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Research Article

A comparative study of microplastic detection in *Nemipterus japonicus*, *Rastrelliger kanagurta*, *Arius* sp. and *Scylla olivacea* from Chennai Coastal Region, India using ATR-FTIR spectroscopy

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ABSTRACT

Microplastics (<5 mm) are omnipresent pollutants produced directly or generated because of larger plastic particle breakdown. The challenge of microplastic pollution is an emerging global concern, with India being no exception. This study investigated the prevalence and characteristics of microplastics in four commercially important aquatic species from two distinct ecosystems in Tamil Nadu, India viz., the Ennore Creek (brackish water) and the Kasimedu landing center (marine). The species examined were catfish (*Arius* sp.), mud crab (*Scylla olivacea*), Japanese threadfin bream (*Nemipterus japonicus*) and Indian mackerel (*Rastrelliger kanagurta*). Microplastics were detected in 78.57% of the 70 samples analyzed, with *Nemipterus japonicus* and *Arius* sp. showing the highest average ingestion of 5±3 and 4±2.5 microplastic items per individual respectively. A distinct organ-specific trend was observed, with gills harboring slightly more microplastics (0.35 items/gills) compared to guts (0.21 items/gut). Fibers and fragments were the predominant microplastic shapes, while off-white (translucent), white, blue and black were the most common colors detected. ATR-FTIR analysis identified low-density polyethylene (LDPE) and polyamide (nylon) as the primary polymer types. The research underscores considerable interspecies and species-specific variations in microplastic accumulation and dispersion, underscoring the necessity for precise, species-specific evaluations to comprehend the potential ecological and anthropogenic health ramifications of this escalating environmental issue. Recommendations include establishing comprehensive monitoring programs, implementing source reduction strategies, enhancing habitat conservation, and fostering collaborative research to address microplastic pollution in the studied ecosystems.

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INTRODUCTION

The environmental issue that is currently creating waves, is plastic pollution. According to Lampitt et al. [1] around 400 million tons of plastic waste is being generated every year. The first contamination caused by microplastics in water bodies, was recorded by Carpenter and Smith et al. [2] during the year 1972, detected microplastics on the Sargasso Sea surface. The plastic production increased its peak during the 1940's and it became the fastest growing industry worldwide. All developing countries contributed to the major production of plastics. During the year 2013, Asia alone contributed (45.6%) that is quarter of the world plastic production. This was followed by Europe which contributed 22.9% of plastic produce, North America with 19.4%, middle East and Africa, 7.3% and finally Central and South America with 4.8%. Packaging was the major factor for the rise of plastic manufacturing. Consumer and household products (like appliances, toys, plastic cutlery, and furniture) also act as sources for the production. During the year 2012, approximately 10 –12 million tons of plastic was found, dumped in the ocean. The Indian Ocean had about 60000 tons of plastic accumulated during the year 2014 and the Pacific Ocean had about 100000 tons of plastic in the sea, which was alarming [3–7]. According to Geyer [8], 6300 metric tons of plastic waste was generated during the year 2016. 79% of waste was being dumped in the landfills without proper disposal. Borelle et al. [9] estimated that about 19 to 23 metric tons of plastic waste was generated worldwide during 2017. One of the notable events that occurred in plastic production worldwide was during the year 2017–2018 when China imposed a plastic waste import ban, which was reported by Wen et al. [10]. This resulted in a sharp decrease in the plastic production worldwide. The usage of single use plastics (SUP) trended during the year 2019, again contributing to 50% of plastic produce worldwide. According to Chen et al. [11], 360 million metric tons of SUP's were mass produced, globally. Due to the prevalence of the COVID-19 pandemic, use of SUP's such as gloves (2.5 million per day), masks (4.6 million per day) and disposable gowns (20 million per day) peaked during the year 2020 in India. This statistical data was provided by Shams et al. [12]. According to Walker et al. [13], 8300 million tons of plastic was produced during the year 2022 to 2023, out of which 6300 million tons of plastic was not disposed properly. By the year 2050, the plastic production would be doubled to billions of metric tons per year.

Plastic waste is broadly categorized into four classes based on size: macroplastics (>25 mm), mesoplastics (5–25 mm), microplastics (<5 mm) and nanoplastics (<100 nm) [14–17]. Microplastics are further distinguished as primary or secondary based on their origin [18, 19]. Primary microplastics are miniscule particles intentionally manufactured for microscopic applications, commonly found in personal care products and synthetic textiles [20]. Secondary microplastics, conversely, originate from the fragmentation of larger plastic debris due to environmental factors [21]. These microplastics (MPs) can be of different shapes like microbeads, fibers, fragments, film foam pellets and filaments [22, 23]. They are omnipresent leading to contamination of diverse global ecosystems from the deep ocean trenches to land-based waterways and adjacent sediments [24–27]. Studies have also reported the presence of microplastics inside the aquatic organisms such as fishes [28, 29] and other invertebrates such as in bivalves - green mussel [30], edible oysters [31], great clam [30], crustaceans - white shrimps [32], mud crabs [33], cephalopods - squid [34] and even in zooplankton [35]. Ingestion of these microplastics (MP) can lead to significant health risks, causing gut blockages, oxidative stress, impaired growth, genotoxicity behavioral alterations and reproductive issues [36–38]. Moreover, microplastics act as a carrier, accumulating persistent organic pollutants, hydrophobic chemicals and heavy metals, amplifying the toxicity threats to ingesting organisms [39–41].

Plastic Overshoot Day Report for 2023 by EA-Environmental Action indicates that India is estimated to have released around 330,000 tons of microplastic and 44,000 tons of chemical pollutants into its waterways by the end of the year [42]. This poses a significant threat to the health of aquatic ecosystems and potentially to human health as well. The complete measurement of microplastic contamination and its resulting effects in India have not yet been entirely determined. Hence, regular monitoring of microplastic contamination is crucial.

This study investigates microplastic ingestion by two commonly consumed brackish water organisms, catfish (*Arius* sp.) and mud crab (*Scylla olivacea*) sourced from the Ennore creek and the two commercial important fishes Japanese threadfin bream (*Nemipterus japonicus*) and Indian Mackerel (*Rastrelliger kanagurta*) sourced from Kasimedu landing center of Tamil Nadu. The proximity of the study area to plastic industries, fishing activities, industrial discharges and domestic/urban waste inputs provides context for the potential sources of microplastics in that environment. Investigating microplastic ingestion in these specific locations can help identify hotspots and contribute to the understanding of regional microplastic pollution patterns. The species that were chosen to investigate microplastic ingestion in two commonly consumed brackish water organisms, catfish (*Arius* sp.) and mud crab (*Scylla olivacea*) from Ennore. This is relevant as these species are important for the local food supply and economy and understanding their microplastic exposure is valuable. Additionally, the two commercially important marine fish species, Japanese threadfin bream (*Nemipterus japonicus*) and Indian mackerel (*Rastrelliger kanagurta*), are appropriate for understanding microplastic contamination in seafood. Examining a range of species from different trophic levels and habitats (brackish and marine) provides a more comprehensive understanding of microplastic pollution in the local aquatic ecosystem. Hence this study aims to explore and compare the abundance of microplastics in these distinct ecosystems, shedding light on potential variations in microplastic contamination based on habitat and species characteristics.

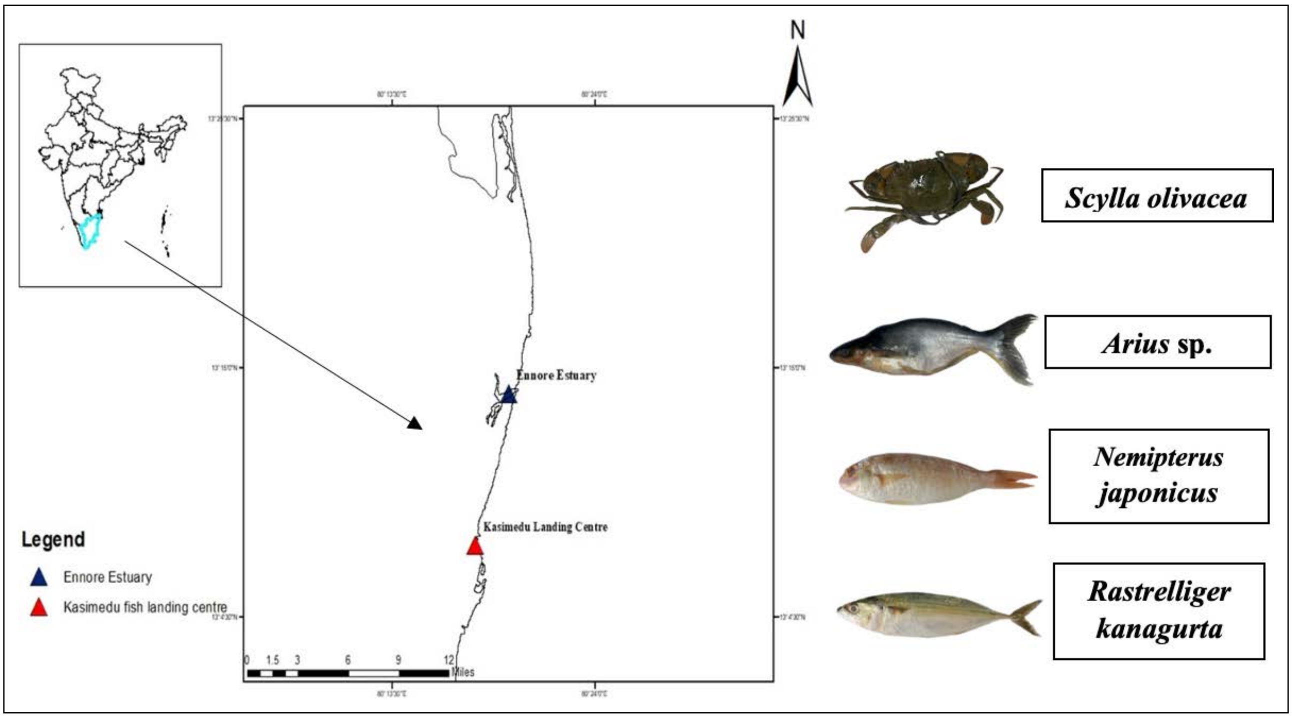

Figure 1. Indicates the sampling sites: Ennore Creek (13° 15′ 26.28″ N, 80° 20′ 16.08″ E) and Kasimedu landing centre (13° 07′ 42″ N, 80° 17′ 38″ E).

Figure 2. Ennore creek.

Figure 3. Kasimedu fish landing centre.

MATERIALS AND METHOD

Study Area

The present study focuses on the geographical coordinates of Ennore Creek (13° 15′ 26.28″ N, 80° 20′ 16.08″ E) and Kasimedu landing centre (13° 07′ 42″ N, 80° 17′ 38″ E) (Fig. 1). Ennore creek, located in Ennore, Chennai, along the Coromandel Coast of the Bay of Bengal, is identified as a backwater with distinct features such as lagoons, marshes, and submerged areas during high tide. This water body serves as a tidal arm connecting to the Bay of Bengal through its outlet at the creek. Shanthi and Gajendran [43] worked on water pollution at the Ennore creek stated that the major industrial installations that produce fertilizers, agrochemicals and petroleum were situated closer to the Ennore belt, making the estuary vulnerable and prone to contamination. Kasimedu, considered as one of the largest fish landing centre of the Chennai coast. It comprises of about 457 trawlers, engaging in fishing activities daily. The fishermen engage in both deep-sea trawling as well as inshore fishing, there by Kasimedu serves as a major hub for export of marine products from India [44]. Stretched along the shoreline for approximately 2 km, North of Chennai Port, Kasimedu plays a pivotal role in supporting the local fishing community.

Ennore Creek has been designated as the first sampling site (Fig. 2) for this microplastic study, focusing on two species: Catfish (*Arius* sp.) and mud crab (*Scylla olivacea*). In contrast, Kasimedu landing centre is identified as the second sampling site (Fig. 3), where the species under investigation are the Japanese threadfin bream and Indian mackerel. The rationale behind selecting these specific locations lies in the diverse ecosystems they represent. Ennore Creek, being a brackish water environment, serves as a habitat for organisms like Catfish and mud crab, which are not only indigenous to the region but also widely consumed by the local population. On the other hand, Kasimedu features marine habitat conditions, and the species chosen, Japanese threadfin bream and Indian mackerel, are commercially significant and readily available in the area.

Collection, Processing, and Quantification of Microplastics

Wild caught, fresh specimens were directly collected from the fishermen at the study areas mentioned above. The collected samples were placed in the icebox, to avoid any sort of damage or contamination. Then they were transferred to the laboratory, for the next step. Each specimen underwent individual measurements for length and weight, and photographs were taken. The gastro-intestinal (GI) tracts and gills of fishes, as well as the hepatopancreas and gills of crabs were isolated, weighed and stored separately in a sterile aluminum foil foam bag at -20°C.

After storage, systematic thawing done by placing the samples in a sealed metal container. Temperatures were monitored hourly using probe thermometer until the samples came to a room temperature. For fishes, the tracts and gills were homogenized separately, while for crabs, both hepatopancreas and gills were homogenized together using an autoclaved mortar and pestle. Alkaline digestion, involving approximately three times the weight/volume of 10% aqueous KOH (potassium hydroxide), was applied to the homogenized organic material, maintaining the solution at 60°C for 72 hours [45–47]. The resulting material underwent filtration through two sets of filters with mesh sizes of 1 mm and 125 µm (Whatman No. 1) using distilled water. They were inspected for the presence of microplastics under a Unilab binocular research microscope and suspected particles were isolated for further analysis.

Precaution and Safety Measures

To minimize microplastic contamination throughout the process, precaution measures were implemented as mentioned by Kumar et al. [46]. The dissection area was maintained in a clean environment to minimize airborne microplastics with a dedicated lab. All the instruments that were used during the process were properly sterilized. A dedicated, stainless-steel dissection kit was used to avoid potential microplastic shedding from plastic tools. Dissections were performed using double-distilled water to minimize any trace contaminants that might interfere with microplastic analysis. To minimize plastic usage, non-plastic containers and tools were used whenever possible and to avoid potential contamination from microplastic leaching. Rigorous cleaning was done. All the equipment used during the entire process, including containers and instruments, was thoroughly cleaned with filtered water followed by laboratory-grade ethanol to remove any residual tissue and potential microplastic contaminants.

Characterization and Identification of Microplastics

Microplastic identification initiates with a thorough visual inspection, enabling the initial recognition of particles through assessments of size, shape and color. This qualitative evaluation acts as an initial screening technique, facilitating the differentiation of potential microplastics from organic substances. After the primary confirmation test, the isolated microplastic particles underwent imaging procedures and their respective diameters were measured. Then the isolated suspected plastic particles underwent FTIR - ATR (Fourier Transform Infrared Spectroscopy - Attenuated Total Reflectance) spectrophotometer analysis.

FTIR Analysis

Obtained plastic samples from the different species were placed in the ATR window. For each sample, multiple ATR-FTIR measurements were taken at different points to ensure to capture data from as many particles as possible, including smaller ones.

ATR-FTIR spectra of samples were recorded at 4000–500 cm^{-1}. FTIR spectra were obtained using the Perkin Elmer spectrum ATR-FTIR spectrometer available at the Sophisticated Analytical Instrumentation Facility at St. Peter's Institute of Higher Education and Research, Chennai. The instrument was under continuous dry air purge to eliminate atmospheric water vapor. Interferograms were averaged for 32 scans at 4.0 cm^{-1} resolution. Perkin Elmer Spectrum software was used for the frequency measurements. The collected spectra were further analyzed by Origin 8.0 software. Microplastics are characterized by comparing the FTIR results of each functional group of spectra with standard values available from the literature.

Statistical Analyses

Statistical analyses were performed to evaluate the distribution of microplastics among species and to assess relationships between various factors. All statistical tests were conducted using IBM SPSS Statistics 27. Descriptive statistics (mean±standard deviation) were calculated for the length and weight measurements of each species. One-way Analysis of Variance (ANOVA) was used to test for significant differences in the number of microplastics found among the four species studied, with a significance level set at $p < 0.05$. To examine the relationship between microplastic abundance in gills and guts, Pearson correlation analysis was performed. The Pearson correlation coefficient (r) was calculated to determine the strength and direction of the linear relationship between these two variables. All graphs and figures were created using Microsoft Excel.

RESULTS

Abundance of Microplastics in Fish and Crab Organs

In this study 70 samples from four commercially important species (Japanese threadfin bream, Indian Mackerel, Catfish and Mud Crab) were investigated. The Japanese threadfin bream had an average length of 15.7±17.7 cm

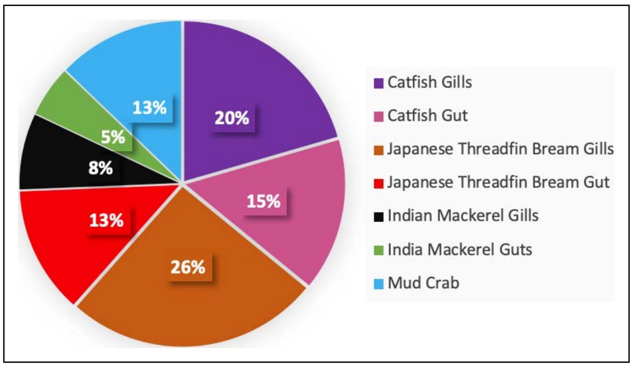

Figure 4. Overall distribution of microplastics among four species.

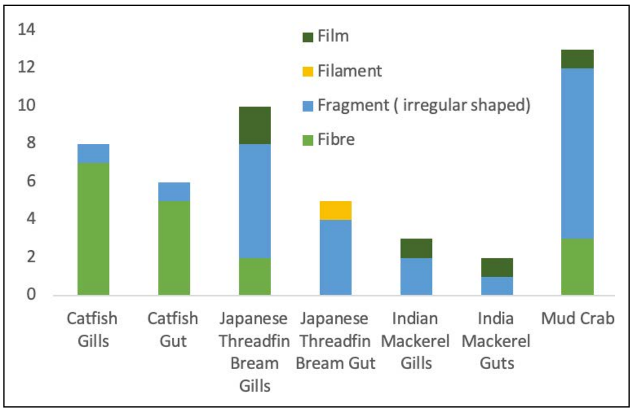

Figure 6. Different shapes of microplastics and their distribution among four species.

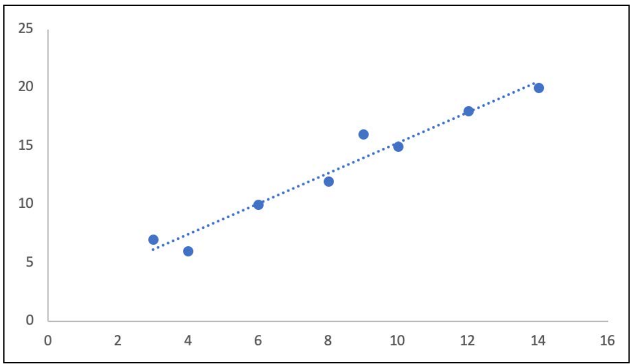

Figure 5. Correlation of microplastic abundance between gills and guts where x- axis depicts the microplastic abundance in the guts and the y-axis depicts the microplastic abundance in the gills.

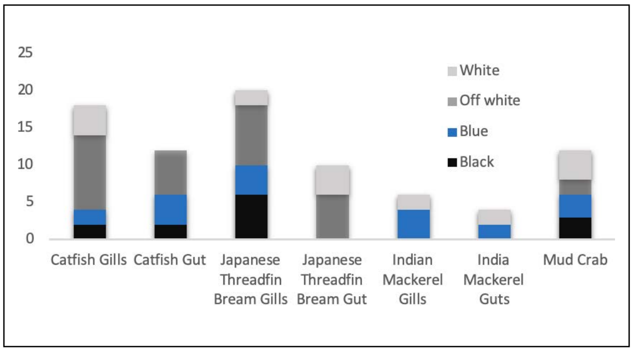

Figure 7. Different colors of microplastics and their distribution among four species.

and a gut weight ranging from 4 to 9 g. Indian Mackerel exhibited an average length of 19±21 cm with a gut weight ranging from 5 to 10 g. Catfish displayed an average length of 17±20.5 cm and a gut weight ranging from 4 to 10 g. The mud crab showed a carapace width of 10±13 cm and a hepatopancreas weight ranging from 13 to 16 g. Microplastics were detected in 55 (78.57%) samples, highlighting their widespread occurrence in these species. There were species-specific variations in distribution of microplastics as *Nemipterus japonicus* and *Arius* sp. had the highest average of 5±3 microplastic items/individual and of 4±2.5 microplastic items/individual respectively. While *Rastrelliger kanagurta* and *Scylla olivacea* showed lower averages of 2±1 microplastic items/individual each. A distinct organ-specific trends were noticed, with gills harboring slightly more microplastics (0.35 items/gills) than guts (0.21 items/gut). Statistical analysis using one-way ANOVA (Analysis of Variance) indicated a significant difference ($p<0.05$) in the number of microplastics found among the four species studied (Fig. 4).

Pearson correlation analysis of the relationship between microplastic abundance in the gills and microplastic abundance in the guts of the studied species reveals a very strong positive correlation. A Pearson correlation coefficient (r) of 0.978 was calculated, indicating a highly linear relationship between these two variables. These results demonstrate that as the microplastic levels increase in the gills, they tend to increase correspondingly in the guts and vice versa, suggesting a strong link between the accumulation of microplastics in these two organs within the studied population (Fig. 5).

Characteristics of Microplastics in Fishes and Crab

The morphological characteristics of microplastics in four different species were studied. Fibers and fragments were the predominant microplastic shapes observed in all four organisms, with thin filaments and film following. Catfish and Japanese threadfin bream displayed a notable dominance of fibers and fragments. The highest deposition occurred in gills, with catfish gills having the highest fiber concentration and Japanese threadfin bream gills and mud crab's hepatopancreas containing the highest fragment concentration (Fig. 6). Notably, microplastics in guts were generally larger than those in gills and the average size across all species ranged from 700 µm to 1 mm.

Microplastics were detected in four colors: black, blue, white and off-white (translucent). The most common color was off-white (translucent) followed by white, blue and black. White microplastics were the most dominant colour present in the mud crab's hepatopancreas. Translucent microplastics were notably concentrated in the gills of catfish and Japanese threadfin bream. In the gut of catfish, the prevailing color was blue, while black color was observed in higher proportions only in Japanese threadfin bream gills (Fig. 7).

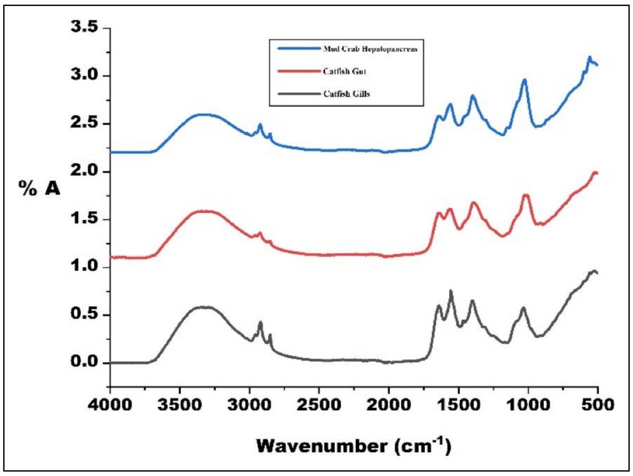

Figure 8. FT-IR spectra of catfish gill, catfish gut and mud crab hepatopancreas (Recorded in the region 4000–500 cm^{-1}).

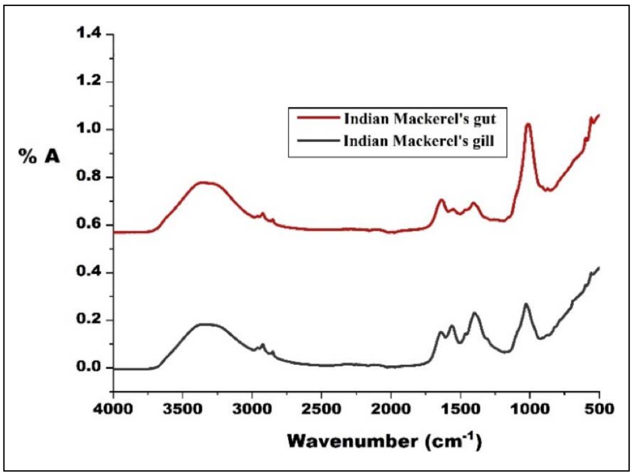

Figure 9. FT-IR spectra of Indian Mackerel gill and gut (Recorded in the region 4000–500 cm^{-1}).

FTIR Frequency Assignment of Microplastics Extracted From Different Species

Figures 8–10 shows average FTIR spectra of extracted microplastics from the gill and gut of the respective species. The spectra were recorded in the region 4000–500 cm^{-1}. Table 1 shows the tentative frequency assignment of the microplastics obtained from gill, gut of Indian mackerel, Japanese threadfin bream, catfish and crab hepatopancreas respectively. As observed from the figure and table a very weak peak ~2915 cm^{-1} corresponds to asymmetric C-H stretching occurs for the samples studied. A weak intensity 2920–2840 cm^{-1} observed corresponds to asymmetric C-H stretching of low-density polyethylene (PE). The gill and gut of the catfish have a strong and medium peak ~2915 cm^{-1} observed and for the rest of the samples, weak peaks were observed. Symmetric C-H stretching corresponds to ~2850 cm^{-1} occurs for the sample except a strong band occurs for the gill of the catfish as seen in the Table 1 and Figure 6. C=O stretching of weak to very weak appears ~ 1653 cm^{-1} and NH bend and C-N stretching frequency assigned to ~1532 cm^{-1} as observed in the samples. NH bend and C-N stretching of low-density polyethylene occurs in the range of 1530–1560 cm^{-1}. A medium to weak peak of ~ 1462 cm^{-1} occurs at the most of samples corresponding to CH_2 bending. CH_3 bending of PE occurs at 1390–1400 cm^{-1}. Our study supports the other findings showing these peaks observed are characterized as polyamide (nylon) [48–51].

A strong band of ~1015 cm^{-1} corresponds to C-O stretching polyethylene occurs at the gut of Japanese threadfin bream and weak peaks were observed in the gut of catfish were observed (Fig. 8, 10). The peaks occur in the region of 1124–1060 cm^{-1} related to photo-aging polyethylene. C-O stretch polyethylene occurs in the region 1010–1080 cm^{-1} which are the characteristics of microplastics observed in the study. The peaks ~780 cm^{-1} and ~690–710 cm^{-1} corresponds to CH_3 and CH_2 rocking respectively. This band characteristics the low-density polyethylene (LDPE) which matches with the LDPE other researchers [49–52].

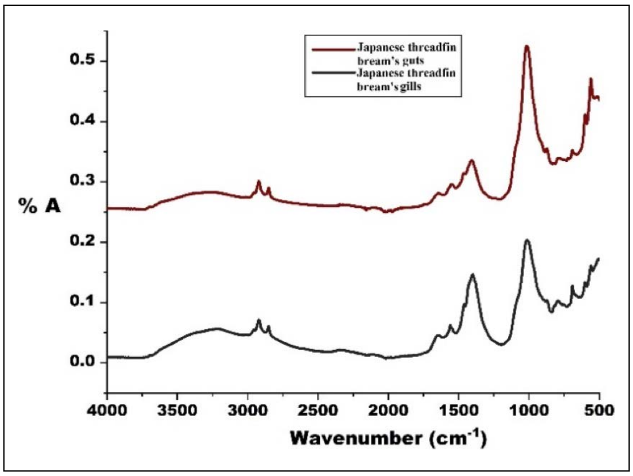

Figure 10. FT-IR spectra of Japanese threadfin bream gills and guts (Recorded in the region 4000–500 cm^{-1})

Polymer Identification

The ATR FTIR analysis identified two microplastic types: Low-density polyethylene (LDPE) and polyamide (nylon). LDPE consistently appeared in all samples where its highest concentration was seen in the gills of Japanese threadfin bream compared to its guts. In contrast, Indian mackerel displayed similar LDPE values in both gut and gill samples, indicating negligible changes in spectral intensity. The observed poor quality of plastics in the mackerel's gut may be linked to effective absorption during digestion. Japanese threadfin bream exhibited the highest LDPE absorption, implying the inefficiency of crab hepatopancreas and gills in LDPE absorption, accompanied by poor plastic quality.

This study advances our comprehension of species-specific digestion and absorption mechanisms in the presence of microplastics, emphasizing the variability among different species. Furthermore, minimal absorption bands characteristic of polyamide (nylon) was discerned in catfish gills and guts, with a significantly higher peak intensities in gills, suggesting preferential accumulation and higher abundance in this tissue. These findings underscore potential

Table 1. FT-IR spectral peaks on gill, gut of Indian Mackerel, Japanese threadfin bream, cat fish and crab hepatopancreas

Frequency assignment	Functional group	Gill of mackerel	Gill of Japanese threadfin bream	Gut of mackerel	Gut of Japanese threadfin bream	Gill of catfish	Gut of catfish	Crab Hepatopancreas
2908	Asymmetric C-H stretching	2915 (vw)	2917 (vw)	–	2925 (vw)	2916 (s)	2920 (m)	2925 (w)
2845	Symmetric C-H stretching	2850 (vw)	2850 (vw)	2856 (vw)	2868 (vw)	2858 (s)	2846 (w)	2842 (w)
	C=O stretching	1653 (vw)	1639 (vw)		1647 (vw)	1645 (vw)	1642 (w)	
						1753 (s)	1749 (m)	1642 (w)
	NH bend and C-N stretching	1532 (vw)	1556 (vw)	–	1537 (vw)	1545 (s)	1557 (m)	1554 (m)
	CH_3 bending					1455 (w)	1454 (w)	
	CH_3 bending						1405 (w)	
1463	CH_3 bending	1408 (w)	1406 (s)	–	1416 (m)	1379 (vw)	1378 (vw)	1394 (s)
	Related to photo-aging polyethylene (1124–1060 cm^{-1})	1092 (s)				1158 (vw)	1154 (vw)	
	C-O stretching polyethylene				1015 (S)		1092 (w)	
	C-O stretching			1083 (vw)		1042 (vw)	1045 (s)	
	C-O stretch polyethylene		1012 (S)			1022 (m)	1027 (s)	1029 (s)
	C-O stretch		1006 (s)	1081 (s)				
	CH_3 rocking,		794 (w)	787 (m)	784 (m)			
717	CH_2 rocking		693 (m)	693 (vw)	698 (w)	715 (vw)	715 (vw)	

S: Strong; M: Medium; w: Weak: Vw: Very weak.

variations in defense mechanisms and biological functions across species in response to microplastics, providing valuable insights for further research in this field.

Table 1 shows tentative frequency assignment of the four species. A weak intensity 2920–2840 cm^{-1} observed corresponds to asymmetric C-H stretching of low-density polyethylene (PE). The very weak intensity of C=O stretching occurs in the range of 1630–1640 cm^{-1}. NH bend and C-N stretching of low-density polyethylene occurs in the range of 1530–1560 cm^{-1}. CH_3 bending of PE occurs at 1390–1400 cm^{-1}. The peak occurs at 1120–1060 cm^{-1} related to photo-aging polyethylene. The strong to peaks occurs at 1000–1030 cm^{-1} C-O stretching polyethylene. A week peaks 680–780 cm^{-1} CH_3/CH_2 rocking polyethylene (Fig. 8–10).

DISCUSSION

Microplastics pollution is one of the major global phenomena of the current decade. Several studies have been carried out all over the world to detect the presence of microplastics and analyzing them and the impacts they cause on living organisms. They were found accumulated in the gills, gastrointestinal tract and organs of various biota. It consists of plankton with accumulation of 164 particles/m^3 [53], corals consist of 50 µg plastic cm^{-1} h^{-1} [54], marine fishes had 112 pieces of microplastics on an average [55], mangrove fishes were found to have 0.79 to 1.00 particles [56]. So, understanding the microplastics abundance is essential.

Abundance of Microplastics in Fish and Crab Organs

This study highlights the concerning prevalence of microplastics in commercially important aquatic species with detection in 78.57% of samples across four species. This widespread occurrence demands closer inspection, especially considering the observed species-specific variations and organ-specific distribution. These findings raise critical questions about the potential ecological and human health implications of microplastic pollution. The study identified *Nemipterus japonicus* (Japanese threadfin bream) harboring 5±3 microplastic items/individual and *Arius* sp. (catfish) containing 4±2.5 microplastic items/individual, showing that they have the highest microplastic accumulation rate comparatively. This may likely due to their benthic feeding habit [57] near microplastic-laden sediments. *Rastrelliger kanagurta* (Indian mackerel) and *Scylla olivacea* (mud crab) showed lower microplastic averages of 2±1 microplastic items/individual each, potentially due to their pelagic or intertidal habitats. This trend of higher abundance of microplastics were also seen in similar recent studies such as in Japanese threadfin bream (*Nemipterus japonicus*) had 28.33±8.11 particles/individual out of 9 samples, Indian Mackerel (*Rastrelliger kanagurta*) had 20.89±8.79 particles/individual out of 9 samples [58], Catfish (*Arius* sp.) showed 1.77±0.25 particles/individual out of 35 samples [59] and several other crab species also showed the presence of microplastics, emphasizing the organisms nature of microplastic consumption. However, further research considering factors like feeding ecology, prey preference and microplastic characteristics is needed [60]. Furthermore, the study revealed organ-specific distribution of microplastics. The higher microplastic presence in gills (0.35 items/gills) compared to guts (0.21 items/gut) suggests potential direct filtration and accumulation in gill structures. This aligns with previous studies reporting gills as primary accumulation sites [23]. Understanding the potential physiological impacts and translocation pathways within organisms is crucial to fully understand its effect [61]. The significant difference in microplastic distribution among species ($p<0.05$) underscores the critical need for species-specific assessments and targeted management strategies. Ignoring these variations could lead to underestimating risks for vulnerable populations and hindering effective mitigation efforts.

Sources of Microplastics

The sources of microplastics in Ennore creek and Kasimedu fish landing center are multifaceted, reflecting the heavily industrialized and urbanized nature of the region. Ennore creek, a polluted industrial estuary in Chennai, Tamil Nadu, encounters microplastic inputs from wastewater discharge by nearby industrial units and residential areas [47], runoff from roads and urban areas carrying plastic litter [62], atmospheric deposition of airborne microplastics [63], as well as fishing activities and lost fishing gear [64]. Kasimedu, the largest fish landing center in Chennai located along Ennore creek, faces microplastic pollution from discarded and lost fishing nets, lines, ropes and other gear, microplastics entering the food chain from the contaminated creek waters [65], improper waste disposal by fish vendors and workers [66] and urban runoff carrying plastic waste [62]. The high levels of industrialization, urbanization and fishing activities in the region are significant contributors to the microplastic contamination of these aquatic ecosystems [67], highlighting the need for effective waste management and better regulation of industrial effluents to address this growing environmental issue.

Characteristics of Microplastics in Fishes and Crab

The microplastics observed in four aquatic species, revealed intriguing patterns in their shapes, sizes and colors. The origin of different microplastic sizes in the samples were due to several factors, in ocean plastic particles deteriorate according to its power function resulting in different size range. Environmental degradation plays a vital role as the larger plastics break down into smaller pieces due to UV radiation, mechanical abrasion, and biological processes [14]. Different environmental conditions (e.g., temperature, salinity) affect the rate and extent of plastic fragmentation [68]. Several studies have classified microplastics based on its size such as Gad et al. [69], worked on microplastic extraction from two commercialized fishes, hardhead catfish (*Ariopsis felis*) and southern flounder (*Paralichthys lethostigma*) of the Gulf of Mexico. The microplastics were categorized into three different categories based on different sizes as small (20–1000 μm), medium (1000–2500 μm), and largest (2500 μm). Ranjani et al. [70], extracted microplastics from two major harbours, the Ennore and Chennai harbour. These microplastics were classified into five categories based on their size range. Ineyathendral et al. [71], collected four major commercial fishes *Sphyraena jello*, *Nemipterus japonicus*, *Leiognathus species*, *Sardinella longiceps* of the Chennai Coast. Each fish had different feeding habits which played a major role in microplastic accumulation in the gut region. A total of 607 microplastic pieces were observed within size range 0.1 mm to 5 mm.

Fibers and fragments were the two most dominant shapes found in all the four organisms; this is likely due to their abundance in the environment from larger plastic breakdown [20]. Interestingly, catfish and Japanese threadfin bream seem particularly susceptible to these shapes, potentially reflecting their feeding habits or habitat characteristics [60]. With larger microplastics (>700 μm) residing mainly in the guts, suggests potential ingestion and incomplete digestion. In contrast, gills harbor smaller ones, possibly due to direct filtration or passive adsorption [23]. This highlights the varying uptake pathways and potential impacts depending on the microplastic size. A range of color variations in microplastics were observed. Translucent microplastics seem favored by catfish and Japanese threadfin bream gills, while white microplastics dominates the mud crab's hepatopancreas. The transparent fragments and fibers could be derived from the decomposing of fishing nets/lines according to their features. These results were similar to Beibu Gulf [72] indicating that the plastics products from fishery activities played an essential role in microplastics pollution. These variations could be linked to factors like production processes, biofouling or selective accumulation mechanisms [73]. According to Marti et al. [74], the color of the plastic pieces fade over time when they are exposed to sunlight for long periods of time. Photo oxidative damage resulted in fragmentation of plastic pieces and color changes to white and yellow to brown color in over period. These factors affect both the mechanical properties as well as color of the plastics pieces. Additionally, blue dominates catfish guts, potentially reflecting specific prey preferences or environmental exposure. Blue could be sourced back to the fishing nets. The scarcity of black microplastics suggests lower environmental presence or different uptake pathways. Investigating the potential impacts of different shapes, sizes and colors on these species is crucial to assess the full scope of microplastic threats [75].

Polymer Identification

The ATR-FTIR analysis conducted identified two primary types of plastics: low-density polyethylene (LDPE) and polyamide (nylon). LDPE was consistently found in all four samples, indicating its widespread presence. The potential sources of LDPE include plastic bags and packaging, fishing gear and single-use plastics. On the other hand, nylon was also detected and its possible sources include fishing gear, textiles and clothing, wastewater, various industrial and commercial applications. The distinct LDPE concentrations in Japanese threadfin bream (gills > gut) vs. mackerel (gut ≈ gills) highlight species-specific variations in digestion process to breakdown poor - quality plastic and absorption mechanisms [76]. Higher LDPE concentration in Japanese threadfin bream gills than guts suggest inefficient absorption, potentially due to differences in gill morphology or filtration mechanisms impacting particle capture [20] and variations in digestive enzymes or gut microbiota composition affecting breakdown [73]. Similar LDPE levels in mackerel gut and gills imply efficient absorption, possibly linked to gut morphology or enzymes facilitating LDPE uptake [77]. Interestingly, crab hepatopancreas and gills exhibited minimal LDPE absorption, suggesting species-specific defense mechanisms or digestive processes. These findings highlight the need for further research into the factors influencing LDPE absorption and its potential ecological impacts.

Polyamide, commonly known as nylon had minimal absorption bands in catfish gills and guts and significantly higher intensities were observed in gills. This suggests preferential accumulation of nylon in gills due to their filtration function or specific binding mechanisms [61]. Further research is needed to understand the implications of polyamide accumulation in gills and its effects on fish health as highlighted in the study. This study insights are in line with previous research highlighting the intricate interplay between microplastics and aquatic organisms' physiological processes [61]. Future research needed in understanding the digestive and absorption mechanisms employed by different species, exploring factors like gut morphology, enzymatic activity and plastic characteristics [18].

Potential Risks to the Studied Species and Human Consumption

The widespread presence of microplastics observed in this study raises concerns about the potential health impacts on the targeted marine species and the humans who consume them. Microplastic ingestion has been shown to have physiological and ecological consequences, such as impaired growth, reduced reproductive fitness and disruption of overall organismal function, in similar marine organisms [60]. Given the high levels of microplastic accumulation found in species like *Nemipterus japonicus* and *Arius* sp., further research is needed to elucidate the specific health impacts on these species and the implications for their population dynamics and the broader ecosystem. Additionally, the presence of microplastics in the studied species poses a potential risk to human consumers, as these contaminants may bioaccumulate and transfer up the food chain. Existing literature suggests that microplastic ingestion in humans can lead to gastrointestinal, immunological and endocrine-disrupting effects [23, 77], highlighting the importance of monitoring microplastic levels in seafood and establishing safe consumption guidelines.

Limitation of the Study

The study is limited to the accumulation of MPs in specific species. Broader ecological and health impacts of MPs, particularly within the framework of the One Health approach, have not been addressed. This represents a significant research gap that needs to be explored in future studies. The MPs were separated into size classes and concentrated ATR-FTIR analysis were done only on the larger fractions (>500 μm), where the technique is most effective. The findings are based on samples collected within a specific time frame and geographic area. MP pollution can vary significantly over time and across different locations, so the results may not be generalizable to other regions or time periods. Longitudinal studies are needed to understand the chronic impacts of MP exposure.

CONCLUSION

The study investigated the presence and characteristics of microplastics in four commercially important aquatic species - Japanese threadfin bream, Indian mackerel, catfish, and mud crab from two locations in Tamil Nadu, India. Microplastics were found in 78.57% of samples, indicating their widespread presence. Significant species-specific variations in accumulation of microplastics were observed with *Nemipterus japonicus* and *Arius* sp. showing the highest levels of microplastic deposition which is likely influenced by their feeding habits. Gills exhibited slightly more microplastics than guts, implying direct filtration and accumulation. Fiber and fragment shapes were the most prevalent microplastic shapes, reflecting their environmental abundance, while the most common microplastic colors were off-white (translucent), white, blue and black. The determination of microplastic colors and their origin is a complex issue because these colors may not necessarily reflect the original color of the plastic products. The colors that were observed were not bright but merely faded due to possible several factors such as photo oxidation, weathering, additive leaching etc. that can influence the observed colors. Larger microplastics were in guts, while smaller ones were in gills, suggesting different uptake pathways. FTIR analysis identified two main polymer types – low density polyethylene (LDPE) and polyamide (nylon). Species-specific differences were observed in the absorption and accumulation patterns of these polymers. Further research is essential for understanding digestive and absorption mechanisms, potential health impacts and population dynamics. The results of this study highlight the need for targeted, species-specific assessments and management strategies to address the growing issue of microplastic pollution in the studied ecosystems. To facilitate immediate action for mitigating microplastic pollution in the studied areas, an establishment of comprehensive monitoring programs are recommended, implementing source reduction strategies, enhancing habitat conservation and restoration, developing fisheries management plans that incorporate microplastic contamination as a key consideration and fostering collaborative research to better understand the fate, transport and biological impacts of microplastics in the studied ecosystems. By implementing these targeted actions, tailored to the specific ecological and socio-economic contexts of the study regions, proactive steps can be taken to address the microplastic pollution challenge and safeguard the health of the studied marine species and the communities that depend on them.

DATA AVAILABILITY STATEMENT

The author confirm that the data that supports the findings of this study are available within the article. Raw data that support the finding of this study are available from the corresponding author, upon reasonable request.

CONFLICT OF INTEREST

The author declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

USE OF AI FOR WRITING ASSISTANCE

Not declared.

ETHICS

There are no ethical issues with the publication of this manuscript.

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