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Original Article

Microplastics contamination in the gastropod, *Telescopium telescopium*, from the mangrove area of Versova Creek, Mumbai, India

Yousuf Dar Jaffer[*1,5], Mahua Saha[2], Zahra Taghizadeh Rahmat Abadi[3], Ganesan Kantharajan[4,5], Kooloth Valappil Rajendran[5]

[1]ICAR-Central Soil Salinity Research Institute, Karnal, India.
[2]CSIR-National Institute of Oceanography, Goa, India.
[3]Faculty of Life Sciences and Biotechnology, Shahid Beheshti University, Tehran, Iran.
[4]ICAR - National Bureau of Fish Genetic Resources, Lucknow, India.
[5]ICAR-Central Institute of Fisheries Education, Mumbai, India.

Abstract: Microplastic (MP) content in the gastropod, *Telescopium telescopium*, collected from the mangrove forest of Versova Creek, Mumbai was investigated. In total, 60 specimens were collected and pooled into six groups of 10 animals, each according to their weight and size. The concentration of extracted MP was expressed as the number of MP particles g^{-1} soft tissue (wet weight) and $Ind.^{-1}$ (individual). MP was detected in all six groups and ranged from ~1 to 4 MP/g soft tissue and ~4 to 23 MP/individual. The minimum number of MP both in soft tissue and in each individual were 1.12 MP/g and 3.6 MP/Ind, respectively, and were found to be present in the lowest wet weight group (3.21±0.33 g). The size of the longest dimension of MP varied from 21-435 µm, most of which were smaller than 100 µm. The majority of the MP found in each weight group were colorless and transparent fragments were the most prevalent shape (55.20%). FTIR analysis showed polyethylene, polypropylene, and polyurethane were the major polymer types. The study reports the microplastic content in a gastropod, Telescopium inhabiting the mangroves of Mumbai, India. As an algal feeder/detritivore, the presence of MP in its soft tissue suggests molluscans are prone to consuming MP, relative to the environmental availability. They had a higher proportion of MP than body weight, indicating the potential transfer of MP into higher trophic levels of the mangrove ecosystem. Irregular fragment MP dominance indicates Telescopium graze on weathered plastic items covered by fouling algae and contributes to MP formation.

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Introduction

Plastic production has increased from 1.5 million metric tons (MT) in 1950 to 360 MT in 2018, and consequently, plastic debris has been recognized as an emerging pollutant of environmental concern (Geyer et al., 2017; Pastics Europe, 2019). Plastic waste from the land reaches the marine environment through various routes and forms the largest proportion of oceanic plastic debris (75-90%), especially along the shallow coastal areas where sensitive ecosystems are located (Andrady, 2011; Mehlhart and Blepp, 2012). The significant impact of plastic pollution is mainly due to its persistence and widespread dispersal (Thompson et al., 2004; Nelms et al., 2019; Coyle et al., 2020). Of all the plastic wastes ending up in water bodies, particles 1 µm to 5 mm in size are defined as microplastics (MP), which pose the biggest ecotoxicological concern (Cesa et al., 2017; Coyleokd et al., 2020). MP can be of primary origin that is produced and released into the environment as micro-size particles from plastic industries such as virgin resin pellets used as precursors in plastic manufacturing processes, and microbeads in cosmetic products. Secondary MP forms the majority of the group and is formed as a result of the fragmentation of larger plastic materials by physical, chemical, and biological weathering (Duis and Coors, 2016). Fragmentation of parent plastic materials is the most likely process for the generation of secondary MP in the marine environment (Andrady, 2011) and MP

[*]Correspondence: Yousuf Dar Jaffer
E-mail: dar.yousuf@icar.gov.in
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presence in the West Indian environment is progressively being researched by scientists (Maharana et al., 2020a; Manickavasagam et al., 2020; Vaid et al., 2021; Gurjar et al., 2022).

The range of MP distribution varies from glaciers (Ambrosini et al., 2019) to the world oceans, including islands (Lavers and Bond, 2017), profound marine ecosystems (Jamieson et al., 2019), and coastal areas (Yu et al., 2018; Hale et al., 2020). The abundance of MP has been reported in different parts of the world ranging from 62.7-7900 particles kg^{-1} dry sediment (Nor and Obbard, 2014; Garcés-Ordóñez et al., 2019; Zhou et al., 2020; Zuo et al., 2020). Although, in water channels connected to the Juhu Creek in Mumbai, India, mostly macro- and mega plastics and a minor amount of MP (1-2 g/Kg debris) were found (Manickavasagam et al., 2020); a recent study in the coastal waters of Mumbai showed 372±143 MP/L and 9630±2947 MP/kg in water and dry sediment samples, respectively. The coastal areas around Mumbai include the mangroves that act as a buffer and natural entrapment zone of debris and silt particles originating from the land and marine environments. The first description of MP presence in mangroves was reported from seven intertidal mangrove habitats of Singapore where the majority of MP was found <20 μ (Nor and Obbard, 2014). While still, very little data are available about the presence and composition of MP in mangrove forests of Mumbai creeks.

The mangrove habitat is dwelled by various infaunal and epifaunal species and supports motile life forms such as prawns, crabs, and fishes by acting as breeding, nursing, and feeding grounds for the animals (Nagelkerken et al., 2008). Concurrently, these significant ecological areas have shallow water levels and slow water flow and, hence, can result in chronic exposure of ambient fauna to the MP transporting from the terrestrial and marine environment (Cannicci et al., 2008). However, there has been no evidence to date on the presence of MP in mangrove-associated fauna in India and the reports of MP in Indian waters are scant.

Mollusca phylum is one of the important aquatic and terrestrial fauna of mangroves represented by predators, herbivores, detritivores, or filter-feeders in the food web (Cannicci et al., 2008). The gastropod mollusc, *Telescopium* from the family *Potamididae* lives throughout the mud flats of mangrove forests along the West and Central Indo-Pacific regions (Alexander et al., 1979; Reid et al., 2008). Commonly known as mangrove horn snails or mud whelks, *Telescopium telescopium* Linnaeus, 1758 is the only living organism of this genus, and is an algal feeder and detritivore. They have an evolutionary association with the mangroves due to shelter, predator protection, and substrate and food provided by the ambient environment (Houbrick, 1991; Reid et al., 2008). Furthermore, the studies conducted so far on MP reporting in the Indian context are lacking information on biota. Against this background, the present study explores the MP contamination in mangrove-dwelling gastropod mollusc, *T. telescopium,* through microscopy and FTIR analysis.

Materials and Methods

Description of the sampling area and sampling procedure: Mumbai is surrounded by a number of Creeks, including Manori, Versova, Mahim, Mahul, and Thane which support mangrove growth with an area of about 66.3 km^2 (Forest Survey of India, 2019). The mangrove habitat of Mumbai is mostly characterized by creek mudflats, rocky cliffs, bays, and open shorelines (Kantharajan et al., 2017). The mangrove area Dharavali (19°10'10"N and 72°47'49"E) in Versova Creek, Mumbai, along the West Coast of India, was selected for this study. The Versova creek has mudflats with the dominance of Grey Mangrove, *Avicennia marina* (Fig. 1).

A 100 m line transect was chosen in all sampling times from seaward to mangrove direction, and the samples were collected at three to four intervals in a 1×1 m quadrant (Andrés Satizabal et al., 2012). *Telescopium telescopium* specimens were quite visible within the quadrats and were randomly collected by handpicking. They were chosen as the representative species due to their higher abundance in the sampling area and were mostly found lying over the muddy substratum rich in detritus. A total of 60

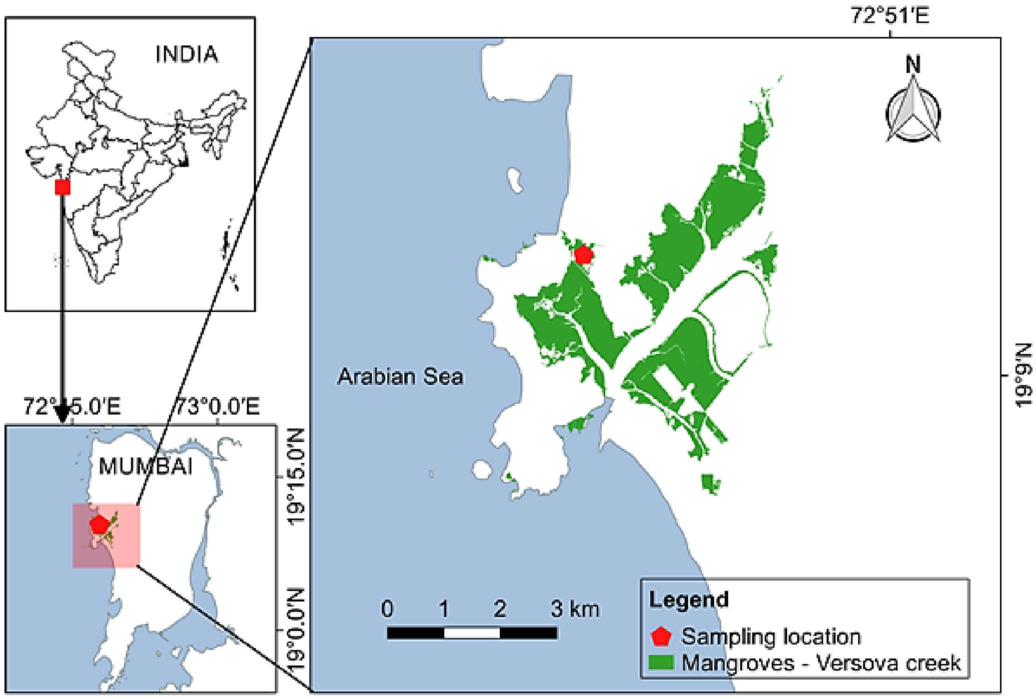


Figure 1. A map developed by using Arc-GIS Ver. 10.5 showing the geographical location of Dharavali mangrove area in Mumbai, India.

alive specimens were collected, stored at -20°C, and subsequently thawed before further examination.

Quality assurance and quality control of analysis: To avoid contamination, all the experiments were done in a clean laboratory, while the technician wore a white cotton lab coat. The samples did not leave without cover and measures were taken to avoid the contamination of airborne fibers. The blank sample without tissue was run in parallel to correct the handling and procedural contamination. Water and other chemicals used were filtered with filter paper before use. All the containers and beakers were rinsed with distilled filtered water whenever in use.

Sample preparation, extraction, observation, and quantification: For each specimen, the morphological characteristics such as total shell length and width, total weight, and soft tissue weight were recorded (Supplementary data, S1) (Fig. 2). The digestion, separation, and collection of MPs from molluscs were based on a modified protocol reported by Li et al. (2018). First, the soft tissue was removed from the shell by cracking the surface carefully and washed thoroughly with double-distilled water to remove the broken intact shell debris, and MP, if any, on the outside. Around ten individuals of similar weight (soft tissue weight) were placed into a 1 L flask as a replicate/group, and subjected to digestion with 200 mL of 10% potassium hydroxide (KOH). As the specimens present a complex stomach structure (Zaman and Jahan, 2013), the whole soft tissue was subjected to the digestion process instead of the gastrointestinal tract only. The flasks were covered with aluminum foil and placed on a shaking incubator at 100 rpm for a week at room temperature (30-35°C).

The previous studies on the extraction of MP from shellfishes (Table 1) mostly used H_2O_2 and HNO_3 or a mixture of acids (HNO_3 and $HClO_4$) (De Witte et al., 2014), but in the present study, KOH was used for the digestion of gastropod soft tissue. The KOH has shown the best performance in the extraction of MP from biotic samples and is the most effective technique for extracting MP. It is cost-efficient, requires a simple sampling procedure, and allows filtration of digestates at ≤25 μm. The chemical is found to be efficient in retaining the size, shape, color, and polymer of each MP particle (Dehaut et al., 2016; Thiele et al., 2019).

The complete digestion of tissues was confirmed

Figure 2. The gastropod, *Telescopium telescopium* collected from the coastal mangroves of Mumbai for determination of microplastic pollution.

intermittently by the absence of any tissue in the shaking flasks. The MP from the supernatant was collected by adding high-density salt of 4 M sodium iodide (NaI) (around 800 mL) to each flask. During the whole process, the working space was cleaned thoroughly with distilled water and maintained in a way to avoid contamination with airborne MP.

Considering the density of NaI solution (3.67 g/cm³), it is expected that the majority of plastic particles be concentrated on the supernatant layer of the solution. Therefore, the supernatant solution was taken after 1 hour and stored in a pre-cleaned glass container for further experiments. The suspended MP particles were counted with a Sedgewick-rafter cell etched with a 20x25 grid repeatedly. The solution containing MP was then collected in a separate pre-cleaned container. Size and particle count measurements were conducted at 20X and 40X magnifications with an optical microscope (Axioskop 40 FL, Carl Zeiss, Micro Imaging Gmbh Germany) attached to an ISH 500, 5 Mega Pixel camera. MP was characterized based on their shapes into filaments, multifilament brunch, strips, and irregular fragments; their color into colorless and colored ones; and their size. Filaments possessed straight and tapering, angular, and twisted forms. The concentration of MP was indicated either as average particles individual^{-1} (i.e., the total number of MP in a pooled sample/number of individuals in the pool) or particles g^{-1} soft tissue (wet weight) (i.e., the total number of MP in a pooled sample/total soft tissue weight of the pool) (Naji et al., 2018).

FTIR characterization of polymers: To identify the polymers in MP samples, Fourier Transformed Infrared (FTIR) spectroscopy analysis was carried out. For this, the collected supernatant containing the MP (approximately 10 mL) was vacuum-filtered through a hydrophilic 0.22 µm filter paper (47 mm diameter) (Durapore GVWP) after 24 h of floatation when the suspension was apparent. Each filter paper was placed in a clean Petri dish for further analysis. The polymer identification was carried out using a Hyperion 3000 FTIR-ATR spectrometer (Bruker, Germany) equipped with a focal plane array (128x128) at the Sophisticated Analytical Instrument Facility (SAIF) of the Indian Institute of Technology Bombay, India. The IR spectra were recorded in transmission mode as average spectra by calculating the arithmetic mean of 16 scans in the range of 4000-900 cm^{-1} with a spectral resolution of 0.2 cm^{-1}. A background scan without a sample was performed on the blank sample carrier before sample measurement (Löder and Gerdts, 2015). Peak identification was made using the range of wavenumbers (cm^{-1}). The interpretation of bands was made using the wavenumbers concerning the transmittance. The identification of polymers was made by the presence of specific absorption bands following the literature (Gulmine et al., 2002; Charles and Ramkumaar, 2009; Chang, 2012; Jung et al., 2018).

Statistical analysis: For comparing MP numbers between replicates/groups, a one-way analysis of variance (ANOVA) was performed using the software IBM SPSS Statistics V21.0. To measure the specific difference between pairs of means, a post-hoc Duncan's Multiple Range Test was used. The significance level of all the results was considered at $P<0.05$. A principal component analysis (PCA) was also conducted using correlation and the Varimax method with Kaiser Normalization as the rotation method to the shapes of MP. PCA was used to observe trends in the shapes of MP.

Results and Discussion

The *T. telescopium* specimens collected from the mangroves had a thick, cone-shaped, and striped shell, and blackish or murky brown color (Fig. 2). The shell length and longest width varied from 60.70±2.54 to 73.56±6.04 mm and 26.65±1.01 to 35.12±1.37 mm, respectively. The size of an average adult shell can reach 90-100 mm in length; however, it has also been recorded to be 130 mm in size (Houbrick 1991; Adriman et al., 2020). There was a significant difference ($P<0.05$) in soft tissue weight (wet weight) among the groups, with the lowest and the highest mean weights of 3.21±0.33 g and 6.80±0.82 g, respectively (Supplementary data, S2).

The suitability of gastropods as ecological indicators is due to their highly tolerant and sedentary mode of life, abundance in coastal environments, easy identification, and deposition of pollutant concentration to the level of reflecting exposure (Maher et al., 2016). Gastropods have been used as a potential biomonitor of toxic trace element pollution in various instances (Yap et al., 2009; Maher et al., 2016; Krupnova et al., 2018). *Telescopium telescopium* dwells mainly in muddy areas rich in mangrove leaves and decomposed organic matter, ingests mud and digests the detritus and other organic matter. These activities, i.e. grazing the surface of firm mud or sand grains, ingesting fine organic detritus, and excreting fecal pellets can cause micro-bioturbation which affects the properties of the grazing substrate. Their feeding behavior is reflected in their morphological trait, i.e., a very small radular structure that functions as proboscis (Zaman et al., 2011; Zaman and Jahan, 2013). Moreover, rhythmic feeding and digestion have been demonstrated in *T. telescopium* and other gastropods which mainly inhabit the upper littoral zone, and are underwater only for 3-4 hours during one tidal cycle. They mostly prefer to graze on exposed mudflats during low tide, but they can even feed during the high tide when covered with water due to extensible snout (Zaman and Jahan, 2013). Their non-selective feeding strategy and morphological characters expose them readily to the available plastic items in their habitat, whether they are settled in mud or particulate form, MP (Putri and Patria, 2021). If they graze upon microalgae from hard surfaces, including decaying plastic items, the same as what happens to decayed leaves, they can remove tiny pieces of plastics, ingest, accumulate, and then excrete them. Therefore, they could actively contribute to the formation of small and smaller pieces of microplastics. This contribution of animals in MP forming is a possible way of forming smaller pieces of plastics that need more attention to clarify their possible paths including enzymatic degradation or mechanical pressures.

Concentration of MP in *T. Telescopium*: MPs were detected in almost all the specimens collected from the mangrove area. The mean number of total MP in specimens ranged from ~1 to 4 particles g^{-1} soft tissue (wet weight), and ~4 to 23 particles $ind.^{-1}$. A study conducted on the coast of Rambut Island, Jakarta reported about 765 MP particles $individual^{-1}$ in *T. telescopium* which is much higher than what we observed in our samples. They also reported 31.7 particles/g in surrounding sediment and 15.46 particles/mL in water samples which reflects the bioaccumulation in the soft tissue of *T. telescopium,* especially in their respiratory organ (Putri and Patria, 2021). Both of these numbers are much higher than reported in sea snails, *Ellobium chinense* body (external and internal) from mangrove forest on the north of Beibu Gulf ranging from 38.0±2.0 to 320.0±3.0 items kg^{-1} (Li et al., 2020).

The abundance of MP in mangrove areas seems to be generally higher than in the surrounding zones (Zhou et al., 2020). Moreover, the higher concentration is believed due to the vicinity of the sampling location to the urban runoff. Along the four sandy beaches of Mumbai, India, 7.49 g and 68.83 plastic litter items per square meter were recorded on average, which comprised 41.85% MP (Jayasiri et al., 2013). In a previous study (Maharana et al., 2020a), 498 MP items m^{-2} were reported at Aksa Beach, Mumbai, which is close to the mangrove area where the gastropods were collected. In a survey conducted along the northeast coast of the Arabian Sea (located in the northwestern part of the Indian Ocean), plastic-

Table 1. Comparison of abundance of MP, polymer type, the shape of MP, color, and size in molluscs reported in the peer-reviewed literature from different regions of the world.

Study area	Name of molluscan species	Polymer type	Number of MP (particles g^{-1} soft tissue (ww)/particles individual^{-1})	Shape of MP	Color	Size range (μm)	Extract solution	Reference
Belgium[ab]	*Mytilus edulis*	-	0.26-0.51 g^{-1} soft tissue	Fibers	black, red, blue, purple, translucent, transparent, orange, green and yellow	200 - 1500	Nitric acid (VWR, 65%) and perchloric acid (VWR, 68%), HNO3:HClO4 (4:1 v:v)	(De Witte et al., 2014)
Halifax Harbor, Nova Scotia, Canada[b]	*Mytilus edulis*	-	nd/34-178	Fibers	Clear, black, other colors	9 - 219	H2O2 treatment	(Mathalon & Hill, 2014)
Germany (North Sea)[a]	*Mytilus edulis* *Crassostrea gigas*	-	0.36 ± 0.07 particles g^{-1} soft tissue, 0.47 ± 0.16* particles g^{-1} soft tissue	Particles without observed fibers	Red, blue	5 –20	69% nitric acid	(Van Cauwenberghe & Janssen, 2014)
Coastal water of China[a]	*Mytilus edulis*	CP, PET, PES	0.9-4.6 g^{-1} soft tissue and 1.5-7.6 particles individual^{-1}	Fiber, fragment, sphere, flake	-	5 - >5000	30% H2O2	(J. Li et al., 2016)
Coastal water of The Persian Gulf, Iran[a]	*Amiantis umbonella*, *Amiantis purpuratus*, *Pinctada radiate*; *Cerithidea cingulata*, *Thais mutabilis*	PE, PET, PA	12.8-20.0 g^{-1} soft tissue and 3.7-17.7 particles individual^{-1}	Fiber, film, fragment, pellet	Black, white, transparent, red, pink and green.	10 - >5000	30% H2O2	(Naji et al., 2018)
Three major cities in south Korea[b]	*Crassostrea gigas*, *Mytilus edulis*,Tapes philippinarum, *Patinopecten yessoensis*	PE, PP; PS, PES	0.15±0.20 g^{-1} soft tissue and 0.97±0.74* particles individual^{-1}	fragment, fiber, film and sphere	Colorless	<300	10% potassium hydroxide (KOH)	(Cho et al., 2019)
15 Oregon coast, U.S.A.[a]	*Crassostrea gigas*, *Siliqua patula*	PET, acrylic, aramid, zein, cellophane cellulose-based material	0.35±0.04 g^{-1} soft tissue and 0.16±0.02 particles individual^{-1}, 10.95±0.77 g^{-1} soft tissue and 8.84±0.45 particles individual^{-1}	Microfibers (99%)	Colorless, blue, gray, and black	100 – 8720	10% potassium hydroxide (KOH)	(Baechler et al., 2020)
Mangrove forest of Beibu Gulf[a]	*Ellobium chinense*	PP/PE	0.038-0.32 g^{-1} soft tissue	-	White	500 - 1000	10% potassium hydroxide	(R. Li et al., 2020)
Sal estuary, Goa India[a]	*Crassostrea* sp., *Perna viridis* and *Paphia malbarica*	PAM, EVOH, PVC and PA	4 ± 2, 3.2 ± 1.8 and 0.7 ± 0.3 g^{-1} body weight	fibres, fragments, films or other plastics	Black, blue, green, red, orange, pink, yellow, brown, white and transparent	10 - 428	30% H2O2	(Maharana et al., 2020b)
Mangroves in Mumbai – India[a]	*Telescopium* sp.	PE, PP, PU	1-4 g^{-1} soft tissue and 4-23 particles individual^{-1}	Filament, fragment, strips	Clear, red and blue	<100	10% potassium hydroxide	Present study

Note: CP: Cellophane, PET: Polyethylene Terephthalate; PES: Polyester; PP: Polypropylene; PS: polystyrene; PU: Polyurethane; PAM: Polyacrylamide; EVOH: Ethylene Vinyl alcohol; PA: Polyamide. a = wild; b = farmed, * = mean concentration; - = not defined

based debris formed the major proportion (87.1%) of total debris recorded and was mainly comprised of plastic bags and food wrappers (Selvam et al., 2021). The above numerical figures of MP concentration explain the higher exposure rates of MP to the fauna inhabiting mangroves, either the detritivores or the filter-feeding molluscs. A comparison of MP concentration, polymer type, dominant shapes, color, and sizes in worldwide shellfish species with different feeding strategies, including our findings is summarized in Table 1.

Also, concentration values were higher in all the replicates when expressed as the number of particles g^{-1} soft tissue (wet weight) than as particles individual^{-}

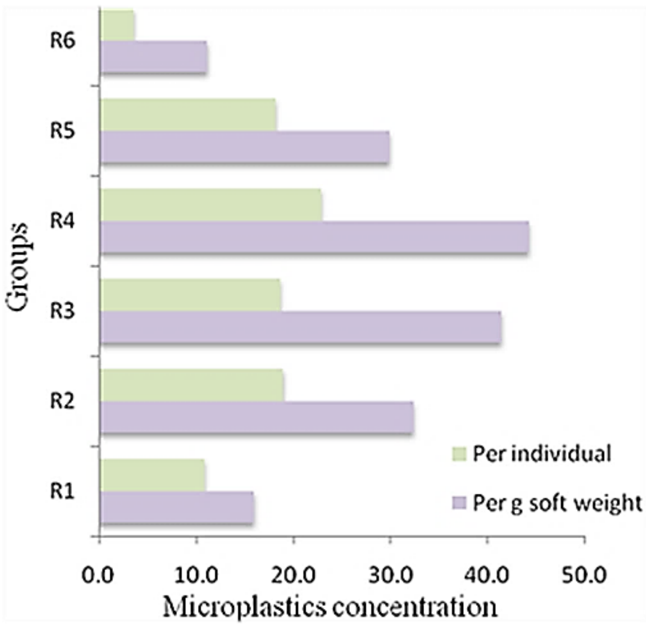


Figure 3. Microplastics concentration found in in the 10% NaOH digested gastropod, *Telescopium* from coastal mangroves of Mumbai. Results estimated by soft tissue weight (number g^{-1} soft weight) and by individual (number $individual^{-1}$) on average. R6 contained the lowest weight group.

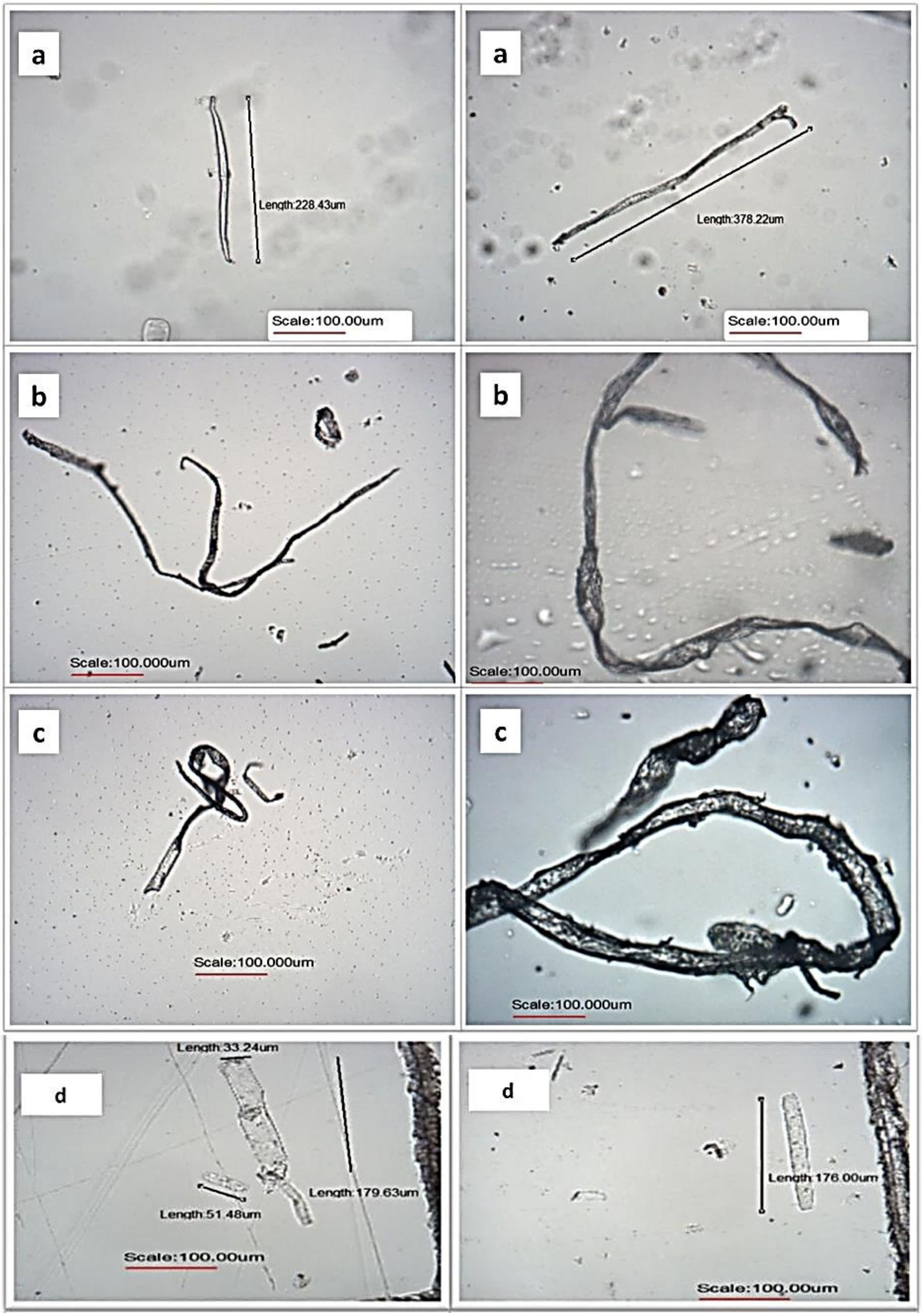
[1]. The minimum number of MP in both g^{-1} soft tissue (wet weight) (1) and $ind.^{-1}$ (4) was found to be present in the lowest wet weight (3.21±0.33 g) group (Fig. 3). If we assume that the members of the lowest weight group are younger molluscs than the heavier ones, then the minimum number of MP in the lowest weight group indicates that the ingestion rate increases with size/age or perhaps due to bioaccumulation through time. Putri and Patria (2018) reported a significant positive correlation (r = 0.744) between the amount of MP in *T. telescopium* soft tissue and the body mass of the specimen, too.

Size and shape of microplastics collected from *Telescopium*: The size range of MP varied from 21 to 435 µm, while most of them were in the size range of <100 µm. In mangrove biota MPs <1 mm size are mostly reported (Deng et al., 2021) because microplastics of smaller sizes are readily taken up by a wide range of organisms. The majority of MP recorded from mangroves in Singapore were fibrous and <20 µm in size (Nor and Obbard, 2014). The size range of MP in the mangrove snails of Beibu Gulf was a bit larger, 500-1000 µm (Li et al., 2020). The size range of ingested MP may reflect the available MP size in their grazing or feeding area. However, the role of degradation and breakage processes under the influence of mechanical movements of intestines or enzymatic activity inside the body of animals should be considered.

Two major types of MPs, filaments, and irregular fragments were observed under the light microscope while almost no microbeads or films were found. The shape, number of fibrils, color, bending, or straight nature of fibers reflect their diverse sources. Although it is not usual, we categorized the observed filaments into three subclasses: filaments were straight and tapering (SFT), angular (AF), and twisted (TF). Moreover, the irregular fragments were separated based on their shape and color characteristics into multifilament bunch (MB), rectangular strip (RS), discolored or transparent fragments (F), and colored fragments (CF) (Fig. 4).

There was a significant difference in the number of MP of fragment shape as compared to the other forms ($P<0.05$) (S2). Except for MB, which was absent in three groups (replicates), the other shapes were present in all the groups. Putri and Patria (2018) found the film shape of MP is dominant in their *Telescopium* samples (about 368.51 MP/Ind.). The degradation of plastics can happen via UV radiation and oxidative properties of the atmosphere (e.g., thermal oxidation), hydrolytic properties of seawater, hydrodynamic phenomena (e.g., effects of winds, waves, and currents), and biological degradation (Andrady, 2011; Webb et al., 2012; Koelmans et al., 2014) (GESAMP, 2015; Chamas et al., 2020). Consistent with our results, the dominance of irregular fragments and films may reflect the grazing habitat and possible MP fragmentation through grazing along with their accumulation in the target tissues.

The Majority of the MPs, including all filaments, were transparent, and only two colors, red and blue were detected in the proportion of 22.08% of the total MP (Fig. 4). The transparent fragments (F) (55.20%) and a multifilament bunch (MB) (0.32%) formed the highest and the lowest proportions of total shapes, respectively. In mangrove biota, MPs possess various colors mainly black, white, transparent, red, pink,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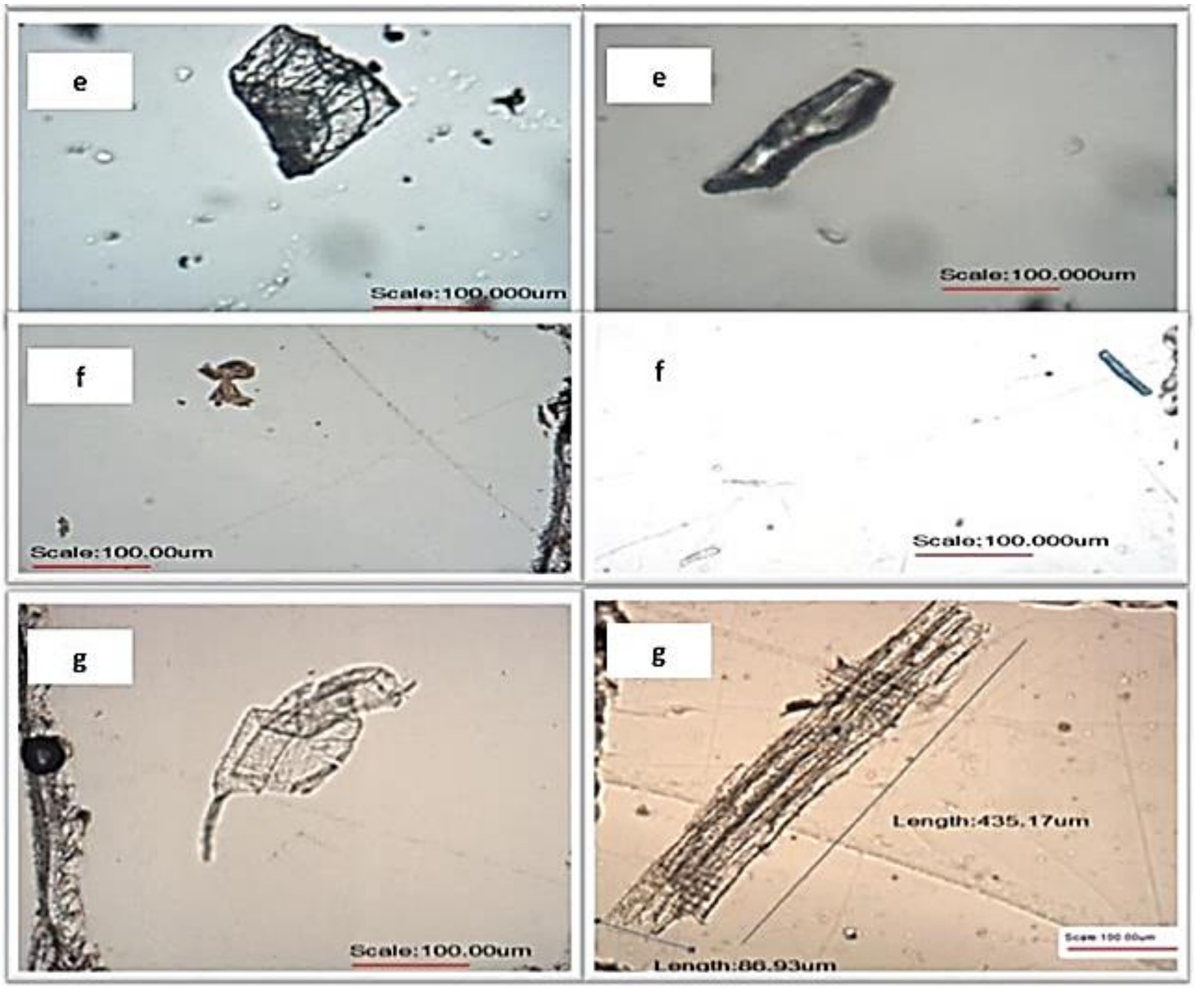

Figure 4. Microscopic images of varios shapes of MP, found in gastropod (*Telescopium telescopium*), collected from Versona coastal mangroves of Mumbai. a; straight and tapering filament, b; angular filament, c; twisted filament, d; rectangular strip, e; transparent fragment, f; colored fragment, g; multibunch fragment.

and green have been reported (Deng et al., 2021). The color pattern of MP gives a broader idea of the range of sources of MP. As an example, MP noticed in snails from mangrove forests located in the north of Beibu Gulf were mostly white (41-90%) (Li et al., 2020).

PCA taking data co-linearity into account was applied to determine which shapes of MP best accounted for the most variability among the different weight groups. The PCA was able to distinguish and highlight the importance of shapes of MP found in the *Telescopium* based on their distribution across other groups (Table 2, Fig. 5). In the PCA of 7 variables, four PCs had eigenvalue>1 and explained 87.3% of the variance in data (Table 2). Highly weighted variables under PC1, PC2, PC3, and PC4 included straight and tapering filaments (SFT), twisted filament (TF), colored fragment (CF), and transparent fragment (F), respectively that best represented the system attributes. PCA is an exploratory data analysis tool that analyses the data variance and identifies the independent principal components (PC) that account for maximum variance within the set. This technique has been applied to identify polymer types, and differentiate plastic with non-plastic particles and associated dyes (Van Cauwenberghe and Janssen, 2014; Imhof et al., 2016; Halstead et al., 2018). However, in our study, PCA is being used to highlight the variance in the shapes of the MP. It is proposed that using PCA for variance in shapes will provide the maximum information possible regarding the distribution and identification of MP. Further study needs to be conducted to confirm if any correlation exists between the shape of MP and polymer type so that an MP shape library can be developed for easy identification of polymer type.

Table 2. Results of principal components analysis of different shapes of microplastics in gastropod mollusk *T. Telescopium.*

PC'S	**PC1**	**PC2**	**PC3**	**PC4**
Eigen Value	2.251	1.616	1.18	1.061
% of Variance	32.158	23.09	16.857	15.159
Cumulative %	32.158	55.247	72.104	87.263
Factor loading/eigen vector Variable				
Straight filaments and tapering (SFT)	0.977	0.006	0.096	-0.108
Angular filaments (AF)	0.92	0.24	-0.111	0.127
Twisted filaments (TF)	0.02	0.976	0.17	0.081
Rectangular strips (RS)	0.368	-0.201	-0.212	0.234
Fragment	-0.006	0.098	-0.078	0.98
Colored fragment (C_F)	0	0.046	0.984	-0.079
Multifilament bunch (MB)	0.561	0.744	-0.332	0.077

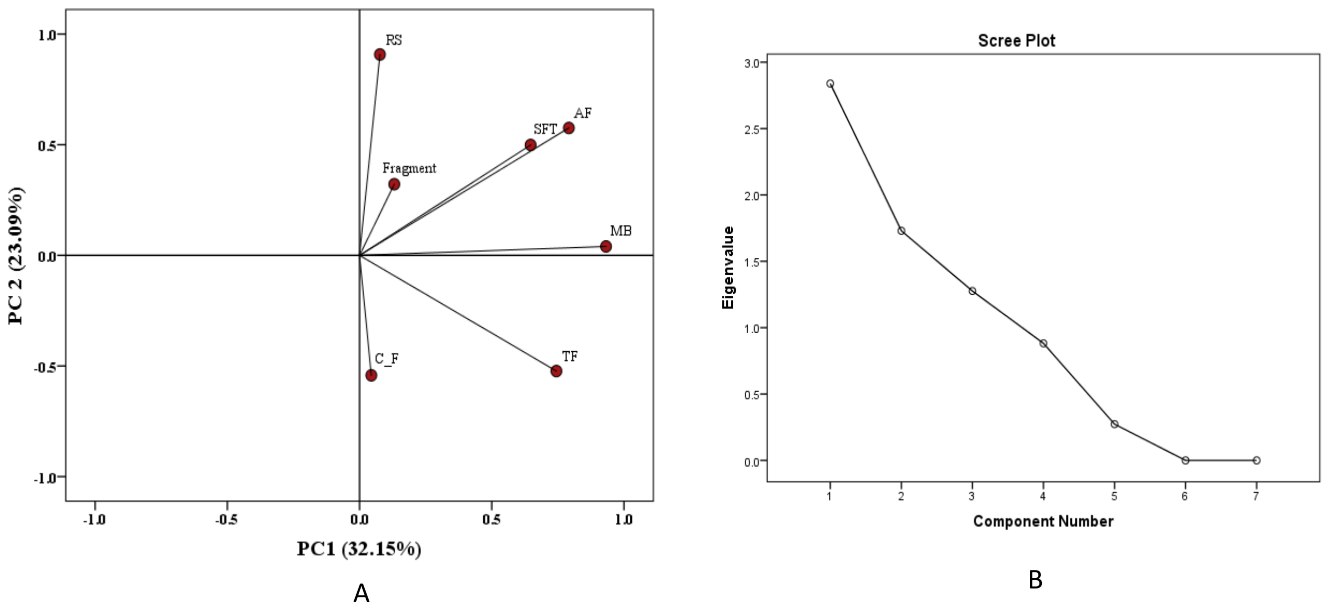

Figure 5. A: Loadings plotted on principal component (PC) PC1 (32.15%) vs PC2 (23.09%) obtained by creating a principal component analysis (PCA) correlation matrix to show the relation between parameters (shapes of MP) and PCs. SFT: straight and tapering filament; AF: Angular filament; TF: twisted filament; RS: rectangular strip; F: transparent fragment; C_F: colored fragment; MFB: multifilament bunch. B: Scree plot of the eigenvalues of factors of principal components in an analysis.

Identification of polymer type: In order to confirm the polymer type of MP in the specimens, some representative samples were chosen for FTIR analysis. The chemical characterization indicated that these particles were composed of polyethylene (PE), polypropylene (PP), and polyurethane (PU) (Fig. 6).

Among the various available spectroscopic methods, FTIR has gained wide acceptance for polymer identification by producing spectra that can be matched to spectral libraries representing distinct chemical functionalities present in the material (Zarfl, 2019). The persistent plastics like PE and PP have mostly been distributed in aquatic ecosystems released mainly during fishing activities. They are also used in packaging, industrial insulation, and facial scrubs (Pruter, 1987; Browne et al., 2011). The polymer type of MP found along the seven intertidal mangroves of Singapore was PE, PP, nylon (PA), PVC, and PE/PP in mangrove snails of Beibu Gulf (Nor and Obbard, 2014; Li et al., 2020). PE and PP of micro and macroplastics were predominant over the SE coast beaches of India with the maximum percentage in Mumbai (Maharana et al., 2020a). The MP particles

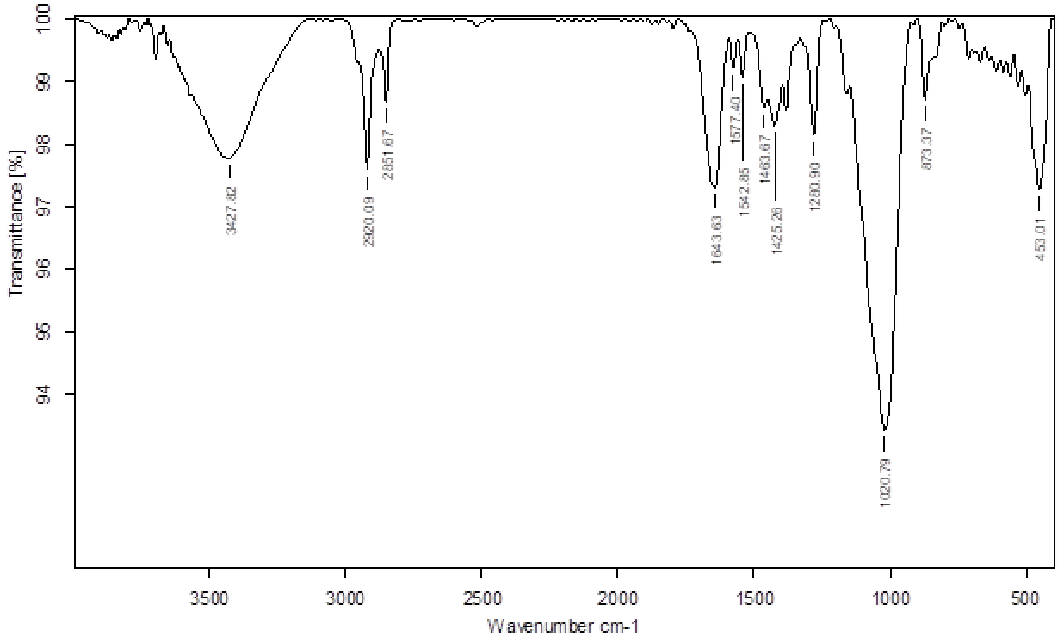

Figure 6. FTIR spectra of polymers found in the gastropod, *Telescopium telescopium,* from coastal mangroves of Mumbai. The peaks are related to the Polyethylene (PE), polypropylene (PP) and polyurethane (PU) polymers.

found in different molluscan species along the Persian Gulf were PE, polyethylene terephthalate (PET), and PA in nature (Naji et al., 2018).

Conclusion

The present study is the first to report on the microplastic content in a gastropod, *T. telescopium* inhabiting the mangroves of Mumbai, west coast of India. A total MP content of ~1 to 4 MP/g wet weight and ~4 to 23 MP/Ind was measured in the gastropod. More importantly, the lowest weight group was found to possess the minimum number of MP indicating the ingestion rate increases with size/age and/or bioaccumulation occurs over time. Two major MP shapes, i.e. fiber and irregular fragment were detected in which fragments, especially transparent ones were the most common (55.20%). The size range of MP was from 21 to 435 µm, and most of them were smaller than 100 µm. The spectroscopy analysis indicated the presence of three polymer types; polyethylene (PE), polypropylene (PP), and polyurethane (PU). They could originate from different sources, including decaying fishing gear, wastewater treatment plants, or beach litter. Since this species is an algal feeder/detritivore, the presence of MP in its soft tissue suggests mollusca are prone to consuming MP, relative to the environmental availability. However, the type and size of MP in the digestive tract of biota can reflect the available plastic and microplastic sources in their surrounding nature, the dominance of irregular fragments and films (Putri and Patria, 2021) in *Telescopium* may be a result of their non-selective grazing on decaying plastic items covered by fouling algae. They had a nearly higher proportion of MP compared with bodyweight which indicates the potential for transfer of MP into higher trophic levels of the mangrove ecosystem. Perhaps collecting larger pieces of plastics and trapped wind or water-drifted plastic sheets and bags from the muddy flats and mangrove areas could be effective in decreasing the exposure of biota to MP contamination.

More knowledge on other aspects of the basic ecology such as temporal distribution, life stage analysis, and age effect would help in assessing if these gastropods can indeed serve as key biomonitors of MP in any selected coastal region. The findings of this study may also help to elucidate the mechanisms underlying the trophic transfer of MP from gastropods to organisms at the higher trophic level.

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Supplementary Materials: S1: Morphometric characters of the samples., S2: Statistical analysis

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