Marine Pollution Bulletin 173 (2021) 113067

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

Baseline

Microplastics in rivers and coastal waters of the province of Esmeraldas, Ecuador

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ARTICLE INFO

Keywords:
Plastic contamination
Pacific Ocean
Urban beaches
Estuaries

ABSTRACT

This study represents the first assessment of microplastic (MP) contamination in the coastal area of the Esmeraldas Province, Ecuador. MPs were quantified in 14 coastal waters in beaches with different urbanization level and in 10 rivers. The most abundant MP types were transparent fibres, brown fragments, grey fragments, transparent fragments, and black fragments, which together represented 84% of the total count. Coastal waters presented significantly higher quantities of MP than rivers. No difference in microplastic abundance was detected between beaches with higher and lower urban occupation, nor between beaches facing North or West. Our results indicate that MP contamination is widespread, and most likely transported from multiple sources. Our results can serve as a baseline for future MP monitoring in the area.

Microplastics (MPs) are ubiquitously detected in all ecosystems and represent a major contamination issue. In coastal areas, MP contamination has been increasingly detected (De-la-Torre et al., 2020; Garcés-Ordóñez et al., 2021). There are reports of MP pollution in estuaries buried in tidal flats and in estuarine beaches (Browne, 2015), and along rivers that ultimately connect with coastal waters (Eerkes-Medrano et al., 2015). When plastics are disposed inland, rivers are considered the main vector of MP into the marine environment (Lebreton et al., 2017). Some continental sources of MPs transported by rivers to the oceans include urban waste, sewage from treatment plants, sewage system overflows, industrial waste and the breakage of larger plastic materials present in the water column (Browne, 2015; Prata, 2018). Also, because MP is widely present in ocean waters, it can be transported over long-distances and deposited in coastal areas (van Sebille et al., 2019). Along coastal areas, industrial and urban sites and estuaries have been identified as sites with high MP contamination (Browne, 2015; Haddout et al., 2021).

In the Tropical Eastern Pacific, MP contamination has been reported in the Galápagos Islands (Alfaro-Núñez et al., 2021; Jones et al., 2021; van Sebille et al., 2019), in the estuary of the Guayas River (Villegas et al., 2021), and in coastal areas of Peru (De-la-Torre et al., 2020) and Colombia (Garcés-Ordóñez et al., 2021). However, data on plastic debris in areas such as the northern coast of Ecuador is scarce (Gaibor et al., 2020). The northern coast of Ecuador is an important national and international tourism destination, and a biodiversity priority area for conservation due to the presence of threatened ecosystems (Cuesta et al., 2017). Moreover, it is a high productivity zone (Chinacalle-Martínez et al., 2021) visited by several migratory marine species (Oña et al., 2017).

Given the scarcity of studies on MP contamination in the continental coast of Ecuador, this study aims at providing a baseline of MP contamination in the rivers and coastal waters of the Esmeraldas

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https://doi.org/10.1016/j.marpolbul.2021.113067
Received 24 August 2021; Received in revised form 11 October 2021; Accepted 14 October 2021
Available online 20 October 2021
0025-326X/

province. Moreover, we aim at searching for differences between areas with difference urbanization levels and between areas with facing different geographic aspects (i.e., facing North or West). Our results can be used to track the origin and distribution of MP in the Tropical Eastern Pacific.

The Esmeraldas province is located in the northwestern coast of Ecuador and occupies an area of 15 km^2, with a population of ca. 644,000 inhabitants (INEC, 2020). It is limited to the north with Colombia and to the west with the Pacific Ocean, with a coastal strip of about 230 km. The coastal area of Esmeraldas receives cold waters from the southern pacific Humboldt current and warm waters from the northern Equatorial current.

Sampling was done in November 2020. A total of 24 surface water samples (14 sampled taken in the coastal waters and 10 samples taken in the rivers that drain to the coast) were taken along the Esmeraldas coast (Fig. 1). Sampling sites were selected according to the level of urbanization, classified as high level (sites Las Palmas, Tonsupa, Atacames, Sua, Same, Tonchigüe, Muisne) and low level (sites Las Peñas, África, Paufí, Galera, Estero de Plátano, Mompiche, Portete) (Table 1). Sampled rivers were those of the main hydrographic basins (Cayapas, Santiago, Rioverde, Esmeraldas, Teaone, Atacames and Muisne) and the rivers that flow into the coastal areas (Colope, Ostiones and Galera).

At each sampling site, two 500 mL amber glass bottles of water were taken. In the coastal areas, samples were obtained slightly away from the shore (~10 m), where the water depth was approximately 1 m. In the rivers, samples were taken in the centre of the main channel, at the middle point of the water column, against the water current. MPs extraction followed the methodologies described by Masura et al. (2015) and Villegas et al. (2021). First, water samples were filtered using a sieve of 63 μm. Then, the particles retained in the mesh were transferred using deionized water to 100 mL glass collection jars. This solution (particles retained and deionized water) was dried at 60 °C for 24 h and then digested with a solution of hydrogen peroxide (H_2O_2) 30% in an oscillation incubator (60 °C at 100 rpm for 2 h). The microparticles were subject to density separation using 15 mL of saturated sodium chloride solution (NaCl, 1200 kg m^{-3}) previously filtered on glass fibre filters (0.22 μm pore size Whatman GF/A 47 mm) due to the presence of suspended solids. The solution with the microparticles was allowed to stand for 24 h. Finally, this solution containing the floating particles was filtered with a membrane filter (0.45 μm pore size) in a vacuum filtration system. The filters were stored in capped glass Petri dishes for further visual identification.

Each filter containing MP was divided in four sections to facilitate the counting. MPs were counted using a stereomicroscope Amscope with 20× magnification, equipped with a 10 MP digital camera and the software AmScope. The patterns used to identify MPs were based on literature descriptions (Gimiliani et al., 2020; Masura et al., 2015; Mohamed Nor and Obbard, 2014), as well as on visual inspection. MPs were categorized by colour and shape. Under the stereomicroscope, the fragments were manipulated or dragged around with the aid of tweezers to confirm the static electricity property of the plastic particles. If the materials crumbled or were easily crushed, they were not considered as plastic compounds. If the particles kept their shape, they were included in the counting.

Precautions were taken to avoid background plastic contamination during sample treatment and analytical steps. All sampled bottles and laboratory materials were rinsed with Milli-Q water and then with ethanol prior to usage. Clean filter papers were placed in Petri dishes and exposed to the air in the laboratory during the processing time to account for atmospheric contamination. Blank samples were prepared with 1 L of Milli-Q water following the same methodologies used for the collection of field samples.

We applied a principal component analysis (PCA) to search for natural groups among the study sites. Variables were centred and scaled prior to the analysis. MP counts were compared by generalized linear models with nested factors (Location/Aspect/Urbanization/Site) with

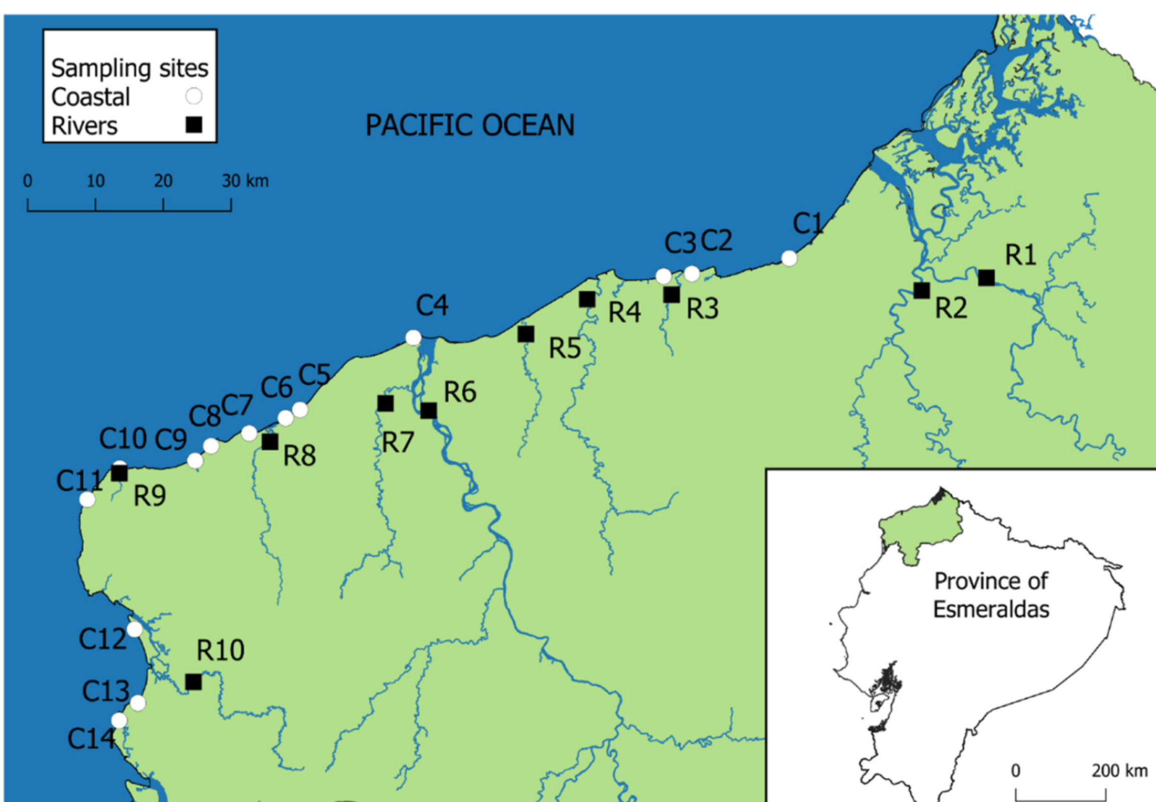

Fig. 1. Location of sampling points in coastal waters (C) and in rivers (R) in the study area.

Table 1
Site information. Coordinates are for the UTM 17S zone. Land uses categories were determined in a 2 km-buffer around the sampling points. Land use categories include forestry plantations, infrastructure and urban areas, agricultural areas, aquaculture, and water. The category "Other" include cland use classes without classification and areas with no vegetation.

	ID	Site	UTM X	UTM Y	Urban (%)	Forest (%)	Agricultural (%)	Aquaculture (%)	Water (%)	Other (%)
Rivers	R1	Santiago	733903	10118580	<1	46	47	–	7	–
	R2	Cayapas	724347	10116682	–	30	60	–	9	–
	R3	Ostiones	687475	10116098	2	7	86	3	<1	<1
	R4	Rio verde	675053	10115416	–	30	65	4	<1	–
	R5	Colope	666013	10110262	<1	42	54	2	–	1
	R6	Esmeraldas	651683	10098965	4	9	71	–	16	<1
	R7	Teaone	645334	10100019	35	15	48	–	–	<1
	R8	Atacames	628292	10094371	16	8	49	26	<1	<1
	R9	Galera	606086	10089716	1	28	70	–	<1	<1
	R10	Muisne	617010	10058982	–	4	92	2	2	–
Coast (high urbanization)	C4	Las Palmas	649427	10109728	65	13	9	–	10	3
	C5	Tonsupa	632717	10099096	64	5	28	–	–	1
	C6	Atacames	630588	10097869	54	9	24	10	3	0
	C7	Súa	625225	10095623	18	24	56	2	–	18
	C8	Same	619599	10093741	17	10	73	–	<1	17
	C9	Tonchigüe	617262	10091566	12	1	76	11	<1	12
	C12	Muisne	608344	10066724	15	10	43	4	18	15
Coast (low urbanization)	C1	Las Peñas	704830	10121448	5	1	94	–	–	–
	C2	África	690473	10119175	<1	18	77	4	–	–
	C3	Paufí	686303	10118794	<1	2	98	–	–	–
	C10	Galera	606149	10090438	2	20	77	–	<1	<1
	C11	Estero de Plátano	601359	10085849	2	45	50	–	3	–
	C13	Mompiche	608826	10055884	2	19	74	2	<1	4
	C14	Portete	606055	10053323	3	7	83	–	1	6

an inverse binomial error model. The Location factor consisted of two levels: coast and river. The Aspect factor consisted of two levels: West, that included all coastal sites south of Cape San Francisco (sites C11-C14) and North, that included the remaining coastal sites. Cape San Francisco could be assumed as a division point for the areas affected by northern or southern sea currents. Urbanization factor consisted of two levels: coastal sites with less than 10% of urban land (low) and sites with more than 10% of urban land (high), as determined by land use maps for

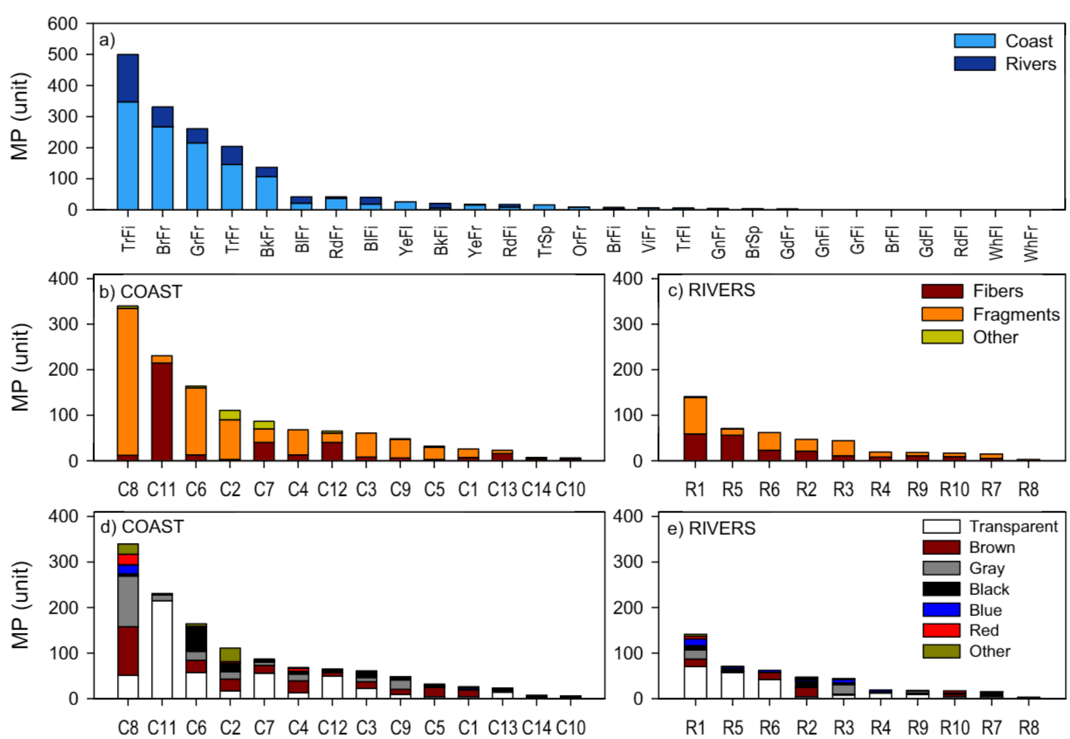

Fig. 2. a) Total count of microplastics as classified by colour and shape in costal (C) and river (R) sites. b–c) Total count of microplastics by shape. d–e) Total count of microplastics by colour. TrFi, transparent fibres; BrFr, brown fragments; GrFr, grey fragments; BkFr, black fragments; BlFr, blue fragments; red fragments; BlFi, blue fibres; YeFm, yellow film; BkFi, black film; YeFr, yellow fragments; RdFi, red film; TrSp, transparent spheres; OrFr, orange fragments; BrFi, brown fibres; VFr, violet fragments; TrFm, transparent film; GnFr, green fragments; BrSp, brown spheres; GdFr, gold fragments; GnFi, green fibres; GrFi, grey fibres; BrFm, brown film; GdFm, gold film; WhFm, white film; WhFr, white fragments. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

the area (Table 1). ANOVA-like tables were created, and multiple comparisons were done with the Tukey test. All statistical analyses were performed with R (R Core Team, 2020), using the functions *prcomp* of base R, *glm.nb* function of the MASS package (Ripley et al., 2021), *Anova* function of the car package and *pairs* and *emmeans* functions of the emmeans package (Lenth et al., 2018).

A total of 1706 MPs were registered. MP from four coastal sites (C2, C6, C8 and C11) represented 50% of the total microplastic count. The most abundant types were transparent fibres, brown fragments, grey fragments, transparent fragments, and black fragments, that together represented 84% of the total count (Fig. 2a). Among them, transparent fibres alone represented 29% of the total count. Other 22 plastic types were present in the samples, but their relative abundances were below 3%. The amount of MPs varied between 6 and 340 in coastal samples and between 3 and 141, in river samples (Fig. 2b and c).

Plastic fragments and fibres accounted for 61% and 36% of the collected MPs respectively, but their abundance varied greatly among the studied sites. Other plastics in the form of films and spheres represented less than 4% of the total count. In coastal sites, fragments were 62% of all MP types, while fibres were 33%. In rivers, fragments were 58% of all MP types, while fibres were 43% (Fig. 2b, c). Transparent plastics were the most abundant (42% of total count, Fig. 2d–e), followed by brown (20%), grey (15%) and black microplastics (9%): MP from coastal sites showed more colour variability than those from river sites. Coloured MPs were found in all river samples. Red-coloured MPs were the only type not found in river samples.

The first component (PC1) explained 28% of the data variability and was positively correlated with the percentage of MP fragments and negatively correlated with the percentages of MP fibres and transparent MP (Fig. 3). The second component (PC2) explained 16% of the data variability and was positively correlated with the percentages of black and red MP in and negatively correlated with the total count and the percentage of grey MP. River sites were located in the centre of the PCA space, while coastal sites were located in the edges of the PCA space, which suggests that rivers samples showed similar MP abundance and types among them than coastal samples. Also, high urbanized coastal sites showed lower scores along the PC2 axis than low urbanized coastal sites, which suggest higher MP abundance in high urbanized sites. Most coastal sites with a western aspect (sites C11, C12, C13) were located in group A, while most coastal sites with a northern aspect were located in group B, which suggest higher fibre percentages and lower fragment percentages in coastal sites with a western aspect.

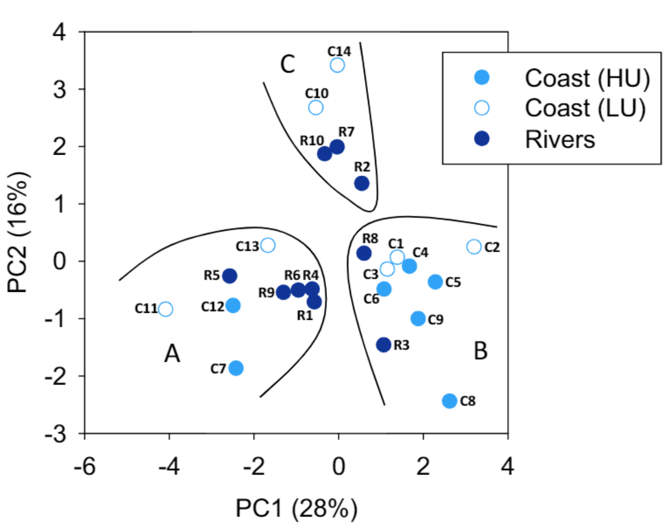

Fig. 3. Results of PCA analysis on the MP found at the study sites. Coastal sites were classified as sites with low (LU) and high urbanization (HU). Inset lines show the distribution of three natural clusters, as detected by visual inspection. Along the PC1 axis, group A, that contained samples with high percentages of fibres and transparent MPs differed from group B, that contained samples with high percentages of MP fragments. Along the PC2 axis, group C, that contained samples with low MPs abundances and with higher percentages of black and red MPs differed from the other two groups. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Samples from river sites showed significantly lower MP abundances than coastal sites ($F_{1,24} = 15.4$, $p < 0.001$, Fig. 4a) and significantly lower abundance of fragments ($F_{1,24} = 13.5$, $p < 0.01$). There were no statistically significant differences in fibre abundances between coastal and river sites ($F_{1,24} = 2.23$, $p > 0.05$). River sites also had lower abundance of transparent and coloured MPs than coastal sites (transparent: $F_{1,24} = 4.57$, $p < 0.05$; coloured: $F_{1,24} = 18.5$, $p < 0.001$).

There were no statistically significant differences in MP abundance between coastal sites with western or northern aspects ($F_{1,24} = 0.32$, n.s., Fig. 4b). However, MP composition changed as a function of the coastal aspect. There were significantly higher abundance of fragments and coloured MP and significantly lower abundance of fibres and transparent MP at the coastal sites with a northern aspect (fragments: $F_{1,24} = 20.8$, $p < 0.001$; fibres: $F_{1,24} = 50.0$, $p < 0.001$; transparent: $F_{1,24} = 10.3$, $p < 0.01$; coloured: $F_{1,24} = 29.8$, $p < 0.001$).

Multiple comparisons showed that the effect of urbanization differed also as a function of coastal aspect (Fig. 4c, d). Among sites with a northern aspect, high urbanized sites showed significantly higher abundances than low urbanized sites (total: $z = 4.08$, $p < 0.001$; fragments: $z = 3.29$, $p < 0.01$; coloured: $z = 3.60$, $p < 0.001$), but there were no statistically significant differences in fibre abundances between low and high urbanized sites ($z = 2.17$, $p > 0.05$). On the contrary, there were no statistically significant differences in MP abundances between high and low urbanization coastal sites with a western aspect (total: $z = 1.55$, $p > 0.05$; fragments: $z = 1.94$, $p > 0.05$; fibres: $z = 0.95$, $p > 0.05$; coloured: $z = 1.04$, $p > 0.05$).

Both samples from rivers and coastal sites presented elevated levels of MP compared to the studies carried out in the Tropical Eastern Pacific (Alfaro-Núñez et al., 2021; Garcés-Ordóñez et al., 2021; Jones et al., 2021; van Sebille et al., 2019; Villegas et al., 2021). Fragments and fibres were the most common MP types found, being fragments generally more abundant than fibres. MP fragments could originate from plastic disposal items associated with touristic activities, as the discarded plastic that breakdown into smaller pieces (Andrady, 2017; Gaibor et al., 2020), while the presence of fibres could be associated to untreated wastewaters that are discharged into coastal areas and estuaries (Haddout et al., 2021). The distribution of fragments and fibres in our study area could be due to the movement of suspended solids in both coastal waters and rivers (Azidane et al., 2021; Haddout et al., 2021). MP fibres tend to be transported by turbidity currents (Pohl et al., 2020) and to be removed from the suspension by sinking in the presence of suspended solids. Fragments, on the other hand, have high buoyancy and low density (Pohl et al., 2020), and could be more abundant than fibres resulted in surface waters.

No significant differences between the MP contamination in coastal areas with different urbanization levels was detected. This result contrasts with similar assessments done in beaches with various degrees of visits and access level (Abude et al., 2021; Hidalgo-Ruz et al., 2018), as MP contamination seemed to be related to the increasing of both factors. High MP contamination has been also reported in coastal areas near populated areas (Garcés-Ordóñez et al., 2021) or in areas with intensive tourism activities (Ormaza-Gonzaìlez et al., 2021). The absence of differences between MP contamination in areas with different urbanization levels could also be because we restricted our sampling to the surface water rather than to the sediments. MP contamination in water is more likely to be influenced by local marine conditions, while MP tend to accumulate over time in the sediments.

Rivers had lower abundance of MP than coastal waters. Our results contrast with the MP contamination assessment made by (Luo et al., 2019), as these authors identified a higher level of MP contamination in

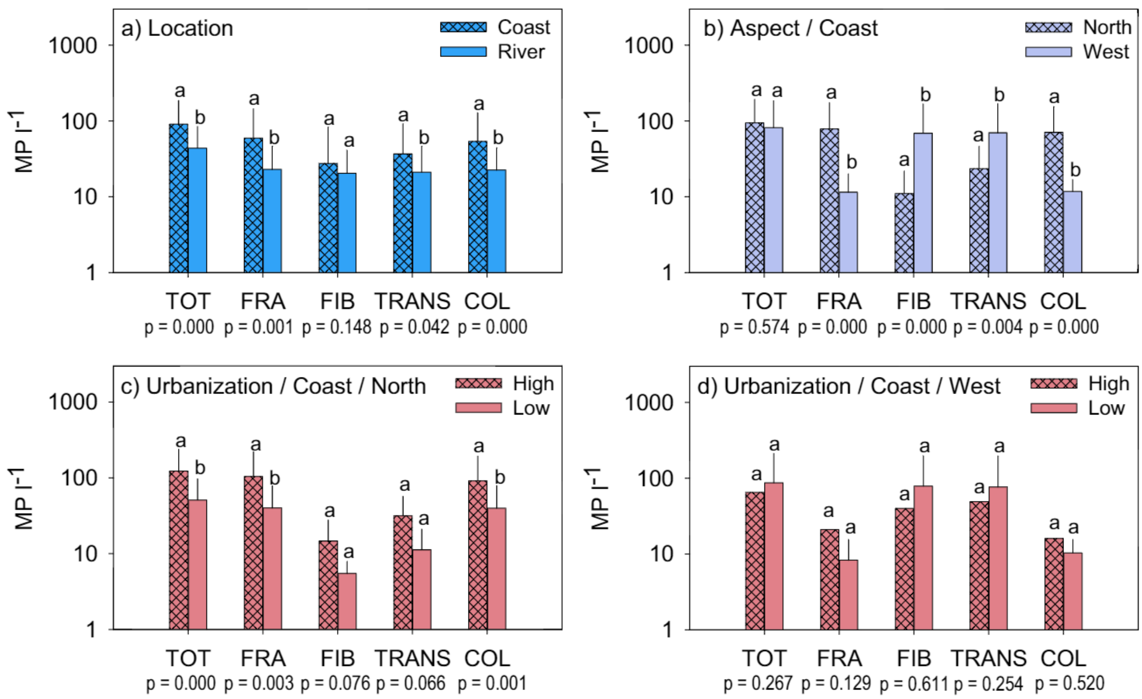

Fig. 4. Results of the nested ANOVA, indicating the comparisons of MP abundances (mean ± standard deviation, MP L^{-1}) between (a) coastal and river sites; (b) among coastal sites as a function of coastal aspect; (c) among coastal sites with a northern aspect as a function of coastal urbanization; and (d) among coastal sites with a western aspect as a function of coastal urbanization. There were no significant differences between levels with the same letter. TOT = total count; FIB = fibres; FRA = fragments; TRANS = transparent; COL = coloured.

rivers than in coastal areas in intensively urbanized and industrial watersheds. The Esmeraldas province is not highly industrialized. The rivers of this province drain through predominantly agricultural areas and low-populated urban centres (Table 1). Up to date, other contaminants, such pesticides, and agrochemicals have been of main concern than MP in rivers that drain agricultural areas in Ecuador (Deknock et al., 2019). However, given the widespread occurrence of MP in the rivers detected in our study, MP contamination in rivers should not be neglected. However, the relative contribution of inland contamination to coastal areas of Ecuador still requires further studies.

Significant differences between sites with western or northern aspects were only found regarding MP shape among sites located facing North. The absence of differences between sites with different aspects suggests that MPs are being transported from multiple sources. It is also possible that contamination in coastal waters is not necessarily brought from the closest continental contamination source, but from other continental or and offshore regions (Lusher, 2015), as the transportation process of floating plastics in the oceans is determined by dynamic marine conditions, such as wind forcing and geostrophic circulation (Li, 2019). For instance, studies have reported widespread MP contamination in coastal areas of Colombia (Garcés-Ordóñez et al., 2021), at the northern border of our study area, and in the offshore waters of Ecuador (Alfaro-Núñez et al., 2021). Thus, it is very likely that macro and microplastic and reach the coastal area of Esmeraldas after being transported by the sea currents existent near the Ecuadorian coast (Gaibor et al., 2020; van Sebille et al., 2019).

Detailed quantitative information of MP contamination can aid conservation actions towards controlling plastic disposal into coastal areas. In this study, we report a small part of an undocumented issue. Therefore, further studies are needed on the MPs removal capacity by wastewater plants and on the effective management of plastic residuals in coastal urban areas. We suggest the continuous monitoring of MP contamination along the coastal area of Ecuador.

CRediT authorship contribution statement

Mariana V. Capparelli: Conceptualization, Writing – original draft, Writing – review & editing, Supervision, Funding acquisition. **Jon Molinero:** Methodology, Formal analysis, Software, Data curation, Writing – original draft, Writing – review & editing, Funding acquisition. **Gabriel M. Moulatlet:** Validation, Data curation, Writing – original draft, Writing – review & editing. **Miren Barrado:** Investigation, Data curation, Formal analysis. **Santiago Prado-Alcívar:** Investigation, Formal analysis. **Marcela Cabrera:** Methodology, Investigation, Supervision. **Giovana Gimiliani:** Methodology, Writing – original draft, Writing – review & editing. **Carolina Ñacato:** Investigation, Formal analysis. **Veronica Pinos-Velez:** Resources, Writing – review & editing, Funding acquisition. **Isabel Cipriani-Avila:** Conceptualization, Resources, Writing – original draft, Supervision, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

We would like to thank the Corporación Ecuatoriana para el Desarrollo de la Investigación y Academia—CEDIA for their contribution through the CEPRA project CEPRA-XIV-2020-09: “Determinación del impacto y ocurrencia de Contaminantes Emergentes en ríos de la Costa Ecuatoriana y propuestas de tratamiento para su remoción”. We also thank the grant provided by Pontificia Universidad Católica del Ecuador as part of the Research Project QINV0151-IINV529020200. We are grateful to Oscar Lucas-Solis for his help in the sample processing.

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