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36 **ABSTRACT**

37 An emerging concern of today's world, due to their universal dispersion worldwide, is the
38 environment's microplastic pollution. The Sundarbans, the world's largest mangrove, have
39 unique and dynamic environmental settings with numerous pollution risk exposures,
40 including microplastics (MPs). Thus, the present study has focused on the MP pollution in
41 water, sediment, and fish samples of the Sundarbans of Bangladesh for the first time. Water
42 and sediment samples were collected ($n=30/\text{each}$) from sampling locations along the Pasur
43 river (Bangladesh). Furthermore, nine species of fish samples were collected from a local
44 fish market situated at the Mongla port. Results show that 100% of the analyzed samples
45 have evidence of MPs. On average, 2.66×10^3 plastic particles/L and 1.57×10^5 particles/kg
46 were found in water and sediment samples, respectively. Furthermore, results show a
47 higher number of MPs in the animals' gastrointestinal tract (GIT) (10.41 particles/g),
48 concerning the average concentration recorded in the muscles (4.68 particles/g). *O. pama*
49 and *H. nehereus* were the species that showed the highest MPs accumulation in the GIT. In
50 the muscles, the highest MP levels were observed in *T. ilisha* and *L. calcarifer*. Most of the
51 particles were smaller than 1 mm; black-colored particles dominated the pool. FT-IR
52 analysis revealed the presence of seven polymer types where polyamide was abundant in
53 water and sediment samples. SEM analysis showed morphological structures and adsorbed
54 particles on the surface of plastic samples, and the spatial distribution of MPs indicates that
55 the location with high human intervention has elevated levels of MPs. Therefore, our study
56 demonstrates that Sundarbans mangrove forests are highly contaminated with MPs and
57 that its fisheries can be a potential source of human exposure to these pollutants.

58 **Keywords:** Microplastics, mangrove, sediment, polymers, FT-IR, SEM, spatial distribution.

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63 **1. INTRODUCTION**

64 Microplastics (MPs) have been established as one of the most concerning problems in
65 today's world. It is used in everyday commodities for its lower cost and broader applications
66 (Thompson et al., 2009, Cordova et al., 2021). It has been stated in most common
67 environments and rare places on the earth (Barnes et al., 2009). A ubiquitous element of the
68 environment, the particles of plastic known for their microscopic sizes are microplastics,
69 which are widely used in industrial manufacture (Cole et al., 2011; Vermaire et al., 2017). It is
70 an emerging synthetic plastic pollutant of less than 5 mm to not more than 1 micrometer in
71 size that needs to be tackled in no time (Afrin et al., 2020; Guo et al., 2020; Lebreton et al.,
72 2018; Thompson et al., 2004). It is a general term for microscopic plastic particles, including
73 their polymer types, size, origin, and shapes (Laermanns et al., 2021).

74 MPs are formed by the breakdown and fragmentation of larger plastic particles due to
75 various processes like waves and UV radiation from the sun (LI et al., 2016). The larger plastic
76 particles are less harmful than the microplastics as these can hamper microscopic organisms,
77 which further may build up in higher organisms by adopting bioaccumulation mechanisms
78 (Mohamed Nor & Obbard, 2014). MPs can be of many diverse types. However, depending on
79 their origin, they are mainly divided into primary and secondary types (Garcés-Ordóñez et
80 al., 2020). Primary microplastics are micro-sized particles typically used in cosmetic products
81 and medicines and reported by some studies in air blasting technologies (Cole et al., 2011;
82 Derraik, 2002; Patel et al., 2009). They are derived from synthetic textiles, car tires, and
83 products used to manufacture other plastic items (Duis & Coors, 2016). When these plastic
84 particles break down even further, the smaller particles are known as secondary
85 microplastics, which are generally used in agricultural sectors and industrial processes;
86 these are also derived from primary plastic particles, illegal waste dumping, and
87 mismanagement of sanitation (Ryan et al., 2009; Thompson et al., 2004; Afrin et al., 2020;
88 Barnes et al., 2009; Deng et al., 2021). Weathering plays a role in changing MPs' size, shape,
89 and polymer structures (Lehtiniemi et al., 2018). According to their shapes, microplastics can
90 be of several types, of which the most common are fragment, fiber, film, pellet, foam, etc.
91 (Arias et al., 2019; Lambert et al., 2017; Wu et al., 2018). Their chemical bonds and polymer
92 types can be distinct and identifiable using infrared radiation. Polyethylene (PE), Polystyrene
93 (PS), polypropylene (PP), high-density polyethylene (HDPE), low-density polyethylene
94 (LDPE), Polyvinyl chloride (PVC), etc., are some of the common types of microplastics found
95 in the environment (Rakib et al., 2022).

96 The MP pollution issue applies to Bangladesh, especially in the Sundarbans mangrove
97 forest. It is the world,' yet it is affected by the intrusion of microplastics in every
98 environmental aspect. Mangrove forests are the intermediate area adjoining terrestrial lands
99 with the coastlines, which nurtures various life forms of land and water (Deng et al., 2021;
100 Zuo et al., 2020). Mangroves are significant in the sense of food providers to diverse life forms
101 and as a protection from natural disasters (Bayen, 2012; Kulkarni et al., 2018). The
102 Sundarbans support diverse local and exotic wildlife; it is also home to many people who
103 depend on the forest for their food and livelihood. Recently microplastic pollution has been
104 established as a global concern as well as a national concern from the perspective of our
105 country (Parvin et al., 2021). Although a significant sink of microplastics, there have been
106 fewer studies involving mangrove forests, presumably due to the hostile environment, risk
107 of wildlife, and sampling challenges (Mohamed Nor & Obbard, 2014). But this has only made
108 it more significant to identify the elements contaminated by MPs.

109 In addition to the lack of research concerning the mangrove environment, especially
110 the Sundarbans, little and unclear information was found on the spatial distribution of the
111 MPs. MPs in sediment have very limited research when compared with aquatic MPs. No
112 universal method was developed for the sampling and extracting of MPs from soil and
113 sediments. The Fourier transform infrared spectroscopy (FTIR) analysis of polymer types,
114 and their peaks are also not evident in the studied research. There are assumptions that MP
115 sources from the atmosphere; however, very little research has been done.

116 As microplastic pollution has gained attention recently, it is plausible that the largest
117 mangrove forest, Sundarbans, has also been affected by its presence. The study hypothesizes
118 that a higher number of MP particles are present in the Sundarbans mangrove forest's
119 sediment, water, and aquatic life forms. Specifically, the study aims to identify and quantify
120 the presence of MPs in the sediment, water, and fish samples of the Pasur river, to
121 characterize the identified MPs according to their shapes, sizes, and polymer types; and, to
122 represent the spatial distribution of microplastics in the study area.

123

124 **2. MATERIALS AND METHOD**

125 **2.1. Study area**

126 Being the world's largest delta blessed with mangrove forests, Sundarbans is located
127 on the mouth of Brahmaputra, Ganges, and Meghna, three mighty rivers (Getzner & Islam,
128 2020). The total forest area is about 10,284 km, with 58.5% in Bangladesh (Chowdhury et

al., 2016). Sundarbans is located between 22.29713E and 89.56718 N. About 117 river flows over Sundarbans to mix with the Bay of Bengal, of which, Pasur is one of the major river systems in Sundarbans, an extension of the Rupsa River. Pasur river is about 48 km south of Khulna district (Shil et al., 2017). The left bank of the river falls within the Khulna district, and the right side is in the Bagerhat district (Figure 1). Plastic pollution is also increasing with the high tourism load and recent developments. Microplastics of a higher concentration could alter the ecosystem and damage the natural balance of the forest, including the food web and ecosystem (Hitchcock, 2022). With all these in mind, the Sundarbans mangrove forest has been chosen as the primary area of this thesis.

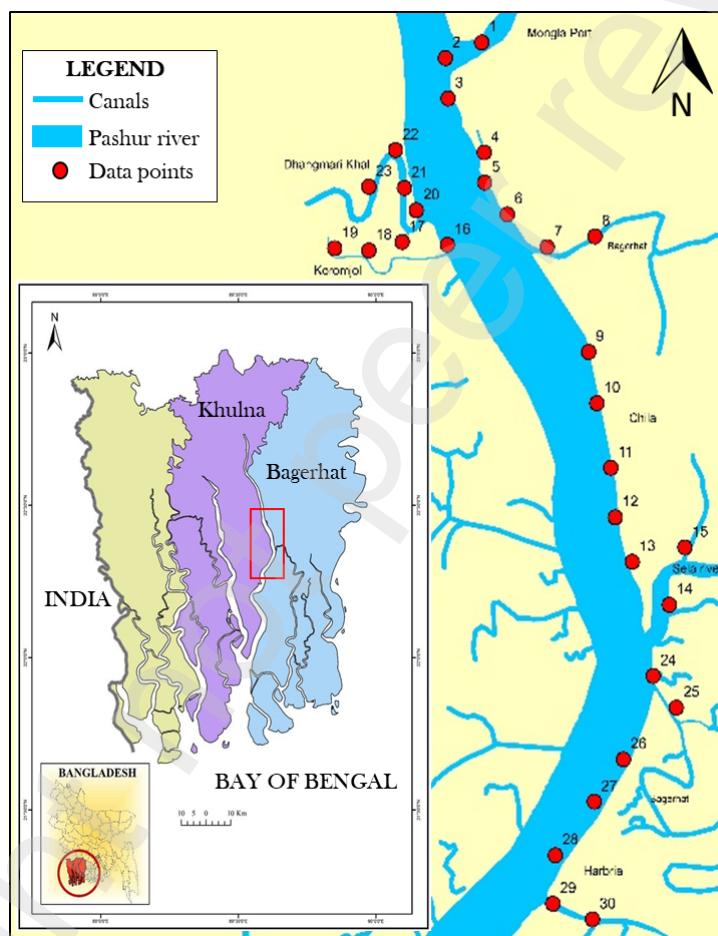


Figure 1. Data collection locations in the Pasur river and its adjacent canals (Bay of Bengal, Bangladesh).

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139 **2.2. Sampling and data collection**

140 During the study in mid-February 2022, sediment and water samples were collected
 141 from the Pasur river of Mongla city (Shil et al., 2017). Thirty sampling locations from Mongla
 142 port to Herbaria were selected beforehand, keeping approximately 1 km distance between

143 each point (Figure 1). A record of the GPS location, time, and name of the local place with a
144 short description of the area was kept during the sample collection. All samples were
145 collected during the daytime for two days, facing both high and low-tide situations. Sixteen
146 samples were collected from the mainstream river, while 14 samples were taken from
147 adjacent canals. Fish samples were collected from Mongla Bazar, located on the opposite side
148 of Mongla port. An inventory was kept for each fish species collected with their local name,
149 scientific name, and feeding zones; afterward, 3-4 individual fish from each sample were
150 weighed and measured for analysis (Table 1).

151

152 **2.3. Pre-processing, digestion, and filtration**

153 **2.3.1. Water**

154 Pre-preparation of water samples was studied from different literature (Laermanns et
155 al., 2021; McCormick et al., 2016; Scherer et al., 2020; Vermaire et al., 2017). One hundred
156 milliliters of water were kept in 150 mL beakers, and 5 mL of concentrated nitric acid (68%)
157 was added. The solution was kept aside for seven days at room temperature. Afterward, the
158 samples were filtered using nylon filter paper with a 0.45 µm mesh size. The filter papers
159 were air-dried for 24 hours for visual observation in glass Petri dishes.

160

161 **2.3.2. Sediment**

162 Pre-processing and preparation of sediment samples were studied from recent
163 literature (Afrin et al., 2020; Cordova et al., 2021; Garcés-Ordóñez et al., 2019; Horton et al.,
164 2017; Laermanns et al., 2021; Mai et al., 2018; Mohamed Nor & Obbard, 2014; Zhou et al., 2020;
165 Zuo et al., 2020). The sediment samples were dried at 65°C for 24 hours in a drying oven. A
166 mortar pestle and sieve shaker were used to separate fine-grained particles with a mesh size
167 of 75 µm. Each sample was shaken for 3 min in a sieve shaker. Twenty milliliters of hydrogen
168 peroxide solution (at 30%) was added to each gram sample and digested in the oven at 80°C
169 for four hours. For density separation, 3 g ZnCl was added to the beaker and kept aside for 48
170 hours. Afterward, the samples were filtered using a vacuum filtration pump using 0.45 µm
171 mesh-size nylon filter paper. The filter papers were air-dried for 24 hours before visual
172 observation

173

174 **Table 1.** Attributes of collected fish samples in Mongla Bazar, located on the opposite side of Mongla port (Bangladesh).

Local name	Specie	Family	Status (IUCN Red List)	Environment: climate zone and depth range	No. of individual
Ilish	<i>Tenualosa ilisha</i>	Clupeidae	Least concern (Freyhof, 2014)	Brackish and pelagic-neritic	3
Bhetki	<i>Lates calcarifer</i>	Latidae	Least concern (Pal & Morgan, 2019)	Brackish and demersal	3
Poa	<i>Otolithoides pama</i>	Sciaenidae	Data deficient (Hasan et al., 2020)	Brackish and benthopelagic	4
Tengra	<i>Mystus vittatus</i>	Bagridae	Least concern (Ng, 2010)	Brackish and demersal	3
Payra	<i>Scatophagus argus</i>	Scatophagidae	Least concern (Collen et al., 2019)	Brackish and reef-associated	3
Loitta	<i>Harpodon nehereus</i>	Synodontidae	Near threatened (Russel et al., 2019)	Brackish and benthopelagic	4
Chemo	<i>Pseudapocryptes elongatus</i>	Oxudercidae	Least concern (Jenkins & Kullander, 2009)	Brackish and demersal	3
Bele	<i>Awaous grammepomus</i>	Gobiidae	Least concern (Larson, 2019)	Brackish and benthopelagic	3

175 Note - IUCN Red List: International Union for Conservation of Nature's Red List of Threatened Species

176 (<https://www.iucnredlist.org/about/background-history>).

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181 **2.3.3. Fish**

182 Pre-processing of fish samples was studied from previous research (Cole et al., 2014;
183 Daniel et al., 2020; Dehaut et al., 2016; Parvin et al., 2021; Sarker et al., 2019; Selvam et al.,
184 2021). Fish samples were washed with ethanol and cut using stainless steel equipment to
185 extract the gastrointestinal tract (GIT) and muscle tissues; afterward, each sample was
186 weighed and refrigerated at -20°C. Alkali digestion was applied for digesting fish samples
187 (Daniel et al., 2020; Dehaut et al., 2016; Tanaka & Takada, 2016). Ten milliliters of KOH solution
188 (at 10%) was added to each beaker per gram of fish sample. The samples were incubated at
189 50°C for four days in a shaker incubator. Upon digestion, the samples were filtered using 0.45
190 µm mesh-sized nylon filter paper. The filter papers were air-dried in glass Petri dishes for 24
191 hours until visual observation.

192

193 **2.4. Visual observation and spatial distribution**

194 The water, sediment, and fish samples were analyzed under a digital fluorescence
195 microscope (Motic B410E, Germany) using 10x zoom, for visual identification of MP particles.
196 The particles were classified into five shapes (fragment, fiber, foam, film, pellet), three sizes
197 (1-5 mm, 0.5-1 mm, less than 0.5 mm), and nine colors (violet, red, blue, brown, black, yellow,
198 orange, grey, white, transparent) (Garcés-Ordóñez et al., 2020; Hossain et al., 2020; Jabeen et
199 al., 2017; Parvin et al., 2021; Tanaka & Takada, 2016; Zhou et al., 2020).

200

201 **2.5. Quality control**

202 Water and sediment samples were collected in marked bottles and airtight bags,
203 respectively, and the openings were sealed. All collection materials were washed with
204 ethanol and distilled water before collecting. Fish samples were weighted and measured at
205 the earliest convenience, and GIT and muscles were dissected with stainless steel equipment.
206 Fish bodies and dissecting equipment were washed with ethanol and distilled water before
207 dissection to avoid contamination (Hossain et al., 2020; J. Li et al., 2016; Parvin et al., 2021).
208 Further, the samples were correctly marked and kept frozen until analysis. While measuring,
209 digestion, and filtration, samples were handled in a glass beaker and Petri dishes.

210

211 **2.6. Fourier Transform Infrared Spectroscopy (FT-IR) and Scanning Electron
212 Microscopy (SEM)**

213 Selected samples were analyzed in FT-IR to identify MP polymers using an FT-IR
214 spectrometer (PerkinElmer). The transmission mode was applied, and wavenumbers were
215 kept at 400-4000 cm⁻¹ (Daniel et al., 2020; Garcés-Ordóñez et al., 2019; Hossain et al., 2020;
216 Parvin et al., 2021). The resulting peaks were matched to standards from other literature, and
217 most matched peaks were accepted to be the specific polymer type (Afrin et al., 2020; Jung et
218 al., 2018; Selvam et al., 2021). Some samples were selected for high-resolution SEM analysis
219 for elemental composition and topography detection by taking photographs. Information on
220 surface topography, morphology, and composition of the surface of MPs can be acquired
221 through an electron beam of SEM (Akhtar et al., 2018; Parvin et al., 2021). In this study,
222 Trinocular Stereo Microscope with Stemi 508 model was used. A built-in camera ‘Axiocam
223 105’ was used to compute images of plastic particles. Exposure and white balance were kept
224 in ‘auto’ mode.

225

226 **2.7. Data analysis**

227 The statistical analysis of the abundance of MPs was shown using data analysis and
228 visual representation using GraphPad Prism software version 9.0 and ArcMap 10.3. Google
229 Earth was used to extract the locations of sampling sites. Using the sampling data and
230 interpolation command, a spatial distribution of MPs in water and sediment was prepared to
231 illustrate the occurrence of the pollutant.

232

233 **3. RESULTS**

234 In this study, we observed that the water and sediment samples showed varying levels
235 of plastic particulate contamination (Figure 2A and 2C, respectively). On average, 2.66×10^3
236 plastic particles/L and 1.57×10^5 particles/kg were found in water (Figure 2B) and sediment
237 samples (Figure 2D), respectively, which may originate from various sources, including
238 terrestrial sources of household wastes, marine litters, tourism, and industrial waste.
239 Furthermore, domestic wastes are an important contributor to mangrove forests (Deng et al.,
240 2021; Garcés-Ordóñez et al., 2019; Naji et al., 2017). As shown in Figure 3, more MP
241 contamination was observed near Mongla port, Mongla-Chila boat line, and Chila bazar,
242 suggesting that tourism and fishing activities contribute to the number of MPs in the adjacent
243 sediment and waters, like previous reports that also identified higher levels of MP
244 contamination in populated areas (Garcés-Ordóñez et al., 2019; R. Li et al., 2020; Zhou et al.,
245 2020; Zuo et al., 2020).

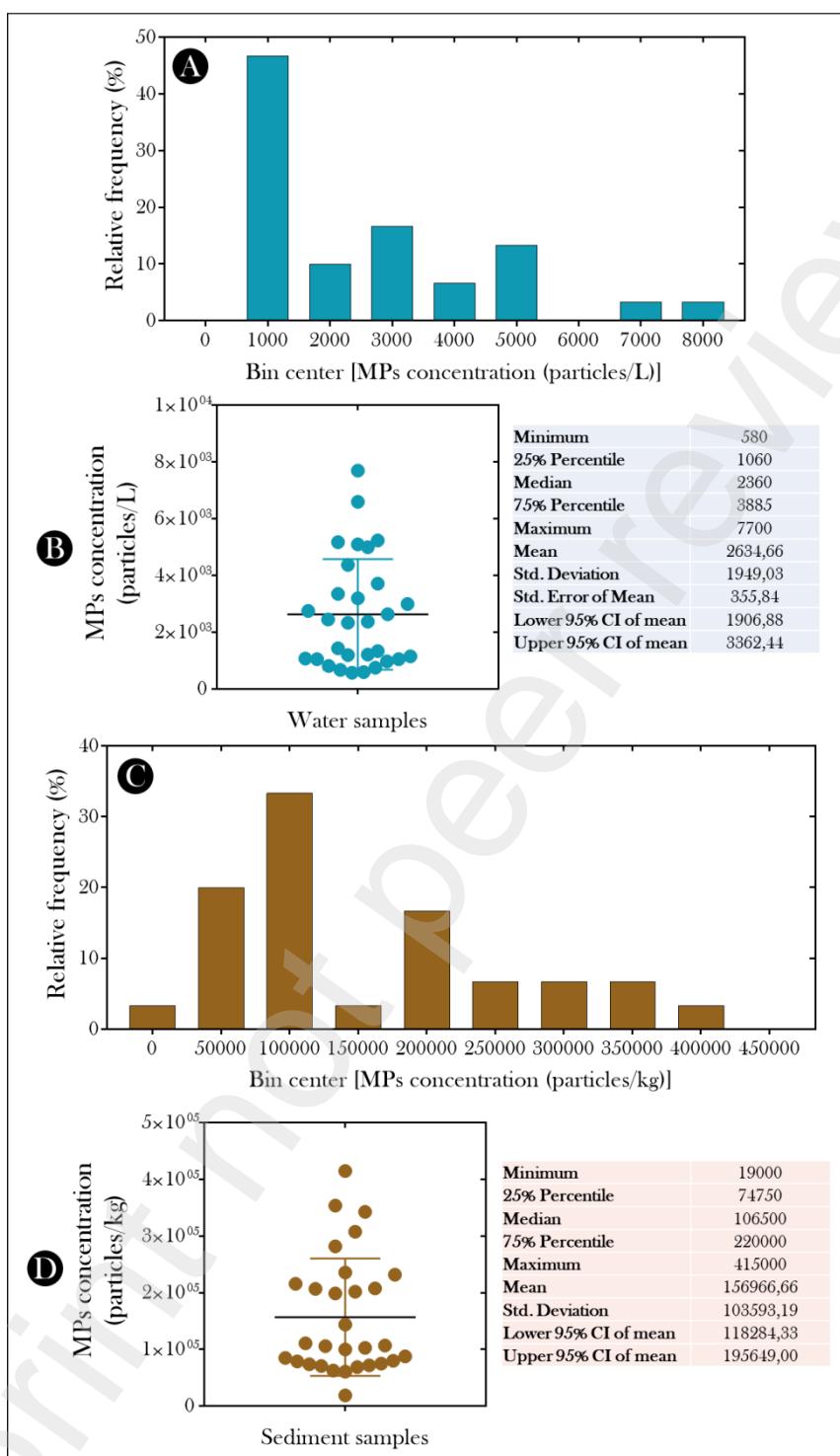


Figure 2. Distribution histogram of plastic particle contamination levels in (A) water and (C) sediment samples (Pasur river, Mongla, Bangladesh). (C-D) Scatter plot and summary of descriptive statistics of concentrations recorded in water and sediment samples, respectively.

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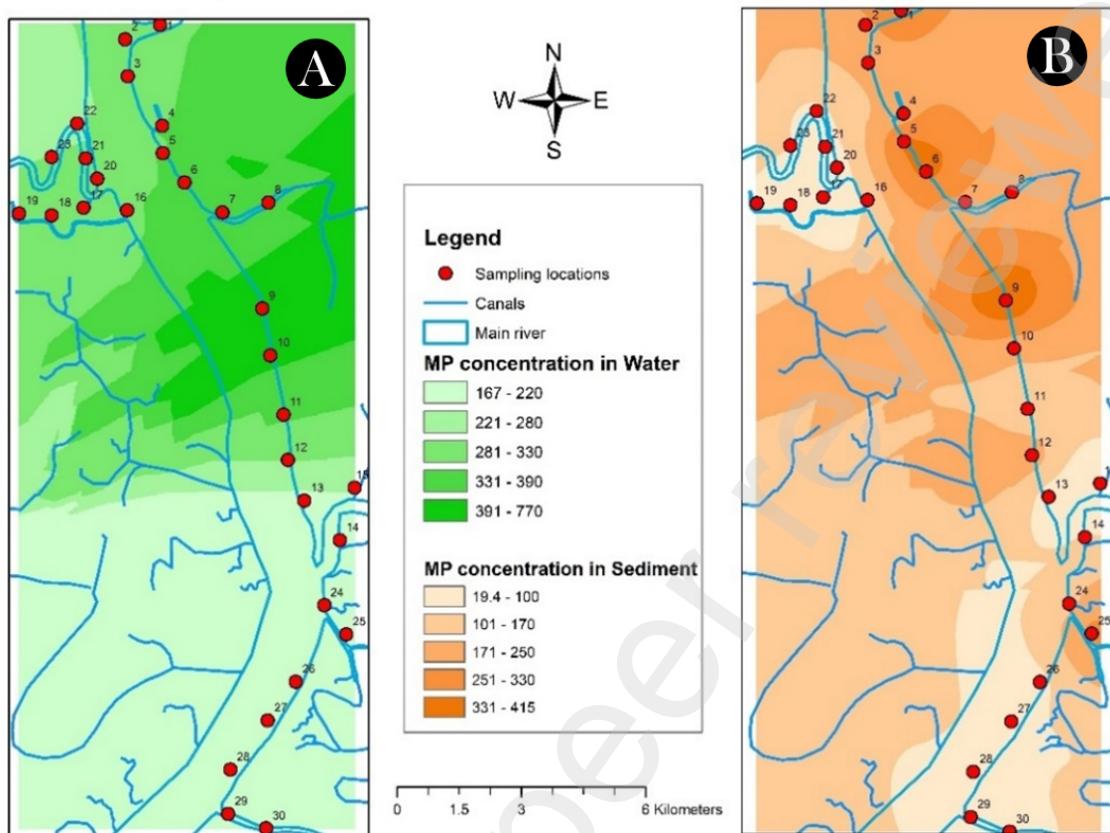


Figure 3. Spatial distribution of microplastics in (A) water and (B) sediment from the study area [Pasur river and its adjacent canals (Bay of Bengal, Bangladesh)].

250

The abundance of MPs in both sediment and water showed a decreasing trend starting from Mongla port toward the Herbaria ecotourism center. The highest number of MPs in water was found at the 6th sampling location, 7.7×10^3 particles/L, beside the riverbank of Mongla-Chila road, forwarding toward Chila bazar. The second highest number was found at point 16th, 5.18×10^3 particles/L near Karamjal, and the lowest MP contamination was recorded in water at Dhangmari Khal (at point 21st, comprising only 5.8×10^2 particles/L) (Figure 4A). Already the major MP number in sediments was present in Chila, point 9, with 4.15×10^5 particles/kg, and second highest at point 6, with 3.54×10^5 particles/kg (Figure 4B). The 6th sampling point is common for both water and sediments with greater MP contamination levels. On the other hand, sampling points 21-22 are lower in MPs for both water and sediments. The lowest number of particles in sediments are present in sampling point 15, with 1.9×10^4 particles/kg only, near Sela River (Figure 4). The abundance of microplastics is lower for both water and sediments near herbaria (Figures 3 and 4).

264 A relationship between the profusion of MPs between water and sediments shows a
 265 decreasing pattern from the northern part of the river toward the southern part (Figure 3).
 266 A similarity in the way of increase and decrease can be observed from the curve, which means
 267 that areas with greater MPs in water are also greater in number in the case of sediments, and
 268 vice versa for the areas with a lower number of MPs (Figure 4). The population density is also
 269 a driver in the number of MPs found. Points a few kilometers south of Mongla port, where
 270 Chila is located, are seen to have a greater number of MPs both in water and sediment
 271 samples. This might result from extreme fishing activities and improper disposal of waste
 272 and boat gear.

273 The spatial distribution of MPs with five classes in water and sediments (Figure 3)
 274 shows similarity with the distribution pattern observed in Figure 4; the northern portion of
 275 the study area is more polluted with plastics than the southern portion. Most Sundarbans'
 276 villages are near Mongla port, Karamjal, and Chila. The spread of microplastics in water is
 277 comparatively lower in the points with limited access to people (points 24 to 30) (Figure 4).

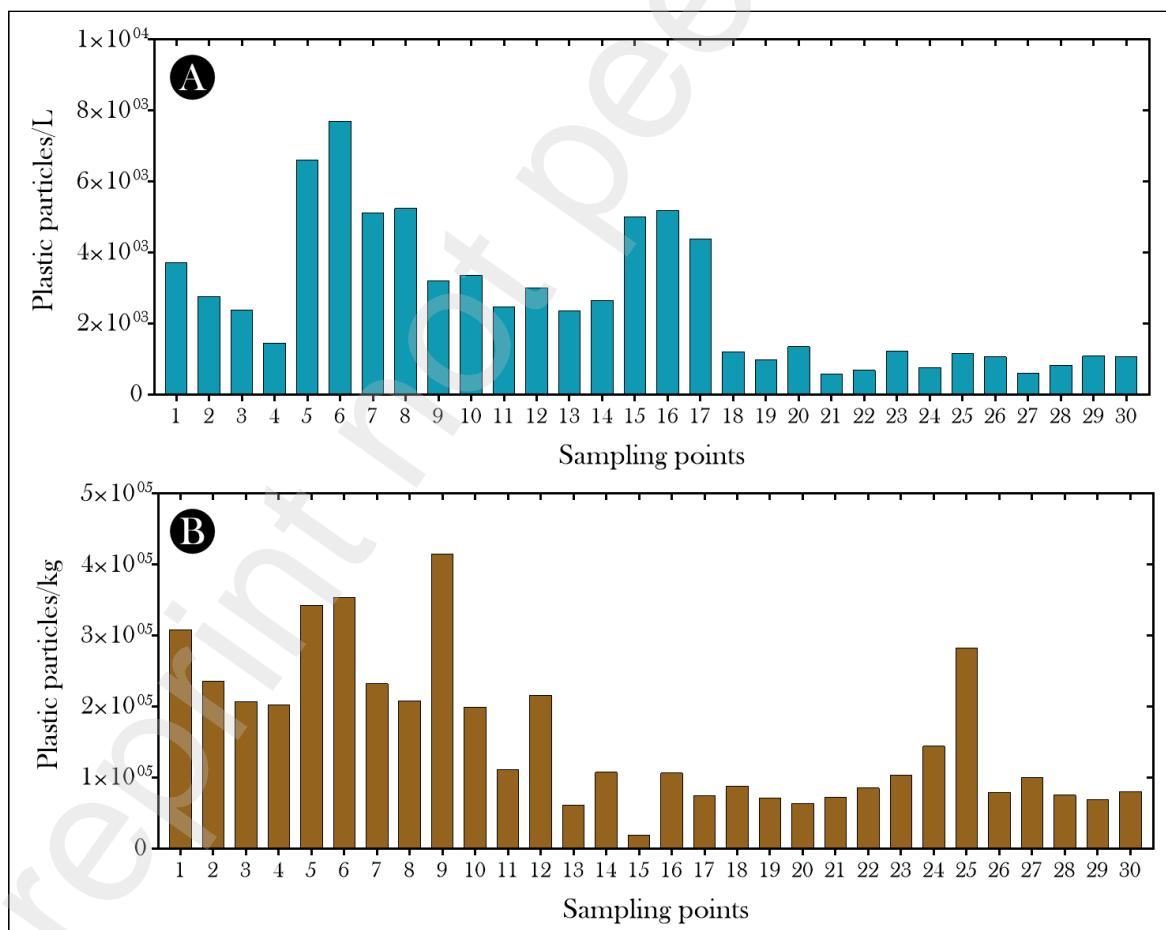


Figure 4. Microplastic abundance in all analyzed (A) water and (B) sediment samples [Pasur river and its adjacent canals (Bay of Bengal, Bangladesh)].

279 MP_s were also observed in all analyzed fish samples, considering 3-4 samples/species.
280 Results show a higher number of MP_s in the animals' GIT (10.41 ± 7.43 particles/g – median ±
281 SEM), concerning the average concentration recorded in the muscles (4.68 ± 1.85 particles/g
– median ± SEM) (higher difference at 50%) (Figure 5A). *O. pama* and *H. nehreus* were the
283 species that showed the highest MP_s accumulation in the GIT (Figure 5B). In the muscles, the
284 highest MP levels were observed in *T. ilisha* and *L. calcarifer* (Figure 5C).

285 The fish species were divided into different feeding zones according to their food
286 habit. The fish from the benthopelagic zone is mostly contaminated with MP_s (Parvin et al.,
287 2021), which was confirmed in our study (Figure 6A). Although the MP concentrations in the
288 GIT did not differ significantly in fish when we performed the analysis considering the
289 feeding zone of the animals, we noticed that the evaluated benthopelagic species [especially
290 *Otolithoides pama* and *Harpodon nehreus* (Table 1)] showed MP_s accumulation in the GIT, on
291 average 80.57% higher than that observed in the other species (Figure 6A). In addition,
292 expressive MP concentration in demersal fish was identified, which confirms previous
293 studies reporting that the ultimate sink for MP_s in water is the benthic and demersal zones
294 (Goldberg, 1997; Pauly et al., 1998; Woodall et al., 2014). A similar pattern was observed when
295 we analyzed the MP concentrations in the muscles of the animals (Figure 6B), which suggests
296 that the uptake of MP_s via oral ingestion is a preponderant factor for the absorption of these
297 pollutants and accumulation in the muscles. Overall, we observed a statistically significant
298 correlation between the MP concentrations in the animals' GIT and muscles, as well as a
299 linear increase in the presence of MP_s in the muscle as there is a greater uptake of MP_s via
300 oral ingestion, thus confirming this hypothesis (Figure 5D).

301 The presence of high MP contamination in *O. pama* and *H. nehreus*, in particular,
302 constitutes a worrying result since *O. pama* [earlier known as *Pama pama* (*Sciaenoides pama*)]
303 and *H. nehreus* (the most famous member of this genus is the Bombay duck), forms an
304 important sciaenid fishery in the Asiatic estuarine systems (Liang & Pauly, 2017; Gao et al.,
305 2018; Bhakta et al. 2019ab; Bhakta et al., 2022). Therefore, the consumption of these animals
306 represents a potential route of human MP contamination.

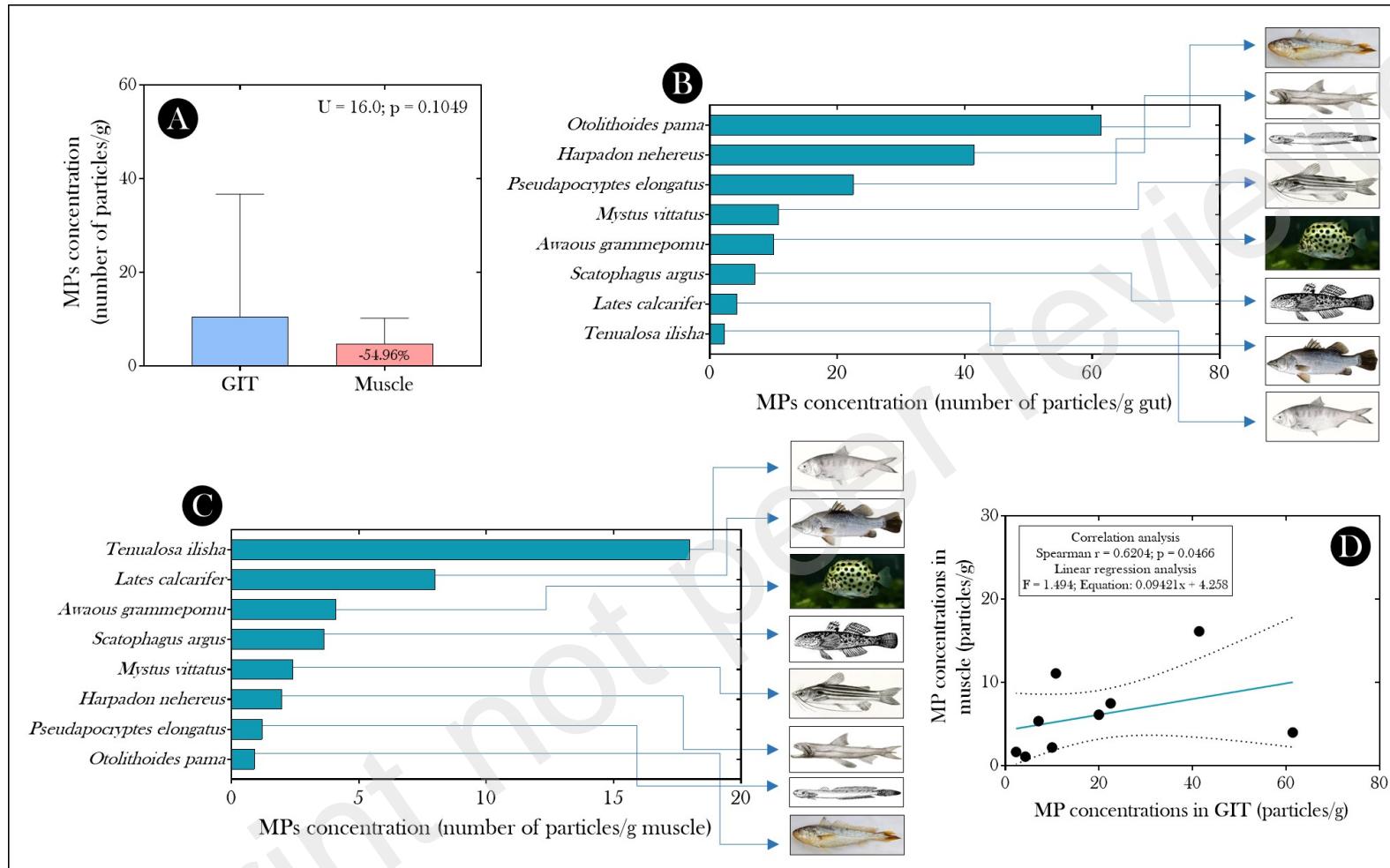


Figure 5. Microplastic concentrations in the gastrointestinal tract (GIT) and muscle samples of fish collected in the Pasur river of Mongla City (Bangladesh). (A) Mean of MP concentrations in GIT vs. muscle; (B and C) MP concentrations in GIT and muscle samples, respectively, of the different fish species. (D) Correlation and linear regression analysis between MP concentrations in GIT and muscles. In “A”, non-parametric data are presented by the median and interquartile range, and the summary of the statistical analysis is shown at the top of the graph.

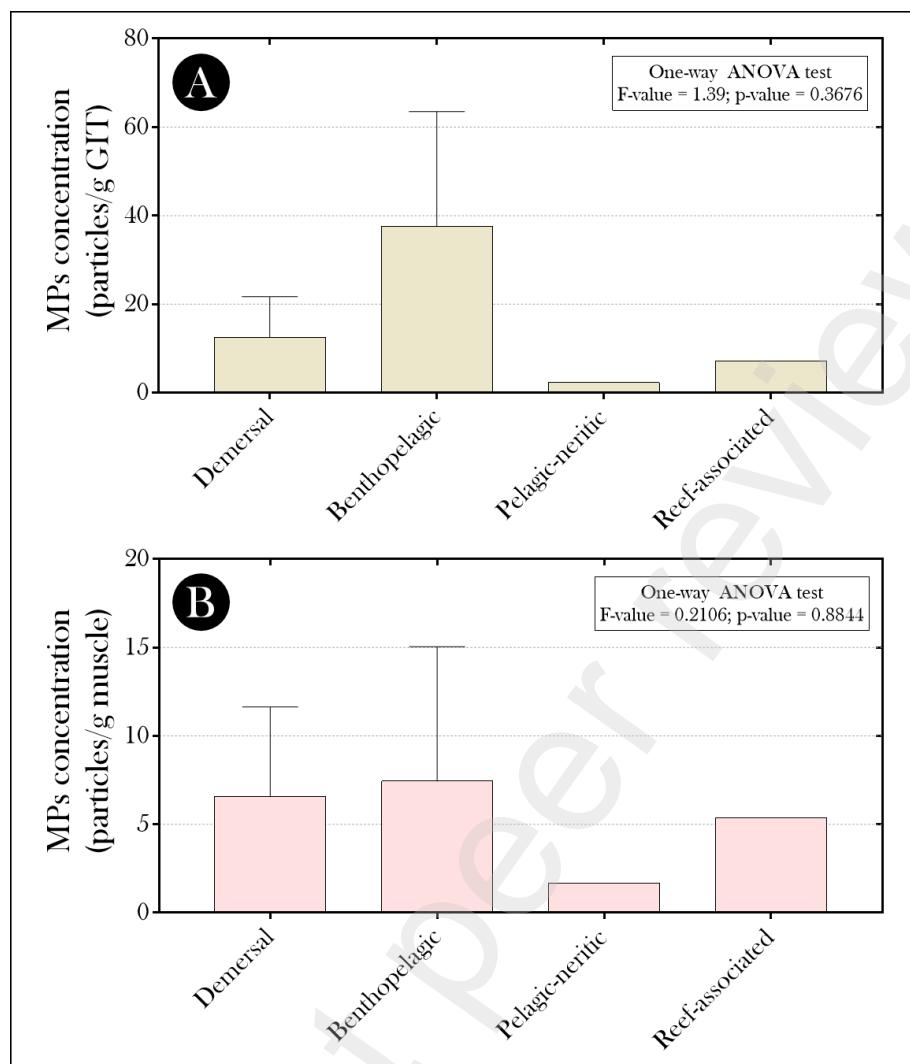


Figure 6. Microplastic (MP) concentrations in (A) gastrointestinal tract (GIT) and (B) muscle of fish separated by habitat type (demersal, benthopelagic, pelagic-neritic, and reef-associated) [Pasur river of Mongla City (Bangladesh)]. Parametric data are presented by the mean + standard deviation, and summaries of statistical analyzes are shown at the top of the graphs.

310

311

312 Regarding the characterization of the MPs identified in our study, we observed that most of
313 the particles found were secondary type, as they do not have any symmetrical shapes, unlike
314 primary particles (Galgani et al., 2015). MPs were differentiated among five forms in this
315 study, like Zhou et al. (2016), with the fragments dominant in the water and sediment samples
316 (Figure 7A-B, respectively). On the other hand, the MP types most abundant in the GIT and
317 muscle samples were the fibers and fragments (Figure 7C-D, respectively). While fragments
318 were more frequent in sediment samples than in water samples (Figure 8A), fibers were more
319 abundant in water samples (Figure 8A). In the biological samples, the differences between the
320 types of MPs in the GIT and muscle samples were not observed (Figure 8B).

321 Similar research also found abundant fragments in sediments (Horton et al., 2017;
322 Selvam et al., 2021; Zuo et al., 2020). Fragment particles occur due to the breakdown of plastic
323 particles (Cole et al., 2011). Some secondary MPs of asymmetrical shape are released from
324 packaging materials such as water bottles, juice bottles, milk cans, garbage bags, polybags,
325 etc. (Wang et al., 2019). Pellets or granules generally come from the presence of fragments to
326 prove that the particles found are of secondary origin (Lehtiniemi et al., 2018). It is reported
327 that microorganisms living in the mangrove can degrade plastics into fragment particles
328 (Deng et al., 2021). Many studies found an abundance of fiber and fragments in water (Aliabad
329 et al., 2019; Nabizadeh et al., 2019).

330 The abundance of fiber particles in fish GIT was also reported by other studies (Daniel
331 et al., 2020; Garcés-Ordóñez et al., 2020; Karlsson et al., 2017; R. Li et al., 2020; Parvin et al.,
332 2021). These particles may come from extreme fishing activities, an everyday livelihood in
333 the Mongla district, because of fishing nets, nylon nets, and gill nets (Garcés-Ordóñez et al.,
334 2020; R. Li et al., 2020). Fibers in GIT may also come from textiles and clothes, sewage water
335 from cloth washing, etc. (Browne et al., 2011; Park et al., 2020). Other sources of fiber found
336 in fish GIT can be from household wastewater and textile wastes (Hossain et al., 2020).

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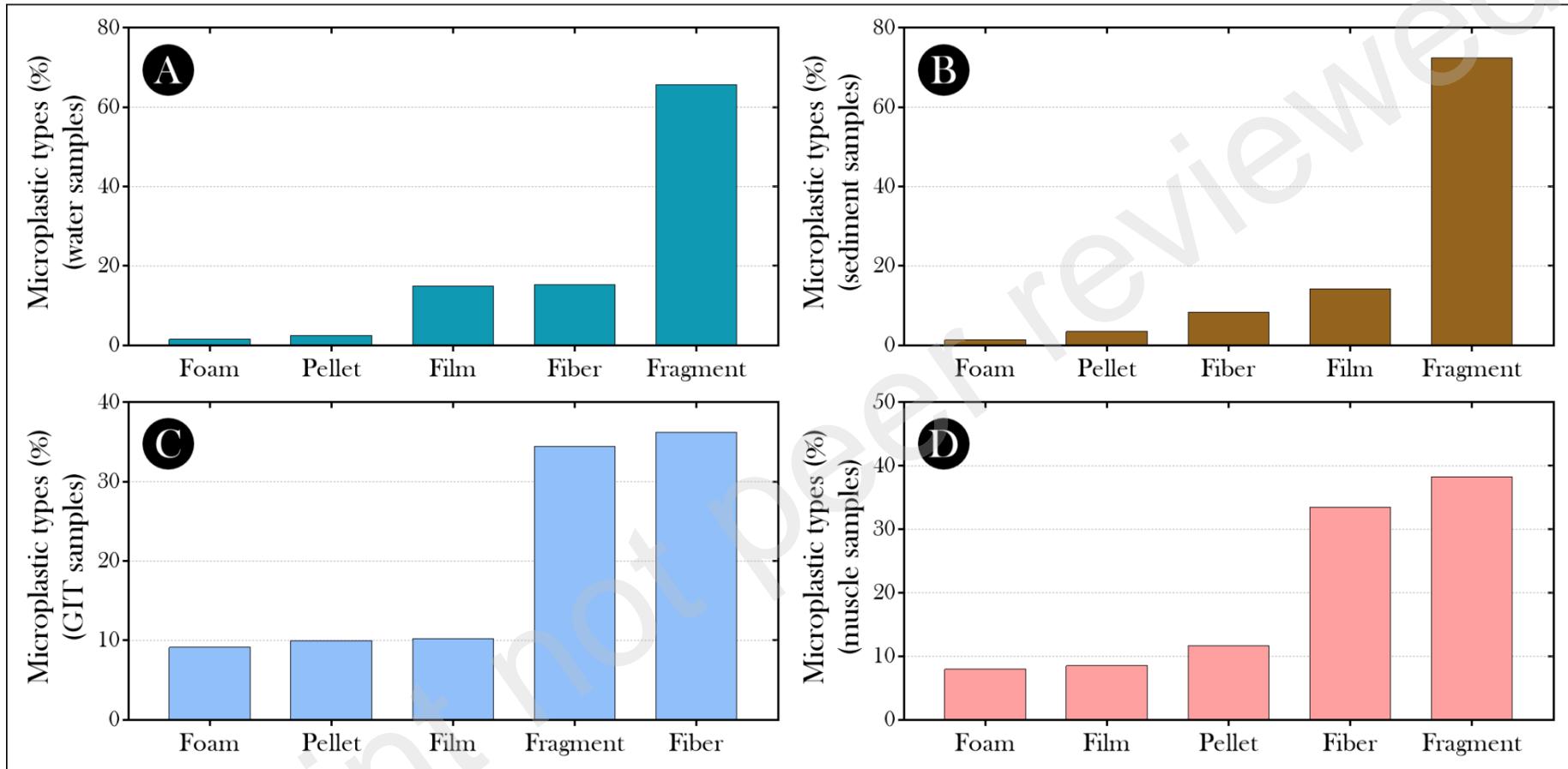


Figure 7. Microplastic types (%) identified in the present study in (A) water, (B) sediment, (C) gastrointestinal tract (GIT), and (D) muscle samples.

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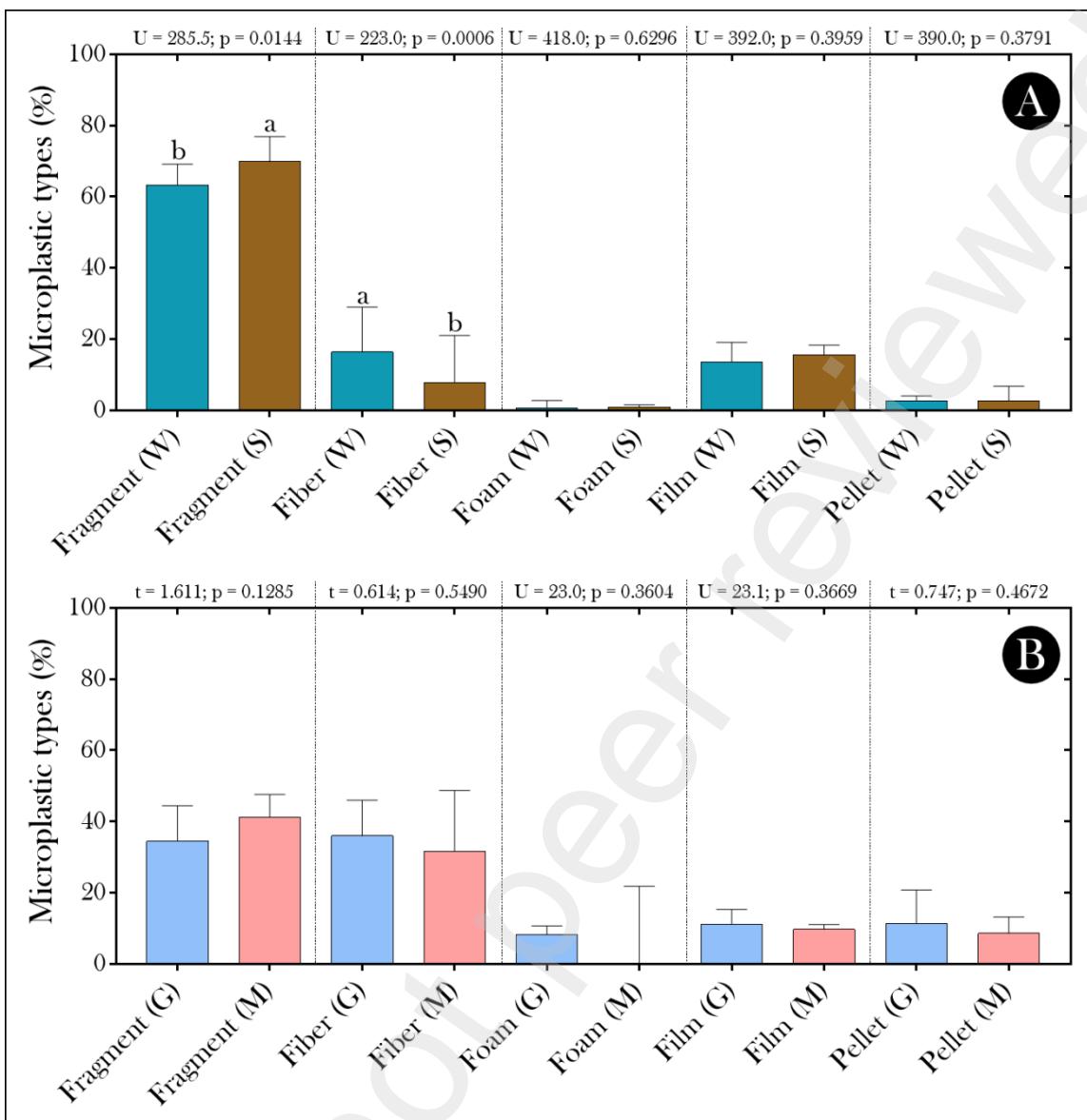


Figure 8. Microplastic types (%) in the different samples analyzed in our study. (A) Water (W) and sediment (S) samples and (B) gastrointestinal tract (GIT) and muscle (M) samples. Parametric data are presented by the mean + standard deviation, whereas non-parametric data are presented by the median and interquartile range. Summaries of statistical analysis are shown at the top of the graphs.

342

343 Microplastics were diversified among the samples, and ten color schemes were
 344 observed (Figure 9A). Water samples showed the most diversified color combinations among
 345 the water, sediment, and fish samples, with more violet, red, blue, and orange colored
 346 particles than the others. Although in all samples, black and brown were dominant, ranging
 347 between 23-39% and 18-23% particles, respectively. The yellow color was found at the lowest
 348 percentage, less than 2% in the samples. Sediment samples had the least diversified plastic

349 particles according to color, with most of the particles being black or brown (Figure 9A). Most
350 sediment samples were brown, although transparent films and white-ish colored pellets were
351 seen.

352 The black color-dominated particles were also found by other researchers (Hossain et
353 al., 2020; Zuo et al., 2020). Several studies have differentiated plastic particles to be ‘colored’
354 and ‘non-colored’; while some researchers found white and transparent colors to be
355 dominant among MPs, others have seen red and blue colors to be prevalent (Deng et al., 2021;
356 J. Li et al., 2018; R. Li et al., 2019, 2020; Mohamed Nor & Obbard, 2014). The source of the
357 colored particles may come from the coloring materials of fishing gears and vessels and
358 packaging (Aliabad et al., 2019; R. Li et al., 2020). Fishing activities are common occupations
359 of the people in Mongla. From dingy boats to engine boats and speed boats, varieties of colors
360 are used; colors from fishing gears are a possible source of colored particles. The colored
361 particles are likely to be ingested by marine biota as it resembles food particles for fishes;
362 thus, attraction by color is a cause of ingestion (Abayomi et al., 2017; Mohamed Nor & Obbard,
363 2014; Rakib et al., 2022).

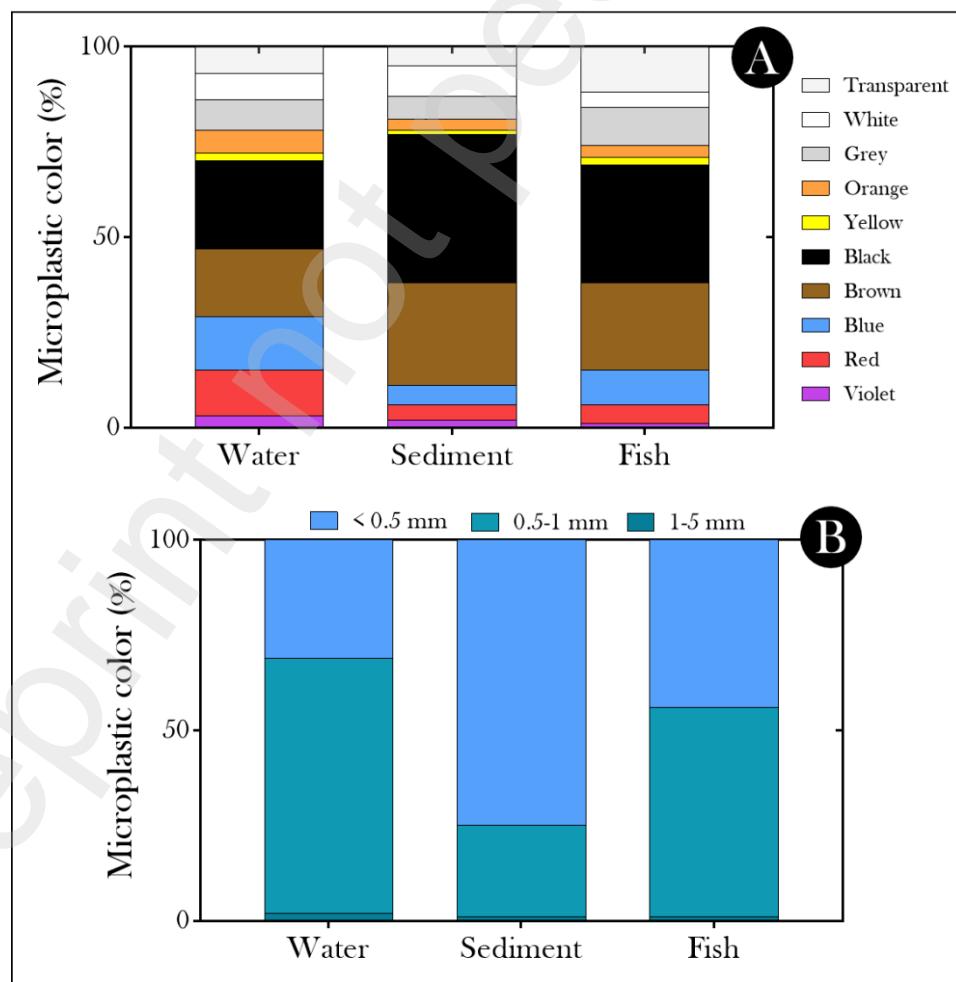


Figure 9. Microplastic (A) color and (B) size percentages identified in the water, sediment, and fish samples analyzed in the present study.

364

365 Furthermore, MPs were found among three size ranges (Figure 9B). Most particles in
366 water and fish samples, 67% and 55%, respectively, were found to be in ranges 0.5-1 mm; but
367 in sediment samples, about 75% of particles were of size ranges below 0.5 mm. Larger
368 particles of 1-5 mm sizes were scarce, about 1-2%, in all three samples. These particles are
369 visible to the naked eye, and most of the observed filter paper was clear, indicating the
370 scarcity of larger particles. Related studies show similar results in the abundance of MPs
371 ranging below 1 mm in size (Hamid et al., 2020; J. Li et al., 2018; R. Li et al., 2020; Naji et al.,
372 2019; Q. Wang et al., 2020; Zhou et al., 2020; Zuo et al., 2020). The smaller particles are more
373 likely to be uptake by a marine organism (Mohamed Nor & Obbard, 2014). The small size also
374 depicts the particle as a secondary MP, which occurs through the breakdown of primary
375 plastic particles. Microscopic analysis shows vibrant colors and various shapes of
376 microplastics present in the studied samples (Figure 10).

377

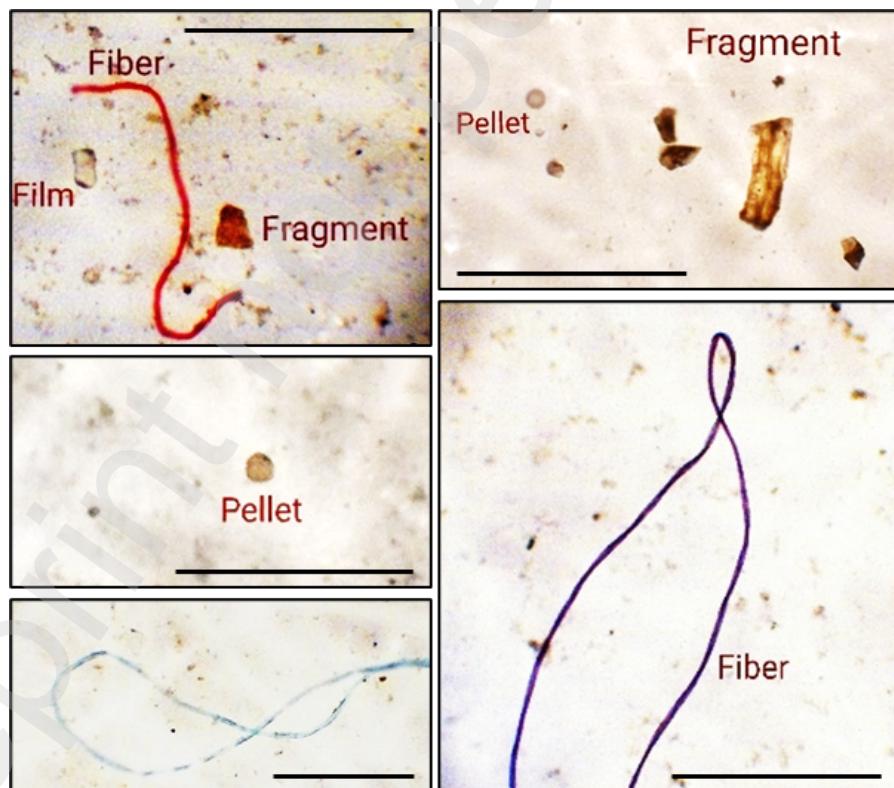


Figure 10. Representative photomicrographs of the types of microplastics identified in the present study. Barr scale = 0.1 mm.

378

379 Regarding the identification of polymers in the analyzed samples, the results of the
380 analysis via FT-IR revealed the presence of seven polymeric types [polyamide (PA),
381 polystyrene (PS), polyethylene (PE), high-density polyethylene (HDPE), low-density
382 polyethylene (LDPE), polypropylene (PP), and polyvinylchloride (PVC)], by comparing the
383 spectra obtained in our study (Figures 11A, 12A, 13A, and 14A) with those available in the
384 literature (Chércoles Asensio et al., 2009; Daniel et al., 2020; Jung et al., 2018; Nishikida &
385 Coates, 2003; Noda et al., 2007; Selvam et al., 2021). While in the GIT and muscle samples, the
386 percentages of the polymeric types did not differ (Figures 11B and 12B, respectively), in the
387 water and sediment samples, PA was the most abundant polymer in these samples (Figures
388 13B and 14B, respectively). Furthermore, we did not notice significant differences between
389 the percentages of each polymeric type between the different samples analyzed (Figures 15
390 and 16), which confirms the relationship between the MPs available in water and sediments
391 and those ingested by the animals.

392 The polymer's PP, PE, and PS abundance, in particular, can be due to the location of
393 aquaculture around the sampling areas, as the aquaculture can contribute to these
394 polymers (J. Li et al., 2018; Q. Wang et al., 2020). The abundance of HDPE in fish can be a
395 consequence of the usage of HDPE in daily activities, including packaging of bags, bottles of
396 milk, detergents, any other food or juice products, pipes, etc. (Parvin et al., 2021).

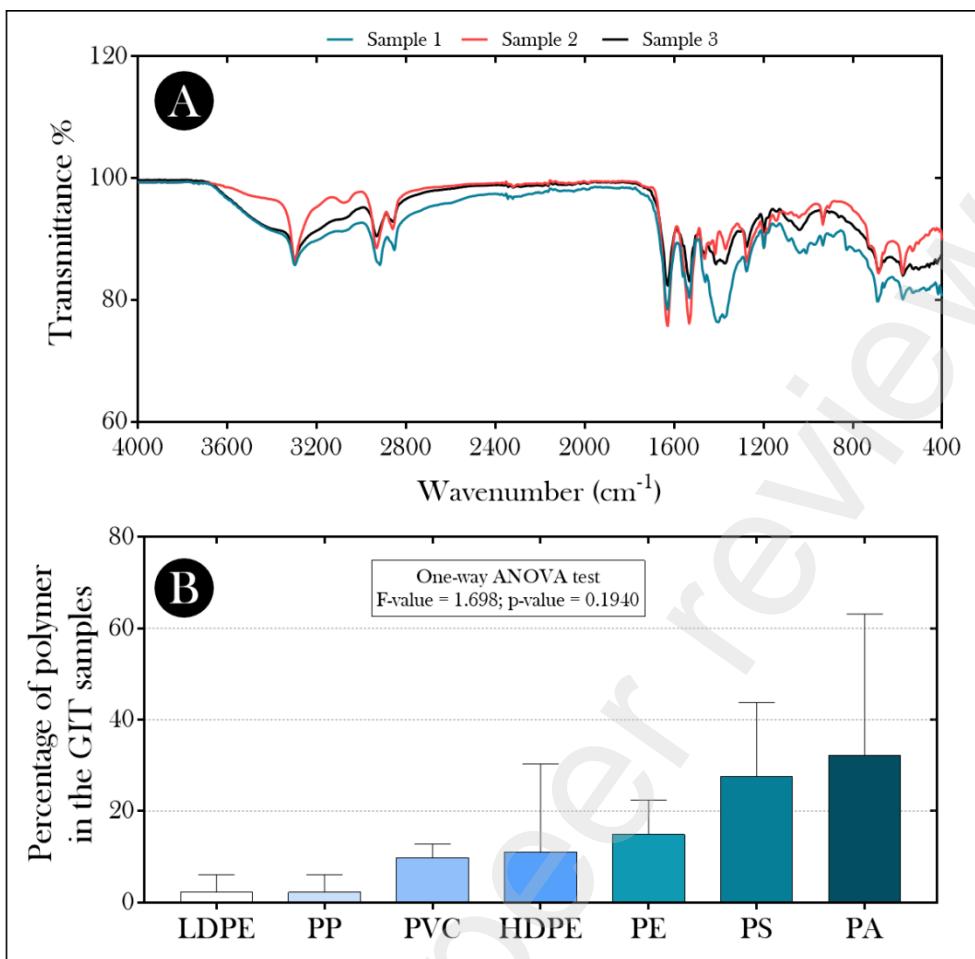


Figure 11. (A) Representative FT-IR spectra and (B) percentage of polymers identified in the gastrointestinal tract (GIT) samples of fish [Pasur river of Mongla City (Bangladesh)]. In “B”, parametric data are presented by the mean + standard deviation, and the summary of statistical analysis is shown at the top of the graph. Note: polyamide (PA), polystyrene (PS), polyethylene (PE), high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), and polyvinylchloride (PVC).

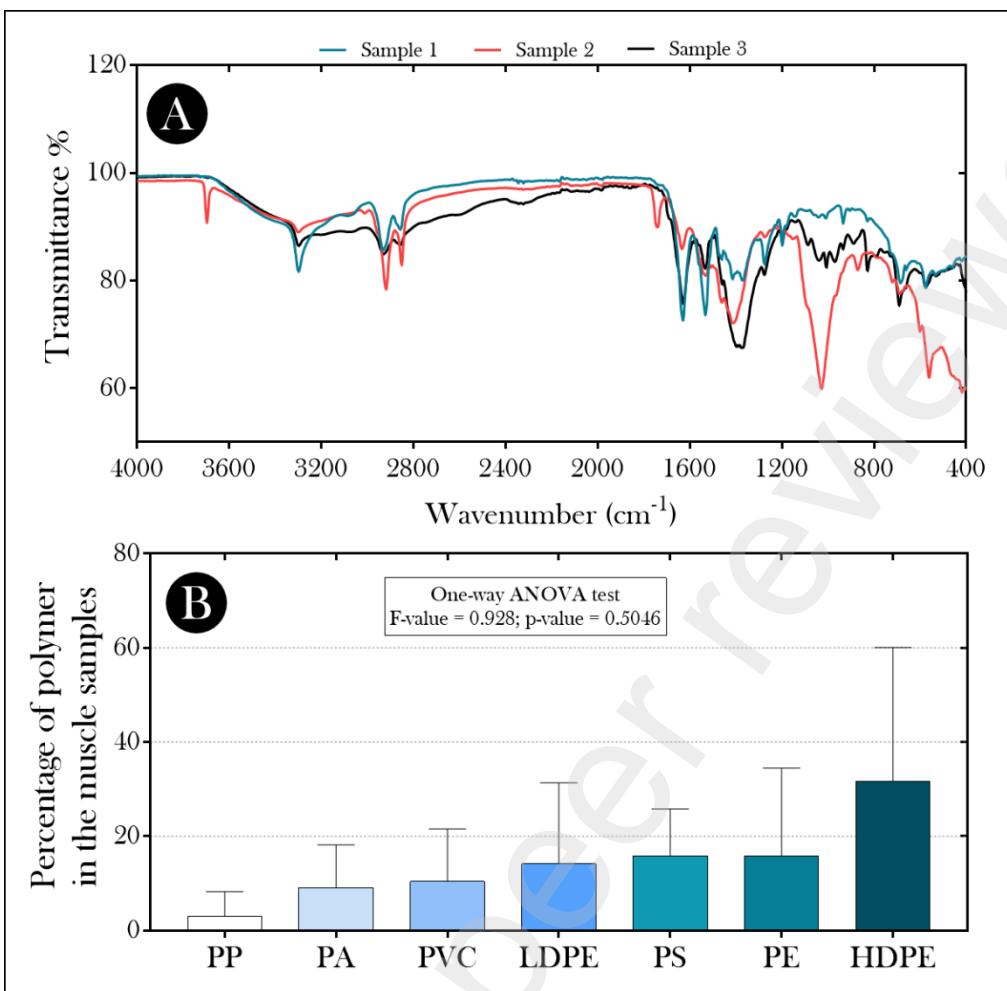


Figure 12. (A) Representative FT-IR spectra and (B) percentage of polymers identified in the muscle samples of fish [Pasur river of Mongla City (Bangladesh)]. In “B”, parametric data are presented by the mean + standard deviation, and the summary of statistical analysis is shown at the top of the graph. Note: polyamide (PA), polystyrene (PS), polyethylene (PE), high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), and polyvinylchloride (PVC).

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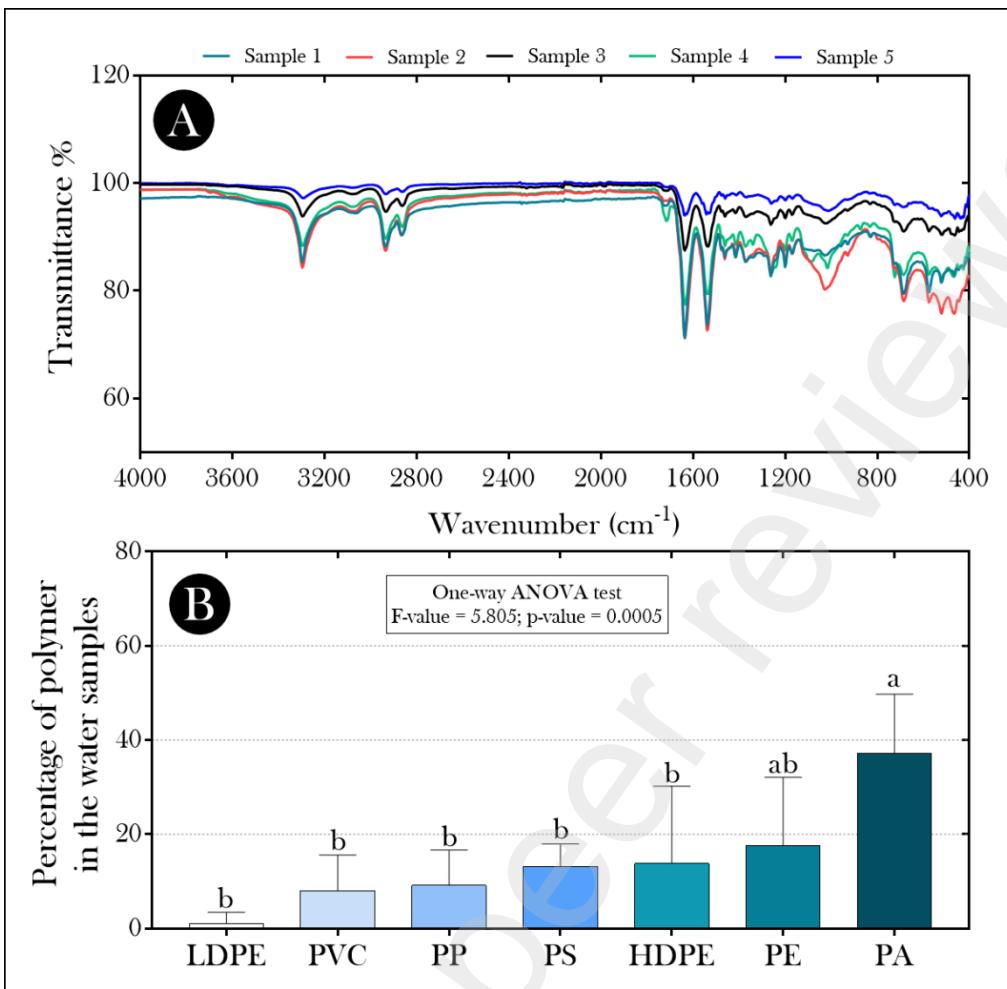


Figure 13. (A) Representative FT-IR spectra and (B) percentage of polymers identified in the water samples. In “B”, parametric data are presented by the mean + standard deviation, and the summary of statistical analysis is shown at the top of the graph. Distinct lowercase letters indicate significant differences. Note: polyamide (PA), polystyrene (PS), polyethylene (PE), high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), and polyvinylchloride (PVC).

401

402

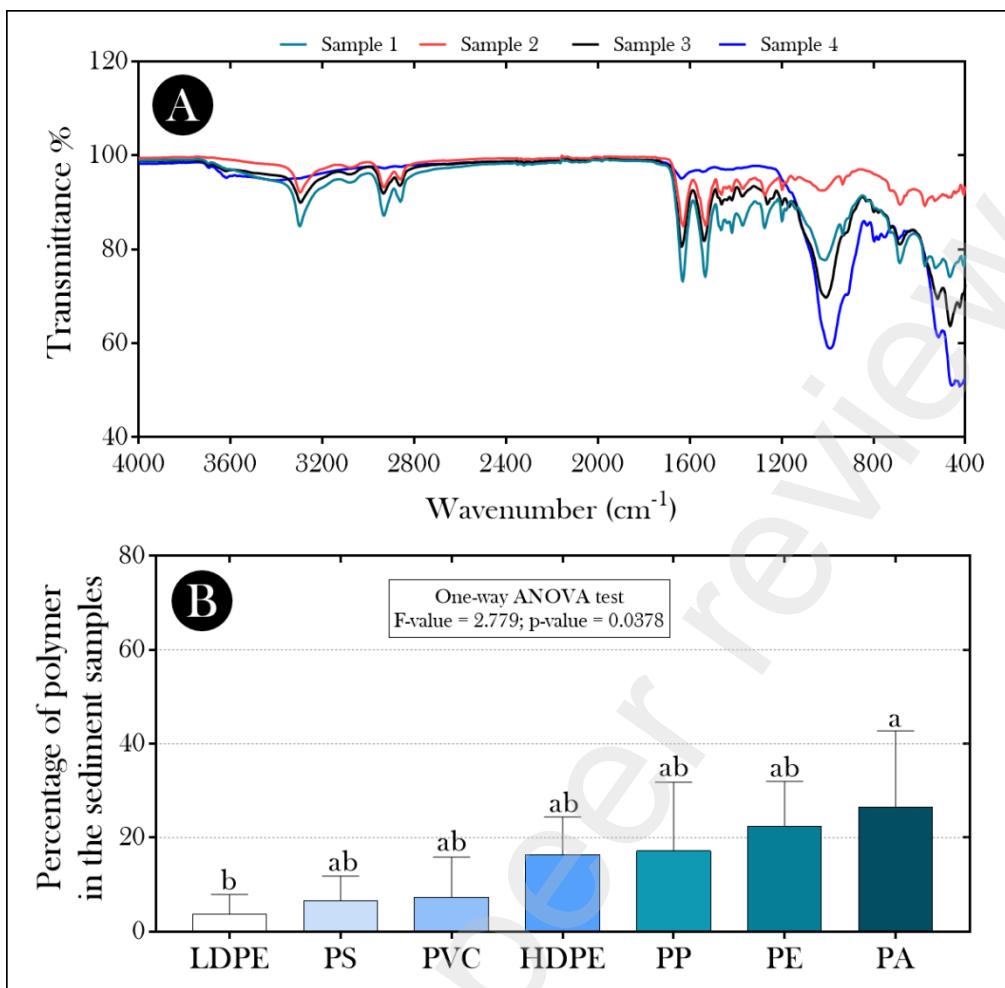


Figure 14. (A) Representative FT-IR spectra and (B) percentage of polymers identified in the sediment samples. In “B”, parametric data are presented by the mean + standard deviation, and the summary of statistical analysis is shown at the top of the graph. Distinct lowercase letters indicate significant differences. Note: polyamide (PA), polystyrene (PS), polyethylene (PE), high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), and polyvinylchloride (PVC).

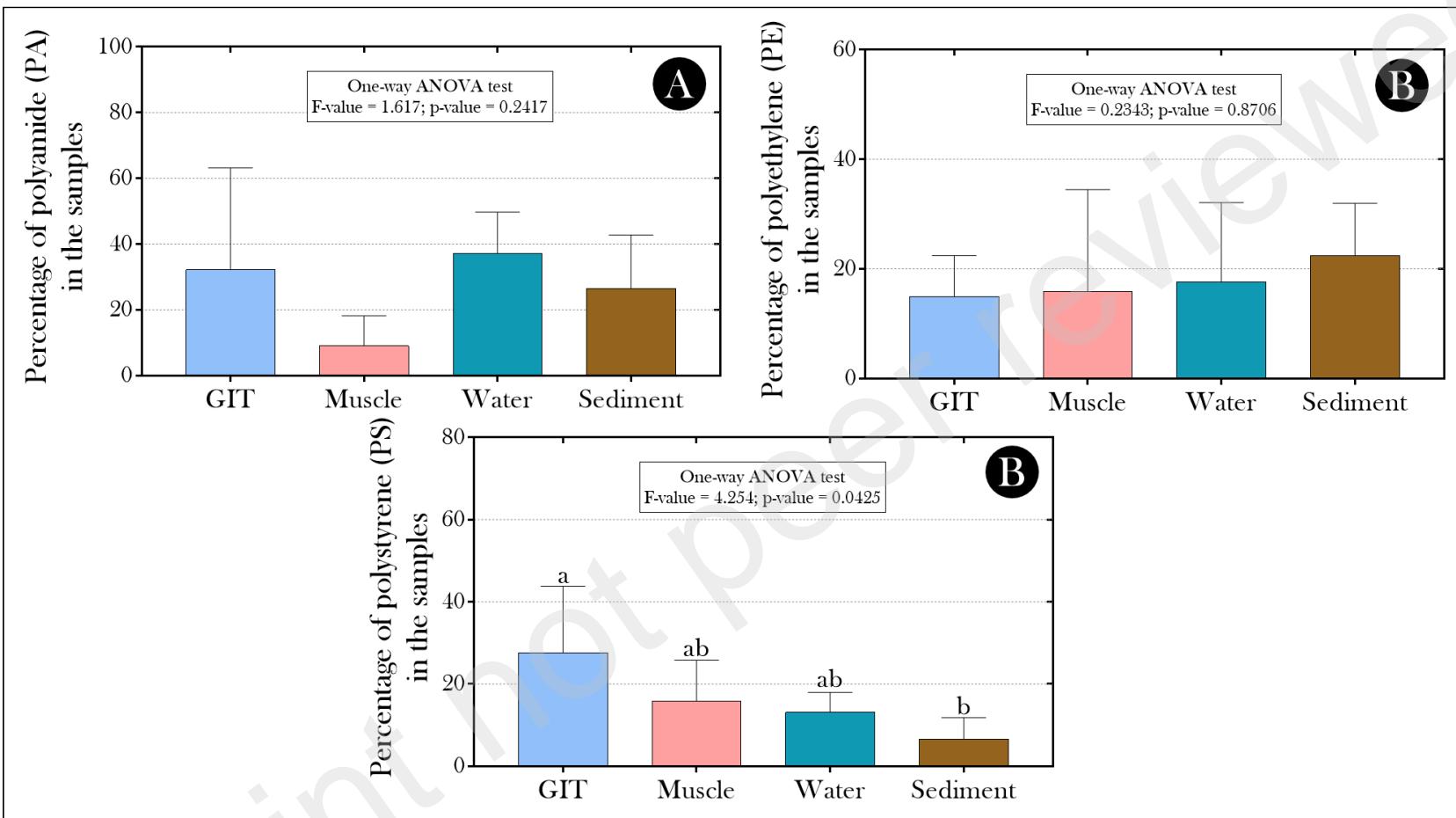


Figure 15. Percentage of (A) polyamide, (B) polyethylene, and (C) polystyrene identified via FT-IR analysis in the different samples analyzed in the present study. Parametric data are presented by the mean + standard deviation, and the statistical analysis summaries are shown at the top of the graphs.

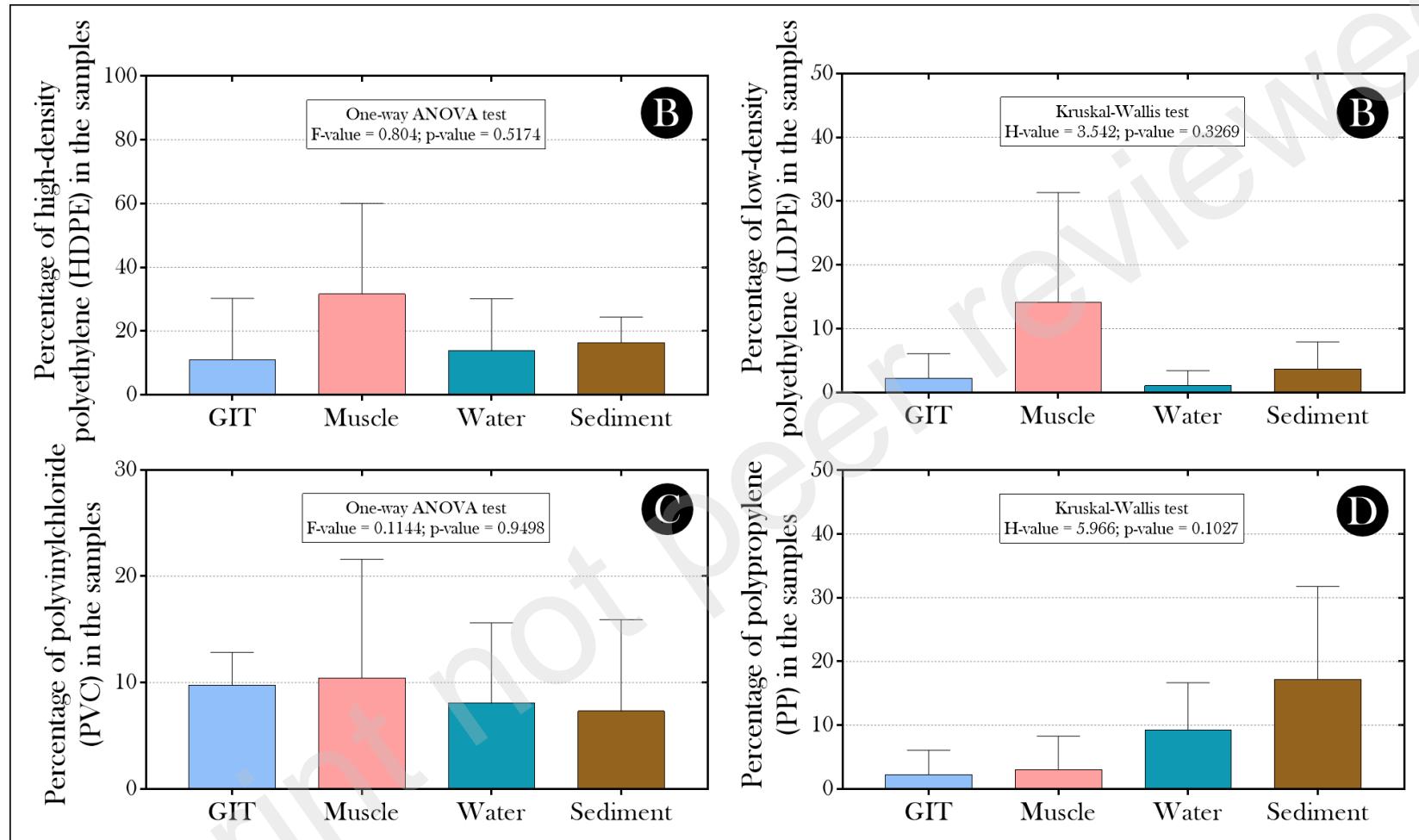


Figure 16. Percentage of (A) high-density polyethylene, (B) low-density polyethylene, (C) polyvinylchloride, and (D) polypropylene identified via FT-IR analysis in the different samples analyzed in the present study. Parametric data are presented by the mean + standard deviation, and the statistical analysis summaries are shown at the top of the graphs.

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406 Different exposures to the environment may have an impact on the structural
407 morphology and additive content of microplastics. The chemical and morphological
408 characteristics can be identified through scanning electron microscopy (Fries et al., 2013).
409 Some samples were selected for SEM analysis, although it is challenging to identify the
410 sources of fiber and fragments through the images (Parvin et al., 2021). SEM images showed
411 several fractures, cracks, flakes, and scratches on the surface of MPs. Some images showed
412 particles adhering to the surface of MPs (Figure 17). MPs not only remain in the
413 environment for extended periods than other pollutants but also absorb other harmful
414 inorganic and organic pollutants like PAHs, pathogens, and heavy metals on their surface.
415 Its larger surface is one reason for this absorbance, leading to toxicity (Rochman et al., 2013;
416 Schrank et al., 2019; Yang et al., 2014). Flakes may result from the particle's oxidative
417 weathering (Parvin et al., 2021). The features observed in the polymers prove that these
418 plastics have gone through mechanical degradation, oxidative weathering, and aging
419 (Wagner et al., 2014).

420

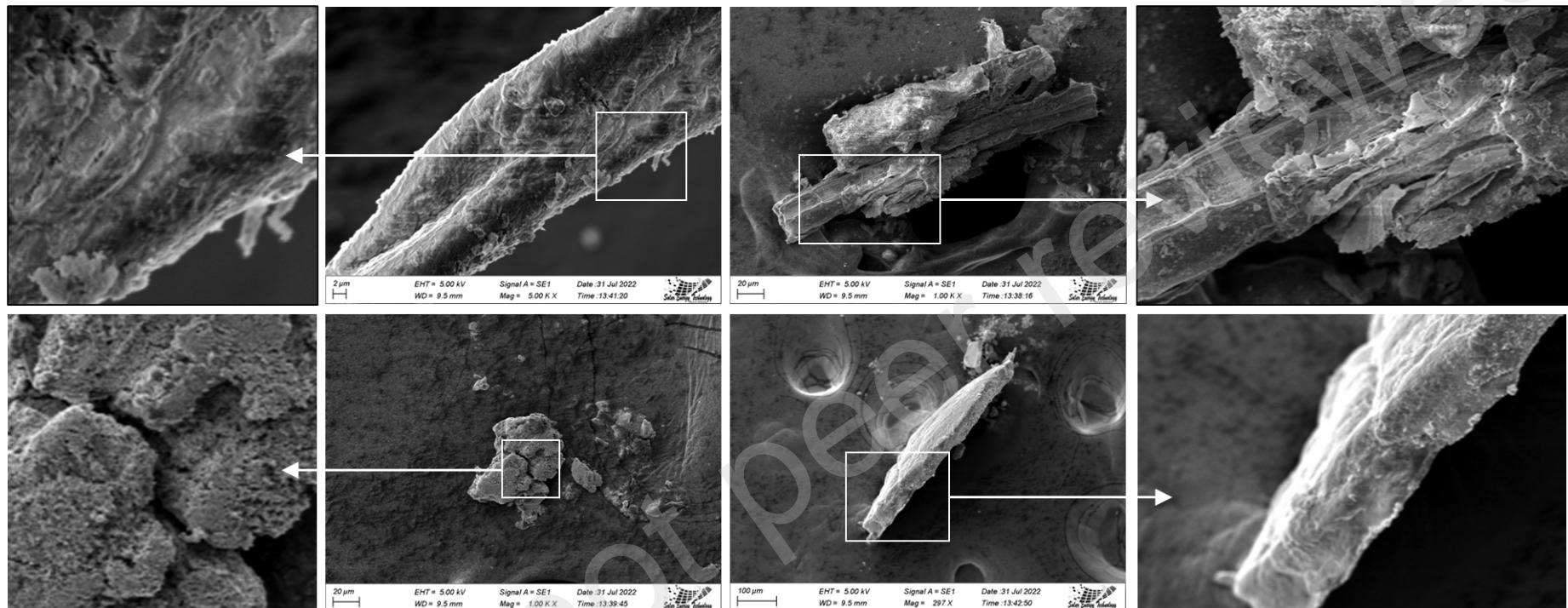


Figure 17. Representative photo electron micrographs of microplastics identified in the different samples analyzed in the present study, at different magnifications.

421

422 **4. CONCLUSION**

423 MP pollution is getting profuse attention at present in the world. Starting from
424 sediment to human blood, it has announced its presence in every possible environment.
425 Holding the ability to alter environmental processes in both terrestrial and aquatic settings,
426 this pollutant is omnipresent. The present study is the first to report the presence of
427 microplastics in the mangrove forest of Bangladesh and has found that the Sundarbans is
428 contaminated with MPs in water, sediment, and fish body at a more significant amount than
429 any other study described. 100% of the analyzed samples showed the presence of MPs, with
430 sediments mainly being contaminated. Fish feeding from the benthic zone of water also
431 showed a greater number of microplastics in their GIT than other fish species. It is also
432 possible to bioaccumulate through fish muscle and may end up in the human digestive
433 system through the food chain. MP contamination on the river, sediment, and fish pose a
434 greater risk to the ecology and wildlife of Sundarbans, as well as the inhabitants. Further
435 research on the spatial and temporal distribution of MPs along mangrove rivers is needed
436 to understand the occurrences of microplastics more precisely and provide better
437 mitigation measures to control pollution.

438

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444 productivity scholarship from CNPq (Proc. No. 308854/2021-7).

445

446 **6. DECLARATION OF COMPETING INTEREST**

447 We certify that the study's findings were not compromised by any known ethical or
448 financial conflicts of interest. We verified that all named authors had read, assessed, and
449 approved the article to see if there were more writers who matched the criteria but were not
450 mentioned. We also confirm that the authors' names appear in the article in the order in
451 which they were approved by one another. The integrity of the work has undergone a
452 thorough inspection.

453

454 **7. ETHICAL ASPECTS**

455 All experimental procedures adhered to the ethical standards for using animals in
456 research, and careful precautions were taken to lessen the suffering of the animals and their
457 exposure to outside sources of stress, pain, and discomfort. The number of animals employed
458 in the current study is within the acceptable range to provide reliable scientific results. The
459 authors of this paper do not cite any of their human studies.

460

461 8. DATA AVAILABILITY

462 The data used to support the findings of this study are available from the
463 corresponding author upon request.

464

465 9. CREDIT AUTHOR STATEMENT

466 **Nowshin Nawar:** conceptualization, sample collection, analysis, and manuscript
467 preparation. **Md. Mostafizur Rahman:** conceptualization, sample collection, analysis,
468 manuscript preparation, supervision of the study. **Shumayta Marzia:** sample collection and
469 analysis. **Mir Mohammad Ali:** sample collection, manuscript review, and editing. **Md.**
470 **Ahedul Akbor:** SEM analysis and revision of the manuscript. **Md. Abu Bakar Siddique:** SEM
471 analysis and improvement of the manuscript. **Mst Afifa Khatun:** FTIR analysis. **Md. Shajalal:**
472 FTIR analysis. **Roksana Huque:** FTIR analysis. **Guilherme Malafaia:** analysis, interpretation
473 of results, and draft manuscript preparation. All authors reviewed the results and approved
474 the final version of the manuscript.

475

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