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# Biological Sciences

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Effect of Dietary Incorporation

Highlights

Composition and Ecological

Lands through Plantation Forests

Discovering Thoughts, Inventing Future

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# Butterfly as Pollinating Insects of Flowering Plants

# By Pollobi Duara & Jatin Kalita

Gauhati University, Assam, India

*Abstract-* The research showed that butterflies are the main pollinators of Ixora coccinea in Nambor Wild Life Sanctuary, Assam. The family of Papilionidae (6 species), Pieridae (3 species) and Nymphalidae (2 species) are mainly found as insect visitors. The time of the day had a significant effect on the number of butterflies that visited the flowers. Afternoons had more visitors than mornings suggesting that the butterflies become active as the day warms up. The frequency of butterflies visited the flowers was high during 09:00-13.00 hour and month of april to august. Flower colour had a positive influence on the number of visitors. The flowering season of I.coccinea is mainly summer and butterflies are deriving most of their heat from the sun.

GJSFR-C Classification : FOR Code: 820209, 069999



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# Butterfly as Pollinating Insects of Flowering Plants

Pollobi Duara <sup>a</sup> & Jatin Kalita <sup>o</sup>

Abstract- The research showed that butterflies are the main pollinators of lxora coccinea in Nambor Wild Life Sanctuary, Assam. The family of Papilionidae (6 species), Pieridae (3 species) and Nymphalidae (2 species) are mainly found as insect visitors. The time of the day had a significant effect on the number of butterflies that visited the flowers. Afternoons had more visitors than mornings suggesting that the butterflies become active as the day warms up. The frequency of butterflies visited the flowers was high during 09:00-13.00 hour and month of april to august. Flower colour had a positive influence on the number of visitors. The flowering season of I.coccinea is mainly summer and butterflies are deriving most of their heat from the sun.

#### I. INTRODUCTION

Plants and animals have a close interrelationship for their survival, propagation and control. Berenbaum (1995) states that "Sexual reproduction is just as important for plants as it is for animals when it comes to sex they can't just get up and find themselves a mate." Plants must rely on pollen vectors, from wind to insects to birds, to transport their pollen to another individual.

The process of transportation of pollens from stamens to the ovary is called pollination. The insects that visit flowers belongs to the group Hymenoptera, Leopidoptera, Diptera, Coleoptera, Thysanoptera and Hemiptera. Very scanty works have been done on pollinating insects of North Eastern states. However, it is generally only adult winged insects that specialise in visiting flowers. Bhattacharjee (1985a, 1985b) studies the taxonomy and distribution of Nymphalidae, Pieridae and Lycanidae butterflies in North Eastern region of India. North East India accounts for nearly a two-third (962 species) (Evans, 1932) of the India's total butterfly species (Kunte et.al, 1999) Plant diversity influences the diversity of pollinating insects like butterfly. The present study is conducted on Pollinating insects of Ixora coccinea.

#### II. MATERIALS AND METHOD

Study Site: Study was conducted at Nambor Doigrung wild life sanctuary which is situated in the Golaghat district of Assam. This sanctuary shares its boundaries with the Nambor Reserve Forest and Garampani wild life sanctuary. It covers and entire area

*Authors* α σ: Department of Zoology, Gauhati University. e-mail: pallu111.111@gmail.com of 97.15 sq. km. Study was conducted from January 2011 to december 2011.Nambor Doigrung Wildlife sanctuary is geographically located between 92o 52`to 92o 53`east longitude and 26o 22` to 26o 24` North latitude.

The area is in tropical basin of India and as a result of that the temperature are never too high or low with a very heavy monsoon. The maximum/minimum temperature remains in between 80 to 300c. Annual rainfall is 2500mm.

Study plant: The study was conducted on lxora coccinea. Ixora is a genus of flowering plants in the Rubiaceae family. It consists of tropical evergreen trees and shrubs and holds around 500 species. The plants possess leathery leaves, ranging from 3 to 6 inches in length, and produce large clusters of tiny flowers in the summer. I. coccinea is a dense, multi-branched evergreen shrub, commonly 4–6 ft (1.2–2 m) in height, but capable of reaching up to 12 ft (3.6 m) high. It has a rounded form, with a spread that may exceed its height. The glossy, leathery, oblong leaves are about 4 in (10 cm) long, with entire margins, and are carried in opposite pairs or whorled on the stems. Small tubular, scarlet flowers in dense rounded clusters 2-5 in (5–13 cm) across are produced almost all year long.

*Pollination Syndrome:* Pollination syndrome study include flower shape, size, colour, odour, reward type and amount, nectar composition, timing of flowering, etc. Pollination syndromes reflect convergent evolution towards forms (phenotypes) that limit the number of species of pollinators visiting the plant.

*Medicinal value:* The flowers, leaves, roots, and the stem are used to treat various ailments in the Indian traditional system of medicine, the Ayurveda, and in various folk medicines. The fruits, when fully ripe, are used as a dietary source. Phytochemical studies indicate that the plant contains the phytochemicals lupeol, ursolic acid, oleanolic acid, sitosterol, rutin, lecocyanadin, anthocyanins, proanthocyanidins, and glycosides of kaempferol and quercetin.[1]

*Flower Phenology :* Flower phenology was observed at both plant and inflorescence level with reference to day to day flowering pattern. Flower phenology is determined by observations made atleast 3times per week, flowering time, time of opening and closing of flowers (Mark and Francoise, 2005) The flowering season of lxora coccinea was recorded.The

phenological traits were estimated by counting flower heads in anthesis on individual plants every seven days.

*Pollination syndrome*: Pollination syndrome study include flower shape, size, colour, odour, reward type and amount, nectar composition, timing of flowering, etc. Pollination syndromes reflect convergent evolution towards forms (phenotypes) that limit the number of species of pollinators visiting the plant.

*Insect Pollinators Diversity :* Diversity of insect pollinators was observed using line transect and point transect method. Several insect visitors were collected for species identification purpose.

*Insect Pollinator visiting Frequency:* Observations of insect flower visiting frequency were conducted by scan sampling methods (Martin and Bateson, 1993). The observations included foraging rate (number of flowers/minute), flower handling time (seconds/flower) and plant handling time (seconds/plant) [Dafni 1992]

*Data Analysis:* Measures used were Visitor abundance, number of flower visitors seen per transect, and visitor species richness, number of insect species visiting flowers in each transect in each week.

#### III. Result

Table 1 : Family and species of butterfly as pollinator for 12 month observation

Taxon	Family	Species	Percentage
	Papilionidae	1.Atrophaneura varuna 2.Papilio clytia 3.Papilio nephelus 4.Papilio helenus 5.Papilio polytes 6.Papilio mormon	54.54%
Lepidoptera	Nymphalidae	1.Melantis leda 2.Ypthima huebneri	18.18%
	Pieridae	1.Hebomoia glaucippe 2.Ixias pyrene 3.Ixias moriame	27.27
Total	3	11	100%















Time block (h)	Family	Species	
7.00-8.00	2	3	
8.00-9.00	1	4	
9.00-10.00	2	5	
10.00-11.00	2	7	
11.00-12.00	3	8	
12.00-13.00	2	8	
13.00-14.00	1	5	
14.00-15.00	2	2	

Table 2 : Total species of insect visitor in time blocks for 12 month observation

*Table 3*: Temperature and Rainfall of the study area during the study period

Month	Minimum Temperature(degree celcius)	Maximum Temperature(degree celcius)	Rainfall
January	10	24	1 cm
February	15	30	2 cm
March	15	30	5 cm
April	20	30.5	15 cm
May	21	31	23 cm
June	25	31	30 cm
July	25	32 c	30 cm
August	24	31.5	25 cm
September	24	30	15 cm
October	21	29.5	5 cm
November	15	26	2 cm
December	11	25.5	2 cm

*Discussion:* Butterflies are the most frequent pollinators of I.coccinea. Similar findings were reported by S.V.A.Hameed(2012). Bees, wasps, moths and other insect groups were also observed visiting I.coccinea flowers, but were less frequent pollinators, so the study was conducted mainly on butterflies as pollinating insect. The family of Butterfly that act as pollinators of I.coccinea are Papilionidae(6 species), Pieridae(3 species) and Nymphalidae(2 species).

Ixora are tubular and bloom in dense rounded clusters about 2 to 5 inches across. The tubular shape of Fragrant ixora flowers prevents many insects from gaining access to the nectar that is stored at the base of the floral tube. The nectar is only accessible to insects, such as hawkmoths, whose mouthparts are long enough to reach to the base of the floral tube. As these insects reach into the floral tube to obtain the nectar they touch the pollen producing structures, or stamens, and transport that pollen to other flowers they visit to obtain more nectar.But when the suitable insect is absent then the pollination mechanism is brought about by the insect that is available in the surrounding.As the body of butterfly is large enough so pollen stuck to it and help in transfer of pollen.Without the specialist insect pollinators to move pollen between flowers, fruit, which only develop following fertilization (of the ovule by the pollen), are not produced.

Data obtained in the present study showed that flowering season of Lcoccinea is mainly the summer.Earlier research showed that warmth is essential. These plants cannot tolerate temperatures below 15°C (59°F). The present study also report similar findings(Table4). Temperature has a profound effect on pollination particularly in poikilothermic insects. Butterflies are mainly diurnal and are mostly active in bright sunshine with relatively low humidity. Butterflies are deriving most of their heat from the sun (Owen, 1971). and are inactive early in the morning, late in the evening, at night, and during cold and wet weather (Larsen, 1991). According to our observations the frequency of butterflies visited the flowers was high during 09:00-13.00 hour (table2) and month of april to august.

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# Floristic Composition and Ecological Characteristics of Shahbaz Garhi, District Mardan, Pakistan

By Musharaf Khan, Farrukh Hussain & Shahana Musharaf

Federal Government College Mardan, Pakistan

*Abstract-* The study was designed to explore the floristic composition and biological characteristics of the area. A record of plant species of Shahbaz Garhi, Mardan was organized during 2009 – 2010. A record of plant species was organized on the source of field trips conducted in winter, summer and monsoon and identified with available literature. The plants were classified into different life form and leaf size classes after standard methods. The flora consisted of 132 plant species belonging to 104 genera and 47 families. Asteraceae and Poaceae are the dominant families. The biological spectrum explains that therophytes (63 spp., 47.73%) were the dominant followed by chamaephytes (24 spp., 18.18%), magaphanerophytes (15 spp., 11.36%), hemicryptophytes (13 spp., 9.85%), nanophanerophytes (12 spp., 9.09%), geophytes (4 spp., 3.03%) and parasite (1 spp., 0.76%). Leaf size classes of plants consisted of microphylls (62 spp., 46.97%), mesophylls (28 spp., 21.21%), nanophylls (18 spp., 13.64%), leptophylls (15 spp., 11.36%) and megaphylls (9 spp., 6.82%). Analysis of the study reveals the phytoclimate to be of therophytic type. The domination of therophytes indicates that the investigated area is under deep biotic stress.

Keywords: flora, ecological characteristics, shahbaz garhi, mardan, pakistan.

GJSFR-C Classification : FOR Code: 069999



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# Floristic Composition and Ecological Characteristics of Shahbaz Garhi, District Mardan, Pakistan

Musharaf Khan <sup>a</sup>, Farrukh Hussain <sup>o</sup> & Shahana Musharaf <sup>o</sup>

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#### I. INTRODUCTION

axonomists are naturally interested to record flora of different geographical areas. Since very long time many attempts have been through by different workers in searching away Flora of our dear native soil, Pakistan. The effort of both Pakistani and Foreign Taxonomists is basic approach. Different workers have worked in different parts of Pakistan still when it was part of United India. The area under discussion is typically unfamiliar and very a small number of reports are originated. Khan (2004) has effort on the flora of Tehsil Banda Daud Shah Karak, Khan (2007) has work on ethnobotany of Tehsil Karak. The floristic composition of Dureji (Khirthar range) was reported by Parveen et al., (2008). They recorded 74 species belonging to 62 genera and 34 families. Qureshi, (2008) identified 120 plant species belonging to 84 genera and 39 families of

Chotiari Wetland Complex, Nawab Shah, Sindh. Pakistan. Hussain, et al., (2009) reported 62 species including 15 monocots and 01 pteridophyte of 24 families from Azakhel Botanical Garden. University of Peshawar. Muhammad, et al., (2009) reported 67 weed species out of which 2 belonging to monocot families, and 27 to dicot families from wheat, maize and potato crop fields of Tehsil Gojra, District Toba Tek Singh, Puniab, Qureshi and Bhatti (2010) recorded 93 plant species belonging to 67 genera and 30 families of Pai forest, Nawab Shah, Sindh, Pakistan. Khan et al., (2011a & b), designed the ethnobotany of halophyte of Tehsil Karak and dara Adam Khel. Khan et al., (2011c) reported 161 plant species in the Tehsil Takht-e-Nasratti, District Karak where 25 monocotyledonous and 136 dicotyledonous species belonging to 52 families. Biological spectrum of vegetation is the index of the phytoclimate of the site, deduction of which is based on diverse life-forms composing the flora of the site. The life-form in its turn is the ultimate manifestation of the sum of all the adaptations undergone by a plant the climate in which it resides. Raunkiaer (1934) to proposed the term "Biological Spectrum" to express both the life-form distribution in a flora and the phytoclimate under which the prevailing life-forms evolved. Life-form study is thus an important part of vegetation description, ranking next to floristic composition. Leaf size classes have been set up to be very positive for plant links. The leaf size knowledge may help out in the accepting of physiological processes of plants and plant communities (Oosting, 1956). Life form and leaf size spectra indicates climatic and creature fracas of a particular area (Cain & Castro, 1959). The life form and leaf size spectra are significant physiognomic feature that comprise generally in vegetation studies. The life form spectra are supposed to be the signal of micro and macroclimate (Shimwell, 1971). Disturbances can have an unfathomable outcome on life forms, phenology and distribution of plant populations. Disturbances caused by man and animals such as fire, scraping and profound grazing frequently reappear within the life period of a plant and may comprise significant constituent of its life cycle (Agrawal 1989). Literature dealing with the life form and leaf size spectra shows that very little work has been made in Pakistan i.e. Abbas et al., (2010), Qureshi & Ahmad (2010) and

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Khan et al., (2011a,b, 2012, 2013). The biological spectrum is thus useful as an index of the health status of a forest. When worked out at periodic intervals, biological spectrum may set the guidelines for ecorestoration and optimization of a community. y. In view of this, the present work was under taken in the forested areas of Shahbaz Garhi, Mardan.

#### a) Location of the study area and physiography

The district lies from 34°12'0"N 72°2'24"E. The elevation of the valley is 1000 to 2056m above sea level. The total area of the district is 1632 kilometers. Mardan district may broadly be divided into two parts, North-Eastern hilly area and south western plain (Figure 1). Shahbaz Garhi is situated on the junction of three ancient routes i.e. Kabul to Pushkalavati, Swat through Buner and Taxila through Hund on the bank of Indus River. The town was once a thriving Buddhisy city surrounded by monasteries and stupas. The Emperor Babar in his book Tuzk-e-Babri has given reference of this monastery. It has also been stated that this village has named with the name of a famous religious person. In the ancient books the name of this village is Varsha pura. In 7th century, a Chinese pilgrim Mr.Haven Sang visited this monastery and recorded this polosha in his book. In local language it is called Shahbaz Garha. This is the place to take a break or rest when you are tired. It has beautiful mountains, green trees, open fields and a small river in the centre of the village. In old times all



Fig. 1 : Map of District Mardan showing research area

these facilities made it attractive for the army and travelers to dig in their tents here, stay for few days and organize their further strategy. The historic Stones of Ashoka (Figure 2) and other sites like Mekha Sanda are worth visiting. The most attractive building of the new era is the high school, this has given a new look to the ancient stones of Ashoka. The local people had put their efforts and resources in building the school. Many sites have been discovered in Mardan and it looks as Mardan was the heart of Gandhara civilization. One of the Buddhist monasteries is of Mekha Sanda, which is located 17 km from Mardan in the North Eastern side in the Hills of Shahbaz Garha (Figure 3, 4). This site was surveyed and excavated by a team of Japanese archaeologists between 1959 and 1965. During courses of excavations a good number Gandhara art sculptures, main stupa, votive stupas, monastery, chapels and Monks' chambers were found. This site became a place for research and a tourist spot. The name is derived from Pushto language. Mekha means a female buffalo and Sanda means a male buffalo. The arrangement of the stones is in such a way that it looks like buffaloes. Unfortunately some treasure hunters illegally dug out the site in search of antiques and it has been spoiled. It is the utmost responsibility of the government to provide guards, restore this site and protect it from further destruction. So far there is no sign of it happening. (Khan et al., 2011d).



Fig. 2 : Historic Stones of Ashoka in research site



Fig. 3 : View of Research area

#### II. MATERIALS AND METHODS

The study area was thoroughly surveyed during the year 2009 - 2010 from time to time to learn the botanical and biological situation with students of biology, Federal government collage Mardan. It presents a prospect to compose plant compilation and field interpretation throughout the flowering and fruiting of maximum quantity of species. Plant specimens



Fig. 4 : View of Research area

collected from the area were dried and preserved (Figure 5). They were identified from first to last available literature Nasir & Ali (1970-1994) and Ali & Qaisar (1971-2006). The plants were classified into different life form and leaf size classes as follows after Raunkiaer (1934), Muller and Ellenberg (1974) and Hussain (1989). These plant specimens were submitted to the Herbarium, Department of Botany, Federal Government College Mardan, Pakistan.



Fig. 5: Collection of plant species in research area

#### III. Result

Field survey and collection of plants were completed 2009 -2010. The current result reveled that 132 plant species belonging to 47 families and 104 genera were initiate in the area (Figure 6). Along with these presented 16 trees, 10 shrubs, 106 herb species (Figure 7). Asteraceae and Poaceae were the dominant with 15 species then Amaranthaceae, Solanaceae both by 7 species, Cucurbitaceae, Euphorbiaceae, Laminaceae and Moraceae by means of 6 species. Polygonaceae had 5 species. Chenopodiaceae and Zygophyllaceae had 4 species each. Brassicaceae, Cyperaceae and Malvaceae had 3 species each one. Apiaceae, Boraginaceae, Caesalpinaceae, Liliaceae, Myrtaceae, Nyctaginaceae, Papaveraceae, Papilionaceae, Rosaceae and Verbenaceae each and every one had 2 species. Adiantaceae, Asclepiadaceae, Cactaceae, Canabinaceae, Caryophyllaceae, Commelinaceae, Convolvulaceae, Crassulaceae, Cuscutaceae, Fabaceae, Fumariaceae, Meliaceae, Mimosaceae, Oxalidaceae, Portulacaceae, Punicaceae, Rhamnaceae, Rubiaceae, Rutaceae, Sapindaceae, Scrophulariaceae, Simaroubaceae and Tamaricaceae had 1 specie each one (Table 1).

The biological spectrum explains that therophytes (63 spp., 47.73%), chamaephytes (24 spp.,

18.18%), megaphanerophytes (15 spp., 11.36%), hemicryptophytes (13 spp., 9.85%), nanophanerophytes (12 spp., 9.09%), cryptophytes (Geophytes) (04 spp., 3.03%), parasite (1 spp, 0.76%) had originated in the investigated area (Table. 3). Leaf spectra of plants consisted of microphylls (62 spp. 46.97%), mesophylls (28 spp. 21.21%), nanophylls (18 spp. 13.64%) leptophylls (15 spp. 11.36%) and megaphylls (9 spp. 6.82%) (Table 2; Figures 8, 9).

SN	Family	Species	Habit	Life Form	Leaf size classes
1	Adiantaceae	Adiantm capillus veneris L	Н	Hem	Na
		Achyranthus aspera L	Н	TH	Mes
		Aerva javanica(Burm.f.) Shult	Н	СН	Mic
		Alternanthera sessil(L.) R.Br. ex DC	Н	СН	Mic
2	Amaranthaceae	Amaranthus spinosus L	Н	СН	Mg
		Amaranthus torreyi Benth. Exs.Watson	Н	NP	Mic
		Amaranthus viridis L	Н	TH	Mic
		Digera muricata (L.)	Н	СН	Mic
3	Aniaceae	Coriandrum sativum L.	н	TH	Lep
5	Aplaceae	Eryngium bourgatii L	Н	NP	Mg
4	Asclepiadaceae	Calotropis procera (Wight.) Ali	S	СН	Mes
		Carthamus oxycantha M. Bieb.	Н	TH	Mic
		Carthamus tinctirius L	Н	СН	Mes
		Centaurea calcitrapa L.	н	TH	Mes
		<i>Conyza aegyptiaca</i> (L.) Aiton	Н	СН	Mes
		Echinops carnigerus DC.	н	СН	Mes
	Asteraceae	Launea procumbens Roxb.	Н	TH	Mes
5	Astoração	Onopordum acanthium L.	н	СН	Na
5	Asteraceae	Parthenium hysterophorus L	н	TH	Mes
		Silybum marianum (L.) Gaertn.	н	TH	Mes
		Sonchus arvensis L.	н	TH	Mes
	2 Amaranthaceae 3 Apiaceae 4 Asclepiadaceae 5 Asteraceae 6 Boraginaceae 7 Brassicaceae	Sonchus asper (L.) Hill	н	TH	Mic
		Sonchus auriculata L	н	TH	Mic
		Sylibum marianum (L) Graertn	н	СН	Mic
		Taraxacum officinale Weber.	н	TH	Mic
		Xanthium strumarium L.	Н	СН	Mes
6	Boraginaceao	Heliotropium europaeum L.	Н	TH	Na
0	Durayinaceae	Heliotropium strigosum Willd	Н	TH	Lep
		Capsella bursa-pestoris Medic.	Н	TH	Mic
7	Brassicaceae	Descurainia sophia (L.) Webb.	Н	TH	Na
		Eruca sativa Mill	Н	TH	Mic

Table 1 : Florist	ic list of Shahbaz	Garhi, District	Mardan
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8	Cactaceae	<i>Opuntia littoralis</i> (Engelm.)	S	NP	Lep
		Cassia fistula L.	Т	MP	Mes
9	Caesalpinaceae	Cassia occidentalis L.	Н	TH	Mes
10	Canabinaceae	Cannabis sativa L.	Н	TH	Mic
11	Caryophyllaceae	Stellaria media (L.) Cry	Н	TH	Na
-		Chenopdium ambrosiodes L	Н	TH	Mic
10		Chenopodium album L.	Н	TH	Mic
12	Chenopodiaceae	Chenopodium murale L.	Н	TH	Lep
		, Spinacea oleracea L.	Н	TH	Mic
13	Commelinaceae	, Commelina communis L.	Н	TH	Mic
14	Convolvulaceae	Convolvulus arvensis L.	Н	TH	Mic
15	Crassulaceae	Sedum acre L	Н	TH	Mic
-		<i>Citrullus lanatus</i> (Thunb.) Mats	Н	TH	Mes
		Cucimus prophetarum L.	Н	TH	Mes
10	Our such its seaso	Cucurbita maxima Duchesne.	Н	TH	Mg
10	16 Cucurbitaceae	Cucurbita pepo L.	Н	TH	Mg
		Luffa cylindrica (L.) Roem.	Н	TH	Mg
		Momordica charantia L.	Н	TH	Mes
17	Cuscutaceae	Cuscuta reflexa Roxb.	Н	Р	Lep
		Cyperus compresus L	Н	Hem	Lep
18	Cyperaceae	Cyperus rotundus L.	Н	Hem	Lep
		<i>Cyperus scarlosus</i> R.Br.	Н	Hem	Lep
		Chrozophora oblique (Vahl) A. Juss.	Н	СН	Mes
		Chrozophora tinctoria (Linn) Raffin.	Н	NP	Mic
10	Cyperaceae	<i>Euphorbia helioscopia</i> Mewski	Н	TH	Na
19		Euphorbia hirta L.	Н	TH	Mic
		Euphorbia prostrata L.	Н	TH	Lep
	19 Euphorbiaceae 20 Fabaceae	Riccinis communis L.	S	NP	Mg
20	Fabaceae	Indigofera hirsute L	Н	СН	Na
21	Fumariaceae	Fumaria indica (Hausskn) Pugsley	Н	TH	Lep
		Ajuga bractiosa Wall. Benth.	Н	TH	Mic
		Ajuga parviflora Benth	Н	TH	Mic
22	Laminaceae	Mentha arvensis L.	Н	Geo	Mic
22	Laminaceae	Mentha longifolia L.	Н	Geo	Mic
		Ocimum basilicum L	Н	СН	Mic
		Selvia moorcroftianaWall. ex Benth	Н	СН	Mg
23	Liliaceae	Allium sativum L.	Н	Geo	Mic
20		Oxalis caniculata L	Н	TH	Na
		Abelmoschus esculentus L.	Н	TH	Mic
24	Malvaceae	Malva neglecta Wallr.	Н	TH	Mic
		Malvastrum coromandelianum(L.) Garcke	Н	TH	Mic

25	Meliaceae	Melia azedarach L.	Т	MP	Mic
26	Mimosaceae	Acacia modesta Wall.	Т	MP	Lep
		Broussonitia papyrifera(L.) Vent	Т	MP	Mg
		Ficus carica Hausskn. Ex. Boiss.	Т	MP	Mes
07	Managara	<i>Ficus palmata</i> Forssk.	Т	MP	Mes
27	Moraceae	Ficus religiosa L.	Т	MP	Mes
		Morus alba L.	Т	MP	Mes
		Morus nigra L.	Т	MP	Mes
00	Murtaaaaa	<i>Eucalyptus camaldulensis</i> Dehnh.	Т	MP	Mic
28	Mynaceae	Eucalyptus lanceolatushoney	Т	СН	Mic
		<i>Boerhaavia procumbens</i> Banks ex	Ц	СЦ	Mic
29	Nyctaginaceae	Nuxu.		СП	Maa
30	Oxalidaceae	Will addits jarapa L.			Mia
00	Oxandaoodo		н	Geo	IVIIC
31	Papaveraceae	Papaver moeas L	<u> </u>		IVIIC
		Papaver somniterum L.	H		MIC
32	Papilionaceae	Ainagi maurorum Medic.	5		Na
			<u>Н</u>	IH 	Na
		Avena sativa L.	H	IH	Mic
		Bromus japonicus Thumb ex Murr	H	Hem	Mic
		Cenchrus ciliaris L.	H	TH	Na
		Chymbopogon jawaracosa L.	Н	TH	Mic
		<i>Cymbopogon distans</i> (Nees ex Steud.)Watson	Н	Hem	Mic
		Cynodon dactylon L. Pers.	Н	Hem	Lep
33	Poaceae	Daicanthium annulatumForssk.) Stapf	Н	TH	Na
00	TUaceae	Desmostachya bipinnata(L)	Н	Hem	Mes
		Hardeum murinum L	Н	TH	Na
		Hordeum vulgare L	Н	TH	Mic
		Imperata cylindrica (L.) P. Beauv	Н	Hem	Mic
	3 Poaceae	Phalaris minor L	Н	СН	Mic
		Saccharum spontaneum L.	S	Hem	Mic
		Sorghum halepense (L.) Persoon	Н	Hem	Mic
		Zea mays L.	Н	TH	Mg
		Polygonum barbatum L.	Н	СН	Mic
34	Polygonaceae	Polygonum plebjum R. Br.	Н	СН	Mic
		Rumex dentatus L.	Н	TH	Mes
		Rumex hastatus D.Don	Н	TH	Na
		Rumix dantatus L.	Н	TH	Lep
35	Portulacaceae	Portulaca olearaceae L.	Н	Hem	Na
36	Punicaceae	Punica granatum L.	Т	MP	Na
37	Rhamnaceae	Ziziphus jujuba Mill.	т	MP	Mic

38	Bosaceae	Prunus persica (L.) Batsch	Т	MP	Mic
00	nosaccac	Rosa indica L.	S	NP	Mic
39	Rubiaceae	Gallium aparine L.	Н	TH	Lep
40	Rutaceae	Citrus aurantifolia Christmann	S	NP	Mic
41	Sapindaceae	<i>Dodonaea viscosa</i> (L.) Jacq.	S	NP	Mic
42	Scrophulariaceae	Verbascum traipses L	Н	NP	Mic
43	Simaroubaceae	Alianthus althesema (Mill.) Swingle	Т	MP	Mic
		Datura metel L.	S	NP	Mes
		Datura stramonium L.	Н	NP	Mes
44	44 Solanaceae	Physalis minima L	Н	СН	Mic
		Solanum nigrum L.	Н	TH	Mic
		<i>Solanum surattense</i> Burm.f	Н	TH	Mic
	<i>Withania somnifera</i> (L) Dunal.	S	СН	Mes	
45	Tamaricaceae	<i>Tamarix indica</i> Willd.	Т	MP	Na
46	Verbenaceae	Lantana camara L.	Н	СН	Mic
40	Verbendeede	Verbena hastata L.	Н	NP	Mic
		<i>Fagonia cretica</i> Burm.	Н	TH	Na
47	Zugophyllaceae	Peganum harmala L	Н	Hem	Mic
77	Zygoprynaceae	Tribulus terrestris L.	Н	TH	Mic
	45Tamaricaceae46Verbenaceae47Zygophyllaceae	Zygopylum simplex L.	Н	TH	Lep

#### IV. DISCUSSION

The work will indubitably present much help out to future investigator assets trying in this field in this area. The area consists of both hills and plains, differing much in floristic composition. Irrigation facilities are very less in the area, depending on rainfall. Due to lack of irrigation conveniences the Flora, particularly cultivated Flora has much difference from highly irrigated areas of Khyber Pakhton Khawa. The chief Agriculture crops are Wheat, different legumes, fodder crops and barely, grown. On hills different grasses, Acacia modesta, Achyranthus aspera, Calotropis procera, Carthamus oxycantha, Conyza aegyptiaca, Xanthium strumarium, Opuntia littoralis, Sorghum halepense and Fagonia cretica etc are commonly found. Mostly the Xerophytes such as Broussonitia papyrifera(L.) Vent, Ficus carica Hausskn. Ex. Boiss., Ficus palmata Forssk., Morus alba L., Eucalyptus camaldulensis Dehnh., Eucalyptus lanceolatus honey etc are found on road sides while Melia azedarach L., Ficus religiosa L., Prunus persica (L.) Batsch, Alianthus althesema (Mill.) Swingle, Tamarix indica Willd etc. are commonly found in Grave-yards. Such type of study was also taken by Khan et al., (2011a,b, 2012, 2013). With the passage of time, increase in population and rising in need of facilities in the culture declining the natural habitats. Our result is similar with that of Khan et al., (2012). The natural assets are being over-used, unclear and spoil. In the research

area, commonly people depend on agricultural and domestic animals. They also collect medicinal plants, fodder, fuel wood and timber.

According to the Raunkiaer (1934) that climate of a region is characterized by life form. Plant species were identified and classified into major life forms to build biospectrum. The biological spectra is helpful to comparing geographically far and wide separated plant communities and used as an indicator of prevailing environment. Biological spectrum may be significantly changed due to preface of therophytes like annual weeds, biotic pressure like agricultural practices and grazing, deforestation and trampling etc. The dominance of therophytic life form showed that the area was under heavy biotic pressure. Our results agree with that of Khan et al., (2011a,b) and Khan, et al., (2012). Comparisons of the percentage of the life form classes of the research area with Raunkiaer standard biological spectrum (RSBS), therophyte form the largest life form class and their percentage is more than thrice (47.73%) that of the RSBS (13.0%). The phanerophytes forms, the second highest class with (21.21%). Their percentage was 46.0 in the RSBS. Thus, the biological spectrum of the research area marker "Therophytic" Phytoclimate at the same time as this class proves the greatest deviation from the standard spectrum. Hemicryptophyte is equal (9.85 %) with that of the RSBS (9.00 %). Cryptophytes was less percentage 3.03 than in the RSBS (6.00 %) (Table. 3). In this study, the domination

of therophytes and phanerophytes over other life forms give the idea to be a response with to the warm dried up weather, topographic divergence, human being and creature disturbance. The dominance of therophytes occurs due to un-favorable environment conditions as definite by a lot of research (Shimwell, 1971, Khan et al., 2011c, 2012). The current results in this regard also agree with them. Khan et al., (2012) considered chamaephytes and therophytes as the major life form in unfavorable environment in desert region. In the investigated area arid conditions, low temperature in winter, high temperature in summer, wind and biotic factors result in un-favourable conditions paving way for Saxina et al.. therophyte. (1987) stated that hemicryptophytes dominated temperate zone in overlapping and loose continuum. Therophytes continue in unfavorable condition during seeds production. The predominance of therophytes in unstable conditions such as dry, hot or cold met for low to higher elevation might be the reason for their higher percentage in the present study.

The present study shows that leptophylls were high at the hilly area while microphylls and nanophylls were present in plain area. Species with large leaves take place in warmer wet climates while smaller leaves are characteristic of cold and arid climates and degraded habitats. A high percentage of microphylls might be due to dry climate in area. Leaf size spectrum of the plant revealed that microphyllous species followed by nanophylls species were dominant in the investigated area. Microphylls are usually characteristic of steppes while nanophylls and leptophylls are characteristic of hot deserts (Khan et al., 2013; Tareen & Qadir, 1993). The soil was poorly developed with thin sheet that banned root penetration. Furthermore, roots absorb low moisture and nutrients under dry conditions. In this region's the plant face drought during winter especially in dry soil. The species with microphyllous

leaves were abundant due to ecological adaptation for these arid conditions. The present findings agree with those of Khan 2013 who reported high percentage of microphylls in the dry climate of Tehsil Takht-e-Nasratti. These data indicated that the percentage of various leaf form classes varied with increasing altitude. Khan (2013) and Khan et al., (2013) also observed that the percentage of microphylls was positively linked with the increasing altitude and this also hold up our findings. According Dolph & Dilcher (1980 a, b) large leaved species were dominant in tropical wet forest. This difference is mainly due to climatic variation such as temperature and wet tropical condition. The situation in our case is far more xeric than in the wet tropics. The size of leaves alone could not be used to identify specific leaf zone or climates. Other features of plants such as habit and root system might also play important role in biodiversity.

An ecologically operating problem of the area is grazing, browsing, and trampling by domestic animals (Figure 10). These elements cause species not to reach its climax stage. Grazing is one of the depressing aspects, which has caused the reduction in vegetation (Khan and Hussain, 2012). In these processes the palatable species are selected and these make the nonpalatable species to increase. This can be noticeably seen in many places, which results in stunting growth and not reaching to flowering stage: so these are a danger of their extinction. The most important factors disturbing the Flora of area are humidity, light, temperature, soil conditions, topography, elevation from sea level, rain fall and other forms of precipitation. On soil having high Nitrogen content are found Malva neglecta, Chenopodium album etc, as occurring near human duellings, on compost heaps and in back yards. The finding is similar with that of Khan et al., (2012, 2013), Khan and Hussain (2013) and Khan (2013).

*Table 2 :* Total number of plant species and percentage of life-form and leaf size classes of Shahbaz Garhi District Mardan

Life-form classes	No. of species	Percentage	Leaf size classes	No. of species	Percentage
Therophytes	63	47.73	Microphylls	62	46.97
Chamaephytes	24	18.18	Mesophylls	28	21.21
Megaphanerophytes	15	11.36	Nanophylls	18	13.64
Hemicryptophytes	13	9.85	Leptophylls	15	11.36
Nanophanerophytes	12	9.09	Megaphylls	9	6.82
Geophytes	4	3.03			
Parasite	1	0.76			

Table 3 : Comparison of biological spectrum of the area with Raunkiaer's Standard Biological Spectrum (SBS).

Spectrum	PP	ChP	ТР	HP	CrP	Total	
RSBS	46	26	13	9	6	100	
Current study	21.21	18.18	47.73	9.85	3.03	100	
Deviation in Percentage	24.79	7.82	-34.73	-0.85	2.97	0	



Fig. 6: Family of plant species recorded in research area





*Fig. 7:* Habit of plant species in research area

Fig. 8 : Life form classes of plant species in research area



Fig. 9: Leaf size classes of plant species in research area

#### V. Conclusion

The region is extremely prosperous in biodiversity. In the current study, the high percentage of therophyte is supported in the study region for the reason that the region is semiarid zone of Khyber Pakhtonkhawa. The dominance of therophytes indicated that the investigated area was under heavy biotic pressure due to deforestation and over grazing. Most of the plants were uprooted for burning purposes and grazed by the livestock. Many plant species were decreasing in the area and special care is needed for their plant life conservation. Many fruits are worn out annually due to non-availability of marketplace. The market convenience has fine result on plants and on nation. Medicinal farm should be set up in the area to support the essential importance of the plants and its conservation.

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Fig. 10 : Goat graze plant species in research area

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# Restoration of Degraded Lands through Plantation Forests

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*Abstract-* Degradation of soil is a matter of serious concern. Vast area of land all over the world has been converted into unproductive and degraded lands. Eco-restoration through plantation forests is the most effective technique to reclaim the degraded ecosystem. Six dominant species viz, Dalbergia sissoo, Pongamia pinnata, Tectona grandis, Gmelina arborea, Azadirachta indica and Cassia siamea were studied for restoration of degraded ecosystem. No amendment was given during plantations except farm yard manure (FYM), Urea and Aldrin as soil insecticide before planting the seedlings. The density of plants was 3333 ha-1. The present paper deals with the edaphic development of degraded coal mine spoil through establishment of six dominant tree species at Northern Coalfield Limited, Singrauli. The results indicated that the bulk density of the reclaimed sites was gradually reduced with the age of the plantations. The soil organic carbon, pH, EC, water holding capacity and nutritional status were found increasing with the age of the plantations. As regards the organic carbon in mine spoil under different tree cover, it was found improved to the maximum extent during 16 years interval in Dalbergia sissoo (358%) followed by Azadirachta indica (317.8%), Pongamia pinnata (273.8%), Tectona grandis (233.3%) and others. The similar increasing trend was found in pH.

Keywords: restoration, plantation forests, degraded lands, productive ecosystem, dominant tree species.

GJSFR-C Classification : FOR Code: 829999

# RESTORATION OF DEGRADED LANDSTHROUGHPLANTATIONFORESTS

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# Restoration of Degraded Lands through Plantation Forests

Priyanka Bohre<sup> a</sup> & O.P. Chaubey<sup> o</sup>

Abstract- Degradation of soil is a matter of serious concern. Vast area of land all over the world has been converted into unproductive and degraded lands. Eco-restoration through plantation forests is the most effective technique to reclaim the degraded ecosystem. Six dominant species viz, Dalbergia sissoo, Pongamia pinnata, Tectona grandis, Gmelina arborea, Azadirachta indica and Cassia siamea were studied for restoration of degraded ecosystem. No amendment was given during plantations except farm yard manure (FYM), Urea and Aldrin as soil insecticide before planting the seedlings. The density of plants was 3333 ha-1. The present paper deals with the edaphic development of degraded coal mine spoil through establishment of six dominant tree species at Northern Coalfield Limited, Singrauli. The results indicated that the bulk density of the reclaimed sites was gradually reduced with the age of the plantations. The soil organic carbon, pH, EC, water holding capacity and nutritional status were found increasing with the age of the plantations. As regards the organic carbon in mine spoil under different tree cover, it was found improved to the maximum extent during 16 years interval in Dalbergia sissoo (358%) followed by Azadirachta indica (317.8%), Pongamia pinnata (273.8%), Tectona grandis (233.3%) and others. The similar increasing trend was found in pH. The electrical conductivity was the maximum in Tectona grandis followed by Azadirachta indica, Dalbergia sissoo, Gmelina arborea, Cassia siamea and Pongamia pinnata. There was gradual increase in microbial biomass from younger to older plantations in different dominant species. It ranged from 40.2 (2 years old plantation) to 51.5 mg kg-1 (18 years old plantation) in T. grandis, from 32.5 (2 years old plantation) to 66.6 mg kg-1 (18 years old plantation) in D. sissoo, from 21.2 (2 years old plantation) to 52.7 mg kg-1 (18 years old plantation) in A. indica, from 35.5 (2 years old plantation) to 50.3 mg kg-1 (19 years old plantation) in C. siamea, from 24.2 (2 years old plantation) to 54.7 mg kg-1 (18 years old plantation) in P. pinnata and from 31.7 (6 years old plantation) to 42.6 mg kg-1 (10 years old plantation) in G. arborea. As far as, the concentration of heavy metals like Cu, Zn, Fe, and Mn was concerned, their concentration in soil of rhizosphere decreased with the age of the plantations.

*Keywords:* restoration, plantation forests, degraded lands, productive ecosystem, dominant tree species.

#### I. INTRODUCTION

Restoration is defined as a tactic employed to return degraded lands to its original condition. With the planned increase in coal production, more and more land is being brought under mining operation. The most serious impact of mining is the land

Authors α σ: State Forest Research Institute, Jabalpur. e-mails: pribohre@gmail.com, chaubey.dr@gmail.com degradation, and ecosystem as a whole. Overburden degraded soils consist of several impediments for plant growth in respect of physical, chemical and biological properties, and also associated with deficiency/non availability of organic matter. The heavy metal toxicity of mine spoil inhibits uptake of nutrients, plant growth and microbial populations.

Selection of ideal species for restoration of mined out areas is very important step in degraded ecosystem (Mukhopadhyay et al., 2013). Soil nutrients can be taken as a functional index of soil development after reclamation. Soil microorganisms play significant role in soil fertility and ecosystem functioning. The demand for a particular mineral nutrient depends on plant internal requirements, while the supply of that nutrient primarily depends on its availability and mobility in soils (Allen et al., 1979; Parfitt and Russell, 1977; Marschner, 1995, Marschner and Timonen, 2004; Chiti et al., 2007). Without microbes and their functions, plant species could not be supported by the soil alone (Kennedy and Smith, 1995; Filcheva et al., 2000). Mineral nutrients such as phosphorus have very limited mobility in soils (Parfitt and Russell, 1977; Insam and Domsch, 1988; Marschner, 1995; Booze et al., 2000; Boswell et al., 1998, Bucking and Shachar, 2005). Thus to obtain more phosphorus, plants must bypass the depletion zones by further root activity elsewhere in the soil. The outcome of this quest for phosphorus (and other relatively immobile soil resources) should largely be determined by the surface area of a plant's root system. The most important role of mycorrhizal fungal hyphae is to extend the surface area of roots. The capacity of plants to influence nutrient availability in soils will also depend on the extensiveness and activity of their root system, since young roots are the primary source of exudates (Curl and Truelove, 1986; Uren and Reisenaur, 1988; Warcup 1990; Nguyen, 2003; Jeffries et al., 2003). Soil microbial biomass measurements were useful in determining the degree of disturbance as well as subsequent recovery of degraded ecosystems (Sandra Brawn, 2002; Chaubey et al., 2012; Tran Van Con, 2001; Vo Dai Hai, 2009; Singh et al., 2012a, b).

The present paper deals with the restoration of degraded ecosystem aiming at studying the changes brought about in physical, chemical and biological properties of soil under the tree cover of dominant tree species in age series of plantations carried out at Northern Coalfield Limited (NCL), Singrauli, India.

#### II. MATERIALS AND METHODS

Singrauli (24° 46' 60"- 24° 78' 33"N, 82° 49' 59"- 82° 83' 30"E, 275 -500m AMSL) was granted district status on 24th May 2008, with its headquarter at Waidhan. Climate of the area is tropical with mean maximum and minimum temperature of 48°C and 21°C respectively, with average rainfall of 1000 mm. 95% precipitation occurs in rainy season. Vegetation during pre-mining period was very dense and covered with northern tropical dry sal forests and northern tropical dry mixed deciduous forests. Due to mining, the large forest areas were clear felled. The present study was carried out in age series of plantations raised on different dumps of mined out Northern Coalfield Limited (NCL) area.

A large number of over burden mixed and monoculture plantations are being continuously raised by M.P. State Forest Development Corporation/ U.P. forest department in different areas of Singrauli coalfields. The species planted were mainly Acacia catechu (L. f.) Willd, A. mangium Willd, A. nilotica (L.) Del., Aegle marmelos (L.) Correa, Albizia procera (Roxb.) Benth., Anthocephalus kadamba (Roxb.) Mig., Azadirachta indica A. Juss., Bauhinia variegata Linn., Bombax ceiba (L.) Gaertn, Cassia fistula L., C. siamea Lamk., Dalbergia sissoo Roxb. ex DC., Delonix regia (Hook.) Raf., Emblica officinalis Gaertner., Eucalyptus camaldulensis Dehnh., Gmelina arborea Roxb., Holoptelia integrifolia (Roxb.), Leucaena leucocephala (Lam.) de Wit, Madhuca indica Roxb., Mangifera indica L., Peltaphorum sp (Miquel) Kurz, Pongamia pinnata (L.) Pierre, Prosopis juliflora (Sw.) DC., Syzygium cumini (L.) Skeels, Tectona grandis L. f., Terminalia arjuna (Roxb.) Wight & Arn., T. belerica (Gaertner) Roxb., etc. The spacing of plants between row to row and plant to plant was 1.5 m and 2 m, respectively. The plants were raised in pits of size 45cm3. No amendment was given during plantations except farm yard manure (FYM), Urea and Aldrin as soil insecticide. Poly-potted seedlings of 6 months old were taken for plantations.

The phyto-sociological study was carried out for determining the dominant species in the coal mine areas of Singrauli, India (Mishra, 1989). Importance Value Index and Shannon Wiener Index of diversity were used for determining the dominant species in different plantations. On the basis of Importance Value Index (IVI) and diversity index (H), the dominant species, viz, Dalbergia sissoo (IVI- 35.80, H- 0.2537), Pongamia pinnata (IVI - 26.52, H - 0.2144), Tectona grandis (IVI-23.82, H- 0.2011), Gmelina arborea (IVI - 22.60, H - 0.1948), Azadirachta indica (IVI - 22.55, H - 0.1945) and Cassia siamea (IVI - 16.40, H - 0.1589), were selected for the study.

To know the effect of plantations on soil properties, soil samples from rhizosphere of six dominant species were collected from surface soil up to the depth of 30 cm. Five samples from different aged plantations of each species were collected and mixed thoroughly to get a composite sample and then, were divided into three replicates for analysis of physicochemical properties and microbial biomass. Soil organic carbon was determined by Black (1956) method. The physico-chemical and nutritional properties (N, P, K) of soil were analyzed using soil testing methods by Jackson (1976) and Piper (1950). The chloroform fumigation extraction method (Carter, 1991) was used for estimation of microbial biomass. The microbial biomass was expressed on the oven dry (105°C for 24 hours) soil basis. Microbial biomass was correlated with the nutritional characteristics using IBM SPSS statistics 19 software.

#### III. Results

Soil nutrients can be taken as a functional index of soil development after reclamation. The slightly acidic to neutral pH under the different plantation forests was suitable for greater availability of nutrients, decomposition of litter and microbial activity especially bacteria and VAM fungi that decomposed organic matter and released nitrogen (Chaubey et al., 2004). On perusal of results (Table - 1), the pH has improved to a great extent in Dalbergia sissoo (30.5%) followed by Azadirachta indica (28.1%), Pongamia pinnata (23.8%), Tectona grandis (22.5%) and others after 16 years interval of plantations. The electrical conductivity has improved to a great extent in 16 years interval in Tectona grandis (233.3%) followed by Azadirachta indica (220%), Dalbergia sissoo (142.9%), Cassia siamea (90%) and Pongamia pinnata (83.3%) and others. The bulk density reduced to the maximum extent in Dalbergia sissoo (180.9%) followed by Azadirachta indica (133.3%), Tectona grandis (63.6%), and others during 16 years interval of plantations. The nutritional status was also showing more or less similar trend in different dominant species and was found to be increasing with the advancement of age of different dominant species. As regards the organic carbon in mine spoil under different tree cover, it was found improved to the maximum extent during 16 years interval in Dalbergia sissoo (358%) followed by Azadirachta indica (317.8%), Pongamia pinnata (273.8%), Tectona grandis (233.3%) and others. The improvement during the period of 16years in terms of available nitrogen was maximum in Pongamia pinnata (227.9%) followed by Azadirachta indica (143.9%), Dalbergia sissoo (157.7%), Tectona grandis (127.8%) and others. The improvement during the period of 16-years in terms of available phosphorus was maximum in Dalbergia sissoo (199.8%) followed by Azadirachta indica (62.1%), Pongamia pinnata (52.4%), Tectona grandis (17.7%) and others. The improvement during the period of 16-years in terms of available potassium was maximum in Dalbergia sissoo (262.2%)

followed by Pongamia pinnata (190.3%), Tectona grandis (133%), Cassia siamea (121.1%), Azadirachta indica (92.3%), and others. The improvement during the period of 16-years in terms of calcium was maximum in Tectona grandis (154%) followed by Dalbergia sissoo (115.9%), Pongamia pinnata (113%), Cassia siamea (35.9%), Azadirachta indica (33.8%) and others. As far as the concentration of heavy metals like Cu, Zn, Fe, and Mn are concerned, it was found that their concentration in soil of rhizosphere decreases with increasing the age of the plantations. There was gradual increase in microbial biomass from younger to older plantations in different species. It ranged from 40.2 (2 years old plantation) to 51.5 mg kg-1 (18 years old plantation) in Tectona grandis, from 32.5 (2 years old plantation) to 66.6 mg kg-1 (18 years old plantation) in Dalbergia sissoo, from 21.2 (2 years old plantation) to 52.7 mg kg-1 (18 years old plantation) in Azadirachta indica, from 35.5 (2 years old plantation) to 50.3 mg kg-1 (19 years old plantation) in Cassia siamea, from 24.2 (2 years old plantation) to 54.7 mg kg-1 (18 years old plantation) in Pongamia pinnata and from 31.7 (6 years old plantation) to 42.6 mg kg-1 (10 years old plantation) in Gmelina arborea.

The nutritional characteristics like organic carbon, available nitrogen and available phosphorus maintained significant positive correlation with microbial biomass. The most significant correlation between organic carbon and microbial biomass was found in Azadirachta indica (95%) followed by Gmelina arborea (93%) Pongamia pinnata (79%), Cassia siamea (79%), Tectona grandis (63%) and Dalbergia sissoo (51%). However, the most significant correlation between available nitrogen and microbial biomass was found in Gmelina arborea (99%) followed by Tectona grandis (94%), Pongamia pinnata (91%), Cassia siamea (81%), Dalbergia sissoo (80%) and Azadirachta indica (75%). In terms of available phosphorus and microbial biomass, the most significant correlation was found in Gmelina arborea (99%) followed by Pongamia pinnata (92%), Azadirachta indica (91%), Cassia siamea (82%), Dalbergia sissoo (82%) and Tectona grandis (72%) (Table 2).

*Table 1 :* Physico-chemical and biological properties of soil in plantation forests of Tectona grandis Linn. f., Dalbergia sissoo Roxb., Azadirachta indica A. Juss., Cassia siamea Lamk, Pongamia pinnata (Linn) Pierre and Gmelina arborea Roxb.

Species	Age		Physico-chemical properties and microbial biomass												
	(yr)	Bulk	Water	pН	EC (ms	Organi	Availab	Availa	Availabl	Calcium	Cu	Zn	Fe (mg	Mn (mg	Microbi
		density	holding		cm⁻¹)	С	е	ble	е	(kg ha <sup>-1</sup> )	(mg	(mg	kg⁻¹)	kg⁻¹)	al
		(g cm <sup>-3</sup> )	capacity			carbon	nitroge	phosp	potassi		kg 1)	kg 1)			biomas
			(%)			(%)	n (kg	horus	um (kg						s (mg
							ha⁻')	(kg ha⁻¹)	ha")						kg⁻')
T. grandis	2	1.80	18.16	5.96	0.03	0.82	180	8.60	63.5	167	0.83	2.98	23.50	0.72	40.2 ±
_		±0.07	±0.91	±0.42	$\pm 0.003$	±0.03	±7.54	±0.70	±4.45	±13.36	±0.07	±0.24	±2.12	$\pm 0.06$	2.41
T. grandis	8	1.60	20.15	6.01	0.04	0.90	345	8.82	69.3	235	0.69	2.50	12.30	7.40	46.1
		±0.08	±1.41	$\pm 0.48$	$\pm 0.004$	$\pm 0.05$	$\pm 10.00$	$\pm 0.45$	±6.24	$\pm 16.45$	$\pm 0.06$	±0.23	$\pm 0.98$	±0.67	±3.23
T. grandis	9	1.50	21.29	6.06	0.05	0.93	355	8.85	70.3	256	0.64	2.61	14.30	7.90	47.4
		±0.09	±1.28	$\pm 0.42$	$\pm 0.004$	±0.06	$\pm 10.00$	$\pm 0.40$	±5.62	±15.36	$\pm 0.04$	±0.26	$\pm 1.05$	±0.55	±3.79
T. grandis	10	1.40	22.72	7.24	0.09	1.84	398	9.50	137.0	381	0.61	2.10	12.06	6.35	48.7
		±0.13	±1.82	$\pm 0.36$	$\pm 0.006$	±0.16	±11.13	$\pm 0.47$	±8.22	±19.05	$\pm 0.07$	±0.19	$\pm 0.96$	±0.51	±3.90
T. grandis	18	1.10	28.25	7.30	0.10	2.20	410	10.12	148.0	425	0.40	1.70	8.65	4.35	51.5
		±0.08	±1.98	$\pm 0.51$	$\pm 0.008$	±0.09	±18.02	$\pm 0.96$	±13.32	$\pm 34.00$	$\pm 0.05$	±0.14	±0.87	±0.44	±4.64
D. sissoo	2	1.77	26.80	5.31	0.07	0.36	187	6.67	50.8	176	1.38	2.36	13.64	14.61	$32.5 \pm$
		±0.14	±3.22	±0.42	$\pm 0.006$	±0.04	±15.23	±0.48	±3.56	±15.84	±0.15	±0.24	±1.23	±1.17	2.28
D. sissoo	4	1.60	32.94	5.68	0.07	0.48	215	7.60	69.0	222	1.39	2.29	13.46	14.57	$34.3 \pm$
		±0.14	±2.64	±0.62	±0.008	±0.06	±20.41	±0.54	±4.14	±22.20	±0.14	±0.25	±1.48	±1.02	2.40
D. sissoo	5	1.50	33.10	5.81	0.07	0.50	267	10.90	74.0	267	1.36	2.26	13.41	14.49	36.4 ±
		±0.11	±2.98	±0.58	±0.007	±0.07	±18.32	±0.78	±7.40	±21.36	±0.11	±0.18	±1.07	±1.30	2.91
D. sissoo	6	1.32	34.75	5.73	0.07	1.00	225	11.90	78.0	292	1.29	2.17	12.93	14.02	38.6 ±
		±0.12	±3.82	±0.69	±0.008	±0.013	±20.55	±085	±7.02	±26.28	±0.14	±0.20	±0.91	±1.47	2.70
D. sissoo	7	1.09	36.60	6.44	0.08	1.44	278	12.90	89.0	334	1.17	2.10	12.40	12.57	39.7 ±
		±0.11	±3.66	±0.71	±0.009	±0.19	±29.57	±0.93	±9.79	±36.74	±0.14	±0.25	±1.49	±1.51	2.78
D. sissoo	8	1.06	39.50	6.46	0.09	1.44	380	13.40	94.0	338	0.98	1.90	11.67	11.79	40.2 ±
		±0.13	±4.74	±0.65	±0.009	±0.19	±33.15	±0.96	±9.40	±33.80	±0.12	±0.23	±1.17	±1.36	3.22
D. sissoo	9	0.96	37.66	6.49	0.09	1.46	286	14.45	95.5	351	0.93	1.73	10.58	10.71	41.8 ±
		±0.08	±3.01	±0.78	±0.011	±0.19	±30.48	±1.04	±7.64	±28.08	±0.10	±0.14	±0.85	±0.86	2.93

(n=3 in each plantations; values are mean $\pm$ standard deviation	on)
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Species	Age	Physico-chemical properties and microbial biomass													
	(yr)	Bulk	Water	pН	EC (ms	Organi	Availabl	Availa	Availabl	Calcium	Cu	Zn	Fe (mg	Mn (mg	Microbi
		density	holding		cm⁻¹)	C	e.	ble	e	(kg ha <sup>-1</sup> )	(mg	(mg	kg⁻¹)	kg⁻¹)	, al
		(g cm™)	capacity			carbon	nitroge	pnosp	potassi		kg⁻')	kg⁻')			biomas
			(%)			(%)	n (Kg ha <sup>-1</sup> )	lloius (ka	ha <sup>-1</sup> )						s (mg ka <sup>-1</sup> )
							114)	ha <sup>-1</sup> )	110 )						'' <b>'</b> 9)
D. sissoo	10	0.92	38.20	6.51	0.09	1.49	288	14.52	96.8	357	0.91	1.58	9.85	10.65	42.5
Daiaaaa	44	±0.08	±4.20	±0.72	±0.010	±0.21	±32.80	±1.08	± 10.65	±39.27	±0.10	±0.17	± 1.03	±1.31	±3.40
D. SISSOO	11	0.87 +0.10	38.75	0.55 +0.66	0.10	1.52	290 +33.10	14.80	98.0 +0.80	360	0.87 +0.00	1.47 +0.10	9.20	10.57	43.6 ±
Π είεερο	13	0.84	39.67	6.63	0.010	1.56	328	15.80	100.0	368	0.09	0.84	7.05	<u> </u>	3.00
<i>D.</i> 313300	10	±0.08	±4.76	±0.80	±0.010	±0.23	±34.25	$\pm 1.17$	$\pm 13.08$	±44.16	$\pm 0.09$	±0.10	±0.87	±1.01	3.56
D. sissoo	14	0.76	42.20	6.67	0.11	1.60	362	16.30	138.0	373	0.64	0.76	6.70	6.90	47.2 ±
		±0.07	±3.88	±0.60	±0.011	±0.23	±35.11	±1.17	±12.42	±39.17	±0.05	±0.08	±0.67	±0.62	3.78
D. sissoo	15	0.70 +0.06	42.30 +3.38	6.70 +0.74	0.11	1.64 +0.23	363 +37.12	17.60 +1.22	147.0 +16.17	376 +41.36	0.63 +0.07	0.60	5.44 +0.49	6.19 +0.47	51.3 ±
D sissoo	16	0.70	42.30	671	0.16	1.64	367	17.90	156.0	376	0.63	0.50	4.60	842	545+
D. 010000	10	±0.07	±3.81	±0.87	±0.021	±0.26	±38.68	±1.30	±15.60	±37.60	±0.08	±0.04	±0.44	±0.70	3.27
D. sissoo	18	0.63	44.00	6.93	0.17	1.65	482	20.00	184.0	380	0.61	0.20	3.54	4.70	66.6 ±
		±0.06	±4.84	±0.76	±0.019	±0.23	±40.40	±1.45	±20.24	±41.80	±0.05	±0.01	±0.30	±0.45	5.33
A. indica	2	1.75	17.43	5.13	0.05	0.73	164	8.33	91.0	157	1.74	2.68	13.83	11.28	21.2
A ' I'	_	±0.16	±1.83	$\pm 0.52$	$\pm 0.005$	$\pm 0.06$	±11.48	$\pm 0.75$	$\pm 7.92$	$\pm 14.44$	$\pm 0.14$	±0.21	$\pm 1.13$	±1.04	±1.48
A. INGICA	5	1.40 ±0.15	21.30 ±2.45	5.59 ±0.63	0.12 ±0.009	$1.35 \pm 0.09$	225 ±18.00	9.55 ±0.96	115.0 ±11.04	$\pm 212$	1.74 ±0.17	1.67 ±0.15	$\pm 0.91$	9.20 ±0.80	25.1 ±2.01
A. indica	6	1.15	23.61	5.68	0.13	1.69	267	9.90	131.0	332	1.56	1.32	9.99	7.71	29.3
		±0.10	±2.24	±0.48	±0.011	±0.10	±26.70	±0.89	±11.00	±26.23	±0.12	±0.09	±0.92	±0.59	±2.64
A. indica	7	1.03	25.75	5.81	0.14	1.73	329	10.70	140.0	385	1.52	1.08	7.07	6.42	30.7
		±0.07	±2.21	±0.54	±0.013	±0.14	±23.03	±0.64	$\pm 10.50$	$\pm 31.96$	±0.12	±0.08	$\pm 0.62$	±0.61	±2.46
A. indica	8	0.98	26.06	5.89	1.42	1.79	358	11.12	156.0	387	1.45	1.05	6.23	5.90	32.6
		±0.09	±2.06	±0.48	±0.144	±0.11	±28.68	±1.11	±12.79	±29.41	±0.17	±0.09	±0.49	±0.51	±2.28
A. Indica	10	0.95	27.30 +2.59	5.97 +0.55	0.15 +0.018	1.95 +0.14	365 +29.20	12.30	160.0 +11.04	389 +35.79	1.39 +0.14	1.03	5.10 +0.42	5.60 + 0.59	36.2 +3.26
A indica	14	0.85	32.40	6.25	0.15	2.25	390	12.60	166.0	395	1.19	0.93	4.65	3.55	42.5
		±0.07	±2.66	±0.63	±0.014	±0.16	±35.10	±1.26	±13.78	±44.24	±0.10	±0.10	±0.35	±0.40	±3.40
A. indica	18	0.75	33.11	6.57	0.16	3.05	400	13.50	175.0	210	0.73	0.78	4.00	2.00	52.7
		±0.06	±3.05	$\pm 0.58$	±0.012	±0.27	$\pm 38.50$	±1.49	$\pm 14.70$	±18.27	$\pm 0.06$	±0.07	$\pm 0.39$	±0.17	±4.22
C. siamea	2	1.80	20.23	5.25	0.10	0.23	137	17.50	95.0	217	1.85	2.98	14.30	11.60	35.5
O stance	~	±0.17	±1.72	$\pm 0.48$	$\pm 0.009$	$\pm 0.02$	$\pm 6.02$	±1.58	$\pm 7.79$	$\pm 19.96$	±0.15	$\pm 0.22$	±1.19	±1.01	±2.49
C. siamea	6	1.70 ±0.15	22.65 ±1.70	5.74	0.11	0.25	142 + 7.10	18.00	103.0	230 ±16.70	1.82	2.90	14.50	11.50 +1.16	41.6 + 2.01
C ciamoa	11	±0.15	±1.72	±0.00	$\pm 0.010$	$\pm 0.03$	±7.10	±1.20	$\pm 7.52$	±10.79	±0.14	±0.23 ೧.07	$\pm 1.13$	± 1.10	±2.91
C. Slainea		±0.12	±2.64	±0.60	$\pm 0.009$	±0.02	±11.92	±1.54	±11.13	±19.43	±0.16	±0.20	±1.07	±1.21	±3.56
C. siamea	14	1.51	24.90	6.10	0.12	0.39	161	19.57	135.0	265	1.67	2.70	13.80	9.80	46.4
		±0.14	±3.14	±0.68	±0.010	±0.03	±11.27	±1.56	±9.05	±24.38	±0.12	±0.22	±1.26	±1.22	±4.18
C. siamea	15	1.41	30.53	6.16	0.15	0.47	181	21.52	166.7	281	1.46	2.33	12.97	8.50	48.7
O stance	10	±0.12	±2.66	$\pm 0.55$	$\pm 0.014$	$\pm 0.03$	±14.48	±1.72	$\pm 13.84$	±20.51	±0.16	±0.22	±1.45	$\pm 0.74$	±4.38
c. siamea	19	±0.13	32.75 ±3.14	±0.60	0.19 ±0.017	0.56 ±0.04	±15.44	22.95 ±1.84	≥10.0 ±22.05	295 ±21.24	1.24 ±0.11	∠.10 ±0.18	12.00 ±1.04	о.40 ±0.44	50.3 ±5.03
P. pinnata	2	1.79	21.06	5.25	0.06	0.42	136	8.53	68.2	216	0.93	3.65	11.17	12.26	24.2
,		±0.19	±1.94	±0.46	±0.005	±0.03	±10.88	±0.68	±6.27	±18.79	±0.08	±0.30	±1.03	±1.07	±1.74
P. pinnata	4	1.67	22.72	5.61	0.06	0.89	164	9.20	91.0	242	0.85	3.41	10.50	10.31	26.3
		±0.13	±2.61	±0.63	$\pm 0.004$	±0.07	±13.12	±0.83	±6.83	±18.15	±0.06	±0.26	±0.91	$\pm 1.00$	±2.05
P. pinnata	5	1.62	23.17	5.83	0.07	0.96	184	10.20	121.0	258	0.72	3.21	9.37	9.76	27.6
		±0.19	±1.81	±0.48	±0.005	±0.08	±16.54	±0.71	±8.11	±19.61	±0.04	±0.22	±0.98	±1.05	±2.26
P. pinnata	6	1.54 +0.15	23.58	5.06	0.08 +0.009	0.99	258 + 22 22	10.30	124.0	282	0.69	2.71	9.21	8.70 +0.07	29.4
P ninnata	8	1 49	26.52	5.89	0.008	1 15	268	10.00	160.0	208	0.00	2 40	8.36	8.40	32 7
, . pii ii aid	0	±0.11	±1.78	±0.66	±0.008	±0.09	±18.76	±0.97	±17.92	±27.42	±0.04	±0.20	±0.63	±0.73	±2.91

Species	Age		Physico-chemical properties and microbial biomass												
	(yr)	Bulk	Water	pН	EC (ms	Organi	Availabl	Availa	Availabl	Calcium	Cu	Zn	Fe (mg	Mn (mg	Microbi
		density	holding		cm <sup>-1</sup> )	С	е	ble	е	(kg ha⁻¹)	(mg	(mg	kg⁻¹)	kg⁻¹)	al
		(g cm-3)	capacity			carbon	nitroge	phosp	potassi		kg⁻¹)	kg⁻¹)			biomas
			(%)			(%)	n (kg	horus	um (kg						s (mg
							ha⁻¹)	(kg ha <sup>-1</sup> )	ha⁻¹)						kg⁻¹)
P ninnata	0	1.40	33.03	5.05	0.00	1 16	208	11.00	170.0	310	0.57	2 3 2	7.26	7 30	35.8
τ. ριπιαια	9	+0.12	+2.58	+0.53	+0.09	+0.10	+20.86	+0.99	+22.73	+21.08	+0.07	+0.16	+0.70	+0.55	+3.29
P ninnata	10	1 35	12.00	6.00	0.007	1 22	305	11 20	183.0	330	0.51	2 27	6 50	6.40	37.2
1 . pii ii lata	10	+0.12	+3.90	+0.00	+0.03	+0.12	+21.35	+1.20	+15.02	+41 91	+0.01	+0.24	+0.60	+0.54	+3.46
P ninnata	13	1.30	40.49	6 14	0.10	1.26	326	11 60	189.0	.348	0.49	2 20	5.30	5 70	40.4
, . p lata	10	±0.14	±3.16	$\pm 0.48$	$\pm 0.011$	±0.13	$\pm 26.08$	$\pm 0.81$	$\pm 12.29$	$\pm 33.06$	$\pm 0.03$	±0.26	±0.46	±0.43	±3.80
P. pinnata	14	1.27	44.75	6.16	0.10	1.29	330	12.10	191.0	380	0.46	2.15	4.70	4.90	43.9
, , <i>p., , ,</i> , , , , , , , , , , , , , , , ,	•••	±0.11	±4.12	±0.70	±0.011	±0.13	±29.70	±0.85	±20.06	±43.70	±0.04	±0.16	±0.49	±0.50	±4.48
P. pinnata	15	1.25	49.01	6.25	0.10	1.32	334	12.40	193.0	410	0.43	2.00	4.10	4.20	46.6
,		±0.08	±5.39	±0.66	±0.009	±0.13	±30.06	±1.12	±14.67	±31.98	±0.05	±0.19	±0.39	±0.36	±4.80
P. pinnata	18	1.20	50.39	6.50	0.11	1.57	446	13.00	198.0	460	0.37	1.90	3.90	4.00	54.7
		±0.10	$\pm 4.59$	$\pm 0.60$	±0.010	±0.14	±31.22	±1.04	±24.75	±51.29	±0.02	±0.15	$\pm 0.30$	±0.32	±6.02
G. arborea	6	1.76	22.72	5.18	0.04	0.30	150	9.40	37.1	207	0.72	2.90	8.76	8.42	31.7
		±0.15	±1.98	$\pm 0.58$	$\pm 0.003$	±0.02	±9.5	$\pm 0.70$	±3.15	±23.18	$\pm 0.06$	±0.28	$\pm 0.76$	$\pm 0.73$	±2.95
G. arborea	9	1.58	34.47	5.58	0.06	0.68	245	9.85	46.8	253	0.68	2.45	7.13	6.24	39.4
		±0.12	±3.34	±0.49	$\pm 0.005$	$\pm 0.05$	±20.83	±0.89	±3.56	±22.01	$\pm 0.05$	±0.26	$\pm 0.54$	$\pm 0.47$	$\pm 3.70$
G. arborea	10	1.20	48.39	5.86	0.10	1.11	270	10.00	58.6	380	0.61	2.20	4.96	4.26	42.6
		±0.10	±5.23	$\pm 0.45$	$\pm 0.008$	±0.11	±22.95	$\pm 0.80$	±5.92	$\pm 28.88$	$\pm 0.06$	±0.25	$\pm 0.42$	$\pm 0.36$	$\pm 4.35$

Table 2: Correlation between nutritional and microbial biomass of tree species

		Corre	ations			
			Organic carbon (%)	Available nitrogen (kg ha <sup>-1</sup> )	Available phosphorus (kg ha <sup>-1</sup> )	Microbial biomass (mg kg <sup>-1</sup> )
	Organic carbon (%)	Pearson correlation	1	0.701**	0.783**	0.779**
		Sig. (2-tailed)		0.004	0.001	0.001
lis I		N	15	15	15	15
	Available nitrogen	Pearson correlation	0.701**	1	0.614*	0.948**
ang	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.004		0.015	0.000
gri		N	15	15	15	15
na	Available phosphorus	Pearson correlation	0.783**	0.614*	1	0.766**
ecto	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.001	0.015		0.001
Τε		N	15	15	15	15
	Microbial biomass	Pearson correlation	0.779**	0.948**	0.766**	1
	(mg kg <sup>-1</sup> )	Sig. (2-tailed)	0.001	0.000	0.001	
		N	15	15	15	15
	Organic carbon (%)	Pearson correlation	1	0.776**	0.912**	0.733**
		Sig. (2-tailed)		0.000	0.000	0.000
		N	42	42	42	42
00	Available nitrogen	Pearson correlation	0.776**	1	0.882**	0.894**
issi	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.000		0.000	0.000
a s		N	42	42	42	42
l <i>G</i> lé	Available phosphorus	Pearson correlation	0.912**	0.882**	1	0.913**
elbe	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.000	0.000		0.000
Dé		N	42	42	42	42
	Microbial biomass	Pearson correlation	0.733**	0.894**	0.913**	1
	(mg kg <sup>-1</sup> )	Sig. (2-tailed)	0.000	0.000	0.000	
		N	42	42	42	42
rac a	Organic carbon (%)	Pearson correlation	1	0.897**	0.947**	0.966**
adi: hta idic		Sig. (2-tailed)		0.000	0.000	0.000
4Z, ij		N	24	24	24	24

		Coner				
			Organic carbon (%)	Available nitrogen (kg ha <sup>-1</sup> )	Available phosphorus (kg ha <sup>-1</sup> )	Microbial biomass (mg kg <sup>-1</sup> )
	Available nitrogen	Pearson correlation	0.897**	1	0.958**	0.859**
	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	.000		0.000	0.000
		N	24	24	24	24
	Available phosphorus	Pearson correlation	0.947**	0.958**	1	0.935**
	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.000	0.000		0.000
		N	24	24	24	24
	Microbial biomass	Pearson correlation	0.966**	0.859**	0.935**	1
	(mg kg <sup>-1</sup> )	Sig. (2-tailed)	0.000	0.000	0.000	
		N	24	24	24	24
	Organic carbon (%)	Pearson correlation	1	0.952**	0.954**	0.896**
		Sig. (2-tailed)		0.000	0.000	0.000
		N	18	18	18	18
Ë	Available nitrogen	Pearson correlation	0.952**	1	0.984**	0.892**
neć	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.000		0.000	0.000
siar		N	18	18	18	18
assia s	Available phosphorus	Pearson correlation	0.954**	0.984**	1	0.915**
	(kg ha <sup>-1</sup> )	Sig. (2-tailed)	0.000	0.000		0.000
<i>C</i>		Ň	18	18	18	18
	Microbial biomass	Pearson correlation	0.896**	0.892**	0 915**	1
	$(ma ka^{-1})$	Sig (2-tailed)	0.000	0.000	0.000	•
	(	N/	18	18	18	18
	Organic carbon (%)	Pearson correlation	1	0.934**	0.960**	0.874**
		Sig (2-tailed)	1	0.000	0.000	0.000
		N	33	33	33	33
ita	Available nitrogen	Pearson correlation	0.934**	1	0.948**	0.950**
nna	$(kg ha^{-1})$	Sig (2-tailed)	0,000		0.000	0.000
iid	(	N	33	33	33	33
nia	Available phosphorus	Pearson correlation	0.960**	0.948**	1	0.924**
gaı	$(kg ha^{-1})$	Sig (2-tailed)	0,000	0,000		0.000
μο	(	N	33	33	33	33
4	Microbial biomass	Pearson correlation	0.874**	0.950**	0.924**	1
	$(ma ka^{-1})$	Sig (2-tailed)	0,000	0,000	0.000	•
	(	N	33	33	33	33
	Organic carbon (%)	Pearson correlation	1	0.999**	0.998**	0 294
		Sig (2-tailed)		0,000	0.000	0.443
		N	9	9	9	9
a	Available nitrogen	Pearson correlation	0.999**	1	0 999**	0.270
Ore	$(kg ha^{-1})$	Sig (2-tailed)	0,000		0.000	0.483
arb	(	N	9	9	9	9
la ć	Available phosphorus	Pearson correlation	0.998**	0 999**	1	0.258
nelii	$(ka ha^{-1})$	Sig. (2-tailed)	0.000	0.000	, i	0.502
Gn	( ) /	N	9	9	9	9
	Microbial biomass	Pearson correlation	0.294	0.270	0.258	1
	$(ma ka^{-1})$	Sig (2-tailed)	0.443	0.483	0.502	
	( e'' e'')	Λ/	Q.440	Q.700	0.002 Q	Q
		/ <b>v</b>	9	3	3	9

#### IV. DISCUSSION

Under different dominant species, the pH range was suitable for greater availability of nutrients, decomposition of litters that decomposed organic matter and

released nitrogen, and is in agreement with the findings of Chaubey et.al. (2004). After 16-year of plantation, the pH improved to maximum extent in Dalbergia sissoo followed by other species. The elemental variation of soil under different species cover was also due to natural
variation of planted sites or dumps. The capacity of species to influence nutrient availability of soil will also depend on the extensiveness and activity of root system, since young roots are the primary source of exudates (Jeffries et. al., 2003). The results may be attributed to the variation in nitrogen content of leaves and rate of litter decomposition, plant canopy and age of different species (George and Kohli, 1957; Puri, 1959; Down, 1975; Rimmer, 1982; Banerjee et al., 1999; Banerjee et al., 2000; Prakasham and Banerjee, 2001; Jha and singh, 1991; Nandeswar et al., 1996). The results were also in agreement with the findings of reclamation of coal mine spoils with Juwarkar and Jambhulkar (2009), Nath (2009), Jain et al. (2009). Moreover, Dutta and Agarwal (2002) assessed soil characteristics of vegetated Northern Coal Field Limited (NCL) under plantations of five exotic species (Acacia auriculiformis, Casuarina equisetifolia, Cassia siamea, Eucalyptus hybrid and Grevelia pleridifolia) and found improved soil status under different plantation stands of 4-year compared to over burden and Eucalyptus hybrid, Acacia auriculiformis and Casuarina equisetifolia were most suitable in terms of modification of spoil characteristics during the revegetation process. The nutritional status was showing increasing trend with the age of plantations of dominant species. As far as the variation in soil nutrients among different species is concerned, it may be due to the plant and microbial interactions occur in different way with the advancement of the age. Moreover, the bulk density values of the reclaimed sites have gradually reduced with increasing the age of the plantations. As regards the concentration of heavy metals like Cu, Zn, Fe, and Mn was concerned, it was found that their concentration in soil of rhizosphere decreased with the increase in the age of the plantations. It may be due to sorption and desorption characteristics of soil and substantial amount of organic matter (Krishnamurti et al., 1999; Chaubey et al., 2012). The metals can also be sequestered in cellular structure becoming unavailable for translocation to the shoot (Lasat et al., 1998).

These species were good for nitrogen fixation. With increasing the age of species, the microbial biomass also increased in different species. The findings were comparable with the observations recorded by Daft and Nicoloson (1974), Gupta and Shukla (1991), Jamaluddin and Chandra (2009), Chaubey et al. (2012) in different studies.

The results indicated that organic carbon, available nitrogen, available phosphorus were the good indices of microbial biomass. However, the best positive correlation between microbial biomass and nutritional characteristics was found with available phosphorus followed by available nitrogen and available carbon in different dominant species (Curl and Truelove, 1986; Uren and Reisenaur, 1988). The results were also in agreement with the observation of Banerjee et al. (2000), who reported a significant positive correlation between the number of organisms and organic carbon in coal mine spoil of Gevra colliery. Soil microbial biomass was useful in determining the degree of recovery of degraded ecosystem and nutritional budget. Microbial activity is reported to improve gradually during restoration of mine spoils (Stroo and Jeneks, 1982).

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# Study and Optimization of Palm Wood Mechanical Properties by Alkalization of the Natural Fiber

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Abstract- This Study is devoted to the characterization of mechanical properties of a date palm Wood fiber (DPF).We propose to measure mass loss, tensile strength, Young modulus and elongation at failure. The use of natural fibers requires specific chemical treatments to address mechanical performance due to water absorption. For this reason an alkaline (NaOH) treatment of different samples at different concentrations was carried. We submit after that the samples to a Thermo gravimetric analysis (TGA) to measure the influence of soda treatment on the mass loss. In a second time, mechanical properties were studied of untreated and treated samples. Thus we can access to the elasticity limit, tensile strength and Young modulus E. The results led us to conclude that treatment of Palm fibers with soda at different concentrations results in a significant improvement of the mechanical properties.

Keywords: mechanical properties; palm fiber; tensile strength; young modulus.

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# Study and Optimization of Palm Wood Mechanical Properties by Alkalization of the Natural Fiber

M. Tlijania <sup>°</sup>, A. Gouadriab <sup>°</sup>, R. Benyounesc <sup>P</sup>, Jf. Durastantid <sup>©</sup> & A. Mazioude <sup>¥</sup>

Abstract- This Study is devoted to the characterization of mechanical properties of a date palm Wood fiber (DPF).We propose to measure mass loss, tensile strength, Young modulus and elongation at failure. The use of natural fibers requires specific chemical treatments to address mechanical performance due to water absorption. For this reason an alkaline (NaOH) treatment of different samples at different concentrations was carried. We submit after that the samples to a Thermo gravimetric analysis (TGA) to measure the influence of soda treatment on the mass loss. In a second time, mechanical properties were studied of untreated and treated samples. Thus we can access to the elasticity limit, tensile strength and Young modulus E. The results led us to conclude that treatment of Palm fibers with soda at different concentrations results in a significant improvement of the mechanical properties.

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#### I. INTRODUCTION

n the recent years, with the strong emphasis on environmental awareness, scientists and technologists have placed so much importance on the application of natural materials. This move has encouraged industries like furniture, automotive, building construction, and packaging to search for new form of berk composites that can substitute the conventional composite materials.

Unfortunately, some drawbacks such as poor wet ability, incompatibility with some matrix and high moisture absorption by the fibers make them undesirable for certain applications [1, 2, 3]. The main problem often encountered in their use is the fiber – matrix adhesion problem due to the incompatibility between the hydrophilic natural fibers and the hydrophobic matrix. This problem may be improved by a chemically treating fiber surface. Therefore, alkaline treatment is a common method to clean and modify the fiber surface to lower surface tension and enhance matrix [4]. That is why several publications have discussed the effects of alkaline treatment on structure and properties of natural fibers, such as kenaf [5], hemp [5], flax [6], jute [7] and sisal [8].

In this context this study was prepared and divided into two major parts:

In the first part stability and durability of the Date Palm Frond (DPF) Fibers are investigated. It is worth noting that one of the difficulties and disadvantages which impedes the development of natural fiber use in industry and in the manufacture of composites is their poor dimensional stability due to water absorption, that is why several authors do have to study the effects of chemical treatments on the properties of natural fibers to improve their characteristics and whence comes the utility of thermo gravimetric analysis performed on samples types. These analyses were carried out on crude and treated fiber to characterize the degradation of the DPF palm fibers and consequently measure the samples mass variation as a function of time or temperature. The mode chosen to analyze the DPF fibers in this case is the isothermal mode in which measurement is done at constant temperature and the measured parameter is the evolution of the mass.

The samples proposed for thermo gravimetric analysis, were extracted from the Date Palm Frond (DPF) (figure 1).

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Fig. 1 : Shematic representation of a Date Palm Frond

This extraction comes from the Stem, the Cluster and the Basel End of the DPF (figure 2).



Fig. 2 : Test Piece of Basel End DPF

And subsequently treated with a sodium hydroxide (NaOH) solution. This alkaline treatment was conducted with concentration of 0.5%, 0.75% and 1% NaOH. The variation of concentration was made to optimize the treatment parameter. The DPF palm fibers were immersed in NaOH solution at various concentrations for an hour and at temperature of 100 ° C , and after that they were rinsed with distilled water until the rinsed solution reached neutral (PH 7). Then, fibers were dried at room under atmospheric pressure, temperature 23  $\pm$  2 ° C and humidity 50 %  $\pm$  5 % for ten days.

While the Second part discusses the influence of chemical treatment on the mechanical properties of the Date Palm Frond Fibers. Therefore, fore specimens of date palm Frond, untreated and treated with concentrations of sodium hydroxide (NaOH: 0.5 %, 0.75 % and 1%) were tested for tensile property determination and were examined under scanning electron microscope (MEB) to study the microstructure of the materials.

### II. Dimensional Stability of the Date Palm Frond (dpf)

#### a) Alkalization treatment (NaOH treatment)

Alkalization is a common preprocessing technique used on base natural fiber to remove hemicelluloses, fats and waxes that may reduce the interfacial strength between the resin and matrix when processed into composite form and often results in a change in fiber surface energy in a polar or dispersive manner. Hemicelluloses, which is thought to consist principally of xylan, polyuronide and hexosan, has been shown to be very sensitive to Caustic Soda. The Caustic Soda (Sodium Hydroxide) is said to exert only minimal influence on the lignin in the fibers and the high strength alpha-cellulose. Therefore ,It is of great interest to understand the effect this treatment has on the base fiber mechanical properties, Indeed, the major effect was the increasing of the resultant composite strength through increasing fiber matrix adhesion. It is additionally beneficial to investigate current literature to aid in understanding other effects that alkalization may have. These include transformation of cellulose type, and also improved ability for microfibrils to rearrange to accommodate loading of the fiber [9-14].

#### b) Thermo gravimetric Analyses

In order to characterize the stability and durability of the Date palm Frond (DPF), a (TGA) Thermo gravimetric analysis was performed on crude and treated fibers with concentrations of sodium hydroxide (NaOH: 0.5%, 0.75% and 1%). This measure is to characterize the degradation of a material with increasing temperature in which the mass variation of a sample is measured as a function of time. The measured parameter is the evolution of the mass.

#### c) Objective

The main purpose of this section was to measure the change in DPF remaining residual mass as the concentration of the NaOH treatment changes.

#### d) Thermal Characterization

Thermo gravimetric analysis (TGA) measurements were performed in air atmosphere using (TA instrument GA 2950) at a heating rate of 10 C/min. At the end of assessing the effect of treatment on different types of samples, we plotted as an example the curve figure (3) which represents the residual mass versus time for different concentration of sodium hydroxide treatment.



*Fig. 3 :* Residual Mass variation of Raw and Treated Basel end DPF with Soda solution (0.5%-0.75%-1%) Versus Time

This figure shows the variations of mass ratios versus time respectively for typical DPF fibers samples of Basel End Palm Wood. In another hand, the figure (4) inform us on how vary the remaining residual mass percentage of the different DPF samples when the amount of NaOH used for the treatment increase. These

curves are represented respectively in black, blue, red and green. The dark curve shows the response of an untreated fiber, the blue curve shows the response of a fiber previously treated at a concentration of 1% NaOH, the red one at 0.5% NaOH while the last green one at 0.75% NaOH.



*Fig. 4 :* Variation of the Residual mass of Cluster, The Stalk and The Basel End DPF Versus Sodium Hydroxide concentration NaOH(%)

By analyzing the curves of the figures mentioned above, we note the following:

- The mass decreases with time until a stable minimum value corresponding to the maximum heating.
- The variation of sodium hydroxide treatment concentration influences the value of the residual mass. In fact, for the three samples of DPF, Basel End, Stem and Cluster, the residual mass increase from 0% NaOH (untreated DPF sample) to 0.75 % NaOH which corresponds to the minimum mass loss. However it decreases for 1% NaOH.
- After stabilization, the difference between the residual mass of an untreated sample (0%) and treated with 0.75% is approximately: 0.486% for the Basel End DPF, 1.06% for the cluster DPF and 1.125% for the stem DPF.
- Beyond a certain concentration of NaOH, we note that mass loss increased significantly ( blue curve: concentration of 1 % NaOH ), this is explained by



Fig. 5 (a) : Untreated Basel End DPF with 0% NaOH



#### Fig. 5 (C) : Basel End DPF treated 0.75%NaOH

## IV. Mechanical Properties of Alkalized Fiber Date Palm Frond

#### a) Tensile test

The trials of push-ups are made according to the standard ISO37/2005 and the method used is (Dynamométrie sur ZWICK), the mechanical properties measured were tensile strength, Young's modulus, and elongation to break of the DPF specimens. This ISO 37/2005 Standard is typically used to quantify the mechanical properties. Tensile tests were performed the fact that the internal structure of wood is quote drops till and starts to degrade. Thus the optimal concentration for treatment in alkaline NaOH is of the order of 0.75 %.

#### III. MORPHOLOGY ANALYSIS

Microscopic examinations were carried out using a HITACHI S3200N scanning electron microscope (SEM) to study fiber morphology. Prior to those analysis, the first step is to take a part of the sample of the wood without altering the structure, the material is immersed in nitrogen liquid for a minute, then a piece of a few centimetres is collected by breaking the structure of the material. This technique helps to avoid the formation of ridges that can interfere with observation so that sample, be well observed in the electron microscope. The SEM micrographs for untreated and treated DPF samples were analyzed, some examples of micrographs for Basel end DPF Fiber are shown in figures (5a, 5b, 5c, 5d)



Fig. 5 (b) : Basel End DPF treated 0.5% NaOH



Fig. 5 (d) : Basel End DPF treated 1%NaOH

using a universal testing machine 1455 WN model 116942. A load-cell with a capacity of 2 KN was used to monitor the applied load to the alkalized fiber; the specimens were tested at 2 mm/min rate. The room temperature tests were carried out at  $23^{\circ}C \pm 2$  with a controlled room humidity of 50 ±5%. Each sample of DPF included three or more specimens. The dimension of the specimen used to carry out test was adapted from ISO 37/2005, for tensile testing. All these testing were carried out for untreated and treated DPF. The last step is to calculate the elastic modulus and tensile strength from the stress–strain curve.

#### *b)* Tensile strength

The specimens were tested for tensile property determination. Consistent results were obtained for tensile Strengths, which proved the effectivness of the treatment. The mechanical properties of the DPF before and after NaOH treatment at different concentrations are shown in Figure (6).



#### Fig. 6: Average tensile strength of Basel End DPF Fiber Versus alkali concentration (NaOH)

The maximum tensile strength was reported at 0, 75% NaOH treatment. As soda concentration increases the fiber become cleaner of its impurities and later improves the tensile strength from 0% NaOH through 0.5% NaOH to 0,75% NaOH treatment to exceed 4,28 Mpa for Basel End DPF Fiber, 62,04 Mpa for Cluster DPF Fiber and135,04 Mpa for Stalk or Stem DPF Fiber.

However, it is interesting to note, as soda concentration increases and reaches 1% NaOH, the solution attacks the main construction components of the fiber and more grooves appear on the surface of the fiber. Improvement in tensile strength of DPF was observed when soda treatment was applied. This results in further weakening in fiber strength, so the tensile strength start to decrease. As it is known, natural fibers are usually composed of cellulosic materials cemented together with weaker materials. The deterioration mechanism has been explained to be due to the attack of the cementing materials rendering the cellulose chains unconnected and hence unable to carry any load. Eichhorn et al. reported in there review that high concentration of caustic soda results in a decrease of fiber tensile strength due to notched grooves at the plant fibers surface [15].

#### c) Tensile modulus

Tensile modulus is a measure of rigidity of the material. The effect of the alkali treatment for the DPF

fiber provides enhancement of their rigidity for all conditions (different alkali concentrations). It means, there is a significantly increasing in tensile modulus with the increase in the alkali concentration. Figure 7 show that the maximum tensile modulus was provided by 0, 75% alkali concentration. But, the most important conclusion from these results is the significant enhancement of the tensile modulus of DPF fiber with the alkali treatment



Alkali Concentration of Sodium Hydroxide (NaOH)

Fig. 7: Average tensile modulus of Basel End DPF Fiber Versus alkali concentration (NaOH)

Rong et al. [16] reported that the alkali treatment for sisal fibers provides the improved crystallinity of cellulose and remove the hemi-cellulose and lignin content. Then, it suggests that sisal fiber becomes relatively ductile after the removal of some hemi-cellulose and lignin. The fibers can result in higher fiber stiffness due to the increased crystallinity of hard cellulose. For the case of DPF fiber, similar reason for the improvement in the tensile modulus is viewed. The average tensile strength, tensile modulus and elongation failure for each sample of groups untreated and treated DPF Fibers were calculated as the mean value of the carried out measurement on all the specimens tested and shown for example for Basel End DPF Palm Fibetr in table 1.

Table 1 : Mechanical properties of Raw and treated Basel End DPF with Soda Solution (0, 5%-0, 75% - 1 %)

Concentration of NaOH treatment	Tensile Modulus (GPa)	Tensile Strength (MPa)	Elongation Failure (%)
Basel End DPF at 0% (Untreated.)	0,23±0,005	0,73±0,260	3,39±1,01
Basel End DPF at 0.5%	0,975±0,181	3,70±0,940	1,06±0,34
Basel End DPF at 0,75%	1,150±0,256	4,28±1,420	0,96±0,58
Basel End DPF at 1%	0,951±0,295	4 <b>.16</b> ±1,240	0,69±0,51

#### V. Conclusion

From this study, we conclude that the alkaline treatment has significantly improved the tensile properties of the DPF. This enhancement in tensile strength and modulus is attributed to the improved wetting of alkali treated fiber by removal of impurities and waxy substances from the fiber surface and the creation of a rougher topography after alkalization, thus the mechanical interlocking and the interface quality will be promoted. The hydrophilic nature of DPF palm fiber has been reduced due to this treatment, the content of hemicellulose and lignin decreased, thereby an increase on the effectiveness of the orientated cellulose fibers , the tensile strength and a considerable improvement in surface morphology were observed. The result indicates that the treatment at the condition of 0.75% NaOH is the optimum treatment which gives the maximum tensile strength, tensile modulus, the minimum mass loss and the better surface morphology of the DPF palm fiber. Thermal analysis of DPF fiber shows that soda treated fibers have better thermal resistance compared to raw fibers which due to the repellent action of the treatment of the sample to the phenomenon of water absorption. However, at higher alkaline concentrations 1% NaOH, the effect of these parameters on tensile properties is so pronounced because at this condition, fiber damages may have been dominant. The results obtained in this study encourage us to integrate DPF Palm fiber as reinforcement in a given matrix by a prior chemical treatment at 0.75% NaOH that we can remedy to its reliable mechanical performance before its integration.

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# Genetic Proof of Chromatin Diminution under Mitotic Agamospermy

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*Abstract-* The previously published data are examined on the base of the hypothesis about the existence of chromosomes differential polyteny and excessive chromatin diminution during the first stages of sugar beet plant embryogenesis. It has been concluded that available data provide the genetic proof of that chromatin diminution is one of the mechanisms underlying the origin of polymorphism in sugar beet agamospermous progenies.

*Keywords:* isozymes; polyteny; diminution; agamospermy; non-mendelian inheritance; sugar beet.

GJSFR-C Classification : FOR Code: 060499



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# Genetic Proof of Chromatin Diminution under Mitotic Agamospermy

Evgenii V. Levites

Abstract- The previously published data are examined on the base of the hypothesis about the existence of chromosomes differential polyteny and excessive chromatin diminution during the first stages of sugar beet plant embryogenesis. It has been concluded that available data provide the genetic proof of that chromatin diminution is one of the mechanisms underlying the origin of polymorphism in sugar beet agamospermous progenies.

*Keywords: isozymes; polyteny; diminution; agamospermy; non-mendelian inheritance; sugar beet.* 

#### I. INTRODUCTION

bulk of evidence has been obtained for polymorphism in agamospermous diploid sugar beet plant (Beta vulgaris L.) progenies. One of the explanations of such polymorphism is based on the recognition of the important role of mixoploidy in plants. Mixoploidy is manifested by an admixture of tetraploid cells among the bulk of diploid archespore mother plant cells (Maletskii, Maletskaya, 1996; Maletskaya E.I., Maletskaya, S.S., 1999). The entering of a tetraploid cell into meiosis leads to the diploid embryo sac formation and, accordingly, to the formation of a diploid egg cell, capable of entering into embryogenesis without fertilization. This mechanism is characteristic for meiotic diplospory which can be also designated as meiotic agamospermy (Levites, 2002). In this case polymorphism is a natural consequence of meiosis and can be designated by the known term "autosegregation" (Gustafsson, 1946-1947; Maletskii et al., 1998). Genetic and cytological data support this hypothesis (Szkutnik, 2010).

At the same time, an additional mechanism has been proposed to explain polymorphism in agamospermous sugar beet plant progenies (Levites, 2005, 2007). It suggests that polymorphism occurs mostly due to the polytenization of chromosomes regions carrying marker loci. Differential polyteny could subsequently lead to a random equiprobable loss of excess chromatin by a cell before it enters embryogenesis.

Theoretical calculations indicate that the differential chromosome polytenization and subsequent diminution of excess chromatin are possible both under meiotic agamospermy and mitotic agamospermy

(adventive embryony) when an offspring arises from the somatic cells which have not undergone meiotic genome transformations. There is also genetic evidence that polytenization can occur in egg cells chromosome regions under sexual plant reproduction (Levites and Kirikovich, 2013a). A genetic proof of this hypothesis has been obtained along with the proof that the polytenization process depends on external conditions (Levites and Kirikovich, 2013b).

The studies of agamospermous progenies, as well as the consideration of chromosome polytenization. provide a new insight into many genetic processes and the causes of numerous variations in the genotype and phenotype ratios of the resulting offspring. A characteristic feature of polymorphism under agamospermy is the mismatch between the identified ratios and the normal Mendelian ratios.

Accounting for the effect of polytenization of chromosome regions carrying marker genes on the respective marker traits segregation expands the boundaries of genetics. At present, trait segregation can be attributed both to changes in the cell chromosomes number (meiosis and gamete fusion) and to other process not attributable to such changes (chromosome endoreduplicated sites diminution).

The facts collected since the early studies have contributed to a gradual shift in our view at the mechanisms underlying agamospermy. At this stage it is necessary to review our earlier data, which is the aim of this article.

Under discussion will be the data presented in the article entitled "Pseudosegregation in the agamospermic progeny of male sterile plants of the sugar beet (*Beta vulgaris* L.)" (1999), (Authors: Levites E.V., Shkutnik T., Ovechkina O.N. and Maletskii S.I.). Isozymes were used as genetic marker traits in this work.

A wonderful peculiarity of isozymes is their codominant inheritance due to which the hybrid plant isozyme spectrum is different from each parent isozyme spectrum (Schwartz, 1966; Scandalios, 1969). For instance, one isozyme with fast (phenotype FF) or slow (phenotype SS) electrophoretic mobility, which corresponds to the genotype of a given locus, is revealed in the electrophoregram for the homozygote *FF* or *SS* on the gene controlling this marker enzyme. But both enzyme allelic variants (isozymes) and also hybrid isozymes are revealed in the heterozygote (phenotype

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FS). This allows one to reveal all 3 phenotypic classes in the progeny of plant heterozygous at the isozyme locus: two homozygous (FF and SS) and one heterozygous (FS) (Schwartz, 1966; Scandalios, 1969).

In the considered paper the genetic methods were used to show that the analyzed sugar beet agamospermous progenies were formed from somatic cells (Levites et al., 1999). This conclusion was based on the monomorphism of the KWS1-5A offspring by heterozygous isozyme spectrum of marker enzyme alcohol dehydrogenase (ADH1). Interesting, the study also revealed the dimorphism of the analyzed progenies, including KWS1-5A, for other marker enzymes. In the progeny the enzymes dimorphism was expressed by the presence of only two phenotypic classes: one homozygous and one heterozygous. Of all the data from the cited article, let us consider two offsprings: KWS1-5A and KHBC2-78A (Table 1).

In the cited article it was assumed that the dimorphism was due to the inactivation in a part of the offspring of one of the alleles at a heterozygous locus. As a result, the seeds with the phenotypes similar to the homozygous one carry one active allele which determines the electrophoretic mobility of the enzyme and one inactivated allele. However, later it was found that phenotypes similar to those of the homozygous are conditioned by homozygous genotypes indeed (Levites, Kirikovich, 2003).

The findings allowed us to hypothesize that the dimorphism of agamospermous progeny is due to the heterozygosity at the marker enzyme locus with one allele represented by a single copy and the other allele represented by three copies arising as a result of polytenization (Levites, 2005, 2007). Polyteny of chromosomes in plants - a well known fact (Carvalheira, 2000). The somatic cells with genotype FFFS lose excessive allelic copies equiprobably before entering embryogenesis. The calculations assisted with hypergeometric distribution formulas (Feller, 1950) indicate that, in this case, only two genotypes, FF and FS, in the ratio of 1:1 are theoretically possible.

The calculation is as follows:

For homozygotes  $FF - C_3^2 \times C_1^0 / C_4^2$  - the number of combinations of the choice two out of three, multiplied by the number of combinations 0 out of 1 and divided by the number of combinations 2 out of 4, i.e., 3x1/6.

For heterozygotes  $FS - C_3^1 \times C_1^1 / C_4^2$  - the number of combinations of the choice one out of three multiplied by the number of combinations one out of one, and divided by the number of combinations 2 out of 4, i.e., 3x1/6.

In the reduced form, this ratio expressed in integers is 1:1.

The *SS* genotype is not formed because it requires two allelic copies while only one copy is present in the genome.

The equiprobable diminution process requires a free exchange of chromatides between chromosomes. The existence of such exchange was demonstrated later on the base of the phenotype ratio observed in the agamospermous colchicine-treated plant progeny (Levites, Kirikovich, 2012).

Therefore, it is interesting to consider the phenotype ratios of the marker enzymes isocitrate dehydrogenase (IDH3) and malate dehydrogenase (MDH1) in agamospermous progeny KHBC2-78A. This offspring is of particular interest because it combines two distinctive traits that are inherent to the agamospermous progeny: the phenotype class ratio for IDH3 corresponding to 3FF:8FS:3SS and dimorphism for MDH1. The MDH1 phenotype class ratio 1FF:1FS indicates that this progeny has originated from somatic cells with different alleles doses at locus *Mdh1*. The somatic origin of these cells implies that no meiotic genome transformations have occurred in such cells nuclei.

Mathematically, ratio 3:8:3 is known to be possible derived if 2 elements are selected randomly out of a sample containing 4 elements of one type and 4 elements of the other type (Feller, 1950).

This occurs, for example, when a heterozygous tetraploid cell of genotype *FFSS* enters into meiosis. Since, at this moment, each chromosome is represented by two chromatids, 8 allelic copies are presented in the nucleus by 4 copies of each of the two alleles. If the frequency of crossing-over between the marker locus and the centromere is 50%, all allelic copies behave independently and the random selection of two copies obeys the probability laws. The frequencies of the resulting gametes can be calculated by the hypergeometric distribution formulas (Feller, 1950). For the above example the gamete frequencies in units fractions can be determined as follows:

For homozygotes  $FF - C_4^2 \times C_4^0 / C_8^2$ , the number of combinations of the choice 2 out of 4 multiplied by the number of combinations 0 out of 4 and divided by the number of combinations 2 out of 8, i.e., 6x1/28.

For heterozygotes  $FS - C_4^1 \times C_8^1 / C_8^2$ , the number of combinations of the choice 1 out of 4 multiplied by the number of combinations 1 out of 4 and divided by the number of combinations 2 out of 8, i.e., 4x4/28.

For homozygotes  $SS - C_4^0 \times C_8^2 / C_8^2$ , the number of combinations of the choice 0 out of 4 multiplied by the number of combinations 2 out of 4 and divided by the number of combinations 2 out of 8, i.e., 6x1/28.

In the reduced form, this ratio expressed in integers is 3:8:3.

From the above-mentioned it can be concluded that the ratio 3:8:3 for *Idh3* observed in the KHBC2-78A

offspring implies that: 1) the offspring emerged from the cells with an increased copies number of each allele at locus Idh3; 2) the number of allelic copies decreases in the moment before cells entering into embryogenesis. On the other hand, the presence in the same seeds of this progeny of two phenotypic classes for MDH1 indicates that this progeny originates from the cells not undergone which have meiotic genome transformations. Therefore, the reduction in the number of allelic copies at the Idh3 locus is not a consequence of meiosis, but it is the result of chromatin diminution only.

Moreover, the phenotype class ratios in both progenies described here can be explained precisely by chromatin diminution.

The presence in one offspring of two complementary traits (somatic origin of the cells entering into embryogenesis and an increased dose of alleles in the cells capable of embryogenesis) confirms both the agamospermous origin of the offspring and the process of chromatin diminution from the cells at the moment before their entering into embryogenesis.

In conclusion, it should also be added that, according to the proposed hypothesis, the equiprobable diminution process of the number of redundant allelic copies is a consequence of equiprobable allelic copies attachment to the nuclear membrane (Levites, 2005, 2007). It is assumed that only one copy from each chromosome out of two homologous ones in a diploid plant attaches to the cell nuclear membrane before its entering embryogenesis. The attached allelic pair determines the genotype of a developing embryo while the unattached allelic copies are lost.

Thus, a new analysis of the previously published data gives a new insight on the complementary genetic facts, which confirm the model describing a specific mechanism of the origin of polymorphism in agamospermous progenies. The available data provide the genetic proof of that chromatin diminution is one of the mechanisms underlying the origin of polymorphism in sugar beet agamospermous progenies.

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*Table1:* Phenotypic classes of marker enzymes in agamospermous progenies obtained from pollen sterile sugar beet plants (Levites et al., 1999)

	Marker enzyme phenotypes of progenies			
Progeny	ADH1	IDH3	MDH1	
	FF : FS : SS	FF : FS : SS	FF : FS : SS	
KWS1-5A	0:78:0	23:41:0*	9:0:0	
KHBC2-78A	-	9:47:10**	45:57:0*	

The probability of affinity with theoretically expected ratio 3:8:3 - \*- P<0.001; \*\* - P>0.05



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# Biochemical Effect of Two Molluscicide Baits against the Land Snail Theba Pisana

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*Abstract-* The present study was investigated the biochemical effect of two molluscicides baits Methomyl and Diazinon on the tissues of the land snail, Theba pisana. The activities of three vital enzymes, total protein (TP) and total lipid (TL) were laboratory tested. The enzymes were aspartate amino transaminase (AST); alanine amino transaminase (ALT), and Alkaline phosphatase (ALK).

Results showed that all tested molluscicides lead to increase the activity of AST, ALT and ALK in the tested land snail, Theba pisana, except Diazinon 1% treatment showed a decrease in AST and ALK when applied against the land snail. On the other hand, the level of total protein was increased after treatment with Methomyl 5% and 3% and Diazinon 5% and 3%, while decreased after treatment with Methomyl 1% and Diazinon 1%. The level of total lipid was increased with Methomyl 5%, 3%, 1% and Diazinon 5%, 3%but decreased with Diazinon 1%. In general, two molluscicides were significantly affected on the activities of enzymes, total lipid and total protein compared with control treatment when applied against the tested snails.

Keywords: theba pisana, methomyl, diazinon, aspartate amino transaminase (ast); alanine amino transaminase (alt) and alkaline phosphatase (alk).

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# Biochemical Effect of Two Molluscicide Baits against the Land Snail Theba Pisana

Sharaf, H.M.<sup> \alpha</sup>, Abdelmonem, M.K. <sup>\alpha</sup>, Salwa, Z.A. <sup>\alpha</sup> Arafa <sup>\overline \alpha</sup> & Aya, A.M. <sup>\delta</sup>

Abstract- The present study was investigated the biochemical effect of two molluscicides baits Methomyl and Diazinon on the tissues of the land snail, Theba pisana. The activities of three vital enzymes, total protein (TP) and total lipid (TL) were laboratory tested. The enzymes were aspartate amino transaminase (AST); alanine amino transaminase (ALT), and Alkaline phosphatase (ALK).

Results showed that all tested molluscicides lead to increase the activity of AST, ALT and ALK in the tested land snail, Theba pisana, except Diazinon 1% treatment showed a decrease in AST and ALK when applied against the land snail. On the other hand, the level of total protein was increased after treatment with Methomyl 5% and 3% and Diazinon 5% and 3% , while decreased after treatment with Methomyl 1% and Diazinon 1%. The level of total lipid was increased with Methomyl 5%, 3%, 1% and Diazinon 5%, 3%but decreased with Diazinon 1%. In general, two molluscicides were significantly affected on the activities of enzymes, total lipid and total protein compared with control treatment when applied against the tested snails.

*Keywords:* theba pisana, methomyl, diazinon, aspartate amino transaminase (ast); alanine amino transaminase (alt) and alkaline phosphatase (alk).

#### I. INTRODUCTION

errestrial white garden snails, Theba pisana (Muller) are considered one of the most common dangerous species in Delta region, esecpcially in northern areas of Egypt. They are known as destructive causing several damage to vegetables, pests ornamental and citrus trees (Hamdy, 1999). E. vermiculata and M. cantiana were recorded with a relatively high population density on the major economic crops at Dakahlia governorate (Awad, 2000; Genena, 2003).

Control of land snails on different crops is heavily dependent on the use of molluscicides that limit the effect of these pests below damaging level. Hence, the synthetic molluscicides are the most effective measures available at present for the control of terrestrial gastropods (Heiba et al., 2002; Genena, 2003; Abd-El-Ail, 2004; Ismail et al., 2005; Zedan et al., 2006; Genena et al., 2008). Bait formulations of molluscicides was the most effective application method in the field for controlling terrestrial gastropods rather any other technique (Kassem, 2004). Carbamate molluscicides are known to act as nerve toxins by inhibition of

Authors α σ ρ Ο ¥: Zoology Department, Faculty of Science, Zagazig University, Egypt. e-mail: sharaf hesham@yahoo.com cholinesterase. Metaldehyde molluscicides caused an excessive increase of fluid excretion in the soft snail body, so leading to snail death (Kassem et al., 1993; El Gohary, R.A. Laila and Marwa A.M. Genena, 2011). Both carbamate and metaldehyde are successfully used in Egypt as well as in many other countries to control land snails (Heiba et al., 2002). Transaminase enzymes and acetylcholine esterase as well as total proteins and total lipids are important in the biological processes in the land snails (Abd-El-Ail, 2004).

The aim of this work was to determine the biochemical effect of two molluscicides namely, Methomyl and Diazinon on the activities of three vital enzymes, Total Protein (TP) and Total Lipid (TL) to throw a light on the toxicity and mode of action of these molluscicides in the land snail, Theba pisana. The enzymes selected for this study were; alanine amino transaminase (ALT), aspartate amino transaminase (AST), and Alkaline phosphatase (ALK).

#### II. MATERIAL AND METHODS

*Tested snails:* Adult snails of Theba pisana collected from infested nurseries and field crops in gardens in (Abees area, few kilometers south of Alexandria and El-Montazah, Alexandria, Egypt. The obtained snails were transferred in plastic bags to the laboratory, then kept in plastic cages (40x30x30 cm, with 100 individuals per cage) filled with moist sterilized sandy loamy soil 1:1 (v:v) and fed on fresh leaves of lettuce (Lactuca sativa L.) for 14 days to be laboratory acclimatized.

*Tested molluscicides:* Two molluscicides belonging to two different chemical groups were tested. The trade name, Common name, chemical name and field recommended rates are shown in Table 1.

*Experimental design:* The experiment took place under laboratory condition at  $22\pm1^{\circ}$ C and  $60\pm2\%$  R.H. Field recommended rate for each molluscicide was introduced to each land snail species. Ten adult snail individuals with approximately similar size were then transferred from stock culture to plastic cups 10 cmdiameter filled with 100 g moist sterilized sandy soil: loamy soil 1:1 (v:v). Each cup was then covered with muslin cloth held by rubber bands. Each of the above mentioned molluscicide and the control were replicated five times. Biochemical studies were made after three days of treatment.

#### a) Biochemical studies

Sample preparation: After three days of treatment, shells of tested snails were removed by making a cut around the whorls in a continuous manner starting at the aperture opening using bone scissors and the broken fragments of the shell were carefully removed. Snail tissues were dissected out and all tissues of each treatment were homogenized in distilled

water (50 mg mL-1). The homogenates were centrifuged at 8000 rpm for 15 min at 5°C in refrigerated centrifuge. The deposits were discarded and the supernatants were kept in a deep freezer till use to determine the activities of alanine amino transaminase (ALT), aspartate amino transaminase (AST), Alkaline phosphatase (ALK),total protein (TP) and total lipid (TL).

Table	1	· List	of	molluscicides	their	r trade name	common n	ame	chemical n	ame
aDic	/	, டாலா		monusciciacs	uicii	trade name,	CONTINUE	ame,	Chemican	anne

Trade name	Common name	Chemical group	Chemical structure
Lannet(90%W.P)	Methomyl	Carbamate	0    CH3-C=N-O-C-NH-CH3   S-CH3
Diazinon ( 60% E.C.)	Diazonix	Organophosphate	H <sub>3</sub> C CH <sub>3</sub> H <sub>3</sub> C CH <sub>3</sub>

*Biochemical measurements:* The activity of AST and ALT were determined according to the method of Reitman and Frankel (1957) using commercial reagents. Total proteins were calorimetrically determined according to Bradford (1976) while total lipids were assayed by the method of Knight et al. (1972).

Data analysis: Data were calculated as Mean±SD and analyzed using analysis of variance technique (ANOVA) followed by Least Significant Difference (LSD). Probability of 0.05 or less was considered significant. All statistical analysis was done with CoHort Software 2004.

#### III. Results

The biochemical effect of six concentrations of molluscicides namely, Methomyl 5%, Methomyl 3%, Methomyl 1%, Diaziono 5%, Diazinon 3% and Diazinon 1% on the activities of five vital enzymes, Total Protein (TP) and Total Lipid (TL) to throw a light on the toxicity and mode of action of these molluscicides in the land snail, T. pisana. The enzymes selected for this study were; aspartate amino transaminase (AST); alanine amino transaminase (ALT) and Alkaline phosphatase (ALK).

Activity of aspartate amino transaminase (AST): Data in Table 2 showed that all tested molluscicides were increase the level of (AST) when applied against Theba pisana. Data also showed that there were not significant increase between Methomyl 3% and methomyl 1% in the activity of enzyme when applied against land snail with mean values,  $432.57\pm2.28$  and  $346.23\pm44.09$  more than control, respectively. Methomyl 5% caused the highest increase in the activity of (AST) with mean value,  $678.47\pm80.21$ .

Table 2 : Aspartate amino transaminase (AST) activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of AST (Mean±SD)
Theba pisana	Methomyl 5%	678.47±80.21
	Methomyl 3%	432.57±2.28
	Methomyl 1%	346.23±44.09
	Control	324.30±21.08
LSD(0.05) = 71.35		

Specific activity of AST: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

Activity of alanine amino transaminase (ALT): Data in Table 3 indicated that, ALT activity was increased in the land snail after treated with all tested molluscicides. There were significant differences between all treatments and control. Methomyl 5% and Methomyl 3% gave the highest increase in ALT activity with mean values, 452.97±14.32 and 387.33±7.70 more than control, respectively. Methomyl 1% gave the

lowest increase in the level of this enzyme with mean value,  $269.93 \pm 19.20$  more than control.

*Table 3 :* Aspartate amino transaminase (ALT) activity in the land snail, Theba pisana after 72 hrs of molluscicides treatment

Snail	Treatment	Activity of ALT(Mean±SD)
Theba pisana	Methomyl 5%	452.97±14.32
	Methomyl 3%	387.33±7.70
	Methomyl 1%	269.93±19.20
	Control	231.77±9.41
LSD(0.05) = 20.39		

Specific activity of AST: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

Activity of Alkaline phosphatase (ALK): Data in Table 4 indicated that, ALK activity was increased in the land snail after treated with all tested molluscicides. There were significant differences between all treatments and control. Methomyl 5% and Methomyl 3% gave the highest increase in ALK activity with mean values,  $173.47\pm20.5$  and  $151.14\pm2.78$  more than control, respectively. Methomyl 1% gave the lowest increase in the level of this enzyme with mean value,  $97.70\pm4.769$  more than control.

Table 4 : (ALK) activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of ALK (Mean±SD)
Theba pisana	Methomyl 5%	173.47±20.52
	Methomyl 3%	151.40±2.78
	Methomyl 1%	97.70±4.76
	Control	88.26±2.96
LSD(0.05) = 16.29		

Specific activity of ALK: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

*Total lipid level:* Data in Table 5 indicated that tested compounds were increase the level of total lipids when applied against land snail and there were no

significant differences between all treatments and control except Methomyl 5%. Methomyl 5% gave the highest increase in total lipids followed by Methomyl 3% and Methomyl 1% with mean values  $33.03\pm0.23$ ,  $31.13\pm0.49$  and  $30.53\pm1.002$  more than control, respectively.

*Table 5 :* Total Lipids activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Total lipid(Mean±SD)
Theba pisana	Methomyl 5%	33.03±0.23
	Methomyl 3%	31.13±0.49
	Methomyl 1%	30.53±1.002
	Control	30.40±0.529
LSD(0.05) = 0.954		

*Total Lipids:* (mg/ snail). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

of total proteins when applied against land snail, with the mean values 2.07±0.55 and 1.72±0.03 more than control, respectively. Methomyl 1% was decrease the level of total proteins with mean value, 1.41±0.13less than control.

*Total protein level:* Data in Table 6 indicated that Methomyl 5% and Methomyl 3% were increase the level

Table 6: Total protein activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Total protein(Mean±SD)
Theba pisana	Methomyl 5%	2.07±0.55
	Methomyl 3%	1.72±0.03
	Methomyl 1%	1.41±0.13
	Control	1.64±0.12
LSD(0.05) = 0.152		

*Total proteins (T.P):* (mg/ snail). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

For Diazinon Activity of aspartate amino transaminase (AST): Data in Table 7 showed that

Diazinon 5% and Diazinon 3% were increase the level of (AST) when applied against Theba pisana. With mean values  $440.000 \pm 44.91$  and  $378.20 \pm 166.46$  more than control, respectively. While Diazinon 1 % was decrease the level of AST with mean value  $254.70 \pm 14.93$ .

*Table 7 :* Aspartate amino transaminase (AST) activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of AST (Mean±SD)
Theba pisana	Diazinon 5% Diazinon 3% Diazinon 1% Control	440.000±44.91 378.20±166.46 254.70±14.93 324.30±21.087
LSD(0.05) = 132.385		

Specific activity of AST: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

Activity of alanine amino transaminase (ALT): Data in Table 8 indicated that, ALT activity was increased in the land snail after treated with Diazinon 5% and Diazinon 3% with mean values,  $386.23\pm31.03$  and  $303.57\pm3.98$  more than control, respectively. Diazinon1% was decrease the level of this enzyme with mean value,  $222.63\pm6.67$  less than control.

*Table 8 :* Aspartate amino transaminase (ALT) activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of ALT(Mean±SD)
Theba pisana	Diazinon 5%	386.23±31.03
	Diazinon 3%	303.57±3.98
	Diazinon 1%	222.63±6.67
	Control	231.77±9.41
LSD(0.05) = 25.32		

Specific activity of ALT: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

Activity of Alkaline phosphatase (ALK): Data in Table 9 indicated that, ALK activity was increased in the land snail after treated with all tested molluscicides. There were significant differences between all treatments and control. Diazinon 5% and Diazinon 3% gave the highest increase in ALK activity with mean values,  $190.30 \pm 4.80$  and  $129.97 \pm 9.21$  more than control, respectively. Diazinon 1% gave the lowest increase in the level of this enzyme with mean value,  $118.87 \pm 0.49$  more than control.

Table 9 : (ALK) activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of ALK (Mean±SD)
Theba pisana	Diazinon5%	190.30±4.80
	Diazinon 3%	129.97±9.21
	Diazinon 1%	118.87±0.49
	Control	88.26±2.97
I SD(0.05) = 8.213		

Specific activity of ALK: (U x 103/ mg protein). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

*Total lipid level:* Data in Table 10 indicated that Diazinon 5% and Diazinon 3% were increase the level of

total lipids when applied against land snail with mean values,  $34.16\pm0.95$  and  $31.93\pm0.55$  more than control, respectively. While Diazinon 1% was decrease the level of total lipids with mean value  $27.33\pm1.20$  less than control.

Table 10 . Total I	inid activity	in the land enail.	Theha nisana afte	er 72 h of molluscicides t	reatment
	ιρία αστίνιτι	in the land shall			reatiment

Snail	Treatment	Activity of T.lipid(Mean±SD)
Theba pisana	Diazinon 5%	34.16±0.95
	Diazinon 3%	31.93±0.55
	Diazinon 1%	27.33±1.20
	Control	30.40±0.529
LSD(0.05) = 0.75		

*Total Lipid:* (mg/ snail). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

*Total protein level:* Data in Table 11 indicated that Diazinon 5% and Diazinon 3% were increase the

level of total proteins when applied against land snail, with the mean values  $2.44\pm0.46$  and  $2.35\pm0.12$  more than control, respectively. Diazinon 1% was decrease the level of total proteins with mean value,  $1.55\pm0.06$  less than control.

Table 11 : Total protein activity in the land snail, Theba pisana after 72 h of molluscicides treatment

Snail	Treatment	Activity of T. protein(Mean±SD)		
Theba pisana	Diazinon 5%	2.44±0.46		
	Diazinon 3%	2.35±0.12		
	Diazinon 1%	1.55±0.06		
	Control	1.64±0.12		
LSD(0.05) = 0.378				

*Total protein (T.P):* (mg/ snail). Values followed by the same letter (s) are not significantly different at the 0.05 level according to Duncan's test.

#### IV. Discussion

The present study revealed that all tested molluscicides increased the activities of AST and ALT when tested against the land snail, Theba pisana. The transaminases enzymes; AST and ALT are not solely located in hepatocytes but rather are also in many body organs. Also, they elevation in their activities could be due to a variety of conditions including muscle damage, intestinal and hepatic injury and toxic hepatitis (Farkas et al., 2004). On the other hand, the decrease activities of AST and ALT may be due to either to leakage of the enzymes into extracellular compartments or to actual enzymes inhibition by these molluscicides. Thus, the deviation of both enzymes activities out of the normal range could lead to biochemical impairment and lesions of the tissues and cellular functions (Radwan et al., 1992). Accordingly, the present elevations or reductions in the activities of AST and ALT enzymes in tissues of the two land snails, E. vermiculata and M. cantiana treated with tested molluscicides could be partially due to cell injury of their different organs and this may be led to disturbances in their enzymatic systems (Mahmoud, 2006). These results support the findings of Radwan et al. (1992) they found that carbamate compounds lead to significant elevation of the activity of AST and ALT when applied against the land snail, Theba pisana.

In general, the present data indicated that all tested compounds increased the level of ALK in land snail, Theba pisana. The current results indicated that increase of total lipids (TL) and total proteins (TP) in the tissues of land snail, T.pisana. But Diazinon 1% was decrease total lipid. The current results are agreement

with the findings of Abd-El-Ail (2004) found that niclosamide molluscicide were increased the level of total lipids and total proteins more than control after 24, 48, 72, 96 h of treatments when applied against the land snail, E. vermiculata.

The decrease in the level of both total protein and total lipids may be partly resulted from imbalance between the rate of synthesis and the rate of degradation. Gabr et al. (2007) reported that the depression in total lipids may be due to decline in lipid synthesizing capacity and/or due to an increase in the hydrolysis of hepatic lipids to combat the stress conditions. The harmful effect of chemical compounds could be attributed to enhancement of energy utilization and/or destruction of cells organelles of treated snails that may be led to inhabitation of protein synthesis (Eissa et al., 2002).

The data presented in this study, provide that these chemical compounds caused an alternation in some biochemical targets which could lead to serious metabolic and cellular damage. In general, the two molluscicides were affected in the activities of three vital enzymes, total lipid and total protein when applied against the tested land snails. Further studies are needed to clearly the most probable mode of action of these chemical compounds on the terrestrial snails.

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# Effect of Dietary Incorporation of Gliricidia Maculata Leaf Meal on Growth and Feed Utilization of Cirrhinus Mrigala Fingerlings

## By S. A. Vhanalakar & D. V. Muley

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Abstract- An eight week feeding trial was conducted to evaluate the potential of Gliricidia maculata leaf meal as dietary protein source in the diet of Cirrhinus mrigala fingerlings. Four experimental diets were formulated to contain 20%, 30%, 40% and 50% G. maculata leaf meal (Diets 1 - 4) to partially replace other protein ingredients in the C. mrigala diet. The diet containing 0% leaf meal served as the control. Each dietary treatment was tested in triplicate groups of 10 fingerlings. The results of the growth and feed utilization responses show that there were no significant differences among the fish fed diets 1 - 3 but were significantly different from fish fed on diet 4 which had lower growth and feed utilization values. The present findings show that G. maculata leaf meal has good potential for use as one of the protein sources in C. mrigala diet up to 40% level without compromising growth.

Keywords: gliricidia maculata; cirrhinus mrigala; growth; feed utilization.

GJSFR-C Classification : FOR Code: 069999

# EFFECT OFDIETARYINCORPORATION OFGLIRICIDIA MACULATA LEAF MEAL ONGROWTH AND FEEDUTILIZATION OF CIRRHINU MRIGALAFINGERLINGS

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# Effect of Dietary Incorporation of Gliricidia Maculata Leaf Meal on Growth and Feed Utilization of Cirrhinus Mrigala Fingerlings

S. A. Vhanalakar <sup>a</sup> & D. V. Muley <sup>o</sup>

Abstract- An eight week feeding trial was conducted to evaluate the potential of Gliricidia maculata leaf meal as dietary protein source in the diet of Cirrhinus mrigala fingerlings. Four experimental diets were formulated to contain 20%, 30%, 40% and 50% G. maculata leaf meal (Diets 1 - 4) to partially replace other protein ingredients in the C. mrigala diet. The diet containing 0% leaf meal served as the control. Each dietary treatment was tested in triplicate groups of 10 fingerlings. The results of the growth and feed utilization responses show that there were no significant differences among the fish fed diets 1 - 3 but were significantly different from fish fed on diet 4 which had lower growth and feed utilization values. The present findings show that G. maculata leaf meal has good potential for use as one of the protein sources in C. mrigala diet up to 40% level without compromising growth.

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#### I. INTRODUCTION

he current trend in fish culture is towards increased intensification of culture systems whereby provision of feeds becomes necessary and success therefore depends significantly on the availability of well-balanced, nutritionally complete and cost-effective feeds. For many years, fish nutritionists has done considerable research on the nutrient requirements of fish, assessment of nutritive value of available ingredients and development of simple and appropriate feeding technology. These are all important factors towards the development of cost-effective feeds and feeding strategy. There is a need, however, for these feeds to be continuously refined, improved and tested for technical and economic feasibility.

In order to reduce the cost of a balanced fish diet, locally available ingredients such as agricultural byproducts and plant proteins should be included in the diet or substituted for expensive protein sources. Research interest has been focused on different leaf meals as protein sources in animal feeds (Wouters, 1994; Agbede and Aletor, 2003).

Author α: Department of Zoology, Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti, Taluka – Bhudargad, Dist – Kolhapur, India. e-mail: sagarayan36@rediffmail.com Author σ: Department of Zoology, Shivaji University, Kolhapur, India. e-mail: drdvmuley@gmail.com The aim of the current study was to assess the potential of Gliricidia maculata leaf meal as an ingredient in practical feeds for freshwater fish, Cirrhinus mrigala. The G. maculata is extensively used for social forestry. This plant grows faster and in some parts of India its leaves are used as feed for goat.

#### II. MATERIALS AND METHODS

The feeding experiment was conducted in triplicate for 8 weeks. Fingerlings of Cirrhinus mrigala were used for the experiment. Four types of pelleted feeds were formulated using different ingredients such as rice bran, groundnut oilcake, fishmeal, guar gum binder, Vitamin – Mineral mixture, fine leaf powder of Gliricidia maculata in different proportions (diet 1 - 4). A diet with all above ingredients except leaf powder is kept as control (Table 1). The diets were analyzed for their proximate nutrient composition.

Fishes were fed at the rate of 5% body weight in two equal rations daily. At fortnightly intervals a minimum of 50% of fishes were sampled to record the growth. At the end of experiment, the growth parameters like mean body weight, specific growth rate (SGR), feed conversion ratio (FCR) and protein efficiency ratio (PER) were estimated. Difference between means of treatments was tested to find out the level of significance by ANOVA.

Table 1 : Formulation and proximate composition of fish diets containing increasing levels of Gliricidia maculata leaf meal

	Control	Diet 1	Diet 2	Diet 3	Diet 4	
Ingredients (%)						
Groundnut oilcake	43	35	29	24	19	
Rice bran	36	27	23	18	13	
Fishmeal	10	09	09	09	09	
Guar gum Binder	10	08	08	08	08	
Mineral – Vitamin mixture	01	01	01	01	01	
G. maculata leaf powder	00	20	30	40	50	
Nutrient content (%)						
Moisture	7.05	6.32	6.93	7.27	7.75	
Total Ash	12.13	12.26	11.59	11.38	10.89	
Protein	26.24	28.30	29.93	30.42	31.10	
Fat	3.81	7.33	6.40	6.26	5.56	
Fibre	10.54	9.21	10.78	11.76	11.60	

#### III. Results and Discussion

The growth study in regard with body weight, specific growth rate (SGR), feed conversion ratio (FCR)

and protein efficiency ratio (PER) were given detailed in the Table 2.

Table 2 : Growth performance and feed utilization in Cirrhinus mrigala fed diets containing Gliricidia maculata leaf

meal

	Control	20%	30%	40%	50%
Initial body weight (gm)	2.1 ± 0.05	2.4 ± 0.02	2.3 ± 0.06	2.3 ± 0.05	2.1 ± 0.04
Final body weight (gm)	14.64 ± 0.42	17.04 ± 0.49 NS	22.85 ± 0.65 ***	29.96 ± 0.86 ***	26.56 ± 0.76 ***
Weight gain	12.54 ± 0.36	14.64 ± 0.42 NS	20.55 ± 0.59 ***	27.66 ± 0.79 ***	24.46 ± 0.70 ***
Specific growth rate (SGR) % day -1	0.89 ± 0.02	0.91 ± 0.02 NS	1.05 ± 0.03 *	1.17 ± 0.03 ***	1.15 ± 0.03 ***
Food conversion ratio (FCR)	2.49 ± 0.07	2.49 ± 0.07 NS	1.98 ± 0.05 ***	1.58 ± 0.04 ***	1.36 ± 0.03 ***
Protein efficiency ratio (PER)	0.65 ± 0.01	0.48 ± 0.01 ***	0.66 ± 0.01 NS	0.85 ± 0.02 ***	0.71 ± 0.02 NS

Fish groups fed with 40% Gliricidia diet showed better growth performance as compared to other diet groups. The final body weight (29.96  $\pm$  0.86), weight gain (27.66  $\pm$  0.79) and SGR (1.17  $\pm$  0.03) were highest in 40% diet group, whereas FCR was highest in control and 20% diet group (2.49  $\pm$  0.07) and PER in 50% diet group (0.71  $\pm$  0.02) (Table 2).

There was significant increase in case of weight gain in all diet groups except 20% compared with control. As compared to control, there was significant decrease in FCR in all diet groups except 20%. The fishes fed with 20% Gliricidia diet showed equal FCR value as control. The increasing SGR was observed at low level inclusion of Gliricidia and decreased SGR as Gliricidia inclusion level increased. The same observations were found for PER.

Utilization of Gliricidia leaf meal diet in the present study showed their effectiveness regarding fish

growth within the inclusion range of 20 – 40%. The best growth was recorded from 40% Gliricidia diet. The use of leaf meal in fish feed at higher inclusion rate always leads to fish growth reduction. A reduction of 60% weight gain at 40% inclusion of moringa leaf meal was reported by Richter et al. (2003) and Afuang et al. (2003). The inclusion of Leucaena leucocephala leaf meal (Wee & Wang, 1987; Santiago et al., 1988), cassava leaf meal (Ng & Wee, 1989), salt bush atriplex leaves (Yousif et al., 1994) and duckweed (Fasakin et al., 1999) at higher level in fish diet lead to significantly lower growth rates in fishes.

Higher inclusion of plant protein in formulated fish diet causes the retarded growth of fish. In the present study, it was observed that incorporation of Gliricidia above 40% impaired the overall growth of experimental fish, Cirrhinus mrigala. The data of the present study agree with the finding of Pereira & Oliva - Teles (2003), who reported that significant decreases were found for both, growth and feed utilization with the highest replacement levels of dietary fish meal with plant proteins for gilthead sea bream.

## IV. CONCLUSION

The present study confirmed that, Cirrhinus mrigala is able to utilize plant based formulated diet. An inclusion level of Gliricidia maculata leaf powder up to 40% in the practical diet for C. mrigala fingerlings had no adverse effects on growth and feed utilization of the fish. From the present work it is concluded that, G. maculata may be a promising source of plant protein; used for partial replacement of fishmeal in the formulated feed. It will definitely help to small scale fish farmers to overcome expenditure on traditional fish feed.

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- 3. Submission of Manuscripts,
- 4. Manuscript's Category,
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Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Yet, use comprehensive sentences and do not let go readability for briefness. You can maintain it succinct by phrasing sentences so that they provide more than lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study, with the subsequent elements in any summary. Try to maintain the initial two items to no more than one ruling each.

- Reason of the study theory, overall issue, purpose
- Fundamental goal
- To the point depiction of the research
- Consequences, including <u>definite statistics</u> if the consequences are quantitative in nature, account quantitative data; results of any numerical analysis should be reported
- Significant conclusions or questions that track from the research(es)

#### Approach:

- Single section, and succinct
- As a outline of job done, it is always written in past tense
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- Center on shortening results bound background information to a verdict or two, if completely necessary
- What you account in an conceptual must be regular with what you reported in the manuscript
- Exact spelling, clearness of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else

#### Introduction:

The **Introduction** should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable to comprehend and calculate the purpose of your study without having to submit to other works. The basis for the study should be offered. Give most important references but shun difficult to make a comprehensive appraisal of the topic. In the introduction, describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will have no attention in your result. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here. Following approach can create a valuable beginning:

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- Present a justification. Status your particular theory (es) or aim(s), and describe the logic that led you to choose them.
- Very for a short time explain the tentative propose and how it skilled the declared objectives.

#### Approach:

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#### Approach:

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Content

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#### Approach:

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