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INSTRUCTION TO AUTHORS

Manuscripts should be submitted in triplicate, typed double spaced on one side of the paper (A4 bond) with 3cms. margin on all sides. The arrangement of the manuscript should be as follows : Title page, Abstract, Key words, Introduction, Methods, Results, Discussion, Acknowl-edgments References, Tables, Figures, Legends and Figures, Full length of paper should not exceed 10 printed pages.

Title Page : It should contain the following information :

(i) The title of the paper which should be concise but informative, (ii) a short running head of not more than 10 words placed at the foot of the title page, (iii) first name, middle initial and last name of each author, (iv) name of department (s) and institution (s) to which the work should be attributed, (v) name and address of author for correspondence.

Abstract : The second page should carry and abstract of not more than 200 words. The abstract should state the purpose of the study, basic procedure, main findings and principal conclusions. Abstract should state the purpose of the study, basic procedure, main findings and principal conclusions, Abstract should be followed by relevant Key Words.

Introduction : This should contain a concise statement of the purpose of the article. Only pertinent references should be given.

Methods : The methodology, apparatus and procedure in sufficient detail should be identified to allow other workers to repeat the experiments. Standard methods can, however, be identified by proper references. The new or substaintially modified methods should be described giving reasons for using them.

Results : This should be quoted in SI units, the results should be presented in logical sequence in the text, tables and illustrations. Unnecessary repetition should be avoided. Only important observations may be emphasized in the text.

Discussion : This should prescisely deal with interpretation of rusults. Emphasis should be given on the new and important aspects of the study and conclusions that follow from them. Recommendations, when appropriate, may be included.

References : Consecutive number in parentheses should be used to indicate the reference in the text. The full reference should be cited at the end of the manuscript. The following forms of citations should be used.

Journals: Bunt, A. H., Lund, R. H. and Lund, J. S. (1994): Retrograde axonal transport of the albino rat retina, Brain. Res. 72: 215-228.

Books: Campbell, A. M. (1984): Monoclonal Antibody Technology. Elsevier, Amsterdam, pp 50-66. Individual Chapters in book : Sladen, G. E. (1975): Absorption of fluid and electrolytes in health and disease. In : Intestinal Absorption in Man (I. McColl and G. E. Sladon, eds.), Academic Press, London, 135-146.

Journal titles should be abbreviated as in Idex Medicus or Biological Abstract.

Tables : Tables should be typed on seperate sheets using double space. Each table should be numbered (Roman numerals) consecutively with a brief caption on the top of the table.

Illustrations : Figures should be numbered and market on the back by author's name. Line drawing sufficiently thick for good reproduction, should be made with Chinese ink on plane white paper or tracing paper.

Legends of the figures should be type written on seperate sheets and their positions in the text should be indicated in the manuscript.

Photographs must be in black and white. A clear print in glossy paper, large enough to be legible after 25% reduction, is necessary for reproduction. These should be submitted along with the paper.

PENAEOID SHRIMP OF DIGHA AND ADJACENT COAST OF MIDNAPORE, WEST BENGAL, INDIA

Angsuman Chanda and Tanmay Bhattacharya

Department of Zoology Vidyasagar University, Midnapore 721 102, West Bengal, India

ABSTRACT

22 species under 10 genera and two families of penaeoid shrimps have been recorded from Digha $(87^{\circ}32N, 21^{\circ}36'30''E)$ and its adjacent coast. Of these, four represent the first record from the study area. A key has also been constructed for ready indentification of the shrimps of this region.

Introduction

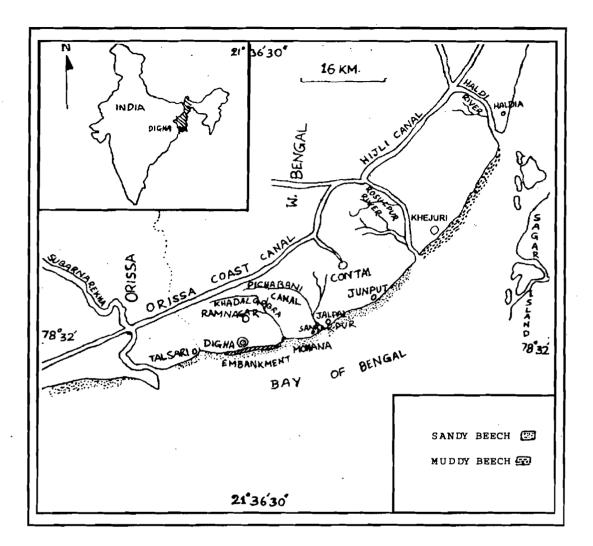
The present study stretches across 50 km long coastal line of the District of East Midnapore from Talsari to Khejuri, which is known for straightness of the coast line, flatness and compactness of the beach (Gupta, 1970). The region is traversed by two irrigation canals viz., Khadalgobra canal and Ramnagar canal, which jointly discharge water into the sea at the point known as Digha Mohana, as a result of which an estuarine zone is created in this area. About 40 km. east from Digha town, near Khejuri, is Rasulpur river which opens onto the Hooghly estuary. Some years back an embankment was constructed from Old Digha to New Digha by rocky boulders. This vast coast line contains a great biogeochemical diversity in respect of soil texture, marine plants and animals.

Socioeconomic status of the people of this coastal belt largely depends upon fishing. Recently, Junput and Sankarpur have appeared as the major landing centres. Shrimp constitute lions share of Sankarpur and Junput trawl landings. Shrimp fishery of this area has grown during the last decade and it has become one of the most valuable contributions to the nation's fishing industry in terms of gross production, gross revenue earnings and export. However, there is no up-to-date comprehensive taxonomic work on the penaeoid shrimps of this region. This work is the first attempt towards a complete taxonomic study of the group.

Bharati Goswami (1992) compiled a list of penaeoid shrimps of Digha beach. This study is based on three lots of collections done by the first author in February, 1995; February, 2000 and March 2000. The specimens were collected from different collection

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Map of Digha Coast and its surroundings

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points like Talsari, New Digha, Old Digha, Junput, Sankarpur and Khejuri. Taxonomic treatment is based on Perez Farfante and Kensley (1997).

An annotated account of the families, genera and species collected during the present study :

Superclass — Crustacea Penant, 1777. Class — Malacostraca Latreille, 1806 Order — Decapoda Latreille, 1803. Suborder — Dendrobranchiata Bate, 1888 Superfamily — Penaeoidea Refinesque – Schmaltz, 1815

Penaeoidea Rafinsque – Schmaltz, 1815 Anal. Nat. Tabl. Univers, : 98

Diagnosis of the superfamily : All five pairs of percopods well developed; pleurobranchia present at least on somite ix; some somites with at least three branchiae on each side; at least 11 well developed gills present on either side of thorax.

Two families viz., penaeidae Rafinesque-Schmaltz, 1815; Solenoceridae Wood-Mason, 1891 have been collected form the study area.

Family - Penaeidae Rafinesque - Schmaltz, 1815.

- Penedia Rafinesque Schmaltz 1815, Anal. Nat. Tabl. Univers : 98 [Subfamily of the Plyonuria].
- Penaedae Bate, 1888, Rep. Scient. Results Voy. Challenger, 24:220; Alcock, 1901, Desc. Cat. Indian Deep-Sea Crust.; De Man, 1911, Siboga Exped., 39:1
- Penaeidae George, 1969, Bull. Cent. Mar. Fish. Res. Inst., 14: 5-48; 1979. Cont. Mar. Sci.,: 21-59
- Penaeidae Perez Farfante, 1978, FAO Sp. Indent. Sh., 6:1; 1988, NOAA Tech. Rep. NMFS, 64 : iii, 8

Diagnosis of the family : Body compressed, slender; well developed rostrum extending to or beyond distal margin of eye; armed with dorsal and sometimes also with ventral teeth; carapace without postorbital spine, post orbital angles sometime prominent; antenal and hepatic spine usually present; cervical sulcus never reaching rostral carina, usually posterior three or four abdominal somite carinate; telson sharply pointed with or without spines.

Optic calathus always present on eye but lacks in mesial tubercle; antennules with well developed prosartema, flagella slender and originating from apex of third segment; third to fifth pleopods biramous; equipod present on first maxilliped and usually on second but lacking on fourth and fifth pereopods.

This family includes six genera in the study area. An account of these are given below.

Genus - Fenneropenaeus Perez Farfante and Kensley, 1997.

Cancer Osbeck, 1765, Aus dem Schw. Uber. J.G. Georgi, Rostock : 552

Peneus Alcock, 1905, Ann. Mag. Nat. Hist., 16: 508-532.

Penaeus De Man, 1911 - Siboga Exp. 39 : 1-131. George, 1969 Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; Muthu and Motoh, 1979, Res. Crust., Carcino. Soc. Japan, 9: 64-70

Penaeus (Fenneropenaeus) Perez Farfante, 1969, Fish. Bull. U.S., 67 (3): 466.

Fenneropenaeus Perez Farfante and Kensley, 1997, Mem. Mus. Nat. d'Hist. Nat., 175:1-233.

Type species : Penaeus indicus H. Milne Edwards, 1837, Hist. Nat. Crust, 2:415.

Diagnosis of the genus : Integument glabrous; rostrum armed with dorsal and ventral teeth, reaching distal end of second antennular segment; carapace with hepatic and antennal spine, lacking orbital and pterigostomial spines; postoccular sulcus and carina absent; post-rostral carina reaching as far as posterior one-eighth of carapace; adrostral carina and sulcus short, usually extending to about the level of epigastric tooth, gastro-frontal carina absent; gastro-orbital carina usually absent, if present occupying middle one-third or not more than posterior two-third of distance between orbital margin and hepatic spine; orbito-antenal sulcus often shallow or poorly marked, cervical carina well marked, but sulcus well defined or feeble; hepatic carina absent or illdefined; branchiocardiac carina absent; sixth abdominal somite with three cicatrices on either side; telson unarmed.

Antennular flagella much shorter than carapace; basial and ischial spines on first percopod, basial spine on second percopod only.

Petasma symmetrical, semiclosed, with distomedian projections usually quite short; appendix masculina subelliptical or subtriangular and armed closely set marginal spines and sometimes with submarginal patches of spinules; thelycum closed, with pair of lateral plates on sternite XIV forming thick mesial lips meeting along midline, shielding simple, median, saclike seminal receptacle.

This genus has only a single species in the study area.

Fenneropenaeus indicus (H. Milne Edwards, 1837)

Penaeus indicus H. Milne Edw., 1837, Hist. Nat. Crust. 2 : 415. Peneus indicus Alock, 1906. Cat. Indian Deca. Crust. Part-III. Mac. Fas. I : 1-55 Fenneropenaeus indicus Perez Farfante, 1969, Fish. Bull. U.S., 67 (3) : 466

Material Examined : 2 Males (60 - 62 mm) and 2 females (60 - 65 mm), Digha Mohana, 10.02.95, A. Chanda, 2 female (55 - 60 mm), Digha Mohana, 20.02.2000. A. Chanda.

Diagnosis of the species : Rostrum slender, long, slightly curved downwards, with 7-9 dorsal teeth and 4-6 ventral teeth; adrostral carina and sulcus extended as far as epigastric tooth; gastrofrontal and hepatic carina absent; gastro orbital carina extending over posterior 2/3 of distance between hepatic spine and orbital margin, distomedian projection of petasma over hanging distal margin of coastae; median margin of lateral plate of thelycum, forming tumid lips with papillae on their inner surface.

Distribution : Indo-West Pacific : East and South East Africa; India to South China, New Guinea and North Australia.

Genus Megokris Perez Farfante & Kensley, 1997

Penaeus (Trachypeneus) Alcock, 1901, Descr. Cat. Indian Deep-Sea Crust., : 15

Trachypeneus Alcock, 1905, Ann. Mag. Nat. Hist. 16 (7): 522

- Trachypeneus De Man, 1911, Siboga Exped., 39 : 87; Dall 1957, Aus. J. Mar. Freshw.
 Res., 8(2) : 136-231; Hall, 1961, Bull. Raffles. Mus., 26 : 76-119., George, 1969,
 Bull. Cent.. Mar. Fish. Res. Inst., 14 : 5-48, 1979, Cent. Mar. Sci., : 21-59, Muthu,
 1971, Indian J. Fish., 15 : 145-154.
- Megokris Perez Farfante & Kensley, 1997. Memoires Du Museum National d'Historie Naturelle, 175 : 233.

Type species : Penaneus granulosus Haswell, 1879, Proc. Linn. Soc. N.S.W., 4:41.

Diagnosis of the genus : Integument thick, densely pubescent; rostrum short, reaching between distal margin of first antennular segment and about end of peduncle, armed only dorsally, epigastric tooth distinctly separated from penultimate tooth; carapace with antennal and hepatic spine, orbital angle sharp or crowned by a small spine; pterygostomian angle blunt; post-ocular sulcus shallow or obsolete; gastro-orbital carina lacking; orbito-antenal sulcus lacking or shallow; cervical sulcus short; hepatic sulcus extending just below hepatic spine; longitudinal suture short; transverse suture distinct; sixth abdominal somaite lacking lateral cicatrix; telson armed with three pairs of movable lateral spines.

Antennular flagella short; basial spine lacking on third maxilleped, present on first and second percopods, small ischial spine on first percopod; lacking on second.

Petasma symmetrical, semiclosed with lateral lobes projecting distolaterally like horn; appendix masculina subquadrangular and rounded corners; thelycum closed, with plate of sternite XIV deeply exacavate anteriorly, anterior plate tongue like.

Thig genus has only one species in the study area.

Megokris granulosus (Haswell, 1879)

Penaeus granulosus Haswell, 1879, Proc. Linn. Soc. N.S.W., 4:41.

Trachypenaeus salaco De Man, 1911, Siboga. Exped., 39 · 87.

Trachypenaeus furcilla Hall, 1961, Bull, Raffles, Mus., 26: 76-119.

- Trachyenaeus gramulosus Dall, 1957, Aust. J. Mar. Freshw. Res. 8 (2) : 136-231; Muthu, 1971, Indian J. Fish., 15 : 145-154
- Megokris gramulosus Perez Farfante & Kensly, 1997, Mem. Mus. Nat. d'Hist. Nat., 175:1-233

Material Examined : 2 males (35-40 mm), Digha mohana, 20.2.2000, A. Chanda.

Diagnosis of the species : Body public pub

be very prominent and is often fused to posterior plate; posterior plate excavated on either side of median convexity.

Distribution : Indo - West Pacific : Kuwait, India, Sri Lanka, Malaya, Indonesia, Taiwan, Australia.

Genus Metapenaeopsis Bouvier, 1905

- Matapenaeopsis Bouvier, 1905, C. r. Herd, Seanc, Acad Sci., Paris, 140 : 981. Hall, 1962, Fish. Publ. Colonia Off. London, 17:1-229
- George, 1969. Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; 1979, Cont. Mar. Sci., 21-59; Muthu, 1971, Indian J. Fish., 15 : 145-154.

Metapenaeus Wood - Mason, 1891, Ann. Mag. Nat. Hist., 187-199

Metapeneus Alcock, 1905, Ann. Mag. Nat. Hist., 16 : 508-532; 1906. Cat. Indian Deca Crust, Penaeopsis De Man. 1911, Siboga Exped Monogr., 39 : 1-131

Type species : *Metapenaeopsis pubescens* Bouvier, 1905, C.r. herd, Seanc. Acad. Sci., Paris, 140 : 981.

Diagnosis of the genus : Integument usually thick and pubescent sometime dense, rostrum with dorsal teeth only, slender not overreaching antennular peduncle, epigastric tooth usually separated from penultimate tooth, carapace with small orbital angle and developed antennular, pterygostomian and hepatic spine, branchiostegal spine absent, postocular sulcus lacking; orbitoantenal and cervical sulcus usually ill defined; hepatic sulcus usually feeble; hepatic carina indistinguishable; sutures absent; in some species stridulating organ present; sixth abdominal somite without cicatrix; telson armed with pair of fixed subapical spines preceded by three pairs of conspicuous movable spines.

Antennule with parapenaeid spine, sometime vestigial; flagella variable in length, usually much shorter but sometime longer than carapace; third maxilliped with a prominent basial spine, first and second percopod so, always lacking on third.

Petasma asymmetrical, divided at about midlength into proximal and distal parts, latter complex, subdivided into several projections and components; appendix masculina trumpet shaped; thelycum closed, with transverse plate on sternite XIV and strongly developed median protuberance on sternite XIII.

Genus Metapenaeopsis has only one species in the study area.

Metapenaeopsis stridulans (Alcock, 1905)

Metapeneus stridulans Alcock, 1905, Ann. Mag. Nat Hist., (7) 16; 581, 526.

Penaeopsis stridulans De Man, 1911, Siboga Exped., 39: 1-131.

Metapenaeopsis stridulans Hall, 1962, Fish. Publ. Colonia, Off., London, 17 : 1-229.

Material examined : 1 Male (57 mm) and 2 female (55-70 mm), New Digha 3.2.95.A. Chanda; 2 Male (50-58 mm), Digha Mohana, 19.03.2000, A. Chanda

Diagnosis of the species : Entire body pubescent; rostrum low, usually straight and directed forward, armed with 7-8 dorsal teeth only, the penultimate tooth generally anterior to orbital margin of carapace; stridulating organ consisting of 5-7 ridges in a wide, straight band; middorsal carina on third abdominal segment with a usually broad sulcus; petasma asymmetrical, right distoventral projection shorter and bearing a few small apical process, left distoventral projection with 5 to 12 large apical processes, inner intermediate strip broadly quadrangular, distomedian lobule slightly shorter but much broader; thelycal plate subquadrate with rounded corners and slightly wider than long, intermediate plate broadly trapezodial and coxal plates of fourth pereopod smaller than thelycal plate.

Distribution : Indo-West Pacific : Arabian sea to the Malay Arclipelago, New Guinea and New Britain

Genus Metapenaeus Wood - Mason, 1891

- Metapenaeus Wood-Mason, 1891. Ann. Mag. Nat. Hist. (6) 8 : 271, George, 1969, Bull, Cent. Mar. Fish. Res. Inst., 14 : 5-48, 1979. Cont. Mar. Sci., : 21-59. Muthu, 1971, Indian J. Fish., 15 : 145-154
- Penaeus Miers, 1878. Proc. Zool. Soc., London, : 301.
- Mangalura Miers, 1878 Proc. Zool. Soc., London, : 203
- Peneus (Metapeneus) Alcock, 1901. Desc. Cat Indian Dec. Sea Crust., : 1-286
- Metapeneus Alcock, 1905, Ann. Mag. Nat. Hist. 16: 508-532. Alcock, 1906, Cat. Indian Dec. Crust. Part-III mac. Fas. 1: 1-55.

Existence of generic name of Metapenaeus Wood-Mason, 1891 over Mangalura

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Miers, 1878, was decided by the International Commission on Zoological Nomenclature, 1969, Opinion 864, Bull. Zool. Nom., 25 (4/5): 140.

Type species : Penaeus affinis H. Milne Edwards, 1837, Hist. Crust. 2 : 416.

Diagnosis of the genus : Integument more or less pubescent, sometimes glabrous; rostrum with dorsal teeth only; epigastric tooth generally separated from penultimate; carapace with sharp orbital angle and prominent antenal and hepatic spine; pterygostomian corner rounded; post occular sulcus deep, gastro-orbitoantenal and cervical sulci well marked; hepatic sulcus placed anterior to hepatic spine and accompanied by ventral carina often descending almost vertically from hepatic spine then turning towards pterygostomian angle, posterior part of hepatic sulcus ill-defined or absent; branchio-cardiac carina variably developed; sixth abdominal somite bearing single continuous long cicatrix or interrupted to two or three parts; telson lacking fixed spine but bearing movable, sometimes minute and numerous.

Antennular flagella shorter than carapace; basial spine present on first through third percopods; exopod lacking on fifth percopod.

Petasma symmetrical, semiclosed, depressed, with median lobes usually produced into simply curved, hoodlike or convoluted distal projections, appendix masculina narrow basally, greatly expanded distally and strongly convex ventrally; thelycum closed, with paired lateral plates of sternite XIV.

Genus Matapenaeus Wood-Mason, 1891 has three species in the study area.

Metapenaeus affinis (H. Milne Edw., 1837).

Penaeus affinis H. Milne Edw., 1837, Hist. Nat. Crust., 2:416

Metapeneus affinis Alcock, 1906. Cat. Indian Dec. Crust. Part-III Mac. Fas-I: 1-55

Metapenaeus affinis, George 1969. Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; 1979, Cont. Mar. Sci. : 21-59; Muthu, 1971, Indian J. Fish., 15 : 145-154.

Material Examined : 3 Male (35 - 45 mm) and 6 female (30 - 50 mm), Junpur, 5.2.95, A. Chanda; 5 Male (19 - 47 mm) and 2 female (30 - 52 mm), Digha Mohana, 19.03.2000, A Chanda.

Diagnosis of the species : Entire body public public public public prostrum armed with 8-11 dorsorostral teeth only; advostral carina ending behind second rostral tooth and sulcus little behind epigastric tooth; telson armed with minute spinules, a short ischial spine present on first percopod; cresent shaped distomedian projection on petasma, disolateral projection directed anterolaterally; anterior plate or thelycum deeply grooved longitudinally and wider posteriorly.

Distribution : Indo-West Pacific : Arabian sea, India, Pakistan to Malay Archipelago and Hong Kong.

Metapenaeus brevicornis (H. Milne Edw., 1837)

Penaeus brevicornis H. Milne Edw., 1837, Hist. Nat. Crust. 2:417

Penaeopsis brevicornis Kemp, 1915, Mem. Indian Mus., 5: 201-325

Metapenaeus brevicornis George, 1969, Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; 1979. Cont. Mar. Sci. : 21-59; Muthu., 1971, Indian J. Fish., 15 : 145-154.

Meterial Examined : 1 Male (52 mm) and 1 female (50 mm) Old Digha, 20.02.2000, A Chanda, 2 female (45 - 55 mm), Digha Mohana, 19.03.2000, A. Chanda.

Diagnosis of the species : Body smooth, rostrum armed with 5-7 dorsal teeth only. distal end toothless about half of rostrum, crest high, adrostral carina and sulcus reaching as far as second rostral tooth; telson armed with two large spine; first pereopod with an ischial spine; distomedian projection of petasma with a long and slender apical filament, anterior plate of thelycum large square and grooved, lateral plates boomerang shaped and enclosing two pearshaped plates.

Distribution : Indo-West Pacific : Pakistan, India to Malaya, Indonesia and Thailand.

Metapenaeus ensis (De Haan, 1844)

Penaeus ensis De Haan, 1844, In Vonsiebold Fauna Japonica, Crustacea (6/7): Pl. 46, Fig 2.

Penaeus mastersi Haswell, 1879, Proc. Linn. Soc. N.S.W., 4 (1): 38-44.

Penaeus incisipes Bate, 1888, Rep. Sci. Res. "Challenger", 24 : 1-942.

Metapenaeus masterisii Haswell. 1879, Proc. Linn. Soc. N.S.W., 4 (1): 38-44

Penaeopsis monoceros De Man, 1911, Siboga Exped., 39: 1-131

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Metapenaeus ensis George, 1969, Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; Muthu, 1971, Indian J. Fish., 15 : 145-154

Material Examined : 2 Male (45 - 50 mm) and female (55 mm) Digha Mohana, 2.2.95, A Chanda.

Diagnosis of the species : Entire body pubescent; rostrum armed with 8 teeth along entire dorsal margin, adrostral carina ending behind second rostral tooth, and sulcus behind epigastric tooth; telson armed with spinules; a small ischial spine of first pereopod; in males, merus of fifth perepod with a proximal notch followed by a long inwardly curved spiniform process and a row of tubercles; distomedian projections of petasma convoluted, greatly swollen and directed forward, triangular in shape; anterior plates with strongly raised lateral margins forming posteriorly two inwardly curved triangular projections.

Distribution : Indo-West Pacific : Pakistan, India to Malaya, Indonesia and Thailand.

Genus Parapenaeopsis Alcock, 1901

Peneus (Parapeneopsis) Alcock, 1901, Descr. Cat. Indian Deep-Sea Crust., : 14; 1906, Cat. Indian Dec., 3 (1) : 34.

Penaeus H. Milne Edwards, 1837, Hist. Nat. Crust., 2:411 [Part].

Penaeopsis Bate, 1881, Ann. Mag. Nat. Hist., 8 (5), 183.

Metapeneus Alcock, 1905, Ann. Mag. Nat. Hist., 16 (7) (95); 516 [part]

Parapenaeopsis [emendment of Parapeneopsis Alcock, 1901, under the plenary powers by the International Commission on Zoological Nomenclature, 1969] - De man, 1911-Kubo, 1949-Barnard, 1950-Dall, 1957-Racek and Dall, 1965-Liu and Zhong, 1988.

Type species : Penaeus styliferus H. Milne Edwards, 1837, Hist. Nat. Crust., 2:418

Diagnosis of the genus : Integument thin; minutely setose-punctate, never dense, rostrum with dorsal teeth only, variable in shape and size; epigastric tooth either closed to dorsal rostral teeth or distinctly separate or vestegial or absent; carapace with small orbital spine, sometimes reduced to tubercle, to form sharp angle; well developed antenal and hepatic spine; pterygostomian spine absent, sometimes angle sharp, postocular sulcus well defined; gastroorbital carina lacking; orbito antenal and cervical sulci feeble; hepatic sulcus usually clearly marked anteriorly, sometimes

feebly posteriorly; hepatic carina often present, occasionally feebly, usually sharp and descending anteroventrally from hepatic spine then extending almost to pterygostomian angle, usually blunt posteriorly, longitudinal suture falling short of posterior margin of carapace, transverse suture usually present; sixth abdominal somite with cicatrix; telson armed or unarmed.

Antennular flagella subequal, may be short or long; basial spine present or absent on first and second percopods; ischial spine present or absent on first percopod.

Petasma symmetrical, semiclosed, variable in shape; appendix masculina simple, or sometimes consisting of two halves, proximal and distal one; thelycum closed, with plate on sternite XIV, broad, sometimes deeply emerginate anteriorly.

The genus Parapenaeopsis Alcock, 1901 has four species in the study area.

Parapenaeopsis hardwickii (Miers, 1878)

Penaeus hardwickii Miers, 1878, Proc. Zool. Soc. Lond., 1878: 300, 306.

- Parapeneopsis sculptilis hardwickii Alcock, 1906, Cat. Indian. Dec. Crust. Part-III. Mac. Fas. 1 : 1-55.
- Parapenaeopsis hardwickii George. 1969. Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; Muthu, 1971, Indian J. Fish., 15 : 145-154.

Material Examined: 1 female (51 mm), Digha, 12.10.95, M. De.

Diagnosis of the species : Rostrum armed with 8 dorsal teeth only. sigmoidal, toothless on distal half and upcurved, epigastric tooth reduced; telson armed with four pairs of movable spines; epipod and basial spine present on first and second pereopod; petasma with wing like distomedian projections, their anterior margin often crenulate, distolateral projections short and directed laterally; anterior plate of thelycum concave, rounded anteriorly, posterior plate flat, with a pair of anterolateral tooth-like projections.

Distribution : Indo-West Pacific : Pakistan, India to China and Indonesia.

Parapenaeopsis sculptilis (Heller, 1862)

Penaeus sculptilis Heller, 1862, Verh. Zool. Bot. Ges. Wien., 12:528

Parapenaeopasis sculptilis cultrirostri Alcock, 1906, Cat. Indian Dec. Crust., Part III. Mac. Fas I : 1-55

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Material Examined : 2 male (35 - 36 mm) and 3 female (30 - 40 mm), Digha Mohana, 20.02.2000. A. Chanda.

Diagnosis of the species : Rostrum armed with 7 dorsal teeth only, in females sigmoid-shaped and in male knife-shaped, longitudinal suture generally reaching three fourth of carapace length; epipod and basial spine present on first and second pereopods; telson unarmed petasma with long, rabbit ear-shaped ditomedian projections, deeply concave ventrally, distolateral projections short, directed anterolaterally; thelycum with anterior plate directly rounded and broadly articulating with posterior plate, the latter with a median tubercle bearing a tuft of long setae.

Distribution : Indo-West Pacific : Pakistan, India to Malay, Hong Kong, Philippines, Indonesia, New Guinea and N. Australia.

Parapenaeopsis stylifera coromandelica Alcock, 1906

Parapenaeopsis stylifera coromandelica Alcock, 1906 Cat. Indian Decap. Crust., 3 (1): 37, George, 1969. Bull. Cent. Mar. Fish. Res. Inst. 14: 5 - 48; Muthu, 1971, Indian J. Fish. 15: 145-154

Material Examined : 2 female (40-42 mm), Sankarpur, 5.2.95, A. Chanda; 2 female (35-40 mm) and 1 male (42 mm) Khejuri Market, 7.2.95, A. Chanda; 1 male (36 mm) Digha Mohana, 2.2.95, A. Chanda; 2 male (30-40 mm), Digha Mohana, 20.2.2000, A. Chanda.

Diagnosis of the species : Rostrum sigmoidal, distal half toothless, armed with 6-8 dorsal teeth only, longitudinal suture reaching 2/3 of carapace length; telson armed with two pairs of subapical fixed spines; epipod and basial spine present on first and second pereopods; distolateral projections of petasma slender, horn-like and straight, directed anterolaterally, with vento-external openings; anterior plate of thelycum square and concave, with a slender stem-like posterior process, posterior plate deeply notched.

Distribution : Indo-West Pacific : India, Sri Lanka, Indonesia.

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Parapenaeopsis uncta Alcock, 1905

Parapenaeopsis uncta Alcock, 1905, Ann. Mag. Nat. Hist. (7) 16 : 522; George, 1969. Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; Muthu, 1971, Indian J. Fish., 15 : 145-154

Parapenaeopsis probata Hall, 1962, Fish. Publ. Colonia Off., 17: 1-229.

Material Examined : 1 male (36 mm) and 2 female (35 - 40 mm) Digha Mohana, 19.3.2000 A. Chanda.

Diagnosis of the species : Rostrum armed with 9 dorsal teeth only; toothless and distal ¼ curved upward; telson unarmed; epipod and basical spine present on first and second perepod, in male basial spine of second pereopod is reduced; distolateral projections of petasma tapering to end each with a long dorsomedian spine-like process; anterior plate of thelycum wider and shorter than posterior plate, rounded anteriorly and with two longitudianl suture is prominent.

Distribution : Indo-West Pacific : Pakistan, India, Bangladesh and Sri Lanka.

Genus Penaeus Fabricius, 1798

Penaeus Fabricius, 1798, Suppl. Entomol. Syst., 408; George, 1969. Bull. Cent. Mar. Fish

Res. Inst., 14; 5-48; muthu. 1971. Indian J. Fish., 15; 145-154

De man, 1911. Siboga Exped., 39; 1-131.

Peneus Alcock. 1901. Des. Cat. Indian Deep Sea Crust. : 1-286

Penaeus (Penaeus) Perez Farfante, 1969. Fish. Bull. U.S., 67 (3); 461-591.

TypeSpecies: Penaeus monodon Fabricius, 1798, Suppl. Entomol. Syst, : 408.

Diagnosis of the genus : Integument glabrous; roustom with dorsal and 2-4 (usually 3) ventral teeth moderately long, reaching to about middle of second antennular segment; carapace with well developed antennal and hepatic spine, lacking orbital and pterygostomian; cervical sulcus shallow; postrostral carina reaching almost to posterior margin of carapace; adrostral carina and sulcus reaching upto the level of epigastric tooth; gastrofrontal carina absent; gastro-orbital carina short, orbito autenal sulcus well marked; cervical and hepatic carina well marked, cervical sulcus

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shallow; branchiocardiac carina absent. Sixth abdominal somite bearing three lateral cicatrices. Telson unarmed. Antennular flagella shorter than carapace. Basial spine on first and second pereopod. Petasma symmetrial, semiclosed with distomedian projections quite short, ventral costae narrowly paping or continuous, long and strongly curving distally. Appendix masculina subelliptical or suboval, armed with numerous marginal spines. Thelycum closed, with pair of lateral plates of sternite XIV meeting along midline, shielding simple median saclike seminal receptacle.

Only one species, *Penacus monodon* Fabricius, 1798 has been collected from study area during the period.

Penacus monodon Fabricius, 1798

Penacus monodon Fabricius, 1798, Suppl. Ent. Syst. 408; George, 1969. Bull. Cent. Mar. Fish. Res. Inst., 14:5-48; 1979, Cont. Mar. Sci., : 21-59; Muthu, 1971, Indian J. Fish, 15: 145-154.

Penacus Carinatus Dana, 1852, Proc. Acad. Nat. Sci. Art., 9(2): 129 - 133.

Penacus tahitensis Heller, 1862, Verh. Zool. Ges. Wien. 12:519-528

Material Examined : 1 Male (162 mm) and 1 Female (170 mm) Digha Market, 20.2.2000. A. Chanda.

Diagnosis of the species : Rostrum typically sigmoidal, armed with 7–8 dorsal and 3–4 ventral teeth; adrostral carina and sulcus extending as far as epigastric tooth; gastro-frontal carina absent; post rostral carina without sulcus; hepatic ridge long, curved, distinctly separate from autenal crest; abdomen with transverse bands.

Distribution : Indo-West Pacific – East and South-East Africa, India, Pakistan, Japan, the Malay Archipelago, Indonesia, New Guinea and Northern Australia.

Family Solenoceridae Wood-Mason, 1891

Solenocerina Wood-Mason, 1891, Ann. Mag. Nat. Hist., (6) 8:275

Peneinae Alcock, 1901, Descr. Cat. Indian Deep Sea Crust., : 13 [part]

Solenocerinae George, 1969, Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48; 1979, Cont. Mar. Sc., 21-59; Muthu, 1971. Indian J. Fish. 15 : 145-154

Solenoceridae Perez Farfante, 1978, FAO Sp. Indent. Sh. 6:2

Diagnosis of the family : Integument thin or firm; rostrum seldom reaching beyond antennular peduncle, armed with dorsal teeth only, carapace with postorbital spine or postantennal spine and hepatic spine, antennal spine always present; orbital, branchiostegal and pterygostomian spines present or absent; cervical sulcus well marked reaching or almost reaching postrostral carina; telson apically acute, usually armed rarely unarmed.

Optic calathus & mesial tubercle always present on eye; prosartema variable in length; flagella usually very long, slender, subcylindrical or flattened; exopods on all maxillipeds and percopods; third through fifth pleopods biramous.

This family has only one genus, *Solenocera* Lucas, 1849 and one species *Solenocera* crassicornis (H. Milne Edwards : 1837) in this area.

Genus Solenocera Lucas, 1849

Penaeus Fabricius, 1798, Suppl. Ent. Syst., : 408

Solenocera Lucas, 1849a, Rev. Mag. Zool, (2) 1:159

Solenocera Alcock, 1901, Descr. Cat. Indian Deep-Sea Crust., : 19. George, 1969, Bull.cent. Mar. Fish. Res. Inst. 14 : 5-48; Muthu, 1971, Indian J.Fish., 15 : 145-154

Philonicus Bale, 1888, Rep. Scient. Results Voy. Challenger, 24: 273

Pleoticus Bate, 1888, Rep. Scient. Results Voy. Challenger, 24 : xii [Part].

Parasolenocera Wood-Mason, 1891, Ann. Mag. Nat. Hist. (16): 276.

Type species : Solenocera philippii Lucas, 1849, Rev. Mag. Zool. Appl., 1 (2) : 298-300.

Diagnosis of the genus : Integument glabrous or sparsely setose on carapace, dorsal patch of setae on rostrum and anterior part of carapace; rostrum never overcome second antennular sgment, armed with dorsal teeth only; epigastric and first rostral teeth variably separated; carapace with orbital spine present or absent; armed with postorbital, antennal and hepatic spines; postantennal spine absent; pterygostomian or branchiostegal spine present or absent; cervical sulcus strongly defined, long, reaching of almost reaching postrostral carina; hepatic sulcus deep, nearly horizontal posterior to level of hepatic spine, from there turning anteroventrally and reaching pterygostomian angle; hepatic carina sharp anteriorly; branchiocardiac carina and

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sulcus present or absent; telson with fixed posterolateral spine, movable spine absent.

Antennule with well developed prosartema; antennular flagella longer than carapace, lamellate, broad, ventral pair follecious; basial and ischial spines present on first percopod; ischial spine usually on second percopod; exopod present on all maxilliped and percopods; petasma semiopen; apendix masculina present; thelycum open.

Only one species, *Solenocera crassicornis* (H. Mine Edwards, 1837) was collected from the study area.

Solenocera crassicornis (H. Milne Edwards, 1837)

Penaeus crassicornis H.MilneEdwards, 1837, Hast. Nat.Crust., 2:418

Solenocera sinensis Yu, 1937, Ibid., 7 : 111-117.

Solenocera indicus Nataraj, 1945, J. Roy. Asiat. Soc. Bengal, 11 (1) : 91-98; George, 1969, Bull. Cent. Mar. Fish. Res. Inst., 14 : 5-48

Solencera subnuda Kubo, 1949, J. Tokyo Coll, Fish., 36 (1): 1-467

Solenocera Kuboi Hall, 1956, Bull. Raffles Mus., 27: 68-70

Solenocera crassicornis Muthu, 1971, Indian J. Fish., 15: 145-154

Material Examined : 2 male (50 - 55 mm) and 1 female (62 mm), Digha Mohana, 10.02.95. A. Chanda.

Diagnosis of the species : Rostrum reaching little beyond the distal margin of eye and armed with 8 dorsal teeth; post rostral crest low and rounded; telson unarmed and not trifurcate.

Distribution : Indo-West pacific : Pakistan, West and East Coast of India, Sri Lanka to Chian, Japan and New Guinea.

		Recorded by Bharati Goswami (1992)	Present Record
1.	Atypopenaeus stenoductylus (Stimpson, 1860)	+	-
*2.	Metapenaeopsis stridulans (Alcock, 1905)	-	+
3.	Metapenaeus brevicornis (H. Milne Edw., 1837)	+	+
4.	M. affinis (H. Milne Edw., 1837)	+	+
5.	M. dobsoni (Miers, 1878)	+	-
6.	M. lysianaessa (De Man, 1888)	+	-
7.	M. Monoceros (Fab., 1798)	+	-
*8.	M. ensis (De Haan, 1844)	-	+
9.	Parapenaeopsis acclivirostris (Alcock, 1905)	+	-
10.	P. stylifera coromandelica (Alcock, 1906)	+	+
11.	P. sculptilis (Heller, 1862)	+	+ ``
12.	P. maxillipedo Alcock, 1905	+	-
13.	P. uncta Alcock, 1905	+	+
*14.	P. hardwickii (Miers, 1878)	-	+
15.	Fenneropenaeus indicus (H. Milne Edw., 1837)	+	+
16 .	Melicertus canaliculatus (Olivier, 1811)	+	-
17.	Penaeus monodon Fab. 1798	+	+
18.	P. semisulcatus De. Haan, 1844	+	-
19.	Trachysalambria curvirostris (Stimpson, 1860)	+	-
*20.	Megokris granulosus (Haswell, 1879)	-	• +
21.	Solenocera choprai Nataraj, 1845	+	-
22.	S. crassicornis (H. Milne Edw., 1837)	+	+
23.	Parapenaeopsis longipes Alcock, 1905	+	-
	Total	19	12

Table 1. A list of shrimps likely to be found in Digha (West Bengal) coast.

* New record from the region

A key to the families, genera and species likely to be found in this region is given below :

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Key to the families of Penaeoidea of Digha coast

1. Cervical groove never reaching more than two-third distance from the hepatic spine to the post rostral carina, antennular flagella short and slender *Penaeidae* Rafinesque Schmaltz 1815.

Family Penaeidae Rafinesque - Schmaltz, 1856.

Key to the Genera of the Family Penaeidae, recorded from Digha

1.	Presence of ventral teeth on rostrum2
	Absence of ventral teeth on rostrum 4
2.	Gastrofrontal carina absent
	Gastrofrontal carina present Melicertus Rafinesque - Schmaltz, 1814
3.	Hepatic carina prominent Penaeus Fabricius, 1798
	Hepatic carina absent or trace of hepatic carina Fenneropenaeus Perez Farfante and Kensley, 1997
4.	Telson with fixed spine, petasma asymmetrical Metapenaeopsis Bouvier, 1905.
	Telson without fixed spine, petasma symmetrical5
5.	Presence of exopod on fifth pereopod
	Absence of exopod on fifth pereopod Metapenaeus Wood-Mason, 1891
6.	Presence of longitudinal suture on carapace7
	Absence of longitudinal suture on carapace Atypopenaeus Alcock, 1905
7.	Longitudinal suture long, extend beyond epigastric tooth, body smooth and no dense hair on abdomen
	Longitudinal suture short, not extend beyond epigastric tooth, body densely hairy
8.	Petasma with distolateral projection curving forward forming a tapering horn

8. Petasma with distolateral projection curving forward forming a tapering horn like extention, anterior plate of thelycum broad with a caudal extension

Genus Metapenaeus Wood Mason & Alcock, 1891

Key to the species

1.	Body smooth; telson with spine
	Body pubesent ; telson without spine2.
2.	Rostrum short, not reaching first segment of antennular peduncle; adrostral crest ending before second rostral tooth
	Rostrum long, exceeding second segment of antennular peduncle; adrostral crest ending beyond second rostral tooth
3.	Branchiocardiac ridge long and reaching hepatic spine, lateral plates of thelycum not like horse-shoe shaped
	Branchiocardiac ridge short and not reaching hepatic spine, lateral plates of thelycum forming horse-shoe like structure
4.	Distomedian projection of petasma crescent shaped M. affinis (H.M. Edw., 1837).
	Distomedian projection of petasma not crescent shaped
5.	Distomedian projection of petasma bulbiform; lateral plate of thelycum with strongly raised lateral margins forming two lingitudinal crest
	Distomedian projection of petasma triangular; lateral plate of thelycum with strongly raised lateral margins forming posteiorly two inwardly curved triangular projections

Genus Parapenaeopsis Alcock, 1901

Key to the species

1. Presence of epigastric tooth; Post-rostral crest ending near margin of carapace; mastigobranchiae present on first and second periopod2.

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Absence of lateral spine on telson; teeth present upto the trip of rostrum 4.

3. Absence of sulcus on post carina; distolateral projection of petasma long hornlike, directed anterolaterally, telson with 1 or 2 pair of fixed spine...... *P. stylifera coromandelica* Alcock, 1906.

Second percopod without basial spine; distolateral projection of petasma tapering at tip *P. uncta* Alcock, 1905

Third percopod without basial spine; tuff of hair present on middle of posterior thelycal plate, distomedian projection of petasma long rabbit ear-like, distolateral projection short, directed anterolaterally *P. sculptilis* (Heller, 1862).

Genus Penaeus Fabricius, 1798

Key to the species :

Family Solenoceridae Wood-Mason, 1891 Genus Solenocera Lucas, 1849

Key to the species

1.	Postrostral crest markedly elevated and laminose, telson with a pair of lateral
	fixed spine
	Postrostral crest low and rounded, telson without spine

Discussion:

The present study has revealed the existence of twelve species of shrimp under seven genera and two families from Digha and its adjacent areas. Of this, one genus and four species are being recorded here for the first time from this region. In an earlier investigation Bharati Goswami (1992) recorded nineteen species under five genera (Table 1). Of the nineteen species recorded by him, ten could not be collected in the present study and the exact identity of one species viz., *P. longipes* Alcock has not been clearly determined validity of which has been doubted by the present authors. It is likely that some of the species might have disappeared from this region in the meantime However, more detailed and extensive study is required to confirm this. It may be concluded that there is a possibility of the presence of 23 species in this region.

Reference

- Bharati Goswami, B.C. 1992 Marine Fauna of Digha Coast of West Bengal, India, J. Mar. Biol, Ass. India. 34 (1 & 2): 115 - 137
- Gupta, A. 1970. The West Bengal Coast : Its nature and evolution. In : A.B. Chatterjee,A. Gupta and P.K. Mukhopadhyuay (Ed.) 'West Bengal'. The Geographical Institute, Presidency College, Calcutta, pp. 30-37.
- Perez Farfante and Kensley, 1997. Penaeoid and Sergestoid Shrimps and Prawns of the World. Keys and Diagnoses for the Families Mem. Mus. Nat. d'Hist. Nat., 175 : 1-233.

WORK STRESS ASSESSMENT OF WORKERS ENGAGED IN A FOOD GRAIN DEPOT

C. K. Pradhan* and S. Thakur

Regional Occupational Health Centre (Eastern) (Indian Council of Medical Research) Block DP, Sector V, Salt Lake Kolkata - 700 091

ABSTRACT

Workers engaged in food grain storage depots carry out different types of jobs like lifting and carrying loaded sacks, cleaning spilled grains, spraying pesticide, administrative work, etc. The objectives of the study were to assess the workload of the workers, identify their work postures and evaluate feelings of pain and discomfort of the workers. The study was carried out with 200 workers (116 load-handling workers, 32 ancillary workers, 13 administration-DMO and 39 administration-Depot workers) engaged in a storage depot. Workload was assessed subjectively on a scale of 1 (light) to 5 (extremely heavy). Peak heart rates of load-handling workers were recorded and energy expenditures were calculated. The work postures were noted by observation and photography. Pain and discomfort in. different body parts reported by the workers were noted. Subjective assessment of the load-handling workers showed the workload as 'heavy' (39%) or 'very heavy' (40%). Ancillary workers, administration workers - DMO and Depot reported their workload as 'moderate' or 'heavy'. The mean peak heart rate of the load handling workers was $125.44.9 \pm 13.14$ b.min⁻¹. Their energy expenditure varied from 12.893 to 23.818 kJ min⁻¹ indicating 'moderate' to 'heavy' workload. Thus the results of subjective assessment were comparable with peak heart rate and energy expenditure. Musculoskeletal pain and discomfort was reported by 55% of the total workers and the most affected body part was back followed by leg. This study showed that workload of the workers may be assessed subjectively as well as from the peak working heart rate and energy expenditure. Change and work practice and support by equipment would help in reducing the work stress.

KEYWORDS : Load handling workers, Ancillary workers, Administrative workers, Subjective perception, Workload, Heart rate, Body pain, Posture.

Dr. C. K. Pradhan, Regional Occupational Health Centre (Eastern) Block DP, Sector V, Salt Lake Kolkata - 700 091, E-mail : <u>pradhanckss@vanl.net</u>

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^{*} Address for Correspondence :

Introduction

Food grains like rice, wheat and sugar are procured and stored by various organisations such as Central Warehouse Corporation, Food Corporation of India, State Warehouse Corporation, etc. These organisations have their own depot consisting of a number of godowns where the food grains are stored. Later, these are distributed to different parts of the districts and states as per requirement. A substantial number of load handling workers remain engaged for handling the food grain sacks. Besides, three other categories of workers are also engaged in food grain depot namely anacillary workers, the administrative workers in depot manager's office (Administration - DMO) and administrative workers working inside the godowns (Administration - Depot).

The load handling workers carry the sacks from a railway wagon or a truck to a godown and vice versa. Sometimes, they shift the sacks from one stack to another place within a storehouse. A small hand tool called hook is used by the workers to facilitate the handling and placing of the heavy sacks. Two persons, each with a pair hooks, lift the sack full of food grains from the stack of floor and place it on the head of a load handling workers. The worker may stand with back and neck straight, bent or twisted depending on the height of the stack. The weight of the sacks varies from 50 to 100 kg; sometimes the workers carry two sacks of 50 kg at a time. They walk a distance of 5 to 50 meter with heavy sacks on head. Often the workers have to climb steps made of food grain sacks and/or walk among an upward slope of wooden platform. They perform their job in a group of ten to fifteen individuals.

The ancillary workers perform different types of jobs, e.g., gathering of grain spilled on the floor, repairing and stitching of sacks, cleaning of grains, brushing, spraying of pesticide, fumigation and opening and closing of railway wagons. The administrative workers mostly perform clerical jobs. The administrative workers (Administration – DMO) do various kinds of paper-work (clerical jobs), which are sedentary in nature. However, besides the clerical job, the Administration – Depot workers also do stock verification of food grains during storage and distribution. Sometimes, they need to get up on a stack of sacs inside the depot and also on loaded trucks or railway wagons for this purpose.

Some studies related to the work stress have been reported for workers engaged in different occupations (Asfour *et al*, 1983; Waikar *et al*, 1991; Pradhan, 1999; Pradhan and Thakur, 2002). The objective of the present study was to assess the workload, work postures, musculoskeletal pain and discomfort of the workers engaged in food grain depot.

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Methods

The study was conducted on a total of 200 workers, involving 116 load handling workers, 32 ancillary workers, 13 administration-DMO and 39 administration-Depot workers. The purpose of the study was explained to them and their consent was taken. Age and physical characteristics like height and weight of the subjects were recorded.

The subjects were interviewed through a pre-designed questionnaire. Subjective assessment of the workload was carried out on a scale of 1 to 5 (Sinclair, 1995) and the verbal expressions, 'light', 'moderate', 'heavy', 'very heavy' and 'extremely heavy' were used. For physiological assessment of workload (Astrand and Rodahl, 1986), working heart rates of 90 load-carrying workers were recorded. Ten heartbeats were counted with the help of a stethoscope placed at the left fifth intercostals space of the chest of the subject and the time (sec) was noted with a stopwatch and then converted into beats per minute. Peak heart rate of the load handling workers was noted during work and energy expenditure was calculated using the equiation of Datta and Ramanathan (1969). The postures adopted by the workers during work were recorded by observation and still photography. The subjects were shown a pictogram of the body and were asked to point out the part(s) where they feel work related musculo-skeletal pain and discomfort (Corlett and Bishop, 1976). For better understanding, the entire body was categorised into four areas -1) head and neck, 2) back, 3) arms and 4) legs, for expression of pain and discomfort.

Body surface area of the subjects was determined by using the equation of Banerjee and Sen (1955). The nutritional statuses of the subjects were assessed from the body mass index (WHO 1995). Statistically significant differences between mean values for variable of physical characteristics were assessed by Student's t-test.

Results

Physical characteristics

The physical characteristics of the subjects have been presented in Table 1. The average age of the load handling workers was the lowest and it was significantly different (p < .001) from other categories of workers. The mean body surface area of administration – depot workers was higher (p < .05) compared to both load handling workers and ancillary workers. The body mass index of the subjects in various categories was above 20 kg. m⁻².

Table 1

Age and other physical characteristics of workers engaged in food grain depot (Mean \pm SD)

Category	No. of Subject	Age (Years)	Height (cm)	Weight (Kg)	Body Surface Area (m ²)	Body Mass Index (kg. m ⁻²)
Load handling	116	32.5	167.99	58.71	1.73	20.80
workers		10.24	5.75	9.77	0.14	3.27
Ancillary	32	48.94***	164.34**	57.91***	1.69	21.39
workers		4.81	7.24	10.88	0.17	3.42
Administration –	13	47.38***	169.54@	59.85@@@	1.75	20.76
DMO		9.23	6.87	10.34	0.17	3.08
Administration –	39	49.94***	168.56@	63.15*@@@	1.79*@	22.11*
Depot		5.70	6.86	10.25	0.17	2.58

* = p<.05, **=p>.01, ***=p<.001 when compared with handling labour

@=p<.05, @@=p<.01, @@@=p<.001 when compared with ancillary worker

Workload assesment

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The assessment of workload of the workers was done by subjective assessment as well as from peak heart rate during work. The workload score varied from 1 (light) to 4 (very heavy). The results of the subjective assessment of different categories of workers have been presented in Table 2. The data showed that workload of the most of the load handling workers was 'heavy' (39%) to 'very heavy' (40%). Majority of the ancillary workers (41%) reported their workload as 'moderate'; however, 'light' (25%) and 'very heavy' (6%) were also expressed by them. Majority of the administrative workers (DMO and Depot) reported the workload as 'moderate'.

Table 2

Subjective assessment of workload of different categories of workers of food grain depot

Workload	Load handling workers (n=116)	Ancillary workers (n=32)	Administration - DMO (n=13)	Administration - Depot (n=39)
Light	7 (6%)	8 (25%)	2 (15%)	2 (5%)
Moderate	17 (15%)	13 (41%) .	7 (54%)	22 (56%)
Heavy	45 (39%)	9(28%)	4 (31%)	15 (39%)
Very Heavy	47 (40%)	2 (6%)	0	0
Extremely Heavy	0	0	0	0

Peak heart rate and energy expenditure of load handling workers have been presented in Table 3. The mean value of peak heart rate was 125.4 ± 13.14 beats/min. It could be interpreted that they were engaged in 'heavy' work (Astrand and Rodahl, 1986), The findings of the present study corroborate with the reported values of Samanta and Chatterjee (1981).

The comparison of workload of load handling workers from subjective assessment and that from peak heart rate during work has been presented in Figure 1. None of the load handling workers expressed their work as 'extremely heavy'; however, workload of 2% workers were in the category of 'extremely heavy' as observed from their peak heart rate. According to subjective assessment, 40% load handling workers were in the 'very heavy' category but peak heart rate showed only 29%.

Table 3

Peak heart rate and energy expenditure during work of the load handling workers (n=90)

Variable	Mean \pm SD	Minimum	Maximum
Peak Heart Rate (b.min ⁻¹)	125.4 ± 13.14	100.0	158.0
Energy Expenditure (kJ. min ⁻¹)	17.673 ± 2.475	12.893	23.818

Work Posture

It was observed that during work the load-handling workers had to bend and twist their neck and back for putting the load on the head. Very often they had to squat depending on the height of the stack from which the sacks are placed on the head. Turning the body with load, the workers moved at a reasonably faster pace towards the place for deposition.

The ancillary workers had to adopt various postures, some of which were awkward, e.g., squatting, stooping, kneeling, etc. Generally, the administrative workers (DMO) performed their jobs in sitting posture. However, those engaged in depot required, several times a day, to move around and also to climb on stack of sacks inside the depot as well as on a truck for verification of stock during loading and unloading jobs.

Pain and Discomfort

The subjects reported pain and discomfort in multiple body parts. The number of workers in different categories reporting pain and discomfort in different body areas has been presented in Table 4. Among all the subjects, the load handling workers reported maximally (62%). When body parts were considered, the load handling workers (42%) and administration workers – DMO (46%) reported their problem in the back. Leg was reported by the ancillary workers (34%) and administrative workers – Depot (31%). Pain in back was reported by a good number of administration workers – DMO. A substantial number (31%) of administration – Depot workers reported pain in the leg compared to those of Administration – DMO (8%).

		Body area reported by subject			
Category of workers	Number of workers reported	Head & Neck	Back	Arms	Legs
Load handling workers	72	23	49	15	25
n = 116	(62)	(20)	(42)	(13)	(22)
Ancillary workers	13	2	8	6	11
n = 32	(41)	(6)	(25)	(19)	(34)
Administration - DMO	6	1	6	4	1
n = 13	(46)	(8)	(46)	(31)	(8)
Administration - Depot $n = 39$	19	1	11	4	12
	(49)	(3)	(28)	(10)	(31)

Table 4

Musculo-skeletal pain and discomfort reported by workers at different body areas

Figures in the parenthesis show the percentage values

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Discussion

With reference to age, the load handling workers were younger than any other category of workers (Table 1). This is due to the fact that those workers are generally recruited at an early age, when they are physically strong so that they can perform the heavy work. With the advancement of age, they work either as gang leaders or as ancillary worker; in either case their job is lighter. There were some differences in height and weight among different categories of subjects; however, it was not reflected much in body mass index, which indicated that most of the subjects of all the categories were normal (>20 kg. m⁻²) from nutritional point of view (WHO 1995).

Most of the load handling workers expressed that their work was in the category of 'heavy' to 'very heavy' (Table 2). While carrying heavy weight, more number of subjects preceived the load as heavier ('very heavy'). However, according to heart rate data, the workloads of a greater number of workers were in the category of 'heavy' (Fig. 1, p-31). When the values of 'heavy' and 'very heavy' categories were considered together, the result of subjective assessment (79%) and that obtained from peak heart rate (84%) were not much different. It has been suggested earlier (Pradhan *et al*, 1994; Lortie *et al*, 1995) that it is essential to take into account the perception of subjects when one attempts to evaluate the difficulty associated with work. It has also been obseved (Pradhan, 1999; Pradhan *et al*, 1994) that subjective perception varies with height of lift and deposition. In the present study, the subjects were carrying loaded sacks after these were put on their head by their coworkers. During deposition, the loaded sacks were either thrown by the movement of their head and/or whole body or these were taken away from their heads by other workers.

Some of the load handling workers (6%) expressed their workload as 'light'. On scrutiny, it was found that these subjects had stopped carrying load for a considerable period of time, as their current job was supervisory in nature. These subjects were not considered for heart rate study.

The ancillary workers used different expressions for workload. Most of them reported their workload as 'light' (25%), 'moderate' (41%) and 'heavy' (28%). Even the term 'very heavy' was also used. This may be due to the fact that that these workers had to carry out different types of jobs. Majority of the administrative workers (DMO and Depot) reported the workload as 'moderate'.

Load handling (lifting, lowering, holding, carrying loads, pushing and pulling weights etc.) requires high muscular effort (Ghosh and Nag, 1986; Yates *et al*, 1980)

in awkward postures, giving rise to musculoskeletal strains and low back signs and symptoms. The force requirements as well as metabolic energy requirements vary with the posture adopted during work (Ayoub and Mital, 1989). Awkward body posture also causes the work to become strenuous and the physiological costs are higher in non-erect postures than in erect postures (Mital *et al*, 1977). The occupational work of handling loads requires high muscular effort (Ayoub and Mc Daniel, 1974; Chaffin, 1974) in awkward postures, giving rise to musculoskeletal strains and low back signs and symptoms According to the classification of work postures by Ovako Work Analysis System (Karhu *et al*, 1977), the postures adopted by the subjects for lifting and carrying 70 to 100 kg of load on their head were very much stressful and these needed immediate consideration for correction (Louhevaara and Suurnakki, 1992). Turning, twisting and bending are also associated with increased incidence of low back disorders like pain, ache and discomfort (Christensen *et al*, 1995).

In the present work set up, the load handling workers bend and twist their neck and back repetitively, the extent of which depends on the height of the stack. Pradhan and Thakur (2002) observed in another group of workers that postures adopted by workers created substantial musculoskeletal stress, causing pain and discomfort. Correction of work posture is possible with the introduction of forklift in the workplace.

The reason for musculoskeletal complain in case of the load handling workers are adoption of awkward posture during lifting and carrying of heavy sack of 50 to 100 kg. repetitively. Although the physical load of the ancillary workers was less, the probable reason of their problem lies with awkward posture and repetitiveness of various types of jobs they performed.

Pain in back as reported by administration-DMO may be due to their prolonged sitting in constrained posture and restricted workspace. On the other hand, repetitive movement of the lower limbs throughout the day, both horizontal and vertical, may be the reason of pain in leg in case of the workers of administration - Depot. Adjustment in work pattern and sharing the job with co-worker may reduce the work stress. The incidence of pain reported in arm (31%) by administration - DMO cannot be explained by the nature of their work. However, this may be due to less number of subjects involved in the study.

In conclusion, it may be stated that the workload of the load handling workers was in the heavy to very heavy category. This has been refelected in both subjective and objective assessments of the workers; it indicated that both the methods were equally reliable. The stress of the different categories of workers had been reflected

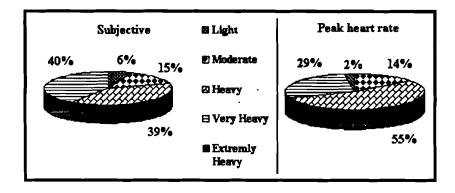


Figure 1

Comparison of workload of the load handling workers assessed subjectively and from peak heart rate

in their subjective preception as a result of integration of physical workload and postural stress. The problems – work stress and musculoskeletal – were related to occupation and not to age (Pradhan and Thakur, 2002). In the present study, objective assessment by measuring the working heart rate was carried out only with the load handling workers. Further study with other categories of workers is necessary. Furthermore, comparatively higher incidence of reported pain and discomfort of the workers in administration-DMO was rather unexpected and the reason remaind unclear. Studies involving more number of workers in that category are needed for clarification.

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References

- Asfour S. S., Ayoub M. M., Mital A and Bethea N. J. (1983). Perceived exertion of physical effort for various manual handling tasks. Am. Ind. Hyg. Assoc. J; 44 : 223 - 228.
- Astrand P. O. and Rodahl K. (1986). *Textbook of Work Physiology*. 3rd edn. New York : Mc-Graw Hill Book Company.
- Ayoub M. M. and Mc Daniel J. W. (1974). Effects of operator stance on pushing and pulling tasks. *AIIE Transactions*, 6: 185 195.
- Ayoub M. M. and Mital A. (1989). Manual Material Handling. London : Taylor & Francis.
- Banerjee S. and Sen R. N. (1955). Determination of the surface area of the body of Indians. J. Appl. Physiol. :7.585 - 588.
- Chaffin D. B. (1974) Human strength capability and low back pain. J. Occup. Med.;16 : 248 - 254.
- Christensen H., Pedersen M. B. and Sjgaard G. (1995) A national cross-sectional study in the Danish wood and furniture industry on working postures and manual material handling. *Ergounomics*; 38 : 793 - 805.
- Corlett E. N. and Bishop R. P. (1976) A technique for assessing postural discomfort. Ergonomics ; 19 : 175 - 182.
- Datta S. R. and Ramanathan N. L. (1969). Energy expenditure in work predicted from heart rate and pulmonary ventilation. J. Appl. Physiol., 25: 297-302.
- Ghosh S. N. and Nag P. K. (1986). Muscular strains in different modes of load handling. *Clin. Biomechanics.*, 1:64 70.
- Karhu O., Kansi P. and Kourinka I (1977). Correcting working postures industry : a practical method for analysis. *Applied Ergonomics*, 8 : 199 - 201.
- Lortie M., Pracdhan C. K. and Kuorinka I (1995). Analyse de la perception associée à une tâche de manutention. Proceedings of the Second International Scientific Conference on Prevention of Work Related Musculoskeletal Disorders (Premus 95), (Montreal : Institutde recherche en santé et en sécurité du travail du Québec), 351 - 353.

- Louhevaara V. and Suurnakki T. (1992). OWAS : A method for the evaluation of postural load during work. Helsinki : Finnish Institute for Occupational Health.
- Mital A, Nicholson AS and Ayoub MM (1977) A Guide to Manual Materials Handling. 2nd Edition, London : Taylor & Francis.
- Pradhan C. K., Lortie M. and Kuorinka I. (1994). Study on subjective perception in manual material handling. *Project Report*, Montreal : Université du Québec à Montréal.
- Pradhan C. K. and Thakur S. (2002). Subjective assessment of work stress in the automobile transport sector. *Ergonomics* SA; 14, 2 7.
- Pradhan C. K. (1999). Subjective perception in load handling tasks. Ind. J. Occ. Environ. Med., 3: 74 - 76.
- Samanta A. and Chatterjee B. B. (1981) Energy expenditure in manual load carriage. Ind Health; 19 : 145 - 153.
- Sinclair M. A. (1995). Subjective perception. In *Evaluation of Human Work*, (Eds.) J. R. Wilson and E. N. Corlett. London : Taylor & Francis, pp. 69 - 100.
- Waikar A., Lee K., Aghazadeh F. and Parks C. (1991). Evaluating tasks using subjective and biomechanical estimates of stress at the lower back. *Ergonomics*, 34: 33 - 47.
- WHO (1995). Physical status : the use and interpretation of anthropometry. WHO Technical Report Series No. 854. Geneva : World Health Organisation.
- Yates J. W., Kamon E., Redgers S. H. and Champney H. (1980) Static lifting strength and maximal isometric voluntary contractions of back, arm and shoulder muscles. *Ergonomics*, 23: 37 - 48.

ANTHROPOMETRIC STUDIES ON THE KOIRENG TRIBE OF MANIPUR

Md. Latif Shah

P. G. Department of Anthropology D. M. College of Science Imphal - 795 001, Manipur

ABSTRACT

It is generally believed that somatometric character is one of the main istruments for determination of morphological variation in a population. In the present study, 13 somatometric measurements and its 9 indices have been applied among the Koireng tribe of Manipur to see the trend of their morphological variation. In this regard, the Koireng males show higher mean values than the females for most of the somatometric characters and indices. The Koireng show the closest affinity with the Chiru tribe and furthest distance with the Tarao tribe of Manipur in most of the characters when a comparison is made between the Koireng and each of 5 population groups of Manipur in this study. This trend of variation might be due to the biological distance prompted by hybridization and isolation among the populations under different ecological conditions.

Introduction

Scientific studies on biological variation among different populations of Manipur have started since 1960s. However, several populations of Manipur are yet to be covered by such studies. The Koireng tribe represent one such population group, about whom information is lacking. Few works available for this tribe are mostly on their ethno-historical profile (Kabui, 1987), although Shah and Singh (1996) have covered some aspects of their biology.

The Koireng are one of the 29 Scheduled tribes of Manipur. The term Koireng is a distorted Meitei version of the word "Kolhen" (Shakespear, 1912) which was probably derived from the word "Kolren" (Grierson, 1908) meaning 'the poeple of the East'. They also believe in a cave origin like other tribes of Manipur as they call themselves "Khurmi" meaning 'Man of Cave'. At present, they are scattered in 9 villages in differenct districts of Manipur at varying distances from Imphal the capital of Manipur. The majority of them are confined in Senapati district of the state. The foothills are their preferred place of habitation. In 1981 census, the total population of the Koireng was 949 with 47.21 per cent males and 52.79 per cent females. The Koirengs are patrilineal in descent and patrilocal in residence. They follow exogamy. But they prefer cross-cousin marriage by tradition. The dialect of Koireng is classified as one of the Kuki languages (Grierson, 1908). The present

^{*} Corresponding Author

study aims at examining the magnitude and trend of morphological variation in the study of somatometric measurements and indices among the Koireng tribe of Manipur. A comparison of the Koireng data with the other available data of 5 populations of Manipur, viz. the Chiru, Kabui, Meitei, Muslim and Tarao has also been attempted.

Materials and Method

A total of 13 somatometric measurements (Table 1) have been made on 106 Koireng individuals representing 57 males and 49 females, belonging to 20 to 49 years of age, from Longa-Koireng and Koireng Colony villages in 1995 following the techniques of Martin and Saller (1956). Longa Koireng is a village which lies on the eastern spurs of Yonga hill in Senapati district, at a distance of about 17 Km. from Imphal — the Capital of Manipur. But, the Koireng Colony is a new settlement at Lamphelpat, Imphal — West district, at a distance of about 3 km. from the heart of Imphal city. Most of the inhabitants of the Koireng Colony are the recent migrants from Longa Koireng. Care was taken not to include closely related and physically handicapped individuals.

A total of 9 indices (Table 3) have also been calculated from the 13 somatometric characters. Various statistical constants have been calculated for somatometric characters and indices (Table 2 and 4). Moreover, t-test has also been applied for the comparison of the morphological characters between the Koireng data and available data of 5 other populations of Manipur to see their relative proximity (Table 6 and 8).

Measurements	Abbrev-	Male	Female
	iations	Mean \pm S. E.	Mean \pm S. E.
Height vertex	HV	161.71 ± 0.86	148.78 ± 0.64
Sittinng height vertex	SHV	85.85 ± 0.52	78.88 ± 0.36
Maximum head length	MHL	19.08 ± 0.12	18.08 ± 0.09
Maximum head breadth	MHB	14.68 ± 0.07	13.73 ± 0.25
Head height	HH	12.84 ± 0.16	12.12 ± 0.14
Least frontal breadth	LFB	10.84 ± 0.07	10.39 ± 0.08
Breadth of bizygomatic arch	BBZA	13.91 ± 0.06	13.06 ± 0.09
Bigonial breadth	BGB	10.76 ± 0.07	10.38 ± 0.07
Total facial height	TFH	11.54 ± 0.09	10.62 ± 0.10
Upper facial height	UFH	6.57 ± 0.04	5.90 ± 0.07
Nasal height	NH	5.01 ± 0.04	4.54 ± 0.05
Nasal breadth	NB	3.82 ± 0.03	3.46 ± 0.03
Body weight	Bwt	57.19 ± 1.06	44.26 ± 0.74

Table – 1

Sex differences of somatometric measurements among the Koireng tribe of Manipur.

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Table - 2

	Ma	le	Fen	nale
Measurements	S. D. ± S. E.	$\overline{\mathbf{C}}$. $\overline{\mathbf{V}}$. \pm S. E.	S. D. ± S. E.	$\mathbf{C.} \ \mathbf{V} \pm \mathbf{S.} \ \mathbf{E}.$
HV	6.61 ± 0.61	4.09 ± 0.38	4.49 ± 0.45	3.02 ± 0.03
SHV	3.96 ± 0.37	4.61 ± 0.04	2.52 ± 0.25	3.19 ± 0.03
MHL	0.87 ± 0.08	4.56 ± 0.04	0.61 ± 0.06	3.37 ± 0.03
MHB	0.54 ± 0.05	3.68 ± 0.03	1.76 ± 0.18	2.77 ± 0.13
HH	1.22 ± 0.11	9.50 ± 0.08	0.96 ± 0.10	7.92 ± 0.08
LFB	0.56 ± 0.05	5.17 ± 0.05	0.57 ± 0.60	5.49 ± 0.06
BBZA	0.49 ± 0.05	3.52 ± 0.03	0.66 ± 0.07	5.05 ± 0.05
BGB	0.52 ± 0.05	4.83 ± 0.04	0.47 ± 0.05	4.53 ± 0.05
TFH	0.66 ± 0.06	5.72 ± 0.05	0.69 ± 0.07	6.50 ± 0.07
UFH	037 ± 0.03	5.63 ± 0.05	0.46 ± 0.05	7.80 ± 0.08
NH	0.31 ± 0.03	6.19 ± 0.05	0.36 ± 0.04	7.93 ± 0.08
NB	0.23 ± 0.02	6.02 ± 0.05	0.24 ± 0.02	6.94 ± 0.07
Bwt	8.02 ± 0.75	14.02 ± 0.12	5.21 ± 0.53	11.77 ± 0.12

Distribution of standard deviation, co-efficient of variation and their respective standard errors for somatometric measurements of the Koireng.

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Table - 3

Sex differences of somatometric indices among the Koireng tribe of Manipur.

Indice	Abbrev-	Male	Female
	iations	Mean \pm S. E.	Mean ± S. E.
Sittinng height vertex index	SHVI	53.09 ± 0.17	52.27 ± 0.76
Lenghth-breadth index	LBI	77.02 ± 0.51	77.70 ± 0.54
Lenghth-height index	LHI	67.34 ± 0.90	67.11 ± 0.91
Breadth-height index	BHI	87.45 ± 1.09	86.10 ± 1.00
Jugo-frontal index	JFI	75.38 ± 0.50	79.72 ± 0.78
Jugo-Mandibular index	JMI	77.40 ± 0.40	79.58 ± 0.70
Total facial index	TFI	83.00 ± 0.56	81.78 ± 0.91
Upper facial index	UFI	47.34 ± 0.41	45.27 ± 0.55
Nasal index	NI	76.38 ± 0.92	76.55 ± 1.23

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Results and Discussion

The mean value of 13 somatometric characters among the Koireng individuals of Manipur show that the male members have larger values than the femals members in all the characters (Table 1) and also in the distribution of the values of Sd, CV and their respective standard errors (Table 2). Moreover, most of the Koireng individuals in both sexes posses 'Short' stature, 'Elongated' head with 'Medium' type of head breadth, least frontal breadth and breadth of bizygomatic arch, 'Low' type of total facial height, 'Below-medium' type of nasal height and 'Above-medium' type of nasal breadth.

For the present study, as many as 9 indices have been calculated from 13 somatometric characters among the Koireng to see the trend of variation and magnitude in between the male and female members (Table 3). The mean values and standard errors of these indices indicate that the male members have larger value than the female members in 5 indices (SHIV, LHI, BHI, TFI, UFI) but the female members also show larger values than the male members in 4 indices (LBI, JFI, JMI, NI) in general (Table 3). The same trend of variation is also observed among them in the S. D., C. V. and their respective standard errors in most of the indices (Table 4).

Table – 4

	Ma	le	Female		
Indices	\overline{S} . D. $\pm \overline{S}$. E.	$\mathbf{C}. \ \mathbf{\overline{V}}. \pm \mathbf{S}. \ \mathbf{\overline{E}}.$	\overline{S} . D. $\pm S$. E.	$\mathbf{C}.\ \mathbf{V}. \pm \mathbf{S}.\ \mathbf{E}.$	
SHVI	1.28 ± 0.12	2.41 ± 0.02	5.30 ± 0.54	10.14 ± 0.14	
LBI	3.32 ± 0.36	4.96 ± 0.04	3.78 ± 0.38	4.36 ± 0.05	
LHI	6.83 ± 0.64	10.14 ± 0.09	6.40 ± 0.65	9.54 ± 0.10	
BHI	8.24 ± 0.77	9.42 ± 0.08	6.99 ± 0.71	8.12 ± 0.08	
JFI	3.74 ± 0.35	4.96 ± 0.04	5.44 ± 0.55	6.32 ± 0.07	
JMI	3.04 ± 0.28	3.93 ± 0.03	4.38 ± 0.49	6.13 ± 0.06	
TFI	4.24 ± 0.40	5.11 ± 0.04	6.34 ± 0.64	7.75 ± 0.08	
UFI	3.11 ± 0.29	6.57 ± 0.06	3.82 ± 0.39	8.44 ± 0.09	
NI	6.92 ± 0.65	9.06 ± 0.08	8.58 ± 0.37	11.21 ± 0.11	

Standard deviation, co-efficient of variation and their respective standard errors of indices for the somatometric characters among the Koireng tribe of Manipur. Moreover, most of the Koireng individuals of both sexes belong to the 'Dolichicephalic' and 'Mesocephalic' head in the LBI, 'Hypsicephalic' and 'Acrocephalic' type, of head in the LHI and BHI, respectively. As such, the Koireng individuals may be classified as 'Narrow' and 'Medium' in JFI, 'Medium' and 'Broad' in JMI, 'Euryprosopic' and 'Mesoprosopic' in TFI, 'Euryne' and 'Mesene' in UFI, and 'Mesorrhine' in NI.

A comparative study is made on the mean values and its standard errors of 12 somatometric characters between the Koirengs and each of 5 other populations of Manipur (Table 5). In this comparison, the Koirengs show colsest affinity with the Chiru in 5 traits (HV, BGB, UFH, NB, Bwt) and furthest distance with the Tarao in 5 (SHV, LFB, BBZA, NH, Bwt) (Table 5). But, the Koirengs show relative proximity with the Meitei in 3 (BBZA, TFH, NH), Kabui (LFB) and Muslim (SHV) in 1 each of the characters in the comparison (Table 5, p-39).

In the t-test comparison between the Koireng and each of 5 other populations of Manipur on somatometric characters, the largest number of characters (HV, SHV, MHB, LFB, BBZA, BGB, UFH, NH, Bwt = 9) are observed as significant differences between the Koireng and Tarao at 5 per cent level of probability (Table 6). However, on the least number of characters (MHB, BBZA, NH=3) significant differences between the Koireng and Chiru populations have been observed in this comparison (Table 6). Moreover, the significant differences are also seen in between the Koireng and Meitei (HV, SHV, MHB, BGB), Kabui (HV, SHV, BBZA, UFH, NB), Muslim (HV, MHL, MHB, LFB, BBZA, BGB, UFH, NB) in t-test comparison (Table 6, p-40). The Koirengs therefore, show the greatest distance and closest affinity with the Tarao and Chiru respectively in all the comparisons made on somatometric characters in the present study.

When a comparison is made on the mean values and its standard errors of 3 indices for 12 somatometric characters in between the Koireng and each of 5 other populations of Manipur, the Koireng data show the closest affinity with the Kabui in 1 (LBI), the Meitei in 2 (TFI, NI) indices (Table 7). However, the greatest distance is observed in between the Koireng and the Muslim in 3 (LBI, TFI, NI) indices (Table 7, p-41).

Moreover, in the t-test comparison between the Koireng and each of 5 other populations of Manipur on 3 indices, the significant differences are found only in respect of 1 (LBI) and 2 (TFI, NI) indices between the Koireng and Chiru, and between Koireng and Meitei respectively (Table 8). But the greatest distance is observed between the Koireng and Muslim in 3 (LBI, TFI, NI) indices in this study

Table	-	5
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Mean values and its standard errors of somatometric characters for comparison between the Koireng and each of 5 other populations of Manipur.

Measurements	Chiru (127) (Singh and Shah, 1997)	Kabui (50) (Singh, 1986)	Muslim (300) (Shah, 1990)	Meitei (50) (Singh, 1986)	Tarao (152) (Devi, 1994)	Koireng (106) (Present Study)
HV	154.14 ± 0.62	159.30 ± 0.54	162.53 ± 0.57	163.30 ± 0.43	151.27 ± 0.86	155.25 ± 0.75
SHV	81.37 ± 0.43	[•] 85.30 ± 0.34	83.28 ± 0.35	85.56 ± 0.22	78.76 ± 0.44	82.37 ± 0.44
MHL	18.46 ± 0.06	18.74 ± 0.06	18.00 ± 0.07	18.46 ± 0.06	18.60 ± 0.26	18.58 ± 0.11
МНВ	14.14 ± 0.07	14.44 ± 0.03	14.67 ± 0.05	14.92 ± 0.05	14.29 ± 0.24	14.23 ± 0.16
LFB	10.41 ± 0.35	10.46 ± 0.03	10.23 ± 0.04	$10.50\pm\!0.03$	11.11 ± 0.16	10.44 ± 0.08
BBZA	13.58 ± 0.06	13.16 ± 0.07	13.19 ± 0.06	13.50 ± 0.08	12.84 ± 0.15	13.49 ± 0.08
BGB	10.53 ± 0.06	9.98 ± 0.03	10.29 ± 0.05	10.25 ± 0.06	10.15 ± 0.14	10.57 ± 0.07
TFH	10.97 ± 0.07	10.00 ± 0.03	11.13 ± 0.05	11.12 ± 0.03	10.98 ± 0.11	11.08 ± 0.10
UFH	6.17 ± 0.05	6.46 ± 0.03	6.66 ± 0.05	6.35 ± 0.03	6.15 ± 0.11	6.24 ± 0.06
NH	4.63 ± 0.04	4.80 ± 0.02	4.68 ± 0.03	4.78 ± 0.02	4.58 ± 0.08	4.78 ± 0.05
NB	3.57 ± 0.04	3.88 ± 0.02	3.32 ± 0.03	3.63 ± 0.02	3.45 ± 0.13	3.64 ± 0.03
Bwt	49.27 ± 0.76		52.45 ± 0.63		47.16 ± 0.90	50.73 ± 0.90

Table – 6

Measurements	Koireng X Chiru	Koireng X Kabui	Koireng X Tarao	Koireng X Meitei	Koireng X Muslim
HV	1.14	4.42 *	3.49 *	9.49 *	7.14 *
SHV	1.61	5.23 *	5.82 *	6.51 *	1.63
MHL	0.92	1.23	0.71	0.92	4.46 *
MHB	5.29 *	1.31	2.07 *	4.06 *	2.59 *
LFB	0.08	0.22	3.72 *	0.67	2.33 *
BBZA	9.00 *	3.00 *	3.82 *	0.13	3.00 *
BGB	0.44	0.76	2.63 *	3.56 *	3.11 *
TFH	0.92	1.80	0.67	0.40	0.45
UFH	0.88	3.14 *	6.92 *	1.57	5.25 *
NH	2.50 *	0.40	2.22 *	0.29	1.67
NB	1.40	6.00 *	1.46	0.25	8.00 *
Bwt	1.24		2.81 *		1.56

t-test values of somatometric characters used for comparison between the Koireng and each of 5 other populations of Manipur.

Key : * indicates statistically significant at 5% level of probability.

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Table -	-7	
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Mean values and its standard errors of Indices for somatometric characters in the comparison between the Koireng and 5 other populations of Manipur.

Indice	Chiru (127) (Singh and Shah, 1997)	Kabui (50) (Singh, 1986)	Meitei (50) (Singh, 1986)	Muslim (50) (Shah, 1990)	Tarao (152) (Devi, 1994)	Koireng (106) (Present Study)
LBI	76.66 ± 0.63	77.03 ± 0.23	81.19 ± 0.23	81.46 ± 0.36	76.19 ± 0.36	77.19 ± 0.53
TFI	80.89 ± 0.58	81.70 ± 0.53	81.90 ± 0.26	84.38 ± 0.49	83.16 ± 0.83	82.39 ± 0.74
NI	77.06 ± 0.88	72.80 ± 0.31	76.30 ± 0.53	71.31 ± 0.73	76.77 ± 0.98	76.47 ± 1.08

(Table 8). Thus, the Koirengs from all aspects show the closest affinity with the Chiru tribe of Manipur in the comparison made here.

Table – 8

t-test values of Indices for somatometric characters in comparison between the Koireng and 5 other populations of Manipur.

Indica	Koireng X Chiru	Koireng X Kabui	Koireng X Tarao	Koireng X Meitei	Koireng X Muslim
LBI	0.85	1.57	1.09	6.71 *	6.41 *
TFI	1.60	0.76	0.69	0.63	2.24 *
NI	0.42	3.28 *	0.21	0.24	3.97 *

Key : * indicates statistically significant at 5% level of probability.

Conclusion

Geographically, Manipur may be considered as having two main parts : (i) The valley, predominantly occupied by the Meiteis, the largest community in the region with a small band of Muslims and a few pockets of tribal people, and (ii) The hills which are completely occupied by the different tribal groups. The Koireng. Chiru, Kabui and Tarao are also tribes of Manipur who have mostly settled on the foothills or spurs or slopes of hills in different districts of Manipur.

In the present study, sexual variation is observed among the Koireng individuals while a discussion is made on the mean values of 13 somatometric characters (Table 1) and 9 indicas (Table 3). When a comparison is made on the mean values (Table 5) and its indices (Table 7) as well as t-test (Table 6 and 8) of the metric characters in each pair between the Koireng and one each of 5 other populations of Manipur, the Koireng show the closest affinity with Chiru and furthest distance from the Tarao in this study. With regard to stature, the Koireng, Chiru, Tarao and Kabui come under 'Short' category while the other two, namely, the Meitei and Muslim have 'Lower-medium' stature in the present study. Different trends of variation between the Koireng and 5 other populations of Manipur may be due to the geographical isolation as well as natural selection, resulting in inbreeding within the populations under consideration. Its social implications need also to be taken into consideration.

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References

- Devi, Kh. S. 1994. Anthropometric characters of Tarao. Jr. Anthro. Soc. Manipur, Vol. 1: 39-54.
- Grierson, G. A. 1908. Linguistic Survey of India. Vol. III, Part II and III, Metilal Banardass, Delhi.
- Kabui, G. 1997. The Koireng of Manipur. Koireng Historical Research Committee, Imphal.
- Shah, M. L. 1990. Bio-anthropology of the Muslims of Manipur. Unpub. Ph.D. Thesis, M. U. Canchipur.
- Shah, M. L. and R. K. R. Singh 1996. The Koireng tribe of Manipur : A Bioanthropological Study. A Project report, Directorate for Develop-ment of Tribals and Backward Classes, Govt. of Manipur.
- Shakespear, J. 1912. The Lushai Kuki Clan. Central Publishing House, Delhi.
- Singh, I. P. and M. K. Bhasin (Reprint) 1989. Anthropometry. Kamal-Raj enterprises, Delhi.
- Singh, L. R. 1986 Anthropometric Variation between the Meiteis and the Plain Kabuis of Manipur. Bull. Deptt. Anthro. Dhbr. U. Dibrugarh, Vol. 15: 28-37.
- Singh, R. K. R. and M. L. Shah 1997. *The Chiru Tribe of Manipur : A Bio-Anghropological Study.* Project report, Directorate for Development of Tribals and Backward Classes, Govt. of Manipur.

ALLELOPATHIC INFLUENCE OF *IPOMOEA PES-CAPRAE* (L.) ROXB. ON *PHASEOLUS MUNGO* ROXB.

R. K. Bhakat, A. Bhattacharjee, U. K. Kanp and R. K. Das

Department of Botany and Forestry, Vidyasagar University, Midnapore - 721 102, West Bengal, India

ABSTRACT

Ipomoea pes-caprae plant was analysed to evaluate the existence of allelopathic effect using fully viable seeds of black gram as the bioassay material. Fresh leaf extract of Ipomoea strongly reduced the percentage germination as well as remarkably extended the time required for 50% germination of seeds. Levels of protein and insoluble carbohydrates as well as activities of catalase and dehydrogenase enzymes were significantly reduced and soluble carbohydrate level was increased in the seed samples pretreated with Ipomoea leaf extracts of two concentration grades in the proportion of 1:5 and 1:7.5 of Ipomoea. Existence of allelopathic effect of Ipomoea was confirmed from the reliable physiological and biochemical data of the present investigation.

KEY WORDS : Allelopathy, Ipomoea, Seed germination, T_{so}, Protein, Carbohydrates, Enzymes.

Introduction

The term allelopathy refers to biochemical interactions between all plant types inclusive of microorganisms. It is different from competition and implies that the effect depends on a chemical constituent escaping into the environment. Since 1960, there has been a spurt in publications dealing with this phenomenon (Garb, 1961; Muller *et al.*, 1964; Tinin and Muller, 1971; Bandyopadhyay, 1983; Nayek, 2000; Bhattacharjee *et al.*, 2001 and 2003). According to Evenari (1961), a pioneer worker in this field of research, allelopathy is the effect of one plant upon another occurring under natural conditions and exerted by chemical means other than nutritional ones. Allelopathy is also an expression of the ecological phenomena which are normal constituents of the environment of the terrestrial plants (Datta and Sinha Roy, 1974).

Considerable evidences have been adduced during the last five decades demonstrating the presence of inhibitory compounds in a wide variety of plant extracts and volatiles (Eberhardt, 1954; Datta and Sinha Roy, 1974; Ghosh and Datta, 1989; Inderjit *et al.*, 1995).

There are some common indices for assessing allelopathic action of plants or plant parts. These include seed germination behaviour (percentage and T_{50} of seed

germination). In the present investigation, an attempt was made to evaluate the allelopathic potential of the coastal species $Ipomoea \ pes-caprae$ (L.) Roxb. using black gram (*Phaseolus mungo* Roxb.) as suitable bioassay material. For precise evaluation of the allelopathic action some selective physiological and biochemical parameters were analysed from the black gram seeds pretreated with the leaf extract of *I. pes-caprae*.

Materials and Methods

Fresh mature and healthy leaves (100g) of *Ipomoea pes-caprae* (L.) Roxb. (Fam. Convolvulaceae), collected from Digha coast, Purba Medinipur were thoroughly homogenized using 100 ml distilled water. The homogenate was strained using a fine cloth and then centrifuged at 5000g for 15 minutes. The supernatant was then made upto 250 ml using distilled water and this was considered as stock solution of leaf extract. From this stock solution two concentration grades in the proportion of 1:5 and 1:7.5 (w/v) were prepared using distilled water. And this was taken as the two different concentrations of leaf extract solution of the plant species.

Fully viable black gram seeds in three lots of 25g each surface sterilized with 0.1% HgCl₂ solution for 90 seconds. The seed lots were then separately presoaked in the leaf extracts or in distilled water for 4 hours and then thoroughly sundried till the original moisture level (6.8%) was achieved. Thus allowed the seeds for various biochemical as well as physiological tests. Data on seed germination behaviour (percentage and T₅₀ value), protein, soluble and insoluble carbohydrate levels, activities of catalase and dehydrogenase enzymes in seeds were recorded.

To analyse percentage germination and T_{50} value from pretreated seeds, three groups of 100 seeds *i.e.* 300 seeds of each treatment were transferred to separate Petri dishes containing filter paper moistened with 10 ml distilled water. Germination data were recorded after 72 hours of seed soaking following the International Rules for Seed Testing (ISTA, 1976).

Protein level was analysed from the seed kernels following the method of Lowry $et \ al.$ (1951). Soluble and insoluble carbohydrate levels were analysed from seed kernels following the method of McCready $et \ al.$ (1950).

Extraction and estimation of the enzyme catalase was done as per the method described by Snell and Snell (1971) modified by Biswas and Choudhuri (1978). For the assay of this enzyme the blank was taken as Zero time control. The activity of this enzyme was expressed as $[(\Delta A \times Tv) / (t \times v)]$, where ΔA is the absorbance of the sample after incubation minus the absorbance of the zero time control, Tv is the total volume of the filtrate, t is the time (minutes) of incubation with the substrate and v is the volume of the filtrate taken for incubation (Fick and Qualset, 1975). For analysing dehydrogenase activity the TTC-stained (formazan formation)

embryonal axes of the seeds of each treatment were extracted with 10 ml of 2- methoxyethanol and O.D. values of the solutions were recorded at 580 nm. This method was adopted after Rudrapal and Basu (1979) with slight modification.

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Statistical analysis of the data was done in terms of least significant different (LSD) which was calculated at 95% confidence limits (Panse and Sukhatme, 1967).

Results

Effect on germination and T_{50} value (Table 1). Data showed that in control sample percentage germination of black gram seed was higher than *Ipomoea* leaf extract pretreated seed. Two concentration grades (1:5 and 1:7.5) of pretreating leaf extracts, regardless of concentration, efficiently enhanced the fall of germination. On the other hand, in treated seeds, time (h) required for 50% germination was noted significantly high than control.

Effect on changes of protein, insoluble carbohydrate and soluble carbohydrate levels in seed kernels (Table 2). Data showed that seed pretreatment with leaf extracts triggered the reduction of protein and insoluble carbohydrate levels in comparison to control sample. Whereas internal soluble carbohydrate level was increased in seed samples irrespective of the treatments.

Effect on changes of catalase and dehydrogenase activities in seed kernels (Table 3). Data showed that activities of the enzymes catalase and dehydrogenase declined in seed kernels which underwent pretreated with the leaf extracts of *Ipomoea*. However, the rate of decreasing the enzyme activities was high in seeds which received pretreatment with more concentrated leaf extract (1:5).

Table 1. Effect of seed pretreatment with leaf extract (two concentration grades in the proportion of 1 : 5 and 1: 7.5) of *Ipomoea* on germination percentage and T_{50} (hours) of black gram seeds.

Treatments	Germination(%)	T ₅₀ (hours)
Control	100.00	13.98
<i>Ipomoea</i> (1:5)	76.03	3 0.73
<i>Ipomoea</i> (1 : 7.5)	80.34	26.28
LSD ($P = 0.05$)	7.21	1.33

Table 2. Effect of seed pretreatment with leaf extract (two concentration grades in the proportion of 1:5 and 1:7.5) of *Ipomoea* on protein, insoluble and soluble carbohydrate levels in black gram seeds.

Treatments	Protein (mg/g frest wt.)	Insol. carbohydrate (mg/g fresh wt.)	Sol. carbohydrate (mg/g fresh wt.)	
Control	218.21	90.76	32.97	
Ipomoea (1:5)	156.25	41.22	68.73	
<i>Ipomoea</i> (1 : 7.	5) 175.33	57.71	59.22	
LSD ($P = 0.05$)	15.39	4.13	3.29	

Table 3. Effect of seed pretreatment with leaf extract (two concentration grades in the proportion of 1 : 5 and 1: 7.5) of *Ipomoea* on catalase and dehydrogenase activities in kernels of black gram seeds.

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Treatments	Catalase [(∆AxTv)/(txv)]	Dehydrogenase (OD/g wet wt.)
Control	122.26	1.56
<i>Ipomoea</i> (1:5)	76.43	0.87
<i>Ipomoea</i> (1 : 7.5)	83.00	0.92
LSD ($P = 0.05$)	8.22	0.78

Discussion

The present investigation showed that pretreatment of black gram seeds with leaf extract of two concentration grades in the proportion of 1:5 and 1:7.5 of *Ipomoea* reduced percentage germination and increased T_{50} value (Table 1), decreased protein and insoluble carbohydrate and increased soluble carbohydrate level (Table 2) and decreased the activities of catalase and dehydrogenase enzymes (Table 3).

Analysis of germination behaviour is considered as a reliable index for evaluation of allelopathic action (Ghosh, 1979; Datta and Chakraborty, 1982; Navek, 2000; Nayek et al., 2002; Bhattacharjee et al., 2003). Reduced seed germinability and slower rate of germination are the two important effects of allelopathic action of plants and such action is chiefly exerted by a number of inhibitors of diverse chemical nature (Bandyopadhyay, 1983; Ghosh and Datta, 1989). In this investigation the leaf extract-induced inhibition of percentage seed germination and increased T_{50} value are clear indicative of the allelopathic action of the test material. High allelopathic potential of Ipomoea was recorded from its stronger germination inhibitory capacity. However, high concentration leaf extract (1:5)was found to be more injurious than diluted leaf extract (1: 7.5). Because highly concentrated plant extract have more inhibitory compounds which easily penetrate into the seed membrane thus resulting quicker deterioration of seeds. Allelopathic potential of *Ipomoea* can further be corroborated from the present data on the leaf extract-induced reduction of protein and insoluble carbohydrate levels as well as activities of catalase and dehydrogenase enzymes. The soluble carbohydrate level was increased in the plant extract-pretreated seeds which might be an indication of degradation of stored carbohydrates by allelochemicals. Various allelochemicals present in plants having allelopathic property reduce the overall metabolism of plant or plant parts and particularly anabolic activities are reported to be strongly impaired (Bandyopadhyay, 1983; Nayek, 2000; Bhattacharjee et al., 2003). Results, therefore, point out that the leaf extract of Ipomoea possesses some chemicals which efficiently render allelopathic action on the bioassay material of this investigation.

References

- Bandyopadhyay, A. (1983). Germination and growth inhibitors in three weeds of West Bengal. Ph. D. Thesis, University of Calcutta, Calcutta.
- Bhattacharjee, A., Bhakat, R. K. and Nayek, A. (2001). Physiological studies on allelopathic potential of *Eucalyptus* and *Parthenium*. Journal of Biological Sciences, V.U., 7, 28-37.
- Bhattacharjee, A., Bhakat, R. K., Kanp, U. K. and Das, R. K. (2003). An investigation on allelopathic action of *Casuarina equisetifolia* J.R. and *Ipomoea pes-caprae* (L.) Roxb. *Environment and Ecology*, **21** (2), 283-289.
- Biswas, A.K. and Choudhuri, M.A. (1978). Differential behaviour of the flag leaf of intact rice plant during ageing. *Biochemic und Physiologie der Pflanzen*, **173**, 220-228.
- Coolbear, P., Francis, A. and Grierson, D. (1984). The effect of low temperature presowing treatment on the germination peformance and membrane integrity of artificially aged tomato seeds. *Journal of Experimental Botany*, 35, 1609-1607.
- Datta, S. C. and Chakraborty, S.D. (1982). Allelopathic potential of *Clerodendron* viscosum Vent. in relation to germination and seeding growth of weeds. Flora, 172, 85-91.
- Datta, S.C. And Singh-Roy, S.P. (1974). Allelopathy and inhibitors. Science and Culture, 40, 47-59
- Eberhardt, F. (1954). Ausscheidung einer organischen verbendung aus den Wurzeln des Hafers (Avena sativa). *Naturwiss*, **41**, 259-260.
- Evenari, M. (1961). Chemical influnce of other plants (Allelopathy). In : Handbuch der Pflanzenphysiologie (Ruhlnd, W, ed.) 16, 691-736. Springer Verlag, Berlin-Gottingen-Heidelburg.
- Fick, N.G. and Qualset, C.O. (1975), Genetic control of endosperm amylase activity and gibberelin responses in standard height and short satured wheat. *Proceedings of National Academy of Science*, USA., **72**, 892-895.
- Garb, S. (1961). Differential growth inhibitors produced by plants. *Botanical Review*, **27**, 422-423.

- Ghosh, K.N. (1979). Allelopathic potential of two species of Chenopodium Linn. Ph. D. Thesis. University of Calcutta, Calcutta.
- Ghosh, K. N. and Datta, S.C. (1989). A glimpse into the phenomenon of allelopathy. Bulletin of Botanical Society of Bengal, 43, 13-25
- Inderjit, Dakshini, K. M.M. and Einhelling, F.A. (Ed.) (1995). Allelopathy Organisms, Processes and Applications. American Chemical Society, Washinton, D.C.
- International Seed Testing Association (1976). International Rules for Seed Testing. Seed Science and Technology, 4, 51-177.
- Lowry, O. H., Rosebrough, N. J., Farr, A.L. and Randall, R. J. (1951). Protein measurement with Folinphenol reagent. Journal of Biological Chemistry, 193, 263-275.
- McCready, R.M., Guggloz, J., Silvirera, V. and Owenns, H.S. (1950). Determination of starch and amylase in vegetables. *Analytical Chemistry*, **22**, 1156-1158.
- Muller, C.H., Muller, W.H. and Haines, B.L. (1964). Volatile growth inhibitors produced by aromatic shrubs. *Science*, 143, 471-473
- Nayek, A. (2000). Studies on allelopathic potential of Eucalyptus and Parthenium. M.Sc. Dissertation. Vidyasagar University, India.
- Nayek, A., Kanp, U. K., Das, R. K., Bhakat, R. K. and Bhattacharjee, A. (2002). A comparative physiological study on allelopathy of *Eucalyptus* and *Parthenium. National Seminar on Environmental Biology and Fish Biology*, Feb 1-3, 2002. Visva Bharati, Santiniketan, India, p-25.
- Panse, V.G. and Sukhatme, P. T. (1967). Statistical Methods for Agricultural Workers, 2nd edition, pp. 150-157. *Indian Council of Agricultural Research*, New Delhi, India.
- Rudrapal, A.B. and Basu, R. N. (1979). Physiology of hydration-dehydration treatments in the maintenance of seed viability in wheat. *Indian Journal of Experimental Biology*, 17, 768-771
- Snell, F.D. and Snell, C.T. (1971). Colorimetric Methods of Analysis, Van Nostrand Reinhold Co., New York. IV AAA, 7-145.
- Tinnin, R.O. and Muller, C.H. (1971). The allelopathic potential of Avena fatua; Influence of herb distribution. Bulletin of Torrey Botanical Club, 98, 243-250

SOME ASPECTS OF PESTICIDAL CONTAMINATION IN FRESHWATER FISHES AND THEIR REMEDIAL MEASURES

Biplab Sarkar', Partha Bandyopadhyay², S. Adhikari', S. Ayyappan³ and Bidhan C. Patra² '

¹Central Institute of Freshwater Aquaculture, Kausalyaganga, Bhubaneswar – 751 002, Orissa, India

² Aquaculture Research Unit, Department of Zoology, Vidyasagar University, Midnapore - 721 102, West Bengal, India

³Deputy Director General (Fisheries), Indian Council of Agricultural Research, Krishi Anusandhan Bhavan, Pusa, New Delhi – 110 011, India.

ABSTRACT

In the last 20th Century, toxicological analysis of different hazardous chemicals and their degradation forms has been done by new techniques and their residual concentration has been quantified which gives a new dimension to toxicological research. Pesticides that are transported to the aquatic environment are primarily of agricultural origin. Fish accumulates pesticide with poor water solubility from water or food. Fishery, especially aquaculture has become a booming industry worldwide and pesticide pollution is a global phenomenon. Multiple cases of mass mortality of fish in tanks have already been recorded through pesticide disposal. Fish has been considered a good species for biosensor in aquatic toxicology and environmental impact assessment. It is a hot topic of several recent reviews and symposia and scientists are concentrating heavily on this avenue. It is necessary to find a suitable antagonistic or antidote to check the poisoning caused due to pesticide pollution and to re-establish the life processes.

KEY WORDS : Pesticide, Freshwater Fishes, Toxicity, Antidote

Correspondent author

Telephone: Office : (91) 03222-275297 Fax : (91) 03222-275329 Residence : (91) 03222-266035 e-mail : bcpatra@yahoo.com

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Introduction

The application of chemicals to plants as a protection against insects is an age old practice. Early part of nineteenth century was the first time when serious attempts were made to control some of the injurious insects by the use of chemicals. Agricultural chemicals came into commercial practice soon after the Second World War. In India, with the occurrence of green revolution the demand of pesticides has been increasing day by day. National data survey depicts that there are more than 68,000 species of pest and disease causing insects and annual loss of food grains due to pests and disease is about Rupees 6000 crores and out of it 26 percent of loss is alone caused by insects and rodents and for this, in 1977 about 38,000 tonnes of insecticides were used in India.

The average consumption of pesticides in our country increased from 3.2 g-lha in 1954-55 to 336 g-lha in 1980. These pesticides have been divided into diferent types viz. insecticide, herbicide, fungicide, nematicide, rodenticide, molluscide, avicide, repellant, attractant etc. Now-a-days carbamate and pyrethroid insecticides are getting more preference over traditional or organochlorine and organophosphates, because of their special characterisation. Both are extremely high insecticide even at low doses and biodegradable in nature.

Fish accumulate pesticides, especially those with poor water solubility, from water or food. The uptake from water occurs because of the very intimate contact with the medium that carries the chemicals in solution or suspension and also because fish have to extract oxygen from the medium by passing enormous volumes of water over the gills. Water that is transported actively (marine fish) or passively (freshwater fish) into the body may also contribute to the uptake from water during the lifetime of a fish. The process of uptake in fish body is influenced by several factors such as chemistry of the molecule, physical condition of medium, and the fish itself—its lipid content, size, stages of development, physiological activity etc.

Fishery, especially aquaculture has become a booming industry worldwide and pesticide pollution is also a global phenomenon. Nevertheless, aquaculture is also sometimes using pesticide, i.e. cypermethrin is applied for argulus-control, some pesticides to kill undesirable fishes, carbofuran is important for paddy-cumfish or fish predator like lamprey or to control aquatic snails that carry various disease like Bilharzia etc. Universally now a days, fish has been considered a good species for biosensor in aquatic toxicology and environmental impact assessment because firstly, fish is the terminal member of aquatic food chain. Secondly, it reacts sensitively to changes in its environment. Thirdly, it suffers damages in gill, skin, digestive tract etc. Fourthly, gasping for air swaying, loss of orientation, lethergy are the other indicators demonstrated by it. It has been well said that in order to

eliminate a species, a pollutant need not be lethal. The survival of a species is affected if its reproductive capacity is affected. Thus, from a practical fisheries standpoint, this aspect of physiology has prime ecological significance. Considering all the above points, some plant extracts and organic matters have been applied as antidotes to counteract pesticidal action. But it needs more studies to come out successfully with some antidotes against these pesticides.

Acute Toxicity and Test Animals

Though many reports have been published describing detailed methods and procedures of use in acute toxicity test with aquatic biota (Jones 1940, Katz 1961), recommended procedures are provided by the U.S. Environmental Protection Agency (1978), The American Public Health Association (1985), The Organization of European Community Development (OECD, 1981).

A large number of papers had been published on the acute toxicity of pesticides to fishes (Mukhopadhay *et.al.*, 1980; Domitrovic 1997; Abdelghani *et.al*, 1997, Farrell *et.al.*, 1998; Pathiratne *et.al.*, 1998; Patra, 2002 a, b.)

Many researchers has studied several kinds of fish species for some of their merits. Lloyd (1963) worked on the toxicity of Salmo gairdneri and Lepomis macrochirus; Gomez el.al. (1998) on tench (Tinca tinca L.); Pathiratne and George (1998) on Nile tilapia (Oreochromis niloticus); Dalvi and Davis (1998) on Channel catfish; Soengas et.al. (1997) on rainbow trout, Tsuda et.al. (1997) on killifish (Oryzias latipes) etc.

In India, test species selected by Bhattacharya et.al (1992) was Channa punctatus, by Mukhopadhyay et.al. (1984) was Clarias batrachus, and by Fernandez et.al. (1996) was Etroplus maculatus etc.

Jhingran (1982) has made a literature survey of Indian scientists and published a variable acute toxicity chart of different fish sepcies and analysed their prospective results.

Safe Concentration and Chronic Toxicity

Safe concentration is the maximum concentration of a toxicant that has no observable harmful effects after long-term exposure over one or more generations. Sprague (1971) emphasized that the term "Safe level" is not an entirely satisfactory one, but in the absence of a better term, it has gained wide acceptance. A number of scientists and many organizations have given formula and concrete suggestions for the estimation of safe concentration. Hart *et.al.* (1948) proposed the formula which is already established and popular. European Fisheries Adviosory Commission (EFAC, 1979) suggested that a concentration less than four orders of magnitude lower than the 96h LC_{so} is an acceptable or safe concentration. Another

approach to arrive at safe level is to use the one tenth of the 48h LC_{50} value.

Mount and Stephan (1969) proposed to use Laboratory Fish Production Index (LFPI), which incorporates data generated over at least one generation of the test species, regarding effects on survival, growth, reproduction etc. to calculate the Maximum Acceptable Toxicant Concentration (MATC). Johnson and Julin (1980) considered the MATC is acceptable more than other methods because it is less time consuming, less expensive and is widely accepted.

Pesticides

Carbamates are moderately toxic to fish, but highly toxic to invertebrates, which were introduced into the late 60's for their modified characters over organophosphates. Among them, Carbofuran and Carbaryl are commonly used (Johnson *et.al.*, 1980). Singh, *et.al.* (1982) worked on the pesticides (carbamate) induced changes in circulating thyroid hormones in the freshwater catfish, *Clarias batrachus.* Belisle *et.al.* (1988) did specific analysis of Carbamate Pesticides in sediments using column extraction and gas chromatography. Bocquene *et.al.* (1993) studied on the effect of carbamate on acetyl cholinesterase system of fish. Hota *et.al.* (1993) analysed the metabolic effects of kilex carbaryl on a freshwater teleost, *Channa punctatus* (Bloch.).

Cypermethrin has been developed in England by Elliot *et.al.*, 1975. There is very little information available regarding the toxicity of cypermethrin to fish. Only some early life stage tests and detection have so far been performed. The acute toxicity of cypermethrin to Brown-trout, Rainbow trout, Tilapia was studied by Stephenson (1982). Singh *et.al.* (1993) worked on the effect of cypermethrin on lactate and succinate dehydrogenase and cytochrome oxidase of fish and Reddy *et.al.* (1995) studied on sublethal concentration of cypermethrin to the protein metabolism level in selected tissues of *Cyprinus carpio*. Davies (1993) described on sublethal effects on Brown trout, Salmo trouts, caused by cypermethrin spraying on Tasmanian stream. Very little or almost no work has been done on the cypermethrin toxicity to Indian major carps and its different sublethal effects. So, after considering the increasing importance of cypermethrin in aquaculture, it may be taken as a new and challenging avenue of research.

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Parameters Related to Environment

Among all the environmental parameters, the effect, of temperature on the toxicity of pesticides have been studied more and it is very important because it directly influences factors like enzyme activity, metabolic rate, oxygen uptake etc. Generally, toxicity increases with the high temperature. Macek (1969) studied the toxicity of 10 pesticides to the rainbow trout and 11 pesticides to the bluegills at different temperatures and found that the toxicity increased with increasing temperature.

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Same kind of results has been reported by Singh *et.al.* (1993) on endosulfan to *Heteropneustes fossilis*, by Spraque (1970) on endosulfan to rainbow trout. by Duangsawasdi *et.al.* (1979) on fenitrothion and acephate to rainbow trout, But a completely opposite results has been shown by DDT and some pyrethroids where toxicity is less at higher temperature.

pH has an excellent role in influencing various physiochemical properties of pesticides like hydrolysis, volatilization and in balancing the dissociated and undissociated forms (Weber, 1972). With few exceptions, the toxicity of organochlorine (OC) and organophophate (OP) compounds are not influenced by pH. The toxic effects of 2, 4-D were reduced when the pH was raised by the addition of sodium chloride (Holcombe *et.al.*, 1980). Malathion is toxic below pH 7, and looses its toxicity in alkaline pH (Bender, 1969). In the case of carbamate compounds, generally tixicity was reported to increase with pH. *Mexacarbate* was 38 times more toxic to bluegills at pH 9.5 than at 7.5 (Mauck *et.al.*, 1977). The toxicity of picloram to cutthroat trout and lake trout increased with a rise in pH from 6.5 to 8.5. (Marking *et.al.*, 1981).

Hardness of water does not seem to influence the toxicity of OC and OP compounds. The toxicity of three formulations for 2, 4-D, three formulations of endothall, fenoprop., PCP and dichlrobenil was not affected by hardness. (Inglis *et.al.*, 1972). Lioyd and Jorban (1964) found that survival time of Rainbow trout in rapidly lethal pH value shortened with decrease of calcium carbonate content of water.

Not much work seems to have been carried out on the influence of low oxygen on the toxicity of pesticides to fish. Davis and Cook (1993) attempted to formulate the criteria for minimum dissolved oxygen requirement of fish.

Histopathology and Stress

Tissue changes due to sublethal doses of toxicants not only give an idea about the nature of toxicant but also predict upon its mode of action or mechanisim of toxicity, about the probable physiological impairment and assist greatly in the diagnosis of nature of toxicant (Nagler, 1984). Histology of fish and aquatic invertebrates is still a less studied discipline compared to counter parts in mammals.

Katz (1961) worked on the exposure of endosulfan and carbaryl on the ovaries of *Channa striatus* (Bloch) and observed reduction in the number of oocytes, increased number of damaged oocytes, development of inter follicular spaces, reduction in gonado-somatic index etc. Shukla *et.al.* (1984) have noted decreased ovarian activity and atretic oocytes in *Sarotherodon mossambica* exposed to Malathion and Ghosh *et.al.* (1985) has described on damage in ovary, viz., degeneration of follicular wall, ooplasm and connective tissues due to malathion toxicity on *Heteropneustes fossilis* and also worked on malathion toxicity on the ovary of zebra fish (*Brachydanio rerio*). Singh *et.al.* (1993) studied extensively on the impact of gamma-BHC to the steroid hormonal profiles of *Heteropneustes fossilis* and correlated it to ovarian damage, and shown that pesticide suppressed the levels of these hormones, viz. testosterone (T), estradiol-17 beta, 17 alpha-OHPg etc. and described that Ovine leutenizing hormone releasing hormone and Mystus gonadotropin has modulatory role on it. Pandey (1988) worked on the impact of endosulfan (thiodon) EC 35 on dynamics of oocyte development in the teleostean fish, *Colisa (Trichogaster) fasciatus* and described that ovarian activity was retarded greatly, the diameter of oogonia and stage I oocytes was greatly reduced and ovarian wall become thicken. Hazarika (1988) worked on the toxicological impact of different sublethal doses of BHC on the ovary of the air-breathing catfish, *Heteropneustes fossilis* (Bloch.) and investigated detailed histopathological changes.

Because of its physiological importance, already some literatures are available on liver. Couch (1975) reviewed the histopathological effects of pesticides and related chemicals on the livers of fish and concluded that many of them were nonspecific. Bhattacharya *et.al.* (1975) observed liver cord necrosis and cytoplasmic and nuclear disintegration of the hepatopancreas in the teleost *Clarias batrachus* due to endrin and reported that chronic exposure to thiodon and agallol caused rapid degeneration and vacuolation of hepatocytes in fish liver. Das (1996) observed coagulation of blood in sinusoids, necrosis and cytoplasmic disintegration in the hepatopancreas of *Channa punctatus* exposed to malathion. Altamirano-Reynoso (1984) analysed that lindane caused hydropic degeneration and the tubular epithelium necrosis were the most frequent microscopic injuries observed in liver of *Tilapia hornorum*. Sahai and Singh (1984) described vacuolation of hepatocytes of *Rasbora dendricus* while exposed to malathion toxicity.

Reproduction and Breeding

Reporductive toxicity indicates changes on the pattern of breeding response, on fecundity, on fertilization rate, hatchability of larvae and above all survivability of larvae, fry etc. Though, it is a prime subject of research because it directly relates to productivity of fishes. Very less researches has been conducted in this field. Very less or no research has been done on the effect of carbamate and synthetic pyrethroid on fishes specially to Indian major carps.

Following the feeding of DDT to brook trout at different doses for 22 weeks, the fishes fed with lower doses showed significantly higher number of ova than those of higher dose (Macek, 1968). In experimental ponds, 2, 4-D butylester at the highest concentration caused a delay in the spawning of blue gills (Cope *et.al.*, 1970). Oviposition of *Oryzias latipes* was suppressed heavily by the 96 hour exposure of chlordecone (Curtis, 1978). Fenitrothion, at sublethal concentration

affected egg production, whereas temephos at sublethyal concentration affected normal birth. Fecundity and fertility of female sheephead minnows exposed to different doses of chlordecone shown marked reduction (Goodman *et.al.*, 1982).

On the countrary, hatchability of coho salmon eggs, exposed to PCBs, was much reduced (Halter, 1974). Eggs of DDT exposed adults showed abnormal gastrulation, and 39% of them had vertebral deformities upon hatching. The percent of deformed larvae was dose-dependent (Smith *et.al.*, 1973). Embryos of Atlantic silverside was treated singly with P.P-DDT, malathion or Carbaryl and was observed that they reduced the survival time of the embryos. These effects were observed even at concentration that may occur temporarily in the environment (Weis *et.al.*, 1976). Embryos, sacfry, and larvae of *Caranx* exposed to lindane had advanced hatching and larval characters. Yolk utilization and sac absorption were also impaired. Hatching success of fish eggs exposed to low concentration of mirex increased and hatching of fathead minnows eggs exposed to different concentration of chlordecone was significantly lower than that of the controls. (Buckler *et.al.*, 1981).

Blood Parameters

These parameters are influenced and modified by many factors, such as, handling, metabolism (Booke, 1964), age (Tugarina and Ryzhova, 1970), maturity (Qureshi *et.al.*, 1971), sex (Steveke and Atherton, 1965), diet (Smith, 1968), stress (Bouch and Ball, 1966) and pollutants (Spraque, 1971). Some of researches has already been conducted on haematological parameters by several types of pesticides on different fish species by the scientists of India and abroad. Witt *et.al.* (1966) found a good correlation between DDT levels in blood and those in adipose tissue. Moss *et.al.* (1966) shown that entering the curculatory system, pesticides rapidly bind to the blood proteins. A sublethal concentration of malathion induced decreased erythrocytes and increased leukocyte count and increased serum-free amino acid in a freshwater catfish (Chliomotvitch *et.al.*, 1977).

Blood glucose and hepatic glycogen can be utilized as a parameter of stress response (Chavin, 1973). Relatively innocuous stimuli such as aquarium transfer (Chavin and Yo ung, 1970), thermal shock (Wedemeyer, 1973) and putting them in a small shaking cages elicited hyper glycemic response in fish.

Blood glucose and hepatic glycogen profile in *Channa punctatus* exposed to non-lethal concentration of single and mixture pollutants respond diversely to two types of treatments (Bhattacharya *et.al.*, 1987). According to them, blood glucose profile can be utilized as indicator of pollutional stress in fish. Generally blood glucose value were elevated after pesticide exposure to fish. These sorts of result has been observed in *Auguilla anguilla* by the application of endosulfan, in *Cyprinus carpio* by deltamethrin (Balint *et.al.*, 1995); (Chandrasekar *et.al.*, 1993) etc.

Study of Antidotes

Zamfir (1979) worked on the possibilities for the removal of pesticide polluted water in treatment stations and described some methodologies, i.e. flocculation and filtration can remove partially-DOT, 2, 4, 5-T, edrin, parathion and lindane, Chlorine oxidation can remove parathion, diuron etc., ozone and pottassium permanganate appear to exert effects similar to those of chlorination. Activated charcoal has positive effects in the removal of absorption of most pesticide and U-V ray also can remove a certain amount of pesticide.

Sado *et.al.* (1992) reported that increased temperature and optimum level of dissolved oxygen (by aerator) can decrease the pesticidal action and Das (1996) suggested application of lime to increase the pH for counteraction. Mukherjee (1996) commented for the efficacy of ascorbic acid (Vitamin-C) for the intoxication of different pollutants including pesticides.

Conclusion

Few researchers have also concentrated their studies on the impact of carbofuran to several species of fishes. But no or very few of them has emphasised on Indian major carps. Pyrethroids are extremely toxic under laboratory conditions; in the field their toxicity is significantly reduced, presumably by absorption of the toxicant to suspended solids. Some literatures are available on the ovarian damage due to pesticide toxicity but very few or no research has done on the histopathological changes of ovarian cycle on Indian major carps. Though physiological effects of toxicants and their use to diagnose the disease are well established in human and veterinary medicine, yet in fish these clinico-pathological tests are still to be standardized. In this background haematological studies presented variation in blood parameters as reliable indices of fish health. But application of different herbal extracts for this purpose is very purposeful step, which can open new avenues of research.

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References

- Abdelghani, A. A., Tehounwou, P.B., Anderson, A. C., Sujono, H., Heyer, L. R., and
 A. Monkiedje (1997). Toxicity evaluation of single and chemical mixtures of round up, Garlon-3A, 2, 4-D and syndets surfactant to channel catfish (*Ictalurus punctatus*), bluegill sunifish (*Lepomis microchirus*) and craw fish (*Procambarus* sp.) Environ. Toxicol. Water-gual. 12(3): 237-244.
- Altamirano-Reynoso, R. (1985). Microscopic liver and kidney injuries in *Tilapia* hornorum caused by an acute intoxication with a commercial product that had lindane. Thesis or Dissertation - Universidad Nac. Autonoma. dc Mexico (Mexico). Fac. De Med. Vet. Zootec.
- Balint, T., Szegletes, T., Szegletes, Z., Halasy, K. and J. Namesok (1995). Biochemical and subcellular changes in Carp exposed to the organophophorus methidathion and the pyrethroid deltamethrin. Aquat. Toxicol. 33 (3-4) : 279-295.
- Belisle, A. A. and D. M. Swineford (1988). Simple, specific analysis of organophosphorus and carbamate pesticides in sediments using column extraction and gas chromatography. *Environ. Toxicol. Chem.* 7(9): 749-752.
- Bender, M. E. (1969). The toxicity of the hydrolysis break down products of malathion to the fathead minow (*Pimephales promelas* Raf). Wat. Res. 3 : 571-582.
- Bhattacharya, S., Mukherjee, S. and S. Bhattacharya (1975). Toxic effects of endrin on hepatopancreas of the teleost fish. *Clarias batrachus* (L). *Ind. J. Experiment Biol.*, 13 : 185-186.
- Bhattacharya, T., Roy, A. K. and S. Bhattacharya (1987). Histopathology of the kidney of *Channa punctatus* (Bloch) exposed to sublethal and chronic levels of Industrial effluents. II. *Ziool. Jb. Anat.* 115 : 263-272.
- Bocquene, G., Galgani, F., Burgeot, T., Le-dean, L. and P. Truquet (1993). Acetylcholinesterase levels in marine organisms along French Coasts. *Mar. Pollut. Bull.* 26(2): 101-106.
- Booke, H. E. (1964). Blood serum protein and clacium levels in yearling brook trout. Prog. Fish. Cult. 26: 107-110.
- Bouck, G. R. and R. C. Ball (1966). Influence of capture methods on blood character and mortality of rainbow trout (Salmo gairdeneri). Trans. Am. Fish. Soc. 95(2): 170-176.

- Buckler D. R., Witt. A. Jr., Mayer, F. L. and J. N. Huckins (1981). Acute and Chronic effects of kepone and mirex on the fathead minnow. *Trans. Am. Fish. Soc.* 110-270.
- Chandrasekar, S. and N. Jayabalan (1993). Haematological responses of the common carp., *Cyprinus carpio* L. exposed to the pesticide endosulfan. *Asian Fish. Sci.* 6(3): 331-340.
- Chavin, W. (1973). In response of fish to invivo-mental changes. Eds. W. C. Chavin. Spring field. Llinoisis. PP. 199.
- Chavin, W. and J. E. Young (1970). Factors in the determination of normal serum glucose levels of gold fish. Carassius auratus J. Comp. Biochem. Physiol. 33 : 629-653.
- Chilamovitch, Y. P. and C. Kuhn (1977). Behavioural, haematological and histological studies on acute toxicity of bis (tri-n-butyltin) oxide on *salmo gairdneri* Richardoon and *Tilapia rendalle* Boulenger. J. Fish. Biol. 10: 1575.
- Cope, O. B., Wood, E. M., and G. H. Watten (1970). Some chronic effect of 2, 4-D on the bluegill (*Lepomis macrochirus*). Trans. Am. Fish. Soc., 99 : 1.
- Couch, J. A. (1975). Histopathological effects of pesticides and related chemicals on the liver of fishes. In : *Pathology of fishes*. Ribelin nand Migaki (eds)(. The University of Wisconsin Press. PP. 559-584.
- Curtis, L. R. and R. J. Beyers (1978). Inhibition of oviposition in the telost Oryzias latipes, induced by subacute kepone exposure, Comp. Biochem. Physio, 61: 15.
- Dalvi, R. R. and S. W. Davis (1998). Role of beta naphthoflavone in the acute toxicity of chlorpyrifos in channel catfish. Bull. Environ. Contam. *Toxicol.* 60(2) : 335-339.
- Das, B. (1996). Ph.D. Thesis. Orissa University of Agriculture and Technology, Bhubaneswar, Orissa.
- Davies, J. S. (1975). Minimal dissolved oxygen requirements of aquatic life with emphasis on Canadian species : a review. J. Fish. Res. Bd. Can. 32(12) : 2295-2332.
- Davies, P. E. and L. S. J. Cook (1993). Catastrophic macroinvertebrate drift and sublethal effects on brown trout, *Salmo trutta*, caused by cypermethrin spraying on a Tasmanian Strcam. *Aquat. Toxicol.* 27(3-4) : 201-224.

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- Domitrovic, H. A. (1997). The use of autochthonous fishes in toxicity tests. Evaluation of the species Acquidens portalegrensis. Rev. Ictiol. 5(1-2): 37-42.
- Duangswawasdi, M. and J. F. Klaverkamp (1979). Acephate and fenitrothion toxicity in rainbow trout : effects of temperature strem and investigations on teh sites of action. Aquat. Toxicol. 35.
- Elliot, M., Farham, A. W. Janes, N. F., Netham, P. H. and B. C. Pearson (1975). Insecticidal activity of the pyrethrins and related compounds. VII. Insecticidal dihalo vinyl analogues of cis and trans Chrysanthemates. *Pest.* Sci. 6:537.
- Farrell, A. P., Stockner, E. and C. J. Kennedy (1998). A study of the lethal and sublethal toxicity of polyphase P-100 an antisapstain fungicide containing 3-iodo-2 propynyl butyl carbamate (IPBC), on fish and aquatic invertebrates. Arch. Environ. Contam. Toxicol. 35(3): 472-478.
- Fernandez, T. V., Thomas, G. Vardhanan, Y. S., R. M. Kumar (1996). An assessment of the toxicity levels of BHC on four aquatic organisms. J. Environ. Biol. 17(1): 21-24.
- Ghazaly, K. S. (1994). Efficacy of Ascorbic acid (Vitamin C) on experimental copper intoxication in *Tilapia zilli. Bull. Natl. Inst. Oceanogr. Fish*, Egypt. 20(2) : 249-257.
- Ghosh. R. and J. Dutta (1985). Malathion toxicity in air-breathing catfish singhi (Heteropneustes fossilis). Bulletin Ind. J. Fish. 32: 447-452.
- Gomez, L., Masot, J., Martinez, S., Duran, E., Soler, F. and V. Roncero (1998). Acute 2,4-D poisoning in tench (*Tinca tinca* L.): Lesions in the hematopoitic portion of the kideny. Arch. Environ. Contam. Toxicol 35(3): 479-483.
- Goodman, L. R. Hansen, D. J., Manning, C. S. and L. f. Favas (1982). Effects of kepone on the sheepshed minnow in an entire life cycle toxicity test, Arch. Environ. Contam. Toxicol., 11: 335.
- Halter, M. T. and H. E. Johnson (1974). Acute toxicities of a Polychlorinated biphenyl (PCB) and DDT alone and in combination to early life stages of Coho salmon (*Oncorynchus Kisutch*).
- Hart, W. B., Weston, R. F. and J. G. Demann (1948). An apparatus for oxygenating test solution in which fish are used as test animals for evaluating toxicity. *Trans. Am. Fish. Soc.* 75 : 228.

- Hazarika, R. and M. Das (1998). Toxicological impact of BHC on the ovary of the air-breathing catfish *Heteropneustes fossilis* (Bloch). Bull. Environ. Contam. and Toxicol. 60(1); 16-21.
- Holcombe, G. W., Flandt, J. T. and G. L. Phipps (1980). Effects of pH increases and sodium chloride addition on the acute toxicity of 2-4 dichlorophenol to the fathead minnow. *Water Res.* 14 : 1073.
- Hota A. K., Mishra D. K. and P. C. Tripathy (1993). Metabolic effects of kilex carbaryl on a freshwater teleost, channa punctatus (Bloch.) Muzaffarnagar-India Society of bioscience. 335-342.
- Inglis, A. and E. L. Davis (1972). Effects of water hardness on the toxicity of several organic and inorganic herbicides to fish. 6 : 23 26.
- Jhingran V. G. (1982). Fish and Fisheries of India. Hindustan Publishing Corporation, New Delhi, India. pp. 666.
- Johnson, W. W. Walter and A. M. Julin (1980). Acute toxicity of toxaphene to fathead minnows, EPA-600/3-80. 005. U. S. Environmental Protection Agency, Duluth : Minn.
- Johnson, W. W. Walter and M. T. Finley (1980). Handbook of Acute toxicity of chemicals to fish and Aquatic invertebrates. Resources publ. 137. Fish and Wild Life Service. U. S. Department of the Interior. Washington D. C.
- Jones, J. R. E. (1940). The toxicity of the double chlorides of mercury and sodium. I. Experiments with the monow *Phoxinus phoxinus* (L.) J. Experiment Biol. 17: 325-330.
- Katz, M. (1961). Acute toxicity of some organic insecticides of three species of salmonids and spike stickleback. *Trans. Amer. Fish. Soc.* 90 : 310.
- Lioyd, R. and D. H. M. Jordan (1963). Predicted and observed toxicities of several sewage effluent to Rainbow trout. J. Proc. Inst. Sewage Purif. 2:16.
- Macek, K. J. (1968). Reproduction in brook trout (Salvelinus fontinalis) fed sublethal concentration of DDT, J. Fish Res. Board Can. 25: 1787.
- Macek. K. J., Hutchinson, C and O. B. Cope (1969). The effect of temperature on the susceptibility of blue gills and rainbow trout to selected pesticides. Bull. Environ. Contam. Toxicol. 4 : 174.
- Marking, L. L. (1981). Toxicity of the synthetic pyrethroid SBP-1382 to fish. Prog. Fish Culturist. 36 : 144.

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- Mauck, W. L., Olson, L. E., and J. W. Hogan (1977). Effects of water quality on deactivation and toxicity of mexacarbate (zectran) on fish. Arch. Environ. Contam. Toxicol. 6: 385.
- Moss, J. A. and D. E. Hathway (1964). Transport of organic compounds in the mammal : partition of dieldrin and telodrin between the cellular eomponents and soluble proteins of blood. *Biochem. J.* 91 : 384.
- Mount, D. J. and C. e. Stephan (1967). A method for establishing acceptable toxicant limits for fish : Malathion and the butoxyethyl ester of 2, 4-D Trans. Amer. Fish. Soc. 96 : 185-193.
- Mukherjee, M. (1996). Antidote to combat white spot disease in culture ponds. Fish. Chimes. 16(5): 30-31.
- Mukhopadhyay, P. K. and P. V. Dehadrai (1980). Biochemical changes in the air-breathing catfish, *Clarias batrachus* exposed to malathion. *Environ. Res.* 22:149-158.
- Nagler, J. J., Aysola, P. and S. M. Ruby (1984). The seasonal effect of sub-lethal pentachlorophenol exposure on ovarian development in rainbow trout (Salmogairdneri). 11th annual aquatic toxicity workship, Richmond, B. C., November 13-15. 1984 (np.)
- OECD, (1979). Report on the assessment of potential environmental effects of chemical. I. OECD, pp. 28.
- Pandey, A. C. (1988). Impact of endosulfan (thiodon) EC 35 on behavirour and dynamics of oocyte development in the teleostean fish, colisa (*Trichogaster* fasciatus). Ecotoxicol. Environ. Saf. 15(2): 221-225.
- Pathiratne, A. and S. G. George (1998). Toxicity of malathion to Nile tilapia, Oreochromis niloticus and modulation by other environmental contaminants. Aquat. Toxicol. 43(4): 261-271.
- Patra, B. C. (2002a). Steriod hormones and ecotoxicological implicatious in the aquatic environment. In Ecology of Polluted Water (Eds. A. Kumar). A. P. H. Publishing Corporation, New Delhi, India. pp. 723-758.
- Patra, B. C. (2002b). Role of Ecotoxicological Research to the protection of our Aquatic Environment. In Ecology and Ethology of Aquatic Biota (Ed. A. Kumar), Daya Publishing House, New Delhi, India, PP. 134-148

- Qureshi, M. D., Heldin, R. V., Vanstone, W. E. and P. A. Anastassiadis (1971). Levels of constitutents of glycoproteins in the sera of pacific salmon. J. Fish. Res. Bd. Can. 28(8): 1173-1179.
- Reddy, P. M. and M. Bashamohideen (1995). Alterations in protein metabolism in selected tissues of fish, *Cyprinus carpio*, during sublethal concentration of cypermethrin *Environ. Monit. Assess.* 36(2): 183-190.
- Sado, E. K. and E. O. Ita (1992). The effects of capture methods, temperature and oxygen on the survival of live freswater clupieds during acclimation and transportation. J. Aquacult. Trop. 7(2): 165-176.
- Sahai, S. and S. Singh (1989). Hepatotoxic effects of some pesticides in *Puntius ticto*. Environment and Experimental Toxicology. Jai Research Foundation, Valvada - 396108.
- Shukla, L., Shrivastava, A., Merwani, D. and A. K. Pandey (1984). Effect of sublethal malathion on ovarian histopathology in Sarothrodon mossambicus. Comp. Physiol. Eco., 9 : 13-17.
- Singh, P. B., Kime, D. E. and T. P. Singh (1993). Modulatory actions of Mystus gonadotropin on gamma-BHC induced histological changes, cholesterol and sex steroid levels in *Heteropneustes fossilis*. Ecotoxicol. Environ. Saf. 25(2) : 141-153.
- Smith, C. E. (1968). Haematological changes in the cohosalmon fed a folic acid deficient diet. J. Fish. Res. Bd. Can. 25 : 151-156.
- Soengas, J. L., Strong, E. F., Aldegunde, M. and M. D. Andres (1997). Effects of an acute exposure to lindane (gamma-hexachlorocyclohexane) on brain and liver carbohydrate metabolism of rainbowtrout. *Ecotoxicol. Environ. Saf.* 38(2) : 99-107.
- Spraque, J. B. (1971). Measurement of pollutant toxicity to fish III. Sublethal effects and "safe" concentration. *Water Res.* 5: 245-266.
- Stephenson. C. E. (1982). In analysis of established pyrethroid insecticides. *Residues Review.* 39 : 188.
- Steveke, E. W. and C. R. Atherton (1965). Use of micro haematocrit value to six largemouth. Bass. Prog. Fish. Cult. 27: 87-90.
- Tsuda, T., Kajima, M., harada, H., Nakajima, A. and S. Aoki (1997). Acute toxicity, accumulation and excretion of organophosphorus insecticides and their oxidation products in Killifish. *Chemosphere*, 35(5): 939-949.

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Vidyasagar University Journal of Biological Sciences. Vol. 8, 2002

- Tugarina, P. Y. and L. N. Ryzhova (1970). Age related feature of the blood of the black Baikal graying *Thymallus arcticos* baicalensis. J. Ichthyol. 10: 384-357.
- Venugopalan, V. K. and P. Sasibhushana Rao (1979). Pesticide induced impairment in incubation and post embryonic development of planktonic eggs of esturine fish of Porto NOVO (South India) Waters. Proc. Symp. Environ. Biol. Academy of Environmental Biology, Muzzaffarnagar, India 397.
- Vittozzi, L. and G. D. Angelis (1991). A critical reiew of comparative acute toxicity data on fresh water fish. Aquat. Toxicol. 19: 167-204.
- Weber, J. B. (1972). Interaction of organic pesticides with particulate matter in aquatic and soil systems, in fate of organic pesticides in the Aquatic Environment (Advances in Chemistry Series III). Faust, S. D., Eds. American Chemical Society Washington D. C. pp. 55.
- Wedemeyer, G. (1973). Some physiological aspects of sublethal heat stress in the juvenile steelhead trout (Salmo gairdneri) and coho salmon (Onchorhynchus kisutch). J. Fish. Res. Bd. Can. 30: 831.
- Weis, J. S. and P.S. Weis (1976). Optical malformations induced by insecticides in embryoes of the Atlantic silverside, *Menidia menidia*, *Fish. Bull.* 74 : 208.
- Witt, J. M., Brown, W. H., Shaw, G. I., Maynard, L. S., Sullivan, L. M., Whiting F. M. and J. W. Stull (1966). Rate of transfer of DDT from the blood compartment. *Bull Environ. Contam. Toxicol.* 1 : 187.
- Zamfir, Gh (1979). Water pollution by pest controlling agents and the possibilities for their removal in surface water treatment stations. *Igiena* 28(2): 97-103.

REGULATION OF TANNASE SYNTHESIS IN AUREOBASIDIUM PULLULANS DBS66

Debdulal Banerjee, Pradeep K. Das Mohapatra, Keshab C. Mondal and Bikas R. Pati *

Microbiology Laboratory, Depertment of Botany and Forestry, Vidyasagar University, Midnapore - 721 102, West Bengal, India, e-mail : brpati@yahoo.com.

SUMMARY

Tannase (Tannin acyl hydrolase; EC 3.1.1.20) has been obtained from a newly isolated soil fungus *Aureobasidium pullulans*. Regulation of tannase biosynthesis in isolated fungus has been studied in presence of different utilizable carbon sources in the culture media containing basal salts. Maximum tannase production occurred in presence of tannic acid compared to gallic acid or in combination of glucose and gallic acid. Catabolic repression of enzyme synthesis was observed in presence of glucose. Among the various concentrations 1% (w/v) tannic acid in the culture medium induced maximum tannase after 48h of cultivation.

KEYWORDS : Aureobasidium pullulans, Gallic acid, Tannase, Tannic acid.

Introduction

Tannase (Tannin acyl hydrolase; E. C. 3.1.1.20), an industrially important enzyme hydrolyses the ester linkages of tannic acid into glucose and gallic acid (Lekha and Lonsane, 1997; Banerjee *et.al.*, 2001). It is widely used in the manufacture of instant tea, clarification of coffee flavored soft drinks, mediarization of wine and fruit juices, and gallic acid (Vermeire and Vandamme, 1988; Bajpai and Patil, 1996).

Tannic acid, a major component of plant tannin induces tannase synthesis in *Bacillus licheniformis* (Mondal *et. al*, 2000), *Aspergillus sp* (Banerjee *et al.*, 2001) and *Penecillium sp* (Bajpai and Patil, 1996). Moreover tannase is a constitutive enzyme and its formation is controlled by catabolic repression but low concentrations of glucose or lactose is not repressive (Mondal *et al.*, 2000). Similar observation was also made by Bradoo *et al* (1997) in *Aspergillus japonicus*. Species of *Aspergillus* are best known producer of this enzyme (Lewis and Starkey, 1969; Lekha and Lonsane, 1997; Banerjee *et al.*, 2001). Synthesis of tannase from *Aureobasidium pullulans* was not reported earlier. However information concerning the detailed regulatory mechanism of tannase synthesis is limited. In the present communication, regulation of tannase synthesis in *Aureobasidium pullulans* DBS66 has been worked out in presence of tannic acid, gallic acid and glucose. Optimization of tannase also been made for tannase biosynthesis.

^{*} Corresponding author

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Materials and Methods

A number of tannase producing fungi were isolated on selective tannic acid agar medium from different forest soils of Midnapore District, West Bengal, India. Sixteen potent tannase producing fungi were selected on the basis of larger clear zone around the colonies. Each fungus was grown on liquid medium containing $(gm/L):(NH_4)_2HPO_4, 3.0; MgSO_4, 1.0; KH_2PO_4, 0.5; CaCl_2, 0.3; and tannic acid (Filter$ sterilized), 10.0. The pH of the medium was adjusted to 5.5. On the basis of tannaseproducing ability one fungus (DBS66) has been selected for the present study. Thesame was preserved in PDA slant at 4° C for further work. Identification of thestrain was made from MTCC, Chandigarh, India, as*Aureobasidium pullulans*andthe same is deposited there as strain no 3828.

An inoculum was prepared by growing the organism in a 250 ml Erlenmeyer flask containing 50 ml of the tannic acid broth (pH 5.5) having identical composition with the isolation medium. A 2% inoculum (v/v) was added and incubated at 30° C for 36 h in a rotary shaker (200 rpm). The culture supernatant obtained by filtration (through Whatman No. 1 paper) was assayed periodically for extracellular tannase activity. Enzyme production has been carried out in presence of different concentrations of tannic acid. Production is also carried out in various combination of tannic acid, gallic acid and glucose. Growth of the organism was estimated on the basis of biomass dry weight (mg/ml). Tannase activity was measured by the method of Mondal et al (2001). The reaction mixture consists of substrate tannic acid 0.5 ml (0.5% w/v in 0.2 M acetate buffer pH 5.5) and 0.3 ml crude enzyme. The enzymatic reaction was stopped by the addition of bovine serum albumin (1mg/ml) which precipitates the unbroken tannic acid. The precipitate was dissolved in SDStriethanolamine (1% w/v, SDS in 5% v/v triethanolamine) and the absorbency was measured with FeCl₃ at 550 nm. One unit of tannase was defined as the amount of enzyme that is able to hydrolyze 1μ -mole of ester linkage of tannic acid in 1 min under the specified condition.

All the experiments have been done in triplicate.

Results and Discussion

Tannase biosynthesis by Aureobasidium pullulans was studied using different utilizable carbon source in the culture media (Table - I). It has been observed that maximum tannase production was occurred in presence of tannic acid compared to gallic acid or in combination of glucose and gallic acid in the medium containing basal salt (Table - I). The enzyme production was not enhanced by simultaneous addition of glucose or gallic acid in the tannic acid medium. This result indicated that tannic acid is the main inducer for tannase synthesis by this organism. The concentration of tannic acid has also been optimized for tannase biosynthesis. It has been observed that 1% (w/v) tannic acid in the culture medium induced maximum tannase after 48h of cultivation (Fig-1), Our result is comparable with the findings of earlier workers (Lekha and Lonsane, 1997; Vermire and Vandamme, 1988). They also mentioned that concentration of tannic acid is a crucial factor for tannase synthesis on different organisms. Maximum growth of the organism (0.88mg/ml) was observed in the presence of glucose containing tannic acid medium. Though the organism showed higher growth in presence of glucose but failed to produce enzyme. Failure of enzyme synthesis in presence of readily available carbon source indicates its catabolic repressive nature (Shih and Labbe, 1994).

The proper cause behind the induction of tannase by tannic acid is not known to us. Earlier Prist (1971) mentioned that microbial exoenzyme synthesis may be induced in two ways, substrate has cell wall binding site, such as penicillinase or microorganism produce a low basal level of constitutive exoenzyme degrades its substrate and the resultant low molecular product enter the cell and induce further exoenzyme synthesis. The regulatory mechanism of tannase synthesis in *Aureobasidium pullulans* by tannic acid may be comparable with the process mentioned by Prist.

Media composition	Growth (mg/ml)	Tannase synthesis (U/ml)
Basal salt (BS)	0.54	ND
Glucose + BS	0.84	ND
Gallic acid + BS	0.79	0.23
Tannic acid + BS	0.78	0.86
Glucose + Gallic acid + BS	0.85	0.65
Glucose + Tannic acid + BS	0.88	0.79
Gallic acid + Tannic acid + BS	0.77	0.68
Glucose + Gallic acid + tannic acid + BS	0.78	0.72

Table – 1, Effect of media composition on growth and tannase production.

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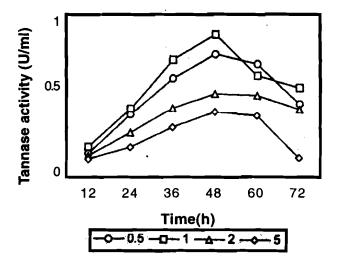


Fig. 1, Effect of tannic acid concentration (g%) on tannase production by A. pullulans DBS66.

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REFERENCES

- Bajpai, B., Patil, S. (1996) Tannin acyl hydrolase (EC-3.1.1.20) activity of Aspergillus, Penicillium, Fusarium and Trichoderma. World Journal of Microbiology and Biotechnology 12, 217-220.
- Banerjee, D., Mondal, K. C., Pati, B. R. (2001) Production and Characterization of Extracellular and Intracellular Tannase from Newly Isolated Aspergillus aculeatus DBF 9. Journal of Basic Microbiology 41(6), 313-318.
- Bradoo, S., Gupta, R., Sexena, R. K. (1997) Parametric optimization and biochemical regulation of extracellular tannase from *Aspergillus japonicus*. *Process Biochemistry* 32, 135-139.
- Lekha, P. K., Lonsane, B. K. (1997) Production and application of tannin acyl hydrolase : state of the art. Advances in Applied Microbiology 44, 215-260.
- Lewis, J. A., Starkey, R. L. (1969) Decomposition of plant tannins by some soil microorganisms. Soil Science 107, 235-241.
- Mondal, K. C., Banerjee, D., Jana. M., Pati, B. R. (2001) Colorimetric Assay Method for Determination of the Tannin Acyl Hydrolase (EC: 3.1.1.20) Activity. Analytical Biochemistry 295(2), 168-171.
- Mondal, K. C., Banerjee, R. and Pati, B. R. (2000) Tannase production by *Bacillus licheniformis*. Biotechnology Letter. 22: 767-769.
- Prist, F. G. (1977) Extracellular enzyme synthesis in the genus *Bacillus*. *Bacteriol.*, Rev., 41, 711-753.
- Shih, N. J., and Labbe, R. G. (1994) Effect of glucose on sporulation and extracellular amylase production by *Clostridium perfringens* Type A in defined medium. *Curr. Microbniol.*, 29, 163-169.
- Vermeire, A., Vandamme, E. (1988) Fungal production of tannin acyl hydrolase. Med Fac Landbouw Rijksuniv Gent 53, 2047-2056.

SMOKING INDUCED OXIDATIVE BURST OF NEUTROPHIL AND RELATED ALTERATION & DAMAGE OF SERUM LIPIDS

Pallab Maity, Tarasankar Maiti, Panchanan Maiti, Somenath Roy*

Immunology and Microbiology Laboratory
 Department of Human Physiology with Community Health
 Vidyasagar University
 Midnapore – 721102
 West Bengal, India.

ABSTRACT

Cigarette smoking is considered as one of the most common causes for oxidative stresses related pathogenesis. Different components of cigarette smoke especially nicotine stimulates the phagocytes to release reactive oxygen intermediates (ROI). Moreover cigarette smoke itself contains several types of reactive oxidants. The relationships between cigarette smoking and serum lipid status, blood leukocyte counts, oxidative burst response of neutrophils and oxidative damage of lipid were examined in 60 adults (50% of whom are smokers). From this study it was evidenced that smoking stimulated the neutrophil oxidative burst response, which is associated with ROI production. Cigarette smoke also altered the serum lipid profile; it increased the total cholesterol, triglyceride and phospholipids levels. The number of blood leukocytes increased due to cigarette smoking. The oxidative damages of serum lipids were more due to cigarette smoking.

KEYWORDS: Smoking, Nicotine, Neutrophil, Reactive Oxygen species, Lipids,

Introduction

Cigarette smoking is the most common and eminently avoidable cause of life style related oxidative stress associated with accelerated onset of degenerative diseases and premature death (Anderson R, 1997). Cigarette smoke is said to contain 4000 or more constituents (e.g., nicotine, acetic acid, CO, CO_2 NO and NO_2) (Wynder EL and Hoffmann, 1979). Cigarette smoke has been implicated as a major risk factor in chronic pulmonary diseases and other several other pathogenesis (Anderson R, 1997).

It was seen that cigarette smoke is responshile for change in lipid profile (Gurdal YE *et al*, 1996). This alteration in lipid profile due to cigarette smoking is thought to be one of the reasons for the occurrence of cardiovascular diseases. Craig *et al* (1989) reported that cigarette smoking resulted in increased serum level of total

^{*} Corresponding Author

cholesterol, triglyceride, LDL-cholesterol and phospholipids and decreased serum level of HDL-cholesterol. Fisher *et al*, (2000) had found that Cigarette smoking was linked to increased cardiac morbidity and mortality, and has been shown to affect lipid profiles. It was also observed that the level of lipid peroxide as elevated in the cigarette smokers, signifying that cigarette smoking is associated with oxidative deterioration of lipid (Banerjee *et al*, 1998).

Numerous studies have found increased leukocyte blood counts in smokers compared with non-smokers (Nobel CN and Penny BB, 1975; Schwartz and Weiss S, 1994). Smoking influences the total number of lymphocytes and the relative distribution of the different lymphocyte subtypes, smoking may also impair lymphocyte function (Tollerud DJ *et al*, 1989; Berntorp K *et al*, 1989). Ludwig and Hoidal (1982) had shown that in cigarette smokers the PMN oxidative metabolism was altered as documented by the increased superoxide production by neutrophil. This study was undertaken in order to find out the effects of cigarette smoking on oxidative metabolism of neutrophils, serum lipid status and blood leukocyte counts.

Methods and Materials

Subject Selection :- For this study, 30 healthy nonsmokers and 30 chronic smokers were selected as the subjects. All the subjects were from same region and same economic group. They are represented in Table 1.

Group	Sample size	Age (years)	Height (cm)	Weight (kg)	No. of cigarette smoked/ day	Duration of smoking (years)
Nonsmokers	30	29.3 ± 3.43	163.2 ± 6.32	61.8 ± 3.52	0	0
Smokers	30	31.4 ± 2.45	161.0 ± 5.24	60.3 ± 4.34	11.2 ± 1.52	9.5 ± 1.35

Table 1 : Subject Characteristics

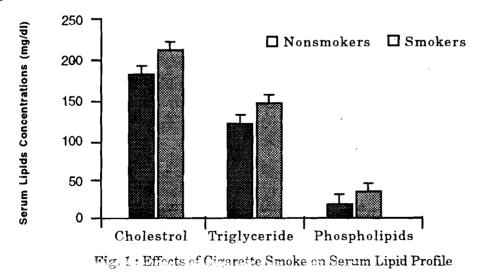
Blood Collection : – The blood samples from the individuals of the control and experimental groups were drawn from the anterior cubital vein. Blood sample from each individual was then divided into two, one was mixed with heparin to prevent clotting, used for the isolation of neutrophils and other hematological experiments and another was used for the preparation of serum, which was used for the biochemical assay.

Experimental procedure :- For biochemical assay, total cholesterol, triglyceride, phospholipids (Biochemical Kits) and lipid peroxidation (Yagi K, 1992), were measured. Total leukocyte blood counts were measured with hemocytometer and leukocyte differential blood counts were determined by counting 200 cells in a smear stained with May-Grunewald-Giemsa solution. Neutrophils were isolated by the method described by Clark RA and Nauseef WM (1996). The respiratory burst response of neutrophils was measured by the Nitroblue Tetrazolium (NBT) test (Park BH *et al*, 1968). The statistical analysis was done by two-tailed t-test (Das D and Das A, 1993).

Chemicals :- Phorbal myristate acetate (PMA), nitroblue tetrazolium (NBT) and dextran were purchased from Sigma Chemical Co. (St. Louis, MO). Ficoll-Hypaque was purchased from Pharmacia (Finland). All other chemicals were of analytical grade and purchased from SRL (India). The biochemical kits were purchased from DiaSys International (Delhi, India).

Results

Effects of Smoking on Serum Lipid profile :- It was seen from the present study that cigarette smoking led to alteration of serum lipid profile (Figure 1). Serum total cholesterol level was found to be significantly higher in smokers than nonsmokers (P<0.001). Cigarette smoking also caused elevation of serum triglycerides (P<0.001). It was also noted that smokers had higher serum phospholipids than nonsmokers (P<0.001). So, smokers tend to have higher serum lipids.



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Effects of Smoking on Blood Leukocyte Counts :- The geometric means of total leukocyte, neutrophil, lymphocyte, monocyte and basophil blood counts in smokers and non-smokers are given in table 2. Besides lymphocytes, all cell counts were higher in smokers than in non-smokers.

Effects of Smoking on Neutrophil Respiratory Burst Response :- Cigarette smoking caused significant increment of the oxidative metabolism of neutrophils (P<0.01), as evidenced by the increased respiratory burst response (Figure 2).

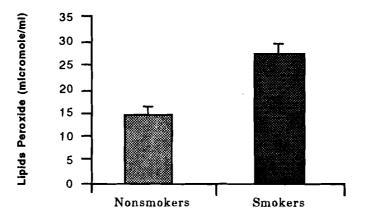


Fig. 1 : Effects of Cigarette Smoking on Serum Lipid Peroxide level

Effects of Smoking on Oxidative Lipid Damage :- Serum lipid peroxide level was found to be higher in smokers (P<0.001) than in nonsmokers (Figure 2).

Discussion

Relationship of Cigarette Smoking with Serum Lipids :- In this present study it was noted that chronic smoking resulted in elevation of serum total cholesterol level, which is the supportive evidence for other workers (Sina *et al*, 1995; Craig WY *et al*, 1989). Umeda *et al*, (1998) reported that serum total cholesterol was increased in direct proportion with increased cigarette smoking.

In this study, the mean serum triglyceride level was significantly higher in smokers than in nonsmokers. The elevation of serum triglycerides due to smoking was reported previously by several workers (Imamura *et al*, 1996; Caraig WY *et al*, 1989). There is a dose-dependent positive relationship between cigarette smoking and triglyceride (Handa K *et al*, 1990; Brischetto CS *et al*, 1983). Prolonged exposure of rats to cigarette smoke resulted in significant alterations in the

metabolism of lipids. There was a significant increase in the concentration of cholesterol, triglycerides and phospholipids in most of the tissues (Latha MS *et al*, 1988).

Although the mechanism by which smoke changes serum lipids levels is not fully understood, some possible explanations have been proposed by Brischetto *et al* (1983). In brief, the nicotine of cigarette smoke stimulates the secretion of catecholamines as well as other hormones such as cortisol and growth hormone, leading to an increased serum concentration of free fatty acids, which stimulates hepatic secretion of phospholipids, triglycerides, cholesterol and lipoproteins (Brischetto CS *et al*, 1983). There was increased cholesterogenesis in the important organs, like heart, lungs and liver, as evidenced by increased activity of HMG-CoA reductase and increased incorporation of labeled acetate into cholesterol. Incorporation of label into the triglycerides also increased in these tissues. Activity of lipoprotein lipase in the extrahepatic tissues was decreased. There was decreased concentration of bile acids in the liver, signifying the lowering of cholesterol degradation (Latha MS *et al*, 1988).

Relationship of Cigarette Smoking with Blood Leukocytes and Respiratory Burst of Nutrophils :- From this present investigation, it was evidenced that chronic cigarette smoking increased the blood leukocyte counts and leukocyte subsets, except lymphocyte. Jensen *et al*, (1998) found that both smoking and nicotine elevated the blood leukocyte counts and leukocyte subsets. In cigarette smokers total leukocyte, neutrophil and lymphocyte blood counts showed a dose dependent relationship with the daily cigarette consumption. After quitting smoking total leukocyte, neutrophil and lymphocyte blood counts decreased during the first 26 weeks (Jensen EJ *et al*, 1998). We confirmed previous reports of a substantial increase in neutrophils in smokers compared with non-smokers (Gillum RF, 1991; Bridges RB *et al*, 1985). There have been reports of both an increase and a decrease in the number of basophils in the blood of smokers compared to nonsmokers (Taylor RG *et al*, 1985 ; Walter S, 1982).

Relationship of Cigarette Smoke Induced Oxidative Burst of Neutrophils and Lipid Damage: In this study, the respiratory burst response of neutrophils was found to be significantly more in cigarette smokers than in nonsmokers, as evidenced by NBT reduction test. Cigarette smoke induced increased oxidative metabolism of neutrophils was reported previously (Anderson R *et al*, 1974; Ludwig PW and Hoidal JR, 1982). Sharma RN *et al* (1997) also found that the neutrophils from smokers had significantly more oxidative burst response than nonsmokers. This increased in oxidative burst of neutrophils is linked with the formation of

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superoxide anion (O_2^0) . This superoxide anion later leads to the formation of other types of ROI (Leto TL, 1999). In smokers the enhanced oxidant generation by neutrophils may be due to the action of nicotine, as both *in vivo* and *ex vivo* nicotine mimicked the ability of tobacco smoke exposure to potentate PMN release of superoxide (Gillespie MN *et al*, 1987; Jay M *et al*, 1986). As these ROI are proxidants in nature, they can able to oxidize cellular macromolecules, thereby causing tissue or cell damage and invites oxidative stress (Bandyopadhyay U *et al*, 1999). One of the well known consequences of ROI induced damage of cellular components are lipid peroxidation (Halliwel B and Gutteridge JMC, 1984). In this study, it was found that in smokers the serum lipid peroxidation was more than nonsmokers. These increased rates of lipid peroxidation in smokers are the supportive evidence for free radical damage to lipids, at least in part (Miller ER *et al*, 1997).

In conclusion, cigarette smoke or nicotine stimulates the number and oxidant generating activities of phagocytes, especially of neutrophils, which induces damages to lipids. Moreover, cigarette smoke also alters the serum lipid status.

Types of Blood Cells	Nonsmokers (Control)	Smokers (Experimental)		
Total Leukocytes	$6.8 \times 10^3 \pm 88.40$	$8.169 \times 10^3 \pm 124.18$ *		
Neutrophils	$4.15 \times 10^3 \pm 93.76$	$5.82 \times 10^3 \pm 100.75$ *		
Lymphocytes	$1.76 \times 10^3 \pm 64.08$	$1.63 \times 10^3 \pm 78.17$		
Monocytes	$0.476 \times 10^3 \pm 47.73$	$0.572 \times 10^3 \pm 64.58$		
Eosinophils	$0.245 \times 10^3 \pm 43.39$	$0.272 \times 10^3 \pm 58.91$		
Basophils	$0.068 \times 10^3 \pm 2.52$	$0.081 \times 10^3 \pm 3.38$ #		

Table 2 : Total leukocyte, neutrophil, lymphocyte, and basophil blood counts (Counts / μ L) of non-smokers and smokers.

Data Mean \pm SEM

* = P < 0.001

$$# = P < 0.02$$

*, # indicates significant difference between two groups

References

- Anderson R (1997). Antioxidant nutrients and prevention of oxidant mediated, smoking related diseases. In *Preventive Nurition* : The Comprehensive Guide for Health professionals. Ed. Bendich A and Deckelbaum RJ. Humana Press Inc., Totowa, NJ. 303 - 315.
- Anderson R, Rabson AR, Sher R and Koornhof HJ (1974). The NBT test in cigarette smokers. *Am J Clin Pathol.* 61 : 879.
- Bandyopadhyay U, Das D and Banerjee RK (1999). Reactive oxygen species : Oxidative damage and pathogenesis. Curr Sci. 77(5): 658 - 666.
- Banerjee K. K., Marimuthu P, Sarkar A, Chaudhuri R. N. (1998). Influence of cigarette smoking on Vitamin C, glutathione and lipid peroxidation status. *Indian J Public Health.* 42(1): 20 - 23.
- Berntorp K, Ekman M, Berntorp E (1989). Cigarette smoke impairment of human lymphocyte function by inhibition of transglutaminase. J Int Med 226 : 73 - 79.
- Bridges R. B, Wyatt R. J, Rhm S. R. Effect of smoking on peripheral blood leukocytes and serum antiproteases. *Eur J Respir Dis.* 1985; 66 : 24 - 33.
- Brischetto C. S, Connor W. E., Connor S. L. and Matarazzo J. D. (1983). Plasma lipid and lipoprotein profiles of cigarette smokers from randomly selected families : enhancement of hyperlipidemia and depression of high-density lipoprotein. Am J Cardiol. 52(7): 675 - 680.
- Clark R. A. and Nauseef W. M. (1996). Preparation and functional analysis of other human and lymphoid and non lymphoid cells. In *Current Protocols in immunology*. Vol. 2 Ed.
- Cohgan J. E., Kruisbeek A. M., Margulies D. H., Shevach E. M. and Strober W. Greene Pub. Associates and wiley Interscience. New York. 7.23.1.
- Craig W. Y., Palomaki G. E. and Haddow J. E. (1989). Cigarette smoking and serum lipid and lipoprotein concentrations : An analysis of published data. *BMJ* 298 : 784 - 788.
- Das D. and Das A. (1993). Tesing of Hypothesis. In Statistics in Biology and Psychology. Academic Publishers, Calcutta, India. 87 111.

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- Fisher S. D., Zareba W., Moss A. J., Marder V. J., Sparks C. E., Hochman J., Liang C., Krone J. R. (2000). Effect of smoking on lipid and thrombogenic factors two months after acute myocardial infection. Am J Carkiol. 86(8): 813 - 818.
- Gillespie M. N., Owasoyo, Kojima S. and Jay M. (1987). Enhanced chemotaxis and superoxide anion production by polymorphonuclear leuckocytes from nicotine – treated and smoke – exposed rats. *Toxicology*. 45(1): 45 - 52.
- Gillum R. F. (1991). Peripheral leukocyte count and pulmonary function in a national sample of women and men Ann Epidemiol. 1 : 525 531.
- Gurdal Y. E., Karadag M., Ozyandimci N., Kunt A. F. U. and Yarkin T. (1996). Cigarette smoking, serum lipids, folate and vitamin B12 in lung cancer. J Environ Pathl Toxicol Oncol. 15(2-4): 161 - 167.
- Halliwell B and Gutteridge J. M. C. (1984). Lipid peroxidation, oxygen radical, cell damage and antioxidant therapy. *Lancet.* 1 : 1396 1397.
- Handa K., Tanaka H., Shindo M., Kono S., Sasaki J and Arakawa K. (1990). Relationship of cigarette smoking to blood pressure and serum lipids. *Atherosclerosis* 84 : 189 - 193.
- Imamura H., Tanaka K., Hirae C et al (1996). Relationship of cigarette smoking to blood pressure and serum lipids and lipoproteins in men. Clin Exp Pharm Physiol. 23: 397 - 402.
- Jay M, Kojima S and Gillespie M. N. (1986). Nicotine potentiates superoxide anion generation by human neutrophil. *Toxicol Appl Pharmacol.* 86(3): 484 487.
- Jensen E. J., Pedersen B., Frederiksen R., Dahl R. (1998). Prospective study on the effect of smoking and nicotine substitution on leukocyte blood counts and relation between blood leukocytes and lung function. Thorax. 53 : 784-789.
- Latha M. S., Vijayammal P. L., Kurup P. A., (1988). Effect of exposure of rats to cigarette smoke on the metabolism of lipids. *Antherosclerosis*. 70(3): 225-231.
- Leto T. L. (1999). The respiratory burst oxidase. In *Inflammation Basic Principal* and Clinical Correlates (JI Gallin and R Snyderman, eds), Philadelphia, P. A., Lippincott Williams and Wilkins, 769 - 787.
- Ludwig P. W. and Hoidal J. R. (1982). Alterations in leukocytes oxidative metabolism in cigarette smokers. Am Rev Resp Dis. 126: 977 - 980.

- Miller E. R., Apple L. J., Jiang L and T. H. (1997). Association between cigarette smoking and lipid peroxidation in a controlled feeding study. *Circulation* 96 : 1097 - 1101.
- Noble C. N., Penny B. B. (1975). Comparison of leukocyte count and function in smoking and non-smoking young men. *Infect Immun.* 12:550-555.
- Park B. H., Fikrig S. M., and Smithwick E. M. (1968). Infection and nitroblue tetrazolium reduction by neutrophils : a diagonistic and *Lancet* ii : 532 534.
- Schwartz J., Weiss S. (1994). Cigarette smoking and peripheral blood leukocyte differential. Ann Epidemiol. 4: 236 242.
- Sharma R. N., Deva C., Behera D and Khanduja K (1997). Reactive oxygen species formation in peripheral blood neutrophils in different types of smokers. *Indian J Med Res.* 106: 475 - 480.
- Sinha A. K., Misra G. C., and Patel D. K. (1995). Effect of cigarette smoking on lipid profile in the young. J Assoc Physicians India. 43(3): 185 188.
- Taylor R. G., Gross E., Joyce H., et al. (1985). Smoking, allergy, and the differential white blood cell count, *Thorax.* 40 : 9 16.
- Tollerud D. J., Clark J. W., Brown L. M., et al. (1989). The effects of cigarette smoking on T cell subsets. Am Rev Respir Dis. 139 : 1446 - 1451.
- Umeda T., Kono S., Samurai Y, et al (1998). Relationship of cigarette smoking, alcohol use, recreational exercise and obesity with serum lipid atherogenicity : a study of self - defense officials in Japan. J Epidemiol. 8(4) : 227 - 234.
- Walter S (1982). Blood basophil counts in smokers and nonsmokers. Indian J Med Res. 72: 317 319.
- Wynder E. L., and Hoffmann D. (1979). Tobacco and health : a social challenge. N Eng J Med. 300 : 894 - 903.
- Yagi K (1992). Assay for serum lipid peroxide level and its clinical significance. In Lipid Peroxides in Biology and Medicine. Ed. Yagi K. Academic Press; New Yourk. USA.

A COMPARATIVE ANALYSIS OF ACACIA AURICULIFORMIS A. CUNN. EX BENTH. AND ACACIA MANGIUM WILLD. WITH REFERENCE TO THEIR CHROMOSOMAL CHARACTERS, SEED PROTEIN CONTENT AND MICROMORPHOLGICAL FEATURES OF SEED SURFACE

Asis Kumar Nandi and Amiyangshu Chatterjee *

Department of Botany and Forestry, Vidyasagar University, Midnapore * Department of Botany, Calcutta University, Kolkata

ABSTRACT

Two Australian species of *Acacia* Miller. (*A. auriculiformis* A. Cunn. ex Benth. and *A. mangium* Willd.) show a good length of resemblance and form as natural as well as artificial hybrids. In this paper the relatedness and variation in two species in terms of chromosomal characters, seed protein features and seed surface ornamentation have been explored. Two species show closeness in respect of diploid chromosome number, total chromosomal length, total chromosomal volume, range of individual chromosomal length, percentage of buffer soluble seed protein content and gross morphology of seed surface, while differ in detail in chromosomal morphology, seed protein profile and vivid ornamentation of seed surface.

Acacia Miller. is the largest mimosoid genus and the second largest in the family Leguminosae. Its two Australian members A. auriculiformis A. Cunn. ex Benth. and A. mangium Willd. are fast growing and are of considerable economic importance. Both are medium sized trees of a height up to 15 m. and resemble in their gross habit, angular outline of branchlet, pinnate leaves, rachis modified to phyllode, flowers arranged in spike, seed characters and dried funicle attached to seed. In having such a magnitude of resemblance they can naturally develop hybrids, which are reported to grow faster than either of two parental species (Tropical Legumes, National Academy of Sciences, Washington, 1979, Sedgley et al 1992 c. Le Dinhkha, 2000.) and so also facilitate to produce such hybrids artificially, techniques for which are being developed (Sedgley et al 1992 a, b,). As a result of better performance of some hybrids at some localities, natural hybridization became a menace for maintaining and identifying the purity of the species. So the studies on different aspects like morphological, anatomical, cytological, biochemical, and genetical features are required to characterize two species properly, the knowledge of which will ultimately help identify true species, their hybrids, and the nature of hybrids as well. Two species show finer difference in morphology, in growth behaviour, and seed as well as seedling characteristics (Hedge, 2000). In this paper a brief account of chromosomal, protein profile and seed micromorphological features of A. auriculiformis and A. mangium has been presented. The study is aimed to characterizing two species at different levels.

Materials and Methods

Seeds of A. auriculiformis A. Cunn. ex Benth. and A mangium Willd. have been taken for the chromosomal, biochemical and seed surface ornamentation study.

Squash technique has been adopted for studying chromosome. Cytological preparation for the study has been made from root tip cells stained with orcein.

Quantitation of seed protein has been made following Lowry *et al* (1951) and qualitative analysis of protein profile has been done by SDS-polyacrylamide gel electrophoresis.

Micromorphological study of seed surface has been carried out under Scanning electron microscope.

Observation

Chromosomal features : Both species show diploid chromosome number to be 26. The range of chromosomal length in *A. auriculiformis* is 1.643 μ m — 3.119 μ m and in *A. mangium* it is 1.667 μ m — 3.095 μ m. The total chromosomal length in the former is 60.334 ± 2.55 μ m., in the latter it is 56.288 ± 4.22 μ m. and the total chromosomal volumes are 30.862 ± 1.818 cu. μ m. and 35.701 ± 3.079 cu. μ m respectively. Number of chromosome pairs with secondary constriction is 3 in *A. auriculiformis* and 4 in *A. mangium*. No chromosome is morphologically identical in two species.

Seed protein contents : The percentage of buffer soluble seed protein is 22.805 ± 0.464 in *A. auriculiformis and* 21.132 ± 0.169 in *A. mangium*. The average amount of protein per seed is 5.631 ± 0.246 mg. in *A. auriculiformis* and 2.389 ± 0.338 mg. in *A. mangium*.

Qualitative analysis through SDS polyacrylamide gel electrophoresis has shown total number of 30 protein bands in *A. auriculiformis* and 46 in *A. mangium*. While cathodic and anodic bands are 18 and 12 in number, respectively, in the former these are 28 and 18 in the latter. The range of molecular weight of polypeptide bands in *A. auriculiformis* is 12 kd. – 1050 kd. and in *A. mangium* it is 11.798 kd. – 1050 kd. *A. auriculiformis* shows 7 marker bands and 10 such bands are present in *A. mangium*, however, no marker band is common in them.

Micromorphological peculiarities of the seed surface : Seeds of both species under SEM show a horse-shoe shaped open fracture line – pleurogram, on both flattened surfaces. The open ends are directed towards the mocropylar end. The rest part of seed surface, excepting the micropylar zone, is heavily fractured forming

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small polygonal areas with entire margin. However, a detailed study under a higher magnification (x 3200) reveals no worthwhole ornamentation in *A. auriculiformis*, instead shows only innumerable meager blocks of elevated polygonal areas; whereas in *A. mangium* a regular reticulate array of rugose ornamentation in a fashion of polygonal areas is evident under higher resolution. Reticulation represents the primary level of ornamentation with prominent rugose structures and the secondary level of ornamentation is formed with irregularly arranged finer rugose, encircled by each polygonal area.

Discussion

The reports of natural hybridization as well as the ease of making of artificial hybridization indicate enough reproductive compatibility between two species, which in turn also signifies their instinct relatedness. Notwithstanding such consanguinity two species show considerable variation at different levels of expressions. The present study shows resemblance between two species in their diploid chromosomal complements in having same diploid number of chromosomes -26, similar range of individual length of chromosomes, similar total chromosomal length and total chromosomal volume, percentage of buffer soluble seed protein and gross features of seed surface, like presence of pleurogram, fractured apprearance, straight nature of fracture lines. Contrary to this, in these two species the number of secondary constriction bearing chromosomes and the detailed morphology of chromosome vary, the average amount of protein per seed differs widely as does the average weight of seeds. Seed protein profiles of two species show least resemblance in regard of the whole spectrum of bands as well as the marker ones. The detailed ornamentation of seed surface even shows marked variation in two species. The overall resemblance of A. auriculiformis and A. mangium expresses the closeness of two species, but still then, variations in detailed characters at different levels of expressions justify the creation of two species, help identify them and enable us to pursue change that may appear in the hybrids of them.

REFERENCES

- Hedge, R., (2000). Variation in seed and seedling characteristics of Acacia mangium Willd. and Acacia auriculiformis A. Cunn. ex Benth. Indian Forester 126 (4) : 382-388.
- Le Dinhkha, (2000). Studies on natural hybrids of Acacia mangium and Acaia auriculiformis in Vietnam. Journal of Tropical Forest Science 12(4): 794-803
- Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., (1951). Protein measurement with Folin phenol reagent. *Journal of Biological Chemistry* 193: 265-275.
- Sedgley, M., Harbard, J. and Smith, R. R. M., (1992a). Hybridisation techniques for Acacias. ACIAR Technical Report 20.
- Sedgley, M., Harbard, J., Smith, R. R. M. and Wickneswari, R., (1992b). Development of hybridization techniques for Acacia mangium and Acacia auriculiformis. ACIAR Proceedings 37: 63 - 69.
- Sedgley, M., Harbard, J., Smith, R. R. M., Wickneswari, R., and Griffin, A. R. (1992c) Reporductive biology and interspecific hybridization of Acacia mangium and Acacia auriculiformis (Leguminosae : Mimosoideae). Australian Journal of Botany 40 : 37-48.
- Tropical Legumes : Resources for the future, 1979 National Academy of Sciences. Washington D. C.

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SCREENING AND ISOLATION OF INDIGENOUS VESICULAR ARBUSCULAR MYCORRHIZAL FUNGI FROM LATERITIC SOIL OF MEDINIPUR

Sudhansu Samanta^{*}, Somdatta Ghosh, N. K. Verma.

Mycorrhizae Laboratory, Department of Botany & Forestry, Vidyasagar University, Medinipur-721 102, West Bengal, India.

ABSTRACT

The work described in this paper deals with the isolation and identification of three indigenous VAM fungi. Acaulospora dilatata, Glomus fasciculatum, Glomus occultum has been identified. Spores have been isolated and inoculated in Sorghum plants for pureculture, which is being maintained in net house and mycorrhizae laboratory of Vidyasagar University.

KEY WORDS: Vesicular arbuscular mycorrhizae, isolation, identification, lateritic soil.

Introduction

VAM are of special significance in low fertility soil (Hatch 1937) where it increases nutrient absorption. Plant grown under phosphorus deficient soil such as acid lateritic soil, have greater dependence on mycorrhizae (Baylis 1972). Vesicular arbuscular mycorrhizae (VAM) act as a bridge between the host and the soil (Jackson et al, 1972) and benefit the plant by procuring P and other nutrients from beyond nutrient depletion zone (Sanders and Tinker 1973, Liu et al 2000).

Mycorrhizae reaches its extensive hyphal system beyond the depletion zone of plant roots and as an obligate symbiont help in absorption and translocation of phosphate and other nutrients. (Sander and Tinker 1973, Jackobsen et al 1992). Besides nutrients they also provide cross protection from disease and better moisture content through hyphal system in rhizospheric soils of plantation. (Baath and Hayman 1983, Dehne 1982, Krishna and Bagyaraj 1983).

It is because of this significance the present work was done to isolate and identify the local VAM spores occurring in the red lateritic soil of this region.

^{*} E-mail- sudhsnsusamanta@yahoo.com

Materials and Methods

Rhizospheric soils were collected from a depth of about 5-10 cm. Minimum of five soil samples were collected from the selected site with roots attachment.

The soil collected in sterile polybags, sealed and brought to the laboratory. The soil samples were air dried and stored until processed.

Isolation and quantitative estimation of VAM fungi was done by "wet sieving and decanting" method (Gerdemann and Nicholson 1963).VAM fungal spores were identified up to species level with the help of standard literature (Schenck and Perez 1987)). Identification and related studies was conducted in pots and fields in the net house and experimental cum botanic garden of the Vidyasagar University. The isolated spores were source of inocula for pure pot culture on Sorghum plants.

Results and Discussion

The following species were identified on the basis of characteristics described in the manual for the identification of VA mycorrhizal fungi by Schenck and Perez (1987) and INVAM (www. INVAM.com) voucher specimens with descriptions.

Acaulospora dilatata. Morton.

Sporocarp unknown. Spores formed singly in the soil, borne laterally on hyphae, each ending in a globose to sub globose hyphal terminus $105-142 \mu m$ diam; contents of terminus sub hyaline to yellow, emptying during spore formation and then collapsing; terminus readily detaching from immature as well as mature spores during extraction from soil. Hyphae at point of attachment 12-25 μm diam, with the distance between spore and terminus 45-49 μm . Spores yellow when examined under reflected light, lighter in transmitted light, mostly globose to sub globose, (78)-106-(130) μm diam. Spore wall structure consisting of 5 walls in 3 groups.

Glomus fasciculatum. (Thaxter sensu Gerdemann) Gerdemann and Trappe emend. Walker and Koske.

Spore-masses spongy, rounded or somewhat elongate or irregular loosely coherent groups, somewhat amorphous. Whole spores colour: pale yellow to pale yellow-brown, shape: globose, subglobose. Size distribution: 60-110 μ m. Structure of subtending hypha: shape: cylindrical to slightly flared. Width: 8-10.2 μ m, composite wall thickness: 1-10 μ m. Hyphal wall structure: two layers are clearly discernible. The spore wall also may continue into the hypha, but more often appears to curve and form a septum-like structure.

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Glomus occultum. (Wallker)

Sporocarps unknown, Chlamydspores borne singly or in loose cluster in the soil, or in compact clusters in the cortex of the roots. Whole spores: Colour: hyaline to pale cream or white. Shape: Globose, subglobose, often slightly irregular, Size distribution: 15-(65)-100 X 20-(65)-100 μ m, mean = 71.5 μ m. Spore Wall : 1 to 2 layered with an additional rough outer deposit of granular materials which sloughs with age. Outer wall when present less than 1 μ m thick, often indistinct. Inner wall I.5 - 2.5 (.5) μ m thick, usually of two sometimes indistinct laminations. Structure of subtending hyphae : straight, simple, 5-50 μ m long, 3-10 μ m wide at spore base, tapering to 2-5 μ m; attached axially or accentrically and re-curved to straight, sometimes closed distally by a septum. Shape: Cylindrical to slightly flared. Width 3.0-10. μ m (mean = 4.1 μ m). The hyphal wall tapers to less than 0.2 μ m at a distance of 5 μ m or more from the spore and becomes difficult to see. NOTES: Spores are nearly transparent and difficult to locate under stereo-microscopes, spores resemblance sands. Spores often float on the surface of sucrose following density gradient centrifugation and also float in water with swirling.

The isolated three VAM strains are native to red-lateritic soil of Medinipur and therefore they are adapted to local agro-climatic condition. Their propagation on large scale for inoculation to locally grown agricultural crops has enough potential in substituting the high cost phosphate fertilizer with added advantage of being eco-friendly. Being low cost it may be accepted by marginal farmers which will be a great help to the poor community.

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Reference

- Baath E., Hayman D.S. 1983. Plant growth response to vesicular-arbuscular mycorrhiza. XIV. Interactions with verticillum wilt on tomato plants. New phytol. 95 : 419-426.
- Baylis G.T.S. 1972. Minimum levels of available phosphorus for non mycorrhizal plants. *Plant Soil.* **36** : 233-234.
- Dehne H.W. 1982. Interaction between vesicular- arbuscular mycorrhizal fungi in plant pathogens. *Phytopathol.* 72 : 1115-1119.
- Gerdemann J.W., Nicolson T.H. 1963. Spores of mycorrhizal endogone species extracted from soil by wet sieving and decanting. *Trans. Brit. Mycol. Soc.* **46**: 235-244.
- Hatch A.B. 1937. The physical basis of mycotrophy in the genus Pinus. *Black rock* For. Bull. **6** : 168.
- Jackson: N.E. Franklin R.E. Millar R.H. 1972. Effects of VAM on growth and phosphorus content of agronomic crops. *Proc. Soil Sci. SOC Am.* **36** : 64.
- Jacobsen I., Abbott L.K., Robson A.D. 1992 External hyphae of VAMF associated with *Trifolium subterraneum* L.I. Spread of hyphae and P inflow into roots. *New Phytol.* 120(3): 371 - 379.
- Krishna K.R., Bagyaraj D.J. 1983. Interaction between Glomus fasciculatum and Sclerotium rolfsii in peanut, Arachis hypogaea. Can. J. Bot. **61** : 2349-2351.
- Sanders F.E., Tinker P.B. 1973. Phosphate flow into mycorrhizal roots. *Pestic. Sci.* **4**: 385-395.
- Schenck N.C; Perez. Y. 1987. Manual for the identification of VA mycorrhizal fungi. INVAM. 1456, Fifield hall University of Florida Gainesvill, Florida.
- Liu A., Hamel C., Hamilton R.I., Ma B.L., Smith D.L. 2000. Acquisition of Cu, Zn, Mn, and Fe by mycorrhizal maize (*Zea mays* .L.) grew in soil at different P and micronutrient levels. *Mycorrhiza*. **9** (6) : 331-336.

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BIOLOGICAL SPECTRUM OF THE VEGETATION IN THE CAMPUS OF VIDYASAGAR UNIVERSITY, MIDNAPORE, WEST BENGAL

* Debabrata Das, * R. B. Ghosh, and T. K. Mishra

Raja N. L. K. Women's College, Midnapore - 721102 (W.B.) * Botanical Sruvey of India, Howrah - 711103 (W.B.)

ABSTRACT

Life forms of the vegetation and billogical spectrum of the flora of the campus of Vidyasagar University have been determined. The comparison with Normal Spectrum shows Therophytic climate, which is contrary to Raunkiaer's hypothesis. This is partly because of plantation activities and other processes of restoration of ecosystem by human activities.

KEYWORDS: Vidyasagar University, Biological Spectrum, Life Form, Therophytes.

Introduction

The classification of vegetation is based on the appearance or general nature of plants indicated as "Growth Forms" or "Life forms" or "Vegetative forms" (Osting 1958). Of several such systems proposed earlier, the system of Raunkiaer (1934) is the most practical one from ecological point of view. His system is based mainly on only one feature *i.e.* adaptation of the plant to survive in unfavourable seasons especially with regard to the degree of protection of the perennating buds of shoot apices. These unfavourable seasons may encompass cold winter period in temperate or hot dry period in the sub-tropics and the tropics. Dombois D. M. and H. Ellenberg (1974) argued that the range of adaptations on the basis of life forms given by Raunkiaer also relate to the mode of shoot withdrawal in the unfavourable seasons. The most complete form of shoot reduction is represented by annuals, which are mostly therophytes and survive only in the form of seeds. Meher-Homii (1964 and 1981) compared the biological spectra of various regions of India and showed that these spectra were related to the bio-climate of the region. Theoretically, Raunkiaer defined the Life Forms as "the sum of the adaptation of the plants to the climate". His system gives definite names to almost every type of plant available.

Study Site

An initiative was taken for establishment of a new university in honour of a great educationist and reformer named Ishwar Chandra Vidyasagar, formerly Ishwar Chandra Bandopadhyaya, during late 70s. The State Government in 1975 took up the idea and the Vidyasagar University Act, 1981 was passed with the report and recommendation of the Bhabatosh Dutta Committee.

Vidyasagar University campus which is in the outskirts of the district town of Midnapore is situated on 22° 14.4" N latitude and 87° 17.5" East longitude. The soil was highly degraded previously but for the last two decades plantation and other processes have more or less reversed the process of degradation. However, the process of laterization is still seen to be in progress, though at a much lower rate than the unmanaged land adjacent to it. Soil is found to be sandy loam of brown to dark brown to reddish brown in colour in the upper layer. Morrum nodules or iron concretions or laterite pan are some times exposed. Soil is well drained due to coarse texture and weathering of soil along with its parent material is still in progress probably due to moderately heavy precipitation followed by prolonged dry season.

Raunkiaer (1934) classified the plants according to their habit of evercoming unfavourable season, thus making the system easily applicable. He believed that the flora of a given region is an exact indicator of its climate. He called the statistical concept of the forms of a flora *i.e.* the percentage distribution of a species within the life forms of various species of any flora as "Biological Spectrum" or "Phytoclimatic Spectrum". According to Raunkiaer, the bio-climate of the region is characterised by the life forms, which is the biological spectrum of the region.

Raunkiaer defined the Normal Spectrum as a particular ratio expressed in percentage of different life forms to all the phanerogams of the world. He grouped the spectra into four types viz. A, B, C, and D. 'A' for the high percentage of phanerogams, 'B' for Hemicryptophytes, 'C' for Therophytes and 'D' for Chamaephytes together with considerable proportion of Hemicryptophytes.

Vidyasagar University campus is in the district of Midnapore, which in its undivided form was considered as one of the biggest district in West Bengal as well as in the country. Studies on the district of Midnapore represent interesting diversity of vegetation due to its variable topography, soil pattern and climate. The campus of the University encompasses nearly 200 acres of land with feeble undulation covered by various taxa of weeds, shrubs, herbs and a good canopy of flowering trees of different families of Angiosperms (Das and Ghosh. 1999). The climate of the vast campus and its surrounding areas is dry tropical. The vegetation is minly deciduous. At some places of the vast campus semi-humid zone is noticeable on account of great shades cast by the flowering trees bearing deep green and thick canopy. Summer, winter and rainy season are moderate, although rain continues sometime up to middle of October. Annual rainfall is approximately 1500 mm and the maximum and minimum temperatures are 40.32°C and 12.24°C respectively. So, with the admixture of climate, the flowering taxa of different ranks perennate with buds at different levels of their trunks thereby manifesting different life forms in the vast stretch of the campus of the University.

Results and Discussion

Life forms of plant species recorded from the various selected topographies of the University campus and their comparison are drawn with Raunkiaer's normal spectrum value. (Table -1)

Table – 1 :

Sl. No.	Life form	Symbol	No. of Species	Life form spectrum at VU (%)	Raunkiaer's normal Spectrum (%)
1.	Phanerophytes	Ph	72	30.77	46.0
2.	Chaemophytes	Ch	24	10.27	9.0
3.	Hemicryptophytes	Н	3	01.20	27.0
4.	Geophytes	G	21	08.97	6.0
5.	Therophytes	Th	119	48.71	13.0

Life form in University campus and its comparison with Raunkiar's normal spectrum :

Biological spectrum is the percentage (percentage of the total species in a community) distribution of species among various life forms as suggested by Raunkiaer (1934). By calculating the percentage value of each life form biological spectrum of an area is obtained. The life forms of the plant species recorded from the various selected sites of the study area *i.e.* campus of the Vidyasagar University were determined and a biological spectrum was proposed following the widely accepted Raunkiaer's system. The life forms of the flora of the study area are given in the Table - 1, which shows the percentage distribution and number of species on the basis of Raunkiar's classification and comparison with Raunkiaer's normal spectrum value. The table presented above reveals that the therophytes were well represented in the areas and form the majority of the species *i.e.* 48.71% followed by Phanerophytes, Chaemophytes, Geophytes and Hemicryptophytes.

In the campus 239 species are found of which 138 are herbs, 43 are shrubs and 53 are trees. Within the plants species found 24 belong to the family Leguminosae, which are nitrogen fixer in nature. 35 species of the campus belong to the family Cyperaceac and Poaceac, which are soil binder in nature. Due to the presence of some species belonging to the above named families the soil formation process has become easier. This shows that there has been an enrichment of the whole phytoclimate of the campus and the vegetation is qualitatively richer.

The phytoclimate has been determined by comparing the biological spectrum of the study area with Raunkiaer's normal biological spectrum of phanerogamic flora of the world (Fig. 1) in order to find out whether the results of the present study support the Raunkiaer's hypothesis that the phytoclimate is an indicator of the vegetation of an area. The biological spectrum of the area *i.e.* University campus as compared to the Raunkiaer's normal spectrum shows that the percentage of Therophytes was nearly four tomes higher (48.71%) than in the normal spectrum (13%). The phanerophytes figure 30.77% and achieve that second position in the study area. However, its value goes lower than the normal value (46%). The percentage value of Chaemephytes in the area under study is 10.27%, which is more or less equal to the normal, value (9%). Geophytes showed a slight higher percentage (8.97%) as compared to normal spectrum value (6.0%). Hemicryptophytes shoed a very low value 1.20% against 27.0% in the normal spectrum. From the above stated observation, plant climate of the area under study may be considered as 'therophytic' with a high percentage of phanerophytes. This goes in contradiction to Raunkiaer's hypothesis and lends support to Beadle (1951) who advocates that climate is not always indicated by vegetation. In the present study, the biological spectrum seems to indicate the most operative factor of the environment, i.e. biotic interference and a prolonged draught period rather than the plant climate. Presence of more therophytes and geophytes is because of its semi-xeric condition of the environment in some parts of the year. The species of therophytes found in the campus, which are xerophytic in habit are Zizyphus numularius, Z. oenoplia, Z. jujuba, Capparis spinosa, Calotropis procera, C. Gigantia, Holarrhena pubescens, Delonix regia, Caesalpinia sappan etc. The higher percentage of therophytes is specifically because of intensive planatation in some selected sites of the University Campus. These plantation sites provide shelter for the therophytes during draught period.

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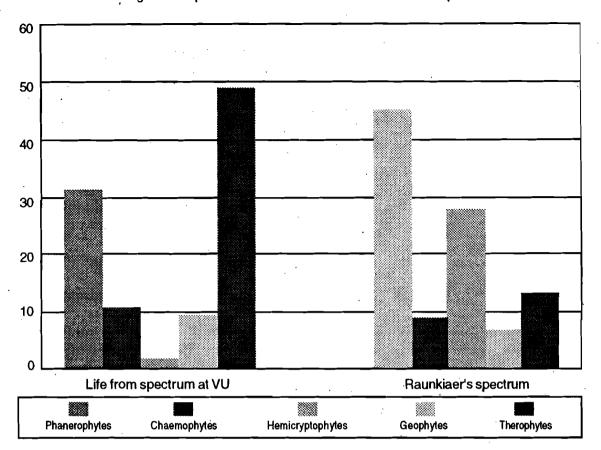


Fig. 1: Comparison between the Normal and observed spectra

REFERENCES

- Beadle, N. C. W. 1961 : The misuse of climate as an indicator of vegetation and soils. *Ecology* : 32 : 343-345.
- Das, Debabrata and R. B. Ghosh 1999: A Preliminary Survey and Taxonomic Census of Flowering trees of angiosperms of the campus of Vidyasagar University, Midnapore, and West Bengal, India. Indian J. Appl. & Pure Biol. Vol-14 (1) : 56-68.
- Dombois D. M. and H. Elenberg 1974 : Aims and methods of vegetation ecology, John Wiley and Sons, New York, USA.
- Meher Homji, V. M. 1964 : Life forms of biological spectra as epharmonic criteria of aridity and humidity in the Tropics. J. Indian. Bot. Soc. 43 : 424-430.
- Meher Homji, V. M. 1981 : Environmental implications of life form spectra from India. J. Econ. Tax. Bot. 2 : 23-30.
- Ousting, H. J. 1958: The study of Plant Communities. Freeman & Co. San Francisco; U.S.A.
- Raunkiaer. C. 1934 : the Life Forms of Plants and Statistical Plants Geography. Oxford University Press, London.

Short Communication

TRADITIONAL KNOWLEDGE FOR CATCHING FISH IN SOUTHERN WEST BENGAL

S. S. Mishra, P. Pradhan, S. K. Chakraborty and R. K. Bhakat*

:

Department of Zoology, Vidyasagar University, Midnapore West Bengal, 721 102, India *Department of Botany and Forestry, Vidyasagar University, Midnapore, West Bengal, 721 102, India

The three southern districts of West Bengal (Bankura, Hooghly and Midnapore) are spread over between 22-23°N latitude and 86-88.3°E longitude. These districts are located very close to Jharkhand and Orissa states on the west and the districts Howrah, North 24 Pargana and Burdwan of West Bengal on the other three sides. The whole area is a brilliant mix of diverse landscapes. The eastern and southern parts harbour highly fertile alluvial and coastal plains respectively. The western and northern portions consist of lateritic soils, densely covered with dry deciduous forests. These three districts are criss-crossed by six main rivers (Dwarkeswar, Kansai, Keleghai, Rupnarayan, Shilabati and Subarnarekha) which support a good number of seasonally and annually flooded riverine water bodies endowed with varied plant and fish species. A large number of rural fishing communities depend on these aquatic ecosystems for their livelihood. The local fishermen capture fishes from these waterlogged areas utilising their indigenous and traditional knowledge of locally available plants used as stupefying agents or agents for poisoning fishes.

Athough there are numerous studies on plant-based traditional knowledges to capture fishes from different parts of India (Chopra et al., 1956 and 1965; Bhat and Singh, 1985; Bhat and Dhyani, 1990; Bhat and Farswan, 1991; Jain, 1991), literature on such studies from West Bengal are few and scattered (Jain, 1991; Mishra et al., 2002). This communication is a rapid field study dealing with the fish-killing plants of three southern districts of West Bengal.

During the surveys conducted at seventeen (17) river based sites covering six rivers across the three districts, the local fishermen were interviewed and crossinterviewed. They were asked about their use of ichthyotoxic plants, plant-parts and methods adopted. The results thus obtained reveal use of various plant parts (leaf, leaf juice; bark; latex; fruit powder and cake; seed powder and whole crushed plant) of twelve (12) locally available species (Table 1). The selected plant parts are thrown into preferably shallow waterbodies and very slow-moving water. The fast-flowing streams are avoided since the risk of plant parts being swept away is greater in lotic systems. The impact of poisonous plant parts renders the target

fish perplexed and restricts its movement all on a sudden, finally forcing it to float on the water surface, thus facilitating its capture. Similar results were also reported by the present workers (Mishra et al, 2002).

The present work, being a preliminary study, provides only a clue to the rural people's indigenous knowledge of fishing. Therefore, it calls for a detailed study of this kind of folk knowledge involving more areas. Simultaneously, the nature of plant poisons involved in fish stupefication needs to be studied.

Study site	River System	Plants Used
Midnapore District		
Gokulpur	Kansai	Bl, Mo, Nt, Ph, Pp, Sn, Ti
Khirai	Kansai	Bl, Mo, Nt, Ph, Po, Pp, Sn, Tp, Vs
Midnapore	Kansai	Bl, Mo, Nt, Po, Sm, Ti, Tp
Dehati	Keleghai	Bl, Ea, Mo, Nt, Pp, Sn, Ti
Khakurda	Keleghai	Bl, Ea, Mo, Nt, Pp, Sn, Ti
Rohini	Subarnarekha	Mo, Ea, Nt, Po, Sn, Ti, Vs
Sonakonia	Subarnarekha	Mo, Ea, Nt, Po, Sn, Ti, Vs
Garbeta	Shilabati	Bl, Mo, Nt, Ph, Po, Sn, Ti, Tp
Ghatal	Shilabati	Bl, Mo, Nt, Ph, Sn, Vs
Bandar	Rupnarayan	Bl, Ea, Mo, Nt, Ph, Po, Sn
Kolaghat	Rupnarayan	Bl, Ea, Mo, Nt, Ph, Po, Sn
Bankura District		
Rajagram	Dwarkeswar	Bl, Mo, Nt, Ph, Ti
Kethardanga	Dwarkeswar	Bl, Mo, Nt, Ph, Ti, Po, Tp
Ekteswar	Dwarkeswar	Bl, Mo, Nt, Ph, Tp, Vs
Hooghly District		
Chandur	Dwarkeswar	Bl, Mo, Nt, Ph, Pp, Ti
Kalipur	Dwarkeswar	Bl, Mo, Nt, Ph, Po, Pp, Sm
Dingdubimore	Dwarkeswar	Bl, Mo, Nt, Po, Pp, Sm, Sn

Table 1: Study sites along six river systems of southern West Bengal showing use of fish-stupefying plants.

 $Bl = Bassia \ latifolia, Ea = Excoecaria \ agallocha, Mo = Moringa \ oleifera,$

Nt = Nicotiana tabacum, Ph = Polygonum hydropiper, Po = Polygonum orientale,

Pp = Pongamia pinnata, Sm = Sapindus mukorossi, Sn = Strychnos nux-vomica,

Ti = Tamarindus indica, Tp = Tephrosia purpurea, Vs = Vangueria spinosa

References

- Chopra, R. N., S. L. Nayer and I. C. Chopra. 1956. Glossary of Indian Medicinal Plants, CSIR, New Delhi.
- Chopra, R. N., M. L. Bhadwar and S. Ghosh. 1965. Poisonous Plants of India, Vol. 1, ICAR, New Delhi.
- Bhat, J. P. and H. R. Singh. 1985. Effect of *Engelhardia colebrookiana* (Lindle) on a freshwater teleost *Barilius bendelisis* (Ham.), *Sci. & Cult.* **51**: 132-133.
- and P. P. Dhyani. 1990. Lethality of piscicidal compounds of plant origin to some freshwater animals, J. Freshwater Biol. 2: 5-12.
- and Y. S. Farswan. 1991. Responses in fish to two piscicidal compounds of plants origin, *Ind. J. Animal Sciences* **61**: 336-341.
- Jain, S. K. 1991. Dictionary of Indian Folk Medicine and Ethnobotany, Deep Publications, New Delhi.
- Mishra, S. S., P. Pradhan, S. K. Chakraborty and R. K. Bhakat. 2002. Plants used for stupefying fish in Southern West Bengal, *Sci. & Cult.* (in press).

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