the loan through milk and obey all cooperation rules. Farmers who get dairy cows must sell the fresh milk production to the cooperation at price as Milk Processing Industry and cooperation agreed. It is seen from the cooperation achievement and ability to maintain capital, number of members, and business success. More member means higher milk production and more cooperation income both through savings, membership payment and milk sales.

Milk price at farmer level

Main income of dairy business is daily milk sales. Income is determined by number of milk produced and the milk sales price. Number of milk produced is determined by number of dairy cow that produce and producing ability. More dairy cow that produce very well, more milk can be sold or marketed. Thus, high income will be achieved if the price offered is high.

High price on dairy business means price will give profit to the dairy business. Milk sales price is determined by production cost, the highest cost is on concentrate feed.

Reduce production cost

Dairy cattle, farmers not only raise lactating and not-lactating cow, but also dairy cow that has not produced yet. Non-productive cow consist of calf, baby or adult cow, non-productive cow raised to replace non-productive cow. Maintain cost of non-productive cow is taken from the cows that produce milk. Thus, in agribusiness calculation, lactating cow both pay itself and non-productive cow. Therefore, more non-productive cow raised will give bad impact on lactating cow and small business profit.

One major cause why dairy farmer's income is too many non-productive cow raised and small number of productive cow (lactating cow). According to Soekartawi (2003) said that production cost is value of all production factors used both in services and thing during the producing process.

Dairy cattle condition in Sinjai regency

One of important element in milk development is dairy cow development both quantity and quality aspect. Sinjai government develops dairy cow in some sub-districts especially in mountain. In 2009 – 2013, growth of milch cow run slowly. The population can be seen on Table 1.

Table 1. Milk cow population per sub-district in Sinjai

No	Sub-districts	2009			2010			2011			2012			2013		
		Male	Female	Total	Male	Female	Total	Male	Female	Total	Male	Female	Total	Male	Female	Total
1.	West Sinjai	39	205	244	23	219	242	30	269	299	35	180	215	30	156	186
2.	Sinjai Borong	0	22	22	0	9	9	0	0	0	0	0	0	0	0	0
3.	South Sinjai	0	0	0	0	0	0	4	4	8	4	4	8	2	3	5
4.	Tellu Limpoe	0	0	0	0	0	0	1	4	5	1	4	5	1	3	4
5.	East Sinjai	0	0	0	0	0	0	1	0	1	0	7	7	0	2	2
6.	Central Sinjai	0	0	0	0	0	0	1	1	2	1	1	2	0	0	0
7.	Bulupoddo	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8.	Sembilan Island	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total Per year		39	227	266	23	228	251	37	278	315	196	196	237	33	164	197
Average Per year		3,25	18,92	22,17	1,92	19	20,92	3,08	23,17	26,25	16,33	16,33	19,75	2,75	13,67	16,42

Source : Livestock Statistic of Sinjai Regency 2010-2014.

Table 1 showed that milch cow growth in Sinjai 2009 – 2013 decreased, milch cow procurment through livestock support from livestock general Instance by applied partnership system which aimed to produce pasteurized milk for people consumption until Makassar.

Dairy cooperation Sintari is one of dairy cattle business that become center of dairy cattle farm as an example for farmers in west sinjai, Sinjai. Therefore, Livestock department of Sinjai opened dairy cattle business.

Dairy cooperation Sintari is the only one dairy cattle cooperation which is in Batu Leppa village, Gunung perak, west sinjai, Sinjai whose milk processed to be Susin traditional drink. The following data is total milk which was accepted from 2008 – 2013 in dairy cooperation Sintari on Table 2.

Table 2 showed that milk received the cooperation in 2008 was 41.740 liter/year, in 2009 decreased 27.731 liter/year. In 2010, milk production increased 32.071 liter/year, however in 2011 till 2013, the production decreased 16.485,5 liter/year. It was caused by the decreasing milk production in some villages and sub-districts and the death of female dairy cow. In some places which has no production, the dairy farmer switch their job to be beef cattle farmer.

Table 2. Milk received (liter) in dairy cooperation Sintari

No	Month	2008	2009	2010	2011	2012	2013
1.	January	2241.5	1495.5	1117	2753.5	1391	1003
2.	February	1968	2280.5	3506.5	1870.5	1280	1306
3.	March	2793.5	2249	4246.5	1902.5	1229	1236
4.	April	3531.5	2352	3761	1810.5	1352	1725.5
5.	May	4972.5	2631	2960	1571	1629	1917.5
6.	June	5694.5	1873	3329	1215	1825	1329.5
7.	July	5294	2118	3144.5	1113	1113	1134
8.	August	4253.5	1869	2325	1380	1269	1345
9.	September	3185	1849	1434	2770	1525	1424
10.	October	2796	2252	2092.5	1341	1235	1093.5
11.	November	2593	2535	1850	1850	1541	1670
12.	December	2417	4227	2305	1660	1223	1301.5
Total Per year		41740	27731	32071	21237	16612	16485.5
Average		3478.33	2310.92	2672.58	1769.75	1384.33	1373.79

Source: Dairy Cooperation Sintari West Sinjai, 2014.

Conclusion

1. Dairy cattle in Sinjai now needs to be revitalized the development, so milk production that has become Sinjai flagship product could fulfill people needs. Decreasing of milk production is caused by farmer's income, so they cannot develop the dairy cattle and quit their job.
2. Obstacles faced by dairy farmer are small business scale, low ability of milk production, unprofitable milk price and high production cost.
3. To improve dairy cattle development which has positive impact to the milk production improvement is by increasing ownership scale of dairy cattle business, improve the ability of dairy cow production and reduce production cost.
4. Enhance the ownership scale of dairy cattle and ability of dairy cow production can be done by increasing the empowerment of dairy cooperation "Sintari", provision of female dairy cow and quality concentrate feed at affordable price, and development of Sintari business network.

References

- BPS. 2014. Statistik Peternakan Kabupaten Sinjai 2010-2014. Badan Pusat Statistik Kabupaten Sinjai, Sinjai.
- Muatip, K. 2008. Kompetensi Kewirausahaan Peternak Sapi Perah Kasus Peternak Sapi perah Rakyat Kabupaten Pasuruan Jawa Timur dan Kabupaten Bandung Jawa Barat. Pasca Sarjana IPB Bogor, Bogor.
- Sirajuddin, N. 2009. Prospek Usaha Sapi Perah di Sulawesi Selatan. Buletin Dinas Peternakan Provinsi Sulawesi Selatan. ISSN 1858 – 0777. Edisi 32, Makassar.
- Soekartawi. 2003. Agribisnis. Teori dan Aplikasinya. PT Raja Grafindo, Jakarta
- Sugiyono. 2004. Statistika untuk Penelitian. ALFABETA, Jakarta.
- Yunasaf, U. 2008. Dinamika Kelompok Peternak Sapi Perah dan Keberdayaan Anggotanya di Kabupaten Bandung. Pasca Sarjana IPB Bogor, Bogor.

Why poultry welfare in Kuwait is an obstacle to trade?

Alsaffar , A. A. & F. Khalil

Kuwait Institute for Scientific Research, ELSRC, 24884 Safat, 13109 Kuwait

Abstract

The poultry industry is one of the most important animal production industries and contributes to approximately 10% of all meat and eggs produced in the world each year. Poultry is highly susceptible to disease outbreaks that may cause irreversible economical losses to the poultry industry. Biosecurity is a modern practice introduced out of a need to protect the poultry from an intentional or unintentional threat of a biological agent. In everyday management, biosecurity is an endless endeavor to keep viral disease agents or the spread of such disease agents at bay. Biosecurity is even more important today because of the avian flu disease, type A influenza, which has been in the global news lately, Wild water birds are the natural carriers of this virus from where it can spread to domestic poultry and become fatal. Type A influenza can occur in many forms. Humans and some other animals are susceptible to some of these forms, but poultry are susceptible to all of them. Any time there is an outbreak, health officials are concerned because influenza viruses continuously change and officials have to determine how it happened and if it can become epidemic. Therefore, biosecurity of poultry farms is an important and vital practice to reduce the burden of any disease producing agent in any commercial operation. Biological hazards or biological agents, infectious/noninfectious, are such things as viruses, bacteria, fungi, and protozoa are responsible for disease outbreak in poultry. Most of the poultry industries in the world have developed biosecurity measures as a part of animal welfare to maintain the safety of poultry from biological hazards and be used for protection and disease control of the poultry. However, in many cases, these program measures including vaccination are not applied or followed properly because a comprehensive program usually is not in effect. The purpose of this study is to provide the poultry industries with a brief account of the most important aspects of poultry biosecurity program so that they may have a better understanding of the farm animal's operations. It is extremely important that poultry industry in Kuwait implement a comprehensive biosecurity program (regulations and recommendations) in their farms to ensure better quality production.

Keywords: welfare, biosecurity program, education, chicken and laying hens

Introduction

The poultry industry, in Kuwait has been applying some kind of welfare measures for many years (Rose & Alsaffar, 2014). It is not well understood that poultry may get diseases from many sources including people, equipment, supplies, trucks, cars, pets, wild animals, dust, feathers, manure, rodents, predators, insects, other poultry, other birds, and contaminated feed and water. Diseases not only increase mortality but also could cause slower growth, lower egg production rates, reduce product quality and lower customer satisfaction (Charles & Ayodele Adekunle, 2014), which all lead to tremendous financial losses to producers and could be reflected in price increase to the consumers.

The Kuwaiti poultry sector is possibly the fastest growing and most flexible of all livestock sectors. Over the last five decades or so it has expanded, consolidated and globalized, driven primarily by very strong demand. Total consumption of poultry meat and eggs has

increased dramatically during the past decades and continues to increase ahead of human population growth (Rose & Alsaffar, 2014). The relative success of the global poultry industry can be attributed to several factors: Technical innovations and the substitution of hand labour by capital supported the development from small back-yard flocks local demand to large scale production for food chains supplying urban consumers; Controlled environment: closed houses with optimized temperature and lighting program to facilitate year-round production; Reduced risk of mortality and /or depressed performance: all-in, all-out management, prophylactic vaccination and improved general hygiene; Better nutrition: balanced feed formulation according to the specific needs of each flock; and last but not least, Improved genetic potential for growth rate in meat-type poultry and eggs in eggs-type chickens (Penz & Bruno 2011). With growing size, companies at all levels of the industry-breeders, multipliers (hatcheries), egg producers and broiler growers- became more professional and market driven.

Since the 1990s, the poultry sector in Kuwait has been part of the "livestock revolution". Demand has grown, driven urbanization, population growth, trade and high expenditure elasticity's for livestock products such as chicken (Rose & Alsaffar, 2014). Supply has been able to match demand, because of technology developments in breeding and nutrition (Upton, 2008; Rose & Alsaffar, 2014). Concentration of the sector, an important trend to increasing requirements to meet poultry health, food safety and quality standards, have raised the issue of poultry welfare needed from the farm, to market, transport and the slaughterhouse.

This study discussed Kuwait status of broilers, laying and broiler breeder hens welfare at the farm level and the impact of establishes new welfare recommendation and requirements in Kuwait. Therefore, the objectives of the study were are to assess the existing poultry welfare practices and developed a comprehensive poultry welfare program for poultry farms and companies.

Materials and Methods

Assessment of the existing poultry welfare in poultry companies

This were included: poultry welfare rules and regulations available such as stocking density, fear and stress, feather pecking, and the status of isolation of different facilities, availability of security procedures for the different facilities, management procedures available, rodents and pest management available, and sanitation and disinfection procedures.

By developing and successfully applying the new poultry welfare program, the incidence of diseases will be significantly reduced leading to a reduction in the operation costs and an increase in profitability of poultry farms and poultry companies in Kuwait. This reduction in production costs is expected to lead to a reduction in prices benefiting consumers in Kuwait and different animal sectors in Kuwait.

Results and Discussion

Development and application of poultry welfare program

Based upon findings of the assessment, a poultry welfare program was designed and developed for poultry farms and companies with the cooperation of the decision makers of the poultry sector as well as with the veterinarians to ensure the success of the program. This program was included the establishment of rules and regulations for the followings:

- Farm management procedures included all practices related to poultry houses welfare, procedures established for the preparation for the receiving of new flocks, the use of all in all out practice, the staff management and behaviours of people working in the poultry houses themselves and other production facilities.

- Isolation procedures included the confinement of the birds within a controlled environment, buildings isolation, and isolation of birds by age.
- Traffic control procedures included check points for the visitors and rules and regulations that must apply before visitors are allowed to visit any of the company's facilities, control any traffic onto the farm and traffic patterns within the farm including movements of staff and workers.
- Sanitation, health and hygiene, and disinfection procedures included the type used the time and frequency of its use, disinfection of materials, people, and equipment in the farm and the ones entering the farm and the cleanliness of the personnel on the farm.
- Follow up and auditing of poultry welfare rules, included daily follow up with staff and workers in all the poultry houses till satisfied of the success of the poultry welfare program.
- Lectures and seminars were conducted on the new poultry welfare program and its procedures, application, advantages and benefits as well as the technical procedures for specific items of the program.

Welfare for broilers

In Kuwait, broilers were generally held in large groups in environmentally controlled housing system. Broilers were usually kept free loose housed on litter with automated of feed and water. Farmers understood that in order to raise the birds with maximum efficiency, many conditions must be fulfilled: stress prevention, supply of good feed and water and sanitation (Alsaffar et al., 2007; Rose & Alsaffar, 2014). In providing these conditions, farmers ensure a basic level of animal welfare (van Horne & Achterbosch, 2008).

Welfare for layers

The majority of all commercial layers in Kuwait were kept in confined housing systems with light control, power ventilation and mechanical feeding. The space per hen in conventional cages was very limited making it impossible to express natural behaviours such as wing flapping (Vitset al., 2005, van Horne & Achterbosch, 2008). To accommodate societal concerns about animal welfare, recommendations of laying hens welfare on housing and cage space allowances from 430 to 550cm² per hen range (UEP. 2000).

Welfare for Broiler Breeders

The two most commonly used commercial restriction breeder feeding programs in Kuwait were skip-a-day, in which amounts of feed calculated to achieve desired body weights were fed on alternate days; and limited every day, in which half of skip amount is fed daily. The skip-a-day program was preferred for males to provide greater body weight than limited every day feeding (Mench, 2002).

Conclusion

The study shows that the Kuwaiti poultry industry plays an important role in providing fresh, high quality food products and also plays an integral part in the nation food security system. One of the most important challenges faced by the people in the poultry industry in Kuwait is the high mortality rate due to the birds being under absent of poultry welfare (stress and diseases) during the bird's life cycle causing significant financial losses to the industry. It was important to note that diseases not only increases mortality but also could cause slower growth, lower egg production rates, reduced product quality and lower customer satisfaction, which all lead to this

tremendous financial losses to producers and could be reflected in price increase to the consumers.

Also, specific biosecurity program should be developed for individual poultry farm according to their particular need and situations with the cooperation of the decision makers and farm veterinarian to ensure the success of the program

References

- Alsaffar, A., A. Alnasser; M. Mashaly; M. Albahou and A. Alhaddad, 2007. Poultry biosecurity- A case Study at KUPCO. Kuwait Inst. Sci. Resc. Report No. KISR 8255, Kuwait.
- Charles, A.O. and Akinwumi, AyodeleAdekunle, A. 2014. Awareness and Practice of Biosecurity Measures in Small Scale Poultry Production in Ekiti State, Nigeria. *J. Ag. Vet. Sci.*7: 24-29.
- Mench, J.A., 2002. Broiler breeders: feed restriction and welfare. *World Poultry Sci. J.*58:23-29.
- Penz-Jr, A.M. and D.G.Bruno, 2011.Challenges facing the global poultry industry until 2020. 22 APSS, Sydney, South Wales, pp 49-53.
- Rose, S.P. and A.A. Alsaffar, 2014. Poultry meat and hatching egg production in Kuwait. *World Poultry Sci. J.* 70:5-14.
- United Egg Producers (UEP), 2000. Recommendations for UEP Animal Welfare Guidelines. Submitted By AWAC.
- Upton, M., 2008. Scale and structures of the poultry sector and factors inducing change: intercountry differences and expected trends. P IntCnf Poultry 21 C:avian influenza and beyond, held 5-7 November 2007, Bangkok, Thailand.
- Van Horne, P.L.M. and T.J. Achterbosch, 2008. Animal welfare in poultry production system: Impact of EU standards on world trade.*World Poultry Sci. J.*64:40-52.
- Vits, A., D. Weitzenburger, and O. Distl, 2005. Comparison of different housing systems for laying hens in respect to economic, health and welfare parameters with special regard organized cages (literature review). *DTW.* 112:332-342.

Nutrition, fatty acid and cholesterol content of Garut lamb meat at different ages fed with diet containing mungbean sprouts waste

Sri Rahayu,, Tuti Suryati & Sita Arum Prabawati

Faculty of Animal Science, Bogor Agricultural University, Bogor 16680 Indonesia

Abstract

Garut lambs from two different ages, under five month old up to eight months old used for meat production, were fed a concentrate diet containing mungbean waste. The effect of different ages on nutrition, fatty acid composition, and cholesterol content were measured. After fattened about 3 months in individual cage, a total of six male lambs (3 lambs under five month old and 3 lambs up to eight month old) were slaughtered. Lambs meat was taken from *Longissimusthoracis et lumborum*. Nutrition content of lamb meat was quantified by proximate analysis. Fatty acid composition and cholesterol content were analyzed by gas chromatography. Analysis of variance was used to compare differences of age effect on nutrition, fatty acid composition, and cholesterol content. The different ages in this study had no significant effect on nutrition content, fatty acid composition, and cholesterol content ($P > 0.05$). The total of SFA was higher than USFA in garut lamb meat.

Keywords: nutrition, fatty acid, cholesterol, lamb meat, mungbean sprouts waste

Corresponding Author: sry19657@yahoo.co.id

Introduction

The lamb meat is one kind of meats that consumed by community in Indonesia. Indonesian people like the lamb meat because of its typical aroma (*muttony*). However, they also afraid to consume lamb meat because the fat and cholesterol of lamb meat were considered to trigger hypertension and coronary heart disease. The content of nutrient, cholesterol and fatty acid composition of meat were determined by the animal species, age, sex, and animal feed (Romans et al., 1994; Salvatori et al., 2004). So the aim of this experiment was to evaluate the nutrition and fatty acid composition and cholesterol content of garut lamb meat at different ages fed with diet containing mungbean sprouts waste.

Materials and Methods

Six male of garut growing lambs (three lambs two month old and three lambs up to eight month old), were used in this experiment. The animals were fattened with complete ration pellet containing 30% of mungbean sprouts waste (87.7% dry matter, 16.7% crude protein, 24.5% fiber, 3.7% extract ether and 72.2% total digestible nutrient). After fattened about 3 months in individual cage, a total of animals were slaughtered. Lambs meat was taken from *Longissimus thoracis et lumborum*. Nutrition content of lamb meat was quantified by proximate analysis. Fatty acid composition and cholesterol content were analyzed by gas chromatography. Analysis

of variance was used to compare differences of age effect on nutrition, fatty acid composition, and cholesterol content.

Results and Discussion

Nutrition content (moisture, ash, protein, fat and carbohydrate) of lamb meat were not significantly different between ages (Table 1). The average of moisture and protein of this study was 70.25 and 21.08%, and they were lower than the result study of Williams (2007), which respectively amounted to 72.90 and 21.90%. However the fat in this study was higher than its study (5.62% and 4.70%).

Table 1. Nutrition content of meat of five and eleven month old garut lamb.

Variables	5 month	11 month	Average
Moisture	69.17 ± 3.17	71.33 ± 4.09	70.25 ± 3.63
Ash	1.88 ± 1.63	1.13 ± 0.19	1.50 ± 0.91
Protein	20.19 ± 2.07	21.9 ± 0.68	21.08 ± 1.37
Fat	5.74 ± 1.12	5.50 ± 3.60	5.62 ± 2.36
Carbohydrate	0.05 ± 0.01	0.05 ± 0.01	0.05 ± 0.01

The fatty acid composition of five month old to eleven month old garut lamb which fed by a diet containing mungbean waste was not significant different. Saturated fatty acid (SFA) which identified by this study from the highest to the lowest were palmitic acid (C16:0), stearic acid (C18:0), myristic acid (C14:0), lauric acid (C12:0), caprylic acid (C8:0), and capric acid (C10:0). Unsaturated fatty acid (USFA) which identified by this study from the highest to the lowest were oleic acid (C18:1), linoleic acid (C18:2) and linolenat acid (C18:3) (Figure 1).

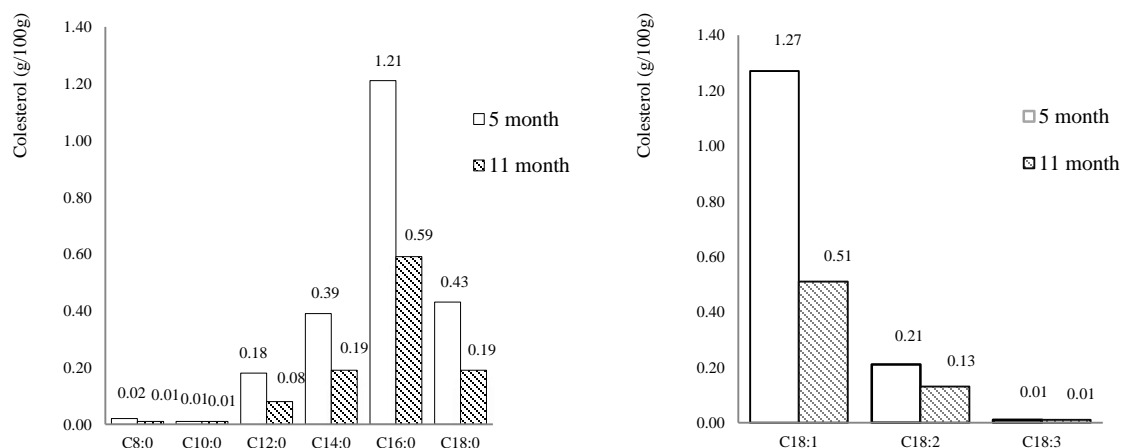


Figure 1. Fatty acid composition of garut lamb meat at different ages

Average of SFA total (palmitic acid, stearic acid, myristic acid, lauric acid, caprylic acid, and capric acid) from five month old and eleven month old lamb meat was 1.485 g/100 g lean meat and the average of USFA (oleic acid, linoleic acid, and linolenat acid) was 1.070 g/100 g lean meat. This result was lower than Williams 2007 that found out the average of SFA was 1.730 g/100 g lean meat and USFA was 2.388 g/100 g lean meat. The total of SFA was higher than USFA in garut lamb.

The age of lamb did not give any effect on lamb meat cholesterol. Average of garut lamb meat cholesterol in five month old and eleven month old lamb was 65.47 ± 22.25 and 71.30 ± 29.90 mg/100 g (the average was 68.38 ± 26.07 mg/100 g) (Figure 2). The cholesterol in garut lamb was higher than the result from William (2007) that found out the average was 66.00 mg/100 g lean meat.

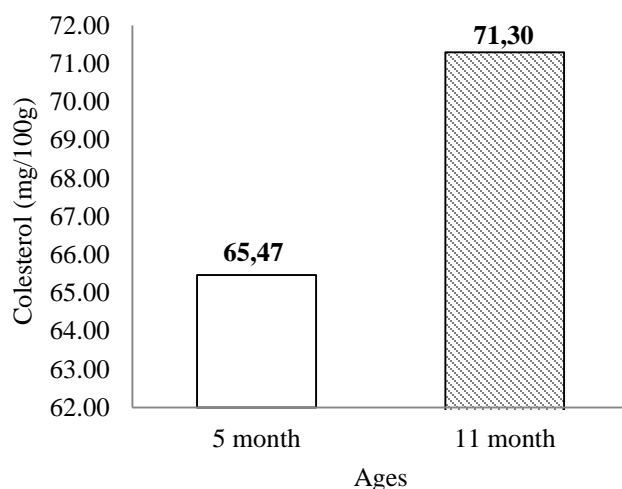


Figure 2. Cholesterol content of garut lamb meat at different ages.

Conclusion

It was concluded that there were no significant effect of different ages on nutrition and cholesterol content and fatty acid composition of garut lamb meat which fed by a diet containing mungbean sprouts waste. The total of SFA was higher than USFA in garut lamb meat.

References

- Purbowati E, E. Baliarti, S.p.S. Budhi, and W. Lestariana, 2005. Fatty acid profile of local malelambreadin in the rural area sonslaughter weight and location of different muscle. *Buletin Peternakan* vol. 29 (2) : 62 – 70.
- Romans, J.R., W.J. Costello, C.W. Carlson, M.L. Greaser dan K.W. Jones, 1994. *The Meat We Eat*. Interstate Publishers, Inc. Danville, Illinois.
- Sunarlim R. and Setiyanto, H., 2005. Cutting of Carcass Male Kacang Goat and Native Sheepon Composition of Physical Carcass, Physical Characteristic and Nutritious Value of Meat. *Prosiding Seminar Nasional Teknologi Peternakan dan Veteriner 2005*. Pp. 666-673
- Forrest, C.J, E.D. Aberle, H.B. Hedrick, M.D. Judge and R.A. Merkel, 2001. *Principle of Meat Science*. W.H. Freeman and Co. San Francisco. p. 4127.
- Salvatori G , L. Pantaleo, C. Di Cesare, G. Maiorano, F. Filetti, G. Oriani, 2004. Fatty acid composition and cholesterol content of muscles as related to genotype and vitamin E treatment in crossbred lambs. *Meat Science*, Volume 67(1) : 45-55. doi:10.1016/j.meatsci.2003.09.004
- Williams, P., 2007. Nutritional composition of red meat. *Nutrition & Dietetics* 64(Suppl. 4): S113–S119 doi: 10.1111/j.1747-0080.2007.00197.x

Some functional properties of beef liver protein concentrates

Al Awwaly^{*1}, K.U., S. Triatmojo², W.T. Artama² & Y. Erwanto²

¹ University of Brawijaya, Malang, 65145, Indonesia, ² Gadjah Mada University, Yogyakarta 55281, Indonesia

Abstract

This research was aimed to examine the functional properties of beef liver protein concentrates. The extraction was conducted as a function of pH and time. The pI method was applied in the purification of proteins from beef livers. Protein content of the beef liver was 68.69%. The functional properties of the beef liver protein concentrates were compared to those of some commercial ingredients such as whey protein concentrates and casein. Protein from beef liver exhibited better foaming properties than casein. Whey proteins exhibited the highest foam stability. The use of by-product proteins appears to be an interesting opportunity to obtain added value slaughterhouse by-products for economic and environmental reasons.

Keywords: beef liver protein, functional properties, foaming ability and stability.

**Corresponding author: aak_umam@ub.ac.id or aak_umam@yahoo.com*

Introduction

The appearance of “mad cow” disease, also called bovine spongiform encephalopathy (BSE) has given a negative brand image of offal to consumers and has strongly restricted its use in pet food. Cholesterol content in the offal is also restricted in the consumption for health reasons. The offal is also considered as place for accumulation of pesticide, antibiotic and drug residues, toxic chemicals and heavy metals contamination from the environment. It is therefore necessary to find new ways to obtain added value of slaughterhouse by-products for economic and environmental reasons. For example, these by-products exhibit a high protein content, between 15 and 20% (w/w), with many essential nutrients such as amino acids, minerals, vitamins and fatty acids (Liu, 2002). Some of them could also present interesting functional properties, but these properties have generally not yet been explored. From the animal and by-product protein sources, Zouari et al. (2011), Oshodi & Ojokan (1997), and Del Hoyo et al. (2008) studied functional properties of protein fraction from turkey liver and bovine plasma protein concentrate, respectively. Selmane et al. (2008; 2010; 2011) reported the extraction, production and functional properties of beef lung protein concentrates. With regard to the functional properties of slaughterhouse by-products, Boles et al. (2000) evaluated the effects of meat proteins on the texture of a finely comminuted sausage product. Conti-Silva et al. (2011) reported the sensory acceptability of raw and extruded bovine rumen protein in processed meat products.

The aim of this research was to examine the functional properties of beef liver protein concentrates. Four functional properties, namely emulsifying and foaming properties, water and oil holding capacities, of the resulting concentrates were compared to those of commercial ingredients from milk (whey protein concentrates and casein).

Material and Methods

Protein Extraction

Beef liver protein concentrate was extracted by alkali according to Selmane et al. (2008). All chemicals used in this research were pro-analysis grade. Proteins obtained after precipitation in the form of a paste were frozen at -20°C and dried using a microwave dryer. The protein contents of both the raw materials and the final powder were determined by using Kjeldahl method. As in the standard method, protein content was deduced from nitrogen content by multiplying the nitrogen mass fraction 6.25. Experiments were done in triplicates.

Functional properties of beef liver protein concentrate

The foaming ability and emulsifying activity (EA) were measured using the method described by Selmane et al. (2008; 2010). The emulsifying activity (EA) was determined from the turbidity of these emulsions, estimated by measuring the absorbance at 500 nm using a UV– Vis spectrophotometer (Cole Palmer). Oil holding capacity (OHC) and water holding capacity (WHC) of the protein concentrates were determined using method described by Subagio (2006). Measurements were done in triplicates.

Result and Discussion

Beef Liver Analyses

The chemical composition of beef liver in term of moisture, protein, lipid, ash and carbohydrate used in this research were listed in Table 1. Table 1 also showed that beef liver constitute the most interesting by-products in terms of recoverable proteins/DM ratio with 68.69±0.66%. This protein content of beef liver suggested that the beef liver can be a source of protein concentrate.

Table 1. Chemical composition of beef liver.^a

Components	Amount ^c
Moisture (%)	70.44 ± 3.21
Protein (%)	20.29 ± 2.03
Lipid (%)	3.95 ± 1.09
Ash (%)	1.68 ± 0.15
Carbohydrate (%) ^b	3.64 ± 0.39
Protein/DM (%)	68.69 ± 0.66

^a calculated on wet basis.

^b calculated using by difference from moisture, protein, lipid and ash.

^c Mean ± SD.

Functional properties of the beef liver protein concentrates

The functional properties of the beef liver protein concentrates obtained in this work were measured for foaming ability (FA) and foaming stability (FS), emulsifying activity (EA) and emulsifying stability (ES), oil and water holding capacity. This functional properties of the beef liver protein concentrates were summarized in Table 2.

Table 2. Functional properties of protein extract from beef liver.

Protein types	Beef liver	Whey protein	Casein
FA (%)	54.6 ± 3	74.4 ± 2	48.5 ± 4
FS (min)	46 ± 2	60 ± 4	18 ± 2
EA	0.52 ± 0.01	0.44 ± 0.03	0.58 ± 0.02
ES (min)	17 ± 4	35 ± 2	16 ± 3
WHC (ml/g)	1.89 ± 0.02	2.16 ± 0.02	1.96 ± 0.01
OHC (ml/g)	6.28 ± 0.03	6.58 ± 0.04	6.42 ± 0.02

Table 2 showed that, except for whey proteins, beef liver proteins exhibit the highest foaming ability. Protein from beef liver exhibited better foaming properties than casein. Whey proteins, as expected, exhibited the highest foam stability, which justifies their wide use as a foaming agent in the food industry. Another protein showed FS values at least 1.5 – 3 times lower. It is possible that this behavior is overshadowed by the high surface-activity of proteins. Another possibility is a stabilizing mechanism involving proteins and protein coated fat droplets, as observed in whipped cream (Selmane et al., 2008). Table 2 showed that caseins were the best emulsifying agents among the selected commercial ingredients, therefore, beef liver proteins exhibited higher EA values than whey protein concentrates. Beef liver proteins exhibited the ES values similar to casein. Such a behavior is not surprising: the low ES values of good emulsifying agents mean only that coalescence is more likely to occur rapidly when the initial turbidity A_o is high because of higher interfacial area and higher droplet number. WHC of the protein concentrate from beef liver was high (1.89 ± 0.02 ml/g), as shown in Table 2. This value was lower than that of whey protein concentrate and casein. The lower WHC suggested the presence of a large proportion of hydrophobic as compared to hydrophilic groups on the surface of protein molecules. Interestingly, OHC of the proteins was also lower, at 6.28 ± 0.03 ml/g.

Conclusion

The beef liver protein concentrates had good functional properties, such as foaming capacity, emulsifying capacity, water and oil holding capacity. Protein from beef liver exhibited better foaming properties than casein. Whey proteins exhibited the highest foam and emulsion stability, and also for water and oil holding capacity.

References

- Boles, J. A., B. M. Rathgeber and P. J. Shand, 2000. Recovery of proteins from beef bone and the functionality of these proteins in sausage batters. *Meat Sci.* 55, 223–231.
- Conti-Silva, A.C., M.E.M.P. Silva, and J.A.G. Areas, 2011. Sensory acceptability of raw and extruded bovine rumen protein in processed meat products. *Meat Sci.* 88:652-656.
- Del Hoyo, P., M. Rendueles and M. Diaz, 2008. Effect of processing on functional properties of animal blood plasma. *Meat Sci.* 78:522-528.
- Liu, D. C., 2002. Better utilization of by-products from the meat industry. <http://www.agnet.org/library/eb/515/>.
- Oshodi, A.A., and E.O. Ojokan, 1997. Effect of salts on some of the functional properties of bovine plasma protein concentrate. *Food Chem.* 59 (3):333-338.

- Selmane, D., V. Christophe and D. Gholamreza, 2008. Extraction of proteins from slaughterhouse by-products: Influence of operating conditions on functional properties. *Meat Sci.* 79, 640–647.
- Selmane, D., V. Christophe and D. Gholamreza, 2010. Production and functional properties of beef lung protein concentrates. *Meat Sci.* 84:315-322.
- Selmane, D., V. Christophe and D. Gholamreza, 2011. Emulsification properties of proteins extracted from beef lungs in the presence of xanthan gum using a continuous rotor/stator system. *LWT Food Sci. Technol.* 44: 1179-1188.
- Subagio, A., 2006. Characterization of hyacinth bean (*Lablab purpureus* (L.) sweet) seeds from Indonesia and their protein isolate. *Food Chem.* 95:65-70.
- Zouari, N., N. Fakhfakh, W.B. Amara-Dali, M. Sellami, L. Msaddak and M.A. Ayadi, 2011. Turkey liver: physicochemical characteristics and functional properties of protein fractions. *Food Bioprod Process.* 89:142-148.

Meat quality assessment in goats subjected to conscious halal slaughter and slaughter following minimal anaesthesia

Sabow¹ A.B., A.Q. Sazili^{*1, 2, 5}, I. Zulkifli^{1, 2}, Y.M. Goh^{2, 3}, M.Z. Ab Kadir⁴ & K.D. Adeyemi¹

¹Department of Animal Science, Faculty of Agriculture, ²Institute of Tropical Agriculture, ³Department of Veterinary Preclinical Sciences, Faculty of Veterinary Medicine, ⁴Department of Electrical and Electronic Engineering, Faculty of Engineering, ⁵Halal Products Research Institute, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia.

Abstract

The study assessed the effect of conscious halal slaughter and slaughter following a minimal anaesthesia on quality of goat meat. Ten Boer cross bucks were divided into two groups and subjected to either halal slaughter without stunning (HS) or minimal anaesthesia prior to slaughter (AS). At pre-rigor, HS had significantly lower ($p < 0.05$) muscle pH and glycogen than AS. The drip loss of HS was significantly lower ($p < 0.05$) than that of AS. However, no significant difference was observed in the pH, glycogen content, MFI, cooking loss and colour between the treatments at 1 and 7 d postmortem. In conclusion, goats slaughtered under fully consciousness and under minimal anaesthesia did not differ in physical and chemical quality of meat during the first seven days postmortem.

Keywords: goat, meat quality, traditional halal slaughter, minimal anaesthesia

Introduction

Minimising stress at slaughter improves productivity, quality and profitability. The welfare of animals during slaughter is protected by the 1958 Humane Slaughter Act. Section of this act makes it compulsory for animals to be stunned before exsanguination to ensure that animals are unconscious to prevent unnecessary suffering and improve meat quality but gives exemption for religious slaughter (College of Law, 2011). Under traditional religious slaughter without stunning, animals must be executed by a throat cut so as to bring the animal to a quick death without agony, through severance of the carotid arteries, jugular veins, trachea and esophagus permitting a rapid and complete bleeding. Although some investigations have been conducted on the efficacy of different slaughter methods on meat quality, most information originates from research in conventional slaughter methods with limited comparison to specifically halal slaughter method. This was due to the limited access to religious slaughter without stunning in most developed countries due to legal and welfare reasons. Animal subjected to minimal anaesthesia model has been identified as one of the best model in terms of animal welfare to study noxious stimulation associated with neck cut slaughter (Gibson et al., 2009). Pain comprises of both sensory and affective components. The minimally anaesthetised method would allow this experiment to study the effects associated only with sensory pain during slaughter, versus both sensory and affective pain when animals are slaughtered fully conscious without any

form of stunning (i.e., halal slaughter) on physical and chemical traits of meat quality. Thus, the effects of conscious halal slaughter and slaughter following minimal anaesthesia on meat quality were examined.

Materials and Methods

This study was conducted following the animal ethics guidelines of the Research Policy of Universiti Putra Malaysia. A total of 10 Boer cross bucks raised under identical conditions and fed the same diet, weighing between 23 and 25 kg were obtained from a commercial farm and transported to the Department of Animal Science abattoir, Faculty of Agriculture, Universiti Putra Malaysia. The goats were randomly assigned to two groups [halal slaughter without stunning (HS) and slaughter following minimal anaesthesia(AS)]. In the religious method, the animals were humanely slaughtered according to the procedure outlined by Department of Standards Malaysia (2009). In the anaesthesia process, animals were anaesthetized using 5mg/kg propofol administered by rapid injection into cephalic vein and maintained with halothane in 100% oxygen (Gibson et al., 2009), slaughtered and subsequently bled. After evisceration and carcass dressing, approximately 20 g of the *Longissimus lumborum* (LL) muscle from the left hind limbs was collected and stored in a 4°C chiller for drip loss determination. Within 30 min postmortem, the right LL between the 6th and 7th lumbar vertebrae was removed, snap frozen in liquid nitrogen before being stored at -80°C and assigned for subsequent determination of pH (pre-rigor) and glycogen content, myofibrillar fragmentation index (MFI) at d 0. The remaining carcasses were then hung in the 4°C chiller and the subsequent sampling was carried out at specific periods (1 and 7 days). Glycogen content in the LL muscles was determined using EnzyChrom™ Glycogen Assay kit Cat# E2GN-100 (BioAssays, USA). Muscle pH, Myofibril fragmentation index (MFI) and colour were determined following the procedure of Nakayinsige et al. (2014). Water holding capacity of meat was measured in terms of drip and cooking losses by calculating a percentage of weight loss, relative to the initial weight (Honikel, 1998). The data obtained were analyzed using the GLM procedure of Statistical Analysis System (SAS) package Version 9.2 software with time as a repeated measure. Duncan multiple range test was used to separate means.

Results and Discussion

The effects of slaughter methods on physical and chemical traits of goat meat are presented in Table 1. Before the onset of rigor, the concentration of glycogen in the muscle of AS goats was significantly higher ($p < 0.05$) than the HS group. The lower glycogen content in the HS could be attributed to the stress during hoisting and restraining prior slaughtering. It is well known that even low level of stress due to pre-slaughter handling can decrease glycogen reserves (Jolley, 1990). These findings are in line with the report of Digre et al. (2011) which indicated that anaesthetised fish had higher muscle glycogen than those not anaesthetised. After rigor, the LL muscle glycogen content was not different for the two slaughter methods. The pre-rigor pH of muscles from AS was significantly ($P < 0.05$) higher than those from HS. However, no significant difference in pH was observed between the treatments at 1 and 7 d postmortem. These findings support the report of Vergara et al. (2005) in lambs, where muscle pH was not influenced by the slaughter method beyond 24 h postmortem. On 0, 1 and 7 d postmortem, results showed that HS

goats produced meat with 92.52, 104.77 and 123.07 MFI while, AS goats produced meat with 91.36, 106.53 and 125.08 MFI, respectively. However, the observed differences were not significant ($P>0.05$). Degradation of inter myofibril linkages occurs as meat ages (Nakyinsige et al., 2014). In this study, the MFI increased significantly ($P<0.05$) with increasing postmortem aging regardless of the slaughter method. There was significant ($P<0.05$) increase in drip loss in goats subjected to AS compared to HS. This may be due to the fast pH decline in AS goats compared to their HS counterparts. There was no significant difference ($P>0.05$) in cooking loss between the treatments throughout the postmortem storage. This could possibly be due to similarity in ultimate pH of HS and AS. Dalle Zotte et al. (1995) reported negative correlation between cooking loss and pHu. On 1 and 7 d postmortem, L* (lightness), a* value (redness), b*(yellowness) were not significantly influenced by slaughter method (Table 1).

In conclusion, the results obtained from this study show that the quality of goat meat from HS was comparable to that from AS. Thus, this study affirms that slaughtering goats following minimal anaesthesia did not affect meat quality compared to those slaughtered fully conscious.

Table 1. Physical and chemical traits of goat meat subjected to different slaughtering methods.

Parameter	Days postmortem	Treatment ¹	
		HS	AS
Glycogen (mg/g meat)	0	1.72 ^b ±0.02	2.23 ^a ±0.18
	1	0.60±0.03	0.71±0.04
	7	0.37±0.02	0.39±0.02
pH	0	6.74 ^b ±0.03	6.90 ^a ±0.04
	1	6.35±0.07	6.33±0.14
	7	6.19±0.15	6.19±0.14
MFI	0	91.52±0.61	91.36±0.65
	1	104.77±0.99	106.53±0.99
	7	123.07±2.26	125.10±2.92
Drip loss (%)	1	1.18 ^b ±0.05	1.61 ^a ±0.08
	7	3.11 ^b ±0.21	3.98 ^a ±0.11
Cooking loss (%)	1	29.11±0.78	28.35±1.23
	7	25.54±0.90	25.20±1.16
Lightness (L*)	1	30.30±1.22	31.58±0.91
	7	32.14±1.10	33.13±1.11
Redness (a*)	1	13.64±0.44	12.02±0.42
	7	11.62±0.37	11.20±0.50
Yellowness (b*)	1	12.09±0.47	11.63±0.43
	7	10.14±0.24	11.22±0.29

¹HS= Halal slaughter; AS= Slaughter following minimal anaesthesia. Least square means with different superscripts in the same row differ significantly at $P<0.05$.

References

- College of law, 2011. Michigan state university. United States Code annotated currentness. Title 7. Agriculture. Chapter48. Humane methods of livestock slaughter.<http://www.animallaw.info/statutes/stusfd7usca1901.htm>.
- Dalle Zotte, A., Parigi Bini, R., Xiccato, G. and S. Simionato, 1995. Proprieta tecnologiche e sensoriali della carne di coniglio. Riv di Coniglicoltura 32: 33-39.
- Department of Standards Malaysia. 2009. MS1500:2009 (1st revision) Halal food-production, preparation, handling and storage-General guideline pp. 1-13.
- Digre, H., Erikson, U., Skaret, J., Lea, P., Gallart-Jornet, L. and E. Misimi, 2011. Biochemical, physical and sensory quality of ice-stored Atlantic cod (*Gadus morhua*) as affected by pre-slaughter stress, percussion stunning and AQUI-S™ anaesthesia. Eur. Food Res. Technol. 233: 447-456.
- Gibson, T., Johnson, C., Murrell, J., Hulls, C., Mitchinson, S., Stafford, K., Johnstone, A., and D. Mellor, 2009. Electroencephalographic responses of halothane-anaesthetised calves to slaughter by ventral-neck incision without prior stunning. N. Z. Vet. J. 57: 77-83.
- Honikel, K.O., 1998. Reference methods for the assessment of physical characteristics of meat. Meat Sci. 49: 447-457.
- Jolley, P.D., 1990. Rabbit transport and its effects on meat quality. Appl. Anim. Behav. Sci. 28: 119-134.
- Nakyinsige, K., Sazili, A., Zulkifli, I., Goh, Y., Fatimah, A. and A. Sabow, 2014. Influence of gas stunning and halal slaughter (no stunning) on rabbits welfare indicators and meat quality. Meat Sci. 98: 701–708.
- Vergara, H., Linares, M., Berruga, M. and L. Gallego, 2005. Meat quality in suckling lambs: effect of pre-slaughter handling. Meat Sci. 69: 473-478.

Effect of dietary blend of canola oil and palm oil on fatty acid composition, color and antioxidant profile of *Biceps femoris* muscle in goats

Adeyemi^{1,4}K.D., A.Q. Sazili^{1,2*} A.B. Sabow¹, R. Karim⁴& M. Ismail¹

¹Department of Animal Science, Faculty of Agriculture, ²Halal Products Research Institute,³Department of Food Science and Technology, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia, ⁴Department of Animal Production, University of Ilorin, PMB 1515 Ilorin, Nigeria.

Abstract

This study examined the effect of blend of 80% canola oil and 20% palm oil (BCPO) on lipid composition, antioxidant status, color and oxidative stability of *Biceps femoris*(BF) muscle in goats. Twenty-four bucks were randomly assigned to diets containing 0, 4 or 8% BCPO, fed for 14 weeks and slaughtered. Diet had no effect on glutathione peroxidase, catalase and superoxide dismutase activities. Goats fed 4% BCPO had higher (P<0.05) CLA cis-9 trans-11 while 8% BCPO had higher C18:3n-3 compared to other treatments. The 4 and 8% BCPO meat had lower (P<0.05) C14:0 and C15:0 and higher C22:5n-3 than did control goats. Dietary BCPO improved α and γ -tocopherol, redness and oxidative stability but did not affect total carotenoids and δ -tocopherol. Dietary BCPO enhanced beneficial muscle lipids without compromising color and lipid stability.

Keywords: lipid oxidation, redness, antioxidants, fatty acids

*Corresponding author: awis@upm.edu.my. awisqurni@gmail.com

Introduction

The link between fatty acid (FA) profile of ruminant meat and incidence of chronic diseases substantiates efforts to modify its lipids (WHO, 2003). However, enhancing the unsaturated fatty acids of red meat can cause lipid oxidation which could impart negatively on the eating and keeping quality of the meat (Nute et al., 2007; Olorunsanya et al., 2012). Thus, enhancing the unsaturated FA in red meat necessitates concomitant enhancement of antioxidants to guard against lipid oxidation. Although dietary supplementation of vitamin E provides a practicable alternative to prevent lipid oxidation, the extra cost of supplementation may not be compensated for by the price of ruminant products (Lauridsen et al., 1999). Supplementing PUFA and antioxidant-rich oils to animals could be a valuable means of preventing lipid oxidation and enhancing PUFA and antioxidants in human diet (Kang et al., 2001). Both palm oil and canola oil are rich in antioxidants and their FA profile (Chawla & Saxena, 2013; Ghazani et al., 2014) has been espoused. Thus, we propose that a blend of palm oil and canola oil will enhance beneficial muscle fatty acids and prevent lipid oxidation. The objective of this study was to assess the effect of blend of 80% canola

oil and 20% palm oil (BCPO) on lipid profile, antioxidant status and color stability of *Biceps femoris* muscle in goats.

Materials and methods

Twenty four Boer goats (4-5 months old, initial body weight of 20.54 ± 0.5 kg) were randomly assigned to diets containing 0, 4, 8% BCPO and fed for 14 weeks following 2 weeks of adaptation. The chemical and fatty acid composition of the diet is shown in Table 1. The animals were slaughtered in a commercial abattoir. All analyses were conducted on *Biceps femoris* muscle. Muscle fatty acid was determined by the method of Folch et al. (1957). Tocopherol and carotenoids were determined by the method of Kamal-eldinet al. (2000) and Okonkwo, (2009) respectively. The experiment followed a completely randomized design. Data obtained for fatty acids were analyzed by GLM procedure of SAS. Data obtained for antioxidants and lipid oxidation was subjected to repeated analysis of variance. Means were separated using Tukey HSD test at significant level of $P < 0.05$.

Results and Discussion

The effect of dietary BCPO on FA composition of *Biceps femoris* (BF) muscle in goats is shown in Table 1. Proportion of C14:0 and C15:0 decreased ($P < 0.05$) as the level of BCPO increased. The decrease in C14:0 could be due to reduction or inhibition of mRNA abundance and activity of lipogenic enzymes such as fatty acid synthase and acetyl coA carboxylase required for the synthesis of medium chain FA (Kim et al., 2007). The C15:0 is one of the odd chain FA derived from rumen microorganisms (Bessa et al., 2007) thus, its depression could be due to the effect of unprotected fat on rumen microbial ecology and metabolism by reducing either the proportion of odd chain FA in microbial biomass or the flow of microbial biomass to the duodenum (Bessa et al., 2007). The higher concentration of C18:1n-9 in meat of goats fed 4 and 8% BCPO could be due to increase in dietary intake of C18:1n-9 as observed in the feeding trial. This finding is consistent with those of Otto et al (2014) who observed that incremental level of degummed canola oil enhanced the concentration of C18:1n-9 in bovine milk. The significant increase in CLA cis-12trans-10 in 8% BCPO might be due to the biohydrogenation of C18:2n-6 and C18:3n-3 (Kim et al., 2007). As observed in *in vitro* rumen fermentation trial (Adeyemi et al., 2015), increasing level of BCPO caused a linear decrease in the biohydrogenation of C18:2n-6 and C18:3n-3. The increase in C18:3n-3 as dietary BCPO increased in diet was likely a result of greater intake and lower extent of biohydrogenation of C18:3n-3. The increase in the proportion of C22:5n-3 in oil fed goats reflects the increase in muscle C18:3n-3 suggesting *in vivo* elongation and desaturation of C18:3n-3.

Table 1. Chemical and fatty acid composition of dietary treatments.

	Levels of BCPO ¹ (%)		
	0	4	8
<i>Chemical composition, % DM</i>			
Dry matter, as fed basis	67.70	67.90	68.07
Crude Protein,	14.27	14.37	14.39
Ether extract	2.30	6.35	11.11
Organic matter	93.16	93.42	93.55
Crude fibre	28.48	27.64	26.81
Nitrogen free extract	16.56	13.97	12.45
ADF	35.04	33.28	32.52
NDF	63.52	62.67	62.06
Metabolizable energy, MJ/Kg DM	11.59	11.61	11.62
Ca	1.02	1.05	1.04
P	0.52	0.54	0.54
<i>Fatty acids (g/kg)</i>			
C12:0, lauric	0.01	0.03	0.04
C14:0, myristic	0.53	0.51	0.51
C16:0, palmitic	2.79	5.98	7.78
C16:1, palmitoleic	0.08	0.11	0.15
C18:0, stearic	0.56	1.12	1.43
C18:1n-9, oleic	3.82	14.87	25.23
C18:2 ω -6, linoleic	7.05	11.87	13.07
C18:3 ω -3, linolenic	1.06	2.61	4.23
Total FA	15.83	37.09	52.27
Σ SFA	3.89	7.79	9.79
Σ PUFA	12.00	29.31	42.52
Σ UFA	8.11	14.49	16.19
ω -6: ω -3	6.65	4.54	3.08
<i>Antioxidant (mg/kg)</i>			
Total carotenoid	14.81	16.71	19.86
α -tocopherol	101.12	112.47	123.21
γ -tocopherol	10.22	34.55	49.17
δ -tocopherol	1.21	3.45	5.93

¹blend of 80% canola oil and 20% palm oil.

Table 2. Effect of diet on fatty acid composition (% of total FA) in *Biceps femoris* muscle in goats.

Parameter	Level of BCPO ¹ (%)			P value
	0	4	8	
C14:0	2.57 ^a	1.97 ^b	1.29 ^c	0.048
C15:0	3.19 ^a	2.50 ^b	1.89 ^c	0.035
C16:0	22.69	20.60	20.06	0.068
C16:1n-7	2.00	1.89	1.97	0.518
C18:0	16.97	16.83	17.35	0.206
C18:1n-9	20.0 ^b	23.11 ^a	23.1 ^a	0.008
C18:1t11vaccenic	2.0	1.9	2.1	0.361
CLA c9t11	1.1	1.3	1.1	0.118
CLA c12t10	1.0	1.2	1.7	0.122
C18:2n-6	16.4	15.2	13.6	0.065
C18:3n-3	1.0 ^b	1.1 ^b	1.9 ^a	0.039
C20:4n-6	7.0	6.9	6.3	0.079
C20:5n-3	2.0	1.8	2.6	0.631
C22:5n-3	1.2 ^b	2.2 ^a	2.4 ^a	0.040
C22:6n-3	1.1 ^b	1.5 ^{ab}	1.9 ^a	0.031
∑SFA	45.4	41.9	40.6	0.063
∑MUFA	24.0 ^b	27.69 ^a	27.3 ^a	0.016
∑PUFA	30.4 ^b	31.2 ^a	32.2 ^a	0.043
∑ω-3	5.3 ^c	6.6 ^b	8.8 ^a	0.010
∑ω-6	23.4 ^a	22.2 ^a	19.9 ^b	0.026
ω-6:ω-3	4.4 ^c	3.3 ^b	2.7 ^a	0.008
UFA:SFA	1.2	1.4	1.5	0.089
PUFA:SFA	0.7 ^a	0.8 ^a	0.8 ^a	0.106
Total FA (mg/g of muscle)	3.6 ^a	3.6 ^a	3.5 ^a	0.791

^{a,b,c} means having different superscript along the same row are significantly different (P<0.05). ¹Blend of 80% canola oil and 20% palm oil.

The antioxidant status of *Biceps femoris* muscle was influenced by dietary treatments (Table 2). Animals cannot synthesize carotenoid and tocopherol hence must be supplied in the diet. Thus, their presence in body tissues indicates dietary availability. The higher (P<0.05) α- and γ-tocopherol and the tendency (P=0.07) for total carotenoid and δ-tocopherol to increase in *Biceps femoris* muscle of goats fed 4 and 8% BCPO compared to the control treatments could be due to the increase in dietary fat in the diets. Both tocopherol and carotenoids are fat soluble vitamins. Thus, increase in dietary fat might have aided the deposition and absorption of the antioxidant vitamins. In line with the present observation, Lauridsen et al. (1999) observed that the *psaos major* and *longissimus dorsi* muscle of pig fed 6% rapeseed oil had higher α-tocopherol content compared with those fed basal diet. This shows that supplementing vitamin E rich oils in diets of livestock can elevate muscle vitamin E. This provides a feasible alternative to prolong the shelf life of meat without dietary vitamin E supplementation whose extra cost to farmers may not be compensated for by the higher price of ruminant products. The decline in the concentration of all antioxidant vitamins as postmortem storage progressed was expected and it reflects breakdown of antioxidant defense system.

The induction of antioxidant enzymes in response to oxidative stress has been established (Renner et al., 1996; 1999). The similarity in the concentration of catalase, superoxide dismutase and glutathione peroxidase among the treatments may be due to the α and γ -tocopherol content in the muscle. This finding suggests that the increment in muscle α and γ -tocopherol in goats fed 4 and 8% BCPO compensated well for the increase in the readily oxidized n-3 PUFA in the muscles. In contrast to the current observation, Renner et al. (1999) observed that dietary rapeseed oil and soybean oil compared with tallow raised GPX and catalase activity in *Pectoralis major* muscle of turkey. It could be inferred from the aforementioned citation that unsaturated fatty acids being unstable every so often elevate antioxidant enzyme activities. This substantiates the earlier assertion that the increase in α and γ -tocopherol in goats fed 4 and 8% BCPO prevented oxidative spoilage in spite of their higher PUFA contents compared with the control treatment. In agreement with the observed stability of antioxidant enzyme activities throughout the 7 d postmortem storage, Renner et al. (1996) reported similarity in the GPX and catalase activities in beef observed on d 1 and 8 postmortem. In contrast, Renner et al. (1999) observed a significant reduction in GPX and catalase activity and a non-significant reduction in SOD activity in the *Pectoralis major* muscle of turkey as storage progressed.

Table 3. Effect of diet and postmortem storage on antioxidants and TBARS value of *Biceps femoris* muscle in goats.

Parameter	Level of BCPO ¹ in %			Storage days		P value		
	0	4	8	0	7	D	D	DxT
Superoxide dismutase ²	2.55	2.55	2.76	2.65	2.46	0.649	0.984	0.366
Catalase ³	1652.9	1693.5	1615.2	1654.9	1667.2	0.671	0.658	0.449
Glutathione peroxidase ⁴	82.33	80.66	76.85	80.45a	75.86a	0.888	0.468	0.139
Total carotenoid (mg/kg)	0.24	0.26	0.29	0.30a	0.18b	0.142	0.030	0.099
α -tocopherol (mg/kg)	2.46 ^c	3.38 ^b	4.02 ^a	3.45 ^a	2.90 ^b	0.001	0.023	0.101
γ -tocopherol (mg/kg)	0.47 ^c	0.77 ^b	0.98 ^a	1.02 ^a	0.80 ^b	0.001	0.008	0.243
δ -tocopherol (mg/kg)	0.06	0.08	0.08	0.07 ^a	0.03 ^b	0.196	0.010	0.311
TBARS (mg MDA/ kg)	0.75	0.71	0.65	0.70 ^b	0.84 ^a	0.054	0.044	0.146

a, b c d means having different superscript along the same row for each factor are significantly different ($p < 0.05$). ¹ = blend of 80% canola oil and 20% red palm oil. ²SOD activity is expressed as Units 50% mg protein. ³Catalase activity is expressed as nmol.H₂O₂/min/mg protein. ⁴GPX activity is expressed as nmoles NADPH oxidized /min/mg protein. D= Diet; S=storage D*S= interaction between diet and storage.

The color coordinates of BF muscle is shown in Table 3. Color plays important role in meat quality assessment due to its indispensable role in the selection and identification of meat (Adeyemi & Sazili 2014). The higher a* in *Biceps femoris* of goats fed 4 and 8% BCPO relative to the control diet could be due to the higher α and γ -tocopherol in the muscle which reduced lipid oxidation (Table 2) and/or myoglobin oxidation. This observation is in tandem with the report of Jensen et al. (1998) who reported that the *longissimus dorsi* muscle of pig fed rapeseed oil had higher vitamin E and redness compared to those fed control diet. Zakrys et al. (2008) observed that changes in a* values are related to lipid oxidation and are strongly correlated with the TBARS values. The increased in L* value as storage progressed is in line with the report of Sabow et al. (2015) and Bressan et al. (2011) which showed that ageing improved lightness of chevon and beef respectively. Zakrys et al. (2008) showed that instrumental a* value had a negative correlation while L* value had a positive correlation with storage days postmortem, suggesting that samples became less red and lighter over the duration of storage.

Table 4. Effect of dietary BCPO and postmortem ageing on color coordinates of *Biceps femoris* muscle in goats.

Parameter	Level of BCPO ¹ (%)			Storage days		P value		
	0	4	8	1	7	D	T	DxT
Lightness (L*)	34.44 ^a	32.23 ^b	32.66 ^b	31.56 ^a	35.27 ^b	0.1384	0.0348	0.0164
Redness (a*)	11.33 ^c	13.46 ^b	14.82 ^a	13.21	10.80	<.0001	<.0001	0.0369
Yellowness (b*)	13.91	13.38	11.98	11.86 ^c	15.09 ^a	0.0524	0.0286	0.1067
Chroma	12.89 ^b	14.38 ^{ab}	15.18 ^a	18.66	15.42	0.034	<.0001	0.4325
Enhanced redness	1.16 ^b	1.55 ^a	1.63 ^a	1.67 ^a	1.19 ^b	0.0001	0.065	0.1044

a, b c d means having different superscript along the same row for each factor are significantly different (P< 0.05).

¹ = blend of 80% canola oil and 20% red palm oil. D= Diet; S=storage D*S= interaction between diet and storage.

Conclusion

Dietary supplementation of blend of 80% canola oil and 20% palm oil in goats' diet enhanced n-3 PUFA without compromising lipid and color stability of chevon.

References

- Adeyemi, K.D. & A.Q. Sazili, 2014. Efficacy of carcass electrical stimulation in meat quality enhancement: A review. *Asian-Austral. J. Anim. Sci.*27:447-456.
- Adeyemi, K.D., M. Ebrahimi, A.A. Samsudin, A. R. Alimon, R. Karim, S.A. Karsani, & A. Q. Sazili, 2015. Influence of carotino oil on in vitro rumen fermentation, metabolism and apparent biohydrogenation of fatty acids. *Anim. Sci. J.* 86:270-278.
- Bessa R.J., S.P. Alves, E. Jerónimo, C.M. Alfaia, J.A. Prates, & J. Santos-Silva, 2007. Effect of lipid supplements on ruminal biohydrogenation intermediates and muscle fatty acids in lambs. *Eur. J. Lipid Sci.Tech.* 109:868-878.
- Bressan M.C., E.C. Rodrigues, L.V. Rossato, E.M. Ramos, & L.T. Gama, 2011. Physicochemical properties of meat from *Bostaurus* and *Bosindicus*. *Revista Brasileira De Zootecnia* 40:1250-1259.
- Chawla S. & S. Saxena, 2013. Red palm oil-health benefits and their molecular executors.*Int. J. Bioassay*2:1223-1231.
- Folch J., M. Lees & G. Sloane-Stanley, 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J. Bio. Chem.*226:497-509.
- Ghazani S.M., G. García-Llatas, & A.G. Marangoni, 2014. Micronutrient content of cold-pressed, hot-pressed, solvent extracted and RBD canola oil: Implications for nutrition and quality. *Eur. J. Lipid Sci.Tech.*116: 380-387.
- Jensen C., M. Flensted-Jensen M., L. Skibsted, & G. Bertelsen, 1998. Effects of dietary rape seed oil, copper (II) sulphate and vitamin E on drip loss, colour and lipid oxidation of chilled pork chops packed in atmospheric air or in a high oxygen atmosphere.*Meat Sci.*50: 211-221.
- Kamal-Eldin A., J. Frank, A. Razdan, S. Tengblad, S. Basu& B. Vessby, 2000. Effects of dietary phenolic compounds on tocopherol, cholesterol, and fatty acids in rats.*Lipids* 35:427-435.
- Kang K.R., G. Cherian, & J.S. Sim, 2001. Dietary palm oil alters the lipid stability of polyunsaturated fatty acid-modified poultry products. *Poult. Sci.*80:228-234.

- Kim S., A. Adesogan A., L. Badinga. & C. Staples, 2007. Effects of dietary n-6: n-3 fatty acid ratio on feed intake, digestibility, and fatty acid profiles of the ruminal contents, liver, and muscle of growing lambs. *J. Anim. Sci.* 85:706-716.
- Lauridsen C., J.H. Nielsen, P. Henckel, & M.T. Sørensen, 1999. Antioxidative and oxidative status in muscles of pigs fed rapeseed oil, vitamin E and copper. *J. Anim. Sci.*77:105-115.
- Nute G., R. Richardson, J. Wood, S. Hughes, R. Wilkinson, S. Cooper, & L. Sinclair L. 2007. Effect of dietary oil source on the flavour and the colour and lipid stability of lamb meat. *Meat Sci.* 77:547-555.
- Okonkwo J, 2009. Effects of breed and storage duration on the beta-carotene content of egg yolk. *Pakistan J. Nutr.* 8:1629-1630.
- Olorunsanya A.O., K.D Adeyemi, & I. A. Babatunde, 2012. Effect of Bamboo (*Bambusavalgaris*) and Elephant grass (*Pennisetum purpureum*) leaf extracts on oxidative stability of cooked and raw broiler meat. *J. Agr. Res.Dev.*
- Otto, J., Malau-Aduli, B., Nichols, P., & A.E.O. Malau-Aduli. 2014, Influence of supplementing pasture-based primiparous holstein-friesian dairy cows with crude degummed canola oil on milk fatty acid composition. *J. Nutr. Therap.* 3:55-66
- Renerre, M., F. Dumont, P. Gatellier, 1996, Antioxidant enzyme activities in beef in relation to oxidation of lipid and myoglobin. *Meat Sci.* 43:111-121.
- Renerre, M., K.Poncet, Y.Mercier, P. Gatellier, B. Métro, 1999. Influence of dietary fat and vitamin E on antioxidant status of muscles of turkey. *J. Agr. Food Chem.*47:237-244.
- Sabow A.B., A.Q. Sazili, I. Zulkifli, Y.M. Goh, M.Z. Ab Kadir M.Z. & K.D. Adeyemi, 2015. Physico-chemical characteristics of *longissimus lumborum* muscle in goats subjected to halal slaughter and anaesthesia (halothane) pre-slaughter. *Anim. Sci. J.* doi:10.1111/asj.12385.
- WHO, 2003. Diet, nutrition and the prevention of chronic diseases.WHO Technical Report Series916:1-60.
- Zakrys P., S.Hogan, M. O'sullivan, P. Allen, & J. Kerry, 2008. Effects of oxygen concentration on the sensory evaluation and quality indicators of beef muscle packed under modified atmosphere. *Meat Sci.*79:648-655.

Meat quality of crossbred fattening pigs sired by Pakchong 5 boars and commercial boars

Lertpatarakomol^{1,2}, R., C. Chaosap³, R. Sitthigripong², R. Limsupavanich², W. Champarat⁴, W. Thaenthane⁴ & K. Chaweewan⁴

¹Faculty of Veterinary Medicine, Mahanakorn University of Technology, ²Faculty of Agricultural Technology, ³Faculty of Industrial Education, King Mongkut's Institute of Technology Ladkrabang, ⁴Department of Livestock Development, Thailand

Abstract

This study evaluated the meat quality of crossbred fattening pigs sired by Pakchong 5 boar and 2 commercial breeding boars. Forty-eight weaning pigs (20 kg body weight) were distributed into 3 treatments with 8 gilts and 8 barrows in each. The experimental treatments consisted of crossbred pigs sired by Pakchong 5 boar (CP5), crossbred pigs sired by commercial boar 1 (CB1), and crossbred pigs sired by commercial boar 2 (CB2). All sires in this study were bred to hybrid sows (Large White x Landrace). The animals were fed with commercial diet and drinking water *ad libitum*. At slaughtering weight (103.54 ± 6.89 kg), all pigs were slaughtered and *Longissimus dorsi* (LD) muscle were collected to compare the physical characteristics of meat. Results showed that breeds had no effect ($P > 0.05$) on pH_{45min}, drip loss, thawing loss and Warner-Bratzler shear force. Crossbred pigs sired by Pakchong 5 boar had a higher value of pH_{24h} ($P < 0.05$) and lower percentage of cooking loss ($P < 0.05$) than crossbred pigs sired by commercial boars. CP5 and CB1 had significantly higher L* values (lighter, $P < 0.05$) than CB2, whereas CP5 and CB2 had significantly lower ($P < 0.05$) a* and b* (less red and less yellow) than CB1. However, the effect of sex and interaction between breed and sex had no significant influence on meat quality attributes ($P > 0.05$). LD from crossbred fattening pigs sired by Pakchong 5 boars was as tender as LD from those sired by commercial boars. But it could be much juicy due to its high ultimate pH and less cooking loss.

Keywords: Pakchong 5 boar, commercial boar, crossbred fattening pig, meat quality

Introduction

Meat quality has been raised as a focal demand amongst all people who are in pig production ranging from customer to researcher. A rise in the lean to fat ratio of pig carcasses have been set as one of the major goal for the swine industry (Alonso et al., 2009). Consequently, the development of body composition of pigs has been quickly taken through genetic selection, with some cases of higher variability among lines within breed than among breeds (Latorre et al., 2003a; Latorre et al., 2003b). The terminal sire can strongly influence pork quality, through it seems that a three-line crossbred pig has intermediate value of parents for meat quality and eating quality traits (Suzuki et al., 2003). Therefore, it is the necessity to study on new genetic lines to meet the market requirement because crossbreeding can be used to enhance lean growth without decreasing pork eating quality (Lo et al., 1992).

Pakchong 5 pigs, have been developed by DLD (Department of Livestock Development) Thailand, are synthetic terminal boar that was established from genetic combination of Duroc and Pietrain. Pakchong 5 is proposed to produce high growth rate and lean crossbred fattening pigs. Previous study has been reported the growth performance of Pakchong 5, but there is a little information about carcass quality and meat quality of those (Chaweewan et al., 2012). Therefore, the aim of this study was to evaluate the meat quality of three-line crossbred fattening pigs derived from Pakchong 5 terminal boars compared with three-line crossbred fattening pigs derived from commercial boars.

Materials and Methods

Animals and Samples Collection

Forty-eight weaning pigs (20 kg body weight) were distributed into 3 treatments with 8 gilts and 8 barrows in each. The experimental treatments consisted of crossbred pigs sired by Pakchong 5 boar (CP5), crossbred pigs sired by commercial boar 1 (CB1), and crossbred pigs sired by commercial boar 2 (CB2). All sires in this study were bred to hybrid sows (Large White x Landrace). The animals were fed with commercial diet and drinking water *ad libitum*. Routine medication, vaccination and husbandry practices were administered. At slaughtering weight (103.54 ± 6.89 kg body weight), all pigs were slaughtered. *Longissimusdorsi* (LD) muscle from left side of each carcass was collected and vacuum packed, then stored at -20°C until subsequent analyses. Physical characteristics, $\text{pH}_{45\text{min}}$, $\text{pH}_{24\text{h}}$ of meat sample were evaluated. After 48 hours post-mortem, meat color (Minolta Chromameter CR-300, Illuminant D65 2° observer), drip loss, thawing loss, cooking loss and Warner-Bratzler shear force (Wheeler et al. 2005) were measured.

Data Analysis

The data was analyzed using the General Linear Models (GLM) procedure in procedure of SAS (SAS Inst., Inc. Cary, NC) by using model as below. All means presented were generated using the LSMEANS option and separated with the PDIFF option.

$$Y_{ijk} = \mu + A_i + B_j + A_iB_j + \epsilon_{ijk}$$

- Y_{ijk} = the observed for sire line i and sex j
- μ = overall mean or grand mean
- A_i = breed effect ($i = 1, 2, 3$), where A_1 is the effect of CP5, A_2 is the effect of CB1 and A_3 is the effect of CB2
- B_j = sex effect ($j = 1, 2$), where B_1 is the effect of gilt and B_2 is the effect of barrow
- A_iB_j = interaction effect
- ϵ_{ijk} = the random error

Results and Discussions

The results of this study were summarized in Table 1. There were no difference ($P>0.05$) on pH_{45min} , drip loss, thawing loss and Warner-Bratzler shear force between breeds. Crossbred pigs sired by Pakchong 5 boar had a higher value of pH_{24h} ($P<0.05$) and lower percentage of cooking loss ($P<0.05$) than crossbred pigs sired by commercial boars. The results of this study are in agreement with the findings of Bulotienė and Jukna(2008) who revealed that pH at 48 h post-mortem was highly correlated ($P<0.01$) with water holding capacity and cooking loss. In this case, Huff-Lonergan and Lonergan (2005);Adzitey and Nurul (2011) explained that lactic acid was built up in the tissue leading to a reduction in pH during the conversion of muscle to meat. Partial denaturation of the myosin head at low pH is thought to be responsible for a large part of the shrinkage in myofibrillar lattice spacing. The result of this study showed that post-mortem pH declined was associated by a significant decrease in lightness (L^*) among different sire lines. However, this results were in contrast with Huff-Lonergan et al., (2002);Bulotienė and Jukna (2008);Karamucki et al. (2013) who indicated that the reduction of pH resulted in the increasing of L^* value. In general, when pH declines rapidly, a partial denaturation of the protein myoglobin occurs. These will depreciate the color intensity of the meat. Under extreme conditions, denaturation of the myofibrillar proteins alters the color by allowing those structures to reflect more light (Adzitey & Nurul, 2011). In addition, CP5 and CB1 had significantly higher ($P<0.05$) L^* values (lighter) than CB2, whereas CP5 and CB2 had significantly lower ($P<0.05$) a^* and b^* (less red and less yellow) than CB1. The results indicated that meat with the highest L^* value had the highest b^* value which is consistent with the findings of Bulotienė & Jukna (2008). The effect of sex and interaction between breed and sex had no significant influence on meat quality attributes in this study($P>0.05$).

In conclusion, results of present study showed that LD from crossbred fattening pigs sired by Pakchong 5 boars was as tender as LD from those sired by commercial boars. But it could be much juicy due to its high ultimate pH and less cooking loss.

Table 1. Effect of breed and sex on physical characteristics of LD muscle of fattening pigs.

Items ²	Group ¹			Sex		SEM	P-value		
	CP5	CB1	CB2	Gilt	Barrow		Breed	Sex	Breed x Sex
pH_{45min}	6.19	6.13	6.21	6.16	6.19	0.04	0.69	0.74	0.68
pH_{24h}	6.06 ^c	5.71 ^b	5.56 ^a	5.77	5.78	0.02	<0.01	0.73	0.59
Meat color									
L^*	50.89 ^b	51.49 ^b	49.16 ^a	49.99	51.04	0.35	0.02	0.13	0.29
a^*	3.42 ^a	4.63 ^b	3.66 ^a	3.70	4.11	0.16	0.01	0.22	0.67
b^*	10.94 ^a	12.31 ^b	10.94 ^a	11.16	11.63	0.13	<0.01	0.08	0.92
DL, %	3.18	3.59	3.25	3.54	3.14	0.16	0.53	0.21	0.74
TL, %	8.84	8.67	7.79	8.38	8.48	0.29	0.29	0.87	0.53
CL, %	20.18 ^b	23.07 ^a	23.88 ^a	22.22	22.54	0.49	0.01	0.75	0.70
WBSF, kg	5.45	5.77	5.95	5.92	5.53	0.16	0.45	0.24	0.52

¹CP5 = crossbred pigs sired by Pakchong 5 boar, CB1 = crossbred pigs sired by commercial boar 1 and CB2 = crossbred pigs sired by commercial boar 2.

² pH_{45min} = pH at 45 min post-mortem, pH_{24h} = pH at 24 h post-mortem, L^* = lightness, a^* = redness, b^* = yellowness, DL = Drip loss, TL = Thawing loss, CL = Cooking loss, WBSF = Warner-Bratzler shear force.

^{a,b,c}Means in the same row with different superscripts are significantly different($P\leq 0.05$).

References

- Adzitey, F., and H. Nurul. 2011. Pale soft exudative (PSE) and dark firm dry (DFD) meats: causes and measures to reduce these incidences - a mini review. *J. Inter. Food Res.* 18: 11-20.
- Alonso, V., M. Campo Mdel, S. Espanol, P. Roncales, and J. A. Beltran. 2009. Effect of crossbreeding and gender on meat quality and fatty acid composition in pork. *Meat Sci.* 81: 209-217.
- Bulotienė, G., and V. Jukna. 2008. The influence of muscle fibre area on pork quality. *Veterinarija Ir Zootechnika. T. 42:* 34-37.
- Chaweewan, K., S. Nakavisut, V. Jumparat and V. Srisuriya. 2012. Genetic parameters of selection for economic traits over five generations of Pakchong 5 swine. Proceedings of the 15th AAAP Animal Science Congress, Thammasat University, Rangsit Campus, Thailand, AAAP: 1347-1351.
- Huff-Lonergan, E. T. J. Baas, M. Malek, J. C. Dekkers, K. Prusa and M. F. Rothschild. 2002. Correlations among selected pork quality traits. *J. Anim. Sci.* 80: 617-627.
- Huff-Lonergan, E., and S. M. Lonergan. 2005. Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. *Meat Sci.* 71: 194-204.
- Karamucki, T., M. Jakubowska, A. Rybarczyk, and J. Gardzielewska. 2013. The influence of myoglobin on the colour of minced pork loin. *Meat Sci.* 94: 234-238.
- Latorre, M. A., R. Lazaro, M. I. Gracia, M. Nieto, and G. G. Mateos. 2003a. Effect of sex and terminal sire genotype on performance, carcass characteristics, and meat quality of pigs slaughtered at 117 kg body weight. *Meat Sci.* 65: 1369-1377.
- Latorre, M. A., P. Medel, A. Fuentetaja, R. Lázaro, and G. G. Mateos. 2003b. Effect of gender, terminal sire line and age at slaughter on performance, carcass characteristics and meat quality of heavy pigs. *Anim. Sci.* 77: 33-45.
- Lo, L. L., D. G. McLaren, F. K. McKeith, R. L. Fernando, and J. Novakofski. 1992. Genetic analyses of growth, real-time ultrasound, carcass, and pork quality traits in Duroc and Landrace pigs: I. Breed effects. *J. Anim. Sci.* 70: 2373-2386.
- Suzuki, K., T. Shibata, H. Kadowaki, H. Abe, and T. Toyoshima. 2003. Meat quality comparison of Berkshire, Duroc and crossbred pigs sired by Berkshire and Duroc. *Meat Sci.* 64: 35-42.
- Wheeler, T. L., S. D. Shackelford, and M. Koohmaraie. 2005. Shear Force Procedures for Meat Tenderness Measurement. Online available: <http://www.ars.usda.gov/SP2UserFiles/Place/30400510/protocols/ShearForceProcedures.pdf>

Comparison muscle fiber size, sarcomere length and tenderness between chicken and duck meat

Khuenklang N.¹, R. Sitthigripong¹ & C. Chaosap²

¹Department of Animal Production Technology and Fisheries, Faculty of Agricultural Technology,²Department of Agricultural Education, Faculty of Industrial Education King Mongkut's Institute of Technology Ladkrabang, Bangkok 10520, Thailand

Abstract

The objective of this study was to investigate the effect animal types (chicken and duck) and cutting parts (breast and thigh) on muscle fiber size, sarcomere length and tenderness. Breast and thigh from 10 Cherry valley ducks and 10 Arbor acres broiler were used in this study. The results showed that muscle fiber diameter (MFD) of chicken meat was statistically longer than duck meat ($P < 0.01$). MFD of thigh meat was statistically longer than breast meat ($P < 0.05$). There was a statistically interaction between animal type and cutting part, which duck breast had longer MFD than chicken breast meat. Duck thigh had the same MFD as chicken thigh. Sarcomere length (SL) was not significant difference between chicken and duck. Thigh meat had longer SL the breast meat ($P < 0.05$). Chicken thigh and duck breast had the longest SL, followed by duck thigh and chicken breast ($P < 0.01$). Duck meat had higher shear force value (SF) than chicken meat ($P < 0.01$). There was a statistically interaction ($P < 0.01$) between animal type and cutting part. Which duck thigh had a higher SF than chicken thigh. Duck breast had the same SF as chicken breast. While chicken thigh showed the lowest SF.

Keyword: chicken, duck, muscle fiber size, sarcomere length, tenderness

Introduction

Consumption of poultry meat is currently growing, and duck production may be able to help meet the consumer demand. Duck meat production is based mainly on commercial hybrid of Pekin duck (Wawro et al., 2004). Currently, modern domestic Pekin duck is not different from modern broiler chicken in terms of gain and feed efficiency to the same live market weight. There are only a few studies carried out on meat characteristics in duck. Therefore, this study was aimed to investigate some meat characteristics of duck compared to chicken. As muscle fiber size and shortening sarcomere can significant influence the meat tenderness (Smith et al., 1993; Dunn et al., 2000). The objective of this study was to compare the microstructure and meat tenderness of breast and thigh in broiler chicken and duck to elucidate the differences between two species.

Materials and Methods

Animal

Ten Cherry valley ducks and 10 Arbor acres broiler chickens (5 males and 5 females in each animal types) aged 42 days were slaughtered using standard commercial procedure. Breast meat and thigh were removed from each carcass at approximately 6 h post-mortem for measuring muscle fiber diameter (Tuma et al., 1962), sarcomere length (Cross et al., 1981) and shear force (Boccard et al., 1981)

Statistical analysis

A general linear model (SAS Inst., Inc., Cary, NC) was used to evaluate the significant differences ($P < 0.05$) between animal types (chicken and duck) and between cutting parts (breast and thigh). The model included the effects of animal type, cutting part and animal type x cutting part. When significant differences ($P < 0.05$) were detected, the mean values were separated by the probability differences (PDIFF) option at a predetermined probability rate of 5%. The results are presented as least - square means (LSMs) for the groups, together with the standard errors of these LSMs. Pearson correlation coefficients were then evaluated to characterize the relationship between traits.

Results and Discussion

The results showed that muscle fiber diameter (MFD) of chicken meat was statistically longer than duck meat ($P < 0.01$). MFD of thigh meat was statistically longer than breast meat ($P < 0.05$). There was a statistically interaction between animal type and cutting part, which duck breast had longer MFD than chicken breast meat. Duck thigh had the same MFD as chicken thigh. Sarcomere length (SL) was not significant difference between chicken and duck. Thigh meat had longer SL the breast meat ($P < 0.05$). Chicken thigh and duck breast had the longest SL, followed by duck thigh and chicken breast ($P < 0.01$). Duck meat had higher shear force value (SF) than chicken meat ($P < 0.01$). There was a statistically interaction ($P < 0.01$) between animal type and cutting part. Which duck thigh had a higher SF than chicken thigh. Duck breast had the same SF as chicken breast. While chicken thigh showed the lowest SF. In agreement with this study, Ali et al. (2007a) reported that duck breast had higher shear force value than chicken breast ($P < 0.05$). Ali et al. (2007b) reported longer sarcomere length of duck breast (1.85 micron at 24 h post mortem) than in this study (1.10 micron at 6 h post mortem). This difference may be caused by difference in the shortening phase of muscle during rigor development. Thompson et al (1987) reported that sarcomere length (1.50 micron) at 24 h post mortem of chicken breast was longer than sarcomere length at 6 h post mortem (1.05 micron) in this study. Smith et al. (1993) reported that duckling *Pectoralis* contained approximately 16% white fibers, and chicken contained 100% white fibers. Duckling white fibers were significantly ($P < 0.05$) smaller than broiler white fibers, 1,809 versus 3,346 μm^2 , respectively. Duckling red fibers, averaged 301 μm^2 , were approximately six times smaller than white fibers.

Table 1. LS Means of muscle fiber diameter, sarcomere length and shear force value of breast and thigh in chicken and duck meat.

Effect	LSMeans		
	MFD (μm)	SL (μm)	SF (kg)
Type			
Chicken (n = 10)	72.00 ^a	1.07	3.34 ^b
Duck (n = 10)	56.27 ^b	1.08	5.25 ^a
P-value	<0.01	0.30	<0.01
Part			
Breast (n = 20)	61.71 ^b	1.06 ^b	4.13 ^b
Thigh (n = 20)	66.56 ^a	1.10 ^a	4.48 ^a
P-value	<0.05	<0.05	0.34
Type x Part			
Chicken Breast (n = 10)	79.05 ^a	1.01 ^c	3.88 ^b
Chicken Thigh (n = 10)	64.95 ^b	1.12 ^a	2.79 ^c
Duck Breast (n = 10)	44.37 ^c	1.10 ^a	4.35 ^b
Duck Thigh (n = 10)	68.17 ^b	1.07 ^b	6.14 ^a
P-value	<0.01	<0.01	<0.01

^{a, b, c} Least square means within a column within each effect with different superscripts are significantly different

¹MFD = muscle fiber diameter, SL = sarcomere length, SF = shear force value.

The relationship between traits in this study was presented in Table 2. In chicken, muscle fiber size negatively correlated with sarcomere length ($r = -0.66$, $P = 0.003$) but positively correlated with shear force value ($r = 0.60$, $P = 0.008$). Similar to chicken, muscle fiber size of duck had negative correlation with sarcomere length ($r = -0.38$, $P = 0.097$) while had positive correlation with shear force value ($r = 0.60$, $P = 0.005$). However, in pool data set from chicken and duck, muscle fiber diameter still had negative correlation with sarcomere length ($r = -0.49$, $P = 0.003$) but there was no significant correlation between muscle fiber size and shear force value ($r = 0.07$, $P = 0.678$). The significant correlation between sarcomere length and shear force value was only found in chicken data set ($r = -0.48$, $P = 0.033$). In agreement with this experiment, Dunn et al. (2000) reported a strong negative correlation between sarcomere length and shear force in Pectoralis major muscle of processed broilers air-chilled at 0 °C and -12 °C. Papinaho et al. (1996) reported that shear force had significantly negative correlation with fiber cross-sectional area ($r = -0.289$), and with fiber diameter ($r = -0.264$).

Table 2. The relationship between study traits (r , P-value).

Trait ¹	Chicken (n = 10)		Duck (n = 10)		All (n = 20)	
	SL	SF	SL	SF	SL	SF
MFD	-0.66 (0.003)	0.60 (0.008)	-0.38 (0.097)	0.60 (0.005)	-0.49 (0.002)	0.07 (0.687)
SL		-0.48 (0.033)		-0.35 (0.125)		-0.25 (0.125)

¹MFD = muscle fiber diameter, SL = sarcomere length, SF = shear force value

Conclusion

Animal type, cutting part and the interaction between them had significantly affected on muscle fiber size, sarcomere length and tenderness. Muscle fiber size had a negative correlation with sarcomere length while had a positive correlation with shear force. There was negative correlation between sarcomere length and shear force in chicken.

Acknowledgements

This work was supported by research grant from the Faculty of Industrial Education, King Mongkut's Institute of Technology Ladkrabang, Thailand and Ducking Co. Ltd.

References

- Ali, S. M., G. H. Kang, H. S. Yang, J. Y. Jeong and Y. H. Hwang, 2007a. A Comparison of meat characteristics between duck and chicken breast. *J. Anim. Sci.* 20: 1002-1006.
- Ali, S. M., H. S. Yang, J. Y. Jeong, S. H. Moon, Y. H. Hwang, G. B. Park and S.T. Joo, 2007b. Quality of duck breast and leg meat after chilling carcasses in water at 0, 10 or 20°C. *J. Anim. Sci.* 20: 1895-1900.
- Boccard, R., I. Buchter, M. Casteels, E. Cosentino, E. Dransfield and D.E Hood, 1981. Procedures for measuring meat quality characteristics in beef production experiments. *Lives. Prod. Sci.* 8: 385-397.
- Cross, H. R., R. L. West, and T.R. Dutson, 1981. Comparison of methods for measuring sarcomere length in beef Semitendinosus muscle. *Meat Sci.* 5(4): 261-266.
- Dunn, A. A., E. L. C. Tolland, D. J. Kilpatrick and N. F. S. Gault, 2000. Relationship between early post-mortem muscle pH and shortening-induced toughness in the Pectoralis major muscle of processed broilers air-chilled at 0°C and -12°C. *Br. Poult. Sci.* 41: 53-60.
- Jing, L., G. B. Chang, Q. Xu, Y. Y. Zhang, H. B. Zhang, Q. Zhou and G.H. Chen, 2009. Study on development rule of muscle fiber in different duck breeds. *Agri. Sci.* 10(5): 107.
- Papinaho, E. A., M. H. Ruusunen, K. Suuronen, D. L. Fletcher, 1996. Relationship between muscle biochemical and meat quality properties of early deboning broiler breast. *J. Appl. Poult. Res.* 5: 126-133.
- Smith, D. P., D. L. Fletcher, R. J. Buhr and R. S. Beyer, 1993. Pekin ducklings and broiler chicken Pectoralis muscle structure and composition. *Poult. Sci.* 72: 202-208.
- Thompson, L. D., D. M. Janky and S. A. Woodwar, 1987. Tenderness and physical characteristics of broiler breast fillets harvested at various times from post-mortem electrically stimulated carcasses. *Poult. Sci.* 66: 1158-1167.
- Tuma, H. J., J. H. Venable, P. R. Wuthier and R.L. Henrickson, 1962. Relationship of fiber diameter to tenderness and meatiness as influenced by Bovine age. *J. Anim. Sci.* 21: 33-36.
- Wawro, K., E. Wilkiewicz-Wawro, K. Kleczek and W.Brzozow-ski, 2004. Slaughter value and meat quality of Muscovy and Pekin ducks and their crossbreds and evaluation of heterosis effect. *Arch. Tierz.* 3: 287-299.

Effect of Immunocastration on Myosin Heavy Chain Expression

Sa-nga-pet^{*1}S., C. Chaosap¹ & P. Sivapirunthep¹

¹Agricultural Education Department, Faculty of Industrial Education, King Mongkut's Institute of Technology Ladkrabang, 10520 Bangkok, Thailand

Abstract

The aim of this study was to examine the effect of immunocastration on myosin heavy chain (MyHC) isoform expression in 3 crossbred pigs [Duroc x (Large white x Land race)]. The animals were used from 2 groups of surgical castration and immunocastration pigs. The pigs were 15 pigs in each group. They were slaughtered at the age of 24 weeks (100-110 live weight). The *Longissimus dorsi* (LD) muscle was taken to process for RNA and cDNA synthesis for measuring the myosin heavy chain isoform expression. The results revealed that the expression of MyHC Ix from immunocastration pigs was significantly higher than from surgical castration pigs ($P < 0.05$). While other three MyHC isoforms MyHC I, MyHC IIa and MyHC IIb were not significantly different between

Keywords: surgical castration, immunocastration, myosin heavy chain, muscle fiber types

Introduction

The surgical castration and immunocastration methods effect on pig growth performance. The immunocastration pigs have leaner carcass compare to the surgical castration pigs (Škrlep et al., 2010). There is a relationship between muscle growth and muscle fiber types. According to the properties of muscle fiber, there are 3 fiber type slow-twitch oxidative, fast-twitch oxidative, and fast-twitch glycolytic fibers (Wimmers et al., 2007). The aim this study was to examine and compare the muscle fiber types as myosin heavy chain (MyHC) isoform expression in surgical castration and immunocastration pigs.

Material and Methods

Animals and Muscle Sampling

In this study 30 of 3 crossbred pigs [Duroc x (Large white x Land race)] were divided into 2 groups of 15 surgical castration pigs and 15 immunocastration pigs. The surgical castration pigs were castrated at the age of 1 week old. The Vaccinations with Improvac[®] were performed to immunocastration pigs at the age of 16 and 20 weeks. All pigs were raised from age of 10 weeks to 24 weeks. They were fed by 12%, 14% and 16% protein commercial pig ration. All pigs were slaughtered at the age of 24 weeks and the slaughter weight was 100-110 kg. Fresh *Longissimus dorsi* (LD) muscle sample taken at the 13th to 14th ribs were stored in liquid nitrogen and further

processed for RNA and cDNA synthesis at laboratory of the Agricultural Education Department, Faculty of Industrial Education, King Mongkut's Institute of Technology Ladkrabang.

Primer used

The list of primers used to quantify myosin heavy chain (MyHC) isoforms were show in the table 1.

Table 1. List of primers used to quantify myosin heavy chain (MyHC) isoforms.

Gene	Primer sequence (5'to3')	Annealing temperature°C	GenBank accession no.
MyHC I	Forward: AAGGGCTTGAACGAGGAGTAGA	60	AB053226
	Reverse: TTATTCTGCTTCCTCCAAAGGG		
MyHC IIa	Forward: GCTGAGCGAGCTGAAATCC	60	AB025260
	Reverse: ACTGAGACACCAGAGCTTCT		
MyHC IIx	Forward: AGAAGATCAACTTGATGAACT	60	AB025262
	Reverse: AGAGCAGAGAACTAACGTG		
MyHC IIb	Forward: ATGAAGAGGAACCACATTA	57	AB025261
	Reverse: TTATTGCCTCAGTAGCTTG		

Source: Wimmers et al. (2007)

Quantitative Real Time PCR

Reverse transcription and PCR were performed on sample from individuals from each group. Real-time PCR reverse transcription-PCR was performed on a CFX96™ Real-Time System (BIO-RAD) in the laboratory of the Agricultural Education Department, Faculty of Industrial Education, King Mongkut's Institute of Technology Ladkrabang. The reaction mixture consisted of cDNA, 0.5 μM of upstream and downstream primers, and SYBR Green Universal PCR Master mix (SensiFast™ SYBR, BIOLINE)

Relative standard curves were generated for each MyHC isoform primer set to calculate PCR efficiency. The C_q values (y-axis) were plotted against log₁₀ ng equivalent RNA (x-axis), and PCR reaction efficiencies (E) were calculated from the standard curve as 10^(-1/slope)-1. (Čikoš et al., 2007). An average C_q value was obtained for each primer and probe set each sample, and this was used to calculate the relative expression ratio (rER)

$$rER = \frac{[1+E(\text{MyHC target gene})]^{-C_q(\text{MyHC target gene})}}{[1+E(\text{MyHC control gene})]^{-C_q(\text{MyHC control gene})}}$$

Because of the nature of the analysis, any one of the primer sets could be set MyHC control gene, with the expression of each other MyHC target gene being calculate relative to this. The rER of the MyHC control gene was set as a constant value of 1. The sum of rER values for the different primer sets was calculate and the relative contribution of each primer set rER was determined as a percentage. Data were expressed as percentage of total adult MyHC expression (Hemmings et al., 2009).

For two castration methods comparison of each isoforms MyHC expression data were analyzed by Independent sample t-test.

Results and Discussion

The results revealed that the expression of MyHC Iix from immunocastration pigs was significantly higher than from surgical castration pigs ($P < 0.05$). There were not significantly different in MyHC I, MyHC Iia, and MyHC Iib ($P > 0.05$) as shown in table 2. However, the MyHC Iia expression of surgical castration pigs was higher than from immunocastration pigs (7.06% vs 1.20%). Regarding surgical castration method, there is no testosterone left in surgical castration pigs which effect on the higher expression of MyHC Iia. The pigs without testosterone grow slow and has slower muscle fiber types fast-twitch oxidative (MyHC Iia).

Table 2. Percentage of myosin heavy chain isoform (MyHC).

Trait	Surgical castration	Immuno-castration	P-value
MyHC I	0.04 \pm 0.03	0.04 \pm 0.03	0.74
MyHC Iia	7.60 \pm 14.26	1.20 \pm 1.42	0.10
MyHC Iix	77.06 \pm 15.70	86.74 \pm 9.16	0.05
MyHC Iib	15.30 \pm 10.22	12.01 \pm 8.36	0.35

In this study, Immunocastration showed higher percentage of expression of fast MyHC Iix than surgical castration pigs.

Acknowledgement

The authors acknowledge the financial support from the Zoetis Thailand.

References

- Čikoš, Š., A. Bukovská and J. Koppel, 2007. Relative quantification of mRNA: comparison of methods currently used for real-time PCR data analysis. *BMC Mol. Biol.* 113.
- Hemmings, K.M., T. Parr, Z.C.T.R. Daniel, B. Picard, P.J. Buttery and J. M. Brameld, 2009. Examination of myosin heavy chain isoform expression in ovine skeletal muscle. *J. Anim. Sci.* 87:3915-3922.
- Wimmers, K., N.T. Ngu, D.G.J. Jennen, D. Tesfaye, E. Murani, K.Schellander and S. Ponsuksili, 2008. Relationship between myosin heavy chain isoform expression and muscling in several diverse pig breeds. *J. Anim. Sci.* 86:795-803.
- Škrlep, M., B. Šegula, M. Prevolnik, A. Kirbiš, G. Faarine and M. Candek-Potoar, 2010. Effect of immunodastration (Improvac[®]) in fattening pigs II: carcass traits and meat quality. *47(2):* 65-72.

Meat composition, fatty acid profile and oxidative stability of meat from broilers supplemented with pomegranate (*Punica granatum* L.) by-products

Ahmed S. T., H. S. Mun, M.M. Islam, A. B. M. R. Bostami, S. Y. Ko, I. H. Bae & C. J. Yang
Department of Animal Science and Technology, Suncheon National University, South Korea

Abstract

The effects of diets supplemented with four levels (0, 0.5, 1.0 and 2.0%) of pomegranate by-product (PB) on meat composition, fatty acid profile and oxidative stability of broiler meat were evaluated. The crude protein and moisture contents were increased, whereas ether extract in breast, thigh meat and cholesterol in breast meat were decreased in response to dietary PB supplementation ($P < 0.05$). In breast and thigh meat, the sum of saturated fatty acids was lower, while the sum of mono-unsaturated and n-3 fatty acids were higher, alongside lower n-6/n-3 ratio in the 1.0 and 2.0% PB supplemented group ($P < 0.05$). The TBARS values of breast and thigh meat were reduced in all the PB supplemented groups ($P < 0.05$). Overall, the results presented herein indicate that supplementation of diets with up to 2% pomegranate by-products improved the meat composition, fatty acid profile and reduced lipid oxidation of broiler meat.

Keywords: broiler chickens, fatty acids, meat composition, pomegranate by-products, TBARS

**Corresponding author:* yangcj@sunchon.ac.kr

Introduction

Owing to the possible carcinogenicity of synthetic antioxidants and residues found in meat products, considerable interest has risen in the use of natural antioxidants as bio-preservatives, such as plant polyphenols, tocopherols, carotenoids, flavonoids, and phenolic acids. The peels of pomegranate have been reported to possess several bioactive compounds including ellagitannin, punicalagin (Kanatt et al., 2010), flavonoids, anthocyanins and other phenolic compounds. The seeds are rich source of total lipids with a high concentration of polyunsaturated fatty acids; PUFAs, protein, vitamins, minerals, polyphenols, and steroids (Prakash and Prakash, 2011). The antioxidant activity of pomegranate juice and peel extract has been demonstrated by many scientific studies. However, there is currently no published research regarding the effects of supplementing broiler diets with pomegranate on its meat quality. Therefore, in this study, we investigated whether feeding pomegranate by-product (PB) in broilers to improves meat composition, fatty acid profile and oxidative stability.

Materials and Methods

The by-products of pomegranate was collected, air dried and grinded to prepare the experimental PB. A total of 320 one-day-old Ross 308 male broilers were randomly allocated into four treatment groups (PB 0%, 0.5%, 1.0% and 2.0%) with ten replicate pens of eight birds in a

completely randomized design. Feed intake and body weight were recorded and the average daily feed intake (ADFI), weight gain (ADG), and FCR (feed : gain) were calculated.

At 36 days of age, two broilers from each replicate cage were randomly selected and slaughtered to separate the breast and thigh meat. The moisture, total ash, crude protein, and ether extract contents of the breast and thigh meat samples were analyzed according to AOAC (2000) methods. Cholesterol was separated from fat after saponification with KOH and extraction with ethyl ether. The fatty acids composition of meat was measured using a slight modification method described by O'Fallon et al. (2007). The thiobarbituric acid reactive substances (TBARS) values of refrigerated meat (4°C) was determined at 1, 3, 5, 7, 14, 21, and 28 day of storage. Data were analyzed by SAS (2003) and level of significance was preset at $P < 0.05$.

Table 1. Effects of dietary pomegranate by-products (PB) on proximate composition and cholesterol content of broiler meat.

Parameter	Pomegranate by-product (PB)				SEM	P-value
	0%	0.5%	1.0%	2.0%		
Breast meat						
Crude protein (%)	26.21 ^b	28.51 ^a	28.19 ^a	28.55 ^a	0.48	0.016
Ether extract (%)	1.50 ^{ab}	1.90 ^a	1.33 ^b	1.19 ^b	0.12	0.015
Moisture (%)	72.38 ^b	72.59 ^b	72.82 ^{ab}	73.29 ^a	0.18	0.033
Crude ash (%)	1.45	1.40	1.39	1.40	0.02	0.359
Cholesterol (mg/100g)	77.44 ^a	63.91 ^b	64.83 ^b	62.80 ^b	3.32	0.048
∑Saturated fatty acid	35.57 ^a	33.47 ^{ab}	30.83 ^c	32.29 ^{bc}	0.64	0.003
∑Monounsaturated fatty acid	43.94 ^c	44.53 ^{bc}	47.46 ^a	45.70 ^b	0.55	0.004
∑Polyunsaturated fatty acid	19.17	20.58	20.31	20.66	0.57	0.33
∑n-3 fatty acid	2.74 ^b	4.04 ^a	3.75 ^a	3.92 ^a	0.19	0.006
∑n-6 fatty acid	17.66	18.36	18.17	18.43	0.57	0.81
PUFA/SFA	0.54	0.62	0.66	0.64	0.03	0.074
n-6/n-3	6.89	4.57	4.87	4.73	0.47	0.057
Thigh meat						
Crude protein (%)	22.18 ^b	24.24 ^a	24.11 ^a	23.44 ^a	0.30	0.002
Ether extract (%)	4.07 ^a	3.03 ^b	3.27 ^b	2.85 ^b	0.18	0.006
Moisture (%)	67.88 ^b	68.30 ^b	71.93 ^a	71.52 ^a	0.86	0.026
Crude ash (%)	1.23	1.11	1.21	1.22	0.04	0.300
Cholesterol (mg/100g)	65.78	63.83	64.54	61.05	1.48	0.263
∑Saturated fatty acid	33.32 ^a	33.54 ^a	31.00 ^b	31.21 ^b	0.38	0.001
∑Monounsaturated fatty acid	46.17 ^b	46.15 ^b	48.69 ^a	49.94 ^a	0.55	0.001
∑Polyunsaturated fatty acid	20.51	20.31	20.31	18.85	0.45	0.13
∑n-3 fatty acid	3.51 ^{bc}	3.45 ^c	3.75 ^{ab}	3.98 ^a	0.09	0.01
∑n-6 fatty acid	17.00 ^a	16.87 ^a	16.56 ^a	14.88 ^b	0.45	0.04
PUFA/SFA	0.62	0.61	0.66	0.61	0.02	0.31
n-6/n-3	4.86 ^a	4.91 ^a	4.43 ^a	3.74 ^b	0.18	0.002

^{a,b,c} Values with different superscripts in the same row differ significantly ($P < 0.05$).

Results and Discussion

Dietary supplementation of PB did not show significant effect on the ADG and ADFI, however FCR was significantly reduced in broiler compared to control (1.73 vs 1.61). The crude protein and moisture contents of breast and thigh meat were increased, whereas ether extract in breast and thigh meat and cholesterol in breast meat were decreased in response to dietary PB

supplementation ($P < 0.05$). Pomegranate peels contain considerable amounts of ellagic acid (1421.6 ± 28.62 mg/100 g) (Kim et al., 2013), which can inhibit the pancreatic lipase activity, thereby reducing abnormal lipid metabolism (Lei et al., 2007). In breast and thigh meat, the sum of saturated fatty acids was lower, while the sum of mono-unsaturated and n-3 fatty acids were higher, alongside lower n-6/n-3 ratio in the 1.0 and 2.0% PB supplemented group ($P < 0.05$). The TBARS values of breast and thigh meat were reduced in the PB supplemented groups ($P < 0.05$), which may be due to the antioxidant property of pomegranate peels (Kannat et al., 2010). Overall, the results presented herein indicate that supplementation of diets with tested level pomegranate by-products can improve the meat composition, fatty acid profile and reduced lipid oxidation of broiler meat, with 2% dose was most effective.

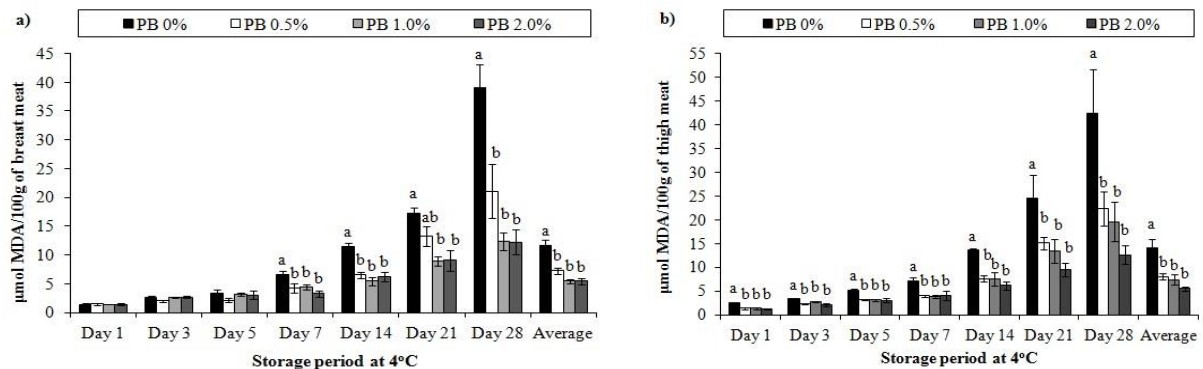


Figure 1. Effects of diet supplemented with or without pomegranate by-products (PB) on TBARS values of broiler breast (a) and thigh (b) meat (35 d) during refrigerated storage.

References

- AOAC, 2000. Official Methods of Analysis of the Association of Official Analytical Chemists, 17th ed., Gaithersburg, MD, USA.
- Kanatt, S. R., R. Chander and A. Sharma, 2010. Antioxidant and antibacterial activity of pomegranate peel extract improves the shelf life of chicken products. *Inter. J. Food Sci. Technol.* 45: 216-222.
- Kim, M. S., S. H. Yun, H. S. Na, H. J. Park, G. C. Choi, S. I. Yang, and J. H. Lee, 2013. Chemical compositions and functional characteristics of Korean and imported pomegranate (*Punica granatum* L.). *Korean J. Food Preserv.* 20: 342-347.
- Lei, F., X. N. Zhang, W. Wang, D. M. Xing, W. D. Xie, H. Su and I. J. Du, 2007. Evidence of anti-obesity effects of the pomegranate leaf extract in high-fat diet induced obese mice. *Inter. J. Obes.* 31: 1023-1029
- O'Fallon, J. V., J. R. Busboom, M. L. Nelson and C. T. Gaskins, 2007. A direct method for fatty acid methyl ester synthesis: Application to wet meat tissues, oils and feedstuffs. *J. Anim. Sci.* 85: 1511-1521.
- Prakash, C. V. S. and I. Prakash, 2011. Bioactive chemical constituents from pomegranate (*Punica granatum*) juice, seed and peel- A review. *Inter. J. Res. Chem. Environ.* 1: 1-18.
- SAS, 2003. SAS User's Guide: Statistics. Version 9. SAS Institute Inc., Cary, NC.

Effect of dried fermented juice of epiphytic lactic acid bacteria on broiler meat quality

Saenmahayak¹, B. *, S. Bureenok¹, K. Ampaporn², K. Vasupen¹, C. Yuangklang¹, E. Vasupen¹, N. Pitiwitayakul¹, & T. Srisapoom¹

¹Rajamangala University of Technology Isan, NakhonRatchasima, Thailand 30000,

²Rajamangala University of Technology Isan, SakonNakhon Campus, Thailand 47160

Abstract

The influence of powdered fermented juice of epiphytic lactic acid bacteria on processing yields, and meat quality of broiler chickens was assessed at 42 d of age. A total of 200 (1-d old) mixed-sex broilers were assigned to 4 dietary treatments: T1) Control diet (basal diet); T2) Control + 0.05% of Oxytetracycline; T3) Control + 0.2% of dried lactic acid bacteria; and T4) Control + 1.0% of dried fermented juice of epiphytic lactic acid bacteria. The birds were arranged in a Completely Randomized Design (CRD; 5 reps/trt and 10 birds/pen). Each treatment was provided in a 2-stage feeding program. Results indicate no differences ($P>0.05$) in body weight, carcass and component yields (wings, leg quarters, drumsticks, thighs, breast fillets and tenders) because of dietary treatments. The incidence and severity of footpad dermatitis did not show any differences ($P>0.05$) due to dietary treatments. Meat quality attributes (pH, drip loss, shear force, and lipid oxidation) were not significantly influenced ($P>0.05$) by treatment regimen. The redness value (a^*) of breast fillets was higher ($P<0.05$) in birds fed 0.2% DLAB compared to those fed control diet. However, dietary treatments had no significant effect ($P>0.05$) on L^* and b^* values. Probiotics can be replaceable to antibiotics in broiler feed in order to enhance meat color.

Keywords: probiotic, lactic acid bacteria, meat quality, broiler

**Corresponding Author: Saenmahayak B.: sbenya@hotmail.com*

Introduction

Antibiotics have been widely used in animal feed as growth-promoting agent. However, several countries have banned certain antibiotics due to the contamination of meat products with antibiotic residues. The use of probiotics has been considered as alternative to antibiotics. Probiotics are microorganisms that fed to animals to promote microflora balance and given beneficially effects on health and nutrition of the host. The advantage effects of probiotics supplementation on carcass and meat quality has been questioned, therefore, the objective of this study was to investigate the influence of powdered fermented juice of epiphytic lactic acid bacteria as probiotics on meat quality of broiler chickens at 42 d of age.

Materials and Methods

A total of 200 (1-d old) mixed-sex broilers were raised in 20 floor pens bedded with rice hulls to 42 d of age (5pens/trt; 10 birds/pen or 0.5 ft²/bird). The birds were arranged in a Completely Randomized Design (CRD). Each treatment was provided in a two stage feeding program. The starter feed was fed on d 1-21 and grower feed on d 22-42. Birds were assigned to 4 dietary treatments: T1) Control diet (basal diet); T2) Basal diet + 0.05% of Oxytetracycline; T3) Basal diet + 0.2% of dried lactic acid bacteria; and T4) Basal diet + 1.0% of dried fermented juice of epiphytic lactic acid bacteria (Bureenok et al., 2011). The incidence and severity of pododermatitis were scored on 42 d of age by using a visual ranking system (Bilgili et al., 2009). Breast fillet from 3 birds per pen were randomly selected for meat quality attributes. Deboned breast fillets (skinless *Pectoralis major* muscle) were individually bagged and stored at 4°C for drip loss and pH at 0 h and 24 h, lipid oxidation, shear force, and color (L*, a* and b*) measurements. The data were statically analyzed by the General Linear Models procedure for the ANOVA using SAS program. The Tukey's test was used to compared and separate means when the main effects were significant (P<0.05).

Results and Discussion

Dietary treatments (Table 1) were calculated according to NRC (1994). The incidence and severity of footpad dermatitis did not show any differences (P>0.05) due to dietary treatments (data not shown). Meat quality attributes (pH, drip loss, shear force, and lipid oxidation) were not significantly influenced (P>0.05) by treatment regimen (Table 2). For meat color, the redness value (a*) of breast fillets was higher (P<0.05) in birds fed 0.2% DLAB compared to those fed control diet. However, dietary treatments had no significant effect (P>0.05) on L* and b* values. The supplementation of single strain (*Lactobacillus plantarum*ST1) and multi strain (fermented juice of epiphytic lactic acid bacteria) probiotics to the diet improved breast meat color of broiler chickens.

Table 1. Composition (%) of dietary treatments.

Ingredients (%)	Starter (1-21 d)				Grower (22-42 d)			
	T1*	T2	T3	T4	T1	T2	T3	T4
Corn	47.15	47.15	46.95	46.15	52.76	52.76	52.56	51.76
Full fat soybean	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Soybean meal	26.50	26.50	26.50	26.50	20.50	20.50	20.50	20.50
Dicalcium Phosphate	2.42	2.42	2.42	2.42	2.26	2.26	2.26	2.26
Limestone	1.70	1.70	1.70	1.70	1.80	1.80	1.80	1.80
DL-Methionine	0.31	0.31	0.31	0.31	0.22	0.22	0.22	0.22
L-Lysine	0.17	0.17	0.17	0.17	0.21	0.21	0.21	0.21
Tallow	1.00	1.00	1.00	1.00	1.50	1.50	1.50	1.50
Salt	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Choline chloride 50%	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Premixed	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
	0.00	0.00	0.20	1.00	0.00	0.00	0.20	1.00
Chemical compositions								
DM (%)	87.86	88.74	88.35	89.47	86.95	87.96	88.54	89.35
CP (%DM)	21.92	22.22	22.09	21.90	21.05	21.10	21.20	21.08
Ash (%DM)	4.19	5.72	5.75	7.33	3.89	4.75	5.62	5.57

*T1 = Control (Basal diet); T2= Basal diet + 0.05% of Oxytetracycline; T3 = Basal diet + 0.2% of dried lactic acid bacteria, and T4 = Basal diet + 1.0% of dried fermented juice of epiphytic lactic acid bacteria.

Table 2. Effect of dietary treatments on meat quality of broiler chickens.

Factors	T1 ¹	T2	T3	T4	SEM ²
Drip loss (%)					
0h	2.0	2.3	2.2	2.5	0.37
24h	2.1	2.4	2.5	2.5	0.31
pH					
0h	6.23	6.30	6.28	6.34	0.08
24h	5.94	5.87	5.85	5.87	0.02
TBARS (mg MDA/kg)	1.76	2.16	1.82	1.83	0.20
Shear force (kgf)	7.89	6.98	6.48	6.63	0.41
Color					
L*	58.04	58.98	58.61	58.38	0.34
a*	5.71 ^b	5.99 ^{ab}	6.39 ^a	6.23 ^{ab}	0.16
b*	12.87	13.25	13.11	13.64	0.39

¹T1 = Control (Basal diet); T2= Basal diet + 0.05% of Oxytetracycline; T3 = Basal diet + 0.2% of dried lactic acid bacteria, and T4 = Basal diet + 1.0% of dried fermented juice of epiphytic lactic acid bacteria.

²SEM = Standard Error of the Mean

^{ab}Means within a column with difference superscripts differ significantly (p<0.05).

Conclusion

Either use DLAB or DFJLB as probiotics had no effect on footpad quality, drip loss, pH, lipid oxidation and shear force of breast fillets. However, redness value (a^*) of breast meat was higher in the birds that received probiotics at 42 d of age.

Acknowledgements

This research was supported by Rajamangala University of Technology Isan, Thailand.

References

- Bilgili, S.F., Hess, J.B., Blake, J.P., Macklin, K.S., Saenmahayak, B., and Sibley, J.L, 2009. Influence of bedding material on footpad dermatitis in broiler chickens. *J. Appl. Poult. Res.* 18: 583-589
- Bureenok, S., Suksombat, W., and Kawamoto, Y, 2011. Effects of the fermented juice of epiphytic lactic acid bacteria (FJLB) and molasses on digestibility and rumen fermentation characteristics of ruzigrass (*Brachiaria ruziziensis*) silages. *Livest. Sci.* 138: 266-271
- National Research Council (NRC), 1994. *Nutrient Requirements of Poultry*. National Academy Press, Washington. pp 176

Prediction of meat and carcass quality traits by real-time Ultrasound in swine

Promket^{1*}, D., K. Nabundi², K. Ruangwittayanusorn¹ & T. Somchan¹

¹ Department of Agriculture Technology, Faculty of Technology, Mahasarakham University, Mahasarakham, Thailand, 44000.

² Graduate student of Animal science, Department of Agriculture Technology, Faculty of Technology, Mahasarakham University, Mahasarakham, Thailand, 44000.

Abstract

The carcass characteristics of 296 crossbreed (large white x landrace x duroc) were evaluated the meat and carcass quality with real-time ultrasound. The body weight pigs were assigned to slaughter between 78 -134 kg. The means of hot carcass weight (CW) was 78.78 kg, chilling carcass weight (CCW) was 76.71 kg and loin eye area (LEA) 40.20 cm². The regressions linear model of ultrasound back fat thickness and ultrasound loin eye area on swine were also developed. LEA was dependent variable and the independent variables were ultrasound of loin eye area (U_LEA), ultrasound back fat thickness positions 3 (U_BF3), measurement of 10th-11th rib (BF10R) and fillet weight (FW), the R² model was high (0.828). The ultrasound measurements are accurate.

Keywords: carcass quality, real-time ultrasound, swine

Introduction

Pork industries, the carcass characteristics, especially the back fat thickness and hams weight, have economic importance for the industry). Ultrasound has been used to predict carcass traits of live animals for many years. The use of ultrasound for measuring carcasses to predict composition dates back to the 1950s. Conventionally, real-time ultrasonic scans of the longissimus muscle and fat layers have been made transverse to the long axis of the animal at the 10th or last rib. These have been shown to be accurate in predicting both back fat and longissimus muscle measurements and carcass composition. Daza et al. (2006) found the R² model between hams weight and slaughter live weight and ultrasound fat thickness was 0.82. Moreover, the correlations between 10th rib back fat measurement by real – time ultrasound and 10th rib back fat was 0.84 (Ragland et al., 2004). Newcom et al. (2014) used of real-time ultrasound to prediction the intramuscular fat percentage in live swine. They found that, the correlation and rank correlation coefficients between predict loin intramuscular fat percentage and carcass loin intramuscular fat percentage were 0.60 and 0.56, respectively. The prediction meat and carcass quality of measurement carried out in live pigs before slaughter is an interesting aspect for producer and for the industry. The objective of this study was to investigate the relationship between ultrasound and carcass quality and to assess the possibility of utilizing the ultrasound technique to predict actual carcass traits.

Materials and Methods

Animals and Ultrasound Image Collection

This study included 296 cross breeds pigs (Large White × Landrace × Duroc) from commercial farm with an initial live weight between 78 and 134 kg. Piglets were raised in standard commercial conditions until the commercial slaughter weight. The ultrasonically scanned 4 h before slaughter, using an HS - 2000 (Honda electronics CO.LTD) with a 10 cm, 3.5 MHz probe and a 13 cm, 1.5-5 probe. All the pig were collect body weight (BW), body length (BL) and scan of loin eye area (U_LAE), back fat thickness 3 positions (U_BF1 , U_BF2 and U_BF3) obtained 2 h before slaughter. The pigs were slaughtered at commercial slaughter house, after electrical stunning.

Carcass Collection

At the commercial slaughterhouse, within 10 min post - mortem, hot carcass weight (CW, kg), pH₀ at 0 h post – mortem (pH₀) and carcass length (CL, cm) were collected. pH at 45 min post – mortem (pH₄₅) were determined using pH meter. The back fat thickness were measurement of first rib (BFFR, mm), measurement of 10th-11th rib (BF10R, mm) and measurement of last rib (BFLR, mm). After chilled, loin eye area (LEA, cm²) were place a plastic grid over the loin eye and count the dots or square the fall within the boundaries of the longissimus muscle convert to square inches by dividing the number of dots or squares by the appropriate conversion factor on the grid, chilling carcass weight (CCW, kg), pH 22 h (pH₂₂), loin weight (LW, kg), ham weight (HW, kg), fillet weight (FW, kg) and shoulder weight (SW, kg) were determined.

Statistical analysis

The means of ultrasound and carcass traits were analyzed using PROC MEANS (SAS, 1998). Multiple linear regressions using to predict the equation model by PROC Stepwise. The prediction model selected was the most right best fit model with a maximum R² and minimum mean square error (MSE).

Results and Discussions

This study showed the means of CW was 78.78 kg, CCW was 76.71 kg and LEA 40.20 cm² previous studies of Schwab et al. (2010) reported ultrasound 10th rib loin muscle areas were 41.05 cm² by showed less than this study. Moreover Ayuso et al. (2013) found the means of 10th rib by ultrasound were 38.12 cm² by larger than this study. When BF was used as the dependent variables and BW, BL, U_BF3, P_DL, BFFR, BFLR, and LW as independent variable R² model was 0.601. The prediction equations of LEA were shown as the dependent variable. The independent variables were U_LEA, U_BF3, BF10R and FW, the R² model was high (0.828) (Table 1). It was implied that independent variables were good predictors of LEA. The variable HW and the independent variable were U_LEA, CW, BFLR, LW and SW (R² model = 0.698). Ayuso et al. (2013) reported the R² model for the prediction of HW were 0.48. The variable LW and the independent variable were BW, BL, LEA, CW, BF10R and HW (R² model = 0.655) and the prediction equations of SW were shown as the dependent variable. The independent variables were BL, HOTW, BFFR and HW (R² model = 0.712) was higher reported with Ayuso et al. (2013) (LW; R² model = 0.27 and SW; R² model = 0.35).

Table 1. Prediction equations for back fat thickness, loin eye area, ham weight and loin weight.

Dependent variables	Independent variable	β	<i>P</i> value	<i>R</i> ² model	MSE
BF	Intercept = 7.718		0.0359	0.601	15.62
	BW	0.049	0.1052		
	BL	-0.180	<.0001		
	U_BF3	0.213	0.0023		
	P_DL	-0.656	0.0475		
	BFFR	0.353	<.0001		
	BFLR	0.218	<.0001		
	LW	0.982	0.0121		
LEA	Intercept = 2.732		0.0176	0.828	4.630
	U_LEA	1.019	<0.0001		
	U_BF3	-0.061	0.1111		
	BF10R	0.055	0.0120		
	FW	-0.886	0.0099		
HW	Intercept = 1.085		0.0108	0.698	0.354
	U_LEA	0.016	0.0362		
	CW	0.061	<.0001		
	BFLR	-0.009	0.0956		
	LW	0.109	0.1022		
	SW	0.4461	<.0001		
LW	Intercept = -3.087		<.0001	0.655	0.244
	BW	-0.009	0.0904		
	BL	0.217	<.0001		
	LEA	0.011	0.0425		
	CW	0.072	<.0001		
	BF10R	0.020	0.0003		
	HW	0.093	0.0395		

Conclusion

The results from this study showed the means of BW was 104.4 kg. Regressions predicting of LEA as the dependent variables and U_LEA, U_BF3, BF2 and SLW as independent variable resulted in the *R*² model of high value (0.82). The use of a dependent variable should be used with BF, LEA, HW and LW values of 0.60, 0.82, 0.69 and 0.65, respectively for *R*² model.

References

- Ayuso, D., A. Gonzalez, F. Hernandez, J. M. Corral and M. Izquierdo. 2013. Prediction of carcass composition, ham and foreleg weights, and lean meat yields of Iberian pigs using ultrasound measurements in live animals. *J. Anim. Sci.* 91: 1884-1892.
- Daza, A., A. Mateos, C. Lopez-Carrasco, A. Rey, I. Ovejero, and C. J. Lopez-Bote. 2006. Effect of feeding system on the growth and carcass characteristics of Iberian pigs, and use of ultrasound to estimate. *Meat Sci.* 72:1-8.
- Newcom, D. W., T. J. Baas, and J. F. Lampe. 2014. Prediction of intramuscular fat percentage in live swine using real-time ultrasound. *J. Anim. Sci.* 80: 3046–3052.
- Ragland, K. D., L. L. Christian, and T. J. Baas. 2004. Evaluation of real-time ultrasound and carcass characteristics for assessing carcass composition in swine. *Zootec.* 41: 2463-2466.
- SAS. 1998. SAS User's Guide. Version 6.12. SAS. Inst., Cary, NC.
- Schwab, C. R., T. J. Baas, and K. J. Stalder. 2010. Results from six generations of selection for intramuscular fat in Duroc swine using real – time ultrasound. *J. Anim. Sci.* 88

Drip and cooking loss of longissimus dorsi muscle under different levels of energy and protein in Iranian kid native breed

Karami M.

Department of Animal Science agriculture and natural resource research center of shahrekord, 415, Iran

Abstract

An experiment was conducted to evaluate the effects of three levels of metabolisable energy (2, 2.4 and 2.8 Mcal/kg DM) and three levels crude proteins (12.6, 14 and 16.8 percents) on drip and cooking loss to identified the optimum levels of dietary energy and protein of indigenous kid meat. One of the importance factors that effect on meat quality in red meat is amount of drip loss and cooking loss of meat. Meat nutrients, water holding capacity, flavor and taste have been important of quality index, health and barbeque index of meat. The 27 kid meat samples were taken of fattened carcasses under complete randomizes design with a factorial experiment of 3×3 with 9 groups for 4 months fattening period. Control group and other experimental groups of 1, 2, 3, 4, 5, 6, 7, 8, and 9 were fed by ration of energy to protein ratio of 1:1, 1:1.2, 1:0.8, 1.2:1, 1.2:1.2, 1.2:0.8, 0.8:1, 0.8:1.2 and 0.8:0.8 respectively by total mixed ration. After 24 hours slaughter were sampled of longissimus dorsi (LD) muscles. The muscle was vacuum- packaged and conditioned for 0, 7 and 14 days in a chiller at 4°C for measuring drip loss and cooking loss. Effect of different levels of energy and protein on drip loss, cooking loss in most of groups were not significant, but post-mortem aging time of different levels of energy and protein were significant ($P < 0.05$). In general, it can be concluded that the diet with 2.4 Mcal/kg DM and 14% crude protein suggested as an appropriate diet.

Key words: drip loss, cooking loss, Iranian native kid, protein and energy levels

Introduction

Water is the major component of lean muscle accounting for approximately 75% of its weight (Aberle et al., 2001). There are three types of water in muscle; each differing in degree of its freedom, most of the water in muscle is held within the cell structures include inside myofibrils and intermyofibrillar spaces, between myofibrils and cell membranes, between muscle cells and between bundles of muscle cells (Kolczak et al., 2007). The bound water has reduced mobility and is very resistant to freezing and evaporation by heat. This water changes very little in post rigor muscles (Offer & Knight, 1988). The second type of water found in skeletal muscle is called immobilized water. A Nuclear Magnetic Resonance study indicated that the amount of water which was tightly trapped in protein networks increases with ageing (Bertram et al., 2004). The ability of fresh meat to retain both inherent and added water content is termed WHC (Aberle et al., 2001). Drip loss is an important quality criterion for the meat processing industry and the consumer. Offer & Knight (1988) described drip loss as a reddish fluid mainly consisting

of water and proteins. It can be expelled from cut surfaces of muscles or pieces of meat without any mechanical force other than gravity. Drip loss occurs from cut surfaces of meat. High drip loss is undesirable because it detracts from the appeal of the meat, and valuable proteins and flavor compound are lost in the exudates (Varnam & Sutherland, 1995). Drip loss is normally in the order of 3% in beef but may be exacerbated by very low pH and by freezing and thawing to as much as 15% (Offer & Knight, 1988). In the past, the assessment of drip loss was done by several methods. For example the filter paper-press method and the bag method of Honikel (1998). Cooking losses from chevon are of interest because, the water that remains in the cooked product is the major contributor to the sensation of juiciness and chevon cooking losses are often close to or over 35% (Webb et al., 2005). High cooking losses not only reduce the size of the meat portion but also result in reduced juiciness, tenderness and loss of flavor. Low energy and low protein portion in animal diets reduced intramuscular fat then effect on meat juiciness. Cooking loss of the meat is possibly exacerbated by its limited fat content (Huff-Lonergan & Lonergan, 2005).

Materials and Methods

The 27 kid meat samples were taken of fattened carcasses under complete randomizes design with a factorial experiment of 3×3 with 9 groups for 4 months fattening period. Control group and other experimental groups of 1, 2, 3, 4, 5, 6, 7, 8, and 9 were fed by ration of energy to protein ratio of 1:1, 1:1.2, 1:0.8, 1.2:1, 1.2:1.2, 1.2:0.8, 0.8:1, 0.8:1.2 and 0.8:0.8 respectively by total mixed ration. After post mortem 24 hours were sampled of longissimus dorsi (LD) muscles. The muscle was vacuum-packaged and put on a plastic hurdle (about 30 g) and conditioned for 1, 7 and 14 days in a chiller at 4°C for measuring drip loss and cooking loss. At the 0, 4 (Second experiment only), 7 and 14 days post mortem, longissimus dorsi muscle sample was weighed (approximately 30g) and put on a plastic hurdle. Then both items (meat samples on the plastic hurdle) were put into sealed polyethylene bags hermetically closed to prevent surface evaporative loss. After a 24 h storage period at 4°C, the meat samples were removed from the bag and reweighed. The difference in the weight of the samples, before and after storage, divided by the sample weight before storage × 100 accounted for the % drip loss (Honikel, 1998).

The measurement of cooking loss was conducted on the longissimus dorsi (LD) (between the 6th and 13th thoracic vertebrae), infraspinatus (IS) and biceps femoris (BF) samples were weighed (about 30g) and held in plastic bags and immersed in a 80°C water-bath until the internal temperature reached 78°C as monitored with a needle thermometer. Then, the bags were cooled under running tap water for 30 min, blotted dry with paper towels, and reweighed. Cooking loss, as percentages, were then calculated from the difference between the weights (Honikel, 1998).

Results and Discussion

Effect of different levels of energy and protein on drip loss, cooking loss in most of groups were not significant, but post-mortem aging time of different levels of energy and protein were significant ($P < 0.05$). In this experiment the least square means for the percent of drip loss and cooking loss in the LD muscle was not affected ($P > 0.05$) by the treatments. The drip loss increased ($P < 0.05$) by days 7 and 14 of the time of display. The post-mortem aging time were significant effect on drip loss and cooking loss in most of groups ($P < 0.05$). The cooking loss

increased ($P<0.05$) by 14 days of the time of display. The post-mortem aging time were significant effect on cooking loss in most of groups ($P<0.05$). Leheska et al. (2002) reported that dietary treatments had not influence on drip loss and cooking loss. According to Kadimet al. (2003) reported ageing had a significant influence on percent cooking loss of the selected muscles. Dietary treatment comprising different level of energy and protein or post mortem aging period did not have any effect on the cooking loss of LD muscle (Kannan et al., 2006). In general, it can be concluded that the diet with 2.4 Mcal/kg DM metabolisable energy and 14 percent crud protein suggested as an appropriate diet.

Table 1. Drip loss and cooking loss in *Longissimus dorsimuscle* of goats under different treatments and post mortem aging period (n=27).

		Post mortem aging periods					
	Treatments	Drip loss			Cooking loss		
		1 day	7 day	14 day	1 day	7 day	14 day
<i>Longissimus dorsimuscle</i> (LD)	1	1.35 ^x	3.65 ^y	3.89 ^y	23.4 ^x	26.2 ^{xy}	28.7 ^y
	2	1.42 ^x	3.73 ^y	3.84 ^y	24.1 ^x	26.5 ^{xy}	29.6 ^y
	3	1.33 ^x	3.48 ^y	3.54 ^y	23.2 ^x	25.8 ^{xy}	28.2 ^y
	4	1.35 ^x	2.59 ^{xy}	3.18 ^y	23.6 ^x	26.9 ^{xy}	29.2 ^y
	5	1.42 ^x	3.05 ^y	3.66 ^y	25.1 ^x	28.3 ^{xy}	30.4 ^y
	6	1.09 ^x	2.46 ^{xy}	3.03 ^y	25.3 ^x	28.5 ^{xy}	30.8 ^y
	7	1.55 ^x	3.26 ^y	3.76 ^y	23.1 ^x	25.7 ^{xy}	28.9 ^y
	8	2.14 ^x	3.42 ^{xy}	4.13 ^y	22.8 ^x	25.6 ^x	27.4 ^y
	9	1.15 ^x	2.78 ^y	3.53 ^y	23.3 ^x	29.1 ^y	31.3 ^y
	±SEM	0.42	0.71	0.69	1.59	1.81	1.73

^{x,y} Means within rows with different superscripts are different among post mortem aging periods ($P<0.05$).

References

- Aberle, E. D., J. C. Forrest, D. E. Gerrard and E. W. Mills. 2001. Structure and composition of animal tissues. In: Principles of Meat Science. pp. 9-116.
- Bertram, H. C., J. Z. Hu, D. N. Rommerein, R. A. Wind and H. J. Andersen. 2004. Dynamic high-resolution ¹H and ³¹P NMR spectroscopy and ¹H T₂ measurements in post-mortem rabbit muscles using slow magic angle spinning. *J. Agr. Food Chem.* 52: 2681-2688.
- Honikel, K. O. 1998. Reference methods for the assessment of physical characteristics of meat. *Meat Science. Meat Sci.* 49: 447-457.
- Huff-Lonergan, E. and S. M. Lonergan. 2005. Mechanisms of water holding capacity of meat: the role of post mortem biochemical and structural changes. *Meat Sci.* 71: 194-204.
- Kadim, I. T., O. Mahgoub, D. S. Al-Ajmi, R. S. Al-Maqbaly, N. M. Al-Saqri and A. Ritchie. 2003. An evaluation of the growth, carcass and meat quality characteristics of Omani goat breeds. *Meat Sci.* 66: 203-210.
- Kannana, G., K.M. Gadiyaram, S. Galipalli, A. Carmichael, B. Kouakou, T. D. Pringle, K. W. McMillin, S. Gelaye. 2006. Meat quality in goats as influenced by dietary protein and energy levels, and post mortem aging. *Small Rumin. Res.* 61: 45-52.
- Kolczak, T., K. Krzysztoforski and K. Palka. 2007. The effect of post-mortem ageing and heating on water retention in bovine muscles. *Meat Sci.* 75: 655-660.
- Leheska, J. M., D. M. Wulf and R. J. Maddock, 2002. Effects of fasting and transportation on pork quality development and extent of post mortem metabolism. *J. Anim. Sci.* 80: 3194-3202.

- Offer, G. and P. Knight. 1988. The structural basis of water-holding in meat part 2: Drip losses. In: Lawrie, R., Editor, 1988.(Vol. 4): Developments in meat science, Elsevier Applied Science, London. pp 172-243.
- Varnam, A. H. and J. P. Sutherland. 1995. Meat and Meat Products: Technology, Chemistry and Microbiology: Chapman and Hall, London. pp 47-119.
- Webb, E.C., N.H. Casey and L. Simela. 2005. Goat meat quality. Small Rumin. Res. 60: 153-166.

Microbiological and chemical characteristics of probiotic goat cheese with mixed cultures of *L.rhamnosus* and *L.plantarum*

Triana Setyawardani, Agustinus HD Rahardjo, Mardiaty Sulistyowati and Juni Sumarmono

Abstract

Probiotic goat cheese is one of probiotic bacteria application in food processing, in order to produce functional food. In this research, the microbiological and chemical characteristics of cheese made from goat's milk with *L.rhamnosus* and *L.plantarum* as adjunct starter were evaluated. The cheese was made by adding mixed culture of *L.rhamnosus* and *L.plantarum* at 2.5%; 5.0 %; 7.5% and 10.0% (v/v). Treatments were arranged in a completely randomized design (CRD). Fresh cheese was then stored for 10, 20 and 30 days in a cold room. The measured variables were the amount of lactic acid bacteria (log cfu/gr), yeast (log cfu/gr), titrable acidity (%) and pH. Each treatment has 3 replicates. Results showed that the effect of different concentration of lactobacilli cultures on cheese characteristics was not significant. The amount of probiotic lactic acid bacteria were ranged from 9.27 to 10.98 log cfu/gr and the amount of yeast ranged from 2.33 to 6.56 log cfu/gr of cheese. In addition, cheese has titrable acidity between 0.06 and 0.23 %, and pH between 4.91 and 6.56. In conclusion, cheese stored for 30 days had the most optimum microbiological and chemical characteristics.

Keywords: cheese, L.rhamnosus TW2, L.plantarum TW14, lactic acid bacteria, yeast

Introduction

Important criteria for probiotic bacteria in functional foods are, among others, the minimum number of viable cells and their survivability during processing and storage. Probiotic bacteria have to be chosen or selected in order to obtain the desired characteristics of the food products. Furthermore, probiotic to be selected must meet minimum criteria through isolation and identification, adhesion, and immune response test. *L.rhamnosus* TW2 and *L.plantarum* TW14 were isolated from goat milk, and have met some obligatory criteria such as resistance against acid and bile salt. This paper presents our observation on microbiological and chemical characteristics of goat cheese added with probiotic bacteria *L.rhamnosus* TW2 and *L.plantarum* TW14 during its making processes.

Methods

Materials. The primary ingredients were fresh goat milk from Ettawah crossbred goats obtained local goat breeders. Other materials included commercial animal rennet, pure culture of *L.rhamnosus* TW2 and *L.plantarum* TW14, microbiological media de *Mann Rogosa Sharp Agar* (MRSB) and de *Mann Rogosa Sharp Broth* (MRSB), aquadest, bufer pH 4 and pH 7, NaOH 0,1 N.

Preparation of culture starter. *L.rhamnosus* TW2 and *L.plantarum* TW14 cultures were refreshed in MRSB media at 37°C within 24 hours before inoculated to goat milk as performance culture by adding 5% (v/v) pure culture in MRSB into goat milk. The milk containing LAB culture was then incubated at 37°C for six hours.

Cheesemaking. Fresh goat cheese was produced following procedure of Walstra *et al.* (1999) and Gardiner *et al.* (1998) on *cheddar* cheese. Probiotic bacteria culture of *L. Rhamnosus* TW2 and *L.plantarum* TW14 were added to start the fermentation process.

Determination of lactic acid bacteria (Burns *et al.*, 2008). LAB stability test comprised of pH test and total LAB. After made, cheese was stored at refrigerator temperature (5°C) for 0 to 30 days. Twenty gr sample was put into 180 ml 2% (b/v) sterile natrium citric, then homogenized in stomacher for 3 minutes. One ml homogenat was taken for 1:10⁸ decimal dilution. One ml samples from three highest dillutions was aseptically taken and put into petri dish. MRSA media was poured and sample was incubated at 37°C for 48 hours.

Determination of cheese pH, and titratable acidity. Cheese pH was measured using glass electrode-pH meter (Hanna Instrument) with 10g cheese pulverized in 10 ml distilled (Shakeel-Ur- Rehman *et al.*, 2003). Titratable acidity (lactic acid content) was measured using Mann's Acid Test method (Sudarmadji *et al.*, 1997) using formula volume of NaOH xN NaOH x 0.09)/(sample weight) x 100%.

Results and discussions

The total number of *L.rhamnosus* TW2 and *L.plantarum* TW14 in the curd was 8,64 log cfu/g and 9,29 log cfu/g, respectively, and the mixed culture was 10,02 log cfu/g (Table 1). Probiotic LAB resistance during fermentation, which was calculated as the initial LAB amount before storage, depended on probiotic strain used, starter culture and milk raw material.

Table 1. Average of LAB amount (log cfu/g) in cheese

Percentage of culture addition	Day			
	0	10	20	30
<i>L.rhamnosus</i> TW2+ <i>L.plantarum</i> TW14, 2.5	9,63 ± 0,38 a	9,27 ± 0,43 a	9,32 ± 0,02 a	9,52 ± 0,19 a
<i>L.rhamnosus</i> TW2+ <i>L.plantarum</i> TW14, 5.0	9,47 ± 0,35 a	9,56 ± 0,28a	10,04 ± 0,82 b	9,77 ± 0,31a
<i>L.rhamnosus</i> TW2+ <i>L.plantarum</i> TW14 ,7.5	9,64 ± 0,19 a	9,49 ± 0,05a	9,57 ± 0,06 b	10,98 ± 0,65ab
<i>L.rhamnosus</i> TW2+ <i>L.plantarum</i> TW14, 10	9,73 ± 0,24 a	9,44 ± 0,28a	9,76 ± 0,07 b	9,64 ± 0,06b

Values bearing different superscript within lines and columns show highly significant difference ($p < 0.01$).

Cheese made with *L. rhamnosus* TW2, *L.plantarum* TW14 cultures and mixed culture (*L.rhamnosus* TW2 and *L.plantarum* TW14) did not experience LAB decrease during 30 day storage, averaged on 9,67 ± 0,48 log cfu/g. it indicated LAB ability to thrive well in cheese product and LAB high adaptability in cheese matrix environment during four week storage at low temperature (5°C). Viability of *Lactobacillus acidophilus* or *Lactobacillus casei* in cheddar cheese during six-month storage still contained 10⁸ cfu/g LAB as tested by Ong *et al.* (2006).

The average count of yeast was 3,83 ± 1,97 log cfu/gr in all treatments. Interaction of culture concentration and storage period significantly affected yeast count, in which the highest count was at 2.5% treatment stored for 20 days, which was 6.56 log cfu/gr and the lowest was at 2.5% treatment stored in 10 days, namely 2,23 log cfu/gr (Table 2). The count of yeast produced was lower than that of LAB. Yeast growth in cheese was evident with increasing cheese pH during ripening (Setyawardani *et al.*, 2012).

Table 2. Average Yeast Amount (log cfu/g) in cheese

Percentage of culture addition	Days			
	0	10	20	30
<i>L.rhamnosus</i> TW2+	7,33 ± 0,14 a	6,33 ± 0,40a	6,57 ± 0,75a	7,06 ± 0,32a
<i>L.plantarum</i> TW14, 2.5				
<i>L.rhamnosus</i> TW2+	3,88 ± 0,42 b	3,65 ± 0,41b	3,85 ± 1,10b	4,27 ± 0,56b
<i>L.plantarum</i> TW14, 5.0				
<i>L.rhamnosus</i> TW2+	4,11 ± 0,59 b	3,85 ± 0,39b	3,71 ± 0,49b	3,62 ± 0,65b
<i>L.plantarum</i> TW14, 7.5				
<i>L.rhamnosus</i> TW2+	4,49 ± 1,39 b	3,73 ± 0,39b	4,33 ± 0,30 b	3,81 ± 0,22b
<i>L.plantarum</i> TW14, 10				

Values bearing different superscript within columns shows significant difference (p<0.05).

Cheese pH observation during storage is presented in Table 3.

Table 3. Average cheese pH value

Culture addition percentage	Day			
	0	10	20	30
<i>L.rhamnosus</i> TW2+	6,51 ± 0,16	5,42 ± 0,20	5,10 ± 0,13	5,03 ± 0,20
<i>L.plantarum</i> TW14, 2.5				
<i>L.rhamnosus</i> TW2+	6,55 ± 0,20	5,51 ± 0,08	5,15 ± 0,05	5,03 ± 0,00
<i>L.plantarum</i> TW14, 5.0				
<i>L.rhamnosus</i> TW2+	6,43 ± 0,17	5,52 ± 0,08	5,28 ± 0,28	4,97 ± 0,13
<i>L.plantarum</i> TW14, 7.5				
<i>L.rhamnosus</i> TW2+	6,47 ± 0,11	5,53 ± 0,20	5,05 ± 0,50	5,05 ± 0,07
<i>L.plantarum</i> TW14, 10				

Values bearing different superscript within line and columns shows highly significant difference (p<0.01).

Thirty-day storage lowered cheese pH, started from day 10 through day 30, during which averaging from 4,97 ± 0,13 to 6,55 ± 0,20, with lowest pH at 7.5% culture and the highest at 5,0% culture without storage (fresh). During the last process of cheese production and initial storage, LAB formed lactic acid and the fermentation activity of lactose into lactic acid. pH product was also affected by added microbial development, so culture LAB addition with different percentage would result in different pH decrease. pH decrease for 21 day storage also occurred in Minas traditional cheese due to continual lactic acid production and other organic acid produced by probiotic started and culture (Buriti *et al.* 2007).

The average titratable acidity was 0,15 ± 0,06 (Table 4). Culture concentration period and storage period affected titratable acidity and its interaction. Lactic acid was metabolite product resulted from lactic acid bacteria. *L.plantarum* TW14 washofermentative LAB group so the main product was lactic acid. Titratable acidity increased after cheese was stored for more than 10 days and the highest was at 20-day storage. Twenty and thirty day storage also produced the highest LAB, namely 10,04 and 10,98 log cfu/gr, respectively. This result was also supported by decreasing pH value up to thirty day storage. Titratable acidity was related to the amount of LAB population.

Table 4. Average titratable acidity of goat cheese

Culture addition percentage	Days			
	0	10	20	30
<i>L.rhamnosus</i> TW2+	0,06 ± 0,03 a	0,15 ± 0,02b	0,23 ± 0,01c	0,21 ± 0,01bc
<i>L.plantarum</i> TW14, 2.5				
<i>L.rhamnosus</i> TW2+	0,06 ± 0,01a	0,11 ± 0,05ab	0,19 ± 0,02b	0,17 ± 0,02a
<i>L.plantarum</i> TW14, 5.0				
<i>L.rhamnosus</i> TW2+	0,07 ± 0,00a	0,11 ± 0,04ab	0,15 ± 0,04a	0,19 ± 0,03a
<i>L.plantarum</i> TW14, 7.5				
<i>L.rhamnosus</i> TW2+	0,07 ± 0,00a	0,13 ± 0,01ab	0,19 ± 0,05b	0,21 ± 0,01b
<i>L.plantarum</i> TW14, 10				

Values bearing different superscript within rows and columns show highly significant difference (p<0.01).

Conclusion

Cheese produced with indigenous probiotic culture of *L.plantarum* TW14 and *L.rhamnosus* TW2 stored for 30 days had the standard amount of LAB, yeast, pH value and titrated lactic acid according to SNI.

References

- Burns, PF, Patrignani D, Serrazanetti GC, Vinderola JA, Reinheimer R, Lanciotti, Guerzoni ME. 2008. Probiotic Crescenza cheese containing *Lactobacillus casei* and *Lactobacillus acidophilus* manufactured with high-pressure homogenized milk. *J Dairy Sci* 91: 500–512.
- Buriti FCA, Cardarelli HR, Filisetti Tullia MCC, Saad SMI. 2007. Synbiotic potential of fresh cream cheese supplemented with inulin and *Lactobacillus paracasei* in co-culture with *Streptococcus thermophilus*. *Food Chemist* 104 :1605–1610.
- Gardiner G, Ross RP, Collins JK, Fitzgerald G, Stanton C. 1998. Development of a probiotic Cheddar cheese containing human-derived *Lactobacillus paracasei* strains. *App Env Microbiol* 64: 2192 – 2199.
- Setyawardani. 2012. Karakteristik dan pemanfaatan BAL asal susu kambing untuk pembuatan keju dengan sifat probiotik. Disertasi, Bogor Agriculture University Indonesia.
- Sudarmadji, S., B. Haryono, and Suhardi. 1997. Prosedur analisis untuk bahan makanan dan pertanian. 4^{ed}. Liberty, Yogyakarta.
- Walstra P, Geurts TJ, Noomen A, Jellema A, van Boekel MAJS. 1999. *Dairy Technology: Principles Of Milk Properties And Processes*. New York: Marcel Dekker, Inc.

Production elasticity of broiler farming in Blitar Regency, East Java, Indonesia

*Budi Hartono**

Faculty of Animal Husbandry, University of Brawijaya, Jl. Veteran Malang 65145 Indonesia

Abstract

The objective of this research was to analyse some factors influencing broiler farming production and its elasticity. The research was conducted in Blitar Regency, East Java, Indonesia and sixty farmers of small broiler farming were chosen by purposive sampling method. Regression function of Cobb-Douglass model was used to determine factors that influenced broiler production and its elasticity. The results showed that number of broiler, feed cost, price of DOC, mortality and cost of production affected the production of broiler farm. Mortality showed negative elasticity but production cost showed unitary elastisty on broiler farming. The number of broiler, purchase of DOC, and feed costs is inelastic.

Keyword : elastisity, small broiler farm, production factors

**Corresponding author: budihartono_ub@yahoo.com*

Introduction

The company acts as a livestock management core to build into the plasma to be independent and more advanced. The character of commercial farming (mainly), is a sideline business and farms that are independent and partnership oriented to maximum advantage. Proper calculation and economic analysis are required to determine the appropriate farming efficiency in order to obtain maximum results. Aspects that need to be analyzed to determine costs and benefits include elasticity in terms of aspects of production (Umar, 2003). It is therefore necessary to conduct an analysis of the productivity of broiler breeding business. The research aims to determine the factors that may affect the production of broiler chicken farming and the elasticity of production of broiler farming in Blitar.

Materials and Methods

Theoretical Framework

The production process for the analysis of the factors affecting the production used a Cobb-Douglas function. Cobb-Douglas function is a function or equation that involves two or more variables, where one variable called the dependent variable, which is described (Y) and the other called independent variables, which explains (X). Analysis of the relationship between Y and X is by using regression method in which the variation of Y will be affected by variations in X. Cobb-Douglas function can be written as equation (Soekartawi, 2003; Beattie and Taylor, 1996; Debertin, 1986; Cramer et al., 1997):

$$Y = a X_1^b X_2^c \dots \dots \dots e$$

$$\ln Y = \ln a + b \ln X_1 + c \ln X_2 + e$$

Where :

- Y : Production
- X₁,.....X_n : Variable input
- e : error

Production elasticity

Elasticity of production is "The degree of sensitivity" production reflected by the presence of an additional percentage of the product for additional input one percent. Ep value greater than 1 indicates the process of production is in the area I, Ep value between zero and one indicates the process of production is in region II, and Ep value smaller than the zero (negative) shows the process of production is in the region III. Ep calculations using simple or multiple linear function by multiplying coefficient "b". In the form of Cobb-Douglas function, then the coefficient "b" already reflects Ep with the following evidence:

$$Y = a X^b$$

$$\frac{\partial Y}{\partial X} = b a X^{b-1} = b \frac{aX^b}{X} = b \frac{Y}{X}$$

$$b = \frac{\partial Y}{\partial X} \frac{X}{Y} = \text{is elasticity of production}$$

Location and Time Research

The study was conducted in the Gandusari, Binangun, and Garum, Blitar Regency in June-July 2014.

Sample Data Collection

Total sample of 60 broiler farmers selected by purposive sampling that broiler farmers who implemented the partnership program. The sample population in this study was people broiler farmers as the plasma in Blitar Regency. Samples taken are farmers who have livestock numbers 4.000-10.000 heads. The overall amount may be sampled population of 60 farmers as a representative sample.

Factors affecting production can be determined by using a model with a Cobb-Douglas equation:

$$Y = a X_1^{b1} X_2^{b2} X_3^{b3} X_4^{b4} X_5^{b5} X_6^{b6} X_7^{b7} X_8^{b8} X_9^{b9}$$

To facilitate the analysis, the model lineared with double log so that the model analysis into:

$$\ln Y = \ln a + b_1 \ln X_1 + b_2 \ln X_2 + b_3 \ln X_3 + b_4 \ln X_4 + b_5 \ln X_5 + b_6 \ln X_6 + b_7 \ln X_7 + b_8 \ln X_8 + b_9 \ln X_9$$

Where :

- Y = amount of production (kg/period)
- X₁ = number of chickens (head)
- X₂ = Day Old Chick purchase (IRD/period)
- X₃ = number of labor (people)
- X₄ = feed cost (IRD/period)
- X₅ = vitamins and medicines(IRD/period)

- X₆ = electricity cost (IRD/period)
- X₇ = Mortality (%/period)
- X₈ = produktion cost (IRD/period)
- X₉ = family size (people)
- a, b = estimated parameter

Results and Discussion

The amount of Broiler

The results showed that there was a positive correlation with the amount of production. Variable number of broiler chickens has a regression coefficient of 0.157 (Table 1) means that if the number of cattle increased by 1%, while other factors held constant, the number of production increased by 0.157% with assumed other factors held constant, thus inelastic.

Table 1. The elasticity use of production factors.

Dependent Variable	Independent Variables	Coefficient
Production amount (Y)	Constant	
	Chickens amount (X ₁)	0.157*
	Day Old Chick purchase (X ₂)	0.257*
	Labor amount (X ₃)	-0.069
	Feed cost (X ₄)	-0.540*
	Vitamins and medicines cost (X ₅)	-0.132
	Electricity (X ₆)	0.016
	Mortality (X ₇)	-2.037**
	Production cost (X ₈)	0.996**
	Family size (X ₉)	-0.076
	Value of R	0.702
	R Square (R ²)	0.752
	Adjusted R Square	0.735
	Value of F	52.087

Notes: ** p<0.01; * p<0.05

Purchase of DOC

Regression coefficient of day old chick (DOC) purchase variable obtained was 0.257 (Table 1), it can be interpreted that if the DOC costs increased by 1%, while other factors held constant, the amount of production will increase by 0.257% that means no elastic.

Feed Cost

Regression analysis showed that feed cost variable has a significant influence on the amount of broiler production farming. The amount of feed costs variable regression coefficient of -0.540 (Table 1), which can be interpreted that if the feed costs increased by 1%, while the number of production will decrease by 0.540% that means no elastic.

Mortality

Mortality variable has a significant influence on the amount of broiler production in partnerships broiler farming. This is indicated by regression coefficient of mortality variable amounts -2.037, that means the elasticity of mortality variable is elastic.

Production Cost

Variable production cost is very significant impact on the number of partnerships broiler production farming, it is associated with a regression coefficient of 0.996 (Table 1). This means that the cost of production is proportional to the amount of chicken production.

Conclusion

1. Mortality and production cost variables are the factors which significantly affect the number of broiler production, while the number of chickens, purchase of DOC, and feed cost are all factors that significantly affect the amount of broiler production.
2. Mortality is negative elastic, and production cost is unitary to broiler production. The number of livestock, purchase of DOC, and feed cost are factors that are not elastic to broiler production.

References

- Beattie, B.R. and C.R. Taylor, 1996. *Ekonomi Produksi*. Cetakan Kedua. Alih Bahasa Dr. Soeratno Josohardjono, MSc. Gadjah Mada University Press. Yogyakarta.
- Cramer, G.L., C.W. Jensen dan D.D. Southgate, 1997. *Agricultural Economics and Agribusiness*. Seven Edition. John Wiley and Sons. Inc.
- Debertin, D.L., 1986. *Agricultural Production*. Economic Macmillan Publishing Company, a division of Macmillan, Inc. New York.
- Soekartawi, 2003. *Teori Ekonomi Produksi dengan Pokok Bahasan Analisis Fungsi Cobb Douglas*. PT. Raja Grafindo Persada, Jakarta.
- Umar, H., 2003. *Studi Kelayakan Bisnis Edisi 2*. PT. Gramedia Pustaka Utama. Jakarta.

The differences of milk density and fat content in Tawang Argo Village compared with Indonesian National Standard

F. T. Saputra

Faculty of Animal Husbandry-University of Brawijaya, Malang 65145, Indonesia,

Abstract

There is still limited information about how much the different between milk quality in local farmer with Indonesian national standard (SNI). Based on it, this study aims to provide some information on how much the differences of milk quality between local milk with SNI. The location used on this study was Tawang Argo Village for one months. The investigation was using field study with total purposive sampling. Variable used was milk density and fat content compared and analyzed separately with Indonesian National Standard using paired t-test using SPSS 16. The result showed that there were high significant different between both milk density and fat content with SNI. For milk density, the different was $0.34 \pm 0.07\%$ below SNI standard while fat content was $48.62 \pm 12.25\%$ above the standard. In conclusion, fresh milk fat content had already met with SNI but not for milk density in Tawang Argo Village.

Keywords: fat content, fresh milk, Indonesian national standard (SNI), milk density

Corresponding author: firmansyahtrisp@ub.ac.id

Introduction

Fresh milk is a very valuable product from livestock and very useful for human. It is produced by dairy cattle including Holstein Friesian Cow. Holstein Friesian Cow in Indonesia is descendant from Native Holstein Friesian from Netherland and Local Cattle called Java cattle which already extinct today. According to Sulastri & Maharjan (2002) Dairy farming was introduced by Dutch in 1905 in order to fulfill Dutch Colony for milk demand and developed in mountain areas such as Boyolali, Salatiga, Ambarawa, Nongkojajar, Malang, and Batu. Unfortunately, quantity and quality of milk from Holstein Friesian in Indonesia is not as good as its ancestor. On this article, study will be specific on milk quality consisting of milk density and fat content. Holstein Friesian is Dairy Cattle with originally produce 3,660 liter milk production each lactation but only 3,660 liter milk each lactation with 3.7 % fat content in Indonesia (Susilorini, Sawitri, & Muharlieni, 2008).

There is international standard and national standard which determining milk quality. International standard is based on Food and Agriculture Organization or FAO, while national standard vary in each nations. In Indonesia, milk standard is based on SNI which created by Indonesian Government ("Standard National Indonesia" or Indonesian National Standard). SNI standard is also based on FAO Standard. According to Draaijer (2004), International fresh milk density standard was 1.028 to 1.033 g/ml, while fat content standard was 3%. Based on BSN (2011), Fresh milk density standard in Indonesia was 1.028 g/ml, while fat content standard was 3%. For fulfilling milk standard, there is a potential problem in local farmer which hold them to make good quality milk which based on SNI. There is also still limited information about this problem. Meanwhile, Tawang Argo Village has many local dairy farm that suitable to answer this question. Based on it, this study aims to provide some information on how many the differences in milk quality between local dairy farmer and Indonesian National Standard.

Materials and Methods

Materials used was fresh milk quality which recorded in 1 months. The investigation was using field study with total purposive sampling. Criteria in milk recorded was milk was successfully passed alcohol test and organoleptic test. Variable measured on this study was milk density and fat content compared and analyzed separately with Indonesian National Standard using paired t-test using SPSS 16.

Results and Discussion

On this study, data had been recorded in Tawang Argo Village for 1 months started in January 30th, 2015 to February 22nd, 2015. Fresh Milk collected had passed alcohol test and organoleptic test. Those two test is very important test which determine whether milk is good or not to be accepted.

Table 1. Paired Sample Test of Both Milk Density and Fat Content with SNI.

		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Milk Fat - Fat Standard	1.45860	0.36747	0.05604	1.34551	1.57170	26.028	42	0.000
Pair 2	Density - Density Standard	-3.36277	.0008611	.0001313	-.0036278	-.0030978	-25.609	42	0.000

Source: Primary data processed using SPSS 16.

After data collected and analyzed there were important result summarized in Table 1 and Table 2. Based on Table 1, the quality of fresh milk in Tawang Argo Village is very significant different with national standard ($P < 0.01$) for both milk density and fat content. Milk density and fat content become very important since they represent milk nutrition and also determining fresh milk basic price as explained by Draaijer (2004), milk fat and density was used to determine standard to pay milk.

Table 2. Comparison of Both Milk Density and Fat Content with SNI

	Tawang Argo	SNI	difference
Milk Density	1.023637 ± 0.0008611	1.027	$-0.34 \pm 0.07\%^{**}$
Fat Content	$4.4586 \pm 0.36747\%$	3%	$48.62 \pm 12.25\%^{**}$

** indicate significant difference ($P < 0.01$)

Source: Processed Primary Data

Based on table 2, milk density of fresh milk collected is $0.34 \pm 0.07\%$ lower than SNI standard (Tawang Argo 1.023637 ± 0.0008611 vs 1.027). It is important value since milk density become representation of milk nutriton beside fat. Milk nutrition represented in milk density including vitamins, protein, calcium, and other minerals. Guetouache, Guessas, and Medjekal (2014) said that milk consisted of water and total solid, but mostly several kinds of nutrient like protein carbohydrate, vitamin, and minerals in total solid. Based on Aquarius National Aeronautics and Space Administration or Aquarius NASA, water density was 1 g/ml (NASA, 2015). Based on this, the value of 1.027 means 1 g/ml water and 0.027 g/ml milk solid. This will explain low milk density means lower nutrition content and higher water content. Based on this, fresh milk in Tawang Argo has lower nutriton content than it should be if SNI standard become benchmark.

Fat content from fresh milk collected in Tawang Argo Village showed differend result with milk density. Based on Table 2, fat content $48.62 \pm 12.25\%$ higher than SNI (Tawang Argo $4.4586 \pm 0.36747\%$ vs 3%). Fat content describes taste and energy content inside milk. Based on Guetouache, Guessas, & Medjekal (2014), fat content was in globule form in milk and consist of saturated and unsaturated fatty acid which giving certain aroma and taste. The higher value of fresh milk content collected in Tawang Argo than SNI describe fat content already fulfilling

criteria in SNI.

In conclusion, milk density in Tawang Argo Village had lower value than SNI while fat content fulfilled SNI. The suggestion for further study is there is need of dairy cattle improvement based on milk density value added so fresh milk in local farmer stage can reach SNI standard in future.

References

- BSN, 2011. Indonesian National Standard: Fresh Milk-Chapter 1: Cow. SNI 3141.1:2011, Badan Standardisasi Nasional, Jakarta. Pp. 2.
- Draaijer, J., 2004. Milk Producer Group Resource Book: A Practical Guide to Assist Milk Producer Groups, Food and Agriculture Organization, Rome. Pp. 32-45.
- Guetouache, M., B. Guessas and S. Medjekal, 2014. Composition and Nutritional Value of Raw Milk. Review. Bio. Sci. and Pharm. Res. 2: 115-122.
- NASA, 2015. Measuring The Density of Water, Aquarius National Aeronautics and Space Administration, http://aquarius.nasa.gov/pdfs/meas_density_water.pdf (Accessed on Monday, May 15, 2015).
- Susilorini, T. E., M. E. Sawitri and Muharlieni, 2008. Farming 22 Potential Livestock Commodity, Penebar Swadaya Grup, Jakarta. Pp.84.
- Sulastri, E. and K. L. Maharjan, 2002. Role of Dairy Cooperative Services on Dairy Development in Indonesia A Case Study of Special Region of Yogyakarta, Int. Dev. 9:17-39.

Marketing analysis of broiler farming system on partnership scheme in Ponorogo Regency, East Java Province, Indonesia

Kusumastuti, A.E*¹

¹Faculty of Animal Husbandry, Brawijaya University, Veteran Street 65145, Malang City, East Java Province, Indonesia

Abstract

Broiler farming partnership become a popular farming scheme in Indonesia at nowadays. This research was conducted at broiler farms member of DMC partnership in Ponorogo Regency from March to April 2013. The purpose of these study were to determine marketing channel, marketing margin, and share profit. Method used in this research was case study. Data were collected by survey, observation and direct interview to farmers (plasma) and broiler traders. Results showed that the partnership scheme has three marketing channel namely: 1) farmers to broiler traders to small slaughterers to retailers to consumers, 2) farmer to broiler traders to small slaughterers to consumers, 3) farmer to chicken carcass traders to consumer. The first marketing channel has a highest (Rp. 7,500/kg) marketing margin than others. While the third marketing channel indicated a highest (69,4%) marketing efficiency.

Keywords: marketing channel, marketing margin, share profit, broiler farming partnership

Introduction

The potential development of the poultry farm commodities has relatively bright prospects, especially East Java Province has substantial resources in particular by-product agro-industry activities as well as a large enough market potential with a population of over 37 million inhabitants. Development efforts are generally associated with small and medium enterprises (SMEs) or co-operatives, through various partnership programs with various patterns which provided a considerable impact on the improvement of farm incomes and the quality of the people around them through a system of cooperation between farmers and industries with the purpose creation of business opportunities and employment, as well as ensuring the availability of animal protein for the society. In regard of this matters, Indonesian Government support is also manifested in the policies contained in the Decree of Agriculture Ministry 472/Kpts/ TN.330 / 6/1996 concerning development broiler farms containing instructions on the implementation of the system of cooperation through broiler business partnership (Anonymous, 1997).

Other factors caused broiler farm business become a leading of small business sector due to it has a wide connection (linkages) both of upstream (backwards) and downstream (forwards). In addition, this broiler farm has also vertically integrated and dynamic situation supported by a capital-intensive company with a modern management system. Livestock, because of their linkages with the overall farming system, make valuable entry points for wider agricultural development programmes. To exploit these opportunities, an integrated approach that combines both technical and institutional interventions is required (Steinfeld and Mack, 1995)

Contribution to the production of broiler meat in Indonesia from 2005 to 2007 always dominate (BPS, 2009). This is proven by the number of broiler production in the top position at 44.5 percent (yr. 2005) and reached 43.5 percent (yr. 2006). Moreover, according to

NationalStatistic Intitution (2009), the growth of broiler production around 10.55 percent (yr 2005 – 2006)and decreased by 6.64 percent (yr 2006 to 2007). Growth of broiler production describe the availability of market and growth rate of consumptionof commodities (Prasetyo, 2008).

One of the factors supporting the success of a broiler farm is well-executed marketing. Differences in marketing costs on producers and marketing agencies led to price differences on each level marketing agencies and produces the final price to be paid by the consumers (Kotler, 1999). The costs incurred will determine the price balance on manufacturers, marketing agencies and consumers, so that each get a decent profit in the operations and obtain the desired product. The purpose of this study were to determine the marketing channels, marketing margins, and profit share of each trader participating in thisbroiler's marketing activities.

Materials and Methods

The method used in this research was a case study. Determining the research location wasby purposive sampling based onbroilers population distribution where Ponorogo City was a center broiler farms in East Java Province (BPS, 2005).The research design was a survey and in- depth interviews. Primary data were obtained through structured questionnaires. While secondary data obtained directly to related institutions such as BPS, Department of Animal Husbandryas well as the private sector, such as partnerships broiler, poultry shop, broiler breeders association. Data were analyzed using descriptive and economic analysis.

Results and Discussions

According to provincial animal husbandry department, Ponorogo City included in one of region as pockets of broiler namely Bojonegoro, Lamongan, Mojokerto, Jombang, Nganjuk, Pamekasan, Blitar, Malang, and Lumajang (BPS , 2005). Annual data recorded from 2000 to 2004 showed that Ponorogo produced about 745 tons of chicken meat per year. In every yearbroiler meat was the largest contributor in the structure of chicken meat production in Ponorogo City which is about 70 % (± 520 tonnes year⁻¹) of the total, compared to broiler about 30 % (± 226 tonnes year⁻¹). The supply of broiler meat come from almost all regions (7 regions) in Ponorogo, namely Sampung District, Balong District, Slahung District, Kauman District, Sukorejo District, Bungkal District, and Jetis District. Therefore, it has the potential opportunities to developed in the wider scale and better management. In 2004, the District Slahung District was the largest contributor broiler meat supply in Ponorogo with a production of about 70 tons, followed by Balong District and Sukorejo District with each supply nearly 51 tons (BPS, 2005).

In regard of broiler marketing, it was company/partnershipresponsibilities. In other words, farmers were not allowed to sell broiler meat to wholesaler other than the merchant that was recommended by the company. In addition, farmers also should not choose marketing channels (traders) who has been in the company. This wasdue to all of the marketing process of broiler meat was the company/partnership responsibilities. Broiler marketing system and broiler marketing channel of small-scale and large scale farmers does not have differences. Traders who would buy a broiler meatwas an active trader who has been working with the partnership.

Result of research indicate there were three types of marketing channels, namely Channel I: Farmers - A big traders - small traders I - Retailers – Consumer; Channel II: Breeders - Traders large B - small traders II – Consumer; and Channel III consists of: Breeders - Wholesalers chicken carcass - Consumers. In general, demand for broiler meat comes from the brokers (wholesalers) from some surrounding cities such as Madiun, Magetan, Kartasura, Yogyakarta, Semarang and Jakarta.

Based on economic analysis showed that marketing margins on Channel I was the largest marketing margin around Rp.7.500,- per kg. While Channel II of marketing margin was around Rp.6.500,- per kg and the third channel was the smallest marketing margin which around Rp. 5.500,- per kg. Kotler (1999) stated that marketing margin represents the price of all services

received by a company marketing in organizing its functions and activities. Therefore, the more middlemen involved in the marketing channels will lead to increasingly enlarge the marketing margin. This was due to the high price difference in the level of each trader. That is why, the greater of marketing margin the higher the price paid by consumers.

Based on the percentage of the marketing margin, it can be determined the marketing efficiency. The highest marketing efficiency received by farmers was on Channel III by approximately 69.4 %.

Table 1. Marketing efficiency analysis received by farmers

Marketing Channel	Price at farmer level (Rp/Kg)	Price at consumer level (Rp/Kg)	Marketing efficiency received by farmers (%)
I	12.500	20.000	62,5%
II	12.500	19.000	65,8%
III	12.500	18.000	69,4%

While the profit share of each trader involved in marketing activities can be seen in Table 2 as follows:

Table 2. Profit share analysis of each trader

Marketing Channel	Profit (Rp/Kg)	Profit Share (%)
Wholeshares A	1.086,6	72,4
Wholeshares B	1.020	68
Wholeshares on broiler carcass	5.110	92,9
Small traders I	2.960	74
Small traders II	3.900	78
Retailers	950	47,5

Based on table 2, it can be seen that the highest profits share were on a broiler carcass wholesalers at 92.9 % while the lowest profit share was the retailers, namely 47.5 %.

Conclusion

It can be concluded that there were three types of marketing channels (Channel I, Channel II, Channel III). The largest of marketing margin was on Channel I around Rp.7500,- and the lowest was on Channel III around Rp. 5500,-. While the highest profits share were on broiler carcass wholesalers at 92.9 % while the lowest profit share was the retailers, namely 47.5 %.

References

- Kotler, P., 1999. Marketing Management in Indonesia. Salemba Publishing. Jakarta. Pp.
- Prasetyo, S., 2008. Substitution effect of commodity prices and complementary to broiler demand in Bogor Regency (*thesis*). Agriculture Faculty. IPB-Bogor. West Java Province.
- Steinfeld, H., and Mack, S., 1995. Livestock Development Strategies. In: word animal review (FAO) 1014-6954. No:84/85; p:83-84.

Physical properties and microstructure of dangkepipening, atraditional cheese of Enrekang Sulawesi Indonesia

Ratmawati Malaka*¹, Sudirman Baco², Kusumandari Indah Prahesti³

¹Laboratory of Biotechnology of Milk Processing, Hasanuddin University, ^{2,3}Department of Animal Production, Hasanuddin University

Abstract

This research aims to determine the ability of edible coating to increasing quality the physical properties and microstructure of Dangke. This study was use agar, bee wax and CMC as edible coating. Dangke was made by clotting of milk using papaya extract as a clotting agent. Curd was formed in coconut shells have perforated and pressing by spoon. Dangke as soft cheese dipped in the edible coating solution, and then ripening at 5°C during 30 days. Parameter was measured hardness, microstructure, and organoleptics. The Data were analyze by Random analyzes. The Result shows that using edible coating can extend the shelf life of Dangke and increase the hardness, and shows the microstructure more compact, furthermore panelist evaluated that Dangke was white color, smelly milk, and smooth texture.

Keywords: Dangke, Soft cheese, edible coating, physical properties, microstructure.

**Corresponding author: E-mail: malaka_ag39@yahoo.co.id; Ratihtamalag39@gmail.com*

Introduction

Dangke is a type of traditional cheese of Enrekang South Sulawesi, Indonesia. It is prepared traditionally by heating milk in open pan and clotting using papaya extract. The coagulated curd placed in perforated coconut shells for drainage of whey and pressing the curd by spoon (Aras, 2009; Malaka et al., 2009; Sukmasari, 2009). Cheese is the generic name for group of fermented milk-based food products, produced in a great range of flavors and forms throughout the world (Fox, 1993). Cheese has been defined by Davis (1976) as a product made from milk by coagulating the casein with rennet or similar enzymes in the presence of lactic acid produced by added or adventitious microorganisms, from which part of the moisture has been removed by cutting, cooking and/or pressing. According this definition so Dangke can be classified as cheese. Dangke is packed with a banana leaf and ready to be consumed or stored in refrigerator. Dangke is made mainly from fresh cow milk or buffalo milk that shelf life is generally two days at room temperature (Hatta *et al.*, 2013). Type of the cheese is classified as fresh cheese durability at temperatures of storage space very quickly spoilage. Dangke similar with 'Dali' a traditional fresh cheese from North Tapanuli, Medan, North Sumatera, Indonesia, made from cow's and buffalo milk using the existing local process, coagulated by pineapple extract and papaya sap (Sirait, 1991). Therefore it is necessary to improve the quality by coating technology in order to reduce the contamination of bacteria from the environment.

Dangke cheese just after its manufacturing is of significant commercial interest as a means of arresting physicochemical changes in cheese during ripening and to extend its self-life. In this study, the manufacture of cheese ripened using edible coating material is beeswax, Agar and CMC (Carboxymethyl cellulose). This research aims to determine the ability of edible coating to increasing quality of physical properties and microstructure of Dangke.

Material and Methods

Preparation of papaya latex and Dangke Manufacture

Milk was pasteurized at 75°C for 20 s, and adding the dried papaya latex of 0.5%. The coagulated milk was poured in the coconut shell with a hole at the bottom where the whey drainage through. After the whey was removed by pressing with spoon, Dangke removed from coconut shell template and dipped in bee wax, Agar, and CMC liquid for 5 s of every sample. The samples were subjected for physical properties analysis for hardness and organoleptics evaluation and microstructure. The manufacturing, hardness and sensory analysis were repeated three times.

Experimental design

A single piece of each cheese Dangke was produced for each sample of this study. There are three group i.e. Group I is Dangke coating by bee wax, Group II is Dangke coating by Agar and Group III is Dangke coating by CMC. Cheese Dangke loaves were cut into 5 x 5 x 5 cm block, sealed in plastic cheese packing bags and stored in refrigerator at 5°C for 30 days.

Hardness

Hardness Cheese Dangke measurement was use CD-Shear Force. The data was analyze by equation

Hardness (kg/cm^2) = $\frac{A}{L}$, where A=Loading stress; L= sectional area of sample, $\pi = 3.14$; r= radius of sample (0,635).

Organoleptics Evaluation

The product was evaluated by conducting sensory evaluation of panel of 30 semi-trained judges. The flavor (1-5), body and texture (1-5), color and appearance (1-5), modification according by Gaikwad & Hembade (2011).

Microstructure

Microstructure preparation methods were according the histological preparation methods (conventional methods) and view the microstructure by light microscopic magnification 1000 (objective 100 and ocular 10).

Data Analyses

Data was analysis by descriptive analysis according by Gasperz (1991).

Result and Discussion

Hardness

Fig. 1 shows the overage of Dangke hardness after ripening until 30 days at refrigerator and evaluated the value each 10 days. Random analysis indicated that the different until 30 days ripening shows significant result on Dangke Hardness, but by Agar Coating indicated the best result ($61.3 \text{ kg}/\text{cm}^2$). This kind of traditional cheese tended to become compact with progress of ripening.

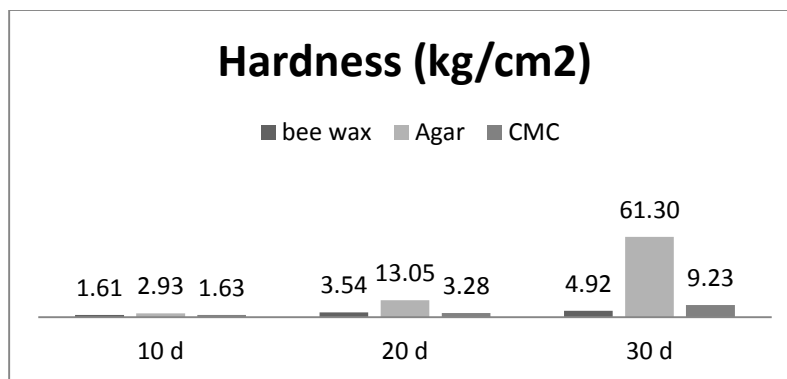


Fig. 1. Hardness of Dangke Cheese by different of coating and long life ripening.

Organoleptics evaluation

The results of sensory evaluation of Dangke samples are shown in Table 1. The ripening time and edible coating were not significantly ($P>0.01$) affected with increase in concentration hence panelists have given of score, but were significantly affected on flavor of Dangke. The product made with all processing due to color of Dangke product same with milk color (white color).

Table 1. Effect of edible coating on sensory quality of Dangke.

Attributes	Bee wax	CMC	Agar	SD	SE
Flavor	3.17	3.87	3.67	0.343	0.094
Body and texture	3.43	3.27	3.47	0.094	0.067
Color and appearance	3.97	4.07	4.03	0.083	0.047

Score: 1 = not good ----- 5 = very good ; SD=standard deviation; SE = Standard Error

The body and texture of Dangke after ripening to 30 days similarly with Mozzarella cheese that uniform, smooth and meaty (not too firm or too soft and pasty) (Mijanet al., 2010). The Dangke was made from cow milk, the cheese made from cow milk generally had appearance, smooth surface, free from cracks and practically free from moulds. The Dangkecolor made by coating Bee wax, CMC or Agar was similarly are yellowish-white and white, the difference might be due to carotene in milk.

Microstructure of Dangke

During the ripening process of Dangke at 5°C, then Dangke have a process of fermentation by lactic acid bacteria which are naturally present in milk. From a fermentation point of view lactose is the most important constituent of milk, which is in solution in the milk composed of one molecule the isomers α or β -glucose linked to one molecule of β -galactose. Lactose hydrolysis can be take place due to enzymatic reaction which is particularly important in the cheese making process. In the natural ripening the action of bacteria ferment the lactose to lactic acid, at the same time, may be responsible for the production of other substance such as acetylmethyl-carbinol and siacetyl which are responsible for flavor production. This causes the longer of ripening can increase the hardness of Dangke cheese.

During the press the curd in the coconut shell with a hole at the bottom as a template form of cheese Dangke (traditional template), the protein form into fibers in a roughly parallel manner. Consequently, fat and water accumulate between the long strands of protein. The microstructure of cross-sectionally samples were different between one edible coating and another edible coating. The microstructure of uncoating Dangke at 10 days after manufacture,

there were a large number irregular cavities dispersed randomly throughout the protein matrix. Some of these cavities may represent the site originally occupied by the extracted fat, and some were originally occupied by whey (Kuo & Gunasekaran, 2009).

The structure of cavities and protein matrix continued to change through 30 days post manufacture, thin strands of protein material encroached from all sides, connected the cavity walls that the spherical shape of fat globules were distinctly defined, but position of the fat globules does not change during aging. The water originally at the irregular cavities, so in the microstructure of Dangke appears to become absorbed into the protein matrix during the early stage of maturation. This was accompanied by a swelling of the protein matrix that continued until the spaces between the fat globules were completely filled with protein matrix as evidenced by the formation of reticular structure.

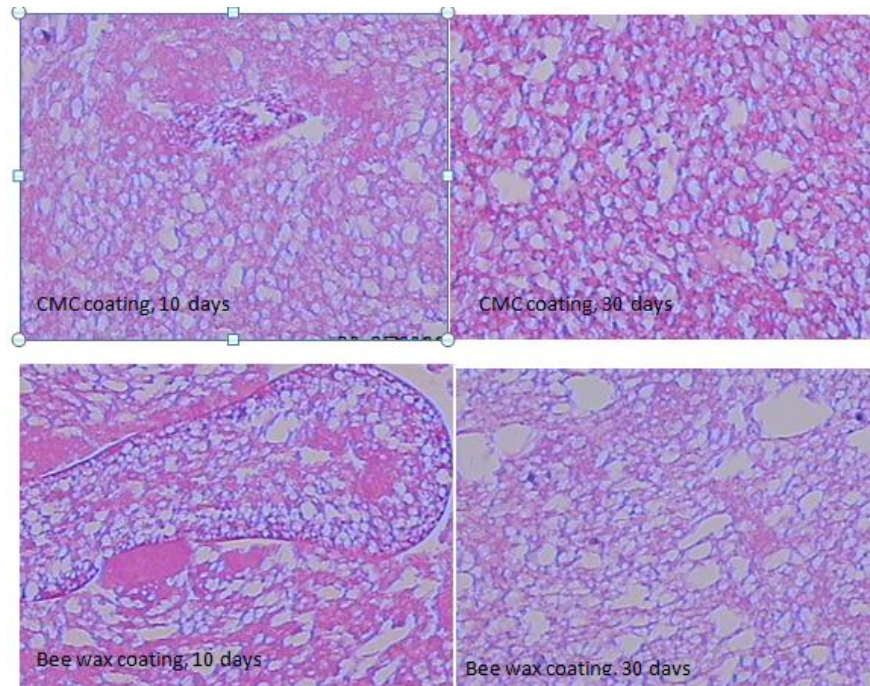


Fig. 2. Microstructure of Dangke cheese by different edible coating at 10 and 30 days ripening

The differences between the milk species (casein) in fat globules size are best appreciated by comparisons of the actual frequency distributions rather than by calculated average (El-Zeini, 2006). The structure of fat globules showed as imperfect spheres which could have length and width as imperfect rectangles. For all milk species, the average volume was the highest for buffalo milk fat globules ($344.9 \mu\text{m}^2$); cow ($32.28 \mu\text{m}^2$); Sheep ($28.29 \mu\text{m}^2$); goat ($18.65 \mu\text{m}^2$) and camel ($13.99 \mu\text{m}^2$) (El-Zeini, 2006).

Conclusion

Dangke quality can increase by enhance cheese-dangke-making which stipulated the concentration of papaya wax will be add and the ripening time. Using edible coating can extend the shelf life of Dangke and increase the hardness, and shows the microstructure more compact, furthermore panelist evaluated that Dangke was white color, smelly milk, and smooth texture.

Acknowledgements

Authors gratefully acknowledge the financial assistance receiver from Ministry of Education and Research and Technology and Hasanuddin University, Indonesia.

References

- Aras, W., 2009. Effect of crude papain level and temperature heating on Dangke Quality. Thesis. Faculty of Animal Husbandry, Hasanuddin University.
- Davis, J.G., 1976. Cheese. J and A Churchill Ltd, London, UK.
- El-Zeini, H.M., 2006. Microstructure, Rheological and Geometrical Properties of fat globules of milk from different animal species. Polish Journal of Food and Nutrition Sciences, 15 (56/2): 147-154.
- Fox, P.F., 1993. Cheese: An overview. In: P.F. Fox. (Eds), Cheese: Chemistry, Physics and Microbiology, p. 1-36. Chapman and Hall. London.
- Gaikwad, S.M. and A.S. Hembade, 2011. Standardization and Production of traditional Indian Milk product 'Ujanibasundi' from Buffalo milk. International Journal of Livestock Production, 2(8): 129-133.
- Gasperst, V., 1991. Metode Perancangan Percobaan. CV Armico. Bandung
- Hatta, W., M. Sudarwanto, I. Sudirman and R. Malaka, 2013. Prevalence and sources of contamination of *Escherichia coli* and *Salmonella* spp. in cow milk dangke, Indonesian Fresh Soft Cheese. Global Veterinaria 11 (3): 352 – 356.
- Kuo, M.I. and S. Gunasekaran, 2009. LWT Food Science and Technology 42: 9-16.
- Malaka, R. 2009. Mechanism of Gelation of Markisa Cheese by Physical Properties and Microstructure analysis. Paper of Seminar National Faculty of Animal Husbandry Padjadjaran University. Yogyakarta.
- Mijan, M.A., M.A. Haque and M.A. Wadud, 2010. The Bangladesh veterinarian, 27 (1): 36 – 42.
- Sirait, C.H. 1991. Evaluasi produk dadi di daerah Porsea-Sumatera Utara. Proceeding Seminar Nasional "Usaha Peningkatan Produktivitas Peternak dan Perikanan". Universitas Diponegoro. Semarang.
- Sukmasari, D. 2009. Effect of Ripening by *Lactococcus lactis* ssp. *lactis* 527 on Dangke Quality. Skripsi. Hasanuddin University.

Studies in urban geography of Laturcity of Maharashtra state of India with special reference to demographic features

Godbole B. M.¹ & Kankure K.B.²

¹ *Department of Geography, Mahatma Basweshwar College, Latur (MS) India,* ² *Principal, Maharashtra Udaygiri College, Udgir, Dist. Latur (MS) India*

Abstract

The present investigation was carried out to study and focus attention on the demographical problems of small town. The degree of the problems remains acute to a great extent ever in these small towns like Latur. Urban problems are acute because of lack of resources available with the municipal authorities. Various aspects of urban demographies have here been studied with the background of the region as a whole. It is concluded from the present investigation that the growth of population of Latur city shows ups and downs. The population distribution of Latur is not uniform throughout the city. In general the concentration of population is heavy in the CBD region and thins out to the peripheries from the core.

Keywords: urban geography, demography, sex ratio

Corresponding author: balasahebgodbole@gmail.com

Introduction

A city essentially distinguishes itself from a rural habitat by its population density characteristic and many other population attributes. Lack of accommodation, congestion in the city, existence of slums inadequacy of public utility services and unemployment, all are necessarily linked with the population problem. Population study offers an index of the cumulative effect of many factors in the growth of the city. Change in population is no doubt the result, but could be also a cause affecting city life (Sawant, 1978).

The study of the population of Latur city is very important because it promotes our understanding of the growth of the city. It also brings into focus the rapidly growing socio economic problems. In this research paper growth of population, density of population, sex ratio, etc. points are analyzed in detail.

Materials and Methods

Data used for the present research work was collected from both the sources i.e. primary and secondary. Secondary data was collected from different sources, basically from Gazetteers of Osmanabad and Latur district, District Census Handbooks of 1981, 1991 and 2001, Annual Reports published by Latur Municipal Council, Silver Jubilee Report of Latur district, Vision – 2032 Latur district, Town Planning Office of Latur, data regarding social and educational collected from Latur Municipal Council and Zilla Parishad, Latur. Primary data was collected through the especially prepared questionnaires, interviews with different officials and citizens of Latur and by visiting different sectors of Latur city. Different research methods employed for the calculation of population density, sex-ratio, population projection and other essential factors related with the research work.

Results and Discussion

As per sector wise distribution of population, 54474 out of the total 197408 persons are concentrated in sector one. This is maximum in Latur city. More than 1/4th population of Latur city concentrated in sector one. Remaining 3/4th population distributed in remaining 10 sectors. Lowest concentration of population observed in sector six. Only 1878 persons concentrated in this sector. More than 50% of the population of Latur city concentrated in three sectors i.e. sector one, sector three and sector four. Remaining 50% population concentrated in eight sectors.

Table 1. Demographic features of Latur city.

Sector	Population	Total Area (Hectare)	% Population to the total area	Density per Hectare	Sex ratio
Sector -I	52474	353.27	26.58	148.53	956
Sector -II	16820	190.32	8.85	88.37	952
Sector -III	25862	249.06	13.10	103.83	947
Sector -VI	21075	183.44	10.68	114.88	928
Sector -V	3432	317.45	1.74	7.66	936
Sector -VI	1878	198.48	0.95	9.46	914
Sector -VII	17708	212.30	8.97	83.41	929
Sector -VIII	16615	379.39	8.42	43.79	921
Sector -IX	11752	276.34	5.95	42.52	934
Sector -X	17105	714.83	8.66	23.92	944
Sector -XI	12687	181.37	6.43	0.41	935
Total	197408	3255.95	100.00	60.62	936

The density of population, usually calculated very crudely by following the P/A ratio, often leads to wrong conclusions. Latur is one such classic example. Latur, with a total municipal area of 3255.95 hectare has got a total population of 197408 in 1991 and thus per hectare density works out be 60.62 persons. It will be wholly wrong to jump to the conclusion that Latur population density poses no problem as out of a total urbanizable area of 3255.95 hectare only 798.51 hectare are urbanized. The rest area is yet to be inhabited. This has happened only because other periphery areas are either far away from the present CBD or present a less hospitable terrain compared to the existing urbanized areas.

A sector wise study of the sex ratio reveals that the numbers of females per 1000 males are below the average for the city in case of IV, VI, VII and VIII sector, while those for II sector and Gandhinagar stand higher than the average. The presence of a large number of industrial and commercial establishments in the VII, VIII and X sector and also a large number of hostels for male students in the VI and VII sector, is responsible for a comparatively low sex ratio in these sector, sector VIII has the lowest number of females per 1000 males, for the reasons mentioned above. Sector III, IV, V and X includes such suburbs as agricultural in character, and therefore a higher sex ratio is expected in these suburbs. A study of the sex ratio of Latur city shows that it has been changing unevenly from decade to decade. During the decade the spread of education was more rapid among the males than in female. Hence there was a preponderance of male students coming to the city from the rural areas. The addition of rural population to that of the city might have been responsible to some degree.

Conclusion

From the present investigation it is concluded that the growth of population of Latur city shows ups and downs. It will be wholly wrong to jump to the conclusion that Latur population density poses no problem as out of a total urbanizable area. A study of the sex ratio of Latur city shows

that is has been changing unevenly from decade to decade. During the decade the spread of education was more rapid among the males than in female. Hence there was a preponderance of male students coming to the city from the rural areas. The increase can be partly explained on the ground that the limits of Latur city were extended far and wide to include large rural areas in the neighborhood of the city. The addition of rural population to that of the city might have been responsible to some degree.

References

- Bharadwaj Prashant, AsimIjazKhwaja and Atif R. Mian, 2009. The partition of India: Demographic consequences, *J. Physical Anthro.*, 3(2):183-189.
- Census of India, 1991 and 2011.
- Gazetteer of Latur District, 1991 and 2011.
- Mallick, Javed and Atiqur Rahman (2012). Impact of population density on the surface temperature and micro-climate of Delhi, *Current Sci.*, 102(12):1708-1713.
- RoumiDeb, Bhatnagar P., and Avasthy D., 2012. Female foeticide in Delhi/NCR: Exploring the socioeconomic and cultural dimensions, *National J. Comm. Med.*, 3(3):548-550.
- Sawant, B.S., 1978. The city of Poona a study in Urban Geography.
- Socio economic Survey, 1991 and 2011.

Body growth of Thai wild boar (*Sus scrofa jubatus*) within a yearling age in a deep litter pig production system

Phoowadon Prapruetdee

*King Mongkut's Institute of Technology Ladkrabang, Prince of Chumphon campus,
Chumkoh, Pathiu, 86160, Thailand*

Abstract

The objective of this study was to investigate development of body growth of Thai wild boar (*Sus scrofa jubatus*) within a yearling age in a deep - litter pig production systems. A total of 10 piglets, at 151 days of age (5 months), at 11.51 kg of average body weight were placed into a deep - litter house, received commercial diet and mixed local roughage; used grated coconut, Napier grass banana stalk and palm kernel (20/80 as fed) and fill bedding materials with 5 parts of coconut coir, 5 parts of coffee hulls and 1 part of soil and topping by salts about 500 g and addition of a bacterial mixture as 30 cm height layer and repeated 2 times to completed 90 cm. height. At the end of the first phase of the experiment at 365 days of age (Yearling). The study reveal that the body weight of Thai wild boar in a deep - litter pig production systems were very low in all age categories. Mean body weight was 17.6 kg. While the maximum and the minimum body weights were 13.4 kg and 25.3 kg. The age categories of deep - litter pig wild boar, as similar to a confined wild boar study, could be defined as piglets (4 - 6 months old with 12.23 kg average body weight), young (7 – 11 months old with 16.64 kg average body weight), and the beginning of subadults (12 months old with 17.6 kg average body weight). The average final weight was 17.6 kg as presented as subadults. The average daily gain was 29 g during 214 days of the study.

Keywords: wild boar, body growth, deep – litter pig production, Sus scrofa jubatus

Introduction

Thai wild boars (*Sus scrofa jubatus*) have general characteristics with a wide range of body weight, elongated strong head and strong snout used for digging and a smaller rear part with thin legs and hooves, narrow face, small ears and big black eyes. Their piglets are light brown with lighter stripes across bodies and the bristly hair and the color will become black or brown in the adult (Tanomtong et al., 2007). Chhanganani & Mohnot (2004) reported that wild boars always rooting around wildlife sanctuaries and national parks, even if human habitations in gardens to find their own favors on food and plants which is in concordance with Prapruetdee (2013) that found that pigs fed only 50% commercial diet and 50% grasses/native vegetables, kept in a deep - litter pig production a low cost pig production system using indigenous microbial mixture in drinking water and sprayed on rice hull as bedding material, had the same growth performance as conventional concrete based house that fed 100% commercial diet. Therefore, this is interesting to keep wild boars in a deep - litter pig production due to their behaviors and their natural feed consumption.

The objective of this study was to investigate development of body growth of Thai wild boar (*Sus scrofa jubatus*) within a yearling age in a deep - litter pig production system.

Materials and Methods

Ten wild boar piglets (6 males and 4 females) were transported from their natural free range oil palm plantation at 4 months of age to adjust to a deep litter pig house. At the beginning of the study at 5 months of age, at 11.51 kg of average body weight were placed into a deep - litter house, filled bedding materials with 5 parts of coconut coir, 5 parts of coffee hulls and 1 part of soil and topping by salts about 500 g and addition of a bacterial mixture as 30 cm height layer and repeated 2 times to completed 90 cm. height. Wild boars received commercial diet and local roughage (20/80). Body weights were measured every 21 days and feed intakes were observed daily through the study. The observation collected from the beginning to the end of the first phase of the experiment at 365 days of age (Yearling).

Results and discussions

The body weight of Thai wild boar in a deep - litter pig production systems were very low in all age categories due to the 29 g average daily gain during 214 days of the study. Mean body weight (total) was 17.6 kg. Maximum and minimum body weights (total) were, 22.4 kg and 13.8 kg, respectively. Daily feed intake at piglets, young and subadults were 0.6, 1.0 and 1.2 kg/day, respectively.

Table 1. Mean live weight (in kg) and average feed intake (kg/day) of Thai wild boars within a yearling age in a deep - litter pig production system.

Age category	Sex	Deep – litter pig production system			
		Max	Average	Min	Average feed intake
Piglets (4 - 6 months)	♂	14.6	12.76	6.63	0.6
	♀	15.47	11.44	7.3	
	combined	15.03	12.23	6.97	
Young (7 - 11 months)	♂	21.11	17.78	12.29	1.0
	♀	21.43	14.43	11.23	
	combined	21.27	16.64	11.76	
Subadults (12 months)	♂	25.3	21.03	14.2	1.2
	♀	19.5	16.6	13.4	
	combined	22.4	17.6	13.8	

Maximum and minimum body weights of deep – littered, combined sex wild boars, were 13.03 kg and 6.97 kg, respectively. These were very different from the free – ranging wild boars from the study of Konjevic et al. (2008) that maximum and minimum body weights were 46 kg and 14 kg, this could be described as a behavior of wild boar in natural habitat that they probably sought a variety of feed to meet their nutrition need themselves (Chhangani & Mohnot, 2004). While the wild boars in this case of study, kept in a confined condition, were fed once a day with the same feed provided.

Conclusion

The age categories of deep - litter pig wild boar, as similar to a confined wild boar study (Mattioli & Pedone, 1995), as shown in Table 1, could be defined as piglets (4 - 6 months old with 12.23 kg average body weight), young (7 – 11 months old with 16.64 kg average body weight), and the beginning of subadults (12 months old with 17.6 kg average body weight). The subadult's age category in recent study was just at the beginning period (12 months). Therefore, mean live weight of wild boars kept in deep – litter house seemed lower than others.

References

- Chhangani, A. K and Mohnot. 2004 Crop raiding by wild boars (*Sus scrofa*) in and around aravalli, and its management in Rajasthan, India. Tigerpaper. 31(2): 385 - 389
- Mattioli, S and Pedone, P. 1995. Body growth in a confined wild boars population. IBEX J.M.E. 3: 64 - 65
- Chhangani, A. K and Mohnot. 2004 Crop raiding by wild boars (*Sus scrofa*) in and around aravalli, and its management in Rajasthan, India. Tigerpaper. 31(2): 385 - 389
- Prapruetdee, P. 2013. Conventional and deep-litter pig production system: the effects on growth performance. Proceedings of the 4th international conference on sustainable animal agriculture for developing countries. 27 - 31 July, 2013, Lanzhou, China. 396 – 397.
- Tanomtong, A., Supanum, P., Siripiyasing, P. and Bunjonrat, R. 2007. A comparative chromosome analysis of Thai wild boar (*Sus scrofa jubatus*) and relationship to domestic pig (*S. s. domestica*) by conventional staining, G-banding and high-resolution technique Songklanakarin J. Sci. Technol. 29(1): 1-13

Consumer attitude toward meat consumption in Ghana

Koizumi*¹, Seiichi

Department of Animal Science & Resources, Nihon University, 1866 Kameino, Fujisawa, Kanagawa 252-0880, Japan

Abstract

This study investigated consumer behavior for meat consumption in Ghana. Questionnaire survey were undertaken September in 2013 by placement method. Data was obtained from 28 respondents in University of Ghana. The data were mainly analyzed using chi-square test and multivariate analysis. The average preference rating of respondents was highest for fish, following by goat, bush meat, chicken, beef, pork, and sheep, which was not so high evaluation. Among the average estimated consuming frequency per week of meat and fish; fish was 2.02 times a week, most frequently, following by chicken, and other meat. And the consuming amount eating on each occasion was also fish was most consumed following by chicken. 39.3 percent of respondents did not eat bush meat at all (whereas proportion of respondents did not eat; pork and sheep-28.6%; fish-14.3%; goat-10.7%; chicken-7.3%; beef-3.6%). The average level of uneasiness for eating all of domestic and imported meat and fish indicated less satisfaction, which means respondents worried about safety of all of meat and fish sold. Domestic pork was lowest. There were no difference between each domestic meat and imported meat significantly. Average ratings (semantic differential scale=5points) on plans for future food consumption was highest for vegetable, following fruits, fish, and yoghurt, whereas other meat and food was rated below 3, which indicated low possibility on increasing meat and other animal products.

Keywords: meat consumption, consumer behavior, Ghana

Introduction

In Ghana, the production of all major livestock types used for human consumption increase over the last three decades (MOFA 2002), except pigs. However, the rate of development has been slow as expected. It created a situation whereby large volumes of cattle are imported to meet the demand. It is very important to increase domestic livestock sector for food security, providing good animal protein to people, and employment opportunities for a large part of population. To improve domestic meat consumption, we investigate the consciousness of people toward meat consumption.

Materials and Methods

The data used in this report were taken from pre-test on survey for meat consumption among staff and students of Ghana University. The questionnaire comprised 9 questions on meat and fish preference, using behavior for meat and fish and consumer consciousness about domestic and imported meat, food safety and future attitudes toward meat and other food consumption. Questionnaire surveys were undertaken 27-30, September in 2013 by placement method. Data was obtained from 28 respondents in University of Ghana (Male 21, Female 6, and NA 1).

Results and Discussion

Preference for meat and fish

The extent of preference for meat and fish was classified using a five-point semantic differential (SD) scale: 1=hate; 2=dislike; 3=average; 4=like; 5=love. The average preference rating of respondents was highest for fish (4.12 ± 0.18), following by goat (3.96), bush meat(3.89), chicken(3.85), beef(3.74), pork(3.59), and sheep(3.48), which was not so high evaluation.

Purchasing and using behavior

Table 1 Consuming frequency & amount of meat and fish

	Frequency times/week	Amount g/time
Fish	2.02 ± 0.42	148.21 ± 11.06
Chicken	1.86 ± 0.36	117.59 ± 9.88
Beef	1.27 ± 0.31	100.00 ± 9.45
Goat	1.21 ± 0.30	112.50 ± 8.38
Sheep	1.14 ± 0.32	95.00 ± 9.57
Pork	1.07 ± 0.33	96.74 ± 12.51
Bush meat	1.06 ± 0.41	79.00 ± 10.77

*Average \pm SE

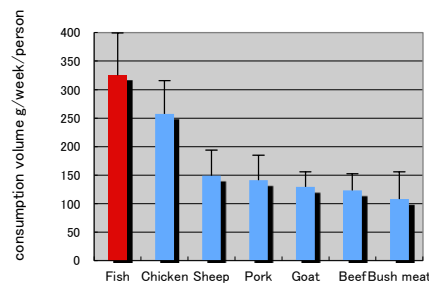


Fig.1 Meats and fish consumption

The average estimated consuming frequency per week of meat and fish were indicated in Table 1. The average estimated amount eating on each occasion for meat and fish were also indicated Table 1. From the result of frequency and amount eating on each occasion for meat and fish, average consumption was estimated as shown in Fig.1. 39.3 percent of respondents did not eat bush meat at all (whereas proportion of respondents did not eat; pork and sheep-28.6%; fish-14.3%; goat-10.7%; chicken-7.3%; beef-3.6%). Rate of respondents level of concern when they purchase each meat and fish were classified using a five-point SD scale: 1=do not care at all; 2=do not care; 3=average; 4=care a little; and 5=care a lot. The average important point index was highest for taste (4.82 ± 0.08), followed by expiration date(4.78), appearance(4.74), safety (4.65), freshness (4.63), and so on.

Safety consciousness

The level of risk perceived by respondents associated with eating domestic and imported meat were classified using a five-point SD scale: 1=worried a lot; 2=worried; 3=average; 4=not worried; and 5=not worried at all. The average level of uneasiness for eating all of domestic and imported meat and fish indicated under 3, which means respondents worried about safety of all of meat and fish sold. Domestic pork indicated by respondent was 1.84 ± 0.24 , which was lowest. There were no difference between each domestic meat and imported meat significantly.

Table 2 Safety consciousness for meat and fish

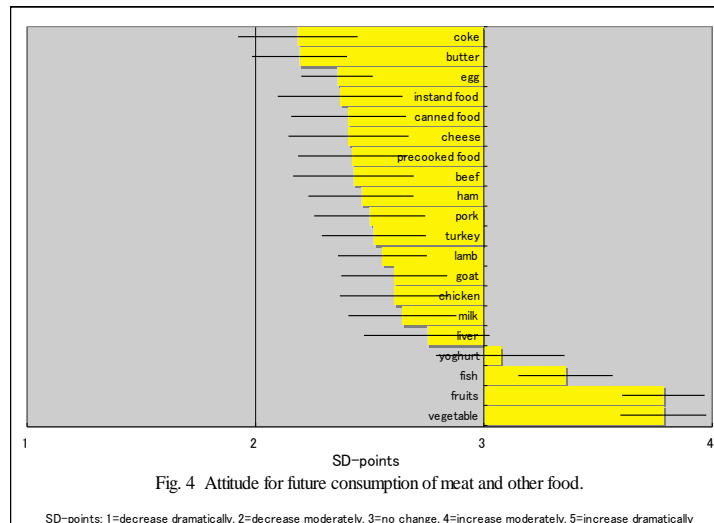
	Domestic	Imported
Fish	2.82 ± 0.24	2.54 ± 0.24
Sheep	2.50 ± 0.24	2.23 ± 0.24
Chicken	2.46 ± 0.24	2.19 ± 0.23
Goat	2.39 ± 0.23	2.31 ± 0.22
Bush meat	2.29 ± 0.21	2.32 ± 0.25
Beef	2.07 ± 0.20	2.04 ± 0.23
Pork	1.84 ± 0.24	2.05 ± 0.26

*Average±SE

** calculated from five-point SD scale: 1=worried a lot; 2=worried; 3=average; 4=not worried; and 5=not worried at all

Attitudes toward future consumption of meat and other food

A five-point SD scale was used to rate respondents on their plans for future consumption of meat and other food. The scale (consumer attitude index) was as follows: 1=decrease considerably; 2=decrease moderately; 3=no change; 4=increase moderately; and 5=increase considerably. Average ratings on plans for future food consumption was highest for vegetable, following fruits, fish, and yoghurt, whereas other meat and food was rated below 3 (Fig.4).



Conclusion

Koizumi et al. (2002) reported there were strong positive correlation between preference for meat and actual consumption. Therefore, it is important to improve evaluation for meat taste and quality in order to increase meat consumption. Evaluation for safety of both of domestic and imported meat were very low. Hence, to wipe out and to improve consumers' low evaluation is needed. In a previous study (Koizumi, Kobayashi, Pan et al. 2000); we reported the possibility of predicting future trends in food consumption by using a consumer attitude index from survey in Japan. Therefore, in the future, meat consumption could not increase in Ghana. Hence, to make promotion policy to link with consumption of meat are needed.

References

- Animal Production Directorate, Veterinary Service Directorate, Livestock Planning & Information Unit. 2002. Livestock development in Ghana Policies and Strategies, Ministry of Food and Agriculture, Ghana. pp 1-122.
- Koizumi, S., S. Kobayashi, In-Jen Pan, S. Takaku, M. Nishino, and M. Nagano. 2000. Study on consumer behavior for meat consumption in Japan (Tokyo, Osaka). *J. Anim. Sci.* 71:621-633.
- Koizumi, S., S. Kobayashi, In-Jen Pan, S. Takaku, M. Baba, R. A. Jussaume Jr., P. Riethmuller, J. Morison, J.C. Ryoo, C.W. Lin, I.H. Lin, H. Zhou, A. Ratanawaraha, and M. Nagano. 2002. Similarities and differences in meat consumption patterns in Asia and Pacific-rim countries. *J. Anim. Sci.* 73:293-300.

Effect of herbal immunostimulant formulation with Newcastle disease oral pellet vaccine on immune response in native chicken

Sathya¹, B., S. Murugesan², M. Veeraselvam³, K. Kanmani⁴, N. Sri Balaji*⁵ & G.Punitha⁶

^{1,6}Shri Sakthikailassh Women's College, Salem 636003, ^{2,3,4,5}Veterinary University Training and Research Centre, Bretts Road, Salem 636001, Tamil Nadu, India.

Abstract

Newcastle disease is endemic in many parts of the world and causes severe economic losses due to high mortality and reduced production. Vaccination has been reported as the only practical approach against endemic ND. In circumstances where the cold chain is weak or absent, the only reliable option will be the use of thermostable ND vaccines like oral pellet vaccine. Hence, the present study was designed to assess the effect on post vaccinated immune response of ND oral pellet vaccine in native chicken and correlation of immune responses with a herbal immunostimulant formulation. A total of 200 native chicks were allocated into 4 groups having fifty chicks in each group. The chicks in the first (I) group served as unvaccinated control and the chicks in the second (II) group was administered ND oral pellet vaccine produced by Tamil Nadu Veterinary and Animal Sciences University, India on day 15 through oral route manually. The chicks in the third (III) group was administered Immular[®]; a herbal immuno-stimulant formulation at the rate of 3ml/50 birds/day from day 15 to 36 in drinking water. The chicks in the fourth (IV) group was administered ND oral pellet vaccine on day 15 with Immular[®] of 3ml/50 birds/day from day 15 to 36. All the chicks were given feed and water *ad libitum* from day 1 to 36. Serum samples were collected on day 15, 22, 29 and 36 for antibody titre analysis through haemagglutination inhibition (HI) test. The results showed that the mean antibody titer of Group II, III & IV were elevated from day 22, but the difference was significant only on the day 29 and 36. The maximum level of antibody titre against ND was shown in group IV on day 36 which received oral Pellet vaccine supplemented with herbal immuno-stimulant formulation (Immular[®]). It could be concluded that the herbal immuno-stimulant formulation can improve the immune response of ND oral pellet vaccine.

Key words: Newcastle disease, immune response, herbal immuno-stimulant, native chicken

Introduction

Newcastle disease is endemic in many parts of the world and cause severe economic losses due to high mortality and reduced production. In India and as in many other developing countries Newcastle Disease (ND) is an endemic and a major problem in the commercial and rural poultry production (Spradbrow, 1992). Vaccination has been reported as the only practical approach against endemic ND (Usman, 2002). Newcastle disease oral pellet vaccine is a thermostable live vaccine and in circumstances where the cold chain is weak or absent, the only reliable option will be the use of thermostable ND vaccines (Ideris et al., 1987; Bensink and Spradbrow, 1999), or inactivated vaccines. The use of herbal formulation has many potential benefits, which includes modified host metabolism, immuno-stimulation, anti-inflammatory reactions, exclusion and killing of pathogens in the intestinal tract and reduced bacterial contamination (Rahimi et al., 2011). Hence, the need of the hour is to find a suitable herbal drug which constantly prevents birds from the threat of new infection, alleviates sub-clinical infections and acts as an immune

booster in a natural way. Thus, this study was undertaken to evaluate humoral immune response in native chickens by using oral pellet thermostable ND vaccine in combination with a herbal formulation.

Materials and Methods

A total of 200 native chicks were allocated into 4 groups having fifty chicks in each group. The chicks in the first (I) group served as unvaccinated control. The chicks in the second (II) group was administrated ND oral pellet vaccine containing D58 strain produced by Tamilnadu Veterinary and Animal Sciences University (TANUVAS) Chennai, India on day 15 through oral route manually. The chicks in the third group was administrated Immular[®]; a herbal immunostimulant syrup of TTK health care limited, Chennai at the rate of 3ml/50 birds/day from day 15 to 36 in drinking water which contains total water soluble extracts derived from *Commiphora mukal*, *Phyllanthus niruri*, *Tephrosia purpura*, *Eclipta alba*, *Withania somnifera*, *Piper longum*, *Trachydium lehmanni* and *Orchids latifolia*. The chicks in the fourth group was administrated ND oral pellet vaccine on day 15 with Immular[®] of 3ml/50 birds/day from day 15 to 36. ND oral pellet was administered to the birds manually at the rate of 1 pellet/bird. All the chicks were given feed and water *ad libitum* from day 1 to 36.

Blood samples were collected from the wing vein and allowed to clot in a test tube for each sample. Each sample of serum was separated by centrifugation of the sample at 2,000 rpm for 10 minutes and then were stored at – 20°C until further processing. Serum samples were collected on day 15, 22, 29 and 36 for antibody titer analysis through haemagglutination inhibition (HI) test as specified in OIE (2004) manual followed. The results were analyzed statistically with one way ANOVA test using the SPSS (version 15.0, Chicago.IL) and expressed as the mean ± SE.

Results and Discussion

The mean ± SE values of HI titre of the groups I, II, III and IV have been illustrated in the Table 1.

Table 1. The level of HI titre of the experimental native chicks.

Group	Day 15	Day 22	Day 29	Day 36
I	2.23 ± 0.90 ^{NS}	2.27 ± 0.89 ^{NS}	2.27 ± 0.86 ^{NS}	2.13 ± 0.88 ^{NS}
II	2.50 ± 1.07 ^a	2.70 ± 1.01 ^a	3.97 ± 1.14 ^b	4.33 ± 1.11 ^b
III	2.13 ± 0.91 ^a	2.30 ± 0.96 ^{ab}	2.32 ± 0.96 ^b	2.37 ± 0.96 ^c
IV	2.83 ± 0.97 ^a	3.57 ± 1.14 ^a	5.23 ± 1.14 ^b	6.40 ± 1.32 ^c

^{a,b,c}. The same letters in a row or column indicate no significant differences (P<(0.001)

Although the mean antibody titre of group II and IV were elevated from day 22, the difference was significant only on the day 29 and day 36. Meanwhile the group III showed significant difference only on day 36. Moreover group IV have appreciably higher antibody titre ($P \leq 0.01$) than other groups on day 29 and day 36. The highest titre was showed by Group IV on day 36.

The maximum level of antibody titre (6.40 ± 1.32) against ND was showed in group IV on day 36, which the chicks received ND oral pellet vaccine together with herbal immunostimulant formulation (Immular[®]), comparing with the chicks in the Group II (4.33 ± 1.11) and those in the Group III (2.37 ± 0.96), which received only ND oral pellet vaccine and herbal formulation respectively. The increased antibody titre may be caused by boosting of humoral response attained by the immuno-stimulatory effect of Immular[®]. Similar results have also been

observed by Raj et al. (2013) who stated that supplementation of herbal medicines during vaccination exhibit significant effect on antibody titre against ND in laying chicken. Likewise Garbaa et al. (2013) found that the group received live ND vaccine together with garlic extract showed better immune response than the group which received vaccine alone in broilers. The possible reason of increased level of antibody titre against ND in the chickens may be due to increased activity of neutrophils in blood after vaccination, which could play a major role in immune body production (Qamar et al., 2015).

Conclusions

Based on this study, it may be safely concluded that supplementation of the herbal immunostimulant formulation for native chickens can improve the immune response of ND oral pellet vaccine.

References

- Bensink, Z., and P. Spradbrow, 1999. Newcastle disease virus strain I-2 a prospective thermostable vaccine for use in developing countries. *Vet. Microbiol.* 68: 131–139.
- Garbaa, S, U. M. Merab, H. S. Garbab, U. Musac, A. A. Jimohd and A. A. Rajie, 2013. Effect of garlic and neemleef aqueous extracts on immune response of broilers to live Newcastle disease vaccine. *Sci. J. Vet. Adv.* 2(2): 16-20.
- Ideris, A., A. L. Ibrahim, P. B. Spradbrow and C. Hung Seng, 1987. Development of Food Pellet Newcastle Disease Vaccine. In: Copland J.W. (ed.): *Newcastle Disease in Poultry: A New Food Pellet Vaccine*. Canberra, ACIAR, pp. 20–23.
- OIE, 2004. *Manual of diagnostic tests and vaccines for terrestrial animals*. OIE publications. 2004.
- Qamar, S. H., A. Haq, N. Asghar, S. Rehman, P. Akhtar and G. Abbos, 2015. Effect of herbal medicine supplementations (Arsilvan super, Bedgen 40 and Hepa-cure Herbal medicines) on growth performance, immunity and haematological profile in broilers. *Adv. Zool Bot.* 3(2): 17-23.
- Rahimi, S., T. Zadesh, M. A. K. Torshini, R. Omidbaigi and H. Rokni, 2011. Effect of the three herbal extracts on growth performance, immune system, blood factors and Intestinal selected bacterial population in broiler chickens. *J. Agric. Sci. Technol.* 13: 527-539.
- Raj, P. M, D. Narahari, N. Sri Balaji, 2013. Enhancing the effectiveness of Newcastle Disease vaccination in the laying hens by herbal diet supplementation. *Int. J. Vet. Sci.* 2(3): 103-105.
- Spradbrow, P.,1992. A review of the use of food carriers for the delivery of oral Newcastle disease vaccine. In: Spradbrow P.B. (ed): *Newcastle Disease in village*. 13: 1-26.
- Usman, M., 2002. Effects of vaccination of chickens against Newcastle disease with thermostable V4 and LaSota vaccines using different grains and their brans as vehicle. Available at: Senne, D.A., King, D.J and Kapczynski D.R. (2004). *Control of Newcastle disease*. *Dev. Biol.*, 1(19): 165-70

Effects of probiotics supplementation on nutrient digestibility in captive asiatic elephants in temples of tamilnadu in india

Senthilkumar¹, A., M.G.Jayathangaraj², S.Gomathinayagam³, A.Valli⁴, A.Thangavelu⁵ & N.SriBalaji⁶

Tamil Nadu Veterinary and Animal Sciences University,
Madhavaram Milk Colony, Chennai-600051,
Tamil Nadu,India

Abstract

Probiotics are live microorganisms which have been found to confer a health on the host when administered in adequate amounts. This study was performed with the purpose of investigating effect of combinations of Probiotic preparation Genus *Lactobacillus* and Genus *Bifidobacterium* supplemented in temple elephants of Tamil Nadu. Before supplementation and after supplementation of probiotics, feed and dung samples were collected in 10 elephants the dry matter digestibility was estimated by lignin as a internal marker. Over all Mean \pm S.E. of dry matter digestibility in temple elephants before and after probiotics administration were 31.89 \pm 2.87 per cent and 42.65 \pm 2.55 per cent respectively. Dry matter digestibility values in per cent before and after the supplementation of Probiotic preparations were statistically highly significant variations ($P\leq 0.01$). Mean \pm S.E. of dry matter digestibility in young and adult temple elephants groups before and after probiotics administration were 18.49 \pm 4.18 per cent and 6.82 \pm 1.40 per cent respectively. Subsequent to the supplementation of Probiotics preparations, the dry matter digestibility between young elephant (n=4) group and adult elephant (n=5) group revealed statistically significant variations ($P\leq 0.05$).

Keywords: dry matter digestibility, elephants, probiotics

Introduction

In order to increase the improvement on nutrient conversion, researches are conducted on the feeding technology using new generation probiotics. Generally, probiotics is group of non pathogen microorganisms that have positive effect on physiology and health of gastro intestinal tract of its host if regularly consumed in sufficient quantities (Schrezenmeir and De Vrese, 2001 and Weichselbaum, 2009). Probiotics have been incorporated through diets, with the objective to keep intestinal microbiota balance of animals, preventing digestive tract diseases, improving feed digestibility, leading to a greater use of nutrients and improving animal performance (Fuller, 1992). Multi-strain or multi-species probiotics have been found to have more effective and consistent functionality than mono-strain or single-species probiotics (Timmerman *et al.*, 2004). Microorganisms producing lactic acid, such as *Lactobacilli sp.*, *Bifidobacteria sp.*, *Streptococcus sp.*, *Pediococcus sp.*, as well as yeast and filamentary fungi (Parvez *et al.*, 2006) are commonly used as probiotic preparations. The current study was carried out to evaluate the effects of combinations of Probiotic preparation Genus *Lactobacillus* and Genus *Bifidobacterium* supplemented in temple elephants of Tamil Nadu.

Materials and Methods

Feed (concentrate feed and fodder) and dung samples were collected from each temple elephant (Tiruchendur, Erattaitirupathi, Tirunelveli, Tirukkurugudi, Ilanji, Tiruvanaikovil, Tiruvaiyaru, Tiruvedaimaruthur, Tirunageswaram and Mannargudi) for this study continuously for three days and representative feed and dung sample (n=10) for each elephant was obtained from their respective pooled feed and dung samples. Samples were subjected to the estimation of moisture and lignin content, as per method of AOAC (2006). Using lignin content as an internal marker, the digestibility on dry matter basis was calculated in per cent.

Results and Discussion

Over all Mean±S.E. of dry matter digestibility in temple elephants before and after probiotics administration were 31.89±2.87 per cent and 42.65±2.55 per cent respectively (Table 1 and Figure 1). Dry matter digestibility values in per cent before and after the supplementation of Probiotic preparations were statistically highly significant variations ($P \leq 0.01$). Similar to the present study, Swanson *et al.* (2002); Kumagai *et al.* (2004) and Han *et al.* (2008) observed that administration of probiotics increased digestibility of dry matter, organic matter and crude protein. Moreover, others (Losada and Olleros (2002); Biradar *et al.* (2005); Jouany *et al.* (2008) and Moura *et al.* (2009) have reported the similar effects of enhancement of peristalsis, healthy microbial balance within the intestine, assimilation of different nutrients, in addition to providing of B vitamins, improved digestibility, regularity and re-absorption of proteins, fats and carbohydrates and improvement in the feed-efficiency.

Mean±S.E. of dry matter digestibility in young and adult temple elephants groups before and after probiotics administration were 18.49±4.18 per cent and 6.82±1.40 per cent respectively (Table 2 and Figure 1). Subsequent to the supplementation of Probiotics preparations, the dry matter digestibility between young elephant (n=4) group and adult elephant (n=5) group revealed statistically significant variations ($P \leq 0.05$). The significant variations ($P \leq 0.05$) observed between the selected age groups (Table 9) might be attributed to the change in metabolic events of the body pertaining to the age factor, variations in health status, feed composition of the feed materials offered, variations in the immunity level, type of probiotic microorganism; method and administered amount; host condition; intestinal microbiota condition etc.

Subsequent to the administration of Probiotics preparations, the dry matter digestibility in per cent were (Table 1 and Figure 1) increased in all the study elephants except in one elephant. Reasons for the absence of enhancement of dry matter digestibility in one elephant might be attributed to the factors like probable concurrent administration of antibiotic, concurrent existence of other disease causing pathogens, altered metabolic status of the individual due to multifaceted reasons etc.

The selection of Genus *Lactobacillus* as the component of the Probiotic preparations that were offered to the temple elephants through the concentrate feed materials in this study programme carried out was in agreement with the findings furnished by Coillie *et al.* (2007) who opined that oral administration of *Lactobacilli* inhibited the growth of *Salmonella enteritidis* and would also reduce the *Salmonella enteritidis* in the intestinal segments, possibly resulting in the reduced systemic infections.

Table1 Dry matter Digestibility of temple elephants (n=10) before and after Probiotics supplementation

Elephant No.	Age (Years)	Body weight (kg)	Ration fed	Digestibility Before probiotic supplementation	Digestibility After probiotic supplementation	Per cent difference
1.	11	1550	Rice-4kg, Horse gram-1kg, Nanal grass + Banyan tree leaves-125kg	36.85	54.84	17.99
2.	41	4600	Rice-10 kg, Nanal grass+ Coconut tree leaves-150kg	34.59	43.71	9.12
3.	15	3800	Rice-10 kg, Horse gram 2kg, Greengram-2kg Nanal grass+ CO3 grass+ Banyan tree leaves-250 kg	33.94	37.24	3.30
4.	54	3750	Rice-7kg,Ragi-1kg Nanal grass +Peepal tree leaves Banyan tree leaves-200kg	37.60	37.14	-0.46
5.	25	4240	Rice-10kg, Nanal grass + Peepal tree leaves Banyan tree leaves + Jampu tree leaves+ Coconut tree leaves -200kg	23.44	31.30	7.86
6.	12	2650	White flaked rice-5kg,Milk-2.5 litre,Rice-6kg,Horse gram-3kg Banyan tree leaves + Coconut tree leaves 250k	21.34	51.16	29.82
7.	8	2650	Rice-4kg,Ragi-4kg Nanal grass +Sorghum fodders-250kg	16.17	32.56	16.39
8.	39	4600	Rice-5kg,White Horsegram-1kg Nanal grass+ Banyan tree leaves-120kg	47.00	50.72	3.72
9.	10	2785	Rice-4kg,Brown flaked rice-4kg,Dates-3kg,Green gram-2kg,Ragi-2kg Nanal grass + Peepal tree leaves Banyan tree leaves+Coconut tree leaves200k	35.40	45.14	9.74
10.	15	3250	Rice-6kg,Green gram2kg,Ragi-2kg Nanal grass + Peepal tree leaves Banyan tree leaves +Coconut tree leaves 175kg	32.58	42.70	10.12
Mean ± S.E				31.89 ± 2.87	42.65 ± 2.55	

Paired t-test

Before			After		t – Test	P - Value	RESULT
N	MEAN	±SE	MEAN(Y)	±SE(Y)			
10	31.89	2.87	42.65	2.55	3.88	0.0037	**

Statistically highly significant ($P \leq 0.01$)

Table 2 Percent Dry matter Digestibility improvement in Temple elephants (n=9) subsequent to Probiotic supplementation between age groups

Less than 14 years group	Per cent - improvement
1	17.99
2	29.82
3	16.39
4	9.74
Mean	18.485
S.E.	4.179227
More than 14 years group	
Per cent – improvement	
1	9.12
2	3.3
3	7.86
4	3.72
5	10.12
Mean	6.824
S.E.	1.401148
“t” test	2.92
“p” value	0.0225*
Results	$P \leq 0.05$

Statistically significant ($P \leq 0.05$)

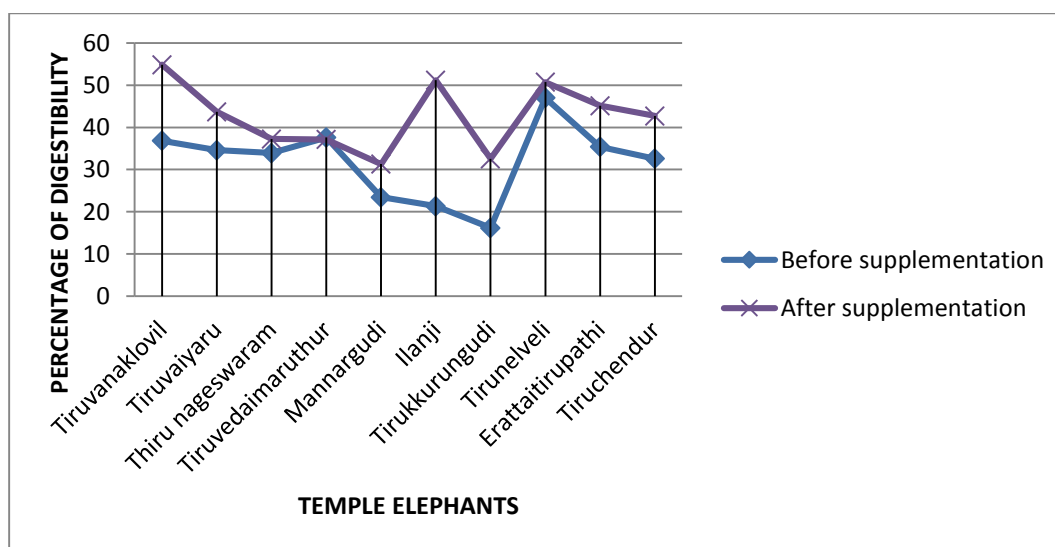


Figure 1 Comparison of Digestibility in per cent on dry matter basis before and after Probiotic supplementation

The administration of the Probiotic leading to overall enhanced dry matter digestibility in the elephants under study led to the conclusion that the elephants esp. with the low digestibility shall be supplemented with the Probiotic preparations containing Genus *Lactobacillus* as well as *Bifidobacterium*. This is indirectly supported by the report furnished by Fowler and Mikota (2006) who reported that the Asian elephants retained food longer in the digestive tract longer, relative to the body size and thus could achieve higher digestibility coefficient, when compared to the case with African elephants due to the ecological adaptations for the divergent dietary strategies and however, all the elephants were generally found to have a low apparent

digestibility. Hence, it might be highly beneficial to provide the supplementation of Probiotic preparations in elephant's esp. with low digestibility, Hence, the Probiotic preparation used in this study with the elephants that were reared in various temples of Tamil Nadu state is to be considered as more significant esp. in the captive elephants. The existence of highly significant variations ($P \leq 0.01$) of the dry matter digestibility in per cent during comparison of the values prior and after the administration of Probiotics (Table 1) might also be attributed to the beneficial effects of Probiotic organism-*Bifidobacterium* which was quoted to be a stimulant of the immune response and further, these organisms are associated with the effective reduction of growth of many potential pathogens as well as the prevention of diarrhoea and constipation, in addition to the improved intestinal functions.

References

- Schrezenmeir, J. And M. De Vrese, 2001. Probiotics, Prebiotics and Symbiotics-Approaching and Definition. *American Journal of Clinical Nutrition* 73: 361S –364S.
- Weichselbaum, E., 2009. Probiotics and health: a review of the evidence. *Nutrition Bulletin* 34:340-373.
- Fuller, R., 1992. Problems and prospects. In: *Probiotics – The scientific basis*, Fuller, R. pp. 377-386, Chapman & Hall, ISBN 0412408503, London
- Timmerman, H. M., C. J. M. Koning, L. Mulder, F. M. Rombouts and A. C. Beynen, 2004. Mono strain, multistrain and multispecies probiotics. A comparison of functionality and efficacy. *Int. J. Food Microbiol.* 96:219-233.
- Parvez, S., S.A., Malik Kang and H.Y. Kim, 2006. Probiotics and their fermented food products are beneficial for health. *Journal of Applied Microbiology*. V. 14. P. 1–15.
- AOAC, 2006. *Official Methods of Analysis*, (18th Ed.). Association of Official Analytical Chemists, Washington, DC.
- Swanson, K., C. Grieshop, E. Flickinger, L. Bauer, B. Wolf, J. Chow, K. Garleb and J. Williams, 2002. Fructooligosaccharides and *Lactobacillus acidophilus* modify bowel function and protein catabolites excreted by healthy humans. *The Journal of Nutrition*, Vol. 132, No. 10, pp. 3042-3050, ISSN 1760-4788
- Kumagai, H., S. Kumagai, M. Katsunosuke and E. Tsuyoshi, 2004. Effects of supplementary probiotics to two different diets on dry matter intake, daily gain, digestibility, ruminal pH and fecal microbial populations and metabolites in female goats. *Anim. Sci. J.*, 75:219-224.
- Han, Y., P. Paengkoum, X.L. Xia, and D.F. Wang, 2008. Effects of probiotics and by-pass fat on rumen metabolism and average daily gain of growing goats fed whole plant corn silage during dry season. *Chinese J. Anim. Nutr.*, 20(3):330-337.
- Losada, M.A. and T. Olleros, 2002. Towards a healthier diet for the colon: the influence of fructooligosaccharides and *Lactobacilli* on intestinal health. *Nutrition Res.*, 22(1-2):71-84.
- Biradar, S.S., S.T. Bahagvati and B. Shegunshi, 2005. Probiotics and Antibiotics: A brief overview. *Internet J. Nutrit. and Wellness* 2(1):1-6.
- Jouany, J.P., J. Gobert, B. Medina, G. Bertin and V. Julliand, 2008. Effect of live yeast culture supplementation on apparent digestibility and rate of passage in horses fed a high-fiber or high-starch diet. *J. Anim. Sci.*, 86: 339-347.
- Moura, R., E. Saliba, F. Almeida, A. Lana, V. Silva and A. Rezende, 2009. Feed efficiency in Mangalarga Marchador foals fed diet supplemented with probiotics or phytase. *Revista Brasileira de Zootecnia*, Vol. 38, No. 6, pp. 1045-1050, ISSN 1516-3598
- Coillie, E.V., J. Goris, I. Cleenweck, K. Grijspeerd, N. Botteldoorn, F.V. Immerseel, J. De Buck, M. Vancanneyt, J. Swings, L. Herman and M. Heyndrickx, 2007. Identification of *Lactobacilli* isolated from laying hens and characterization for potential use as probiotics to control *Salmonella Enteritidis*. *J. Appl. Microbiol.* 102:1095-1106.
- Fowler, M.E. and S.K. Mikota, 2006. *Biology, Medicine, and Surgery of Elephants*. Blackwell Publishing, London.

A retrospective investigation of co-endoparasitism of dog and cat patients in Nongchok subprovince, Thailand

Khumpool, G.

Faculty of Veterinary Medicine, Mahanakorn University of Technology, Bangkok, 10530, Thailand

Abstract

Between 2000 and 2013, dog and cat fecal specimens were analysed for saturated salt centrifugation-flotation and centrifugal sedimentation (n=103). The most commonly detected endoparasites on float and sediment techniques were Strongyle-type egg (40.9%) and *Isosporacanis* (31.8%) of dogs, in cats was Strongyle-type egg (33.3%). Moreover, the characters of co-infection were found 3 types in dog and only a type in cat patients. On the classification, the helminth eggs of the order Strongylida were found higher than the other of the both fecal pets. The Strongyle-type egg was detected in the highest of the proportion of samples in the both population study. This study highlights on the importance of specificity when interpreting diagnostic examinations of the illness domestic pets. Especially, the co-parasitism information could provide to prevention and treatment successfully.

Keywords: nongchok, endoparasitism, dog-cat, co-parasitism

*Corresponding author: kgrisada@hotmail.com

Introduction

The effect of endoparasite infection and the health of dogs and cats generally concerns blood loss, the obstruction of intestine and the loss of nutrients. Occasionally, transmammary and placental transmissions are also reported. Thus, better understanding of the diversity of endoparasite in endemic areas is essential for parasitic prevent and control programs. In Nongchok area, the recording of endoparasitism from owned pets is always rare and lack of clinical update. The endoparasitic prevalence was a wide variety of the formats that have been reported from the endoparasitism situation of 600,000 stray dogs in Bangkok area and that have also related to infection of the other pets in the same area (Sangvaranond, 2003). Additionally, the serious situation was similar to more than 1,485 stray cats in Bangkok area (Jittapalapong et al., 2007). Moreover, the major endoparasites: hookworms, Ascarid worm and protozoa from the stray dogs and cats could infect to human. For this reason, this can cause the risk of zoonosis situation directly. These methods are also economic and simple than the other techniques. Confirming the parasitological procedures of previous studies, Sangvaranond (2003) and Jittapalapong et al. (2007) have reported the use of two conventional techniques for the detection of endoparasitic eggs and protozoan oocysts or cysts in their research. The objective of this study was thus to determine the type of co-infection of endoparasite in the dog and cat patients in Nongchok are based on retrospective analysis of diagnostic examination results of Mahanakorn veterinary hospital and diagnostic center.

Materials and methods

From January, 2000 to December, 2013, faecal samples from 78 owned dogs and 25 cats that showed the sign of diarrhea or gastrointestinal tract disorder were collected from the rectum directly in Mahanakorn veterinary hospital and the samples were submitted to Mahanakorn veterinary diagnostic center. Each 5 g of fecal samples were put into plastic bags and bound at the top by elastic ring and kept in refrigerator at 5-7 °C until processing, which were carried out within 24 hours. The fecal samples were subsequently examined for two times by using centrifugal flotation technique (Jittapalapong et al., 2007) and centrifugal sedimentation technique (Sangvaranond, 1998) for investigation and classification of the endoparasite eggs as well as protozoan oocysts or cysts. The samples were analyzed using the centrifugal flotation technique in saturated NaCl solution and centrifugal sedimentation technique (Sangvaranond, 1998). Evolutive forms were identified according to their morphological characteristics and by micrometric measurement. The results of the coproscopic were recorded and analyzed by Microsoft Access 2007. The endoparasite eggs and protozoan oocysts/cysts found in this study were expressed as number and percentage of prevalence.

Results

In this investigation, the overall prevalence of endoparasite eggs and protozoan oocysts/cysts in owned dog patients of Mahanakorn veterinary hospital was 28.2% (22/78). The single infective type included 3 types: with Strongyle-type egg, *T. canis* egg and *I. canis* oocyst, respectively. This study presented that the dogs infected with Strongyle-type egg was the highest 40.9%. Moreover, the results also showed that the co-infective type with more than one species were 3 types and the all diversity co-infective types were Strongyle-type egg+*Giardia* cyst, Strongyle-type egg+*I. canis* and *T. vulpis*+*S. lupi* (Table 1). For distribution of positive samples and parasite species by the local endoparasite diversities were Strongyle-type, *T. canis*, *T. vulpis*, *S. lupi*, *I. canis* oocyst and *Giardia* cyst. The results presented that the prevalence of Strongyle-type egg was the highest 40.9% and with *I. canis* oocyst was 10.3 %, respectively (Table 2). The overall prevalence of endoparasite eggs and protozoan oocysts/cysts in owned cat patients was 24% (6/25). The single infection of cats was as in the dogs that included 3 types: with Strongyle-type, *G. spinigerum* and *I. felis* oocysts. The single infection with only one species Strongyle-type egg was the highest 33.3%. These results shown that the co-infection was one type: Strongyle-type egg+ *T. cati* (Table 3). The local endoparasite diversities were Strongyle-type, *T. cati*, *G. spinigerum*, *I. felis*. The results presented that the prevalence of Strongyle-type egg was the highest 12% (Table 4). The classification on the orders of endoparasite eggs were the highest on the order Strongylida (Strongyle-type eggs) 46.4%, 39.3 % on Protozoa (oocysts/cysts) and 10.7% on order Ascaridida, respectively (Figure 1).

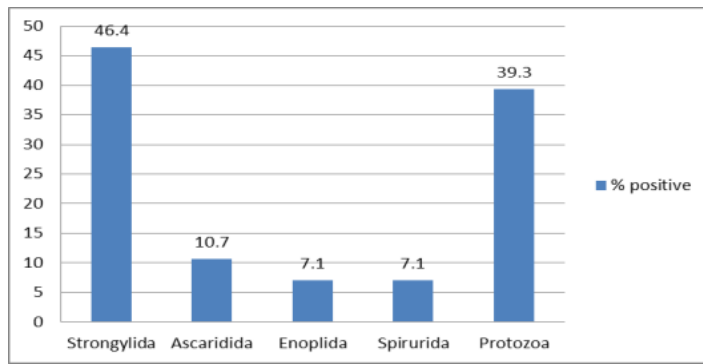


Figure 1. Positive sample percentages of both species patients on the orders of endoparasite classification (n=28).

Table 1 Characters of endoparasite infections of positive dog patients (n=22).

No.	Endoparasites (eggs)	Positive samples	Positive number (%)
1	Strongyle-type	9	40.9
2	Strongyle-type + <i>Giardia</i> cysts	1	4.6
3	Strongyle-type + <i>I. canis</i> oocysts	2	9.1
4	<i>T. vulpis</i> + <i>Slupei</i>	1	4.6
5	<i>T. canis</i>	1	4.6
6	<i>I. canis</i> oocysts	7	31.8

Table 2 The local endoparasite diversities in dog fecal samples, submitted (2000-2013) to the MVDC for saturated salt centrifugal flotation and centrifugal sedimentation techniques (n=78).

No.	Endoparasites (eggs)	Positive samples	Positive number (%)
1	Strongyle-type	10	12.8
2	<i>T. canis</i>	2	2.6
3	<i>T. vulpis</i>	2	2.6
4	<i>Slupei</i>	1	1.3
5	<i>I. canis</i> oocysts	8	10.3
6	<i>Giardia</i> cysts	1	1.3

Table 3 Characters of endoparasite infections of cat patients (n=6).

No.	Endoparasites (eggs)	Positive samples	Positive number (%)
1	Strongyle-type	2	33.3
2	Strongyle-type + <i>T. cati</i>	1	16.7
3	<i>G. spinigerum</i>	1	16.7
4	<i>I. felis</i> oocysts	1	16.7

Table 4 The local endoparasite diversities in cat fecal samples, submitted (2000-2014) to the MVDC for saturated salt centrifugation-flotation and centrifugal sedimentation (n=25).

No.	Endoparasites (eggs)	Positive samples	Positive number (%)
1	Strongyle-type	3	12
2	<i>T. cati</i>	1	4
3	<i>G. spinigerum</i>	1	4
4	<i>I. felis</i> oocysts	1	4

Discussion

An understanding of the parasitic diagnosis, the fecal examination is the essential method for investigation the Helminthic eggs inside the hosts (Sangvaranond, 1998). The results of the coproscopic examinations were recorded from parasite section data base of Mahanakorn veterinary diagnostic center. In this study, there was not at all allowed to necropsy the owned dogs and cats. The results reported that the co-infection of endoparasites of the order Strongylida was the optimal helminthic diversity that included a lot of nematodes in the same group. After the fertilization, the female nematodes lay the typical egg "Strongyle-type egg" that egg have the similar morphology of all species in this order. In 1998 and 2007, Sangvaranond & Jittapalaponget al. reported that the strogyle-type eggs were detected in the fecal samples of stray dogs and cats and the percentage detection was higher than the other as this study. In this study, *T. vulpis* (whipworm) eggs were also detected and the percentage of

prevalence was nearby to *T. canis* (2.6%) and *S. lupi*(1.3%) as well. These results completely agree with the data reported in dog by Pullola et al. (2006) that the *T. vulpis* prevalence of 541 owned dogs and stray dogs in Finland was 0.2% and *T.canis* was 3.1% from 296 owned dogs. Additionally, Sangvaranond (2003) presented that the worms *S. lupi* and *G. spinigerum*were detected in oesophageal wall and the abnormal mass in stomach, respectively and the protozoan *I. canis* oocysts was also identified in the fecal samples of stray dogs in Bangkok areas. Another more,in this study presented that the percentage of *Giardia* cysts was 1.3%. This result was agree with data of Riggioet al.(2013) that reported the owned dogs in the central Italy were detected *G. duodenalis* 3.8% from the fecal sample. In the examined results of owned cats, the *T.cati* eggs and protozoan *I. felis*oocysts were found in the similar percentages. This correlated with the observations of McGladeet al. (2003) in Perth cats, Western Australia and Jittapalaponget al. (2007) in Thai stray cats. Moreover, many protozoan *Isospora* spp. oocysts were reported in German young cats up to one year of age also (Barutzki&Schaper, 2013). This study showed the examined results thatthelocal endoparasitediversity were two types: a single and co-infections. The single infection by endoparasitism in owned dogs included with Strongyle-type *T. canis*, *T. vulpis*, *S. lupi*, *I. canis*oocysts and *Giardia* cysts. The co-infections were infected by Strongyle-type +*Giardia* cysts, Strongyle-type + *I. canis* oocysts and *T. vulpis* + *S. lupi*, respectively. In the owned cats, the single infection types were with Strongyle-type, *T. cati*, *G. spinigerum*, *I. felis* oocysts, respectively. Theco-infections were only infected by Strongyle-type+*T. cati*. These results indicate that the owned dogs and cats with diarrhea or gastrointestinal tract disorder in Nongchok area should have the combine treatment with against protozoan drugs and helminthocides and the zoonotic character of some nematodes found in this study must serve pet owners and veterinarians.

Acknowledgements

Author thanks Mahanakorn Veterinary Hospital and Mahanakorn Veterinary Diagnostic Center for their cooperation and Dr. Sitthichon Rattanachan for reviewing and proofreading.

References

- Barutzki, D. and Schaper, R.2013.Age-Dependant Prevalence of Endoparasites in Young Dogsand Cats up to One Year of Age.Parasitol. Res.112:112-131.
- Bowman, D.D. and Lynn, R.C.1999.Georgis' Parasitology for Veterinarians.7 thed.W.B.Saunders Company.Philadelphia.U.S.A.414 pp.
- Hoopes, J.H., Polley, L., Wagner, B. and Jenkins, E.J.2013.A retrospective investigation of feline gastrointestinal parasites in western Canada.Can. Vet. J.54:359-362.
- Jittapalapong, S., Inparnkaew, T., Pinyopanuwat, N., Kengradomkij, C., Sangvaranond, A. and S. Wongnakphet, 2007.Gastrointestinal Parasites of Stray Cats in Bangkok Metropolitan Areas,Thailand. Kasetsart J.(Nat. Sci.)41:69-73.
- Kaufmann, J.1996.Parsitic infections of Domestic animals: A Diagnostic Manual. Birkhaeuser Verlag.Berlin.Germany.423pp.
- Lapage, G., 1962. Mönning's veterinary helminthology and entomology, 5th end. Baillie`re,Tindall&Cox,London,UK.1182pp.
- McGlade, T.R., Robertson, I.D., Elliot, A.D., Read, C. and R.C.A., Thompson, 2003. Gastrointestinal parasites of domestic cats in Perth, Western Australia. Vet.Parasitol.117:251-262.
- Pullola, T., Vierimaa, J., Saari, S., Virtala, A.M., Nikander, S. and A., Sukura, 2006.Canine intestinalhelminths in Finland: Prevalence, risk factors and endoparasite control practices. Vet. Parasitol.140:321-326.

- Riggio, F., Mannella, R., Ariti, G. and S. Perrucci, 2013. Intestinal and lung parasites in owned dogs and cats from central Italy. *Vet. Parasitol.* 193:78-84.
- Sangvaranond, A., 1991. Internal parasites of dogs and cats in Bangkok Metropolitan and Vicinity areas. *Thai. J. Hlth. Resch.* 6(1):21-31.
- Sangvaranond, A., 1992. *Veterinary protozoology: Internal organ Protozoa.* 4th ed. Kasetsart University. Bkk. 318pp.
- Sangvaranond, A., 2000. *Veterinary Parasitology.* Kasetsart University. Bkk. 412pp.
- Sangvaranond, A., 2003. Helminths among Stray Dogs in Bangkok. *Kasetsart Veterinarians.* 13(1):30-42.
- Sommerfelt, I.E., Cardillo, N., Lopez, C., Ribicich, M., Gallo, C. and A., Franco, 2006. Prevalence of *Toxocaracati* and other parasites in cats' faeces collected from the open spaces of public institutions: Buenos Aires, Argentina. *Vet. Parasitol.* 140:296-301.
- Urquhart, G.M., Aromor, J., Duncan, J.L., Dunn, A.M., Jennings, F.W. 1987. *Veterinary Parasitology Logman scientific & technical.* New York, U.S.A. 133 pp.

Quality of chicken eggs in the flea market, KamphaengSaen district, NakhonPathom province, Thailand

Rattanatabtintong^{*1}, S., O. Songserm¹, K. Thoosai¹ & J. Mitunawong¹

¹Department of Animal Science, Faculty of Agriculture at KamphaengSaen, Kasetsart University, KamphaengSaen campus, KamphaengSaen district, NakhonPathom province, 73140, Thailand.

Abstract

The objective of this study was to evaluate the quality of chicken eggs sold in the flea market. One hundreds of chicken eggs were randomly sampled from each of ten (10) flea markets located in the area of KamphaengSaen district, NakhonPathom province, Thailand. The quality of egg was evaluated in terms of albumen height (mm.), haugh unit, albumen pH, yolk color, egg shell thickness (mm.), specific gravity, and the ratio (%) of yolk, albumen, and egg shell per egg. The results showed that the values of mean (\pm SD) and maximum-minimum of albumen height, Haugh unit, albumen pH, egg shell thickness, specific gravity, and yolk color were 4.13(\pm 0.45) and 5.89-2.69mm., 56.05(\pm 6.39) and 74.26-36.47, 9.19(\pm 0.02) and 9.39-8.34, 0.36(\pm 0.03) and 0.38-0.35mm., 1.089(\pm 0.01) and 1.100-1.072, and 10.55(\pm 0.77) and 12.00-9.00, respectively. Additionally, the average values (\pm SD) of the ratio (%) of yolk, albumen, and egg shell per egg were 27.76 \pm 0.59, 59.55 \pm 0.31, and 12.69 \pm 0.24, respectively. The parameters of albumen height and Haugh unit are important keys to describe the freshness of edible egg. The result of this study indicates that chicken eggs sold in the flea markets, settled temporarily outdoors, have high variations of freshness. These are possibly affected by the factors of temperature control and time during the transportation, storage, and distribution.

Keywords: chicken egg, quality, Haugh unit, flea market

**Corresponding author: agrsura@ku.ac.th*

Introduction

Chicken egg is a crucial nutritious source for consumers and the most popular choice for consumption. In Thailand, there are different distribution systems of eggs from layer farm to market (Issaree et al., 2011). The retail eggs are currently distributed in the supermarket, fresh market, farm shop and flea market. Nowadays, quality of egg has been concerned by consumers. The grading for quality of eggs is the classifying of the individual egg according to established standards. In the U.S., standards for quality of individual eggs have been developed on the basis of such interior quality factors as condition of the albumen and yolk, size of the air cell, and the exterior (USDA, 2000). The Internal egg quality is affected by various factors, such as temperature, and period of transportation and storage. The purpose of this study was to evaluate the quality of chicken eggs distributed in the flea markets, KamphaengSaen district, Nakhon Pathom province.

Materials and Methods

Egg sample collection

A total of 1,000 chicken eggs were purchased from the flea markets. One hundreds of chicken eggs were randomly sampled from each of ten (10) flea markets located in the area of KamphaengSaen district, NakhonPathom province, Thailand. Each set of chicken egg samples was labeled with the location of flea market and purchase date. All samples were directly transferred to the laboratory and immediately evaluated for egg quality.

Egg quality and egg composition measurements

Individual egg sample was weighed by digital scale. The specific gravity of egg was subsequently evaluated. The egg was broken and then albumen height was measured by digital albumen height gauge and yolk color score was determined by the Roche yolk color fan. Haugh unit value was then calculated (Haugh, 1937). The egg yolk was separated from albumen. The egg yolk, albumen, and shell were weighed by digital scale. Therefore, the ratio (%) of egg composition was calculated. Albumen pH was measured by pH metermodel HI 111 (HannaInstruments, USA).

Statistical analysis

All data of egg quality and ratio of egg composition were analyzed by using descriptive statistical method. The values were shown as mean, standard deviation, maximum, minimum and coefficient of variation.

Results and Discussion

In this study, chicken eggs sold in the flea market, a type of bazaar that temporarily rents space to people who want to sell goods, items, agricultural produces and products, were directly bought from layer farm or egg collecting and grading center. The results of interior egg quality and egg compositions showed that the values of mean (\pm SD) and maximum-minimum of albumen height and Haugh unit were $4.13(\pm 0.45)$ and $5.89-2.69$ mm., and $56.05(\pm 6.39)$ and $74.26-36.47$, respectively (Table 1). Additionally, the average values (\pm SD) of the proportion of egg yolk, albumen, and shell per egg (%) were 27.76 ± 0.59 , 59.55 ± 0.31 , and 12.69 ± 0.24 , respectively (Table 2). The parameters of albumen height and Haugh unit are important keys to describe the freshness and internal quality of edible egg (Kul & Seker, 2004). The high variations of both variables ($>10\%$) were observed in this study. Scott & Silversides (2000) reported that the albumen heights of chicken eggs stored more than 10 days decreased from 9.16 to 4.75 mm. Chicken eggs kept in a long period had higher albumen pH (Samliet al., 2005). These are possibly affected by the factors of temperature control and time during the transportation, storage, and distribution. Control of these conditions potentially keeps high quality of edible egg sold in the market.

Table 1. Quality of chicken eggs sampled from 10 flea markets in KamphaenSaendistrict, NakhonPathom province.

Parameters of egg quality	Mean	SD ¹	Max	Min	C.V. ² (%)
Albumen height (mm.)	4.13	0.45	5.89	2.64	10.90
Haugh unit	56.05	6.39	74.26	36.47	11.40
Albumen pH	9.19	0.20	9.39	8.34	2.18
Yolk color	10.55	0.77	12.00	9.00	7.30
Egg shell thickness (mm.)	0.36	0.03	0.38	0.35	8.33
Specific gravity	1.089	0.01	1.100	1.072	0.92

¹ Standard deviation

² Coefficient of Variation

Table 2. Ratio of composition of chicken eggs sampled from 10 flea markets in KamphaenSaen district, Nakhon Pathom province.

Parameters of egg composition	Mean	SD ¹	Max	Min	C.V. ² (%)
Yolk (%)	27.76	0.59	30.29	25.34	2.13
Albumen (%)	59.55	0.31	62.35	57.16	0.52
Egg shell (%)	12.69	0.24	13.17	12.29	1.89

¹ Standard deviation

² Coefficient of Variation

Conclusion

As a conclusion, the wide range of albumen height and Haugh unit values are detected in chicken eggs sold in the flea markets settled temporarily outdoors. This indicates that chicken eggs in the flea market have high variations of freshness attributes.

References

- Haugh, R. R., 1937. The Haugh unit for measuring egg quality. U.S. Egg Poult. Mag. 43:552-555
- Kul, S., and I. Seker, 2004. Phenotypic correlation between some external and internal egg quality traits in the Japanese quail (*Coturnixcoturnix japonica*). Department of Zootechnia, Faculty of Veterinary, Firat University. Elazig, Turkey. 3(6): 400-405.
- Samli, H.E., A. Agma, and N. Senkoylu, 2005. Effects of storage time and temperature on egg quality in old laying hen. J. of Appl. Poult. Res. 14: 548–553.
- Sayraumyart, I, S. Rattanatabtimtong, S. Attamangkune, and Y Ruangpanit, and O. Jintasataporn, 2011. A Survey of Chicken egg quality from farm to market. J Sci Technol MSU (special ed.) p 96-105.
- Scott, T.A, and F.G. Silversides. 2000. The effect of storage and strain of hen on egg quality. Poult. Sci. 79: 1725–1729.
- United States Department of Agriculture (USDA). Egg Grading Manual. 2002. Available from: URL: <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELDEV3004502> Accessed June 25, 2014.

Effect of carbon and nitrogen sources on growth of *Alternaria solani* (Ell. & Mart.) Causing early blight of Tomato

Chate D. B¹. & Ghuge D.S².

¹Mahatma Basweshwar College, Latur. (M.S.) India,²Narayanrao Waghmare College, Akhada Balapur, Dist. Hingoli (MS) India

Abstract

In Present investigation the effects of carbon and nitrogen nutrition on seven isolates of *Alternaria solani* isolated from different varieties of Tomato (*Lycopersicon esculentum* Mill.) was studied at 28°C for ten days at pH 6.0. Among the seven carbon sources glucose for the isolate I, Lactose for isolate III and IV, Gelatin for isolate I and II, Mannitol for III, V, VI and Sucrose for the isolate II, IV and VII were found to be the best sources. Among the seven Nitrogen sources Potassium nitrate was the best sources for the growth of isolate II, IV, VI, Peptone for the Vth isolate, Ammonium Chlorite for VII and Ammonium Molybdate for III isolate were found to be the best Nitrogen sources.

Keywords: Alternaria solani, tomato, carbon, nitrogen.

Introduction

Carbon and nitrogen are essential elements for the growth and sporulation of fungi. Utilization of carbon and nitrogen containing compounds by fungi depends on their ability to assimilate them, directly or after conversion in to simple compounds (Lilly and Barnet, 1951). Tomato (*Lycopersicon esculentum* mill.) is an important commercial vegetable crop grown widely throughout the world including tropical, sub tropical and temperate regions. English traders of East India Company introduced tomato in India in 1822 India is second largest producer of tomato next to China.

The association of phytopathogenic micro-organisms with this crop is perhaps as old as civilization. Among these organism fungi hold a significant place and are important as the pathogens.

Material and Methods

In the present investigation the infected tomato (*Lycopersicon esculentum* Mill) plant material (Leaves and fruits) collected from Marathwada Agriculture University, Parbhani, Dist. Parbhani (MS) The *A. solani* pathogen was isolated (Aneja, 2007) and identified (Subramanian). The pure cultures of the pathogen maintained on P.D.A. The *A solani*. The pathogen isolates were cultured on basal medium. The medium was autoclaved and Ph was adjusted to 6.0. Each flask containing 25 ml of the medium was inoculated with fungal species and incubated for 10 days at 25°C, filtered, mycelial mat was dried and dry weight was recorded.

Results and Discussion

The result obtained are presented in table 1 and 2. Among the seven carbon sources fructose for isolate six and sucrose for isolate seven was found to be superior sources. Among nitrogen

sources all except sodium nitrate for isolate seventh was found to be the inferior to all isolates Hadge et al. (1990) observed the glucose and sucrose as good carbon source for the growth. of *Colletotricum gloeosporides*, isolated from areca nut. Bhandari and Singh (1976) reported glucose as better source of carbon for growth of *Alternaria tritincina*. Pande & Shukla (1978) observed better growth of *Helminthosporium* on sucrose. These findings supports with present study and similar results were also reported by Agarwal & Shinkhede (1959), Kaif & Tarr (1966) and Reddy (1972).

Conclusion

From the present investigation it is concluded that among the seven Nitrogen sources Potassium nitrate was the best sources for the growth of isolate II, IV, VI, Peptone for the Vth isolate, Ammonium Chlorite for VII and Ammonium Molybdate for III isolate were found to be the best Nitrogen sources.

Table 1. Effect of Carbon sources on *Alternaria solani*. (Ell. And Mart).

Sr.	Carbon sources	Dry wt. of mycelial mat(Mg) of different isolates of <i>A.solani</i>						
		Isolate.						
		I	II	III	IV	V	VI	VII
01	Glucose	360	240	335	350	305	265	310
02	Fructose	275	305	310	345	310	370	305
03	Lactose	240	310	320	320	305	270	370
04	Starch	210	250	210	260	235	210	270
05	Gelatin	330	340	315	260	280	310	320
06	Manitol	270	310	370	325	360	380	270
07	Sucrose	265	370	310	370	365	320	410
08	Control	040	035	040	030	060	060	060

Table 2. Effect of Carbon sources on *Alternaria solani*.(Ell.and Mart.).

Sr. No.	Nitrogen sources	Dry wt. of mycelia mat (Mg) of different isolates of <i>A.solani</i> .						
		I	II	III	IV	V	VI	VII
01	Sodium nitrate	165	210	190	210	210	220	310
02	Calcium nitrate	110	130	130	125	120	110	160
03	Potassium nitrate	310	230	190	260	220	260	270
04	Peptone	210	190	170	160	260	265	235
05	Ammonium chloride	160	170	180	205	170	170	270
06	Ammonium molybdate	260	240	220	210	230	240	260
07	Casein	210	230	170	180	140	150	210
08	Control	050	060	050	060	070	060	060

References

- Agrawal, S. C. and C. G. Shinkhede, 1959. Physiological studies of *Helminthosporium rostratum* Drechs. *Phyton*. 13: 45.
- Bhandari, J. K.S. and R. S. Singh, 1976. Effect of carbon and nitrogen sources on the growth of *Altdnraria triticina*, *Indian Phytopath*, 29 (1): 88
- Bilgrami, K. S. and R. N. Verma, 1978. "Physiology of fungi. " Vikas Pub. House Pvt .Ltd New Delhi.
- Hedge, Y.R., P. K. Hedge and S. Kulkarni, 1990. Glucose and sucrose as good carbon source for the growth of *Colletotricum gloeosporides*, isolated from ereca nut. *Mysore J. Agric. Sci.* 24:358.
- Kaif, A. and S. A. J. Tarr, 1966. Growth, sporulation and conidial characteristics of five graminicolous species of *Helminthosporium*. I. Effect of nutrients. *Trans. Brit. Mycol. Soc.* 49:327.
- Lilly V. G. and H. L. Barnett, 1951. "The physiology of fungi " . Mc. Graw-Hill com. Lnc., New York.
- Pandey, S.C. and T. N. Shukla, 1978. Host range studies of six *Helminthosporium* spp. causing leaf spot diseases of sorghum, *Indian J. Mycol. Plant Patho.* 8(2):207-208.

Prevalence of parasite infections of the first 2 month- old puppies in the animal hospital institute, Udon Thani Rajabhat University

Jaiwangdee¹, C., M. Pungpet¹ & N. Anantasook²

¹Animal hospital institute, Faculty of Technology, Udon Thani Rajabhat University, Udon Thani, 41000, Thailand, ²Program in Animal Production Technology, Faculty of Technology, Udon Thani Rajabhat University, Udon Thani, 41000, Thailand

Abstract

This study was to investigate the prevalence of parasite infections of the first 2 month-old puppies in the animal hospital, Udon Thani Rajabhat University. Eighty of the first 2 month-old puppies were chosen for this study and investigated for five months and fecal samples were collected during December 2014 to April 2015. Each month, 80 sample of fecal was collected by rectal sampling at examination room. All samples were done by direct fecal smear with 0.9% normal saline on glass slide and then be closed with cover glass for identify the egg parasites and then examined the samples with 4x ,10x and 40x light microscopy, respectively. It was found that the first 2 month-old studied puppies in the animal hospital institute, Udon Thani Rajabhat University had the prevalences of *Toxocara canis* 46.25% (37/80), *Dipylidium caninum* 6.25% (5/80), *Strongyloides* spp. 7.5% (6/80), *Ancylostoma caninum* 2.5% (2/80) and *Ascarid* spp. 1.25% (1/80), respectively. Based on this study, it could be concluded that the most prevalence of parasite infections of the first 2 month-old studied puppies in the animal hospital institute, Udon Thani Rajabhat University was *Toxocara canis*.

Keywords: parasite, prevalence, poppy dog, animal hospital institute

*Corresponding author: Mr.junt@live.com

Introduction

The domestic dog is generally considered the first domesticated mammal. Pet dogs are often considered to be faithful friends and intimate companions of humans and enjoy life together with human (Awoke et al., 2010). Dogs are the definitive or reservoirs hosts of more parasites, such as *Taenia* spp., *Echinococcus* spp., *Dipylidium caninum*, *Toxocara canis*, *Ancylostoma* spp., (Satyal et al., 2013). Their roles transmitting human infection have been recongnized worldwide (Chen et al., 2012) . Gastrointestinal parasite are one of the main entero-pathogens and cause of mortality in dogs especially in whelp or neonates (Perera et al., 2013) . The clinical sings of parasitic infection in dogs are varied such as vomiting, diarrhea, anemia, anorexia, dermatitis and loss of condition and occasionally some infected animal present no symptoms (Awoke et al., 2010). The dogs and puppy dogs infect parasite via environment contamination such as soil, feces, transmammary and transplacental. Many studies have been conducted to access the situation of parasite in puppy dog at Udon Thani. However, the prevalence of parasite infection of puppy dogs in Udon Thani is still limited. Therefore, the objective of current study was determined the prevalence of parasites infection in puppy dogs Udon Thani.

Materials and methods

Eighty, puppy dog to two month age were used in the experiment. The study was run in five months and fecal samples were collected during December 2014 to April 2015. Each month, fecal samples were collected in puppy dogs by rectal sampling at examination room in animal hospital Udon Thani Rajabhat University. Identification of characteristic parasites was made according to the egg morphological characteristics. All samples were done by direct fecal smear with 0.9% normal saline on glass slide and then be closed with cover glass for identifying egg parasite and then examined by direct fecal smear with 4x, 10x and 40x microscopy, respectively.

Results and Discussion

The prevalence of parasitology in puppy dogs at Udon Thani are showed in Table 1. It was found that during December 2014 to April 2015, prevalence of parasite infection of the first 2 month-old puppies at Udon Thani animal hospital were *Toxocara canis* 46.25% (37/80), *Dipylidium caninum* 6.25% (5/80), *Strongyloides* spp. 7.5% (6/80), *Ancylostoma caninum* 2.5% (2/80) and *Ascaris* spp. 1.25% (1/80). The result revealed that the prevalence of *Toxocara canis* was higher in < 2 months old puppy dogs. These could be due to puppy dogs are higher risk of infection via transplacental and transmammary gland transmission from bitch (Jamal, 2014). The prevalence of *Toxocara canis* was higher in < 6 months old (5.3%) than those > 6 months old (0.65%), similar to investigations in Canada and Ethiopia information is available about the prevalence (Getahun & Addis, 2012). Prevention puppy dogs infection parasite with deworm bitch before pregnancy period and puppy dogs 30 days postpartum period. Represented a significant risk for public health and appropriate measures should be taken to regulate the populations of carnivores. Promote deworming Programs for puppy dogs with contamination in faces from dogs distributions in food borne parasitic infection at the processing.

Table 1. Sample puppies of 2 month age infection parasite groups at Udon Thani during December 2014 to April 2015.

Items	<i>Toxocara canis</i>	<i>Dipylidium caninum</i>	<i>Strongyloides</i> spp.	<i>Ancylostoma caninum</i>	<i>Ascaris</i> spp.
Total	80	80	80	80	80
Positive samples	37	5	6	2	1
Percentages	46.25	6.25	7.5	2.5	1.25

Conclusion

Percentage infection of parasites in puppy dogs at Animal Hospital, Udon Thani Rajabhat University was *Toxocara canis* 46.25% (37/80), *Dipylidium caninum* 6.25% (5/80), *Strongyloides* spp. 7.5% (6/80), *Ancylostoma caninum* 2.5% (2/80) and *Ascarid* spp. 1.25% (1/80). Based on this study could be concluded that the most prevalence of parasite infection of the first 2 month-old studied puppies in the animal hospital institute, Udon Thani Rajabhat University was *Toxocara canis*. The present study demonstrated that Udon Thani, gastrointestinal helminths are present in fecal puppy dogs.

Acknowledgements

The authors would like to express their most sincere thanks to staffs of the Animal Hospital, Faculty of Technology, Udon Thani Rajabhat University for financial support, laboratory and research facilities.

References

- Awoke, E., B. Bogale and M. Chanie, 2010. Intestinal nematode parasites of dogs: Prevalence and associated risk factors. *Int J. Anim. Vet. Adv.* 3: 374-378.
- Chen, J. M.J. Xu, D.H. Zhou, H.Q. Song, C.R. Wang and X.O. Zhu, 2012. Canine and feline parasitic zoonoses in China. *Parasit. Vectors.* 5: 152.
- Fang, F., J. li.T. Huang, J. Guillot and W. Huang, 2015. Zoonotic helminths parasites in the digestive tract of feral dogs and cats in Guangxi, China. *BMC veterinary Research.* 11: 211.
- Getahun, Z. and M. Addis, 2012. Prevalence of gastrointestinal helminthes among dogs in Bahir Dartown, Ethiopia. *World Appl. Sci. J.* 19: 595-560.
- Jamal, G., 2014. Study on Gastrointestinal Zoonotic Parasities in Pet Dogs in Western Iran. *Gharekhani J.* 38: 172-176.
- Satyral, R.C., S. Manandhar, S. Dhakal, B.R. Mahato, S. Chaulagain and L. Ghimire, 2013. Prevalence of gastrointestinal zoonotic helminthes in dogs of Kathmandu, Nepal. *Int J. Infect Microbiol.* 2: 91-94.
- Perera, P.K., R.P.V.J. Rajapakse and R.S. Rajakaruna, 2013. Gastrointestinal parasites of dogs in Hantana area in the Kandy District. *J. Natn. Sci. Foundation. Sri lanka.* 41: 81-91.

SCIENTIFIC PROMOTION CO., LTD.

บริษัท ไซแอนติฟิกโปรโมชั่น จำกัด



Betagro Public Co., Ltd.

บริษัท เบทาโกร จำกัด (มหาชน)



BETAGRO

History

Founded in 1967 with a registered capital of Baht 10 million, the Betagro Group began as a single entity, Betagro Company Limited, to produce and distribute animal feed. Its headquarters were originally located in Pom Prab, Bangkok, and its first feed mill in Prapadaeng, Samut Prakan province.

Betagro Group focuses on the food business in both domestic and international markets with an integrated agro-industrial production base. Betagro classifies its operation in five major categories:



The Thailand Research Fund; TRF

สำนักงานกองทุนสนับสนุนการวิจัย; สกว.



The Thailand Research Fund (TRF) was established in response to the 1992 Research Endowment Act and although it is part of the government system, it lies outside the government administrative bureaucracy. This freedom allows great efficiency in research support. Since its creation, TRF has remained focused on its main duty: supporting at both local and national levels the creation of a knowledge-base to help those tackling societal problems.





AGRICULTURAL RESEARCH DEVELOPMENT AGENCY (PUBLIC ORGANIZATION)
สำนักงานพัฒนาการวิจัยการเกษตร (องค์การมหาชน)



Background

Due to the economic crisis in B.E. 2540 (1997), the government has set austerity measure in spending the budget. Resulting many projects has to balk. The government has agreed to borrow the funding from abroad to continue the project. The Cabinet has approved to abide by the conditions of policy matrix. Loan program to restructure the agricultural sector to enhancing the competitiveness of the export on 6th July B.E. 2542 (1999)

Agricultural Research Development Agency (Public Organization), ARDA was established as an autonomous agency in the operating fund as the decree establishing the Agricultural Research Development Agency that become effective from 15th March B.E. 2546 (2003) (with the initial capital of 3,000 Million Baht)

MISSION

- *PROMOTE AND SUPPORT AGRICULTURAL RESEARCH*
- *PROMOTE AND SUPPORT PERSONNEL DEVELOPMENT IN AGRICULTURAL RESEARCH*
- *PROMOTE AND RESEARCH AND DEVELOPMENT, DISSEMINATION AND AGRICULTURAL RESEARCH INFORMATION TECHNOLOGY*

ARDA

CPF (Thailand) Public Co., Ltd.

บริษัท ซีพีเอฟ (ประเทศไทย) จำกัด (มหาชน)



Charoen Pokphand Foods Public Company Limited is the leading agro-industrial and food conglomerate in the Asia Pacific region. The Company operates in both the livestock (swine, broilers, layers, and ducks) and aquaculture (shrimp and fish) businesses. The vertically integrated businesses incorporate the manufacturing of animal feed, animal breeding and animal farming; meat processing, the manufacturing of semi-cooked meat and fully-cooked meat; food products and ready meal products, as well as the meat and food retailer and restaurant businesses.

Feed Business



Farm Business



Food Business



Lannacom Co., Ltd.

บริษัท ลานนาคอม จำกัด



บริการให้คำปรึกษา ออกแบบ จำหน่าย ติดตั้ง ซ่อมแซมและบำรุงรักษา ที่เกี่ยวกับระบบเทคโนโลยีสารสนเทศ และการสื่อสาร ได้แก่ โครงสร้างพื้นฐานของระบบเครือข่ายคอมพิวเตอร์ ระบบรักษาความปลอดภัยระบบเครือข่ายคอมพิวเตอร์ ระบบโสตทัศนอุปกรณ์สารสนเทศ และระบบโทรศัพท์

Dutch Mill Co., Ltd.

บริษัท ดัชมิลล์ จำกัด



Office of the Higher Education Commission; OHEC

สำนักงานคณะกรรมการการอุดมศึกษา; สกอ.



Iken Scientific Limited Partnership

ห้างหุ้นส่วน จำกัด ไอ เค็น ไซเอนทิฟิค



Behn Meyer Chemicals (T) Co., Ltd.

บริษัท เบ็นไมเยอร์ เคมีคอลส์ (ที) จำกัด



Beef Cluster Cooperative Limited

สหกรณ์เครือข่ายโคเนื้อ จำกัด



K.M.P. BIOTECH CO., LTD.





5th

SAADC 2015

The 5th International Conference on Sustainable Animal Agriculture for Developing Countries

October 27-30, 2015, Dusit Thani Pattaya Hotel, THAILAND

SPONSORS

