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Marine microplastic pollution in Muttom: Detection and characterisation

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Abstract

The study examines the techniques for isolating, detecting, and analyzing microplastics (MPs) in marine habitats, specifically in the coastal waters of Muttom, India. Water samples were subjected to density separation filtration and sieving, which identified the presence of microplastics primarily as fibers, pellets, filaments and fragments. Density separation used the buoyancy of microplastics, resulting in high-density MPs settling as pellets while low-density MPs remained suspended in the supernatant. Filtration of isolated MPs yielded approximately 40 mg per 5 liters of seawater. Visual and microscopic identification classified microplastics by size, shape, and color, with fibers being the predominant kind. Scanning Electron Microscopy (SEM) yielded high-resolution images, detecting microplastics as tiny as 500 nm and exposing rough, porous, and flaky surfaces. FT-Raman spectroscopy verified the existence of polymers, specifically polystyrene and polycarbonate, through distinctive vibrational frequencies. The results highlight the dangers that microplastics provide to marine ecosystems and human health, stressing the need for efficient waste management and legislative actions to alleviate microplastic pollution.

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Introduction

The presence of microplastics in marine environments and its detrimental impact on biota and ecosystem functionality has been a significant focus of global study in recent decades. The extensive presence of plastic in marine environments is particularly significant when considering small-sized plastics, defined as microplastics (MPs), which range from 1 µm to 5 mm (Gago *et al.*, 2018).

Microplastics often aggregate at the water column's surface, although they are also vertically transferred to the seabed via several processes (Wang *et al.*, 2016). Plastic introduced into the marine ecosystem may originate from terrestrial sources (e.g., home or personal care goods that enter the sea primarily via waste treatment) or marine sources (e.g., nylon fishing nets) (Duis and Coors, 2016).

The problem with microplastics, similar to other plastics, is their durability in the environment. Plastics need an exceedingly prolonged period to disintegrate, often spanning hundreds or thousands of years, during which they may adversely affect ecosystems (Karbalaei *et al.*, 2018). Microplastics are diminutive in size and may originate from several sources, including the breakdown of bigger plastic materials.

The beaches are the most probable locations for the production of microplastics in the marine ecosystem. Plastics exposed to sunshine and air on beaches is more susceptible to breakdown, resulting in the creation of smaller particles. Furthermore, coastal activities, including fishing techniques, aquatic tourism, and marine businesses, augment the prevalence of microplastics in the marine environment.

Microplastics are observable on beaches as tiny, colorful plastic particles inside the sand. Microplastic contamination is often consumed by marine organisms in the seas (Rist *et al.*, 2018). Microplastics have been detected in several marine creatures, ranging from small plankton to massive whales. This makes microplastics a troubling and intricate environmental issue.

Moreover, there are deliberately engineered minuscule plastic particles referred to as "microbeads," frequently utilized in various health and beauty products (Cincinelli *et al.*, 2017). Microbeads are included in products such as face cleansers and toothpaste to provide exfoliating or abrasive properties. A troubling aspect of microbeads is their diminutive size, which allows them to circumvent water filtration systems and be released into aquatic environments, ultimately reaching the ocean. A significant environmental issue associated with microplastics, such as microbeads, is that aquatic organisms and avian species may erroneously identify these minuscule particles as sustenance (Courtene-Jones *et al.*, 2017).

Ingestion of microplastics may adversely affect marine habitats and creatures. Initiatives have been undertaken to limit or prohibit the use of microbeads in personal care products throughout many locations to alleviate their environmental consequences and diminish the prevalence of microplastics in aquatic environments.

The domain of microplastics study is very nascent, and several aspects about their effects remain unidentified (Masura *et al.*, 2015). Ultimately, these field and laboratory techniques will provide worldwide comparisons about the volume of microplastics entering the environment. The cumulative effects of microplastics on aquatic ecosystems and species constitute an escalating environmental issue, underscoring the need to mitigate plastic pollution and curtail the discharge of microplastics into the Muttom Coast.

Materials and methods

Study area

Muttom is a coastal village located in the Kanyakumari district of Tamil Nadu, India. It lies at latitude 8.1364°N and longitude 77.3122°E, approximately 200 meters (660 feet) above sea level (Fig. 1). Known for its scenic beauty, Muttom is characterized by its rocky shoreline, black sand beaches and unique geological formations. The region is part of the southernmost tip of the Indian subcontinent, where the Western Ghats meet the Arabian Sea. Muttom is not only a popular tourist destination but also a vital fishing hub for the local community, with fishing being a primary livelihood. The coastal region is rich in mineral deposits, particularly in the black sand that lines the beaches. The area experiences a tropical climate with annual rainfall averaging around 1,450 millimeters (57 inches), primarily during the monsoon season.

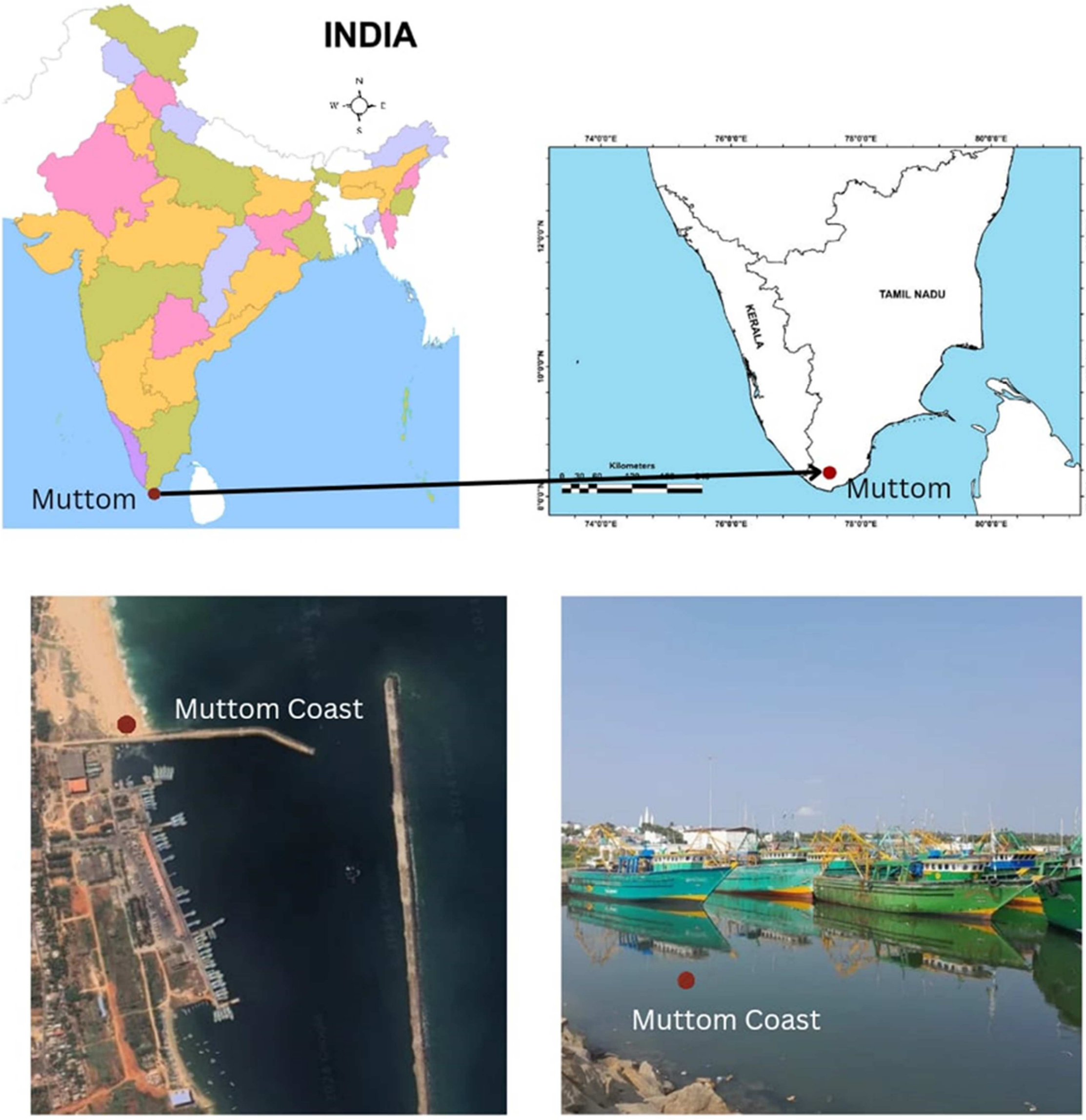

Fig. 1. Map showing the location of Muttom coast

Temperatures range between 20°C and 35°C (68°F to 95°F), offering a moderate coastal climate throughout the year. The fishing season in Muttom typically runs from June to March, while fishing activities are restricted during April and May to allow fish populations to reproduce. This regulation helps sustain marine biodiversity and supports the livelihoods of the local fishing community. Muttom's ecological significance, coupled with its distinctive geographical features, makes it an ideal region for studying the environmental impact of microplastics in coastal waters, which is the focus of this research. The area's natural and economic importance highlights the need for preserving its marine ecosystem.

Analysis of microplastics

The analysis of microplastics typically involves several methods and materials for sampling, extraction and identification.

Sampling procedure

Five litres of water sample were collected from the Muttom coast in plastic bottles to distinguish, recognize, and describe the microplastic in the provided seawater sample. Sieving, density separation, and filtration are three categories under which separation techniques can be grouped.

Preparation of the collected sample

Isolation of microplastic by sieving method

The first step in sample preparation is a visual sorting process to remove any visible microplastic particles larger than 5 mm. Following this, the isolation of microplastics from the provided seawater sample was executed using a sieving technique. Five litres of seawater passed through a 5 mm metal sieve to exclude larger microplastics and retain particles less than 5 mm in size. Subsequently, the sieved water sample was subjected to a density separation process for further analysis.

Isolation of microplastics by density separation method

The collected sieved sample was centrifuged at 3000 rpm for 10 minutes to separate high- and low-density particles. After the centrifugation, the supernatant and pellet were collected.

The pellet was washed thoroughly with distilled water to remove the salt content. After that, the collected pellet was dried in a hot air oven at 40°C. The dried pellet was viewed under an inverted light microscope for microplastic contents, and the collected supernatant sample was used for further filtration.

Isolation of micro plastics by filtration method

The microplastics were separated using the filtration method. The collected supernatant sample was filtered using a sterile membrane filtration apparatus equipped with cellulose nitrate membrane filters of 0.2 µm pore size. A suction pump was used to facilitate this process. Following filtration, the filter paper was thoroughly rinsed with distilled water to prevent the formation of salt crystals on the dried filters. The particles retained on the filter paper, along with the filter itself, were placed in a petri dish and dried in a hot air oven set at 40°C. The dried filter paper was subsequently examined for the presence of microplastics under an inverted light microscope and weighed to determine the mass of microplastics, paving the way for further analyses.

Light microscopic observation

The dried filter was taken for microscopic study, which observes at 10X magnification. Physical methods were firstly applied for fast characterization, and photomicrographs were taken. The suspected particles were analyzed with SEM and FT-Raman spectroscopy for further analysis.

Sample preparation in SEM analysis

After the filtration, the microplastics collected on the filter paper were subjected to SEM analysis. A certain portion of filter paper was loaded on a stub provided for SEM analysis. The stub is made of copper; in the shape of a small cylinder about the size of 1 mm in diameter, one side of the stub was stuck with double-sided carbon material.

After loading the sample on the carbon material, the stub was fixed to a holder. The holder accommodates about 4 samples at a time for 10 minutes twice, and then they were collected for further characterization. The images of microplastics were obtained in the scanning electron microscope (FEI-Quanta FEG 200F Secondary Electron Microscope, Indian Institute of Technology, Chennai).

Identification of microplastics in FT-Raman spectroscopy

Raman spectroscopy is based on Raman effective measurement of the wavelength and intensity of inelastically scattered light from molecules or crystal lattices. The energies of molecular vibrations shift the wavelengths of scattered light from the incident light. Polystyrene and naphthalene standards are regularly used for testing signal-to-noise ratio and wavenumber accuracy.

Results and discussion

Laboratory methodologies for MP analysis

Microplastic samples require one or more separation techniques to extract the microplastic from seawater. According to Hidalgo Ruz *et al.* (2012), the following are the categories that may be used to categorize separation methods: density separation, filtering, and sieving systems. Samples can be visually sorted before filtration or sieving to eliminate microplastics larger than 5 mm, which are discernible to the naked eye (Maes *et al.* 2017). Alternatively, this sorting can occur after sieving by discarding the microplastics that remain on the sieve (Viršek *et al.*, 2016).

Isolation of microplastics

Density separation

The density separation method plays a vital role in the precise evaluation of microplastic composition within marine ecosystems, given that density can greatly affect the behaviour, transport, and bioavailability of microplastics (Thompson *et al.*, 2004). The process of density separation effectively isolates microplastics from the remainder of the sample by utilizing the buoyancy characteristics of microplastics in various solutions that possess a higher density than the microplastics themselves. MPs that contain supernatant are gathered and subjected to filtration (Barrows *et al.*, 2017). The microplastics in the current study are progressively separated based on their density. The supernatant contained the low-density microplastics, whereas the high-density microplastics were found in the pellets. Browne *et al.* (2011) conducted a study using density separation to examine microplastic concentrations in different water bodies, demonstrating that substantial amounts of microplastics could be retrieved from both supernatants and pellets. Microplastics with high density typically settle at the bottom or exist as sediment, whereas those with low density tend to remain suspended in the surface water. Various factors, such as their specific gravity (Rocha-Santos and Duarte, 2017), microbial colonization (Harrison *et al.*, 2018), their morphology (Zhao *et al.*, 2015) and water waves (Ballent *et al.*, 2012), influence the buoyancy and sinking of MPs.

Filtration

Various types of filters are employed to isolate MPs from seawater, such as polycarbonate, polyamide, nylon, glass fiber, cellulose acetate, and cellulose nitrate (Enders *et al.*, 2015; Pan *et al.*, 2019). The current investigation utilized a cellulose nitrate filter membrane. The filters employed for MP samples exhibited pore sizes of 0.2 µm. The concentration of dried microplastic particles collected from the Muttom coast was measured at 40 mg per 5 liters. Fig. 2 illustrates the distribution of microplastics collected from the Muttom coast, revealing that fibers constitute 59%, pellets account for 16%, filaments make up 12%, and fragments represent 13% of the total. The Muttom coast, known for its fishing activities and tourism, is experiencing a significant influx of plastic waste into the coastal region. Research conducted by Shahul Hamid *et al.* (2018) indicates that tourism and fishing are significant contributors to the elevated levels of microplastic abundance observed at tourist beaches and fishing ports.

Sieving

The process of sieving facilitates the separation of microplastics from seawater through the utilization of one or more metal sieves. The quantity of sieves employed and their respective mesh sizes are contingent upon the objectives of the sieving process, which may include the selection of a particular microplastic size range, the elimination of certain fractions of microplastics (for instance, plastic particles exceeding 5 mm), or the categorization of microplastics into distinct size classes (Masura *et al.*, 2015).

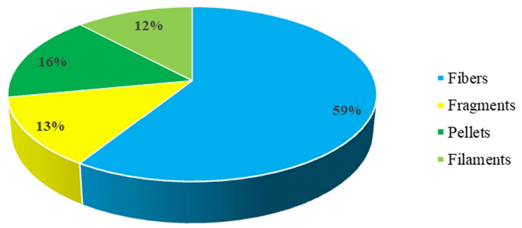

Fig. 2. Percentage of microplastics collected in the Muttom water samples

Sieving can be conducted on the MP sample after it has been isolated from seawater using various techniques, including visual sorting, density separation, or digestion methods (Masura *et al.*, 2015; Kroon *et al.*, 2018).

The sieving process demonstrated the presence of microplastics in the water samples collected from Muttom. The observation of microplastic particles measuring less than 5 mm indicates that larger plastic items have probably broken down into these smaller fragments.

Substantial plastic materials are subject to environmental degradation mechanisms, including photodegradation and mechanical wear, which result in the formation of microplastics (Andrady, 2011). Akhter and Panhwar (2022) emphasized the influence of physical forces, including wave action and sediment abrasion, in hastening the fragmentation of larger plastics into microplastics.

Visual identification

Techniques for visual identification are mainly employed to recognize and categorize MPs. In this study, two distinct microscopes were utilized.

1. Microplastics classification

Microplastics acquired from the Muttom coast underwent microscopic analysis to classify them according to their size, shape, colour and morphology. Generally, the MPs were classified according to their shape into categories such as fibre, foam, pellet, fragment, and film (Ruggero *et al.*, 2020). The morphological characteristics, seen under an inverted light microscope, indicate that the Muttom Coast includes microplastics in the forms of shards, fibers, pellets, and filaments (Fig. 3). The most observed form of microplastics in the Muttom water samples is fibers, consistent with findings from other researchers (Lusher *et al.*, 2013; Neves *et al.*, 2015).

The regular fishing and port operations in the Muttom coastal region may have led to elevated fiber concentrations in the water resources. Fibers are often associated with the deterioration of textiles and synthetic materials, as shown by the study conducted by Napper *et al.* (2022), which observed that laundering synthetic garments is a major contributor to microplastic fibers in aquatic habitats. Wright *et al.* (2013) indicated that fibrous microplastics are mostly found in aquatic environments. The cellulosic fibers produced from this work exhibit vibrant color, suggesting they have undergone industrial processing (Courtene-Jones *et al.*, 2022).

Browne *et al.* (2011) indicated that residential sewage, which encompasses laundry waste, could be a considerable contributor to the presence of synthetic microfibers in marine ecosystems. Moreover, the fishing and shipping activities in the region are well-established, and fishing practices could contribute to the presence of fibers in the marine ecosystem (Cole *et al.*, 2011).

Plastic fishing gear breakdown, domestic wastewater (such as from laundry), and industrial textile manufacturing represent several possible origins of filaments found in surface water (Mistri *et al.*, 2018). Savoca *et al.* (2019) reported the detection of microplastics, particularly fragment types originating from polyvinyl chloride (PVC) and low-density polyethylene (LDPE) materials in the Tyrrhenian Sea, which is an area of the Mediterranean. The shape of the fragments, specifically the surface area to volume ratio, may explain why these particles are able to float on the surface of water. Fragments and filaments are likely to originate from the breakdown of larger plastic items, such as bags and bottles, as well as from pellets, which serve as the raw materials for plastic products (Rocha-Santos and Duarte, 2017).

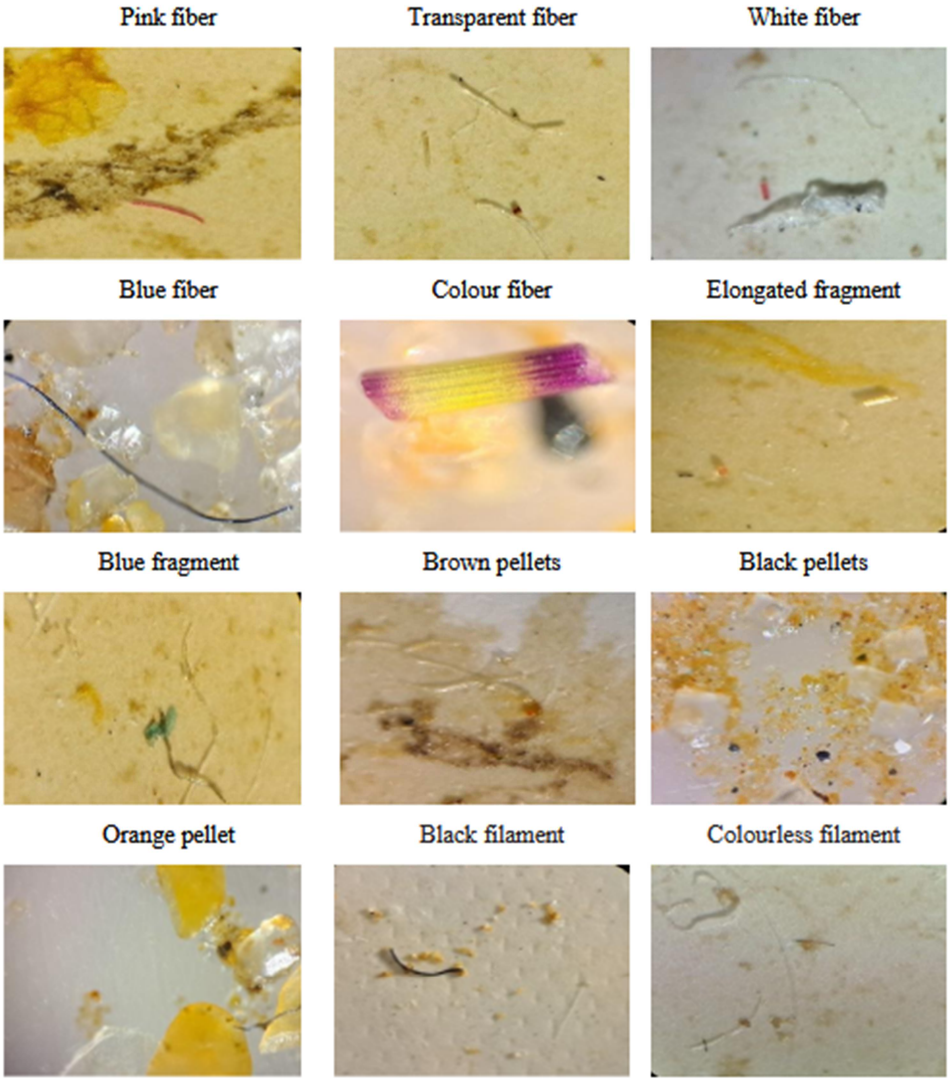

Fig. 3. Various types of microplastics observed in the Muttom coast

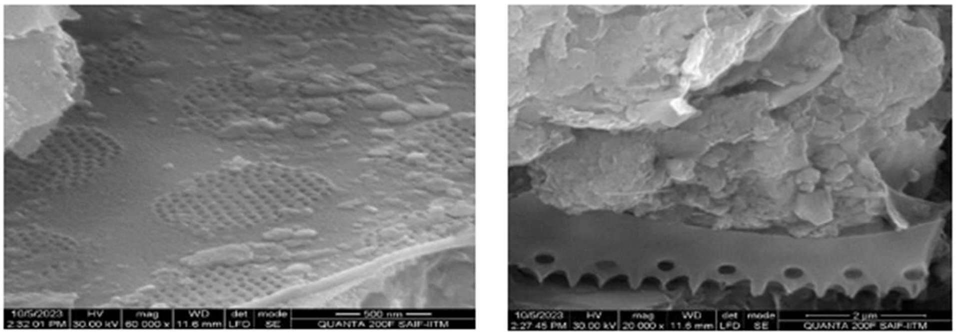

Fig. 4. Scanning microscopic images of microplastics

MP colours are utilized to conduct a preliminary evaluation of the types of chemical compounds present, as well as the origins of MPs (Rocha-Santos and Duarte, 2017). The microplastics found in the Muttom water samples exhibit a variety of colours, including pink fiber, transparent fiber, white fiber, blue fiber, coloured fiber, elongated fragment, blue fragment, brown pellets, black pellets, orange pellets, black filament and colourless filament.

The occurrence of coloured fibers and filaments may stem from human activities, including synthetic textiles like clothing, carpets and ropes, which can enter the wetland via direct surface runoff or rivers (Rasta *et al.*, 2020). Coloured fragments likely stem from the degradation of consumer goods and plastic debris originating from boats. The colour of microplastics influences the marine species that consume them. Species like fish exhibit a lack of discernment when consuming particles that resemble the coloration of their prey (Bessa *et al.,* 2018).

2. Scanning electron microscope (SEM)

Scanning electron microscopy has demonstrated its efficacy in characterizing the morphology and size distribution of microplastics. The examination utilized a scanning electron microscope, renowned for its high resolution, enabling the most accurate identification of MPs (Sagawa *et al.,* 2018). The examination of microplastics in Muttom water samples through Scanning Electron Microscopy (SEM) indicated particle sizes ranging from 500 nm to 2 µm in diameter (Fig. 4). The observed size distribution in Muttom indicates that these microplastics could stem from the breakdown of larger plastic debris, supporting the conclusions drawn by Andrady (2011). Particle size is a critical determinant of the bioavailability and bioaccumulation of microplastics. Organisms can consume microplastics that are comparable in size to their food (Ory *et al.*, 2017). Moreover, the diminutive size of the identified microplastics carries significant consequences for their engagement with marine organisms; for instance, the consumption of particles within the nano- and sub-micron spectrum may result in detrimental impacts on the well-being of marine species (Browne *et al.*, 2011).

Morphological analyses of MPs by SEM imagery displayed rough, porous and flaky surfaces.

In a similar manner, cracked, rough and edged surfaces were noted in the isolated polymeric fragments obtained from water samples of the Caspian Sea (Manbohi *et al.*, 2022). Similarly, research conducted by Zbyszewski revealed that 78% of the microplastic particles examined from the Great Lakes exhibited varying levels of mechanical weathering, characterized by surface scratches (Zbyszewski *et al.*, 2014). The weathering process can lead to roughness and flakiness on microplastic surfaces, potentially increasing the surface area. This change may facilitate higher concentrations of persistent organic pollutants (POPs) adhering to these surfaces (Wardrop *et al.*, 2016).

Analytical identification

FT-Raman spectroscopy analysis

In the realm of spectroscopy techniques, instruments are integrated with an FT-Raman system to facilitate the identification of microplastics with dimensions less than 1 mm. The application of analytical techniques yielded a spectrum corresponding to each particle under examination. Each spectrum was compared with a reference to determine the polymer composition of the particles. Consequently, these methods facilitated a definitive assessment of whether particles are composed of plastic.

The study of microplastics using Fourier Transform Raman (FT-Raman) spectroscopy used to analyze microplastics shows that the atoms' masses and the strength of their atomic bonds affect the vibration frequencies. In particular, Raman shifts are larger for lighter atoms and stronger bonds and lower for heavier atoms and weaker bonds (Chakraborty *et al.*, 2022).

The functional groups were identified based on the varying wavelengths observed in the FT-Raman spectrum data. The analysis indicates the existence of various functional groups, including alkyne ($C\equiv C$), C–H, P-H (aromatic), alkyne ($C=C$), alkanes (CH_3, CH_2), carboxylate acid (C-S) aromatic, carboxylic acid dimer, and C-C aliphatic chains, which are observed in the polymers of microplastics. The spectrum analysis of the Muttom sample indicates the presence of polystyrene and polycarbonate polymers, as shown in Table 1.

Table 1. Characteristic Data of FT- Raman spectrum

Wave number cm^{-1}	Functional group	Bond	Intensity
3288.87	Alkyne	–C≡ C– stretch	Very weak
2910.01	Alkanes	C–H stretch	Strong
2649.59	Aldehyde	H–C=O: C–H stretch	Weak
2416.08	Aromatics	P-H	Very weak
2077.24	Alkyne	C≡ C	Strong
1965.64	-	-	-
1625.45	Aromatics	C–C stretch (in–ring)	Very strong
1441.97	Alkanes	CH_3, CH_2 bend	Weak
1300.26	carboxylic acids, esters, ethers	C–O stretch	Medium
1131.31	aromatic	C-S stretch	very weak
951.64	Carboxylic acid dimer	O–H bend	Weak
632.02	Aliphatic chains	C-C	Medium
469.4	-	-	Strong
101.6	Lattice vibrations	-	Strong

Polystyrene

The spectrum exhibits high-frequency carbon-hydrogen (C-H) vibrations at approximately 3000 cm^{-1}. The vibrations of carbon-carbon (C-C) bonds at low frequencies are around 900 cm^{-1}. The frequency of C-H vibrations surpasses that of C-C vibrations due to the lighter mass of hydrogen compared to carbon. The vibrational frequencies of two carbon atoms connected by double bonds (C=C) occur around 1600 cm^{-1}, whereas the vibrations of two carbon atoms joined by a single bond (C-C) are observed at approximately 900 cm^{-1} (Fig. 5). Polystyrene exhibits unique vibrational properties, featuring high-frequency C-H vibrations close to 3000 cm^{-1} and low-frequency C-C vibrations approximately at 900 cm^{-1}. This pattern reflects the structure and stability of the polymer (Choong *et al.*, 2021).

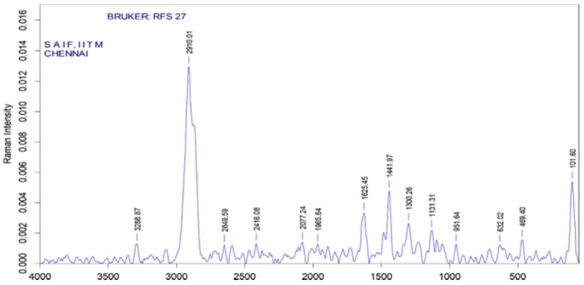

Fig. 5. Analysis of Raman shift and intensity spectrum

Polycarbonate

Polycarbonates are acknowledged as belonging to polyester resins, with various characteristics, and the bands C-C, C=C, and CH_3 carboxyl linkages are recognized as belonging to plastics. The one that is generated from bisphenol A is the most significantly important. According to Choong *et al.* (2021), polycarbonates that are generated from bisphenol A exhibit vibrational bands that are connected with C-C, C=C, and CH_3 carboxyl bonds. This highlights the significance of these compounds in environmental research.

Based on the unique vibrational frequencies of several polymers, Lenz *et al.* (2016) were able to identify numerous different kinds of polymers, including polystyrene and polycarbonate.

The results of their investigation demonstrated the existence of alkynes (C≡C), C-H groups, and aromatic structures, all of which are essential indications of microplastic polymers that are more often seen.

According to Coyle *et al.* (2020), the presence of microplastics in Muttom water may not only make marine creatures susceptible to harm via ingestion and accumulation, but it may also put human health at risk through the eating of seafood. The continuation and expansion of research activities is very necessary in order to address the ecological and human health problems that are associated with the presence of microplastics in the marine environment.

Furthermore, there is a need for regulatory measures and public awareness programs to be directed at the local population. These efforts will assist in reducing the amount of microplastics that are released into the environment and will also encourage responsible waste management practices.

Conclusion

This investigation highlights the significant occurrence of microplastics in the coastal waters of Muttom. The amount of weight of the desiccated microplastic particles obtained from the water samples is roughly 40 mg per 5 liters. The microplastics found in the Muttom water samples exhibit a variety of colours, including pink fiber, transparent fiber, white fiber, blue fiber, coloured fiber, elongated fragments, blue fragment, brown pellets, black pellets, orange pellets, black filament and colourless filament. The SEM analysis indicated the presence of microplastic particles measuring 500 nm, whereas the Fourier-transform Raman spectroscopy identified specific polymers, including polystyrene and polycarbonate. The samples predominantly exhibit microplastics in the form of fibers. The results of this study demonstrate the presence of microplastics along the Muttom coastline. The main source of income for the residents of Muttom is derived from fishing activities. Consequently, it is essential to tackle the issue of microplastic pollution. Effective waste management practices, enhancing the longevity of plastic products, and increasing awareness can facilitate this outcome. Such initiatives have the potential to greatly diminish the influx of waste into natural surroundings, facilitating the restoration of aquatic habitats.

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