Paulo Cezar Silva, Mariana Pacheco Almeida, Rayane Sorrentino, Luis Felipe Skinner

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Brief Communication

Baseline survey of microplastics in *Phallusia nigra* (Ascidiacea) in different sites along the Brazilian Coast, Central, and Southwest Atlantic

Paulo Cezar Azevedo da Silva[1,2*], Mariana Pacheco de Almeida[1], Rayane Sorrentino[3], Luis Felipe Skinner[1,2]

P.C.A.S.: ORCID® https://orcid.org/0000-0002-6896-8363
M.P.A.: ORCID® https://orcid.org/0009-0006-9857-3828
R.S.: ORCID® https://orcid.org/0000-0002-0937-0214
L.F.S.: ORCID® https://orcid.org/0000-0003-0971-4870

1 Universidade do Estado do Rio de Janeiro – Laboratório de Ecologia e Dinâmica Bêntica Marinha (Rua Dr. Francisco Portela 1470 – sala 172 – 24435-005 – São Gonçalo – RJ – Brazil).
2 Universidade do Estado do Rio de Janeiro – Programa de Pós-Graduação em Oceanografia (Rua São Francisco Xavier 524 – PJLF – bl. E – sala 4018 –20550-013 – Rio de Janeiro – RJ – Brazil).
3 Universidade Federal do Estado do Rio de Janeiro – Departamento de Ecologia Aquática Experimental e Aplicada (Av. Pasteur 296 – 22290-250 – Rio de Janeiro – RJ – Brazil).

* Corresponding author: bio.paulo.a@gmail.com

ABSTRACT

Microplastics (MPs) pose a growing threat to marine ecosystems, yet their accumulation in tropical ascidians remains understudied. This study investigated MPs in the digestive tract of *Phallusia nigra* from 15 sites along the Brazilian coast (75 individuals). Tissues were digested by alkaline solution, and MPs were quantified, measured, and characterized via microscopy and ATR-FTIR spectroscopy. MPs were detected in 64% of individuals (mean: 1.21 particles/individual), predominantly as fragments (63%) and fibers (37%), with an average of 0.77 ± 0.81 MP/g. The most abundant polymers were polyethylene (PE), polypropylene (PP), and nylon (PA), consistent with global coastal pollution trends. The prevalence of PE, PP, and PA, polymers widely linked to fishing gear, packaging, and sewage, underscores the role of anthropogenic sources in tropical marine pollution. Besides these findings, this study emphasizes the need for standardized methodologies to assess MPs impacts on understudied filter-feeders and the urgency of extending monitoring efforts to tropical ecosystems.

KEYWORDS: MARINE POLLUTION; FILTER-FEEDERS; POLYMER COMPOSITION; TROPICAL ECOSYSTEMS

Ascidians are marine invertebrates commonly found in benthic communities, occurring in solitary, colonial, and social forms (Zeng & Swalla, 2005). They are distributed globally and inhabit diverse ecosystems, from intertidal zones to abyssal regions (Rocha et al., 2024). These sessile organisms filter organic particles by pumping water through their incurrent siphon via cilia lining the stigmata of the branchial basket. Particles are trapped in a mucus net, rolled into a food cord along the dorsal lamina, and transported to the esophagus (Petersen, 2007; Wotton, 2020). The ascidian *Phallusia nigra* Savigny, 1816 is a solitary species characterized by its smooth black tunic, typically devoid of epibionts, often exceeding 10 cm in size (Hirose et al., 2001). Originally described in the early 19th century in the Red Sea, this species has since been recorded across tropical and subtropical regions of the Indian and Atlantic Oceans, including warm waters off Bermuda, eastern South Carolina (USA), and along the Brazilian coast from the Northeast to Santa Catarina in the south (Bonnet and Rocha 2011; Vandepas et al., 2015).

Ascidians are valuable models for studying environmental contamination, including metals, chemical pollutants, and, more recently, microplastics (MPs) (Messinetti et al., 2018a; Gallo & Tosti, 2015; Gonzalez-Pineda et al., 2025). Microplastics (MPs) (< 5 mm) are widely distributed across diverse environments and contaminate marine species through ingestion and filtration behaviors (Silva et al., 2021). Recent studies report impacts of MPs contamination on representatives of the class Ascidiacea, including digestive tract blockages, altered feeding patterns, and larval deformities (Messinetti et al., 2018a; Messinetti et al., 2019). *P. nigra* inhabits both natural and artificial substrates and has previously been documented as contaminated by microplastics (Silva et al., 2021). Therefore, this study aimed to quantify and identify the predominant synthetic polymers (via ATR-FTIR) in the digestive tracts of the ascidian *P. nigra*, an abundant, easily identifiable, and insufficiently researched species as a bioindicator, along a central stretch of the Brazilian coast, providing a basis for future monitoring studies.

Fifteen localities (Figure 1) across the states of São Paulo, Rio de Janeiro, Bahia, and Alagoas were investigated. Sampling was conducted between the summer seasons of 2021 and 2022. Five individuals were collected in each site, totaling 75 *P. nigra*. Specimens were collected manually via snorkeling at depths of 1–5 m. Sterile microbiological bags (Kasvi) were used for specimen collection and they were anesthetized with menthol diluted in seawater. Organisms were preserved in 10% formaldehyde to enable subsequent microplastic extraction. Formaldehyde fixation does not affect microplastic particles, as demonstrated by Courtene-Jones et al. (2017).

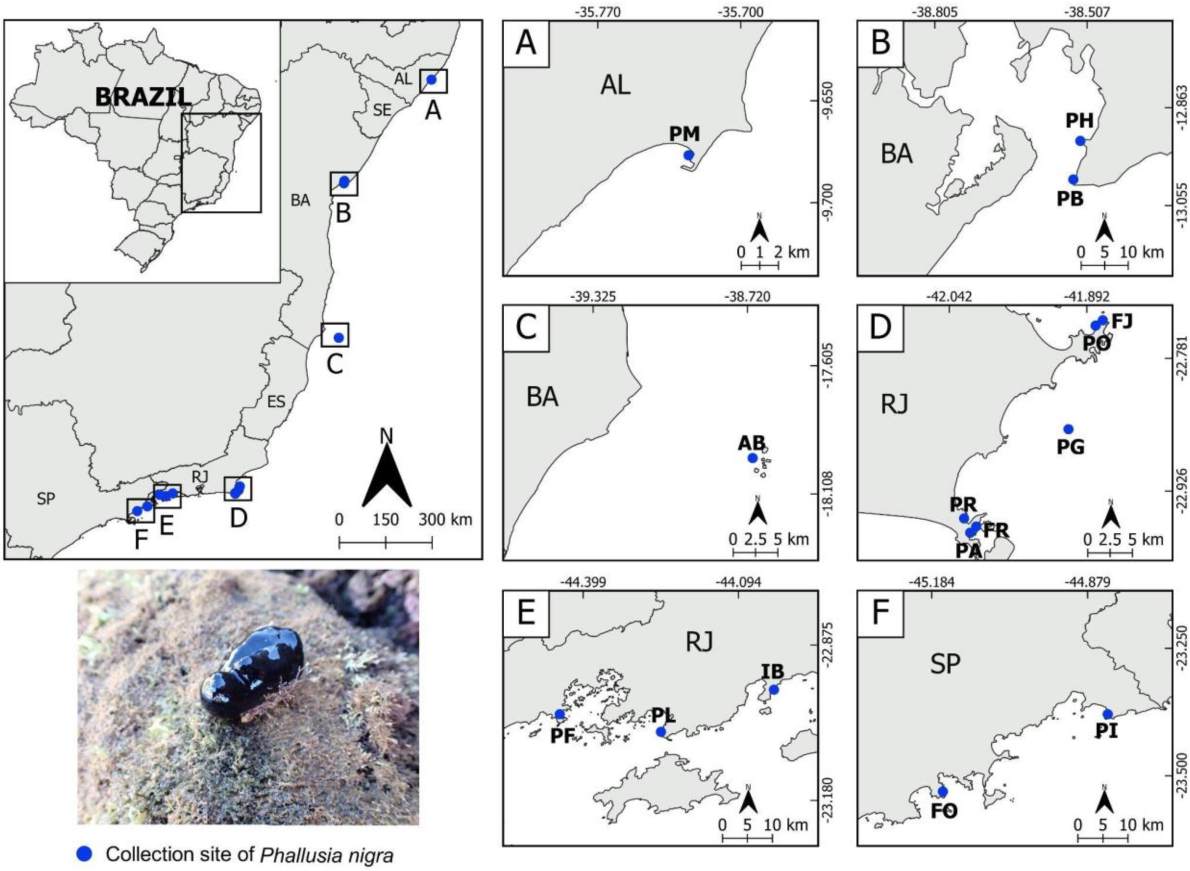

Figure 1. Collection sites of *Phallusia nigra* along the Brazilian coastline. In (A), AL stands for the state of Alagoas and (PM) for Porto de Maceió; in (B) and (C), BA stands for the state of Bahia, and PH, PB, AB refer to Porto de Humaitá, Porto da Barra, and the Abrolhos Archipelago, respectively. In (D) and (E), RJ stands for the state of Rio de Janeiro, with the locality abbreviations corresponding to FJ (Praia de São Fernando), PO (Praia dos Ossos), PG (Ilha dos Pargos), PR (Praia da Prainha), FR (Praia do Forno), PA (Praia dos Anjos), (IB) Ibicuí, (PL) Ponta Leste, and (PF) Piraquara de Fora. In (F), SP stands for the state of São Paulo, and the abbreviations (PI) and (FO) refer to Praia de Picinguaba and Praia Fortaleza, respectively.

To characterize the environmental heterogeneity of the sampling sites, we compiled key descriptors, including protection status (protected or non-protected areas), environment type (harbors, rocky shores, reefs), distance from urban areas and the coast, and potential pollution sources visually identified through satellite imagery (Google Earth, images from the 2021-2022 period). The classification of these anthropogenic sources of microplastics was based on global studies and reviews on the main entry pathways of MPs into the marine environment (Yang et al., 2021; Pourebrahimi & Majid, 2023). The predicted categories were: (1) maritime traffic (cargo ships and smaller vessels), (2) port infrastructure (including commercial ports, tourist marinas, and shipyards), (3) industrial activities (presence of nearby industries), (4) sewage and stormwater (urban morphology and land use patterns), (5) tourism (visitor waste, hotel and recreational infrastructure), and (6) nonpoint sources (e.g., atmospheric deposition and coastal roads). We emphasize that this approach is descriptive and aims to complement ecological results and discussions. It was not integrated into the statistical analyses due to the limited number of replicates per site, which precludes robust spatial comparisons. Table 1 summarizes these environmental characteristics for the 15 sites.

Table 1. Morphometric characteristics of *Phallusia nigra* individuals and quantification of microplastics (MPs) detected at each sampling site along the Brazilian coast. For each site, the following information is presented: site name and abbreviation (Site), mean body length (mean ± SD, in centimeters), mean body weight (mean ± SD, in grams), total number of detected MPs, mean number of MPs per gram of tissue (mean ± SD), mean number of MPs per individual (mean ± SD), and the percentage of individuals contaminated with MPs. Sampling sites are grouped by state: AL – Alagoas, BA – Bahia, RJ – Rio de Janeiro, SP – São Paulo. SD: standard deviation.

Locality - State	**Site**	**Environmental Protection Area (Yes or No)**	**Type of environment**	**Distance from the coast (km)**	**Distance from urban area (km)**	**Possible sources of pollution (e.g., sewage, marinas)**
Porto de Maceió - AL	PM	No	Harbor	<1	<1	Maritime traffic; Port infrastructure; Industrial activities; Sewage and rainwater; Nonpoint sources
Humaitá - BA	PH	No	Rocky coast / Bay	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Porto da Barra - BA	PB	No	Rocky coast	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Abrolhos - BA	AB	Yes (APA)	Coral reef	>60	>65	Tourism; Maritime traffic; Nonpoint sources
Praia de João Fernandinho - RJ	JF	Yes (APA)	Rocky coast	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Praia dos Ossos - RJ	PO	Yes (area of integral protection)	Rocky coast	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Ilha dos Pargos - RJ	PG	No	Rocky coast	<10	<12	Tourism; Maritime traffic; Nonpoint sources
Prainha - RJ	PR	Yes (APA)	Rocky coast	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Praia do Forno - RJ	FR	Yes (APA)	Pier	<1	<1	Tourism; Maritime traffic; Sewage and rainwater; Nonpoint sources
Praia dos Anjos - RJ	PA	Yes (Extractive Reserve)	Rocky coast	<1	<1	Tourism; Maritime traffic; Port infrastructure; Sewage and rainwater; Nonpoint sources
Ibicuí - RJ	IB	Yes (APA)	Rocky coast / Bay	<1	<1	Maritime traffic; Port infrastructure; Sewage and rainwater; Nonpoint sources
Ponta Leste - RJ	PL	Yes (APA)	Rocky coast / Bay	<1	<1	Maritime traffic; Port infrastructure; Sewage and rainwater; Nonpoint sources
Piraquara de Fora - RJ	PF	Yes (APA)	Rocky coast / Bay	<1	>2	Maritime traffic; Port infrastructure; Industrial activities; Sewage and rainwater; Nonpoint sources
Picinguaba - SP	PI	Yes (APA)	Rocky coast	<1	<1	Maritime traffic; Sewage and rainwater; Nonpoint sources
Praia da Fortaleza - SP	FO	Yes (APA)	Rocky coast	<1	<1	Maritime traffic; Sewage and rainwater; Nonpoint sources

In the laboratory, the digestive tract (stomach and intestine) of each specimen was dissected, measured, and weighed on a high-precision balance. For each specimen, the height and width of the stomach were measured, and the total area was referred to as a substitute for digestive tract area, hereafter referred to as "length" throughout the text. This metric does not correspond to the total body length of the organism, but rather to a morphometric descriptor of the digestive system specifically related to its potential particle retention capacity. The decision to isolate the digestive tract rather than analyze the entire organism was based on three interrelated considerations. First, this approach minimizes potential artifacts from external contamination, which is particularly relevant for benthic marine organisms that often have particles and epibionts adhered to their tunic. Second, focusing solely on the digestive system allows for more direct correlations between morphometric parameters and particle retention patterns, eliminating interference from other tissues. Lastly, as demonstrated by Ramos et al. (2015) and Sorrentino & Senna (2025), the presence of symbiotic organisms (amphipods and copepods) in the branchial cavity can introduce exogenous microplastics that do not represent actual ingestion by the ascidian. Silva et al. (2021) conducted a prior study in Ilha Grande Bay comparing *P. nigra* and amphipods, which showed that both groups were contaminated by microplastics. Similar methodologies have been applied to other marine invertebrates, such as sea urchins and sea cucumbers (Fagiano et al., 2023), and even to other ascidian species (Gonzalez-Pineda et al., 2025). To digest organic matter, the material was immersed for 48 hours in a 10% potassium hydroxide (KOH) solution (1:10 w:v), following Rios-Fuster et al. (2022) and Ribeiro et al. (2024a) protocols.

For filtration, qualitative filter paper (125 mm, 100 µm mesh) was used with a Prismatec 131 2VC vacuum pump and compressor. The filters were stored in Petri dishes and dried at room temperature for 24 hours. Microplastics were quantified, measured, and classified as fragments or microfibers using a Bel Photonics STEREO-ZOOM SZ/SZT Series microscope (5.6x magnification) connected to an EUREKAM 5.0 MP camera. All plastic particles larger than 5 mm were excluded from the analysis.

Microplastics were also analyzed using the available Fourier-transform Infrared Spectroscopy with an Attenuated Total Reflectance diamond crystal (ATR-FTIR), a PerkinElmer model Frontier (version 10.7.2). Spectra were obtained in the range of 400 to 4000 cm^{-1}, with a spectral resolution of 4 cm^{-1}, and eight scans were accumulated per analysis. The ATR-FTIR technique is limited by particle size and is recommended for samples larger than 500 µm (Montagner et al., 2021). Due to this limitation, only 61% of the microplastics that were measured and photographed could be analyzed by ATR-FTIR. The resulting spectra were compared with reference libraries, and particles showing consistent spectral patterns with polymeric similarity above 60% were classified accordingly. Those with lower similarity scores or inconclusive spectra were grouped as non-identified (IND), and particles identified as non-plastic materials, such as cellulose-based fibers, were excluded from the final analyses. Language grammar from the original manuscript was supported by AI-assisted tools (e.g., ChatGPT and Language Tool). All content was critically reviewed and validated by the authors.

The microplastic recovery test followed a methodology similar to that of Ribeiro et al. (2024b), conducted with 5 replicates of digestive tissue from the ascidians, with the addition of five nylon fibers (density 1.14 g/cm³) and five polystyrene fragments (density 1.05 g/cm³), both microplastics differentiated by their color. These replicates underwent the same process as the other samples. Four replicates showed 100% recovery of the added microplastics, while one replicate showed 90% recovery. During all analysis procedures, Petri dishes with distilled water were exposed as controls for airborne microplastics. The samples were then filtered, counted, and categorized. Microplastics found in control samples and visually similar to those identified in ascidians were excluded from the final results, as described by Vered et al. (2019). Preventive measures were taken to mitigate external contamination by microplastics. These included reducing the number of people in the environment, requiring researchers to wear cotton lab coats, and cleaning materials with distilled water. Additionally, bottles and containers were wrapped in aluminum foil to prevent cross-contamination by airborne particles, as described by Paiva et al. (2022).

A multiple linear regression (Python 3.9, statsmodels v0.13.2) was used to assess the influence of body weight (g) and length (mm) on microplastic accumulation in *P. nigra*. Standardized coefficients (β) and p-values (α = 0.05) were used to evaluate predictor contributions. Due to the low number of replicates per site (n = 5) and high intra-site variability (Table 1), spatial comparisons of MPs concentration (MP/g) via one-way ANOVA (scipy.stats v1.7.3) should be interpreted as exploratory; Tukey post hoc tests were applied only to significant results. An interaction term between weight and site was tested to explore potential spatial effects, but sample size limits its interpretation. Environmental descriptors (e.g., pollution sources and distance from the coast) were not included in the statistical models, which reinforces the need for future studies integrating these variables.

The size of the digestive tracts ranged from 2.5 to 6.5 cm (mean: 4.8 cm), and their weight varied between 0.64 g and 3.82 g (mean: 1.6 g) (Table 2). 89 microplastic particles (Table 2) comprising 56 fragments and 33 fibers, were identified in ascidians, and a corresponding overall mean of 1.21 ± 0.71 MPs per individual and an average of 0.77 ± 0.81 MP/g. At least one individual was contaminated at each sampling site, resulting in 64% of all individuals. The average length of fragments was 0.75 ± 0.45 mm (min = 0.09 mm; max = 2.08 mm), and microfibers was 1.78 ± 1.14 mm (min = 0.3 mm; max = 3.1 mm). 62% of all microplastics were fragments, differing from other studies investigating MPs in marine biota (Wang et al., 2019; Silva et al., 2021; Fagiano et al., 2023). However, Bonello et al. (2018), in the northern Tyrrhenian Sea, found up to 75% of microplastics were fragments in filter-feeding marine invertebrates (ascidians, mussels, and oysters). The depth of sampling and the hydrodynamic conditions at each site contribute to different concentrations of MPs in waters (Erni-Cassola et al., 2019), and according to Fagiano et al. (2023), microfibers tend to sink and are more frequently reported in sediments than in superficial waters.

Table 2. Environmental characteristics of the sampling sites along the Brazilian coast. For each site, the following information is presented: locality and state, site abbreviation (Site), presence of an Environmental Protection Area (yes or no), type of environment, approximate distance from the coast (km), approximate distance from the nearest urban area (km), and potential pollution sources. Symbols “<” and “>” indicate approximate values lower or higher than the specified distance,

respectively. APA: Environmental Protection Area.

Locality - State	**Site**	**Mean length ± SD (cm)**	**Mean weight ± SD (g)**	**Total number of MPs**	**Mean MP/g ± SD (g)**	**Mean MP/individual ± SD**	**% of individuals with MPs**
Porto de Maceió - AL	PM	5.1 ± 0.93	2.0 ± 0.6	1	0.07 ± 0.15	0.2 ± 0.45	20%
Ponta de Humaitá - BA	PH	3.18 ± 0.33	0.92 ± 0.19	1	0.22 ± 0.5	0.2 ± 0.45	20%
Porto da Barra - BA	PB	3.52 ± 0.6	0.93 ± 0.3	3	0.72 ± 0.72	0.6 ± 0.55	60%
Abrolhos - BA	AB	3.46 ± 1.04	0.75 ± 0.65	2	0.91 ± 1.31	0.4 ± 0.55	40%
Praia de João Fernandinho - RJ	JF	4.68 ± 1.07	1.46 ± 0.85	4	0.47 ± 0.65	0.8 ± 1.30	40%
Praia dos Ossos - RJ	PO	4.36 ± 0.63	1.48 ± 0.57	10	1.24 ± 0.58	2.0 ± 1.73	100%
Ilha dos Pargos - RJ	PG	4.51 ± 0.96	1.5 ± 0.45	8	1.24 ± 0.7	1.6 ± 0.55	100%
Prainha - RJ	PR	5.6 ± 1.89	2.24 ± 0.69	7	0.57 ± 0.55	1.4 ± 1.52	80%
Praia do Forno - RJ	FR	7.0 ± 1.84	2.22 ± 0.41	7	0.65 ± 0.74	1.4 ± 1.52	60%
Praia dos Anjos - RJ	PA	4.88 ± 1.56	1.74 ± 0.78	5	0.64 ± 0.84	1.0 ± 1.00	60%
Ibicuí - RJ	IB	4.82 ± 1.53	2.42 ± 0.82	11	0.87 ± 0.38	2.2 ± 1.30	100%
Ponta Leste - RJ	PL	5.58 ± 1.6	2.04 ± 0.65	9	0.92 ± 0.67	1.8 ± 1.48	80%
Piraquara de Fora - RJ	PF	3.96 ± 0.36	0.92 ± 0.15	6	1.13 ± 1.65	1.2 ± 1.79	40%
Picinguaba - SP	PI	5.6 ± 1.57	2.1 ± 0.89	7	0.75 ± 0.61	1.4 ± 1.14	80%
Praia da Fortaleza - SP	FO	3.71 ± 0.55	1.28 ± 0.28	8	1.19 ± 1.03	1.6 ± 1.34	80%

In this study, the average microplastic concentrations in *P. nigra* were consistent with those reported for Ascidia spp. in natural environments (Bonello et al., 2018; Silva et al., 2021). These results are also aligned with the global average of 1.78 ± 1.12 MPs/individual for ascidians, as reviewed by Miller et al. (2020), reinforcing that this group tends to accumulate fewer MPs than other marine invertebrates. For instance, holothurians such as *Holothuria tubulosa* and mussels like *Mytilus galloprovincialis* in the Mediterranean show significantly higher concentrations, with averages of 15.60 ± 7.44 and 5.00 ± 4.51 MPs/individual, respectively (Fagiano et al., 2023). However, this variation is considerable, as evidenced by studies in the southern Mediterranean, where *M. galloprovincialis* exhibited averages as low as 1.7 ± 0.2 MPs/individual (Digka et al., 2018). Furthermore, methodological and environmental variations, as well as differences between similar species, also influence the results. The review by Bajt (2021) on bivalves revealed a wide global variation in MPs quantities, with averages ranging from 0.23 to 12.4 MPs/individual and from 0.3 to 11.4 MPs/g wet weight, as well as particle sizes ranging from 0.005 to 4.8 mm. The average MPs per individual vary among mollusks (7.81 ± 20.67), arthropods (7.80 ± 10.05), and echinoderms (6.58 ± 5.06), which may also be related to differences in feeding strategies Miller et al., 2020. This high variability underscores the need for standardized methodologies and expanded assessments of understudied taxa, such as *P. nigra*, particularly in tropical regions where data remain scarce.

Our analysis revealed distinct patterns in MPs accumulation in *P. nigra*. A strong positive correlation was found between organism weight and MPs counts (β = 0.932; p < 0.001), indicating that heavier individuals accumulated higher quantities of particles. In contrast, body length showed a negative correlation (β = −0.511; *p* = 0.036), suggesting that larger individuals retained fewer MPs. These results align with previous findings in other ascidians, where body mass positively correlates with particle retention capacity (Jiang et al., 2008). However, the negative relationship with body length proposes a divergent mechanism, possibly involving size-dependent MPs rejection. Similar responses have been observed in *Halocynthia pyriformis* exposed to non-nutritive particles (Armsworthy et al., 2001).

While this size-related pattern is reminiscent of observations in benthic fish from China (Wang et al., 2019), the underlying mechanisms likely differ across taxa, especially given the ecological and geographic disparities. In ascidians, changes in siphon morphology and filtration efficiency across life stages (Armsworthy et al., 2001; Sherrard, 2005) may explain the negative correlation between body length and MPs retention. Although we did not distinguish organisms by age, our findings could reflect ontogenetic shifts in particle processing, which should be addressed in future targeted experiments. Beyond ingestion, evidence from other ascidian species suggests that digestive processes can alter MPs properties and promote the formation of fecal pellets with enhanced sinking rates (Harel et al., 2024). This indicates that *P. nigra* may also contribute to MPs removal from the water column, acting both as a sink and a transporter of MPs to deeper layers. Such a role underscores the importance of investigating its contribution to vertical MPs flux in marine ecosystems.

Notably, microplastics concentrations did not vary significantly across collection sites (ANOVA, *p* = 0.189), though this result should be interpreted with caution due to the low sample size per site (n = 5) and high environmental heterogeneity (e.g., proximity to anthropogenic activity, oceanic vs. coastal influences). Exploratory analyses detected localized weight × site interactions (significant in Piraquara de Fora and Praia dos Ossos), but the lack of consistent spatial trends and standardized environmental metadata precludes further interpretation. Given this heterogeneity and the absence of standardized environmental data, we refrain from interpreting site-specific effects.

Of the particles analyzed by ATR-FTIR (n = 58), 8.6% were identified as non-plastic materials (e.g., hydroxypropyl cellulose and sodium alginate salt) and were excluded from the polymeric analysis. The presence of these cellulose-based items has been reported in similar studies (Remy et al., 2015), likely originating from textile or biopolymer-derived sources, also detected in the marine environment. Future research should consider the use of complementary methodologies, such as Raman or μFTIR, for a more accurate identification of these materials. Among the remaining particles (n = 53), 64.8% were characterized as high-density polyethylene (PE-HD), 16.7% as polypropylene (PP), 7.5% as polyamide 6 (PA6), 5.5% as ethylene-vinyl acetate (EVA), 3.7% as polyethylene terephthalate (PET), and 1.8% as low-density polyethylene (LDPE) (Figure 2).

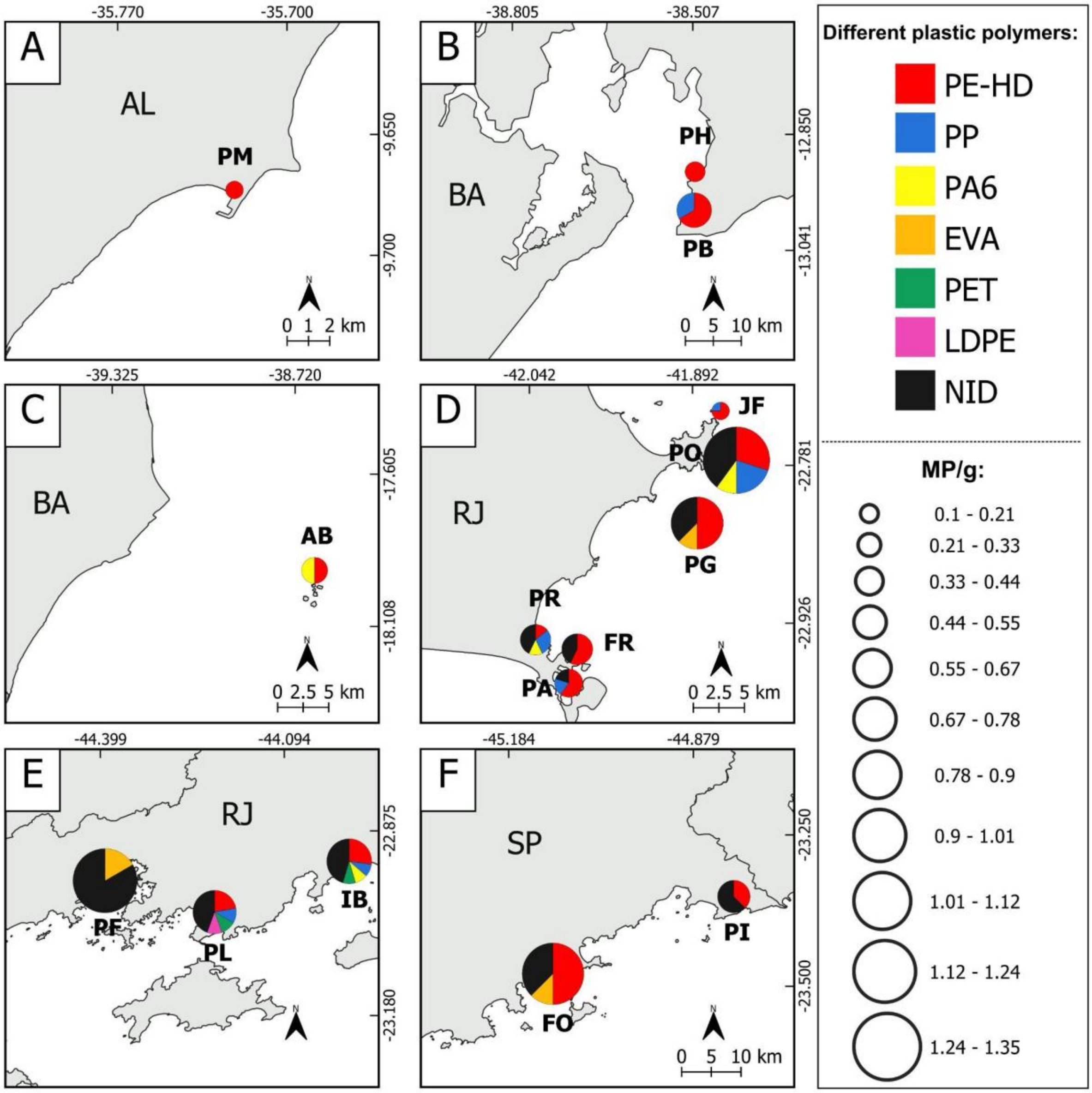

Figure 2. Spatial distribution and polymer composition of plastic particles found in *Phallusia nigra* samples along the Brazilian coastline. At each collection site, the pie charts represent the percentage of particles of different types of plastic polymers identified: PE-HD (red), PP (blue), PA6 (yellow), EVA (orange), PET (green), LDPE (pink), and NID (black, non-identified polymers based on ATR-FTIR technique). The size of the circles is proportional to the average microplastic concentration per gram of tissue (MP/g) in the organisms, as indicated by the scale on the right. Collection areas are grouped by state: (A) AL – Alagoas (PM: Porto de Maceió); (B–C) BA – Bahia (PH: Porto de Humaitá, PB: Porto da Barra, AB: Abrolhos Archipelago); (D–E) RJ – Rio de Janeiro (JF: Praia de São Fernando, PO: Praia dos Ossos, PG: Ilha dos Pargos, PR: Praia da Prainha, FR: Praia do Forno, PA: Praia dos Anjos, IB: Ibicuí, PL: Ponta Leste, PF: Piraquara de Fora); (F) SP – São Paulo (PI: Praia de Picinguaba, FO: Praia da Fortaleza).

Although PE-HD was the only polymer identified in *P. nigra* from Porto de Maceió (Alagoas), this finding is consistent with Dantas-Filho et al. (2024), who reported the presence of nanoplastics in the gastrointestinal tract of shrimp from Pontal do Peba, predominantly polyethylene, nylon (PA6), and polyester, in an area also influenced by port activities and riverine discharges. Surprisingly, despite the expected influence of port areas, neither Porto de Maceió nor Porto da Barra (Bahia) showed elevated MP concentrations in our results compared to more preserved sites. Sorrentino et al. (2025) highlighted that, in Bahia, particularly in Todos os Santos Bay, factors such as tourism, urban expansion, high-energy estuaries, deposition of fine sediments, and proximity to major population centers and urban drainage contribute to the accumulation of MPs in sediments.These findings are also consistent with Maynard et al. (2021), who reported the predominance of polyethylene (PE) and PP in coastal sediments of Bahia, emphasizing the combined influence of local land-based sources and hydrodynamic transport in shaping MP distribution. Despite this, these factors were not sufficient to increase MP concentrations in *P. nigra* compared to other sites studied. These observations, although descriptive, suggest that the polymeric composition found in the gastrointestinal tracts of P. nigra may integrate signals from both local sources and large-scale transport processes and/or reflect differences in sampling characteristics such as seasonality, collection depth, and high stress levels during sampling.

A subtle increase in the mean MP concentration per gram of tissue was observed from Alagoas toward southeastern sites (RJ and SP), and beyond PE and PP, a greater variety of plastic polymers was observed. This pattern may be related both to the greater intensity of coastal exploitation and tourism in these regions and to oceanographic factors that redistribute MPs along the Brazilian coast. As described by Ivar do Sul et al. (2014), currents and wind-driven processes transport buoyant polymers from distant sources to coastal and oceanic areas, including Abrolhos. Similar results were reported by Imsaurriaga et al. (2024) for islands in southeastern Brazil, where local fishing activity, maritime traffic, and hydrodynamic retention act in combination with regional-scale transport processes of microplastics.

Nevertheless, the polymeric composition observed in this study is strongly dominated by conventional plastics, particularly PE-HD and PP, reflecting their extensive industrial use and predominance in the global plastics market. In 2023, these two polymers accounted for 31.2% of global plastic production (PLASTICS EUROPE, 2024), and in Brazil, they are the leading thermoplastic materials, particularly in key sectors that account for over 35% of national plastic consumption, including the construction sector (28.32%) and vehicle components (7.25%) (ABIPLAST, 2023). This correspondence between production and consumption patterns and the polymer profiles found suggests that the composition of microplastics in the environment may be closely linked to the industrial and commercial flows of these materials.

Microplastics (MPs) commonly found on beaches and coastal areas tend to reflect global patterns of production and disposal, with polyethylene (PE) and polypropylene (PP) being the most abundant in the marine environment (Andrady, 2015). This trend is supported by studies in various regions: Hawaii (PE: 85%; PP: 14%) (Carson et al., 2011), Italy (PE: 48%; PP: 34%) (Vianello et al., 2013), and coastal sediments in Belgium (PP, PS, and PE) (Claessens et al., 2011). In Brazil, the patterns are consistent, with Pelegrini et al. (2024) and Ricardo et al. (2024) reporting high concentrations of PE, PP, and nylon (PA) on the southern coast, while Nolasco et al. (2022) identified PP, PA, and PE as dominant in Fortaleza (Northeast). The latter highlights that the entry of these polymers into the marine environment is linked to fishing activities, packaging waste, and domestic sewage, reinforcing the role of anthropogenic sources. Additionally, studies in the coastal areas of Rio de Janeiro (Southeast) (Silva et al., 2022; Carvalho et al., 2025) corroborate the prevalence of PE, PP, and polystyrene.

A recent meta-analysis revealed that PE and PP exhibit higher relative abundance in the surface layer (PE: 42%; PP: 25%) compared to the water column (PE: 9%; PP: 3%) and intertidal zones (PE: 18%; PP: 5%) (Erni-Cassola et al., 2019). Despite this prevalence, studies indicate that these polymers are also widely distributed across different environmental compartments, including sediments and marine biota. Frère et al. (2017) in France, Wang et al. (2019) in the Yellow Sea (China), and Fagiano et al. (2023) in the Western Mediterranean Sea observed that PE and PP were the most abundant polymers in the marine environment, but with low occurrence in invertebrates (holothurians, urchins, and mussels) and fish (Fagiano et al., 2023). In contrast, Rios-Foster et al. (2022) in Spain reported high concentrations of PE and PP in holothurian and mussel species. A review by Bajt (2021), which compiled data on MPs contamination in various benthic filter-feeding invertebrates (such as *Mytilus spp*., *Crassostrea gigas*, and *Perna viridis*), identified the presence of multiple polymer types in these organisms, with a predominance of PE and PP, but also including PET, PS, PVC, EVA, polyester, and nylon, among others. This diversity of MPs detected in organisms with similar ecology to ascidians further emphasizes that plastic contamination, particularly PE and PP, is a widely distributed phenomenon in marine filter feeders.

The chemical components of MPs has been indicated by several authors as toxic for ascidians, like the reduction of larval metamorphosis rates in *Ciona robusta* (Messinetti et al., 2018a) caused by polystyrene MPs Plastic additives may also produce taxon-specific responses to plastic additives, particularly during larval development of *C. robusta* and *C. intestinalis* (Mercurio et al., 2022). Exposure to bisphenol A (BPA) may cause embryonic developmental disorders, phenotypic alterations, and pigmented organ malformations in *Phallusia mammillata* (Messinetti et al., 2018b). In addition to their physical and cumulative environmental impacts, microplastics (MPs) can serve as substrates for adsorbing persistent organic pollutants (POPs), heavy metals, and pharmaceuticals (Guzzetti et al., 2018), thereby amplifying risks to marine biota. Despite these findings, key physiological aspects, including filtration selectivity, retention efficiency, and bioaccumulation dynamics, remain poorly understood, especially under varying environmental conditions (Vered et al., 2019).

In Brazil, most studies on microplastics in marine invertebrates have focused on filter-feeding species consumed by humans, particularly bivalves (Bom et al., 2022; Ribeiro et al., 2023; Neves et al., 2024; Jankauskas et al., 2024). Although less frequently, some research has focused on investigating MPs contamination in other marine invertebrates such as gastropods, sponges, and ascidians (Silva et al., 2021; Soares et al., 2022; Ribeiro et al., 2024b). This narrow taxonomic focus presents significant gaps and limitations that neglect ecologically important organisms whose unique physiological traits, trophic roles, and ecosystem functions could serve as valuable bioindicators, potentially providing novel perception into the dynamics of plastic contamination in coastal ecosystems.

Although the ANOVA did not reveal significant differences among sites (p = 0.189), the detection of MPs even in protected areas, including Parque Nacional Marinho de Abrolhos (Bahia), Piraquara de Fora (Estação Ecológica de Tamoios, Rio de Janeiro), and Picinguaba (Parque Estadual da Serra do Mar, São Paulo), suggests that: (i) the low replication (n = 5) and high intra-site variability may have limited our statistical power, and/or (ii) factors beyond simple geographic location (e.g., biological traits of *P. nigra*) may regulate MPs accumulation. The limited sample size may have obscured more subtle spatial patterns, as evidenced by the high variation in MPs concentrations among individuals from the same site, with differences of up to fivefold between the minimum and maximum values.

Notably, the polymeric profile was dominated by PE and PP, followed by smaller proportions of PA6, PET, and EVA, mirroring the patterns reported in global coastal environments and reflecting their extensive industrial use in packaging, fishing gear, and consumer goods. This reinforces the need for integrated approaches that not only quantify microplastic abundance but also characterize their composition to better trace potential sources and ecological impacts.

To advance our understanding of the drivers of MPs accumulation in tropical ascidians, future studies should: (i) substantially increase sampling effort, (ii) standardize collection protocols (e.g., organism size range, sampling depth), (iii) incorporate quantitative metrics of anthropogenic pressure and oceanographic variables, and (iv) conduct controlled experiments to clarify species-specific particle retention mechanisms. This integrated approach will be essential to properly evaluate the bioindicator potential of *P. nigra* and other neglected taxa in tropical coastal ecosystems.

AI USE DISCLOSURE

Artificial intelligence tools (ChatGPT and Language Tool) were used exclusively to refine the English language of this manuscript. The content was carefully reviewed by the authors to ensure consistency and correctness, and the authors are fully responsible for the final version of the manuscript.

DATA AVAILABILITY STATEMENT

All data are available from the corresponding author upon reasonable request.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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AUTHOR CONTRIBUTION

P.C.A.S.: Conceptualization; Formal Analysis; Investigation; Writing – original draft; – review & editing.
M.P.M.: Conceptualization; Methodology, Writing – original draft; Writing – review & editing.
R.S.: Conceptualization; Investigation; Writing – original draft; Writing – review & editing.
L.F.S.: Conceptualization; Supervision; Project Administration; Funding Acquisition; Writing – review & editing.

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