DETECTION OF MICROPLASTIC FROM THREE IMPORTANT CRAB SPECIES FROM THE COAST OF BANGLADESH

A THESIS

BY

MD. MEHEDI HASAN

Registration No. 1706101

MS Session: 2022

Thesis Semester: January - June 2024

MASTER OF SCIENCE (M.S.)

IN

AQUACULTURE

DEPARTMENT OF AQUACULTURE

HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY, DINAJPUR

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Dedicated

To

My Beloved Parents and

Honourable Teachers

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At the very beginning of my thesis, I would like to memorize Almighty Allah who always kept his hand over my head in any problematic situation. Without his blessing, I couldn't perform my given work successfully. So, all praise and gratitude of this work is dedicated to the supreme ruler of the universe.

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ABSTRACT

In this study, the abundances and characteristics of microplastics in different tissues of three species of wild crabs (*Scylla olivacea*, *Portunus pelagicus*, and *Portonus sanguinolentus*) were investigated across three important fishing grounds (Chattagram, Cox's Bazar and Saint Martin Island region) of Bangladesh. Each region consists of three distinct fishing grounds. The result of the study showed that the highest detection of microplastics was recorded in Cox's Bazar followed by Saint Martin and Chattagram. The crab species *Scylla olivacea* and *Portunus pelagicus* were most abundant in Cox's Bazar and Chattagram while *Portonus sanguinolentus* in Cox's Bazar and Saint Martin Island. The microplastics size was also varied among the crab samples. Comparatively larger microplastics were recorded in the crab samples in Chattagram region. Black coloured microplastics were the most prominent in gill and guts of the samples (45.45% and 42.37%, respectively). Among various types of shapes about (56.92%) of thread was most abundance in gill where thread was (53.33%) in gut. The 50% gill tissue showed fragment microplastics followed by microbead (6.25%). The 49.18% gut tissue showed microbead microplastics followed by fibre (8.20%). Overall, the study reported that the distribution pattern, abundance, size, color and shape of microplastics in the studied crab samples which will ultimately help the biologist to get insight into the plastic pollution in marine ecosystems by taking crabs as model species.

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LIST OF ABBREVIATIONS

ANOVA	: Analysis of variance
BBE	: Bahia Blanca Estuary
BFRI	: Bangladesh Fisheries Research Institute
DMRT	: Duncan's Multiple Range Test
et al.	: et alia (and others)
etc.	: et cetera
g	: Gram
HSTU	: Hajee Mohammad Danesh Science and Technology University
kg	: Kilogram
km	: Kilometer
mg	: Milligram
MS	: Master of Science
NST	: National Science and Technology
PAHs	: Polycyclic Aromatic Hydrocarbons
POPs	: Persistent Organic Pollutants
sp.	: Species
SPSS	: Statistical Package for Social Sciences
µm	: Micro Meter

CHAPTER I

INTRODUCTION

Crabs are an indispensable component of the benthic ecosystem and represent a type of seafood that is easily obtained and frequently eaten by humans. However, little is known about microplastic accumulation in different tissues of crabs important fishing areas. The coastal zone is defined as the area along the seashore where the living and non-living elements of marine and terrestrial environments interact to create intricate ecological and economic resource systems (Protocol on Integrated Coastal Zone Management in the Mediterranean, 2012). The area is well-suited for human activities, including housing complexes, commercial industries, and human recreation (Nordstrom, 2000). As a result, it is subject to numerous disturbances, such as pollution (Brown and McLachlan, 2002).

Plastic is widely utilised in many items and is a significant contributor to consumer goods packaging due to its affordability, adaptability, lightweight nature, durability, and ease of production (Thomson et al. 2004; Plastics Europe, 2010; Jambeck et al. 2015); Andrady, 2015. Plastic is highly accessible to manufacture and convenient to utilise, which explains its widespread adoption. As a result, the amount of plastic garbage discarded by humans into the environment has grown rapidly, having a detrimental impact on coastal and marine ecosystems. Consequently, plastic accounts for around 89% of coastal and marine waste, with an estimated yearly global average of 45,000 pieces of plastic per square mile (Central database system and data standard for marine and coastal resources, 2013). Plastic trash in the coastal environment is a result of activities in the tourism, agriculture, aquaculture, fisheries, and industrial sectors (Nagelkerken et al. 2001; Fujieda and Sasaki 2005; Oigman-Pszczol and Creed 2007).

Microplastics are plastics that have a particle size smaller than 5 mm, as described by Thomson et al. (2004) and Moore (2008). Microplastics have a greater tendency to disseminate in the environment than larger plastic particles, mainly because of their smaller size. Microplastics are widely present in several environmental media, including seas, sea water, sea ice, and sea sediments (Derraik, 2002; Lusher et al. 2013; Zhu et al. 2018; Feng et al. 2020). Furthermore, numerous marine creatures, including fish (Foekema et al. 2013; Feng et al. 2019; Su et al. 2019), marine worms (Besseling et al. 2012), bivalves (Von Moos et al. 2012; Li et al. 2018), and sea cucumbers (Mohsen et al. 2019), consume microplastics due to their environmental attributes. Microplastics originate from the breakdown of larger plastic materials and can result from several processes, such as wave action, abrasion by sand or sediment, photodegradation due to sunlight exposure, biodegradation induced by living organisms, and other similar mechanisms (Boucher, 2017). The many forms of microplastics include fragments, fibres, pellets, and films (Sari et al. 2015). Due to their small size, currents are more likely to transport microplastics and lodge them in aquatic organisms like crustaceans (Datu et al. 2019). Research has demonstrated that microplastics possess the capacity to assimilate noxious hydrophobic substances from their surroundings (Cole et al. 2011). Moreover, microplastics have the ability to build up in marine species, leading to a range of negative consequences. These include physical harm, such as blockage and abrasion in the intestines, as well as oxidative damage, inflammation, liver strain, and reduced growth. (Von Moons et al. 2012; Rochman et al. 2013). Because of their strong hydrophobic properties, microplastics have the ability to absorb persistent organic pollutants (POPs) and serve as vehicles for the spread of pollutants (Rochman et al. 2013).

Marine environments are home to crabs. The crab is a member of the Portunidae family under the crustacea class, which also includes lobster and shrimp. Bangladesh's coastline is abundant with crabs, especially in estuaries, Sundarbans mangrove swamp, and coastal ghers

(Khan and Alam, 1992). People frequently refer to the crab as the "Green Crab" or "Mangrove Crab," and locally, they call it "Habba Kankra," "Silla Kankra," or "Kankra" (Shafi and Quddas, 1982). They have a wide distribution and are capable of consuming both plant and animal matter. Researchers frequently use crabs as indicators of pollution levels due to their high sensitivity to environmental changes (Aheto et al. 2011; Jonah et al. 2015). Furthermore, crabs, being one of the prominent species, have a significant impact on the preservation and equilibrium of the benthic environment (Botto and Iribarne, 1999; Eça et al. 2013). Furthermore, individuals extensively consume crabs, which provide significant nutritional value and serve as a crucial protein source. Fishing grounds are the primary source of these wild crabs.

Crab has become a promising source of export revenue due to its high global demand and the expansion of aquaculture in Bangladesh's coastal region. Dhaka transports crabs from the southern region, freezes them, and then sends them to either direct export markets or major export destinations. During the peak season, traders assess the quality of the catch and sell it at elevated prices, reaching up to $45 per kilogram. Traders export crabs in two distinct states: either alive or frozen. Asian countries typically import live crabs because of their strong demand and transportation limitations. Western countries, including Australia and the United Kingdom, have a high demand for frozen soft-shell crabs. In the fiscal years 2018–2019, crab exports generated a total revenue of $42.8 million. The sale of frozen crab generated $33 million, while the sale of live crab generated $9.8 million (BER 2019). Among the crab samples the three studied samples brief description are as follows -

Scylla olivacea, commonly known as the orange mud crab, is a commercially important species of mangrove crab in the genus *Scylla*. It is one of several crabs known as the mud crab and is found in mangrove areas from Southeast Asia to Pakistan, and from Japan to northern Australia. Along with other species in the genus *Scylla*, it is widely farmed in aquaculture using wild-caught stocks. They can be differentiated from other species of *Scylla* by having blunted spines on the dorsal distal corner of the palm (propodus) of the claw, and by the rounded frontal lobe spines with shallow separations in between the eyes.

Figure 1.1 *Scylla olivacea*

Scientific Classification

Kingdom: Animalia

Phylum: Arthropoda

Class: Malacostraca

Order: Decapoda

Family: Portunidae

Genus: *Scylla*

Species: *Scylla olivacea*

Portunus pelagicus, also known as the flower crab, blue crab, blue swimmer crab, blue manna crab or sand crab is a species of large crab found in the Indo-Pacific, including off the coasts of Indonesia, Malaysia, Cambodia, Thailand, the Philippines, and Vietnam; and in the intertidal estuaries around most of Australia and east to New Caledonia.

Figure 1.2 *Portunus pelagicus*

Scientific Classification

Kingdom: Animalia

Phylum: Arthropoda

Class: Malacostraca

Order: Decapoda

Family: Portunidae

Genus: Portunus

Species: *Portunus pelagicus*

Portunus sanguinolentus, the three-spot swimming crab , blood-spotted swimming crab or red-spotted swimming crab, is a large crab found throughout estuaries of the Indian and West Pacific Oceanic countries.

Figure 1.3 *Portunus sanguinolentus*

Scientific Classification

Kingdom: Animalia

Phylum: Arthropoda

Class: Malacostraca

Order: Decapoda

Family: Portunidae

Genus: Portunus

Species: *Portunus sanguinolentus*

Crabs, as integral members of marine ecosystems, are particularly susceptible to the impacts of microplastic pollution. With their diverse habitats ranging from intertidal zones to deep-sea environments, crabs bioaccumulate microplastics through various pathways, including ingestion, adhesion to their exoskeletons, and absorption through their gills. Consequently, the accumulation of microplastics in crab tissues can have profound physiological, biochemical, and ecological implications. This research will make a significant contribution to the preservation of marine ecosystems and the assurance of seafood safety. However, there has been no investigation on the contamination of wild crabs in primary fishing areas of microplastics, and there has been limited research on the deposition of microplastics in various crab tissues. The goal of this study is to find out how common and what kinds of microplastics are in the digestive system (gut) and respiratory organs (gills) of three species of crabs: *Scylla olivacea, Portunus pelagicus*, and *P. sanguinolentus*, at three important fishing spots. They inhabit a wide range of coastal habitats and play integral roles in marine food webs. However, their exposure to microplastic pollution can induce a variety of physiological responses with potentially deleterious effects on individual health and population dynamics. Studies have documented the uptake and accumulation of microplastics in crab tissues, including the hepatopancreas, gills, and digestive tract, leading to impaired feeding, energy allocation, and reproductive fitness. The mechanisms underlying the toxicity of microplastics in marine crabs are multifaceted and encompass physical, chemical, and biological pathways. Physical interactions between microplastics and crab tissues can cause mechanical damage, obstruction of digestive tracts, and interference with respiratory surfaces. Moreover, the leaching of chemical additives and adsorption of persistent organic pollutants (POPs) onto microplastic surfaces can exacerbate toxicological effects through bioaccumulation and biomagnification within crab trophic networks. The impact of microplastic pollution on marine crab tissue extends beyond individual organisms to encompass broader ecological dynamics within marine ecosystems. Crabs serve as important prey for various predators, including fish, birds, and mammals, thus facilitating the transfer of microplastics across trophic levels. Furthermore, the disruption of crab physiology and behaviour by microplastic exposure may alter ecosystem functions such as nutrient cycling, benthic community structure, and fisheries productivity.

This study aimed to ascertain the attributes of microplastics contamination and the distribution of microplastics in several crab species. Furthermore, this study provides evidence to support future assessments of the ecological hazards posed by microplastics in marine ecosystems and their impact on seafood health in significant fishing regions.

Objectives of the study

- To identify the size, color, shape of microplastics on three crab species in different costal region of Bangladesh; and
- To explore the distribution of micro plastics on three crab species in different coastal regions of Bangladesh.

CHAPTER II

REVIEW OF LITERATURE

Takarina et al. (2023) highlighted that microplastics are a type of contaminant that can be generated by the breakdown of plastic debris near coastal areas. These microplastics have the potential to pollute water, sediment, and organisms. The minuscule dimensions of microplastics facilitate their transportation by currents and subsequent entrapment inside the biota. Therefore, they conducted a study to examine the prevalence of microplastics in the water, soil, edible crabs, and gastropods found in the Blanakan coastal areas. They employed the random purposive sampling approach to collect water, sediment, and biota samples. The sampling was conducted at three stations, with three repetitions at each station. The overall concentration of microplastics varied from 320 to 380 particles per liter in the water, 280 to 2200 particles per kilogram in the soil, 3.5 to 25 particles per individual in crabs, and 5.5 to 8 particles per individual in gastropods, as documented by the researchers. The microplastics detected in the water, soil, and biota on the Blanakan coast encompassed shards, films, fibres, and pellets. The pellets were frequently observed in the sediment samples, while the fibres were detected in the water samples, as well as in crabs and gastropods.

Angelica et al. (2022) made the initial observation that microplastics were found in the gills and digestive system of the mangrove crab U. occidentalis obtained from local markets in Tumbes, Peru. They detected microplastics in all of the crabs that were examined, with a total of 921 pieces. Of these, 475 pieces (52.57%) were located in the gills, while 446 pieces (48.43%) were found in the digestive tract. The size range was defined as 2 to 250 µm, 250 to 500 µm, 500 to 1 mm, and 1 to 5 mm. Microplastics measuring between 2 and 250 µm were the most prevalent, accounting for 53.79% in the digestive system and 90% in the gills. There were six distinct types of microplastic identified, with fibres (59.64%–61.05%) and films

(19.28%−36.63%) being the most common in both tissues. Clear fibres were the predominant type of microplastic found in both tissues. Microplastics measuring less than 250 µm were discovered in the gills of 90% of the samples and in the digestive system of crabs at a rate of 53.79%.

Siddiquee (2022) stated that microplastics pollution is now a serious environmental threat for both freshwater and marine ecosystems. The presence of microplastics (1 µm to 5 mm) in the gastrointestinal tracts of the mud crab (*Scylla serrata*) in Sundarbans coastal regions was reported. The author examined the microplastics pollution for two seasons such as monsoon and winter. He found the number of ingested microplastics was 154 in *S. serrata* and the number of ingested microplastics per *S. serrata* (n = 20 per season) were 4.7 ± 1.39 during monsoon and 3 ± 0.98 during winter, which did not differ significantly between the two seasons. Fibres were the most abundant types of microplastics in 84% in *S. serrata*. Since fibres were mostly filament-shaped, filament was the dominant shapes of microplastics in the mud crab. The findings indicated ubiquitous distribution of microplastics in the waters of Sundarbans aquatic biota, as well as their presence in water column and sediments where crab species dwell. These findings raised concerns that microplastics contaminations in the Sundarbans waters of Bangladesh and their ingestions by fish and crustacean species were posing threat to aquatic food chain and public health.

According to Horn (2021), microplastics are widely present in marine ecosystems, but our understanding of their impact on marine animals is poor. Plastic waste carried by the ocean gathers in coastal habitats around the world. Invertebrate organisms living in these environments can consume tiny plastic particles, mistakenly thinking they are food. Research has indicated that microfibres are the most common kind of microplastic found in sandy beach habitats. To investigate this, the author analyzed the impact of microplastic fibres on the physiological and reproductive results of a nearshore organism. This was done by exposing Pacific mole crabs (*E. analoga*) to micro-sized polypropylene rope fibres at quantities that are often found in the environment. The researcher conducted a comparison between mature gravid female crabs exposed to microfibres and a control group. The comparison focused on death rates, reproductive success, and the rate of embryonic development. As a result, Pacific mole crabs that were exposed to polypropylene rope saw higher death rates among adult crabs and lower ability to retain egg clutches, leading to variations in the rates of embryonic development. The author reported that the impacts of consuming microplastics by a prey species in nearshore areas have consequences for predators in those areas, such as surf perf and shore birds. This is particularly relevant as the usage of plastic and the subsequent abundance of microplastics in nearshore ecosystems continue to rise.

Dorothy et al. (2020) investigated the impact of microplastic fibres on the physiological and reproductive results of Pacific mole crabs (*E. analoga*) in a nearshore environment. The crabs were exposed to microsized polypropylene rope fibres at concentrations that are often found in the environment. The researchers conducted a comparison between adult gravid female crabs that were exposed to microfibres and those that were not exposed (control group). The author examined the death rates, reproductive success, and rates of embryonic development in these crabs. Exposing Pacific mole crabs to polypropylene rope resulted in higher mortality rates among adult crabs and reduced their ability to retain egg clutches, leading to variations in the rates of embryonic development. The ingestion of microplastics by a prey species in nearshore areas has consequences for predators as surf perf and shore birds. This is particularly relevant as the usage of plastic and the subsequent prevalence of microplastics in nearshore ecosystems continues to rise. Microplastics are widely present in marine and sandy beach settings, and they have a substantial influence on Pacific mole crabs.

Gerrit et al. (2020) observed the effect of that exposure to microplastics resulted in a notable decline in the duration of startle response at the population level, along with a decrease in the variation within individuals. To put it simply, crabs exhibited reduced aversion to risk on average and their behaviour became more predictable as the concentrations of microplastics increased. Overall, their research suggests that microplastic contamination could heighten the vulnerability of hermit crabs to being preyed upon.

Research by Gündoğdu et al. (2020) found that microplastic ingestion by green crabs (*Carcinus aestuarii*) resulted in the accumulation of polycyclic aromatic hydrocarbons (PAHs) in crab tissues, potentially posing risks to higher trophic levels. Furthermore, microplastic ingestion may alter crab-mediated ecosystem processes, such as sediment bioturbation and nutrient cycling, with broader implications for coastal ecosystem functioning.

Villagran et al. (2020) has shown that microplastics are accessible to a diverse array of marine creatures, with filter-feeding bivalves and crabs being particularly susceptible. Neohelice granulata plays a crucial role in shaping the ecosystem of the Bahía Blanca Estuary (BBE) in the South-western Atlantic. Due to its ecological significance, this species is particularly susceptible to many pollutants. Various forms of microplastics were discovered in both the crabs and water column samples. However, the most commonly seen microplastics were fibres measuring between 500 and 1500 micrometers, predominantly blue in color. This study is the first to discover microplastics in both the gills and digestive tract of *N. granulata*. Furthermore, the gills exhibited greater overall quantities of microplastics compared to the digestive system. This indicates that in this scenario, the primary method of microplastic uptake would be through adherence to the gills.

According to Waddell et al. (2020), Blue crabs (*Callinectes sapidus*) are vulnerable to this contamination because they consume sediment where there is a high concentration of plastics. The researchers evaluated the consumption of microplastics by blue crabs in Corpus Christi Bay, Texas. The researchers collected and digested the stomachs of crabs using a tissue destruction process that relies on hydrogen peroxide. They then confirmed the composition of the collected material using micro attenuated total reflectance Fourier transform infrared spectroscopy (μ-FTIR). Out of the 39 blue crabs that were examined, it was discovered that 28 of them had completely artificial shards and fibres in their stomachs, while 24 of them had partially artificial fibres. After accounting for potential contamination, 36% of the blue crabs that were gathered were found to have totally synthetic fragments and fibres, as well as semisynthetic fibres, with an estimated average of 0.87 items per crab.

Forbes and Rosch (2019) conducted a study to investigate the presence of microplastics in fiddler crabs (genus Uca) taken from the marsh at Waties Island, SC. The crabs dissected their faecal samples and analyzed them under a microscope to determine the quantity and composition of microplastics present. Microplastics were detected in nearly every sample collected, suggesting that microplastics are abundant even in this seemingly unspoiled environment. The consumption of microplastics by fiddler crabs at Watie's Island, SC will have significant consequences for their survival and reproductive success. These effects will also extend to other organisms through predation and other ecological processes, given that fiddler crabs play a crucial role as a low trophic organism in the ecosystem.

According to Horn et al. (2019), microplastics are frequently found in marine environments, and their presence in coastal sediments puts invertebrate in fauna at risk of ingesting plastic trash and the hazardous substances that come with it. The researchers evaluated the occurrence of microplastics in the sands of sandy beaches along the California coast, covering a distance of over 900 km. They then examined whether Pacific mole crabs (*E.*

analoga) had consumed these microplastics. On average, 35% of Pacific mole crabs were found to have microplastics in their digestive systems.

Waite et al. (2018) did a study to quantify and assess the variety of microplastics present in the water and soft tissues of eastern oysters (*Crassostrea virginica*) and Atlantic mud crabs (*Panopeus herbstii*) in Mosquito Lagoon. Mosquito Lagoon is a shallow estuary with little tidal fluctuations located along the central Florida east coast. They received an item. The average number of microplastic fragments per individual in a sample of 90 adult oysters was 16.5. The majority of our collections were comprised of fibres, primarily in a royal or dark blue hue. Crabs had 100 times more microplastic fragments than oysters when measured per gram of tissue.

The ingestion of microplastics by coastal crabs can disrupt feeding behaviour, nutrient uptake, and reproductive success, thereby influencing population dynamics and community structure within coastal ecosystems. Additionally, microplastic accumulation in crab tissues can be bioaccumulated up the food chain, posing risks to predators and human consumers (Galloway et al. 2017).

Studies have demonstrated various physiological impacts of microplastic exposure on coastal crabs. For instance, research by Sussarellu et al. (2016) showed that exposure to microplastics led to altered energy metabolism and oxidative stress in shore crabs (*Carcinus maenas*). Similar findings indicated that ingestion of microplastics by green crabs (*Carcinus maenas*) resulted in histopathological alterations in the hepatopancreas and intestine, indicative of potential tissue damage. These physiological disruptions can compromise the health and fitness of coastal crab populations, with potential cascading effects on ecosystem dynamics.

The ecological consequences of microplastic pollution on coastal crab populations extend beyond individual physiological and behavioural effects. Microplastic ingestion by crabs can contribute to biomagnification and the transfer of contaminants through marine food webs.

Exposure to microplastics can alter the behaviour of coastal crabs, affecting their foraging patterns, locomotion, and predator avoidance strategies. Studies have documented decreased activity levels and impaired sensory perception in crabs exposed to elevated concentrations of microplastics, compromising their ability to thrive in their natural habitats (Sussarellu et al. 2016).

According to Watts et al. (2015), microscopic plastic fragments measuring less than 5 mm are a global conservation concern, since they are degrading coastal and marine habitats. Fibres are the predominant plastic type found in the digestive systems of marine species. The researchers examined the destiny of polypropylene rope microfibres (measuring 1-5 mm in length) that were consumed by the crab *Carcinus maenas*, and assessed the impact on the crab's energy allocation. During extended feeding studies lasting 4 weeks, crabs that consumed food containing microfibres (0.3-1.0% plastic by weight) exhibited decreased food intake (from 0.33 to 0.03 g d(-1)) and a substantial decline in the amount of energy available for growth (scope for growth) from 0.59 to -0.31 kJ crab d(-1) in crabs fed with 1% plastic. The polypropylene microfibres underwent physical modifications as they passed through the foregut, resulting in a reduction in overall size and length. These modified fibres were then expelled and formed into separate balls, as indicated by the findings. Their findings provided evidence for the developing notion that the ingestion of microplastics has a significant biological impact, specifically by reducing the energy budgets of marine organisms affected by it.

Watts et al. (2014) investigated the notion that the shore crab (*Carcinus maenas*) can absorb microplastics by inhaling them through their gills, as well as by consuming food that has already been exposed to microplastics. The researchers utilized fluorescently labelled polystyrene microspheres (8-10 μm) to demonstrate that the crabs retained the ingested microspheres in their body tissues for a maximum of 14 days after ingestion and up to 21 days after inhalation through the gills. The uptake of microspheres was notably higher in the posterior gills compared to the anterior gills. Their Multiphoton imaging indicated that the majority of microspheres were trapped in the foregut after being exposed to the diet, primarily due to their attachment to the hair-like setae. Additionally, the microspheres were observed on the outer surface of the gills after being exposed to water. The findings were utilized to develop a basic conceptual framework of particle movement throughout the gills and the stomach. The results revealed that ventilation serves as a pathway for the absorption of microplastics into a widely found marine species that does not engage in filter feeding.

Microplastics, defined as plastic particles less than 5 mm in size, are ingested by coastal crabs through filter feeding, ingestion of contaminated prey, or direct uptake from sediments. Once ingested, microplastics can cause mechanical damage to the digestive tract, leading to abrasions, blockages, and perforations. Furthermore, the leaching of chemical additives from microplastics can induce toxic effects, disrupting cellular function, and compromising immune responses in coastal crabs (Wright et al. 2013).

Previous study demonstrated that the short-term exposure to polystyrene microspheres (8 μm) with various surface coatings had notable albeit temporary impacts on bronchial function. After inhaling microspheres into the gill chamber, there was a noticeable and proportional impact on oxygen consumption within 1 hour of exposure. However, the oxygen consumption levels returned to normal after 16 hours. The process of ion exchange was also impacted, as there was a notable reduction in sodium ions and a rise in calcium ions in the hemolymph after 24 hours of exposure. In order to evaluate the impact on osmoregulation, the researchers subjected crabs to lower salinity levels following their exposure to microplastics. The crab's response to osmotic stress was not affected by either microspheres or natural sediments, regardless of the concentration of plastic applied. Carboxylated (COOH) and aminated (NH_2) polystyrene microspheres exhibited distinct distribution patterns on the gill surface, although neither had a notable negative effect on gill function. Their findings demonstrated the magnitude of the physiological impact of microplastics on shore crabs, as well as the ability of these crabs to retain their osmoregulatory and respiratory functions even after being exposed to both man-made plastics and natural particles.

Microplastics which are plastics smaller than 5 mm, pose a possible danger to marine biodiversity. In order to investigate the impact of microplastic pollution on animal behaviour and cognition, researchers conducted a study using common European hermit crabs (*Pagurus bernhardus*) and observed their shell selection process. The goal was to determine whether exposure to microplastics affects their ability to perform crucial survival behaviours such as contacting, investigating and entering an optimal shell. A total of 64 female hermit crabs were housed in tanks, with 35 crabs in tanks containing polyethylene spheres and 29 crabs in tanks without any plastic. After 5 days, the crabs were moved to substandard shells and placed in an observation tank with a better alternative shell. The study demonstrated that hermit crabs exposed to plastic experienced a decline in their ability to choose suitable shells. These crabs were less inclined than the control group to make contact with optimal shells or enter them. Furthermore, they exhibited a delay in contacting and entering the most advantageous shell. Exposure to plastic had no impact on the duration of time spent examining the most suitable shell. Their findings demonstrated that microplastics hinder cognition, specifically the ability to acquire and interpret information, hence compromising crucial survival behaviour in hermit crabs.

CHAPTER III

MATERIALS AND METHODS

3.1 Materials and reagents needed

The following instruments were used in the study-

1. Metallic tray
2. Compass
3. Scale
4. Needle
5. Magnifying glass
6. Camera
7. Pin
8. Scale
9. Stereo-microscope
10. Scissors
11. Scalpel
12. Forceps
13. Deep freeze
14. 75% alcohol
15. 10% KOH
16. Distilled water
17. H_2SO_4

3.2 Study period and research stations

The experimental sample collection, species identification, separation, research work, analysis, and thesis writing were conducted for a period of one year, which was effective from July 2023 to June 2024. Sample dissection and digestion, observation, and identification of microplastics were conducted at the Bangladesh Fisheries Research Institute's Marine Station Laboratory in Cox’s Bazar.

3.3 Sample collection

The crab samples were collected at nine locations in the coastal areas of the Bay of Bengal. Samples were collected from three different locations in Chattagram (Chakaria-S1, Shagorika-S2, Bashbaria-S3), Cox's Bazar (Bakkhali-S4, Sonadia-S5, Ghotivangga-S6) and Saint Martin Island (East part of the island-S7, West part of the island-S8, Middle part of the island-S9) regions (Figure 3.1). The traditional method was used to catch the crabs. Each crab collected was packed in aluminum foil bags and temporarily stored in a cooler at -50°C). Then, it was stored at -20°C for processing and analysis. Before dissection and digestion, all crab samples were stored for no more than one week.

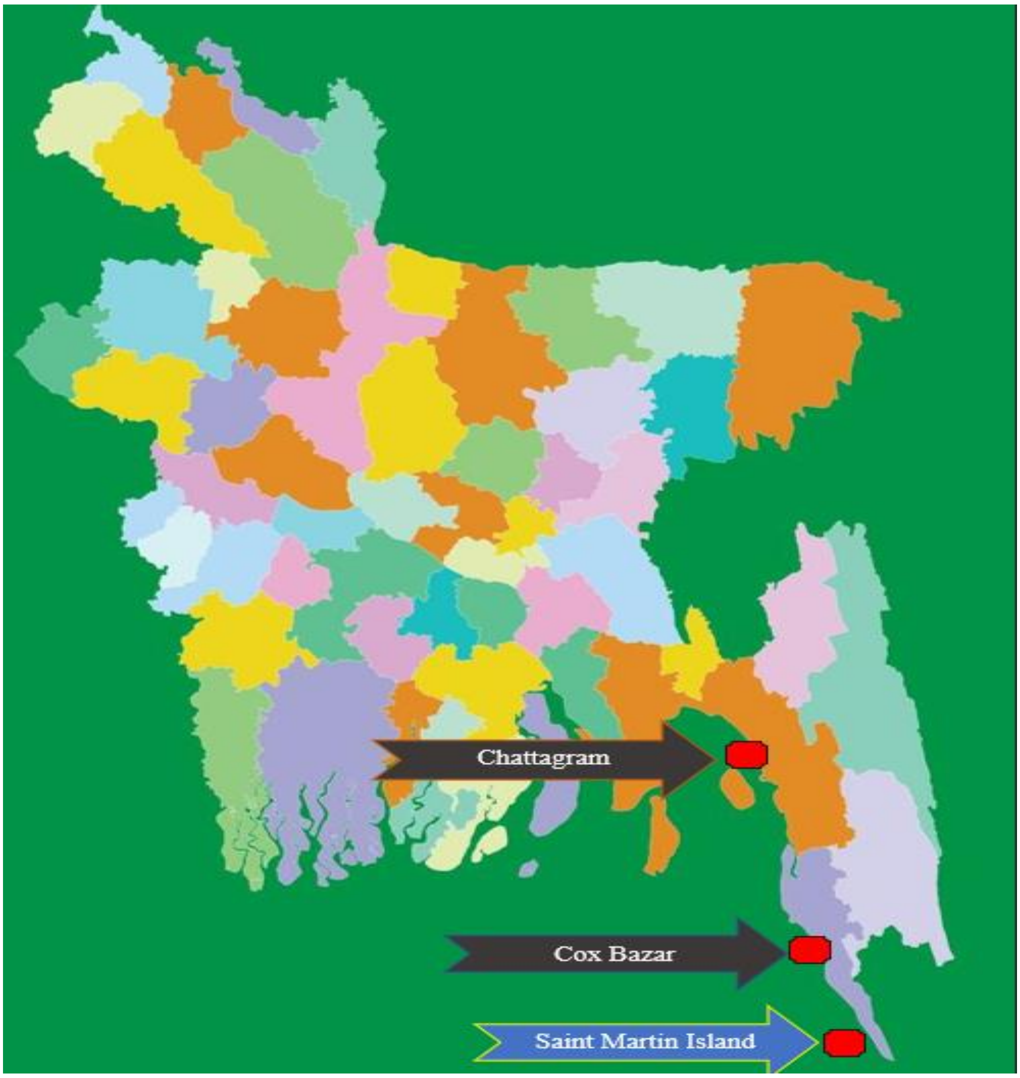


Figure 3.1 The sampling locations in Bangladesh (Red Marked Area)

3.4 Quality assurance and control

During sampling, aluminium foil bags were used quickly to pack the collected samples separately to reduce atmospheric contamination. Suitable preventive measures were applied to each possible step to prevent plastic and fibre contamination, for instance, during laboratory processing, and all operations, including crab dissection and microplastics observations, were carried out in a laminar flow cabinet. Minimal personnel were ensured in the laboratory, air circulation to the outdoors was minimized, and all chemical reagents were filtered with 8-mm glass microfibre filters before use. The forearms of the laboratory personnel were scrubbed three times with 75% alcohol before the laboratory procedures were performed, and white cotton lab coats, disposable latex gloves, and face masks were provided throughout sample manipulation and processing. In addition, all instruments and equipment will be thoroughly cleaned three times with 75% alcohol. The process blank was maintained.

3.5 Sample dissection and digestion

After thawing, the surface of each crab was carefully washed three times with filtered distilled water to remove foreign bodies. The total weight of the crab and the wet weight of each separated tissue were determined with a precision electronic balance. Each crab was dissected on a metal tray using a scalpel, forceps, and scissors, and the gut, gills, and muscles (not including leg muscles) were separated and placed in a clean beaker and covered with aluminium foil (Figure 3.2 and 3.3). The dissected and separated tissues were digested with 10% KOH in oscillation incubators at 40 °C and 60 rpm (Foekema et al. 2013; Karami et al. 2017) (Figure 3.4). After 24-48 hours of digestion, the clear and yellow digestion solution was filtered using a glass microfibre filter (2.7 mm pore size, Whatman Grade GF/D) (Figure 3.5). Then, the filter was placed in a clean Petri dish with a lid and dried completely at 40 °C.

Figure 3.2 Dissection process of crab samples for microplastics identification

Figure 3.3 Crab samples for microplastics identification

Figure 3.4 Digestion process of crab samples for microplastics identification

Figure 3.5 Filtration process of crab samples for microplastics identification

3.6 Observation and identification of microplastics

A DM1000 stereomicroscope was used to separate the suspected microplastics, take pictures of and measure their maximum cross-section (select and measure the two points with the largest distance by stereomicroscope), and classify them according to their size, shape, and color.

3.7 Data Analysis

Collected data were statistically analysed to find out the significant difference among the treatment by using the SPSS computer package program. The mean values of all the characters were calculated and analysis of variance was performed. For descriptive statistical analysis (Mean and Stander Error of Mean), where the mean differences were adjudged with Duncan's Multiple Range Test (DMRT), Analysis of Variance (ANOVA), and trait correlation analysis. Microplastics detection rate (%), abundance of size, color distribution, shape distribution of three crab species were detected.

CHAPTER IV

RESULTS

4.1 Microplastic detection rate (%)

The microplastics detection rate is presented in Figure 4.1.

The microplastic detection rate showed significant variation over the fishing ground of the various locations of the study (Figure 4.1). Among the microplastic collection sites, the highest detection was recorded in 15.2% in Bakkhali (S4) and Sonadia (S5) fishing ground followed by West Part (14.4%) and Middle Part (13.6%) of the fishing ground of the Saint Martin Island (S8 and S9 respectively). The lowest microplastic detection was recorded in Shagorika (S2) and Bashbaria (S3A) (6.4%) fishing ground area of Chattagarm.

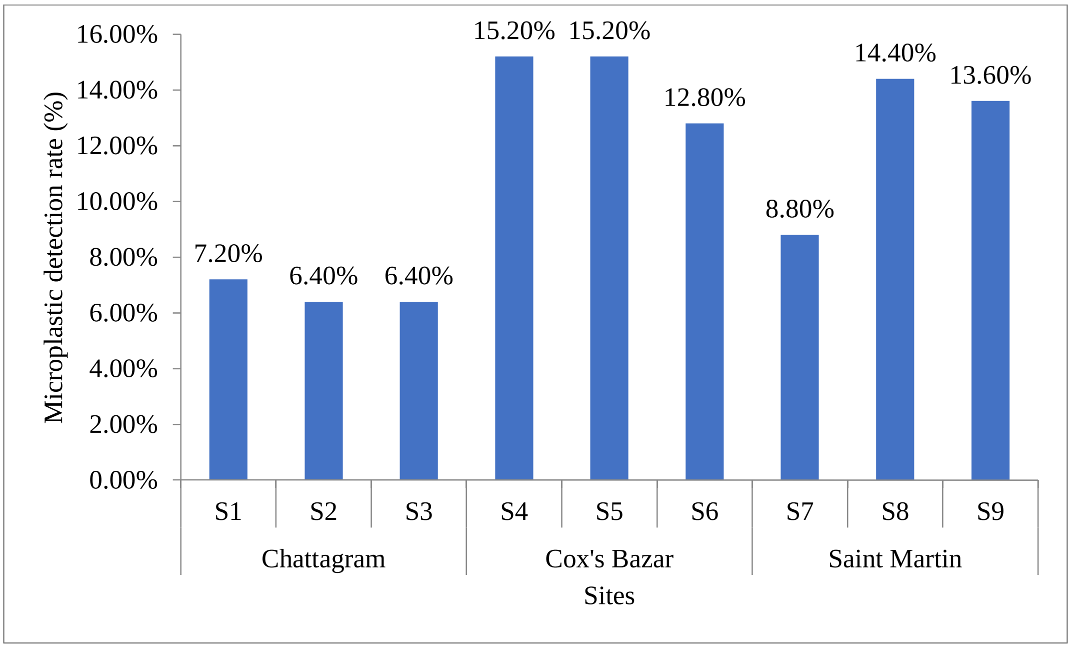


Figure 4.1 Microplastic detection rate (%)

Figure 4.2 Microplastics identified in *S. olivacea*

Figure 4.3 Microplastics identified in *P. pelagicus*

4.2 Abundance of microplastics (%)

The abundance of microplastics is presented in Figure 4.4.

The study found significant variation in the extent of microplastics present in *S. olivacea* across different fishing grounds (Figure 4.4 (A)). Out of the 9 places examined, only 6 demonstrated the presence of microplastics in *S. olivacea*. Nevertheless, the greatest amount of microplastics was found in the gut of *S. olivacea* in the Bakkhali (S4) fishing ground (35.30%) of the total Microplastics in Cox's Bazar. On the other hand, the highest proportion of Microplastics in the gills was seen in the Sonadia (S5) fishing ground, also accounting for 35.30% of the total microplastics in the same region. The Shagorika (S2) fishing ground (17.66%) in the Chattagram region had the second greatest number of microplastics in gill while same amount was recorded from Ghotivangga (S6) of Cox's Bazar.

The study found significant heterogeneity in the amount of microplastics in *P. pelagicus* across different fishing grounds (Figure 4.4 (B)). Out of the 9 fishing ground locations, only 5 demonstrated the presence of microplastics in *P. pelagicus*. However, the maximum Microplastics were recorded in the gut of *P. pelagicus* in the Bashbaria (S3) (30.00%) fishing grounds of Chattagram followed by Chakaria (S)1, Shagorika (S2) and Ghotivangga (S6) (20.00%) of Chattagram and Cox's Bazar respectively. The highest microplastics were recorded in the gill of *P. pelagicus* in the Bashbaria (S3) of Chattagram (30.00%) followed by Chakaria (S)1, Shagorika (S2) and Ghotivangga (S6) (20.00%) of Chattagram and Cox's Bazar respectively. The Sonadia (10.00%) fishing site in Cox's Bazar had the lowest incidence of microplastics.

There was significant diversity in the abundance of microplastics in *P. sanguinolentus* across different research locations (Figure 4.4 (C)). Out of the total of 9 locations, only 6 exhibited the presence of microplastics in *P. sanguinolentus*. The greatest quantity of microplastics was found in the gut of *P. sanguinolentus* in the Sonadia (S5) fishing ground of Cox’s Bazar and the West Part of the Saint Martin Island (S8) fishing ground, accounting for 27.78% and 27.77%, respectively. The East Part of the Saint Martin Island (S7) fishing ground had the lowest incidence (5.56%). The microplastics in gill was recorded in Middle Part of the Saint Martin Island (S9) fishing ground (24.00%) followed by East Part of the Island (S7) (20.00%) and West Part of the Saint Martin Island (S8) fishing ground and Bakkhali (S4) of Cox’s Bazar (16.00%). The Sonadia fishing site (S5) in Cox’s Bazar had the lowest frequency of microplastics (8.00%).

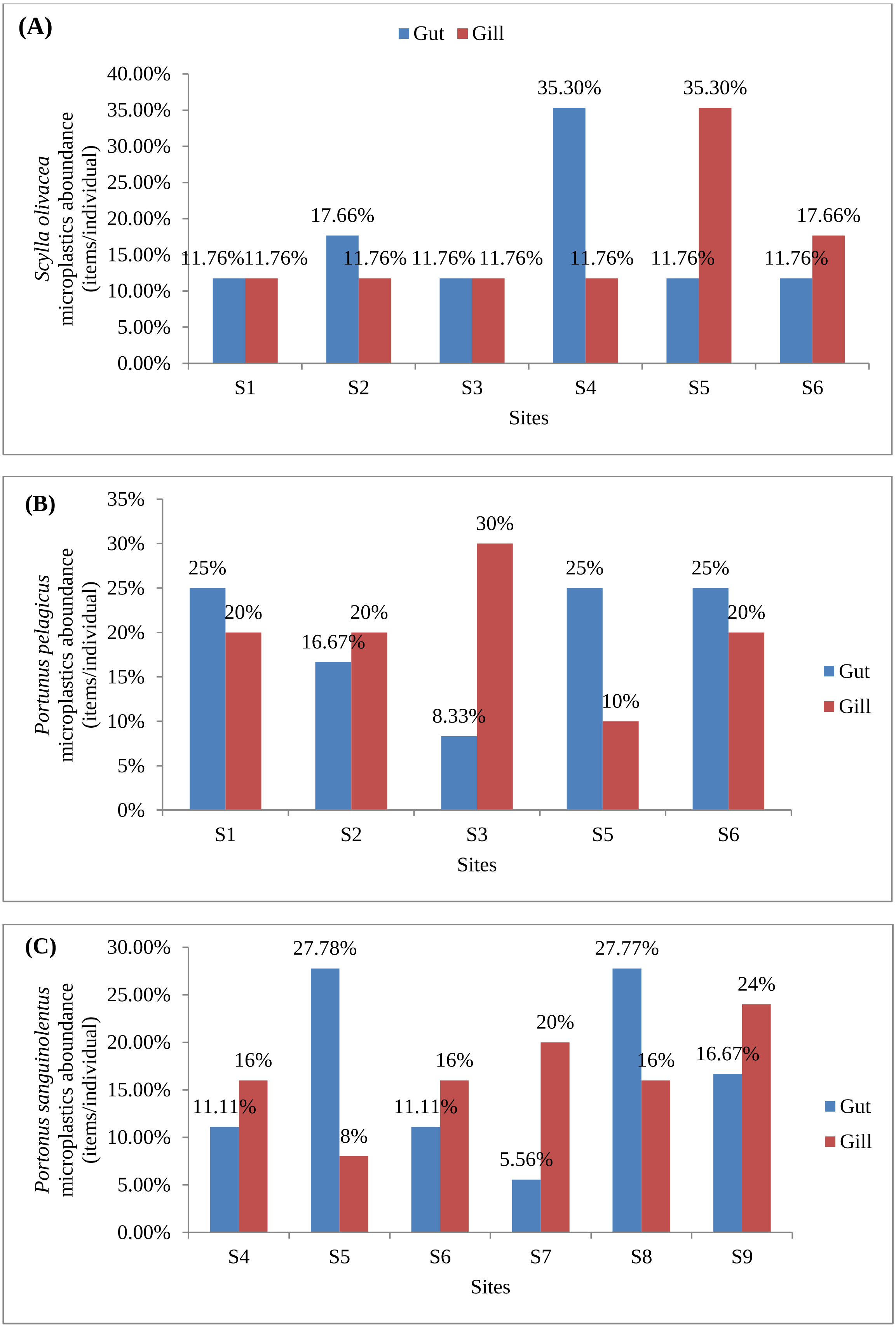


Figure 4.4 Abundance of microplastics (%) in two tissues (gut and gills) of three crab species (A) *S. olivacea*, (B) *P. pelagicus*, (C) *P. sanguinolentus*

4.3 The size of microplastics identified in the study areas

The size of the microplastics is presented in figure 4.5.

There was notable variance in the size of the microplastics in *S. olivacea* across different research locations, as depicted in Figure 4.5 (A). The largest microplastic sizes were found in the gut and gills of *S. olivacea* samples of the Chakaria fishing ground (S1) in Chattagram (0.22 mm) statistically alike to the samples of Sonadia fishing ground (S5) in Cox's Bazar (0.15 mm), and the Bashbaria fishing ground (S3) in Chattagram, where the microplastics size was 0.09 mm. The smallest microplastics were discovered at the samples of Sagorika fishing ground (S5) site (0.03) in the Chattagram region which is statistically alike to rest of the all samples except Chakaria fishing ground (S1) in Chattagram.

The size of the microplastics in *P. pelagicus* exhibited substantial variance across the different study locations (Figure 4.5 (B)). However, the maximum sized microplastics were recorded in the samples of *P. pelagicus* in the Bashbaria (S3) fishing ground of Chattagram (0.22mm) followed by Ghotivangga (S6) fishing ground (0.1676 mm) samples of Cox's Bazar. The minimum sized microplastics value was recorded from the samples of Shagorika (S2) (0.03mm) and Chakaria (S1) fishing ground (0.03mm) of Chattagaram region which is statistically alike to rest of the all samples except Bashbaria (S3) fishing ground in Chattagram. The Size of the Microplastics in *P. sanguinolentus* showed significant variation over the various locations of the study (Figure 4.5 (C)). The largest microplastic particles were found in the gut samples of the samples of *P. sanguinolentus* of the Bakkhali (S4) fishing ground (0.228 mm) statistically similar to Ghotivangga (S6) (0.16 mm) and East Part of the Island of Cox's Bazar and Saint Martin (0.13 mm) respectively. The smallest recorded values were obtained from the Western (S8) and Middle parts (S9) of Saint Martin Island (0.36 mm and 0.34 mm), respectively.

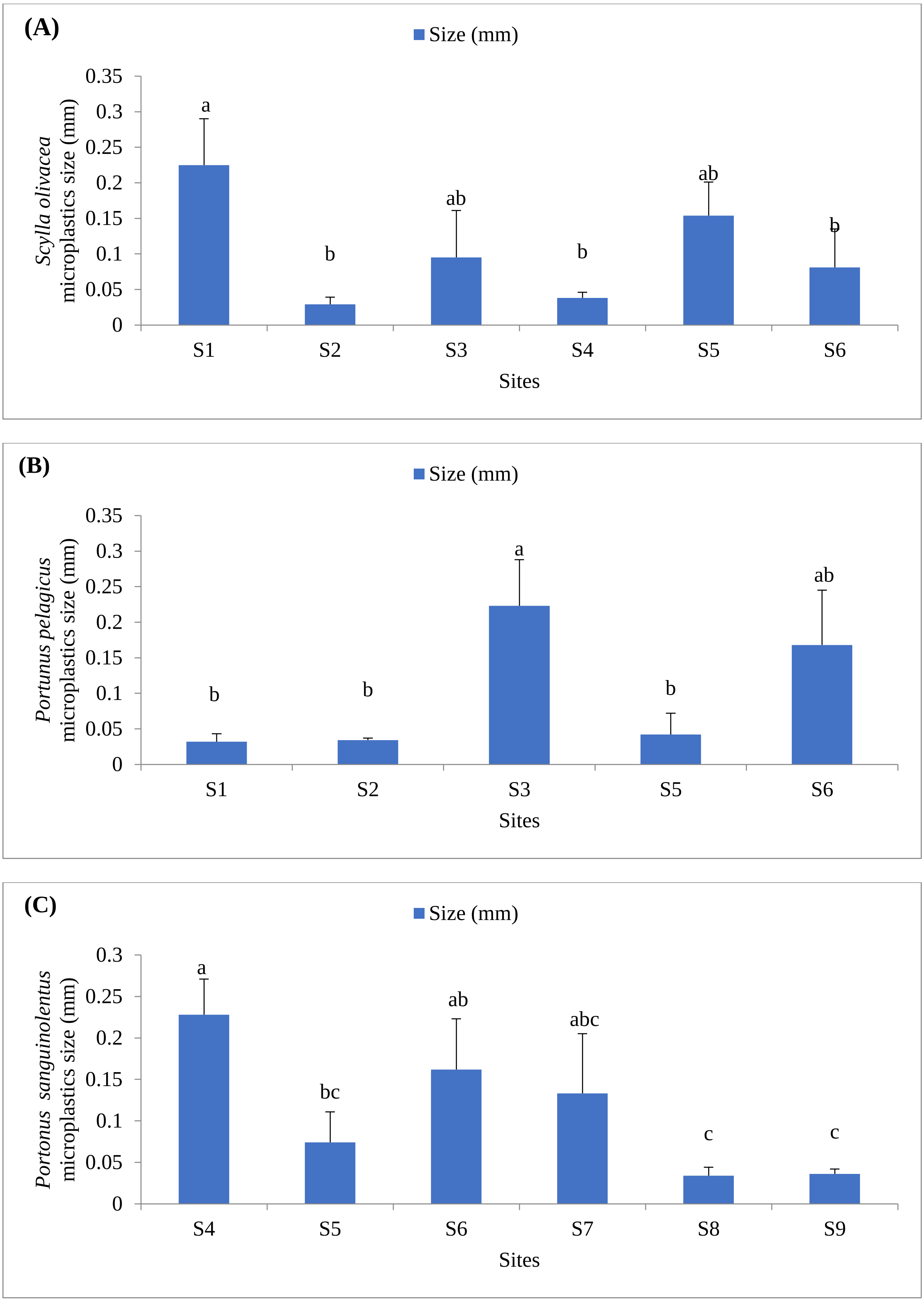


Figure 4.5 The size of microplastics identified from (A) *S. olivacea*, (B) *P. pelagicus*, (C) *P. sanguinolentus*

4.4 Color distribution of the microplastics in different tissues (gut and gill) of the Crab Samples

Color distribution of the microplastics in different tissues of the Crab Samples is presented in figure 4.6.

In the study, the various microplastics colors revealed significant variation in the gut of the studied crab samples (Figure 4.6 (A)). The study recorded four types of color (black, red, purple, and black grey) in the crab samples' guts. The maximum crab sample had the highest number of black-type microplastics, which significantly differed from the other types (42.37%). Subsequently, red-colored microplastics were identified in the crab samples, which possessed the second-highest position (28.81%). The purple microplastics generally constitute 16.95% of the identified sample. However, the lowest sample was recorded for black grey (11.86%).

The various microplastics color found significant variation in the gill of the crab samples of the study (Figure 4.6 (B)). The study documented four distinct color variations (black, red, purple, and dark grey) observed in the gill of the crab samples. The crab sample with the highest quantity had a predominant presence of black-colored microplastics, accounting for 45.45% of the total sample. This proportion differed considerably from the other types of microplastics present. Adjacent to it, crab samples were found to include red-colored microplastics, which accounted for the second greatest proportion (28.79%). The microplastics that are black or grey in color typically make up 16.67% of the identified sample. Nevertheless, the purple colour exhibited the lowest recorded sample at 9.09%.

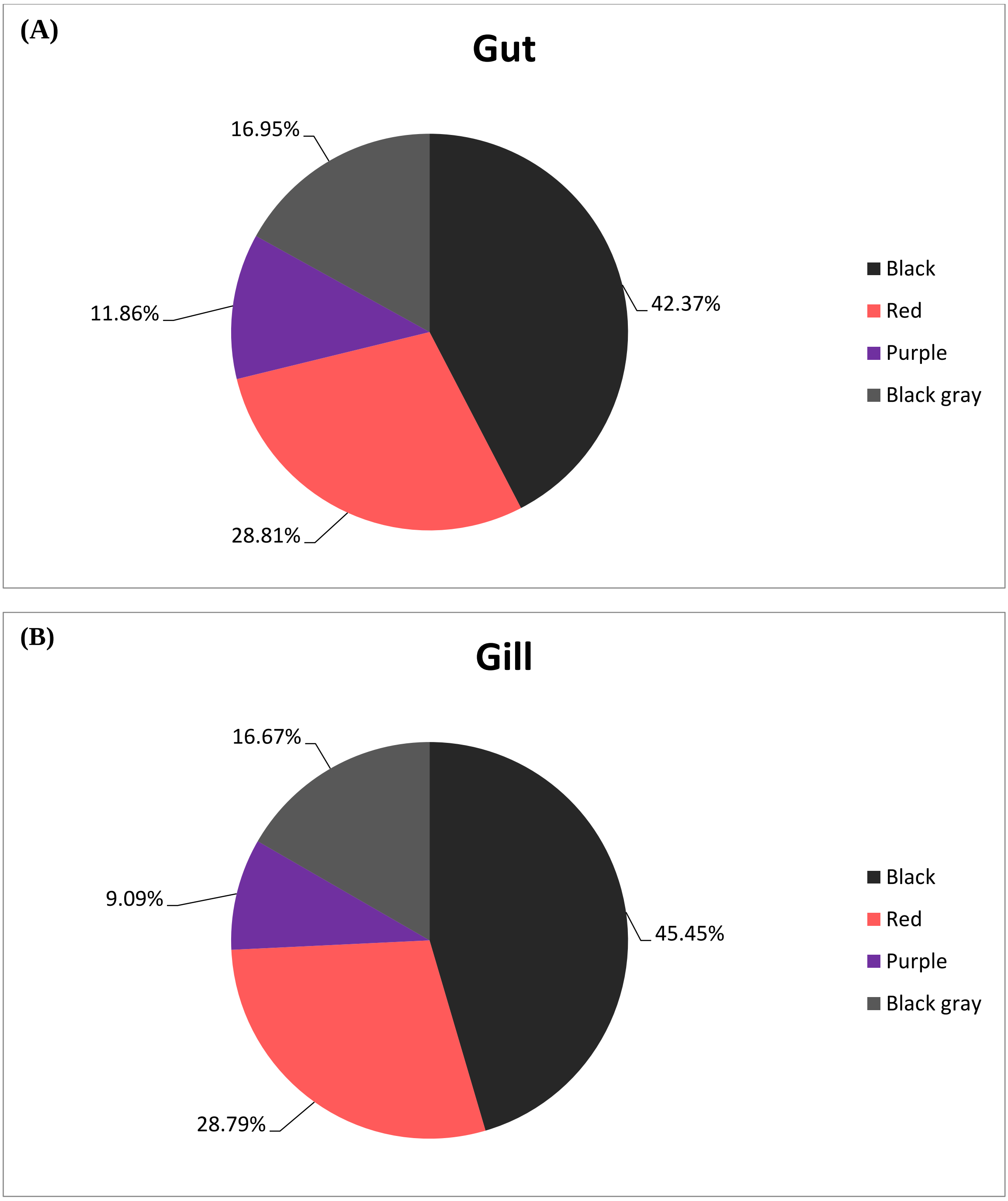


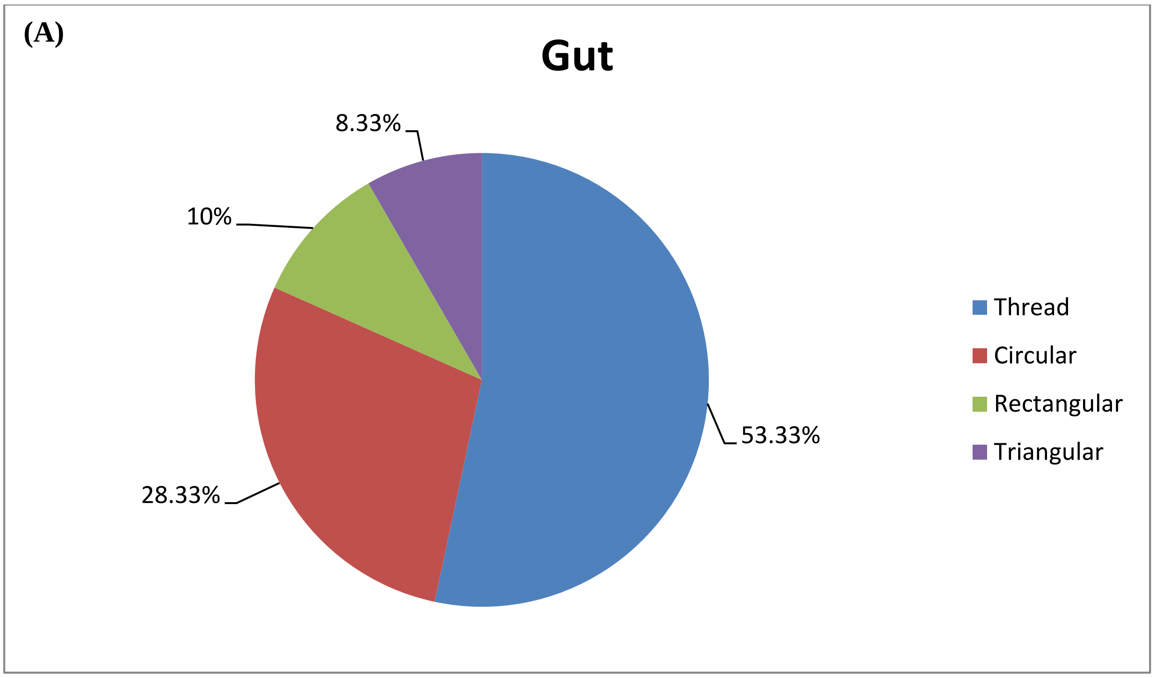
Figure 4.6 Color distribution of the microplastics in different tissues (A) Gut and (B) Gill of crabs

4.5 Shape distribution of the microplastics in different tissues (gut and gill) of crabs

Shape distribution of the microplastics in different tissues of crabs is presented in figure 4.7.

The study observed significant diversity in the gut of the crab samples in terms of the different types or forms of microplastics present (Figure 4.7 (A)). Crab samples were found to have four distinct types of shapes for microplastics. These are thread, circular, rectangular and triangular shaped. The crab sample with the highest quantity exhibited thread-like microplastics, which considerably differed from the other samples (53.33%). Next to it, crab samples were found to contain a circular kind of microplastics, which accounted for the second greatest proportion (28.33%). Rectangular microplastics typically make up 10% of the identified sample. However, the triangular forms had the lowest reported percentage (8.33%).

The various types or forms of microplastics found significant variation recorded in the gill of the crab samples of the study (Figure 4.7 (B)). Generally, four types or forms of microplastics were identified in the crab samples. These are thread, circular, rectangular and triangular type. The largest crab sample had a predominant presence of thread type microplastics, accounting for 56.92% of the total amount, which considerably differed from the other types. Adjacent to it, circular type of microplastics was identified in the crab samples which possess the second highest position (26.15%). The microplastics those are triangular type generally constitute the 9.23% of the identified sample. However, the lowest sample was recorded for types (7.69%).



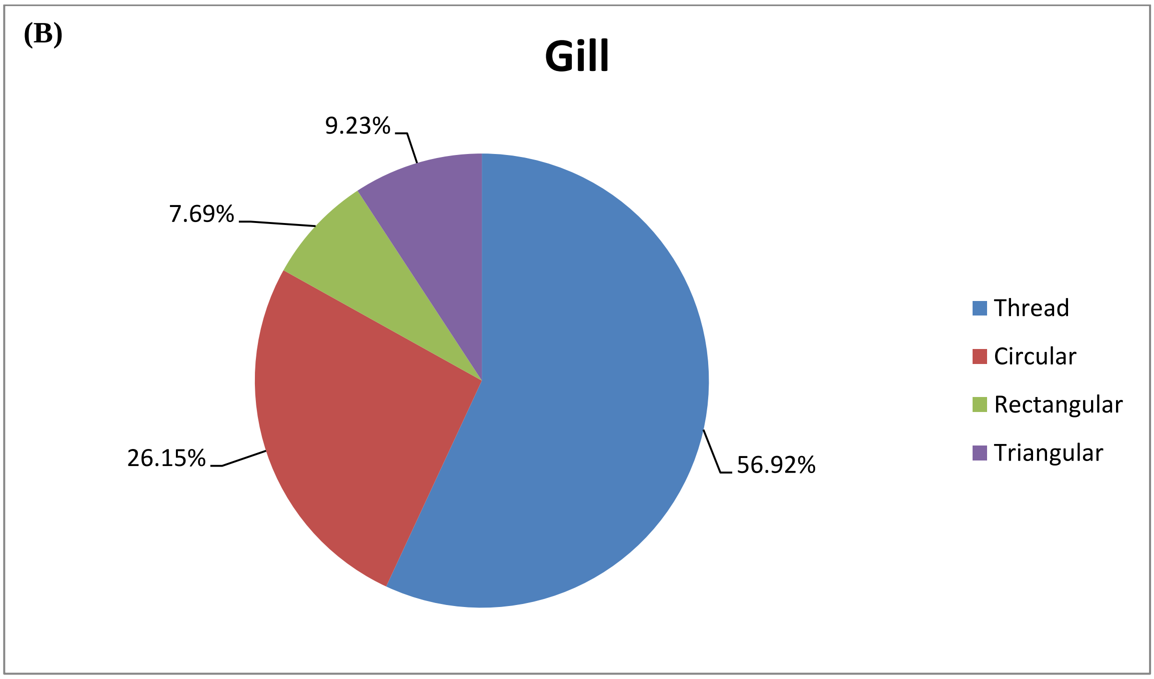


Figure 4.7 Shape distribution of the microplastics in different tissues (A) Gut and (B) Gill of crabs

4.6 Types of microplastics in different tissues (gut and gill) of crabs

Types of microplastics in different tissues of crabs is presented in figure 4.8.

The various types or forms of microplastics found significant variation recorded in the gut tissues of the crab samples of the study (Figure 4.8 (A)). Crab samples were found to contain four distinct types or forms of microplastics. The types include fibre, sheet, fragment, and microbead. The crab sample with the highest quantity exhibited the highest proportion of fragment type microplastics, which substantially differed from the other types (49.18%). The crab samples were found to have a fibre type of microplastics, which ranked second highest at 34.43%. The microplastics that are in the form of sheets and microbeads typically make up an equal proportion (8.20%).

The study observed significant diversity in the gill tissues of the crab samples in relation to the different types or forms of microplastics (Figure 4.8 (B)). Crab samples were found to contain four separate kinds or forms of microplastics. The types include fibre, sheet, fragment, and microbead. The maximum crab sample showed fragment microplastics which is maximum in amount and significantly varied from others (50.00%). The crab samples were found to have a fibre type of microplastics next to them, which ranked second highest at 34.38%, followed by the sheet sample at 9.38%. The microbead type often makes up the smallest percentage among all the samples (6.25%).

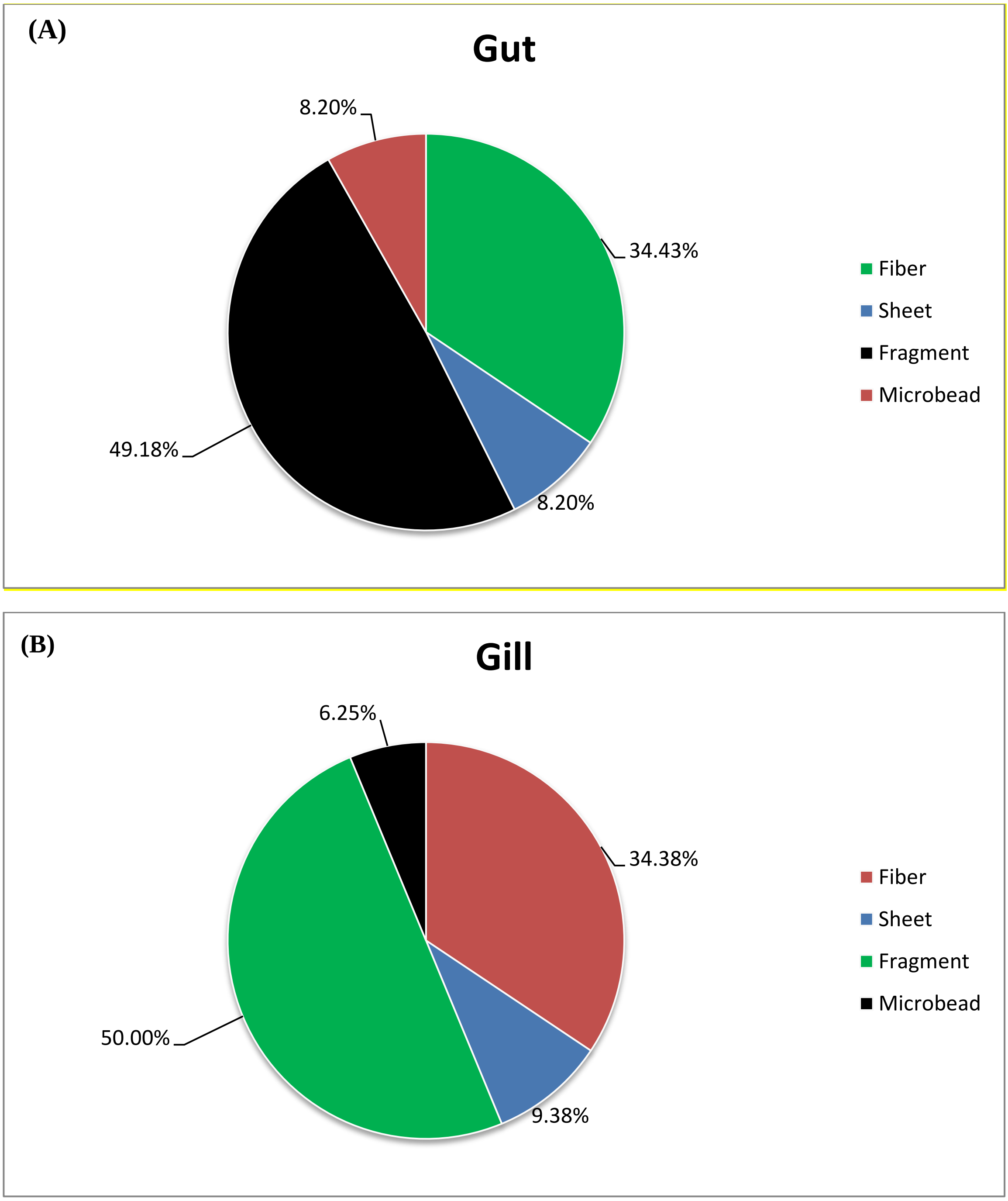


Figure 4.8 Types of microplastics in different tissues (A) Gut and (B) Gill of crabs

CHAPTER V

DISCUSSION

The results showed that the microplastics are most abundant in Cox's Bazar followed by Saint Martin and Chattagram region. Different crab species contain varying amounts of microplastic in different coastal regions of Bangladesh due to differences in their feeding habits and habitats. Crabs that dwell in or near heavily polluted areas, such as urban or industrial regions, are more likely to ingest microplastics. Additionally, the varying water currents, tides, and local waste management practices influence the distribution and concentration of microplastics in different coastal areas. The specific morphology and behaviour of each crab species also play a role in how much microplastic they accumulate. Zhang et al. (2021) recorded the identification variation of crab samples in at three important fishing grounds in China.

Microplastics were found in the gut and gills of *S. olivacea* in all other locations that were examined. Interestingly, none of the samples taken from Saint Martin Island contained any trace of microplastics in *S. olivacea*. Different crab species, including *S. olivacea*, are found in varying amounts in coastal regions of Bangladesh due to differences in habitat preferences, availability of food sources, and environmental conditions such as salinity and temperature. The distribution is also influenced by human activities like fishing pressure and habitat destruction, which can affect population sizes and distribution patterns. Additionally, the presence of predators and competition with other species can impact the abundance of *S. olivacea* in certain areas. Lastly, variations in reproductive cycles and larval dispersal mechanisms contribute to the uneven distribution of these crabs along the coast. Interestingly, no samples of *P. pelagicus* from Saint Martin Island showed the presence of microplastics. Chattagram region offers a more suitable habitat for the crab species *S. olivacea* due to its extensive mangrove forests and estuarine conditions, which provide ideal breeding and feeding grounds. Cox's Bazar and Saint Martin Island, while coastal, lack the dense mangrove ecosystems that are crucial for the crabs' lifecycle. Additionally, the nutrient-rich waters and sediment types in Chattagram support higher crab populations compared to the relatively less diverse marine environments of the other two regions may be the reasons of its dominance in Chattagram. Interestingly, no samples of *P. sanguinolentus* from Saint Martin Island showed the presence of microplastics. The abundance of microplastics in *P. sanguinolentus* in Bangladesh varies due to differences in pollution levels across different coastal areas, as regions with higher human activity and industrial discharge tend to have higher concentrations of microplastics. Additionally, the feeding habits and habitat preferences of *P. sanguinolentus* influence their exposure to microplastics, as crabs in areas with more sediment and organic matter may ingest more microplastic particles. Seasonal variations, such as monsoon rains, can also affect the distribution and abundance of microplastics, impacting the levels found in marine organisms. The abundance of microplastics in *S. olivacea* of Bangladesh varies due to factors such as the proximity to urban and industrial areas, which increases microplastic pollution from waste discharge. Additionally, variations in fishing locations and water currents can influence the concentration of microplastics ingested by these crabs. Seasonal changes and human activities, such as tourism and fishing practices, also contribute to the fluctuating levels of microplastics in their habitat. The abundance of microplastics in *P. pelagicus* in Bangladesh varies due to differences in pollution levels across various coastal regions, influenced by local human activities such as industrial discharges, fishing, and waste management practices. Additionally, ocean currents and tides can redistribute microplastics, leading to variability in their concentration in different habitats. Seasonal changes and monsoonal impacts further contribute to this variability by affecting the distribution and accumulation of microplastics in the marine environment. Microplastics were found in the gills and intestines of Chub mackerel, a marine fish species, along with the stomach, indicating contamination in multiple organs (Hasanah et al. 2023; Villagran et al. 2019). Microplastics were found in the intestines of marine fish species in the Northern Bay of Bengal, posing a public health threat through the food chain (Hossain et al. 2019).

The size of microplastics in *Portunus sanguinolentus* from Bangladesh varies due to several factors, including differences in the sources and types of plastic pollution, varying environmental conditions like currents and tides that influence plastic degradation, and the feeding habits and habitats of the crabs which affect their exposure to different sizes of microplastics. Additionally, microplastic fragmentation processes, both physical and chemical, contribute to the diversity in sizes encountered within these crabs. This variability highlights the complex interactions between marine organisms and their polluted environments. microplastics were found in the gills and guts of the crabs. The abundance and size of the microplastics in the guts were significantly higher than those in the gills (Zhang et al. 2020). Compared with large plastics, microplastics are more easily dispersed in the environment due to their small size. The study found that microplastics smaller than 1000 mm were the most prevalent. The presence of this finding was also detected in the saltwater and sediments of the Yellow Sea and the East China Sea in China (Feng et al. 2019; Zhu et al. 2019). Regrettably, these diminutive microplastics appear to have a particularly harmful effect on organisms. Lee (2013) discovered that tiny microplastics have highly harmful impacts on the organs of adult and juvenile copepods. Moreover, the microplastics present in the stomach are primarily consumed during crab feeding activities, and certain smaller particles can be transported to the hemolymph or other tissues (Farrell and Nelson, 2013).

The abundance and color distribution of microplastics in different gut tissues of crab samples from Bangladesh may vary due to the differing environmental conditions and sources of plastic pollution in their habitats. Different gut tissues might accumulate and retain various types and sizes of microplastics differently, influencing their color distribution. Additionally, dietary habits and feeding mechanisms of crabs can lead to the ingestion of microplastics with distinct characteristics depending on the surrounding microplastic availability and composition. The color of microplastics found in marine organisms, such as crabs in Bangladesh, varies across different studies. In freshwater fish species in Bangladesh, black-colored microplastics were the most dominant, followed by white, blue, red, and green (Khan and Setu, 2022).

The abundance of shape distribution of microplastics in different gut tissues of crabs in Bangladesh varies due to factors such as differences in feeding habits, habitat conditions, and the size and type of microplastics present in the environment. Crabs may ingest microplastics of varying shapes based on their feeding strategies and the specific ecological niches they occupy, leading to discrepancies in microplastics shape distribution across gut tissues. Additionally, environmental factors like water currents and sediment composition influence the types and abundance of microplastics available for ingestion by crabs in different regions. The variation in the shape distribution of microplastics across different gill tissues in crabs from Bangladesh may stem from diverse factors. These could include variances in the types and sources of microplastics present in the environment, variations in the filtering mechanisms of different gill tissues, and potential differences in the accumulation and retention capacities of microplastics within specific tissues. Additionally, local environmental conditions and anthropogenic activities might contribute to the heterogeneous distribution patterns observed. Zhang et al. (2021) recorded the alike result. Research on microplastics in marine environments in Bangladesh has shown that these particles can be in the form of fibres, films, lines, fragments, and foam (Khan and Setu, 2022). Additionally, studies on microplastics in the sediments of the Karnaphuli river estuary revealed that the predominant shape of microplastics was films, with white being the most common color and sizes ranging from 1-5 mm (Rakib et al. 2022).

The variance in the type distribution of microplastics among different gut tissues of crabs in Bangladesh likely arises from variations in ingestion mechanisms and digestive processes. For instance, certain gut tissues might have higher affinity or capability for retaining specific types of microplastics due to their composition or function. Environmental factors such as the concentration and type of microplastics present in the crabs' habitat could also influence the distribution pattern across different gut tissues. The variation in the abundance of different types of microplastics across various gill tissues of crabs in Bangladesh can be attributed to differences in exposure routes and filtration efficiency. For instance, fibre microplastics might be more prevalent in certain gill tissues due to their ability to be trapped during filter feeding processes. Additionally, factors such as water currents and particle size distribution can influence the retention of various MP types in different gill structures, leading to heterogeneous distributions. In the Yellow Sea and East China Sea, fibres are the most prevalent shape because plastic fishing gear, such as nets and ropes, is used so frequently (Browne et al. 2008; Feng et al. 2019; Zhang et al. 2019).

CHAPTER VI

SUMMARY AND CONCLUSION

Crabs are an indispensable component of the benthic environment and serve as readily available and commonly consumed seafood for humans. Nevertheless, there is limited knowledge regarding the accumulation of microplastics in various tissues of crabs inhabiting significant fishing regions. This study examined the quantities and properties of microplastics in various tissues of three species of wild crabs (*S. olivacea, P. pelagicus,* and *P. sanguinolentus*) in three significant fishing areas (Chattagram, Cox's Bazar, and Saint Martin Island region) in Bangladesh. Every region is comprised of three separate fishing grounds. The study revealed that Cox's Bazar detected Microplastics most, followed by Saint Martin and Chattagram. The crab species *S. olivacea* and *P. pelagicus* were found in the highest numbers in Cox's Bazar and Chattagram. On the other hand, *Portunus sanguinolentus* was most plentiful in Cox's Bazar and Saint Martin Island. The size of the Microplastics also differed among the crab samples. Larger Microplastics were seen in the crab samples collected from the Chattagram region. The prevalence of black-colored microplastics was highest in the gill and gut samples, accounting for 45.45% and 42.37% respectively. The thread type is the most prevalent shape in the gill, accounting for 56.92% of the total. On the other hand, the thread type is most commonly found in the gut, representing 53.33% of the total. The 50% gill tissue showed fragment microplastics followed by microbead (6.25%). The 49.18% gut tissue showed microbead microplastics followed by fibre (8.20%).

The microplastic collection sites showed the greatest detection rate of 15.2% in Bakkhali (S4) and Sonadia (S5) fishing ground, followed by West Part (14.4%) and Middle Part (13.6%) of the fishing ground of Saint Martin Island. The fishing grounds of Bashbaria (S3) and Shagorika (S2) in Chattagarm recorded the lowest detection rate of microplastics at 6.4%.

The highest concentration of microplastics was discovered in the gut of *S. olivacea* in the Bakkhali (S4) fishing area, representing 35.30% of the overall microplastics in Chattagram. Conversely, the Sonadia (S5) fishing site had the greatest percentage of microplastics in the gills, making up 35.30% of the total microplastics in the region. The highest microplastics concentration was found in the gut of *S. olivacea* in the Chakaria (S1), Sonadia (S5), and Ghotivangga (S6) (25%) fishing grounds in the Chattagram and Cox's Bazar region. This was followed by the Shagorika (S2) (20%) and Bashbaria (S3) (30%) fishing grounds. In terms of gill contamination, the highest quantity of Microplastics was recorded in the Bashbaria (S3) (30%) fishing ground in Chattagram. Only six of the nine locations showed evidence of microplastics in *P. sanguinolentus*. The greatest number of Microplastics (27.78% and 27.78%, respectively) was discovered in the gut of *P. sanguinolentus* in the Sonadia (S5) fishing ground in Cox's Bazar and the West Part (S8) of the Saint Martin Island fishing area. The Bashbaria (S3) fishing field in Chattagram was where the microplastics in Gill were detected (11.76%). Sonadia (S5) fishing ground (35.30%) in the Cox's Bazar region had the second-highest microplastics in Gill, followed by Chakaria (S1), Shagorika (S2) fishing ground (11.76%) in Chattagram, and Ghotivangga (S6) fishing ground (17.66%) in the same region. The gut and gills of *S. olivacea* samples from the Chakaria (S1) fishing site in Chattagram comprised the greatest microplastic sizes (0.22 mm). The largest microplastics, however, were found in *P. pelagicus* samples from the Bashbaria (S3) fishing ground in Chattagram (0.22mm), followed by samples from the Ghotivangga (S6) fishing ground (0.17 mm) and Sonadia (S5) fishing ground (0.04mm) in Cox's Bazar. The samples from the Chattagaram region's Shagorika (S2) (0.34mm) and Chakaria (S1) fishing site (0.03mm) yielded the minimum-sized microplastics value. The gut samples of *P. sanguinolentus* from the Bakkhali (S4) fishing site contained the biggest microplastic particles, measuring 0.23 mm. The second-largest microplastics (0.16 mm) were found in the crab samples from the

Ghotivangga (S6) fishing ground in Cox's Bazar. The samples from the East Part (S7) of Saint Martin Island fishing ground had microplastics of 0.13 mm. The Western (S8) and Middle regions (S9) of Saint Martin Island yielded the lowest levels ever measured, measuring 0.34 and 0.36 mm, respectively. In gut, there were substantially more black-type microplastics (42.7%) in the maximum crab sample than in any other form. Then, red-colored microplastics were found in the crab samples, which had the second-highest percentage (28.81%). Generally, 16.95% of the identified sample consists of black gray microplastics. On the other hand, the purple sample had the lowest percentage (11.86). In gill black-colored microplastics predominated in the largest quantity of crab samples, making up 45.45% of the sample as a whole. This percentage was significantly different from the other microplastics types. Red-colored Microplastics were discovered in crab samples next to it, making up the second-highest percentage (28.79%). Typically, 16.67% of the detected sample consists of black gray microplastics. In contrast to the other samples, the crab sample that had the largest quantity of thread-like microplastics (53.33%) showed a notable difference. The next highest percentage (28.33%) was found to be accounted for by a circular type of Microplastics identified in crab samples. Approximately 10% of the identified sample are rectangular Microplastics. The triangular forms, however, had the lowest percentage (8.33%) that was reported. In contrast to the other categories, thread-type microplastics were primarily present in the largest crab sample, making about 56.92% of the total amount. In the crab samples, a circular type of Microplastics was found next to it, accounting for the second greatest proportion (26.15%). The Microplastics that have a triangle shape typically make up 9.23% of the detected sample. The crab sample that had the greatest number displayed the largest proportion of microplastics of the fragment type, which significantly varied from the other types (49.18%). The crab samples were discovered to include a fragment type, which had the second-highest percentage (34.43). The microplastics, which exist in the form of sheets and microbeads, normally constitute an equal proportion of 8.20%. The crab sample with the highest quantity had the greatest proportion of fragment-type microplastics (50.00%), which significantly differed from the other samples. The crab samples were discovered to be accompanied by a fibre type known as Microplastics, which had the second greatest proportion of 34.38%, followed by the sheet sample with a proportion of 9.38%. The microbead type typically constitutes the smallest proportion among all the samples, accounting for 6.25%. The study provided comprehensive information on the distribution pattern, number, size, colour, and shape of microplastics in the crab sample under investigation. This data can assist biologists in gaining a deeper understanding of plastic pollution in coastal ecosystems, using crabs as a model species.

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