Occurrence of microplastics in the gastrointestinal tracts of edible fishes from South Indian Rivers

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Abstract

The occurrence of microplastics MPs in the gastrointestinal (GI) tracts of the five different fish species (*Chanos chanos*, *Chanda nama*, *Chelon macrolepis*, *Gerrus filamentosus*, and *Carangoides malabaricus*) from the Kollidam River and Vellar River of Tamil Nadu, Southern India were evaluated. A maximum of 23 individual fishes was sampled from both Rivers and alkaline digestion was performed to extract the MPs in the GI tract of fishes. The results revealed that a total of 315 MPs were isolated from the GI tracts of all fishes sampled from both Rivers with fibres and fragments. The average size of ingested MPs was ranged between 108.72 to 129.11 µm and 181 to 284 µm in the fishes sampled from Kollidam and Vellar River respectively. In the morphological examination of MPs, fishes from both Rivers showed the maximum level of fibres, followed by fragments. The colour pattern of the ingested MPs was dominated by blue, transparent, red and other colours in both rivers. In this study, MPs in the GI tract of fishes indicated that the MPs pollution in studied Rivers can cause significant threats to aquatic fauna.

Introduction

Plastics are synthetic polymers that originated from the derivatives of fossil fuels and are used widely due to their higher durability with great applications on commercial products. At present, plastic industries have become a major contributor to the applications of various industries, such as the pharmaceutical industry, energy generation, aerospace research, automotive, construction, electronics, packaging, textiles, etc (Plastic Europe 2019). On another hand, plastic pollution has become a very serious threat to the environment all around the globe, due to mismanagement and poor recycling rates of plastics. India was lifted to the 15th position of the plastic polluting country in the list of nations with the lower amount of recycling rates in terms of plastic waste treatment (Geyer et al. 2017). Microplastics (MPs) are very tiny particles that are all less than 5mm in size and widespread in all water bodies throughout the globe. Primary MPs were originated and reduced in size by the environmental processes, in order of photo-oxidation, thermal stress and wave actions (de Sá et al. 2018). MPs have been reported for several detrimental effects, on feeding, reproduction, intestinal damage, and disturbances of energy metabolism in various aquatic animals (Campanale et al. 2020; Hurley et al. 2017; Jiang et al. 2020; Koelmans et al. 2016; Xu et al. 2018). MPs have been encountered in the following invertebrates the zooplankton, amphipods, polychaete worms, tubifex worms, molluscs, echinoderms, etc. It is also encountered in chordates such as fishes, birds and mammals (Desforges et al. 2015; Fossi et al. 2018; Herzke et al. 2016; Hurley et al. 2017; Long et al. 2015; Rummel et al. 2016). Nearly 20% of MPs came from Fishing practices and 80% of MPs from were came from terrestrial activities which include personal care products, air-blasting process, improperly disposed plastics and leachate from landfills (Cole et al. 2011).

Microplastics entered to river ecosystem led to transfer to the ocean which can disturb the river and marine ecosystems (Dris et al. 2015; Free et al. 2014). A wide range of organisms ingests the MPs which accumulate and creates an adverse effect on the biology and physiology of organisms. Most of the effects are often related to a disruption to the digestive system like blockage of the intestinal tract, false sensation of satiation, etc (Cole et al. 2013; Farrell and Nelson 2013). Earlier studies reported that the accumulation of MPs in marine organisms including fish and shellfishes (Jonathan et al. 2021; Murray and Cowie 2011; Selvam et al. 2021; van Cauwenberghe et al. 2015). Nevertheless, the information on the accumulation of MPs in the organisms from the freshwater environment is limited that also can be applied to the freshwater bodies in India. Accumulation of MPs in the digestive tracts of animals including fish, molluscs, crustacean can affect their nutritional gain, energy reserves and poor survival and growth due to the reduction in feeding (Cole et al. 2013; Lu et al. 2016; von Moos et al. 2012; Wright et al. 2013).

In India, most of the studies focused on the occurrence of MPs pollution in the marine environment, while the impact of MPs pollution in the freshwater environment is very limited. Hence, the present study aims to evaluate the occurrence of MPs in the gastrointestinal tracts of edible fishes from Kollidam and Vellar Rivers in South India. The selected Rivers for the study, are serving as a habitat for the various faunal diversity and livelihood for the local fishermen communities until they end up in the Bay of Bengal Sea Kollidam and Vellar Rivers are flowing in sub-parallel patterns to each other. Both Rivers are geographically located in the Cuddalore District along the southeast coast of Tamil Nadu, India. Kollidam River in the Northern distributary of Cauvery River, which splits from Cauvery near Srirangam. It flows about 150 km and empties into the Bay of Bengal near Sirkali in Nagapattinam District. Vellar River originates in Chittori hills of the Eastern Ghats in the Salem district of Tamil Nadu at an elevation of 900 m, and it has a total length of 210 km runs through Salem and Cuddalore districts in Tamil Nadu enter into the Bay of Bengal near Parangipettai, in Chidambaram district.

Materials And Methods

Study area and sample collection

Fishes were collected from the selected sites of Vellar (11°26'23.00" to 11°26'27.15" N, 79°33'53.35" to 79°33'57.05" E) and Kollidam (11°19'58.30" to 11°20'23.19" N, 79°42'42.82" to 79°43.07'71" E) Rivers (Fig .1) with the help of local fishermen during February to May 2020. Collected fishes were anaesthetized and stored in a sterile sample container and transported to the laboratory for further examination. Fishes were cleaned by using de-ionized water to remove the debris and minimize external MPs contamination. The lengths and weights of sampled fish were ranged between 11.7 to 27.8 cm and 21.77 to 217.72 g respectively.

Identification of fishes

Collected fishes were identified based on their morphological features using standard manuals and the data obtained from the Fish Base website Froese and Pauly (2021). Identification keys of length, shape, depth, mouth, distribution and nature of fish spines, scales were used to identify and categorize the taxonomical information of these collected fishes. Totally 23 fishes were examined and identified as five different species. Among these, *Chanos chanos* and *Chanda nama* were identified from the Kollidam River, *Carangoides malabaricus* and *Gerrus filamentosus* were identified from the Vellar River, and *Chelon macrolepis* identified from both Kollidam and Vellar Rivers.

Sample processing for extraction of MPs

The gastrointestinal tract (GI) from each fish was dissected out in an aseptic condition and rinsed by using ultrapure water to reduce the chances of plastic contamination, dissected guts were transferred into sterile glass containers for further digestion. Digestion of fish guts was done by using the alkaline digestion method of Karami et al. (2017). In brief, dissected guts of the fishes were transferred into sterile 100 ml wide mouth glass beakers, followed by 10% of KOH w/v solution was added and the mixture was kept at 40ºC for 72 hours in an incubator to compete for the digestion process. The digested mixture was filtered by using a 0.8 µm polycarbonate membrane filter to isolate the remaining residues from the mixture. Density separation was performed by the addition of 15 ml of 4.4 M sodium iodide into a beaker containing filter membrane with residues and kept for 5 min- at room temperature. Further, the filter membrane containing beaker was sonicated at 50 Hz for 5 min using Labman Ultra Probe Sonicator, followed by the beaker was agitated by using orbital shaker at 200 rpm for 5 min, and the solution was centrifuged at 500× g for 2 min. The supernatant was obtained from the centrifuge tube was vacuum filtered using a vacuum filtration unit (0.8µm pore size). The density separation was repeated on the settled precipitate remains from the centrifugation step.

Morphological observation of extracted MPs

Filter membranes were obtained from the vacuum filtration unit and viewed under the Magnus stereomicroscope with 20-40× magnification. Suspected particles were treated with a hot needle test to observe the melting points of the particles to identify the MPs (de Witte et al. 2014).Photographs of the suspected particles were taken by using Ultrascope 9.0v camera and the particles sizes were measured by using ImageJ software v.1.50i, (http://imagej.nih.gov). The morphological characterization of MPs was done by using the distinguishing method criteria like size, shape, colour, etc (Hidalgo-Ruz et al. 2012).

Scanning electron microscope analysis of extracted MPs

Extracted MPs from both rivers were quantified and picked up by using 0.3×0.15mm tip-sized micro forceps and segregated for surface morphological analysis using a Field Emission Scanning electron microscope (FE-SEM) (FEI Quanta 250 FEG).

Data Analysis

The data obtained from all the parameters were expressed as mean ± SD. Significance variations in MPs, colour, size and shape were determined by one-way analysis of variance (ANOVA) followed by Duncan's multiple range test. The significance level was fixed at $p < 0.05$. All the statistical analysis was performed using SPSS (16.0) software.

Contamination Measures

Adequate care was taken from the sample collection GI tracts of fishes were dissected out within a sterile condition in order to avoid cross contamination. Along with that, cotton lab coats were used to wear and clean, and acid-washed glassware was used while processing the GI tract samples. Samples were not exposed to any open environmental conditions. Types of equipment were cleaned by using 90% ethanol and kept in an aseptic condition before use. In sample processing, solvents and chemicals were prepared by using pre-filtered de-ionized water. Digestion, density separation, filtration and microscopic observations are carried out within a laminar cabinet system. Procedural blanks and controls were prepared and used to examine and minimize the aerobic contamination of MPs.

Results And Discussion

Abundances of MPs in the GI tracts of fishes

In the present study, a total of 315 MPs was isolated from the GI tracts of fishes sampled from both Rivers sites. Totally 128 MPs were isolated from GI tracts of fishes collected from the Kollidam River and 187 MPs were isolated from GI tracts of fishes collected from the Vellar River. Among the examined fishes from the Kollidam River, the maximum number of MPs were isolated from *C. chanos* (10.00 ± 5.5), followed by *C. macrolepis* (7.80 ± 3.11), and *C. nama* (13.00 ± 14). In the case of fishes sampled from the Vellar River, a higher abundance of MPs was found in the GI tract of *G. filamentosus* (22.75 ± 4.92), followed by *C. macrolepis* (14.75 ± 5.56), and *C. malabaricus* (18.50 ± 7.77) (Table 1). The MPs concentration on fish/individuals did not show any significant ($P > 0.05$) difference in-between the species collected from each sampling location. However, the total number of MPs per individual was significantly ($P < 0.05$) higher in the fishes collected from the Vellar River than that of Kollidam River, which denotes the higher amount of MPs pollution in the Vellar River compared to the Kollidam River (Table 1). The abundance of MPs in the examined fishes from this study showed that higher rates than the previous literature, on MPs in the fishes from other locations. This might be due to the small-scale examination measurements. Similarly, MPs accumulation the been reported in the GI tracts of fishes such as *C. carpio, Carassius auratus, Hypophthalmichthys molitrix, Pseudorasbora parva, Megalobrama amblycephala*, and *Hemiculter bleekeri* sampled from Taihu Lake, China which showed the MPs pollution in the sampled environments (Jabeen et al. 2017). Similarly, 10-13 MPs in the fishes such as *Dorosoma cepedianum*, *Catostomus commersonii*, *Pimephales promelas*, *Carpoides cyprinus*, *Notropis stramineus*, *Notropis hudsonius*, *Fundulus diaphanus*, *Micropterus sp.*, *Notropis atherinoides*, *Neogobius melanostomus*, and *Cyprinella spiloptera* has been noticed from the major tributaries of Lake Michigan, the USA which is similar to the outcome of our present study (McNeish et al. 2018). Likewise, the mean abundance of MPs in fish guts showed 4.3 items per grey mullet (*Mugil cephalus*) (Cheung et al. 2018), and 2.4 pieces of MPs per individual in Demersal fishes (*Evynnis cardinalis, Inegocia japonica, Repomucenus richardsonii, Solea ovata*, and *Lutjanus stellatus*) were observed from a fish farm in Cheung Sha Wan at Hong Kong, China (Chan et al. 2019). MPs ingestion in the fishes (*C. carpio, Pelteobagrus fulvidraco, Mystus macropterus*, and *Pelteobagrus* vachelli) from Lijiang River in Guangxi, Southwest China has also been studied (Zhang et al. 2021). In this study different fish species exhibited different levels of MPs ingestion which suggests that the ingestion of MPs by fishes may vary based on the size, density, and colour. Moreover, an inspection of the fish GI tract is a more realistic way instead of tissues for MPs examination. A piece of supporting evidence to this, the high level of MPs abundance in the GI tract of fishes (*Prochilodus magdalenae* and *Pimelodus grosskopfii*) has been observed than tissues (Garcia et al. 2021).

Table 1
Abundances of MPs in the GI tracts of fishes collected from the Kollidam and Vellar River

Site	Species	No. of MPs	No. of MPs/ Indiv.	F- Value	Level of Sig. (<0.05)
Kollidam	*C. chanos*	50	10.00 ± 5.5^{a}	0.460	0.644
	C. macrolepis	39	7.80 ± 3.11^{a}		
	C. Nama	39	$13.00 \pm 14^{a*}$		
	∑	128			
Vellar	*C. macrolepis*	59	$14.75 \pm 5.56^{a*}$	1.984	0.208
	G. filamentosus	91	$22.75 \pm 4.92^{a*}$		
	C.malabaricus	37	$18.50 \pm 7.77^{a*}$		
	∑	187			
∑ Kollidam + Vellar		315	Overall	2.716	0.056

Each value at the number of individuals is mean ± SD of the respective grouped fish species, (n=23) Mean values within the same column of both sites sharing same alphabetical letter superscripts are not statistically significant at P<0.05; Whereas *, denotes the significant difference of MPs in fishes between locations (one-way ANOVA and subsequent post hoc multiple comparisons with DMRT).

Lengthwise distribution of MPs in the GI tract of fishes

The mean length of 128 MPs isolated from the fishes *C. nama, C. chanos* and *C. macrolepis* from the Kollidam River was recorded. The length of MPs lay within the ranges from 32.14 to 267.083 μm, 11.602 to 273.389 μm, and 18.40 to 333.916 μm in *C. nama, C. chanos* and *C. macrolepis* respectively. In the case of MPs isolated from fishes *C. macrolepis, G. filamentosus* and *C. malabaricus* sampled from Vellar River had shown length between 13.01 to 506.248 μm, 19.404 to 377.536 μm, and 19.637 to 487.611 μm respectively (Table 2). In this study, the mean length of MPs isolated from fishes of Kollidam River showed an insignificant (P > 0.05) difference when compared to each other. Whereas, mean length of MPs isolated from *G. filamentosus* showed a significant (P < 0.05) increase when compared to MPs isolated from other fishes from the Vellar River. However, the mean length of MPs isolated from *C. macrolepis* and *C. malabaricus* showed an insignificant (P > 0.05) difference sampled from the Vellar River (Table 2) (Figure 5 and 7). Moreover, the mean length of MPs was significantly (P< 0.05) higher in the fishes collected from the Vellar River than that of Kollidam River which indicates the ingestion of a high range of MPs by fishes in the Vellar River compared to the Kollidam River (Table 3; Fig. 5, 6 and 7). Also, the much deviation in the MPs isolated from the GI tract of fishes indicated that the high rate of fragmentation led to ingestion by fishes as false feeding in the sampled environments. Similar to our study, 0.3 to 0.6 mm sized MPs has been observed in the fishes *C. Carpio, Carassius cuvieri, Lepomis macrochirus, Micropterus salmoides, Silurus asotus*, and *Channa argus* from Han River, South Korea (Park et al. 2020). A report from South-West Nigerian Eleyele Lake revealed that the fishes (*Coptodon zillii* and *Oreochromis niloticus*), ingested about 124 and 126 μm sized MPs (Adeogun et al. 2020). Also, the majority of MPs in the GI tracts of *Oreochromis niloticus, Clarias gariepinus*, *C. carpio*, and *Carassius carassius* at Lake Ziway, Ethiopia which had a length between 0.2 to 5 mm (Merga et al. 2020). Microplastics from the GI tracts of fish *C. auratus* caught from the Poyang Lake, China, has shown majorly above 0.5mm length (Yuan et al. 2019). The size of MPs ranged between 1.2 to 4.68 mm in the GI tract of different marine fish species from the Red Sea, Saudi Arabia has been reported earlier (Baalkhuyur et al. 2018). In the present study, the size of MPs is much lower than the above-cited previous studies which suggested that the continuous fragmentations of plastics led to a higher ingestion rate. Moreover, the smaller plastics particles can serve as a vector for carrying toxic metals to the organisms, and size determination factors that also determine the metal pollution carriage by the MPs (Hildebrandt et al. 2021).

Table 2
Length-wise distribution of MPs among the fish species collected from the Kollidam and Vellar River

Location	**Species**	**No. of MPs**	**Length of MPs (μm)**			**F- Value**	**Sig.(P<0.05)**
			Minimum	**Maximum**	**Mean length**		
Kollidam	*C. chanos*	50	18.404	333.916	120.66 ± 72.62^{a}	0.754	0.473
	C. macrolepis	39	11.602	273.389	108.72 ± 74.05^{a}		
	C. nama	39	32.14	267.083	$129.11 \pm 75.01^{a*}$		
Vellar	*C. macrolepis*	59	13.01	506.248	$201.993 \pm 97.75^{b*}$	5.694	0.004
	G. filamentosus	91	19.404	377.536	$284.233 \pm 83.26^{a*}$		
	C. malabaricus	37	19.637	487.611	$181.29 \pm 95.43^{b*}$		
Overall						2.716	0.056

Each value at the number of individuals are mean ± SD of the respective grouped fish species, (n=23) Mean values within the same column of microplastic mean length sharing same alphabetical letter superscripts for the Kollidam site are not statistically significant at (P>0.05) and the mean values of the MPs length of Vellar river sharing different alphabetical letter superscripts are statistically (P<0.05) significant; Whereas *, denotes the significant difference of MPs in fishes between locations (one-way ANOVA and subsequent post hoc multiple comparisons with DMRT).

Table 3
Morphological distributions of MPs in the GIT of fish species from the Kollidam and Vellar River according to their Shape

Location	Species	No. of MPs	Shape of MPs	
			Fibre	Fragment
			No. of MPs/Fish	No. of MPs/Fish
Kollidam	*C. chanos*	50	13	-
			3	1
			13	2
			13	1
			4	-
	C. macrolepis	39	8	5
			3	2
			6	1
			4	2
			5	3
	C. nama	39	25	4
			3	-
			7	-
	Total	128	107	21
Vellar	*C. macrolepis*	59	12	-
			7	4
			11	12
			13	-
	G. filamentosus	91	21	-
			19	-
			27	3
			20	1
	C. malabaricus	37	12	1
			21	3
	Total	187	163	24
Overall percent			85.70%	14.30%

Values of each shaped microplastic fibres and Fragments were provided with their percentage for both Kollidam and Vellar Rivers

Morphological distribution of MPs in the GI tract of fishes

In the present study, the morphological distribution of MPs indicated that the fibres were the dominant shapes in the GI tract of fishes sampled from both Rivers which constituted nearly 85.71% of the total 315 MPs, followed by fragments with 14.29%. MPs isolated from fishes belonging to the Kollidam River was categorised as fibres and fragments with 83.59% and 16.41% respectively, Among the fish species from the Kollidam River, *C. chanos* had the maximum number of fibres, followed by *C. nama* and *C. macrolepis*, whereas, the fragments were found to be higher in the order of *C. macrolepis* > *C. chanos* > *C nama* (Table 3; Fig. 8). In Vellar River, the GI tract of *G. filamentosus* showed a maximum level of fibres, followed by *C. malabaricus* and *C. macrolepis*, whereas, the fragments level was maximum in the GI tract of *C. macrolepis* compared to other fishes (Table 3; Fig. 8) In the present study, a maximum level of fibres suggests that the mismanaged fishing nets and gears are the major source for the MPs invasion in fish guts. Rivers can be a major contributor to the discharges of plastic litter into marine environments (Napper and Thompson 2016; Roch et al. 2019). The earlier study also indicates that the abundance (>80%) of MPs fibres in the GI tracts of fish *Rutilus rutilus* sampled from River Thames in the UK (Horton et al. 2018). The fish *Gambusia holbrooki* from wetlands of Melbourne, Australia ingested MPs were majorly found in the fibre shape (Su et al. 2019), which is similar to our present study. Similarly, the alien fish *Piaractus brachypomus* sampled from Ramsar wetland Vembanad Lake, Kerala India was accounted for 50% of fibres in their GI tracts (Devi et al. 2020). Report on the fish species *Platichthys flesus* and *Osmerus eperlanus* collected from the River Thames, London was revealed higher fibre particles rather than other types of MPs (McGoran et al. 2017). Also, the GI tracts of fishes (*Esox lucius*, *Catostomus commersoni*, *Notropis atherinoides*, *Pimephales promelas*, and *Eucalia inconstans*) were sampled from Prairie Creek in Saskatchewan, Canada showed an elevated level of fibres and fragment shaped MPs (Campbell et al. 2017). Defragmentation of plastic wastes into secondary MPs are the major contributor to the plastic litters of the rivers. In the study areas of the present study, the fibre shaped MPs were occupied a higher level of distribution in the GI tract of all sampled fish species, this might be due to plastic debris originating from commercial fishing gears and urban activities.

Colour pattern of MPs in between in the GI tract of fishes

In the current study, the colour-wise distribution of MPs in fish guts showed major differences in their pattern of accumulation among fish species of both Kollidam and Vellar Rivers. The blue-coloured MPs, followed by transparent colours were dominant in the GI tract of fishes sampled from both Rivers (Table 4; Fig. 9). MPs in the GI tract of fishes sampled from Kollidam River showed the colours in the order of Blue > transparent > red > white > yellow, whereas, the colour pattern of MPs in the fishes sampled from Vellar River showed in the order of blue > transparent > red > yellow > white (Table. 4 and Fig. 9). In this study, the dominant blue colour of MPs in the GI tract of fishes suggests that the false feeding of fishes on fragments of fishing gears that look like copepods. The blue colour of MPs was similar to the copepods in aquatic conditions, which led to more false feeding by fishes (Ory et al. 2018). The remaining-coloured MPs might be direct false feeding or transferred from other prey. Similarly, a different colour pattern of MPs has been reported in fish species including *Argyrosomus regius, Caranx crysos, Dentex dentex* sampled from the Mediterranean Sea Turkey (Güven et al. 2017). Previously on fish species of *Dicentrarchus labrax, Diplodus vulgaris, Platichthys flesus* from Mondego estuary western coast of Portugal showed dominant blue, transparent and black coloured MPs particle (Bessa et al. 2018). Colour pattern mediated alterations in the ingestion rate of MPs has been reported in fish species such as *Myctophum aurolanternatum, Symbolophorus californiensis, Cololabis saira, Hygophum reinhardtii, Loweina interrupta*, and *Astronesthes indopacifica* collected from North Pacific Central Gyre (Boerger et al. 2010). The colour distribution blue, clear, yellow, grey, green, red, black, and white of ingested MPs in fishes *Coregonus wartmanni, Lota lota, Perca fluviatilis, Alburnus alburnus, Gymnocephalus cernua*, *Squalius cephalus*, *Rutilus rutilus*, *Gasterosteus aculeatus*, *Abramis brama*, *Esox lucius*, *Stizostedion lucioperca*, *Leuciscus leuciscus*, *and Barbatula barbatula* from the lakes and river sampling sites located at Baden-Württemberg (Germany) which are similar to the present study (Roch et al. 2019). Further, different proportions of colours on ingested MPs from the GI tracts were observed on fish species of *Oreochromis niloticus* and *Cirrhinus molitorella* from the Rivers of Guangdong province, south China (Sun et al. 2021). The colour distribution of MPs can greatly affect the ingestion rate in fishes due to the difficulty in differentiating the feeds. Moreover, transparent MPs are non-identical in the aquatic systems and blue colour is predominately used in fishing nets and their different colours and buoyance have resembled the planktonic feeds.

Table 4

Colour distribution of MPs extracted from the GI tracts of fishes from both Kollidam and Vellar River

Study site	Species	Color of MPs											
		Transparent		Blue		Red		Black		White		Yellow	
		No	%	No.	%	No.	%	No	%	No.	%	No.	%
Kollidam	*C. chanos*	1	22	8	48	4	18	-	4	-	4	-	4
		1		2		-		1		-		-	
		3		9		1		1		1		-	
		4		5		4		-		1		-	
		2		-		-		-		-		2	
	C. macrolepis	2	17.94	5	35.89	-	10.25	-	12.82	5	15.4	1	7.7
		1		3		-		-		1		-	
		3		3		1		-		-		-	
		-		1		2		1		-		2	
		1		2		1		4		-		-	
	C. nama	-	5.12	7	30.76	5	12.28	8	23.07	5	15.4	4	2.83
		-		3		-		-		-		-	
		2		2		-		1		1		1	
Total		24	18.75	50	39.06	18	14.06	16	12.5	14	10.93	6	4.68
Vellar	*C.macrolepis*	-	18.64	10	49.15	-	8.47	-	19.94	-	-	2	6.8
		3		4		2		1		-		1	
		8		9		1		4		-		1	
		-		6		2		5		-		-	
	G. filamentosus	9	26.37	2	28.57	3	9.89	6	29.67	-	1.10	1	4.4
		4		8		2		4		-		1	
		9		3		1		16		-		1	
		2		13		3		1		1		1	
	C. malabaricus	3	21.62	7	43.24	3	18.91	-	10.81	-	-	-	5.4
		5		9		4		4		-		2	
Total		43	22.99	71	37.96	21	14.22	41	5.88	1	0.53	18	9.62
Overall		67	21.26	121	38.41	39	12.38	57	8.57	15	4.76	24	7.6

Values of each coloured MPs extracted from the fishes collected from both Kollidam and Vellar River along with their Percentage.

SEM- analysis of extracted MPs

The surface morphological characteristics of MPs was carried out with a cluster of MPs picked up from the filter obtained from the extraction step. Fig. 10a denotes the complex surface topography of the extracted MPs which indicates that the linear strings due to the heavy of accumulation of fibres in the GI tracts of fishes collected from both sites. Surface of the MPs strings were convex, rough non-porous and with many folds. These strings were appeared in irregular in shape, brittle body with both sharp and blunt edges, these damages may be caused by the environmental factors like continuous mechanical disturbances caused by water current flow in the river and photo-oxidative weathering of MPs caused by UV radiation etc (Kalogearkis et al. 2017; Ding et al. 2019; Zbyszewski et al. 2014). The damages and peeled spots were also observed on the surface of the MPs strings in the FE-SEM analysis (Fig. 10b).

Conclusions

Results of the present study revealed that the overall occurrence of MPs per individual fish, mean length of MPs were significantly higher in the GI tract of fishes sampled from the Vellar River compared to fishes sampled from the Kollidam River which indicates the higher level of microplastic pollution in Vellar River than that of Kollidam River. Further, the fibre content was higher in the GI tract of fishes sampled from both Rivers, it suggested that the mismanaged fishing gears might be the major sources of fibres in the GI tract of fishes. Therefore, these findings suggest that the MPs were added to the diets of fishes, which may carry to the next level of the food chain in the ecosystems with potential risks. Moreover, both Kollidam and Vellar Rivers are the sources of the Bay of Bengal Sea, and they can serve as a carrier of pollutants along with their water current, which may carry the plastic litter into the marine environment which seems to be serious threats to the marine biota.

Declarations

Ethics approval: This paper does not need any ethical approval.

Consent to participate: Not Applicable

Consent to Publish: Not Applicable

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Data availability statement: Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

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Figures

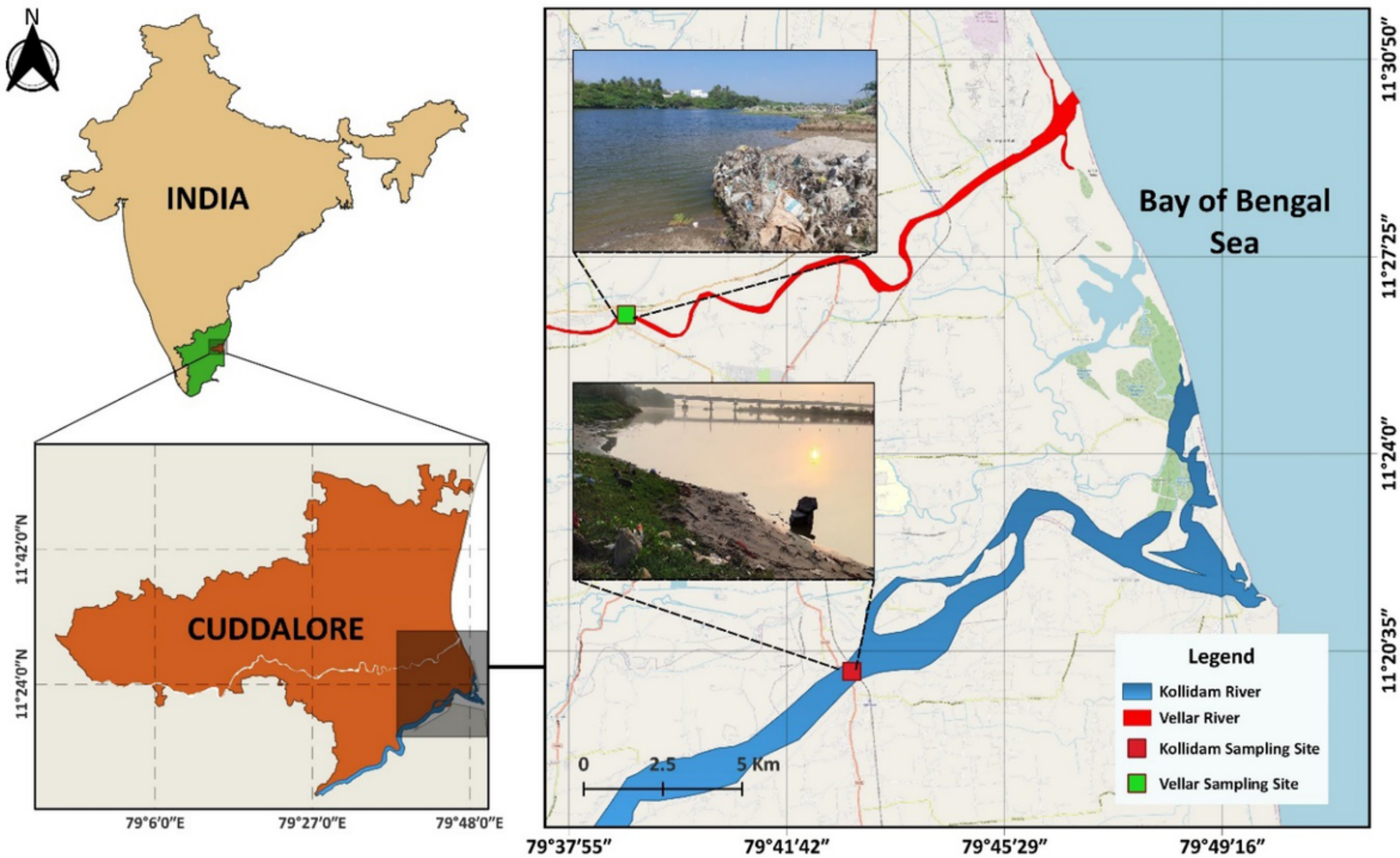

Figure 1

Geographical data of the selected study sites of both Vellar and Kollidam Rivers

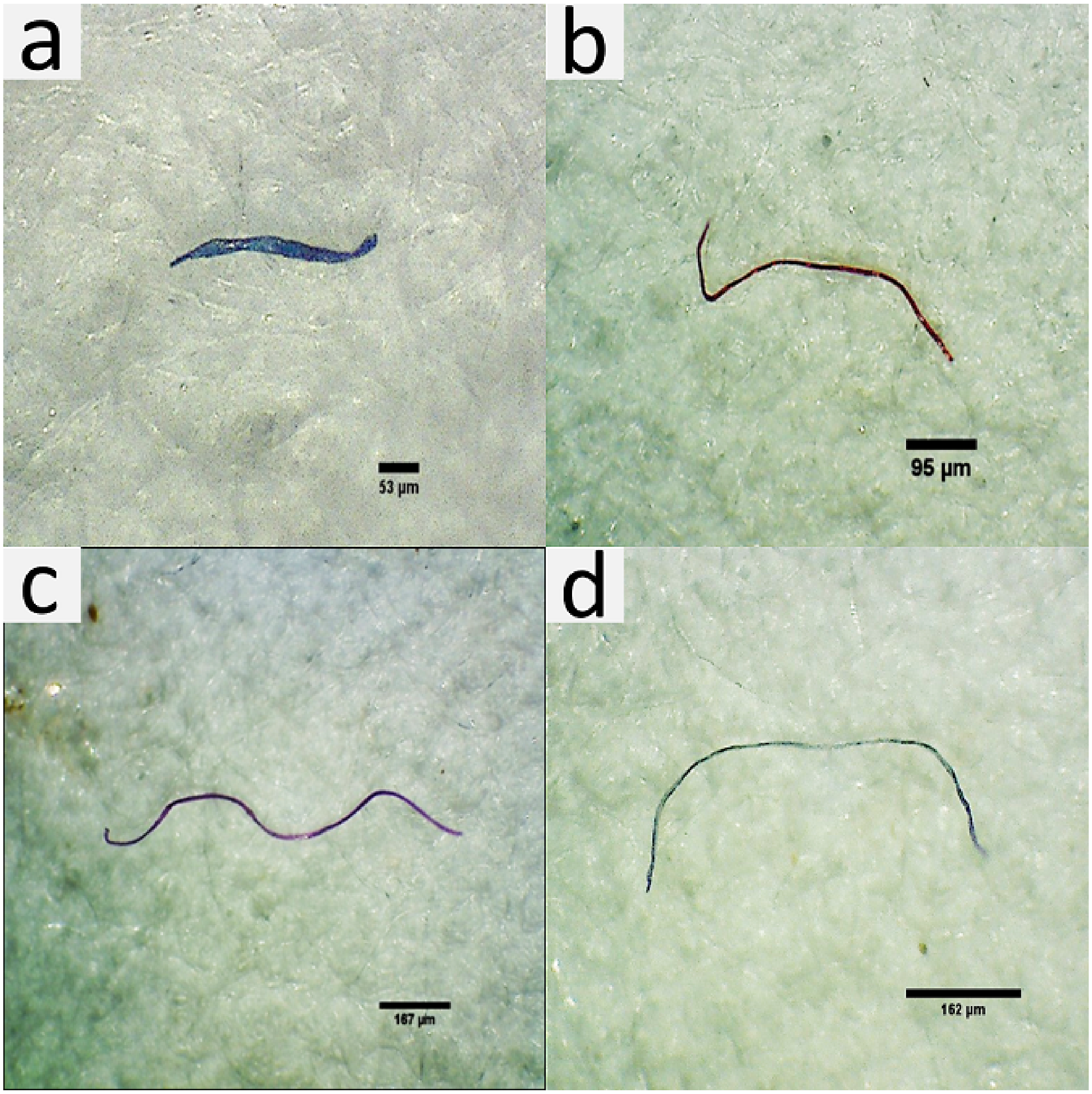

Figure 2

Extracted MPs from the GI tracts contents of the fish *C. chanos* sampled from the Kollidam River site, **(a)**, **(c)** and **(d)** are Blue Fibres, **(b)** Red Fibre

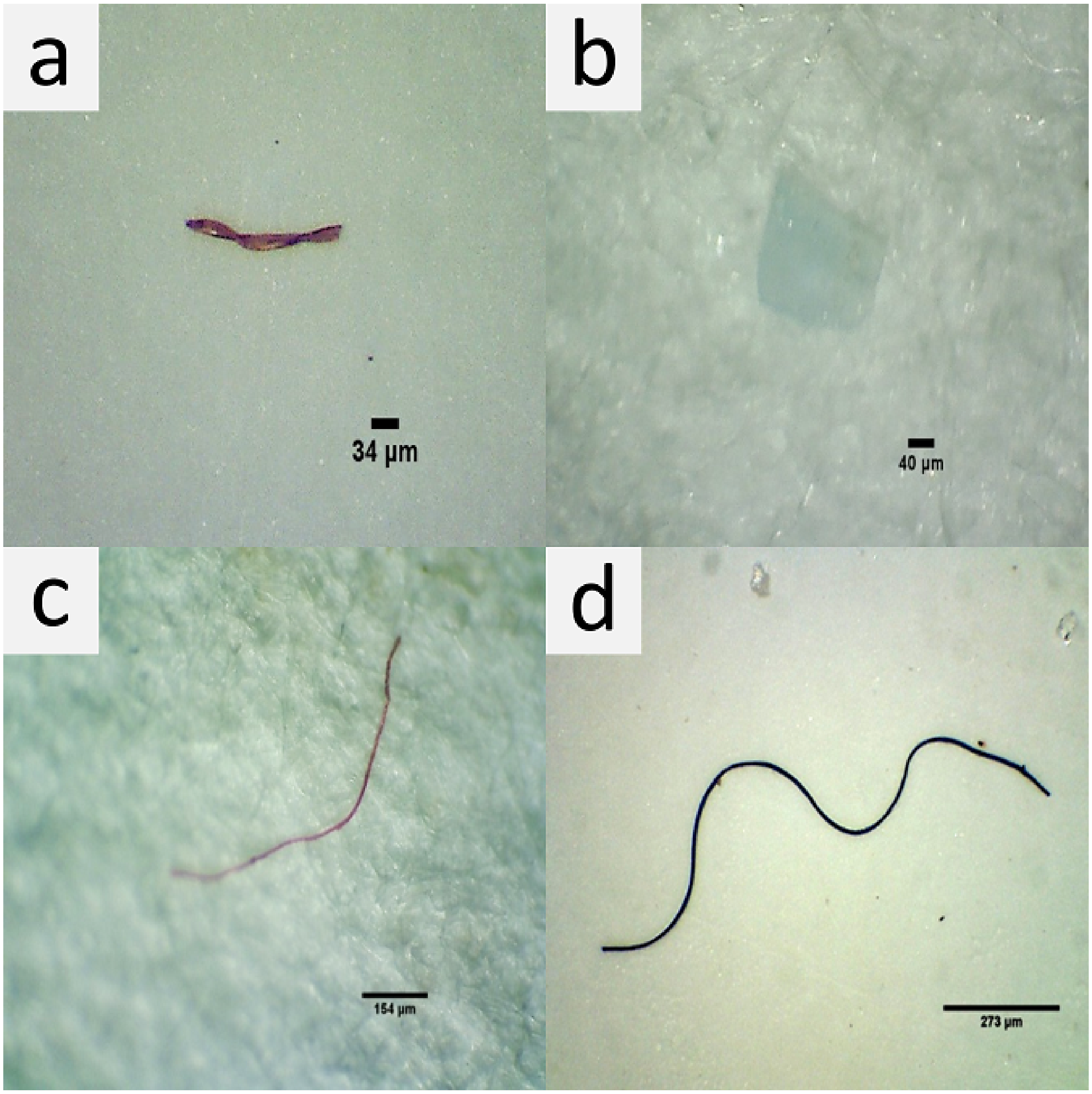

Figure 3

Extracted MPs from the GI tracts contents of the fish *C. macrolepis* sampled from the Kollidam River site, **(a)**, **(c)** are Red Fibres **(b)** Transparent Fragment, **(d)** Black Fibre

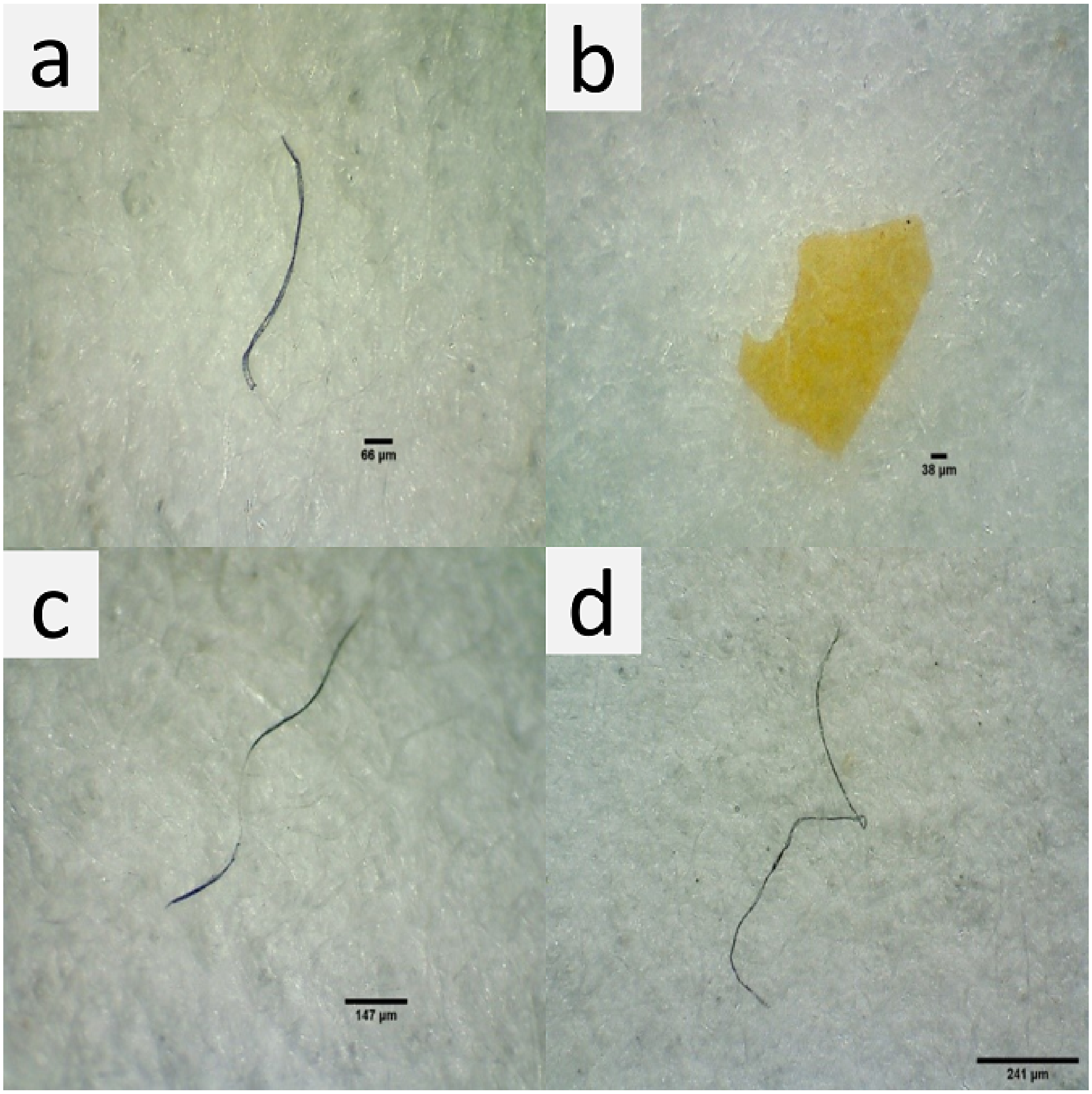

Figure 4

Extracted MPs from the GI tracts contents of the fish *C. nama* sampled from the Kollidam River site, **(a)**, **(c)** are Blue Fibres **(b)** Yellow Fragment, **(d)** Black Fibre

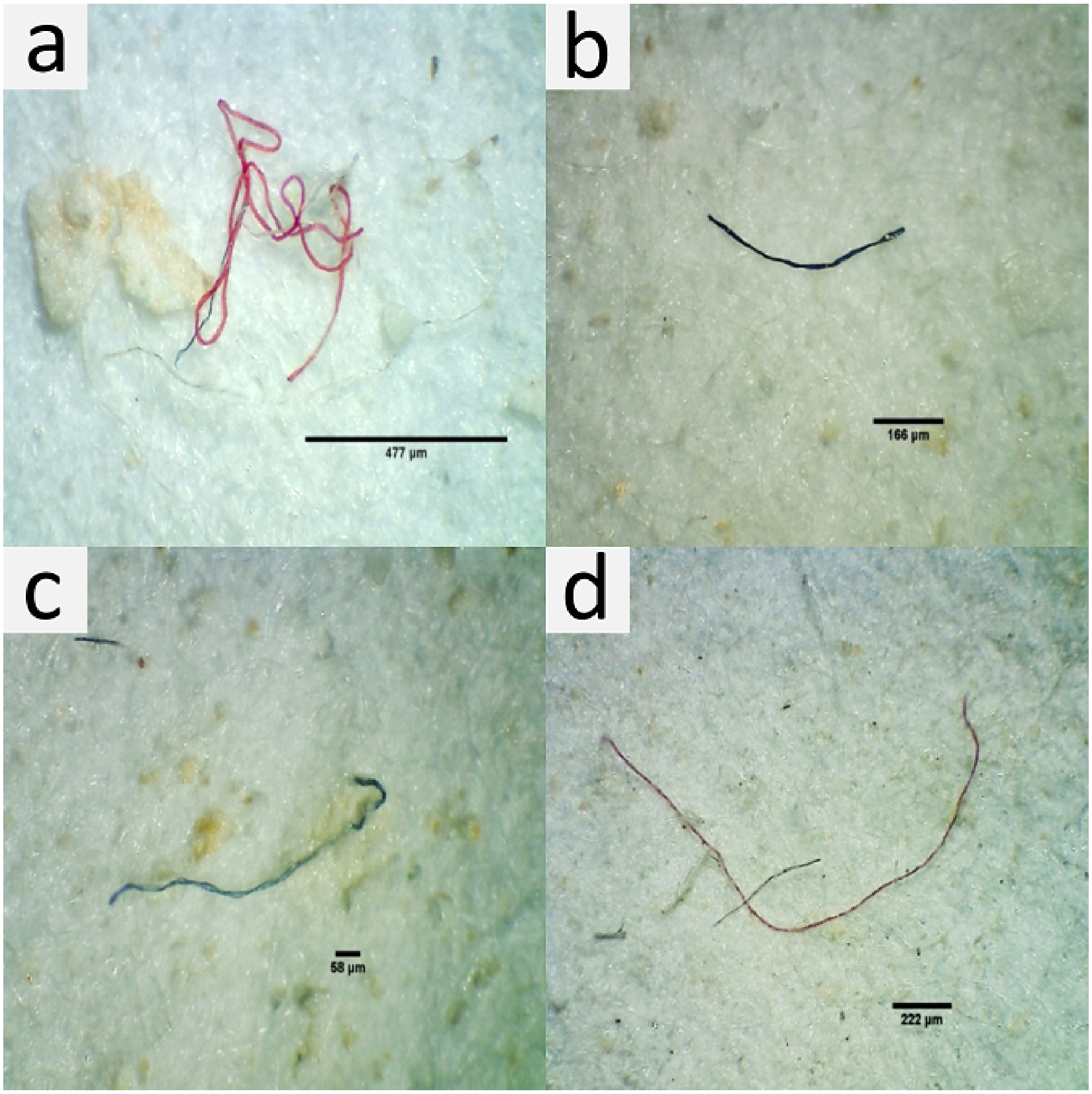

Figure 5

Extracted MPs from the GI tracts contents of the fish *C. macrolepis* sampled from the Vellar River site, **(a)**, and **(d)** are Red Fibres, **(b)** Black Fibre, **(c)** Blue Fibre

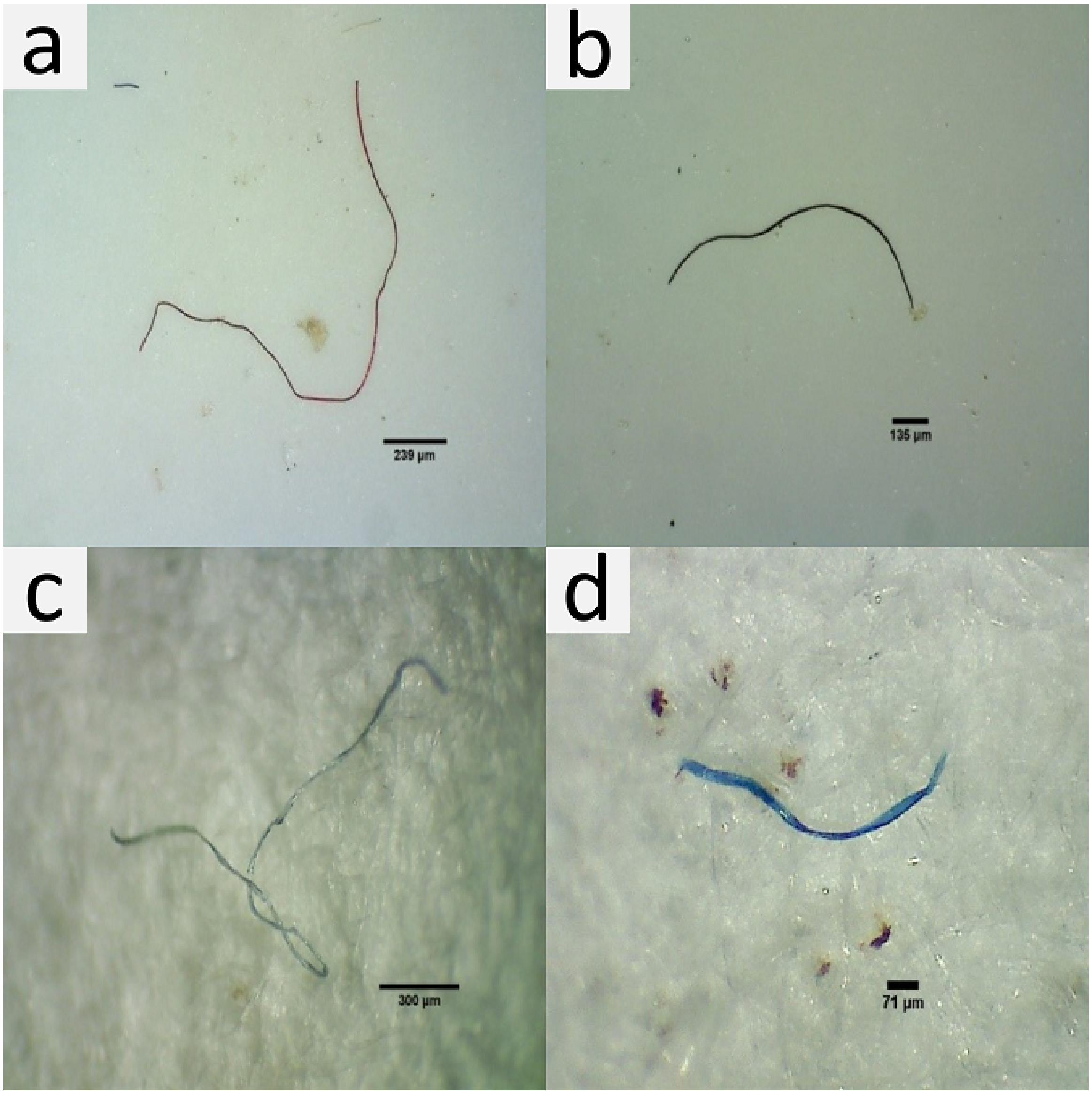

Figure 6

Extracted MPs from the GI tracts contents of the fish *G. filamentosus macrolepis* sampled from the Kollidam River site, **(a)** Red Fibre, **(b)** Black Fibre **(c)** Transparent Fibre, **(d)** Blue Fibre

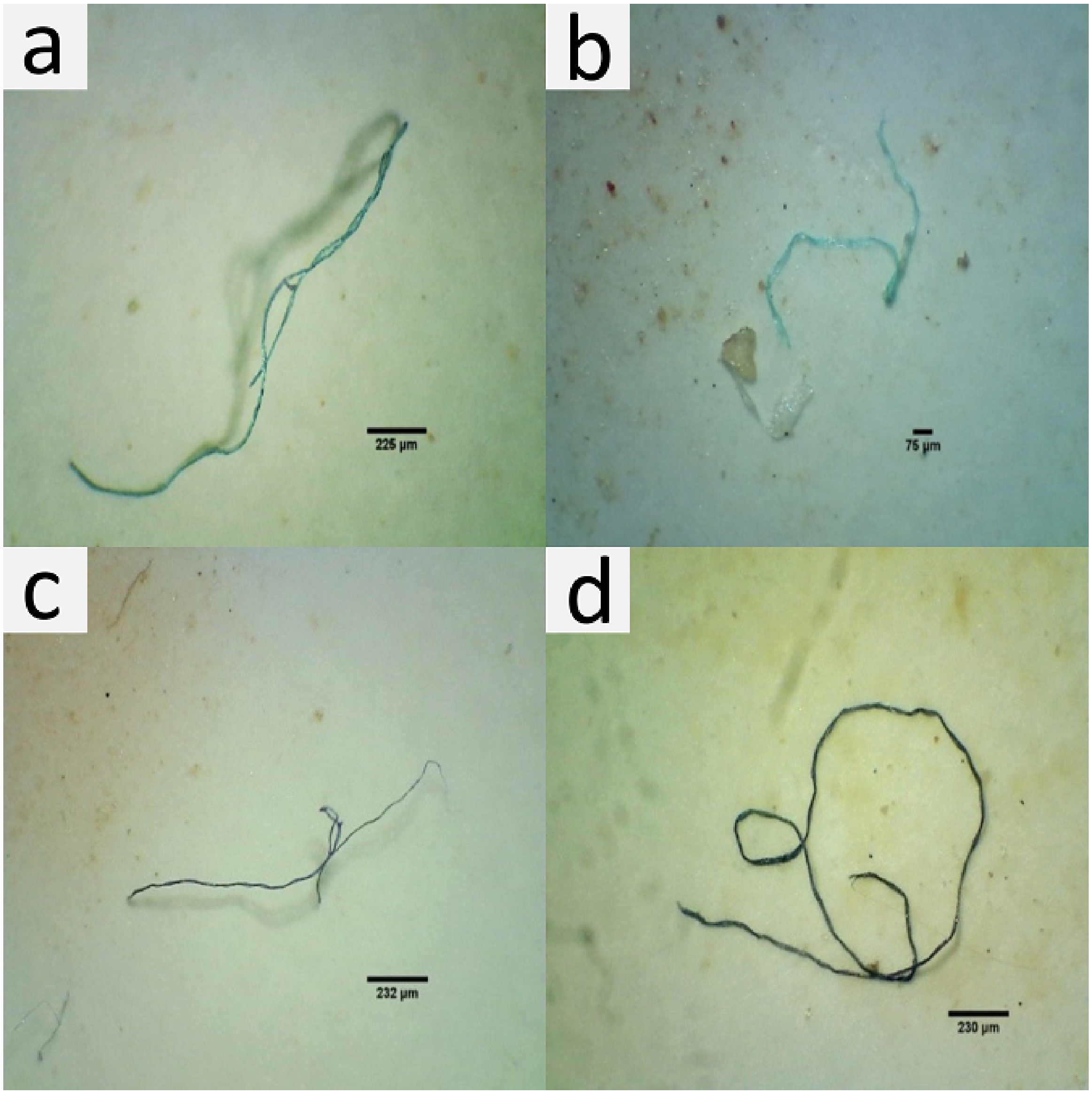

Figure 7

Extracted MPs from the GI tracts contents of the fish *C. malabaricus* sampled from the Kollidam River site, **(a)**, **(b)** and **(c)** are Blue Fibres, **(d)** Black Fibre

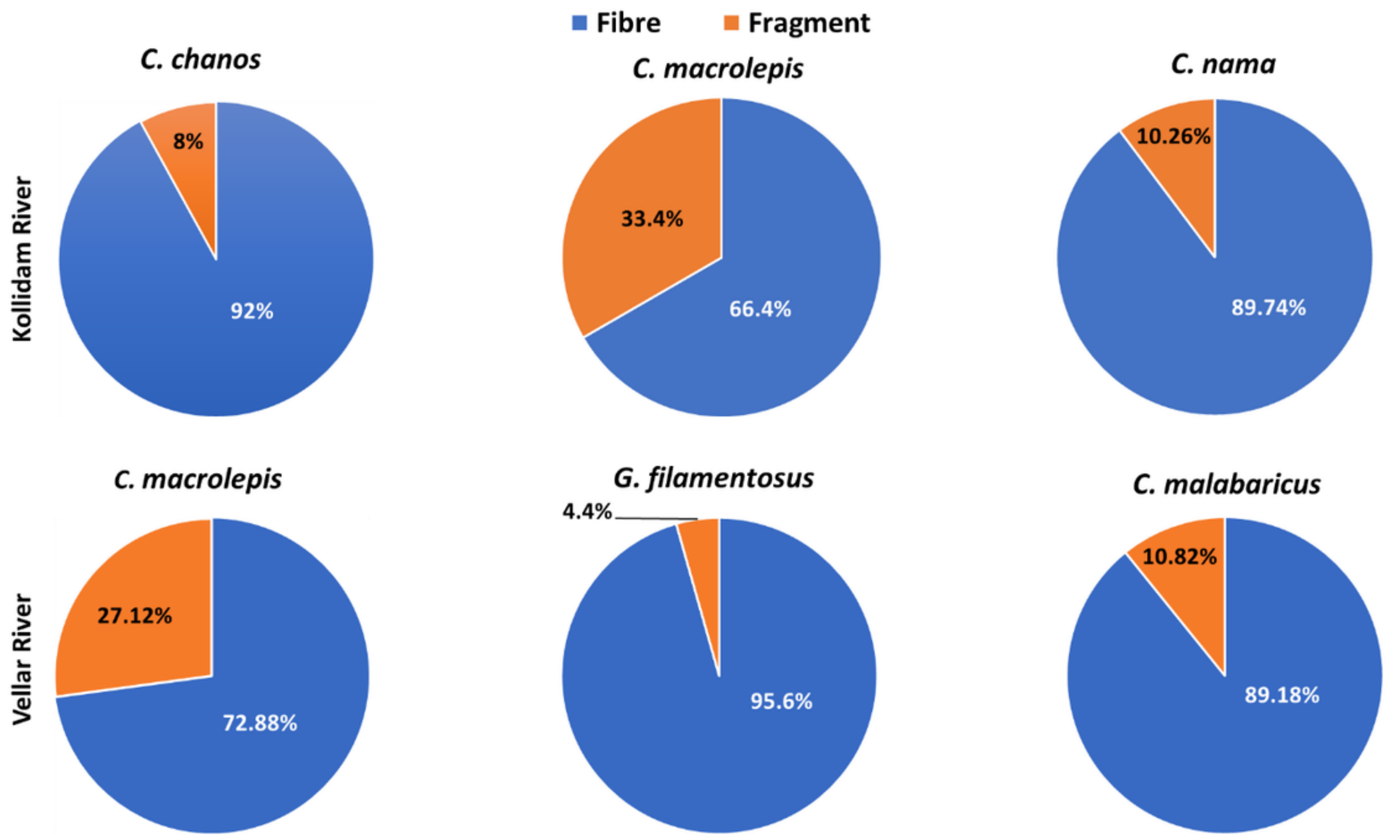

Figure 8

Shape Distribution of MPs in the fish species collected from the Kollidam and Vellar Rivers

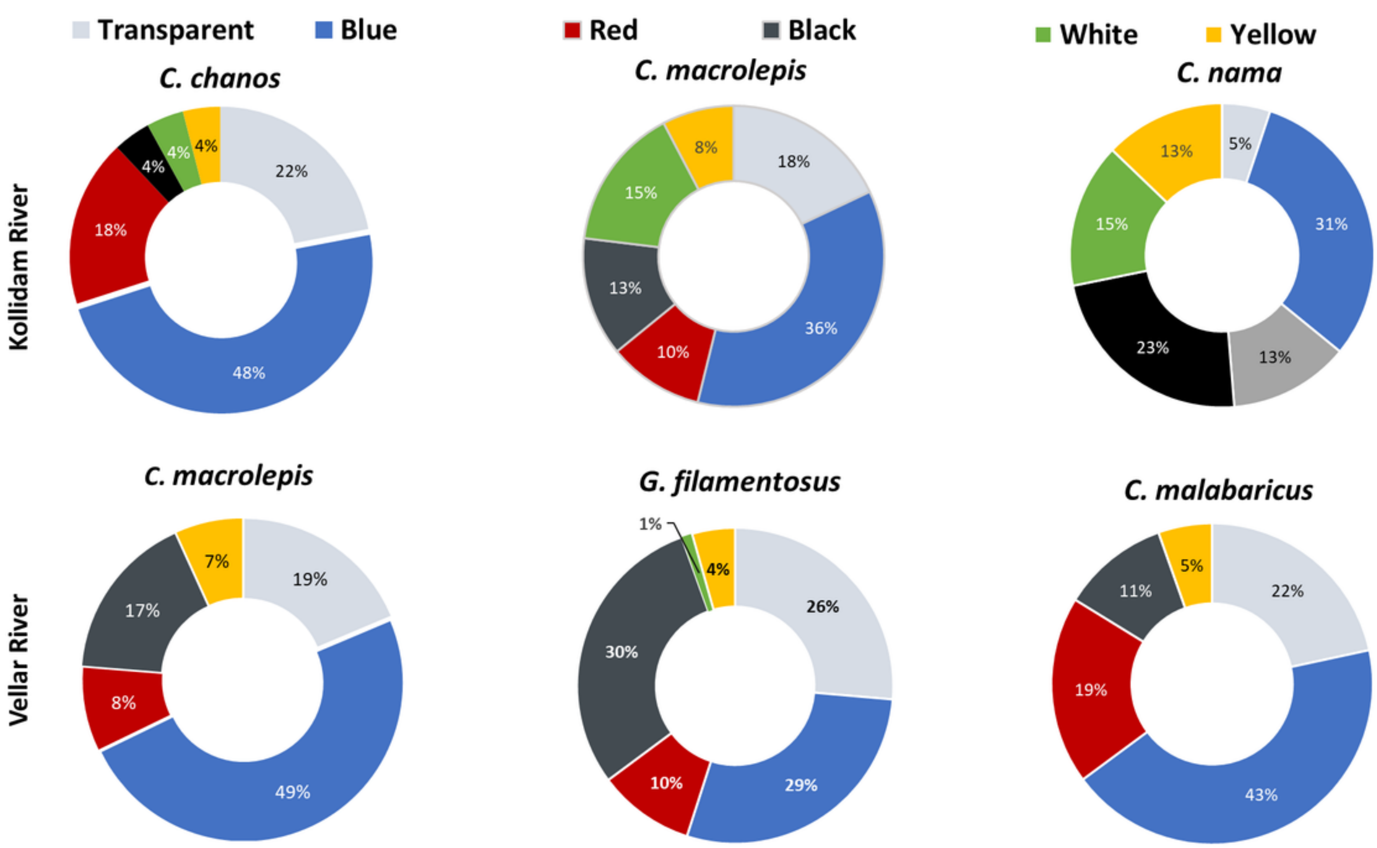

Figure 9

Colour pattern of MPs in the fish species collected from the Kollidam and Vellar Rivers

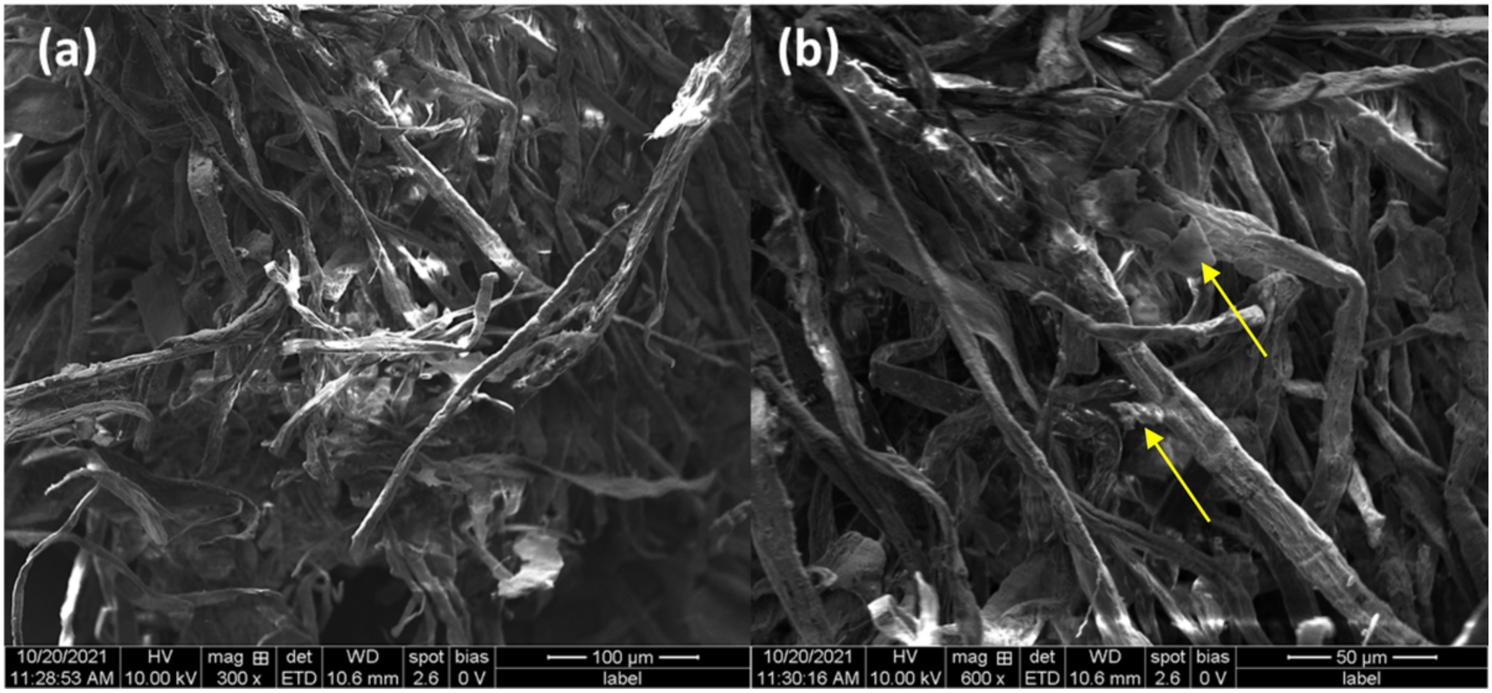

Figure 10

FE-SEM image of isolated MPs in the GI of fish species collected from the Kollidam and Vellar Rivers. (a) Cluster of MPs strings (b) Damaged surface parts of MPs strands marked as yellow arrow