BRACHYURAN CRABS OF COCHIN BACKWATERS- DIVERSITY, HABITAT ECOLOGY AND SYSTEMATICS, WITH SPECIAL EMPHASIS TO GENUS SCYLLA

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in

Marine Biology Under the Faculty of Marine Sciences

Ву

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Brachyuran Crabs of Cochin Backwaters- Diversity, Habitat Ecology and Systematics, with special emphasis to Genus *Scylla*

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Eertificate

This is to certify that the thesis entitled " **Brachyuran Crabs of Cochin Backwaters- Diversity, Habitat Ecology and Systematics, with special emphasis to Genus** *Scylla*" is an authentic record of the research work carried out by Ms. P. Lakshmi Devi under my supervision and guidance in the Department of Marine Biology, Microbiology and Biochemistry, School of Marine Sciences, Cochin University of Science and Technology, in partial fulfillment of the requirements for the degree of **Doctor of Philosophy in Marine Biology** of Cochin University of Science and Technology, and no part thereof has been presented for the award of any other degree, diploma or associateship in any university. All the relevant corrections and modifications suggested by the audience during the pre-synopsis seminar and recommended by the Doctoral committee have been incorporated in the thesis.

Kochi - 682 016 24th February 2015 Dr. Aneykutty Joseph (Supervising Guide)

Declaration

I hereby declare that the thesis entitled "Brachyuran Crabs of Cochin Backwaters – Diversity, Habitat Ecology and Systematics, with special emphasis on Genus *Scylla*" is an authentic work carried out by me, under the supervision and guidance of Prof. (Dr.) Aneykutty Joseph, Department of Marine Biology, Microbiology and Biochemistry, School of Marine Sciences, Cochin University of Science and Technology for the partial fulfilment of the requirements for degree of Doctor of Philosophy in Marine Biology of Cochin University of Science and Technology, and no part thereof has been presented for the award of any other degree, diploma or associateship in any university.

Kochi - 682 016 24th February 2015 P Lakshmi Devi

Dedicated to

My Family for Their Love and Endless Support

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Preface

Among the decapod crustaceans, brachyuran crabs or the true crabs occupy a very significant position due to their ecological and economic value. Crabs support a sustenance fishery in India, even though their present status is not comparable to that of shrimps and lobsters. They are of great demand in the domestic market as well as in the foreign markets. In addition to this, brachyuran crabs are of great ecological importance. They form the conspicuous members of the mangrove ecosystems and play a significant role in detritus formation, nutrient recycling and dynamics of the ecosystem. Considering all these factors, crabs are often considered to be the keystone species of the mangrove ecosystem.

Though several works have been undertaken on brachyuran crabs world –wide as well as within the country, reports on the brachyuran crabs of Kerala waters are very scanty. Most of the studies done on brachyuran fauna were from the east coast of India and a very few works from the west coast.

Among the edible crabs, mud crabs belonging to genus *Scylla* forms the most important due to their large size and taste. They are being exported on a large scale to the foreign markets like Singapore, Malaysia and Hong Kong. Kerala is the biggest supplier of live mud crabs and Chennai is the major centre of live mud crab export. However, there exists considerable confusion regarding the identification of mud crabs because of the subtle morphological differences between the species.

In this context, an extensive study was undertaken on the brachyuran fauna of Cochin Backwaters, Kerala, India, to have a basic knowledge on their diversity, habitat preference and systematics. The study provides an attempt to resolve the confusion pertaining in the species identification of mud crabs belonging to Genus *Scylla*.

The Thesis is divided into three parts

Part. I comprises three general chapters.

Chapter 1: General Introduction gives a general account of the brachyuran crabs, rationale of the present study and the objectives set for the work.

Chapter 2: Review of Literature covers the previous works carried out by various authors in relation to various aspects of brachyuran crabs.

Chapter 3: Materials and Methods gives an account of the study area, Cochin backwaters and the sampling stations selected for the study. The chapter also explains in detail the methods and protocols followed during the study.

Part. II comprises four working chapters

Chapter 4: Diversity of Brachyuran crabs of Cochin Backwaters is a documentation of the brachyuran diversity of the Cochin backwaters and the associated mangrove patches.

Chapter 5: Habitat Ecology and Habitat Preferences of Brachyuran crabs of Cochin Backwaters is an attempt to describe the distribution as well as habitat preferences of the brachyuran crabs that are enlisted from the Cochin backwaters.

Chapter 6: Taxonomic account of Brachyuran crabs of Cochin Backwaters provides a systematic, figurative key for the identification of brachyuran crabs occurring in the Cochin backwaters.

Chapter 7: On the systematics of Genus *Scylla* **De Haan,1833 of Cochin Backwaters** is an attempt to describe the *Scylla* spp from the Cochin backwaters on the basis of morphological characters including the description of the male gonopods and the third maxillipeds, morphometry as well as molecular tools.

Part III comprises Salient findings of the study, Bibliography and Publications.



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Part - I

Chapter 1 GENERAL INTRODUCTION

Chapter 2 REVIEW OF LITERATURE

Chapter 3 MATERIALS AND METHODS

Chapter **1** GENERAL INTRODUCTION

India is bestowed with a rich fauna of edible crustaceans, several of them supporting the commercial fisheries. At present, around 150 species of this group form the major part of the commercial catches either on regular basis or as occasional inclusions (Suseelan, 1996). During the past 3-4 decades, exploitation of edible crustaceans along the Indian coast has increased, due to their sustainable demand in the international markets. The extension of fishing activities into deeper waters and the capturing of nonconventional species, have added up more number of species into the above faunistic list of commercially important crustaceans. Among them, brachyuran crabs form the most important group.

Brachyuran crabs or the true crabs are characterized by a hard exoskeleton and a reduced abdomen, which is most often referred to as its short tail, which is entirely hidden under the thorax. Hence, they got the name Brachyura, which means short-tailed. They are armed with a single pair of massive chelae, which is often used for prey capture, communication, mating, defense and offense. They vary in size from a few millimeters width as in the pea crabs to 4 meters leg span (13 feet) as in the Japanese spider crab.

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Crabs are adapted to survive in almost every habitat of earth, except the air and the polar ice caps. They are found to exist in various biotopes, such as marine, estuarine, fresh water, intertidal and even terrestrial habitats. They are even found to inhabit the abyssal zone, hydrothermal vents, river swamps, rain forests, mountain tops and the deserts. However, certain landscapes exhibit domination of certain group of crabs. For instance, mangrove habitats are usually dominated by Sesarmids and Ocypodids, the coral reefs by Xanthid crabs and their kin, the tropical forests by the terrestrial Gercarcinids, and the fresh water systems by Potamids and their allies (www.nio.org).

Crabs often show marked sexual dimorphism. The most conspicuous among the sexual dimorphism being the shape of the abdominal flap. In males, abdominal flap is narrow and triangular in shape, while in females it is broader, rounder abdomen, enabling them to brood the fertilized eggs on their pleopods or the abdominal appendages. In some group of brachyurans, males possess larger claws than the females. This type of sexual dimorphism is particularly pronounced in the fiddler crabs of genus *Uca* belonging to the family Ocypodidae. The male fiddler crabs possess unequal chelipeds, one of the chelae greatly enlarged into pincers, which they primarily use for communication. They exhibit waving displays to attract females towards them for courtship.

Crabs are generally omnivorous, feeding on both plant matter and animal matter depending on their availability. However, some groups of brachyura, for instance the mangrove crabs exhibit herbivory. They feed on the leaf litter from the mangroves. Some crabs like grapsid crabs are found to be detritivorous, feeding on the decaying plant matter present in the sediments (Ajmal Khan and Ravichandran, 2009).

Biologists find brachyurans as very interesting creatures, due to their typical and complex behavioral pattern, particular modes of life, association with other invertebrates etc. Crabs typically walk sideways, which can be attributed to the peculiar articulation of their legs, which makes a sidelong gait more efficient. However, some crabs including the Raninids are capable of forward or backward movement.

Usually, they are found to be aggressive towards each other and it is observed that the males often fight over the females. On rocky seashores, crabs also fight among themselves over hiding holes. Some crabs like the *Uca*, exhibit waving displays of chelate legs, some crabs can even produce sound, by stamping their feet or banging their pincers on the ground.

The brachyurans are also found to retain commensal association with many organisms including algae, sea weeds and invertebrates like sponges, coelenterates, barnacles, holothurians and bivalves.

Brachyuran crabs are of appreciable importance in the present scenario due to its economic and ecological value. Crabs support a sustenance fishery in India, even though its present status is not comparable to that of shrimps and lobsters. Annual landings of crabs in the Indian waters are estimated to be 13,000 tonnes (www.mpeda.com/mudcrabreports.pdf). The most important species contributing to the fishery are *Scylla serrata*, *S. olivacea*, *Portunus pelagicus*, *P. sanguinolentus*, and *Charybdis feriata*. Crabs are being exported

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to foreign countries on a large scale in the form of crab meat, cut crab as well as in live condition. Live mud crabs are exported to Singapore and Hong Kong, which fetch better prices than the swimming crabs belonging to *Portunus spp*. India earns about 18 million US\$ as foreign exchange from the export of live mud crabs (www.mpeda.com/mudcrabreports.pdf).

Crab meat is found to be very tasty and in nutritional point of view, it is rich in proteins, vitamins, glycogen and minerals. Crab meat is also believed to stimulate brain cells and also possess some medicinal value. It is believed to cure asthma and chronic fevers (Chopra, 1939; Chidambaram and Raman, 1944). Mud crab meat is effective in curing diarrhea and dysentery, while the swimming crabs *Portunus sanguinolentus* and *P. pelagicus* are usually served to people just recovered from malaria and typhoid (www.nio.org).

In addition to this, many small-sized intertidal crabs which are not of direct economic value are being used in the preparation of high energy yielding; cheaper artificial pellet feeds for the cultivation of edible varieties of sea food.

Crabs are the conspicuous members of the mangrove ecosystems (Verwey, 1930, Macnae, 1968) and are ecologically significant in many ways. Crabs have a significant role in detritus formation, nutrient recycling and dynamics of the ecosystem. It is found that their burrows alter the topography and grain size of the sediments (Warren and Underwood, 1986). Their digging behavior enhances aeration and facilitates drainage of mangrove soils (Ridd, 1996). They keep much of the energy within the mangrove forest by burying and consuming the leaf litter. Furthermore, their

faeces form the basis of coprophagous food chain, contributing to mangrove secondary production (Lee, 1997). Hence, crabs are often considered to be the keystone species in the mangroves (Holling, 1992) because of their role in carbon recycling (Schories *et al.*, 2003).

Brachyuran crabs play a very dominant role in the marine food web. These crabs and their larvae feed on lower organisms like algae, molluscs and other crustaceans and the crab larvae are in turn preyed upon by larger predators like omnivorous fishes. Thus they play a vital role in the transfer of energy through the food chain. Robertson *et al.* (1992) reports that crab larvae form the major food source for the juvenile fishes and the crabs themselves form the food for many carnivorous fishes and birds including many threatened species like the crab plover (Seys *et al.*, 1995).

It is quite surprising that despite of all these facts, this group lacks priority in our investigations and therefore there is an urgent necessity to initiate more works on this group.

Hence an extensive study was undertaken on the brachyuran fauna of the Cochin backwaters, Kerala, India, to have a basic knowledge on their diversity, habitat preferences and systematics. The study provides an attempt to resolve the confusion pertaining in the species identification of mud crabs belonging to Genus *Scylla*. The objectives set for the study were as follows.

Objectives of the study

- To study the diversity of brachyuran crabs of Cochin backwaters.
- To have a basic knowledge on the habitat ecology, habitat preferences, abundance and distribution of brachyuran crabs of Cochin backwaters.
- To provide a systematic account on the brachyuran crabs of Cochin backwaters
- Revalidation of the commercially important mud crabs of genus Scylla using molecular methods to resolve the problems of species ambiguity.

Though several works have been undertaken on brachyuran crabs world –wide as well as within the country, only little have been studied on the brachyuran crabs of Kerala waters. Most of the studies done on the brachyuran fauna were from the east coast of India and a very few works from the west coast. The earliest available record on brachyuran faunal studies from Kerala waters was that done by Pillai (1951) who dealt with the decapods of Travancore waters. Eventually, Antony and Kuttyamma (1971), Rao and Kathirvel (1972), Radhakrishnan and Samuel (1982), Devasia and Balakrishnan (1985), Thomas *et al.* (1987), Kurup *et al.* (1990), Anil (1997) dealt with the brachyuran fauna of the Cochin backwaters. Succeeding them, no further studies were initiated on the brachyuran fauna of Cochin backwaters for a long time till 2008. Roy and Nandi (2008) examined the brachyuran biodiversity of Vembanad Lake and stated that despite its high productive status, the lake support a low brachyuran diversity. They also pointed out the requirement of a season-wise survey to be conducted on the Vembanad Lake to assess the exact brachyuran diversity of the area. In this context, the present work was undertaken to study the diversity of brachyuran crabs in Cochin backwaters employing a month-wise survey for a period of two years from June 2010- May 2012.

Cochin backwaters is characterized by wide salinity gradient and varying habitat types, like low lying swamps, tidal creeks and the mangrove patches, which enable the backwater to support diverse flora and fauna, including the brachyuran crabs. The distribution of crabs is influenced by habitat characteristics such as vegetation, substratum, food, salinity and the presence of other animals (Aspey, 1978; Icely and Jones, 1978; Rabalais and Cameron, 1985; Ewa-oboho, 1993; Thurman, 1998; Caesar *et al.*, 2005, Pandya and Vachharajani, 2010). Hence, in addition to the diversity assessment, the habitat ecology and habitat preferences of the brachyuran crabs encountered in the Cochin backwaters were also dealt with. Knowledge on the ecology and the habitat type preferred by the crabs is very important to conserve the crab population, since destruction of their habitat is the most important factor responsible for the deteriorating population and diversity of crabs.

Studies related to taxonomy and systematics of crabs is very limited, though numerous works are being carried out in various other aspects. The greater degree of adaptation of brachyuran crabs to different environments and the resulting individual variability within the species as well as population render taxonomic works difficult. Discontinuous distribution, allometric growth pattern which results in dynamic morphology, polymorphisms

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and sexual dimorphisms add more confusion in species identification of brachyuran crabs (Sethuramalingam and Ajmal Khan, 1991). Usually, the brachyuran crabs are distinguished based on minor morphological characters like spination – its presence or absence, orientation and arrangement or the number, transverse ridges, specific markings etc. But, in many cases, these characters are influenced by the growth. Therefore, besides the morphological characters, some other powerful taxonomic tools are needed to be employed for the proper identification of species. In the case of brachyuran crabs, first male pleopods or the third maxillipeds can be used as taxonomic tool (Chhapgar, 1957; Sethuramalingam and Ajmal Khan, 1991). The present study provides an extensive systematic account of the brachyuran crabs found in the Cochin backwaters, employing the difference in first male pleopods, shape of the male abdomen and the third maxillipeds.

Mud crabs are commercially important due to their large size and nutritive value. Hence, they are of great demand in the domestic as well as foreign market. Kerala is the biggest supplier of live mud crabs and Chennai is the major centre of live mud crab export (www.mpeda.com /mudcrabreports.pdf). The species identification of mud crabs belonging to genus *Scylla* has been controversial, for many years (Fuseya, 1998). Attempts to identify *Scylla* have led to much confusion because of the subtle morphological differences between the species. Taxonomy of mud crabs of the Cochin backwaters was first dealt with by Kathirvel (1981) and he recorded two species of *Scylla*, from Cochin backwaters; larger species, *Scylla oceanica* and smaller species, *Scylla serrata*. Later in 1982, Radhakrishnan and Samuel reported the occurrence of a subspecies *Scylla* *serrata serrata* from Cochin backwaters, which was denied by the other carcinologists (Joel and Sanjeevraj, 1983; Kathirvel and Srinivasagam (1992). They opined that the sub species *S. serrata serrata* reported by Radhakrishnan and Samuel from the Cochin Backwaters could be *S. serrata* only. Joel and Raj (1983) and Kathirvel and Srinivasagam (1992) reported that the two species of mud crabs occurring in the Indian waters is *S. serrata* and *S. tranquebarica*. However, according to the new reports, (Shaji *et al.*, 2006; Devi and Joseph, 2013 a, b; Mandal *et al.*, 2013, 2014 a, b; Balasubramanian *et al.*, 2014; Devi and Joseph, 2015), the mud crab species that are found to exist in the Indian waters is *S. serrata* and *S. olivacea*. Hence, it is very crucial to resolve the problems pertaining in the species identification of genus *Scylla*, since mud crabs hold appreciable importance in the inland fishery of the Cochin backwaters.

Chapter **2** REVIEW OF LITERATURE

- 2.1 Indian Brachyura A Review
- 2.2 Diversity of Brachyuran crabs- A Review
- 2.3 Habitat Ecology of Brachyuran Crabs- A Review
- 2.4 Systematics of Brachyuran Crabs- A Review
- 2.5 Systematic Account of mud crabs belonging to Genus Scylla
 A Review

2.1 Indian Brachyura - A Review

Studies on brachyuran fauna of Indian seas were initiated by Milne Edwards (1834), Henderson (1893) and de Man (1888 a, b) and they mostly dealt with the deep sea species. Later several authors dealt with various aspects of the brachyuran crab fauna of Indian sub continent including the fishery aspects, biological studies, diversity, systematics, ecological aspects and culture techniques.

The important notable works related to the brachyuran fauna of Indian waters include that of Kemp (1917, 1919,); Gravely (1927); Chopra (1931), Chopra and Das (1930, 1937); Pearse (1932), Panikker and Aiyyer (1937); Pillai (1951); Prasad and Thampi (1952, 1953); Menon (1952); Jones and Sujansinghani (1982); Chacko *et al.* (1953), Naidu (1953); Chacko (1956, 1957), Chhapgar (1957, 1962); George and Nayak (1961); Sankarankutty (1965); Thomas (1971); Ramamurthy (1972); Rao *et al.* (1973); Raman and
Srinivasagam (1978); Jameson *et al.* (1982); Srinivasagam and Raman (1985); Chakraborthy *et al.* (1986); Thomas *et al.*, (1987); Joel and Raj (1987); Manna (1988); Prasad and Neelakantan (1989); Balasubramanian (1993); Sukumaran and Neelakantan (1997); Roy and Das (2000); Ravichandran *et al.* (2001); Bijukumar *et al.* (2007); Soundarapandian *et al.* (2008); Funde *et al.* (2009); Josileen (2001, 2011).

Most of the earlier workers concentrated mostly on the taxonomic aspects of brachyuran crabs of Indian waters (Milne Edwards, 1834; Henderson, 1893, Alcock, 1899; De Man 1888 a, b). Notable contributions on the crab fishery related studies from Indian waters was made by Jones and Sujansinghani (1952), Prasad and Thampi (1953), Chhapgar (1962), Chacko *et al.* (1953), Chacko (1957), George and Nayak (1961), Rao *et al.* (1970), Thomas (1971), Ameer Hamsa (1978), Srinivasagam and Raman (1985), Joel and Raj (1987), Kurup *et al.* (1990), Anil (1997) and Josileen (2001). Studies related to the biological aspects of crabs in India are those of Menon (1952), Prasad and Thampi (1953), Rekha (1968), Pillai and Nair (1968, 1976), Simon and Sivadas (1979), Aruldhas *et al.* (1980), Prasad and Neelakantan (1989), Josileen (2001, 2011), Funde *et al.* (2009).

Ecological aspects of crabs including the zonation, habitat preferences, distribution and abundance of crabs in varying environmental conditions were dealt with by Pearse (1932), Balasubramanyan (1966), Rajinder *et al.* (1976), Roy and Das (2000), Ravichandran *et al.* (2001), Ajmal khan and Ravichandran (2009), Pandya and Vachhrajani (2010), Trivedi *et al.* (2012), Devi *et al.* (2013).

Studies related to larval development of crabs was dealt with by many workers including Naidu (1953), Anil (1997), Kathirvel *et al.* (2004), Josileen (2001).

Faunistic accounts of Brachyuran crabs of Andaman-Nicobar Islands have been dealt with by Alcock (1899), Chopra (1935), Sankarankutty (1961a), Premkumar and Daniel (1971), Reddy and Ramakrishna (1972), Kathirvel (1983), Pretzmann (1984), Deb (1985a, b) and Kumaralingam *et al.* (2012) while Borradile (1902), Sankarankutty (1961b), Meiyappan and Kathirvel (1978) and Suresh (1991) dealt with the crabs of Lakshadweep Islands.

2.2 Diversity of Brachyuran crabs- A Review

The earlier works on the brachyuran crab diversity were mostly restricted to the eastern coast of India and Bay of Bengal (Kemp, 1917, 1919; Gravely, 1927; Chopra and Das, 1930, 1937). The brachyuran fauna of west coast of India was first dealt with by Pillai (1951), who studied the crabs of Travancore waters. Later, Chhapgar (1957) gave a detailed account on the marine crabs of Bombay state along with their geographic distribution. An extensive study on the brachyuran crabs of Killai backwaters was done by Sethuramalingam (1983) and recorded 10 species of crabs from the backwaters. Later in 1991, Sethuramalingam and Ajmal Khan dealt with the brachyuran crabs of Parangipettai coast and provided descriptive keys with detailed figures. As a part of the biodiversity project on Gulf of Mannar biosphere reserve, Jayabaskaran *et al.* (1999) studied the brachyuran crabs of Gulf of Mannar.

A Comparative study on the brachyuran crab diversity of Pichavaram mangroves and artificially developed mangroves of Vellar estuary was done

by Ajmal Khan et al. (2005). Similar work was later conducted by Ravichandran and Kannupandi (2007) and Soundarapandian et al. (2008), who documented the biodiversity of brachyuran crabs in Pichavaram mangroves. Bijukumar et al. (2007) documented the brachyuran crab diversity landed as by-catch from the trawlers in the Kerala coast and recorded forty three families represented under 11 families and 22 genera, including 8 commercially exploited species. Roy and Nandi (2008) undertook a comparative study on the brachyuran crab diversity of three major brackish waters of India, viz., Chilka Lake, Pulicut Lake and Vembanad Lake. Brachyuran crab diversity of sub-littoral zones and mangrove regions Karwar estuary of Karnataka was studied by Haragi et al. (2010) and Bandekar et al. (2011) respectively. An annotated checklist of brachyuran crabs from the Pondicherry mangrove regions was provided by Satheeshkumar and Khan (2011). Kumaralingam et al. (2012) recorded the brachyuran diversity of Ritchieo's archipelago, Andaman and Nicobar Islands; meanwhile the brachyuran diversity of Mudasal Odai and Nagapattinam coast of Tamil Nadu was dealt with by Sakthivel and Fernando (2012). While studying the decapod diversity from the mangrove ecosystem of Uran, Maharashtra, Pawar (2012) recorded 13 species of brachyuran crabs, which accounts to 50% of the total decapods fauna of the region. Trivedi et al. (2012) and Trivedi and Vachhrajani (2012) dealt with the diversity of brachyuran crabs of Gulf of Kutch as well as the Saurashtra coast of Gujarat state respectively, while the diversity of brachyuran crab fauna of Mahi and Dhadar estuaries of Gujarat was studied by Shukla et al. (2013). An extensive study on the diversity of brachyuran crabs of the mangrove ecosystems of Tamil Nadu was conducted by Frederick and Ravichandran (2013). Roy and Nandi (2013)

studied in detail the diversity of marine brachyuran crab communities of the west coast of India, taking into account all the maritime states located on the west coast of India.

The comparative study on the brachyuran crab diversity of Chilka Lake, Pulicut Lake and Vembanad Lake, conducted by Roy and Nandi (2008) revealed the occurrence of 18 species of brachyuran crabs from Vembanad Lake, which is much lower when compared with the other two brackish water lakes. Hence they pointed out the need for a season-wise survey for a period of at least two years to assess the exact brachyuran diversity of Vembanad Lake. In this context, Devi *et al.* (2014) conducted an extensive study on the Cochin backwaters, located at the northern tip of the Vembanad Lake for a period of two years and could record a total of 24 species from the Cochin backwaters.

2.3 Habitat Ecology of Brachyuran Crabs- A Review

Wide range of studies is available on the brachyuran crabs regarding the ecology, habitat characteristics, its interactions with the environment, the influence of environmental parameters on their life activities, effects of changing environmental conditions on them etc. The feeding ecology and behavioural ecology of some selected crab species from the Kenyan mangroves was studied by Dahdough-Guebas (1994). Percival (2002) made an extensive report on the substratum preferences of Japanese shore crab *Hemigrapsus sanguineus*. Shirley *et al.* (2004) studied the effect of salinity fluctuations on the population of oyster reef crabs. Lee (2004) dealt with the ecological factors determining the settlement of post larvae and the dynamics of adults of velvet swimming crab, *Necora puber*. Mokhtari *et al.* (2008)

reported the population ecology of the fiddler crab *Uca lactea annulipes*, while the feeding ecology of the American crab *Rithropanopeus harisii* was studied by Hegele-Drywa and Normant (2009). Influence of substratum characteristics on the spatial distribution of Ocypodid crabs has been dealt with by several carcinologists, viz., Lim *et al.* (2005) who conducted the studies on *Uca annulipes* and *U.vocans* from Pulau Hantu Besar, Singapore; Bezzera *et al.* (2006) on the *U. leptodactyla*, *U. rapax*, *U. thayeri* and *U. maracoani* from the tropical mangroves of Brazil, Saher and Qureshi (2012) on *U. sindensis* from the Pakistan coast. The substratum preferences of mud crab, *Scylla paramamosain* was studied by Xue-lei *et al.* (2009) and stated that the settlement of juveniles and the burrowing habits of crabs depend on the particle size of the sediments.

An extensive study on the habitat preferences of crabs in the Pichavaram mangrove environment of south-east coast of India was done by Ravichandran *et al.* (2007). Spatial distribution and substratum preferences of the Ocypodid crab *Macrophthalmus depressus* was studied by Pandya and Vachhrajani (2010) from the Gujarat coast. Ethnoecology of *Scylla serrata* was studied in detail by Nirmale *et al.* (2012). Trivedi *et al.* (2012) studied the diversity and habitat preferences of brachyuran crabs in the Gulf of Kutch. A detailed account of the habitat ecology of the herring bow crab, *Varuna litterata* of the Cochin backwaters was given by Devi *et al.* (2013). Arya *et al.* (2014) carried out an extensive study on the role of brachyuran crabs for the assessment of chemical pollution in the environment and described them as an effective bio monitoring tool.

2.4 Systematics of Brachyuran Crabs- A Review

A perusal of the available literature on the brachyuran crabs would reveal that most of the earlier works on the brachvuran crabs were on the taxonomic aspects. The earliest classic works on the taxonomy of crabs include that of Linnaeus (1758), Fabricius (1798), De Haan (1833), Dana (1852a, b,c), Wood Mason (1871), Boas (1880), Miers (1886) Ortmann (1892), Henderson (1893) and Alcock (1895, 1896, 1898, 1899, 1900, 1901), Bouvier (1896), Borradile (1907), Balss (1957), Glaessners (1969) from different parts of the world including the Indian sub continent. Later, several carcinologists from different parts of the world, paid attention on the brachyuran fauna belonging to different families. Stephenson (1972) provided a checklist of the Portunid crabs of the Indo-west Pacific region, and according to his findings there are as many as 270 valid species existing under this family in this region. An extensive study on the fiddler crabs belonging to Family Ocypodidae was done by Crane (1975). Guinot (1977, 1978) proposed a new classification of brachyuran crabs based on the male and female genital openings, while Rice (1980) classified them on the basis of larval characteristics. A detailed monograph of the crabs of Japan and the adjacent seas was provided by Sakai (1976), which is widely referred in the systematic account of brachyuran crabs. Lucas (1980) dealt with the spider crabs of the family Hymenosomatidae, with special reference to Australian species, while Davie (1982) dealt with the mangrove crabs of Australia. Jones (1984) provided a comprehensive review of crabs inhabiting the mangrove ecosystem. A review of the grapsid crabs of American mangroves was studied by Abele (1992). Tan and Ng (1994) provided an annotated checklist of brachyuran crabs from the mangroves of Malaysia and Singapore. Ng *et al.* (2008) gave an annotated checklist of the extant brachyuran crabs of the world, which comprises over 10,500 names treated, including 6,793 valid species and sub species, 1271 genera.

In the national scenario, Chhapgar (1957) gave a detailed description on the marine crabs of Bombay state along with a valid identification key, which is widely used currently. Sethuramalingam and Ajmal Khan (1991) dealt with the brachyuran crabs of Parangipettai coast including the mangrove species and provided descriptive keys with detailed figures. As a part of the biodiversity project on Gulf of Mannar biosphere reserve, Jayabaskaran *et al.* (1999) studied the brachyuran crabs of Gulf of Mannar and provided figurative keys for the identification of marine crabs.

However, several researchers have dealt with the taxonomy of the commercially important mud crab species (Kathirvel, 1981; Radhakrishnan and Samuel, 1983; Joel and Sanjeevaraj, 1983; Kathirvel and Srinivasagam, 1992; Kathirvel *et al.*, 2004; Devi and Joseph, 2013a; Trivedi and Vachhrajani, 2013; Padate *et al.*, 2013; Balasubramanian *et al.*, 2014; Mandal *et al.*, 2014 a, b; Devi and Joseph, 2015).

2.5 Systematic Account of mud crabs belonging to Genus *Scylla* – A Review

The first record of mud crabs was made by Forskal (1775) and he designated this newly found crab as *Cancer serratus*. Later several carcinologists from different parts of the Indo-Pacific region concentrated on this species (Herbst, 1796; Fabricius, 1798; De Haan, 1833; Milne Edwards, 1834; Dana 1852a,b,c; Stimpson, 1907). Estampador (1949) gave a detailed taxonomic account of genus *Scylla*, where he recognized three

species and a new sub species from Philippines. Serene (1952) also reported the occurrence of four forms of *Scylla* in Vietnam. These works were subsequently reviewed by Stephenson and Campbell (1960) and Holthius (1978) and they pointed out the inconsistent and variable nature of the distinguishing characters. Fushimi (1983) and Oshiro (1988) reported the presence of three forms of mud crabs in Japan. The genetic variability analysis of mud crabs were studied by Fuseya and Watanabe (1996) and they proposed three species for genus *Scylla* on the basis of variations noticed through electropohoretic analysis. Fushimi and Watanabe (1998) described the major problems persisting in the identification of mud crabs. Mean while, Keenan *et al.* (1998) collected mud crab specimens from the Red Sea and throughout the Indo-Pacific regions and classified them into four species, *Scylla serrata*, *Scylla tranquebarica*, *Scylla olivacea* and *Scylla paramamosain*, on the basis of morphological, morphometric and molecular analysis.

In India, reference to the occurrence of mud crabs was first reported by Fabricius (1798), who described the mud crabs specimens collected from Tranquebar (Tharagambady of Tamil Nadu coast) and designated them as *Portunus tranquebaricus*. Succeeding this, many authors viz., Alcock (1899), De Man (1909), Kemp (1915), Gravely (1927), Pearse (1932), Chopra and Das (1937), Panikker and Aiyyer (1937), Pillai (1951), Naidu (1953), Chhapgar (1957, 1962), Balasubramanian (1966), Rekha (1968), Premkumar and Daniel (1971) have mentioned the existence of mud crabs, which was designated as *Scylla*, while dealing with various aspects such as fishery, biology etc.

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Later, the taxonomy of mud crabs of Indian waters was dealt with by Kathirvel (1981), Radhakrishnan and Samuel (1982), Joel and Sanjeevaraj (1983) and Kathirvel and Srinivasagam (1992). Kathirvel (1981) recorded two species of Scylla from Cochin backwaters, larger species being Scylla oceanica and smaller one being Scylla serrata. Later in 1982, Radhakrishnan and Samuel reported the occurrence of a subspecies Scylla serrata serrata from Cochin backwaters on the basis of morphological characters. Joel and Sanjeevaraj (1983) studied the taxonomy of Scylla from Pulicut lake and reported that the occurrence of two species, namely S. tranquebarica and S. serrata. Taxonomy of mud crabs from India were then critically analysed by Kathirvel and Srinivasagam (1992) and stated that the two species of mud crabs found to occur in Indian waters is S. serrata and S. tranquebarica. In contrast to their findings, Shaji et al. (2006) stated that the two species occurring in the Indian waters is S. serrata and S. olivacea. However, some of the recent studies also claims the occurrence of more than two apparent mud crab species from Indian coastal waters, namely, S. serrata, S. tranquebarica and S. olivacea, on the basis of external morphology (Devi and Joseph, 2013 a; Trivedi and Vachhrajani, 2013; Devi et al., 2014). Quite recently, the studies conducted by Mandal et al., (2013, 2014 a, b), Balasubramanian et al (2014) and Devi and Joseph (2015) confirms the findings of Shaji et al. (2006), i.e., the two species of mud crabs that exist in the Indian waters is S. serrata and S. olivacea.

Chapter **3 MATERIALS AND METHODS** 3.1 Study Area

- 3.2 Sampling stations
- Sampling Techniques
- Analytical Techniques
- Statistical Analysis

3.1 **Study Area**

The area selected for the study is the Cochin Backwaters, Kerala, South west coast of India.

The Cochin backwaters is situated at the tip of northern Vembanad Lake and a tropical positive estuarine system extending between $9^{0}40$ ' & $10^{0}12$ 'N and $76^{0}10$ ' & $76^{0}30$ 'E, with its northern boundary at Azhikode and southern boundary at Thanneermukkom.

The salinity gradient of the Cochin backwaters supports diverse species of flora and fauna depending on their capacity to tolerate oligohaline, mesohaline or marine conditions. Low lying swamps and tidal creeks, dominated by sparse patches of mangroves with their nutrient rich physical environment, support larvae and juveniles of many commercially important species. The backwaters also act as the nursery grounds of commercially important fin fishes and shell fishes (Menon et al., 2000).

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Chapter 3

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Plate 3.1: A view of the study area, the Cochin backwaters



Plate 3.2: Mangrove patch along the Cochin backwaters.

3.2 Sampling stations

An extensive field survey was conducted along the study area and based on the survey, 15 sampling stations (Fig.3.1) were fixed along the backwaters. The selection of sampling stations was done on the basis of salinity, habitat type, presence of mangroves as well as the availability of crabs.



Fig. 3.1: Map showing the study area Cochin backwaters with sampling stations.

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The sampling stations selected were:

1) Station 1: Thanneermukkom (North): The station located on the northern side of Thanneermukkom barrage, forms the southern boundary of Cochin Backwaters. The influence of tides and currents arising at the mouth of the estuary situated at Cochin can be observed in this zone. Intense fishing activity- stake nets and cast nets were observed in this area. Besides, prawns and the pearl spots, mud crabs also contribute a major share in the local fishery of this area. No mangrove patches are found to occur in this area.



Plate 3.3: Station 1: Thanneermukkom North.

 Station 2: Vaikom – The station is located about 13 km north to Station 1. The station is observed to have intense fishing activity, sand mining and inland water transport activities. No mangrove patches are observed in this zone.

- Station 3: Aroor The station is highly influenced by the tidal action, currents and saline water from the Arabian Sea. The sampling site has an extensive stretch of mangroves.
- 4) Station 4: Valanthakad The station is an isolated island, despite of its proximity to the National Highway. The island has a very rich mangrove stretch and has gained much interest among the ecologists and the biologists.
- 5) Station 5: Kumbalanghi is a fishing village, and has a very vast backwater stretch. Here the backwater is in an enclosed condition with a rather narrow opening into the main lake. This site too has a rich diversity of mangroves and mangrove associates.
- Station 6: Kandakkadavu lies at the western part of the Cochin backwaters. It is a typical coastal area ornamented with lush green mangroves.
- Station 7: Thevara The station is highly influenced by the tidal action and the currents from the Arabian Sea. The zone exhibits sparse patches of mangroves.
- 8) Station 8: Thoppumpady The station is influenced by the tidal influx and currents emerging from the Arabian Sea. Intense fishing activity and inland water transport activities are actively seen in this zone. The zone is also very close to the Cochin Fisheries Harbour.

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Plate 3.4: Station 8: Thoppumpady

- 9) Station 9: Barmouth- The zone connects the Cochin Backwaters with the Arabian Sea, hence highly influenced by the tidal influx and currents emerging from the sea. Intense fishing activities, inland water transport activities as well as dredging activities are observed in this zone.
- 10) Station 10: Marine Science Jetty- The zone is in the vicinity of the oil tanker berth of Cochin, due to which, the zone exhibits pollution due to oil spills at times. This zone is also influenced by the tides and currents from the sea.
- Station 11: Vallarpadam- The station has a close proximity to the Cochin Container Trans-shipment terminal and the zone has experienced indiscriminate mangrove destruction, during the past few years. Still, some scattered patches of mangroves are found to observe in this area.

12) Station 12: Vypin- Vypin is the biggest among the group of islands in the Cochin backwaters and is bound by the Arabian Sea on its western side and the backwaters on its eastern side. The station had a luxuriant mangrove patch earlier, but the recent LNG and Petroleum tanks and terminals built at the heartland of mangroves in Vypin has destroyed the vegetation on a mass scale.



Plate 3.5: Station 12: Vypin.

- 13) Station 13: Varappuzha The zone is in the vicinity of the industrial area, Eloor, where Fertilizers and Chemicals Travancore (FACT), Travancore Cochin Chemicals, Hindustan Pesticides Limited (world's largest manufacturer of DDT) are located. The zone is thus an industrially polluted area.
- 14) Station 14: Munambam Munambam is a coastal village, situated at the northern point of the backwaters, surrounded by the Arabian Sea on the west, Periyar on the east and a mouth opening to the sea on the north. The site does not have a luxuriant mangrove vegetation, only a sparse patch is found.

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15) Station 15: Azhikode – The station is located in Thrissur District and the Azhikode old harbor is located in this zone. The station forms the northern boundary of the Cochin Backwaters, where the backwaters opens in to the Arabian Sea.



Plate 3.6: Station 15: Azhikode.

3.3 Sampling Techniques

Sampling was done along the 15 stations of the study area on a monthly basis, for a period of two years from June 2010 to May 2012. During the sampling, crab specimens were collected from all the 15 stations, along with water and sediment samples of the area.

Different methods were employed for the collection of crabs. Samples, especially, that of commercially important crabs like the mud crabs and the marine crabs were collected from local markets and local fishermen- stake net operators, Chinese dip net operators, gill net operators as well as from the indigenous crab traps or the 'Njandara' operators of that particular area.



Plate 3.7: A fisherman operating the indigenous 'Njandara'.



Plat 3.8: A Chinese dip net along the Cochin backwaters

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Those small sized crabs from the mangrove regions were collected by hand picking or scoop nets. The burrowing crabs from the mangrove sediments were captured using knots made of rachis of coconut leaflets. In addition to these methods, boat sampling was also conducted in the study area on a monthly basis in order to collect the bottom crawling benthic crabs. The benthic crabs were collected using a Van Veen grab $(0.4m^2)$.



Plate 3.9: Collection of benthic crab sample with a Van Veen grab

The crabs were bought to the lab alive and its colour, carapace width, carapace length, and the morphological features like shape of the carapace, markings on the body, number of anterolateral teeth, features of frontal lobe, pubescence, spination on the chelae and other legs etc were noted. The crabs were then numbed by freezing and later on preserved in 10% formaldehyde solution for further identification and the taxonomic studies.

The samples needed for the molecular analysis of Genus *Scylla* was collected from station 12: Vypin. Live mud crabs were collected from the local fishermen and they were packed carefully in perforated cardboard boxes and taken to the Central Genetics Lab, RGCA, Sirkazhi. Muscle tissue was dissected out from the live mud crabs and the DNA was isolated immediately after the dissection.

3.4 Analytical Techniques

3.4.1 Diversity study

The crab specimens collected from the sampling sites were brought to the lab alive and its colour, carapace width, carapace length, and the morphological features like shape of the carapace, markings on the body, number of anterolateral teeth, features of frontal lobe, pubescence, spination on the chelae and other legs etc were noted. It was then numbed by freezing and preserved in 10% formaldehyde for further identification. The collected specimens were then identified up to species level using standard keys.

The identification keys provided by Chhapgar (1957), Sakai (1976) and Ng *et al.* (2008) were referred for the identification of marine species. The detailed account on the fiddler crabs provided by Crane (1975) was made use for the identification of Ocypodid crabs. The benthic crabs collected from the sediments were identified using the key proposed by Lucas (1980) and the mangrove crabs were identified referring to the keys

and descriptions provided by Sethuramalingam and Ajmal Khan (1991), Jayabaskaran *et al.* (1999) and Ajmal Khan and Ravichandran (2009). The identification of crabs belonging to genus *Scylla* was done using the keys proposed by Keenan *et al.* (1998), since the identification key provided by Keenan *et al.* (1998) is widely accepted by the carcinologists world-wide as well as by the FAO.

3.4.2 Ecological study

As a part of the ecological study, water quality parameters like water temperature, salinity, pH and sediment quality parameters like sediment temperature, moisture content, organic carbon and sediment texture of the stations were also analysed using standard techniques.

3.4.2.1 *Water Temperature:* using a centigrade thermometer

- **3.4.2.2** Salinity: using salino-refractometer (ERMA Japan)
- **3.4.2.3 pH:** using a digital pH meter(EUTEC Instruments pH Tutor Model No: 1548649)
- **3.4.2.4 Sediment Temperature:** using a centigrade thermometer

3.4.2.5 *Moisture content: Evaporation method (Jackson, 1973)*

5 gm of sediment was taken in a previously weighed pertridish and the combined weight of petridish and the sediment was noted. It was then kept for drying in a hot air oven at 60° C for 5 hours. After 5 hours, the petridish was taken out and checked whether the sediment had been dried completely. Usually, the sediment which is clayey contains more moisture and hence requires more time to get the sediment completely dried. In that case, the petridish containing the sediment was again kept for drying and checked after every 1 hour. The process was repeated till the water content is

completely removed. When the sediment was dried completely, without any moisture, the petridish was weighed again.

Calculation:

Water loss = Sample wet weight – Sample dry weight Percentage of water content = $\frac{\text{Sample wet weight} - \text{Sample dry weight}}{\text{Sample wet weight}} \times 100$

3.4.2.6 Estimation of Organic carbon (Walkey- Black method)

The sediment was first ground finely to pass through the 0.5 mm sieve. Weighed about 0.5 g of finely ground sediment into a 500 ml wide mouthed Erlenmeyer flask. To this added 10 ml potassium dichromate solution and swirled gently, followed by the addition of 20 ml conc. H_2SO_4 carefully through the sides of the flask. It was then mixed by swirling for 1 min and kept for 30 min. Then added about 250 ml distilled water and allowed to cool. 1 ml ferroin solution was added as indicator and titrated with freshly prepared ferrous solution while the mixture was being stirred, till the end point was reached. The colour change during titration will be from greenish blue to reddish brown.

Calculation:

Organic carbon (%) =
$$\frac{(BV-TV) \times 0.003 \times 100}{W}$$

Where, BV = blank value, TV = titre value,

M = Molarity of ferrous solution, W = Weight of the soil

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3.4.2.7 Sediment texture analysis

50 gm of sediment was taken in a 100 ml beaker and 10% sodium hexametaphosphate solution was added to it in order to disperse the particles. It was then kept overnight, so that the whole particles are separated completely. The sediment was then sieved in a 5mm sieve, with minimal amount of water, say 50 ml. The particles which were retained in the sieve were the sand content of the sediment. The sand particles were transferred into a watch glass and weighed. Now, the remaining sediment solution contained silt and clay particles. The amount of silt and clay was estimated by running the sediment solution in the particle size analyser (Sympatec-Germany).

3.4.2.8 Substratum and Distribution of crabs

i) Similarity indices:

For finding out the similarities between seasonal and spatial aspects in the distribution of crabs, Bray-curtis similarity plots with SIMPROF tests and multi-dimensional scaling (MDS) were made.

 SIMPROF test: The test involves cluster analysis. Cluster analysis provides a summary of the similarity in species composition of the different sampling stations. Clusters are formed of the sampling stations that are similar in species composition, as measured by a chosen ecological distance. Sites that are grouped into the same cluster are more similar in species composition than the sites that are grouped into different clusters.

- Non-Metric Multi-Dimensional Scaling plots
 MDS is an ordination technique, where similarities between the sampling stations in terms of species composition is depicted in as few dimensions as possible. As in the case SIMPROF test, similar stations are grouped together in an MDS plot.
- ii) Bubble plots: Bubble plots were constructed for finding out the relative abundance of crab species in different sampling stations.

3.4.3 Taxonomic study

For taxonomic studies of crabs, usually the first male pleopod or the third maxillipeds is studied in detail. Only the left pleopod or third maxilliped, i.e., right when viewed from the ventral side of the crab was removed carefully using a clean forceps, needle and scissors. It was then mounted temporarily with glycerine and alcohol (Ajmal Khan and Ravichandran, 2009) and then studied under a stereo microscope or compound microscope depending on the size. The structures were measured and drawn with the help of a micrometer and a camera lucida respectively.

Apart from these two structures, the shape of the male abdomen can also be considered for taxonomic studies. It is observed that the shape of the male abdomen too varies with different species. In that case, crabs can be identified and distinguished without causing any harm to the crab. In the present study, the male abdomen is studied carefully using a hand lens in the case of larger species and that of smaller species is studied under a stereo microscope. The structure was not dissected out for the study, but was kept intact on its body.

3.4.4 Species Identification of Genus Scylla

3.4.4.1 Morphological study

The morphological characters include colour and shape of the carapace, shape of the frontal lobes, spination of the anterolateral borders and the chelate legs and the presence of hexagonal markings. The crabs were identified using the identification key provided by Keenan *et al.* (1998). Though several identification keys are available by various authors, the key provided by Keenan *et al.* (1998) is widely accepted by crab taxonomists as well as by FAO (Ng, 1998).

The first and second male pleopods from the male specimens with a size range of 95- 128 mm carapace width (110.94 ± 10.2) and the third maxillipeds were cut carefully from both sexes were dissected out carefully using a forceps and scissors. They were observed under a stereo microscope to study the variations among the species. The pleopods and the maxillipeds were then drawn and described using a camera lucida.

3.4.4.2 Multivariate Analysis of Morphometric characters

Twenty four morphometric characters were noted for 365 mud crabs (211 males and 154 females) with a size range of 80 mm- 128 mm carapace width (100.8±10.17), measured using vernier calipers. The characters include, Carapace width (CW), Internal carapace width (ICW), Carapace width on 8th spine (8CW), Posterior carapace width (PCW), Carapace length (CL), Frontal width (FW), Frontal median spine height (FMSH), Distance between frontal median spines (DFLS), Sternum width (SW), Abdomen width (AW), 5th periopod

dactyl length (5PL), 5th periopod dactyl width (5PW),3rd merus length (3PML), Dactyl length (DL), Propodus depth (PD), Inner carpus spine (ICS), Inner propodus spine (IPS), Outer carpus spine (OCS), Outer propodus spine (OPS), Merus length (ML), Propodus length (PL) and Propodus width (PW).

Crabs with carapace width above 80 mm were selected and the morphometric measurements of these crabs were considered for tabulating the following 27 ratios for statistical analysis. The ratios include 9th carapace spine height/ ICW, CW/ 8CW, PCW/ICW, CL/ICW, FW/ICW, PWC/FW, FMSH/FW, FMSH/DFMS, DFMS/FW, DFMS/DFLS, SW/ICW, AW/SW, PL/ICW, DL/PL, PW/PL, PD/PL, PW * PD * 0.7854/ PL, IPS/PL, OPS/PL, IPS/OPS, ICS/PL, OCS/PL, ICS/OCS, ML/PL, 5PW/5PL and 3PML/ICW.

After tabulating the 27 ratios for all the three morphologically distinct species, discriminant function analysis of these 27 ratios was carried out using SPSS ver. 20.0.

3.4.4.3 Molecular study

In the present study taxonomic identification of *Scylla* spp. was based on amplification of the ITS-1 region and the molecular sequence data from the mitochondrial cytochrome c oxidase subunit I (COI) gene.

Live samples of *Scylla* species with a size range of 80 mm- 128 mm carapace width (100.8±10.17), were collected from Station 12 Vypin. Muscle tissue from thoracic appendages was dissected from the live mud crabs and DNA isolation was done immediately after the dissection. Total

genomic DNA was isolated from 10 mg of muscle tissue. Tissue was digested by incubating with proteinase K/SDS solution at 37°C for two hours. DNA isolation was carried out following standard phenol: chloroform extraction and ethanol precipitation technique (Sambrook et al., 1989). Purity and quality of DNA was checked on 0.8% agarose gel. concentration of dissolved DNA was estimated using UV The spectrophotometer. DNA was diluted so as to obtain a final concentration of 100 ng/µl. Polymerase chain reaction (PCR) amplification was carried out to obtain sequences of the partial mitochondrial COI gene, in a total of 25 µl volume containing 1x standard Taq buffer (10 mMTris-HCl, 50 mMKCl, pH 8.3), 3.5 mM MgCl₂, 200 µM dNTPs, 0.4 µM each primer, 1U Taq DNA polymerase (Fermentas, Inc.) and 1 µl DNA template (100 ng). Amplification was done using Universal primer as well as Species specific primer (Keenan primer), designed by Central Genetics Lab, Rajiv Gandhi Centre for Aquaculture, Sirkazhi, Tamil Nadu. The thermal profile used was 94^oC for 5 min (initial denaturation) followed by 35 cycles of 94 °C for 15 sec (denaturation), 50° C for 30 sec (annealing) and 72° C for 30 sec (extension) and a final extension at 72° C for 10 min. 10 µl of the amplified PCR product was analyzed by electrophoresis in 1.5 % agarose gel in TBE buffer, stained with ethidium bromide and visualized under UV light. PCR products were purified using Isopropanol precipitation technique and the purified PCR products were sequenced with CO1 primers using ABI Prism Sequencing kit (BigDye Terminator Cycle). The homologue searching of the nucleotide sequence (using blastn suite) was performed with the Basic Local Alignment Search Tool (BLAST) through NCBI server (http://www.ncbi.nlm.nih.gov/blast).

In addition to the sequencing of Cytochrome oxidase-1 gene, amplification of the ITS-1 region is also done in order to confirm the results. The bands obtained after the PCR amplification of ITS-1 region was compared with the bands proposed by Imai *et al.* (2004).

3.5 Statistical Analysis

The software programmes employed for the univariate and multivariate analysis of the data includes SPSS ver.20.0 (Statistical Programme for Social Sciences version 20.0) and PRIMER ver. 6 (Plymouth Routines In Multivariate Ecological Researches version 6).

The station-wise and season-wise variations of the ecological parameters were statistically analysed by two-way ANOVA and their means were compared by Tuckey's HSD using SPSS ver.20.0. The correlation between the ecological parameters and the distribution of crabs was established by Best analysis using PRIMER 6. The BEST routine available in PRIMER ver. 6 combines the BIOENV and BV STEP procedures of PRIMER ver. 5. This routine uses all the available environmental variables to find out the variables that 'best explains' the patterns in biological data. Starting with the variable showing the maximum matching coefficient, variables were successively added, the combinations tested at each stage. The variables contributing the least were eliminated. Several iterations of the procedure were carried out from a random selection (=6) variables to ensure the best match found.

In order to discriminate the species belonging to the Genus *Scylla*, Stepwise Discriminant Function analysis was done using SPSS ver. 20.0 to determine the characters which contributed the most. These characters, which contributed the most, were taken into consideration while distinguishing between the species.



Part - II

Chapter 4 DIVERSITY OF BRACHYURAN CRABS OF COCHIN BACKWATERS

Chapter 5 HABITAT ECOLOGY AND HABITAT PREFERENCES OF BRACHYURAN CARBS OF COCHIN BACKWATERS

Chapter 6 TAXONOMIC ACCOUNT OF BRACHYURAN CRABS OF COCHIN BACKWATERS

Chapter 7 ON THE SYSTEMATICS OF GENUS SCYLLA DE HAAN,1833 OF COCHIN BACKWATERS

DIVERSITY OF BRACHYURAN CRABS OF COCHIN BACKWATERS



Introduction 4.1 4.2 Materials and Methods Results Discussion #

4.1 Introduction

Studies on the local checklist of fauna are of great importance for the formulation of conservation policies (Fransozo et al., 1992, Hebling et al., 1994), because these studies lead to the structure, ecological processes and problem faced by ecosystem. The Indian coastal or marine environments comprise some of the high biodiversity areas (Ajmal Khan et al., 2005). The environs contain specialized habitats like salt marshes, coral reefs and mangroves and each habitat has its specific animal community. Crustaceans are the most important groups of tropical benthic communities. The larger and more abundant species are important for human consumption while the incredible variety of small species contribute importantly to the complexity and functioning of tropical ecosystems (Hendrickx, 1995); for e. g., on rocky shores, crabs are prime predators on molluscs, small crustaceans and other invertebrates, but on the other side they also provide prey base for

fish, decapods and some terrestrial vertebrates (Siddon and Witman, 2004). So as a prey and predator they have potential influence on the behaviour, distribution and abundance of their own as well as neighboring communities (Seeley, 1986; Trussell and Nicklin, 2002).

Among all benthic macro fauna which dwells in the intertidal zone, brachyurans are the most prominent because of great diversity, comprising of about 700 genera and 5000 species (Melo, 1996). About 640 species of crabs are listed from Indian waters, among which 12 species are regarded as commercially important, inhabiting the coastal waters and adjoining brackish water environment. They occupy a very important position in the commercial crustacean fishery on account of their export potential and high nutritive value. In sea food export, among crustaceans, crabs occupy third position based on their external market demand. Even though its present status is not comparable to that of shrimps, crabs support a sustenance fishery of appreciable importance (Rao et al., 1973). In nutritional point of view, crab meat is not inferior to any sea food items, as it is rich in vitamins, glycogen, protein, fats and minerals. Crab meat is also considered to possess some medicinal value. It is believed to cure asthma and chronic fevers (Chopra, 1939; Chidambaram and Raman, 1944). In addition to the value of the larger and more abundant species for human consumption, a tremendous variety of small species contribute to the complexity and functioning of tropical ecosystems. In mangrove environment, crabs represent the predominant group. Crabs have a significant role in detritus formation, nutrient recycling and dynamics of the ecosystem. Their digging behavior enhances aeration and facilitates drainage of mangrove soils (Ajmal Khan and Ravichandran,

2009). They are also found to retain commensal association with algae, seaweeds, coelenterates, barnacles, bivalves and holothurians.

This work has been undertaken to document the brachyuran diversity of the Cochin backwaters and the associated mangrove patches.

4.2 Materials and Methods

The protocol followed has been described in chapter 3 (Section 3.4.1).

4.3 Results

A total of 23 species of crabs belonging to 16 genera and 8 families were collected from Cochin backwaters during the present study. Among the families, highest number of species was recorded from Family Portunidae (8 species) followed by Grapsidae (7 species).

Systematics:

Phylum: Arthropoda Superclass: Crustacea Pennant, 1777 Class : Malacostraca Latreille,1806 Subclass: Eumalacostraca Calman,1904 Order : Decapoda Latreille, 1803 Suborder : Pleocyemata Burken Road, 1963 Infraorder : Brachyura Latreille, 1802.

Description of the species

Scylla serrata (Forskal, 1775)
 Family : Portunidae
 Subfamily : Portuninae (Plate 4.1)

These are edible crabs, commonly known as the mangrove crab or mud crab (green morph of the mud crabs). They are the largest known species from the near shore and brackish water habitats of India. Carapace of the crab is smooth with conspicuous transverse ridges. Colour of the crab is pale green to olive green and the fingers of the chelae pale orange. Hshaped gastric groove is deep. The frontal margin of the carapace is cut into bluntly pointed teeth, with rounded interspaces. Anterolateral margin of the carapace is cut into 9 narrow teeth. Chelipeds are large and strong with its propodus inflated. The distinguishing character of the species is the presence of two prominent sharp spines on the distal half of outer margin of the carpus of the cheliped. The limbs of the crabs exhibited conspicuous hexagonal patterning in both sexes. This is found to be the largest among the *Scylla* species.

It occurs fairly abundantly in the backwaters and the associated mangrove mud flats, throughout the year and has a great tolerance to fluctuations in salinity. They constitute an important fishery and were caught in crab traps, gill nets, Chinese dip nets and stake nets.

They are of great demand in the foreign markets and therefore the lion share of the green mud crabs caught in the area is being exported in live condition. Kerala forms the biggest supplier of live mud crabs and Chennai forms the main centre of export, from where the crabs are being taken to other countries, mainly the South East Asian countries like Singapore, Philippines, and Malaysia.

Scylla olivacea (Herbst, 1796)
 Family : Portunidae
 Subfamily : Portuninae (Plate 4.2)

Carapace of the crab is smooth. Colour of the carapace is crab grayish green, fingers and the last segments of the walking legs are pale orange. The frontal margin is cut into rounded lobes. Anterolateral margin of the carapace is cut into 9 broad teeth. Chelipeds are large and the propodus are inflated. The species is characterized by the presence of single spine on the distal half of outer margin of the carpus of the cheliped. Palm of the cheliped exhibits a pair of prominences on the dorsal margin behind the insertion of the dactyl. The inner prominence is larger than the outer one. Hexagonal markings are weak or absent.

Though *S. olivacea* are found to occur in the backwaters and the associated mangrove mud flats, throughout the year, like the *S. serrata*, they are recorded mostly from the low saline areas. They are even caught from the Pokkali rice fields and swamps of the region. They constitute an important fishery and were caught in crab traps, gill nets, Chinese dip nets and stake nets. They are of great demand in the domestic market, since most of the green mud crabs are being exported to foreign market.
Portunus(Portunus) pelagicus (Linnaeus, 1798)
 Family : Portunidae

Subfamily : Portuninae (Plate 4.3)

They are edible crabs, commonly known as the blue swimmer crabs. Carapace is blue coloured in males and sand coloured in female, with an extensive spread of irregular white spots. The distal segments of the legs are pinkish in colour. The anterolateral border of the carapace has nine teeth of which the posterior most one is much larger and laterally prolonged. The last pair of legs is paddle shaped and adapted for swimming. It is a marine migrant species and occurs in the backwaters during the late post monsoon and pre monsoon periods when, the salinity is comparatively high. They were caught in stake nets, gill nets and Chinese dip nets.

4) *Portunus (Portunus) sanguinolentus* (Herbst, 1783)Family : Portunidae

Subfamily : Portuninae (Plate 4.4)

They are edible crabs, commonly known as blood-spotted swimmer crab. They are comparatively smaller in size. Carapace is sand coloured and characterized by three large reddish round spots on the posterior half of the carapace, of which one is median and the other two lateral, each spot encircled by a white ring. The carapace is much broader than the length. Its anterolateral margin is cut into nine spines of which, the posterior most is the longest. The fifth pair of legs is paddle shaped and adapted for swimming. Like, *P. pelagicus*, this species too is marine, and occurs in the backwaters during post monsoon and pre monsoon periods when salinity is high. They were caught in Chinese dip nets, stake nets and gill nets.

5) Charybdis (Charybdis) feriata (Linnaeus, 1758)Family : Portunidae

Subfamily : Portuninae (Plate 4.5)

These are edible crabs, and fairly larger in size. Carapace brown in colour with purple tinge and conspicuous yellow markings, the central one resembling a cross; chelipeds and limbs brownish with yellow and white spots and the tips brownish pink. Carapace is broad, slightly convex and smooth, length as much as its width. Anterolateral borders with six spines, the first one is truncated and notched anteriorly and the rest acuminate. The last pair of legs paddle shaped and adapted for swimming. It inhabits offshore waters, but also found to occur in the backwater during the pre monsoon period when the salinity is relatively high. Though it is an edible species, it is not consumed in some parts of the country, while it contributes to the crab fishery of the Cochin region. They are caught in Chinese dip nets from the backwaters.

6) Charybdis (Charybdis) lucifera (Fabricius, 1798)

Family : Portunidae

Subfamily : Portuninae (Plate 4.6)

Carapace is yellowish brown with two large white spots on either branchial region. Chelipeds, scarlet pink, the fingers brownish and the extreme tips whitish. The right cheliped is markedly larger than the left cheliped. The last pair of legs is paddle shaped, for swimming and the posterior border of its propodite is found to be serrated. They are well distinguished with the presence of a sharp median lobule on lower border of the orbit. There is no spine on the posterior margin of carpus of natatory leg. These were found to occur during the post monsoon and pre monsoon period and were caught in stake nets and Chinese dip nets from the backwaters.

7) Charybdis (Goinohellenus) hoplites (Wood mason, 1877)

Family : Portunidae

Subfamily : Portuninae (Plate 4.7)

Carapace of the species is light brown in colour. The most distinguishing characters of this species is that the posterior border of the carapace forms an angular junction with the postero-lateral borders and the last tooth of the anterolateral borders is a long spine, about twice as long as those in front of it., which is usually observed in the Genus *Portunus*. It is also found to possess a spine on the posterior border of the arms of the cheliped. The last pair of legs paddle shaped and adapted for swimming. The specimens were caught in a Chinese dip net during the pre monsoon season during the sampling period.

8) Thalamita crenata (Latreille, 1829)

Family : Portunidae

Subfamily : Portuninae (Plate 4.8)

Colour of the carapace brownish or greenish grey, claws pinkish, tips brown and the extreme tips white. The front is cut into six lobes, excluding the inner supra-orbital tooth. The anterolateral borders are cut into five equal teeth. The transverse ridge on the carapace appears faint. The outer surface of the propodus of the cheliped is smooth. The last pair of legs paddle shaped and adapted for swimming. This estuarine crab inhabits mud flats, sandy beaches and mangroves. They occur throughout the year and were caught in stake nets, Chinese dip nets and van veen grab during the sampling.

9) Uca (Celuca) lactea annulipes (H.Milne Edwards, 1837)

Family : Ocypodidae

Subfamily : Ocypodinae (Plate 4.9)

They are commonly known as the 'Porcelain fiddler crab'. Colour of the carapace black with white or yellow stripes and the chelipeds pinkish or whitish. Carapace is subquadrilateral with moderately convergent lateral borders. Front broad, supraorbital border oblique and the external orbital angle pointed obliquely outward. The most striking feature of this species is the enlarged pincers of the males, which they use for courtship. The pincers or the major cheliped is smooth on the outer surface, while the lower border has a faint ridge, extending upto the base of the immovable lower finger. There is a wide gap between the fingers and the movable upper finger is curved inward at the tip and it extends past the immovable lower finger. They inhabit damp ground, found on the shores and near the mangroves. They were caught by handpicking and using knots made of coconut leaflets from the study area during the sampling period.

10) Macrophthalmus depressus (Ruppell, 1830)

Family : Ocypodidae

Subfamily : Macrophthalminae (Plate 4.10)

Carapace grey in colour; broader than long; rectangular in shape; surface studded with minute pearly granules. The anterolateral borders parallel and the anterolateral angles with a square cut lobe, rather than a tooth. Eyestalks are remarkably long and slender and almost reaching the end of the orbital angles. The inner surface of the palm is smooth, and ornamented with thick hairs. Hairs are also present on the ambulatory legs. They are found to be mud dwellers and occur in mangrove regions. They were caught in stake nets. Handpicking was also done more often during the sample collection.

11) Nanosesarma (Beanium) batavicum (Moreira, 1903) Family : Grapsidae Subfamily : Sesarminae (Plate 4.11)

Colour of the carapace mottled grey, chelipeds cherry red. Front broad and deflexed. Upper surface of the palm of cheliped with two oblique pectinated crest. The distinguishing feature of the species is the presence of three acute spines on the posterodistal border of pereopod four. They are found to occur in the mangrove regions and the samples were collected by hand picking and with knots made of coconut leaflets.

12) Nanosesarma (Beanium) andersonii (De Man, 1888) Family : Grapsidae Subfamily : Sesarminae (Plate 4.12)

Colour of the carapace dark brown, chelipeds reddish. Front broad and deflexed. Upper surface of the palm of cheliped with numerous striae, one of which forms pectinated crest. They are distinguished by the presence of four strong spines on posterodistal border of merus of pereopod four. They are abundantly found in the mangrove regions. Handpicking and knots made of coconut leaflets were the methods employed to collect the samples during the study.

13) Neoepisesarma (Neoepisesarma) mederi (H.Milne Edwards, 1854)

Family : Grapsidae

Subfamily : Sesarminae (Plate 4.13)

Colour of the carapace black or dark brown, Chelipeds reddish. Front broad and deflexed. Carapace quadrangular, with an anterolateral teeth, slightly convergent posteriorly. The surface of the carapace have tufts of hair on the anterior portion. The distinguishing feature of this species is that, above the transverse dactylar tubercles, a sulcus runs about one third of total length of the tubercles and the vertical granular crest of the the inner palm is salient. They are found in the muddy substratum of mangrove regions and were caught by handpicking and knots made of coconut leaflets during the study period.

14) Neoepisesarma (Selatium) brockii (De Haan, 1887)

Family : Grapsidae

Subfamily : Sesarminae (Plate 4.14)

Colour of the carapace dark brown, chelipeds orange coloured with white tips. The distinguishable feature of the species is that the pectinated crest on upper palm reach the margin and the dactylar tubercles are well separated from one another without any tranverse sulcus above. They are semi terrestrial forms, found in mangrove mud, fissures of rocks, stone embankments, backwater shores etc. They are very vigilant and intelligent and they trust their speed and craft to escape the enemies, thus they are hard to pursue. The samples were captured by handpicking during the study.

15) *Metapograpsus messor* (Forskal, 1775)

Family : Grapsidae

Subfamily :Grapsinae (Plate 4.15)

Colour of the carapace bottle green, chelipeds appears light violet. The legs are striped with alternate dark and light bands. Carapace is four-fifths as long as broad and the lateral margins markedly converging backwards. Front more than half the width of the carapace. The post frontal region appears to have some transverse markings. The propodus of the chelae inflated. Walking legs are short and the dactylus of the legs are nearly as long as the propodus. The last segment of the abdomen appears triangular in males. They are mud dwellers and inhabit the mangroves, subtidal region etc. During the sample collection, the specimens were caught by handpicking and knots made of coconut leaflets.

16) Metapograpsus maculatus (H.Milne Edwards, 1873)

Family : Grapsidae

Subfamily :Grapsinae (Plate 4.16)

Carapace is black in colour and chelipeds are bright violet. Carapace is comparatively elongated, being seven-eighth as long as broad, with less convergent sides. The post-frontal region is devoid of any transverse markings or ridges. The fingers of the chelipeds are much longer than the upper border of the palm. Walking legs are larger and the dactylus of the legs distinctly shorter than the propodus. The striking feature of this species is that the last segment of male abdomen is trilobed. They inhabit the mangroves and the intertidal region. The specimens were caught by handpicking during sampling.

17) Varuna litterata (Fabricius, 1798)

Family : Grapsidae

Subfamily : Varuninae (Plate 4.17)

They are commonly known as the 'herring bow crab'. Colour of the carapace light brown to brownish grey. Carapace is squarish, with a smooth surface. Anterolateral borders are cut into three broad, but less sharp teeth. Dactylus, propodus and carpus of legs are laterally flattened like paddles and are fringed with long, densely packed hairs. With its legs shaped as paddles used for swimming, it is sometimes called the 'Paddler crab'. They are estuarine species and are found to occur in the mangrove regions and intertidal areas. They were caught in large numbers in stake nets and Chinese dip nets during all the seasons of the sampling period.

The present study is the first time report on the occurrence of *V. litterata* from the study area, the Cochin backwaters.

18) Heteropanope indica (De Man, 1858)

Family : Xanthidae

Subfamily : Pilumnidae (Plate 4.18)

These are small sized crabs, with transversely oval shaped carapace, markedly oval and glabrous. Frontal lobe is straight and truncate. Anterolateral borders are cut into four teeth. The dorsal surface of the carapace with two parallel transverse ridges, which are beaded with fine granules. Chelipeds are extremely unequal and the carpus and the propodus of the chelipeds are found to be studded with pearly granules. The ambulatory legs are slender and sparingly haired. They are estuarine and found to inhabit the mangrove regions. During sampling, these specimens were caught in a Van Veen grab

19) Xenophthalmus pinnotheroides (White, 1846)

Family : Pinnotheridae

Subfamily : Xenophthalminae (Plate 4.19)

Colour of the carapace is dirty white. Carapace is subtrapezoid in shape, slightly broader than long, with the anterolateral angle bluntly rounded. Frontal region is narrow and strongly deflexed. Orbits situated longitudinally and parallel to each other. Chelipeds are found to be symmetrical, its palm longer than the fingers. Propodus of the first walking leg as long as broad. Carpus and propodus of second leg are armed with a tuft of dense pubescence. The third and the last legs longer and more slender covered with short hairs. The third leg is the longest among all. They were caught in large numbers from the backwater, where the substratum is muddy, throughout the year. They were caught in Van Veen grab as well as in stake nets during sample collection. 20) Doclea gracilipes (Stimpson, 1857)
 Family : Majidae
 Subfamily : Pisinae (Plate 4.20)

Carapace is discoid and have a series of blunt spine like structures arranged longitudinally along the middle line. The body and limbs are covered with thick hairs and the chelipeds are shorter than the other legs. The second pair of legs is the longest and are about three to four times the length of the carapace. Colour of the carapace and the segments of the legs covered with hair, is greenish brown, which the segments devoid of hair is pink in colour. They are marine species, occur in 30 to 50 meters depth in sandy or muddy substrata. The specimens were caught in Chinese dip net from the Cochin backwaters during the pre monsoon season, when the salinity was high in the sampling location.

21) Ebalia malefactrix (Kemp, 1915)

Family : Leucosiidae

Subfamily : Ebaliinae (Plate 4.21)

The carapace is subcircular in shape, with a separate facet on the side wall of the carapace at the hepatic region. Eyes and orbits are too small. Chelipeds are stouter and longer than the legs. These tiny crabs inhabit the sediments of the backwaters and were collected by operating a Van Veen grab. They occur in large numbers during the post monsoon and the premonsoon period when the salinity is comparatively higher.

22) Halicarcinus messor (Stimpson, 1858)

Family : Hymenosomatidae (Plate 4.22)

These are small sized, benthic crabs. Carapace is suboval and as long as broad. Lateral walls of the carapace with two teeth, one at the anterolateral angle and the second at the posterolateral angle. The front region is extended to form a rostrum. The rostrum is trilobed, the lateral lobes are small and acute and the medium one is long and spatuliform, broadly rounded apically. The median lobe is broadest at halfway along its length and are devoid any setae. Chelipeds are heavy, slightly longer than the legs. Palm of the chelipeds markedly swollen. These are benthic species and were caught in Van Veen grab during sample collection.

23) *Elamenopsis alcocki* (Kemp, 1917)Family : Hymenosomatidae (Plate 4.23)

These are small sized, benthic crabs with a circular carapace. Frontal region extended to form a rostrum. Excluding the rostrum, the carapace width to length ratio is 1:1. The anterolateral border cut into a single tooth. Chelipeds are heavy and the fingers of the chelae with a wide gap at the base. The ambulatory legs are long and slender. During sample collection, they were caught from the muddy sediments of the backwaters by operating a Van Veen grab.

4.4 Discussion

Among the benthic communities, crustaceans are important because more number of species are used for human consumption and a large variety of small species contributing to the complexity and functioning of tropical ecosystem (Hendrickx, 1995). Brachyuran crabs form the most diverse group among the decapods crustaceans, comprising about 700 genera, and 5000 to 10,000 species worldwide (Kaestner, 1970; Melo, 1996; Ng, 1998; Martin and Davis, 2001; Ng *et al.*, 2008; Yeo *et al*, 2008). Indian waters harbor rich variety of brachyuran crabs of about 640 species and most of them are either commercially or ecologically important or both. Crabs occupy a very important position in the commercial crustacean fishery on account of their export potential and high nutritive value. In sea food export, among crustaceans, crabs occupy the third position based on their external market demand. Even though its present status is not comparable to that of shrimps, crabs support a sustenance fishery of appreciable importance. (Rao *et al*, 1973). Besides commercial importance, crabs are also considered to be ecologically important too. In mangrove environment, crabs represent the predominant species and are often considered to be the keystone species.

Checklist of biodiversity of an area plays significant role in the conservation of habitat and species. It provides a better way for different kinds of research studies like biogeography, biodiversity and environment monitoring activities and these on other hand provide crucial information for policy makers in the designing of better conservation strategies (Shukla *et al.* 2013). So far, several workers have undertaken the assessment of brachyuran biodiversity on their area of interest. Reports of Kemp (1917, 1919); Gravely (1927); Chopra and Das (1930, 1937) forms the earliest works regarding the brachyuran crab diversity of eastern coast of India and the Bay of Bengal, while the brachyuran fauna of west coast of Travancore waters. The diversity study of brachyuran fauna was undertaken by several

authors later on which includes Chhapgar (1957), Sethuramalingam (1983), Sethuramalingam and Ajmal Khan (1991), Jayabaskaran *et al.* (1999), Ajmal Khan *et al.* (2005), Ravichandran and Kannupandi (2007), Roy and Nandi (2008), Haragi *et al.* (2010), Bandekar *et al.* (2011), Kumaralingam *et al.* (2012), Sakthivel and Fernando (2012). Trivedi *et al.* (2012), Trivedi and Vachhrajani (2012), Shukla *et al.* (2013), Frederick and Ravichandran (2013), Roy (2013), Devi *et al.*, (2014).

Although there are lots of works being carried out on the benthic fauna, mangrove diversity, fish diversity etc in the Cochin Backwaters, a comprehensive study of the brachyuran fauna has not been attempted. Some of the previous works reports that the Cochin backwaters supports a low diversity of crabs (Pillai, 1951; Rao and Kathirvel, 1972; Devasia and Balakrishnan, 1985, Roy and Nandi, 2008). Being the latest study on the brachyuran diversity, Roy and Nandi (2008) reported 18 species from the Vembanad Lake and pointed out the requirement of a season wise survey for about two years to assess the exact brachyuran biodiversity status of Vembanad Lake. The present study was carried out on the Cochin Backwaters, for a period of two years and the survey was conducted on a monthly basis, rather than season wise, and could collect a total of 23 species of brachyuran crabs. Among the 23 species, five of them, namely Scylla serrata, S. olivacea, Portunus pelagicus, P. sanguinolentus and Charybdis feriata are edible and commercially important. Menon et al. (2000) reported that S. serrata, P. pelagicus and P. sanguinolentus constitute 4% of the commercial fishery of Vembanad Lake. Sheeba (2000) recognized four species viz., S. serrata, P. pelagicus, P. sanguinolentus and C. feriata as the commercially important crab species of the Cochin backwaters.

Among the brachyuran crab resources of the study area, mud crabs holds the prime position in terms of abundance as well as its economic importance. Mud crabs belonging to Genus Scylla contribute considerably to the estuarine fisheries of Cochin Backwater area. Earlier, there has been much confusion regarding the species of Scylla, which were found to occur in the backwaters. Kathirvel (1981) recorded two species of Scylla from Cochin backwaters, larger species being S. oceanica and smaller one being S. serrata. Later in 1982, Radhakrishnan and Samuel reported the occurrence of a subspecies Scylla serrata serrata from the Cochin backwaters on the basis of morphological characters, but their findings were denied by Joel and Sanjeevaraj (1983) and Kathirvel and Srinivasagam (1992) stating that only two species are found to occur in the Indian waters, viz., S. serrata and S. tranquebarica. They also opined that the species which have been identified as S. serrata serrata by Radhakrishnan and Samuel could be S. serrata only. Later on Devi et al. (2014) could identify three species of Scylla from the Cochin backwaters, based on the morphological variations. Beside S. serrata and S. tranquebarica, they also recorded a third variety of Scylla sp. However, in the present study, only two species of Scylla were found to occur in the Cochin Backwaters namely, S. serrata and S. olivacea. The findings were confirmed with the help of morphological characters proposed by Keenan et al. (1998), morphometric comparisons as well as molecular analysis. Hence, the present work is the first report on the occurrence of S.olivacea in the Cochin Backwaters.

S. serrata, commonly known as the green mud crab occurs throughout the year in the Cochin Backwaters. Though they are estuarine species and are reported to tolerate wide range of salinity, they are caught only from

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those areas of backwaters where mesohaline conditions prevail. During the two year sampling period, they were not reported from low saline areas. The findings are in accordance with Kurup *et al.* (1990), who reported the occurrence of *S. serrata* in the backwaters on a year round basis where a mesohaline condition was prevailing over. However, *S. olivacea* was found to be available throughout the backwaters, and they are abundant in those areas, where the backwater turns into a freshwater basin during monsoon season, rather than the mesohaline areas. *S. serrata* and *S. olivacea* are commercially important due to their large size and good taste. *S. serrata*, being larger are tastier, are of great demand in the foreign markets, while *S. olivacea*, being smaller species fetches good price in the domestic market.

During the sampling period, was observed that some of the marine migrant species invade the high saline areas of the backwaters during the summer months. They include *Portunus pelagicus*, *P. sanguinolentus*, *Charybdis feriata*, *C. lucifera*, *C. hoplites* and *Doclea gracilipes*. Menon (1952) reported the occurrence of *P.sanguinolentus* in large numbers in the Malabar coast during the period from January –April, i.e., the pre monsoon months. The occurrence of *P. pelagicus* in the Cochin backwaters during the post monsoon was reported by Rao and Kathirvel (1972), while Kurup *et al.* (1990) reported its appearance during the high saline premonsoon period. Raj (2006) observed that *C. feriata* comes up occasionally in the shore seines and are caught in the Pulicut lake. The findings of the present study is totally compatible with the findings of previous studies regarding the invasion of marine migrants into the backwater areas occasionally. These marine migrant species are found to invade the backwaters during the post monsoon and pre monsoon periods and tend to disappear when the salinity falls during

the monsoon season. Among these, *P. pelagicus*, *P. sanguinolentus* and *C. feriata* are commercially important species and contribute to the fishery of the backwaters during the post monsoon and pre monsoon periods. Though *C. lucifera* is considered as edible in some parts of the country, especially the east coast, they are not consumed in the Cochin area.

Thalamita crenata is another Portunid crab, which is found to occur in the Cochin Backwaters. Manickaraja and Balasubramaniam (2009) reported *T. crenata* to be a deep sea crab, but observed to occur in shallow water gill net operation along the north Tuticorin region. However, in the present study, *T. crenata* was found to occur throughout the year in the mangrove dominated areas of the Cochin backwaters. In accordance with the reports of Manickaraja and Balasubramaniam (2009), this species never formed a fishery and lacks any economic importance in the Cochin backwater areas. Besides these, *Charybdis hoplites* and *Doclea gracilipes* are the other marine migrant species which are found to occur in the Cochin backwaters during the summer months.

Among the 23 species recorded from the Cochin backwaters, 13 species belonging to four families were found to occur in the mangrove dominated regions. They include 3 Portunids, 7 Grapsids, 2 Ocypodids and 1 Pilumnid, namely, *Scylla serrata*, *S. olivacea*, *Thalamita crenata*, *Nanosesarma batavicum*, *N. andersonii*, *Neoepisesarma brockii*, *N. mederii*, *Metapograpsus messor*, *M. maculatus*, *Varuna litterata*, *Macrophthalmus depressus*, *Uca annulipes* and *Heteropanope indica*. Except, *V. litterata*, the other 6 grapsid crabs were found exclusively in the mangrove regions and nowhere else. However, the brachyuran crab diversity on the mangrove

ecosystems of the Cochin backwater was low when compared to other regions. Joel et al. (1985) could record the occurrence of 29 species of mangrove crabs from Pulicut Lake. The mangrove regions of Sunderbans recorded the occurrence of 18 crab species (Chakraborthy and Choudhary, 1992). The diversity of brachyuran crabs of Pichavaram mangroves was 23 (Ravichandran *et al.*, 2001), however an extensive study conducted by Frederick and Ravichandran (2013) to assess the brachyuran crab resources of mangrove ecosystems of Tamil Nadu, reported 46 species from Pichavaram mangroves and 17 species from Vellar estuary. The mangrove regions of Karwar estuary were found to harbor 13 species of crabs (Bandekar et al., 2011), similar to the mangrove regions of the Cochin backwaters. The mangrove mud flats of Gulf of Kutch recorded only 10 species of brachyuran crabs (Trivedi et al., 2012), which was lower when compared to the brachyuran crab resources of the Cochin backwaters (13 species). Mangrove crabs are ecologically significant due to the role they play in nutrient recycling, balancing the ecological equilibrium, soil aeration etc. Hence they form the keystone species of the mangrove ecosystem.

Varuna litterata, is a grapsid crab, commonly known as the 'herring bow crab'. They usually inhabit the mangroves, estuarine and freshwater environments, in shallow sub tidal regions and usually found under rocks, logs and dead leaves and lives in burrows along the embankments or sides of pools, creeks and shallow banks. *V. litterata* has been reported from all over the Indo-West Pacific. In India, it is found to be very abundant in the Sunderbans of West Bengal, estuarine regions of Hoogly and in Gulf of Mannar and Palk Bay (Rao *et al.*, 1973). In the West coast of India, *V. litterata* was first collected by Pillai (1951) from the open sea at Trivandrum, but was not reported from the Travancore backwaters. Later Chhapgar (1957) reported V.litterata from Bombay and Kolak. The species was found to occur in the waters along the Karnataka coast by Dinesh babu et al. (2011). A comparative study conducted by Dev and Roy (2008) on the brachyuran biodiversity of three brackishwater lakes, viz. Chilka Lake, Pulicut Lake and Vembanad Lake revealed that they are present in Pulicut and Chilka Lake but are absent in Vembanad Lake. However, the species was found to be available plentifully in the Cochin backwaters throughout the year. They were caught in large numbers from the mangrove regions of backwaters, which exhibited good tidal influx and close proximity to the sea. Hence, the present work is the first report on the occurrence of V. litterata in the Cochin Backwaters. V. litterata has been recorded as commercially important species in Bengal, India, where it is eaten by poor people, and its numbers compensate for its small size (Hora, 1933). Rao et al. (1973) also reported it as an edible species and as a fishery resource of India. However, this species is not considered for food in the Cochin backwater area. They are usually caught as by-catch in the stake nets, which are being laid for capturing prawns in the Cochin backwaters and the crabs found entangled in the net are taken and discarded.

In the present study, six benthic species were observed, viz. *Ebalia* malefactrix, Elamenopsis alcocki, Halicarcinus messor, Heteropanope indica, Xenophthalmus pinnotheroides, Macrophthalmus depressus, mainly where the substratum is muddy in nature. They were caught by operating a Van Veen grab. *E. malefactrix*, belonging to Family Leucosiidae, *E. alcockii* and *H. messor*, belonging to the family Hymenosomatidae are very tiny creatures to be distinguished between. These microscopic crabs were found

abundantly in the muddy sediments of backwaters throughout the year, from the mesohaline areas of the backwaters. *Heteropanope indica* is a small sized crab species, which were found in the vicinity of concrete structures, pillars etc, attached to the stones and concrete parts. *Xenophthalmus pinnotheroides* is yet another small sized crab, found abundantly in the Cochin backwaters. These filthy looking crabs were caught in stake nets and grabs, covered in mud. Unlike the other 5 species, *Macrophthalmus depressus* is not too small a creature. They are mud dwellers and were caught in stake nets from the mangrove regions of the Cochin backwaters. Pillai (1977) also reported six benthic species from the sediments of the Cochin backwaters, viz. *Litocheira* sp, *Viaderiana* sp, *Rynchoplax, Macrophthalmus sp, Ebalia malefactrix, Eriphia smithii*, while Batcha (1984) could find only two benthic species, viz., *Rynchoplax* and *Viaderiana* sp.

Though the brachyuran diversity of the Cochin backwaters is not comparable with that of other regions of the country like Pichavaram mangroves, Sunderbans, Vellar estuary etc, it is definitely endowed with good brachyuran crab resources, with a total of 23 species. However, destruction of mangroves, filling of lands, waste disposal and other human interventions are leading to the deterioration of the backwaters. This would affect the diversity of brachyuran crabs in the near future itself, since their habitat is being destroyed. Hence, it is so crucial to conserve the habitat of the crabs, in order to conserve their diversity. Therefore, there is an urgent need to adopt suitable measures to prevent the habitat from deterioration and the species diversity from decline.





Plate 4.7: Charybdis (Goinohellenus) hoplites (Wood mason)



Plate 4.2: Scylla olivacea



Plate 4.4: Portunus (Portunus) sanguinolentus (Herbst)



Plate 6: Charybdis (Charybdis) lucifera (Fabricius)



Plate 4:8 Thalamita crenata (Latreille)

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Plate 4.9: Uca (Celuca) lactea annulipes (H.Milne Edwards) Plate 4.10: Macrophthalmus depressus (Ruppell)





Plate 4.11: Nanosesarma (Beanium) batavicum (Moreira) Plate 4.12: Nanosesarma (Beanium) andersonii (De Man)





Plate 4.15: Metapograpsus messor (Forskal)



Plate 6:14: Neoepisesarma (Selatium) brockii (De Haan)



Plate 4.16: Metapograpsus maculatus (H.Milne Edwards)

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Department of Marine Biology, Microbiology and Biochemistry, School of Marine Sciences, CUSAT



Plate 4.23: Elamenopsis alcocki (Kemp)

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HABITAT ECOLOGY AND HABITAT **PREFERENCES OF BRACHYURAN CARBS OF COCHIN BACKWATERS**

- 5.1 Introduction
 - Materials and Methods

Results

Discussion

Introduction 5.1

The environmental parameters determine the existence of communities in an ecosystem and a comparative knowledge about these parameters are imperative in understanding the ecosystem. Aquatic ecosystem is a complex one comprising interacting physicochemical and biotic components whose dynamics are often integrated (John, 2010). The estuarine system is subjected to distinct seasonality in its physicochemical parameters and hence the estuarine organisms tend to have different tolerance levels and responses to different physicochemical parameters. The seasonal variations in the environmental parameters are clearly evident in the case of Cochin backwaters due to the influence of strong south west monsoon. The hydrographical parameters such as salinity, water temperature and pH play a very significant role in the distribution and abundance of flora and fauna in aquatic ecosystems. The sediment characteristics have been regarded as the cardinal factor, which affects the benthic community structure. Hence a comprehensive knowledge on the water and sediment characteristics is a pre requisite to understand the ecology of brachyuran crabs.

Benthic invertebrates show peculiar habitat and substratum preference and are often considered to be the keystone species of the environment because of the role they play in the different functions of the community. Wide range of studies is available on macro invertebrates as indicator species of aquatic habitat, but amongst them specifically, brachyuran crabs is an indicator of changes in biotic and abiotic factors. Being benthic invertebrate, brachyuran crabs have a significant influence of the substratum characteristics. Hence, an extensive study on the diversity status, distribution, density and population dynamics of crabs can denote the health of the environment, where they live.

The inhabitation patterns and habitat preferences are found to be varying among the brachyuran crabs. They are found to be adapted to various biotopes, such as marine, estuarine, fresh water, intertidal and even terrestrial habitats. The existence and distribution of crabs are influenced by the environmental factors prevailing in the area.

Among the ecological parameters the distribution of crabs is influenced by sediment characteristics, rather than the water quality parameters. Botto and Iribane (2000) opined that activities of burrowing crustaceans like crabs are mainly influenced by sediment characteristics. Pandya and Vachhrajani (2010) are also of the opinion that the density and distribution of crabs are controlled by sediment composition and overall habitat characteristics. As per the findings of Aspey (1978), Icely and Jones (1978), Rabalais and Cameron (1985), Ewa-oboho (1993), Thurman (1998) and Caesar *et al.* (2005), the distribution of crabs is influenced by habitat characteristics such as vegetation, substratum, food, salinity and the presence of other animals. According to Arya *et al.* (2014) crabs are considered to be the indicators of sediment quality.

Knowledge on the ecology and the habitat type preferred by the crabs is very important to conserve the crab population, since the destruction of their habitat is the most important factor responsible for the deteriorating population and diversity of crabs. In turn, the health status of the benthic environment of a particular area can be well determined by analyzing the crab population of the area- its characteristics, behavioral patterns and their life activities. The present study is an attempt to describe the distribution as well as habitat preference of the brachyuran crabs that are enlisted from the Cochin backwaters.

5.2 Materials and Methods

The protocols followed for the analysis of water quality and sediment quality parameters have been described in Chapter 3 (section 3.4.2)

The station-wise and season-wise variations of the ecological parameters were statistically analysed by two-way ANOVA and their means were compared by Tuckey's HSD using SPSS ver.20.0. The correlation between the ecological parameters and the distribution of crabs was established by Best analysis using PRIMER 6. To find out the similarities between seasonal and spatial aspects in the crab distribution, Bray-curtis similarity plots with SIMPROF tests and multi-dimensional scaling (MDS) were made.

Using the abundance data, bubble plots were constructed for showing the relative abundance of prominent crab species in different stations.

5.3 Results

5.3.1 Ecological parameters

The seasonal average values of water quality parameters, viz., salinity, water temperature and pH for two consecutive years are presented in Figs. 5.1, 5.2, 5.3 and that of sediment quality parameters, viz., sediment temperature, moisture content and organic carbon content are presented in Figs. 5.4, 5.5 and 5.6. Fig. 5.7 depicts the percentage composition of sand, silt and clay particles in the sediments of 15 stations of the study area.

The mean variations (Mean \pm SD) in the water quality and sediment quality parameters for the 15 sampling stations (pooled data of two consecutive years) are presented in Figs. 5.8, 5.9, 5.10, 5.11, 5.12, 5.13 and 5. 14. The station wise mean and standard deviation of water quality and sediment quality parameters in all the 15 stations are given in Table 5.1a and 5.1b, 5.1c, respectively. The Draftsman plot showing the interactions of ecological parameters (season wise) are shown in Fig. 5.15 a, b, c. Results of ANOVA and correlation are given in Tables 5.2a, 5.2b, 5.2c, 5.2d, 5.2e, 5.2f, 5.2g, 5.2h, 5.2i and 5.3.

5.3.2 Water quality parameters

<u>Salinity</u>: Salinity exhibited wide fluctuations both station-wise as well as season-wise. During the study period, the salinity ranged from 0.5 to 33 ppt. The lowest salinity was observed during the monsoon period, while salinity gained momentum during post monsoon months and recorded its maximum value during pre monsoon period.

During the first year (June 2010- May 2011), the monsoon period exhibited a low salinity range of 0.5 to 10, while it increased in post monsoon period with a salinity range of 1.5 to 27. Salinity was its maximum during the premonsoon period with a range of 6 to 33. During the second year (June 2011- May 2012), monsoon period recorded a salinity range of 1.5 to 11, post monsoon period recorded 1.5 to 24, and the premonsoon period recorded a salinity range of 11 to 32.

Salinity exhibited a well marked variation among the 15 sampling stations. Spatially, the stations which are closer to the sea and have higher tidal influence exhibited a higher salinity, when compared to those stations which are away from the sea. The lowest salinity was recorded at Station.1: Thanneermukkom with a salinity range of 0.5 to 13 with a mean of 5.39 ± 0.16 and the highest was at Station 15: Azhikode with salinity ranging from 10 to 33 and a mean value of 18.71 ± 0.18 . Station-wise mean and standard deviation of salinity is given in Table. 5.1a

<u>Water Temperature</u>: During the study period, water temperature exhibited marginal fluctuations both season wise as well as station- wise. When the first year's data was considered, Water temperature fluctuated between 24° C- 28° C during the monsoon months, 26° C to 30° C during the post monsoon and 26° C to 32° C during the premonsoon period. During the second year, monsoon months recorded a water temperature range of 23° C to 27° C, post monsoon months recorded 25° C to 30° C and the premonsoon months recorded a temperature range of 25° C to 33° C.

Among the 15 stations, Station 11: Vallarpadam, Station 14: Munambam and Station 15: Azhikode exhibited a slightly higher water temperature than the other 12 stations. Station 11: Vallarpadam and Station 15: Munambam recorded a water temperature ranging from 28° C to 32° C with a mean of 29.46 ± 0.06. Station 15: Azhikode exhibited a range of 28° C to 33° C with a mean of 29.62 ± 0.29. Station-wise mean and standard deviation of water temperature is given in Table. 5.1a

<u>pH</u>: The pH of the study stations was almost nuetral throughout the study period, though, Station 13: Varappuzha exhibited a slightly lower or acidic range. In the present study, season wise variations in pH was not found to be significant, since the stations exhibited a steady pH range through out the year, irrespective of the seasons.

However, spatial variations were recorded. When all the stations exhibited a nuetral pH range, Station 13: Varappuzha exhibited an acidic pH range of 5.12- 6.82 with a mean of 5.87 ± 0.32 . Station-wise mean and standard deviation of pH is given in Table. 5.1a



Fig. 5.1: Seasonal Variation of Salinity during the study period

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Fig. 5.2: Seasonal Variation of Water Temperature during the study period



Fig. 5.3: Seasonal Variation of pH during the study period

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Sl No:	Stations	Salinity		Water Temperature		рН	
		Mean	± SD	Mean	± SD	Mean	± SD
1	Thannrmukkm	5.39	0.16	25.71	0.18	7.05	0.04
2	Vaikom	5.84	0.38	25.54	0.41	7.11	0.07
3	Aroor	7.77	0.50	24.79	0.18	6.87	0.27
4	Valnthakad	8.33	0.11	25.42	0.12	6.96	0.31
5	Kmblngi	14.50	0.00	26.46	0.18	6.95	0.11
6	Kndkdvu	15.21	0.06	26.62	0.29	7.09	0.03
7	Thevara	13.21	0.29	26.17	0.24	7.49	0.21
8	Thpmpdy	15.75	0.00	27.17	0.00	7.31	0.15
9	Brmouth	15.71	0.53	27.04	0.18	7.61	0.07
10	Mar Sci	14.37	0.06	27.75	0.12	7.58	0.00
11	Vllrpdm	14.25	0.12	29.46	0.06	7.65	0.21
12	Vypin	14.83	0.12	28.75	0.12	7.51	0.19
13	Vrppzha	5.90	0.09	26.21	0.06	5.87	0.32
14	Munbm	17.54	0.06	29.46	0.06	7.91	0.19
15	Azhikde	18.71	0.18	29.62	0.29	7.95	0.16

 Table 5.1 a: Station wise Mean and Standard deviation of water quality parameters

5.3.3 Sediment quality parameters

Sediment temperature: Sediment temperature too followed the similar pattern of variation as observed in the case of water temperature. Both station wise and season wise fluctuations were evident in the case of sediment temperature. During first year, the monsoon months exhibited a low range of sediment temperature, 21° C to 27° C, while a higher range of 24° C to 28° C was recorded during the post monsoon and pre monsoon months. During the second year, sediment temperature ranged between 23° C to 26° C in the monsoon period, 24° C to 28° C in the post monsoon and 25° C to 32° C in the pre monsoon period.

The station wise variations were also very much similar to the variations observed in water temperature. Those stations which exhibited a higher range of water temperature recorded maximum range in the case of sediment temperature also. To be more precise, as in the case of water temperature, three stations out of the 15 sampling stations, viz., Station. 11: Vallarpadam, Station 14: Munambam and Station 15: Azhikode recorded a higher range of sediment temperature when compared to the other 12 stations. Station 11: Vallarpadam exhibited a sediment temperature range of 26° C to 29° C with a mean of 27.08 ± 0.82 ; Station 14: Munambam recorded 26° C to 28° C with a mean of 27.42 ± 0.12 and Station 15: Azhikode recorded 26° C to 30° C with a mean of 27.54 ± 0.29 . Station-wise mean and standard deviation of sediment temperature are given in Table. 5.1b

It was also observed that in most of the cases, the sediment temperature of a sampling station as slightly lower than the corresponding water temperature of that station.

<u>Moisture content</u>: Moisture content was determined by the water retention capacity of the sediments and it is influenced by the sediment texture as well as the organic matter content in the sediments. From the season wise data analysis, it was observed that the there is no significant and conspicuous fluctuation in the moisture content of the sediments of the sampling stations seasonally. However, moisture content varied considerably between stations. The sediments of Station 1: Thanneermukkom and Station 2: Vaikom recorded less moisture, and it varied within the range of 3.01% to

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6.83% with a mean of 4.81 ± 0.53 and 3.02% to 7.63% with a mean of 5.01 ± 0.24 , respectively. Moisture content in the sediments of 6 sampling stations viz., Station 7: Thevara, Station 8: Thoppumpady, Station 9: Barmouth, Station 10: Marine Science Jetty, Station 11: Vallarpadam and Station 12: Vypin were found to be higher, when compared with other sampling stations. Station 7: Thevara recorded a range of 10.07% to 12.67% with a mean of 11.46 ± 0.03 ; Station 8: Thoppumpady recorded 10.24% to 12.76% with a mean of 11.53 ± 0.15 ; Station 9: Barmouth recorded 10.14% to 12.9% with a mean of 11.17 ± 0.25 ; Station 10: Marine Science Jetty recorded 10.18% to 12.9% with a mean of 11.6 ± 0.02 ; Station 11: Vallarpadam recorded 10.2% to 12.21% with a mean of 11.16 ± 0.01 and Station 12: Vypin recorded a range of 10.24% to 12.13% with a mean of 11.42 ± 0.0 . Station-wise mean and standard deviation of moisture content are given in Table. 5.1b

<u>Organic carbon</u>: During the study period, organic carbon content in the sediments did not show seasonal variations as in the case of moisture content. However, considerable variation in the organic carbon content occurred spatially. When station wise data were analysed, maximum range is recorded from Station 3: Aroor and Station 4: Valanthakad, owing to the luxuriant mangrove patches contributing leaf litter to the sediments, which in turn decomposes and adds to the organic carbon content of the sediments. The organic carbon content in the sediments at Station 3: Aroor was within the range of 3.99% to 5.37% with a mean of 4.68 ± 0.11 and that of Station 4: Valanthakad the range was within 4.21% to 4.76% with a mean of 4.39 ± 0.02 . Station-wise mean and standard deviation of organic carbon is given in Table. 5.1b







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Fig. 5.5: Seasonal Variation of Moisture content during the study period






Fig. 5.6: Seasonal Variation of Organic carbon during the study period

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SI		Sedi	ment	Mois	Moisture		Organic Carbon	
No	Stations	Temperature (^v C)		conten	content (%)		(%)	
110.		Mean	± SD	Mean	± SD	Mean	± SD	
1	Thannrmukkm	25.25	0.24	4.81	0.53	0.66	0.00	
2	Vaikom	24.88	0.18	5.01	0.24	0.42	0.00	
3	Aroor	23.96	0.18	7.40	0.18	4.68	0.11	
4	Valnthakad	24.00	0.12	9.01	0.13	4.39	0.02	
5	Kmblngi	24.58	0.35	7.73	0.03	1.20	0.05	
6	Kndkdvu	24.92	0.00	7.41	0.04	1.23	0.00	
7	Thevara	24.71	0.29	11.46	0.03	0.95	0.03	
8	Thpmpdy	25.75	0.12	11.53	0.15	0.65	0.00	
9	Brmouth	25.83	0.24	11.17	0.25	0.35	0.01	
10	Mar Sci	25.83	0.00	11.60	0.02	0.63	0.02	
11	Vllrpdm	27.08	0.82	11.16	0.10	1.24	0.16	
12	Vypin	27.08	0.35	11.42	0.00	1.39	0.03	
13	Vrppzha	25.25	0.00	6.82	0.07	0.41	0.02	
14	Munbm	27.42	0.12	6.47	0.06	0.69	0.01	
15	Azhikde	27.54	0.29	6.74	0.21	0.70	0.00	

 Table 5.1 b: Station wise Mean and Standard deviation of Sediment quality parameters

<u>Sediment Texture</u>: The texture of sediments of the sampling stations did not exhibit conspicuous variations with seasons. Though the percentage composition of the sediments varied with seasons, the variations were not considerable and insignificant. The dominant particle of a particular station remained the same throughout the year.

However, sediment texture of different stations disclosed marked variations in the percentage composition of sand, silt and clay. The stations and the corresponding textural class are presented in Table 5.4. The seasonal average of sediment texture of the 15 sampling stations is presented in Fig. 5.7. Station wise mean and standard deviation of sediment particles are given in Table 5.1c



Fig. 5.7: Seasonal Variation of Sediment Texture during the study period

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SI	Stations	%sand		%silt		% Clay	
No:		Mean	± SD	Mean	± SD	Mean	± SD
1	Thannrmukkm	32.02	0.19	46.81	1.39	21.18	1.20
2	Vaikom	53.95	0.25	35.16	4.24	10.90	4.00
3	Aroor	9.96	1.07	27.10	0.30	62.94	0.78
4	Valnthakad	39.89	16.66	25.45	4.79	34.66	11.87
5	Kmblngi	50.31	0.09	20.09	0.23	29.61	0.14
6	Kndkdvu	62.04	2.69	21.97	1.27	16.00	1.42
7	Thevara	9.00	0.06	41.18	1.25	49.83	1.19
8	Thpmpdy	15.95	0.71	37.61	2.03	46.45	2.74
9	Brmouth	41.04	0.18	40.84	0.05	18.13	0.23
10	Mar Sci	9.14	0.31	35.44	1.73	55.43	1.42
11	Vllrpdm	32.19	1.37	42.08	0.14	25.73	1.51
12	Vypin	26.80	1.22	24.27	1.32	48.94	0.10
13	Vrppzha	43.93	0.17	32.60	2.19	23.47	2.36
14	Munbm	49.30	1.38	36.01	2.69	14.70	1.31
15	Azhikde	41.36	1.09	51.75	1.86	6.90	0.77

 Table 5.1c: Station wise Mean and Standard deviation of percentage composition of sediment



Fig. 5.8: Mean variations (Mean ±SD) of salinity for all the 15 stations during the study period



Fig. 5.9: Mean variations (Mean ±SD) of Water Temperature for all the 15 stations during the study period





Fig. 5.10: Mean variations (Mean ±SD) of pH for all the 15 stations during the study period



Fig. 5.11: Mean variations (Mean ±SD) of Sediment Temperature for all the 15 stations during the study period



Fig. 5.12: Mean variations (Mean ±SD) of Moisture content for all the 15 stations during the study period



Fig. 5.13: Mean variations (Mean ±SD) of Organic carbon content for all the 15 stations during the study period

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Fig. 5.14: Mean variations (Mean ±SD) of Sediment Texture for all the 15 stations during the study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	3355.734	14	239.695	15.794	.000
Season	7775.020	2	3887.510	256.163	.000
Error	2473.678	163	15.176		
Total	13604.432	179			

Table 5.2a: ANOVA of salinity during the study period

Table 5.2b: ANOVA	of Water	Temperature	during	the study	period
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Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	470.478	14	33.606	14.311	.000
Season	1.478	2	.739	.315	.730
Error	382.772	163	2.348		
Total	854.728	179			

Table 5.2c: ANOVA of pH during the study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	36.546	14	2.610	11.743	.000
Season	.624	2	.312	1.404	.249
Error	36.236	163	.222		
Total	73.406	179			

Table 5.2d: A	ANOVA of Sedim	ent Temperatur	e during the	e study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	294.144	14	21.010	9.319	.000
Season	1.678	2	.839	.372	.690
Error	367.489	163	2.255		
Total	663.311	179			

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Table 5.2e: ANO	VA of Moisture conf	tent during the	study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	1035.134	14	73.938	81.704	.000
Season	16.143	2	8.071	8.919	.000
Error	147.508	163	.905		
Total	1198.785	179			

Table 5.2f:	ANOVA	of Organic	carbon	during t	he study	neriod
1 abic 3.21.		or organic	carbon	uuring u	nestuay	periou

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	313.586	14	22.399	953.062	.000
Season	.056	2	.028	1.181	.310
Error	3.831	163	.024		
Total	317.472	179			

Table 5.2g: ANOVA of %sand during the study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	48561.518	14	3468.680	256.808	.000
Season	121.757	2	60.878	4.507	.012
Error	2201.624	163	13.507		
Total	50884.899	179			

Table 5.2h: ANOVA of % silt during the study period

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	12169.271	14	869.234	332.951	.000
Season	40.740	2	20.370	7.803	.001
Error	425.543	163	2.611		
Total	12635.555	179			



Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Station	50997.389	14	3642.671	267.732	.000
Season	37.542	2	18.771	1.380	.255
Error	2217.719	163	13.606		
Total	53252.651	179			

Table 5.2i: ANOVA of % clay during the study period



Fig. 5.15a: Draftsman plot showing the interactions of environmental variables for all the 15 stations during the monsoon period.



Fig. 5.15 b: Draftsman plot showing the interactions of environmental variables for all the 15 stations during the Post monsoon period.



Fig. 5.15 c: Draftsman plot showing the interactions of environmental variables for all the 15 stations during the Pre monsoon period.



Table 5.3.	Correls period.	ation betw	veen the v	vater and	sediment	quality f	arameter	s during	the study
				Pearson (Correlation				
	Salinity	W.Temp	рН	S.Temp	Moisture	Org. C	%sand	%silt	%clay
Salinity	1	0.279**	0.386**	0.192**	0.264**	-0.146	-0.043	0.116^{**}	-0.015**
W.Tmp	0.279**		0.362**	0.684**	0.190*	-0.316**	0.073**	0.217	-0.177**
Нd	0.386**	0.362**	1	0.395**	0.268**	-0.269**	-0.081**	0.367**	-0.100
S.Temp	0.192**	0.684**	0.395**	1	0.158*	-0.294**	0.002**	0.316**	-0.156**
Moistur	0.264**	0.190*	0.268**	0.158*	1	-0.007	-0.590**	0.123*	0.517**
Org.C	-0.146	-0.316**	-0.269**	-0.294**	-0.007	1	-0.286	-0.492**	0.520^{**}
%Sand	-0.043	0.073	-0.081	0.002	-0.590**	-0.286**	1	-0.202	-0.879
%Silt	0.116	0.217**	0.367**	0.316**	0.123	-0.492**	-0.202	1**	-0.289**
%Clay	-0.015	-0.177*	-0.100	-0.156*	0.517**	0.520^{**}	-0.289*	-0.289*	
**Significat	ntly differe tly differer	ent at 1% levent at 5% levent	vel (P<0.01) sl (P<0.05)						

Sl No:	Stations No	Station Name	Texture
1	Station.1	Thanneermukkom	Silty
2	Station.2	Vaikom	Sandy
3	Station.3	Aroor	Clayey
4	Station.4	Valanthakad	Sandy
5	Station.5	Kumbalangki	Sandy
6	Station.6	Kandakkadavu	Sandy
7	Station.7	Thevara	Clayey
8	Station.8	Thoppumpady	Clayey
9	Station.9	Barmouth	Sandy
10	Station.10	Marine Science Jetty	Clayey
11	Station.11	Vallarpadam	Silty
12	Station.12	Vypin	Clayey
13	Station.13	Varappuzha	Silty
14	Station.14	Munambam	Sandy
15	Station.15	Azhikode	Silty

Table 5.4: Sampling stations and their predominant textural class

5.3.4. Substratum and Distribution of crabs

From the diversity study, it was observed that, a total of 23 species of brachyuran crabs belonging to 16 genera and 8 families occur in the Cochin backwaters. Each crab exhibited different inhabitation patterns and are distributed in the backwaters according to their habitat preferences. It was observed that there existed seasonal as well as spatial variations in the distribution and abundance of crabs, which in turn was influenced by the ecological parameters.

The Global BEST analysis was conducted to establish the relationship between the environmental parameters and the distribution of crabs in seasonal patterns, which are presented in Tables. 5.5 a, b, c. The results showed that the best correlation coefficient (Rho) for the distribution of crabs was 0.52 during the monsoon (Fig.5.16), 0.639 during the post monsoon (Fig.5.17) and 0.508 during the pre monsoon period (Fig.5.18).

	Distribution of Crabs						
SI No:	Variables	Variables Selected	Best Correlation Values (Rho)				
1	Log (Salinity)	1,5,6,8	0.572				
2	Water Temperature	1,3,5,6,8	0.545				
3	pН	1,5,6,8,9	0.541				
4	Sediment Temperature	1,3,5,6,8,9	0.530				
5	Moisture content	1,5-8	0.529				
6	Organic Carbon	5,6,8	0.528				
7	Log (Sand)	1,3,5-8	0.518				
8	Log (Silt)	1,5,6	0.518				
9	Log (Clay)	1,6,8	0.499				

 Table 5.5 a: BEST (Biota and environmental Matching) Results for distribution of crabs during Monsoon Season

 Table 5.5 b: BEST (Biota and environmental Matching) Results for distribution of crabs during Post monsoon Season

	Distribution of Crabs						
SI No:	Variables	Variables Selected	Best Correlation Values (Rho)				
1	Log (Salinity)	1,2,5,6,8,9	0.631				
2	Water Temperature	1,2,5,6,8	0.607				
3	pН	1,2,5,6,9	0.601				
4	Sediment Temperature	1,2,5,8,9	0.597				
5	Moisture content	1,2,5,6	0.582				
6	Organic Carbon	1,2,5,9	0.580				
7	Log (Sand)	1,2,5,8	0.577				
8	Log (Silt)	1-3,5,6,8,9	0.571				
9	Log (Clay)	1,5,8,9	0.569				

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Distribution of Crabs							
SI No:	Variables	Variables Selected	Best Correlation Values (Rho)				
1	Log (Salinity)	1,3,9	0.508				
2	Water Temperature	1,9	0.508				
3	рН	1,3,5,9	0.481				
4	Sediment Temperature	1,3,4,9	0.471				
5	Moisture content	1,4,9	0.467				
6	Organic Carbon	1,3,8,9	0.465				
7	Log (Sand)	1,3-5,9	0.464				
8	Log (Silt)	1,3,4,8,9	0.458				
9	Log (Clay)	1,3,7,9	0.454				

Table 5.5 c:	BEST	(Biota	and	environmental	Matching)	Results	for
distribution of crabs during Pre monsoon Season							



Fig. 5.16: Histogram showing the BEST results during the monsoon season (Rho value = 0.52)



Fig. 5.17: Histogram showing the BEST results during the Post monsoon season (Rho value = 0.639)



Fig. 5.18: Histogram showing the BEST results during the Pre monsoon season (Rho value = 0.508)

Similarity indices:

1) SIMPROF Test

The significance of the clusters created was tested by Similarity Profile (SIMPROF test). The clusters formed based on the seasonal

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pattern are given in Fig. 5. 19 (Monsoon), Fig. 5.20 (Post Monsoon) and Fig. 5.21 (Pre Monsoon).

2) Non-Metric Multi-Dimensional Scaling plots

Multi-dimensional scaling (MDS) plot was constructed for finding out the similarity ranking of sampling stations with respect to crab distribution, in seasonal pattern. The similarity obtained by the Bray-Curtis similarity plots are strongly supported by the MDS plots. MDS plot for the distribution of crabs gave a good ordination having the stress value of 0.08 in monsoon (Fig. 5.23), 0.14 (Fig.5.24) in post monsoon and 0.12 (Fig. 5.25) in pre monsoon months.

Bubble plots were constructed using the abundance data, for showing the relative abundance of prominent crab species in different stations (Fig. 5.22a- 5.22w)



Fig. 5.19: Dendrogram showing the cluster similarity of stations with respect to crab distribution during monsoon.



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Fig. 5.20: Dendrogram showing the cluster similarity of stations with respect to crab distribution during Post monsoon.



Fig. 5.21: Dendrogram showing the cluster similarity of stations with respect to crab distribution during Pre monsoon.

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Habitat Ecology and Habitat Preferences of Brachyuran Carbs of Cochin Backwaters



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Habitat Ecology and Habitat Preferences of Brachyuran Carbs of Cochin Backwaters



























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Fig. 5.22: (a) - 5.22 (w). Bubble plots showing the relative abundance of crabs with respect to the sampling stations.





Fig. 5.23: Non-Metric Multi Dimensional Scaling (MDS) ordination plot (stress = 0.08) for monsoon.



Fig. 5.24: Non-Metric Multi Dimensional Scaling (MDS) ordination plot (stress = 0.14) for Post monsoon.



Fig. 5.25: Non-Metric Multi Dimensional Scaling (MDS) ordination plot (stress = 0.12) for Pre monsoon.

5.4 Discussion

Habitat characteristics clearly affects the distribution of crabs, while the reverse is also true (Ravichandran *et al.*, 2007). It is clearly evident from the findings of the present work that the distribution of brachyuran crabs of a region is highly dependent on the habitat type, ecological parameters, substratum characteristics as well as the presence of mangroves in that region. Crabs depend directly on mangroves for survival and are adapted to the special sediment conditions, tidal fluctuations and varying salinities (Cocelho, 1967).

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During the study period, the water quality parameters viz., salinity and water temperature showed variations both seasonally and spatially, while pH exhibited spatial variation only. Salinity is one of the most important environmental variables which influence the life activity of aquatic organisms. It acts as a limiting factor for their survival, distribution and the life activities including feeding, reproduction etc. In the present study, salinity exhibited wide fluctuations both seasonally and spatially. In all the stations, during the monsoon period, salinity showed a comparatively lower value due to the dilution of water, owing to the heavy rainfall and strong inflow of the fresh water. The post monsoon period presented a trend of recovery and then salinity reached its maximum range during pre monsoon months. The study also revealed that those stations, which are closer to the sea and have higher tidal influence tends to exhibit a higher salinity, when compared to those stations which are distant from the sea. The stations located closer to the sea viz., Station 8: Thoppumpady, Station 9: Barmouth, Station 14: Munambam, Station 15: Azhikode showed a higher salinity throughout the year due to the higher tidal influence from the sea, when compared to Station 1: Thanneermukkom, Station 2: Vaikom, Station 13: Varappuzha, which are far away from the marine environment. The present findings are in affirmative with Chandramohan (1990), Saravanan (1999) and Menon et al. (2000).

Besides salinity, water temperature is an important ecological factor influencing the distribution and activities of an organism. In the present study, water temperature showed monsoonal minimum and pre monsoonal maximum. The fall in water temperature in the monsoon months can be attributed to the heavy rainfall and the resultant cold weather and the higher temperature during pre monsoon is due to the intense solar radiation. When observed spatially, three stations viz., Station 11: Vallarpadam, Station 14: Munambam and Station 15: Azhikode exhibited a higher temperature range. John (2010) also claims that the northern limb of the Cochin estuary registered a slight increase in the water temperature, when compared to the southern limb, due to its proximity to industrial discharge.

pH was found to be neutral throughout the year, at all the stations, except at Station 13: Varappuzha. Station 13: Varappuzha exhibited a lower pH range, throughout the year, owing to its vicinity to the industrial area Eloor and the resultant industrial pollution of the region. John (2010) also observed an acidic pH range in Varappuzha in her study conducted on Cochin backwaters.

Like the water temperature, sediment temperature also followed the same pattern of variations, both station-wise and season-wise. The sediment temperature was observed to be minimum during the monsoon period due to the precipitation and cold weather, while it showed its maximum range during pre monsoon, which can be attributed to the intense heat of sun solar in summer season. Spatial variation of sediment temperature also followed the same pattern as in the case of water temperature. When the 15 sampling stations were taken into account, those three stations namely, Station 11: Vallarpadam, Station 14: Munambam and Station 15: Azhikode, which recorded higher water temperature registered higher sediment temperature also. It was also observed that in most of the cases, the sediment temperature of a sampling station was slightly less than the corresponding water temperature of that station.

Moisture content showed wide fluctuations, station-wise, but did not exhibit season-wise variations. According to John (2010), the fine fractions of sediment retain more water than the coarser fraction and seasonal fluctuations in moisture content were not conspicuous within the stations. Moisture content of the sediments is greatly influenced by the texture as well as the amount of decaying plant matter of the sediments. It is evident that more the amount of fine clay matter and decaying plant matter, more will be the water retention capacity. The stations with a muddy substratum and those stations with luxuriant mangroves, recorded higher moisture content in the sediments. The stations which recorded high moisture content are Station 7: Thevara, Station 8: Thoppumpady, Station 9: Barmouth, Station 10: Marine Science Jetty, Station 11: Vallarpadam and station 12: Vypin. From the sediment analysis, it was observed that sediments of the stations 7, 9 and 10 are dominated by clay particles. Clay has maximum water retention capacity due to its smaller pore size, which enables them to hold the water within itself. Station 11 and Station 12 wass characterized by rich and lush mangroves. There are several virgin mangrove patches ornamenting these two stations and therefore experiences more litter fall. The decaying plant matter in the sediments traps more water and hence the water retention capacity reached its maximum value.

Similarly, the organic carbon content in the sediments varied significantly among the study stations, while it followed the same pattern throughout the year. It is obvious that organic carbon content was found to be maximum in the sediments of those stations ornamented with mangroves. The leaf litter from the mangroves decomposes in due course of time and adds up to the organic carbon content of the sediments. Station 3: Aroor and Station 4:

Valanthakad exhibited maximum organic carbon content than the other sampling stations. These stations are ornamented with owing to the luxuriant mangrove patches and hence the maximum organic carbon in these sediments is well justified.

The percentage compositions of sediments also showed high degree of variations among the stations, though season-wise variations were not significant. Although the percentage composition of the sediments varied with seasons, the variations were not considerable and insignificant. To be precise the dominant particle of a particular station remained the same. It is seen that the percentage of clay was slightly higher in the post monsoon and pre monsoon periods than the monsoon period. This may be due to the wilted off *Eichornia* which gets deposited to the substratum. The decaying plant matter adds to the sediments making it more clayey. A comparison of data on the sediment texture and organic carbon content revealed that finer the sediment, higher the organic carbon content. Similar result was observed earlier by Murthy and Veerayya (1972) and John (2010).

Based on the two-way ANOVA the variations in the water quality parameters, viz., salinity, water temperature and pH and the sediment parameters viz., sediment temperature, moisture content, organic carbon and sediment texture (%sand, % silt and % clay) are significant at 1% significance.

The results of the Pearson's correlation of the pooled data showed that salinity had a positive correlation with temperature, pH, sediment temperature and moisture content at 1% level (r = 0.279, 0.386, 0.192 and 0.264 respectively). It was also observed that water temperature and pH also exhibited a positive correlation at 1% level (r = 0.362). Besides salinity,

sediment temperature was positively correlated with water temperature and pH at 1% level (r = 0.682 and 0.395 respectively). From the comparatively higher r value (r = 0.682) between sediment temperature and water temperature, it is evident that these two parameters were strongly correlated. The sediment temperature of a sampling station was often found to record slightly lower value than the corresponding water temperature of that station. It was also observed that higher the water temperature resulted in higher sediment temperature and vice-versa. Moisture content in the sediments was found to be positively correlated with pH at 1% level (r = 0.268). The organic carbon content in the sediments gave a negative correlation with water temperature, pH and sediment temperature (r = -0.316, -0.269 and 0.294 respectively).

In the case of sediment texture, each constituent particles, viz., sand, silt and clay were separately correlated with the other environmental variables. The Pearson's correlation results showed that the percentage composition of sand particles in the sediments was positively correlated with water temperature and sediment temperature (r = 0.073 and 0.002 respectively) and negatively correlated with pH and moisture content (r = -.0.081 and -0.590 respectively) at 1% level. The percentage composition of silt particles showed a positive correlation with salinity, pH and sediment temperature (r = 0.116, 0.367 and 0.317 respectively) at 1% level and with moisture content (r = 0.123) at 5% level). The percentage composition silt particles exhibited a negative correlation with organic carbon content at 1% level (r = -0.492) and with the percentage composition of clay particles at 5% level (r = -0.289). The clay particles showed a positive correlation with moisture content and organic carbon at 1% level (r = 0.512 and 0.520

respectively), while a negative relation was observed with salinity, water temperature, sediment temperature and silt content at 1% level (r = -0.0.15, -0.177, -0.156 and -0.286 respectively). This was strongly supported by the Draftsman scatter plot (Fig 5.).

The Global Best study revealed that, rather than the water quality parameters, it was the sediment quality parameters, which influenced the distribution of crabs, which is in accordance with the findings of Ewa-oboho (1993), Botto and Iribane (2000), Pandya and Vachharajani (2010). Ravichandran *et al.* (2007) and Bandekar (2011) opined that the possible factors influencing the distribution of crabs are substrate suitability, tidal inundation, mangrove distribution and salinity.

The influence of sediment quality parameters on the distribution of crabs can be very well justified by the crawling and burrowing behavior of the crabs. The nature of the substratum, which helps in easy movement and burrowing, will be preferred by most the crabs, especially those crabs which exhibits a crawling movement like the grapsid crabs. Distribution of brachyuran crabs was observed to be the highest at the stations with muddy substratum namely, Station 3: Aroor and Station 12: Vypin with high organic carbon content and comparatively high amount of moisture. The muddy substratum enables ease in burrowing as well as crawling movement of the crabs, while organic carbon content influences the feeding habit. The burrowing crabs are mostly filter feeders and they mostly feed on the substratum available (Arya *et al.*, 2014). The correlation between the available nutrients in the substratum and the crabs is well established by Lee (1998) and Nordhaus *et al.* (2006). According to their findings, the

bioturbation activities of the crabs help to retain the organic matter and nutrients within the mangrove ecosystem. The influence of moisture content may be attributed to the requirement of oxygen for respiration in the case of burrowing crabs. Generally, crabs are capable of breathing both on land and water, but oxygen can often become a limiting factor in the case of burrowing crabs. The moist substratum provides fresh supply of oxygen for burrowing crabs, which is taken in with the help of hair tufts at the leg bases (Ravichandran *et al.*, 2007)

The percentage composition of sand, silt and clay is another factor which influences the crab distribution. According to Joel et al. (1985), the substratum suitability controls the zonation of crabs. It is also observed that, different crabs prefer different substratum type. In the present study, most of the crabs were found to prefer muddy substratum. This might be attributed to the ease in crawling and burrowing. The mud crabs, Scylla olivacea and S. serrata, Thalamita crenata, Uca annulipes, Macrophthalmus depressus, Neoepisesarma andersonii. Nanosesarma batavicum. N_{\cdot} mederii. *Metapograpsus* messor, M_{\cdot} maculatus, Heteropanope indica and Xenophthalmus pinnotheroides were found to prefer muddy sediments, while D. gracilipes and V. litterata were found to prefer sandy substratum. The sesarmid crab Neoepisesarma brockii was collected from all the stations irrespective of the sediment type which prevailed in the area. Though S. olivacea was also present throughout the study area, it was observed that they were abundant in the mangrove mud flats, where they occurred throughout the year, while at other stations; S. olivacea was found in low numbers and did not occur every time. The benthic crabs Elamenopsis alcockii, Ebalia malefactrix and Halicarcinus messor were
also found to prefer a muddy substratum. Ravichandran *et al.* (2007) reported that *Macrophthalmus* species prefer a muddy substratum, similarly *Uca* species also found to prefer a muddy substratum (Ravichandran and Kannupandi, 2007). Devi *et al.* (2013) reported that *Varuna litterata* inhabits regions dominated by mangrove patches and are found to be distributed in areas with a sandy substratum and higher organic carbon content and they prefer areas with close proximity to the sea and with good tidal influx.

Apart from the substratum characteristics, salinity also plays a significant role in the distribution of crabs in an area. According to Jones and Simons (1982), Omori et al. (1998), Mia et al., (2001), salinity is one of the most important factors controlling the vertical and horizontal distribution of grapsid crabs. In the present study the swimming crabs especially the, Portunus pelagicus, P. sanguinolentus, Charybdis feriata, C. lucifera and C. hoplites did not exhibit any peculiar inclination towards a particular sediment type. These are marine migrant species which invaded the Cochin backwaters during the post monsoon and pre monsoon periods when salinity reached its peak and disappeared when salinity fell. They are caught from the high saline areas of the backwaters, namely Station 5: Kumbalanghi, Station 6: Kandakkadavu, Station 7: Thevara, Station 8: Thoppumpady, Station 9: Barmouth, Station 12: Vypin, Station 14: Munambam and Station 15: Azhikode, irrespective of the sediment characteristics and the habitat type prevailing in these stations. The distribution of these marine migrants was solely influenced by the salinity and tidal influx which can be attributed to the proximity to the sea. Rao and Kathirvel (1972) reported the occurrence of the marine inhabitant P.

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pelagicus in the Cochin backwaters during the post monsoon, while Kurup *et al.* (1990) reported its appearance during the high saline premonsoon period. Devi *et al.* (2014) reported the invasion of six species of marine crabs viz., *Portunus pelagicus, P. sanguinolentus, Charybdis feriata, C. lucifera, C. hoplites* and *Doclea gracilipes* into the backwaters, when salinity shoots up. Besides the water and sediment quality parameter, the most important factor influencing the distribution of crabs is the presence of mangroves. From the present study, diversity as well as distribution of crabs were found to be the maximum in mangrove zones viz., Station 3: Aroor, Station 4: Valanthakad, Station 5: Kumbalanghi, Station 6: Kandakkadavu Station 11: Vallarpadam and Station 12: Vypin when compared to other stations which were devoid of mangroves. Crabs form the most abundant macrofauna in the mangroves and are considered to be the valuable asset to the mangrove ecosystem (Verwey, 1930, Macnae, 1968).

It is evident that the environmental parameters play a critical role in the distribution and abundance of crabs. Therefore, it is assumed that the sampling stations with similar environmental conditions exhibit similarity in crab distribution also. From the SIMPROF test, it is clear that the stations which exhibited similar environmental conditions and habitat type formed a cluster with respect to the diversity and distribution of crabs. Bray-curtis similarity plots for the monsoon season gave 11 clusters of similarity, having the similarity percentage ranging from 35.69- 90.21%. The highest similarity of 90.21% was obtained between Station 1: Thannermukkom and Station 2: Vaikom, followed by 83.25% between Station 5: Kumbalanghi and Station 6: Kandakkadavu. The similarity could be attributed to the similarity in environmental parameters, which in turn influence the crab resource of the area. Station 1: Thannermukkom and Station 2: Vaikom exhibited an oligohaline condition during the monsoon months with a mean salinity of 0.91 ± 0.3 and 1.19 ± 0.27 respectively, owing to the distance from the sea. The substratum characteristics were also found to be similar for both the stations. Station 1 recorded a mean moisture content of 4.77 ± 0.62 , while station 2 recorded 4.46 ± 0.31 . Another important factor which influences the crab biota is the organic carbon content of the sediments. Both the stations exhibited a lower organic carbon content in the sediments, the mean value being 0.61 ± 0.01 for Station 1 and 0.38 ± 0.00 for Station 2. However, the percentage composition of sediments was found to be varying for both the stations. The substratum of Station 1 was found to be dominated by silt followed by sand, while of Station 2, it was dominated by sand followed by silt. When the habitat type of Station 1 and Station 2 were considered, both the stations were located upstream and devoid of mangroves. Only two species of crabs were found to representing at both these stations, namely, Neoepisesarma brockii and Scylla olivacea. When Station 5: Kumbalanghi and Station 6: Kandakkadavu (83.25% similarity) were considered, the environmental variables, which influenced the crab distribution was found to be similar. Station 5 recorded a mean salinity of 5.38 ± 0.18 , while station 6 recorded 6.5 ± 0.35 . Likewise, the moisture content and the organic carbon content of the sediments were also comparable. Station 5 recorded mean moisture of 8.11 ± 0.01 % and station 6 recorded $7.3 \pm 0.17\%$. The mean organic carbon content of station 5 was 1.25 ± 0.12 , while that of Station 6 was recorded as 1.19 ± 0.02 . The sediment texture of both the stations was sandy in nature. All these factors were supplemented by the presence of luxuriant mangroves in both these

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stations. Also, the stations experienced a good tidal influence from the sea. Hence, it is obvious that these stations exhibit similarity in distribution of crabs too. During the monsoon season, 12 species of crabs were reported from Station 5, viz., *S. serrata, S. olivacea, U. annulipes, N. mederii, N. brockii, N. batavicum, N. andersonii, M. depressus, V. litterata, X. pinnotheroides, E. malefactrix, E. alcockii* and *H. messor*, while at Station 6 11 species of crabs were recorded in total. Except *U. annulipes*, all the other crab species recorded from Station 5 has been collected from Station 6. The third cluster was represented by Station 7: Thevara and Station 9: Barmouth with 77.74% similarity. These stations possess similar ecological parameters viz., salinity, pH and muddy substratum with low organic carbon, and high moisture content, hence the distribution of crabs was also found to be highly similar. The crabs representing this cluster were *Ebalia malefactrix, Elamenopsis alcockii, Halicarcinus messor* and *Xenophthalmus pinnotheroides*.

The Bray-curtis similarity plot for the post monsoon season gave 14 clusters of similarity having the similarity percentage ranging from 34.04-89.58%. Among these, 5 clusters gave a similarity of above 80%. The highest similarity of 89.58% was obtained between Station 1: Thannermukkom and Station 2: Vaikom, followed by 85.39% between Station 7: Thevara and Station 10: Marine Science Jetty. The clusters include Station 5: Kumbalangi and Station 12: Vypin with 84.78% similarity, Station 14: Munambam and Station 15: Azhikode with 83.83% similarity, Station 8: Thoppumpady and Station 9: Barmouth with 82.77% similarity. As described earlier, Stations 1 and 2 recorded a similar range of salinity, moisture content and organic carbon content in its sediments. Two species of crabs were found to occur in these stations during the post

monsoon period namely, N. brockii and S. olivacea. Hence the highest similarity ranking for these two stations is well justified. Station 7 and 10 was characterized by muddy substratum with low organic carbon, and high moisture content. Salinity, another environmental variable was also comparable in both these stations and hence the distribution of crabs was also found to be highly similar. The crabs representing this cluster were Ebalia malefactrix, Elamenopsis alcockii, Halicarcinus messor and Xenophthalmus pinnotheroides. The third cluster is Station 5: Kumbalangi and Station 12: Vypin with 84.78% similarity. Aside from the similar environmental parameters, these two stations are characterized with rich mangroves and also found to experience good tidal influx from the sea. The fourth cluster was formed by Station 14: Munambam and Station 15: Azhikode with 83.83% similarity. These two stations record a high salinity during the post monsoon season; devoided of mangrove patches and the sediments characterized with low moisture content and moderate amount of organic carbon. A few marine migrant species were recorded from these two stations during the post monsoon season, which may be attributed to the high salinity which almost reaches the marine condition and their proximity to the sea, which enables good tidal influx in both these stations. The marine migrants collected include P. pelagicus, P. sanguinolentus, C. feriata, C. lucifera, C. hoplites and D. gracilipes. All these factors contributed to the cluster formation in the case of Stations 14 and 15. The fifth cluster was formed by Station 8: Thoppumpady and Station 9: Barmouth with 82.77% similarity. Apart from the similarity in environmental parameters, both these stations are devoid of mangrove patches but exhibited excellent tidal influx from the sea. During the post monsoon period, these two

stations also reported the invasion of few marine migrant species namely, *P. pelagicus, P. sanguinolentus* and *C. feriata*, which can be attributed to the high salinity which almost reached the marine condition as well as the tidal influence from the sea.

The Bray- Curtis similarity plot for the pre monsoon period gave 14 clusters of similarity having the similarity percentage ranging from 27.43-97.05%. As in the case of monsoon and post monsoon months, the highest similarity was recorded between Station 1: Thanneermukkom and Station 2: Vaikom (97.01%) followed by 85.39% at Station 5: Kumbalanghi and Station 12: Vypin. Both these clusters records similar environmental conditions as described earlier and therefore the distribution of crabs are also found to be similar.

The similarity obtained by the Bray-Curtis similarity plots are strongly supported by the MDS plots. Multi-dimensional scaling (MDS) plot was constructed for finding out the similarity ranking of sampling stations with respect to crab distribution, in seasonal pattern. Sampling stations showing similarities were grouped together and dissimilar ones, far away. Stress value < 0.1 corresponds to a good ordination with no real prospects of misinterpretation. The results showed a good ordination between the stations for post monsoon and pre monsoon with a stress value of 0.14 and 0.12, respectively, but for monsoon, stress value was 0.08. When the MDS plots of monsoon season is taken into account, Station 1: Thanneermukkom and Station 2: Vaikom showed 80% similarity. Likewise, Station 5: Kumbalanghi and Station 6: Kandakkadavu too exhibited 80% similarity. When the MDS plot of post monsoon period was considered, 5 clusters gave

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80% similarity. The clusters were Station 1: Thanneermukkom and Station 2: Vaikom; Station 8: Thoppumpady and Station 9: Barmouth; Station 7: Thevara and Station 10: Marine Science Jetty; Station 5: Kumbalanghi and Station 12: Vypin and Station 14: Munambam and Station 15: Azhikode. The MDS plot for pre monsoon season gave 2 clusters with 80% similarity, which included Station 1: Thanneermukkom and Station 2: Vaikom and Station 5: Kumbalanghi and Station 12: Vypin. This similarity can be attributed to the similarity in environmental conditions, which in turn influenced the similarity in crab distribution. The relative abundance of the 23 crab species at the 15 sampling stations is represented with the help of bubble plots, which is dependent on the varying ecological conditions of the stations.

Chapter **6** TAXONOMIC ACCOUNT OF BRACHYURAN CRABS OF COCHIN BACKWATERS

- 6.1 Introduction
 6.2 Materials and Methods
 6.3 Results
 - 6.4 Discussion

6.1 Introduction

Knowledge on taxonomy is very important as proper identification of species enables their right classification and placing them into different hierarchical levels. For economically important species, taxonomical identification is essential for developing successful culture techniques, stock assessment and larval recruitments. In ecological point of view, proper taxonomic study is a pre-requisite for understanding the evolutionary patterns and conservation of species, especially threatened ones. Taxonomy is often considered as a relatively unexciting topic for various reasons. However, taxonomic study serves an important function of ordering nature into a practical and generally useful system (Jayabaskaran *et al.*, 1999). Systematics in the wide sense also has the more challenging task of understanding the mechanisms of phylogeny (Sneath and Sokal, 1973).

Classification of decapod crustaceans was first attempted by Latreille (1802), who divided the decapods into Macrura (long tailed decapods) and Brachyura (short tailed decapods). A third group was added to this classification by Milne Edwards (1834), and designated them as the Anomura (differently tailed). The members belonging to the Infra order Brachyura are considered as the 'true crabs'. The brachyuran crabs are characterized by a reduced abdomen, which is often referred to as its short tail, which is entirely hidden under the thorax and have 4 pairs of well developed walking legs.

Further classification of the brachyurans was done by Milne Edwards (1834), who divided this group into four divisions viz., Oxyryncha, Cyclometopa, Catometopa and Oxystomata. Meanwhile De Haan (1833-1849) proposed another classification of Brachyurans into two divisions, Oxystomata and Brachygnatha, based on its mouth parts. Later, Several carcinologists dealt with Brachyuran taxonomy and proposed various types of classifications. Dana (1852 a, b,c) classified the true crabs into five subtribes namely Maioidea, Cancroidea, Corystoidea, Grapsoidea and Leucosoidea. A completely new classification of Brachyura was then given by Miers (1886), who grouped Brachyura into four sub-tribes, viz., Oxystomata or Leucosiidae, Oxyrhyncha or Maioidea, Catometopa or Ocypodidae and Cyclopmetopa or Cancroidea. Bouvier (1896) classified Brachyura in five tribes viz., Dromiacea, Oxystomata, Corystoidea, Brachyrhyncha and Oxyryhncha. Later, Borradile (1907) classified them into three sub tribes as Dromiacea, Oxystomata and Brachygnatha, which is widely accepted by the Zoologists. With a slight modification to Borradile's scheme of classification, Balss (1957) proposed another classification, which included four sub-tribes of Brachyuran crabs, namely, Dromiacea, Brachygnatha, Oxystomata and Hapalocarcinoidea. Later in 1969, Glaessner provided another classification scheme, where Brachyuran crabs were classified into 5 sections, Dromiacea, Brachyrhyncha, Oxystomata, Oxyrhyncha and Corystoidea. Sakai (1976) classified Brachurans into seven sub-sections, viz., Dromiacea, Brachyrhyncha, Oxystomata, Oxyrhyncha, Gymnopleura, Hapalocarcinoidea and Corystoidea. Guinot (1977, 1978) proposed a new classification of Brachyura, where this group is divided into three sections namely, Podotremata, Heterotremata and Thoracotremata, on the basis of the position of female and male genital openings. Later, another classification was provided by Rice (1980) on the basis of the brachyuran larval characteristics.

In India, taxonomic studies on crabs are very scanty and sparse. Several workers have dealt with the taxonomy of the commercially important mud crab species (Kathirvel, 1981; Radhakrishnan and Samuel, 1983; Joel and Sanjeevaraj, 1983; Kathirvel and Srinivasagam, 1992; Kathirvel *et al.*, 2004; Devi and Joseph, 2013a; Trivedi and Vachhrajani, 2013; Padate *et al.*, 2013; Balasubramanian *et al.*, 2014; Mandal *et al.*, 2014 a, b; Devi and Joseph, 2015). However, an extensive study on the taxonomy of crabs belonging to other families has not been attempted much. A perusal on the previously published works proves that the available taxonomic works on brachyuran crabs of Indian waters with a valid identification key include the works of Chhapgar (1957), Sethuramalingam and Ajmal Khan (1991) and Jayabaskaran *et al.* (1999). In this context, the present study holds much significance, as it provides a proper systematic account of brachyuran crabs of Cochin Backwaters.

6.2 Materials and Methods

The brachyuran crabs are usually distinguished on the basis of morphological characters such as their spination, transverse ridges and specific markings. The morphological characters are highly influenced by the growth and life stages of the species. Hence, besides these morphological characters, various other powerful taxonomic tools are needed to be employed for the proper identification of species. In the case of brachyuran crabs, first male pleopods or the third maxillipeds can be used as an important taxonomic tool (Chhapgar, 1957, Sethuramalingam and Ajmal Khan, 1991).

The materials and methods followed for the study has been described in Chapter 3 (section 3.4.3)

6.3 Results

The present study reveals that the first male pleopods and the shape of the male abdomen serve as a powerful taxonomic tool in the species level identification of brachyuran crabs.

Based on the morphological characters coupled with the structure of third maxillipeds, first pleopods of males and the shape of male abdomen, an identification key is presented for the brachyuran crabs that are found to occur along the study area the Cochin backwaters and the associated mangrove patches. The identification key includes the details of 23 species of brachyuran crabs which belongs to 16 genera and 8 families, which have been enlisted from the study area.

6.3.1 Key to the families of Brachyuran crabs of Cochin Backwaters

- 1. Carapace transversely hexagonal or transversely ovate (Fig.6.1a, 6.1b).

- 2. Carapace transversely rectangular or squarrish or trapezoidal (Fig.6.7 a,b,c).
- 2A. Orbital region elongated and cover a major portion of the anterior margin of the carapace, making the frontal region very narrow. Eye stalks are slender and long. The third maxillipeds with a quadrate merus and ischium. The merus is smaller than the ischium. Merus and ischium is completely fused to each other. Carpus of the third maxilliped remain separated from the merus (Fig 6.8)
- 2B. Orbits are short. Frontal region broad and slightly deflexed downwards. Eyestalks are short and broad. Chelipeds are equal in

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size. The third maxillipeds with a subcircular merus and ischium, however narrow. (Fig. 6.9) Third maxillipeds leave a prominent gap in between and bears minute hairs all over......Grapsidae

- 3. Carapace circular, sub-circular or pyriform (Fig 6.12 a, b)

- 3C. Carapace circular or sub circular, poorly calcified. Frontal region narrow and not modified to form rostrum. The third maxillipeds is broad and flattened. The merus and the ischium fused together to form a single structure (Fig 6.15).....Leucosiidae



Fig. 6.1 a: Carapace type- Transversely hexagonal



Fig. 6.1 b: Carapace type- Transversely ovate



Fig. 6.2: Spinous frontal region of Portunid crabs





Xanthidae





Fig. 6.7 a: Carapace type- Transversely rectangular

Fig 6.7 b: Carapace type- Squarrish



Fig. 6.7 c: Carapace type- Trapezoidal





Fig. 6.9: Third maxilliped of Family Grapsidae

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Fig. 6.15: Third maxilliped of Family Leucosidae

6.3.2. Key to the sub families, genera, sub genera and species of Brachyuran crabs of Cochin Backwaters

6.3 2.1. Family Portunidae

Key to subfamily of Family Portunidae

Anterolateral borders of the carapace cut into 4 to 9 teeth. Frontal region broad and spinous. Chelipeds longer than the walking legs. Fifth pair of legs paddle shaped, adapted for swimming......Subfamily Portuninae

Key to the genera and species of the subfamily Portuninae

- I Anterolateral border cut into 9 teeth (Fig 6.16)

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Key to the species of Genus Scylla





Fig. 6.17: Infated propodus of Genus *Scylla*



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- IB. 9th anterolateral teeth elongated and modified to spine-like......Portunus

Key to the species of Portunus

- IB.2. Carapace characterized with three large blood red spots (Fig 6.29). No spine on the posterior border of the cheliped. The male abdomen is narrow; third, fourth and fifth segments fused and immovable (Fig 6.30). The first male pleopod is quite slender and straight and have no sharp bend on near the tip. The margins of the tip is characterized with fine hairs (Fig (6.31)...... Portunus sanguinolentus
- II. Anterolateral teeth cut into 6 teeth (Fig 6.32).....Charybdis

Key to subgenus Charybdis

- IIA. Posterior border of the carapace curved (Fig 6.33).....Subgenus *Charybdis*
- IIB. Posterior border of carapace straight and form an angular junction with the posterolateral border(Fig 6.34).Subgenus *Goniohellenus*

Key to the species of Subgenus Charybdis.

Key to the species of Subgenus Goniohellenus

Last anterolateral teeth elongated to form a spine, which resembles Genus *Portunus* (Fig 6. 42). The male abdomen is triangular, broader at the base and pointed while reaching the tip. (Fig 6.43). The first male pleopod is broad at the base, having elegant curves to the middle portion and blunt tip. The tip is fringed with hairs (Fig 6.44).....Charybdis (Goniohellenus) hoplites

III. Anterolateral teeth cut into 5 teeth (Fig 6.45).....Thalamita

Key to the species of Genus Thalamita



Fig. 6.23: Two prominent spines on the outer margin of the carpus of the cheliped in *Scylla serrata*

Fig. 6.24: Bluntly pointed frontal lobes in Scylla serrata



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Fig. 6.34: Angular junction formed by the posterolateral border with the posterior region of carapace in sub genus *Goniohellenus*



Fig. 6.35: Carapace marked with a 'cross' sign in *Charybdis (Charybdis) feriata*

Fig. 6.36: First pair of anterolater teeth bifurcated in *Charybdis* (*Charybdis*) feriata



2 cm

Fig. 6.37: Male abdomen- Charybdis (Charybdis) feriata

Fig. 6.38: First male pleopod-Charybdis (Charybdis) feriata

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Fig. 6.39: Carapace marked with 4 white spots in Charybdis (Charybdis) lucifera





Fig. 6.40: Male abdomen- Charybdis (Charybdis) lucifera





Fig. 6.42: Portunus like spine in subgenus Charybdis (Gonioehellenus) hoplites



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1.5 cm







Fig. 6.46: Male abdomen- *Thalamita* crenata



Fig. 6.45: Carapace of Genus Thalamita



Fig. 6.47: First male pleopod-Thalamita crenata

6.3.2. 2. *Family Xanthidae* Key to the subfamily of Family Xanthidae

I. Carapace transversely oval. Anterolateral border of carapace not longer than the posterolateral border.....Subfamily Pilumninae

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Key to the Genera and species of Subfamily Pilumninae

IA. Carapace convex and glabrous. Frontal region straight and truncate. Anterolateral borders cut into four teeth......*Heteropanope*

Carapace with two parallel transverse ridges and beaded with fine granules (Fig 6.48). Chelipeds extremely unequal and studded with pearly granules (Fig 6.49) The male abdomen is pyramid shaped, with second to fifth segments fused and immovable (Fig 6.50). The male pleopod is slender and curved elegantly at the tip, with few hairs at the inner margin. (Fig 6.51)

6.3.2.3. *Family Ocypodidae* <u>Key to subfamily of Family Ocypodidae</u>

 Carapace subquadrilateral, Anterolateral region of the carapace broder than the posterolaterals. Orbits large. Eyestalks long and slender. Chelipeds unequal. Flagella of antennae reduced and completely hidden under the frontal region......Subfamily Ocypodinae

Key to the genera and species of subfamily Ocypodinae

Key to the species of Genus Uca

Carapace black with cream or yellow dotted stripes. Chelae rosy pink in colour, and are enormously unequal. A tooth is present near the tip of the large chelae, giving the finger tip a notched truncate appearance (Fig 6.52). The male

II. Carapace transversely rectangular. Orbits large, reaches extreme width of carapace with remarkable long eyestalk. Chelipeds are sub equal in size.....Subfamily Macrophthalminae

Key to the genera and species of subfamily Macrophthalminae

Key to the species of Genus Macrophthalmus



Fig. 6.52: Notched tip of the larger chelae of Uca annulipes



Fig. 6.57: First male pleopod- Macrophthalmus depressus

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6.3.2.4. Family Grapsidae

Key to subfamily of Family Grapsidae

Key to the genus and species of subfamily Grapsinae

Key to the species of genus Metapograpsus

Lateral margin of the carapace less converging towards the posterior region (Fig 6.62). Chelipeds are slender and bright violet in colour. Walking legs are large. Dactylus shorter than the propodus. Segments of the male abdomen from the first to penultimate are of same width, sides being parallel, last segment suddenly narrow to a point (Fig 6.63). Male pleopod coarse and thick. Tip of the pleopod form a separate hammer head shaped lobe with serrated margins (Fig 6.64)......*Metaposgrapsus maculatus*

II. Carapace transversely rectangular. Frontal region broad and markedly deflexed downwards. The third maxillipeds are slender and the ischium exhibits transverse hairy crestsSubfamily Sesarminae

Key to the genus and species of subfamily Searminae

Carapace without any anterolateral teeth. Posterolateral border of merus of the 4th and 5th pereopod with denticulation (Fig 6.65)*Nanosesarma*

Carapace with 1 or 2 anterolateral teeth. Posterolateral border of merus of the 4th and 5th pereopod without denticulation (Fig 6.66) *Neoepisesarma*

Key to the subgenera and species of Genus Nanosesarma

Key to the species of Genus Nanosesarma

Upper surface of the chelipeds show two oblique pectinated crest (Fig 6.67). Merus of the 4th pereopod with three acute spines (on long and two short) posterodistally (Fig 6.68). The male abdomen is bell shaped, with segments of equal width and telson too small (Fig 6.69). The first male pleopod is slender, getting broader at the middle portion and tapering at the tip. The pleopod is fringed with hairs at both the margins. Fig (6.70) *Nanosesarma (Beanium) batavicum*

Upper surface of palm of the cheliped with single pectinated crest, associated with numerous striae (Fig 6.71). Merus of the 4th pereopod with

four strong spines on its posterodistal border (Fig 6.72). The male abdomen is bell shaped, with segments of equal width and telson too small (Fig 6.73). The first male pleopod is stouter with broad and rounded tip (Fig 6.74). *Nanosesarma (Beanium) andersoni*

Key to the subgenera and species of Genus Neoepisesarma

Upper surface of the palm of the cheliped with pectinated crest, which runs continuously from the distal end to proximal end. Numerous long swollen dactylar tubercles arranged transversely in a continuous pattern (Fig 6.75)......Subgenus *Neoepisesarma*

Upper surface of the palm of the cheliped with pectinated crest only at the proximal region......Subgenus *Selatium*

Key to the species of Genus Neoepisesarma

Carapace quadrangular. A sulcus runs above the transverse dactylar tubercles, continuously about one third of the total length of the tubercles (Fig 6.76). The male abdomen is bell shaped, with segments of equal width and telson too small (Fig 6.77). The male pleopods stouted with round blunted tip and fringed with hairs (Fig 6.78)...... *Neoepisesarma (Neoepisesarma) mederi*

III. Carapace squarrish. Front broad, but not deflexed. The third maxillipeds with broad exopod and long ischium; No gap left in between; close the buccal cavern completely (Fig 6.82)Subfamily Varuninae

Key to the genus and species of Subfamily Varuninae

Key to the species of genus Varuna



Fig. 6.58: Rhomboidal gap in between the third maxillipeds in subfamily Grapsinae

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Fig. 6.61: First male pleopod-Metapograpsus messor



Fig. 6.63: Male abdomen-Metapograpsus maculatus



Fig. 6.60: Male abdomen-Metapograpsus messor



Fig. 6.62: Carapace- Metapograpsus maculatus



Fig. 6.64. First male pleopod-Metapograpsus maculatus

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Fig. 6.71: Pectinated crest on the upper surface of the palm of the chelipeds in *Nanosesarma (Beanium) andersoni*



Fig. 6.72: Merus of the 4th pereopod showing denticulation in Nanosesarma (Beanium) andersoni



Fig. 6.73: Male abdomen-Nanosesarma (Beanium) andersoni



Fig. 6.74: First male pleopod-Nanosesarma (Beanium) andersoni



Fig. 6.75: Pectinated crest on the upper surface of the palm of the chelipeds in sub genus *N. Neoepisesarma*

Fig. 6.76: Chelae of *Neoepisesarma* (*Neoepisesarma*) mederi showing the pattern of crests

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Fig. 6.77: Male abdomen-Neoepisesarma (Neoepisesarma) mederi



Fig. 6.78: First male pleopod-Neoepisesarma (Neoepisesarma) mederi





Fig. 6.79. Chelae of *Neoepisesarma* (*Seletium*) brockii showing the pattern of crests







Fig. 6.86: Male abdomen- Varuna litterata



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6.3.2. 5. *Family Pinnotheridae* <u>Key to subfamily of Family Pinnotheriade</u>

Carapace subtrapezoidal. Orbits situated longitudinally and parallel to each other......Subfamily Xenophthalminae

Key to the species of sub family Xenophthalminae





Fig. 6.90. First male pleopod- Xenophthalmus pinnotheroides

6.3.2.6. Family Majidae

Key to the subfamily of Family Majidae

Key to genus and species of subfamily Pisinae

Key to the species of Genus Doclea

Carapace discoid with a series of blunt spines arranged longitudinally along the middle line. Body and limbs covered with thick hairs. Frontal region extended to form rostrum. Chelipeds shorter than the other legs; second pair of legs is the longest and reaches about three to four times the length of the carapace (Fig 6.91).





Fig. 6.93: First male pleopod- Doclea gracilipes

6.3.2.7. Family Leucosiidae

Key to the species belonging to the family Leucosiidae



Fig. 6.94: Male abdomen- *Ebalia malefactrix*



Fig. 6.95: First male pleopod- *Ebalia* malefactrix

6.3.2.8. Family Hymenosomatidae

Key to the species of Family Hymenosomatidae

Carapace suboval and lateral walls of the carapace with two teeth, one at the anterolateral angle and the second at the posterolateral angle. The front region is extended to form a trilobed rostrum; the lateral lobes small Carapace circular with well defined grooves. Frontal region modified to form trilobed rostrum. Lateral margin cut into a single tooth on the anterior region. Chelipeds are heavy and the fingers of the chelae with a wide gape at the base. Male abdomen is dome shaped with a broad base and gradually narrowing towards the tip (Fig 6.98). The first male pleopod is stouter, with a broad base and appears twisted at the middle portion. The tip of the pleopod blunt (Fig 6.99)......*Elamenospsis alcocki*



Fig. 6.96: Male abdomen-Halicarcinus messor



Fig. 6.97: First male pleopod-Halicarcinus messor



6.3.3. Systematic Account on the brachyuran crabs of Cochin backwaters *Scylla serrata* (Forskal, 1775)

Frequent synonyms: *Cancer serratus* Forskål, 1775: 90; *Portunus (Scylla) serrata* De Haan, 1833-1849 (1835): 44.; *Scylla tranquebarica* var. *oceanica* Dana, 1852: 270. *Scylla serrata*, Miers, 1886: 185, De Man, 1888: 332, Alcock, 1899: 27, Chopra & Das, 1937: 391, Chhapgar, 1957: 17, pl. 5 a-c., Stephenson & Campbell, 1960: 111, fig. 2N, pl. 4, fig. 4, pls 5 N, Guinot, 1967: pl. 1, fig. 1., Sakai, 1976: 335, pl. 115. *Scylla oceanica* (Dana, 1852) Estampador, 1949: 101, pl. 1, fig.2. *Scylla serrata* var. *paramamosain* (Estampador, 1949), Serène, 1952: 134, fig. 1D, pl. 1, fig. 4, pl. 2, figs 4, D. *Scylla tranquebarica* (Fabricius, 1798) Joel & Raj, 1983: 39, figs 1, 3.

Materials Examined: Numerous specimens of both sexes (84 males and 62 females) from all the 15 sampling stations of the study area.

Measurements: 95mm-135mm carapace width for males

95mm-142mm carapace width for females

Diagnostic features: Carapace of the crab is smooth with conspicuous transverse ridges. Colour of the crab is pale green to olive green and the fingers of the chelae pale orange. H-shaped gastric groove is found to be deep. The frontal margin of the carapace is cut into bluntly pointed teeth, with rounded interspaces. Anterolateral margin of the carapace cut into 9 narrow teeth. Chelipeds are large and strong with its propodus inflated. The distinguishing character of the species is the presence of two prominent sharp spines on the distal half of outer margin of the carpus of the cheliped. The limbs of the crabs exhibited conspicuous hexagonal patterning in both sexes. This is found to be the largest among the *Scylla* species.

The first male pleopod of *S. serrata* exhibited two patches of thin and inconspicuous setae on the inner margin on its posterior end. The second male pleopod is smaller than the first male pleopod. The tip of the second male pleopod is bilobed and the pleopod lacks any setation.

Remarks: Taxonomy of *Scylla* has been controversial for a long time, since the morphological features which separate the species are rather subtle and sometimes difficult to recognize in smaller specimens. *S. serrata*, the largest among the 4 *Scylla* species was often misidentified to be *S. tranquebarica*. However, *S. serrata* can be easily distinguished by the two prominent spines on the carpus of its chelipeds, bluntly pointed frontal teeth and the conspiucuous hexagonal markings on its limbs.

Habitat: Brackish waters and mangrove mud flats

Distribution: Abundant in the backwaters and the associated mangrove mud flats, where mesohaline conditions are prevailing. They are found to

occur in the backwaters, even during the monsoon regions, when the salinity almost reaches zero, but only in small numbers. Collected from Aroor, Valanthakadu, Thevara, Thoppumpady, Marine Science Jetty, Barmouth, Vypin, Kumbalanghi, Kandakkadavu, Vypin, Vallarpadam, Munambam and Azhikode

Scylla olivacea (Herbst, 1796)

Frequent synonyms: *Cancer olivaceous* Herbst, 1794: 157, pl. 38, fig. 3. *Scylla serrata* (Forskål, 1775) Estampador, 1949: 99, pl. 1, fig. 1., Serène, 1952: 134, fig. 1C, pl.1, fig.3, pl.2, figs3,C., Joel & Raj, 1983: 39, figs.2, 4, 6, 8, 10 a-b. *Scylla olivacea* Keenan *et al.*, 1998: 233, figs. 7D, 8D, 9D, 14., Ng, 1998: 1118 (key), 1127,

Materials Examined: Numerous specimens of both sexes (127 males and 92 females) collected from all the 15 sampling stations of the study area.

Measurements: 95mm-115mm carapace width for males

95mm-107mm carapace width for females

Diagnostic features: Carapace of the crab is smooth. Colour of the carapace is crab grayish green, fingers and the last segments of the walking legs pale orange. The frontal margin is cut into rounded lobes. Anterolateral margin of the carapace cut into 9 broad teeth. Chelipeds are large and the propodus are inflated. The species is characterized by the presence of single spine on the distal half of outer margin of the carpus of the cheliped. Palm of the cheliped exhibits a pair of prominences on the dorsal margin behind the insertion of the dacytyl. The inner prominence is larger than the outer one. Hexagonal markings are weak or absent. The first male pleopod of the species exhibited a single patch of setae, which is thick and prominent. The second male pleopod is bilobed at its tip. Unlike, *S. serrata*, the pleopods are found to possess setation on the inner margin on its posterior region.

Remarks: *S. olivacea,* being the smaller species was initially misidentified as *S. serrata*. The species can be easily distinguished from *S. serrata* by the form of its frontal margin (rounded frontal teeth) and the cheliped armature (single prominent spine on the carpus of the chelipeds)

Habitat : mangrove mud flats.

Distribution: Usually occurs in the mangrove mud flats and the backwater region where a low salinity prevails. They are found to occur throughout the backwaters and are collected from all the study stations.

Portunus(Portunus) pelagicus (Linnaeus, 1758)

Materials Examined: Numerous specimens of both sexes (69 males and 53 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 104mm-121mm carapace width for males

93mm-112mm carapace width for females

Frequent Synonyms: Portunus pelagicus Fabricius, 1798: 367; Sankarankutty, 1961: 102(list), 103; Guinot, 1967: 10; Neptunus (Neptunus) pelagicus Miers, 1886: 173; Ortmann, 1893: 74; Kemp, 1915: 248; Balss, 1922: 107; Chhapgar, 1957.

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Diagnostic features: Carapace is blue coloured in males and sand coloured in female and exhibit a granulose pattern all over the carapace. Frontal region with four acute spines. Anterotaleral region cut into nine teeth of which the ninth teeth is elongated to form spine like. Fifth pair of walking legs are flattened to form a paddle shape, which is an adaptation for swimming.

Remarks: *P. pelagicus* is often confused with *P. trituberculatus*, which resembles the former one. However, *P. pelagicus* can be easily distinguished by the presence of 4 spines on the inner margin of merus of the cheliped. Moreover, in the study area, the occurrence of *P. trituberculatus* has not been reported so far, hence the question of confusion is not valid here.

Habitat: Marine.

Distribution: The lower reaches of backwaters, where salinity is comparatively high. Collected from Thoppumpady, Barmouth, Kandakkadavu, Kumbalanghi, Vypin, Munambam and Azhikode.

Portunus (Portunus) sanguinolentus (Herbst, 1783)

Materials Examined: Numerous specimens of both sexes (92 males and 81 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 72mm-93mm carapace width for males

67mm-89mm carapace width for females

Frequent Synonyms: Lupa sanguinolenta Milne Edwards H., 1834: 451; Neptunus sanguinolentus de Haan, 1833-1849 (1835): 38, de Man, 1888: 328, Alcock, 1899: 32. *Neptunus (Neptunus) sanguinolentus* Miers, 1886: 174, Ortmann, 1893: 75, Balss, 1922: 106, 107, Chhapgar, 1957: 18, pl. A, fig. 3, pl. 4 m-o; *Portunus (Portunus) sanguinolentus* Sakai, 1976: 338.

Diagnostic features: Carapace sand coloured, finely granulose and characterized by three large reddish round spots on the posterior half of the carapace, of which one is median and the other two lateral, each spot encircled by a white ring. Anterolateral margin cut into nine teeth of which, the ninth teeth elongated to form a spine. The fifth pair of legs is paddle shaped and adapted for swimming.

Remarks: This species can be easily distinguished from all the other crabs by its distinctive blood red spots on the carapace.

Habitat : Marine species.

Distribution: The lower reaches of backwaters, where salinity is comparatively high. Collected from Barmouth, Thoppumpady, Munambam and Azhikode

Charybdis (Charybdis) feriata (Linnaeus, 1758)

Materials Examined: Numerous specimens of both sexes (175 males and 132 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 66mm-127mm carapace width for males

54mm-113mm carapace width for females

Frequent Synonyms: Cancer cruciatus Herbst, 1794: 155, pl. 38, fig. 1; Portunus crucifer Fabricius, 1798: 364; Portunus (Oceanus) crucifer de Haan, 1833 pl. A. Charybdis crucifera Dana, 1852c: 286, Dana, 1852a: pl. 17, fig. 11a-c; *Charybdis (Goniosoma) crucifera* Alcock, 1899: 51. *Charybdis cruciata* Rathbun, 1910: 362. *Charybdis (Charybdis) cruciata* Leene, 1938: 39C, p.24; *Charybdis (Charybdis) feriata* Sakai, 1976: 357.

Diagnostic features: Carapace brown in colour, characterized with a cross mark on the carapace. Anterolateral borders cut into six teeth and the first one exhibits a bifurcation and the rest acuminate. The last pair of legs paddle shaped and adapted for swimming.

Remarks: This species can be easily distinguished from all the other crabs by its distinctive cross mark on the carapace as well as the bifurcation on the first anterolateral teeth.

Habitat : Marine. Inhabits off shore waters.

Distribution: The lower reaches of backwaters, where salinity is the highest. Collected from Thoppumpady, Barmouth, Munambam and Azhikode

Charybdis (Charybdis) lucifera (Fabricius, 1798)

Frequent Synonyms: Portunus lucifer Fabricius, 1798: 364; Goniosoma luciferum de Man, 1888: 83 Henderson, 1893: 374; Charybdis (Goniosoma) quadrimaculata Alcock, 1899: 54; Charybdis lucifera Rathbun, 1910: 364, pl. 2, fig. 10. Charybdis (Goniosoma) lucifera Chopra & Das, 1937: 392, fig. 3, Chhapgar, 1957: 21, pl. A, fig. 5, pl. 6 d-g. Charybdis (Charybdis) lucifera Leene, 1938: 57, figs 23-25.

Materials Examined: Numerous specimens of both sexes (21 males and 14 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 53mm-76mm carapace width for males 42mm-79mm carapace width for females

Diagnostic features: Carapace yellowish brown with two large white spots on either branchial region. Right cheliped is markedly larger than the left cheliped. They can be well distinguished with the presence of a sharp median lobule on lower border of the orbit. The last pair of legs is paddle shaped, for swimming.

Remarks: This species can be easily distinguished from all the other crabs by its distinctive white spots on the carapace and the shape median lovule on the lower border of the orbit.

Habitat : Marine species. Inhabits off shore waters.

Distribution: Collected from Munambam and Azhikode

Charybdis (Goniohellenus) hoplites (Wood-Mason, 1877)

Materials Examined: Four specimens (3 males and 1 female) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 34mm-58mm carapace width for males

33mm carapace width for female

Frequent Synonyms: Charybdis (Goniohellenus) hoplites, Alcock, 1899: 66, Leene, 1938: 99; Charybdis hoplites, Rathbun, 1911: 207; Charybdis (Charybdis) hoplites, Chhapgar, 1957: 24

Diagnostic features: Carapace light brownish. Anterolateral teeth cut into 6 teeth of which, the last tooth forms a long *Portunus*-like spine, Posterior

border of the carapace forms an angular junction with the postero-lateral borders. This species can be easily distinguished from all the other crabs by its distinctive

Remarks: The species can be often mistaken as a *Portunus* species, due to its long anterolateral spine, which is a characteristic feature of Genus *Portunus*. However, it can be distinguished by observing the number of anterolateral teeth, which is six in this species (characteristic feature of Genus *Charybdis*), while Genus *Portunus* bears nine anterolateral teeth.

Habitat : Marine species. Inhabits off shore waters.

Distribution: Collected from Munambam

Thalamita crenata (Latreille, 1829)

Materials Examined: Eleven specimens (4 males and 7 females) collected from the high saline areas of the Cochin backwaters.

Measurements: 34mm-71mm carapace width for males

45mm-79 carapace width for female

Frequent synonyms: *Thalamita crenata* Milne Edwards H., 1834: 461, de Man, 1888: 79, Alcock, 1899: 73(key), Rathbun, 1910: 365, Rathbun, 1911: 207, Kemp, 1915: 249, Balss, 1922: 111. Chhapgar, 1957: 25, pl. 7 l-n. Sankarankutty, 1961: 102(list), 106; *Thalamita crenata* Sakai 1976: 369

Diagnostic features: Carapace brownish or greenish grey, with faint transverse ridges. front cut into six lobes, while the anterolateral margins cut into five equal teeth. The last pair of legs paddle shaped and adapted for swimming.

Remarks: On a preliminary view, *T. crenata*, resembles mud crabs belonging to Genus *Scylla* due to the similar colouration. However, it can be easily distinguished by the number of anterolateral spines, which is five in this species, while Genus *Scylla* possess nine anterolateral teeth.

Habitat: Estuarine. Occurs in mud flats, sandy beaches and mangroves

Distribution: Thevara, Thoppumpady, Barmouth, Munambam

Uca (Celuca) lactea annulipes (H.Milne Edwards, 1837)

Materials Examined: Eight specimens (All males) collected from the mangrove areas of the Cochin backwaters through out the year.

Measurements: 23mm-38mm carapace width for males

Frequent synonyms: *Gelasimus annulipes* de Man, 1888: 69, Kemp, 1915: 221, Chhapgar, 1957: 46, pl. B, fig. 7, pl. 13 j-o, Crane, 1975: 301; *Uca annulipes* Rathbun, 1910: 305, Balss, 1922, Panikkar & Aiyar, 1937: 295, 301, Sankarankutty, 1961: 113; *Uca (Celuca) lactea annulipes* Crane, 1975: 299, figs 18A-C, 19I-N, 20D-K, 24N-O, 27I-J, 29D, 32L-M, 37L, 46L, 54I-II, 69D, pls 39A-D, 45A, 47C, 50A, Sakai, 1976: 608.

Diagnostic features: Colour of the carapace black with white or yellow stripes and the chelipeds pinkish or white. The most striking feature of this species is that one of the chelae of the males has been modified and enlarged to form pincers and found to exhibit waving displays with the pincers. Eye stalks are elongated and prominently seen projecting diagonally.

Remarks: The species can be easily distinguished by its extremely large pincers and the long eyestalk. They are observed to prefer damp grounds

and hence found on the shores, muddy substratum of mangrove regions, near the water channels. These were the most difficult species to be captured while sampling.

Habitat : Damp ground, found on the shores and near the mangroves.

Distribution: Aroor, Valanthakad, Vypin, Vallarpadam, Kumbalanghi

Macrophthalmus depressus (Ruppell, 1830)

Materials Examined: Five specimens (All males) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 31mm-38mm carapace width for males

Frequent synonyms: *Macrophthalmus depressus* De man, 1888: 124, Henderson, 1893: 389, Alcock, 1900: 280, Kemp, 1919, Gravely, 1927: 150, Chhapgar, 1957: 52.

Diagnostic features: Carapace grayish, not smooth and surface studded with minute pearly granules. Eyestalks remarkably long, slender and reach the end of the orbital angles. Palm with inner surface smooth and hairy. Hairs present on the ambulatory legs.

Remarks: The species is characterized with large orbits, remarkably long eye stalks and the inner palm and limbs hairy. They are strict mud dwelling forms and the specimens collected have mud stained to it whenever collected.

Habitat : Mud dwelling forms and occurs in mangrove regions.

Distribution: Valanthakad, Kumabalanghi

Nanosesarma (Beanium) batavicum (Moreira, 1903)

Materials Examined: Numerous specimens of both sexes (39 males and 57 females) collected from the mangrove regions of the Cochin backwaters through out the year.

Measurements: 23mm-28mm carapace width for males

22mm-29mm carapace width for females

Frequent synonyms: *Nanosesarma (Beanium) batavicum*, Serene and Soh, 1970:394, Sethuramalingam and Ajmal Khan, 1991: 20.

Diagnostic features: Carapace mottled grey in colour and chelipeds cherry red. Front broad and deflexed. Upper surface of the palm of cheliped with two oblique pectinated crest. The distinguishing feature of the species is the presence of three acute spines on the posterodistal border of the fourth pereopod.

Remarks: The sesarmid crabs are very difficult to distinguish between. They exhibit similarity in colouration, habitat, behavioral pattern etc. They can be primarily distinguished into genus level by observing the denticulation on the fourth and fifth pereopod. Sesarmid crabs belonging to genus *Nanosearma* posess denticulations on the fourth pereopods. In the case of *N. batavicum*, the denticulation can be explained by the three acute spines on the posterolateral border of the fourth pereopds. Another unique feature of this species being the presence of two oblique pectinated crests on the upper surface of the palm. These are found to occur in the mangrove regions abundantly.

Habitat : Mangrove region

Distribution: Aroor, Valanthakad, Vypin, Vallarpadam, Kumbalanghi, Kandakkadavu

Nanosesarma (Beanium) andersoni (De Man, 1888)

Materials Examined: Numerous specimens of both sexes (19 males and 14 females) collected from the mangrove regions of the Cochin backwaters through out the year.

Measurements: 25mm-19mm carapace width for males

22mm-29mm carapace width for females

Frequent synonyms: *Sesarma andersoni* De Man, 1888: 657; *Nanosesarma (Beanium)andersonii*, Serene and Soh, 1970:394, Sethuramalingam and Ajmal Khan, 1991: 20.

Diagnostic features: Carapace dark brown with reddish chelipeds. Numerous striae are present on the upper surface of the palm of cheliped, one of which forms pectinated crest. They are distinguished by the presence of four strong spines on posterodistal border of merus of pereopod four.

Remarks: Species level differentiation is almost impossible on primary observation. Genus level identification involves the observation of denticulation on the fourth pereopod. Once the genus level identification is confirmed, *N. andersoni* can be distinguished by the four numbers of strong spines on the merus of fourth pereopod. This can be supplemented with presence of numerous striae ornamenting the upper surface of the palm of the cheliped, of which one o the striae if modified to form a pectinated crest. Like other sesarmids, *N. andersoni* inhabits the mangrove regions of the study area and has been recorded in abundance.

Habitat : Mangrove region

Distribution: Aroor, Valanthakad, Vypin, Vallarpadam, Kumbalanghi, Kandakkadavu

Neoepisesarma (Neoepisesarma) mederi (H.Milne Edwards, 1854)

Materials Examined: Numerous specimens of both sexes (13 males and 19 females) collected from the mangrove regions of the Cochin backwaters through out the year.

Measurements: 23mm-26mm carapace width for males

20mm-29mm carapace width for females

Frequent synonyms: *Sesarma mederi* Milne edwards, 1834:185; *Neoepisesarma (Neoepisesarma) mederi* Serene and Soh, 1970:405, Sethuramalingam and Ajmal Khan, 1991: 20.

Diagnostic features: Carapace black or dark brown and chelipeds red in colour. The surface of the carapace have tufts of hair on the anterior portion. The distinguishing feature of this species is that, above the transverse dactylar tubercles, a sulcus runs about one third of total length of the tubercles and the vertical granular crest of the inner palm is salient.

Remarks: The genus *Neoepisesarma* is characterized by the absence of denticulation on the merus of the fourth pereopod. In the case of *N. mederi*, preliminary identification is possible, unlike the other sesarmids. The species can be firstly identified by the tufts of hair on the frontal region of the carapace. The species is characterized a sulcus which runs above the dactylar tubercles. These crabs inhabit the mangrove mud flats are found in abundance throughout the year.

Habitat : Mangrove regions

Distribution: Aroor, Valanthakad, Vypin, Vallarpadam, Kumbalanghi, Kandakkadavu

Neoepisesarma (Selatium) brockii (De Haan, 1887)

Materials Examined: Numerous specimens of both sexes (34 males and 29 females) collected from all the 15 stations of the study area throughout the year .

Measurements: 33mm-42mm carapace width for males

31mm-39mm carapace width for females

Frequent synonyms: *Sesarma brockii* De Man 1888: 651; *Neoepisesarma (Selatium) brockii* Serene and Soh, 1970:405, Sethuramalingam and Ajmal Khan, 1991: 20.

Diagnostic features: Colour of the carapace dark brown and the chelipeds orange coloured with white tips. The pectinated crest on upper palm reach the margin and the dactylar tubercles are well separated from one another without any tranverse sulcus above.

Remarks: They can be identified primarily by its semi terrestrial mode of life. However, genus level identification can be made by the absence of denticulation on the merus of fourth pereopod. The species is characterized by well separated dactylar tubercles, with the transverse sulcus. They are found to inhabit the mangrove mud, fissures of rocks, stone embankments as well as backwater shores. It is observed that they are very vigilant and intelligent creatures and they are hard to pursue.

Habitat: Semi terrestrial form, found in mangrove mud, fissures of rocks, stone embankments, backwater shores etc.

Distribution: Throughout the study stations

Metapograpsus messor (Forskal, 1775)

Materials Examined: Numerous specimens of both sexes (12 males and 8 females) collected from the mangrove regions of the Cochin backwaters throughout the year.

Measurements: 33mm- 39mm carapace width for males

32mm- 37mm carapace width for females

Frequent synonyms: *Cancer messor* Forskål, 1775: 88; *Grapsus Savignyi* de Haan, 1833-1849 (1835): 59; *Grapsus messor* Milne Edwards H., 1834: 88; *Metopograpsus messor* Alcock, 1900: 397. Balss, 1922, Chhapgar, 1957: 54, pl. B, fig. 4, pl. 15 n-q, Sakai, 1976: 633, pl. 216, fig. 2.

Diagnostic features: Carapace bottle green in colour and the chelipeds appears light violet. The legs are striped with alternate dark and light bands. The post frontal region appears to have transverse markings. The propodus of the chelae inflated.

Remarks: There exists two species belonging to genus *Metapograpsus*, in the study area. *Metaposgrapsus messor* and *M. maculatus* have a common colouration and similar habitat preference. However, *M. messor* is characterized with pale violet coloured chelipeds which is stout. The post frontal region of the carapace bears some transverse markings too. Another important character which can be considered for the distinction between both the species is the shape of the male abdomen and first male pleopods.

Habitat : Mud dweller and inhabits the mangroves, subtidal region.

Distribution: Aroor, Valanthakad, Vypin, Vallarpadam, Kumbalanghi, Kandakkadavu

Metapograpsus maculatus (H.Milne Edwards, 1873)

Materials Examined: 2 specimens (male) collected from the mangrove regions of the Cochin backwaters during the post monsoon season.

Measurements: 33mm-36mm carapace width for males

Frequent synonyms: Metopograpsus latifrons Rathbun, 1910: 308;
Metopograpsus maculatus, de Man, 1888: 145, pl. 10, figs 1-3, Alcock, 1900:
398, Pillai, 1951: 35, Chhapgar, 1957: 55, pl. 15 r-u

Diagnostic features: Carapace black in colour and chelipeds bright violet. Carapace is comparatively transversely elongated, on the anterior region alone. The post-frontal region is devoid of any transverse markings or ridges. The chelipeds are slender and not inflated. The striking feature of this species is that the last segment of male abdomen is trilobed.

Remarks: *M. maculatus* can be distinguished from *M. messor* by its slender non-inflated chelae, which is bright violet in colour. The carapace is smooth and no transverse markings is seen on the post-frontal region. The important character which can be considered for the distinction between both the species is the shape of the male abdomen and first male pleopods

Habitat : mangroves and the intertidal region

Distribution: Vypin, Vallarpadam

Varuna litterata (Fabricius, 1798)

Materials Examined: Numerous specimens of both sexes (110males and 97 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 19mm-42mm carapace width for males

21mm-39mm carapace width for females

Frequent synonyms: *Cancer litteratus* Fabricius, 1798: 342, *Varuna litterata* De Man, 1888: 698(list), Alcock, 1900: 401, Chhapgar, 1957: 56, pl. B, fig. 3, pl. 15 v, Sakai, 1976: 644, pl. 220, fig. 3.

Diagnostic characters: Carapace light brown to brownish grey, with a smooth surface and well-defined postero-lateral facets. Anterolateral borders cut into three broad, but less sharp teeth. Frontal region broad and without any spines. Dactylus, propodus and carpus of legs are laterally flattened like paddles and are fringed with long, densely packed hairs.

Remarks: *V. litterata* is characterized by well defined posterlateral facets on the carapace and the laterally flattened pereopods, which is fringed with dense hairs. The present study can be counted as the first report on the existence of this species in the study area, since no previous works records its presence in the study area before. These are estuarine crabs, however prefer areas with close proximity to the sea having good tidal influx.

Habitat : Occur in the mangrove regions and intertidal areas

Distribution: Vypin, Kumbalanghi, Kandakkadavu and Munambam

Chapter 6

Heteropanope indica (Stimpson, 1858)

Material Examined: Single specimens (Male) collected from the mangrove region of the Cochin backwaters during premonsoon period.

Measurements: 18mmcarapace width

Frequent synonyms: *Heteropanope indica* de Man, 1888: 53, pl. 3, figs 1-2, Alcock, 1898: 208. *Pilumnopeus indica* Sakai, 1976: 500, pl. 178, fig. 2. *Benthopanope indica* Davie, 1982: 144(list).

Diagnostic features: Carapace transversely oval and glabrous. Anterolateral borders is cut into four teeth. The dorsal surface of the carapace and the carpus and the propodus of the chelipeds are found to be studded with pearly granules. Chelipeds are extremely unequal.

Remarks: The species can be readily distinguished by the unequal chelipeds and the pearly granules ornamenting the carapace and the chelipeds.

Habitat : mangrove regions

Distribution: Vallarpadam

Xenophthalmus pinnotheroides (White, 1846)

Material Examined: Eighteen specimens (16 Males and 2 females) collected from the mangrove region as well as the muddy sediments of the Cochin backwaters during post monsoon and pre monsoon period.

Measurements: 19mm-25mm carapace width for males

13mm-21mm carapace width for females

Frequent synonyms: *Xenophthalmus pinnotheroides* Rathbun, 1910: 338, fig. 22a-e, Sankarankutty, 1966: 347, 350, Sakai, 1976: 591, pl. 203, fig. 4. *Xenophthalmus pinnoteroides* Alcock, 1900: 332.

Diagnostic features: Carapace dirty white in colour and subtrapezoid in shape, Frontal region is narrow and strongly deflexed. Orbits situated longitudinally and parallel to each other. Propodus of the first walking leg as long as broad. Carpus and propodus of second leg possess dense pubescence. The third and the last legs longer and more slender covered with short hairs.

Remarks: The species can well distinguished by its white colour and the third and fifth pair of percopods which is longer than the other legs. The most striking character of this species is longitudinal orbits, which is parallel to each other.

Habitat : Mesohaline regions of the backwaters where the substratum is muddy in nature.

Distribution: Aroor, Thevara, Thoppumpady, Marine Science Jetty, Kandakkadavu, Kumbalanghi, Vypin

Doclea gracilipes (Stimpson, 1857)

Materials Examined: Four specimens (All males) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 29mm-55mm carapace width for males

Frequent synonyms: *Doclea sp.* De man, 1888:13; *Doclea gracilipes* Alcock, 1895: 229, Laurie, 1906: 381, Chopra, 1935: 470, Shen, 1940:80, Chhapgar, 1957:13.

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Diagnostic features: Colour of the carapace and the segments of the legs covered with hair, is greenish brown, while the segments devoid of hair is pink in colour. Carapace discoid with a series of blunt spine like structures arranged longitudinally along the middle line. The body and limbs are covered with thick hairs and the chelipeds are shorter than the other legs. The second pair of legs is the longest and is about three to four times the length of the carapace.

Remarks: The species can be readily distinguished by the serious of blunt spines arranged longitudinally along the middle line of the carapace. The frontal region has been extended forward to form a rostrum. Another striking feature of this crab, chelipeds are the shortest among the legs and the longest is the second pair of pereopods. The crab is covered with dense hairs.

Habitat : Marine species actually occurring in 30 to 50 meters depth in muddy or sandy substratum.

Distribution: Munambam, Azhikode

Ebalia malefactrix (Kemp, 1915)

Frequent synonyms: Philyra malefactrix, Kemp 1915.

Materials Examined: Six specimens (4 males and 2 females) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 2.1mm-2.3mm carapace width for males

1.3mm-2.1mm carapace width for females

Diagnostic features: Carapace is subcircular in shape, with a separate facet on the side wall of the carapace at the hepatic region. Eyes and orbits are too small. Chelipeds are stouter and longer than the legs. **Remarks**: These are benthic crabs dwelling the muddy sediments of the backwaters. They can be distinguished from the other two benthic species by the presence of three lobes on the frontal region. The chelipeds are stout and longer, however not as prominent as the other two benthic species.

Habitat : The muddy sediments of the backwaters.

Distribution: Aroor, Thevara, Thoppumpady, Marine Science Jetty, Kandakkadavu, Kumbalanghi, Vypin, Barmouth.

Halicarcinus messor (Stimpson, 1858)

Materials Examined: Four specimens (All males) collected from the high saline areas of the Cochin backwaters during summer months.

Measurements: 2mm-2.2mm carapace width for males

Frequent synonyms: *Cancer planatus*, Fabricius, 1775; *Hymenicus*, Dana, 1851: 253

Diagnostic features: These are small sized benthic crabs. Carapace suboval with lateral walls characterized with two teeth, one at the anterolateral angle and the second at the posterolateral angle. The front region is extended to form a rostrum. The rostrum is trilobed, the lateral lobes are small and acute and the medium one is long and spatuliform, broadly rounded apically. The median lobe is broadest at halfway along its length and are devoid any setae. Chelipeds are heavy, slightly longer than the legs. Palm of the chelipeds markedly swollen.

Remarks: It is quite a tedious task to distinguish between the two hymenosomatid crabs of the study area. The most important character of the hymenosomatid crabs from the study area is that the frontal region has been

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extended to form a trilobed rostrum. *H. messor* is characterized with two lateral spines, one on the anterolateral region and the other at the posterolateral angle. Chelipeds are long and prominent with an inflated propodus.

Habitat : The muddy sediments of the backwaters.

Distribution: Aroor, Thevara, Thoppumpady, Marine Science Jetty, Kandakkadavu, Kumbalanghi, Vypin, Barmouth.

Elamenopsis alcocki (Kemp, 1917)

Frequent synonyms: *Elamenopsis lineatus*, Milne Edwards, 1834: 324-5, pls.18, fig.4; *Rynchoplax alcocki*, Kemp, 1917:250; *Neorhynchoplax alcocki*, Sakai, 1976.

Diagnostic features: Carapace pyriform and the frontal region extended to form a rostrum. Excluding the rostrum, the carapace width and carapace length are of the same proportion .The anterolateral border cut into a single tooth. Chelipeds are heavy and the fingers of the chelae with a wide gape at the base. The ambulatory legs are long and slender.

Remarks: *E. alcocki* can be distinguished by a single tooth on the anterolateral border of the carapace. The chelipeds are long and prominent as in the case of H messor, however, the propodus is not inflated, but characterized by a wide gape between the fingers.

Habitat : The muddy sediments of the backwaters.

Distribution: Aroor, Thevara, Thoppumpady, Marine Science Jetty, Kandakkadavu, Kumbalanghi, Vypin, Barmouth.



6.4. Discussion

Studies related to taxonomy and systematics of Indian Brachyura are limited, though several studies are being carried out in other areas like their fishery, biology and culture techniques. Discontinuous distribution, allometric growth pattern which results in dynamic morphology, polymorphism and sexual dimorphism make classification of brachyuran crabs highly confusing. This is supplemented by the adaptive features of brachyuran crabs to varying environmental conditions and the resulting individual variability or clinal variability with the species and populations (Jayabaskaran *et al.*, 1999). However, proper taxonomic studies hold much significance in both economical and ecological point of view.

In the case of commercially important species like mud crabs belonging to genus *Scylla, Portunus pelagicus, P. sanguinolentus* and other Portunids, proper taxonomical identification is essential in order to develop successful culture techniques, stock assessment and larval recruitment. In ecologically important crab species like the Sesarmids, Grapsids and Ocypodids proper taxonomic study is essential for understanding the evolutionary patterns and conservation of species, especially which exhibits a declining population.

Earliest classic works on the taxonomy of Indian brachyura include works of Linnaeus (1758), Fabricius (1798), De Haan (1833), Dana (1852a, b,c), Wood Mason (1871), Boas (1880), Miers (1886) Ortmann (1892), Henderson (1893) and Alcock (1895, 1896, 1898, 1899, 1900, 1901), Bouvier (1896), Borradile (1907), Balss (1957) and Glaessners (1969), who conducted an extensive study on the crabs world wide including the Indian sub continent. Meanwhile some Indian workers also conducted studies related to crab taxonomy viz., Pillai (1951) and Chhapgar (1957). However, crab taxonomy remained a relatively unexciting topic for many in the field of study.

Taxonomy of the commercially important mud crab species have been dealt with by many workers (Kathirvel, 1981; Radhakrishnan and Samuel, 1983; Joel and Sanjeevaraj, 1983; Kathirvel and Srinivasagam, 1992; Kathirvel *et al.*, 2004; Devi and Joseph, 2013a; Trivedi and Vachhrajani, 2013; Padate *et al.*, 2013; Balasubramanian *et al.*, 2014; Mandal *et al.*, 2014 a, b; Devi and Joseph, 2015). However, taxonomy of crabs belonging to other families has not been attempted much in the Indian scenario. The available taxonomic works on brachyuran crabs of Indian waters with a valid identification key include the works of Chhapgar (1957), Sethuramalingam and Ajmal Khan (1991) and Jayabaskaran *et al.* (1999), which are considered as standard references in the present work.

Though crabs exhibit conspicuous morphological variations, right from family to species level, these variations cannot be fully relied up on for their classification, as the morphological characters are influenced by age and growth. To be precise, morphological features are subjected to growth variations (Jayabaskaran et al., 1999). Hence, some other unique structures have to be considered along with the morphological characters for the identification proper of crabs. According Chhapgar (1957),to Radhakrishnan and Samuel (1982), Joel Sanjeevaj and (1983),Sethuramalingam and Ajmal Khan (1991), Keenan et al. (1998) first male pleopods or the third maxillipeds can be used as a powerful tool in crab
taxonomy. Aside from these two structures, the shape of the male abdomen is also observed to vary considerably among the brachyuran crab species. Therefore, it can be also considered for taxonomic studies, along with the third maxillipeds and the first male pleopods.

As suggested by other workers (Chhapgar, 1957; Sakai, 1976; Lucas, 1980; Sethuramalingam and Ajmal Khan, 1991, Jayabaskaran et al., 1999; Ajmal Khan and Ravichandran, 2009) brachyuran crabs exhibit remarkable variations in the morphological features like colour, carapace shape, markings on the carapace, frontal region, spination on the lateral margins, size, shape and spination on the cheliped and other walking legs. In the present study, crabs were primarily classified on the basis of carapace shape. Sethuramalingam and Ajmal Khan (1991) and Jayabaskaran et al. (1999) have employed carapace shape as an important feature to classify the brachyuran crabs into different families. They have also taken into account the characteristics on the chelae and pereopods for the classification. In the present study, the brachyuran crabs enlisted from the Cochin backwaters have been placed into different families after considering the morphological characters including carapace shape, frontal spines, features on the chelipeds and the percopods coupled with the third maxillipeds. Ajmal Khan and Ravichandran (2009) suggested that third maxillipeds can be used for the identification of brachyuran crabs.

When the third maxillipeds were studied in detail, it was found that these structures do not show marked variation in species level, but it can be considered for family or sub family level identification. For instance, the third maxilliped of Portunid crabs is characterized by broad, quadrate and flattened merus and ischium, lined with thick hairs on the inner margin. The third maxillipeds of Genus *Scylla, Portunus* and *Charybdis* were found to follow the same structural pattern (Fig 6.100 a, 6.100b, 6.100c), from which it is clear that no genus level variation do exist for third maxillipeds in Portunid crabs. Devi and Joseph (2013a; 2015) as a part of their attempt to revalidate the *Scylla* species occurring in the Cochin backwaters, made an extensive study on the third maxillipeds of *Scylla* species. They failed to find any considerable variation in the third maxillipeds of two *Scylla* species viz. *S. serrata* and *S. olivacea*. Their findings are in accordance with the present work and inferences as per their studies. Hence it can be concluded that in Portunid crabs, third maxillipeds do not show genus level as well as species level variation. A similar trend has been observed among the members of other crab families too.

In the case of Grapsid crabs, third maxillipeds are characterized with a subcircular and narrow merus and ischium, and they were found to leave a prominent rhomboid gap in between, through which the buccal cavern is visible (Fig 6. 58). The grapsid crabs belonging to sub family Grapsinae, which includes genus *Metapograpsus* and sub family Sesarminae, comprising genus *Nanosesarma* and genus *Neoepisesarma* tend to follow the same structural pattern for the third maxillipeds. It is also found that, no conspicuous variations do exist in species level for the grapsid crabs. All the six crabs, *Metapograpsus messor*, *M. maculatus*, *Nanosesarma* (*Beanium*) *batavicum*, *N. (Beanium) andersoni*, *Neoepisesarma* (*Neoepisesarma*) *andersoni*) and *N. (Seletium) brockii* exhibited same structural pattern for the third maxillipeds. However, the third sub family of family Grapsidae, sub family Varuninae showed deviation from this structural pattern. In Varunid crab,

the rhomboidal gap was not prominent and the third maxillipeds were found to close the buccal cavern completely (Fig 6.82). Hence, it can be concluded that third maxillipeds vary at the sub family level also. The findings are totally in agreement with the findings of Sethuramalingam and Ajmal Khan (1991).

In the case of Family Hymenosomatidae, the third maxillipeds is characterized with a subovate merus and a quadrate ischium, where the merus is larger than and both the structures are covered with hairs. The two members of the Family Hymenosomatidae, *Elamenopsis alcocki* and *Halicarcinus messor*, followed this same structural pattern (Fig 6.101 a, 6.101b) for the third maxillipeds irrespective of the genera they belong to.

From the present study, it is observed that the structural pattern of third maxillipeds is unique for different crab families. They exhibit a peculiar pattern, which varies considerably with different families. For instance, in the case of family Majidae, third maxillipeds is narrow and slender, with a quadrate merus and ischium (Fig 6.13), but in the case of Family Leucosiidae, it is characterised with a sub ovate merus and quadrate ischium fused together to form a single structure (Fig 6.15). In Family Xanthidae and Family Ocypodidae, merus is found to be smaller than the ischium (Fig 6.6, Fig 6.8), while in the case of Family Hymenosomatidae (6.14), the third maxillipeds is characterised with larger merus and smaller ischium.

All these observations claims that the third maxillipeds can be studied for the identification of crabs up to family or sub family level, but it cannot be considered as reliable tool for genus level or species level identification.

Another important taxonomic tool suggested by earlier workers for the identification of crabs is the first pair of pleopods in males (Chhapgar, 1957; Stephenson and Campbell, 1960; Joel and Sanjeevaraj, 1983; Sethuramalingam and Ajmal Khan, 1991; Fuseya, 1998; Keenan et al., 1998; Jayabaskaran et al., 1999; Ajmal Khan and Ravichandran, 2009; Devi and Joseph, 2013a, 2015). In the accordance to their findings, the shape of the first male pleopods and the male abdomen were found to exhibit considerable variation in size, shape, structure, setation etc, even among the closely related species. In the case of genus Scylla, it is observed that the first male pleopods of S. serrata and S. olivacea were similar in shape. However, they showed variation in the pattern of setation. The first male pleopod of the first S. serrata exhibited two patches of thin and inconspicuous setae on the inner margin on its posterior end (Fig 6.22), while that of S.olivacea exhibited a single patch of setae, which is thick and prominent (Fig 6.26). The findings are comparable with the illustrations provided by Keenan et al. (1999). Radhakrishnan and Samuel (1982) and Joel and Raj (1983) noticed some minor variations in the apex region of male pleopods in the two species of Scylla, but in the present study, no such remarkable variations have been noticed.

It has been noticed that the first male pleopods exhibited wide degree of variation in species level, in some cases. For instance, the first male pleopods of the two species belonging to genus *Metapograpsus* is markedly very different. *Metapograpsus messor* is characterised with tubular male pleopods with a bilobed tip and the distal end of the pleopod is fringed with hair on both the margins (Fig 6.61), while the male pleopods of *M. maculatus* is coarse and thick and the tip of the pleopod form a separate hammer head shaped lobe with serrated margins (Fig 6.64). The observations are in confirmation with the observations of Chhapgar (1957).

Similar observations were made in the case of Genus *Charybdis* also. The first male pleopods of the species belonging to genus *Charybdis* differ markedly. In *Charybdis (Charybdis) feriata*, the first male pleopod is straight and little stout, with a blunt and notched tip, fringed with thick hairs on the extreme tip, while that of *Charybdis (Charybdis) lucifera* is narrow and the tip fringed with thick hairs. The male pleopod of *Charbdis (Goniohellenus) hoplites* is markedly different from the other two *Charybdis species*. The first male pleopod is broad at the base, having elegant curves to the middle portion with blunt tip ornamented with hairs. The observations made in the present study are totally in agreement with the findings of Chapgar (1957).

Sesarmids crabs were also found to follow the trend of species level variations in the shape of first male pleopods. The two species belonging to genus *Nanosesarma* showed variations in the shape of male pleopods. In *Nanosesarma (Beanium) batavicum*, the first male pleopod is slender, getting broader at the middle portion and tapering at the tip (Fig 6.70), while that of *Nanosesarma (Beanium) andersoni* is stouter with broad and rounded tip. The male pleopods is stouter with round blunted tip and fringed with hairs in *Neoepisesarma (Neoepisesarma) mederi*(Fig 6.78), while it is relatively narrow with bent and pectinated tip in the case of *Neoepisesarma (Seletium) brockii* (Fig 6.81).

Elamenopsis alcocki and *Halicarcinus messor*, belonging to Family Hymenosomatidae are two benthic crab species found to occur in the sediments of Cochin backwaters. Lucas (1980) opined that the hymenosomatid crabs were closely related to each other though they belong to different genera. They are found to be morphologically very similar to be distinguished between. However, it is noticed that their male pleopods show greater degree of variation. The first male pleopod of *Halicarcinus messor* is long, slender and curved with a bifurcating tip (Fig 6.97), while that of *Elamenopsis alcocki* is characterised with a broad base and appears twisted at the middle portion, with a blunt tip (Fig 6.99).

The first male pleopod *Varuna litterata* is stout with a uniform width throughout the length and the tip of the pleopods are found to be rounded and are fringed with hairs (Fig 6.87). *Varuna yui* is another Varunid crab belonging to genus *Varuna*, which is endemic to the continental waters of South East Asia. According to Tan and Ng (1994), both *V. litterata* and *V. yui* are morphologically very similar and can be distinguished only by means of their male pleopods.

In certain cases, it was observed that the male pleopods show variations, however not conspicuous. For instance, the crabs belonging to genus *Portunus, Portunus pelagicus* and *P. sanguinolentus* is characterised with pleopods without conspicuous variation. The first male pleopod of both the species is slender and straight without any bend near the tip. The pleopods of *P. pelagicus* bears stout spinules at the margins of the tip (Fig 6.28), while that of *P. sanguinolentus* bears fine hairs at the tip (Fig 6.31).

The male pleopod is characterised with a broad base, which become slender towards the length of the pleopod and a notched tip, fringed with hairs in the Majid crab *Doclea gracilipes*. (Fig 6.93) The first male pleopod of the porcelain fiddler crab *Uca annulipes* is slender with a bilobed tip (Fig 6.54 a, b). Both these observations are in conformity with the illustrations provided by Chhapgar (1957).

From the present study, it can be concluded that crabs tend to exhibit uniqueness in the shape of first male pleopods, which varies with species to species. Hence, the first male pleopods can be considered as a powerful taxonomic tool in the identification of crabs, up to species level.

Aside from the first male pleopods, it was noticed that the shape of male abdomen too showed marked difference with species. The variations in the shape of male abdomen can be considered along with the variations of male pleopods to confirm the identification.

For instance, *Metapograpsus messor* and *M. maculatus* are two mangrove species, characterised by violet chelipeds. This similarity can lead to confusion in distinguishing between these two species. It is observed that the shape of the male abdomen show conspicuous variation among these two species, which can be considered for their distinction in the preliminary examination itself. In *M. messor* and *M. maculatus*, segments of the male abdomen from the first to penultimate are of same width, sides being parallel, but they can be distinguished by noticing the last segment of the pleopod. The last segment is triangular, making the male abdomen tapering at its end in *M. messor* (Fig 6.60), while in *M. maculatus*, last segment suddenly narrow to a point (Fig 6.63). This significant variation has been pointed out by Chhapgar (1957) also. During the course of study, it was noticed that, it is difficult to identify the juvenile crabs by noticing the common morphological features, since most of these features are highly influenced by the growth stages. The Portunid crabs, *C. feriata* and *C. lucifera* are primarily identified by the conspicuous markings on their carapace. *C. feriata* possess a cross mark on its carapace, while *C. lucifera* can be identified by noticing the four white spots on its carapace. It is observed that, the markings on the carapace are not conspicuous in the juvenile crabs and many a times, it has lead to much confusion in identification. However, variation in the shape of the male abdomen was clearly visible even among the juvenile crabs (Fig 6. 37, 6.40). In that case, shape of the male abdomen can be studied to distinguish between the two juvenile crabs.

The first male pleopod is undoubtedly a powerful tool identification of crabs at species level however; shape of the male pleopod is preferable over the former as it helps in the identification of crabs in the preliminary examination itself. Crabs can be identified readily at the field itself by observing the shape of the male abdomen. The male pleopods can be studied only after taking the specimens to the lab and dissecting out the structure to be examined under the microscope to note the minor characters. The shape of the male abdomen can be studied without making any harm to the crab. This is very much applicable in the case of mangrove species, as it is a tedious task to identify the mangrove crabs, especially the Sesarmids, since they are morphologically very similar. Moreover, the Sesarmid crabs are considered to be a declining population, and sacrificing the crabs of studies can be avoided to some extent.

Chapter **7 ON THE SYSTEMATICS OF GENUS SCYLLA DE HAAN, 1833 OF COCHIN BACKWATERS**



Introduction 7.2 Materials and Methods Results Discussion

Introduction 7.1

Mud crabs belonging to the genus Scylla inhabit brackish waters, such as estuaries and mangrove areas. They are widely distributed throughout the Pacific and Indian Ocean, from Tahiti, Australia and Japan to Southern Africa (Chhapgar, 1957; Hill, 1975; Sakai, 1976). In India, mud crabs are reported from the estuaries of the rivers of Ganga, Mahanadi, Krishna and Cauvery and the brackish water lakes viz., Chilka and Pulicut on the east coast, the estuaries of Narmada and Tapti and the brackish waters of Kerala on west coast. They are also found to inhabit the mangrove regions of Andaman and Nicobar Islands, Andhra Pradesh, Tamil Nadu and Kerala (Anil, 1997).

Mud crabs are commercially important due to their large size and good taste. Hence, they are of great demand in the domestic as well as

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foreign market. India earns about US\$ 18 million as foreign exchange from the export of live mud crabs (www.mpeda.com/mudcrabreports.pdf). The export of live mud crabs from India to countries like Singapore, Malaysia and Hong Kong stimulated increased exploitation of mud crabs from their natural habitats during the past few years. The attractive prices offered for live crabs in the export market encouraged farmers to culture mud crabs in some parts of India as practiced in a more organized manner in South East Asian countries (Suseelan *et al.*, 1996).

Prior to the classification of Keenan et al. (1998), there was considerable confusion regarding the taxonomy of mud crabs, belonging to genus Scylla. Estampador (1949) recognized three species of Scylla namely, S. serrata, S. oceanica, S. tranquebarica and one variety, and S. serrata var. paramamosain, based on the external morphology. These four morphotypes of Scylla were further classified into two groups: 'Banhawin' and 'Mamosain'. Banhawin crabs are free swimming forms characterized with green colour and polygonal markings on all legs and chelipeds, while the Mamosain crabs inhabited holes and possessed dark brown colour, without any markings on legs and chelipeds. In accordance with the findings of Estampador (1949), Serene (1952) reported the occurrence of four forms in Vietnam. These works were subsequently reviewed by Stephenson and Campbell (1960) and Holthius (1978) and they pointed out the inconsistent and variable nature of the distinguishing characters. Fuseya and Watanabe (1996) proposed three species for genus Scylla, viz., S. serrata, S. tranquebarica and S. oceanica on the basis of variations noticed through electrophoretic analysis. Keenan et al. (1998) collected mud crab specimens from the Red Sea and throughout the Indo-Pacific

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regions and classified them into four species, *Scylla serrata*, *Scylla tranquebarica*, *Scylla olivacea* and *Scylla paramamosain*, on the basis of morphological, morphometric and molecular analysis.

Despite of the importance of mud crabs in both coastal aquaculture and artisanal fisheries, studies conducted on the Indian mud crab population is very scanty. In India, studies on mud crabs have been dealt with by Kathirvel (1981), Radhakrishnan and Samuel (1982), Joel and Sanjeevaraj (1983) and Kathirvel and Srinivasagam (1992). Kathirvel (1981) recorded two species of Scylla from Cochin backwaters, larger species being Scylla oceanica and smaller one being Scylla serrata based on the morphology. Later in 1982, Radhakrishnan and Samuel reported the occurrence of a subspecies Scylla serrata serrata from Cochin backwaters on the basis of morphological characters. Joel and Sanjeevaraj (1983) studied the taxonomy of Scylla from Pulicut lake and reported the occurrence of two species, namely S. tranquebarica and S. serrata. Taxonomy of mud crabs from India were then critically analysed by Kathirvel and Srinivasagam (1992) and stated that two species are found to occur in Indian waters, S. serrata and S. tranquebarica, which are characterized by the differences in size, spines on the outer border of the carpus of the cheliped and habitat preferences. In contrast to their findings, Shaji et al. (2006) stated that S. serrata and S. olivacea are the most common species occurring in India, based on the taxonomic identification of Keenan et al. (1998).

Knowledge on taxonomy is important for the development of a more successful aquaculture industry and management of wild stock. There is a clear need to identify the mud crab species in India using molecular tools, as morphological diagnostic characteristics of mud crabs are rather weak or specific to life stages or sex. It is crucial to investigate which species is dominant in coastal aquaculture in India, as an incorrect name application can affect the success of aquaculture industry, and it can lead to the farming of a wrong species (Wowor and Ng, 2007), or utilizing the fund to develop a technology for the aquaculture of an economically or biologically unsuitable species (Balasubramanian *et al.*, 2014)

Quite recently, Mandal *et al.* (2014 a) dealt with the taxonomic uncertainty of mud crabs commonly available in the Indian coastal waters using molecular genetic markers, ITS-1 and sequencing of COI gene combined with traditional morphometry. A similar study was conducted by Balasubramanian *et al.* (2014) on the basis of two mitochondrial genes, 16S rRNA and COI of *Scylla* populations collected from different locations along the Indian coast. Mandal *et al.* (2014 b) identified the *Scylla* species from the Indian waters using RAPD and PCR-RFLP markers.

The present chapter is an attempt to describe the *Scylla* spp from Cochin backwaters on the basis of morphological characters including the description of the male gonopods and the third maxillipeds, morphometry as well as molecular tools.

7.2 Materials and Methods

7.2.1 Morphological study

The protocol followed has been mentioned in chapter 3 (section 3.4.4.1).

7.2.2 Multivaraite Analysis of Morphometric characters

24 morphometric characters (Fig. 7.1) were recorded for 365 mud crabs in total (211 males and 154 females) using vernier calipers to the nearest 0.1mm and the characters were then size standardized through the creation of 27 ratios (Table.7.1) (Keenan *et al.*, 1998).

Step wise Discriminant function analysis using SPSS ver. 20.0 was conducted using the 27 ratios to determine the characters that best discriminate the morphologically recognized species (Keenan *et al.*, 1998).

For Discriminant Function Analysis, only adult crabs with an internal carapace width 95mm were used in order to avoid the juvenile ontogenic changes. Crabs with broken or missing appendages and spines were avoided to obtain a reliable data.







Fig. 7.1: Details of the morphological characters considered for morphometric analysis; (A) Carapace, (B) Frontal Lobe, (C) Abdomen (D) Periopods and (E) Chelipeds

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Table 7.1: Size standardized mud crab morphometric data used for discriminant function analysis

A.	Ca	arapace Data
	1.	9^{th} Lateral spine height (LSH)/ Internal Carapace width (ICW), where LSH = (CW/ICW)/2
	2.	Carapace width (CW)/ Carapace width at 8 th spine (8CW)
	3.	Carapace length (CL)/ Internal Carapace width (ICW)
	4.	Posterior carapace width (PCW)/ Internal Carapace width (ICW)
	5.	Carapace Frontal Width (FW)/ Internal Carapace width (ICW)
	6.	Posterior carapace width (PCW)/ Carapace Frontal Width (FW)
	7.	Frontal median spine height (FMSH)/ Carapace Frontal Width (FW)
	8.	Frontal median spine height (FMSH)/ Distance between frontal median spines (DFMS)
	9.	Distance between frontal median spines (DFMS) / Carapace Frontal Width (FW
	10.	Distance between frontal lateral spines (DFLS) / Carapace Frontal Width (FW)
	11.	Distance between frontal median spines (DFMS) / Distance between frontal lateral spines (DFLS)
	12.	Sternam width (SW)/ Internal Carapace width (ICW)
	13.	Abdomen width/ Sternam width
B.	Cl	heliped Data
	14.	Propodus Length (PL)/ Internal Carapace width (ICW)
	15.	Dactyl Length (DL)/ Propodus Length (PL)
	16.	Propodus Width (PW)/ Propodus Length (PL)
	17.	Propodus Depth (PD)/ Propodus Length (PL)
	18.	(Propodus Width (PW)* Propodus Depth (PD) * 0.7854) / Propodus Length (PL)
	19.	Inner propodus spine (IPS)/ Propodus Length (PL)
	20.	Outer propodus spine (OPS)/ Propodus Length (PL)
	21.	Inner propodus spine (IPS)/ Outer propodus spine (OPS)
	22.	Inner carpus spine (ICS)/ Propodus Length (PL)
	23.	Outer carpus spine (OCS)/ Propodus Length (PL)
	24.	Inner carpus spine (ICS)/ Outer carpus spine (OCS)
	25.	Merus Length (ML)/ Propodus Length (PL)
B.	Pe	eriopod Data
	26	5^{th} maximum of Destrul width $(5\text{DW})/5^{\text{th}}$ maximum of Destrul low oth (5DL)

- 26. 5th periopod Dactyl width (5PW)/ 5th periopod Dactyl length (5PL)
- 27. 3rd periopod merus width (3PML)/ Internal carapace width (ICW)

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7.2.3 Molecular study

7.2.3.1 DNA isolation

Muscle tissue from the third thoracic appendage of live mud crabs was dissected and DNA isolation was done immediately after the dissection. Total genomic DNA was isolated from 10 mg of muscle tissue. Tissue was digested by incubating with proteinase K/SDS solution at 37°C for two hours. DNA isolation was carried out following standard phenol: chloroform extraction and ethanol precipitation technique (Sambrook *et al.*, 1989). Purity and quality of DNA was checked on 0.7% agarose gel. The concentration of dissolved DNA was estimated using UV spectrophotometer at 260 nm. DNA was diluted so as to obtain a final concentration of 75 ng/µl (Mandal *et al.*, 2014a)

7.2.3.2 Amplification of First Internal Transcriber Spacer (ITS-1) region

The ITS-1 region was amplified using the primer sets designed by Central Genetics Lab, Rajiv Gandhi Centre for Aquaculture (Mandal *et al.*, 2014a) and following the protocol described by Imai *et al.* (2004).

7.2.3.3 Amplification and sequencing of cytochrome oxidase c- oxidase sub unit I (COI) gene

The protocol followed has been described in Chapter3.

The homologue searching of the nucleotide sequence (using blast suite) was performed with the Basic Local Alignment Search Tool (BLAST) through National Centre for Biotechnology Information (NCBI) server (http://www.ncbi.nlm.nih.gov/blast).

7.3 Results7.3.1 Morphological Study

The initial examination of the morphological characters, viz., shape of the frontal lobes, spination of the carpus of the chelae and the pattern of hexagonal markings indicates the presence of three morphotypes of *Scylla* in the Cochin Backwaters, viz., *Scylla serrata*, *Scylla tranquebarica* and *Scylla olivacea* based on the identification key provided by Keenan *et al.* (1998).

The first morphotype, which is identified to be *S. serrata* exhibits pale green to olive green colouration and the dactylus of the chelae pale orange. H-shaped gastric groove is found to be deep. The frontal margin has bluntly pointed spines, with rounded interspaces. Anterolateral margin of the carapace has 9 narrow teeth. Chelipeds are large and sturdy with its propodus inflated. The distinguishing character of the species is the presence of two prominent sharp spines on the distal half of outer margin of the carpus of the cheliped (Plate 7.1).



Plate 7.1: Morphology of first morphotype (assigned to be *S. serrata*) male, 117.23±0.56 mm- A, closer view of the pointed frontal lobes and the narrow anterolateral spines- B, two obvious carpal spines-C.

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The limbs of the crabs exhibited conspicuous hexagonal patterning in both male and female. This is found to be the largest among the three *Scylla* species.

The second morphotype, which is assigned to be *S. tranquebarica* is grayish green in colour and the limbs have a pale orange tinge. The propodus of the chelae and the last segment of the paddle legs exhibited orange colouration. The frontal margin of the carapace has blunt spines, with round interspaces. Anterolateral margin of the carapace has 9 broad teeth. Chelipeds are large with propodus inflated. The carpus of the cheliped possesses two spines on the distal half of outer margin (Plate 7.2.). Hexagonal markings are weak and inconspicuous on the chelae and paddle legs.



Plate 7.2: Morphology of second morphotype (assigned to be S. tranquebarica), male, 97.85 ± 0.31 mm-A; closer view of the bluntly pointed frontal lobes and the broader anterolateral spines-B, two carpal spines-C.

The third morphotype, identified to be *S. olivacea* is grayish green in colour, while the propodus of the chelae and the last segments of the walking legs are pale orange. The frontal margin is cut into rounded lobes. Anterolateral margin of the carapace cut into 9 broad teeth. Chelipeds are large and the propodus are inflated. The species is characterized by the presence of single spine on the distal half of outer margin of the carpus of the cheliped. Palm of the cheliped exhibits a pair of prominences on the dorsal margin behind the insertion of the dactyl. The inner prominence is larger than the outer one (Plate 7.3). Hexagonal markings are weak or absent.



Plate 7.3: Morphology of third morphotype (assigned to be S. olivacea)male, 97.11± 0.01 mm- A; closer view of the rounder frontal lobes and the broader anterolateral spines, C- single carpal spine, D- the pair of prominences on the dorsal margin behind the insertion of the dacytyl; inner prominence larger than the outer one.

7.3.2 Description of the Third maxillipeds and the first and second male pleopods

The camera lucida study of the first and second male pleopods exhibited variations among the three morphotypes, while the third maxillipeds was found to be similar (fig.7.2). The third maxillipeds have a broad and flattened ischium and are lined with thick hairs on its inner side.



Fig. 7.2: Third maxillipeds of *Scylla* spp – characterized by broad and flattened ischium, lined with thick hairs on its inner side

The male pleopods of the three morphotypes were similar in shape, but showed variation in the pattern of setation. The first male pleopod of the first morphotype (assigned to be *S. serrata*) exhibited two patches of thin and inconspicuous setae on the inner margin on its posterior end. The second male pleopod is smaller than the first pleopod. The tip of the second male pleopod is bilobed and the pleopod lacks any setation (fig.7.3).



Fig. 7.3: Male gonopods of first morphotype (assigned to be *S. serrata)*male, 117.23mm- First pleopods showing two tufts of thin and inconspicuous setae on the inner margin-A1, second pleopods showing its bilobed tip-A2. Scales (1) = 10 mm; (2) = 0.25 mm

The first male pleopod of the second morphotype (assigned to be *S. tranquebarica*) has two patches of setae on the inner margin on its posterior end. Unlike the first morphotype, the setation is thick and conspicuous. The tip of the second male pleopod is bilobed and the pleopods lack setation, similar to the first morphotype (fig.7.4).

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Fig. 7.4: Male gonopods of second morphotype (assigned to be S. tranquebarica)- male, 97.85mm- First pleopods showing two tufts of thick and conspicuous setae on the inner margin-B1, second pleopods showing its bilobed tip-B2.Scales (1) = 10 mm; (2) = 0.25 mm

The first male pleopod of the third morphotype (assigned to be *S.olivacea*) exhibited a single patch of setae, which is thick and prominent. The second male pleopod is bilobed at its tip. Unlike, the first and second morphotype, the second pleopods are found to possess setation on the outer margin on its posterior region (fig.7.5).



Fig.7.5: Male gonopods of third morphotype (assigned to be *S. olivacea*), male, 97.11mm - First pleopods showing single tuft of thick setae on the inner margin-C1, second pleopods showing its bilobed tip and the setae-C2. Scales (1) = 10 mm; (2) = 0.25 mm

7.3.3 Multivariate analysis of Morphometric characters

The forward stepwise Discriminant Function Analysis gave two canonical discriminant functions incorporating 7 morphometric ratios, which together provided 94.9% of discrimination between the three morphotypes of *Scylla*. The 7 morphometric ratios contributing the most to discriminate between the species are ICS/OCS, PW/PL, LSH, AW/SW, ML/PL, OCS/PL and 3PML/ICW (Table.7.2). The group centroid reveals that the first morphotype (*S. serrata*) is entirely different from the other two morphotypes viz., *S. tranquebarica* and *S. olivacea*. A mixing between the second morphotype *S. tranquebarica* and third morphotype (*S. olivacea*) is clearly visible (fig.7.6).

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Table 7.2:	Discriminant	Function	Analysis	(DFA)	table	showing	the	7
	morphometric	characters	which co	ontribute	most	in discrim	inatin	ıg
	the Scylla spp.							

	Entered	Wilk's Lambda						
Step		Statistic dfl	161	df2	df3	Exact F		
			an			Statistic	df1	df2
1	Ratio 24	0.042	1	2	291.000	3360.491	2	291.000
2	Ratio 16	0.024	2	2	291.000	794.852	4	580.000
3	LSH	0.020	3	2	291.000	582.026	6	578.000
4	Ratio 13	0.018	4	2	291.000	464.317	8	576.000
5	Ratio 25	0.016	5	2	291.000	392.092	10	574.000
6	Ratio 23	0.015	6	2	291.000	342.486	12	572.000
7	Ratio 27	0.014	7	2	291.000	297.829	14	570.000



Fig. 7.6: Diagram showing the group centroid of the morphometric characters of the three morphotypes of genus *Scylla*.

7.3.4 Molecular Study

7.3.4.1 First Internal Transcribed Spacer (ITS-1)

The ITS-1 marker produced only two types of band when tested with three morphotypes. The sample identified to be *S. serrata* (with two prominent carpus spines) produced a 1,474bp band, whereas the sample identified to be *S. olivacea* (with single carpus spine) and *S. tranquebarica* (with two carpus spines, one bud like and the other prominent spine like) produced a 1,282 bp band (Fig. 7.7), analogous to *S. serrata* and *S. olivacea* respectively. No amplified product of 1,618 bp was found to match the band reported for *S. tranquebarica* (Imai *et al.*, 2004)

The amplification of ITS-1 region confirms the occurrence of two species of *Scylla* viz., *Scylla serrata* and *S. olivacea* in Cochin Backwaters. The sample identified as *S. tranquebarica* came out to be *S. olivacea* in the molecular study.

7.3.4.2 Cytochrome oxidase c- oxidase sub unit I (COI) gene

When the COI region of the DNA samples isolated from the three morphotypes were amplified and sequenced (using the species specific pimers), a partial COI readable sequence of 658bp were obtained for *S. serrata* and *S. olivacea* (Fig. 7.8). BLAST analysis of the nucleotide sequence of *S. olivacea* showed 98 % similarity to *Scylla olivacea* (GenBank ID KC154078) and *S. serrata* showed 99% similarity to *Scylla serrata* (GenBank ID KC154082). The nucleotide sequences obtained were submitted to GenBank database (*Scylla serrata* GenBank ID AB861521) and *Scylla olivacea* GenBank ID AB861522). The sequences had a GC content of 35.7 and 37.4 respectively.





Fig. 7.7: Electrophoretic pattern from the ITS-1 region of three morphotypes of genus *Scylla* collected from Cochin Backwaters, Lane 1: Blank. Lane 2,9 & 14: 100bp ladder. Lane 3,4,5,6 : First morphotype assigned to be *Scylla serrata*. Lane 7&8: Second morphotype assigned to be *Scylla tranquebarica*. Lane 10, 11,12,13: Third morphotype assigned to be *Scylla olivacea*. Lane 14: Negative control.



Fig. 7.8: Electrophoretic pattern from the CO-1 region (Keenan Primer) of three morphotypes of genus *Scylla* collected from Cochin Backwaters. Lane 1: Blank. Lane 2& 11: 100bp ladder. Lane 3,4,5,6 : First morphotype assigned to be *Scylla serrata*. Lane 7, 8, 9,10: Third morphotype assigned to be *Scylla olivacea*

7.4 Discussion

Attempts to identify *Scylla* species have led to much confusion because of the subtle morphological differences between the species. According to Balasubramanian *et al.* (2014), identification of *Scylla* species has been contentious because of the morphological plasticity, overlapping morphological and morphometric traits.

Initially, mud crabs were originally assigned into three species namely, S. serrata, S. oceanica, S. tranquebarica and one variety, and S. serrata var. paramamosain, based on its external morphology (Estampador, 1949). These four morphotypes were further classified into two groups: 'Banhawin' and 'Mamosain'. Banhawin crabs are free swimming forms characterized with green colour and polygonal markings on all legs and chelipeds, while the Mamosain crabs inhabited holes and possessed dark brown colour, without any markings on legs and chelipeds (Estampador, 1949). In accordance with Estampador's findings, Serene (1952) also recognized the existence of four forms in Vietnam; which were further categorized into the marked and unmarked forms. The marked form comprises a single species S. oceanica and its variety S. oceanica tranquebarica, while the unmarked form comprises S. serrata and its variety S. serrata paramamosain. However, Stephenson and Campbell (1960) considered the four forms as a single species, S. serrata and suggested that the morphological differences may be the result of environmental changes. Ong (1964) also noticed four forms of the genus Scylla from the Malayan waters. Fushimi (1983) reported the presence of three forms of mud crabs, viz., S. serrata, S. tranquebarica and S. oceanica in Hamana Lake, Japan. Later on Oshiro

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(1988) also recognized three species of mud crabs from Japan, which is in agreement with the findings of Fushimi (1983). The genetic variability analysis was carried out by Fuseya and Watanabe (1996) employing horizontal starch gel electrophoresis and they confirmed the existence of three species of mud crabs namely, *S. serrata, S. tranquebarica* and *S. oceanica* in Japan. Keenan *et al.* (1998) made a revision of the genus *Scylla* using specimens collected from the Red Sea and throughout the Indo-Pacific region. Aside from the morphometry and morphological characters, molecular method was used for the identification of four species of *Scylla* viz., *S. serrata, S. tranquebarica, S. olivacea* and *S. paramamosain*.

Several workers have dealt with the genus Scylla from Indian waters like Henderson (1893), Alcock (1899), De Man (1909), Kemp (1915), Gravely (1927), Pillai (1951), Chhapgar (1957). Most of these studies referred to the occurrence of Scylla serrata only. Neither of these works indicated the occurrence of any other forms of *Scylla* in the Indian waters. The validity of their identifications were questioned by Serene (1952) and he opined that the Scylla species commonly found in the Madras markets is S. tranquebarica and the specimens identified as S. serrata consists of at least two forms. Later on, Kathirvel (1981) identified two species of Scylla, a larger and a smaller species from Cochin backwaters and designated them as S. oceanica and S. serrata respectively. Subsequently, Radhakrishnan and Samuel (1982) described the occurrence of a sub species in the Cochin backwaters, which was designated as S. serrata serrata. Joel and Sanjeevaraj (1983) reported the existence of two species of genus Scylla, viz., S. serrata and S. tranquebarica from Pulicut lake. The findings of Radhakrishnan and Samuel (1982) were critically analysed by Kathirvel and

Srinivasagam (1992) and stated that *S. serrata serrata* described by them could be *S. serrata* only.

The previous works on the taxonomy of genus *Scylla* claims that only two species of mud crabs exist in the Indian waters, namely *S. serrata* and *S. tranquebarica* (Joel and Sanjeevaj, 1983, Kathirvel and Srinivasagam, 1992, Kathirvel *et al.* (2004), Mohanty *et al.* (2006), Ajmal Khan and Ravichandran (2009). According to their findings, *S. serrata* forms the smaller species and characterized by brown to dark greenish brown colouration, devoid of hexagonal markings on the limbs, blunt frontal teeth, and a single carpus spine, while *S. tranquebarica* forms the larger species, characterised by grayish green colouration, conspicuous hexagonal markings on the limbs, on the limbs, sharp frontal teeth and presence of two prominent spines on the carpus of the cheliped.

However, according to Keenan *et al.* (1998), Sangthong and Jondeung (2006), S. *serrata* is characterized with bluntly pointed frontal lobes, two spines on the outer margin of the carpus of the cheliped and conspicuous hexagonal markings on chelipeds and limbs. *S. tranquebarica* are also found to possess two spines on the carpus of cheliped, but they can be distinguished with their blunt frontal lobes with rounded interspaces and the pattern of hexagonal markings, i.e weak and inconspicuous markings on the chelipeds and first two pairs of limbs and stronger markings on the last two pairs. *S. olivacea* and *S. paramamosain* are characterized by single spine on the outer margin of the carpus of the cheliped (Keenan *et al.*, 1998; Sangthong and Jondeung, 2006). The observations of Keenan *et al.* (1998), since he has given more concrete evidences on the basis of morphological, morphometric

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and molecular analysis. In the present study three morphotypes of *Scylla*, viz. *S. serrata*, *S. tranquebarica* and *S. olivacea* were recorded in the Cochin backwaters based on the morphological details provided in the identification key of Keenan *et al.* (1998). Some of the recent studies also claims the occurrence of more than two apparent mud crab species from Indian coastal waters, namely, *S. serrata*, *S. tranquebarica* and *S. olivacea*, on the basis of external morphology (Devi and Joseph, 2013a; Trivedi and Vachhrajani, 2013; Devi *et al.*, 2014).

The first and second pairs of male pleopods and the third maxillipeds have been recognized as of taxonomical value by several carcinologists. (Stephenson and Campbell, 1960, Joel and Sanjeevaraj, 1983, Fuseya, 1998, Keenan et al., 1998). The first male pleopods were found to vary in S. serrata and S. tranquebarica (Joel and Sanjeevaraj, 1983). Fuseya (1998) examined the first and the second male pleopods of S. serrata, S. tranquebarica and S. oceanica and found these clearly distinguishable. Keenan et al. (1998) stated that the shapes of first male pleopods are similar in the Scylla spp. examined, however there exist some minor variations which are not clear enough to distinguish between the species easily. According to his observations, the first male pleopods showed variations in the apex region and the pattern of setation. The first male pleopods of Scylla olivacea exhibited long and slender apex, while it is more sinuous in the other three species. As per the illustrations provided by Keenan et al. (1998) the first male pleopods of S. serrata and S. tranquebarica exhibited double setation pattern, while S olivacea and S. paramamosain exhibited single setae on the inner margin, which is in accordance with the observations of the present study. In the present study also, the first male pleopods of the three species were found to be similar in shape, but variations were observed in the pattern of setation (Keenan *et al.*, 1998). The first and second morphotypes assigned to be *S. serrata* and *S. tranquebarica* showed two patches of setae on the inner margin on its posterior end, which is thin and inconspicuous in the former, while thick and conspicuous in the latter. The third morphotype identified to be *S. olivacea* was found to possess only a single tuft of thick setae on the inner margin of the first male pleopod. The second male pleopods were similar in the first and second morphotypes, while found to possess setation on the outer margin on its posterior region of the third morphotype (*S. olivacea*), which is totally absent in the first and second morphotype (*S. serrata* and *S. tranquebarica* respectively)

The third maxillipeds were observed to be an important taxonomic tool in the identification of closely related species of crabs. However, in the present study, no significant variations were observed among the three morphotypes of genus *Scylla*, found in the Cochin backwaters. It was observed that the third maxillipeds of three morphotypes of *Scylla* were characterized by broad and flattened merus and ischium, lined with thick hairs on the inner margin (Fig.7.2).

Multivariate analysis of morphometric characters has shown to be a rapid and effective technique in providing an insight into the discrimination of many animal species, including various types of crustacean; moreover, the hard, well-defined body parts of crustaceans facilitate the collection of accurate data (Overton *et al.*, 1997). Previously, Weber and Galeguillos (1991) confirmed the existence of two species of *Liopetrolisthes mitra*, a

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porcellanid commensal crab from its four morphotypes. Allegrucci et al. (1992) have made use of multivariate analysis of morphometric data to show the interspecific variation between two undescribed species of crayfish, Procamburus spp, discovered from a Mexican Cave and they could confirm them as two sympatric species. In the present study, the multivariate analysis of 27 morphometric ratios formed from 24 morphometric characters were studied to discriminate between the Scylla spp. The study reveals that out of the 27 ratios, 7 ratios contribute mostly to discriminate between the species. The 7 ratios include ICS/OCS, PW/PL, LSH, AW/SW, ML/PL, OCS/PL and 3PML/ICW (Table.7.2). Keenan et al. (1998) also observed seven useful characters to distinguish between the Scylla species viz., ICS/OCS, FMSH/FW, FW/ICW, ML/PL, AW/SW, PL/ICW and IPS/PL. Sangthong and Jondueung (2006) noted nine morphometric characters including FMSH, DFMS, External carapace width (ECW), Frontal margin (FM), Right carpus length (RCL), Right dactyl length (RDL), Right propodus width (RPW), Upper paddle length (UPL) and Body depth (BD) which exhibiting distinctive difference among Scylla species as well as Scylla population. Mandal et al. (2014a) claimed that three ratios viz., ICS/OCS, FMSH/FW and FW/ICW can be considered to discriminate between S. serrata and S. olivacea (all P < 0.01).

Discriminant Function Analysis (DFA) is a statistical application, which is used discriminate between two naturally occurring groups. Quite recently, Jaiswara *et al.* (2013) examined the effectiveness of DFA in the cluster analysis of male field crickets and they claim that the accuracy of discrimination using DFA is high and are not influenced by the number of taxa used. In the present study, when the scattering pattern of Discriminant Function Analysis is taken into account, the group centroid reveals that the

first morphotype (S. serrata) is entirely different from the other two morphotypes viz., S. tranquebarica and S. olivacea. A mixing between the second morphotype (S. tranquebarica) and third morphotype (S. olivacea) is clearly visible in the above analysis. Therefore, from the morphometric study, it is observed that two morphotypes assigned to be S. tranquebarica and S. olivacea are very similar to each other and at the same time, those are clearly different from the first morphotype, which is assigned to be S. serrata. Though a number of ratios are found to contribute in discrimination between different species, Keenan et al. (1998) adds a cautionary note that no single characters can be considered to provide clear discrimination information between the species. Jirapunpipat et al. (2008) conducted a similar morphometric analysis to discriminate between the Thai mud crab populations, but came out to be unsuccessful. They found an overlap of S. serrata and S. tranquebarica although these two species were clearly distinguishable from S. olivacea and S. paramamosain in the Discriminant Function analysis.

The molecular analysis clearly indicates the possibility of the existence of two species of *Scylla* viz., *S. serrata* and *S. olivacea* in the study area Cochin Backwaters. ITS-1 region is the spacer between18s and 5.8s ribosomal RNA. Sequence comparison of ITS-1 region has wide applications in taxonomy and molecular phylogeny related studies since it is easy to amplify even from smaller quantities of DNA and has a high degree of variation even between closely related species. Several workers has made use of ITS-1 region to identify fish and shell fish species (Chow *et al.*, 1993; Bouchon. *et al*, 1994; Masuda *et al.*, 1995; Fernandez *et al.*, 2001). Murphy and Goggin (2001) reported a high divergence rate in the ITS-1 region

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among species of Sacculina carcini. Chu et al. (2001) demonstrated that the PCR product from the ITS-1 region exhibits a high degree of length polymorphism among the crustaceans. Tang et al. (2003) made use of ITS-1 region to resolve the phylogenetic relationship among the Asian mitten crabs of Genus Eriocheir and seven species of Grapsoidea. ITS-1 region was again considered by Perez -Barroz et al. (2008) to study the taxonomic status of two sympatric squat crabs Munida gregaria and M. subrugosa. In the present study, the amplification of ITS-1 region was performed to distinguish between the three morphotypes of Scylla. The ITS1 marker produced only two types of bands, one with a fragment size 1,474 bp and the other with a fragment of 1,282 bp which have concordance with S. serrata and S. olivacea respectively (fig 7.7), as reported by Imai et al. (2004), Mandal et al. (2014a). Imai et al. (2004) made use of ITS-1 region and 16S rDNA to identify the species origin of brood stock and larvae of four Scylla species, viz., S. serrata, S. tranquebarica, S. olivacea and S. paramamosain. When the ITS-1 region of the four Scylla species were amplified, they could observe that S. serrata and S. olivacea were clearly distinguishable from the other two species, S. tranquebarica and S. paramamosain based on the species-specific fragment lengths. S. serrata produced bands with fragment length of 1,474 bp, S. olivacea with a fragment length of 1,282 bp while S. tranquebarica and S. paramamosain produced bands with fragment length of 1618bp. Quite recently, an extensive study was conducted by Mandal et al. (2014a) to resolve the taxonomic uncertainty of mud crabs commonly available in Indian coastal waters using the molecular genetic markers, ITS-1 and CO1 gene, combined with traditional morphometry. In their study, they could observe only two genotypes viz., Genotype A with

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a fragment length of 1,474 bp and Genotype B with a fragment of 1,282 bp which is analogous to *S. serrata* and *S. olivacea* respectively. Hence, the amplification of ITS-1 region of three morphotypes in the present study reveals the existence of only two species of *Scylla*, namely *S. serrata* and *S. olivacea* in Cochin Backwaters. It is also noticed that No amplified product of 1,618 bp was found to match *S. tranquebarica* as reported by Imai *et al.* (2004). The morphotype identified as *S. tranquebarica* produced that same band as *S. olivacea*.

The amplification and sequencing of CO1 gene also confirms the existence of only two species of Scylla, S. serrata and S. olivacea in the Cochin backwaters. When the COI region of the DNA samples isolated from the three morphotypes were amplified and sequenced (using the species specific pimers), a partial COI readable sequence of 658bp were obtained for S. serrata and S. olivacea (Fig.8). BLAST analysis of the nucleotide sequence of S. olivacea showed 98 % similarity to Scylla olivacea (GenBank ID KC154078) and S. serrata showed 99% similarity to Scylla serrata (GenBank ID KC154082). The sequences produced with S. serrata and S. olivacea samples in the present study show enough barcoding gap required for interspecific divergence between the two species. Normally, the barcoding gap between interspecific species is demonstrated to be larger than 0.03 (3% threshold) in more than 98% of closely related lepidopteran species pairs (Hebert et al., 2003) using a CO1- based identification system. Keenan et al. (1998) reported less than 2% sequence difference within and more than 8% between mud crab species over a wide geographic range to conclude the existence of four mud crab species. The sequences of S. serrata had a GC content of 35.7, while that of S. olivacea

had a GC content of 37.4. Mandal *et al.* (2014a) observed a GC content of 36.0 in the sequences of *S. serrata* and 37.7 in the sequences of *S. olivacea*. Molecular Identification of mud cabs using RAPD- PCR-RFLP markers also indicates that only two species of mud crabs *S. serrata* and *S. olivacea* are commonly present in the Indian coastal waters (Mandal *et al.*, 2014b). Balasubramanian *et al.* (2014) also reports two species of *Scylla*, *S. serrata* and *S. olivacea* in the Indian waters after 16s rRNA and CO1 gene analysis.

The morphotype identified to be *S. tranquebarica* came out to be *S. olivacea* in the molecular study and in the multivariate analysis of morphometric characters, it is observed that two morphotypes assigned to be *S. tranquebarica* and *S. olivacea* are very similar to each other and at the same time, those are clearly different from the first morphotype, which is assigned to be *S. serrata*. Therefore, it is likely that the morphotype assigned to be *S. tranquebarica* may be the juveniles of *S. olivacea* originally. Studies conducted by Fuseya and Watanabe (1996), Keenan (1999), Ma *et al.* (2006), Balasubramanian *et al.* (2014) also reports close similarity of *S. tranquebarica* and *S. olivacea* are the result of very recent speciation (Balasubramanian *et al.*, 2014).

The molecular study including the amplification of ITS-1 region and the sequencing of CO1 gene clearly indicates the non existence of *S. tranquebarica* in Cochin backwaters. Regional taxonomic studies carried out in Vietnam (Macintosh *et al.*, 2002); South Africa (Davis, 2004); China (Ma *et al.*, 2006, 2012); Thailand (Somboonna *et al.*, 2010) indicates the
absence of *S. tranquebarica* in these regions. Recently, Mandal et al (2013; 2014a, 2014b), Balasubramanian *et al.* (2014) also reported the absence of *S. tranquebarica* from Indian waters. However, the fact cannot be denied that *S. tranquebarica* was originally described from India by Fabricius (1798) probably from a specimen obtained from Tranquebar (Taragambadi). Later Keenan *et al.*, (1998) also reported existence of *S. tranquebarica* in the Indian sub continent (type locality: South India). Therefore, it is unlikely to exclude the possibility of existence of *S. tranquebarica* is relatively less abundant even in its distributional range. The absence of *S. tranquebarica* in Indian waters (Balasubramanian *et al.* 2014).

To conclude, the present study clearly indicates the occurrence of two species of mud crab namely, *S. serrata* and *S. olivacea* in the Cochin Backwaters. Shaji *et al.* (2006), Mandal *et al* (2014 a,b), Balasubramanian *et al* (2014) and Devi and Joseph (2015) also claims that *S. serrata* and *S. olivacae* are the most common species occurring in India, based on the morphological characters, morphometry as well as molecular studies. It can be concluded that the green morph of Indian mud crab is *S. serrata* and the brown morph is *S. olivacea*

Moreover, the present work is the first time report of the occurrence of *S. olivacea* from Cochin backwaters. The occurrence of *S. olivacea* in the Indian waters was first reported by Shaji *et al.* (2006). Quite recently, Trivedi and Vachharajani (2013) recorded the existence of *S. olivacea* from Gujarat and Padate *et al.* (2013) claimed the new record of *S. olivacea* from

Goan waters. However, the existence of *S. olivacea* from Cochin Backwaters has not been reported before. Hence the present work can be counted as the first report of the occurrence of *S. olivacea* from the Cochin backwaters.

Part - III

Chapter 8 SALIENT FINDINGS OF THE STUDY

✤ BIBLIOGRAPHY

✤ PUBLICATIONS

Chapter **8**

SALIENT FINDINGS OF THE STUDY

- Diversity study revealed the occurrence of 23 species of brachyuran crabs belonging to 16 genera and 8 families in the study area Cochin Backwaters.
- Among the families, the highest number of species was recorded from Family Portunidae (8 species) which was followed by Grapsidae (7 species). The list also included crabs belonging to Family Ocypodidae (2 species belonging to 2 genera), Family Hymenososmatidae (2 species), Family Majidae (1 species), Family Pinnotheridae (1 species), Family Leucosidae (1 species) and Family Xanthidae (1 species).
- Among the 23 crab species enlisted from the Cochin backwaters, 5 species are of commercial importance and contribute a major share to the crustacean fishery of the Cochin region. They are *Scylla serrata*, *S. olivacea, Portunus pelagicus, P. sanguinolentus* and *Charybdis feriata*.
- It was observed that, the Cochin backwaters is invaded by certain marine migrant species during the Post monsoon and Pre monsoon

periods and they are found to disappear with the onset of monsoon. These marine migrant species include *P. pelagicus*, *P. sanguinolentus*, *C. feriata*, *C. lucifera*, *C. hoplites* and *Doclea gracilipes*.

- Among the 23 species recorded from the Cochin backwaters, 13 species were found to occur in the mangrove dominated regions, belonging to four families. They include *Scylla serrata*, *S. olivacea*, *Thalamita crenata*, *Nanosesarma batavicum*, *N. andersonii*, *Neoepisesarma brockii*, *N. mederii*, *Metapograpsus messor*, *M. maculatus*, *Varuna litterata*, *Macrophthalmus depressus*, *Uca annulipes* and *Heteropanope indica*. Except, *V. litterata*, the other 6 grapsid crabs were found exclusively in the mangrove regions and nowhere else.
- The study reports the occurrence of the 'herring bow crab' Varuna litterata in the Cochin backwaters for the first time.
- The analysis of water quality and sediment quality parameters indicated seasonal as well as spatial variations in these ecological variables, which in turn influenced the distribution and abundance of crabs.
- Ecological studies showed that the substratum characteristics influence the occurrence, distribution and abundance of crabs in the sampling stations rather than water quality parameters.
- The variables which affected the crab distribution the most were Salinity, moisture content in the sediment, organic carbon and the sediment texture.

- Besides the water and sediment quality parameters, the most important factor influencing the distribution of crabs is the presence of mangroves. The study indicated that the diversity as well as distribution of crabs was found to be maximum in mangrove zones when compared to other stations devoid of mangroves.
- The study also revealed that most of the crabs encountered from the study area preferred a muddy substratum, with high organic carbon content and high moisture content.
- The shape of the male pleopods and male abdomen act as a powerful taxonomic tool for the species level identification of brachyuran crabs.
- Though third maxilliped is also considered to be an important taxonomic tool by other carcinologists, in the present study it was observed that this structure did not show variation in species level, but it can be considered for family or sub family level identification.
- In the present study, an identification key is presented for the brachyuran crabs occurring along the study area the Cochin backwaters and the associated mangrove patches, taking into account the morphological characters coupled with the structure of third maxillipeds, first pleopods of males and the shape of male abdomen.
- The initial examination of the morphological characters indicated the presence of three morphotypes of genus *Scylla* in the Cochin backwaters, viz., *Scylla serrata*, *S. tranquebarica* and *S. olivacea* based on the identification key provided by Keenan *et al.* (1998). However, the morphometric study and the molecular analysis involving the

amplification of ITS-1 region and sequencing of CO1 gene confirms that only two species of *Scylla* exist in the Cochin backwaters, namely *S. serrata* and *S. olivacea*.

- Though morphological examination indicated the existence of a morphotype which is comparable with the morphological features of *S. tranquebarica*, the morphometric study and the molecular analyses confirmed the non existence of *S. tranquebarica* in the Cochin backwaters. The morphotype identified to be *S. tranquebarica* came out to be *S. olivacea* in the molecular study and in the multivariate analysis of morphometric characters, it was observed that the two morphotypes assigned to be *S. tranquebarica* and *S. olivacea* were very similar to each other. At the same time, the second and third morphotypes were clearly different from the first morphotype, which was assigned to be *S. serrata*. Therefore, it is likely that the morphotype identified to be *S. tranquebarica* in the initial examination may be the juveniles of *S. olivacea* originally.
- Recently, the existence of *S. olivacea* is being reported from several different regions of India; however the existence of *S. olivacea* from Cochin backwaters has not been reported yet. Hence the present work can be counted as the first report of the occurrence of *S. olivacea* from Cochin Backwaters.

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Publications

List of Publications

Published Manuscripts – 1 Journal publication, 1 Book chapter and 1 Proceedings paper

- 1) **Devi PL** and Joseph A (2013) Habitat ecology and food and feeding of the herring bow crab *Varuna litterata* (Fabricius, 1798) of Cochin Backwaters, Kerala, India. *Arthropods*, 2(4), 172-188.
- 2) Devi PL, A. Joseph and S. Ajmal Khan (2014) Diversity of brachyuran crabs of Cochin backwaters, Kerala, India. In: K. Venkataraman and C. Sivaperuman (eds.) Marine faunal Diversity in India: Taxonomy, Ecology and Conservation, Zoological Survey of India, Academic Press, Elsevier, Oxford, UK. 75- 87.
- 3) Joseph A, **Devi PL** and Anup Mandal. (2014). On the record of the brown mud crab *Scylla olivacea* (Herbst, 1796) from Cochin Backwaters, South West coast of India. Proceedings, 24th Swadeshi Science Congress. Pp. 489-495. ISBN: 978-81-928129-2-2.

Manuscripts accepted for publication

- A note on the taxonomy of the Genus *Scylla* (De Haan) of Cochin Backwaters, Kerala, India
 P.Lakshmi Devi and Aneykutty Joseph Accepted for publication in *Indian Journal of Fisheries*
- On the record of herring bow crab Varuna litterata from Cochin Backwaters, Kerala, South India
 P. Lakshmi Devi and Aneykutty Joseph Accepted for publication in Indian Journal of Geo-Marine Sciences
- Distribution and Habitat of Brachyuran Crabs of Cochin Backwaters, Kerala, South India
 P.Lakshmi Devi and Aneykutty Joseph Accepted for publication in *Fishery Technology*
- Scylla species found in Cochin Backwaters, Kerala, South India.
 P.Lakshmi Devi and Aneykutty Joseph
 Full length paper in the proceedings of International Seminar-Workshop on 'Mud crab Aquaculture and fisheries Management' held at Rajiv Gandhi Centre for Aquaculture, (ISMAF-2013), Sirkazhi, Nagapattinam, Tamil Nadu, from 10-12 April, 2013.

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Morphometry of the Herring bow crab, *Varuna litterata* (Fabricius, 1798) from Cochin Backwaters, a South Indian Estuary.

Deepthi Gopi Nair, P.Lakshmi Devi, Alphi Korath and Aneykutty Joseph

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Article

Habitat ecology and food and feeding of the herring bow crab *Varuna litterata* (Fabricius, 1798) of Cochin backwaters, Kerala, India

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Abstract

Habitat ecology and food and feeding of the herring bow crab, *Varuna litterata* of Cochin Backwaters, Kerala, India were investigated for a period of one year (April 2011-March 2012). Among the 15 stations surveyed, the crabs were found to occur only in 4 stations, which had a close proximity to the sea. Sediment analysis of the stations revealed that the substratum of these stations is sandy in nature and is rich in organic carbon content (0.79% to 1.07%). These estuarine crabs is euryhaline and are found to be distributed in areas with a sandy substratum, higher organic carbon content and more tidal influx. The stomach contents analysis of crabs examined showed that their diet included crustacean remains, plants, sand and debris, fishes, miscellaneous group and unidentified matter. In adults and sub-adults, crustaceans formed the dominant food group, while in juveniles, sand and debris formed the dominant group. From the present study, *V. litterata* was found to be a predatory omnivore capable of ingesting both animal and plant tissues.

Keywords habitat ecology; food and feeding; Varuna litterata; Cochin backwaters.

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1 Introduction

Varuna litterata, is a grapsid crab, commonly known as the 'herring bow crab'. They usually inhabit the mangroves, estuarine and freshwater environments, in shallow sub tidal regions and usually found under rocks, logs and dead leaves and lives in burrows along the embankments or sides of pools, creeks and shallow banks. *V. litterata* is euryhaline and can be found in rivers, brackish waters or at sea. With its legs shaped as paddles used for swimming, it is sometimes called the 'Paddler crab'. After a flood, it can be washed far away in the sea and observed there drifting with pieces of debris (Bouchard et al., 2011). They are found to prefer areas faced to more oceanic waters (www.sealifebase.fisheries.ubc.ca). It has been reported from all over the Indo-West Pacific (Alcock, 1900; Sakai, 1939, 1976; Barnard, 1950; Crosnier, 1965; Holthius, 1978).

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Though there are lots of works being carried out in brachyuran crab fauna throughout the world, the compilation of studies conducted on V. litterata is comparatively few in number particularly with reference to India. One of the first studies on V. litterata is by Kemp (1915), who reported the mass migration of the megalopae in the Hooghly River near Calcutta, India. A Similar phenomenon was observed in Fiji in 1987 (Ryan and Choy, 1990). Panikker (1951) and Gross et al. (1996) dealt with the osmoregulation in V. litterata and proved that they show strong osmoregulation even in 150% seawater. The metabolic effect of eyestalk removal in V. litterata was studied by Madhyastha and Rangneker (1974). Behavioral study of V. litterata in the coastal waters of Bay of Bengal was carried out by Manna (1988). Tu Chin-Hung (1992) documented the whole zoeal development of V. litterata. Jameieson et al. (1996) analysed the spermatozoa ultrastrucure of V. litterata. It has been recorded as commercially important species in Bengal, India, where it is eaten by poor people, and its numbers compensate for its small size (Hora, 1933). Rao et al. (1973) also reported it as an edible species and a fishery resource of India. In the West coast of India, V. litterata was first collected by Pillai (1951) from the open sea at Trivandrum and later by Chhapgar (1957) from Bombay and Kolak. Dineshbabu et al. (2011) reported the species from waters along Karnataka coast. It was neither reported from Travancore backwaters (Pillai, 1951) nor Vembanad Lake (Roy and Nandi, 2008). Devi et al. (2013) studied the brachyuran crab diversity of Cochin backwaters and reported that this species is available plentifully in certain sampling localities of the area studied. However, little has been reported on the habitat ecology and food and feeding of the grapsid crabs of Indian waters.

Several authors have dealt with the ecological aspects of various crab species. Dahdough-Guebas (1994) studied the feeding ecology and behavioral ecology of some selected crab species from the Kenyan mangroves. Feeding ecology of velvet swimming crab, *Necora puber* was studied by Freire and Gonzalez-Gurriaran (1995), while Lee (2004) dealt with the ecology and behavior of postlarvae and juveniles of *Necora puber*. Mokhtari et al. (2008) dealt with the population ecology of the fiddler crab *Uca lactea annulipes*. Feeding ecology of the American crab *Rithropanopeus harisii* was studied by Hegele-Drywa and Normant (2009). Pandya et al. (2010) studied the spatial distribution and substratum preference of *Macrophthalmus depressus*. Ethnoecology of *Scylla serrata* was studied by Nirmale et al. (2012). Trivedi et al. (2012) studied the diversity and habitat preference of brachyuran crabs in the Gulf of Kutch.

There are several reports on the food and feeding of various species like, *Scylla serrata* (Williams, 1978; Hill, 1976, 1980; Joel and Raj, 1986; Prasad and Neelakantan, 1988), *Portunus spp* (Hill, 1980; Stephenson et al., 1982; Campbell, 1984; Wassenberg and Hill, 1987; Sumpton and Smith, 1990; Josileen, 2011), *Thalamita crenata* (H.Milne Edwards 1834) (Canicci et al., 1996), *Callinectes sp.* (Tagatz, 1968; Paul, 1981; Laughlin, 1982; Stoner and Buchanan, 1990; Rosas et al., 1994). *Chionocetes opilio*, (Wieczorek & Hopper, 1995). Jewett and Feder (1982, 1983) analysed the food of the king crab, *Paralithodes camtschatius* and that of the tanner crab, *Chionocetes bairidi*. The natural diet and feeding habits of two species of *Liocarcinus* was described by Choy (1986) and that of *Cancer* spp. and *Ovalipes ocellatus* was dealt with by Stehlik (1993). However, there is no much information on the habitat preference, diet and preferred food items of *V. litterata*. Hence, the study has been undertaken to investigate the habitat ecology and food and feeding habits of *V. litterata* from Cochin backwaters, Kerala, India.

2 Materials and Methods

2.1 Description of the study area

Cochin backwaters situated at the tip of northern Vembanad Lake is a tropical positive estuarine system extending between $9^{0}40'$ & $10^{0}12'$ N and $76^{0}10'$ & $76^{0}30'$ E, with its northern boundary at Azheekode and southern boundary at Thannirmukham bund (Fig. 1). The salinity gradient of Cochin backwaters supports

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diverse species of flora and fauna depending on their capacity to tolerate oligohaline, mesohaline or marine conditions. Low lying swamps and tidal creeks, dominated by sparse patches of mangroves with their nutrient rich physical environment, support larvae and juveniles of many commercially important species. Backwaters also act as nursery grounds of commercially important fin fishes and shell fishes. (Menon et al., 2000).

On the basis of salinity, 15 study stations was selected along the study area, namely, Thannermukham, Vaikom, Aroor, Valanthakad, Kandakkadavu, Kumbalanghi, Thevara, Thoppumpady, Barmouth, Marine Science Jetty, Varappuzha, Vallarpadam, Vypin, Munambam and Azheekode. A detailed survey along these stations was conducted to find the availability of this species (Fig. 1).

2.2 Physiochemical parameters

Standard techniques were employed for the analysis of water quality parameters viz. temperature (centigrade thermometer), salinity (salino-refractometer), pH (digital pH meter) and the sediment quality parameters, viz. Organic carbon content (Walkley-Black method, Jackson, 1973) and the sediment texture (particle size analyzer., Sympatec – Germany).

2.3 Food and feeding studies

Specimens of *V. litterata* were collected once a month for a period from April 2011-March 2012. After recording the carapace width (CW), carapace length (CL), total weight and sex for all the captured specimens, the dorsal side of the body was cut open and foregut was removed carefully. A visual estimate of the fullness of the stomach was made immediately after its removal as about 100%, 75%, 50%, 25% and 0% according to the degree of fullness. The foreguts were transferred in 70% alcohol (Williams, 1981) and all the stomachs were subsequently opened and their contents were washed with alcohol into a petridish and gut contents were separated and identified into different food groups under a binocular microscope.

As characteristic of brachyurans, most of the food items were found to be highly crushed down into small fragments and hence only those structures that could be identified were relied upon for determining food composition and evaluation. Gut contents were broadly classified as suggested by Sukumaran and Neelakantan (1997), Josileen (2011).

- 1) Crustacean remains : Penaeid prawns body parts, appendages, telson, eggs, crabs and stoamatopods.
- 2) Fish remains : fins, scales, bones and vertebrae.
- 3) Molluscan remains : gastropods and bivalves.
- 4) Miscallaneous items : algal filaments, nematodes, polychaetes, ophiuroides and unidentified items.
- 5) Debris : sand and mud

Only stomachs with food (n=145) were considered for calculation. The whole stomach contents of all the specimens were segregated food-group wise and each group's contribution was assessed visually. Dominance of food groups was evaluated by ranking them by its percentage frequency of occurrence and percentage points (Sukumaran and Neelankantan, 1997). As described in detail by Wear and Haddon (1987), in the Percentage Points method each of the more common food categories (in this study) is given a value, ranging from 0 to 100, according to the percentage of content it represents within each stomach. Then the number of points each category received are weighted according to the real fullness of the stomach in which it was found. For instance, in a stomach half full, containing 25% bivalves and 75% algae, the bivalves were scored 12.5 and algae 37.5 points, respectively (Cannicci et al., 1996).

Frequency of occurrence is calculated by dividing the number of stomachs which contained a food category by the total number of stomachs observed. The following percentages were calculated for each prey type (Williams, 1981):

Percentage points for i^{th} prey = $(\sum_{i=1}^{n} a_{ij}/A)*100$

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Percentage occurrence for i^{th} prey = $(b_i / N)100$

where a_{ij} = the number of points for prey item i in the foregut of the jth crab; A = the total points for all the crabs and all the prey items in all the foreguts examined; N = the number of crabs examined with food in the foregut; and b_i = the number of crabs with foreguts containing prey category i.

3 Results

A detailed survey was conducted along the study area, to find the availability of the species. Among the 15 stations fixed, *V. litterata* was found to occur only in 4 stations namely, Kandakkadavu, Kumbalanghi, Vypin and Munambam. (Fig. 1-higlited with arrow marks). A detailed study was then conducted on the location type, habitat and the physico-chemical parameters of all the 15 stations, to find the peculiarity of the four above said stations and thus draw an inference on the habitat type preferred by the species.

Site 1- Kandakkadavu, which lies at the west part of the backwaters is a typical coastal area ornamented with lush green mangroves. Site 2- Kumbalanghi is a fishing village, and has a very vast backwater stretch, Here; the backwater is in an enclosed condition with a rather narrow opening into the main lake. This site too has a rich diversity of mangroves and mangrove associates. Site 3- Vypin is the biggest among the group of island in the Cochin backwaters and is bound by Arabian Sea on its western side and the backwaters on its eastern side. The station had a luxuriant mangrove patch earlier, but the recent LNG and Petroleum tanks and terminals built at the heartland of mangroves in Vypin has destroyed the vegetation on a mass scale. Site 4-Munamabam is a coastal village, situated at the Northern point of the backwaters, surrounded by Arabian Sea on the West, Periyar on the East and a mouth opening to the sea on the North. The site does not have luxuriant mangrove vegetation, only a sparse patch is found.

3.1 Physiochemical parameters

Water quality and sediment quality parameters of the 15 stations were anlaysed on a monthly basis, however, emphasis is given to those four stations, viz. Kandakkadavu, Kumbalanghi, Vypin and Munambam, where *V. litterata* is found.

In general, a wide fluctuation was observed in the salinity and temperature during the study period. A low salinity was recorded throughout the study area during the monsoon season and it increased during the post monsoon period and the salinity ranged between 0-17 ppt. Highest salinity was observed during the post monsoon period. The salinity ranged from 2-31 ppt with a mean of 12.08 ppt at site-1, Kandakkadavu, 1-22ppt with a mean of 10.5 ppt at site-2, Kumbalanghi; 0-16 ppt with a mean of 6.08 ppt at site-3, Vypin and 4-29 ppt with a mean of 15.83 ppt at site-4, Munambam during the study period. Temperature too showed a wide fluctuation. Site-1 recorded a temperature range of 23-30^oC with a mean 26.91^oC, site- 2 recorded a range 25-28^oC with a mean of 26.41^oC; site-3, 23-27^oC with a mean of 25.25^oC and site-4 with a range of 28-32^oC with a mean of 29.41^oC. pH was almost neutral throughout the sites, throughout the study period. It fluctuated between 6.43-7.89 with a mean of 6.78 in site-1, 6.18-7.98 with a mean of 6.96 in site-2, 6.09-7.33 with a mean of 6.71 in site-3 and 7.23-8.21 with a mean of 7.76 in site-4. The range of variation in the salinity, temperature and pH of the study stations, during the study period is presented in Table.1.

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Sl.No	Sampling stations	Salinity (ppt)	Temperature (⁰ C)	рН
1	Thannermukham	0-13	24-28	6.32-7.58
2	Vaikom	0-12	20-29	6.43-7.9
3	Aroor	0-16	21-29	6.09-7.45
4	Valanthakad	0-16	23-28	6.18-7.98
5	Kandakkadavu	2-31	23-30	6.43-7.89
6	Kumbalanghi	1-22	24-28	6.17-7.98
7	Thevara	0-22	23-29	6.45-7.97
8	Thoppumpady	1-26	25-30	6.56-7.85
9	Bar Mouth	2-28	26-29	6.54-7.94
10	Marine Science Jetty	0-23	25-29	6.50-8.01
11	Varappuzha	0-8	23-28	4.92-7.01
12	Vallarpadam	2-18	28-32	7.09-7.90
13	Vypin	0-16	23-27	6.22-7.11
14	Munambam	4-29	28-32	7.25-8.88
15	Azheeekode	4-33	28-32	7.25-8.88

Table 1 Range of salinity, temperature and pH of the study stations during the study period.





Fig. 1 Map of the study area, Cochin Backwaters with the sampling stations.

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The sediment analysis of the sampling stations revealed three types of sediments in the study area- sandy. silty and clayey. The substratum of 5 stations, viz Vaikom, Kandakkadavu, Kumbalanghi, Vypin and Munambam was found to be sandy in nature and its composition is 53.77%, 50.24%, 63.94%, 43.81% and 48.32% respectively . The stations with a silty substratum are Thanneermukham, Valanthakad, Thevara, Varappuzha and Azheekode with a composition of 45.83%, 43.05%, 48.99%, 41.98% and 53.06% respectively and the 4 stations, viz Aroor Thoppumpady, Marine Science Jetty and Vallarpadam was found to be clearly clayey in nature with a composition of 62.39%, 44.91%, 56.43% and 49.01% respectively. The substratum of the station Barmouth is found to have an equal composition of silt and clay (40.91% silt and 40.8% clay). The percentage composition of sand, silt and clay of the sampling stations are given in Fig.2. The sediment of the stations showed varying amount of organic carbon content. Out of the 15 stations, Aroor (1.08), Valanthakad (1.04), Kandakkadavu (1.00), Kumbalanghi (1.02), Vallarpadam (0.99), Vypin (1.07) and Munambam (0.79) recorded higher organic carbon content, owing to the litter fall from the mangroves while Thanneermukham (0.66), Thevara (0.59), Thoppumpady (0.57), Barmouth (0.67), Marine Science Jetty (0.73) and Azheekode (0.72) exhibited medium organic carbon content. Vaikom (0.49) and Varappuzha (0.48) are the two stations which reported least amount of organic carbon. Luxuriant mangrove patches are observed from Aroor, Valanthakad, Kandakkadavu, Kumbalanghi, Vallarpadam, Vypin and some sparse patches from Munambam. The average organic carbon content of the sampling stations are given in Fig.3

3.2 Food and feeding studies

The stomach contents of *V. litterata* appeared to contain highly digested matter and hence identification of food organisms was found difficult. However, the natural diet of the animal was found to consist mainly crustacean remains, fish, plants, sand and debris and miscellaneous group of items but could not find the molluscan remains. Out of the total 321 stomachs analyzed, 13.79% were 100% full, 6.89% were 75% full, 9.65% were 50% full, 24.13% were 25% full and 45.15% were empty. The month wise and size wise details on the stomach fullness are given in Fig.4 and Fig.5.

When percentage of frequency of occurrence is analyzed, the crustacean remains comprised 98.46%; sand and debris 81.53%; plant remains 44.61%, and the miscallaneous items comprised 23.07%.(Fig.6). The points of major food groups (size wise and month wise) were estimated and the results are depicted in Fig.7& Fig.8. In percentage of points method using the overall sample, crustacean remains was the most dominant food. It was found in almost all stomach analyzed and consisted primarily of decapods (parts of shrimp rostrum, exoskeleton, appendages etc.), amphipods, isopods and stomatopods. The maximum percentage of points (93.33%) was observed in July. In different size groups, crustacean remains varied from 5.68% to 88.23%.

The second dominant item of food was sand and debris. It ranges from 4.19% to 56.1% in different months. The maximum amount was found in the month of September and the least was observed in July. Percentage points varied from 5.41% to 72.41% in various size groups, the maximum found in size class 1.5 - 2.5 cm class and minimum found in 3.5 - 4.0 cm size class. This group consists mainly of sand particles and detritus especially of plant origin.

The third predominant item of food was plant remains. It ranges from 0% to 21.65% in different months. The maximum amount plant remains was found in the gut during May and no remains found in the gut during September. Percentage points varied from 1.27% to 18.09% in various size groups, the maximum found in size class 3.5 - 4.0 cm class and minimum in 4.0 - 4.5 cm size class. This group consists mainly of algal filaments, mangrove leaf litter and other partially digested matter.

The fourth predominant item of food was fish remains. It ranges from 0% to 18.75% in different months. The maximum amount was found in November, while no fish remains is recorded in January, September and December .In various size class, percentage points varied from 0% to 4.11%, the maximum found in size class

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2.5 - 3.0 cm class and minimum found in 1.5 - 2.0 cm size class. This group consists mainly of fins, spines, scales, bones and vertebrae.

The fifth predominant item of food was miscellaneous items. It was rarely found in the analyzed stomachs. It ranges from 0% to 43.75% during the study period. The maximum amount was found in January and no such content was observed in the month of October. Percentage points varied from 0% to 25%, the maximum found in size class 4.0 - 4.5 cm class and no miscellaneous items found in 1.5 - 2.0 cm size class. This group consists mainly of nematodes, helminthes, polychaetes and other unidentified matter.

When the quantity of food and points of major food groups' sex wise were estimated, the results showed that there is no significant difference, that it is almost similar.







Fig. 3 Average organic carbon content in the sediments of the study area.

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Fig. 4 Stomach fullness during various months in V.litterata.



Fig. 5 Stomach fullness in various size groups in *V.litterata*.

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Fig. 6 Frequency of occurrence of food items in the gut content of V.litterata.



Fig. 7 Percentage points of major food groups during various months in V.litterata.

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Fig. 8 Percentage points of major food groups in various size groups of V.litterata.

4 Discussion

In the present study, *V. litterata* is found to occur only in four stations among the 15 stations surveyed, namely Kandakkadavu, Kumbalanghi, Vypin and Munambam, which exhibited a sandy substratum with high organic carbon content and good tidal influx due to more proximity to the sea. The higher amount of organic carbon in the sediments can be attributed to the litter fall from mangroves. Vaikom is one another station observed to have a sandy substratum, but when the organic carbon content of the sediment is considered, it exhibited a lower value. The lower organic carbon content may be due to the absence of mangrove patches in the area. It also exhibits poor tidal influx, since it is much away from the sea. Besides, these four stations, where *V. litterata* is found to be available, mangrove patches are also observed from three other stations viz. Aroor, Valanthakad and Vallarpadam. The substratum of these three stations. This may be due to the sediment texture prevailing in these three stations, Aroor being clayey and Valanthakad and Vallarpadam being silty in nature. These stations also records lower tidal influx due to the distance from the sea.

V. litterata inhabit mangrove regions (http://mangrove.nus.edu.sg/pub/seashore/text/197.htm) and prefer areas faced by more tidal waters, (www.sealifebase.fisheries.ubc.ca), which is in complete agreement with the present study. The four stations where the crabs are found to occur are mangrove regions with a close proximity to the sea. Though stations like Barmouth, Thevara, Thoppumpady, Marine Science Jetty and Azheekode are also closer to the sea, the substratum characteristics, amount of organic carbon in the sediments and the absence of mangrove patches make it unsuitable for the species. Therefore, the present study reveals that *V. litterata* is an inhabitant of mangrove regions with sandy substratum, rich in organic carbon and prefer areas with close proximity to the sea having good tidal influx.

There is remarkable fluctuation in the physico chemical parameters, temperature and salinity of the study stations during the study period, however, the stations showed a neutral pH during the period. The occurrence of *V. litterata* in the study area throughout the study period shows that, it has a wide range of tolerance to temperature and salinity. This remarkable species is euryhaline and can live in both fresh water and saltwater,

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being found not only in streams and ponds far inland, but also in mangroves, and the edge of the shore (http://mangrove.nus.edu.sg/pub/seashore/text/197.htm). Panikker (1951) reported that they are able to regulate their internal salinities and can accomplish osmoregulation in a variety of salinity and temperature combinations to a certain extent. Gross et al. (1996) proved that they show strong osmoregulation even in 150% seawater.

Knowledge on natural diet is essential for studies of its nutritional requirements, its interactions with other organisms and its potential for culture (Williams, 1981). Though the diet of decapods is primarily macroscopic items, their mouth parts and gastric mill ossicles reduce the food to small fragments and hence the identification of food types and quantities is often difficult. The points method and the occurrence method are the only methods which can be applied readily to the analysis of the gut contents in crabs. The structure of each type of food and the way in which it is manipulated prior to ingestion, however, affect the accuracy of the percentage points and, to a lesser extent, percentage occurrence of some types of food. Percentage points is an approximate volume measure and since the soft parts of most foods rapidly become unrecognizable after ingestion, it is inaccurate for most foods. The points method is however is suitable for foods which are ingested in large recognizable pieces or in their entirety. Percentage occurrence is a measure of the regularity of inclusion of a food in the diet of a sample. It is inaccurate only for foods which have no recognizable hard parts but it may give an inflated relative importance to small food items.

In the present study, it was observed that *V. litterata* is an opportunistic omnivore, crustaceans being the most dominant item followed by sand and debris and plant remains. Crabs are opportunistic omnivores with preferences for animal prey, but rarely fed on more mobile prey such as fish and prawns (Williams, 1982). Warner (1977) is also of the opinion that crabs carry over the primitive behavior of being opportunistic omnivores with a preference of animal food in conjunction with predatory propensity. On comparing it with the studies conducted by other workers in different crabs, Sukumaran and Neelakantan (1997) reported that *P. pelagicus* from the Mangalore coast preferred crustaceans followed by fishes and molluscs. Josileen (2011) observed a similar result for the *P. pelagicus* collected from Mandapam coast All studied crabs are reported to consume mixed diets of molluscs, crustaceans, fishes, polycheates etc. (*S. serrata:* Hill, 1976; Joel and Raj, 1986; Prasad and Neelakantan, 1988), *C. sapidus:* Laughlin, 1982; *S. tranquebarica:* Joel and Raj, 1986; *Thalamita crenata:* Cannici et al., 1996).

Presence of debris and detritus in the stomach contents of V. litterata suggested that these crabs are omnivorous consuming plant as well as animal components. The detritus found in the stomachs examined mainly belonged to plant matter. It is increasingly recognized that coastal and estuarine food chains are based to a significant extent on detritus and dissolved organic matter produced from the breakdown of rooted and attached macrophytes of intertidal and shallow subtidal habitats. In the present study, the specimens have been collected from the sites where there is lush green mangrove patches. Therefore the detritus of plant origin found in the gut content of the crabs will be owing to the mangrove leaf litter of the area. Ground-dwelling grapsid crabs have been reported to be the main consumers of fresh mangrove litter, particularly in Indo-Pacific systems (Lee, 1998; Ravichandran et al., 2009) and have identified as keystone species in the tropical Indo-west Pacific mangrove ecosystems (Ajmal Khan and Ravichandran, 2008). Fresh mangrove litter has low nutrient (C/N ratio > 50) and high feeding deterrent (e.g. soluble tannins) content, making it an unattractive staple food item (Wolcott & O'Connor, 1992). Some grapsid crab species prefer leaf litter with specific C/N ratios (Lee, 1993), while others feed indiscriminately (Micheli, 1993). From the present study, it is observed that V.litterata inhabit areas which are rich in organic carbon content. The presence of considerable amount of sand particles in the gut is in total agreement with the sediment texture of the study sites, since sand was the main constituent of the sediment in all the four study sites. Therefore, it is obvious to find good amount of

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sand particles in the gut contents, which may be ingested accidently while gleaning the food particles from the substratum.

Molluscs form an important component in the gut contents of most of the omnivorous crabs, like Scylla serrata (Hill, 1976) and Portunus pelagicus (Sukumaran and Neelakantan, 1997; Josileen, 2011). In contradictory to their findings no molluscan remains were found in the gut contents of V. litterata throughout the study. This can be attributed to the cheliped strength of the crabs. The chelipeds of V. litterata may not be strong enough to crush the shells of the molluscs or it is also possible that, the crabs do not prefer sedentary organisms like molluscs. Arimoro and Idoro (2007) also reported the absence of molluscs from the gut contents of Callinectes amnicola, but they have not furnished the possible reasons for the absence of molluscs. Generally feeding takes place every day throughout the year except during few days of moulting and mating when feeding ceases or it is at its minimum. The study period has been divided into premonsoon, monsoon and post monsoon and the majority of the empty stomachs encountered in the study were during the period from April to July, i.e., during the late premonsoon and early monsoon period. Choy (1986) reported empty stomachs in gravid females and parasitized crabs during the winter. He also observed the newly moulted crabs having empty stomachs during the summer season. Jewett and Feder (1982) reported that in king crab Paralithoides sp, feeding increases immediately after spawning during spring and summer. In contrast to this, Jewett and Feder (1983) observed that the tanner crabs, Chionocetes bairdi feed more intensely during the nonspawning periods (i.e., November to February). The presence of empty stomachs during the late premonsoon and early monsoon period in the present study indicates that V. litterata breeds during this period. Kemp (1915) observed that every year at the commencement of monsoon, the waters of Hooghly river near Calcutta, India, teem with the megalopes of V. litterata. The behavioural study of V. litterata carried out by Manna (1988) in the coastal waters of Bay of Bengal reports that V. litterata occur abundantly once in a year on the new moon day of July from midnight to 0900 hours. On that day numerous crabs float on the surface of water embracing themselves like clustering balls. This unusual physiological behavior is possibly due to the sudden increase of hormonal levels stimulating them for mating. Tu, Chin Hung (1992) observed that the features of ovaries of V. litterata caught from Kao-ping river, change from brown and inseparable to black and disperse the diameter increase from 0.2mm to 0.32 mm during March to June. This further proves that this species breeds during the late premonsoon and the monsoon periods. However, no berried females were captured from the study area, during the period of study. Kemp (1915) observed mass migration of megalopes in the Hughli river near Calcutta as well as the Gangetic delta and stated that the adult migrate to the estuaries to breed. However, he could not find any ovigerous females of V. litterata from the Chilka lake, which is in accordance to the present study. He reported that they are found to occur in myriads, where the current is sluggish and in small creeks. The sampling sites from where the V. litterata samples are observed and captured have good tidal influx and are open waters, devoid of any creeks. This fact gives the evidence for the absence of ovigerous females in the samples collected from the study area, Cochin backwaters.

In the present study, there is remarkable variation in stomach fullness and stomach contents in various size groups. It was observed that the maximum number of empty stomachs among the specimens analysed were between the size class of 2.5 - 3.0 carapace width. This size class may be considered as the optimum size of breeders, since majority of the crabs belonging to this size class were found in the period from April-July, which is supposed to be the breeding season of *V. litterata*. The number of empty stomachs were found to decrease in lower size classes. It is observed that the crabs of class 1.5- 2.0 and 2.0- 2.5 carapace width exhibited stomachs almost filled with food. Balasubramanian (1993) reported that the feeding intensity is comparatively low among adult crabs of *Charybdis smithii*. Jewett and Feder (1983) concluded that small crabs feed more intensively than larger crabs since moulting efficiency among smaller crabs is greater and

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energy demand is more. It is observed that the juvenile crabs (1.5-2.0 class carapace width) consumed more amount of sand and the debris, followed by plant remains. Crustaceans formed the most dominant food in the larger crabs. Difference in diet composition in different size groups could be related to the change in cheliped strength and foraging behavior (Sukumaran and Neelakantan, 1997). It is not possible to confirm from the stomach contents whether the prey had been alive or not when preyed. Caine (1974) had explained a prey catching mechanism in the portunid crab *Ovalipes guadulpensis*, but Hill (1976) could not observe such technique in *S. serrata. Gaetice depressus*, a grapsid crab has been described to be capable of suspension feeding (Depledge, 1989).

There was no significant difference in the quantity of the food consumed by males and females in the present study, as reported by Williams (1981), Jewett and Feder (1982), Sumpton and Smith (1990), Wieczorek and Hooper (1995) and Josileen (2011).

Crabs occupy different niches and inhabit many different habitats in a variety of geographical areas and this is reflected in the variety of food consumed by them. (Chande et al., 1999; Dahdough-Guebas et al., 1999; Kyomo, 1999; Bryceson and Massinga, 2002). The feeding strategies of the crabs are found to be diverse, and the type of food consumed is significantly dependent on the locality inhabited as reported by Hegele-Drywa and Normant (2009). Freire and Gonsalez-Gurriaran (1995) states that the diet variability of the crab is influenced by its habitat. Varuna litterata, a grapsid crab, is observed to be an inhabitant of regions dominated by mangrove patches and are found to be distributed in areas with a sandy substratum and higher organic carbon content. They are euryhaline in nature; however, prefer areas with close proximity to the sea and with good tidal influx. The gut content analysis of the crabs revealed that its habitat has a great influence on its food and feeding habits. Their diet included crustaceans, detritus, fishes, sand, debris and miscellaneous items. Crustaceans were the most dominant food item, followed by sand and debris, detritus of plant origin, fish remains and miscellaneous food items. The sand and debris found in its gut can be correlated with the sandy substratum they prefer to thrive. The guts also contained good amount of plant remains, which can be attributed to the mangrove leaf litter in their habitat. However, no molluscs were recorded from the dietary components of the species. From the present study, it is concluded that V. litterata is an opportunistic omnivore capable of ingesting both animal and plant tissues, with a preference to diets of animal origin.

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Chapter 5

Diversity of Brachyuran Crabs of Cochin Backwaters, Kerala, India

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INTRODUCTION

Among the decapods, crustaceans, the brachyuran crabs belonging to the infraorder Brachyura, are important in view of their rich diversity besides ecological and economic consequences. These crabs generally have a hard exoskeleton and are armed with a single pair of chelae. They typically have a short projecting tail (the reduced abdomen) which is entirely hidden under the thorax. They range in size from the pea crab of few millimetres wide to the Japanese spider crab, with a leg span of up to 4 metres (13 feet). They are adapted to various biotopes, such as marine, estuarine, fresh water, intertidal and even terrestrial habitats. Brachyurans are quite diverse, comprising about 700 genera and 5000 species worldwide. About 640 species of crabs are listed from Indian waters, among which 12 species are regarded as commercially important, inhabiting the coastal waters and adjoining brackish water environment. They occupy an important place in the crustacean fishery on account of their export potential and high nutritive value. The crab meat is not inferior to any sea food item, as it is rich in vitamins, glycogen, protein, fats and minerals. Crab meat is also considered to possess some medicinal value. It is believed to cure asthma and chronic fevers (Chopra, 1939; Chidambaram and Raman, 1944). Among crustaceans, crabs occupy the third position in terms of external market demand. Even though the volume of export is not comparable to that of shrimps, crabs support a sustenance fishery of appreciable importance (Rao et al., 1973). In the mangrove environment, crabs represent predominant taxa. They play a significant role in detritus formation, nutrient recycling and dynamics of the ecosystem. Their burrowing behavior enhances aeration and facilitates free circulation of

Marine Faunal Diversity in India. DOI: 10.1016/B978-0-12-801948-1.00005-7 Copyright © 2015 Elsevier Inc. All rights reserved. water (Ajmal Khan and Ravichandran, 2009). They are also found to have commensalism with algae, seaweeds, coelenterates, barnacles, bivalves and holothurians.

It is quite surprising that, despite so much of importance, this group is not given priority in investigations. In particular the diversity of brachyuran crabs in mangrove areas is poorly known. In view of this fact, the present study was undertaken to document the brachyuran crab diversity in the mangrove patches of the Cochin backwaters.

METHODS

Study Area

The study area, Cochin backwaters situated at the tip of northern Vembanad Lake, is a tropical positive estuarine system extending between 9° 40' and 10° 12' N and 76° 10' and 76° 30' E, with its northern boundary at Azheekode and southern boundary at Thannirmukham bund (Figure 5.1). Fifteen stations were selected in the study area on the basis of salinity. Crab specimens were collected from the above stations every month. They were collected from local markets, local fishermen, stake net operators, Chinese dip net operators and the indigenous 'Njandara' operators. Various methods were employed to collect the crabs from mangrove regions, including handpicking, scoop nets, knots made of co-conut leaflets, etc. Sampling was also done every month using a boat at selected stations of the study area to collect the bottom crawlers using a Van Veen grab.

The specimens collected were then brought alive to the laboratory and details regarding their color, carapace width, carapace length, and other morphological features like shape of the carapace, markings on the body, number of anterolateral teeth, features of frontal lobe, pubescence, spination on the chelae and other legs etc. were noted. The specimens were preserved in 10 percent formaldehyde for further identification. Identification was done up to species level following Chhapgar (1957), Crane (1975), Sakai (1976), Lucas (1980), Sethuramalingam and Ajmal Khan (1991), Keenan *et al.* (1998), Jayabaskaran, (1999); Ng *et al.* (2008), Ajmal Khan and Ravichandran, (2009).

RESULTS

A total of 24 species of brachyuran crabs belonging to 16 genera and 8 families was recorded from Cochin backwaters. Among the families, the highest number of species was recorded from the family Portunidae (9 species) followed by Grapsidae (7 species).

in

Systematics

Phylum: Arthropoda Superclass: Crustacea Pennant, 1777



FIGURE 5.1 Study area map.

Class: Malacostraca Latreille, 1806 Subclass: Eumalacostraca Calman, 1904 Order: Decapoda Latreille, 1803 Suborder: Pleocyemata Burken Road, 1963 Infraorder: Brachyura Latreille, 1802

Description of the Species

Scylla serrata (Forskal) Family: Portunidae Subfamily: Portuninae

It is an edible crab, commonly known as the mangrove or mud crab and found to be the largest known species from the near shore and brackish water habitats of India. Carapace is pale brown to greenish brown, hands red colored at the outer surface and the dactylus of chela greenish. Frontal margin is usually with sharp teeth. The anterolateral border is cut into nine narrow spines. The chelipeds are very strong and the carpus of the chelipeds has two spines on the distal half of the outer margin. The fifth pair of legs is paddle shaped and adapted for swimming. It occurs fairly abundantly throughout the year in the backwaters and the associated mangrove mud flats and has a great tolerance to fluctuations in salinity. It constitutes an important fishery stock and is caught in crab traps, gill nets, Chinese dip nets, and stake nets.

Scylla tranquebarica (Fabricius)

Family: Portunidae Subfamily: Portuninae

It is an edible crab and also known as mud crab. Carapace is greyish green in color. Frontal lobe is sharp and acuminate. Anterolateral border is cut into nine broad teeth. Carpus of the chelipeds has two obvious spines on the distal half of the outer margin. The frontal margin is usually with rounded teeth. The chelipeds and the limbs exhibit conspicuous polygonal patterns in both sexes. The fifth pair of legs is paddle shaped and adapted for swimming. Like *S. serrata*, this crab is also present abundantly throughout the year in the backwaters and the mangrove mud flats. It is adapted to a wide range of salinity and it contributes to the crab fishery of the region. It is caught using crab traps, gill nets, Chinese dip nets and stake nets.

Scylla sp.

Family: Portunidae Subfamily: Portuninae

It is comparatively small in size with morphological characters distinct from the other two *Scylla* species. Research works are in progress to determine whether it is a separate species or a subspecies. It usually occurs in the mangrove mud flats. It is caught in crab traps and Gill nets.

Portunus (Portunus) pelagicus (Linnaeus)

Family: Portunidae Subfamily: Portuninae It is an edible crab, commonly known as the blue swimmer crab. Carapace is blue colored in males and sand colored in females, with an extensive spread of irregular white spots. The distal segments of the legs are pinkish in color. The anterolateral border of the carapace has nine teeth of which the most posterior one is much larger and laterally prolonged. The last pair of legs is paddle shaped and adapted for swimming. It is a marine migrant species and found to occur in the backwaters during late post-monsoon and pre-monsoon period, when the salinity is comparatively high. It is caught in stake nets, gill nets and Chinese dip nets.

Portunus (Portunus) sanguinolentus (Herbst)

Family: Portunidae Subfamily: Portuninae

It is an edible crab, commonly known as blood-spotted swimmer crab. It is comparatively small in size. Carapace is sand colored and characterized by three large reddish round spots on the posterior half of the carapace, of which one is median and the other two lateral, each spot encircled by a white ring. The carapace is much broader than long. Its anterolateral margin is cut into nine spines, of which the most posterior is the longest. The fifth pair of legs is paddle shaped and adapted for swimming. Like *P. pelagicus*, it is also marine, and occurs in the back waters during post-monsoon and pre-monsoon period, when salinity is high. It is caught in Chinese dip nets, stake nets and gill nets.

Charybdis (Charybdis) feriata (Linnaeus)

Family: Portunidae Subfamily: Portuninae

It is an edible crab, commonly known as crucifix crab, and is fairly large in size. Carapace is brown in color with a purple tinge and conspicuous yellow markings, the central one resembling a cross; chelipeds and limbs are brownish with yellow and white spots and the tips brownish pink. Carapace is broad, slightly convex and smooth, length as much as its width. Anterolateral borders have six spines, the first truncated and notched anteriorly and the rest acuminate. The last pair of legs is paddle shaped and adapted for swimming. It inhabits offshore waters, but is found to occur in the backwater during the pre-monsoon period, when the salinity is relatively high. Though it is an edible species, it is not consumed in some parts of the country, while it contributes to the crab fishery of the Cochin region. It is caught in Chinese dip nets from the backwaters.

Charybdis (Charybdis) lucifera (Fabricius)

Family: Portunidae Subfamily: Portuninae Carapace is yellowish brown with two large white spots on either branchial region. Chelipeds are scarlet pink, the fingers brownish and the extreme tips whitish. The right cheliped is markedly larger than the left cheliped. The last pair of legs is paddle shaped, for swimming, and the posterior border of its propodite is found to be serrated. They are well distinguished with the presence of a sharp median lobule on the lower border of the orbit. There is no spine on the posterior margin of carpus of natatory leg. It is found to occur during the post-monsoon and pre-monsoon period and is caught in stake nets and Chinese dip nets from the backwaters.

Charybdis (Goniohellenus) hoplites (Wood Mason)

Family: Portunidae Subfamily: Portuninae

Carapace of the species is light brown in color. The most distinguishing character of this species is that the posterior border of the carapace forms an angular junction with the posterolateral borders and the last tooth of the anterolateral borders is a long spine, about twice as long as those in front of it, which is usually observed in the Genus *Portunus*. It is also found to possess a spine on the posterior border of the arms of the cheliped. The last pair of legs is paddle shaped and adapted for swimming. The specimen was caught in a Chinese dip net during the pre-monsoon season.

Thalamita crenata (Latreille)

Family: Portunidae Subfamily: Portuninae

Color of the carapace is brownish or greenish grey, claws pinkish, tips brown and the extreme tips white. The front is cut into six lobes, excluding the inner supra-orbital tooth. The anterolateral borders are cut into five equal teeth. The transverse ridge on the carapace appears faint. The outer surface of the propodus of the cheliped is smooth. The last pair of legs is paddle shaped and adapted for swimming. This estuarine crab inhabits mud flats, sandy beaches and mangroves. It occurs throughout the year and is caught in stake nets, Chinese dip nets and rarely in a Van Veen grab.

Uca (Celuca) lactea annulipes (H. Milne Edwards)

Family: Ocypodidae

Subfamily: Ocypodinae

It is commonly known as the 'Porcelain fiddler crab'. Color of the carapace is black with white or yellow stripes and the chelipeds pinkish or white. Carapace is subquadrilateral with moderately convergent lateral borders. Front is broad, supraorbital border oblique and the external orbital angle pointed obliquely outward. The most striking feature of this species is the enlarged pincers of the males, which it uses for courtship. The pincers or the major cheliped is smooth on the outer surface, while the lower border has a faint ridge, extending up to the base of the immovable lower finger. There is a wide gape between the fingers, and the movable upper finger is curved inward at the tip and extends past the immovable lower finger. It inhabits damp ground, found on the shores and near the mangroves. It is caught by handpicking and by using knots made of coconut leaflets from the study area.

Macrophthalmus depressus (Ruppell)

Family: Ocypodidae Subfamily: Macrophthalminae

Carapace is grey in color; broader than long; rectangular in shape; surface studded with minute pearly granules. The anterolateral borders are parallel and the anterolateral angles have a square cut lobe, rather than a tooth. Eyestalks are remarkably long and slender and almost reach the end of the orbital angles. The inner surface of the palm is smooth, and ornamented with thick hairs. Hair is also present on the ambulatory legs. It is found to be a mud dweller and occurs in mangrove regions. It is caught in stake nets. Handpicking is also used more often.

Nanosesarma (Beanium) batavicum (Moreira)

Family: Grapsidae Subfamily: Sesarminae

Color of the carapace is mottled grey, chelipeds cherry red. Front is broad and deflexed. Upper surface of the palm of cheliped has two oblique pectinated crests. The distinguishing feature of the species is the presence of three acute spines on the posterodistal border of pereopod four. It is found to occur in the mangrove regions and collected by hand picking and with knots made of coconut leaflets.

Nanosesarma (Beanium) andersonii (De Man)

Family: Grapsidae Subfamily: Sesarminae

Color of the carapace is dark brown, chelipeds reddish. Front is broad and deflexed. Upper surface of the palm of cheliped has numerous striae, one of which forms a pectinated crest. It is distinguished by the presence of four strong spines on the posterodistal border of merus of pereopod four. It is abundantly found in the mangrove regions. Handpicking and knots made of coconut leaflets are the methods employed to collect this species.

Neoepisesarma (Neoepisesarma) mederi (H. Milne Edwards)

Family: Grapsidae Subfamily: Sesarminae Color of the carapace is black or dark brown, Chelipeds reddish. Front is broad and deflexed. Carapace is quadrangular, with an anterolateral tooth, slightly convergent posteriorly. The surface of the carapace has tufts of hair on the anterior portion. The distinguishing feature of this species is that, above the transverse dactylar tubercles, a sulcus runs about one third of the total length of the tubercles and the vertical granular crest of the inner palm is salient. It is found in the muddy substratum of mangrove regions and is caught by handpicking and knots made of coconut leaflets.

Neoepisesarma (Selatium) brockii (De Haan) Family: Grapsidae Subfamily: Sesarminae

Color of the carapace is dark brown, chelipeds orange colored with white tips. The distinguishable feature of the species is that the pectinated crests on the upper palm reach the margin and the dactylar tubercles are well separated from one another without any transverse sulcus above. It is a semi-terrestrial form, found in mangrove mud, fissures of rocks, stone embankments, backwater shores, etc. It is very vigilant and intelligent and it trusts its speed and craft to escape its enemies, thus it is hard to pursue. It is captured by handpicking.

Metapograpsus messor (Forskal)

Family: Grapsidae Subfamily: Grapsinae

Color of the carapace is bottle green, chelipeds appears light violet. The legs are striped with alternate dark and light bands. Carapace is four-fifths as long as broad, and the lateral margins markedly converge backwards. Front is more than half the width of the carapace. The post-frontal region appears to have some transverse markings. The propodus of the chelae is inflated. Walking legs are short, and the dactylus of the legs are nearly as long as the propodus. The last segment of the abdomen appears triangular in males. It is a mud dweller and inhabits the mangroves, subtidal region, etc. It is caught by handpicking, knots made of coconut leaflets, etc.

Metapograpsus maculatus (H. Milne Edwards)

Family: Grapsidae Subfamily: Grapsinae

Carapace is black in color and chelipeds bright violet. Carapace is comparatively elongated, being seven-eighths as long as broad, with less convergent sides. The post-frontal region is devoid of any transverse markings or ridges. The fingers of the chelipeds are much longer than the upper border of the palm. Walking legs are larger, and the dactylus of the legs distinctly shorter than the propodus. The striking feature of this species is that the last segment of the male abdomen is trilobed. It inhabits the mangroves and the intertidal region. The specimens were caught by handpicking.

Varuna litterata (Fabricius)

Family: Grapsidae Subfamily: Varuninae

It is commonly known as the 'herring bow crab'. Color of the carapace is light brown to brownish grey. Carapace is squarish, with a smooth surface. Anterolateral borders are cut into three broad, but less sharp teeth. Dactylus, propodus and carpus of legs are laterally flattened like paddles and are fringed with long, densely packed hair. With its legs shaped as paddles used for swimming, it is sometimes called the 'Paddler crab'. It is an estuarine species and is found to occur in the mangrove regions and intertidal areas. It is caught in large numbers in stake nets and Chinese dip nets throughout the year.

Heteropanope indica (De Man)

Family: Xanthidae Subfamily: Pilumnidae

It is a small crab, with transversely oval shaped carapace, markedly oval and glabrous. Frontal lobe is straight and truncate. Anterolateral borders are cut into four teeth. The dorsal surface of the carapace has two parallel transverse ridges, which are beaded with fine granules. Chelipeds are extremely unequal, with the carpus and the propodus of the chelipeds being studded with pearly granules. The ambulatory legs are slender and sparingly haired. It is estuarine and found to inhabit the mangrove regions. The specimen was caught in a Van Veen grab.

Xenophthalmus pinnotheroides (White)

Family: Pinnotheridae Subfamily: Xenophthalminae

Color of the carapace is dirty white. Carapace is subtrapezoid in shape, slightly broader than long, with the anterolateral angle bluntly rounded. Frontal region is narrow and strongly deflexed. Orbits are situated longitudinally and parallel to each other. Chelipeds are found to be symmetrical, with palm longer than the fingers. Propodus of the first walking leg is as long as broad. Carpus and propodus of second leg are armed with a tuft of dense pubescence. The third and the last legs are longer and more slender and covered with short hair. The third leg is the longest of all. The crab is caught in large numbers from the backwater, where the substratum was found to be muddy, throughout the year. It was caught in a Van Veen grab as well as in stake nets.

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ON THE RECORD OF THE BROWN MUD CRAB SCYLLA OLIVACEA (HERBST, 1796) FROM COCHIN BACKWATERS, KERALA, SOUTH WEST COAST OF INDIA.

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ABSTRACT

The present work is on the record of brown mud crab, Scylla olivacea from Cochin Backwaters, Kerala, South India. Solivacea contributes a major share to the crab fishery along the Cochin region and they are of great demand in the domestic as well as the foreign markets due to their large size and taste. Identification of the species was done on the basis of morphological characters as well as the description of the first and second male pleopods and the third maxillipeds, which is then confirmed using the molecular technique, sequencing of cytochrome c oxidase sub unit 1. The gene sequence then obtained after the sequencing of cytochrome c oxidase subunit 1 (CO1) gene was submitted in the GenBank (accession number - KF612463). The present study is the first time report on the occurrence of S. olivacea from Cochin Backwaters.

Keywords: Mud crab, Scylla olivacea, taxonomy, new record

INTRODUCTION

Mud crabs belonging to the genus *Scylla* inhabit brackish waters, such as estuaries and mangrove areas. They are widely distributed throughout the Pacific and Indian Ocean, from Tahiti, Australia and Japan to Southern Africa^{1,2,3}. In India, mud crabs are reported from the estuaries of the rivers of Ganga, Mahanadi, Krishna and Cauvery and the brackish water lakes viz., Chilka and Pulicut on the east coast, the estuaries of Narmada and Tapti and the brackish waters of Kerala on west coast. They are also found to inhabit the mangrove regions of Andamans and Nicobar Islands, Andhra Pradesh, Tamil Nadu and Kerala⁴. Due to their large size and taste, mud crabs are of great demand in the domestic as well as the foreign markets. India earns about US\$ 18 million as foreign exchange from the export of live mud crabs⁵.

Knowledge on taxonomy is important for the proper identification of the species and placing them into the right systematic classification. Proper

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identification and classification of species, especially those species which holds economic importance, is cardinal for the management of wild stock and the development of a more successful aquaculture industry. This also enables the adoption of proper conservation measures in the case of threatened species, thereby improve the population size. Prior to the classification of Keenan et al.⁶, there was a considerable confusion regarding the taxonomy of mud crabs, belonging to the genus Scylla, world wide. In India, studies on mud crabs have been dealt with by Kathirvel⁷, Radhakrishnan and Samuel⁸, Joel and Sanjeevraj⁹ and Kathirvel and Srinivasagam¹⁰. Kathirvel⁷ recorded two species Scylla from Cochin Backwaters on the basis of morphological characteristics, larger species being Scylla oceanica and smaller one being Scylla serrata. Later in 1982. Radhakrishnan and Samuel⁸ reported the occurrence of a subspecies Scylla serrata serrata from Cochin Backwaters on the basis of morphological features. Joel and Sanjeevaraj⁹ studied the taxonomy of Scylla from Pulicut lake and reported the occurrence of two species, namely Scylla tranquebarica and S. serrata. Taxonomy of mud crabs from India were then critically analysed by Kathirvel and Srinivasagam¹⁰ and stated that two species are found in the Indian waters, S. serrata and S. tranquebarica, which are characterized by the differences in size, spines on the outer border of the carpus of the cheliped and habitat preferences. In contrast to their findings, Shaji et al.¹¹ claimed that S. serrata and S. olivacae are the most common species occurring in India, on the basis of taxonomic identification of Keenan et al.6. The occurrence of S. olivacea in the Gujarat waters was reported by Trivedi and Vachharajani¹², based on the morphological features as well as the morphometric analysis. Padate et al.13 claimed the new record of S. olivacea from Goa coast along with the comparative diagnosis with S. serrata from the region. Devi et al. 14 reported three species of Scylla from Cochin Backwaters, viz., S. serrata, S. tranquebarica and Scylla sp based on the morphological features. The present study confirms the third species to be S. olivacea on the basis of the morphological features proposed by Keenan et al.6, the morphology of the first and second pleopods and the third maxillipeds as well as the molecular analyis. The present work is the first time report on the existence of S. olivacea, a commercially important mud crab species from Cochin Backwaters, Kerala, South west coast of India.

S. olivacea, commonly known as the brown mud erab, is distributed throughout the Indo-west Pacific. They are associated with mangrove forests and coastlines inundated with low salinity. These estuarine species are euryhaline in nature, however they are found to prefer a low salinity. These are procured in considerable amount where oligohaline condition is prevailing and these are also found to exist where salinity is almost zero. Being commercially important, these are of great demand in the domestic as well as the foreign markets due to their large size and taste. Along with *S. serrata* and *S. tranquebarica*, *S. olivacea* is also being exported to foreign countries especially the south east Asian countries on a large scale.

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MATERIALS AND METHODS

As a part of the diversity study conducted on Cochin Backwaters, a positive tropical estuarine system, located at the south west region of the Indian sub continent, 15 sampling stations were fixed along the study area on the basis of salinity, habitat type, presence of mangroves and the availability of crabs; viz., Thanneermukkom, Vaikom, Aroor, Valanthakad, Kumbalanghi, Kandakkadavu, Thevara, Thoppumpady, Marine Science Jetty, Varappuzha, Vallarpadam, Vypin, Munambam and Azheekode. Sampling of crabs was conducted monthly for a period of two years from June 2010 to May 2012. Live mud crabs with a size range of 80-128mm carapace width, procured from local market, local fishermenoperating the indigenous crab traps known as 'Njandara', Chinese dip net operators and other collectors were brought to the laboratory. The morphological characters of the crabs such as the colour and shape of the carapace, shape of the frontal lobes, spination of the anterolateral borders and the chelae and the presence of hexagonal markings were noted. The crabs were then identified using the identification key provided by Keenan et al.6, which has been widely accepted by crab taxonomists as well as by FAO15.

The specimens were numbed by freezing and preserved in 10% formaldehyde¹⁶. The first and second ploeopds from the male specimens with a size range of 95-128 mm carapace width (110.94 \pm 10.2) and the third maxillipeds were cut carefully using a forceps and scissors and examined under a binocular microscope. The pleopods and the maxillipeds were then drawn using a camera lucida.

The species identification was then confirmed using the sequence data obtained from the mitochondrial cytochrome c oxidase subunit 1 (CO1) gene. Muscle tissue from the thoracic appendages was dissected out from live samples of *Scylla*, for DNA extraction. DNA extraction was carried out following standard phenol:chloroform extraction and ethanol precipitation technique, followed by PCR amplification to obtain the sequences of the partial mitochondrial CO1 gene. Amplification was done using the universal primers as well as the species specific primers. The purified PCR products were then sequenced with CO1 primers using ABI Prism sequencing kit (BigDye Terminator cycle) and the homologue searching of the nucleotide sequence (using blast suite) was performed with the Basic Local Alignment Search Tool (BLAST) through NCBI server¹⁷.

RESULTS

The study reveals the occurrence of the brown mud crab *S. olivacea* from the study area Cochin backwaters, Kerala, aside from *S. serrata* and *S. tranquebarica*, on the basis of morphological features proposed by Keenan *et al.*⁶ and the molecular analysis. The gene sequence obtained after the sequencing of cytochrome oxidase-1 gene was submitted to the GenBank database (accession number- KF612463).

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A. Distinguishing characters

Carapace of the crab is smooth. The carapace is grayish green, propodus of the chelae and the last segments of the walking legs are pale orange. The frontal margin is cut into rounded lobes. Anterolateral margin of the carapace cut into 9 broad teeth. Chelipeds are large and the propodus are inflated. The species is characterized by the presence of single spine on the distal half of outer margin of the carpus of the cheliped. Palm of the cheliped exhibits a pair of prominences on the dorsal margin behind the insertion of the dactyl. The inner prominence is larger than the outer one. Hexagonal markings are weak or absent.

The first male pleopod of *S.olivacea* is elegantly bent, with a broad base and then tapering towards its apex. It exhibits small spinules like structures on its outer margin, while the inner margin of the pleopod is characterised with a single patch of setae, which is thick and prominent. The second male pleopod is slender throughout its length and broad at the base. The tip of the pleopod is bilobed and the pleopod is found to possess setation on the inner margin on its posterior region. The third maxilliped of *S. olivacea* showed the characteristic broad and flattened ishium. The maxillipeds are also found to possess thick hairs on the inner margin.

B. Molecular Analysis

Partial CO1 readable sequence of 597 bp was obtained and the BLAST analysis of the nucleotide sequence showed 98% similarity to *S. olivacea* (GenBank ID KC154082). The nucleotide sequences obtained were submitted to GenBank database (accession number - KF612463).

DISCUSSION

The species identification of mud crabs belonging to genus Scylla has been controversial, for many years¹⁸. Attempts to identify Scylla spp. have led to much confusion because of the subtle morphological differences between the species. The confusion in identification has led to the proliferation of available names, and this in turn has led to the inconsistent use of name between authors.⁶

The findings of the earlier workers who dealt with the mud crabs of Cochin backwaters were contradictory to each other, leaving the classification of mud crabs even more confusing. Kathirvel⁷ came across one larger and one smaller species from the Cochin backwaters and identified the larger one as *S. oceanica* and the smaller one as *S. serrata*. Subsequently, Radhakrishnan and Samuel⁸ described the occurrence of a sub species, which was observed to possess a convex carapace, with blunt frontal teeth and a single spine on the outer margin of the carpus and this subspecies was designated as *S. serrata*. Joel and Sanjeevaraj⁹ reported the existence of two species of genus *Scylla*, viz., *S. serrata* and *S. tranquebarica* from Pulicut lake. However, their findings were critically analysed by Kathirvel and Srinivasagam¹⁰ and stated that *S. serrata serrata* could be *S. serrata* only. They also opined that only two species of *Scylla* do exist in the

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Indian waters, namely *S. serrata* and *S. tranquebarica*. Later on, Devi *et al.*¹⁴ could record a third species of *Scylla*, in addition to *S. serrata* and *S. tranquebarica* from Cochin backwaters, based on the morphological variations proposed by Keenan *et al.*⁶

In the present study the morphology of crabs were analysed following the identification keys provided by the carcinologists viz., Stephenson and Campbell¹⁹, Joel and Sanjeevaraj²⁰, Sethuramalingam and Ajmal Khan²¹, Fuseya¹⁸, Keenan *et al.*⁶ . The identification key proposed by Keenan *et al.*⁶ is considered to be feasible and generally accepted by crab taxonomists as well as by FAO¹⁵, since he has given more concrete evidences on the basis of morphological, morphometric and molecular analysis. When the observations of the present study were compared with the findings of Keenan *et al.*⁶, it was confirmed that the third species, which is found to exist in Cochin Backwaters aside from *S. serrata* and *S. tranquebarica* is *S. olivacea*.

The genetic variability analysis carried out by Fuseya and Watanabe²² employing horizontal starch gel electrophoresis confirmed the existence of three species of mud crabs namely, S. serrata, S. tranquebarica and S. oceanica in Japan. Later, Keenan et al.⁶ made a revision on genus Scylla using specimens collected from the Red Sea and throughout the Indo-Pacific region. In their study, two independent genetic methods, allozyme electrophoresis and sequencing of two mitochondrial DNA genes viz., cytochrome oxidase 1 and 16s RNA was employed in an attempt to differentiate the species. Aside from the molecular analysis, they had also made use of external morphology, morphology of first male pleopods as well as morphometry to distinguish the species and they could recognize four species of Scylla namely, S. serrata, S. tranquebarica, S. olivacea and S. paramamosain. In the present study, the species specific primers developed by Keenan et al. has been employed for the amplification of Cytochrome oxidase I gene and the gene sequences obtained showed 98% similarity with that of S. olivacea (GenBank ID KC154082) when the sequences were underwent BLAST analysis, confirming the species to be S. olivacea.

Though, *S. serrata* and *S. tranquebarica* are reported from the Cochin backwaters from the earlier studies⁷, the occurrence of *S. olivacea* was not yet reported. The present study records the occurrence of *S. olivacea* in Cochin backwaters, Kerala, South India as for the first time on the basis of valid classical as well as molecular taxonomic evidences.

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