AQUAFISH COLLABORATIVE RESEARCH SUPPORT PROGRAM

IMPLEMENTATION PLAN 2007-2009

MARCH 2008

AquaFish CRSP
College of Agricultural Sciences
Oregon State University
418 Snell Hall
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The mission of the AquaFish Collaborative Research Support Program (CRSP) is to enrich livelihoods and promote health by cultivating international multidisciplinary partnerships that advance science, research, education, and outreach in aquatic resources. Bringing together resources from host country institutions and US universities, the AquaFish CRSP emphasizes sustainable solutions in aquaculture and fisheries for improving health, building wealth, conserving natural environments for future generations, and strengthening poorer countries' ability to self-govern.

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INTRODUCTION

The AquaFish CRSP *Implementation Plan 2007-2009* provides six brief project summaries along with descriptions of each project's investigations to be undertaken through July 2009. Projects are authorized for 30 months, ending 30 September 2009.

The six research projects that form the foundation for this *Implementation Plan* were selected in response to the first AquaFish Request for Proposals. The RFP was released and widely distributed in November 2006 by Oregon State University, which serves as Lead Institution for the overall AquaFish CRSP. Throughout the RFP period, which closed on 30 January 2007, OSU updated its matchmaking service and FAQ on the RFP web page. Nineteen eligible proposals were competitively reviewed following an NSF-style, external peer-review process in March 2007; finalists were then further reviewed by USAID. The RFP and supporting documentation can be found at http://pdacrsp.oregonstate.edu/afcrsp/rfp/.

The six projects described herein represent all of the key regions, themes, and topic areas called for in the RFP. As a group they include 13 countries, 12 US universities, and over 20 HC institutions in formal funded partnerships, plus more than 70 additional collaborators in informal partnerships. The six projects contain 38 investigations accounting for nearly US$2.4 million from USAID, not including management costs or other outreach expenditures. Each project additionally provides over 50% in matching funding for each federal dollar received.

The overall CRSP is managed in a manner to achieve maximum program impacts, particularly for small-scale farmers and fishers, in Host Countries and more broadly. CRSP program objectives address the need for world-class research, capacity building, and information dissemination. Specifically, the AquaFish CRSP strives to:

- Develop sustainable end-user level aquaculture and fisheries systems to increase productivity, enhance international trade opportunities, and contribute to responsible aquatic resource management;
- Enhance local capacity in aquaculture and aquatic resource management to ensure long-term program impacts at the community and national levels;
- Foster wide dissemination of research results and technologies to local stakeholders at all levels, including end-users, researchers, and government officials; and
- Increase Host Country capacity and productivity to contribute to national food security, income generation, and market access.

USAID (May 2006, RFA) looks at the AquaFish CRSP to "develop more comprehensive, sustainable, ecological and socially compatible, and economically viable aquaculture systems and innovative fisheries management systems in developing countries that contribute to poverty alleviation and food security."

The overall research context for the projects described in this *Implementation Plan* is poverty alleviation and food security improvement through sustainable aquaculture development and aquatic resources management. Research that generates new information should form the core of projects. Projects must also include institutional strengthening, outreach, and capacity building activities such as training, formal education, workshops, extension, and conference organizing to support the scientific research being conducted.

Global AquaFish CRSP Project Themes (Goals)

A. Improved Health and Nutrition, Food Quality, and Food Safety
B. Income Generation for Small-Scale Fish Farmers and Fishers
C. Environmental Management for Sustainable Aquatic Resources Use
D. Enhanced Trade Opportunities for Global Fishery Markets

Each project identified one AquaFish CRSP theme as its primary focus, but also addressed all four themes in an integrated systems approach. The global themes of the CRSP are cross-cutting and address several specific USAID policy documents and guidelines, including the *Policy Framework for Bilateral Foreign Aid*, *Agriculture Strategy*, EGAT Offices of Agriculture & Natural Resource Management Strategic Objectives, and IEHA (Initiative to End Hunger in Africa). The themes also address global initiatives and strategies that relate to the overall program goal.

AquaFish CRSP Topic Areas for Research, Outreach, and Capacity Building

Thematic projects contain work plans (investigations) organized around a number of specific areas of inquiry called Topic Areas. Projects contain between five and eight investigations. Projects focus on more than one topic area in describing aquaculture research that will improve diets, generate income for smallholders, manage environments for future generations, and enhance trade opportunities. Projects were formed around *core program components*, as identified by USAID:

- a systems approach
- social, economic, and environmental sustainability
- capacity building and institution strengthening
- outreach, dissemination, and adoption
- gender integration

A systems approach requires that each CRSP project integrate topic areas from both *Integrated Production Systems* and *People, Livelihoods and Ecosystem Interrelationships.* USAID also encourages the CRSP portfolio (the sum of all funded projects) to address biodiversity conservation and non-GMO biotechnology solutions to critical issues in aquaculture. While not every investigation individually addresses each element recommended by USAID, each overall project describes a comprehensive development approach to a problem.

Topic Areas pertain to aquaculture and the nexus between aquaculture and fisheries. Some of the following topic areas overlap and are interconnected. Investigations in this *Implementation Plan* identify a single topic area that best describes each individual investigation. The text under each topic area is provided for illustrative purposes and is not prescriptive. Specific fisheries issues were not funded in the current RFP per guidance from USAID.

Research and Outreach Topic Areas: Integrated Production Systems

- **Production System Design & Best Management Alternatives (BMA)**
  Aquaculture is an agricultural activity with specific input demands. Systems should be designed to improve efficiency and/or integrate aquaculture inputs and outputs with other agricultural and non-agricultural production systems. Systems should be designed so as to limit negative environmental impacts. CRSP research should benefit smallholder or low- to semi-intensive producers, and focus on low-trophic species for aquaculture development. Research on soil-water dynamics and natural productivity to lessen feed needs were fundamental to the Aquaculture CRSP; critical new areas of research may be continued. Interventions for disease and predation prevention must adopt an integrated pest management (IPM) approach and be careful to consider consumer acceptance and environmental risk of selected treatments.

- **Sustainable Feed Technology (SFT)**
  Methods of increasing the range of available ingredients and improving the technology available to manufacture and deliver feeds are an important research theme. Better information about fish nutrition can lead to the development of less expensive and more efficient feeds. Investigations on successful adoption, extension, and best practices for efficient feed strategies that reduce the "ecological footprint" of a species under cultivation are encouraged. Feed research that lessens reliance on fish meals/proteins/oils and lowers feed conversion ratios is desired, as is research on feeds (ingredients, sources, regimes, formulations) that result in high quality and safe aquaculture products with healthy nutrition profiles.

- **Indigenous Species Development (IND)**
  Domestication of indigenous species may contribute positively to the development of local communities as well as protect ecosystems. At the same time, the development of new native species for aquaculture must be approached in a responsible manner that diminishes the chance for negative environmental, technical, and social impacts. Research that investigates relevant policies and practices is encouraged while exotic species development and transfer of non-native fishes are not encouraged. A focus on biodiversity conservation, and biodiversity hotspots, as related to the development of new native species for aquaculture is of great interest. Aquaculture can be a means to enhance and restock small-scale capture and wild fisheries resources (Aquaculture-Fisheries Nexus Topic Area). Augmentation of bait fisheries through aquaculture to support capture fisheries is an area of interest, provided there are no net negative environmental effects.

- **Quality Seedstock Development (QSD)**
  Procuring reliable supplies of high quality seed for stocking local and remote sites is critical to continued development of the industry, and especially of smallholder private farms. A better understanding of the factors that contribute to stable seedstock quality, availability, and quantity for aquaculture enterprises is essential. Genetic improvement (e.g., selective breeding) that does not involve GMOs may be needed for certain species that are internationally traded. All genetic improvement strategies need to be cognizant of marketplace pressures and trends, including consumer acceptance and environmental impacts.

<u>Research and Outreach Topic Areas: People, Livelihoods, & Ecosystem Interrelationships</u>

- **Human Health Impacts of Aquaculture (HHI)**
  Aquaculture can be a crucial source of protein and micronutrients for improved human health, growth, and development. Research on the intrinsic food quality of various farmed fish for human consumption is needed—this might include science-based studies of positive and negative effects of consuming certain farmed fishes. Patterns of fish consumption are not well understood for many subpopulations. Human health can be negatively impacted by aquaculture if it serves as a direct or indirect vector for human diseases. There is interest in better understanding the interconnectedness of aquaculture production and water/vector-borne illnesses such as malaria, schistosomiasis, and Buruli ulcer and human health crises such as HIV/AIDS and avian flu.

- **Food Safety & Value-Added Product Development (FSV)**
  Ensuring high quality, safe, and nutritious fish products for local consumers and the competitive international marketplace is a primary research goal. Efforts that focus on reducing microbial contamination, HACCP controls and hazards associated with seafood processing, value-added processing, post-processing, and byproduct/waste development are of interest. Consumers and producers alike will benefit from research that contributes to the development of standards and practices that protect fish products from spoilage,

adulteration, mishandling, and off-flavors. Certification, traceability, product integrity and other efforts to improve fish products for consumer acceptance and international markets are desired. Gender integration is important to consider as women are strongly represented in the processing and marketing sectors. **(**Aquaculture-Fisheries Nexus Topic Area)

- **Technology Adoption & Policy Development (TAP)**
  Developing appropriate technology and providing technology-related information to end-users is a high priority. The program encourages research that results in a better understanding of factors and practices that set the stage for near-term technology implementation and that contribute to the development of successful extension tools and methods. Areas of inquiry can include institutional efforts to improve extension related to aquaculture and aquatic resources management; science-based policy recommendations targeting poor subpopulations within a project area, or more broadly (for example, national aquaculture strategies); methods of improving access to fish of vulnerable populations including children (e.g., school-based aquaculture programs); science-based strategies for integrating aquaculture with other water uses to improve wellbeing, such as linkages with clean drinking water and improved sanitation. Policy initiatives that link aquaculture to various water uses to improve human health are needed. Additionally, social and cultural analyses regarding the impacts of fish farming may yield critical information for informing policy development.

- **Marketing, Economic Risk Assessment & Trade (MER)**
  Aquaculture is a rapidly growing industry and its risks and impacts on livelihoods need to be assessed. Significant researchable issues in this arena include cost, price, and risk relationships; domestic market and distribution needs and trends; the relationships between aquaculture and women/underrepresented groups; the availability of financial resources for small farms; and the effects of subsidies, taxes, and other regulations. Understanding constraints across value chains in local, regional, and international markets is of interest, especially as constraints affect competitiveness, market demand, and how to link producers to specific markets. **(**Aquaculture-Fisheries Nexus Topic Area)

- **Watershed & Integrated Coastal Zone Management (WIZ)**
  Aquaculture development that makes wise use of natural resources is at the core of the CRSP. Research that yields a better understanding of aquaculture as one competing part of an integrated water use system is of great interest. The range of research possibilities is broad—from investigations that quantify water availability and quality to those that look into the social context of water and aquaculture, including land and water rights, national and regional policies (or the lack thereof), traditional versus industrial uses, and the like. Water quality issues are of increasing concern as multiple resource use conflicts increase under trends toward scarcity or uneven supply and access, especially for freshwater. Ecoregional analysis is also of interest to explore spatial differences in the capacities and potentials of ecosystems in response to disturbances. Innovative research on maximizing water and soil quality and productivity of overall watersheds is of interest. Pollution is a huge concern, as over 50% of people in developing countries are exposed to polluted water sources. Additionally, aquatic organisms cannot adequately grow and reproduce in polluted waters, and aquaculture may not only be receiving polluted waters, but adding to the burden. Rapid urbanization has further harmed coastal ecosystems, and with small-scale fisheries and aquaculture operations in the nearshore, integrated management strategies for coastal areas are also important. **(**Aquaculture-Fisheries Nexus Topic Area)

- **Mitigating Negative Environmental Impacts (MNE)**
  With the rapid growth in aquaculture production, environmental externalities are of increasing concern. Determining the scope and mitigating or eliminating negative

environmental impacts of aquaculture—such as poor management practices and the effects of industrial aquaculture—is a primary research goal of this program. A focus on biodiversity conservation, especially in biodiversity “hotspot” areas, as related to emerging or existing fish farms is of great interest. Therefore, research on the impacts of farmed fish on wild fish populations, and research on other potential negative impacts of farmed fish or aquaculture operations is needed, along with scenarios and options for mitigation. **(**Aquaculture-Fisheries Nexus Topic Area)

Program Regions

Projects were selected that focused on one USAID-eligible country within a region, but had activities in nearby countries within the same region. Proposed activities received USAID country-level concurrence prior to award. Non-concurrence meant that a project or investigation was not approved for funding, as was the case with an investigation that included Bangladesh. The USAID Mission in Bangladesh did not concur due to perceived management overload; another CRSP was also denied the privilege of working in Bangladesh. Each project site will be described in a separate volume of site descriptions due for completion in 2008.

Rules of Conduct

Rules of conduct are described in greater detail in each project’s subcontract with the Management Entity and in other program documents. The following subset of rules especially pertains to the *Implementation Plan*.

Fostering Respectful Partnerships: Projects aim to foster linkages with organizations including US minority-serving institutions, non-governmental organizations (NGOs), national agricultural research institutions, other CRSPs, international centers, private businesses, and others as desired. Projects that link Host Country researchers from one CRSP site to another CRSP site are encouraged. US and Host Country PIs share in budgetary decisions and overall priority setting for the project, as well as in other collaborative activities related to the CRSP. Proposals, work plans, and project budgets must be developed collaboratively between HC and US researchers. US PIs must actively establish an effective working relationship with the ME and other CRSP US and Host Country PIs and program participants.

Memoranda of Understanding: Upon award selection, the Lead US Institution of each project is required to enter into Memoranda of Understanding (MOUs) with institutions at Host Country sites. Subcontracting US institutions under the US Lead institution may also enter into MOUs with HC partners to strengthen institutional relationships and streamline administrative processes. MOUs between Host Country institutions are not discouraged but will not take the place of MOUs between US and Host Country institutions. MOUs must provide the opportunity for other CRSP projects to function under the authority of the agreement and must provide for joint authorship of reports and site visits at the discretion of the CRSP Management Entity. Draft MOUs are submitted to the ME for review prior to execution.

The following USAID environmental restrictions apply to the projects and the overall program:

- Biotechnical investigations will be conducted primarily on research stations in Host Countries.

- Research protocols, policies, and practices will be established prior to implementation to ensure that potential environmental impacts are strictly controlled.

- All training programs and outreach materials intended to promote the adoption of CRSP-generated research findings will incorporate the appropriate environmental recommendations.

- All sub-awards must comply with environmental standards.
- CRSP Projects will not procure, use, or recommend the use of pesticides of any kind. This includes but is not limited to algaecides, herbicides, fungicides, piscicides, parasiticides, and protozoacides.
- CRSP Projects will not use or procure genetically modified organisms (GMO).
- CRSP Projects will not use, or recommend for use, any species that are non-endemic to a country or not already well established in its local waters, or that are non-endemic and well established but are the subject of an invasive species control effort.

From April through May 2007, the six projects were evaluated for environmental compliance, and by late May, all six projects had made the necessary changes to receive concurrence from USAID environmental officers. The investigations presented herein are in compliance with USG environmental regulations. Each project PI must seek additional environmental compliance approval if significant changes are made through the course of the project.

At least 50% of funds must be expended in or on behalf of the Host Country or region. Each project must supply an additional 50% or more of matching funding from participating institutions. Collaborative efforts that involve undergraduate students, graduate students, and post-doctoral fellows are encouraged. CRSP funds will not be used to support US expatriate personnel or consultants, as the CRSP model is intended to build institutional networks and capacities. In furtherance of the Title XII initiative that authorizes all CRSPs, projects must demonstrate return benefits to the US. Under Title XII, CRSP has responsibility to provide mutual benefits and discoveries that can apply to the HC region and US and that will support future development of sustainable aquaculture and fisheries.

Investigations

Investigations that generate new information form the core of projects. Each investigation is clearly identified as an experiment, study, or activity, based on the following definitions:

Experiment A scientifically sound investigation that addresses a testable hypothesis. An experiment implies collection of new data by controlled manipulation and observation.

Study A study may or may not be less technical or rigorous than an experiment and may state a hypothesis if appropriate. Studies include surveys, focus groups, database examinations, most modeling work, and collection of technical data that do not involve controlled manipulation (e.g., collection and analysis of soil samples from sites without having experiments of hypothesized effect before collection).

Activity An activity requires staff time and possibly materials but does not generate new information like an experiment or a study. Conference organization, training sessions, workshops, outreach, and transformation and dissemination of information are examples of activities.

Investigations provide a transparent means for evaluating different types of work under the CRSP, be they quantitative, empirical, biologically-based, qualitative, policy-based, or informal. Each project was required to include at least one *experiment* or *study*. Projects were also required to include outreach *activities* such as training, formal education, extension, and conference organizing to supplement the scientific research being proposed.

In addition to the investigations presented in Part II of this *Implementation Plan*, projects also submitted to USAID and the lead US Institution (or Management Entity at OSU):

- A plan for outreach and dissemination. The CRSP seeks to build capacity of HC researchers, farmers, and other stakeholders through improved understanding of aquaculture technologies, including soft technologies such as best practices and knowledge-based systems, as well as hard technologies.

- A gender "inclusivity" plan. Projects identified intended beneficiaries, stakeholders, and end-users in each investigation or for their projects in their entirety.

<u>**Research Priorities**</u>

All six projects address the following general research priorities:

- **Priority Ecosystems**
  Freshwater and brackish water ecosystems for aquaculture and aquaculture-fishery nexus topic areas. Marine ecosystems are also included in the aquaculture-fishery nexus topic areas.

- **Priority Species**
  Low-trophic level fishes; domesticated freshwater fishes; non-finfishes (e.g., bivalves, seaweeds); aquatic organisms used in polycultures and integrated systems; native species. Food fishes are a priority but species used for non-food purposes (e.g., ornamental, pharmaceutical) may also be included as a priority if they are a vital part of an integrated approach towards food security and poverty alleviation.

- **Target Groups**
  Aquaculture farms (small- to medium-scale, subsistence and commercial) and aquaculture intermediaries, policy makers, and others in host countries.

- **Key Partners**
  University, government, non-government, and private sector.

ACRONYMS

Program-Related

ACRSP	Pond Dynamics/Aquaculture CRSP
A&F CRSP	Aquaculture & Fisheries CRSP
AquaFish	Aquaculture & Fisheries CRSP
CRSP	Collaborative Research Support Program
HC	Host Country
ME	Management Entity
MOU	Memorandum of Understanding
NGO	Nongovernmental organization
PD/A CRSP	Pond Dynamics/Aquaculture CRSP
PI	Principal Investigator
RFA	Request for Assistance
RFP	Request for Proposals

General

FAQ	Frequently Asked Questions
KSh	Kenya Shillings
NB	Nota Bene, note well
PDF	Portable Document Format

Institutions, Organizations, Government Entities & Programs

ACIAR	Australian Centre for International Agricultural Research
AIT	Asian Institute of Technology
APEC	Asia-Pacific Economic Cooperation
ASEAN	Association of Southeast Asian Nations
ATA	American Tilapia Association
AwF	Aquaculture without Frontiers, USA
BAU	Bangladesh Aquacultural University
BFAR	Bureau of Fisheries & Aquatic Resources, Philippines
BIOTECMAR	Cultivos & Biotecnologíca Marina C.A., Venezuela
CESASIN	Comite Estatal de Sanidad Acuicola de Sinaloa (Sinaloa State Committee for Aquaculture Sanitation
CETRA	Centro de Transferencia Tecnológica para la Acuacultura (Center for Aquaculture Technology Transfer), Mexico
CI	Conservation International, Mexico
CIAD	Centro de Investigación de Alimentos y Desarrollo (Research Center for Food & Development), Mexico
CIDEA-UCA	Centro de Investigación de Ecosistemas Acuáticos de la Universidad Centroamericana (Center for Research on Aquatic Ecosystems-Central American University), Nicaragua
CIFAD	Consortium for International Fisheries & Aquaculture Development
CIMMYT	International Wheat & Maize Improvement Center, Mexico
CLAR	Central Laboratory for Aquaculture Research, Egypt
CLSU	Central Luzon State University
CRC/URI	Coastal Resources Center/University of Rhode Island
CTU	Can Tho University, Vietnam
DASP	Department of Animal Sciences & Production, SUA
DA-BFAR	Department of Agriculture-Bureau of Fisheries & Aquatic Resources, Philippines
EGAT	Bureau for Economic Growth, Agriculture, & Trade (USAID)

EPA	US Environmental Protection Agency
EU	European Union
FAC	Freshwater Aquaculture Center, Central Luzon State University, Philippines
FAO	Food & Agriculture Organization, United Nations
FD	Fisheries Department, Kenya
FDA	US Food & Drug Administration
FDAP	Fisheries Development Action Plan, Cambodia
FiA	Fisheries Administration, Cambodia
FISH	The FISH Project (Fisheries Improved for Sustainable Harvest), Philippines
FIU	Florida International University
GESAMP	Joint Group of Experts in the Scientific Aspects of Marine Environmental Protection, FAO
GIFT	Genetically Improved Farmed Tilapia
GOP	Government of Philippines
GTIS	Guyana Trade & Investment Support Project
IAAS	Institute of Agriculture & Animal Science, Nepal
IARC	International Agricultural Research Center(s)
ICLARM	International Center for Living Aquatic Resources Management (= The WorldFish Center), Malaysia
IDRC	International Development Research Centre, Canada
IEHA	Initiative to End Hunger in Africa
IFREDI	Inland Fisheries Research & Development Institute, Cambodia
ISSC	Interstate Shellfish Sanitation Conference
ISA	Sinaloa Institute for Aquaculture, Mexico
ISTA	International Symposium on Tilapia in Aquaculture
KBDS	Kenya Business Development Services
KNUST	Kwame Nkrumah University of Science & Technology
LAC	Latin America & Caribbean Regions
LSU	Louisiana State University
MARENA	Nicaraguan Ministry of the Environment
MRC	Mekong River Commission
MSU	Michigan State University
NAAG	National Aquaculture Association of Guyana
NACA	Network of Aquaculture Centers in Asia, Thailand
NARS	National Agricultural Research System (of Host Countries)
NCSU	North Carolina State University
NIC	National Investment Center
NOAA	National Oceanographic & Atmospheric Administration (US)
NPRS	National Poverty Reduction Strategy, Cambodia
NSF	National Science Foundation, USA
NSSP	National Shellfish Sanitation Program
OSU	Oregon State University
PACRC	Pacific Aquaculture & Coastal Resources Center/University of Hawaii at Hilo
RIDS-Nepal	Rural Integrated Development Society-Nepal
SEAFDEC/AQD	Southeast Asian Fisheries Development Center/Aquaculture Department, Philippines
SEDPIII	Third Five-Year Socioeconomic Development Plan, Cambodia
SEMARNAT	Secretariat of Natural Resources, Mexico
SUA	Sokoine University of Agriculture
SUCCESS	Sustainable Coastal Communities & Ecosystems (EGAT/USAID)
TIES	Training, Internships, Education & Scholarships Program (USAID-Mexico)
TNC	The Nature Conservancy

TTU	Texas Tech University, Lubbock
UA	University of Arizona
UAPB	University of Arkansas, Pine Bluff
UAS	Universidad Autónoma de Sinaloa (Autonomous University of Sinaloa)
UAT	Universidad Autónoma de Tamaulipas (Autonomous University of Tamaulipas)
UCA	Universidad Centroamericana (Central American University)
UG	University of Georgia
UHH	University of Hawaii at Hilo
UJAT	Universidad Juárez Autónoma de Tabasco (Autonomous University of Juarez, Tabasco)
UJAT-CPSR	Cooperativa Pesquera San Ramón (San Ramón Fisheries Cooperative)
UBAC	Ujung Batee Aquaculture Center
UM	The University of Michigan
URI	University of Rhode Island
US	United States
USG	United States Government
USAID	United States Agency for International Development
USEPA	US Environmental Protection Agency
VT	Virginia Polytechnic Institute & State University
WAS	World Aquaculture Society
WWF	World Wildlife Fund

Topic Areas

BMA	Production System Design & Best Management Alternatives
FSV	Food Safety & Value-Added Product Development
HHI	Human Health Impacts of Aquaculture
ISD	Indigenous Species Development
MER	Marketing, Economic Risk Assessment & Trade
NE	Mitigating Negative Environmental Impacts
QSD	Quality Seedstock Development
SFT	Sustainable Feed Technology
TAP	Technology Adoption & Policy Development
WIZ	Watershed & Integrated Coastal Zone Management

Project & Investigation Terms

AOP	Advanced Oxidation Process
BMP	Best Management Practice
BOD	Biochemical Oxygen Demand
BSE	Bovine Spongiform Encephalopathy
BW	Brackish Water
cDNA	complementary DNA (Deoxyribonucleic acid)
CFU	Colony Forming Units
CG	Compensatory Growth
DO	Dissolved Oxygen
EC	*E. coli*
EPT	*Ephemeroptera*, *Pleocoptera* & *Trichoptera*
FCR	Food (Feed) Conversion Ratio
GIFT	Genetically Improved Farmed Tilapia
GIS	Geographic Information System
GLM	Generalized Linear Model
GMO	Genetically Modified Organism
GnRHa	Gonadotropin Releasing Hormone Analogue

HACCP	Hazard Analysis & Critical Point Control
HIV/AIDS	Human Immuno Virus/Acquired Immune Deficiency Syndrome
HPLC	High Performance Liquid Chromatography
HSD	Hepatosomatic Index
IGF-I	Insulin-like Growth Factor-I
IPM	Integrated Pest Management
LC/MS	Liquid Chromatography/Mass Spectrometry
LCA	Life Cycle Assessment
LCCA	Life Cycle Cost Analysis
LST	Lauryl Sulfate Tryptose
MC	Microcystins
mRNA	messenger RNA (Ribonucleic Acid)
MT	17α-Methyltestosterone
NL	Notochordal
PDI	Pellet Durability Index
PRCA	Participatory Rural Communication Appraisal
RIA	Radioimmunoassay
RRA	Rapid Rural Appraisal
SGR	Specific Growth Rate
SPE	Solid Phase Extraction
SL	Standard Length
SR	Sex Reversed
SS	Salmonella-Shigella
TN	Total nitrogen
TP	Total phosphorus
TSS	Total suspended solids
UV	Ultraviolet
XLD	Xylose Lysine Desoxycholate

PART I. RESEARCH PROJECT SUMMARIES

LEAD US INSTITUTION: NORTH CAROLINA STATE UNIVERSITY

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PROJECT TITLE
IMPROVED COST EFFECTIVENESS AND SUSTAINABILITY OF AQUACULTURE IN THE PHILIPPINES AND INDONESIA

AQUAFISH PROJECT THEME
INCOME GENERATION FOR SMALL-SCALE FISH FARMERS AND FISHERS

Investigations

1. 07QSD01NC	Broodstock Seed Quality and Fingerling Production Systems Rearing for Nile tilapia in the Philippines
2. 07SFT02NC	Feeding Reduction Strategies and Alternative Feeds to Reduce Production Costs of Tilapia Culture
3. 07TAP02NC	Internet-based Extension Podcasts for Tilapia Farmers in the Philippines
4. 07SFT03NC	Alternative Feeding Strategies to Improve Milkfish Production Efficiency in the Philippines
5. 07MNE02NC	Training in Sustainable Coastal Aquaculture Technologies in Indonesia and the Philippines
6. 07MER04NC	Implications of Export Market Opportunities for Tilapia Farming Practices in the Philippines

US & Host Country Institutions

USA	North Carolina State University (lead US institution) University of Arizona
Philippines	Central Luzon State University (lead Host Country institution) Southeast Asian Fisheries Development Center (SEAFDEC) AQD, Iloilo Bureau of Fisheries and Aquatic Resources (BFAR)
Indonesia	Ujung Batee Aquaculture Center, Banda Aceh

Other Collaborators and Linkages

Aquaculture without Frontiers (AwF), USA
Florida International University
Genetically Improved Farmed Tilapia (GIFT) Foundation International, Inc., Munoz, Philippines
Indonesian Department of Fisheries, Indonesia

Ladong Fisheries College, Indonesia
World Aquaculture Society (WAS), Baton Rouge, Louisiana
U.S. Department of Commerce, Milford Connecticut
Department of Agriculture, Philippines

Project Summary

Aquaculture in the Philippines and Indonesia is a high food security priority particularly in the light of the countries' rapidly growing populations and their continued dependence on fish protein. The incomes from family farming, however, are generally poor with 43% of small-scale tilapia farmers in Central Luzon, Philippines, falling below the poverty line. The difficult socioeconomic conditions are even more pronounced for fishers in coastal regions where traditional livelihoods have been lost, and many seek transition to milkfish farming. In Indonesia, a tsunami eliminated shrimp farms, and the livelihoods of entire communities must be rebuilt.

The project will develop and implement strategies to improve the cost effectiveness and sustainability of fish farming in the Philippines and Indonesia, and subsequently improve the livelihood of their people. Project investigations will assess key areas of research and extension that include methods to reduce farming costs for tilapia and milkfish, a marketing analysis to address the opportunities and constraints of expanding tilapia culture to fillet production for the export markets, development of a tilapia podcast as an emerging extension tool for the farming community of Central Luzon, and integration of polyculture for sustainable aquaculture in the Philippines and the tsunami-devastated Aceh region of Indonesia. Extension activities will provide five workshops and train approximately 30 students. The proposed project investigations incorporate specialists from Central Luzon State University, the Southeast Asian Fisheries Development Center (SEAFDEC AQD), Ujung Batee Aquaculture Center, North Carolina State University, University of Arizona, their collaborators, and the farming communities of the host country.

Tilapia and milkfish are the two most prominent finfish species cultured in the Philippines throughout inland and coastal areas. Fish feed is one of the most costly aspects of fish farming, representing as much as 80% of total production costs for tilapia. This emanates in part from feed wastage and the rising cost of fishmeal in commercial diets, whose sources are in rapid decline and in high demand. Studies aim to develop better management strategies to reduce feed usage and costs, a requirement for controlling production costs and increasing income for small-scale farmers. Limiting nutrient load from feed wastage will also help mitigate the environmental imprint of fish farming and promote its sustainability. Research will continue previous work that showed delayed or reduced feeding schedules can limit feed usage for growout of freshwater tilapia and milkfish in brackish and marine waters, without significantly altering yield. Tilapia farmers are excited by this research and several will participate in the studies. Least cost formulation and feed manufacturing technology will be used to limit the usage of fishmeal by development of tilapia feeds formulated from locally available Philippine ingredients. To address the USAID priority area of establishing suitable biotechnologies for the advancement of aquaculture, a study will be conducted on a highly promising protein as an instantaneous biomarker of growth rate in both tilapia and milkfish. This technology does not involve genetic modification of any organisms, but rather is a new tool for rapidly testing specific parameters of tilapia and milkfish performance for use by scientists and industry personnel.

Due to the projected increase in demand for tilapia and expansion into the export market, the need for seed of reliable quality is expected to triple. Consistent, high quality seed must be available at reasonable costs to increase both the profitability of farming and reduce the risks for entry of new farmers. One investigation is aimed at improving hatchery technologies for tilapia seed production. This study will assess broodstock age relative to fecundity, and also ascertain the relative effectiveness of three types of fry/fingerling rearing systems on tilapia growout. In collaborative work with the Genetically Improved Farmed Tilapia (GIFT) Foundation

International, experiments will use an extremely popular GIFT strain of tilapia seedstock that has been selectively bred for high performance and whose lineage has been distributed throughout the Philippines and Southeast Asia.
There is currently a strong desire to expand tilapia culture in the Philippines for the production of fillets for export markets and for supermarket retail. This would both increase income for farmers and promote further expansion of tilapia culture in the Philippines. Toward the goal of developing a viable export market for tilapia, a study will determine requirements for export opportunities of tilapia in the Philippines, assess implications of export markets on production systems, and provide recommendations for facilitating the development of a Philippines export market of tilapia.

In Indonesia and the Philippines, interest has heightened in diversifying aquaculture crops, following the realization that intensive shrimp farming practices contributed to the deterioration of water quality in the mangrove coastal habitat. Extension activities proposed in this project will utilize the expertise of scientists at SEAFDEC AQD and Central Luzon State University in the Philippines to assess, train, and help implement polyculture of tilapia, shrimp, and seaweed as a more sustainable and environmentally benign form of aquaculture in both Indonesia and the Philippines. Polyculture workshops in Indonesia will assist in rebuilding tsunami-devastated communities with more sustainable forms of agribusiness and those in the Philippines will complement ongoing USAID mission work geared to improving sustainable fisheries and the environment under the Fisheries Improved for Sustainable Harvest (FISH) project.

Podcasting will be developed as an extension technology. This powerful approach to information distribution has been used barely, if at all, in the aquaculture industry and is virtually nonexistent in the Philippines. Internet-based communication methods are increasing sharply in popularity, as is access for the people of developing countries that include the Republic of the Philippines. Podcasts as a mode of communicating news and technical developments are the wave of the future, and a tilapia podcast will be launched at Central Luzon State University, Philippines, so that farmers can gain the latest practical news on tilapia farming.

Long-range goals will focus on the excessive production costs associated with commercial feeds in finfish aquaculture. Continuation of refinements of feed strategies and formulations for tilapia and milkfish should directly benefit farmers and their capacity to improve incomes. Research and implementation of water reuse technologies in tilapia culture should limit nutrient outflow in waterways. An evaluation of a milkfish-like, progressive culture strategy to provide continuous rather than periodic harvest of tilapia seems a high priority as this could limit seasonal fluctuations in market supply and prices. This culture strategy could also reduce the need for application of chemicals and provide a more "organic" product for the people. Other areas of research could include better development of technologies for all-male tilapia production, which would limit the need for hormone-based, sex-reversal feed applications that are widely used but also necessary for the tilapia industry. Other goals include development of hatchery/nursery management technologies for both tilapia and milkfish to maintain stable seed supplies and the growth sustainability of both industries. Tilapia is the focus as it represents one of the highest sources of protein consumed by the world's population. Because of the wide popularity of tilapia, the management strategies applied to tilapia production in the Philippines will be applicable to addressing similar constraints in other underdeveloped countries in Africa, Asia, and Central/South America.

LEAD US INSTITUTION: UNIVERSITY OF ARIZONA

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PROJECT TITLE
DEVELOPING SUSTAINABLE AQUACULTURE FOR COASTAL AND TILAPIA SYSTEMS IN THE AMERICAS

AQUAFISH PROJECT THEME
ENVIRONMENTAL MANAGEMENT FOR SUSTAINABLE AQUATIC RESOURCES USE

Investigations

1. 07BMA03UA	Co-sponsorship of "Second International Workshop on the Cultivation and Biotechnology of Marine Algae: An Alternative for Sustainable Development in Latin America and the Caribbean"
2. 07SFT04UA	Utilization of Local Feed Ingredients for Tilapia and Pacu Production
3. 07IND01UA	Development of Snook (*Centropomus spp*) Seed Production Technology for Application in Aquaculture and Restocking of Over-fished Populations
4. 07MNE06UA	Elimination of MT from Aquaculture Masculinization Systems: use of Catalysis with Titanium Dioxide and Bacterial Degradation
5. 07IND02UA	Incorporation of the Native Cichlids, tenhuayaca, *Petenia splendida* and Castarrica, *Cichlasoma urophthalmus* into Sustainable Aquaculture in Central America: Improvement of Seedstock and Substitution of Fish Meal Use in Diets
6. 07HHI02UA	Food Safety Study of Leafy Greens Irrigated with Tilapia Farm Effluents
7. 07SFT05UA	Local Ingredients Substituting for Fishmeal in Tilapia and Pacu Diets in Guyana
8. 07TAP03UA	AquaFish CRSP Sponsorship of the Eighth International Symposium on Tilapia in Aquaculture to be Held in Egypt

US & Host Country Institutions

USA	University of Arizona (lead US institution) Texas Tech University, Lubbock
Guyana	Department of Fisheries
Mexico	Universidad Autónoma de Tamaulipas, Ciudad Victoria & Reynosa Universidad Juárez Autónoma de Tabasco

Other Collaborators and Linkages

Academy of Scientific Research & Egyptian Universities, Egypt
American Soybean Association
American University of Beirut, Lebanon

BIOTECMAR, Caracas, Venezuela
Central Administration of Agricultural Foreign Relations, Egypt
Central Laboratory for Aquaculture Research (CLAR), Abbassa, Egypt
Cornell University
Delaware State University
Egyptian Society of Agribusiness, Egypt
Instituto Sinaloense de Acuacultura, Mazatlán, Mexico
Maharaja Oil Mill, Guyana
Mariano Matamoros Hatchery, Teapa, Tabasco, Mexico
Ministry of Agriculture & Land Reclamation, Egypt
Mon Repos Aquaculture Center, Department of Fisheries, Guyana
National Aquaculture Association of Guyana (NAAG), Guyana
Oregon State University
San Carlos University, Guatemala
Sinaloa State Fisheries Department, Mexico
Texas Parks & Wildlife Department, Texas
United Animal Feed Producers
United Cooperative of Fishermen
United States Agency for International Development (USAID)/GTIS Programme, Guyana
University of Costa Rica, Costa Rica
University of Texas
US-Mexico Aquaculture TIES Program
Von Better Aquaculture, Guyana
WorldFish Center (ICLARM), Penang, Malaysia
Zamorano University, Honduras
Texas A&M University

Project Summary

The recent rapid growth of aquaculture has been a boon to many developing countries. Local consumption of aquaculture products and export of surplus production has improved both household nutrition and standard of living. Most cultured species are popular foods in local cuisines, representing multi-billions of dollars in local sales and international exports that are critical to developing economies. Unfortunately, the environmental impacts of aquaculture practices have been considerable. Demand for fishmeal and fish oil for aquaculture diets has grown to be a significant percentage of global markets. Effluents from aquaculture farms have contributed to eutrophication of receiving waters. As the economic and social importance of aquaculture products grows, it is imperative that scientists in both producing and consuming countries collaborate to develop diets and production systems that mitigate environmental impacts and reduce demands on limited resources.

This project builds on the former ACRSP's research and outreach by linking a series of research investigations and activities focused on (1) developing multiple use and polyculture systems to reduce impacts of effluents on the environment, with special focus on marine macroalgae (seaweeds); (2) developing indigenous species of cichlids and snook for aquaculture; (3) examining the bacterial degradation of methyltestosterone (MT) from hatchery effluent; (4) determining the food safety aspect of leafy vegetable crops grown with aquaculture effluents; (5) testing locally available protein and lipid sources to replace fish meal and fish oils in practical diets for tilapia, native cichlids, snook, and pacu; and (6) developing and providing improved lines of tilapia for aquaculture across Central America. Associated outreach activities include workshops, training sessions, and regional and international symposia. Under this project, AquaFish CRSP is sponsoring the 8th International Symposium on Tilapia in Aquaculture and organizing a special session at the 2008 World Aquaculture Meetings in Busan, Korea.

Mexico, the primary host country, is partnering with Guyana as a new CRSP country. Faculty from Delaware State University, Cornell University, and Oregon State University are participating as co-PI's or advisors to the US partners. This project also builds on collaborations developed within the former Aquaculture CRSP and US-Mexico Aquaculture TIES program, which is also supported by USAID. The US-Mexican network in TIES will also be tapped as project partners. At UJAT, the CETRA center provides a means for information exchange and training via the Internet and workshops. Making use of CETRA's network, research results will be distributed on production of snook, native cichlids, and seaweeds in recirculating systems.

Aquaculture has been touted as the solution to overfishing, depleted natural stocks, wasteful by-catch, and an opportunity to protect and enhance threatened and endangered species. It has also been assailed as a threat to the environment, a source of low quality or contaminated seafood, and responsible for the transfer of land and fishing rights to the wealthy from native peoples. Of course there are elements of truth in both camps. Sloganeering, "The Blue Revolution" versus "Friends don't let friends eat farmed fish" does us little good. Instead, our role as scientists is to utilize our skills, technology, and the scientific method to develop and share techniques and knowledge that will enable farmers to produce more aquatic foods while mitigating the impacts of their activities on the environment. Coastal zone management has developed as a discipline to coordinate the competing demands on coastal resources. In many developing countries, aquaculture has become one of the primary demands for water resources. Shrimp farming in particular has devastated mangrove forests and left abandoned ponds. In the last workplan of the Aquaculture CRSP, several investigations and activities demonstrated the benefits of more sustainable aquaculture systems beyond shrimp monoculture. Tilapia and other species could be reared in abandoned shrimp ponds; polyculture systems of tilapia and shrimp could mitigate some of the environmental impacts and disease problems endemic to the shrimp industry; and mangroves and seaweeds could absorb nutrients.

Some of the proposed investigations would expand the polyculture research and incorporate seaweed culture as well. Seaweed culture is especially important as seaweeds act to absorb nutrients, thereby mitigating effluent impacts, while also providing a nutritious and highly marketable product.

Scientists and institutions in the host countries need help to develop, share, and demonstrate sustainable aquaculture techniques. Local demand for aquaculture knowledge and technology must be increased. International buyers must be informed of new supplies of aquaculture product from sustainable farming systems.

The overall vision is to improve the welfare of aquaculturists in Mexico, Guyana, and their neighboring countries, while reducing their impacts on the environment. This vision will be achieved primarily by improving the knowledge and capacity of the producers and their local technical support. Considerable contributions will be made to improving the physical capacity of Universities and a research center.

The first set of investigations will demonstrate significant progress with experimental results and extend knowledge through several avenues of outreach. A second set of investigations will provide even more direct support and training for small fish producers who need to continue to improve their productivity and technology if they wish to compete in the markets, while protecting the native habitat. Integrated systems, with seaweeds, aquatic plants, and halophytes will presumably become the preferred method of treating fish effluents. A&F CRSP will continue to be the global leader in developing these concepts.

One primary objective is to leverage project funds with significant support from partners. The Guyana project has local in-kind support from farmers and government as well as continued interest from the local USAID mission, which has already provided significant financial support to the local fish farmers cooperative. Partners in Egypt have prepared a budget for the next International Symposium on Tilapia in Aquaculture (ISTA8) with $100,000 of Egyptian government support. This would match what was received from the Mexican government when the Aquaculture CRSP co-sponsored ISTA7.

Building on these partnerships should reduce our requests for CRSP funds while continuing collaboration in future years. As capacity is improved in the host countries, CRSP funds will be more highly leveraged. For example, in the early years of the PD/A CRSP, significant resources were used to build aquaculture capacity in Egypt, specifically at Abbassa. Today, Egypt has the second largest tilapia production (2005 = 445,000 metric tons) in the world. The PD/A CRSP clearly played an incubator role in this success, building physical and intellectual capacity. Now, Egyptian partners, like their Mexican colleagues, have the ability and interest to provide significant resources to partner with sustainable aquaculture efforts. ISTA 8 should highlight this progress and gain the attention of European vendors who may jump-start the international trade of Egyptian tilapia.

The goal to improve sustainability is especially important in the Americas where tilapia and shrimp are now the two major aquaculture crops. Even incremental improvements in tilapia productivity are magnified to affect thousands of farmers and employees. Abandoned shrimp ponds are targets for further conversion for urbanization unless they are restored into a mangrove-friendly aquaculture system.

LEAD US INSTITUTION: UNIVERSITY OF MICHIGAN

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PROJECT TITLE
IMPROVING SUSTAINABILITY AND REDUCING ENVIRONMENTAL IMPACTS OF AQUACULTURE SYSTEMS IN CHINA, AND SOUTH AND SOUTHEAST ASIA

AQUAFISH PROJECT THEME
ENVIRONMENTAL MANAGEMENT FOR SUSTAINABLE AQUATIC RESOURCES USE

Investigations

1. 07MNE03UM	Impact of Introduction of Alien Species on the Fisheries and Biodiversity of Indigenous Species in Zhanghe Reservoir of China and Tri An Reservoir of Vietnam
2. 07MNE04UM	Assessing Effectiveness of Current Waste Management Practices for Intensive Freshwater and Marine Pond Aquaculture in China
3. 07MNE05UM	Determining the Ecological Footprint of Shrimp Aquaculture Through Life Cycle Analysis of Outdoor Pond Systems
4. 07HHI01UM	Monitoring and Reducing Microcystins in Tilapias and Channel Catfish Cultured in a Variety of Aquaculture Systems
5. 07BMA02UM	Polyculture of Sahar (*Tor putitora*) with Mixed-sex Nile Tilapia (*Oreochromis niloticus*)
6. 07MNE07UM	Workshop on Aquaculture, Human Health and Environment

US & Host Country Institutions

USA	University of Michigan (lead US institution)
Asia	World Wildlife Fund in Asia, Vietnam staff
China	Shanghai Fisheries University (lead Host Country institution) Hainan University Huazhong Agricultural University Wuhan University
Nepal	Institute of Agriculture & Animal Science
Vietnam	University of Agriculture & Forestry

Other Collaborators and Linkages

Dong Nai Fisheries Company, Ho Chi Minh City, Vietnam
Huiting Reservoir Fisheries Management Company, Jingmen City, China
Rural Integrated Development Society-Nepal (RIDS-Nepal), Nepal
Sichuan Aquacultural Engineering Research Center, Chengdu, China
Zhanghe Reservoir Fisheries Management Company, Jingmen City, China

Project Summary

This project represents a collaboratively designed series of six investigations with host country counterparts in China, Nepal, and Vietnam. The host country scientists, in consultation with their university and government colleagues, were largely responsible for defining these experiments and studies as the most currently important research priorities. This project encompasses work on better aquaculture management practices, effects of effluents and invasive species on natural systems in the region, human health effects of aquaculture-related toxins, and methodology to quantitatively evaluate sustainability of different aquaculture systems in the study region and the world.

Investigation 1 evaluates effects of past introductions of exotic fish species (icefish in China; tilapia in Vietnam) on the local fishery and fish community in three reservoirs (Zhanghe and Huiting Reservoirs in China; Tri An Reservoir in Vietnam). This investigation focuses on existing natural systems with already introduced alien fishes and draws on comparative studies or historic data to determine changes in the fishery and indigenous fish community. The three study reservoirs are used for small-scale fisheries that provide economic benefits mainly to the regional rural poor. Changes in fish catch and fish communities will negatively affect, or already have affected, these poor fishers.

Investigation 2 evaluates effluent releases and pond water quality in a variety of intensive aquaculture systems used in freshwater, brackish water, and marine areas of China. The problem of surface water quality and the influence of aquaculture in degrading water quality is a primary concern of the Chinese government. However, the extent of this problem, as well as possible solutions, remains elusive. Tilapia production is important since China produces approximately 50% of the world's tilapia. Most of the production is in extensive to intensive systems managed by small-scale farmers. Field surveys and tests will evaluate the influence of these farmers on local water quality.

Investigation 3, while occurring in China, is global in focus and is associated with another current study on indoor recirculating systems. This assessment study applies well-established quantitative methods—life cycle assessment (LCA), life cycle cost analysis (LCCA), and mass balance modeling—to outdoor shrimp culture. Our intent is to produce clear metrics of environmental impacts and to compare the ecological footprint of two very different aquaculture systems. While LCA has been applied sporadically to fishery and aquaculture systems, the methodology to combine LCA and LCCA with mass balance analysis, has not yet been developed. Application of all three methods is important for both quantifying impacts and for making fair comparisons between aquaculture and other food production systems.

Investigation 4 focuses on the health-related issue of algal blooms and cyanotoxins in the low trophic-level species culture of tilapia, using current, mostly semi-intensive systems as test cases. This experiment investigates the prevalence of microcystins (the most controversial cyanotoxin) in culture water from various types of tilapia aquaculture, how concentrated they are in fish flesh, and if they can be cleared by depuration. These studies will help determine if further action, beyond depuration, is needed and at the same time help small-scale farmers in their sale of fish.

Investigation 5 focuses on the interaction between fisheries and aquaculture in Nepal. Sahar is an important Nepalese game fish that is declining in abundance. It becomes piscivorous at larger sizes. Tilapia is commonly cultured in Nepal, but sex control is not practiced, resulting in a mixed-sex culture of stunted tilapia. Polyculture of sahar and tilapia may provide a tilapia reproduction and stunting control option. Tilapia consumption by sahar would provide a production control method while sahar production would offer a valuable pond-produced, alternative fish for sale, thus reducing demand for sahar from wild populations. This experiment is an attempt to initiate such a polyculture system, which could spur a whole new industry for small-scale culturists in Nepal.

Investigation 6 offers a concluding workshop that shares the project's research results with a wider audience from the host institutions, government fisheries and public health agencies, private sectors, and NGOs. Local experts will also be invited to share their experiences on related topics.

All the project collaborators share in the dual mission of the AquaFish CRSP, which focuses on solving critical problems facing global aquaculture development and aquatic resource management in lower income countries. From a philosophical and methodological perspective, this mission provides a platform for collaborative research with the intent to develop host-country capacity for better problem solving. The project vision is for current and future work to focus both on solving important aquaculture problems in the region and developing the capacity for host-country investigators and their students to contribute to future research. This project continues a research interaction that was begun over 20 years ago with the goal to better understand regional resources and train the next generation of researchers to face the critical issues of aquaculture development.

Over its first five years, this project is designed for successful research outcomes to lead to future work. For example, additional experiments on sahar-tilapia polyculture will likely be undertaken to either refine best possible systems of production or to extend that technology to poor farmers. Following the previous research model, NGOs and government organizations will continue in their role of offering extension outreach of important components of this work. Future research developing from this project will focus on the next emerging issues. Some outcomes may indirectly lead to further project progress such as additional life cycle assessments of aquaculture and fishery crops and the impacts of invasive species. In this regard, the near-term vision is to continue with this research focus, either in clear next steps or in following a similar research philosophy applied to different systems.

In the long term, aquaculture systems that deal with eutrophication of natural waters in China and the region will be developed. This work may encompass water quality issues in rivers and lakes, with or without cage culture, as well as the development of cage culture systems to better retain nutrients. With Investigation 2, the focus is on effluent levels and management practices. Reduction of conflicts between aquaculture and invasive species is another area of mutual research interest. The investigation of invasive species effects on indigenous fish communities and aquatic ecosystems fits the goals of the current CRSP program mission.

LEAD US INSTITUTION: UNIVERSITY OF HAWAII AT HILO

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PROJECT TITLE
HUMAN HEALTH AND AQUACULTURE: HEALTH BENEFITS THROUGH IMPROVING AQUACULTURE SANITATION AND BEST MANAGEMENT PRACTICES

AQUAFISH PROJECT THEME
IMPROVED HEALTH AND NUTRITION, FOOD QUALITY, AND FOOD SAFETY

Investigations

1. 07IND03UH	Spat Collection, Growth Rates and Survival of the Native Oyster Species, *Crassostrea corteziensis* at Santa Maria Bay, Mexico
2. 07IND04UH	Oyster-relaying and Depuration in Open-water Locations.
3. 07WIZ02UH	Determination of Carrying Capacity of the Boca Camichin Estuary in Reference to Oyster Culture
4. 07HHI03UH	International Workshop for Aquaculture Sanitation
5. 07HHI04UH	Regional Workshop on Shellfish Culture and Sanitation
6. 07BMA04UH	Training in Best Management Practices for the Production of Molluscs in the States of Nayarit and Sinaloa.
7. 07HHI05UH	Microbiological Quality of Shellfish Growing Waters and Tissues
8. 07BMA05UH	Intensive Training and Internship in Bivalve Culture and Shellfish Sanitation

US & Host Country Institutions

USA	University of Hawaii at Hilo (lead US institution) Louisiana State University
Mexico	Research Center for Food & Development (CIAD), Mazatlán Universidad Autónoma de Sinaloa, Culiacán Campus Universidad Autónoma de Sinaloa, Mazatlán Campus
Nicaragua	Centro de Investigación de Ecosistemas Acuáticos-Universidad Centroamericana (Center for Research of Aquatic Ecosystems-Central American University: CIDEA-UCA)

Other Collaborators and Linkages

Comite Estatal de Sanidad Acuicola de Sinaloa (State Committee for Aquaculture Sanitation of Sinaloa [CESASIN])
Coastal Resources Center/University of Rhode Island (CRC/URI)
Ecocostas, Ecuador
Federation of Shrimp Cooperatives, Mexico
Fisheries Industry Technology Center/University of Alaska
Nicaraguan Ministry of the Environment (MARENA), Nicaragua

Pacific Aquaculture & Coastal Resources Center/University of Hawaii at Hilo (PACRC/UHH)
Pacific Shellfish Growers Association, Olympia, Washington
Sinaloa Institute for Aquaculture (ISA), Sinaloa, Mexico
SUCCESS project (USAID funded)
U.S. Food & Drug Administration (FDA), Washington, DC
Women's Oyster Culture Cooperatives of Nayarit, Mexico
Women's Oyster Culture Cooperatives of Puerto Penasco, Mexico

Project Summary

The project's research, training, and outreach investigations will add components of aquaculture research, development, and training to existing integrated coastal zone management programs for five estuaries in Mexico and Nicaragua. Design of the research activities is based on prior needs assessments, feasibility studies, management plans, and previous research findings. The overall goal is to increase capacity to implement best management practices in aquaculture sanitation as a means to improve human health through disease prevention and product quality and safety. This work also aims to develop bivalve culture as a means of increasing utilization of indigenous species, which are low on the food chain, have low technology requirements and have high value. Other species such as finfish and small-scale shrimp culture are also considered where appropriate. Work will focus on research to develop shellfish sanitation schemes to improve bivalve fisheries and farming; estimation of carrying capacity for a nationally important estuary to aid in planning for bivalve and other forms of aquaculture development; training in best management practices focused on food quality, safety, and shellfish sanitation; and training in a wide range of bivalve production methods. Two trials will be conducted to determine feasibility of cultivating a local oyster species and using relaying sites for oyster depuration. Expected outcomes include information critical to decision making and planning for coastal communities and economic development; increased capacity for extension agents and researchers to work in bivalve culture, fisheries management, and shellfish sanitation; improved extension services benefiting coastal communities; developing the basis for shellfish sanitation plans and classification of shellfish growing waters; improved food quality and safety for shellfish and other aquaculture products; improved prices and markets for products; and reduction in the incidence of food-borne illnesses related to aquaculture.

Improving the health and well being of stakeholders is the fundamental justification for aquaculture development. Aquaculture can affect human health through a wide variety of direct and indirect causal pathways, including but not limited to the relationship with environmental quality; use of natural resources (e.g., water, land, inputs); consumption of safe, high protein food products; increased household revenues to improve food security; and involvement of women, youth, and marginalized groups.

The ways in which users and resources are affected by and affect aquaculture are complex, not completely understood, and are dynamic in nature. Workers in this area must constantly update their knowledge and understanding of the processes involved, new technology, and the changing socioeconomic framework. CRSP stakeholder and expert panel meetings of the Africa, Asia and Latin America/Caribbean regions (2002) revealed two critical trends; (1) research and development of new aquaculture technology has been effective in laying the informational basis for development of subsistence aquaculture; and (2) the ability of researchers and extension agents to transfer and implement the outcomes of research and development has not kept pace with the rate of technological innovation or with the rapidly changing socioeconomic milieu of most developing nations and their communities. Technology transfer commonly lags technology development in any economic sector, but an opportunity exists to significantly strengthen the collective CRSP and associated stakeholders' ability for technology transfer in human health themes.

Similar issues affect the on-going, community-based coastal management efforts in Pacific Mexico. For this reason, the partners involved in the Santa Maria Bay Management Initiative in Sinaloa, Mexico, the Universidad Autónoma de Sinaloa (UAS), Coastal Resources Center/University of Rhode Island (CRC/URI), and the Pacific Aquaculture and Coastal Resources Center/University of Hawaii at Hilo (PACRC/UHH) have chosen to work closely with issues of aquaculture, environmental health, and associated socioeconomic aspects as part of a larger coastal management initiative. The Santa Maria Bay Management Plan and related initiatives in other coastal watershed areas such as Marismas Nacionales (National Wetlands) in Nayarit State encompass work to manage economic activities, including aquaculture, to maximize economic and social benefits for local residents, minimize environmental impacts, and improve productive efficiency for long-term sustainability. The investigations will be closely tied to on-going activities and will expand the breath of current initiatives. Similar coastal zone management efforts and development of sustainable, alternative livelihoods are taking place in Nicaragua, led by Central American University under sponsorship from USAID[1], the EU, and Japan. Much of this work has focused on conducting research and outreach that lays the foundation for development of bivalve culture, improvement of bivalve fisheries management, and best management practices for sanitation and food quality. Links between colleagues in Mexico and Nicaragua have been built through past CRSP-sponsored work, and efforts in both countries are moving forward in a parallel and complementary manner.

The project will build on current coastal and aquaculture management efforts to: (1) develop bivalve aquaculture; (2) determine the carrying capacity of a nationally-important estuary in relation to bivalve culture; (3) conduct research needed to establish shellfish sanitation plans and implement classification of shellfish growing waters; (4) build capacity through workshops, training, extension, and educational opportunities; (5) build networks of practitioners in these fields to enhance learning and collaboration; and (6) improve access to key information for decision-making and planning through publications, outreach, extension, and exchanges.

This project aims to further current efforts to develop indigenous species in Mexico and Central America focusing on bivalves such as clams, oysters, and scallops as a low-impact alternative to shrimp aquaculture and to more directly benefit poor coastal communities. A thriving bivalve fishery and aquaculture industry in Mexico and Nicaragua that yields safe, high quality products will create jobs, improve food security, and reduce the incidence of shellfish-borne illnesses. Training and extension in general food safety and quality for all aquaculture products will build capacity among producers and vendors to reduce risks and improve the value of their products. Additionally, this work will contribute to improving national capacity in Mexico and Nicaragua by training professionals (including one graduate student) to increase their knowledge in these fields.

U.S., regional, and global benefits are as follows:

- Development of bivalves as an important species group for the aquaculture industries in two countries, potentially establishing a model for alternative aquaculture that utilizes low technology, low-input forms of aquaculture.

- The LAC region benefits from the approaches that emphasize the use of native bivalve species for culture rather than importing bivalve seed.

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[1] The coastal program in Nicaragua led by Central American University is a five-year effort funded by the United States Agency for International Development Bureau for Economic Growth, Agriculture and Trade, Office of Natural Resource Management through an award to the University of Rhode Island with involvement of the University of Hawaii at Hilo and other partners such as TNC, WWF, and CI. This global project named "SUCCESS" (Sustainable Communities and Coastal Ecosystems) is carried out in Ecuador, Nicaragua, Tanzania and Thailand.

- Since few developing nations have shellfish sanitation plans, developing and implementing shellfish sanitation in Nicaragua and Mexico establishes a model for the LAC and other regions.

- Dissemination of current and innovative shellfish culture methods is enhanced by availability of training and outreach materials.

- Two countries benefit from developing professional capacity for bivalve culture and shellfish sanitation, as there are few professionals currently in the LAC region with this expertise.

- There is direct benefit to the U.S. as many US citizens visit Mexico and Nicaragua and consume shellfish there. Improving shellfish sanitation benefits US citizens who might be vulnerable to contaminated shellfish. Also, shellfish are transported within each of the target countries and within the LAC region (often illegally), so improvements in shellfish sanitation lessen the risk when consuming bivalves throughout the region.

LEAD US INSTITUTION: UNIVERSITY OF CONNECTICUT-AVERY POINT

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PROJECT TITLE
DEVELOPMENT OF ALTERNATIVES TO THE USE OF FRESHWATER LOW VALUE FISH FOR AQUACULTURE IN THE LOWER MEKONG BASIN OF CAMBODIA AND VIETNAM: IMPLICATIONS FOR LIVELIHOODS, PRODUCTION AND MARKETS

AQUAFISH PROJECT THEME
ENHANCED TRADE OPPORTUNITIES FOR GLOBAL FISHERY MARKETS

Investigations

1. 07MER01UC	Competition and Impacts Between use of low Value/Trash Fish for Aquaculture Feed Versus use for Human Food.
2. 07MNE01UC	Assessment of Diversity and Bioecological Characteristics of low Value/Trash Fish Species.
3. 07SFT01UC	Alternative Feeds for Freshwater Aquaculture Species
4. 07TAP01UC	Feed Technology Adoption and Policy Development for Fisheries Management
5. 07FSV01UC	Maximizing the Utilization of low Value or Small Size Fish for Human Consumption Through Appropriate Value Added Product Development

US & Host Country Institutions

USA	University of Connecticut-Avery Point (lead US institution) University of Rhode Island
Cambodia	Inland Fisheries Research & Development Institute (IFREDI), Phnom Penh
Vietnam	Can Tho University

Other Collaborators and Linkages

Australian Centre for International Agricultural Research (ACIAR), Nelson Bay, Australia
Fisheries Administration in Cambodia (FiA), Cambodia
International Development Research Centre (IDRC), Ottawa, Canada
National Oceanic and Atmospheric Administration (NOAA), International Sea Grant
Network of Aquaculture Centers in Asia (NACA), Bangkok, Thailand
Southeast Asian Fisheries Development Center-Aquaculture Philippines
The WorldFish Center, Malaysia

Project Summary

There is increasing demand and trade in the lower Mekong region of Cambodia and Vietnam for low value/trash fish for (1) local consumption (e.g., fresh, dried); (2) direct feed (e.g., livestock, high value species aquaculture); (3) fish meal production (e.g., poultry, aquaculture); and (4) value-added products (e.g., fish sauce). This project envisions sustainable freshwater aquaculture development in the Lower Mekong basin region of Cambodia and Vietnam, taking into consideration the balancing of social, economic, and environmental/natural resource needs and implications. The main driver of this project is the continued expansion of aquaculture and its dependency on capture fisheries for low value/trash fish for feed. This project also takes into account that: capture and culture fisheries continue to play an important role in the food security, poverty alleviation, and economies of both countries; the strong interdependency between capture fisheries and aquaculture; management of these two sub-sectors cannot be carried out in isolation of each other; there is increasing intra-regional trade; and there is increasing competition and conflict between the use of low value/trash fish for feed and human consumption. This project will address these issues through five separate but complementary investigations: (1) analysis of supply and demand, and support policy development addressing aquaculture/capture fisheries interactions; (2) assessment and management of the abundance and status of low value/trash fish fisheries; (3) development of alternative feeds and feeding strategies; (4) outreach and feed technology adoption; and (5) trade and value-added product development.

In the Asia-Pacific region, many capture fisheries resources have been largely overexploited and, as a result, development of aquaculture has been encouraged to provide the protein, income, employment, and export earnings for some countries. In Cambodia, for example, freshwater aquaculture production has increased rapidly over the last two decades, with an average growth rate of 16.3%. In 2004, aquaculture represented 8.3% of total inland fisheries production. In Vietnam, the annual growth of aquaculture has been about 10-13% during the last decade. The Mekong Delta region of Vietnam often contributes about 55-60% of the total aquatic production and more than 60% of total aquatic production for export of the whole country. Such a development trend implies that sufficient feed for aquaculture production will be available. One source of feed is low value/trash fish, (defined as fish that have a low commercial value by virtue of their low quality, small size, or low consumer preference). They are either used for human consumption (often processed or preserved) or used to feed livestock/fish, either directly or through reduction to fish meal/oil. There is a general lack of accurate information on how much low value/trash fish is currently used in the Asia-Pacific region, but a conservative estimate of 25% for livestock and aquaculture feed has been put forward. The uses of low value/trash fish are diverse and include: (1) local consumption, e.g., fresh, dried; (2) direct feed, e.g., livestock, high value species aquaculture; (3) fish meal production, e.g., poultry, aquaculture; and (4) value-added products, e.g., fish sauce.

Demand and trade in the region for low value/trash fish is increasing for both aquaculture and animal feeds. In Cambodia, for example, estimates indicate that at least 62 freshwater low-valued or small-sized fish species are used to feed inland aquaculture. These fish species represent both adult species that are commonly used as food fish, and also juveniles of commercially important fish species. Cage culture uses as much as 50% low value/trash fish in the total feed. In Vietnam, at least 11 species of freshwater, and increasingly a number of marine, low value/trash fish are used to feed inland aquaculture. The price of low value/trash fish has tripled since 2001 and it is predicted to continually rise as aquaculture expands. The use of artificial fish-based feeds and/or fresh fish resources has further increased pressure on wild fish stocks. Inevitably, a dangerous spiral has evolved where the demand for low value/trash fish for aquaculture feed has supported increased fishing pressure on already degraded resources. As aquaculture grows in the region, it probably will be difficult to meet the demand for low value/trash fish. There is a general concern that the rapid expansion of aquaculture may ultimately be constrained by the dependence on low value/trash fish and fish meal, popularly referred to as the “fish meal trap”. The Asia-Pacific countries may need to increase imports of fishmeal from the global market for the aquaculture industry, or replace these with other feed materials. There is a need to address the increasing aquaculture demand for low value/trash fish by improving feeds for aquaculture through replacement of direct feeding by pellet feeding, and reduction of fishmeal content by substitution of suitable ingredients in pellets.

Conflict is increasing between the use of low value/trash fish for feed and for human consumption. In some cases, such feeds are comprised of fish species traditionally used as cheap food for people, and this allocation of fish resources to aquaculture may result in negative impacts of food security and livelihoods. The economics of the different uses of low value/trash fish in different localities direct the fish one way or the other. There are also trade-offs between direct food benefit and the indirect employment and income generation opportunities afforded by feeding to aquaculture. There is debate that it would be more efficient and ethical to divert more of the limited supply to human food, using value-added products. Proponents of this suggest that using low value/trash fish as food for domestic consumers is more appropriate than supplying fishmeal plants for an export, income-oriented aquaculture industry that produces high-value commodities. On the other hand, food security can also be increased by improving the income generating abilities of poor people. The large volume of people employed in both fishing and aquaculture also has a beneficial effect. This raises some important questions regarding the social, economic, and ecological costs and benefits of aquaculture, as well as its sustainability and future trends.

This project focuses equally on the aquaculture of carnivorous fish and the management of lower value/trash fish. Investigations 1, 2, and 5 address the uses and bioecological characteristics of low value/trash fish. Investigations 3 and 4 address alternative feeds for freshwater aquaculture and feed technology adoption. This project will address the USAID Foreign Assistance Framework objective of economic growth, specifically in the program areas of agriculture (agriculture enabling environment and agriculture sector productivity) and environment (natural resources and biodiversity). The global research theme of the project is Enhanced Trade Opportunities for Global Fishery Markets. Human capacity development in aquaculture and fisheries, a goal of USAID foreign assistance, will be a central focus of this project. The project will also address USAID focal areas for broadening market access, improving nutrition and health, food quality, processing, and food safety of fishery products.

This project envisions sustainable freshwater aquaculture development and innovative fisheries management systems in the Lower Mekong basin region of Cambodia and Vietnam, taking into consideration the balancing of social, economic, and environmental/natural resource needs and implications. The main driver of this project is the continued expansion of aquaculture and its dependency on capture fisheries for low value/trash fish for feed. This project also takes into account: capture and culture fisheries continue to play an important role in the food security, poverty alleviation and economies of both countries; the strong interdependency between capture fisheries and aquaculture; management of these two sub-sectors cannot be carried out in isolation of each other; there is increasing local and intra-regional trade for low value/trash fish products; and there is increasing competition and conflict between the use of low value/trash fish for feed and human consumption. The work undertaken will be sustained after the life of the project by the partners in Cambodia and Vietnam, and through the development of partnerships with other regional organizations such as the Network of Aquaculture Centers in Asia (NACA), the Southeast Asian Fisheries Development Center/Aquaculture Department (SEAFDEC/AQD), and The WorldFish Center. Additional funding will be secured through such sources as the Australian Centre for International Agricultural Research (ACIAR), International Development Research Centre (IDRC), Association of Southeast Asian Nations (ASEAN), Asia-Pacific Economic Cooperation (APEC), and each country. Future activities associated with the project are the development of feed and feeding strategies for other fish species; further on-farm trials of feed formulations, policy and technology for trade and value-added product development for low value/trash fish, development of farm-made feeds, improved management strategies for capture fisheries, and policy development for sustainable aquaculture and capture fisheries.

LEAD US INSTITUTION: PURDUE UNIVERSITY

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PROJECT TITLE
IMPROVING COMPETITIVENESS OF AFRICAN AQUACULTURE THROUGH CAPACITY BUILDING, IMPROVED TECHNOLOGY, AND MANAGEMENT OF SUPPLY CHAIN AND NATURAL RESOURCES

AQUAFISH PROJECT THEME
INCOME GENERATION FOR SMALL-SCALE FISH FARMERS AND FISHERS

Investigations

1. 07MER02PU	Developing Supply Chain and Group Marketing Systems for Fish Farmers in Ghana and Kenya
2. 07QSD02PU	Development of Small-scale *Clarias* Fingerlings as Bait for Lake Victoria Commercial Fisheries in Western Kenya
3. 07WIZ01PU	Characterization of Pond Effluents and Biological and Physiochemical Assessment of Receiving Waters in Ghana
4. 07SFT06PU	Development of Locally Available Feed Resource Base in Tanzania
5. 07MER03PU	On Farm Verification of Tilapia-catfish Predation Culture

US & Host Country Institutions

USA	Purdue University (lead US institution) Virginia Polytechnic Institute & State University (VT) University of Arkansas at Pine Bluff (UAPB)
Ghana	Kwame Nkrumah University of Science & Technology (KNUST)
Kenya	Moi University
Tanzania	Sokoine University of Agriculture (SUA) Ministry of Natural Resources & Tourism, Aquaculture Development Division

Other Collaborators and Linkages

Fisheries Department, Ministry of Food & Agriculture, Ghana
Kenya Business Development Services (KBDS)
Kingorwila National Fish Center, Tanzania
Lake Victoria Environmental Management Project
Mbegani Fisheries Development Centre
National Investment Center (NIC)
Nyegezi Fisheries Institute
Tanzania Fisheries Research Institute, Tanzania
United Nations Food & Agriculture Organization (FAO)

University of Dar-es-Salaam, Tanzania
Water & Sewerage Company, Ghana

Project Summary

Most sub-Saharan African nations are net food importers due to rapid population growth, low agricultural productivity, high post-harvest losses, environmental degradation, political conflicts, and periodic natural disasters such as floods and droughts. African governments acknowledge in National Development Plans that urgent poverty reduction measures are needed to achieve the UN Millennium Development Goals by 2015, with most focusing on national poverty eradication strategies and improvements in food security.

Fish has always been an important part of the diet of the people of the continent but until recently, fish has primarily been harvested from the wild. Total fish output in some African nations such as Nigeria and Egypt continues to grow at accelerating rates and fish cultivation is now part of many rural agricultural enterprises. This has been encouraged by expansion of NGO developmental activities on aquaculture, improved aquaculture production technologies, recognition of over-exploitation of natural fisheries, decline in natural resources, and increased nutritional requirement of a rapidly growing population. These factors combine to make aquaculture an economically attractive agricultural production alternative in sub-Saharan Africa.

As the continent makes strides in aquaculture, there is the need to enhance capacity in value chain and aquatic resource management to ensure the long-term impact of aquaculture on rural communities and the nation. Project investigations cover the development of aquaculture supply chain for fish farmers in Ghana and Kenya, the development of catfish fingerling production as baits for the Lake Victoria commercial fisheries in Kenya, the study of pond effluents on watersheds in Ghana, the development of local fish feeds in Tanzania, and on-farm verification of tilapia-catfish culture in Tanzania.

The project vision is agribusiness-focused aquaculture in sub-Saharan Africa made possible through physical and human capacity development; new and better technology of fish production; growth of a whole chain of activities from farm to the consumer; better management of the natural resources; and increased profitability of fish production at the farm level. The whole chain of activities, beginning from management of natural resources, production and marketing of fish fingerlings and food fish to transportation and retail sales will significantly contribute to employment and income generation in these linked activities. Agribusiness-focused aquaculture will vitalize rural aquaculture entrepreneurship by providing capacity and opening up a larger market for rural aquaculture producers. Results from the investigations will help to achieve this goal for aquaculture in rural sub-Saharan Africa, and will provide additional employment and income generation that will create demand for other products, and thus support the growth of other rural economic activities.

PART II. INVESTIGATIONS

TOPIC AREA
PRODUCTION SYSTEM DESIGN & BEST MANAGEMENT ALTERNATIVES

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POLYCULTURE OF SAHAR (*TOR PUTITORA*) WITH MIXED-SEX NILE TILAPIA (*OREOCHROMIS NILOTICUS*)

Production System Design & Best Management Alternatives/Experiment/07BMA02UM

Collaborating Institutions & Lead Investigators

University of Michigan (USA) — James S. Diana
Institute of Agriculture & Animal Sciences (Nepal) — Madhav K. Shrestha
Shanghai Fisheries University (China) — Yang Yi

Objectives

1. Assess growth and production of Nile tilapia and sahar at different stocking ratios of sahar. Assess the appropriate stocking ratio of sahar to control recruitment of tilapia.
2. Develop the appropriate tilapia-sahar polyculture system for farmers.

Significance

Sahar or golden mahaseer (*Tor putitora*) is considered as the king of the Himalayan fishes (Bhagat 1992) and contributes a significant proportion of the natural stock of fish in Bangladesh, India, Nepal, and Pakistan (Islam 2002; Joshi et al. 2002). It is a high valued indigenous fish from hill streams of Nepal (Shrestha 1981). The fish possesses both food and recreational value, as it is renowned for its taste and sport. It also has potential for aquaculture development in the country (Rai et al. 2001). Despite its high value, the fish is in threat (Rajbanshi 2001). Its population is declining from its natural habitats due to overfishing; improper fishing practices like blasting, electric fishing, and poisoning; damming the water course; industrial effluents; and road construction (Rai et al. 2001). The declining trend in the population of sahar needs immediate attention for its conservation and rejuvenation in natural waters (Bista and Shrestha 2000). There are regular government programs to release sahar seeds to some lakes and rivers to maintain its population in nature. Therefore, culture of sahar in ponds may help to conserve and decrease fishing pressure in natural waters.

This popular game fish attains a maximum size of 270 cm and weight of more than 50 kg (Shrestha, 1990; Shreshta and Jha 1993). It can withstand wide range of water temperatures from 6 to 35°C (Petr 2002), providing the possibility of its culture in subtropical climates. Being an omnivore, it feeds on filamentous algae, insect larvae, small mollusks, periphyton on rocks, and locally made pelleted feeds (Shrestha 1997). However, growth of fish in captive condition is slower than in the wild (Shrestha 1997). Attempts have been made to breed and develop culture techniques for sahar, mainly in the mid hill regions (Shrestha 1986, 1997, 2001). These experiments have also been limited to monoculture of sahar. Growth of sahar is much more rapid in the southern, warmer climate of Nepal compared to the mid hills (Shrestha et al. 2004). Larger sahar also develop a predatory feeding habit useful in controlling tilapia recruitment in ponds (Paudel 2003, Acharya 2004, Yadav 2006). However, sahar need more than 2 years to attain 500 g and perhaps need 3 years to reach 1 kg, sizes that are appropriate for tilapia control.

Nile tilapia (*Oreochromis niloticus*) is the most important freshwater aquaculture species in the world (Chen et al. 2003). It is considered an ideal species for rural fish farming because of its desired characteristics (Pillay 1990, Teichert-Coddington et al. 1997). This species was introduced in Nepal in 1985 (Pantha 1993), however, its culture is limited to government farms and in research stations during this experimental stage (Shrestha and Bhujel 1999). Since Nile tilapia is a prolific breeder, recruitment in mixed sex culture causes stunting and reduced fish production for market (Pillay 1990, Focken et al. 2000). Nile tilapia, feeding low in the food chain (Welcomme 1996), is becoming popular with small farmers of southern Nepal. Proper management of mixed-sex culture can provide consistent production of seed and save seed cost. The predatory habit of sahar may be important in controlling tilapia recruitment by co-culture of sahar with mixed-sex tilapia. The combination of sahar and tilapia in a proper ratio may provide multiple harvesting of tilapia (two-three crops) with new recruits for continuous seeding. Sahar consumption may also limit tilapia recruitments and stunting in the ponds. Sahar may be harvested after two years. The purpose of this study is to evaluate co-culture of Nile tilapia and sahar.

Quantified Anticipated Benefits

The results of this study will provide an alternative strategy to monoculture mixed-sex tilapia for small-scale, resource poor farmers. It will add high valued fish in the culture system and will supplement income. It will also help in production of sahar and decrease fishing pressure in nature. It will benefit fish culturist in south Asia and other countries where mixed-sex tilapia is commonly cultured. Immediate impact will be measured by the increased production and economic returns in on-farm trials for the different culture systems.

Research Design & Activity Plan

Location**:** Institute of Agriculture & Animal Science (IAAS) and culture ponds in Chitwan, Nepal

Methods
1. Pond Research
1.1. Pond facility: 12 earthen ponds of 100 m^2 will be used for both on-station and on-farm trials.

1.2. Culture period: one year each for on-station and on-farm trials.

1.3. Test species: sahar (*Tor putitora*) and Nile tilapia (*Oreochromis niloticus*).

1.4. Stocking density: Mixed-sex Nile tilapia (10-20 g), 2 fish/m^2; sahar (20-30 g) at different treatment densities.

1.5. Nutrient input: Initial fertilization and alternate day feeding with locally made feed at 2%BW to sahar and tilapia once sahar have achieved 100 g.

1.6. Water management: maintain at 1 m deep.

1.7. Sampling schedule
Water quality: Standard CRSP protocol, biweekly water sampling and monthly diel analysis at various depths.
Fish growth: monthly sampling and at tilapia harvest (>200 g size) every four months.
Partial enterprise budget: variable costs and value of fish crops.

1.8. Statistical design, null hypothesis, statistical analysis:

On-station trial: There will be four treatments in triplicate each: (A) tilapia only (control); (B) tilapia plus sahar at 0.125 fish/m^2); (C) tilapia plus sahar at 0.25 fish/m^2; and (D) tilapia plus sahar at 0.5 fish/m^2. The trials will be conducted in completely randomized design, and data will be analyzed using one-way ANOVA and linear regression.

Null hypothesis: Stocking ratio of sahar to tilapia has no effect on growth, production, and economic return of both tilapia and sahar.

On-farm trial: On-farm trials will be conducted in collaboration with a NGO-Rural Integrated Development Society-Nepal (RIDS-Nepal), which promotes Women in Aquaculture, thus more than 50% women will be involved in the trial. There will be two treatments with six replicates each: (A) tilapia only (control); (B) tilapia plus sahar at the best stocking density determined in the on-station trial. The on-farm trial will be conducted in two locations with three replicates in each location using randomized complete block design. Data will be analyzed using two-way ANOVA.

Null hypothesis: There are no significant differences in the growth, production, and economic return of tilapia and sahar between tilapia monoculture and tilapia-sahar polyculture.

2. Workshop: Upon the completion of the analyses, a one-day workshop will be organized jointly with RIDS-Nepal to present findings from this project as a continuing consultation process with farmers and government officials. At least 50% of the workshop participants will be women.

<u>Schedule</u>

April 2007 – September 2009. Report submission: not later than 30 September 2009.

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CO-SPONSORSHIP OF "SECOND INTERNATIONAL WORKSHOP ON THE CULTIVATION AND BIOTECHNOLOGY OF MARINE ALGAE: AN ALTERNATIVE FOR SUSTAINABLE DEVELOPMENT IN LATIN AMERICA AND THE CARIBBEAN"

Production System Design & Best Management Alternatives/Activity/07BMA03UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)	Kevin Fitzsimmons
Universidad Autónoma de Tamaulipas, Ciudad Victoria (Mexico)	Pablo Gonzalez Alanis
Universidad Autónoma de Tamaulipas, Reynosa (Mexico)	Mauricio A. Ondarza
Instituto Sinaloense de Acuacultura, Mazatlán (Mexico)	Roberto Arosemena
BIOTECMAR (Venezuela)	Raul Rincones

Objectives

1. Organize and conduct a workshop focused on the use of marine algae in sustainable coastal aquaculture systems.
2. Provide travel support for CRSP partner biologists from Guyana and Mexico to participate.
3. Print and publish the proceedings of the workshop.
4. Build and maintain a website for on-line submission of papers and eventual web posting of PDF versions of papers and PowerPoint presentations from the workshop.

Significance

Marine macro-algae, seaweeds, comprise the largest category of aquaculture product on a global basis. Seaweeds provide a significant component of the diet for most Eastern Asian consumers and the production of agar and other phycocolloids from red algae provides employment for tens of thousands of small producers across Southeast Asia. With the exception of Chile, there are no significant seaweed farming areas in the Americas (Buschmann et al., 2001; Rincones, 2000). Seaweed farming is widely recognized as the most sustainable of any aquaculture production as it fixes nutrients and carbon dioxide from the environment and provides habitat for many marine species during its growth cycle. Seaweeds provide many edible products for direct human consumption, several products used in processed foods, and is a common ingredient in animal feeds.

Integration of seaweed culture with aquatic animal culture has been suggested as a method for reducing the environmental impact of aquaculture effluents (Yang, 2005; Nelson et al., 2001). Seaweeds provide substrate for nitrifying and heterotrophic bacteria needed to convert ammonia and organic compounds and then utilize the nitrogen, phosphorus and other nutrients in the effluent from the farms. The first International Workshop on Marine Algae was held ten years ago in Venezuela. South America, especially Chile, has seen some benefits from the workshop and now we hope to provide a further advance in the technology to Mexico and neighboring countries.

Quantified Anticipated Benefits

Integrated seaweed and animal culture would improve the sustainability of fish and shrimp farms in coastal areas and provide an additional cash income to the farmers. The seaweed farmers could be part of, or separate from the aquatic animal farm.

Specifically regarding the conference, two scientists from Guyana and two from Mexico will be provided with travel support. Also, two students from Mexico will be provided with travel support. All of the conference participants will benefit from the organization provided by the PI's and their students. The attendees and aquaculture practitioners will benefit from the conference proceedings. Two hundred copies of the proceedings are expected to be printed. We expect 100 people to attend and receive copies, the other hundred will be for later distribution. In addition, a website will be developed with an electronic version of the proceedings posted.

Activity Plan

Location: Mazatlán, Sinaloa, Mexico

Methods: The PI's have made preliminary plans and have gotten preliminary support from the State of Sinaloa fisheries department. Brochures and invitation for submission of papers are being printed and will be distributed at the World Aquaculture Meetings in Texas and at several meetings and workshops in Mexico. An effort will be made to widely announce the meeting at academic and governmental institutions in Latin American countries. The steering committee includes scientists from Venezuela, Mexico, and the US.

Schedule

July 2007	Planning workshop, submission of papers,
August 2007	Make travel plans for those with travel support
July 2007	Prepare website with workshop details and on-line submission
September 2007	Conference in Mazatlán, Sinaloa
October 2007	Posting of PDF versions of papers and PowerPoint presentations
December 2007	Final Report submission

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TRAINING IN BEST MANAGEMENT PRACTICES FOR THE PRODUCTION OF MOLLUSCS IN THE STATES OF NAYARIT AND SINALOA

Production System Design & Best Management Alternatives/Activity/07BMA04UH

Collaborating Institutions & Lead Investigators

Pacific Aquaculture & Coastal Resources Center University of Hawaii at Hilo (USA)	Maria Haws
Louisiana State University (USA)	John Supan
Centro de Investigación de Alimentos y Desarrollo (CIAD), Mexico	Omar Calvario Martínez
Universidad Autónoma de Sinaloa, Culiacán Campus (Mexico)	Eladio Gaxiola Camacho
Universidad Autónoma de Sinaloa, Mazatlán Campus (Mexico)	Guillermo Rodriguez Domínguez

Objectives

Disseminate and provide training in Best Management Practices (BMPs) for molluscan culture in two Mexican states with shellfish aquaculture industries to improve key aspects of production and sanitation. The end goal is to enable farmers and vendors to produce and sell shellfish that is high quality and safe to consume, as well as to improve farm efficiency and profitability

Significance

CIAD has researched methods and produced manual in Best Management Practices for bivalve, trout and shrimp culture (Garcia-Ortega 2007; 2003; Calvario 2007; 2003) and has been able to provide a limited amount of training to producers and vendors. Other team members (UHH, UCA) have also produced a wealth of research and training materials (Boyd et. al. 2001; Calvario and Montoya 2003; Haws and Martinez 2005; Rojas et. al. 2005) related to best management practices for various forms of aquaculture. Training in these topics has proved to be of value in locations such as Honduras, Nicaragua, Marshall Islands and Tanzania in encouraging the adoption of BMPs if coupled with on-going extension assistance. This work will focus largely on aspects of BMPs related to sanitation, although other topics will be included as appropriate. The human health and aquaculture case studies (Haws et. al. 2006) identified lack of awareness and capacity for implementation as a key obstacle to adequate food safety and sanitation levels. There is also a wealth of other resources for training in this topic and related production aspects; lack of information is not an obstacle, but rather dissemination to key target groups and on-going, hands-on training. Some materials such as the Interstate Shellfish Sanitation guidelines also need to be translated to Spanish.

In this activity, researchers and extension agents from UAS and CIAD will work together with assistance from the UHH and UCA partners to develop training materials, conduct training workshops for farmers and other stakeholders such as processors and vendors, conduct follow-up monitoring to assess the success of technology transfer and adoption, and provide reinforcement training where necessary. CESASIN will also play a key role in this work as they are the principal providers of technical assistance and diagnostic services in Sinaloa for human and animal health topics related to aquaculture. They provide on-going extension services to most of the aquaculture producers in this region.

This work will focus on small-scale oyster farmers, women's groups that work mainly in post-harvest tasks and development of products such as pickled oysters, and vendors (largely women) who sell live, raw shellfish and fresh products such as ceviche (raw seafood cocktail) and pates.

This work is particularly crucial given the local preference to consume live, raw shellfish products which are most vulnerable to contamination by *E. coli*, salmonella, and Hepatitis. The women's groups involved in this work also produce a wide range of products that are also likely to be affected by shellfish-borne disease as well as contamination incurred during the processing and preservation stages. Training materials will be shared with any other interested institutions but particularly in Nicaragua and Ecuador where extension agents are working with similar community groups.

This work continues previous efforts for implementation of the Bahia Santa Maria Bay Management Plan where the primary stakeholder groups are women's groups. These women work primarily in shrimp and fish processing plants and as fishers. In the latter role, they primarily gather bivalves when not working at home or in the processing plants. These women's groups have for many years expressed the desire to begin growing oysters as they view this as a more profitable activity that would allow them more flexibility with fulfilling their work as mothers and home-makers. The project's work includes strategies to enhance the participation of women using simple methods such as appropriate scheduling to prevent conflicts with other work and home duties, liaising directly with the women community leaders, and working to involve male companions and leaders in activities which benefit women. In the case of Boca Camichin, oyster production has been in place for about 30 years. Women do work nearly co-equally with men on the oyster culture raft systems, but take on a larger role in the post-harvest activities such as processing and sales and thus have a direct role in sanitation issues. This work has a large emphasis on training stakeholders in improved sanitation methods for all stages of production, and thus women are the co-equal target audiences and participants in this work. The fundamental driving force in much of aquaculture sanitation is improved community sanitation.

As awareness is raised regarding the need to improve sanitation for all aspects of community life and production, women have been shown to be particularly critical actors. For example, in past work in these areas, it has been women who have organized and taken responsibility for community clean-ups, development of solid waste management systems and the building of latrines. Women and children are also thought to be particularly vulnerable to diseases carried by contaminated shellfish. In both Mexico and Nicaragua, women and children often gather shellfish, generally near inhabited areas, largely to satisfy household needs. They are therefore more likely to consume contaminated shellfish and may have other nutritional and health issues that lower immunity.

Quantified Anticipated Benefits

Training and on-going extension support will increase producers, processors and vendor's capacity to produce and sell healthy, safe shellfish products. Value will also be added to the products through consumer perception that safe products are produced, and through extending the shelf-life of the products. The danger of serious diseases associated with shellfish and products will be significantly reduced.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; State Committee for Aquatic Sanitation (CESASIN); and the Federation of Shrimp Cooperatives. Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries.

Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems" (SUCCESS).

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of training sessions: 6
- Number of institutions directly or indirectly benefiting: 19
- Number of individual participants in technical training: 20-30 each site
- Number of communities benefiting: 20
- Students involved: 1 graduate student
- Training modules produced: 3
- Publications: 1

Activity Plan

Three, two day workshops will be held in both Nayarit and Sinaloa in communities where shellfish are farmed, fished and processed. One course will be held every three months. Training materials will be developed by UAS and CIAD and jointly implemented. Between courses, UAS, CESASIN and CIAD will provide extension support for monitoring and reinforcement to assure that recommended methods and processes are adopted.

Drs. Haws and Supan will assist and participate in this work. The UAS Masters degree student will assist with this work.

Schedule

Preparation for training will begin in May 2007. The workshops will be held in August and November (2007) and February (2008) to prepare for, and coincide with harvest and processing seasons which are timed for the Christmas and Easter season when seafood consumption peaks. Training materials will be published by May 2008.

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INTENSIVE TRAINING AND INTERNSHIP IN BIVALVE CULTURE AND SHELLFISH SANITATION

Production System Design & Best Management Alternatives/Activity/07BMA05UH

Collaborating Institutions & Lead Investigators

University of Hawaii at Hilo (USA)	Maria Haws
Louisiana State University (USA)	John Supan
Central American University (Nicaragua)	Agnes Saborio Coze Nelvia Hernandez
Universidad Autónoma de Sinaloa, Culiacán Campus (Mexico)	Eladio Gaxiola Camacho
Universidad Autónoma de Sinaloa, Mazatlán Campus (Mexico)	Guillermo Rodriguez Domínguez

Objectives

This work aims to increase capacity among host country professionals and scientists in bivalve culture and shellfish sanitation.

Significance

Although shellfish fisheries and culture are of increasing importance and concern, capacity among host countries in Latin America is generally low. This work will increase capacity for three host country nationals (1 from Mexico, 1 from Nicaragua and 1 from Ecuador) in a wide range of topics related to bivalve culture (hatchery, grow-out, farming, hurricane resistance) and shellfish sanitation (microbiological, legal, economic and implementation).

Activity Plan

Louisiana is the largest oyster producing state in the U.S. and Louisiana State University is uniquely suited for training in topics related to oyster culture and shellfish sanitation due to the research, extension and industry support it has traditionally provided. State agencies are also active in supporting the oyster culture industry. Fishing, aquaculture, and processing facilities are also conveniently located and accessible to LSU staff. A two-week workshop will be held at the LSU Grand Isle oyster hatchery, followed by two weeks of practical training in the form of internships for host country nationals. Topics will include: hatchery production, nursery culture, innovative oyster culture methods, shellfish sanitation, commercial oyster farming, oyster processing, water quality monitoring for shellfish sanitation, and post-harvest handling of oysters. Most of the work will occur at the LSU oyster hatchery, but visits will be made to other professionals involved in the industry (i.e., State agencies), commercial oyster farming operations, processing plants, retail venues and other industry facilities. Dr. John Supan will be primarily responsible for planning, design, and execution of this work.

The UAS and UHH Masters degree students will participate in the workshop and the LSU student will both participate and assist. Ms. Nelvia Hernandez from CIDEA/UCA will also attend the workshop and will stay on for an additional two weeks of practical work.

Schedule

This work is tentatively scheduled for May 2008.

TOPIC AREA
SUSTAINABLE FEED TECHNOLOGY

❖

ALTERNATIVE FEEDS FOR FRESHWATER AQUACULTURE SPECIES

Sustainable Feed Technology/Study/07SFT01UC

Collaborating Institutions & Lead Investigators

University of Rhode Island (USA) David Bengtson
Can Tho University (Vietnam) Tran Thi Thanh Hien

Objectives

The objective of the study is the development of cost-effective alternative feeds for carnivorous freshwater species to replace or reduce the dependence on low value/trash fish.

Significance

Aquaculture of freshwater carnivorous and omnivorous fish species in Cambodia and Vietnam is highly dependent on inland fisheries of low value fish for sourcing key dietary nutrient inputs. In Cambodia, the use of low value/trash fish applies mainly to commercial-scale cage culture rather than small-scale pond culture (Heng et al. 2004). Commercial-scale cage culture in Cambodia contributes about 70% of the country's total aquaculture production (DOF 2005). The major fish species for cage culture are river catfish (*Pangasius hypophthalmus*, *P. bocourti* and *P. larnaudii*) and snakehead (*Channa micropeltes*). The main feed for pangasid catfish and snakehead is low value/trash fish. Feed conversion ratios range from 1.3 to 1.5. Fresh fish feeds are available seasonally and regionally, which has led to problems of overfeeding in times of plenty and underfeeding when feeds are scarce. There is plenty of fresh fish for feed available during the peak period of fish catch in the country, particularly from November to January. Small cyprinids are caught along the Tonle Sap River. For the rest of the year, some farmers use dried fish but others are not able to afford this input. During the peak fishing season, a large bulk of low value fish is sun-dried. This processing sometimes leads to contamination with sand and dust, which reduces the quality of the dried fish produced. Manufactured pellet feeds are expensive and imported. Only a very few producers can afford the high cost of the feeds and they are primarily used for ornamental fish.

There have been very few studies conducted on feed and feeding in Cambodia (Heng et al. 2004). There is no tradition of on-farm feed formulation that can be widely used in aquaculture systems. Pond fertilizer techniques are well understood by farmers but organic manures are scarce since they are needed for agricultural crops. The market price for farmed fish, especially in relation to the cost of feed, is a major problem. Prices are very low when fish are plentiful from capture fisheries and consumers prefer wild captured fish to cultured fish.

Heng et al. (2004) recommend cooperation on feeds and feeding technologies through research and experimentation, especially suitable feed and feeding strategies. In Vietnam, more than 70% of the fish species cultured are omnivores (Hung 2004). Catfish of the *Pangasius* and *Clarias* genera are the most important omnivores cultured in Vietnam. The omnivores are cultured in cages in the Mekong River, mainly in An Giang and Dong Thap provinces. Traditional feeding is largely based on trash fish, a protein source that is quite limited in Vietnam. The traditional feeding method is to prepare and mix ingredients on the spot and cook them to produce a wet, sticky paste. Feed conversion ratios vary from 1.5 to 2. Floating pellet feed is expensive and is not suitable for small-scale farmers who cannot afford it.

The carnivores (approximately 10% of fish species cultured) are all indigenous species with a high market value (Hung 2004). The two main species are *Channa micropeltes* and *C. striatus*, estimated at 90-95% of cultured carnivores. Low value/trash fish is the only feed source for the carnivorous fish. Low value fish are caught in reservoirs and in the Mekong River during the flood season. Farmers also use marine low value/trash fish. The feed conversion ratio is approximately 4 to 5. The pressure on the low value fish supply increases the price and makes the production system "unstable and unsustainable" (Hung 2004 p77).

Hung (2004) recommends that research is needed on:

- Alternative feeds for carnivores to replace or reduce the dependence on trash fish
- Identification of locally available ingredients from which home-made feed can be prepared.

During the past three years, scientists at Can Tho University (CTU) have been involved in an ACIAR project to study the use of alternative feeds for omnivorous fish, primarily *Pangasius* catfish and have made much progress. The study proposed here will therefore concentrate on use of alternative feeds for production of carnivorous species, primarily the *Channa* snakeheads. Also during the past three years, scientists at the University of Rhode Island (URI) have studied the replacement of fish meal with plant proteins in diets for juvenile summer flounder, *Paralichthys dentatus*. Because this is a carnivorous fish with protein requirements similar to that of snakehead (about 50% protein in the diet), we expect that the experimental methods and findings with flounder may transfer to some degree to the snakehead. We are now able to replace 40% of the fish meal in the flounder diet with a combination of soybean meal, corn gluten meal and canola protein concentrate and achieve the same growth as with fish meal alone. When one also adds certain feed additives, food conversion ratio (FCR) is also significantly improved. Such changes could reduce the feed costs for flounder by 10-20%. In Vietnam, a number of plant products (rice bran, cassava meal, etc.) are cheaply available and soybean meal is imported and the ACIAR study has already characterized the availability and composition of these products. Thus, an investigation of the use of fish meal and plant proteins for snakehead diets to replace the use of trash fish seems to be a worthwhile strategy. As mentioned above, use of trash fish to feed aquaculture species is an unsustainable strategy and research must be conducted now to find alternatives for the future.

It should be noted that fish meal replacement by plant protein may have an effect on the taste and flavor of the fish and on consumer acceptance. This potential taste and flavor change and consumer acceptance will need to be studied.

Quantified Anticipated Benefits

The results of this study will provide information on alternative diets for snakehead, especially those diets that incorporate locally available plant materials, in order to build a long-term sustainable industry. Through an economic analysis of costs of the diets (based on costs of fish meal and plant proteins vs. trash fish) and the risks of the unavailability of trash fish in the future, the information provided from this study will allow decisions to be made about the development of feed mills for local production of diets for the snakehead industry.

Research Design & Activity Plan

Location of work: Formulation of diets will be done at the URI based on information about chemical composition of locally available plant products in Vietnam. Manufacture of the diets will be done at CTU All feeding trials will be conducted in a wet lab at Can Tho University, which has a small fish feed mill (for sinking feed, 50-200kg/hour), as will analysis of diet composition: protein. lipid, mineral, fiber, and energy.

Methods: Diets will be formulated based on what little is known about the nutritional requirements of snakehead (Samantaray and Mohanty, 1997; Arockiaraj et al., 1999), with supplemented information from other carnivorous species to fill in gaps where snakehead requirements are unknown. A basal diet will include fish meal as the only protein source and alternative diets will include fish meal replaced by various levels of plant proteins. A series of experiments (feeding trials) will be conducted at CTU with two snakehead species, *Channa micropeltes* and *Channa striata*, that are being raised widely in the Mekong Delta). In the first, rice bran will be used to replace fish meal at replacement levels of 0, 20, 30, 40 and 50% of the fish meal. Thus, the experiment will have five treatments, each with three replicates. In addition there will be a control treatment, in which the snakehead are fed their current normal diet of trash fish. The experiment will last 12 weeks and fish will be fed to satiation every day, with uneaten food removed from the tank daily for calculation of food consumption and FCR. The second and third experiments will be the same as the first, except that cassava meal and soybean meal will be used as the replacements for fish meal. These experiments, all performed during year 2 of the study, will provide background information on growth of snakehead with a) fish meal compared to trash fish, and b) different levels of three kinds of plant protein replacing fish meal. We expect that different plant proteins might be able to replace fish meal at different levels in the diet (e.g., soybean meal might be able to replace 30% of the fish meal in the diet with no reduction in growth compared to fish meal alone, cassava meal might replace 40%, rice bran might replace 20%, etc.).

During year 3 of the study, the diets that allowed maximum levels of replacement of fish meal will be modified by adding certain components to them (e.g., phytase and taurine) and trying to increase the levels of fish meal replacement. Carnivorous fish that have difficulty with plant proteins in the diet can often be helped if phytase is added to the diet (phytin in plant material can bind up necessary minerals). Recent evidence has also shown that taurine, which is abundant in fish meal but not considered an essential amino acid, might limit growth of fish whose diets have reduced levels of fish meal (Gaylord et al., 2006). Identification of specific studies for year 2 will depend on the results of the feeding trials in year 1, but will generally include the following types of treatments: treatment 1) control diet with maximum addition of plant material as determined in year 1 (e.g., 30%), treatments 2-5) higher levels of fish meal replacement (e.g., 40, 50, 60, 70%) plus phytase. The same approach can be taken with additions of taurine rather than phytase. Finally, we will use combinations of plant proteins (e.g., rice bran plus cassava meal plus soybean meal) to increase the maximum levels of fish meal replacement.

To assess the impact on consumer tastes and preferences for fish fed with plant protein rather than fish meal, a consumer food choice and acceptance analysis will be undertaken. Consumer studies have shown that taste and flavor are important factors in consumer food choice (Clark 1998, Shepherd 1985). A number of both qualitative techniques (observation, group discussion, projective techniques, in-depth interviews, repertory grid, laddering) and quantitative techniques (preference mapping, conjoint analysis, scanned panels) are available to study consumer food choices. Due to limited resources, this project will rely on test marketing to assess consumer tastes and preferences for the two different fed fish. A sample of students at Can Tho University will be provided with samples of similarly cooked fish of the two types and asked to complete an anonymous self-administered questionnaire. They will be asked questions on aroma, taste, texture, and appearance. They will also be asked whether or not they would purchase the fish for their own use. A statistical analysis of responses will be conducted.

Schedule

Year 1 — One URI PI to travel to Vietnam to observe facilities and prepare for conduct of experiments, formulation of diets, and ordering of supplies.

Year 2 — Six feeding trials (three with each species) of 12 weeks duration each (= 36 weeks + time for setting up and breaking down experiments, analyzing data, etc).

Year 3 Six feeding trials (three with each species) of 12 weeks each. From year 2 results, preparation of publications for peer-reviewed journals, dissemination of information via workshops, fact sheets, etc. Consumer food choice study will be undertaken.

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FEEDING REDUCTION STRATEGIES AND ALTERNATIVE FEEDS TO REDUCE PRODUCTION COSTS OF TILAPIA CULTURE

Sustainable Feed Technology/Experiment/07SFT02NC

Collaborating Institutions & Lead Investigators

North Carolina State University (USA)	Russell Borski Peter Ferket Charles Stark
Central Luzon State University (Philippines)	Remedios Bolivar Emmanuel Vera Cruz
GIFT (Genetically Improved Farmed Tilapia) Foundation International, Inc (Philippines)	Hernando Bolivar

Objectives

1. Assess combined feed reduction and feeding delay strategies on growth performance of Nile tilapia.
2. Assess reduction of the daily feed ration as a function of body size in the grow-out culture of Nile tilapia on the farm.
3. Reduce feed cost and wastage by utilizing least cost formulation and feed manufacturing technology that limit fishmeal and maximize usage of local Philippine ingredients.
4. Field-test the efficacy of using IGF-I as a biomarker of growth in pond-cultured tilapia.

Significance

Feed is widely recognized as the most costly component of fish farming. A cost-farm budget analysis shows that feed constitutes 60-80% of total production costs for small-scale, rural farmers in the Philippines (ADB 2005). Because of this, any reductions of feed costs can increase income effectively for Philippine farmers. Reductions in both the amount of feed used for growout of marketable fish and in the cost of formulated feeds are two approaches to containing feed costs.

The Aquaculture CRSP at CLSU has generated grow-out tilapia feeding options, which have been found to be cost-saving strategies (see Bolivar et al. 2006). First, by delaying the onset of supplemental feeding to either 45-days or 75-days in fertilized ponds we reported that tilapia fingerling feed consumption was significantly reduced without any negative impact on the production of marketable tilapia (Brown et al., 2000). Secondly, feeding at a sub-satiation level did not reduce fish yield; a 100% satiation diet reduced to 67% did not significantly reduce measurable production of marketable fish (Bolivar et al., 2003). Feeding only on alternate days saved approximately half of feed cost without a significant reduction in growth, survival, or market yield of Nile tilapia in growout ponds (Bolivar et al. 2006). In the proposed studies we will examine the utility of combined feeding delay and reduction as they may affect growth performance, as an additional feeding option for tilapia farmers to reduce costs. Through information founded on the CLSU studies above and there own pragmatism, Philippine tilapia farmers believe reducing daily feeding as a percentage of biomass may also yield feed cost savings relative to traditional full feeding schedules, which will be tested on farm in the proposed studies.

Commercial diets are rising sharply in cost as a consequence of market demand and declines in the fisheries that are the source of fishmeal. Around 33-44% of feed costs are attributable to fishmeal that constitutes15-20% of feed formulation. Much of the fishmeal used in the Philippines is imported from Peru and costs are expected to rise in the future as global supplies become constrained by increasing demands from other aquaculture and declines in commercial bait fisheries. Because tilapia are omnivorous fish which naturally feed on plankton, diatoms, small

crustaceans, algae, higher plants and decomposing vegetable matter, they do not require fish in their diet and they are an ideal group of species to recycle food by-products into high quality food protein for humans (Brown, 1983). Tilapia are capable of digesting high levels of carbohydrate in their diet (Anderson et al. 1984; National Research Council 1993), and effectively utilizing alternative feed ingredients, such as rice bran, cocoa, various flowers,
soya, nut oil, milling waste, brewer's wastage, poultry by-product meal and feather meal, cassava, and ipal-ipal leaf (Jackson et al., 1982). All of these lower-cost ingredients are readily available in the Philippines to completely replace or significantly reduce fishmeal in formulated tilapia diets. Indeed, various animal (meat, poultry, feather, blood and bone meal by-products) and plant proteins (soya, copra, cottonseed and others) have been shown to be either partially or in a few cases, namely with animal by products, completely effective in replacing fishmeal in tilapia diets (El-Sayed 1998; for reviews see Lim and Webster, 2006 and El-Sayed 2006). We have determined that tilapia could be fed diets containing up to 33% sweet potato and lactic acid-stabilized poultry carcasses (60:40 co-extruded blend) without adverse effects on growth performance or consumer panel sensory indices (aroma, flavor and texture) (Middleton et al., 2000). However, the use of food by-products to produce least-cost fish feed is primarily
constrained by their poor nutrient content, poor digestibility, or poor functional properties in manufacturing feed that can withstand the rigors of pond feeding (Li et al., 2006). Few studies have addressed the combinations of animal and plant protein types that might suffice in replacing or significantly reducing fishmeal in tilapia feed. Also, most investigations focus on the performance and nutritional characteristics of different protein sources rather than their ability to improve profit margins in tilapia production (see El-Sayed 2006).

Better feeds require high water stability and durability; pellets that break up will leach nutrients leading to poor feed conversion and performance (Leonard, et al. 2002). Diet formulation (40%), ingredient particle size (20%), conditioning (20%), die specs (15%), and cooling (5%) are the major factors that influence pellet quality (Axe, 2002). Nutritionists control 60% of the factors related to pellet quality through formulation (40%) and ingredient particle size (20%). Proteinaceous and starch materials have important implications in terms of producing a water-stable feed and in calculating least-cost formulations to meet nutrient specifications (Rokey and Huber, 2005). Fishmeal replacement strategies must therefore address the role of ingredients in the feed manufacturing process and ultimately their impact on feed water stability. Ironically, diets formulated on a least-cost basis often lead to greater feed wastage and higher production costs to the farmers. We propose to develop tilapia feed formulations and feed manufacturing technology to maximize the dietary inclusion of low-cost agricultural by-products at the expense of high-cost fishmeal.

Evaluation of the growth of a particular fish strain or of any parameter on growth performance usually requires numerous and costly production trials. Such growth trials are expensive, time-consuming and labor-intensive. Research progress is also limited by the time required to see gross changes in body weight or length for a specific growth trial. There is a need, therefore, for development of a means for rapid and direct assessment of growth of fish exposed to a particular test parameter over short periods. Insulin-like growth factor-I (IGF-I) is a highly promising candidate to be used as an instantaneous growth indicator in fish (see Picha et al. 2006; Luckenbach et al. 2007; Vera Cruz et al. 2006 and citations therein). Laboratory tests show circulating IGF-I levels can change within 4 days and that these changes closely parallel subsequent changes in growth (Picha et al. 2006). Our previous Aquaculture CRSP research demonstrates that tissue expression of the IGF-I gene is strongly correlated with Nile tilapia growth status under different temperature and feeding manipulations in the laboratory. In the present investigation we will build on this technology and test the efficacy of tissue IGF-I gene expression, along with blood circulating hormone levels (a non-lethal measure), as a corollary to the growth rate of tilapia in the field, *i.e.* in ponds.

Quantified Anticipated Benefits

- Research will take place at CLSU and on at least seven commercial tilapia farms. It will establish the utility of practical feeding strategies in containing feed costs without negatively impacting yield.
- The work will provide research training and educational experiences of an additional five graduate and seven undergraduate students at CLSU and NCSU. An important aspect will be the synergistic experiences among CLSU students, staff, faculty and farmers in the region.
- The field-tested assays for IGF-I could provide a good measure of growth status in tilapia precluding the need for costly and time-consuming grow-out experiments when testing specific parameters of performance. Establishment of biotechnology-based tools such as this can be used to advance aquaculture in the developing world. This agrees with current USAID goals, which specifically refer to the promotion of the development of sustainable agriculture technologies without use of GMOs.
- The proposed work will develop least-cost feed formulations and feed manufacturing technologies that minimize or eliminate the inclusion of fishmeal in favor of agricultural by-products available in the Philippines. The proposal will provide feed manufacturers in the Philippines feed formulation and manufacturing specifications developed and training material for the production of least-cost tilapia feed.

Research Design & Activity Plan

Location: This investigation consists of a series of studies that will follow-up on our previous CRSP-sponsored research to assess the utility of feed reduction and delayed feeding strategies in containing feed costs in the production of tilapia. Additional studies will determine the value of the least-cost feeds including several locally available feed ingredients in place of fishmeal, and develop feed manufacturing technology that will minimize feed wastage in pond-reared tilapia. In each of these studies, growth and feed conversion will be monitored at the Freshwater Aquaculture Center or at nearby tilapia farms in Central Luzon. Direct measurements of growth parameters will be supplemented by the assessment of
IGF-I, to field-test this method as a rapid indicator of growth rates. Here we will seek correlations between growth under various farm conditions in the Philippines with the hepatic expression of IGF-I mRNA and circulating IGF-I. Development and initial testing of feeds will be done at NCSU.

Methods:
1. Growth Performance of Nile Tilapia in Ponds Using Combined Feed Reduction Strategies: This study will combine the different feeding strategies that have been tested in a previous CRSP work plan into one feeding schedule that incorporates both delayed and reduced feeding. There will be two (2) treatments. This study will be done on-farm for 120 days using fingerling size #24 (0.1 g).

T1: 60 days delayed feeding (delayed from the typical period where fish are fed commercial feed after stocking of the fingerlings — fish under these conditions graze on algae and zooplankton resulting from the primary productivity of fertilized ponds); 30 days alternate day feeding (the amount of feed will be given in full amount but on alternate day), and 30 days full feeding on a daily basis but at a sub-satiation level of 67%.
T2: as a control, 100% amount of feed based on fish biomass will be given using commercial feed.

Ponds will be stocked with sex-reversed Nile tilapia (*Oreochromis niloticus*) fingerlings at a density of 4 fish m^{-2}. All ponds will be fertilized weekly using inorganic fertilizers (urea [46-0-0] and ammonium phosphate [16-20-0]) at a rate of 28 kg N and 5.6 kg P ha-1 week-1). Pond primary productivity will be monitored weekly by a combination of chlorophyll-a measurement and the Secchi disc visibility test from water sampled from ponds at 9 am each morning. Water quality will be monitored weekly (Bolivar et al., 2007). Fish will be fed twice daily. Fish will be sampled every two weeks using cast net method. Fifty fish will be weighed individually for growth rate determinations (weight and length) as well as for feeding adjustment (Bolivar et al. 2007). A subset of animals will be sampled for tissue (liver) and blood (fish >10 g) for IGF-I mRNA (Taqman real-time PCR; Vera Cruz et al. 2006) and circulating IGF-I (using `Universal' IGF-I reagents; Picha et al. 2006) determinations respectively, corollaries with growth. After 4 months of culture fish will be harvested by seining and complete draining of the pond. The total weight of fish stocks will be recorded at harvest and a subset of fish will be measured for circulating levels and gene expression of IGF-I. Feed conversion ratio (FCR), feed input, specific growth rates, and total production biomass will be calculated. A basic marginal cost-benefit analysis will determine if the mixed feeding strategy can provide savings (Bolivar et al. 2007). Data will be analyzed by a combination of repeated measures analysis, t-test and regression analyses.

Null Hypotheses: (1) The mixed feeding schedule will not affect growth performance or other production parameters of tilapia growout relative to continuous full daily feeding; and (2) Circulating IGF-I or hepatic IGF mRNA levels will not correlate with growth rates under the different feeding regimes.

2. Reduction of the daily feed ration in the grow-out culture of Nile tilapia on the farm:
Using alternate-day feeding, farmers have realized a cost savings for feed of more than 50% (Bolivar et al., 2007). Philippine tilapia farmers, in an effort to find ways to reduce the amount of feeds without compromising yield and profit, will use a feeding rate based on fish biomass with the daily ration divided by half (T1) as compared with a control fed at the full amount based on fish biomass (T2). This comparison of treatments T1 and T2 will be conducted on-farm, using two ponds at each farm (7-9 separate farms). Fingerlings (size #22), fertilizers and feeds will be provided to farmers for the 120-day growout study. Growth, FCR, IGF's/growth correlations, and total production will be assessed as above.

*Null Hypotheses: (*1) Sub-satiation feeding at 50% level will not affect growth performance/production efficiency parameters of tilapia relative to continuous full daily feeding, and (2) Circulating IGF-I or hepatic IGF mRNA levels will not correlate with growth rates under the different feeding regimes.

3. Effect of Formulated Feeds with Varying Fish Meal and Alternative By-products on Growth of Nile Tilapia Cultured in Earthen Ponds:
3A. Tilapia diets will be formulated to reduce fishmeal usage in combination with the use of alternative ingredients from food by-products (e.g., rice bran, cocoa, various flowers, soya, nut oil, milling waste, brewer's wastage, poultry by-product meal, feather meal, cassava, ipal-ipal leaf, etc.) commonly found in the Philippines. The primary by-products to be considered as alternatives to fish meal will include protein by-product meals from the poultry and swine industry, and yeast by-products from the brewing industry that are located in Central Luzon. All grower diets will be formulated to meet the nutrient requirement of tilapia (NRC, 1993; Li et al., 2006) using least-cost linear programming.

3B. Develop feed manufacturing specifications for the formulas developed in 3.1 at NCSU. The formulas will be processed using pellet-extrusion technology to develop specifications that are economical for Filipino farmers. Each treatment will be evaluated to determine the optimal processing parameters and cost based on production rate and energy usage. The treatment pellets will be evaluated for pellet quality (PDI) and nutrient leaching over 6 hr. The pelleted treatment feeds will be fed to fish in test tanks at NCSU to determine palatability, feeding behavior, growth performance, IGF/growth corollaries, FCR over 1-month trials in sex-reversed juvenile Nile tilapia.

3C. We will then test the most promising treatment feeds developed and tested in 3.1 and 3.2 in earthen ponds at FAC, CLSU. This experiment will use twelve 500 m^2 earthen ponds at the FAC, CLSU. It will have 4 treatments and 3 replicates. Ponds will be stocked with sex-reversed Nile tilapia (*Oreochromis niloticus*) fingerlings at a density of 4 fish m^{-2}. Treatments will be as follows:

1) Control Diet containing 15% fishmeal.
2) Treatment 1 Diet with 25% reduction in fishmeal
3) Treatment 2 Diet with 50% reduction in fishmeal
4) Treatment 3 Diet with 100% reduction in fishmeal

At the termination of the growth trial, fish will be sampled for proximate analysis to determine if body composition is similar among fish on the treatment diet versus those on the control diet containing fishmeal. A marginal, cost-benefit analysis will also assess the potential for fishmeal-free or –reduced diets in improving cost savings for pond production of tilapia.

Null Hypothesis: Decreasing feed formulation costs by replacing fishmeal with low-cost by-products will have no effect on cost per kg of tilapia produced.

<u>Schedule</u>

April 2007 – September 2007	Study 3A
October 2007 – March 2008	Studies 1 and 3B
April 2008 – September 2008	Study 3B
October 2008 – March 2009	Studies 2 and 3C
April 2009 – September 2009	Write extension brochures/manual and final reports

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ALTERNATIVE FEEDING STRATEGIES TO IMPROVE MILKFISH PRODUCTION EFFICIENCY IN THE PHILIPPINES

Sustainable Feed Technology/Experiment/07SFT03NC

Collaborating Institutions & Lead Investigators

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Objectives

The overall goal of this investigation is to develop feeding strategies as potential cost containment options for milkfish (*Chanos chanos*) production in both brackish water (BW) and marine environments. The specific objectives are as follows:

1. Compare alternate day feeding with traditional daily feeding on milkfish production characteristics (growth, feed conversion) in tanks in brackish and marine water.
2. Assess feeding schedules of longer feed restriction and refeeding increments on compensatory "catchup" growth responses and milkfish production characteristics in marine and BW environments.
3. Test daily, alternate day, and the best-determined feed restriction/refeeding period of Objective 2 on milkfish growout in BW ponds and cages in marine waters.
4. Evaluate the potential for insulin-like growth factor-I (IGF-I) as a biomarker of growth status in milkfish.

Significance

Milkfish has been cultured in the Philippines for almost a century (Bagarinao, 1999). It is the largest finfish aquaculture industry in the Philippines with a harvest area of 280,000 ha and total production of 300,000 metric tons annually (BAS, 2006). There is also a growing export market for milkfish at one million Kg annually and a value of almost $3M, with the U.S. topping imports among all countries. As part of the Philippine government's food security and poverty alleviation programs, expansion of milkfish culture is a high priority (Rosario, 2006) both to wean fishers off capture fisheries by providing them with fish farming as an alternative/ supplemental form of living, and to increase income of farmers whose poverty levels are disproportionately high (Rivera *et al*., 2006). Milkfish production is increasing at 5% annually, with much of the production moving away from traditional culture in BW ponds to fish cages in marine waters, and with a 14% increase in marine cage culture seen in 2005 alone (BAS, 2006). Cage culture of milkfish in marine environments is done at high densities and with significantly greater inputs of artificial feeds. The use of this practice, however, has led to wastage of artificial feeds and excessive nutrient loading in receiving waters (Sumagaysay-Chavoso *et al*., 2004), exacerbating problems with pollution and possibly periodic fish kills that have been observed in areas of marine milkfish culture clusters.

Sixty percent of milkfish farming expenses are attributable to feed costs (Rosario, 2006, Rivera *et al*., 2006). Research supported by Aquaculture CRSP has demonstrated that feeding tilapia only on alternate days saves almost 50% on feed costs with little effect on growth, survival or market yield (Bolivar *et al*., 2006). Like the tilapia, milkfish is a low trophic species and plankton eater. Our previous research and that of others also demonstrates that longer-term feed restrictions followed by a refeeding period can elicit a compensatory growth response characterized by a 2-3 fold increase in specific growth and feed conversion in both tanks (Skalski *et al*., 2005; Picha *et al*., 2006) and ponds (Turano *et al*., 2007; see Ali *et al*., 2003 for review). Cyclic feeding was also shown to reduce total phosphorous load in ponds by 40% (Turano 2006). Although general feed methods and nutritional requirements to raise marketable sized milkfish have been established, largely through research at SEAFDEC AQD (Juario *et al*., 1984; Alava and Lim, 1988; Sumagaysay, 1995; Sumagaysay, 1998), studies have not assessed whether periodic episodes of feed restriction/refeeding can induce compensatory growth and improve production efficiency of this cultivar. In the proposed investigations we will examine whether feeding schedules that incorporate feed restriction/refeeding increments might limit overall feed input without significantly impacting fish yield. We propose to conduct studies in both marine water cage culture and in BW ponds, as each may require different feed inputs due to natural productivity (BW ponds) or lack thereof (marine fish cages). Also, BW ponds represent 70% of total production and marine cage culture is on the rise. The sustainability of these two culture methods and farmer's profitability could be enhanced through methods that both reduce feed input and limit nutrient loading in receiving waters through excessive feed wastage.

Insulin-like growth factor-I (IGF-I) is the primary hormonal mediator of growth in fish. We and others have found that it may serve as a robust biomarker of growth rates in various fish species that include tilapia, hybrid striped bass, flounder and salmonids (Beckman *et al*., 2004; Picha *et al*., 2006; Vera Cruz *et al*., 2006; Luckenbach *et al*., 2007). Its levels change rapidly prior to and in parallel with specific growth responses (Picha *et al*., 2006). Considering traditional methods of conducting lengthy and costly growth trials, a biomarker of growth such as IGF-I could prove valuable for researchers for the rapid assessment of the numerous, and often confounding number of environmental and nutritional parameters that regulate growth. We will undertake studies under controlled growout environments to evaluate the utility of either circulating IGF-I or expression of its gene as an instantaneous indicator of growth in milkfish.

<u>Quantified Anticipated Benefits</u>

- Direct evidence of the effectiveness of pulsed versus daily feeding could provide milkfish farmers practical methods to contain feed costs and increase profitability while limiting the environmental impacts of their activities in BW ponds and cages in the marine environment.
- The investigations will take advantage of the strong research and extension mission of SEAFDEC AQD to promote environmentally friendly culture of milkfish in the Philippines and Southeast Asia. This will include development of an extension manual, detailing alternative feed protocols for milkfish growout.
- A bioassay for rapid evaluation of growth rates will be developed for the largest finfish culture industry in the Philippines. Establishment of an IGF-I based assay as a rapid growth rate indicator should expedite assessment of factors that optimize management and growout of milkfish. SEAFDEC is the Southeast Asian hub for aquaculture technology development and is well positioned to adopt the biomarker assay for use in milkfish, and for future testing in other marine and coastal fisheries.
- The investigations will contribute to the research training of at least 4 undergraduate students.

<u>Research Design & Activity Plan</u>

Location: Experiments will initially be conducted in tanks to test the efficacy of different feed schedules on milkfish production parameters in well-controlled environments at Tigbauan Main Station of SEAFDEC AQD.

These initial growth trials in both BW and seawater are important for obtaining precise measures of feed consumption and conversion ratios, while simultaneously assessing compensatory growth responses and the establishment of IGF-I as a useful biomarker of growth. Following these initial studies, which will aid in defining the feeding regimens that might best work for milkfish, we will conduct field production trials in BW ponds and in fish cages in marine waters at the Dumangas Brackishwater Station and Igang Marine Station of SEAFDEC AQD, respectively.

Methods:
1. Evaluate the efficacy of feed schedules that incorporate different increments of feed restriction and refeeding on milkfish production characteristics and the compensatory ("catch-up") growth response in marine and brackish water tanks.

For these experiments, 5 treatment groups will be used and each treatment will have 3 replicates. The studies for BW (15 ppt) and seawater culture will be done separately over a culture period of 60 days in 5-ton concrete tanks. The treatments are as follows:

T1: control, normal daily feeding using current commercial farming feeding schedules for marine cage culture where feeding rate begins at 10% bw and gradually decreases with increased fish biomass. We cannot replicate feeding in brackish water ponds because of the primary productivity found there.
T2: alternate day feeding at normal levels
T3: 4 weeks restricted feeding, 4 weeks normal feeding
T4: 4 weeks restricted feeding, 2 weeks normal feeding, 2 weeks alternate day feeding
T5: 2 weeks restricted feeding, 2 weeks normal feeding, 2 weeks restricted, 2 weeks normal

Our previous studies in hybrid striped bass indicate that 4 weeks of feed restriction followed by refeeding can elicit a robust compensatory growth (CG) response that is accompanied by higher specific growth rates and feed conversion relative to animals fed continuously (Skalski et al. 2005; Picha et al. 2006; unpublished data). The degree of the CG response also appears depend on a significant but not overly excessive catabolic state achieved with restricted feeding. Therefore, both a 2- and 4-week feed restriction has been incorporated into the investigations.
Milkfish fingerlings produced at the SEAFDEC AQD fish hatchery (20 g body weight) will be stocked at a density of 25 fish/m3 and maintained in a recirculating system at salinity of 20 ppt. for BW and 33 ppt for SW. Fish will be fed SEAFDEC AQD formulated feeds 4 times daily (Santiago *et al*., 1983; Alava and Lim, 1988; Sumagaysay, 1998; Sumagaysay and Borlongan, 1995). Body weight measurements of at least 20 fish will be taken every 2 weeks to monitor growth and adjust for feed ration. During sampling, 3-5 fish from each treatment will be sacrificed for measurements of IGF-I mRNA expression in the liver and plasma IGF-I levels. IGF-I mRNA expression will be measured by real time PCR (Picha et al. 2006; Ayson et al., 2007) using fluorescent probes designed against milkfish IGF-I cDNA for which we already have a partial sequence (our unpublished data). Plasma levels of IGF-I will be measured by radioimmunoassay at NCSU. We will subsequently establish a time-resolved fluoroimmunoassay, at SEAFDEC AQD, for plasma IGF-I measures. Specific growth rates (SGR), food conversion ratio (FCR), and hepatosomatic index (HSI) will be ascertained and differences among treatments analyzed by our previously published methods (Picha et al. 2006, Turano et al. 2007). Significant corollaries between growth and IGF will be assessed by regression analysis.

Null hypotheses: (1) The mixed feeding schedule will not affect the growth performance of milkfish and other production parameters relative to normal daily feeding; (2) Plasma levels of IGF-I and hepatic IGF-I mRNA levels will not correlate with growth rates under the different feeding schedules.

2. Evaluate the efficacy of alternate feed schedules on full grow-out trials in brackish water ponds and fish cages in the marine environment.
Milkfish brackish water pond growout: We will test the efficacy of feed schedules tested in concrete tanks under commercial growout in brackish water. Three treatment groups will be used and each will be done in triplicate ponds over a 4-month growout. The treatments will be as follows:

T1: control, natural food ("lablab" algae) for the first 45 days and supplemental daily feeding at 3% body weight
T2: natural food for the first 45 days and alternate daily feeding of artificial diet at 3% body weight
T3: natural food for the first 45 days and the feed strategy most effective among the T3-T5 groups established in tank trials with fish being fed at 3% body weight during the normal feeding periods (the cyclic feed schedule will be repeated twice)
For this experiment, 9 units of small-sized ponds (500 m^2) in the Dumangas Brackishwater Station of SEAFDEC AQD will be used. Ponds will be properly prepared for the growth of lablab, the

natural food of milkfish in ponds. Organic (chicken manure, 0.5 ton/ha.) and inorganic fertilizers (urea at 25 kg/ha and 16-20-0 at 50 kg/ha) will be applied to initiate growth of natural food ("lablab" algae). Milkfish fingerlings will be stocked at densities of 10,000/ha in 500 m^2 pond. Water quality (salinity, temperature, dissolved oxygen level and transparency) will be monitored 3 times weekly. Standard water quality parameters, including ammonia, nitrate, nitrite and phosphorous levels will be measured weekly. At least 50 fish will be sampled every 2 weeks to monitor growth and to adjust the feed ration. During sampling, 3-5 fish from each treatment and pond will be sacrificed for measurements of liver IGF-I mRNA expression and plasma IGF-I levels. Total yield, SGR, FCR, and HSI will be calculated and analyzed along with IGF-I/growth corollaries as previously described (Picha et al. 2006; Turano et al. 2007).

3. Milkfish growout in fish cages in marine waters: Experiments will also be conducted in fish cages in marine waters to test the efficacy of the feeding schedules tested in study #1. Three treatment groups will be used and each treatment will have 3 replicates during a 4-month growout. The treatments will be as follows:

T1: control, normal daily feeding under milkfish feeding schedules used by farmers, which is a feeding rate of 10% body weight with gradual decreases to 3% bw for larger fish.
T2: alternate day feeding at normal rates
T3: the feed strategy most effective among the T3-T5 groups from tank trials in study #1 only the cycle will be twice repeated

For this experiment, 9 units of fish cages measuring 4 x 4 x 2m will be set up in Igang Marine Station of SEAFDEC AQD. Milkfish fingerlings (15-20 g body weight) will be stocked at a density of 15-20 fish/m3. Fish will be fed SEAFDEC AQD formulated feeds 3 times daily. Water quality parameters including dissolved including oxygen level, chlorophyll a and phosphorous will be monitored in relation to establish potential relationships with feed input and fish biomass. Body weight measurements of at least 50 fish will be taken every 2 weeks to monitor growth and adjust the feed ration. Sampling and analyses will be conducted as outlined above.
Null hypotheses: (1) The mixed feeding schedule will not affect the growth performance of milkfish and other production parameters relative to normal daily feeding; (2) Plasma levels of IGF-I and hepatic IGF-I mRNA levels will not correlate with growth rates under the different feeding schedules.

Schedule

May – July 2007	Procurement of materials and supplies
August 2007 – February 2008	Tank experiments
March – August 2008	Trials in brackish water ponds
September 2008 – February 2009	Trials in fish cages in SW
March – May 2009	Extension manual writing
June – September 2009	Final report writing

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UTILIZATION OF LOCAL FEED INGREDIENTS FOR TILAPIA AND PACU PRODUCTION

Sustainable Feed Technology/Activity/07SFT04UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)	Kevin Fitzsimmons
Mon Repos Aquaculture Center Department of Fisheries (Guyana)	Tejnarine S. Geer Kamila Singh

Objectives

1. Discuss semi-intensive culture of Tilapia and Pacu, with a focus on nutritional requirements.
2. Identify and examine local ingredients suitable for use in formulation of Tilapia and Pacu feed.
3. Examine simple feed formulation techniques using identified ingredients.

Significance

Tilapia and pacu aquaculture has been practiced for several years in Guyana. However, in the last few years the growers have organized into a farmer's cooperative, the National Aquaculture Association of Guyana. With the assistance of the Ministry of Agriculture and the USAID/GTIS Program, the farmers have begun to develop local markets and have been able to participate in the 7th International Symposium on Tilapia in Aquaculture. The Mon Repos Aquaculture station has worked with the farmers to provide technical assistance and training (Geer 2004, 2006).

In Guyana, aquaculture is now starting to emerge as an industry. Existing feed and oil manufacturers are willing to engage in fish feed manufacture. However, they require assistance in suitable ingredient identification, feed formulation, manufacture of low cost feed, producing with existing machinery, and training in aquatic feed production techniques. Development of economic diets utilizing locally available ingredients will be a central focus point. The pacu and tilapia producers have several advantages over regional producers that they feel will allow them to compete effectively in local markets as well as in North American markets. There is an abundance of high quality water with ideal temperatures, a stable government and economic system, a well-educated, native English-speaking workforce and a local population that has already proven the acceptance of tilapia and other fish as a staple in the diet.

Sustainability has been identified as a priority for the aquaculture producers. Utilization of local feed ingredients, minimizing the use of fish meal, and increasing the efficiency of the feed that is input to the system are immediate targets.

Quantified Anticipated Benefits

The fish farmer's group anticipates that several dozen of their management and staff will attend the work activities. They believe that with additional training and instruction they could operate in a more efficient manner and improve both profitability and environmental sustainability. Specifically, they would like to receive advice and guidance on locally available ingredients that could be substituted into practical feeds and how to process them into usable pellets utilizing existing feedmill infrastructure. This should be done in a cooperative manner with both the farmers, who have concerns of cost and quality and the feedmill operators who have concerns over expenses and operational characteristics. Of course both groups are also interested in shelf life of the feed, percent of fines in the products and other details. One important parameter to measure will be the change in sales volume of locally produced aquatic feeds and the amount of local ingredients incorporated. Another will be the number of participants in the workshops and the number of farmers who incorporate changes arising from the workshop. We anticipate that 20 to 40 people will attend.

Another area of interest is operational characteristics of feeding and especially the Best Management Practices associated with feeding practices. The farmers report that they would like to incorporate BMP's both for their own farm improvement, but also as a step towards certification of their product as being more sustainable and capable of receiving a higher price on international markets.

Activity Plan

Locations: Mon Repos Aquaculture Station and Maharaja Oil Mill with contributions from:

- Mon Repos Aquaculture Station (facilities, equipment, personnel)
- Maharaja Oil Mill (facilities, equipment, personnel)
- NAAG (transportation, accommodation)
- USAID/GTIS (Training materials and curriculum preparation)

Participants: Tilapia and Pacu farmers, Feed producers, Fish Processors, Government Representatives, Kevin Fitzsimmons (University of Arizona: Presenter), Dennis McIntosh (Delaware State University: Presenter)

Presentations:

- Aquaculture Theory: pond design, water quality, Tilapia biology, feeding, fingerling production, grow-out, health and disease, etc.
- Practical Aquaculture: plankton examination, male and female Tilapia ID, Tilapia dissection, transportation, acclimatization, pond fertilization, etc.
- Feed Ingredient ID: examining local feed materials, and determining suitable ones for feed ingredients
- Feed Formulation Theory: calculation of protein and other nutrient content, balancing of ingredients, etc.
- Practical Feed Formulations: preparation and mixing of ingredients, mixing, pelletizing and drying of feed.

Schedule

July 2007	Confirm locations and make announcements
August 2007	Make travel arrangements and prepared presentations
August 2007	Confirm attendees; finalize preparations
October 2007	Workshop Duration: 3 – 4 days

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Geer, T. 2004. Report on the activities of the Mon Repos Freshwater Aquaculture Demonstration Farm and Training Centre - July 2001 to December 2003. Guyana Ministry of Agriculture, Department of Fisheries.

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LOCAL INGREDIENTS SUBSTITUTING FOR FISHMEAL IN TILAPIA AND PACU DIETS IN GUYANA

Sustainable Feed Technology/Experiment/07SFT05UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)	Kevin Fitzsimmons
Delaware State University (USA)	Dennis McIntosh
Mon Repos Aquaculture Center Department of Fisheries (Guyana)	Kamila Singh

Objectives

Test experimental diets on Tilapia and Pacu reared in cages and ponds.

1. Trial one will replace 20%, 50% and 80% of fish meal with locally derived poultry by-product meal (Null hypothesis – growth rates and Feed Conversion Ratios will not be significantly different).
2. Trial two will replace 20%, 50% and 80% of fish meal with locally derived shrimp by-product meal (Null hypothesis – growth rates and Feed Conversion Ratios will not be significantly different).
3. Trial three will incorporate both poultry by-product and shrimp meal in a combination suggested by the results of the earlier trial.

Significance

Tilapia (*Oreochromis niloticus*) and pacu (*Colossoma macropomum*) aquaculture has been developing in recent years in Guyana. In 2006 the growers organized into a farmer's cooperative, the National Aquaculture Association of Guyana. With the assistance of the Ministry of Agriculture and the USAID/GTIS Program, the farmers have begun to develop local markets and would like to develop an export trade. The Mon Repos Aquaculture station has worked with the farmers to provide technical assistance and training (Geer 2004, 2006).

Feed costs represent 60-75% of the variable production costs in Guyana. The currently available, locally produced, commercial feed has ingredient costs of approximately $0.36/kg ($360/mt) and contains almost 25% fish meal. Fish meal prices are rising and the sustainability of fishmeal harvest is questionable. Use of alternatives ingredients, especially locally available by-product meals would be preferable for economic as well as environmental reasons.

One of our goals is to work collaboratively with the commercial feed mill and the fish farmers to develop economically attractive feeds that contribute to sustainability, which has been identified as a priority for the aquaculture producers. Utilization of local feed ingredients, minimizing the use of fish meal, and increasing the efficiency of the feed that is input to the system are immediate targets.

Reducing costs, using local ingredients, and reducing fishmeal usage are all laudable goals, but will be meaningless if the feed quality or efficiency are reduced. Fish effluents leaving a cage or being discharged from a pond will be increased if food is not converted to fish biomass. Our goal is to improve the diets nutritional efficiency, by being more nutrient dense, that is matching the nutritional need with the dietary ingredients (Lim and Webster 2006). Poultry by-product meal has been shown to be an effective protein replacement for up to 50% of fish meal in tilapia diets (Viola and Zohar 1984) and Williams et al. (1998) demonstrated that shrimp meal could also be used as a suitable ingredient to replace fishmeal.

Quantified Anticipated Benefits

The National Aquaculture Association of Guyana believes that development of practical diets incorporating local ingredients to replace fishmeal would improve profitability and sustainability.

Currently over a dozen farms utilize a commercial diet containing 25% fish meal. The present project expects to work with two commercial feedmills to develop practical diets that will reduce or even eliminate fishmeal usage.

The second important goal is to gain certification of the tilapia producers for following Best Management Practices. Producers recognize that this would provide a marketing advantage in local and international markets.

Research Design & Activity Plan

Locations: Mon Repos Aquaculture Station, Maharaja Oil Mill and private fish farm (Von Better Aquaculture or similar)

Methods: Substitution of fish meal with poultry by-product and shrimp meals on *O. niloticus* and *C. macropomum* juveniles. Growth of juveniles will be evaluated using practical iso-caloric and iso-nitrogenous diets containing 25% fish meal and 0% poultry by-product meal (Control diet), 20% FM and 5% PBM, 12.5% FM and 12.5% PBM and 5% FM and 20% PBM. The second trial will utilize the same substitution plan with shrimp by-product meal.

1. Project Coordinator: Ms. Singh will be the coordinator, supervising staff and students who will conduct the feeding trials and then preparing the analytical analyses and project report.

2. Culture Period: 100 days.

3. Stocking rate: 40 fish per cage with four cages fed each of the four diets at the Mon Repos Aquaculture station in one pond stocked with tilapia (16 cages). A second pond will be stocked in a similar fashion with pacu. At the private farm facility, three ponds of tilapia will be fed each diet following the standard farm operating procedures. Tilapia and pacu of approximately 40 grams will be stocked initially. The fish will be fed with experimental diets two times a day. Fish will receive rations containing 5% of their body weight initially. Daily rations will be adjusted down to 4% and 3% daily as the fish grow.

4. Water quality: Dissolved oxygen, temperature, and pH will be monitored on a daily basis. Ammonia, nitrite and nitrate will be determined on a weekly basis.

5. Sampling Schedule: Tilapia and Pacu juveniles will be sampled at the beginning of the experiment and every 21 days. Total length and weight will be measured to the nearest 0.1 cm and g. Mortality will be recorded daily.

6. Statistical Methods and Hypothesis: Hypothesis: Substitution with poultry meal in the diets produces unequal growth and survival. To test the null hypothesis, growth data will be compared using a one-way ANOVA for each species. Survival will be compared using a Chi-square test.

7. Economic analyses: A simple enterprise budget will be developed to compare a theoretical farm operation using the control diet compared to the best performing of the experimental diet.

8. Results will be shared initially through regular meetings of the Mon Repos staff and the NAAG. Formal results will be presented at a workshop to be held at Mon Repos Aquaculture Station in July 2009. Formal results will be prepared for publication in professional journals and presented at a professional aquaculture conference (WAS or ISTA8). An electronic presentation will be provided through a project website or at the A&F CRSP website.

Schedule

November 2007	Prepare diets and stock ponds and cages
March 2008	Harvest ponds and cages and
June 2008	Restock ponds and cages
September 2008	Harvest ponds and cages
October 2008	Adjust diet formulations
November 2008	Prepare diets and stock ponds and cages
March 2009	Harvest and begin analyses of results
July 2009	Workshop Duration: 3 – 4 days

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Geer, T. 2004. Report on the activities of the Mon Repos Freshwater Aquaculture Demonstration Farm and Training Centre - July 2001 to December 2003. Guyana Ministry of Agriculture, Department of Fisheries.

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DEVELOPMENT OF LOCALLY AVAILABLE FEED RESOURCE BASE IN TANZANIA

Sustainable Feed Technology /Study/07SFT06PU

Collaborating Institutions & Lead Investigators

University of Arkansas at Pine Bluff (USA)	Carole R. Engle Rebecca Lochmann
Sokoine University of Agriculture (Tanzania)	Sebastian Chenyambuga
Ministry of Natural Resources & Tourism - Aquaculture Development Division (Tanzania)	Kajitanus O. Osewe

Objectives

The project is developed to promote and enhance the adoption of mono-sex tilapia culture by creating an enabling environment for aquaculture production system that is capable of producing quality all-male tilapia fingerlings at a stable and sustainable manner, produce fish feeds at low cost, produce marketable fish to meet consumer demand, and develop human capital for both research and extension to support all-male tilapia culture in Tanzania. This vision will be achieved through the following implementation plans or investigations and activities.

1. Identify local plant species that can be a protein sources for small-scale tilapia culture by determining the suitability of *Moringa oleifera*, *Leucaena leucocephala, Jatropha curcas*, and *Sesbania sesban* as protein sources for fish feed.
2. Determine the nutritional content of these plant species.
3. Evaluate the suitability and economic profitability of different feed forms that include mash, crumbs and pellets.
4. Develop feeding strategies to minimize adverse effects on water quality.
5. Support a graduate student at the MSC level in fish nutrition.

Significance

Feeds account for more than 50% of production costs in semi and intensive aquaculture operations. Protein feed resources constitutes a major cost component of fish feed. This is compounded in some instances when suitable protein feed sources is scarce. Furthermore, the situation is made worse for resource-poor fish farmers in developing countries where most of the proteins feed sources are also food for humans, and hence their use in fish feed conflicts with food security interests. Due to these factors, most farmers tend to use nutritionally low quality feeds such as maize bran, wheat bran, rice polishing, and brewery residues, which supply energy and not protein. This results in poor fish yields due to imbalanced energy-protein ratio. In view of the above considerations, the need to identify appropriate alternatives or complementary sources of protein is imperative. It is highly desirable that the selected protein sources do not conflict with human food security interests, as is the case with fishmeal. Moreover, recent BSE disease outbreaks have aroused doubts on the wisdom of feeding animal derived-protein feed resources to non-carnivorous species. Moreover, there is an environmental push to move fishmeal from fish diets. The selected plant species have long been established in Tanzania under the pastures and forages development program. The plants are used by small-scale dairy farms as protein supplement for lactating cows. Plants have therefore become a preferred alternative to animal protein sources. In the past decades several attempts have been made to study the suitability of plant derived protein sources for aquaculture. Many of these studies were centered on materials like soybean (Kotaro 1999), cottonseed meal (El-Sayed 1990) and corn gluten (Wu et al 1996). However, most of these plant protein sources require high external energy subsidies and environmental and soil conditions that restrict the scope for increasing their production under the small-scale aquaculture. In addition, there is an increasing direct human demand for nutrients derived from these sources; hence they are not expected to contribute greatly as alternative sources of protein for aquaculture development in Tanzania. Therefore, there is a need to study plant protein sources for small-scale aquaculture, which are able to grow under adverse soil and climatic conditions and require low external energy subsidies.

Quantified Anticipated Benefits

The study will provide knowledge on plant species with high potential as protein sources for aquaculture production system in Tanzania. Taking into consideration the fact that in most developing countries animal feed is very expensive relative to fish prices, the use of plant-based protein will lower feed costs. This will enable the formulation of low cost diets with a relatively high quality, which can be easily prepared by fish farmers themselves. This is anticipated to have a positive impact on the improvement of fish nutrition and aquaculture productivity in Tanzania.

Research Design & Activity Plan

The following experiments will be conducted at SUA and will form a base for a MSc student thesis in Fish Nutrition. A student will be selected among interested graduate students enrolled in the Aquaculture Sciences Program in the Department of Animal Science and Production (DASP). Since there is a close collaboration between DASP and the Faculty of Aquatic Sciences and Technology of the University of Dar-es-Salaam, recruitment will be extended to include women students in the faculty in case there is no qualified female student at SUA. In additional, the Kingolwira National Fish Center occasionally accepts students from both Universities to undertake research and other special projects. The student should have finished all coursework and have an average GPA of more than 3.0. The preference will be given to female students and those who are currently employed as aquaculture extension agents by the Ministry of Natural Resources and Tourism or aquaculture development agents by NGOs operating in Tanzania. The selection process will involve oral interview and subject matter presentation as related to this experiment. Drs. Chenyambuga and Lochmann will guide the student on the fish nutrition experiments and Drs. Engle and Kaliba will guide the student on economic analysis. The project team in collaboration with the graduate committee members shall guide the selected student to produce research results that will fulfill some of the project's objectives and meet the requirements for the Department of Animal Sciences and Production. The project will provide financial support for materials and all research related to activities, publication of research results and attendance to one international or regional meeting. Whenever possible, qualified host country PI's in Ghana and Kenya will be include in the graduate committee of a student or will be involved as external examiners. This is to promote collaboration and spillover effect.

Determination of nutritional content of the selected plant species: This will be achieved through laboratory analysis of plant material including leaves and seeds to determine proximate composition, mineral content, amino acid profile, and anti-nutritional factors.

Determination of feeding value of the selected plant species: This will be achieved through feeding trials at SUA using a completely randomized design. Cylindrical concrete tanks with a surface area of $7m^2$ and height of 1m will be used for the trials. Water will be refreshed daily at a rate of 15-20% in order to ensure adequate amounts of dissolved oxygen. Fingerlings with an average weight of 10g will be stocked at a rate of two fingerlings/m^2. Diets will consist of either plant material or soybean or both as protein sources and rice polishings/maize bran as energy sources. Minerals will also be included in the diets. Fish will be fed twice a day at 0900hrs and 1600hrs at a rate of 10% of tank biomass and the amount of feed will be adjusted according to monthly sample weights. Trials for each plant material will run for a period of 90 days i.e., within the active growing phase of Nile tilapia, to avoid metabolic complications brought about by onset of maturity in Nile tilapia. Five dietary treatments, each replicated three times, will be imposed for each leaf plant species. The method of preparing the form of feeds to be used in the study will be standardized and the feed will be formulated to meet diet requirement for Tilapia. Feed formulation will be done in consultation with Dr. Lochmann, who has a lot of experience in fish nutrition.

Diet	% Digestible Crude Protein from Plant	% DCP from soybean[1]
1	0	100
2	25	75
3	50	50
4	75	25
5	100	0
6	Maize bran	

[1]This will be based on other studies. For examples see Mlay et al (2006), Murro et al (2003) and Ndemanisho et al (1998).

Body weight, fork length, width, survival rate and feed conversion ratio will be recorded at day 30, 60 and 90. Prior to weighing, fish will be starved for at least 15hrs. Water quality parameters such as temperature, dissolved oxygen, pH and Nitrogenous compounds (NH4-N, NO2-N and NO3-N) concentration will be measured daily before feeding. At the end of the experiment five fish will used for proximate analysis to determine the effects of feed forms on dry matter, crude protein, crude fat and gross energy concentration. Data will be analyzed using the SAS GLM procedure. The GLM procedure uses the method of least squares to fit general linear models. Among the statistical methods available in PROC GLM are analysis of variance, analysis of covariance, and partial correlation. The GLM procedure performs analysis of variance from a wide variety of experimental designs. In analysis of variance, a continuous response variable, known as a dependent variable, is measured under experimental conditions identified by classification variables, known as independent variables. The variation in the response is assumed to be due to effects in the classification, with random error accounting for the remaining variation. The GLM procedure is designed to compare several groups of treatments and to handle both balanced and unbalanced data (i.e., data with equal and unequal numbers of observations for every combination of the classification factors).

Evaluation nutritional suitability and economic profitability of different feed forms: This study shall be carried out using diets that will have been shown to support good performance in terms of growth of fish and water quality. For each selected diet, three forms of fishmeal from leaves will be evaluated (i.e., mash, crumbs, and pellets). The experimental procedure and design and data analysis shall be as described in section b above. Water quality parameters such as temperature, dissolved oxygen, pH, temperature, alkalinity, Nitrogenous compounds (NH4-N, NO2-N and NO3-N), Phosphates and secchi disk visibility will be measured twice a week before feeding. Physical environment measurements will involve solar radiation (light), rainfall, wind speed, and air temperature. Water samples will be taken for qualitative and quantitative analysis of plankton once per month. The data collected will be analyzed using the SAS GLM procedure explained above to determine difference in growth performance.

Economic profitability of each feeding strategy and associated economic gain of adopting the most superior feeding practices by small-scale farmers will be determined through partial budgeting. This is to compare costs and returns of alternative feeding strategy and evaluate the economic effect of adjustments in fish farming by introducing plant feed resources. Partial budget analysis is a simple but effective technique for assessing the profitability of new technology for new enterprises. Results of partial analysis provide the foundation for comparing the relative profitability of alternative treatments and associated risk due to changing product or input prices. The study will follow the procedure on partial budget analysis as published in the manual by the International Wheat and Maize Improvement Center (CIMMYT 1988). Boughton et al (1990) reviews many examples of the economic analysis of on-farm trials, with special attention to the CIMMYT method and acknowledges the importance of CIMMYT's contribution, particularly in regard to the use of field prices for inputs and outputs. The procedures presented in the CIMMYT manual were developed to guide agricultural scientists as they develop recommendations for farmers from on-station research-based data. The guidelines are also used to help design trials in the earlier stages of research. Based on these guidelines the economic analysis will proceed through the following stages:

1. Preparation of a partial budget in order to estimate the net benefits arising from alternative treatments. For each treatment, including the control, the following three values must be calculated: value of production to the farmer, including by-products; value of all inputs used across treatments, and calculation of the net benefit.
2. Identification of inferior treatments or dominated treatment, i.e., those treatments that involve higher cost but does not generate higher benefits through dominance analysis.
3. Marginal analysis will be used to assess relative profitability among alternative treatments. Marginal analysis is used to identify the treatment, which gives the highest net return and a marginal rate of return greater than the minimum considered acceptable to farmers. This minimum rate of return reflects the opportunity cost of resources to the farmer, i.e., the return that can be earned in alternative activities. Minimum returns and sensitivity analyses shall be carried out to assess the robustness of the candidate recommendation given variation in yields and input or output prices.

Impact Indicators:

- A Nile tilapia feeding protocol using plant materials developed
- Graduation of a M.SC student in Fish Nutrition with knowledge on development and formulation of plant-based fish feeds and using experimental data for simple economic analysis.
- Training of 50 farmers on the formulation and feeding of plant-based protein supplement to fish.

Schedule

Time	Activities
August – December 2007	- Selection of students - Building infrastructure - Purchase of materials - Laboratory analysis of plant materials
January – March 2008	- Feeding value experiment
April 2008	Data analysis for the feeding value experiment
May – August 2008	- Conducting feeding trial
September – December 2008	- Data analysis of the feeding trial
January – March	
January – March 2009	- Thesis writing
April – May 2009	- Training of farmers - Research results dissemination
June – September 2009	- Thesis defense - Writing of project final report

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TOPIC AREA
INDIGENOUS SPECIES DEVELOPMENT

❖

DEVELOPMENT OF SNOOK (*CENTROPOMUS SPP*) SEED PRODUCTION TECHNOLOGY FOR APPLICATION IN AQUACULTURE AND RESTOCKING OF OVER-FISHED POPULATIONS

Indigenous Species Development/Experiment/07IND01UA

Collaborating Institutions & Lead Investigators

Texas Tech University (USA) — Reynaldo Patiño

Universidad Juárez Autónoma de Tabasco (Mexico) — Wilfrido M. Contreras Sánchez, Ulises Hernández Vidal, Carlos A. Alvarez-González, Arlette Hernández Franyutti, Alejandro Mcdonal Vera

Objectives

1. Develop techniques for the production of good quality snook eggs:
   - Hypothesis 1: Injections of GnRHa will produce good quality eggs.
   - Hypothesis 2: Implants of GnRHa will produce good quality eggs.
2. Develop techniques for the production of snook seed:
   - Hypothesis 3: Initial stocking rates of snook larvae will not affect their growth or ability to wean from live zooplankton to prepared diets.
3. Conduct an international workshop on snook biology and culture.

Significance

Most fisheries stocks in southeastern Mexico have been reduced or depleted due to an increase in the demand for fish products and in the number of fishermen, and to anthropogenic alterations of aquatic habitats. Despite this situation, artisanal fisheries based on the capture of wild fish continue to be the primary source of fish for the food market in the gulf coast and southeastern regions of Mexico. The declining populations of native fishes are raising concern among fishermen. A large number of these fishermen live in isolated, poor communities; and the loss of their primary income is contributing to the migration of rural inhabitants into major Mexican cities and to the United States in search of alternative employment (Vidal, 2002).

Aquaculture is a viable option to capture fisheries. The development of aquaculture techniques for native species not only would help reduce fishing pressures on wild stocks, but it also would provide a source of fingerlings for the development and implementation of plans to restore depleted stocks. In turn, a sustainable fishery for important native species coupled with the development of their aquaculture potential could help solve the problem of rural emigration by providing new and better opportunities for employment or income.

Snooks are the most valuable fishery species in southeastern Mexico. At least three species are found in the Gulf of Mexico: common snook (Centropomus *undecimalis*), Mexican snook (*C. poeyi*), and fat snook (*C. parallelus*). The natural range of common snook extends from North Carolina to Brazil (Muller et al., 2001), making the status of wild snook populations of international concern in the Americas. Snooks enjoy one of the highest prices in the Mexican national market (about 6.2 D/kg; beach price), which creates an incentive for fishermen to engage in its commercial and artisanal fisheries (a single fish captured represents an important income for a family). The annual catch for common snook, which is perhaps the most economically important of the snooks, is approximately 5,000 tons for Mexico and 900 tons for the state of Tabasco (Anonymous, 2002). However, there is an overall national trend for diminishing catch volumes, a situation that has led to concerns for the health of the snook fisheries in Mexico and to calls for improved management practices. Common snook were also once abundant off the Texas coast and supported commercial and recreational fisheries, but the Texas common snook population is now characterized by low abundance and erratic recruitment (Pope et al., in press). The Rio Grande/Bravo is a shared resource of the US and México, and our research suggests that this river provides critical nursery and feeding habitat for the Texas-northeastern Mexico snook populations (C. Huber et al., unpublished data).

Due to its geographic and hydrological features, the state of Tabasco is one of the most promising areas in Mexico for the development of aquaculture. However, the implementation of aquacultural activities strongly depends on the availability of fingerlings (seed); which until now has primarily focused on exotic tilapias. Recently, some fishermen in the region successfully raised snooks in freshwater earthen-ponds utilizing wild-caught seed. This practice was subsequently banned by the Secretariat of Natural Resources (SEMARNAT) and by state agencies because it was rapidly spreading, raising concerns about the potential impact on wild stocks. Despite its short life, this experience justified the development of snook seed production technologies because it showed (1) an interest in and a demand for snook seed for the purpose of aquaculture, (2) the feasibility of growing snook juveniles in freshwater ponds; and (3) a commitment by state and federal management agencies to manage the snook fisheries and aquaculture industry on the basis of ecologically sound principles.

Reproductive Biology and Spawning of Snooks: Knowledge of the reproductive biology of snooks is limited. Most information available concerns the common snook. Snooks are considered to be protandric hermaphrodites (Taylor et al., 2000). Spawning has been successfully induced by hormone treatment in wild-caught common snook broodstock (Neidig et al., 2000) and in captive fat snook broodstock (Alvarez-Lajonchere et al.., 2002). However, currently available technologies need to be validated on site at the target locations to confirm their general applicability.

Status of Larviculture and Seed Production for Snooks: Snook larvae are small (approximately 1.5 mm) and require small prey organisms for their survival and growth. The rearing of larval fish is the most critical stage in the production cycle for many species, and the primary obstacle is that of an adequate food supply (Léger et al., 1986; Abi-Ayad and Kestemont, 1994). A diet which is ready and available and of high nutritional quality, and that is accepted and digested by larval fish must be used (Kim et al., 1996). Feeding protocols using microalgae, rotifers and *Artemia nauplii* are currently used for many species. At first feeding, adequate live food density and particle size are also essential for larval fish survival during this critical stage (García-Ortega and Lazo, 2004). Little information is currently available on the dietary requirements of captive common snook larvae. However, fat snook larvae have been successfully reared on a combination of rotifers and *Artemia nauplii* (Alvarez-Lajonchere et al., 2002), or solely on *Artemia nauplii* (Alves et al., 2006).

The next step towards the establishment of an aquaculture for new species is the successful weaning and growth of juveniles for seed production. Seed production of fat snook at an experimental scale in Brazil and of the related species, Barramundi (*Lates calcarifer*) at a commercial scale has been successful achieved (Alvarez-Lajonchere et al.., 2002). The laboratory of aquaculture at UJAT also has conducted pilot tests that demonstrate the feasibility of rearing common snook juveniles on artificial diets (Silvercup trout pellets; J.M. Vidal et al., unpublished data).

Rationale for Research and Workshop Activities

Induced ovulation (egg production). Common snook broodstock has been difficult to maintain in captivity and thus the few available hatchery-spawning programs have mostly relied on wild-caught fish. Wild-caught broodstock are either immediately processed upon capture to obtain gametes for in vitro fertilization, or they are brought to the hatchery where they are promptly injected with hormones to induce spawning (Anonymous, 2001). Alvarez-Lajonchere and colleagues (2002) developed spawning techniques for fat snook that are based on chronic and acute hormone treatment methods. However, spawning methods used elsewhere (or for other species) need to be tested on site to determine the precise conditions that result in larvae of good quality. Information regarding the timing, hormone type and dose and water quality conditions is particularly needed. There is the possibility of differences between species in their "manageability."

Larval rearing. Although fat snook larvae have been successfully reared and weaned in other places (Alvarez-Lajonchere et al., 2002; Alves et al., 2006), the applicability of these techniques to new production sites needs to be validated. Also, larviculture techniques for common snook have not been established. In addition, information is needed concerning the weaning of common snook larvae to grow-out diets.

International workshop. In spring 2004, TTU and UJAT organized a successful international workshop on the biology and culture of snooks. Presenters included leading world experts on these topics. Since then, we have generated new information on the use of artificial diets for juvenile common snook (J.M. Vidal et al., unpublished data), status of common snook in Texas (Pope et al., in press), and freshwater habitat for juvenile common snook (C. Huber et al., unpublished data); and important information on the larval rearing of snooks has been developed by others (e.g., Alves et al., 2006). Moreover, we anticipate the additional generation of information from the research effort proposed here and from the ongoing efforts of other investigators. Thus, as part of this proposed project we intend to organize the Second International Workshop on Snook Biology and Culture for the purpose of collecting and widely disseminating the latest information on snooks.

Our proposed project is based on the premises that (1) there is an increasing food-market demand for snooks in Mexico; (2) there is an ecological need for alternatives to the capture of wild snooks; (3) the development of aquaculture for valuable native species, such as snooks, is preferable for the region in the context of both market acceptability and ecological compatibility; and (4) the sustainable development of aquaculture coupled with the sustainable management of capture fisheries will provide more and better employment and income opportunities for artisanal as well as commercial fishermen.

Quantified Anticipated Results

Culture techniques for snooks will benefit farmers and fishermen by promoting aquacultural diversification while avoiding the potentially harmful ecological effects associated with the use of exotic species. If the proposed protocols provide positive results, farm trials will be set up in small farms located at "Ejido Cucuyulapa" and "Ejido Morelos" in Tabasco to assess the transferability of laboratory results into the field. The Laboratory of Aquaculture at UJAT disseminates information to an average of three farmers/fishermen a day; we thus anticipate that 800-900 people will receive information on snook culture in this manner.

One UJAT PI (U. Hernandez) will travel to TTU for training and information exchange. One graduate student and one undergraduate student will be closely involved with the project, and many more undergraduates (up to 10 students) are also anticipated to participate during the course of the study. Although the students have not yet been selected, the Laboratory of Aquaculture at UJAT has an established record of training female students and we anticipate a balanced ratio between males and females. UJAT students will also interact with at least one TTU graduate student via exchange information on laboratory techniques and data analysis. At least two referred publications in scientific journals are expected from the results of this proposed study.

The Second International Workshop on Snook Biology and Culture will be conducted in Villahermosa, Tabasco and is expected to attract over 100 participants including farmers, fishermen, researchers, students, technicians, and federal and state agency officials from Mexico, Central and South America. The information generated at the workshop will be used to develop an informational brochure on the biology and culture of snooks. This brochure will be made available as hard copy and on the internet (on UJAT and TTU websites).

This project will strengthen ties between TTU and UJAT and foster continued collaborative research projects between the two institutions. Also, because snooks are geographically widespread and are the target of fishery interests in several countries of the Americas, the technologies developed by this project are expected to benefit aquaculturists and fishermen in coastal Mexico as well as other countries. We have met with hatchery managers from Tabasco, Veracruz, Campeche and Chiapas and they have expressed their interest in collaborations. In Central America there is strong interest in the culture of snooks and we have already communicated with researchers from Zamorano University (Honduras), San Carlos University (Guatemala) and the University of Costa Rica to express our intentions to exchange information and technology transfer. We also plan to share our experiences with hatchery managers and workers in those countries and potentially establish experimental trials in their farms by implementing thesis projects with undergraduate students. Our target population will be small-scale farmers and poor fishermen. In Texas, the Texas Parks and Wildlife Department has been interested in development of snook spawning techniques, and we have made contact with researchers from The University of Texas who share similar interests. The information generated from this project also will be made available to any interested US agency or individual.

Research Design & Activity Plan

Objective 1: Develop techniques for the production of good quality snook eggs. The experimental protocols described below are generally based on work with snooks of Alvarez-Lajonchere et al. (2001, 2002), Neidig et al. (2000), Alves et al. (2006), and Vidal et al. (unpublished data from our laboratory).

Experiment/Hypothesis 1. Injections of GnRHa will produce good quality eggs.
Location of work: Recently built snook research facility in the coastal county of Centla, in the fishermen community of Jalapita, Tabasco. This facility is a rural, low budget project initiated by UJAT researchers and fishermen from Cooperativa Pesquera San Ramón (UJAT-CPSR).

Test species: common snook and fat snook.

General methods: Wild broodstock will be collected during the natural spawning season (June-September) and initially maintained in the laboratory in 25 m^3 holding tanks. Female maturity will be confirmed by observing the location of the oocyte´s germinal vesicle and by measuring egg diameter before they are transferred to holding tanks (males are not expected to be a problem). At least 36 adults per species will be collected (24 females and 12 males). Spawning will be induced within 12 h of collection by injecting different doses of GnRHa (Argent Labs). However, if necessary, injections may be performed immediately upon capture to increase treatment effectiveness (H. Grier, personal communication). Eggs and sperm will be obtained by stripping. Artificial insemination will be conducted using the Alvarez-Lajonchère & Hernandez Molejón (2001) method. The buoyant eggs will be stocked at 500-1000 eggs/l in 120-L cylindroconical incubation tanks.

Treatments and replications: Females will be injected with vehicle (saline), 75 or 150 µg’kg GnRHa/fish. All males will be injected with 50 μg/kg GnRHa/fish. There will be at least four replicates per treatment, each with two females and one male per spawning tank (diameter, 2m). Females will be monitored for maturation and ovulation. Sperm activation will be evaluated in males under microscopic examination. The incidence of ovulated females per replicate will be monitored. Lack of ovulation in any of the two females in a replicate will be considered a "negative-result" and will be recorded. A "positive-result" replication will be considered as one where eggs are produced regardless of their number or quality.

Water management: Marine water (35 ppt) will be used in all tanks. Initially, 50% of the water will be exchanged twice weekly; after treatment, 80% water will be replaced every other day. Water pH, dissolved oxygen, and temperature will be monitored daily.

Sampling schedule: Samples for macroscopic inspection of the developing oocytes will be collected from each female before treatment. After injection, oocytes will be sampled for monitoring ovulation every 24 hours.

Endpoints and statistical analysis: Negative- and positive-result replicates for each treatment will be assigned a score of 0 or 1, respectively, and analyzed by non-parametric Kruskal-Wallis ANOVA followed by separation of mean treatment scores using Dunn's post-test. For positive-result replications, egg size and number will be analyzed by 1-way nested ANOVA (tank nested into treatment) and mean treatment values will be analyzed using Tukey's HSD. Egg quality will be assessed by determining rates of fertilization, hatching, and survival to first feeding; these results will be compared among treatments by Chi square test using contingency tables.

<u>Experiment/Hypothesis 2</u>. Implants of GnRHa will produce good quality eggs.
Except for the treatment, all other procedures and analyses will be as described in the preceding experiment. Also, the outcomes of the two treatment strategies (injection and implant) will be conducted by gross comparison.

Treatments: Females will be implanted with pelleted vehicle (no GnRHa), 100 μg GnRHa pellet/fish, or 200 μg GnRHa/fish pellet. All males will be implanted with 100 μg GnRHa/male pellets

Objective 2. Develop techniques for the production of snook seed.
<u>Experiment/Hypothesis 3</u>. Initial stocking rates of snook larvae will not affect their growth or ability to wean from live zooplankton to prepared diets

Location of work: Induced ovulation, rural snook facility; larval rearing, UJAT's Aquaculture Laboratory.
Tests species: given the limited time and resources available, we will be able to conduct this experiment only with one of the species (common or fat snook). The selection will be made depending upon best performance in experiments 1 and 2. However, preference will be given to common snook.

General methods: Eggs will be obtained from an induced spawning using the technique (injection or implant) and GnRHa concentration that yielded optimal results (see preceding experiments). After removing unfertilized eggs, embryos will be placed in a graduated cylinder to estimate the total number (by volume displacement). Embryos will be transported to the laboratory and incubated for 12 h at 24 ºC and 35 ppt salinity in an aerated in 120-L cylindroconical incubation tank with flow-through water circulation. The number of yolk-sac larvae will be estimated according to survival at hatching. Yolk-sac larvae will be transferred from the incubator to a closed recirculating system composed of fifteen 70-L fiberglass tanks at desired treatment densities (see below; densities will be estimated by volumetric method).
The water in the recirculating system will be filtered with a Jacuzzi sand filter (5-20 μm) and a biological filter, and passed through an UV lamp filter. Water will be recirculated in the tanks

with a continuous exchange. The airflow to each tank will be approx. 0.5 cm^3/s and will be increased gradually until the end of the experiment. A photoperiod of 13:11 (L:D) will be used. The following water quality variables will be measured daily: temperature and DO using a YSI Model 58; salinity using a refractometer; and nutrient level using colorimetric techniques.

The general feeding schedule for the lowest stocking density treatment (20 larvae/L) will be as follows: L-type rotifers *Brachionus plicatilis* (50-300 µm lorica length) during days 2-14 posthatch. Prior to presentation to larvae, rotifers will be intensively fed with *Nannochloris* spp. and enriched with Selco (Artemia Systems) for 4 h. Rotifer stocking density will be 7-15/mL. Snook larvae will be also fed newly hatched *Artemia* spp nauplii during days 11-30 at a density of 2-10/mL. The Artemia will be also enriched with Selco for 12 h prior to presentation to larvae. Artemia juveniles will be also added directly to the tanks during days 23-30. The larvae will be fed five times daily. Larvae will be gradually weaned to a formulated diet (grounded Silvercup trout pellets) during days 30-45 and grown for an additional 30 days until an age of 75 days posthatch. Fish will receive daily rations containing 10% of their wet weight. Rations will be estimated using a spread-sheet constructed with previous growth data. The same feeding schedule will be used for the other experimental densities except that the quantity of food will be increased proportionally to match fish density.

Treatments: Initial stocking densities for snook larvae will be 20, 40, 60, 80, and 100 larvae/L. Each treatment will be conducted in triplicate.

Water management: Water for rearing larvae will be recirculated using bio-filters, UV filtration, mechanical filtration, 25% of the volume will be exchanged daily.

Sampling Schedule: Random samples of approximately 10 larvae per tank (30 per density) will be taken every five days. They will be first anaesthetized with MS-222 and fixed with 4% formaldehyde. Larvae will be measured from a digitized image (Sigma Scan Pro V. 4.0) captured with an optical microscope Zeiss (8X). Depending on the developmental stage of the larvae, notochordal (NL) or standard length (SL) will be recorded to the nearest 0.001 mm. Fish will be weighed with an analytical balance to the nearest 0.0001 g. Survival will be evaluated by making daily counts of dead larvae collected from the bottom of the tanks by siphoning.

Endpoints and statistical analysis: Growth and length data will be compared using a two-way nested ANOVA (treatment x time; tanks nested into treatment) followed by Tukey's HSD. Survival to weaning (yolk-sac to 45 days) and to seed stage (yolk-sac to 75 days) and will be compared using Chi-square tests.

Objective 3. Conduct an international workshop on snook biology and culture
The Second International Workshop on Snook Biology and Culture will be organized at UJAT on the basis of our successful experience with the first (2004) workshop. It will be widely advertised by using listserv, newsletter, and web site outlets that target fisheries and aquaculture audiences as well as by personal contacts. The 3-day workshop will consist of lectures, panel discussions, demonstrations and field trips and will be led by experts in the field from North, Central and South America and the Caribbean. We expect an attendance of 100 to 150 participants. In addition to a small fund request from CRSP, costs for the workshop will be partly covered by registration fees. An informational brochure (fact sheet) will be developed based on workshop discussions for hardcopy and internet distribution.

<u>Schedule</u>

Experiment 1

April – May 2007	broodstock collection
June – September 2007	spawning induction
September – December 2007	egg incubation and larval rearing

January – June 2008	data analysis and preparation of reports and publications

Experiment 2

April – May 2007	broodstock collection
June – September 2007	spawning induction
September – December 2007	egg incubation and larval rearing
January – June 2008	data analysis and preparation of reports and publications

Experiment 3

January – June 2008	algae, rotifer and copepod culture
June – September 2008	collection of larvae
September – December 2008	egg incubation and larval rearing
January – September 2009	data analysis and preparation of reports and publications

Workshop

Spring 2008 (tentative date)	a fact sheet will be developed within 6 months

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Incorporation of the Native Cichlids Tenhuayaca, *Petenia splendida*, and Castarrica, *Cichlasoma urophthalmus* into Sustainable Aquaculture in Central America: Improvement of Seedstock Quality and Substitution of Fish Meal Use in Diets

Indigenous Species Development/Experiment/07IND02UA

Collaborating Institutions & Lead Investigators

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Objectives

1. Improve seedstock quality based on a genetic improvement program for *P. splendida* and *C. urophthalmus*.
2. Determine the effect of the substitution of fish meal for poultry meal on growth, survival, apparent digestibility, and chemical composition of *P. splendida* and *C. urophtzahalmus* juveniles.

Significance

In Southeastern Mexico, a growing number of fish producers are requesting the development of alternative culture techniques that involve native species. The culture of native species of fish is important from an economic and a conservation standpoint at a time when the local and foreign demand has imposed great pressures on their natural populations. In Tabasco and Chiapas, several species of native cichlids have been proposed for aquacultural purposes. Among those species, *Petenia splendida* and *C. urophthalmus* are of special interest because of their local demand and cultural value (Mendoza, 1988). Few studies have evaluated the basic biological parameters for incorporation of new species into aquaculture processes and information is needed in order to participate in the recovery of their populations and launch a different kind of aquaculture; one based on sustainability of native species and food security.

During the last decade the laboratory of aquaculture at UJAT has conducted research supported by the PDA/CRSP to develop techniques that allow the culture of native species such as the tropical gar (*A. tropicus*) and the cichlids *P. splendida*, *C. synspillum* and *C. urophthalmus*. These studies have valuable information regarding reproductive performance in captivity, effective masculinization and larval culture. We have found that among the cichlids; *P. splendida* and *C. urophthalmus* are very good candidates for aquaculture purposes. Commercially, these species are the most desirable, they have the highest values among cichlids in the local market and the demand is very high.

To achieve fish culture to a commercial scale, basic and applied research is needed to complete the life cycle of the species in captivity. However, in Mexico the impulse for this activity has been given for introduced species such as tilapia and trout, because the technology is already known (Rojas y Mendoza, 2000). The potential for *C. urophthalmus* was highlighted by Martínez in 1987. In our laboratory, we have started studying the acceptance of commercial feeds and growth performance of this species. So far we have compared growth performance of fry fed *Artemia* nauplii against fry fed commercial feed. Finding a significant difference in the growth of fish that are fed nauplii during the first 30 days of feeding; However, fry accepted inert food and had relatively good growth. Real (2003) successfully masculinized *C. urophthalmus* obtaining between 95 to 100% males feeding larvae with *Artemia* nauplii enriched with Methyltestosterone.

For *P. splendida* several studies that include ecology, habitat, feeding content and distribution have been conducted (Reséndez and Salvadores, 1983; Ferreira et al., 1988; Caro et al., 1994; Domínguez and Rodiles, 1998). Regarding aquaculture, the studies include, García (2003), Jiménez (2004), and Chan (2004), who determine the maximum interval of temperature resistance (30.68 and 31.95 Celsius), and the thermal preferendum (28-30 ºC) for the best metabolism and growth of juveniles. Contreras (2003) and Vidal-López (2004), obtained 95 to 100% males feeding larvae with trout commercial diet and Artemia nauplii enriched with MT using a concentration of 30 mg/L for 45 days. Martínez (2004) described the morphological development of larvae, while Uscanga (2006) determined the protein requirements for masculinized and non-masculinized juveniles using semi-purified diets. All this information is promising; but most fry produced in captivity has been produced utilizing wild broodstock that is domesticated and introduced into spawning systems. The fry production numbers are encouraging and very valuable, but genetic improvement has not been approached, and quality of the fry can be significantly improved. It is well known that genetic improvement in aquaculture can increase phenotypic characteristics of fish from one generation to the other, but this process is significantly higher during the first rounds of selection. Therefore, we expect that a minimum of 10% increase in yield can be achieved in the first generation (F1). Furthermore, homogeneity in fry size and quality will be obtained in a short period of time.

Quantified Anticipated Results

The establishment of good quality broodstock treatments, their distribution to local hatcheries, and the implementation of intensive masculinization programs are basic steps for sustainable aquaculture. These actions can significantly improve the production of high quality fingerlings and have a favorable impact on more than 5,000 subsistence farmers and medium-scale producers. Well-supported aquacultural practices can help secure good quality food products in the near future, especially in the proposed site of study where a large portion of the population is composed of extremely poor campesinos.

On the other hand, commercial feeds currently used for native species are formulations designed for tilapia, catfish or trout. In the case of *Petenia*, feeds formulated for trout are currently used for grow-out. This artificial feeds contain a high quantity of proteins and lipids, increasing significantly the cost for large-scale production. Use of locally derived, lower cost ingredients would contribute to sustainability, efficiency, and profitability. We expect to several dozen to 100 farmers will adopt these fish and diets before the end of the project.

Research Design & Activity Plan

Experiment 1. Genetic improvement of *Petenia splendida*, and Castarrica, *Cichlasoma urophthalmus* using total length and condition factor

Site: Reproduction and progeny testing will be conducted at the Mariano Matamoros Hatchery, Teapa, Tabasco, Mexico. Groups of broodstock will also be kept at UJAT as a backup.

Activities. The first group of broodstock for fry production and growth comparisons will be obtained from the current broodstock used in the laboratory of Aquaculture. One hundred and fifty females and 50 males will be selected from the broodstock pools. Female selection will be based on the best total length measurements, and male selection will be performed using individuals with the best condition factor. Each selected broodstock group will be placed in a 2.5m-diameter tank. Each tank will contain 30 females and 10 males/tank, fish will be stocked at a sex ratio of 3:1 female:male in five spawning tanks. Fry to be used for grow-out trials will be collected from spawning tanks and stocked in grow-out ponds at a density of 14 fish/m^2. To eliminate age variability, fish stocked in a single pond will have a maximum difference of three days of age.

Growth Performance. Fry will be collected from the spawning ponds pooled and 28,000 fish will be stocked in a 2,000 m^2 grow-out earthen pond. This procedure will be repeated five times to assure five groups of fry. Fish will be fed five times a day providing a daily ration of 5% of the estimated biomass. Feeding charts will be constructed from samplings performed every two weeks. Sampling will be conducted after 60 days of grow-out and statistical comparisons will be made.

Line Selection. After three months of growth; fish will be collected and measured. All fish will be divided in three groups using weight as the selection variable: 1) Fry which are 33% above the median value, 2) fry which are 33% around the median value, and 3) fry which are 33% below the median value. Group 1 will be reserved for follow-up studies, and group 2 will be used for line selection. All fish in group 3 will be discarded. From group 2 (of each replicate), 10,000 fish will be stocked in 1,000 m^2 earthen ponds (14 fish per m^2) and grown-out for 3 months. At the end of the grow-out phase, 3,000 females and 1,000 males with the highest length will be selected and placed (separated by sex) in 1,000 m^2 earthen ponds. After three months of growth, fish will be selected to produce the F1 generation. Males will be selected based on highest condition factor, and females will be selected based on highest length. One thousand females and 330 males will be selected from each replicate.

Laboratory and Pond Facility. The State government officials responsible for the Mariano Matamoros Hatchery have set aside four concrete ponds (200 m^2) for spawning, four grow-out concrete ponds (1,000 m^2), and four grow-out earthen ponds (2,000 m^2) for UJAT's line selection investigation. If needed, more ponds can be used at the hatchery.

Universidad Juárez Autónoma de Tabasco (DACB). Ten spawning tanks (2.5 m diameter), 50 net cages (1 m^3) for fry grow-out, three grow-out ponds (200 m^2).

Universidad Juárez Autónoma de Tabasco (DACA). Two grow-out ponds (50 m^2), 20 net cages (1 m^3) for grow-out.

Test Species: Castarrica (*Cichlasoma urophthalmus)* and tenhuayaca (*Petenia splendida*).

Water Management: 10% water exchange per day.

Sampling: The following variables will be sampled: Initial weight and length, survival, final weight and final length. The following values will be estimated: Growth rate, condition factor and food conversion factor.

Statistical Methods and Hypotheses. Hypothesis: Broodstock selected from the F1 generation will produce fry with better quality and quantity than those produce from the original stock. To determine differences in growth performance and fry production, ANOVA will be performed using weight, length, and mean number of fry per female as the dependent variables. This methodology will be repeated in 2008 to obtain the F2 generation, using the F1 group as the starting stock.

Disposition of broodstocks and improved strains: All of the breeders and progeny will be held at UJAT. No releases of fry or fingerlings are anticipated within the scope of this investigation.

Experiment 2: Effects of the substitution of fish meal with pork meal on *P. splendida* and *C. urophthalmus* juveniles.

Site. Experiment will be conduced at the Laboratory of Aquaculture at UJAT, Tabasco, México.

Methods: Growth of *P. splendida* and *C. urophthalmus* juveniles will be evaluated using practical isocaloric and isoproteic diets containing 0, 25, 50, 75 and 100% fish meal protein that will be substituted with pork meal.

Laboratory and Pond Facility at UJAT. 5 rearing recirculating systems (33, 100-l tanks each); 1 rearing recirculating systems (21, 70-l tanks each).

Culture Period.90 days or until juveniles reaches 200% of the initial size.

Stocking rate. 50 fish/tank.

Tests Species. *Petenia splendida* and *Cichlasoma urophthalmus.*

Water management. Water for rearing will be recirculated using bio-filters, 25% of the volume will be exchanged twice a week.

Sampling Schedule. Tenhuayaca and Castarrica juveniles will be sampled at the beginning of the experiment and every 15 days. Total length and weight will be measured to the nearest 0.001 mm or g. Mortality will be recorded daily. Feces recollection for the measure of apparent digestibility recorded daily. Some samples of fish will be taken at the beginning and the end of the experiment to determine chemical composition in whole fish (AOAC, 1990). Experimental treatments will be as follows:

1) Tenhuayaca juveniles fed with a diet containing 0% fish meal and 100% pork meal.
2) Tenhuayaca juveniles fed with a diet containing 25% fish meal and 75% pork meal.
3) Tenhuayaca juveniles fed with a diet containing 50% fish meal and 50% pork meal.
4) Tenhuayaca juveniles fed with a diet containing 75% fish meal and 25% pork meal.
5) Tenhuayaca juveniles fed with a diet containing 100% fish meal and 0% pork meal.
6) Tenhuayaca juveniles fed with a commercial diet (Control).

A similar experimental design will be executed using tenhuayaca juveniles.
Each treatment will consist of three replicates. First feeding juveniles will be fed with experimental diets four times a day. Fish will receive rations containing 10% of their weight. Daily rations will be estimated using a spread-sheet constructed with previous growth data.

Statistical Methods and Hypothesis. Hypothesis: Substitution with pork meal in the diets produces unequal growth, survival, apparent digestibility and chemical composition of *P. splendida* and *C. urophthalmus* juveniles. To test the null hypothesis growth data will be compared using a one-way ANOVA for each species. Survival will be compared using a Chi-square test.

Economic analyses: A simple enterprise budget will be developed to compare a theoretical farm operation using the control diet compared to the best performing of the experimental diet.

Experiment 3: Effects of the substitution of fish meal with poultry meal on *P. splendida* and *C. urophthalmus* juveniles.
Site. Experiment will be conduced at the Laboratory of Aquaculture at UJAT, Tabasco, México.

Methods. Growth of *P. splendida* and *C. urophthalmus* juveniles will be evaluated using practical isocaloric and isoproteic diets containing 0, 25, 50, 75 and 100% fish meal protein that will be substituted with pork meal.

Laboratory and Pond Facility at UJAT. 5 rearing recirculating systems (33, 100-l tanks each); 1 rearing recirculating systems (21, 70-l tanks each).

Culture Period. 90 days or until juveniles reaches 200% of the initial size.

Stocking rate. 50 fish/tank.

Tests Species. *Petenia splendida* and *Cichlasoma urophthalmus.*

Water management: Water for rearing will be recirculated using bio-filters, 25% of the volume will be exchanged twice a week.

Sampling Schedule: Tenhuayaca and Castarrica juveniles will be sampled at the beginning of the experiment and every 15 days. Total length and weight will be measured to the nearest 0.001 mm or g. Mortality will be recorded daily. Feces recollection for the measure of apparent digestibility recorded daily. Some samples of fish will be taken at the beginning and the end of the experiment to determine chemical composition in whole fish (AOAC, 1990). Experimental treatments will be as follows:

1) Tenhuayaca juveniles fed with a diet containing 0% fish meal and 100% poultry meal.
2) Tenhuayaca juveniles fed with a diet containing 25% fish meal and 75% poultry meal.
3) Tenhuayaca juveniles fed with a diet containing 50% fish meal and 50% poultry meal.
4) Tenhuayaca juveniles fed with a diet containing 75% fish meal and 25% poultry meal.
5) Tenhuayaca juveniles fed with a diet containing 100% fish meal and 0% poultry meal.
6) Tenhuayaca juveniles fed with a commercial diet (Control).

A similar experimental design will be executed using tenhuayaca juveniles.
Each treatment will consist of three replicates. First feeding juveniles will be fed with experimental diets four times a day. Fish will receive rations containing 10% of their weight. Daily rations will be estimated using a spread-sheet constructed with previous growth data.

Statistical Methods and Hypothesis: Hypothesis: Substitution with poultry meal in the diets produces unequal growth, survival, apparent digestibility and chemical composition of *P. splendida* and *C. urophthalmus* juveniles. To test the null hypothesis growth data will be compared using a one-way ANOVA for each species. Survival will be compared using a Chi-square test.

Economic analyses: A simple enterprise budget will be developed to compare a theoretical farm operation using the control diet compared to the best performing of the experimental diet.

Schedule

Experiment 1	
June – July 2007	System setup, broodstock collection
July – September 2007	Spawning
October 2007 – June 2008	Grow-out trials; report results through CETRA and annual reports
July – September 2008	Spawning
October 2008 – June 2009	Grow-out trials
January – June 2009	Data analysis and preparation of reports and publications
August 2009	Workshop for local fish farmers, academics, and governmental agencies
Experiment 2	
March – July 2008	Data collection
July 2008	Technical report (with posting at CETRA) Include in August
August 2009	Workshop
Experiment 3	
August – December 2008	Data collection
December 2008	Technical report
August 2009	Workshop

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SPAT COLLECTION, GROWTH RATES AND SURVIVAL OF THE NATIVE OYSTER SPECIES, *CRASSOSTREA CORTEZIENSIS* AT SANTA MARIA BAY, MEXICO

Indigenous Species Development/Experiment/07IND03UH

Collaborating Institutions & Lead Investigators

University of Hawaii at Hilo (USA)	Maria Haws
Louisiana State University (USA)	John Supan
Centro de Investigación de Alimentos y Desarrollo (CIAD) (Mexico)	Omar Calvario
Universidad Autónoma de Sinaloa, Mazatlán Campus (Mexico)	Guillermo Rodriguez Domínguez
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Objectives

This work will assess the biological feasibility and cost-effectiveness of oyster culture in a new location working in conjunction with the local women's groups. Currently only the introduced oyster (*Crassostrea gigas*) is cultured in Santa Maria Bay, but spat must be purchased from overseas hatcheries at relatively high cost and logistical difficulties. It would be desirable to cultivate the local oyster species (*Crassostrea corteziensis*) as is done further south in the state of Nayarit, if local spat fall is dependable and cost-effective.

1. Spat fall rates will be measured using three spat collection methods for the native oyster, *C. corteziensis*, in Santa Maria Bay.
2. Collected spat will then be grown out in suspended culture with data collected on growth rates, survival, shell-to-meat ratios and other culture parameters.
3. Economic data will also be collected in order to assess cost effectiveness of this culture model.
4. This work will also inform efforts to establish culture of the native oyster species in Nicaragua.

Significance

Most oyster culture in Sinaloa State and northwards uses almost entirely an imported species, *C. gigas* (Japanese oyster) and producers are highly dependent on *C. gigas* seed imported primarily from one large U.S.-based oyster hatchery. The goal is to work towards reducing dependency on exotic species and foreign suppliers. The native oyster, *C. corteziensis*, is very similar in taste and appearance to *C. virginica* (American Oyster) produced on the East Coast and Gulf Coast of the U.S., is in high demand by local consumers and commands a higher price. *C. corteziensis* is now cultivated largely in Nayarit state and southward, so introduction of the southern methods to Sinaloa offers potential to establish a new aquaculture industry and benefits to local takeholders, particularly women's fisher groups who have a long-standing interest in oyster cultivation.

Quantified Anticipated Benefits

Anticipated benefits include: lessening of dependence of an exotic species, increased utilization of a highly desirable local species; diversification alternative to shrimp culture; development and application of an appropriate culture methods; information for decision-making; and development of a new source of income for local fisher women's groups.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; State Committee for Aquatic Sanitation (CESASIN); and the Federation of Shrimp Cooperatives. Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of institutions directly or indirectly benefiting from the training: 7
- Number of individual participants in extension and technical training: estimated at 65
- Number of communities benefiting from extension services: 6
- Number of cooperatives or community groups benefiting from improved extension services: 4
- Students involved: 2
- Training modules produced: 1
- CRSP newsletter articles: 1 (in combination with results from Investigation 2)
- Peer-reviewed journal article: 1 (in combination with results from Investigation 2)

Research Design & Activity Plan

This work aims to transfer spat collection methods to Santa Maria Bay stakeholder groups. Natural spat fall of *C. corteziensis* appears to be locally abundant in Santa Maria Bay where it is widely gathered from mangrove roots by women and other local residents for consumption. Sales of oysters fished in this way are not common because the size is much smaller than when cultured. It is mainly consumed in households and often constitutes an emergency food when other food sources run short. Prior feasibility studies suggest that culture of this species is biologically feasible and that communities are highly interested, particularly fisher women's groups who need alternative livelihoods. Additionally, State support may be possible if the first trials succeed. Spat fall will be tested using three methods: direct collection from mangrove roots, wooden stakes and oyster shell hung on collection lines. The latter is the most commonly used method in Nayarit (Marcet et. al. 1992). Spat fall abundance, spat size, survival and cost-effectiveness of the three methods will be assessed. Spat collected will then be grown out in suspended culture on rafts, with data collected on important culture parameters such as survival, growth rate, meat-to-shell ratios, meat quality and costs of production. The oysters produced in this manner will be test-marketed at local shellfish kiosks and restaurants. The final report will include a cost-benefit analysis. Maria Haws (UHH) and John Supan (LSU) will advise on this work.

Security for the oysters being depurated is assured under the same system currently used by the community which has some oyster growing rafts in the same area and within eye-sight. Community members take turns watching the area at night although they report that few problems with security have occurred over the last 30 years that they have been growing oysters as access to the area can only occur through one road or through the mouth of the estuary, both of which are constantly monitored by the community.

Schedule

This work will begin in May, 2007 with spat fall experiments with a duration of six months, followed by grow-out trials to begin in October 2007 with a duration of 18 months. Completion of data analysis and reporting will then conclude in June 2009. Community members will be trained during each visit by researchers as each field visit includes a community meeting, short training event and active participation by community members in all field research activities. The UAS Masters degree student will assist with this work.

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OYSTER-RELAYING AND DEPURATION IN OPEN-WATER LOCATIONS

Indigenous Species Development/Experiment. 07IND04UH

Collaborating Institutions & Lead Investigators

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Objectives

1. Determine if selected open water areas near oyster grow-out grounds can be utilized for oyster depuration. Oysters grown in Boca Camichin estuary will be transported (relayed) to nearby, calm open water areas and held for depuration.
2. Determine whether oysters grown in demonstrably cleaner waters have added-value through consumer preference.

Significance

Water quality monitoring of two large estuary areas in Mexico (Santa Maria Bay and Boca Camichin) during 2006 under previous CRSP funding has shown that *E. coli* levels in Santa Maria Bay sites are sufficiently low that oysters and other bivalves that are safe for human consumption can be grown and harvested in this estuary throughout the year. In the case of Boca Camichin, *E. coli* levels are more variable over the annual cycle and in some cases come close to exceeding official standards or exceed them (Gaxiola et. al. 2006). Since 2006 was a drier than normal year, it is assumed that during rainier years when run-off increases, that *E. coli* levels will rise and prohibit safe consumption of oysters and other bivalves in Boca Camichin. This is unfortunate given the thriving small-scale oyster industry operating in Boca Camichin; hence the need to locate areas where harvested oysters can be transported ("relayed") and held in water where *E. coli* levels are acceptable for at least two weeks in order to assure consumer safety. Relaying is method commonly used in the Gulf Coast States such as Louisiana and Texas where oysters are harvested from less than optimal growout waters and transferred to areas with waters certified to have acceptable levels of pathogens such as E. coli, where oysters can be purged of suspected pathogens before sale. The two week depuration period is that recommended by the U.S. National Shellfish Sanitation Program (NSSP 2006). Additionally, given the growing awareness of shellfish-borne disease in Mexico, it is hypothesized that shellfish grown in monitored waters may have added value, particularly for the large tourist market that exists in towns such as Mazatlán and Puerto Vallarta. The depurated shellfish will be test marketed in nearby markets to assess this.

Quantified Anticipated Benefits

Given that oysters and other bivalves are widely grown, fished and consumed along the Pacific coast of Mexico, and exported to other regions of Mexico such as Guadalajara and Mexico City, shellfish sanitation is of regional and national issue importance.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; State Committee for Aquatic Sanitation (CESASIN); and the Federation of Shrimp Cooperatives. Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of new technologies developed: 2 (relaying and monitoring of depuration waters)
- Number of institutions directly or indirectly benefiting : 9
- Number of individual participants technical training: 20
- Number of communities benefiting: 26
- Students involved: 1 graduate, 2 undergraduate
- CSRP newsletter articles: 1 (in conjunction with Investigation 1)
- Peer-reviewed journal article: 1 (in combination with results from Investigation 1)

Research Design & Activity Plan

Open water areas near the shellfish growing grounds of Boca Camichin have been pre-selected. Oysters will be harvested from Boca Camichin and held in suspended culture in these areas for two weeks, as is recommended by the U.S. NSSP (NSSP 2006). The water in this area will be monitored on a bi-monthly based for a period of six months during which multiple batches of oysters will be held, tested for microbial levels and then test-marketed. Marketing will be conducted in nearby tourist areas with the oysters being branded with the Boca Camichin name and with testimony that they were harvested from monitored waters. Data will be collected on survival, meat weight loss or gain, costs of relaying oysters, and revenues obtained from "certified" oysters as compared to oysters taken directly from the grow out sites. Maria Haws (UHH) and John Supan (LSU) will advise on this work.

Security for the oysters being depurated is assured under the same system currently used by the community which has some oyster growing rafts in the same area and within eye-sight. Community members take turns watching the area at night although they report that few problems with security have occurred over the last 30 years that they have been growing oysters as access to the area can only occur through one road or through the mouth of the estuary, both of which are constantly monitored by the community.

Schedule

This work will begin in November 2007 and terminate in May 2008 to coincide with the periods of highest shellfish consumption (December and April). Community members will be trained during each visit by researchers as each field visit includes a community meeting, short training event and active participation by community members in all field research activities. The UAS Masters degree student will assist with this work.

Literature Cited

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TOPIC AREA
QUALITY SEEDSTOCK DEVELOPMENT

❖

BROODSTOCK SEED QUALITY AND FINGERLING PRODUCTION SYSTEMS REARING FOR NILE TILAPIA IN THE PHILIPPINES

Quality Seedstock Development/Experiment/07QSD01NC

Collaborating Institutions & Lead Investigators

North Carolina State University (USA)	Russell Borski
Central Luzon State University (Philippines)	Remedios Bolivar
GIFT (Genetically Improved Farmed Tilapia) Foundation International, Inc (Philippines)	Hernando Bolivar

Objectives

1. Examine the effect of broodstock age on seed production and fingerling growout performance of genetically improved tilapia strains.
2. Assess the size distribution, growth and survival of fry and fingerling growout performance of tilapia seed produced from artificial incubation units, hapas, and ponds.

Significance

Nile tilapia (*Oreochromis niloticus*) seed production in the Philippines is estimated at over 1.2 billion annually with demand expected to triple (Bureau of Fisheries and Aquatic Resources, BFAR; ADB 2005). There is a strong push and government effort to improve food security through increased hatchery development and enhanced technologies for consistent and quality seed production. The goal is to provide year-round, high-quality seed that can be widely distributed at reasonable costs to both increase income and reduce perceived risks to tilapia farmers. Fry and fingerlings either produced on farm or purchased represent a considerable investment to farmers, constituting around 15% of total cash costs for crop production, second only to feed (ADB 2005). Here, we aim to assess seed production efficiency as a function of broodstock age and different rearing production systems to evaluate better the hatchery and nursery practices intended to increase seed yield without a negative impact on fish growout performance.

It is well established that broodstock age and size, among various other factors, can affect seed quality and fecundity in farmed fish (Bromage, 1995; Green *et al*., 1997; Siddiqui and Al-Harbi, 1997). Previous studies show that small Nile tilapia broodstock produce more eggs than larger fish while larger broodstock produce more eggs per clutch than smaller ones. Work in other species, *T. zillii*, *T. tholloni*, and *S. melanotheron,* shows no direct correlation between fish size and fecundity (see El-Sayed, 2006). With regard to age, seed production per female was higher for 3-year-old compared with 1-year-old hybrid tilapia bred in concrete tanks. However, seed production as a function of broodstock biomass or density was best in 1-year-olds compared with 2-, 3-, and 4-year olds (Siddiqui and Al-Harbi, 1997). In red tilapia and *O. spilurus* 1-year old broodstock produce a greater number of seed than older age class fish (Ridha and Cruz, 1989; Smith et al. 1991). To our knowledge an evaluation of broodstock age on gamete quality (survival and growth) has never been done for the genetically improved GIFT (Genetically Improved Farmed Tilapia) strains of Nile tilapia. The GIFT strain is selectively bred for rapid growth and presently distributed by the GIFT Foundation International, Inc. It is widely used throughout the Philippines and Southeast Asia and its rapid expansion as a choice of tilapia farmers is supported by estimates that it can produce cost savings as much as 25% (Dey 2000). The GIFT Foundation is an independent organization that works with and refines genetically improved strains of Nile tilapia for food production (see Dunham et al., 2000). These fish enjoy widespread popularity because of their enhanced growth and feed utilization parameters, but additional refinements in hatchery technology would be beneficial; for this reason our studies will evaluate the effects of age of broodstock on the viability of progeny.

Additional studies will assess the efficacy of three artificial and natural seed rearing systems (artificial incubation units, hapas and ponds) on fingerling production characteristics (survival, sex inversion, and yield) and the subsequent growout performance of tilapia. These systems encompass the growout methods being used on either family farms or small/medium-scale hatcheries in the Philippines, and there is considerable interest by farmers and state and private hatcheries on the relative impact each might have on fingerling production and subsequent growout.

Quantified Anticipated Benefits

The broodfish age range that effectively produces the highest yield of seed will be known in tilapia strains selectively bred for superior growth characteristics and used throughout the Philippines and Southeast Asia. Most Nile tilapia farmed in the Philippines are derived from or hybridized with the GIFT strains (ADB 2005). For this reason, our results are likely to be of interest to many small-scale farmers who produce their own seed from purchased broodstock. Considering the latest estimate of over 2,000 tilapia hatcheries and nurseries of all size scales (ADB 2005), the impact of this work could be profound.

- The work will provide research training and educational experiences to four graduate and four undergraduate students at the Central Luzon State University in the Philippines.

- This investigation will provide qualitative information on the seed rearing system(s) most effective for producing fingerlings and harvest fish with more uniform size and yield.

- The research will foster collaborations between a non-profit organization and CLSU in extending better management technologies to farmers, hatchery and nursery operators in the Philippines.

Research Design & Activity Plan

Location: This investigation consists of a series of four studies, which will be carried out at the Central Luzon State University (CLSU) with an active collaboration between the CRSP group and the GIFT Foundation based at the CLSU research station.

Methods:
1. Effect of age of broodfish on the seed production of GIFT strain of Nile tilapia: Different ages of broodstock fish will be evaluated for seed production yield in one breeding cycle in ponds and hapas (fine mesh net enclosures) during the wet season (beginning in June). There will be four treatments in this study: 8-month old; 1-year old; 2-year old; and mixed-aged broodstock. Breeding will occur in 3 x 10 x 1 m hapas (within a 2500 m^2 pond) and in 200 m^2 earthen ponds, as these are the two most common breeding systems used by small-scale tilapia hatcheries/nurseries. The sex ratio will be maintained at 1:3 male:female and fish densities at around 0.3 Kg m^{-2}. Eggs from mouthbrooding females and fry will be collected after 14 days from initial stocking of hapas. Eggs will be incubated in artificial units to the swim-up fry stage and included with fry counts. For ponds, schools of fry will be collected with a dip net as they swim along the edge of the pond. Fry collection will be done four times daily for 10 consecutive days beginning 10 days after breeder stocking. Total fry numbers will be counted as an index of seed production by each treatment (broodstock age). Each treatment (age group) will be carried out in triplicate ponds and hapas. Differences in mean fry yield with broodstock age will be analyzed separately for ponds and hapas by ANOVA, and the least-significant difference test for

predetermined comparisons.

Null Hypothesis: There is no effect of broodstock age on fry yield in either hapas or ponds.

2. Effect of age of broodfish on the grow-out performance of Nile tilapia fingerlings: The objective of Study 2 is to compare the growth performance of Nile tilapia fingerlings produced from two age categories; young (8 month-old) and old (2 year-old) broodstock. Sex-reversed male fingerlings of size# 22 from the two age classes raised in hapas from the fry stage will be stocked in 500 m^2 ponds at a density of 4 fish m^{-2}, in triplicate. Growth (length and weight) will be monitored under controlled conditions at monthly intervals (subsampling 50 fish) over the four-month growout cycle. Fish will be fed daily using a progression of starter and growout feeds (Bolivar et al. 2006). Fish will be harvested after four months for total biomass, number, and survivorship determinations. Weights and lengths will also be obtained from 100 randomly selected fish. Differences in growth parameters among fish produced from different age class broodstock will be assessed by Student's t-test.

Null Hypothesis: There is no effect of broodstock age on growout performance variables in tilapia.

3. Size distribution, sex conversion rate, growth and survival of fry collected from artificial incubation units, hapas and ponds: Most farmers prefer monosex culture of male tilapia because of the substantial advantages of better growth and limited reproductive recruitment. In the Philippines, fry/fingerling production is largely done in hapas and ponds, although flow-through hapas are generally preferred over free distribution in ponds by a significant percentage of farmers. This is because fry collection is easier and there is less variability in seed size/age distribution in hapas than ponds (see also Green 2006). In either system fry production relies on the natural incubation of eggs by the female brood fish. Fry are then collected and sex-reversed by hormone-impregnated (SR) feed to produce fingerlings. Interest is currently growing in the use of artificial incubation units like conical-shaped plastic (soda bottles) containers in which eggs are incubated under constant motion to produce fry that are then subsequently sex-reversed. Our preliminary evidence indicates that size variability of fry/fingerlings might be reduced under artificial as compared with natural incubation. The production of more uniformly-sized/aged fry could prove advantageous for more effective and complete sex reversal and for the production of a more consistent, high-quality fingerling. To our knowledge no studies have evaluated fingerling production variables from fry produced in hapas, ponds and artificial incubation units.

Again in collaboration with the GIFT Foundation, the size distribution, sex conversion rate, growth and survival of fry collected from artificial incubation units, hapas and ponds will be evaluated in hapa enclosures. The hapas used in these studies will typify the commercial type and configuration, with dimensions of 2 x 5 x 1 m and will be installed in a 2500 m^2 pond at the GIFT Foundation area. Fry will be collected from hapas and ponds as above. Eggs will be collected from mouthbrooding females and placed in artificial incubation units (2-liter capacity hatching jars) and raised in flow-through freshwater to the swim-up fry stage (eight-nine days post yolk-sac absorption). Fry from each rearing condition will be stocked in hapas at a density of 600 m^{-2}. Each treatment (rearing unit type) will have 3 replicates. The rearing period for fry to reach fingerling stage will be 25 days. Fry will be fed with SR-treated fry mash. Data to be gathered will include growth, survival, size distribution and sex conversion level (determined after further rearing of the fingerlings to about three-five grams). Differences in these parameters among the different fry rearing systems will be analyzed by ANOVA followed by Fisher's least significant test for predetermined comparisons.

Null Hypothesis: There is no effect on fingerling survival, sex conversion, growth or size distribution between fry collected from incubation units, hapas and ponds.

4. Growout performance of Nile tilapia fingerlings produced from fry collected from artificial

incubation units, hapas and ponds: Fingerlings produced from fry collected in artificial incubation units, hapas and ponds as described above for Study 3 will be evaluated for growout performance. After 25 days of rearing in hapas, tilapia fingerlings will be collected and sorted according to the common size (size #22; with weight range of 0.20g–0.35g) that farmers typically raise or purchase from hatcheries, and stocked at a density of 4 per m^{-2} in 500 m^2 ponds. The treatments (fingerlings produced from fry collected in the three different rearing conditions) will be performed in triplicate ponds. Fish will be fed with commercial feeds that vary with the progression of culture as practiced by tilapia farmers (i.e., 1st month - Tilapia Fry Mash; 2nd month-Tilapia Starter Feeds; 3rd and 4th months-Tilapia Grower). Monthly sampling will be done to monitor growth of fish over a 120-day culture period. Mean weight, yield, and size distribution of the fish will be determined at harvest. Water quality parameters will be monitored and corrected as needed throughout all four of the proposed studies in this investigation, according to our previously published procedures (Bolivar et al. 2006, 2007).

Null Hypothesis: There is no effect on fingerling growout performance, survival or size distribution from fish collected as fry from incubation units, hapas, and ponds.

<u>Schedule</u>

April 2007 – September 2007	Studies 1 & 2
November 2007 – April 2008	Study 2
June 2008 – November 2008	Study 3
January 2009 – June 2009	Study 4
July 2009 – September 2009	Write technical report and CRSP final report

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DEVELOPMENT OF SMALL-SCALE *CLARIAS* FINGERLINGS AS BAIT FOR LAKE VICTORIA COMMERCIAL FISHERIES IN WESTERN KENYA

Quality Seedstock Development/Activity/07QSD02PU

Collaborating Institutions & Lead Investigators

Purdue University (USA) — Kwamena Quagrainie
Moi University (Kenya) — Charles Ngugi

Objectives

Our overall objective is to increase catfish fingerling production as bait to feed commercial fishing in Lake Victoria to reduce overexploitation of indigenous species and conserve the diversity of Lake Victoria Fisheries. This project therefore seeks to:

1. Train small- and medium-scale fish farmers as well as fisheries extension officers on hatchery technology, pond operation and management as well as business plans for catfish fingerling production.
2. Organize fish farmers into production clusters to produce catfish fingerlings as baitfish for Lake Victoria commercial fisheries.
3. Develop the baitfish market in the Lake Victoria region for catfish fingerlings (will become part of Investigation 1 (07MER02PU). This will help to enhance biodiversity habitats and populations.
4. Provide training for fisheries extension officers on technology transfer mechanism to fish farmers

Significance

In spite of several decades of fish culture, Kenya's aquaculture has not come far and remains a young industry, practiced mainly on a relatively small scale using tilapia (Oreochromis niloticus) and catfish (Clarias gariepinus). The artisanal fishery of Lake Victoria, the largest commercial fishery in Kenya, has been degraded by environmental deterioration (water hyacinth) and a decrease in the number of smaller food species following introduction of the voracious Nile perch (Lates niloticus). The stock of catfish in Lake Victoria has also been drastically reduced because wild-caught juveniles are used as bait for Nile perch hooks deployed daily in the commercial fisheries. Although trawling for Nile perch was practiced by some fishers in the past, it is now illegal in the Winam Gulf, a main fishing zone in Kenya. So, some fishers have resorted to longline fishing, using fingerling-sized Clarias as bait. The traditional supply of fingerlings is wild-caught from Lake Victoria, but this supply is intermittent and seems to be related to the extent of floating and drifting water hyacinth mats in near-shore areas, with Clarias being numerous under the thick growth. Fishers usually use small-mesh beach seines and small seine nets to catch fingerlings for bait, but beach seining is highly destructive of the spawning habitats of native cichlids and is illegal. The Fisheries Department (FD) has imposed a fishing moratorium for small seine nets from 1st April to 31st August every year thereby rendering bait fisher unemployed for over four months. Fishermen also find themselves in a difficult situation because they need the bait on a daily basis at an affordable price to be able to continue fishing. Income opportunities therefore exist for current agricultural farmers to diversification into small-pond aquaculture to provide farm-raised catfish fingerlings as alternative supply source of bait for the commercial fisheries on Lake Victoria.

The African catfish, Clarias gariepinus, is endemic to the Lake Victoria region. It is popular with the communities living around the lake. Clarias is popularly farmed in polyculture with Nile tilapia (Oreochromis niloticus) to control unwanted tilapia populations. Catfish are also grown in monoculture as food fish. Clarias culture is being recognized for its importance as bait fish for the Lake Victoria Nile perch fishery.

Frame Survey results for the year 2000, 2002, 2004 and 2006 showed that there were between 2.5 million and 3.0 million long line hooks operated on the Kenya side of Lake Victoria. This gives the number of boat using an average of 1,000 hooks per day to be 2,500 to 3,000 requiring a similar number of baits on daily basis. At 300 fishing days per year, it is estimated that there is an annual demand of between 750 and 900 million fingerlings at the optimum. At the reported selling price of KShs 5.00 to 8.00 per fingerling and an estimated production cost of about KShs 0.50 per fingerling, farm-based production of Clarias fingerlings could be a highly profitable business for fish farmers. Despite the huge demand for Clarias bait fish in the region, production has been very limited.

Spawning of Clarias is not a major problem, but they generally have a very low survival of the juveniles. However, studies such as Ngugi et al. (2004, 2005) have reported successes in survival of catfish juveniles from appropriate stocking densities for fry nursed indoors in 30-L glass aquaria, as well as studies on appropriate stocking densities and varied amounts of cover provided for fry reared in hapas in outdoor ponds. Results from the studies and others suggest that additional work is needed on fingerling survival to increase catfish production for bait as well as food fish. Increased production of catfish fingerlings will raise farm income and contributing to food security in the area.

Quantified Anticipated Benefits

- Farmers will be able to evaluate and compare alternative fingerling production technologies and apply suitable technologies to produce *Clarias* fingerlings for the Lake Victoria commercial fisheries.
- Fish Farmers will learn how to keep good records regarding the operation of fish ponds so that evaluations can be based on facts and so that there are records of the actions that did or did not result in increased fish production. This will in turn result in higher fish production and increased revenues from fish sales for participating farmers.
- Other farmers who know about the project may elect to adopt the new technologies or apply the improved management practices they have observed from neigbors.
- There will be spin-off farmer-to-farmer contacts increasing dissemination of information and reducing government cost on extension.
- Data recorded during the course of these activities will be useful for the development of enterprise budgets and business plans for the types of pond systems in use.
- Fisheries extension officers will develop the knowledge and skills on fish hatchery and technology transfer for effective extension services to fish farmers.

Anticipated Benefits

- New information or technologies resulting from this project will be transferred to farmers producing *Clarias* fingerlings through training courses conducted at the FD fish farm/Moi University fish farm and at the farms of cluster *Clarias* producers.
- Increased supplies of *Clarias* fingerlings will provide Lake Victoria Nile perch fishers with a reliable source of bait.
- Fishing pressure on immature *Clarias* in Lake Victoria will be reduced. Reduction in beach seining will result in reduced habitat destruction on native fishes in Lake Victoria.
- Net income to fishers may increase if baitfish is more available and if costs are kept down through competition among bait producers.
- The number of women taking up this activity as a proportion of males (trained and nontrained through extension) will increase.
- A steady supply of *Clarias* fingerlings will also help producers in areas where *Clarias* is gaining popularity as a cultured food fish.

Impact Indicators and Targets

- The number of small-scale farmers producing catfish bait on commercial and sustainable basis will be monitored. The baseline will be set at zero and a benchmark of 70% of trained cluster farmers will be used for evaluation.
- The target is to provide about 5% of all bait fish required in Lake Victoria basin or 130,000 fingerlings per day, since production of the catfish bait is expected to commence immediately after training and putting in place the basic production facilities.
- With quarterly production of 960,000 per annum, there will be an annual production of 3.84 million with a turnover of KShs. 7.68 million (US $1.1million) assuming a sale value of KShs. 2.00 per fingerling.
- This level of production can support about 18 traders handling an average of 2,000 fingerlings per day.

Activity Plan

Fish farmers will be selected from Western Province, a high aquaculture concentration area. The area has good climate and optimum temperatures for catfish and tilapia growth. The region is very close to the Lake Victoria, the where catfish fingerling market can be developed for Nile perch fishery. Marketing channels will be easy to establish because of the proximity. Transportation of inputs from major town such as Kisumu city will be cheaper and transportation of produce from fish farms to the end markets will be faster.

There will be six cluster sites selected based on static pond and catfish fingerling production suitability. These sites will be selected from six districts in western Kenya. Farmers will be selected according to their willingness and ability to participate. A rapid needs assessment on the capabilities of cluster farmers, basic requirements such as minimum number of ponds, ability to source required inputs such as organic fertilizer will be carried out. Experimental and production ponds for the proposed project will be selected based on:

- Farmers interested in participating in the cluster and growing cattish fingerling mainly to supply the baitfish market
- Surface area per pond — should have a minimum of 200 m^2, and a maximum of 1,000 m^2 and the farmer must have not less than two ponds.

Farmers willing to construct ponds to meet the above criteria will be allowed to participate. Farmers should be willing to work in a group and also be willing to be trained.

Six cluster groups will be formed with each cluster having up to ten (10) members. In areas where groups do not have leadership structures, they will be assisted to put them in place and start the process of registering with the Social Service Department to enable them open bank accounts. Cluster formation will be sequenced with each cluster identification and formation taking five days. The first cluster will then proceed to start catfish fingerling production. This will ensure that no two clusters perform tasks at the same time. The sixty (60) fish farmers selected as cluster members will undergo site based training for each cluster to prepare them for running their project on profitable basis.

Training will cover various topics such as hatchery management; methods of seed production (catfish and tilapia); applications of various hatchery production techniques including broodstock collection, fertilization and spawning techniques, incubation and hatching, egg mortality and their treatment, larval rearing and mass catfish fry production; fish nutrition and feed; fish health management; as well as transportation of live fish. Extension officers will also learn leadership skills, and communication. From this training, the HCPI will be able to identify areas of project development and implementation concern and generate mitigation measures.

Schedule

Start date: June 1, 2007
End date: September 30, 2009

June 2007 – August 2007	Site visits Identification of cluster farmers, Cluster mobilization and sensitization Group dynamics Selection and registration of graduate students
September 2007 – March 2008	Organize pre-production workshops Identification of end markets and linkages Graduate student course work Develop fact sheet, technical materials
April 2008 – September 2008	Conduct six residential Trainings for farmers on pond management and record keeping Procurement of hatchery accessories and catfish brooders. Develop brochure Farmers' pond preparation and construction of cluster collection centers.
September 2008 –June 2009	Video recording and posters Seed production at Moi University Linking farmers to markets and end-users Graduate student thesis field research Develop monitoring and evaluation instruments
July 2008 – September 2009	Production monitoring and recording Graduate student's theses defense Post –production Farmers' workshop Submission of project report

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TOPIC AREA
HUMAN HEALTH IMPACTS OF AQUACULTURE

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MONITORING AND REDUCING MICROCYSTINS IN TILAPIAS AND CHANNEL CATFISH CULTURED IN A VARIETY OF AQUACULTURE SYSTEMS

Human Health Impacts of Aquaculture/Experiment/07HHI01UM

Collaborating Institutions & Lead Investigators

University of Michigan (USA)	James S. Diana
Hainan University (China)	Lai Qiuming
Shanghai Fisheries University (China)	Yang Yi
Wuhan University (China)	Song Biyu

Objectives

1. Determine the content of microcystins in aquaculture ponds/tanks at different levels of intensification and different culture environments.
2. Evaluate body burdens of microcystins in the flesh of tilapia cultured under different systems.
3. Test possible depuration strategies to eliminate microcystins from the flesh of cultured tilapias.

Significance

Microcystins (MCs) or nodularin cyclic peptide hepatotoxins are cyanobacterial (blue-green algal) metabolites found world-wide in freshwater, brackish and marine environments. They are produced by natural blooms of *Microcystis, Anabaena, Oscillatoria and Nodularia* species (Carmichael 1994, Lindholm et al. 1989, Paerl et al. 2001, Pires et al. 2004, Soares et al. 2004). These blooms are often dominated by *Microcystis* spp. (Orr et al. 2001, Park 2001). Microcystins are considered to be the most common and dangerous group of cyanotoxins (Sivonen and Jones 1999) and are potentially hazardous to human health, especially by inducing tumors through consumption of MCs-contaminated food products such as fish (Nishiwaki-Matushima et al. 1991, 1992; Codd et al. 2005). No case of human death has yet been documented from oral consumption of cyanobacteria toxins, but chronic toxic effects from exposure in food needs to be considered, especially if there is long-term and frequent exposure (Chen et al. 2006, Xie 2006).

There have been extensive studies on MC bioaccumulation in fishes (Tencalla et al. 1994, Soares et al. 2004, Zhao et al. 2006) and mussels (Vasconcelos 1995, Amorim and Vasconcelos 1999, Williams et al. 1997, Pires et al. 2004) under laboratory conditions. Fish species such as rainbow trout (*Oncorhynchus mykiss*), *Tilapia rendalli*, channel catfish (*Ictalurus punctatus*) and Nile tilapia (*Oreochromis niloticus*) were shown to accumulate microcystins in internal organs as well as in muscle (studies listed above). Therefore, the occurrence of toxic cyanobacterial blooms producing microcystins in aquaculture ponds could represent a risk to the quality of fish flesh to consumers. On the other hand, only a few studies on MC accumulation have been conducted in the field (Chen et al. 2006). For example, MC content was examined in liver, kidney, gut, and muscle of Nile tilapia in an Egyptian fish farm (Mohamed et al. 2003), and a survey was done on eight species of fish in Lake Chaohu, China (Xie et al. 2005). Their results indicated that Nile tilapia, Chinese carps and other commonly cultured species from aquaculture or cultured-based fisheries did contain microcystins at significantly elevated levels in various fish organs. Furthermore, the study conducted by Mohamed et al. (2003) revealed that Nile tilapia can depurate and excrete microcystins into the bile and surrounding water as a way to avoid toxicity from such a potent hepatotoxin.

The eutrophication of freshwater ecosystems normally results in frequent cyanobacterial algal blooms, and as a result aquatic crops from these systems may be contaminated by microcystins. On the other hand, filtering-feeding species such as silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*) as well as Nile tilapia are used by managers to consume and control these blooming algae (Xie 2003). China has the largest production of aquatic crops among all countries in the world. About 50% (0.9 million metric tons) of the world's total tilapia production originates in China (FAO 2004), which is also the largest supplier of tilapias to U.S. markets (FAO 2004). The purpose of this study is to examine the content of microcystins in the flesh of tilapias cultured in different types of culture systems at different intensification levels.

Quantified Anticipated Benefits

The results of this study will provide information on status of microcystins in cultured tilapias and possible management strategies to avoid accumulation of MCs. It will allow government agencies to establish policies and plans for the management of microcystins in cultured tilapias. Management strategies for microcystins will secure aquatic product safety and benefit human health. Immediate impact of this study includes the number of culture systems in which microcystins have been detected in both water and tilapia flesh. Also, depuration studies will estimate the times required for holding tilapias in clear water to reduce or eliminate microcystins in their flesh.

Research Design & Activity Plan

Locations: Sichuan and Hainan provinces; particularly, Shanghai Fisheries University, Wuhan University, and Hainan University.

Methods:
1. Farm selection: Tilapia culture systems in Hainan province will be classified as semi-intensive freshwater ponds, intensive freshwater ponds, intensive indoor freshwater tanks, semi-intensive brackish/seawater ponds, intensive brackish/seawater ponds, and intensive indoor brackish/seawater tanks, while channel catfish culture systems will be classified as semi-intensive freshwater ponds, intensive freshwater ponds, intensive indoor freshwater tanks, cage culture in rivers, and cage culture in reservoirs. Ten farms will be selected randomly from each of the above types of ponds/tanks or cages for the examination of microcystins in both fish flesh and water.

2. Assessment of microcystins in the water and fish:
2.1. A column water sample will be collected from each of the selected ponds/tanks/rivers/reservoirs for the analysis of microcystins using solid phase extraction (SPE), enrichment and ultra performance liquid chromatography (HPLC) and liquid chromatography/mass spectrometry (LC/MS) (Nicholson and Burch 2001).

2.2. Five fish will be sampled randomly from each of the selected culture systems at harvest. Fish will be filleted and fillets used for analysis of microcystins in flesh using the above methods.

2.3. Depuration experiment: One hundred tilapias or channel catfish contaminated by microcystins will be collected from the culture systems with the highest microcystin content in fish flesh identified from the above determination. They will be held in an indoor recirculating tank supplied with clear water. The identified culture system will serve as a control. Three tilapia or catfish will be sampled and killed daily from both the indoor recirculation tank and the identified culture system for the analyses of microcystins in fish flesh until the level of microcystins in fish flesh from the recirculating tank becomes zero or constant.

3. Null hypothesis, statistical analysis:

Null hypotheses: Different culture systems have no effects on microcystins in water or in tilapia or channel catfish flesh; different time periods for holding tilapias in clear water have no effects on the levels of microcystins in tilapia or channel catfish flesh compared to those kept in the original culture system.

Data will be analyzed using ANOVA, t-test, correlation and regression.

4. Workshop: Upon the completion of the analyses, a one-day workshop will be organized to present findings from this project as a continuing consultation process with farmers, private enterprises, and government officials especially those involving in monitoring food quality and safety. At least 50% of the workshop participants will be women.

Schedule

April 2007 – September 2009. Report submission: no later than 30 September 2009.

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FOOD SAFETY STUDY OF LEAFY GREENS IRRIGATED WITH TILAPIA FARM EFFLUENTS

Human Health Impacts of Aquaculture/Experiment/07HHI02UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)	Kevin Fitzsimmons
Delaware State University (USA)	Dennis McIntosh
Universdad Autónoma de Tamaulipas (Mexico)	Pablo Gonzalez Alanis

Objectives

1. Determine the levels of E. coli, fecal coliforms, and salmonella that are present on leafy vegetable irrigated with water from pond and indoor recirculating tilapia production systems.
    - Null hypothesis 1: The levels of *E.coli*, fecal coliforms, and salmonella will be the same between the systems.
    - Null hypothesis 2: The levels of *E.coli*, fecal coliforms, and salmonella will be above a level of concern for human consumption.
2. Determine if the bacterial levels in effluent and/or leafy vegetables are reduced below detectable limits with a simple UV treatment of effluent water.
    - Null hypothesis 3: The levels of *E.coli*, fecal coliforms, and salmonella will not be significantly reduced by the UV treatment of effluent water.

Significance

In 2006, several nation-wide (United States) epidemics of *Escherichia coli* or related gastro-intestinal pathogens were traced to consumption of fresh vegetables (spinach, lettuce, green onions). In several of these cases the vector was thought to be contamination from human or animal wastes applied through irrigation water. As we encourage more organic farming methods and re-use of composted wastes and effluent waters from animal operations to irrigate field crops, we increase the opportunities for these types of contamination. One alternative is to increase the multiple-use of water for production of fish and use of these effluents for irrigation of vegetables that are bound for human consumption.

Across much of Asia, fish is the primary protein component of the diet and integrated fish and vegetable farming has been practiced for centuries. However, very few studies have been conducted to determine any health hazards that may result from this practice. The assumption is that fish being heterotherms (cold-blooded) and obviously not mammals, they are unlikely to be vectors for intestinal pathogens. However, fish farms do have human workers and ponds can be visited by farm animals and pets. Examination of typical aquaculture farm effluents and any residual potential pathogens that might be left on leafy vegetables irrigated with fish farm effluents, bound for direct human consumption would be an advisable precaution.

Bacteria have been identified in aquaculture systems that are considered human pathogens, such as fecal coliforms including *Escherichia coli* (Ogbondeminu, 1993; Flick, 1996; Del Rio-Rodriguez et al., 1997; Pullela et al., 1998), *Clostridium botulinim* (Pullela et al., 1998), Pseudomonas species (Nedoluha and Westhoff, 1995), *Aeromonas hydrophila* (Nedoluha and Westhoff, 1997) and salmonella species (Ogbondeminu, 1993). The microflora of the fish gills, skin, and digestive tract have been shown to reflect the microflora of the water they inhabit and may also pose a threat to humans (Reilly and Käferstein, 1997; Nedoluha and Westhoff, 1997; Ogbondeminu, 1993).

Preliminary studies at the University of Arizona demonstrated that total coliforms levels of 104 CFU's/100ml and fecal coliform levels of 103 CFU's/100ml can be found in two separate recirculating systems rearing grass carp and tilapia (McKeon et al., 2000; McKeon, 1998). Samples from this system were found to contain total coliform levels as high as 104 CFU/100 ml. Fecal coliform levels were found to be variable over a test period of two months. Counts were as low as 1 CFU/100 ml to as high as 103 CFU/100 ml. The mere presence of organisms does not imply that a health threat is present. Furthermore, the Colilert rapid detection test demonstrated a negative result for E. coli presence. The source of the coliforms was unknown.

Reuse of aquaculture effluent for crop irrigation has been suggested as an environmentally appropriate technique. The Environmental Protection Agency has recommended land application and crop irrigation as a Best Management Practice for the disposal of aquaculture effluents (EPA-Federal Register 40 CFR Part 451, 2004). Use on a variety of filed crops seems to be perfectly safe. Use of aquaculture effluent on leafy vegetables, while common across Asia and in numerous aquaculture–hydroponic (aquaponic) systems in the US, has not been closely examined for the presence or absence of gastro-intestinal bacteria. If these bacteria are present, even in very low numbers, a simple UV treatment system may further reduce them to negligible or even non-detectable levels.

Quantified Anticipated Benefits

If levels of pathogenic bacteria in fish effluent or fish effluent treated with a low cost UV system are determined to be low enough for safe use as irrigation water, these results would be useful for developing farm management strategies that will improve food safety for consumers of crops grown in integrated production systems.

Research Design & Activity Plan

Location of work: The greenhouse tilapia production system is at the Veterinary College at the Universidad Autónoma de Tamaulipas. The outdoor system is in the town of Abasolo, outside the city of Ciudad Victoria. The microbiological lab facilities are in the Veterinary College at UAT.

Materials & Methods: We propose that complementary lab and field studies be performed to examine the bacterial flora typically encountered in in-door and out-door aquaculture operations. In conjunction, we would grow lettuce and spinach in greenhouses and in the field which would be irrigated with effluents from attendant fish (tilapia) production facilities.

Greenhouse studies: Tilapia production facilities at the Universidad Autónoma de Tamaulipas utilize recirculation technology to rear fish. These systems incorporate biological filtration, which includes nitrifying and heterotrophic bacteria to treat water so that it can be recycled to the fish tanks for continuous rearing of fish in the same water. The experimental system would utilize a portion of the treated water stream to irrigate vegetable beds. These effluents from the fish would be used to flood irrigate plots of lettuce and spinach, to be grown in summer and winter, respectively. Cool weather and warm weather crops may be more or less susceptible to contamination. Soil beds would be prepared using a standardized experimental soil bed composed of local soil mixed with sand and a commercial potting soil mix.

The effluent water and a control water supply, tap water from the city water source, used for irrigation would be sampled for bacterial contamination during each irrigation event (every 3rd day for the first three weeks and 5th day for the final nine weeks).

At the end of the growing cycle, leaves from the portions of the plant typically harvested for commercial sale will be collected and sampled for total fecal coliforms, *E. coli*, enterococci and salmonella. Total colony forming units (CFU’s) will be determined and reported on a per 100 ml basis.

Field Studies: Tilapia production facilities at Abasolo, Tamaulipas utilize a flow-through system with source water from wells passing through two or more ponds stocked with fish before use as irrigation water for olive trees, alfalfa, or Bermuda grass. This farmer cooperator has agreed to provide matching support to the project and will field test lettuce and spinach during winter, spring and summer respectively, as well as asparagus, to be irrigated with fish effluent and with untreated water directly from the well. The field plots will require more frequent irrigation in the dry desert field environment, with the exact irrigation schedule dependant on weather conditions. Sample schedule will not be a frequent as the lab testing but will encompass at least five sample dates during the irrigation schedule.

At the end of the growing cycle, leaves from the portions of the plant typically harvested for commercial sale will be collected and sampled for total fecal coliforms, E. coli, enterococci and salmonella. Total colony forming units (CFU's) will be determined and reported on a per 100 ml basis.

Lab Analyses: Fecal coliforms will be isolated by membrane filtration of the sample and direct plating of the filter on mFC agar according to Standard Methods (APHA, 1998). For E. coli isolation, a presumptive test involving Lauryl Sulfate Tryptose (LST) broth will be employed. Positive presumptive tests (gas production) will be followed by the tests with *E. coli* (EC) broth (Hitchins et al., 1992).

Enterococci will be isolated according to Standard Methods (APHA, 1998) using membrane filtration and m-Enterococcus agar for determining the presence of fecal streptococci. Confirmation will be performed after transferring typical colonies from a membrane to the surface of a brain-heart infusion (BHI) agar and incubation. A catalase test and gram stain of the BHI isolate will be done as well as observations of growth on bile esculin agar and growth in BHI broth containing 6.5% NaCl to confirm the presence of enterococci (APHA, 1998; Hartman et al., 1992).

Salmonella pre-enrichment will be accomplished using lactose broth (Flowers et al., 1992) and selective enrichment will subsequently be performed using a ducitol selenite broth. After enrichment subcultures will be performed on Salmonella-Shigella (SS) agar (Difco Laboratories, 1984) and Xylose Lysine Desoxycholate (XLD) agar (APHA, 1998). Following incubation, an oxidase test and indole test will be performed. A short set of biochemical tests will be followed by inoculation of an API 20E strip as per directions by API (Flowers et al., 1992; APHA, 1998). Testing with salmonella antiserum will conclude the confirmation tests (APHA, 1998).

Statistical Analyses: We will compare total fecal coliform, enterococci and salmonella densities (Colony Forming Units per 100 ml) among treatments with a two-way ANOVA where season and irrigation treatment are the two independent factors. There will be two levels for the Season factor (winter and summer) and three levels of the Irrigation-treatment factor (tap water/well water, and raw effluent water and effluent water treated with ultraviolet light). Analysis for statistically different levels of potential pathogenic bacteria in the fish effluent, treated effluent and control water used to irrigate vegetables will be determined. Statistically different levels of potential pathogenic bacteria actually collected off the edible portions of the plants will also be examined. SysStat software will be utilized to facilitate analysis.

Schedule

October 2007	Stock ponds and tanks with tilapia and prepare planting beds
November 2007	Pre-irrigate and germinate spinach in beds, collect first effluent samples
March 2008	Harvest first spinach crop and sample inner leaves for bacteria
May 2008	Pre-irrigate and germinate lettuce, collect effluent samples
September 2008	Harvest first lettuce crop and sample inner leaves for bacteria

November 2008	Pre-irrigate and germinate second spinach crop, collect effluent samples
March 2009	Harvest second spinach crop and sample inner leaves for bacteria
April 2009	Pre-irrigate and germinate second lettuce crop, collect effluent samples
August 2009	Harvest second lettuce crop and sample inner leaves for bacteria
September 2009	Prepare final reports

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INTERNATIONAL WORKSHOP FOR AQUACULTURE SANITATION

Human Health Impacts of Aquaculture/Activity/07HHI03UH

Collaborating Institutions & Lead Investigators

Pacific Aquaculture & Coastal Resources Center, University of Hawaii at Hilo (USA)	Maria Haws
Louisiana State University (USA)	John Supan
Centro de Investigación de Alimentos y Desarrollo (CIAD) (Mexico)	Omar Calvario Martínez
Universidad Autónoma de Sinaloa, Culiacán (Mexico)	Eladio Gaxiola Camacho
Universidad Autónoma de Sinaloa, Mazatlán (Mexico)	Guillermo Rodriguez Domínguez

Objectives

1. This workshop will serve to bring together researchers, practitioners and community stakeholders working in the field of aquaculture sanitation to exchange information, research findings and experiences in several aspects of aquaculture sanitation.
2. The workshop will also provide a venue to present prior and current findings of CRSP-sponsored work.

Significance

In 2004-2006, a team of thirty-one researchers from UAS, CIAD, CESASIN, UHH, URI, UCA (Nicaragua) and Ecocostas (Ecuador) collaborated on research into three topics in human health and aquaculture and subsequently produced a book of case studies (Haws et. al. 2006). Not surprisingly, the team found that deficiencies in aquaculture sanitation for all cultured species except shrimp, presented hazards to human health at the individual and community level, as well as affecting the efficiency and profitability of aquaculture operations. Shrimp was largely an exception to this, since most shrimp farming in Mexico is conducted by large commercial operations that must comply with international export standards and have the financial resources to do so. In contrast, small-scale producers of shrimp for the local market, freshwater fish farmers (tilapia, trout, catfish, local cichlids) and shellfish farmers all exhibited severe problems with processes that affect food safety and sanitation. In a few cases, potential life-threatening illnesses or death could result from the lack of awareness and capacity. Examples of this include the growing incidence of gnathosome parasites in freshwater and brackish fish and shellfish-borne diseases (e.g., hepatitis, Salmonella A). The lack of appropriate handling and preservation methods in general leads to contamination and rapid decomposition of products, thus reducing revenues to producers and vendors and possibilities of marketing outside the immediate local area.

Quantified Anticipated Benefits

Intensive, hands-on training in the theory and application of HACCP and related topics will improve the capacity of producers and other stakeholders to produce safe aquaculture products and improve the efficiency and profitability of aquaculture operations.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; State Committee for Aquatic

Sanitation (CESASIN); Sinaloa and Nayarit fish farmeres; and the Federation of Shrimp Cooperatives. Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of institutions directly or indirectly benefiting: 12
- Number of individual participants in technical training: 70
- Number of communities benefiting: 34
- Number of documents produced or contributed to: 1
- Students involved: 1 graduate student
- Training modules produced: 3
- Publications: 1

Activity Plan

The workshop will take place in Culiacán, Mexico and have two components:
1. Presentation of research findings and new methods developed during prior work.
2. Hands-on training in appropriate pre- and post-harvest methods.

The training team will consist of international and national experts drawn from the University of Arizona, Louisiana State University, Texas A&M, University of Hawaii-Hilo, Autonomous University of Sinaloa, Autonomous University of Sinaloa, Autonomous University of Nayarit, CIAD, and the Central American University (UCA). Drs. Haws and Supan will advise and participate in this work. The UAS Masters degree student will assist with this work.

Schedule

The workshop will be held in November 2007. Workshop proceedings will be published in January 2008.

Literature Cited

Haws, M.C., E. Ochoa-Moreno, G. Gaxiola Camacho, G. Rodriguez D. and J.A. Tobey. 2006. (eds.). (submitted November 2006). Human Health and Aquaculture: three case studies of the relationships between human health, the environment and aquaculture in coastal communities of Sinaloa, Mexico. Pond Dynamics/ Aquaculture Collaborative Research Support Program, Oregon State University. In Spanish. 157 pp.

REGIONAL WORKSHOP ON SHELLFISH CULTURE AND SANITATION

Human Health Impacts of Aquaculture/Activity/07HHI04UH

Collaborating Institutions & Lead Investigators

Pacific Aquaculture & Coastal Resources Center, University of Hawaii at Hilo (USA)	Maria Haws
Louisiana State University (USA)	John Supan
Centro de Investigación de Alimentos y Desarrollo (CIAD) (Mexico)	Omar Calvario Martínez
Universidad Autónoma de Sinaloa, Culiacán (Mexico)	Eladio Gaxiola Camacho
Universidad Autónoma de Sinaloa, Mazatlán (Mexico)	Guillermo Rodriguez Domínguez

Objectives

The culture of bivalves and management of molluscan fisheries are important economic and environmental themes in Mexico and the rest of Latin America with close linkages to various dimensions of human health, but relatively little research or intellectual exchange occurs on the topic on a regional basis as compared to other species.

1. This regional workshop is designed to allow researchers, educators, government representatives, fishers and farmers to meet, exchange information and experiences and develop working partnerships.
2. This is also an opportunity for molluscan researchers and stakeholders to establish linkages with international institutions such as the NSSP, FAO and Pacific Shellfish Growers Association.

Significance

More attention is needed for bivalve culture and molluscan fisheries in order to advance shellfish industry development and improve management of molluscan fisheries. This workshop will allow specialists in the fields of shellfish sanitation, culture, ecology and fisheries to exchange experiences, information and lessons learned. This workshop will follow in the model of two prior international workshops held by the team and sponsored by CRSP that focused on aquaculture themes, strategic planning and development of collaborative partnerships. One outcome of this will be the workshop proceedings which will include the workshop presentations, supplementary technical materials and training aids. Past workshop proceedings have been widely read and used in the U.S. and Latin America as reference materials and for training and educational purposes (Haws et. al. 2004; 2005).

Quantified Anticipated Benefits

This work will benefit stakeholders through improving access to new and critical information and methods. Proceedings from this will benefit researchers, educators, government officials and farmers.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; and State Committee for Aquatic Sanitation (CESASIN). Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:

- Number of institutions directly or indirectly benefiting: 19
- Number of individual participants in technical training: 25
- Number of communities benefiting: 34
- Number of documents produced or contributed to: 1
- Students involved: 1 graduate student
- Training modules produced: 3
- Publications: 1

Activity Plan

1. The conference will be held in May 2008 for three days at the Culiacán campus of UAS. Researchers at UAS and CIAD will organize and hold the conference.
2. Field visits will be made to farms and processing sites.
3. Stakeholders and speakers from shellfish farming groups or institutions with expertise in the field will be invited from the Mexican States of Sonora, Sinaloa, Baja California Sur, Baja California Norte, Nicaragua, and Ecuador. US representatives will include M. Haws (UHH) and J. Supan (LSU). Other stakeholders will be welcome to attend although funding may not be provided to cover their attendance.
4. Conference proceedings will be compiled and will include supplemental materials and training aids and will be distributed widely to workshop participants and other stakeholders.
5. Materials will also be posted on institutional websites.
6. Drs. Haws and Supan will advise and participate in this work. The UAS Masters degree student will assist with this work.
7. The workshop will take place in Culiacán, Mexico.

Schedule

To be held for three days in May 2008. Proceedings to be published and distributed by June 2008.

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Haws, M.C., E. Gaxiola, G. Dominguez and J. Tobey. 2004. Proceedings of the first international workshop on human health and aquaculture and extension methodology. May 6-9, 2004. Mazatlán, Mexico. Pacific Aquaculture and Coastal Resources Center, University of Hawaii at Hilo.

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MICROBIOLOGICAL QUALITY OF BIVALVE GROWING WATERS AND TISSUES

Human Health Impacts of Aquaculture/Experiment/07HHI05UH

Collaborating Institutions & Lead Investigators

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Louisiana State University (USA)	John Supan
Centro de Investigación de Ecosistemas Acuáticos, Universidad Centroamericana (Nicaragua)	Agnes Saborio Coze Nelvia Hernandez

Objectives

This work aims to complement current efforts to culture and manage black cockles (*Anadara* spp.) and other bivalves in Nicaragua by monitoring water quality in coastal estuaries where cockles are widely fished by poor communities and monitoring pathogens in cockle tissues in order to provide the long-term data needed to identify suitable areas for shellfish collection and culture.

Significance

Various species of open ocean and estuarine bivalves are a critical fisheries resource for Nicaragua, as well as other Latin American countries, but are threatened by overfishing and suspected increasing levels of contamination as coastal populations and activities grow. Numerous species are fished and consumed in Nicaragua including *Crassostrea rhizophorae* (mangrove oyster), *Crassostrea gigas* (Japanese Oyster), *Ostrea iridescens* (rock oyster), *Anadara tuberculosa*, *Anadara similis* (black cockles), *Iphigenia altior* (beach clam), *Pinctada mazatlanica* (pearl oyster), *Spondylus sp.* (thorny oyster).

Two species of black cockle, *Anadara similis* and *A. tuberculosa*, are particularly important for the poor coastal communities of Nicaragua. Many communities in and around coastal estuaries depend on daily fishing of cockles for income and as a food source. The latter use is particularly important as cockles are the food of last resort when poor families have no other source of food as traditionally they could be readily found and harvested close to home. Single, female heads of households are especially dependent on the cockle resource. The Nicaraguan bivalve populations are also important on a regional basis since fishers from El Salvador and Honduras routinely fish in Nicaraguan waters and sell their catch in their respective countries.

UCA has been leading efforts in recent years to conduct research into basic cockle biology and ecology, develop culture methods for cockles and other bivalves, and test alternative management regimes for bivalve fisheries. The latter is of importance since current regulations depend on a long closed season which does not appear to coincide with the peak reproductive season or other biological attributes, and is based simply on the time during which demand is highest. There is also an overall lack of compliance with the existing regulations as little outreach has been done to the fishers and enforcement capacity is minimal. Current research to improve management involves participatory research with local communities to establish community-controlled no-take zones for cockles and allowable fishing zones, and efforts to elucidate key parameters such as size at maturity to inform efforts to development better management regimes. USAID and other donors have recently funded these efforts which are beginning to yield positive results. The Nicaraguan Ministry of the Environment (MARENA) is a partner in these efforts which emphasize co-management with the local communities.

Shellfish-borne illnesses are of serious concern for shellfish harvested from coastal areas. Gastrointestinal illness due to *E. coli*, and outbreaks of salmonella and *Vibrio parahaemolyticus* appear to be on the rise based on the sparse data collected by rural health clinics and urban hospitals and observations by field workers. This data is probably highly inaccurate due to poor diagnosis and under-reporting, but incidences are high. Infants and children in rural areas appear to be particularly prone to serious illness and death due to these diseases.

A key aspect of long-term sustainability for the culture and fisheries management efforts is the eventual classification of shellfish growing and fishing waters to assure that harvested shellfish are safe to eat. Nicaragua's fished bivalves are exported widely within the country, and allegedly, illegally to other Central American countries, so the food quality and safety of these widely consumed shellfish is of national and international concern. This work aims to establish a long-term monitoring program of shellfish waters in three important estuary areas to determine which areas are best for establishing allowable fishing zones and areas for shellfish culture. Initial testing of monitoring protocols and procedures has resulted in suitable methodology for monitoring of water quality and tissues. At least two years of monitoring over the wet and dry seasons is needed to accurately classify the shellfish grounds and provide a reasonable level of assurance of safety for the consumer. Protocols are similar to those recommended by the U.S. National Shellfish Sanitation Program (NSSP 2006). This work will also be accompanied by outreach activities intended to educate cockle fishers and the public on issues related to shellfish sanitation to reduce the incidence of serious disease. UCA will lead this effort with support from UHH (M. Haws) and LSU (J. Supan).

Quantified Anticipated Benefits

This work will allow researchers and managers to determine which areas are safest for shellfish fishing and culture, and provide this information to stakeholder groups on the coast to guide co-management activities. This information may help reduce the incidence of serious and potentially fatal diseases. It is also thought that due to increasing consumer awareness and fear of shellfish-borne diseases, shellfish that can be certified as being harvested from clean waters may have added market value, and minimally, may guide poor coastal residents who intend to consume shellfish, to collect in areas that are the safest and avoid contaminated areas. It will also benefit current efforts to develop shellfish aquaculture by providing key information to the site selection process. Additionally, it will assist in efforts to clean up coastal areas and with community sanitation programs by demonstrating a clear link between harmful practices and community health and pinpointing areas where improvement is most needed.

Target groups for this work include: aquaculture extension workers and researchers in Nicaragua and key private sector representatives. Approximately 24 communities that depend on bivalve resources surrounding the three estuaries will benefit from this work. Groups benefiting in Nicaragua include extension and research staff at the Central American University and partner organizations such as MARENA, and fishers and women's groups in three Nicaraguan estuaries. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of institutions directly or indirectly benefiting: 9
- Number of individual participants in technical training: 20
- Number of communities benefiting: 24
- Number of documents produced or contributed to: 1
- Students involved: 1 undergraduate

- Publications: 2 (technical report and article for CRSP newsletter)

Research Design & Activity Plan

Water quality monitoring and tissue collection sites in three estuaries (Estero Padre Ramos, Aserradores, and El Realejo; see Map) in Nicaragua have been selected during previous development of monitoring protocols. Monthly samples will be taken from sampling sites within each estuary. Tissues will be collected from the sample areas. Water samples are analyzed for *E. coli*. Tissues are analyzed for *Vibrio parahaemolyticus* using a PCR-based method. Two years of monthly sampling are required to accommodate extreme periods of rain and drought which can affect levels of pathogens, as recommended by the ISSC. Once sufficient data has been gathered, areas that meet minimal requirements for safe shellfish production can be identified although long-term monitoring in these areas will need to continue for as long as shellfish are collected or grown in these waters.

Outreach with communities, responsible government agencies (e.g., Ministry of Health, Environment), fishers, aquaculturists, and others will be conducted to raise awareness of this issue and to promote improved management of these estuary areas.

The UCA undergraduate student will assist with this work.

Schedule

Because sampling sites have already been identified and protocols developed, the work can begin immediately in May 2007, and will continue until June 2009.

Outreach will be conducted to disseminate results and management recommendations once the first year of sampling has begun, and will conclude in August 2009. Community members will be trained during each visit by researchers as each field visit includes a community meeting, short training event and active participation by community members in all field research activities.

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TOPIC AREA
FOOD SAFETY & VALUE-ADDED PRODUCT DEVELOPMENT

❖

MAXIMIZING THE UTILIZATION OF LOW VALUE OR SMALL SIZE FISH FOR HUMAN CONSUMPTION THROUGH APPROPRIATE VALUE ADDED PRODUCT DEVELOPMENT

Food Safety & Value-Added Product Development/Study/07FSV01UC

Collaborating Institutions & Lead Investigators

University of Connecticut-Avery Point (USA)	Robert Pomeroy
Inland Fisheries Research & Development Institute (Cambodia)	So Nam

Objectives

The overall objective of this study is to ensure high quality, safe and nutritious low value or small-size fish processed products for local consumers and the competitive international markets. The specific objectives are as follows:
1. Document and evaluate the low value or small size fish processing practices, and specifically fermenting, for human consumption, and to compare the traditional and modern processing practices.
2. Study the market channels and markets for processed low value fish.
3. Determine problems and issues relating to low value fish processing practices and value-added product development, and marketing and trade.
4. Identify the needs for further studies and activities.
5. Recommend policies and strategies to address the identified problems and issues in order to ensure high quality, safe and nutritious low value fish products for local and international trade, and to support value-added product development.

Significance

Fish is a staple and affordable food and nutrition source for most people in Cambodia and Vietnam and provides food security and livelihoods to millions of people living in rural areas (Svrdrup-Jensen, 2002; So & Haing, 2006). Cambodia has an annual per capita consumption of fish of 30-66 kg per person per year (So & Thuok, 1999; So & Haing, 2006). This comprises over 75% of total annual animal protein intake (Ahmed et al., 1998).

There is an abundance of low value/trash fish harvested using *dai* “bag net” and many other fishing gears in a short period during the peak fishing season (December–February) along the Tonle Sap and Mekong rivers, and their major tributaries. By estimation, at least 16,000 tons were caught by only one commercial type of fishing gear (bag net or *dai*) during 2004 (Hortle et al., 2005) and almost double (i.e., 30,000 tonnes) during 2005 (DoF, 2006). These fish are typically landed in many isolated locations and in a poor condition or severely damaged from the capture methods. Utilization of these fish is either through conversion into fish meal and other animal meal or direct use for livestock and aquaculture in the general vicinity of the landing site (So et al., 2005).

In Vietnam, as the national demand for fish sauce is predicted to double over the next ten years, there appears to be direct competition for mixed low value/trash fish between feeds and those needed to make processed value-added products. In Cambodia, low value/trash fish is used for human consumption, particularly by the poor as it is relatively cheap and provides good sources of protein and other nutrients. The fish is utilized for household food security and income. Artisanal processing is undertaken to make fermented fish or fish paste (*prahoc* in the Khmer language) or processed in other ways (e.g., fish sauce, *te trei* in the Khmer language) since the past centuries using different traditional processing practices and skills/techniques. This processed product is used in the household and sold in local and regional markets (Thailand, Laos, Vietnam) and provides income to the household. Tens of thousands of the Cambodia poor, especially rice farmers, who are living hundreds of kilometers away from the main natural freshwater bodies, come to the Tonle Sap River during December to February every year to buy low value or small-sized fish to make *prahoc*. Vietnamese traders are known to come to Cambodia to purchase both fresh and semi-processed low value/trash fish for further value-added processing in Vietnam. The increasing competition for low value/trash fish for aquaculture has reduced the supply of fish for these value-added products.

Yim & McKenney (2003a, b) studied the domestic fish trade and fish exports of high value fresh fish products from the Tonle Sap Great Lake to Phnom Penh and from the Tonle Sap Great Lake to Thailand, respectively. Recently there were also several case studies on local and international fish markets and trade of processed products of high value fish, e.g., Rab et al. (2006); Rab et al. (2005); Vanna (2005); and DoF/APIP (2001). However, there has been very limited assessment and documentation of processing practices of low value/trash fish and markets for the processed products by rural households in Cambodia. Much of what is known is anecdotal. An important issue is whether better processing can yield a better return for the limited resource. An analysis of processing practices will allow for development of both markets and value-added products.

<u>Quantified Anticipated Benefits</u>

- One student will be supported for their Master's Degree and two undergraduate students will be involved in the project through thesis research.
- 100 scientists, researchers, government fisheries managers and policy-makers, inter-government and non-government staff and extension agents will have a better understanding of low value/trash fish processing practices, including the role of women, and markets for these products and value-added product and market opportunities.
- 5000 poor households in Cambodia who rely on low value/trash fish processed products will have improvements in product quality.
- 2000 poor households in Cambodia, including women members of the households who process low value/trash fish will be better informed about potential improvements in processing practices, value-added product development opportunities, and market opportunities.

<u>Research Design & Activity Plan</u>

Location of work: This study will be undertaken only in Cambodia as resources are limited and there is a priority need within this country for this information. Survey work will be undertaken in five provinces located along the Mekong River, Bassac River, Tonle Sap River and Tonle Sap Great Lake. These provinces include Kampong Cham, Prey Veng, Kandal, Kampong Chhnang and Battambang.

Methods: The study will be comprised of five activities:
1. Literature review and desktop survey to better understand fish processing in Cambodia and markets for processed products.

2 Field survey using a questionnaire of households in the five selected provinces to study processing practices of low value/trash fish and to identify problems and issues related to food safety, processing, value-added product development, and markets. A sample of 50 households which process low value/trash fish in each of the five provinces will be undertaken. The role of women in processing and market activities will be highlighted.

3. Key informant interviews of fish traders at both wholesale and retail levels in the five provinces will be undertaken to identify market channels and markets for processed low value/trash fish. The fish traders will be asked about new opportunities for both value added products and markets locally and in the region.

4. Compilation of data, data analysis, and report preparation.

5. Results of the study in different formats for different audiences will be undertaken. The result formats will include research report, fact sheets, radio interviews, and newspaper articles. The publications will be translated into the local Khmer language.

Schedule

This study is planned to be implemented as below:

Activity	Beginning	Ending
Literature review	04/2007	09/2007
Survey	11/2007	05/2008
Key informant interviews	05/2008	11/2008
Data analysis	01/2009	06/2009
Report preparation and validation	06/2009	09/2009

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TOPIC AREA
TECHNOLOGY ADOPTION & POLICY DEVELOPMENT

❖

FEED TECHNOLOGY ADOPTION AND POLICY DEVELOPMENT FOR FISHERIES MANAGEMENT

Technology Adoption & Policy Development/Activity/07TAP01UC

Collaborating Institutions & Lead Investigators

University of Connecticut-Avery Point (USA)	Tessa Getchis
Inland Fisheries Research & Development Institute (Cambodia)	Prum Somany
Can Tho University (Vietnam)	Le Xuan Sinh

Objectives

The overall objective of this activity is to develop appropriate feed technology and provide technology related information to end-users of aquaculture and aquatic resources to change their use patterns. To achieve the overall objective of the investigation, the following are the specific objectives:

1. To apply the research results, develop, and disseminate appropriate technology to the end-users of aquatic resources and aquaculture practitioners.
2. To train farmers in the project sites on farm made feeds, feed and feeding strategy and benefits of using alternative feed technology.
3. To improve feeding practices and promote adoption and change behavior over alternative feeds.
4. To provide scientific-based strategy for integrating aquaculture with other water uses to improve wellbeing of the rural poor farmers.
5. To provide sufficient information for policy makers to develop policy on aquaculture and aquatic resource management.

Significance

The long-term vision of the Government of Cambodia is to create a cohesive and advanced country, free from the grip of poverty and illiteracy. The long-term strategy to achieve this vision is the Government's Triangle and Rectangular Strategy. The Government's poverty reduction goals are envisioned in the Triangle/Rectangular Strategy, the medium-term Third Five-Year Socioeconomic Development Plan 2006-2010 (SEDPIII): National Economic Growth and Poverty Reduction Strategy, and the National Poverty Reduction Strategy 2003-2005 (NPRS). SEDPIII and NPRS focus on three national development objectives in the context of broader governance reform and poverty reduction strategies:

- Economic growth that is broad enough to include sectors from which the poor derive a livelihood
- Social and cultural development
- Sustainable use of natural resources and sound environmental management

The fisheries sector plays an important role in the food security and the national economy of the country and therefore contributes significantly to the national development objectives. The vision of the fisheries sector as described in the Fisheries Development Action Plan 2005 – 2008 (FDAP,

November 18, 2004) is that "Ensuring the supply of fish and fishery products will keep pace with increasing demands to safeguard the nutritional standards, and the social and economic well-being of communities depending on fisheries for their livelihoods". The goal of the fisheries sector as described in the FDAP is to maximize the contribution of fisheries to the achievement of national development objectives, especially those related to improving rural livelihoods of the poor, enhancing food security and the sustainable development and equitable use of the fisheries resource base.
Cambodia's freshwater capture fisheries are among the largest and most significant in the world. The catch is conservatively estimated between 250,000 and 400,000 tons per year. The freshwater capture fisheries production contributes over 75% of the total fish production. On the other hand, Aquaculture production is only 34,000 tons per year which is relatively low if compared to Vietnam and Thailand.

The Fisheries Administration (FiA) in Cambodia has envisaged a future in which fisheries and aquaculture activities contribute significantly to the social and economic well-being of the people, in particular rural communities, where fisheries is as much a way of life as it is a source of nutrition and income. The FiA has set priority areas in order to achieve the goal as follows:

- Reviewing, revising and improving the policy, plans, legislation, institution and capacity (human and physical) of the fisheries sector.
- Increasing more areas for family fishing through Fishing Lot reforms.
- Stock enhancement through conservation and research study.
- Community based fisheries management to promote local participation in fisheries management linked to livelihood diversification.
- Improving livelihood of poor rural people by enhancing the role of fish in food security, employment and income generation by ensuring the sustainable use of aquatic resources, and by increasing community and household production levels through rural aquaculture development.
- Improving livelihood of poor people by improving their capacity to more effectively use of fish after capture through enhanced post-harvest fisheries development.

This proposed investigation aligns with the short-term and long-term actions of the Fisheries Development Action Plan 2005–2008 (FDAP, November 18, 2004) which (1) focuses on improving livelihood of rural poor people through stock enhancement and conservation of fisheries resources (3rd priority area); (2) enhancing the role of fish in food security, employment, and income generation by ensuring the sustainable use of aquatic resources and by increasing community and household production levels through rural aquaculture development (5th priority area), and (3) improving their capacity to more effectively use fish after capture through enhanced post-harvest fisheries development by disseminating widely and educating people in areas on understanding the importance of fisheries resources, especially, appropriate aquaculture, and fish-made feed technologies. Moreover, this investigation will lead to the establishment of effective linkages between researchers and communicators. The research results will be applied to develop appropriate technologies to disseminate information and to provide awareness and better understanding of the importance of low value fish, feed made technology and feeding practices to the fish farmers, which will significantly reduce the high pressure on capture low value/trash fish for feed and feeding in aquaculture activities. In addition, this investigation will provide functional frameworks and methods to integrate aquaculture with other water uses for local income generation, and to implement the National Aquaculture Development Strategy.

Since 2000 in Vietnam, the importance of aquaculture extension has been recognized due to its role in the economic restructuring in agriculture and rural development. There are various institutions providing different types of extension activities to aquaculture farmers. The most important extension services now are offered by the Provincial Extension Centers and the input suppliers (companies or wholesalers) and processors/exporters. Training on farming techniques and fish feed and health management in association with the provision of reading materials are preferred by the farmers (Sinh 2005). Aquaculture extension programs need to be improved in the man-power, facilities, methods and types of information, as well as the cooperation among the related stakeholders, in order to provide more and better extension services for aquaculture development (Sinh 2005).

Quantified Anticipated Benefits

This investigation will provide direct and indirect benefit to different stakeholder groups:

- One graduate student and one undergraduate Cambodian student will be involved in this project through the thesis research.
- 500 fish farmers in the lower Mekong basin of Cambodia and Vietnam will be informed and trained on farm made feeds, feeds and feeding strategies and the benefits of using alternative feed technology
- 3000 fish consumers, including women and children, in the lower Mekong basin will have indirect benefit indirectly from more fish and lower fish prices due to the increased fish production through appropriate information, fish feed technology, and management of fish feeding practices.
- 400 women, children, and elders, who are often involved in making fish feeds and fish feeding practices, will be informed and trained in the benefits of using alternative feed technology and these methods.
- 50 aquaculture extension specialists will be trained to be trainers and disseminate relevant information and appropriate technologies to 20,000 of fish farmers (aquaculture and aquatic resource users).

Activity Plan

Location of Work: There will be three (3) sites in Cambodia and two (2) sites in Vietnam. In Cambodia the sites will be in Kandal, Kampong Chnang and Siem Reap provinces. In Vietnam, the sites will be in An Giang and Can Tho provinces.

Methods: Since this is an activity, the methods will be in the form of workshops, conference organization, outreach, transformation and dissemination of information, and training sessions for farmers on farm-made feeds, feed, and feeding strategies; benefits of using alternative feed technology; improving feeding practices; and promoting adoption and change over to alternative feeds.

1. Review all related documents and technologies to be disseminated.
2. Conduct Participatory Rural Communication Appraisal (PRCA) and Institutional and Training Needs Assessment.
3. Conduct trainings, training of trainers, workshops, and awareness raising activities.
4. Hands-On Training and Farmers Field School.
5. Produce printed media materials: posters, leaflets, flyers, billboards, and training modules.
6. Produce mass media: video training documentary

Schedule

Phase 1 (6 months) April 1 to September 30, 2007: Development of Organizational and Operational Framework.

1. Organize and activate Investigation Team.
2. Select exact sites for targeted technology adoption and interventions.
3. Make a preliminary assessment of aquaculture and aquatic resources problems at project sites.
4. Conduct Inception Workshop
5. Make a preliminary communication analysis mainly from information gathered from field observation and from secondary data.

6. Prepare detail project work plan and organize a briefing seminar to present work plan and create a collaborative framework among the project participants in both countries Cambodia and Vietnam.
7. Conduct local consultations for start up of field activities.

Phase 2: (18 months) October 1, 2007 to March 30, 2009: Design and Implementation of Information/Communication Plan for Technology Adoption and Policy Development.
1. Review relevant and appropriate technologies to be disseminated.
2. Conduct Participatory Rural Communication Appraisal (PRCA) to assess information, learning, and communication needs of fish farmers and identify specific knowledge-attitude-behavior to be addressed.
3. Design local Information/Communication Plan (Message design, multimedia strategy, targeted training activities).
4. Produce prototype multimedia messages according to local Information/Communication Plan.
5. Pre-test prototype with samples of intended audience group.
6. Prepare final versions of multimedia.
7. Implement local Information/Communication Plan in project sites.
8. Set up networking and linkages for information dissemination and reporting system in project sites.
9. Assess Information/Communication needs and prepare training program for Project Team members.
10. Conduct Training of Trainer for lead fish farmers (Approximately 20 users in each pilot site will be trained).
11. Conduct Hands-on Training and Farmer field schools
12. Conduct Workshops to provide awareness on Appropriate Fish Feed Technology and Good Fish Feeding Practices and Management.

Phase 3: (6 months) April 01, 2009 to September 30, 2009: Monitoring, Evaluation, and Impact Assessment of Technology Adoption and Policy Development and Finalize Investigation.
1. Prepare methodology for comparative attitude-knowledge-practice indicators for monitoring and evaluation of Technology Adoption and Policy Development through Information/Communication interventions.
2. Conduct training workshop on Information/Communication Monitoring and Evaluation (Impact Assessment Method and Techniques for Project Team members)
3. Prepare Impact Assessment report based on the results of monitoring and evaluation
4. Conduct seminar to review impact assessment, discuss outcomes and formulate recommendation to adapt, improve and scale out the application of Technology Adoption and Policy Development through the use of Information/Communication interventions.
5. Prepare Aquaculture Development Policy
6. Prepare final report and organize workshop for Investigation termination.
7. Evaluation studies using surveys of users of the new feed technologies will be undertaken. This will evaluate, among other indicators, reduction in the use of low value/trash fish as feed and adoption of recommended feeding practices. Since many of the impacts of the project, especially on stocks of low value/trash fish, will not occur until after the project life, longer term evaluation will be required to be undertaken by the project team members.

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INTERNET-BASED EXTENSION PODCASTS FOR TILAPIA FARMERS IN THE PHILIPPINES

Technology Adoption & Policy Development/Activity/07TAP02NC

Collaborating Institutions & Lead Investigators

North Carolina State University (USA) — Russell Borski
U.S. Department of Commerce, Milford CT (USA) — Christopher Brown
Central Luzon State University (Philippines) — Remedios Bolivar

Objectives

1. Summarize tilapia-related publications, conference plans, events and news into a series of short (~5 minute), internet-friendly broadcasts.
2. Conduct a workshop and provide this *Tilapia Cast* to the computing center at CLSU on a trial basis, for access to and feedback from appropriate user groups (aquaculture farmers, students, and faculty). This workshop will also serve as a classical outreach activity for providing farmers with the latest information and training on best management practices for pond-cultured tilapia.
3. Refine and broaden this into a series of podcasts and to promote access to this information among aquatic farmers in the Philippines and other users worldwide.

Significance

Podcasting is an internet-based communication method that is increasing sharply in popularity. Contrary to a popular misunderstanding, the use of podcasts is not restricted to owners or users of iPods, and neither an iPod nor other MP3 players is actually necessary. Podcasts are information broadcasts that can be retrieved and played using any computer with ordinary internet access. With a podcast, freshly updated sound and/or images and video can be distributed economically to internet users worldwide, and the use of this means of communication is very much on the rise. To our knowledge, the potential for podcasting for the benefit of aquaculture farmers has not yet been explored.

In the course of our studies, we have generated considerable technical information of practical utility to farmers in the Philippines. For the most part these have been feeding parameters and strategies that enable farmers to reduce production costs without any negative impact on productivity (for example see Brown et al., 2004). We have also developed a molecular method that provides a rapid assessment of growth rate (Vera Cruz et al., 2006) that has the potential to increase the pace of methods testing and practical technical advances that could help farmers. Our approach to extension work has involved active interfaces with farmers on a variety of different levels. We have produced and distributed pamphlets, held training sessions and hosted numerous workshops at the Freshwater Aquaculture Center at Central Luzon State University (CLSU). We have also found that the active participation of farmers in experimental field trials (Bolivar et al. 2006) assists dramatically in getting the news out, particularly when excellent yields and lowered production costs are part of the equation; farming methods established in this manner have been adopted quickly and broadly. In short, we find tilapia farmers in Luzon, the Republic of the Philippines to be accepting and appreciative of extension work from CLSU.

Our extension activities have also included the establishment of an internet-connected computing center, which has drawn the interest of both students and farmers. This center is on campus at the Freshwater Aquaculture Center, which is visited on a regular basis by aquaculture farmers in that region. Use of this center by students, farmers, and faculty has been heavy. Internet access is growing in the Philippines with about nine percent of citizens currently wired for access. This rate is increasing rapidly, and it is supplemented by the appearance of numerous campus computing centers and internet cafes to which farmers have access. Moreover, most Filipinos use wireless phone technology and many use MP3 players where podcasts can be readily uploaded and viewed anywhere. Our proposal in this investigation is to establish a specialized, internet-based delivery system for news and technical developments of interest to tilapia farmers. Our short-term goal is to develop and assimilate tilapia-related news into a series — possibly quarterly — of current announcements, in an objective way that will keep farmers up to date. In the longer term, we plan to deliver these announcements as serial "Podcasts" to which internet users can subscribe at no cost.

<u>Quantified Anticipated Benefits</u>

Farmers in Luzon Island, the Philippines have been the beneficiaries of work from this research group in terms of the development and application of innovative technologies. New feeding strategies and stocking schedules have reduced the effort and cost required to produce tilapia for local consumption, thereby increasing profitability (for further information, see Bolivar et al., 2004; Brown et al., 2004). The proposed investigation is an outgrowth of these successful studies, and one that emphasizes a new application of internet technology to the distribution of practical information to benefit farmers.

An advantage of establishing a Tilapia Production Podcast now, as the popularity of this medium is in the process of being established and while the technology is still under development, is that podcasting can emerge as a thoroughly trusted and convenient source of the most reliable information for farmers. Demand within the United States and worldwide for tilapia products continues to increase (see Hatch and Hanson, 2000; Fitzsimmons, 2002) and we believe that neither interest in tilapia nor internet access will be declining soon. One reasonable expectation is that coming groups of younger farmers will rely increasingly on this sort of media, and a pattern of established credibility could boost the future utility and popularity of tilapia podcasts.

A simple but reliable source of the latest information will have immediate utility in the Philippines, and in terms of both operating costs and practical utility it will prove to be far superior, for example, to the editing and distribution of printed extension pamphlets. The podcast approach is far thriftier, more easily updated, and more efficient than the distribution of printed media. It has been our experience that printed pamphlets are often out-of-date before the considerable invoice for printing them has been processed. Podcasts are not only more economical and easily updated than printed media, they are more far-reaching and vastly less consumptive of natural resources than virtually any other available method of distribution of extension information.

A workshop involving the participation of 20-30 tilapia producers will be incorporated into this project to provide information on the newest methods for improving tilapia production and for introducing the podcast.

The research will incorporate educational and training experiences of two undergraduate students.

<u>Activity Plan</u>

Location: Luzon Island, Republic of the Philippines, will be the initial site targeted for internet-based distribution of informative tilapia broadcasts.

Methods: The most currently available practical information relating to tilapia farming will be gathered by reviewing journals and other publications, monitoring current and upcoming scientific meetings, and by communicating on a regular basis with appropriate academic and professional groups. Among others, these will include Sea Grant offices with an emphasis on tilapia work, appropriate government agencies, universities with relevant aquaculture programs, professional groups, the World Aquaculture Society, participants in the AquaFish CRSP and other stakeholder groups. The three project investigators will collaboratively summarize and distill the information gathered through this international effort.

Initially, we will produce a digitally-recorded (verbal) summary of current events and activities of interest to tilapia farmers. These announcements will include reviews of new publications (e.g., Lim and Webster, 2006; El Sayed 2006), and briefs on the outcome of the World Aquaculture Society (Feb/March 2007, San Antonio Texas) and the Asian Pacific Aquaculture (August 2007, Hanoi) meetings. We will then deliver it by a link on an appropriate "Index" or cover page at the Freshwater Aquaculture Center computing complex at CLSU. We will conduct a workshop at CLSU to both provide classical outreach activities and to help launch the tilapia podcast. During this workshop we will disseminate recent tilapia research activities and findings and demonstrate podcasting to farmers, faculty and students with at least two short podcasts. We will monitor patterns of access and number of hits on the podcast site by farmers and other users. We will also solicit a rating, reactions and suggestions for the podcast from appropriate user-groups. This will be accomplished using a brief questionnaire and "reviewer comment" field built into the Central Luzon State University Aquaculture Computing site in order to poll farmers that visit the CLSU facility. This will include collection of data characterizing the demographics of podcast users. This monitoring will allow an evaluation of the overall effectiveness of the podcast(s), its general usefulness, the identification of groups who might most exploit its use as an extension tool and other assessment measures. This information will be used to improve future access and utility of the podcasts and to establish the suitability of podcasting as an outreach tool to tilapia farmers. Along with the podcast, the workshop will discuss many features of tilapia aquaculture including water quality, pond management, alternative feed strategies, hatchery production, and genetics and breeding as examples. A brochure will be produced to disseminate methods to reduce production costs, e.g. alternative feeding strategies.

Following the initial phases of the project we will expand on the initial trial-based distribution of announcements to initiate periodically updated podcasts. Although the frequency of updates will be determined by the availability of new information or meetings (WAS 2008), updates during this period will include summaries of this and other events/publications.

Podcast content will be expanded to use visual images in the form of photographs and short digital video clips. The geographic target audience will also be expanded to include tilapia farmers and interested academic individuals beyond the Philippines. By the end of the 2.5 years of this project, podcast content will be updated at least quarterly. The availability of tilapia podcasts will be announced at appropriate meetings and through appropriate media for worldwide access.

Schedule

April – September 2007	Collect tilapia information and produce digital audio recordings.
November 2007 – April 2008	Conduct workshop at CLSU, and link podcast to FAC computing center. Expand on announcements and update podcasts.
June 2008 – November 2008	Update and expand podcasts with visual images and short digital video clips for a broader geographic target audience.

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AQUAFISH CRSP SPONSORSHIP OF THE EIGHTH INTERNATIONAL SYMPOSIUM ON TILAPIA IN AQUACULTURE TO BE HELD IN EGYPT

Technology Adoption & Policy Development/Activity/07TAP03UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)	Kevin Fitzsimmons
Universidad Autónoma de Tamaulipas (Mexico)	Pablo Gonzalez Alanis
Central Laboratory for Aquaculture Research (Egypt)	Ahmed Said Diab
American University of Beirut (Lebanon)	Imad Saoud

Objectives

1. Provide travel support for Dr. Gonzalez and four international contributors from other AquaFish CRSP countries to attend ISTA 8.
2. Employee a UAT student to assist with the compilation of papers submitted to the ISTA 8 conference proceedings.
3. Publish and print the Proceedings of the 8th ISTA.
4. Establish an ISTA website for on-line submission of abstracts and papers and eventual e-archiving of paper and presentations in PDF formats.

Significance

The ISTA meetings traditionally were held every four years and are the premier international meeting focused directly on tilapia aquaculture. However, after the Manila meetings the demand for more information and number of other locations vying to host additional conferences led us to decide to hold the future ISTA's within two years. The ISTA's have provided one of the most important outlets for publication and discussion of the findings of Aquaculture CRSP supported research. Aquaculture CRSP has been a co-sponsor of the last four ISTA's (Fitzsimmons 1997; Fitzsimmons and Carvalho 2000; Bolivar, Mair and Fitzsimmons 2004; Contreras and Fitzsimmons 2006). World-Fish Centre and the Department of Aquaculture Central Research Lab will be co-hosting ISTA 8 and will be especially interested to ensure the success of the conference. The original Pond/Dynamics CRSP contributed greatly to capacity building of the aquaculture and especially tilapia production in Egypt. Today, Egypt is the world's second largest producer of tilapia, with production of 445,000 mt in 2005. Much of this success can be attributed to earlier CRSP activities and the ISTA 8 will highlight past achievements that led to current success.

Travel support is critical to the ability of scientists from developing countries to present their findings in international forums. Host country CRSP scientists benefit from the opportunity to discuss their work amongst themselves, with their US colleagues as well as the rest of the international community. The efforts within this proposal fit the Activity description of conference organisation.

Quantified Anticipated Results

We will provide direct financial support for five host country scientists to participate in the ISTA 8. We anticipate that several more of the various A&F CRSP host country and US scientists and students will also participate. Our larger target group is the international community devoted to tilapia aquaculture. We have increased ISTA participation by about 150 persons each over the last four symposia. We expect that the meeting in Egypt could reach 1000 participants. The project would also provide partial financial support for a Mexican graduate student to assist with conference planning.

As at the past ISTA's the sponsorship will publish a large body of information. In this case generated by the new AquaFish CRSP project. This will be an ideal forum to demonstrate the scope of effort within the new A&F CRSP. It is also important for us to establish relationships between tilapia research community and African and Arab farmers

Activity Plan

Location of work: UAT and Egypt

Methods: Drs. Fitzsimmons, Gonzalez, Diab and Saoud are on the international organising committee planning the ISTA 8 symposium. A sub-committee will be formed to set the selection criteria and then determine which applicant scientists will be awarded the travel support. The selection criteria will be based on contributions of papers to the conference, past participation in AquaFish CRSP projects and other available support. If partial support can be generated from other sources, the funds may be split to support additional participation.

A graduate student from UAT will receive assistance to assist with the conference organisation. Specifically, the student will work with the publication committee working on the proceedings.

The local organizing committee has been formed and includes:
Honorary Chair Dr. Fadia Nosir; Central Administration of Agricultural Foreign Relations
Dr. Ahmed Said Diab; Director of CLAR and Egyptian coordinator of the conference.
Representative of Ministry of Agriculture and Land Reclamation.
Representative of Worldfish Center.
Representative of Egyptian Society of Agribusiness
Representative of American Soybean Association
Representative of the United Animal Feed Producers.
Representative of United Cooperative of Fishermen.
Representative of Academy of Scientific Research and Egyptian Universities.

Schedule

June 2007	Local organizing committee meets in Egypt, First call for abstracts
October 2007	Select printer
October 2007	Begin to organise sessions
November 2007	Select exact venue and conference hotels
December 2007	Design and put up ISTA 8 website
January 2008	Begin to receive abstracts
March 2008	Begin to receive and edit papers
May 2008	Select and invite scientists who will receive travel support
June 2008	Determine travel plans for those receiving travel support
June 2008	Provide travel stipend for airfare, registration, and other travel costs
October 2008 2nd week	ISTA 8
Late October 2008	Submit final report

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TOPIC AREA
MARKETING, ECONOMIC RISK ASSESSMENT & TRADE

❖

COMPETITION AND IMPACTS BETWEEN USE OF LOW VALUE/TRASH FISH FOR AQUACULTURE FEED VERSUS USE FOR HUMAN FOOD

Marketing, Economic Risk Assessment & Trade/Study/ 07MER01UC

Collaborating Institutions & Lead Investigators

University of Connecticut-Avery Point (USA)	Robert Pomeroy
Can Tho University (Vietnam)	Le Xuan Sinh
Inland Fisheries Research & Development Institute (Cambodia)	Hap Navy

Objectives

1. Describe and analyze the status and trend of supply and demand of low value/trash fish, and to assess the impacts of utilization of low value/trash fish in aquaculture on food security and livelihoods of households in the lower Mekong basin of Cambodia and Vietnam.
2. Support development of a policy framework addressing aquaculture/capture fisheries interactions, the sustainable exploitation and use of low value/trash fish in the lower Mekong basin, and human food security issues.

Significance

Low value/trash fish is defined as fish that has a low commercial value by virtue of their low quality, small size or low consumer preference. The uses of low value/trash fish are diverse and include: (1) local consumption (e.g., fresh, dried); (2) direct feed (e.g., livestock, high value species aquaculture); (3) fish meal/oil production (e.g., for poultry, aquaculture); and (4) value-added products (e.g., fish sauce, surimi, protein concentrates). The issues related to low value/trash fish uses are underpinned by the rapid development of the aquaculture industry and the increasing demand for fish by consumers, and competition between these two uses. Given the strong interdependency between capture fisheries for low value/trash fish and aquaculture, knowledge of and information on these two sub-sectors cannot be carried out in isolation of each other.

There is in general a lack of accurate information on the supply and demand for low value/trash fish for use in freshwater aquaculture in Cambodia and Vietnam. In Vietnam, for example, no data are available on the proportion of low value/trash fish used for feed production/direct feed for aquaculture and livestock and for the production of fish sauce (FAO-APFIC 2005). In the Asia-Pacific region as a whole, a conservative estimate of 25% for livestock and aquaculture feed has been put forward (FAO-APFIC 2005). Data and information on low value/trash fish production (species, size) and uses are patchy and often of poor quality. The exact portion of fish meal going to aquaculture compared with that going to livestock feed is also unknown, although it is known that the share for aquaculture is increasing.

Over the last decade, the price of low value/trash fish has risen considerably and it is predicted to increase over the next few years due to increased demand for fish meal and fish oil to meet market demands for aquaculture of carnivorous fish (and well as a source of affordable food). At the local level, prices of low value/trash fish vary depending on species, seasons and abundance of other fish and fishery products. Prices also fluctuate with the demand for fish meal in the livestock and aquaculture industry and the availability of raw materials for fish meal production. Given that aquaculture is predicted to grow while capture fisheries remain stable, it will become increasingly more difficult to meet the demand for low value/trash fish.

There is an increasing conflict between the use of low value/trash fish for animals/fish and for human consumption. Supplies of low value/trash fish are finite, and as indicated by a recent increase in price, demand is outstripping supply. It has been argued that it would be more efficient and ethical to divert more of the limited supply to human food, using value-added products, etc. Proponents of this suggest that using low value/trash fish as food for poor domestic consumers is more appropriate than supplying fish meal plants for an export income oriented aquaculture industry, producing high value commodities. On the other hand, food security can also be increased by improving the income generation abilities of poor people, and it can be argued that the large number of people employed in both fishing and aquaculture has this beneficial effect, via income generation, rather than direct food supply.

In general, the disposition of low value/trash fish is market driven and dependent upon local economic mechanisms. Without external interventions (such as incentives and subsidies) it will be the economics of the different uses of low value/trash fish in different localities that will divert the fish one way or the other. For example, in Vietnam, as the national demand for fish sauce is predicted to double over the next 10 years, there appears to be direct competition for mixed low value/trash fish between *Pangasius* feeds and those needed to make low-cost fish sauce. Traditional small-scale pig rearing uses low value/trash fish but large-scale pig farming uses agro-industrial formulated feed containing fish meal.

Low value/trash fish is important to the community and aquaculture, as well as the environment in Cambodia and Vietnam. There is a need to better understand the supply and demand for low value/trash fish in order to support the development of a policy and management framework to address aquaculture and capture fisheries interactions.

<u>Quantified Anticipated Benefits</u>

A total of five students/staff members will be trained and gain experience by participating directly in this study (two Cambodians and three Vietnamese). This helps to strengthen the man-power for both Cambodian and Vietnamese institutions.

- 500 fish farmers in four provinces in Cambodia and four provinces in Vietnam will be informed and have a better understanding of the supply and demand of low value/trash fish and will be able to make more informed production decisions
- 50 government officials, extension agents, researchers, and aquaculture industry stakeholders (marketing, input suppliers) at national, provincial and local government levels will be informed and have a better understanding of the supply and demand of low value/trash fish and will be able to make more informed policy and business decisions concerning use and management of low value/trash fish, including trade-offs between aquaculture and human consumption.
- One student will be supported for their master degree in agricultural and natural resource economics and three undergraduate students (one Cambodian and two Vietnamese).
- 300 scientists, researchers, resource managers, government officials, non-government organizations, and inter-governmental organizations concerned with and working on the issue of low value/ trash fish in the Asia-Pacific region will be better informed and have alternatives for fishery interventions and improved utilization to implement an action plan to address the issues.

<u>Research Design & Activity Plan</u>

Location of work: This study is planned to be implemented in both Cambodia and Vietnam, particularly in selected provinces of the lower Mekong basin where freshwater aquaculture is practiced. These are areas where both pond and cage aquaculture is undertaken. The availability and role of low value/trash fish in freshwater areas may differ by the depth of annual floods in the region. Survey work will be undertaken in four provinces of Cambodia located along the Mekong River, Bassac River and Tonle Sap Lake. These provinces include Kompong Cham, Prey Veng, Kandal, and Siem Reap. In Vietnam, An Giang and Dong Thap provinces are considered to be deep flooding areas, and Can Tho and Hau Giang provinces are considered to be shallow flooding areas.

Methods: The study will be conducted using the following methods. All methods will supply information to assess the growth of aquaculture and the low value/trash fish fishery; timelines; identify the uses of low value/trash fish for feed and human consumption and livelihoods; estimate future growth of aquaculture and the implications for low value/trash fish resources; and document demands, conflicts and impacts of uses of low value/trash fish.

The use of any social science research method, whether survey, semi-structured interview, focus group interview, oral histories, or visualization technique, will depend on the willingness of individuals to provide information to the questions. Social science research methods have been developed to address this issue through various guiding principles of field data collection. This includes respecting the respondents and stakeholders; clarifying the objective of data collection; having an interactive approach; recognizing informant bias; cross-checking data; maintaining anonymity; and using a range of different field data collection methods. In addition, this issue can be addressed through the sampling approach utilized. The dependability of survey findings is affected by the sampling plan and the faithfulness with which it is carried out, and also by procedures used. (Selltiz et al. 1976; Bunce et al. 2000).

1. Literature review: A literature review of secondary data will be undertaken to identify information related to the parameters and sub-parameters on aquaculture, low value/trash fish, and food consumption. This information will be useful during the reconnaissance survey and while planning field data collection. The review will involve compiling all existing information on aquaculture development in the lower Mekong basin of Cambodia and Vietnam to understand feed and feeding practices; use of low value/trash fish in agriculture/aquaculture and for human consumption; sources and supply of low value/trash fish.

2. Rapid Rural Appraisal (RRA): A reconnaissance survey, which is a brief survey of the study areas, will be undertaken using the RRA methodology to provide information to the project team and to help in planning for field data collection. The RRA will help to finalize the selection of study sites, collect preliminary information on stakeholders and aquaculture, identify logistical requirements, and refine study objectives and parameters. A RRA will be conducted in all eight provinces identified above in Cambodia and Vietnam. The role and participation of women and poor or minority groups will be emphasized.

3. Semi-structured interviews: Semi-structured interviews are based on a set of open-ended questions or discussion points to generate qualitative information. This method allows for two-way interaction and exchange of information between the facilitator and the informant. Semi-structured interviews will be utilized to gather information on aquaculture and fisheries practices from resource managers, government officials, extension agents, researchers, and aquaculture industry stakeholders (marketing and input suppliers) at national, provincial and local government levels in Cambodia and Vietnam. Information on aquaculture production technologies, species, feeding practices, sources of feed, current status of low value/trash fish, markets, trade, and issues/problems relating to use of low value/trash fish.

4. Survey: Surveys use questionnaires with highly structured, close-ended questions. The questionnaire will have specific questions with limited answers which result in quantitative data that can be analyzed statistically. (1) A survey of a statistically representative sample of pond and cage fish farmers will be undertaken to better understand the utilization of low value/trash fish.

Questions to be asked will include culture system; stocking density; fish species; feed and feeding practices; feed conversion ratio; use of alternative feeds; feed composition; feed cost; source of feed, use, price and source of low value/trash fish; production/yield; trends in supply and demand; role and participation of women; current status of low value/trash fish; and issues/problems relating to use of low value/trash fish. (2) A second survey of a statistically representative sample of households will be undertaken to better understand food consumption patterns of low value/trash fish. Questions to be asked will include uses, sources, price, importance in diet/nutrition, frequency of use, trends in supply and demand, current status of low value/trash fish; role and participation of women; and issues/problems relating to use of low value/trash fish. (3) A third survey of a statistically representative sample of low value/trash fish fishers will be undertaken to better understand supply and demand issues. Questions to be asked will include fishing methods, location of fishing, years fishing, seasonality of fishing, species, catch rates, markets and marketing, prices, role and participation of women, current status of low value/trash fish; and issues/problems relating to use of low value/trash fish.

5. Data analysis: All data will be compiled, reviewed, prepared, and stored in both a Microsoft Access and Excel format for further analysis. The quantitative data will be analyzed using statistical methods. The analysis of quantitative data will be carefully compared with the data collected from the other sources to check for any discrepancies.

6. Report: Final reports will be prepared. Key learnings will be finalized and policy implications and recommendations prepared. The findings will be validated by presenting them to the stakeholders and informants for comment. Validation will take place through small discussion groups and presentations to specific groups of stakeholders/informants.

7. Dissemination: Results of the study in different formats for different audiences will be undertaken. The results formats will include research report, policy reports, fact sheets, comics, radio interviews, newspaper articles, and scientific journal articles. The publications will be translated into local languages (Khmer and Vietnamese).

Schedule

This study is planned to be implemented as below:

Activity	Beginning	Ending
Literature review	04/2007	08/2007
Rapid Rural Appraisal and reconnaissance	08/2007	01/2008
Semi-structured interviews	02/2008	05/2008
Survey	06/2008	11/2008
Data analysis	01/2009	06/2009
Report preparation and validation	06/2009	09/2009

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DEVELOPING SUPPLY CHAIN AND GROUP MARKETING SYSTEMS FOR FISH FARMERS IN GHANA AND KENYA

Marketing, Economic Risk Assessment & Trade/Study & Activity/07MER02PU

Collaborating Institutions & Lead Investigators

Purdue University (USA)	Kwamena Quagrainie Jennifer Dennis
Kwame Nkrumah University of Science & Technology (Ghana)	Stephen Amisah Paul Sarfo-Mensah
Moi University (Kenya)	Charles Ngugi John Makambo

Objectives

The overall objective is supply chain development and establishment of mechanisms that enhance the management and skills of fish farmers to access and become integrated into urban fish markets in order to realize significant profitability and potential credit. The development of these mechanisms will allow both buyers of farm-raised fish products and small- and medium- scale fish farmers to act to minimize their joint transactions costs. The specific objectives will be:

1. Develop an aquaculture supply chain framework to create value for farm-raised fish chain in Ghana and Kenya.
2. Train small- and medium-scale fish farmers in supply chain management, risk analyses, costs analyses, pricing strategies, quality and cost effectiveness in post-harvest value chain.
3. Build synergies between fish producers and fish vendors for improvements in product and service delivery through cost effective supply-chain coordination.
4. Equip farmers with skills for group marketing, developing new markets, developing distribution and market structures and networks, and identify value-added opportunities for tilapia and catfish.

Significance

Production technology in aquaculture has tremendously improved in Ghana and Kenya resulting from the developmental efforts of NGOs including Aquaculture CRSP but the fish farmers lack the knowledge and skills in supply chain issues and marketing. The growth in aquaculture in these two nations can be sustained if fish farmers engage in a marketing system that involves a chain of links between producers and consumers, including all mechanisms, flows, interchanges, services and operators (Johnson and Hofman, 2004). This provides tremendous earnings opportunities for fish farmers. Flows through a well-functioning farm-raised fish marketing system will include information on prices, market situation, trends, consumer preferences as well as flows of money, and credit. By developing this chain, there will be large potential benefits for aquaculture development, employment, higher income for fish producers, potential microfinancing by fish vendors and middlemen (FAO, 2002). The extent of the potential benefits depends to a large degree upon the efficiency and design of the system. Leyva (2004) demonstrated that a thorough understanding of urban markets and coordinated production and marketing efforts are key factors to profitability of fish farmers in Nicaragua and Honduras. Leyva (2004) found that profitability resulted from primary sales to restaurants, with supplemental sales to supermarkets in relative proximity to the target restaurants. Fish farmers in Ghana and Kenya sell fish at pond banks and in the immediate local market and lack the knowledge and skills required to access lucrative urban markets.

The lifestyles of many sub-Saharan African urban dwellers are increasingly patterned after the West resulting in increased eating out in restaurants and catering operations, which has positively affected demand for fish products. This trend offers a great opportunity for fish farmers to benefit from the growing urban fish market by supplying tilapia and catfish to meet the growing demand for fish. In particular, group marketing could provide consistent supply of tilapia and catfish to meet demand, gain some negotiating power from the seafood trade, spread marketing risks and costs among participants; and have enough volume to add value through processing if that becomes necessary. Economic theory suggests that the development of supply chain mechanisms and minimization of transaction costs are important determinants of supply chain success. Boger, Hobbs and Kerr (2001) found that transaction cost-reducing mechanisms and relationships in Polish pork production were critical aspects of the success of the industry. Consequently, training in supply management will provide fish farmers a better understanding of opportunities and constraints in the fish/seafood value chain, and the effects on competitiveness, demand, and linkage to specific markets. The current fish market in urban Ghanaian and Kenyan markets deals mainly in whole dressed tilapia and catfish. Tapping into this growing fish market will allow fish farmers to grow from subsistence/small-scale operations into commercial moneymaking aquaculture operations and become competitive and an integral part of the seafood supply chain.

Quantified Anticipated Benefits

Supply chain management and marketing are key factors to the development and sustainability of commercial small- and medium-scale aquaculture operations in Ghana and Kenya when local and global issues relating to fish farming are considered. It is anticipated that this activity will develop an aquaculture supply management to enable rural fish farmers access urban markets. Supply-chain coordination would also help to place fish vendors and farmers in a better position to collectively engage in other economic responsible initiatives. This will result in higher incomes for fish producers, potential micro-financing from fish vendors and middlemen, aquaculture development, and rural employment. By demonstrating close cooperation through a common coordination platform will help consolidate and facilitate efficient aquaculture trade in Ghana and Kenya. It is also anticipated that the activity will result in the formation of group marketing activities in major fish producing regions in Kenya and Ghana. Accomplishment of these could result in tilapia and catfish sales values and farmers' revenues increasing by about 300%. For example, a farmer having pond-bank sales value of $100 can easily increase sales value to $400 through a managed supply chain and access to urban seafood markets.

Research Design & Activity Plan

1. Design a questionnaire to elicit physical, financial and social information for a range of typical fish businesses along a chain (e.g., Martin and Jagadish, 2005).
2. Survey a sample of middlemen, fish vendors, restaurants and small- and medium-scale fish farmers to gain further information on a range of market segments and businesses servicing these segments. The survey questionnaire will first be pre-tested to assess its suitability to respondents, verify if respondents understood the questions, eliminate ambiguous questions, and solicit suggestions from respondents. Data collected will be validated and assessed for reliability using a test-retest procedure, i.e., a sample of respondents who have already responded to the questionnaire will be randomly selected and visited again to answer subset of the questions. A comparison of the two responses will then be made using chi-square analysis to examine if there is any significant difference between their previous and current responses to the same questions.
3. Analyze the fish supply chain data in the areas of logistical efficiency, supply chain relationship management, and competitiveness. Questionnaire for farmers will elicit information on species of fish sold, frequency of sales, annual sales, kinds of marketing channels used including middle men, prices obtained per channel, marketing costs associated with channels, fish cleaning methods, fish grading standards, use of ice, types of sales arrangements - open market, contracts, marketing alliances or joint ventures, other logistics associated with marketing, and farm financial data such as costs of farm inputs, location, age,

years of education, gender, farm size, and number of years growing fish. The questionnaire for fish vendors and restaurants will elicit information on species of fish bought, preference for farmed fish versus wild-capture fish, preferred fish sizes, preferred fish quality attributes, volume of fish sales, wholesale and retail prices, marketing costs, transportation modes and costs, fish grading standards, use of ice, frequency of purchase, types of purchase arrangements - spot market, contracts, marketing alliances or joint ventures, and other logistics associated with marketing.

4. Organize workshops for fish farmers, harvesting crew, fish haulers, middlemen, fish vendors, restaurants and individuals involved in the middles industries, on supply chain management, pricing strategies, quality and cost effectiveness in post-harvest value chain.
5. Organize farmers into “study” groups for case-study assignments with middlemen, fish traders, restaurants, and other retail outlets to provide hands-on (practical) experience in value chain management for tilapia and catfish.
6. Develop a brochure for producers presenting the main opportunities and constraints of the voluntary participation in group marketing programs and describing the guidelines for such a program.
7. Develop extension manual on supply-chain case studies illustrating costs and benefits of partnerships, especially economic returns to farmers.

Impact Indicators:
- Number of urban markets developed.
- Number of farmers who have developed direct linkage with urban fish vendors
- Number of brochures developed on “How to Market Your Fish to Urban Retailers.”
- How much increase in fish sales with supply chain management

<u>Schedule</u>

Start date: June 1, 2007
End date: September 30, 2009

June 2007 – August 2007
Develop survey instruments for assembling physical, financial and social information in existing fish marketing chains, marketing mechanisms, structures and agents, credit facilities, preferences, demand and price trends.

September 2007 – March 2008
Interview a sample of middlemen, fish vendors, restaurants, and other urban fish vendors as well as fish farmers; and assemble fish pricing and marketing cost data in rural and urban fish markets.

June 2008
Organize workshop for fish farmers on supply chain management, pricing strategies, quality and cost effectiveness in post harvest value chain; feasibility of formulating group marketing arrangements; and organize farmers into “study” groups for case-study assignments.

July 2008 – June 2009
Use a supply-chain framework to analyze data and assess fish marketing operations, integration processes, logistics and quality control, information flows, vertical integration and relationship management.

July 2008 – September 2009
Develop a brochure for producers on group fish marketing; and an extension manual on farm-raised fish supply-chain case studies.

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ON FARM VERIFICATION OF TILAPIA-CATFISH PREDATION CULTURE

Marketing Economics, Risk Assessment & Trade/Study/07MER03PU

Collaborating Institutions & Lead Investigators

University of Arkansas at Pine Bluff (USA)	Carole R. Engle Rebecca Lochmann
Sokoine University of Agriculture (Tanzania)	Sebastian Chenyambuga Kajitanus O. Osewe

Objectives

1. Validate the research-extension based recommendations on Nile tilapia-African catfish predation culture under small-scale farmers' conditions.
2. Access economic profitability of Nile-tilapia culture under small-scale farmers' condition.
3. Develop a management protocol and identify potential policy options and extension. support for Nile tilapia-catfish culture under small-scale fish farmers' conditions.
4. Train farmers and extension agents on manual separation of sexes at the fingerling stage
5. Support a graduate student at the M.SC level.

Significance

Verification trials are demonstration of the implementation of research-based Extension recommendations in the actual farming environment. It is a methodology used to verify and to determine if the total set of research-based Extension recommendations applied on farmers fields produces yields, feed conversions ratios, survival, and costs consistent with results from onstation research trials. Moreover, it is clear that to produce Nile tilapia in ponds as an economically feasible enterprise there are two basic options: 1) mixed sex or 2) culture of allmale (or predominantly male) populations. The first option is usually limited by uncontrolled reproduction in ponds.

It has been shown in experiments and limited farm trials that the problem of overpopulation can be overcome in three ways. First, fish population can be maintained by using appropriate management practices, which enhances the growth of fingerlings to attain a commercially acceptable size in a short period of time. For this to be successful, the pond should be completely drained after each cropping. It should be left to dry, so as to destroy any fish left. The stocking of the pond should be done with healthy fingerlings of one age class, and intensive feeding should be done with a suitable protein-rich diet. Due to the lack of stable fingerling supply system, this option is not practical under Tanzania's conditions. The selection of strains with fast growth rate and late maturity would obviously contribute greatly to the success of this system. Second, a predator that feeds on fry and fingerlings is introduced in the pond to control the population. Predator species such as African Catfish (Clarias gariepinus) have been used for this purpose in several countries (FAO, 1980). Third, all-male culture can resolve the problem of uncontrolled reproduction in rearing ponds. Since the males have higher growth rates than the females, production of larger fish and higher yields are possible when all-male stocks are grown. All-male tilapia populations can be produced by hand sexing of the fingerlings, crossing of some tilapia species and hormonal sex reversal. Culture of hybrids has not yet been adopted for farming to any appreciable extent in Africa. There are several problems associated with production of hybrids such as maintaining the purity of broodstocks and the limited fecundity of parent fish, which restricts fry production. Though reported to be successful in Asian countries, hormonal sex reversal has yet to be tried on large-scale farming operations in Tanzania. Since the larvae have to be given specially prepared artificial feeds containing the hormone for about six weeks and excluding all natural food in the fingerlings diets, sustainability under small-scale aquaculture may be any issue.

Moreover, in Tanzania, recent studies show that profit margins for mixed-sex culture with catfish predation were high enough to support the economic sustainability of the Nile tilapia culture system

(Kaliba et al, 2006). This was strengthened by the fact that switching from a culture system of no predation-to-predation adds only a minimal increase in operating cost. Mixed-sex culture with catfish predation was recommended to be a middle stage for developing a mono-sex Nile tilapia production system, which is superior to both mixed-sex and catfish-predation cultures. Switching from mixed-sex to catfish predation will allow development of skills and a Research-Extension system that can support mono-sex Nile tilapia culture in Tanzania.

Quantified Anticipated Benefits

Small-scale farmers do not keep production or marketing records. The comprehensive quality of the data to be collected throughout the verification program will provide more accurate estimates of survival, yield, and feed conversion ratios than data obtained from farm surveys. This will allow accurate analysis of Nile tilapia-catfish enterprises under small-scale production system and also provide a basis for quantifying risks involved in small-scale fish farming and for impact assessment. This will also improve and refine Extension recommendations and management protocols for Nile tilapia culture with catfish predation under small-scale farmers' conditions. A Swahili manual on production and hand sexing of Nile tilapia fingerling will be produced for both training and Extension education purposes. In addition, the study would create awareness on the importance and benefits of monosex Nile tilapia culture in Tanzania. At the same time, it would develop the necessary expertise required in hand sexing of Nile tilapia fingerling an important stage towards monosex culture.

Research Design & Activity Plan

A masters student enrolled in Aquaculture Sciences at the Sokoine University Agriculture, Department of Animal Science and Production will be supported for thesis research, research results dissemination and publication. A student will be selected based on their academic qualifications, interest and relevance. The selection procedure is as explained earlier on. Mr. Kajitanus and Drs Chenyambuga and Kaliba will guide the selected student to achieve project's objective and fulfill the requirement of the Department of Animal Science and Production. In addition, students at Sokoine University of Agriculture enrolled in Aquaculture Science in both undergraduate and graduate program, extension agents and small-scale fish farmers in the Morogoro Region, will be trained on production, rearing and hand-sexing of Nile tilapia and catfish fingerlings. Host country PIs and Co-PIs have the required expertise in both hand-sexing of Nile tilapia and production of catfish fingerlings.

The on-farm trial will use six ponds owned by small-scale farmers in Mikindu Village, in the Morogoro Region. The ponds will be selected based on willingness to participate and size of the pond, which will be at least 150m 2. This is the average pond size for most farmers in the Morogoro Region. The ponds will be harvested, drained and all fish removed from the pond. Pond fertilization will be according to farmer's practices. Four ponds will be stocked with fingerlings that have been hand-sexed using family labor and weighing at least 20 g at a rate of 22 fingerlings per 10 m 2 and with two African catfish per fingerlings per 10 m2 and weighing at least 30 g. Two ponds will act as a control (i.e., stocked with mixed-sex culture as practiced by farmers). The farmer will feed the fish using locally available feed resources. The feeding rate will be adjusted to account for the presence of catfish in ponds. Sampling will be twice a month to monitor growth and for adjusting the feeding rate. Measurement of water quality parameters will be done fortnightly and the analysis will be as described in Boyd and Tucker (1992). The experiment will cover two periods of 300 rearing days each. At the end of the first rearing period, data will be collected on the number and weight of marketable fish harvested, overall survival from each of the tilapia-catfish yield verification ponds and the control experiment. In the second rearing period, data will be corrected at 150 and 300 rearing days. These parameters will be used to calculate growth rate of fish, annual marketed yield, overall net yield, and average weight of fish sold and feed conversion ratio (FCR). Performance between the two treatments will be compared using a 2 x 2 contingency table and 2x test.

Under small-scale agriculture most technologies are developed to reduce the likelihood of yield or financial loss. For many small-scale farmers the margin of production over basic needs is so small that even partial failure can result in serious deprivation. The need to reduce risk is reflected both in farmer's traditional practices and in their response to the promotion of new technology packages (Collinson l982). There is strong evidence that technology packages, which increase farmer's exposure to risk, are unlikely to be adopted on a wide scale. There are several ways in which the risk level of alternative treatments can be compared as reviewed in the CIMMYT manual (CIMMYT 1988). These include: determining probability of success and failure, multiple regression analysis and farmer participation in risk evaluation and. Probability analysis is used to indicate whether or not an important number of farmers might lose by adopting the technology. It estimates the probabilities with which a farmer would achieve different levels of net benefits (including negative net benefits). This is to indicate whether or not an important number of farmers might lose by adopting the new technology. Multiple regression analysis is a statistical technique, which can be used to the effect of each environmental and/or management factor while controlling for all the others. For each variable included in the regression model the estimated magnitude of the effect can be compared to its variance in order to determine whether it is statistically significant. Important insights into ways of reducing risk are more often achieved when quantitative analysis has been preceded by effective farmer participation. Willing neighboring farmers will participate in hand sexing, stocking, and harvesting of the ponds selected for the on-farm trials. This is to allow farmer to assess and determine which treatment justifies the investment.

In this study, all three methods will be used in analyzing the performance of Nile tilapia-catfish culture under small-scale farmers conditions. During the experiment, economic parameters to be estimated will be on direct expenses (i.e., expenses that require cash outlays during the operation), fixed expenses (i.e., expenses representing the cost of owning and using the ponds and equipment), and price of different marketable fish sizes. Economic analysis will be conducted to determine total expenses, net returns per area, profit margins, and breakeven prices. Simulation will be conducted to assess and compare risk associated with switching from mixed-sex culture to tilapia-catfish predation in terms of yield variability, income stabilization and probability of enterprise failure between mixed-sex culture without predation and had-sexed tilapia culture with catfish predation using dominance analysis.

Significance & Method: Internal human capacity building and leadership are needed support all-male Nile tilapia culture in Tanzania. The training of trainers is a technique used to build the skills of extension agents, innovators, and other stakeholders so that they can confidently and effectively train others. This will be a two days training workshop conducted in collaboration with SUA and Kingolwira National Fish Center. First, both a Swahili and English manual on hand sexing of Nile tilapia fingerlings will be developed. A practical course on hand-sexing Nile tilapia fingerlings will be introduced in the general aquaculture sciences class. In a two-year period, at least thirst thirty small-scale farmers from the Morogoro Region and twenty aquaculture extension agents from Morogoro, Iringa, Pwani and Dodoma will be trained on the technique. Farmer selection will focus on innovators and women who do most the management activities. Extension agents will come from those areas with high concentration of Nile tilapia fish farming.

Impact Indicators

- New information will be obtained on feasibility of Nile tilapia-catfish polyculture under small-scale farmers condition.
- New information will be generated on the potential of Nile tilapia-catfish polyculture on improving income of small-scale fish farmers.
- Number of aquaculturists trained to hand-sexing Nile tilapia culture in the Morogoro region increase from 0 to more than 50.
- Availability of the materials needed for the training of other stakeholders on hand sexing of Nile tilapia culture and production of catfish fingerlings.

- The 20% of small-scale farmers in the Morogoro Region adopt Nile tilapia-catfish predation culture that incorporate hand sexing in the production system. The 20% is based on the adoption curve or early adopter hypothesis as summarized in Feder, Just and Zilberman (1985).

Schedule

Time	Activities
August–September 2007	-Selection of students -Purchase of materials -Selection of farmers
October 2008-July 2008	-First Experiment
June 2008-March 2009	-Second Experiment
April-May 2009	-Data analysis and report writing
June-September 2009	-Thesis defense -Research results dissemination -Training of farmers -Final reports

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IMPLICATIONS OF EXPORT MARKET OPPORTUNITIES FOR TILAPIA FARMING PRACTICES IN THE PHILIPPINES

Marketing, Economic Risk Assessment & Trade/Study/07MER04NC

Collaborating Institutions & Lead Investigators

North Carolina State University (USA)	Russell Borski Upton Hatch
Central Luzon State University (Philippines)	Remedios Bolivar Wilfred Jamandre

Objectives

The overall goal is to foster the development of a viable export market for tilapia. In this investigation we will conduct studies to:

1. Determine requirements for export opportunities for tilapia in the Philippines.
2. Assess implications of the export markets on production systems.
3. Provide recommendations for facilitating the development of an export market.

Significance

Tilapia have well-established local markets in the Philippines where smaller fish are produced and consumed. Our recent A CRSP investigation has shown the feasibility of growing larger tilapia for the fillet market using a two-stage grow-out system (Brown and Bolivar, 2007). The challenge that will be addressed in this proposal is the requirements to expand Philippine farmed tilapia into high volume, high quality fillet market for export and local supermarkets using larger harvest size (600-800 g) fish. These new markets should provide an excellent opportunity for income growth for aquaculture producers while maintaining their production systems for domestic consumption. Clearly, export markets will have different requirements in the form of species, volumes, product forms and other constraints that have the potential to have important implication for selecting the appropriate farming systems.

There are three principal questions that the proposed effort will address:

- What are the export requirements for tilapia?
- What are the implications for farming system?
- What government strategies are required to foster exports of tilapia?

Important secondary questions will be:

- Under what conditions can small fish farmers profitably participate in the export market?
- Are there strategic investments that can foster expansion of export markets and the ability of small farmers to participate?

Export requirements can be quite imposing, particularly when a new market is being targeted. Generally, these markets will involve many constraints on the appropriate production systems that will tend to make these new systems much more intensive. That is, the volumes, timing and quality requirements will imply a vastly more controlled growing environment. Stocking densities, feeding, water quality and targeted final size and form - virtually all management decisions - will require greater sophistication and financial resources. Although the latter typically will favor medium (6-15 ha farms) or larger (> 15 ha) better-financed operations, small farmer participation is possible.

The ability of smaller farmers (1-5 ha farms) to take advantage of this export market opportunity is a challenge that the Government of the Philippines (GOP) must decide whether it is willing to invest the necessary public funds or grant special concessions to foster. Unfettered market forces tend to lead to production concentration in large economic entities that exclude small producers. Without government intervention, small farmer involvement is most likely to occur through a vertically integrated contract relationship with marketing firms. This type of arrangement would be quite similar to poultry production in many parts of the world and will be evaluated in the proposed study.

Export market requirements

Product size. Export markets are likely to require larger fish size than domestic markets. Two stages of culture – fingerling and grow out – are likely to be needed to reach a size of 600–800 grams. Local Tilapia consumption in many parts of the world consist of small fish in the 100 – 200 gram range whereas export markets will generally target a much larger fish that will be filleted. Dress out losses when filleting will require a fish of approximately 600–800 grams to obtain the appropriate sized fillet (Brown and Bolivar, 2007). There is heterogeneity in export markets resulting in requirements that are not the same for all countries and market niches. Thus, the decisions on exactly what export markets to target will be an important point to address e.g., Japanese, Korean, American, and European markets have major differences and even sub-markets within each country can have important differences.

Seasonality and market windows. The seasonality of export markets can also impose constraints. Targeting a particular market window can prove lucrative, if such windows provide exceptional price premiums. Market windows typically occur based on either production constraints during certain times of year—temperature in northern latitudes and dry seasons in the tropics—or exceptionally high demand, usually related to Easter, New years or other cultural holidays. This seasonality is often exacerbated by the common fact that such seasonalities are being experienced by both the consumer and producer; thus, price premiums persist based on strong economic realities. In addition, market windows can close very quickly, meaning that there may be a very tight time frame that is being targeted and competitors may be attempting a similar strategy.

Volume and product form. The minimum volumes required to meet needs of international markets generally far exceed that of local markets. Not only are volumes higher but these levels often are needed throughout the year. Obvious implications are scale issues (see below) and need to stagger harvests. Scale issues will imply that the volumes of product will most likely require a considerable acreage and the ability to provide a consistent volume every month. These large volumes will require either large producers or well-coordinate small and medium sized producers. If it is a policy of the GOP to foster the involvement of smaller producers there will most likely need to be a concerted effort in establishing cooperatives.

Product form. The alternative targeted product forms - frozen, fillet, live, bulk or individual - will considerably constrain the set of appropriate technologies. Each of these will imply differences in harvested size and quality in terms of time form pond to consumer. Also, and very importantly, will be the location of processing. Fish processing for export markets will foster the development of enterprises that will have further income and employment benefits for the country. In many cases the distance form the pond to the processing facility will not be substantial and specialized vehicles will be needed.

Destinations. Both the length and cost of transport will greatly affect the competitive position of Philippine tilapia exports. In addition, cultural traditions are often important in terms of the product form and time period associated with selected destinations. As mentioned above, cultural events are often times of high fish consumption.

Implications for production systems
Scale issues. The size of the operations sufficient to supply the consistent volumes and quality for export markets. Sufficient acreages in proximity to processing and hatchery operations will be necessary. Supply of skilled farm managers may also be a constraint. The coordination and management associated with export production will far exceed that needed for local consumption largely due to need to stagger harvest to obtain the ended consistent flow of product that meets specifications. For small producers to be involved in such export markets, it is highly likely that some form of cooperatives or vertical integration will be essential.

Farm management. Input intensity will likely increase with the growth in export markets. Increased stocking densities and feeding rates will be needed to meet volume and harvest size requirements. These higher stocking densities and feeding rates will in turn increase need for improved water quality management, particularly aeration and exchange. Power for pumping and aeration will substantially increase costs and equally important economic risk. Increased length of growing cycle will further exacerbate an already high risk farming system. This increased risk is likely to be a further detriment to small farmer participation in this new opportunity.

Strategies to foster growth of export markets
Cooperatives/Vertically Integrated Systems. Facilitation of the formation of cooperatives with the assistance of extension service is an important alternative in increasing small farmer chances of establishing themselves in export markets. Cooperatives could assist with marketing, financial services and processing availability. Because some cooperative based arrangements are unpredictable or have not succeeded, vertically integrated systems are becoming more common worldwide. Hence it is possible that a combination of both cooperatives and vertical integration might arise.

Subsidized loans. Provision of low or no interest loans for operating or investment is a commonly used method of creating incentives for new endeavors. Unfortunately, the history of repayment of subsidized loans to small farmers is not good. Any government programs of this type will need to be prepared the financial impact of high default rates.

Storage, processing and refrigeration. Infrastructure to cover the fish coming out of pond into the product desired at final destination will be an investment that GOP will likely need to foster in some fashion.

Streamline customs. Competitive position will be greatly supported by streamlining customs delay and product quality control. However, export market customers are typically more demanding in terms of quality requirements. The reputation of exports from particular countries can be greatly damaged by publicized incidents of contaminated products.

<u>Quantified Anticipated Benefits</u>

- The work will provide research training and educational experiences for two additional graduate students at Central Luzon State University in the Philippines and at North Carolina State University. Specifically these students will be trained on marketing and production economics.
- This investigation will produce a document with detailed information on production-marketing constraints and opportunities to expand tilapia culture into large volume, high quality, large-sized fillet export markets and local retail supermarkets. Of particular importance will be the relationship between these larger markets and local production.
- The work will foster collaboration among CLSU, small-scale farmers, and large-scale marketing firms and will facilitate improved opportunities for local producers.
- The proposed study will facilitate the development of export markets for tilapia that could improve farm incomes and expand tilapia farming.

The majority of the work will be undertaken in the Philippine tilapia production area in central Luzon. Assessment of potential for growing areas for export in other regions will determine need for travel outside the dominant growing areas in central Luzon. Also, there will be some effort needed in collecting export data and other seafood data in Manila along with interviews of selected experts on export markets.

Research Design & Activity Plan

1. Literature will be reviewed to gain understanding of the challenges of expanding local tilapia production into export markets (Zidack and Hatch, 1991). Of particular relevance will be the experience in Jamaica (Hatch and Hanson, 1991) and other countries (Medley et. al. 1994) that have recently been successful. This literature review will serve as an excellent starting point for the MS theses that will result from this project.

2. Secondary data analyses will be used to begin to characterize the details of the product specifications that will be imposed by export markets in several countries (Hatch and Kinnucan, 1993). That is, data from potential importers will be collected and analyzed to ascertain seasonality, product form, size and other important characteristics that will have implications for Philippine tilapia production systems.

3. Interviews with export brokers will determine the exact specifications of the export products. Several destinations will be targeted, including Korea, Japan, U.S. and Europe. Secondary data (from 2. above) will serve as a starting point in identifying appropriate export experts and other industry professionals for more detailed interviews and possible data obtainment.

4. A survey of tilapia production and processing will be conducted as a basis for data to be included in assessment of exports potential (Olowolayemo et al., 1992; Engle et al., 1988).

5. Existing research results and extension experience will be used to tailor production systems to individual export markets. CLSU researchers and extension professionals will assist in specifying production systems for each of the export markets specified above. Typically export markets will result in more intensive production systems (Hatch et al. 1998; Engle and Hatch, 1988; Hatch and Tai, 1996; Cacho et al., 1990)—ability of Philippine producers to move to these more skill and financially demanding systems will be important. To meet the needs to intensify production processes, multi-stage systems are often required (Agbayani et al., 1996: Hatch et al., 1996). Increased risk can be expected as systems intensify (Hatch et al., 1988). In some cases, these refinements will be minor changes in existing systems, whereas in other cases substantial changes may be required (Nerrie et al., 1990). If changes are substantial, the possibility of entering the selected markets will be greatly diminished in the short term and possibly even in the long term also. This interaction with production researchers and extension experts will be crucial to success in identifying realistic export opportunities for tilapia producers in the Philippines. The ability of small to intermediate producers to participate in this opportunity will be given special scrutiny (Hishamunda et al., 1998; Popma et al., 1995; Hatch et al., 1995).

6. Drs. Jamandre and Hatch will advise MS students in completion of project-related work and completion of their degrees.

Schedule

October 2007 – March 2008
Initiate economic study to analyze tilapia export market potential. Recruit MS student at CLSU. Complete literature review, secondary analysis and initial determination of export requirements to determine data collection needs and interviews to be accomplished during in-country visit and subsequent study.

April 2008 – September 2008
U.S. PI travels to Philippines, finalizes plan of study with HC PI and initiates interviews and surveys. Establish full collaboration with tilapia production researchers so as to facilitate ability to assess implications of export markets on production systems.
October 2008 – March 2009
Complete an initial draft of analysis and develop "Recommendations to Philippine Government" on potential actions that will assist in the development of the capability to compete in export markets.

April 2009 – September 2009
Complete data collection, data analysis and recommendations will be presented to appropriate Philippine officials. Final report will be submitted and MS theses completed.

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TOPIC AREA
WATERSHED & INTEGRATED COASTAL ZONE MANAGEMENT

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CHARACTERIZATION OF POND EFFLUENTS AND BIOLOGICAL AND PHYSICOCHEMICAL ASSESSMENT OF RECEIVING WATERS IN GHANA

Watershed& Integrated Coastal Zone Management/Study/07WIZ01PU

Collaborating Institutions & Lead Investigators

Virginia Polytechnic Institute & State University (USA) — Emmanuel Frimpong

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Objectives

1. Characterize effluent or potential effluent quality according to type of system.
2. Characterize receiving water quality in terms of nutrients, suspended solids, and pathogens.
3. Investigate the biological effects of ponds on receiving waters using structural and functional composition of fish and macroinvertebrate assemblages.
4. Develop best management practices for pond aquaculture in Ghana.

Significance

Aquaculture as an industry is unique in the sense of being a potential polluter of natural water bodies with effluents and a user of polluted water. It has been argued that owing to its tight linkage with natural ecosystems, aquaculture has the potential to have much more profound impact on [aquatic] environmental quality than terrestrial farming systems of similar size, and that to be sustainable, practitioners of aquaculture must place more emphasis on recognizing linkages and protecting the ecosystem which nurture and sustain the industry (Baired et al. 1996). The search for a balance between growth and intensification of aquaculture and protection of natural fisheries and the environment continues. Four main components of aquaculture waste water are of interest; nutrients (including nitrogen (N) and phosphorus (p)), biochemical oxygen demand (BOD), suspended solids, and pathogens (Cripps and Kelly 1996). Additions of nitrogen and phosphorus to natural waters can cause eutrophication. Therefore, these nutrients became an issue in the rule-making process for aquaculture effluents by the United States Environmental Protection Agency (USEPA) (Boyd and Queiroz 2001). High concentration of settleable solids may be generated by harvesting methods and ponds with heavy blooms of algae may discharge organic suspended solids in the form of plankton that will contribute to BOD. Natural organic fertilizers are commonly used in fish ponds. A test of a sample of 11 common feeds and four organic fertilizers used in Ghana indicated three of the feeds (biscuit waste, groundnut husk, and dried termite) and three of the organic fertilizers (cow manure, pig manure, and poultry manure) contained significant counts of fecal coliforms (Ampofo and Clerk 2003). Additionally, four out of 11 feeds (biscuit waste, cassava, groundnut husk, and termites) and all organic fertilizers (poultry manure, cow manure, pig manure, and cow blood) contained fecal streptococci (Ampofo and Clerk 2003). These and many pathogens may be passed on in effluents to receiving waters.

Environmental impacts of aquaculture on aquatic ecosystems are related to the species cultured, location of installations, intensity of operations, the morphology, limnology and hydrology, trophic status, and assimilative capacity of the receiving water (Costa-Pierce 1996; Cripps and Kelly 1996; Boyd and Queiroz 2001). Thus, for example, ponds located in relatively pristine watersheds are likely to alter receiving waters to a larger extent than those located in heavily agricultural watersheds. Consequently, the conservation concerns for these two systems would differ. Based on data synthesis, Costa-Pierce (1996) concluded that during normal operations of channel catfish ponds, total phosphorus (TP) releases are comparable to precipitation whereas during harvesting, mean TP discharges are comparable to concentration in runoff from intensive agriculture. The wide range of factors determining pollution potential of aquaculture necessitates a different way of categorizing aquaculture systems, even for those that culture the same or similar species with emphasis on management practices besides intensive, semi-intensive, and extensive. In Africa, it is already recognized that the strategies for addressing problems arising from small-scale and large-scale commercial aquaculture operations will probably be different (Jamu and Brummett 2004). However, it is not sufficient to argue that systems should be regulated differently simply because of size of operation. Differences between large-and small-scale systems can be specifically demonstrated by characterizing their effluents.

Without understanding of the differences among aquaculture systems, there is a tendency on the part of environmental advocacy groups and consequently decision makers to lump all systems together and exaggerate the impact of the industry. Such was the case after the publication of 'murky waters' by the Environmental Defense Fund and the subsequent decision by the USEPA to regulate virtually all aquaculture operations in the United States (Boyd and Tucker 2000). The growing aquaculture industry in Africa is bound to be confronted with competition for water and regulation of pollution. Characterization of effluents would allow for a proactive management of the environmental effects of aquaculture. Proactive action will forestall the restrictive regulations that could result from regulatory agencies acting on insufficient or even exaggerated assessments of the industry (Cripps and Kelly 1996; Muir 1996).

Pollution can limit the uses of water and aquatic resources, and water quality criteria have therefore been formulated on the bases of various uses (e.g., drinking water supply, agricultural use, bathing and amenity, and aquatic life). Quality criteria for aquatic life are generally considered as the most important and protective of the environment (Biney 1997). In recent years, quality criteria have been established by consolidating the view that an aquatic ecosystem in which structure and functions [ecological integrity] are not disrupted possesses a quality which is immediately suitable or suitable after simple treatment, for a variety of uses (Biney 1997). Such is a receiving water body whose assimilative capacity for pollutants has not been exceeded. It is now recognized that the aquatic biota themselves provide the most reliable signals of the effects of pollutant or habitat alteration, providing the basis for direct biological assessment and monitoring (Karr and Chu 1999). Biological monitoring is a feasible and low-cost alternative or complement to chemical measurements and toxicological bioassays that should be developed for resource-poor countries. Previous successes in application of biological monitoring have been documented from West Africa under the Onchocerciasis Control Program, where fish and benthic macroinvertebrates were used to monitor the effects of pesticides on aquatic communities of rivers (Leveque et al. 2003).

Some general characteristics of aquaculture effluents like relatively low concentrations of pollutants, large volume, and high flow rates, make conventional treatment options cost-prohibitive or even infeasible (Cripps and Kelly 1996). Best management practices (BMPs) are widely proposed as the alternative and these require a thorough understanding of not only the effluent quality but also operational characteristics like amount and frequency of discharges, whether discharges are released from top or bottom of ponds, through drainage ditches or directly into receiving waters, and weather there is discharge after every production cycle or water is reused. For example, most catfish ponds are drained twice in 15-20 years, implying much lower nutrient loading rates than would be assumed from feed conversion ratios (Boyd and Queiroz 2001). Likewise, Baitfish farmers in Arkansas, United States are increasingly reusing water to minimize the cost of pumping dwindling ground water, with a consequence of reducing effluents (Frimpong and Lochmann 2006). Among these farms, the use of pond drainage ditches vary widely as does effectiveness of these ditches in reducing effluent concentrations before they enter receiving waters (Frimpong et al. 2003; 2004). These observations make generalizations on an individual farm's impact difficult, implying that for effective development of BMPs, a thorough understanding of farms and their operational characteristics is required. With understanding of effluent quality, operational characteristics, and alternative uses of receiving waters, recommendations on feeding optimization can be made to curtail nutrient outputs. Pathogen counts in effluents will provide the basis for recommendation on storage, low-cost pretreatment like sun-drying, and the frequency and amount of application of manures. Water reuse and modification of harvesting drainage practices can be encouraged.

Quantified Anticipated Benefits

This study will result in scientific publications that clearly characterize the major practices in Ghana aquaculture, the variety and quality of receiving waters, and how the combination of practices and receiving waters separates systems by potential effluent impacts. Quantitative information on pond effluents will be available for at least 30 ponds grouped into 4-6 subsystems but the entire industry will be the beneficiary. The industry in Ghana will be regulation-ready by the end of 2009 with proactive BMPs developed for 4-6 distinct types of aquaculture to be published and disseminated to the fisheries department for extension by that time. Approximately 10 personnel from the fisheries department, the EPA, Water Company, and the HC university will be invited to participate in this study and be trained thereby (please see Outreach/Training). In addition, one graduate student, most likely a female, from the host country who will work on this project will be funded for 2 years to obtain an MS degree from the US through Virginia Tech. Opportunities exist for training at least one additional graduate student in the host country on this project. Ghana will have a country-specific protocol for fish-based biological assessment of aquatic ecosystems from which other countries in the region can learn. It will be applicable to all polluting industries so that the Ghana EPA will have a scientific basis for fair treatment of aquaculture. This study will also lay a firm foundation for planned future watershed studies (pending availability of funds) in partnership with the United Nations Food and Agriculture organization (FAO) that will develop GIS-based pond and practitioner databases for Ghana and other countries in the region. A spatially explicit inventory of ponds, including types and their effluent and receiving water characteristics will greatly enhance environmental management and the sustainability of aquaculture development in sub-Saharan Africa.

Research Design & Activity Plan

Location: This study will be conducted in Ghana, in the Ashanti and Brong Ahafo regions where most aquaculture is concentrated. It is expected that these two regions will provide the needed variety of aquaculture systems and practices and receiving water characteristics for a study that will be fairly generalizable to the rest of southern Ghana and similar systems in other sub-Saharan African countries.

Methods: Major steps of this study will include (a) pre-stratification of fish farms, (b) physicochemical sampling and analysis of ponds and receiving streams, (c) biotic sampling of receiving streams, (d) analysis of water-quality and biological data, and (e) development of BMPs.

Based on existing inventories, farms will be pre-stratified according to factors such as size of ponds, inputs (e.g., species, feeds, water source), operational methods (e.g., integration, frequency of discharge), and the nature of receiving waters and their watersheds (type, size and major uses besides aquaculture. A random sub-sample of at least five farms will be selected from each stratum in the two regions. Farmers will be contacted to explain objectives of the study and obtain their consent. Effluent draining events happen fortuitously. It is therefore necessary to maintain communication with farmers and schedule sampling around their plans. Anticipated draining events during the year will be noted. If sufficient draining events are not anticipated, more farmers in affected categories will be contacted as backup. It is also possible to do a vertically stratified sampling of water within ponds as a last resort if no draining occurs in the anticipated time. Accessibility of farms to quick transportation will be an important factor in farm selection because samples are subject to deterioration if analyses are delayed.

During draining, grab samples will be taken from pond outlet and about 50 m upstream and downstream of receiving stream. All sampling will be conducted by a graduate student assisted by a technician or undergraduate student. Samples will be transported to the lab for analysis of TN, TP, suspended and settleable solids, BOD, fecal coliforms, and fecal streptococci. Dissolved oxygen, pH, and temperature will be recorded in the field as ancillary data. To the extent feasible, a second sample will be taken during the draining of the last 5-10% of the pond as well to follow changes in effluent quality during draining and provide replicate samples from streams. One aspect of nutrient pollution (eutrophication) potential of effluents may be measured as the ratio of TN to TP. By comparing against a balanced (Redfield) ratio of 7.2 Costa-Pierce (1996) shows that some intensive aquaculture operations produce effluents that are enriched with phosphorus. Such indices will be computed following water-quality analysis. Differences among pond water and upstream and downstream of receiving stream will be compared statistically using Analysis of Variance (ANOVA).

Schedule Start date: July 1, 2007 End date: September 30, 2009 Whereas physicochemical measures provide indication of the transient effects of pond draining activities on water quality, the resident biota provide long-term records of any persistent effects of periodic changes in water quality. To investigate the biological effects of ponds on receiving waters, sites 50-100 m upstream and downstream of entire fish farms will be identified for fish and macroinvertebrate sampling. An additional stream segment close to designated study segments in the same watershed that is not influenced by aquaculture will be selected for each identified farm as a paired control (i.e., site-specific reference). Farms will not have to be those from which effluent is sampled. However, some overlap is expected. About ten farms, each with one reference stream, will be selected and sampled during the summer of 2008. The lead and host country PI and the graduate student supervised by the lead PI will be involved in sampling, sorting, and species identification. The most effective local gear will be used for sampling and sampling effort will be determined based on established bioassessment and monitoring protocols on North America and Europe. Taxanomic and functional composition of fish (e.g., percent omnivores and percent benthic species) and macroinvertebrate (e.g., *Ephemeroptera*, *Plecoptera*, and *Trichoptera* (EPT) and chironomid taxa richness) of assemblages will be calculated and compared between upstream-downstream and study and control streams using ANOVA and Analysis of Covariance (ANCOVA). The combined result of physicochemical water analysis and biological assessment will be used to develop BMPs. The study results, recommended BMPs and biomonitoring protocol will be published and submitted to all relevant institutions.

Schedule

Start date: July 1, 2007
End date: September 30, 2009

Activity/Month	July 07-May 08	June-July 08	August-December 08	January-September 09
Project Preparation	x			
Pond/Water Sampling		x	x	
Stream Biotic Sampling		x		
Analysis/BMPs/Reports			x	x

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DETERMINATION OF CARRYING CAPACITY OF THE BOCA CAMICHIN ESTUARY MEXICO, IN REFERENCE TO OYSTER CULTURE

Watershed & Integrated Coastal Zone Management/Experiment/07WIZ02UH

Collaborating Institutions & Lead Investigators

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Forestry and Natural Resources Management	William Steiner
Universidad Autónoma de Sinaloa, Mazatlán (Mexico)	Guillermo Rodriguez Domínguez
CIAD-Mazatlán (Mexico)	Omar Calvario Martinez

Objectives

This research will collect data to supplement previous collection of hydrographic and physio-chemical data to allow estimation of the carrying capacity for oyster culture to prevent over-expansion of the growing oyster culture industry and allow for planning in this important estuary in Mexico. Results will also allow identification of the most nutrient rich areas of the estuary where oyster farms could ideally be located and indicate possibly problematic areas that farmers should avoid. Findings will be integrated into an on-going effort to develop an integrated management plan for the area.

Significance

Boca Camichin an estuary in the larger wetlands complex of Marismas Nacionales (National Wetlands) which represents the largest and most important Mexican wetland in terms of biodiversity; it is also an important aquaculture and fisheries site. Boca Camichin is the primary site for the culture of the native oyster, *C. corteziensis*. Approximately 50 small-scale farmers now operate family-run oyster farms in this area and rapid growth is projected (Haws et. al 2006). There is wide spread concern that over-expansion or poor siting of oyster farms could lead to environmental impacts and/or affect oyster production if natural productivity of the estuary is over-exploited or if possible accumulations of oyster waste were to occur. Findings from this work will be used in aquaculture planning and for current efforts to develop a management plan for the wetlands, which includes guidelines for economic activities such as aquaculture.

Quantified Anticipated Benefits

This work will benefit small-scale oyster farmers in Boca Camichin, governmental and institutional decision makers such as state regulators and planners, the Committee for Conservation and Management of Santa Maria Bay, and community groups in Santa Maria Bay wishing to begin small-scale aquaculture.

Target groups for this work include: aquaculture extension workers and researchers in Pacific Mexico and Nicaragua; key private sector representatives; oyster growers in Nayarit, Sinaloa and Sonora; Women's oyster culture cooperatives of Nayarit; Women's oyster culture cooperative of Puerto Peñasco; Conservation International; Universidad Autónoma de Sinaloa (UAS, Culiacán and Mazatlán Campuses); Sinaloa Institute for Aquaculture (ISA); Ecocostas, an NGO dedicated to conservation and sustainable development for Latin America; Sinaloa State Committee for Aquatic Sanitation (CESASIN); and the Federation of Shrimp Cooperatives. Linkages will also be made to the NOAA International Sea Grant efforts through participation of Maria Haws and John Supan, Sea Grant personnel/associated faculty from Hawaii and Louisiana and Alaska. This work will inform and complement activities of the USAID coastal zone management projects, "Sustainable Coastal Communities and Ecosystems (SUCCESS)."

Quantifiable benefits will include: new culture methods developed; existing methods transferred to new user groups; increased skill levels for improving areas of health and sanitation related to aquaculture; improved knowledge of the linkages between the environment and health.

Metrics:
- Number of estuary management plans contributed to: 2
- Number of institutions directly or indirectly benefiting: 9
- Number of individual participants in technical training: 15
- Number of communities benefiting from findings: 26
- Number of documents produced or contributed to: 1
- Students involved: 1 graduate student

Research Design & Activity Plan

Extensive hydrological and physio-chemical data is available for the Boca Camichin estuary. This work will focus on sampling of seston and other nutrient sources in the estuary to determine concentrations and variations as measures of the availability of the primary food source for oysters and other bivalves. Other parameters to be monitored will include meteorological conditions, estuary and near-oceanic currents and other inputs such as effluents. Sampling will also be conducted in and around the oyster farming areas to detect possible depletion of nutrient levels by farm aggregations. There will be nine sampling stations for surface waters in the estuary system, drainage areas and two stations in the adjacent ocean area. Sampling will be conducted during the rainy and dry seasons in 2007-2008. At each station, temperature, salinity and dissolved oxygen will be measured *in situ*, with water samples being collected from depth for further laboratory analysis for nitrates, nitrites, ammonia, orthophosphate, total phosphorus and biological oxygen demand (BOD). Four samplings of waters in or near oyster culture areas will also be conducted and will be analyzed for dissolved organic material, particulate organic material, chlorophyll a, primary productivity, temperature, salinity, dissolved oxygen, and turbidity. Oyster growth rates and filtration rates will also be measured. A biogeochemical model developed in previous estuary monitoring efforts will be used estimate carrying capacity and to produce graphic representation of data for use in dissemination to stakeholders (Calvario and Dominguez 2006; Calvario et al. 2006). After data analysis and interpretation, findings will be presented to concerned stakeholders such as government regulators, researchers, oyster farmers and community members. This work will be carried out as a joint effort of researchers from CIAD and UAS, with support from the US PIs (Haws and Steiner). The UAS Masters degree student will assist with this work.

Schedule

Sampling will be conducted for two annual cycles beginning in May 2007 and terminating in July 2009. Data analysis and reporting will be completed by August 2009. Community members will be trained during each visit by researchers as each field visit includes a community meeting, short training event, and active participation by community members in all field research activities.

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TOPIC AREA
MITIGATING NEGATIVE ENVIRONMENTAL IMPACTS

❖

ASSESSMENT OF DIVERSITY AND BIOECOLOGICAL CHARACTERISTICS OF LOW VALUE/TRASH FISH SPECIES

Mitigating Negative Environmental Impacts/Study/07MNE01UC

Collaborating Institutions & Lead Investigators

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Inland Fisheries Research & Development Institute (Cambodia)	So Nam
Can Tho University (Vietnam)	Le Xuan Sinh

Objectives

The overall objective of this study is to investigate the environmental impacts of aquaculture development in the Lower Mekong Basin of Cambodia and Vietnam as the biodiversity "hotspot" areas, and to research potential negative impacts of aquaculture operations on wild fish species diversity and their stocks (i.e., populations). The specific objectives are as follows:

1. Assess the diversity of low value fish in order to characterize these low value fish into different species, genus and family.
2. Estimate the abundance and catches of low value fish in order to determine the current status and trends of low value fish in the total inland fish catch in Cambodia and Vietnam.
3. Assess the share of juvenile of commercially important fish species in low value fish in order to provide foundation for tailoring planning and management of inland fisheries in the Cambodia's and Vietnam's Lower Mekong Basin.
4. Make recommendations to protect the important fish juvenile areas in order to sustain their growth and survivals for human consumptions.
5. Analyze the current harvesting regime in order to assess its sustainability;
6. Study the ecological impacts of the use of low value fish as direct feed on wild fish populations in order to provide measures for mitigation.
7. Build the capacity of local fisheries staff and workers in order to collect biological and ecological data to help local communities to evaluate the importance of low value fish resources and identify negative impacts of farmed fish used these resources as direct feed on wild fish stocks.
8. Establish indicators on the status of low value fish in order to provide information for government fisheries managers.
9. Develop fisheries management strategies/actions for low value fish in order to mitigate negative impacts at national and subregional levels.

Significance

FAO expects that aquaculture will provide 50% of food fish supplies by the year 2030 (Tidwell and Allen 2001) and a large proportion of this production will origin from intensive aquaculture techniques, i.e., farming finfish and shrimps using high input of feed. Besides potential environmental degradation due to untreated effluents, habitat destruction, spreading of pathogens and non-indigenous species (Naylor et al. 2000; Chopin et al, 2001), the use of artificial fish based feeds or fresh fish resources will further increase pressure on wild fish stocks (FAO, 2005). In some cases such feeds comprise of fish species traditionally used as cheap food for people, especially in developing countries, and this allocation of fish resources by the aquaculture industry may therefore cause negative socio-economic effects. The problem with utilization of low valued fish resources in aquaculture has been identified by FAO as a key issue for sustainable aquaculture development. Many recent international symposiums and workshops arranged by FAO have focused on this increasing problem.

Fish is a staple and affordable food source for most people in Cambodia and Vietnam (Svrdrup-Jensen, 2002; So & Haing, 2006). Cambodia has an annual per capita consumption of 30-66 kg per person per year (So & Thuok, 1999; So & Haing, 2006) and Vietnam- Mekong delta 15-60 kg per capita per year (Svrdrup-Jensen, 2002). This comprises over 75 of total annual animal protein intake (Ahmed et al., 1998). Thousands of tonnes of low value fish are caught along their migration route, from Tonle Sap up the Mekong River to Khone Falls, and into tributaries. By estimation, at least 16,000 tonnes was caught by only one commercial type of fishing gear (bag net or dai) during 2004 (Hortle et al., 2005) and more than 30,000 tonnes during 2005 (DoF, 2006). Moreover, fishing in the river system is highly diverse, and over 150 fishing methods are employed (Hortle et al., 2005). No national statistical data of the production of total low value fish (including species, size and composition) are available.

The current reports on uses of freshwater low value/trash fish indicate that the majority of these species (62 species in Cambodia and 11 species in Vietnam) are already being used for fish feed. In Vietnam, increasingly more marine low value/trash fish are being used for freshwater aquaculture feed. There are reportedly few remaining low value/trash fish species that are not being utilized. In this investigation, an assessment of diversity and bioecological characteristics of low value/trash fish species will be undertaken. This study may allow for the identification of other low value species that could be promoted for human consumption, if stocks can support additional harvest rates.

Cambodia aquaculture represents about 10% of the total inland fisheries production (So & Haing, 2006), while the Mekong delta in Vietnam approximately 20% (Phillips, 2002). They have expanded, diversified and intensified; their contributions to aquatic food production have increased gradually and potentially. They are highly diverse and consist of a broad spectrum of systems, practices and operations, ranging from simple backyard small, household pond systems to large-scale, highly intensive, commercially oriented practices. In Cambodia, over 70% of freshwater aquaculture production come from cage culture (total: 4,492 fish cages) operated in Mekong basin, including the Tonle Sap Great Lake (43%), Tonle Sap River (17%), upper stretch of the Mekong River (19%), lower stretch of the Mekong River (14%) and Bassac River (7%) (So & Thuok, 1999). It is entirely dependent on wild fish both as seed and feed (So Nam et al. 2005). In Vietnam, about 4,639 fish cages are operated in four Mekong delta provinces, especially in An Giang and Dong Thap provinces, while about 17,000 ha of earthen ponds are used for fish culture there. The most commonly cultured fish species in the Lower Mekong Basin of Cambodia and Vietnam are snakehead (*Channa micropeltes*), pangasiid catfish (*Pangasianodon hypophthalmus*), hybrid clarias catfish (*C. btrachus* x *C. gariepinus*), and giant freshwater prawn (*Macrobrachium rosenbergii*).

Giant snakeheads are generally cultured in smaller cage of less than 200 m^3 in Cambodia. Feed represent more than 70% of the total operational cost and the main type of feed for giant snakehead culture is low valued fish, representing 60 to 100% of the total feed used depending on feeding strategies adopted by different farmers (So et al., 2005). During the dry season (October to May), the most important source of feed is low value freshwater fish, while more low value marine fish species are used during the rainy season (June to September) (So et al., 2005). Pangasiid catfish and hybrid clarias catfish are commonly cultured in earthen ponds in Cambodia. They are omnivorous and low value freshwater fish are used as direct feed at an average proportion of 20% for pangasiid catfish and 54% of total feed fed for hybrid clarias catfish (So et al., 2005). No or very little information relating to the use of low value fish for giant snakehead, pangasiid catfish and hybrid clariid catfish are available in the Mekong delta in Vietnam. Giant freshwater prawn is carnivorous and it may depend on low value fish used as direct feed. Such information is not available.

The government of Cambodia put a ban on snakehead farming in May 2005 and the reasons for this was the potential negative impacts on wild fish populations from wasteful snakehead seed collection and on other fish species diversity, and also potential negative effects on poor consumer groups from decreased availability of low valued fish. The ban seems, however, not to be effective as resources for implementation are lacking. The incentives for choosing snakehead before other fish species by fish farmers are strong, generating more than 10 times higher profits. Further more, no or very little information of biology and ecology of low value fish resources are available in the Lower Mekong River Basin of Cambodia and Vietnam.

This proposed investigation falls into the Aquaculture-Fisheries Nexus Topic Area of Mitigation Negative Environmental Impacts, which is one of the priorities described in the RFP, relating to "Research and Outreach Topic Areas: People, Livelihoods, and Ecosystem Interrelationships". Moreover, the investigation aligns with the short-term and long-term actions of the Fisheries Development Action Plan (FDAP- 2005-2008) of Cambodia (DoF, 2004), which focuses on improving livelihoods of rural poor people through "Conservation and Research Study to Enhance Fish Stocks" (3rd priority area).

<u>Quantified Anticipated Benefits</u>

The immediate or direct beneficiaries of this research will be:

- 400 scientists, researchers, government fisheries managers and policy markers, inter-government agencies, and non-government staff working on the issues of fish and fisheries, and diversity of low value fish, in the Lower Mekong River Basin of Cambodia and Vietnam, as well as other riparian countries in the Mekong Basin, will be better informed on the status of low value/trash fish stocks and be better able to develop and implement the actions to address the issues.
- 120 fisheries officers, researchers, and extension workers (80 in Cambodia and 40 in Vietnam) will be better informed of the knowledge of biology and ecology of low value fish, and their diversity and wild stocks (including size and species composition) and will be able to better inform the fisheries mangers and policy makers about these concerns, and ecological impacts of harvesting low value fish and juvenile of commercially important species on natural populations.
- One graduate student and two undergraduate students will be supported and trained through their thesis research.
- At least 550 fishers and fish farmers (400 in Cambodia and 150 in Vietnam) will have a better understanding of low value fish species diversity and composition (including size and catch composition), biological and ecological characteristics of these resources, and their stocks.
- 80 research scientists, extension agents, and government fisheries managers in Cambodia and Vietnam, as well as other riparian countries in the Mekong Basin, will have methodologies, individual and participatory stock and catch assessment tools, and taxonomic identification tools designed for application at a local level to assess the importance of local aquatic resources and their habitats.
- 300 fishers operating in fisheries where low value/trash fish are harvested will have individual and participatory stock and catch assessment tools and taxonomic identification tools designed for application at a local level to enable fishing communities to assess the importance of local aquatic resources and their habitats and to develop co-operative village, commune and district level management and conservation agreements.
- 10,000 indirect beneficiaries in Cambodia and Vietnam who rely on fish or low value fish resources for both food and income and for whom management and conservation of these resources are vital, will have great access to fish and lower price fish.

Research Design & Activity Plan

Location of work: The proposed country of work, sampling sites (province/water body) and sample size (number of experienced/knowledgeable fishers/farmers being interviewed in the Lower Mekong River Basin of Cambodia and Vietnam) are presented below.

No.	Country	Province	River	Sample size
1.	Cambodia			
		- Kampong Cham	Upper Mekong	50
		- Prey Veng	Lower Mekong	50
		- Kandal	Bassac	50
		- Kandal	Tonle Sap	50
		- Phnom Penh	Tonle Sap	50
		- Kampong Chnang	Tonle Sap/Lake	50
		- Battambang	Tonle Sap Lake	50
		- Siem Reap	Tonle Sap Lake	50
2.	Vietnam	- An Giang	Bassac/Mekong, Mekong delta	50
		- Dong Thap	Mekong, Mekong delta	50
		- Can Tho	Bassac, Mekong delta	50

Methods: This study will comprise four interrelated parts:
1. Literature review and desk survey (including questionnaire) work.

2. Detailed work plan for inception report preparation, and training and workshop.

3. Field survey: The field survey using a standard semi-open questionnaire which is composed of individual interviews with experienced and knowledgeable fishers and fish farmers and focus group discussions with key informants in locations identified above. The field survey will focus on study of the biological and ecological characteristics of low value fish, ecological impacts of farmed fish fed directly with these low value fish resources on wild fish stocks, and on others which are mentioned in the section (c) Objectives above. The methodology of the use of local knowledge for the study of river fish biology and ecology is presented in the report of Bao et al. (2001). Timetables will be used to obtain information about how the situation today compared with that of ten years ago and operations on what it is likely to look like 10 years from now.

4. Compilation of data, data analysis, and report writing.

Schedule

The duration of implementation of this proposed investigation will be 30 months, starting from 1 May 2007 till 30 October 2009. The schedule for the proposed work is presented in the following table.

Activity	Year 1 (1 April - 30 September 2007)				Year 2 (1 October 2007 – 30 September 2008)				Year 2 (1 October 2008 – 30 September 2009)			
Literature review			x	x		x		x			x	
Workplan and inception report preparation			x	x								
Information/inception report workshop				x								
Questionnaire preparation				x								
Pre-survey and field consultation				x								
Testing of questionnaire				x	x							
Training of local fisheries staff				x	x							
Full field survey					x	x	x	x				
Data compilation, entering and analysis								x	x	x	x	
Reporting writing				x		x		x			x	x
Stakeholder review workshop											x	
Recommended strategies/actions and policy briefs												x
Final national workshop												x

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TRAINING IN SUSTAINABLE COASTAL AQUACULTURE TECHNOLOGIES IN INDONESIA AND THE PHILIPPINES

Mitigating Negative Environmental Impacts/Activity/07MNE02NC

Collaborating Institutions & Lead Investigators

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Objectives

1. Conduct a series of short courses demonstrating alternatives to monoculture of shrimp in affected communities of Aceh Province of Indonesia and the Luzon (Batangas) and Visayas (Panay/Guimaras Island) regions of the Philippines.
2. Transfer techniques developed by researchers and extension specialists in the Philippines for seaweed, oyster, and tilapia-shrimp polyculture to Indonesian and Filipino aquatic farmers.
3. Provide oversight and review of matching funds provided by Aquaculture without Frontiers for training in sustainable coastal aquaculture.
4. Provide training to coastal farmers and to NGO volunteers who are providing assistance to survivors of the tsunami.
5. Determine if farmers receiving training adopt some of the techniques rather than returning to shrimp monoculture.
6. Determine if crop diversification and more sustainable farming techniques have improved household income.

Significance

The 9.0 magnitude earthquake that struck the Aceh Province of Indonesia impacted 30,000 households who had registered with aquaculture being the primary household livelihood. The vast majority of these families had small tambaks (ponds of less than one hectare) that were used for monoculture of penaeid shrimp. The survivors would like to return to aquaculture, but recognize that the removal of mangroves may have contributed to the scope of the disaster. Further, even before the tsunami they were impacted by degradation of water quality, diseases in shrimp, and low prices due to over-production. Similar problems have also occurred with shrimp monoculture in the Philippines.

Researchers at SEAFDEC AQD in the Philippines have pioneered mangrove friendly shrimp farming technologies and have been active in publication and dissemination of these methods (Primavera 2000; Tadokoro et al. 2000). These include use of mangroves, seaweeds and bivalves as biofilters. In an earlier Aquaculture CRSP, CLSU researchers worked with farmers to develop and document the tilapia – shrimp polyculture system in the Philippines. The University of Arizona has developed red algae (*Gracilaria*) farming techniques that are especially useful to integrate with a shrimp and/or tilapia production to remove nutrients from effluents (Nelson et al. 2001). Similar work in China (Yang et al. 2005), Colombia (Gautier 2002) and Thailand (Menasveta 2002) provides additional examples from several distinct environments.

A direct cash match of $41,000 will come from the Aquaculture without Frontiers (AwF) charity. (See letter attached). The donors to AwF requested that all donated funds be expended in Indonesia, but the organization has requested a partner to provide technical expertise for the restoration effort in conjunction with the Indonesian Department of Fisheries and several NGO's. The PI's will provide technical expertise and oversight for the expenditures of the AwF funds.

Changes to more sustainable farming should also translate into improved household income as additional products should be available for sale or consumption and costs for disease treatments and feed associated with monoculture of shrimp are reduced. A final survey will ask if the participants have improved household welfare by implementing the techniques taught in the workshops.

This study addresses issues in the adoption of aquaculture technology applicable to similar situations in other countries. It also addresses questions of coastal disaster rehabilitation and diversification of livelihoods. Through its links with the SEAFDEC AQD, the project is connected with a regional and global research and development network. In particular, the results of the training will be presented at the WAS meeting in Busan Korea, May 2008.

Quantified Anticipated Benefits

- The project will incorporate four workshops, two each in the Philippines and Indonesia that will involve the participation of 20-30 farmers per workshop. Training will benefit tsunami survivors, and other coastal aquaculture farmers throughout Indonesia and the Philippines, especially shrimp farmers who are looking for a more sustainable farming system. Participant lists will be collected at each training event in order to document individuals in attendance, including their association, gender, and whether they represent the primary household earner. Consultations with NGOs and community organizationswill ascertain any potential constraints that might prevent attendance at workshops, for instance travel. Potential constraints will be mitigated as appropriate.
- The researchers will benefit, as they will be able to synthesize their complementary knowledge and experiences to customize the sustainable coastal aquaculture system to the Indonesian and Filipino situations.
- Aquaculture without Frontiers and their donors will benefit as they need experts to oversee and direct the financial support.
- In addition to this direct benefit to coastal and tsunami affected communities, a longer-term and more global benefit is to provide information to further develop sustainable coastal aquaculture in tropical conditions.
- We expect to see that the training and implementation of sustainable coastal aquaculture techniques will improve household welfare.
- The activity will contribute to the training of three graduate students.

Activity Plan

Location: The training efforts will occur in three villages (Ladong, Pedie, Buran) in Aceh Province of Indonesia and at Panay/Guimaras Island and Batangas in the Philippines. A demonstration project, supported by the matching funds, will be in operation in Ladong, near the Ladong Fisheries College. Much of the training materials will be produced at the SEAFDEC AQD in the Philippines.

Methods: We will conduct a series of workshops to train participants in tilapia, tilapia-shrimp polyculture, and seaweed culture techniques. In Indonesia, the trainings will be conducted jointly in English by the project PI's and in Bahasa Indonesia by the professional staffs of the Ujong Batee Aquaculture Center and Ladong Fisheries College (Sugeng Raharjo, see attached letter). The work in the Philippines will be joint workshops with Drs. Bolivar (CLSU), Hurtado/Golez (SEAFDEC

AQD) and Lopez (DA-BFAR).

Some instructional materials will be purchased from SEAFDEC AQD and used in the Philippines and Aceh, Indonesia. Additional materials will be translated by Ujong Batee Aquaculture Center staff and photocopied or printed in Indonesia. Invitations and announcements will be distributed in the week prior to the workshops to each village in both the Philippines and Indonesia. A small box lunch will be provided to each of the participants, with a selection of foods appropriate per local traditions and customs.

A follow up visit and survey of the farmers will be conducted to determine how many of the farmers have implemented the training received. The baseline condition will be considered a return to monoculture of shrimp. Changes including culture of, or polyculture with tilapia, milkfish and/or seaweeds, or other sustainable improvements will be tabulated. Two questions on the survey will inquire if the household income has been increased, decreased or remained the same and second if the household welfare has been increased, decreased or remained the same. Data will be gender disaggregated.

Schedule

June 2007	Project start date, selection of training materials
July 2007	Translation of selected materials to Bahasa Indonesia
August 2007	Training workshops and pond demonstration in Indonesia
January 2008	Training workshop in Batangas province (Luzon) and Panay/Guimaras Island (Visayas) Philippines
February 2008	Survey of application of training in Indonesia
July 2008	Survey of application of training in the Philippines
September 2008	End date and final report

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IMPACT OF INTRODUCTION OF ALIEN SPECIES ON THE FISHERIES AND BIODIVERSITY OF INDIGENOUS SPECIES IN ZHANGHE RESERVOIR OF CHINA AND TRI AN RESERVOIR OF VIETNAM

Mitigating Negative Environmental Impacts/Study/07MNE03UM

Collaborating Institutions & Lead Investigators

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Huazhong Agricultural University (China)	Wang Weimin
Shanghai Fisheries University (China)	Yang Yi
University of Agriculture and Forest (Vietnam)	Le Thanh Hung

Objectives

1. Investigate the changes in population structure of indigenous fish species after the introduction of alien species in the reservoirs.
2. Compare reservoirs in China with and without presence of exotic icefish.

Significance

Alien species are defined as species or subspecies introduced outside their natural past or present distribution. These introductions include any parts, such as gametes, seeds, eggs, or propagules of such species that might survive and subsequently reproduce (Bartley and Fleischer 2005). Among various reasons for introduction of exotic aquatic animals, aquaculture development is a main motive (Welcomme 1998). The introduction of alien species into a new region may cause not only environmental and socio-economic effects, but also be associated with "genetic pollution" (Cripps and Kumar 2003). Invading alien species in the United States cause major environmental damages and losses adding up to almost $120 billion per year (Pimentel et al. 2005). The introduction of alien species into a new region will risk corrupting native fish communities through predation, competition for habitats and food, disease, hybridization and other adverse environmental impacts (Welcomme 2001, Cripps and Kumar 2003), and can also produce imbalances in the fish community, disrupting food chains, and threatening the survival of non-target species (Welcomme 2001).

China has a long history of intentional introductions of alien species, especially species proven to be productive elsewhere and offering potential economic benefits to China. These introductions have changed local ecosystems by modifying species composition, population structure, and food chains (Xie et al. 2001). Significant economic damage has been caused by introduction of alien species in freshwater fisheries, but this damage has not drawn the attention of the public in China (Xie et al. 2001). Although there have been many comprehensive scientific surveys on fish populations conducted in large natural watersheds including lakes, rivers, and estuaries of China, fishery research has focused on the biology and status of economically important fish, while very little research has been conducted on the impact of alien species especially introduced for aquaculture on species population, biological diversity and trophic interactions in freshwater aquatic ecosystems (Liao 1998).

Icefishes (family Salangidae) have a wide natural distribution in the coastal and inland waters of China (Fang 1934a, b) and are important commercially for fishes with high market value in the inland waters, such as Taihu Lake and Puyang Lake (Wang 1980, Chen and Zhang 1990). Overfishing and water pollution have caused the natural production of icefishes to decrease rapidly in late 1970s and early 1980s (Chen and Zhang 1990). Since late 1970s, icefishes have been introduced into many lakes and reservoirs in which they are not native species, in order to increase production of icefish to meet market demands. From 1995 to 1997, more than 150 reservoirs were stocked with 360 million eggs of icefishes (Feng et al. 2002). One of the major stocked icefishes is *Protosalant hyalocranius*.

Tilapias (*Oreochromis* spp.) have been being promoted as poor farmers' fish for food security as well as fish with export potential in many parts of Asia. Tilapias were introduced into Vietnam several times from 1951 to 1997, and have been widely cultured in various systems, such as ponds, cages, and rice-fields (Tu 2003). Escaped tilapias from aquaculture have established populations in natural aquatic environments such as reservoirs (Tu 2003 2006). For example, tilapias accounted for about 4% and 20% of the total catch in Tri An and Thac Mo Reservoirs, respectively (Tu 2003, 2006). Tilapias established in natural waters are enigmatic. Some regard tilapia as beneficial to local fisheries (and sometimes for control of mosquitoes or aquatic plants); some consider them pests with stunted populations that compete with indigenous fish species; and some consider their presence controversial, with benefits and negative effects, depending on geographical area (Lowe-McConnell 2000).

The purpose of this study is to investigate the impacts of both icefish and tilapias on fisheries and biodiversity of indigenous fish species in reservoirs in China and Vietnam.

Quantified Anticipated Benefits

This study will estimate the degree and extent of damages on fisheries and biodiversity of indigenous fish species due to the introduction of alien species in the region. The impact of this study will be evaluated by estimating the number of indigenous species which have been affected in each reservoir. Information on the impacts of the introduced alien species on fisheries and biodiversity of indigenous fish species will allow governmental agencies to establish policies, plans and mechanisms for the management of the introduction of alien species.

As reservoirs are widely distributed in the Asian countries and alien species have been introduced into many reservoirs either intentionally or unintentionally, these case studies and two one-day workshops each in China and Vietnam involving farmers, reservoir management entities and government officials will provide useful information on appropriate management strategies for alien species in reservoirs and general recommendations on policies for the introduction of alien species.

Research Design & Activity Plan

Location: Field sites include Zhanghe and Huiting Reservoirs in Jingmen City, Hubei province, China; and Tri An Reservoir, Hochiminh City, Vietnam. Lab work will be conducted in Huazhong Agricultural University, China and University of Agriculture and Forestry, Vietnam.

Research method:
Location: Field sites include Zhanghe and Huiting Reservoirs in Jingmen City, Hubei province, China; and Tri An Reservoir, Hochiminh City, Vietnam. Lab work will be conducted in Huazhong Agricultural University, China and University of Agriculture and Forestry, Vietnam.

1. Survey in China: Zhanghe Reservoir and Huiting Reservoir are located in Jingmen City, about 100 km away from each other. Icefish were introduced into Zhanghe Reservoir, but not into Huiting Reservoir. Both have similar size and have been subject to similar fisheries management practices under management of the same department of fisheries.

1.1.. Secondary data collection: Historical data on fish catch and fish species composition will be collected from relevant reservoir management agencies, the Zhanghe Reservoir Fisheries Management Company, and the Huiting Reservoir Fisheries Management Company. Huiting Reservoir has not been introduced with icefish and serves as a control site.

1.2. Primary data collection: During the one-year study period, fish catch and fish species composition will be collected through: (1) seining fish at 4, 5, and 4 locations respectively upstream, midstream, and downstream of the two reservoirs for four times each during winter, spring (spring season), summer, and autumn seasons; (2) collecting data at fish landing points; (3) collecting data during the annual harvest by the fisheries management companies; (4) providing record sheets to all the fishermen in the two reservoirs for recording the fish species they catch.

2. Survey in Vietnam:
2.1 Secondary data collection: Historical data on fish catch and fish species composition in Tri An Reservoir will be collected from relevant reservoir management agencies and Dong Nai Fisheries Company.

2.2. Primary data collection: During the one-year study period, the primary data on fish catch and fish species composition will be collected through the same ways as defined in 1.2.

2.3. Data analyses: The secondary and primary data from the two reservoirs with and without the introduction of icefish (Zhanghe and Huiting reservoirs, respectively) will be compared statistically to assess the impacts of the introduction of icefish on the fisheries and biodiversity of indigenous fish species. The secondary and primary data from Tri An Reservoir will be used to analyze statistically the changes of fish catch and fish species composition before and after the introduction of tilapias as well as the trend over time.

Hypotheses: Introduction of icefish does not affect the fish catch and fish species composition of indigenous species. Null hypotheses: There are no significant differences in total fish catch and fish species composition of indigenous species between the Chinese reservoirs with and without icefish introduction, as well as over time. There are no significant changes in total catch and fish species composition of indigenous species over time in Tri An Reservoir.

2.4. Staff and student exchange: two staff/student from China and Vietnam will visit the counterpart institutions to exchange experiences and information about the management of alien species in the respective countries, and to expose students in different academic environment, enhance students' knowledge, and to establish closer collaboration between host institutions through student exchange.

2.5. Workshops: Upon completion of the analyses, two one-day workshops each in China and Vietnam will be organized to present findings from this project as a continuing consultation process with farmers, reservoir management entities and government officials for general recommendations on policies for the introduction of alien species. At least 40% of the workshop participants will be women.

Schedule

April 2007 – September 2009. Report submission: not later than September 2009.

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ASSESSING EFFECTIVENESS OF CURRENT WASTE MANAGEMENT PRACTICES FOR INTENSIVE FRESHWATER AND MARINE POND AQUACULTURE IN CHINA

Mitigating Negative Environmental Impacts/Study/07MNE04UM

Collaborating Institutions & Lead Investigators

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Objectives

1. Identify and quantify existing management practices to reduce effluent and solid waste pollution used by intensive freshwater and marine farmers.
2. Quantify nutrient loading rates, nutrient budgets, sediment accumulation of nutrients, and release of nutrients in effluents for intensive ponds, utilizing different waste management strategies.
3. Determine the best current methods for pollution mitigation in intensive pond aquaculture.
4. Inform small-scale pond farmers about waste management in order to build a consensus on management and to develop more effective government policies.

Significance

Both freshwater and marine aquaculture has developed and intensified rapidly in China since the early 1990s. According to Chinese Department of Fisheries, total fisheries production reached 51 million metric tons in 2005, accounting for one quarter of the world total, while aquaculture contributed 65% to the total fisheries production. As aquaculture production intensifies, feed inputs increase and waste material including organic matter, nutrients, and suspended solids in ponds increases, causing oxygen depletion, eutrophication, and turbidity in receiving waters (Lin and Yi 2003).

The discharge of untreated effluent water into receiving waters is responsible for eutrophication and self pollution. This can be mitigated by developing effective wastewater treatment methods. A number of physical, chemical, and biological treatment methods have been investigated to mitigate pollution caused by the wastes from intensive aquaculture ponds (Boyd 1978, Tunvilai and Tookwina 1991, Chaiyakarm and Dusit 1992, Yahiya 1994, Schwartz and Boyd 1995, Kouka and Engle 1996, Tucker et al. 1996, Ghate et al. 1997, Songsangjinda et al. 1999). Due to the increasingly aggravated environmental problems caused by the wastes discharged from intensive aquaculture ponds, the Chinese government has recently adopted a series of regulations and control measures. Thus, waste treatment and effluent reuse facilities are rapidly being developed, especially for large-scale farms (Fang et al. 2004, Wang et al. 2004, Xiao et al. 2006). However, most treatment methods are still experimental, while effluent water from most small-scale farms is discharged without any treatment. Since most fish farmers in China, as well as in most Asian countries, are small-scale, they contribute a significant proportion of wastes discharged from intensive aquaculture ponds.

To date, no comprehensive studies have been conducted to assess waste management practices and evaluate the efficiency of waste treatment methods for intensive aquaculture ponds, especially small-scale ponds in China. This study will focus on waste management in Hubei and Hainan provinces. Hubei is the largest freshwater aquaculture production province in China, while Hainan is the only tropical province that has major tilapia and marine shrimp production. The proposed study will assess waste management practices for intensive aquaculture ponds, to evaluate the efficiency of waste treatment methods used currently by fish farmers, to recommend appropriate waste treatment methods to different types of farms, and to build the consensus among small-scale pond aquaculture farmers implementing effluent and solid waste management.

Quantified Anticipated Benefits

This study will estimate the degree and extent of environmental damage due to intensive pond aquaculture in the region. The impact of this study will be evaluated by estimating the number of intensive aquaculture ponds in each region and their current average waste loadings as well as the reductions that could be achieved by use of better waste management practices. This study will evaluate different waste management strategies used by small-scale farmers. Information on the effectiveness of these strategies will allow governmental agencies to establish policy and plans for waste management in intensive pond aquaculture. Understanding more effective waste management strategies will benefit thousands of small-scale farmers in particular and the environment in general. Upon completion of the analyses, three one-day workshops will be organized to present findings from this project as a continuing consultation process with farmers, private enterprises, community members, and government officials. Impact here will be the number of participants exposed to workshop training.

Research Design & Activity Plan

This study consists of two components. The first component is a survey to assess the current practices of waste management in both freshwater and marine aquaculture ponds, and the second is field measurements to estimate waste loading from intensive aquaculture ponds to the environment and to identify the appropriate waste management practices.

Location: Hubei, Hainan and Sichuan provinces, particularly Huazhong Agricultural University, Hainan University and Shanghai Fisheries University.

Methods:
1. Survey: One hundred farmers will be selected randomly from each category of pond farmers, including: inland intensive farmers, coastal and inland intensive farmers for freshwater tilapia, and marine shrimp farmers. The 400 farmers will be interviewed using a set of designed questionnaires, including a structured checklist and open-ended questions. The questionnaires will evaluate socio-economic characteristics of farmers, pond culture practices, water supply management, effluent water and solid waste management, as well as knowledge and awareness of environmental implications of pond waste disposal.

Relevant environmental regulations will be reviewed and governmental agencies interviewed for the information about regulations, enforcement, and farmers' attitudes.

2. Assessment of wastes from intensive culture ponds: Fifty ponds will be randomly selected from each of the three types of intensive aquaculture farms to assess effluent water quality, sediment quality, and nutrient budgets.

2.1. Pond water will be sampled during filling and drainage at harvest. A column water sample will be collected prior to pond draining, and effluent water samples will be collected from the pumping pipe when the pond is full, and when pond water is at 50, 50 to 30 cm, and 30 to 0 cm depths (top, middle, and bottom water effluents, respectively). Analyses will include organic matter, total solids, total suspended solids, total ammonia nitrogen, total nitrogen (TN), and total phosphorus (TP), following standard methods (APHA et al. 1985). Dissolved oxygen, pH, temperature, and salinity (for marine ponds) will also be measured in situ.

2.2. Core samples of pond sediments at top 10 cm will be collected from at least nine locations before filling ponds and after draining ponds. Sediments will be analyzed for moisture, bulk density, texture, pH, organic matter, TN, and TP.

2.3. Fish and feeds will be sampled at the beginning and end of the culture cycle for the analyses of proximate composition, TN, and TP.

3. Analyses of data: The null hypothesis for these experiments is there are no differences in water or sediment quality under different pond management practices.

Effluent quality at different water depth will be compared using ANOVA within each category of ponds and among the three categories of ponds. Nutrient budgets, wastes discharged, and nutrients in sediments will be compared using ANOVA among three categories of ponds. Nutrient changes before and after fish or shrimp culture will be compared using paired t-test within each category of ponds. If different waste management strategies are found within each category of ponds, t-test or ANOVA will be used to compare the above data.

4. Workshops: Upon completion of the analyses, three one-day workshops each for carp polyculture, tilapia culture and shrimp culture will be organized to present findings from this project as a continuing consultation process with farmers, private enterprises, community members, and government officials. At least 40% of the workshop participants will be women.

Schedule

April 2007 – September 2009. Report submission: not later than September 2009.

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DETERMINING THE ECOLOGICAL FOOTPRINT OF SHRIMP AQUACULTURE THROUGH LIFE CYCLE ANALYSIS OF OUTDOOR POND SYSTEMS

Mitigating Negative Environmental Impacts/Study/07MNE05UM

Collaborating Institutions & Lead Investigators

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Objectives

1. Perform life cycle assessment (LCA) and life cycle cost analysis (LCCA) for an outdoor, flushed pond shrimp aquaculture system.
2. Develop mass balance models of the shrimp aquaculture system to make it possible to evaluate methods to improve the economics and reduce the environmental impacts of the system.

Significance

As human populations increase, the food production system must intensify to accommodate population growth. Currently, crop lands and pastures occupy nearly 40% of the surface of the earth (Asner et al. 2004), and have changed natural ecosystems into monocultures with intensive growing of crops (Foley et al. 2005). Aquaculture, an alternative to these traditional food production systems, has been the most rapidly growing food production system, with an annual increase of about 10% per year since 1985 (Delgado et al. 2003, Diana 2004). To date, aquaculture expansion has occurred mainly in Asia. There is also demand, since seafood production from wild fish harvest is stabile or declining, global population is increasing, and per-capita seafood consumption will increase about 1.5 kg per person per year by 2025 (FAO 2004). Both population growth and increased individual consumption indicate that seafood products will be more important as food, and aquaculture will play an increasing role in that consumption as natural fish stocks continue to decline (Wijkstrom 2003, FAO 2005). Shrimp stocks in nature have declined due to overfishing and pollution, and shrimp harvesting is one of the fishing methods most destructive to biodiversity (Harrington 2005, Hannesson 2002. Therefore, an efficient expansion of shrimp production in the U.S. and worldwide must occur through aquaculture. We believe this is an achievable goal since shrimp aquaculture is a relatively lucrative business with high demand and an acceptable profit margin.

Aquaculture has the reputation of being a particularly environmentally damaging food production system (Goldburg and Triplett 1997, Naylor et al. 2000) due to the use of fish meal for feed, the location of ponds in sensitive coastal areas, accidental release of exotic species or genotypes, and discharge of excess solids and nutrients causing coastal eutrophication (Boyd and Clay 1998). Consequently, many people have questioned whether sustainable aquaculture can be practiced. In reality, any intensive food production system is not currently sustainable, since they all rely heavily on energy and other inputs and often have a major impact on the environment. Nevertheless, expansion of shrimp aquaculture will need to proceed in a sustainable and environmentally sensitive manner (Clay 1997). Systems proposed for sustainable shrimp aquaculture include indoor recirculating systems (which produce limited or no discharge of water and have no escapees), and outdoor systems with zero discharge (which reuse water in ponds but may have escapement problems). Both of these systems are experimental and at present provide minimal additions to traditional shrimp culture in ponds. These systems vary considerably in energy use and sustainability from perspectives other than discharge effluents.

Balancing the future growth of aquaculture, as well as fisheries, requires a fair comparison of environmental impacts. Life cycle assessment (LCA) is one method to make such a comparison. LCA allows one to calculate the relative energy intensity and material usage in an overall process or in various components of that process. Thus, one can compare various aquaculture systems from an energy efficiency and material use perspective. Life cycle modeling, which evaluates feed production, farming, and harvesting, transport, processing, distribution and sales, consumption, and waste management, provides a useful framework for understanding the limiting steps in energy or material use. Life cycle cost analysis (LCCA) can also be used to study the economic dimension of sustainability. Many effects of aquaculture are based upon widely differing impacts, such as land use, sensitivity of the land used, amount of water used, nutrient discharges, feed use, transportation of aquaculture products, release of exotic species, and genetic alteration of existing species by escapees from culture. These are very difficult characteristics to compare in a common unit of measure. The purpose of our analysis is to begin to build such comparisons, using multiple perspectives, including LCA, LCAA, ecological efficiency, and nutrient dynamics for shrimp aquaculture systems.

To develop a truly efficient aquaculture production system, most of the nutrients applied should be absorbed by crop organisms rather than accumulated in the system, discharged as waste or released to the atmosphere. Mass balance modeling will allow us to determine how much of the different nutrients provided are assimilated directly by shrimp, utilized by bacteria, or released into the environment.

The purpose of this study is to compare the sustainability and environmental impact of a photosynthetic, suspended-growth system applied in outdoor ponds (Hargreaves 2006), the traditional shrimp aquaculture system today, with a zero-exchange, recirculating indoor system, promoted commonly as a more sustainable system. We have received funding from the University of Michigan to complete LCA, LCCA, and mass balance models of an indoor recirculating system. Therefore, this proposal focuses on the more typical outdoor system.

Shrimp are marine, warm-water species, with optimal temperatures for growth around 28ºC (Fast 1991). Thus, to be feasible, shrimp aquaculture in Michigan requires a greenhouse or indoor system with heating and temperature control, salt water prepared by adding salt or by using local brackish water sources, and water treatment to allow recycling of the water. The energy and material costs of such a system may exceed those of outdoor production facilities in warmer areas with ready access to salt water. However, other costs, such as marketing and transportation costs, may be reduced. Therefore, it is necessary to do a comprehensive financial, energy, and material analyses over a production cycle in order to produce an integrated assessment of the overall impact of the indoor system. Our comparison will utilize LCA, mass balance modeling, and cost-benefit analysis of both systems. The indoor shrimp aquaculture system is relatively unstudied, but a prototype of this system has been built in Okemos, Michigan by Russ Allen and is available for data collection. Data will be collected from outdoor shrimp pond facilities to complete the objectives and allow the direct comparison of each system.

Quantified Anticipated Benefits

The results of this study will provide information relative energy intensity, environmental impact, and cost-benefit of two very different aquaculture systems. Currently there are no objective methods to assess sustainability of aquaculture in a quantitative and fair manner. For example, the Watch Program (Monterey Bay Aquarium 2006) and the Seafood Choices Alliance (2006) have no objective method to compare sustainability of captured and cultured seafood, so the actual listings rely on value judgments. This methodology will allow evaluation of different aquaculture products, and by extension wild fishery products as well, in quantifiable terms that are clear indicators of sustainability. The quantifiable benefits will include a developed methodology for analyses and direct comparisons of two divergent systems to allow regulators, governments, and the public to understand and extend more sustainable aquaculture. The measurable benefits will be the amounts of energy used in different components of the market cycle, and potential reduction that could occur with changes in the production system. Also, the mass balance of nutrients and nutrient recovery of different systems will also be compared in terms of eutrophication potential as well as the production of greenhouse gases, and again direct comparison of different components of the system will provide measurable reductions that could occur by changes in production systems.

Research Design

Locations: Hainan province and Hainan University (China).

Research methods:
1. Farm selection: A typical shrimp farm will be selected, and the management options used there compared to other sites locally and in the region.

2. Field and evaluation methods:
2.1. Life cycle assessment: An LCA of shrimp aquaculture requires the availability of data on materials and energy inputs, as well as data on water cycling, water use, water treatment, and water quality. The LCA will be conducted in accordance with ISO 14040 Standards (ISO 1997). A variety of sites for production and consumer markets will be analyzed to study the importance of transportation on the total environmental impact. Life cycle environmental sustainability metrics that will be evaluated for each system include total primary energy use, global warming potential, acidification, eutrophication, human toxicity potential, and solid waste production. In addition, the economic analysis will evaluate the full costs for production, processing, and distribution.

2.2. Mass balance modeling: Mass balance modeling, as an approach, is commonly used in the understanding of energy and material flow as well as in setting limits on natural systems. For example, mass balance models have been used to predict carrying capacity for natural cod populations, and factors influencing their recovery (Bundy and Fanning 2005). Mass balance models have been applied to shrimp aquaculture (Alongi et al. 2000, Epp et al. 2002, Obaldo and Ernst 2002, Alonso-Rodriguez and Paez-Osuna 2003) to understand system efficiency and also to evaluate environmental impacts (Paez-Osuna et al. 1999, Siri 1999, Trott et al. 2004, Schneider et al. 2005). The current project will use mass balance to model the use of nitrogen, phosphorous, and food energy for a traditional system.

3. Null hypothesis, statistical analysis:
Null hypothesis: There are no significant differences in traditional aquaculture systems and indoor recirculating systems for energy efficiency, nutrient retention, or global warming potential in the production of marine shrimp.

Data will be analyzed using ANOVA, t-test, correlation and regression.

4. Student exchange: a PhD student from University of Michigan will be exchanged to Chinese host institution to conduct this research together with the host institution students.

Schedule

April 2007 – September 2009. Report submission: no later than 30 September 2009.

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ELIMINATION OF MT FROM AQUACULTURE MASCULINIZATION SYSTEMS: USE OF CATALYSIS WITH TITANIUM DIOXIDE AND BACTERIAL DEGRADATION

Mitigating Negative Environmental Impacts /Experiment/07MNE06UA

Collaborating Institutions & Lead Investigators

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Objectives

1. Evaluate the effectiveness of MT degradation utilizing catalysis with titanium dioxide and UV.
2. Determine if bacteria obtained from biological filters are capable of degrading MT used in masculinizing systems.

Significance

Agricultural and industrial activities generate a significant number of pollutants that are released to the environment through sewage water, which reach the superficial and underground water reservoirs. Many of these substances are highly toxic and do not degrade easily in nature. As a result of years of negligence, the levels of these substances in rural and industrial areas in Latin America are dramatically high. In order to reduce risks to human health and the environment, water treatment systems have been implemented (Valladares, 1995). Water treatments aim at 1) eliminating waste, floating fats and oils, sand, and all other coarse elements water may contain; 2) eliminating decantable materials, both organic and inorganic; 3) eliminating biodegradable organic matter dissolved in water; and 4) stabilizing and disposing mud extracted as a result of those processes (Crites and Tchobanoglous, 2000).

Traditional technologies used to separate organic substances from water use activated charcoal absorption or air hauling. However, these processes only transfer pollutants from water to other environmental media, which perpetuates the problem. At present there is a group of technologies based on pollutant destruction processes through chemicals known as hydroxyl radicals, which are highly oxidative. In these technologies, called "Advanced Oxidation Processes" (AOP), radicals react with pollutants and transform them into environmentally harmless compounds, mostly CO_2 and water. Implementation of these technologies has begun in North America, Europe, and Japan.

Among the materials used as catalysts are TiO_2, ZnO, CdS, iron oxides, WO_3, and ZnS, among others. These chemicals are financially affordable, easily traceable in nature, and can excite with low-energy light, absorbing part of the sun's spectrum radiation that hits the Earth's surface ($\propto >$ 310 nm). In particular, photocatalytic degradation based in the use crystallized titanium dioxide (TiO_2,) and low-energy (320-390 nm) ultra-violet (UV) sunlight is interesting for water treatment purposes. This method has been tested in laboratories since the mid-80's for hydrocarbons such as chlorate and phosphate compounds contained in pesticides, herbicides, colorants and surfactants. The technique consists in generating hydroxyl radicals which then oxidate organic compounds. Recently, interest in this technology has increased due to its potential solar energy applications, as 5% of the sunlight reaching the troposphere provides enough energy to activate titanium dioxide (Valladares, 1995).

Aquaculture is an area in which AOPs could be applied to effluent treatment. This industry uses steroidal hormones to manipulate phonotypical sex (Yamamoto, 1969). If these chemicals are adequately treated, their use is beneficial and has no adverse environmental effects. However, there seems to be grounds for concern regarding water overuse. In particular, a legitimate international concern is the lack of up-to-date quantitative information regarding the chemicals being used and the lack of efforts oriented to studying and treating the polluting by-products generated by this industry, steroids being one of them. Hormones used in aquaculture are among a list of compounds which environmental effects and possible treatments have been prioritized for research by GESAMP (FAO's Joint Group of Experts in the Scientific Aspects of Marine Environmental Protection).

Recent innovations in hormone applications in aquaculture include sex control, thus producing the so-called mono-sex lines, which result in significant production improvement (GESAMP, 1997). In particular, the molecule 17 α-Methyltestosterone (MT) has been widely used as androgenic agent in masculinization of tilapia (Contreras-Sánchez, 2001). After the hormonal treatment (28 days), the hormone is released to the water through urine and feces and becomes a powerful pollutant capable of producing harmful effects in wild species when effluents are released. In recent studies (Contreras-Sánchez, 2001), we found that masculinization of fry through dietary treatment with MT resulted in the accumulation of MT in sediments which produced both intersex fish and females with altered ovarian development. In systems where substrate was not present, there were higher concentrations of MT in the water and lower (sometimes null) masculinization rates than in systems with either soil or gravel. We found that charcoal filtration of water from systems where substrate was not present lowered the amount of MT in water to almost background levels and the treatment resulted in almost complete masculinization of all three broods tested (100, 98 and 100% males, respectively). Apparently, the recommended dose of MT for masculinizing tilapia is higher than needed and a significant portion of it separates from the food and remains either in suspension in the water for the short term or persists in the sediments over the long term (Contreras-Sánchez, 2001). In the cited study, we recommended the use of filtration systems to eliminate excess MT to increase masculinization, and to prevent potential risks to humans of unintended exposure to MT due to contamination of water and soils in farms. In this sense, research has been published on photocalytic hormone elimination (Tanizaki, 2002). However, the published studies have focused on sewage water estrogen elimination due to the harmful biological effects of the presence of small levels of steroidal estrogens in water (Ike, 2002; Coleman et al., 2000 ; Carballa, 2004). Nevertheless, the elimination of MT in aquaculture using advanced oxidation processes have not been addressed.

Another possible approach to eliminating MT from masculinization systems involves the use of bacterial degradation since it has been reported that some bacteria are capable of degrading steroids (Voishvillo et al, 2004). From this information and results from the previous investigations we hypothesized that MT was being eliminated from water by solar irradiation and/or bacterial degradation within the filtration system. In our latest experiments (unpublished data) we have detected specific bacteria colonies growing in the biological filter of MT-treatment tanks, but not in the control tanks (showing mainly algae). In this particular study, the ultimate goal of our research was to isolate, characterize and cultivate the species of bacteria responsible for degradation of steroids. However, experiments are needed to demonstrate the efficacy of these bacteria.

The goal of this investigation is to determine if the titanium dioxide or bacteria present in biofilters are capable of degrading MT. If either method efficiently eliminates MT from aquaculture effluents, tilapia fry producers could incorporate them into their filtration systems, thus eliminating the steroid in a completely natural way. Removal of MT could both increase masculinization rates and reduce the amount entering substrates which could affect other aquatic organisms. These methods may allow for the production of large numbers of all-male populations of tilapia fry using a reliable technique compatible with the proposed Best Management Practices for aquaculture systems.

Quantified Anticipated Benefits

- Number of tilapia fry hatcheries and tilapia growers in Southeastern Mexico that will benefit from the Implementation of clean technology for masculinization of tilapia fry.
- Number of persons participated in workshops.
- Number of linkages with other organizations completed.
- Number of technical reports and journal articles.

If successful, these methods can be transferred to tilapia hatcheries that play an important role supplying masculinized fry to poor areas of the states of Tabasco and Chiapas, México as well as other countries in Central America. The use of reliable and efficient masculinizing methods in the hatcheries will benefit thousands of small scale fish farmers who currently see their productivity negatively affected by the use of mixed-sex populations of tilapia. A series of training workshops will be developed and offered to different audiences in the communities of southeastern Mexico to ensure that these methodologies are effectively transferred to its final users. Technical workshops will target hatchery managers, extension agents and university students (many of whom will become workshop instructors over time). Public extension workshops will be tailored to the cultural characteristics of the target audience and will be offered to fish farmers, farm workers and selected community leaders.

Research Design & Activity Plan

Experiment 1. Use of TiO_2, UV light and air: To conduct this experiment, a rectangular 30x30x20 cm reactor will be used. Glass plates the size of the reactor's base with immobilized titanium oxide will be prepared. The flux will be recirculated with a peristaltic pump. The volume of water in the reactor will be 3,000 ml. High-pressure mercury light will be set over the reactor to serve as a source of UV light (Luck, 1997).

Fixation of TiO_2 in glass plate. Four successive layers of a TiO_2 5% m/v ethanol suspension will be applied with a foam rubber roller. Following application of each layer, the panel will be sun-dried and, once dry, will be heated at 200° C for one hour in a laboratory stove in order to eliminate any remaining organic matter and contribute to the cohesion of the semiconductor. The panel will then be washed with distilled water in order to eliminate any semiconductor trace that does not adhere after the thermal treatment. After applying the last layer, it will be heated at 280 °C for 2 hours (Byrne, 1998).

Solutions. A stock solution of 17 α-Methyl testosterone (Sigma-Aldrich, Inc.) in ethanol will be prepared with a final concentration of 1 ug/L. A working solution of 3L of water containing MT will be prepared to assure a concentration of 1 ng/ml. The titanium oxide to be used is TiO_2 P25 (Degussa, Corp.) in distilled water (Colleman, 2004).

Experimental design. Experiment will be conduced at the Laboratory of Aquaculture at UJAT, Tabasco, Mexico. The Experiment will consist of a 2 x 2 x 3 factorial experiment where factor A is the air effect, factor B is the effect of UV light and factor C is the type of titanium oxide delivery method (attached to a glass plate, in suspension and absent). All treatments will be evaluated in duplicate. Treatments will be as follows:

FACTOR A (AIR FLOW)	FACTOR B (UV LIGTH)	FACTOR C (TiO_2)	TREATMENT CODE
YES	NO	ABSENT	T1
		IMMOBILIZED	T2
		IN SUSPENSION	T3
	YES	ABSENT	T4
		IMMOBILIZED	T5
		IN SUSPENSION	T6
NO	NO	ABSENT	T7
		IMMOBILIZED	T8
		IN SUSPENSION	T9
	YES	ABSENT	T10
		IMMOBILIZED	T11
		IN SUSPENSION	T12

Water Management. Three liters of water (control or MT-treated) will be placed in the reactor and will be recirculated using a peristaltic pump. Water will be exposed to treatments for 7 hours.

Project Coordinator: Rosa Martha Padrón-López, Hill be responsible for supervising students who will conduct the trial and she will prepare the analytical analyses and report.

Sampling schedule. Water samples will be collected at 0, 1, 3, and 7 hours. Samples will be frozen (–20°C) and preserved until processing. All samples will be extracted using ether and the concentration of MT determined by RIA (at Oregon State University).

Statistical Methods and Hypothesis. Hypothesis: The use of Ti_2O, UV and air supply will eliminate MT from the water; MT detectable in water at any time during treatment independently of the treatment used. Data will be compared with two a factorial ANOVA test and a posteriori Tukey test.

Experiment 2. Use of bacterial degradation: Isolated bacterial colonies obtained from biological filter units in our last experiment will be used. These colonies where obtained by inoculation of microbiological culture media with material (sediments and fouling) from a biological filter unit used in an MT masculinization system at UJAT. Best growing and most abundant colonies where selected.

Microbiological culture media. Standard microbiological culture media will be used. A stock solution of 17 α-Methyl testosterone (Sigma-Aldrich, Inc.) in ethanol will be prepared with a final concentration of 1 ug/L. Culture media solution will be enriched with MT stock solution to assure a concentration of 1 ng/ml.

Experimental design. Site: This experiment will be conducted at the Aquaculture and Microbiology laboratories at UJAT. The experiment will be based on one factor, it will consist of 6 treatments. All treatments will be evaluated in triplicate.
Treatments will be as follows:

1. Control: No bacterial colonies added.
2. BC1: Bacterial colonies type 1 added.
3. BC2: Bacterial colonies type 2 added.
4. BC3: Bacterial colonies type 3 added.
5. BC4: Bacterial colonies type 4 added.

6. BC5: Bacterial colonies type 5 added.

Culture management. Selected colonies will be inoculated in each culture media with a similar cell concentration using turbidimetric methods. All culture media will be placed in a microbiological incubation oven to maintain constant temperature (35 ºC) until the end of the experiment.

Sampling Schedule. Samples for MT determination will be collected before bacterial inoculation and at 12, 24, 48 and 72 hours after treatment (the final sampling time will depend on bacterial culture lifetime). At every sampling samples will be deep-frozen under liquid nitrogen to kill bacteria and stop potential MT degradation. All samples will be frozen (–20°C) and preserved until processing. MT will be extracted from samples using ether and the concentration of MT determined by RIA (at Oregon State University).

Statistical Methods and Hypothesis: Hypothesis. MT will be eliminated from the culture media by bacterial degradation. MT concentrations will be compared using a Kruskall-Wallis test ($p< 0.05$).

Experiment 3. Use of bacterial degradation of MT in masculinizing systems: Best treatment from experiment 2 will be used in a masculinization system. Isolated bacterial colonies will be inoculated to microbiological culture media to produce massive amounts of steroid degrading bacteria. Biological filters used in masculinization systems will be inoculated with this bacteria and MT concentration will be determined.

Experimental design. Site: This experiment will be conducted at the Aquaculture and Microbiology laboratories at UJAT. The experiment will be based on one factor, it will consist of 2 treatments. All treatments will be evaluated in triplicate. Treatments will be as follows: The experiment consists of two treatments; each treatment will be triplicated:

1. Fry fed MT at 60 mg/kg food for 28 days; water recirculated through biofilter without bacterial inoculation.
2. Fry fed MT at 60 mg/kg food for 28 days; water recirculated through biofilter with bacterial inoculation..

Water Management: Water from masculinization systems (8000 l tanks) will be recirculated through biofilters. All tanks will receive MT-treatment recirculated water. After 7 days of fish treatment, bacteria will be inoculated into the biofilters of Treatment 2.

Test Species: Nile tilapia (*Oreochromis niloticus*).

Stocking Rate: 1,500 fry/m3

Sampling Schedule. Samples for MT determination will be collected the day of inoculation and at days 14 and 28 of MT treatment. All samples will be frozen (–20°C) and preserved until processing. MT will be extracted from samples using ether and the concentration of MT determined by RIA (at Oregon State University).

Statistical Methods and Hypothesis: Hypothesis. MT will be eliminated from the masculinization systems where bacteria are inoculated. MT concentrations will be compared using a Kruskall-Wallis test ($p< 0.05$). Sex ratios will be compared by a Chi-square test to determine efficacy of MT treatment.

Destruction of MT in Experiments 1-3: All waters containing MT will be collected during the trials. At the conclusion of the trials, all of the water will be treated with ozone to insure complete destruction of all methyltestosterone.

Schedule

Experiment 1

June – Sep 2007	System setup
Oct – Dec 2007	Experiment
Jan – June 2008	Detection of MT by RIA
June – Dec 2008	Data analysis and preparation of reports and publications

Experiment 2

June 2007	Bacterial colonies selection and culture.
June – Sep 2007	Bacterial MT degradation experiment
Sep – Dec 2007	MT determination by RIA
Jan – June 2008	Data analysis and preparation of reports and publications

Experiment 3

July – Oct 2008	Bacterial culture.
Nov 2008 – Jan 2009	Bacterial MT degradation experiment
Jan – May 2009	MT determination by RIA
June – Sep 2009	Data analysis and preparation of reports and publications

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WORKSHOP ON AQUACULTURE, HUMAN HEALTH, AND ENVIRONMENT

Mitigating Negative Environmental Impacts/Activity/07MNE07UM

Collaborating Institutions & Lead Investigators

University of Michigan (USA)	James S. Diana
Shanghai Fisheries University (China)	Yang Yi
World Wildlife Fund (WWF)	Flavio Corsin

Objectives

1. Present the research results of AquaFish CRSP.
2. Share the experience in research related to aquaculture, human health, and environment.
3. Establish links between AquaFish CRSP host institutions and other institutions in the region.
4. Begin collaboration in aquaculture between Asian CRSP colleagues and WWF.
5. Develop links between research findings and responsible management within the aquaculture sector.

Significance

Aquaculture and culture-based fisheries have been developed and intensified rapidly in Asian countries since late 1970s, and have played important roles in the livelihood of local farmers through employment, income generation, and food security. However, environmental impacts of aquaculture and culture-based fisheries have caused serious concerns. Lack of environmental awareness and environmentally friendly technologies have polluted many public waters such as rivers, lakes, and reservoirs. Several alien species have been introduced to many Asian countries. The potential impacts on natural aquatic ecosystems and native biodiversity urgently need to be addressed and appropriate control mechanisms need to be established. With intensification of aquaculture systems, heavy uses and abuses of chemicals and drugs as well as microcystins produced by natural blooms of bluegreen algae are concerns for food safety and sustainable use of those waters for fisheries production. However, such environmental concerns have not been fully addressed through research and education at various levels. Therefore, the purpose of this activity is to develop a workshop to address environmental issues related to aquaculture and culture-based fisheries that will promote sustainable development of aquaculture and cultured-based fisheries in Asia countries.

Quantified Anticipated Benefits

These include the number of AquaFish CRSP and other invited presentations; number of participants and number of participating institutions at the workshop.

Activity Plan

1. A two-day workshop on aquaculture, human health, and environment will be held in Chengdu, China in August 2009, in cooperation with the Sichuan Aquacultural Engineering Research Center in Chengdu.
2. Two speakers from WWF (most likely Flavio Corsin, the *Pangasius* project leader stationed in Vietnam, and Eric Bernard, shrimp project leader) will present on WWF programs in sustainable aquaculture, human health, and food safety.
3. All host institution PIs will present the results of AquaFish CRSP investigations.
4. Several invited speakers will present their experiences in related topics.
5. The participants will include PIs, researchers and students from host institutions, host country government officers, experts in aquaculture, public health, environment and food safety in host country institutions, and those from private sectors and NGOs, with the total participants of about 40.

Schedule

August 2009. Report submission: no later than 30 September 2009.