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The role of mesopelagic fishes as microplastics vectors across the deep-sea layers from the Southwestern Tropical Atlantic☆

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A R T I C L E I N F O

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A B S T R A C T

Microplastics (MPs; <5 mm) are a macro issue recognised worldwide as a threat to biodiversity and ecosystems. Widely distributed in marine ecosystems, MPs have already been found in the deep-sea environment. However, there is little information on ecological mechanisms driving MP uptake by deep-sea species. For the first time, this study generates data on MP contamination in mesopelagic fishes from the Southwestern Tropical Atlantic (SWTA) to help understand the deep-sea contamination patterns. An alkaline digestion protocol was applied to extract MPs from the digestive tract of four mesopelagic fish species: *Argyropelecus sladeni*, *Sternoptyx diaphana* (Sternoptychidae), *Diaphus brachycephalus*, and *Hygophum taaningi* (Myctophidae). A total of 213 particles were recovered from 170 specimens, and MPs were found in 67% of the specimens. Fibres were the most common shape found in all species, whereas polyamide, polyethylene, and polyethylene terephthalate were the most frequent polymers. The most contaminated species was *A. sladeni* (93%), and the least contaminated was *S. diaphana* (45%). Interestingly, individuals caught in the lower mesopelagic zone (500–1000 m depth) were less contaminated with MPs than those captured in the upper mesopelagic layer (200–500 m). Our results highlight significant contamination levels and reveal the influence of mesopelagic fishes on MPs transport in the deep waters of the SWTA.

1. Introduction

Since its invention, plastic production has risen considerably, reaching up to 348 million tons (Mt) in 2017 (PlasticsEurope, 2018), with a prognosis to hit 1100 Mt by 2050 (Geyer, 2020). Vast quantities of plastic materials are mismanaged or illegally discarded in marine ecosystems (Koelmans et al., 2017; Ostle et al., 2019). Land-based sources contribute to about 80% of plastics entering the oceans (Andrady, 2011) via riverine discharges (Meijer et al., 2021). In marine ecosystems, plastic debris is weathered by natural processes (*e.g.*, hydrodynamics, solar radiation and interaction with biota; Jambeck et al., 2015; Thompson et al., 2004) and eventually fragmented into microplastics (MPs, < 5 mm; Arthur et al., 2009).

MPs are widely distributed all over the marine environment, from urban coastal areas (Lins-Silva et al., 2021) to remote regions such as the Arctic and Antarctic polar seas (Lusher et al., 2015; Waller et al., 2017). MPs accumulate in the ocean gyres (Jiang et al., 2020) due to the interaction of winds and rotatory ocean currents. In the Atlantic Ocean, remote islands are known to be contaminated with MPs, as is the case of Falklands and Ascension Islands (Green et al., 2018); the Canary Islands (Álvarez-Hernández et al., 2019); Abrolhos Archipelago, Fernando de Noronha Archipelago, and Trindade Island (Ivar do Sul et al., 2013, 2014). In the short term, these islands might retain MPs in the nearshore due to the actions of winds, waves, vortices, and eddies surrounding the

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islands (Lima et al., 2016). Nevertheless, not only the sea surface is impacted by MPs, but also the deep sea, which has been pointed out as a major MPs reservoir (Woodall et al., 2014). Indeed, MPs have already been observed in the subsurface waters, sediments, and fauna of the deep sea (Lusher et al., 2016; Courtene-Jones et al., 2017; Choy et al., 2019; Jamieson et al., 2019; Kane et al., 2020). However, processes involved in the dispersion and fate of MPs into deeper ocean layers are still poorly understood.

MPs can be transported from the surface to deep waters through interaction with marine communities. For example, giant larvaceans can pack MPs filtered in the surface into faecal pellets that quickly sink to the seafloor (Katija et al., 2017; Choy et al., 2019). MPs incorporation into marine snow is hypothesised to be the main sinking mechanism for buoyant polymers (Kvale et al., 2020). Additionally, many deep-sea species undertake epipelagic vertical migrations to feed (Eduardo et al., 2020a) and may act as biological plastic transporter whenever contaminated with MPs (Ferreira et al., 2022). Although the role of mesopelagic fishes in the vertical movement of MPs in the water column has been proposed, it is still not well understood (Lusher et al., 2016; Savoca et al., 2021). Thus, widespread MPs pose several threats to marine biota (Galloway et al., 2017), as they can easily be mistaken with prey and ingested by marine species (Boerger et al., 2010). Furthermore, they might be transferred from prey to predator through trophic interactions (Ferreira et al., 2016, 2019; Nelms et al., 2018). Once ingested, MPs can cause digestive damage, decrease predatory efficiency, and induce toxic effects (Teuten et al., 2007; Moore, 2008; de Sá et al., 2015; Barboza et al., 2018). Moreover, MPs can adsorb and concentrate pollutants available in the ocean (*e.g.*, persistent organic pollutants and heavy metals; Oehlmann et al., 2009; Ashton et al., 2010; Rochman et al., 2013c; Jamieson et al., 2017) or release their additive burden (Paluselli et al., 2019; Fauvelle et al., 2021), and may be bioaccumulated and biomagnified in the food web (Teuten et al., 2009; Batel et al., 2016).

The mesopelagic layer (200–1000 m) hosts remarkable marine biodiversity that plays a pivotal role in sequestering carbon, recycling nutrients, and acting as a key trophic link between primary consumers and higher trophic levels (*e.g.*, larger fishes, mammals, and seabirds; Drazen and Sutton, 2017; Eduardo et al., 2020a). Additionally, many mesopelagic species migrate vertically to the upper ocean layers to feed at night and return to deep waters during daylight, contributing to the connection between shallow and deep-sea ecosystems (Davison et al., 2013; St. John et al., 2016; Eduardo et al., 2020b).

MP ingestion by mesopelagic fishes has been already reported all over the world, as observed in the North Pacific Central Gyre (Boerger et al., 2010), North Pacific Subtropical Gyre (Davison and Asch, 2011), North Atlantic (Lusher et al., 2016; Wieczorek et al., 2018), Mediterranean Sea (Romeo et al., 2016), South China Sea (Zhu et al., 2019), and in the South Atlantic, around the Tristan da Cunha and St. Helena islands (McGoran et al., 2021). However, this group is still poorly investigated in deep waters due to sampling difficulties (*e.g.*, high sampling cost and operational complexity), especially in the least developed countries (Howell et al., 2020). To date, no study has investigated MP contamination in fishes inhabiting the mesopelagic zone of the Southwestern Tropical Atlantic (SWTA). Located in the SWTA, the Fernando de Noronha Archipelago (FNA) is essential for the conservation of the marine biodiversity in the tropical oceanic region, as it serves as a shelter, reproduction and nursery area for several species, including the mesopelagic fishes (Lima et al., 2016; Eduardo et al., 2020a; Martins et al., 2021).

Hatchetfishes (Sternoptychidae) and lanternfishes (Myctophidae) are among the most abundant and widespread mesopelagic fish groups in the world (Gjøsaeter and Kawaguchi, 1980; Eduardo et al., 2020a, 2021). These groups present an essential linkage between the epipelagic producers and deep-sea predators since they represent a key energy source in the mesopelagic zone (Eduardo et al., 2020a, 2020b, 2021).

Within the SWTA, four species in the mesopelagic compartment are outstanding in terms of abundance and/or vertical migration: the sternoptychids *Argyropelecus sladeni* Regan, 1908 and *Sternoptyx diaphana* Hermann, 1781; and the myctophids *Diaphus brachycephalus* (Tåning, 1928) and *Hygophum taaningi* Becker, 1965. These species are zooplanktivorous, feeding primarily on fish larvae, amphipods, gelatinous, and euphausiids (Drazen and Sutton, 2017; Eduardo et al., 2020a, 2021). Furthermore, they all perform diel vertical migration, ascending to the epipelagic zone at night mainly to forage and avoid predators (Eduardo et al., 2020a, 2021). However, these species present strong niche segregation, belonging to functional groups with different diet preferences, isotopic composition, and vertical distribution (Eduardo et al., 2020a, 2021). These ecological differences, therefore, might also influence MP uptake.

In this study, we identify the patterns of MP contamination in mesopelagic fishes from the SWTA and their relationship with different ecological habits. Specifically, this study aims (*i*) to describe the occurrence of MP contamination in four mesopelagic species from the SWTA, (*ii*) to identify the main shapes and polymer nature of the ingested particles, and (*iii*) to investigate whether there are differences in MP ingestion rates according to depth and period (day or night).

2. Materials and methods

2.1. Study area

The study area is located along the Fernando de Noronha Ridge, SWTA, with oligotrophic and warm waters influenced by the South Equatorial Current (SEC) and South Equatorial Undercurrent (SEUC) (Assunção et al., 2020), specifically the Fernando de Noronha Archipelago (FNA), Rocas Atoll (RA), and adjacent seamounts (Fig. 1). These areas are important for marine biodiversity and are recognised as an EBSA "Ecologically and Biologically Significant Marine Area" (CBD, 2014). Furthermore, FNA is inserted in a Marine Protected Area (MPA), with a National Marine Park (PARNAMAR) and an Environmental Protection Area (EPA), which is classified as a UNESCO natural heritage. The RA is also inserted in an MPA, and it is situated at the top of a submarine mountain chain, with its base at 4000 m depth, located 148 km west of the Fernando de Noronha Archipelago (Soares et al., 2010).

2.2. Sample collection and laboratory procedures

Mesopelagic fishes were collected using a micronekton trawl (body mesh: 40 mm, cod-end mesh: 10 mm) during the day and at night, from 90 to 800 m depth for 30 min at 2–3 kt (Eduardo et al., 2020b). Samples were collected along the Fernando de Noronha Ridge during the scientific survey ABRACOS 2 (Acoustics along the BRAzilian COaSt), carried out from 9th April to May 6, 2017, onboard the French RV *Antea* (Bertrand, 2017). After each sampling, the specimens were labelled, frozen, and subsequently identified.

Four mesopelagic species were selected for this study: *Argyropelecus sladeni* ($n = 15$); *Sternoptyx diaphana* ($n = 33$); *Diaphus brachycephalus* ($n = 69$); and *Hygophum taaningi* ($n = 53$). Specimens were measured (nearest 0.1 cm of total length and standard length), weighed (nearest 0.01 g of total weight), and dissected (Table 1). The digestive tracts (stomach and intestine) were carefully removed, weighed, and frozen again for the digestion analysis.

2.3. Contamination control

Before the extraction procedures, several steps were carefully carried out to ensure quality assurance/quality control (QA/QC) and avoid possible airborne and cross-contamination, following the protocol described by Justino et al. (2021). This QA/QC includes using 100% cotton lab coats, face masks, and disposable gloves in a cleaned and reserved room, with a limited flow of people during the whole process. Additionally, all solutions were filtered using a vacuum pump system

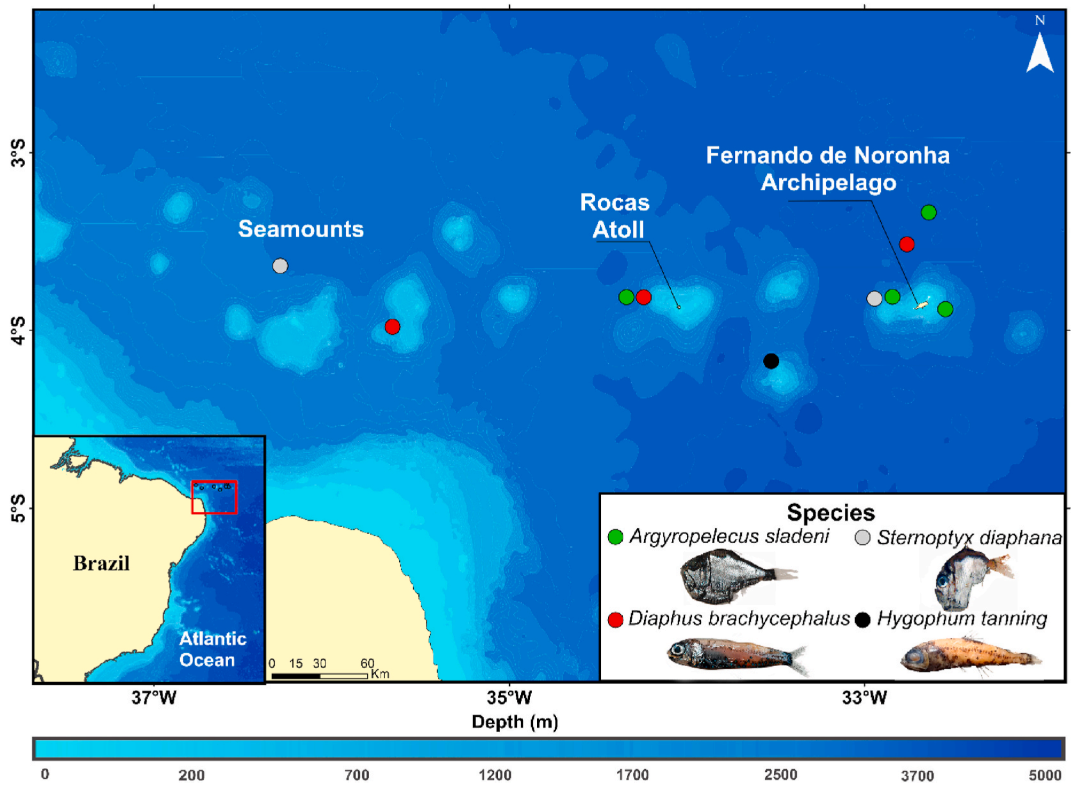

Fig. 1. Fernando de Noronha Ridge, off northeastern Brazil (STWA). Sampling stations for each species are indicated by coloured circles.

Table 1
Biological aspects and sampling data of the species analysed. Abbreviations: SL, standard length; TW, total weight; FO%, frequency of occurrence; SD, standard deviation.

Family/Species	Sampling		Biometry		Microplastics occurrence		
	n	Depth (m)	SL (cm) range	TW (g) range	FO%	MPs mean ± SD	Length (mm) mean ± SD
Sternoptychidae							
Argyropelecus sladeni	15	430; 610; 615; 800	3.00–5.85	0.70–3.18	93	1.66 ± 1.23	0.74 ± 0.53
Sternoptyx diaphana	33	615; 800	1.92–3.06	0.18–0.97	45	0.54 ± 0.71	0.36 ± 0.82
Myctophidae							
Diaphus brachycephalus	69	230; 610; 700	2.51–4.98	0.34–2.15	75	1.63 ± 1.41	0.40 ± 0.55
Hygophum taaningi	53	90	4.13–5.99	1.14–2.68	62	1.07 ± 1.20	0.49 ± 0.80

(equipped with laboratory glassware) through a 47 mm GF/F 0.7 μm pore size glass fibre filter (Whatman). Extraction tools were cleaned with ethanol 70%, rinsed with filtered distilled water and checked for contamination.

Before starting the chemical digestion, blank procedures were done for each set of 10 samples. For the blanks, a beaker was filled with 50 mL of NaOH (1 mol L^{-1}) solution, covered with a glass lid, and then treated with the same protocol applied to the samples (see next section). A total of 4 particles were observed in the blank procedures, of which two were filaments (one red and one white), and two resembled paint chips (blue). The red filament was further identified as polylactic acid (PLA), and the blue particle resembled a paint chip as styrene-butadiene rubber (SBR). Particles identified in the samples with any similarity to those observed in the blanks were excluded from further analysis.

2.4. Microplastic extraction protocol

An alkaline digestion protocol using sodium hydroxide (NaOH) was used for extracting MPs from the digestive tract of fish (Justino et al., 2021). Digestive tract samples were rinsed with filtered distilled water to remove any particles adhering to the external tissue before being placed in a beaker and submerged in NaOH (1 mol L^{-1}; PA 97%) solution (the proportion used was 1:100 (w/v), i.e. 1 g of digestive tract weight for 100 mL NaOH solution), covered by a glass lid and oven-dried at 60 °C for 24 h. After that, samples were filtered using a vacuum pump system through a 47 mm GF/F. After filtration, samples were carefully set in a Petri dish and covered. These filters were oven-dried again at 60 °C for 24 h. Then, filters were visually examined for MPs identification using a stereomicroscope (Zeiss Stemi 508, with 40–50 times magnification with a size detection limit of 0.07–5 mm). The particles suspected to be MPs were photographed (Axiocam 105 Color), counted, and measured in length (mm) (Zeiss Zen 3.2). MPs were categorised according to their shape (Fig. 2; Justino et al., 2021) as fibres (filamentous shape), fragments (irregular shape), films (flat shape), foams (soft with an irregular shape), or pellets (spherical shape).

2.5. Laser direct infrared (LDIR) analysis of MPs polymers

A subset (10% of the total particles extracted) of samples was selected to identify the main types of MPs polymers using the LDIR analyser Agilent 8700 Chemical Imaging System using the Microplastic Starter 1.0 library. The LDIR analyser scans the particles (size range 20–5000 μm) in an automatic mode and obtains a spectral curve using a wavelength range of 1800–975 cm^{-1}. The information is collected with

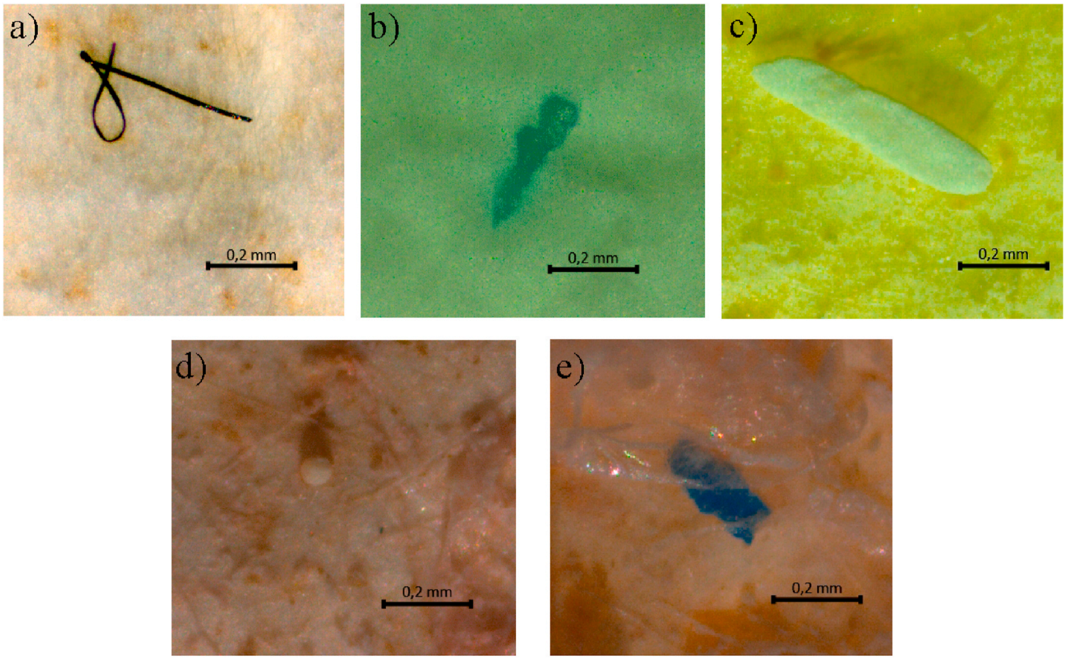

Fig. 2. Shapes of microplastics identified in the mesopelagic fishes: a) fibre; b) fragment; c) foam; d) pellet; e) film.

the Clarity image software (© Agilent version 1.3.9) and compared with the polymer spectrum library (~400 references spectra). A particle was considered as identified if the accordance of its spectrum with the reference spectrum was ≥70% (Ourgaud et al. In prep).

2.6. Data analysis

Kruskal-Wallis test was used to verify whether ingested MPs presented significant differences among species (*A. sladeni*, *S. diaphana*, *D. brachycephalus*, and *H. taaningi*) considering the number and size of MPs. We also used Kruskal-Wallis to test whether the total number of MPs ingested varied according to depth. When the Kruskal-Wallis test presented significant differences, *post hoc* pairwise comparisons, Dunn's test was used to investigate the sources of variance (Dunn, 1964). Mann-Whitney tests were applied to determine differences in the MPs ingested according to the period (day or night). A Spearman's correlation test was used to evaluate the relationship between MPs ingestion and biological parameters of fishes (standard length and total weight). All statistical analyses were performed with the software R version 3.6.3 (R Core Team, 2020) and were conducted considering a level of significance of 5%.

3. Results

A total of 213 microplastic (MPs) particles were recovered from the 170 analysed specimens (frequency of occurrence 67%). MPs were presented in 93% of *Argyropelecus sladeni*, 75% of *Diaphus brachycephalus*, 62% of *Hygophum taaningi*, and 45% of *Sternoptyx diaphana* specimens (Table 1). According to the number of MPs, ingestion significantly differed between species (chi-squared = 20.437, df = 3, $p < 0.05$), with *A. sladeni* being the most contaminated (1.66 ± 1.23 MPs ind.$^{-1}$), followed by *D. brachycephalus* (1.63 ± 1.41 MPs ind.$^{-1}$), *H. taaningi* (1.07 ± 1.20 MPs ind.$^{-1}$), and *S. diaphana* (0.54 ± 0.71 MPs ind.$^{-1}$) (Table 1). Dunn's *post hoc* test showed that *S. diaphana* differed from *A. sladeni* and *D. brachycephalus*. Additionally, there was no relationship between the MPs ingested by fish species and the biological parameters (standard length and the total weight) (Spearman's rank correlation, $p > 0.05$).

In general, the mean size of ingested MPs also varied according to the species (chi-squared = 12.247, df = 3, $p < 0.05$). *Argyropelecus sladeni* (0.74 ± 0.53 mm ind.$^{-1}$) showed the longest size of MPs ingested, followed by *H. taaningi* (0.49 ± 0.80 mm ind.$^{-1}$), *D. brachycephalus* (0.44 ± 0.53 mm ind.$^{-1}$), and *S. diaphana* (0.36 ± 0.82 mm ind.$^{-1}$), with significant differences observed between *A. sladeni* and *S. diaphana* (Table 1). Overall, fish MP contamination levels were not significantly different between day or night sampling, regardless of species (chi-squared = 1.4024, df = 1, $p > 0.05$), and by species individually ($p > 0.05$). However, ingestion differed among the sampling depths (chi-squared = 18.80, df = 6, $p < 0.05$). Fishes were generally most contaminated at 230 m (1.73 ± 1.25 MPs ind.$^{-1}$), followed by 430 m (1.66 ± 0.57 MPs ind.$^{-1}$), and 610 m (1.62 ± 1.44 MPs ind.$^{-1}$), and less contaminated at 800 m (0.57 ± 0.75 MPs ind.$^{-1}$) (Fig. 3). Statistically significant differences were observed between depths of 800 and 230 m and between depths of 800 and 610 m ($p < 0.05$). Regarding the shape of MPs ingested by fishes, most were fibres (64%), followed by fragments (19%), pellets (6%), films and foams (4%). However, the shape of ingested MPs did not vary between the species (chi-squared = 3.1683, df = 4, $p > 0.05$). Fibres were mainly observed in *S. diaphana* (83%), *A. sladeni* (76%), *H. taaningi* (63%), and *D. brachycephalus* (58%), followed by fragments in *D. brachycephalus* (23%), *H. taaningi* (21%), *A. sladeni* (12%) and *S. diaphana* (11%). Pellets were found in *H. taaningi*

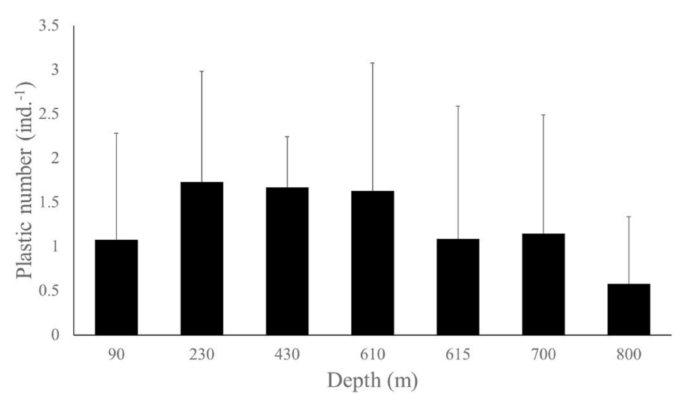

Fig. 3. Mean number (±standard deviation) of MPs ingested per depth strata.

(12%), *S. diaphana* and *D. brachycephalus* (5%), and films were found in *A. sladeni* (12%), *D. brachycephalus* (5%), *H. taaningi* (1%). Foams were only found in *D. brachycephalus* (7%) and *H. taaningi* (1%) (Fig. 4).

Overall, plastic polymers were identified in 80% of particles from the subset of samples. Natural particles identified as cellulose were observed in 15% of all particles, and 5% were unidentified. The most common polymers found were polyamide (PA) at 25% abundance, followed by polyethylene (PE) and polyethylene terephthalate (PET), with a similar abundance at 19%. The other polymers contributed to a similar percentage of 6–7% and included the ethylene-vinyl acetate (EVA), polyvinylchloride (PVC), styrene-butadiene rubber (SBR), polylactic acid (PLA), alkyd varnish and chlorinated polyisoprene (Fig. 5).

4. Discussion

This study confirmed that the mesopelagic fishes from the SWTA are contaminated with MPs. The four species analysed here exhibited a high MP detection frequency in their digestive tract (67%). These findings bring new information into the contamination of the deep sea and shed light on the potential role of marine organisms in MPs sinking.

Worldwide, few studies have documented plastic ingestion by mesopelagic fishes. For example, in the North Pacific Gyre, Davison and Asch (2011) reported an MP detection frequency of 9.2% of the fishes sampled, whereas Boerger et al. (2010) found 35% in the same area. In the Mediterranean Sea, Romeo et al. (2016) found MPs in 2.7% of sampled lanternfishes, whereas Zhu et al. (2019) reported the presence of MPs in more than 90% of the deep-sea fishes sampled in the South China Sea. In the Islands of Tristan da Cunha and St. Helena, McGoran et al. (2021) found 73.3% of species contaminated with MPs; and in the North Atlantic, Lusher et al. (2016) found 11% of individuals contaminated, in contrast with Wieczorek et al. (2018) which detected MPs in 73% of the mesopelagic fish specimens from the same area. The substantial divergence in the frequency of occurrence of MPs recovered in mesopelagic fishes may be due to several factors such as ecological behaviour, site-specific oceanographic differences, laboratory procedures, and sampling methods. However, differences in the extraction methods, an issue previously addressed by Wieczorek et al. (2018), might also influence the contamination rate. A lack of standardisation of the protocols for MPs extraction in organisms is the main issue for comparing studies on plastic contamination. The scientific community emphasises the importance of employing reliable and replicable research methods (Hermsen et al., 2018; Markic et al., 2020; Müller, 2021), not only concerning the choice of a suitable extraction method for MPs (*e.g.*, digestion and QA/QC protocols), but also an adequate sample size (>10; Justino et al., 2021) and size detection threshold of the particles, which is determinant in the number of plastics recovered (Savoca et al., 2021). Such decisions are important to avoid the bias of over/underestimation due to cross-contamination and loss of samples and were carefully considered in the present study.

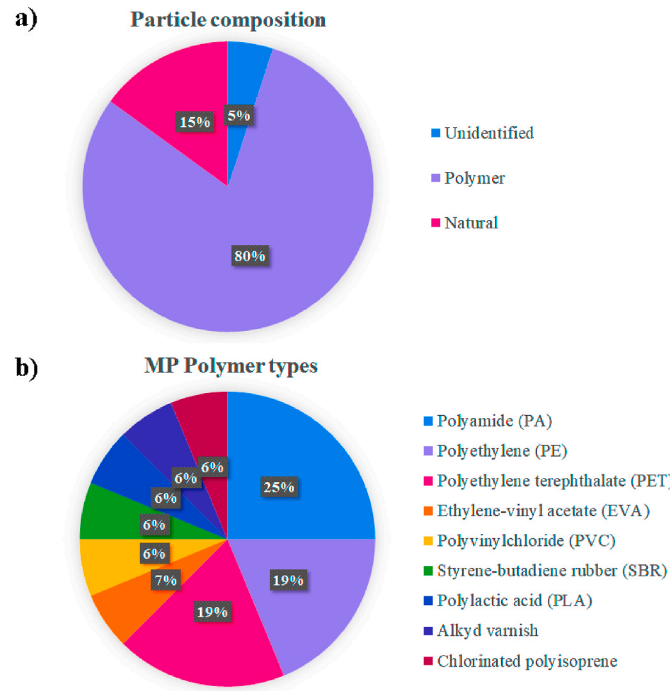

Fig. 5. Polymers identified using the LDIR analyser. a) Particle composition in the samples analysed, and b) Percentage of microplastic polymers found in the samples.

The wide availability of MPs is expected to threaten biodiversity throughout the marine environment. Plastic debris is found all along the coastal zone, continental slope, around oceanic islands, seamounts, and even in the deepest parts of the ocean (Cai et al., 2018; Monteiro et al., 2018; Lins-Silva et al., 2021; Pinheiro et al., 2021). Differences in the ecological habits, such as feeding strategy and migration, might influence the MP uptake by marine species. A clear distinction was observed in our study between the number of MPs ingested by species. For example, *A. sladeni* exhibited the highest number of particles (mean of

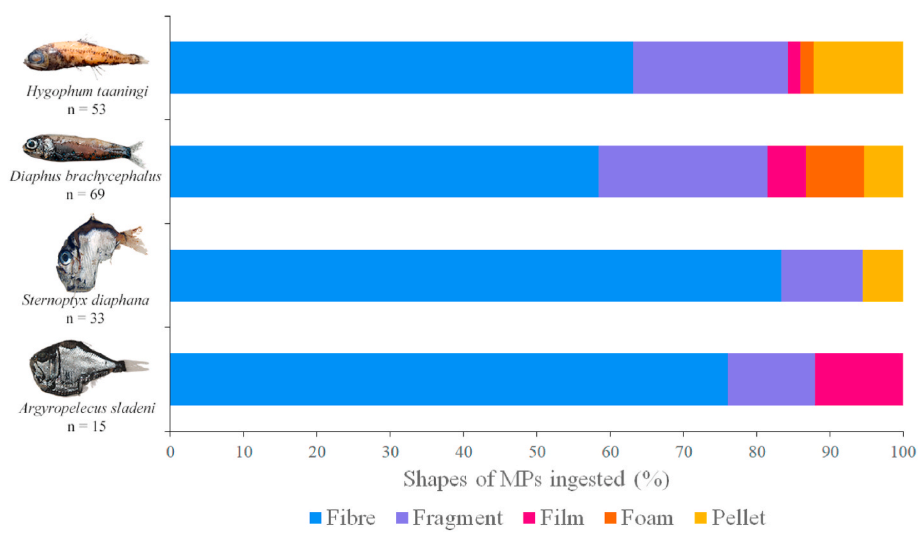

Fig. 4. Relative abundances (%) of MP shapes ingested per fish species.

1.66 ± 1.23 MPs ind.$^{-1}$; FO = 93%), while *S. diaphana* exhibited the lowest number (0.54 ± 0.71 MPs ind.$^{-1}$; FO = 45%). A distinct pattern from that recorded in previous studies on mesopelagic fishes, where two of the most up-to-date references did not observe any differences between species and depths (Lusher et al., 2016; Wieczorek et al., 2018).

The difference observed in MPs ingestion might be explained by the species vertical migration behaviour. For example, in our study area, *A. sladeni* is mostly distributed at 400–500 m during the daytime, mainly feeding on fish larvae and ostracods (Eduardo et al., 2020a). On the other hand, *S. diaphana* is found chiefly in deeper waters (700–900 m), primarily feeding on amphipods (Eduardo et al., 2020a). Likewise, in the daytime, *D. brachycephalus* is mainly distributed in the upper mesopelagic layer at 200–500 m, while *H. taaningi* was predominantly found in deeper waters (700–1000 m) (Eduardo et al., 2020a, 2021). However, the *H. taaningi* analysed in this study were only caught in the epipelagic zone, probably captured during migration towards superficial areas. Even though all species analysed in this study performed diel vertical migration (DVM), we did not observe any significant differences in the MP concentration in specimens sampled day or night. However, differences in MP number were observed depending on the depth strata.

Indeed, the most contaminated species (*A. sladeni* and *D. brachycephalus*) were mainly caught in the upper mesopelagic layer (230–430 m), and *S. diaphana*, which ingested a lower number of MPs particles, was captured in the lower mesopelagic layer (800 m). Therefore, we suggest that when migrating to the upper layers, these species interact with MPs and, when returning, they probably act as vectors of MPs to the deeper ocean layers (Fig. 6). For instance, in the study area, myctophids constitute 85% of the viperfish diet, the most abundant mesopelagic micronektivore fish species (Eduardo et al., 2020b). To our best knowledge, there is no information on MP in sediment and bottom organisms for the SWTA region, making the real impact of MP and their transportation into the deep sea speculative. However, coupling the data gathered in the present study with the widely acknowledged fact that mesopelagic species transport carbon to deep waters (Davison et al., 2013; Drazen and Sutton, 2017; Eduardo et al., 2020a), it seems that these species may also be transporting MPs to the deep sea.

Furthermore, our data support previous hypotheses that the deeper layers are less contaminated (Kvale et al., 2020; Zobkov et al., 2019). In Monterey Bay, California, Choy et al. (2019) also observed a similar pattern: a peak concentration of MPs in the mesopelagic zone at a range of 200–600 m depth. Additionally, the size of MPs ingested was also influenced by the depth in which species were caught (Ferreira et al., 2022). *Argyropelecus sladeni* ingested the longest MPs, whereas *S. diaphana* ingested significantly smaller MPs, coinciding with surveys investigating MP size in the water column (Dai et al., 2018; Zobkov et al., 2019). The ingestion of smaller size plastics was also observed in deep-water species in the North-East Atlantic (Pereira et al., 2020). The sinking of MPs is associated with biological activities such as biofouling, marine snow, faecal pellets, and plastic pump, contributing to the dispersion of smaller particles in the deeper layers (van Sebille et al., 2020). We corroborate previous findings by linking MP size to depth since we found the smallest particles in species inhabiting the deepest layers.

In our study, fibres were the common MP shape for all species (64%), and polyamide (PA), polyethylene (PE), and polyethylene terephthalate (PET) were the most common polymers identified, which are mainly used in the fishery and the textile industry (Lima et al., 2021). Previous research has already found lower density polymers as polyethylene in mesopelagic fishes (Wieczorek et al., 2018); these buoyant microplastics can be ingested by fish when they migrate towards epipelagic areas, thereby transporting these particles to deeper areas. Sources of fibres are related to the release of untreated water from the washing machine into

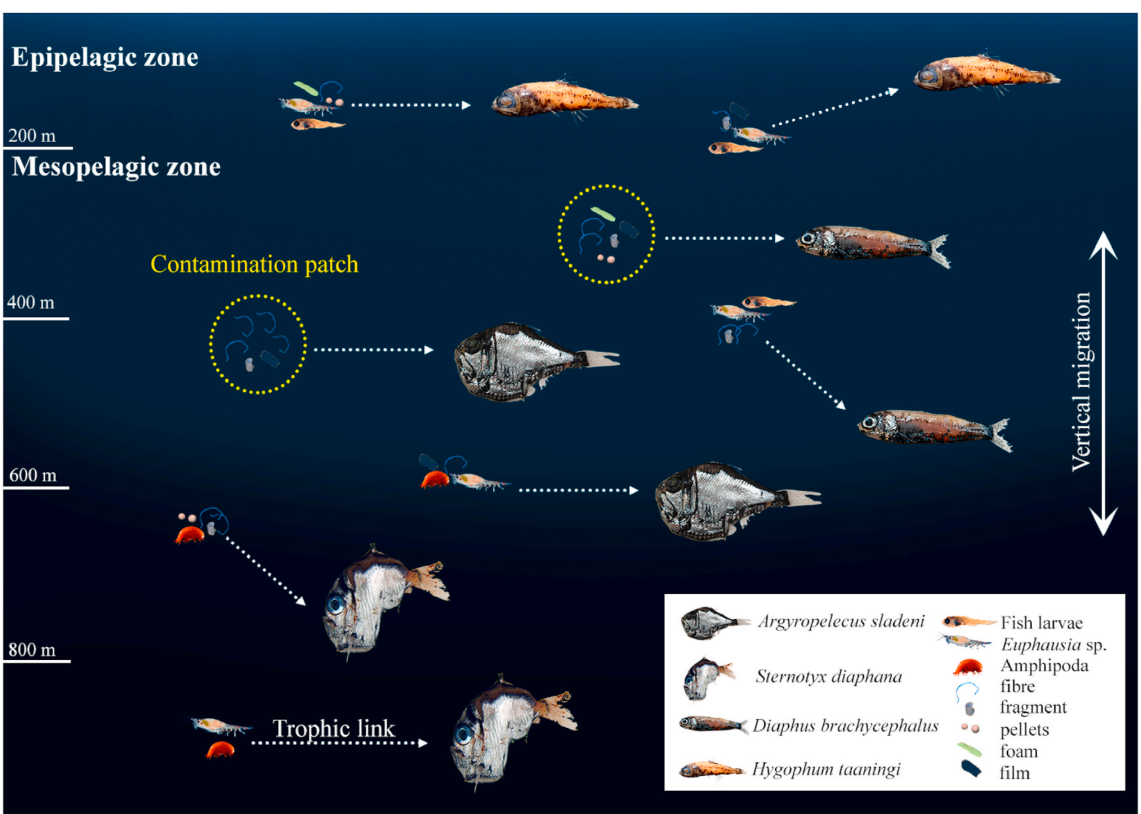

Fig. 6. Schematic representation of the microplastic ingestion by mesopelagic fishes in the Southwestern Tropical Atlantic. White dotted arrows indicate the ingestion by trophic link, and yellow dotted circles the probable microplastic accumulation zone. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

aquatic environments (De Falco et al., 2019) and extensive fishery activities (Chen et al., 2018; Xue et al., 2020). Despite FNA including MPAs, this archipelago has a high influx of tourists and extensive subsistence and recreational fishing activities (Lopes et al., 2017). Nets and fishing lines are known to degrade and fragment in the environment by physical factors, such as solar radiation (Andrady, 2011). Indeed, microfibres are the most common type observed in marine ecosystems (Kanhai et al., 2018; Lima et al., 2021) and recorded in the FNA and nearby islands (Ivar do Sul et al., 2014; Lima et al., 2016). Additionally, the Equatorial Atlantic is not perceived as an accumulation zone of fibres in surface water masses, decreasing the sinking of this type of MPs to deeper layers where fishes were captured (Lima et al., 2021). However, in the short-term, these islands might retain MPs in the nearshore due to the actions of winds, waves, vortices, and eddies surrounding the islands (Lima et al., 2016; Gove et al., 2019). The most contaminated species were captured around the FNA, suggesting that proximity to the MPs sources also influences ingestion rates.

Fibres are reported as the most ingested shape by mesopelagic fishes (Wieczorek et al., 2018; McGoran et al., 2021) and were also found in deep-sea amphipods in the Mariana trench (Jamieson et al., 2019); these tiny zooplankton act as energy sources in the oceanic trophic web. All fish species analysed here are zooplanktivorous, and amphipods are one of their main prey (Eduardo et al., 2020a, 2021). In the Mediterranean Sea, Romeo et al. (2016) observed similarities in the size of MPs and the size of the copepods, prey of lanternfishes, suggesting active and selective ingestion of MPs. We observed a similar pattern, as the dimensions of the MPs found in the SWTA were similar to those of common prey of the species (<2 mm), *e.g.*, amphipods and fish larvae in this region (Figueiredo et al., 2020). Through experiments, Li et al. (2021) demonstrated that fish could capture MPs passively by breathing but that some of them are also ingested inadvertently due to the similarity between their prey or the tiny sizes, which are hard to distinguish. Thus, MPs in mesopelagic fishes analysed here might be accidently consumed when confused as prey or by trophic transfer through ingestion of contaminated prey. However, due to methodological limitations in our study, we cannot state that these species interacted with MP by ingestion through food or swallowed by accident.

Regardless of the uptake routes (ingestion or breathing) of MPs in the mesopelagic fishes, the contamination rates (MP extracted from the digestive tract) observed in this study can be used as an indicator for the levels of MP available in the environment. The less contaminated species, *S. diaphana* captured in the deepest region, is evidence of the lower availability of MP particles in these areas. Additionally, this fact is corroborated by the smaller dimensions of MP extracted from *S. diaphana*, as expected for greater depths.

MPs' wide availability in the deep ocean layers may be harmful to the marine community, which is poorly investigated, but already interacts with these anthropogenic particles. In addition to organic additives (phthalates, OPEs, bisphenols) contained in plastics (Paluselli et al., 2019; Fauvelle et al., 2021), the surface of MPs can adsorb organic pollutants such as polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs; Rochman et al., 2013a), the latter process being enhanced by a longer transit time of MPs in meso- and bathypelagic waters (Rochman et al., 2013b; Jamieson et al., 2017). All of these compounds may very likely migrate into their surrounding environment, such as the digestive tract of biological species. Besides, MPs ingestion can cause adverse effects in fishes, such as physical injuries and blockage of the digestive tract, or even developmental, reproductive and locomotor toxicity (Teuten et al., 2009; Bhagat et al., 2020). Additionally, smaller MPs can bioaccumulate in tissues (Lee et al., 2019; Sökmen et al., 2020).

5. Conclusions

This study was the first to assess microplastic (MP) contamination in mesopelagic fishes in the Southwestern Tropical Atlantic (SWTA). The four species analysed here were contaminated with MPs in their digestive tract. The primary polymer types identified were polyamide (PA), polyethylene (PE), and polyethylene terephthalate (PET). Ingestion rates of MPs varied between species and depth. However, no difference between day or night sampling was observed. Thus, even though all species interact at some level with MPs, individuals caught at the lower mesopelagic zone seem to be less exposed to MPs than those captured in the upper mesopelagic layer.

Mesopelagic fishes may act as a vector of MP to the deep sea as they perform vertical migrations, presenting an important link between epipelagic and lower mesopelagic layers (Lusher et al., 2016; Savoca et al., 2021). They also play an essential role in the energy transfer in the ecosystem, transferring the energy of primary and secondary consumers to the top oceanic predators, which are valuable for the fishery stocks. So, the presence of MPs in the SWTA mesopelagic ecosystem will likely pose several risks to marine ecosystems if high contamination is confirmed in the near future.

Further research on MP contamination is needed, especially concerning the deep-sea community, whose crucial role in the marine ecosystem functioning has been proven. Additionally, including the effects of oceanographic parameters (*e.g.,* oceanic currents, microturbulence, salinity) and ecological interactions (*e.g.*, prey-predator interaction) into the evaluation of MPs uptake is also needed since there are many factors involved in the transport, sinking, and uptake of MPs in the deep ocean. Finally, the pressure of anthropogenic impacts is rapidly increasing in the SWTA, so there is an urgent need to comprehend how contaminations occur and affect the ecosystem to establish mitigation measures.

CRediT authorship

Anne K. S. Justino: Conceptualization, Methodology, Validation, Investigation, Formal analysis, Writing - original draft. **Guilherme V. B. Ferreira:** Methodology, Validation, Investigation, Writing - review & editing. **Natascha Schmidt:** Resources, Writing - review & editing. **Leandro N. Eduardo:** Investigation, Writing - review & editing. **Vincent Fauvelle:** Resources, Writing - review & editing. **Véronique Lenoble:** Supervision, Methodology, Writing - review & editing. **Richard Sempéré:** Resources, Writing - review & editing. **Christos Panagiotopoulos:** Resources, Writing - review & editing. **Michael M. Mincarone:** Resources, Writing - review & editing. **Thierry Frédou:** Supervision, Writing - review & editing, Funding acquisition. **Flávia Lucena-Frédou:** Project administration, Supervision, Resources, Writing - original draft, 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.

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