

Uncovering microplastics contamination in canned seafood

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ABSTRACT

There is limited research on the occurrence of microplastics (MPs) in canned seafood. All types of canned seafood investigated in the present study were contaminated. After sample digestion in 30 % hydrogen peroxide, a total of 40 MPs were recovered. Fibers were the most common type, blue was the dominant colour, and Fourier Transform Infrared Spectroscopy (FTIR) identified polyester as the most common polymer. Considering all samples, an average of 3.5 ± 5.2 MPs/can was obtained, with octopus in tomato sauce and tuna in olive oil presenting the highest contamination (5.2 ± 7.5 MPs/can and 5.2 ± 5.1 MPs/can, respectively). Also, significant differences between the number of MPs in the seafood tissues and immersion liquids were verified. The present study demonstrates MPs occurrence in canned seafood, a potential contamination pathway for humans. More research on the different stages of the canning processing is vital for understanding MPs contamination in cans.

1. Introduction

In recent decades, the global production of plastics has experienced an exponentially rise, now reaching nearly 400 million tons per year. This growing tendency triggered a global concern regarding the occurrence and consequences of plastics in marine environments and services. According to the United Nations Environment Programme (UNEP), 75–199 million tons of plastic are currently found in aquatic system. Additionally, approximately 19–23 million tons of plastic are introduced into these environments each year, with a tendency to increase (UNEP, 2021).

Microplastics (MPs) are small plastic items commonly defined as less than 5 mm in size. There are two types of MPs: i) primary MPs, particles originally manufactured in small sizes, mainly for industrial purposes, and ii) secondary MPs, particles originated from the fragmentation of larger plastics (GESAMP, 2016). Several studies have already demonstrated the widespread presence of MPs in the environment. These investigations documented the presence of MPs in multiple aquatic environments, in organisms that inhabit those ecosystems, and in originated natural and processed foods (Andrady, 2011; Botterell et al., 2019; Bao et al., 2023). In addition, the growing evidence of a

relationship between MPs and surrounding toxic pollutants, such as metals and pesticides, are raising concerns regarding the potential negative impacts of MPs on human health. Preliminary research demonstrated some possible threats, such as enhanced inflammatory response, release of endocrine disruptors, or interferences with the gut microbiome. However, it is crucial to understand that the potential toxicity linked with MPs is dependent on some parameters such as particles size, associated chemicals, and dosage (Smith et al., 2018). Despite the increasing number of studies regarding the presence of MPs in several types of food sources, there is still a lack of knowledge concerning potential risks to human health. Considering the already documented physical and chemical toxic effects in animals (e.g. oxidative stress, genotoxicity, reproductive disorders, mortality) (Qiao et al., 2019), and the potential risks for humans, further research to assess the impacts and toxicity of MPs on humans, as well as the establishment of a clear range of MPs concentration capable of causing adverse effects on consumers is crucial for food security issues (Diaz-Basantes et al., 2022).

Canned seafood corresponds to approximately 10 % of the total 178 million tons of world fishery production. Canned seafood is an important food source, rich in proteins and other essential nutrients such as omega-3 fatty acids, and its extensive consumption is observed in several

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countries across the world (FAO, 2021). There are several types of sea organisms canned (e.g. fish, molluscs, or crustaceans) often combined with additional ingredients and immersion liquids, such as oils (e.g., sunflower, or olive), water, or sauces (e.g., tomato, or escabeche), each one with specific nutritional facts. Canned seafood is one of the most popular methods of fish preservation. It is produced from fresh or frozen processed organisms sealed in hermetic containers, with one of the major advantages being the long shelf life, from 1 to 5 years (Warne, 1988; FAO, 2021). Since canned seafood is typically considered a ready-to-eat product, and most consumers eat these products without additional cleaning, for food security and human health purposes it is crucial to have a better understanding on MPs occurrence in these products.

Although the presence of MPs in seafood organisms has been extensively studied, there is a knowledge gap concerning the contamination of canned seafood. In the literature, only a limited number of studies have addressed the presence of MPs in this important category of seafood products, to our knowledge: Karami et al. (2018), Hussien et al. (2021), Akhbarizadeh et al. (2020), and Diaz-Basantes et al. (2022). Plus, the existing literature focuses on a reduced range of species, lacking comprehensive coverage of crucial organisms for the canning industry, such as octopus, horse mackerel, and mussels – all of which analysed in the present investigation.

Therefore, the main objective of the present study is to investigate the occurrence of MPs in several products of canned seafood, testing the hypothesis that MPs contamination varies between different types of canned seafood, and also between the solid and liquid components of the cans. The selected samples include different seafood types: sardine (in tomato sauce and sunflower oil), octopus (in tomato sauce), mussels (in escabeche sauce), tuna (in olive oil), and chub mackerel (in sunflower oil). Some of these types have not yet been tested, such as molluscs. This investigation will provide more comprehensive understanding of MPs occurrence in this important seafood source, contributing valuable insights to human health concerns and food safety regulations. Moreover, this research aims to overcome a data limitation by distinguishing, and comparing, the occurrence of MPs in the solid (seafood tissues) and liquid (immersion liquids) components of canned products, since consumers can consume the two components or discard the immersion liquids.

2. Materials and methods

2.1. Canned seafood sampling

In total, sixty seafood cans from 4 popular brands were purchased from two supermarkets in the city of Porto, Portugal. Brands A, B, C are manufactured in Portugal, while brand D is manufactured in Spain. Six different types of canned products, selected accordingly to the most popular and consumed items, were investigated for MPs contamination: i) sardine in tomato sauce; ii) sardine in sunflower oil; iii) octopus in tomato sauce; iv) mussels in escabeche sauce; v) tuna in olive oil; and vi) chub mackerel in sunflower oil. For each seafood type, 10 cans from the same brand were studied (Table 1).

Table 1

Characteristics of the sampled canned seafood: seafood species, brand, number of cans (N), net weight (g), liquid weight (mL), type of immersion liquid and ingredients.

Species	Brand	N	Net W (g)	Liquid W (mL)	Immersion liquid	Ingredients
Sardine (<i>Sardina pilchardus</i>)	A	10	120	78	Tomato sauce	Water, tomato, sunflower oil, salt, sugar
Sardine (<i>Sardina pilchardus</i>)	A	10	120	36	Sunflower oil	Sunflower oil, salt
Octopus (<i>Octopus</i> spp.)	B	10	120	48	Tomato sauce	Soy oil, tomato, onion, salt, garlic, spices, pepper
Mussel (<i>Mytilus galloprovincialis</i>)	B	10	111	42	Escabeche sauce	Sunflower oil, wine vinegar, salt, spices
Tuna (<i>Katsuwonus pelamis</i>)	C	10	120	42	Olive oil	Olive oil, salt
Chub Mackerel (<i>Scomber colias</i>)	D	10	115	34	Sunflower oil	Sunflower oil, salt

2.2. Microplastics analysis

2.2.1. Microplastics quantification

The quantification of MPs from canned seafood samples was performed according to the methodology previously optimized and fully described in Silva et al. (2022). Thus, a subsample of 10 g of seafood tissue from each can was randomly weighted in an analytical balance (Kern ALJ 200-5DA, Germany), placed in a cleaned Schott flask, and left drying overnight at 40 °C in a laboratory oven (Heraeus D-6450, Germany). After, 30 % hydrogen peroxide (H₂O₂) (Fisher Chemical, UK) solution was added to each flask in a volume sufficient to cover the entire sample. Flasks were capped with aluminium foil and placed in the laboratory oven at 65 °C for 24–48 h, until the complete digestion of the organic component. For each can, the same protocol with exception of the overnight step was applied for the total amount of the immersion liquid (analysed volumes described in Table 1). After sample digestion, the product was directly vacuum filtered (pump KNF Neuberger D-79112, N035 AN.18, Germany) through 0.45 µm cellulose nitrate membrane filters (Sartorius, Germany), or subjected to a fat reduction step prior to filtration (see detailed description in Silva et al., 2022). During filtration, flasks were rinsed with deionized water to remove any attached residues. When necessary, more than one filter was used for the same sample due to filter clogging. Filters were placed in cleaned glass Petri dishes (VWR International, USA) and air-dried at room temperature for at least two days, prior to observation.

2.2.2. Microplastics characterization and identification

Filters were visualized under a stereomicroscope (Leica EZ04, Germany), and MPs were photographed and classified according to the type (e.g., fiber, fragment, film), colour and size. Also, MPs were collected for further polymer identification using Fourier Transform Infrared Spectroscopy (FTIR) analysis, in a PerkinElmer Spectrum Two™ (USA), Attenuated Total Reflectance (ATR) spectrometer. ATR-FTIR spectroscopy operates by measuring the absorption of infrared light by a sample placed in contact with a crystal. This technique produces stable spectra from irregular MPs surfaces and can be used to examine a wide variety of solids, detecting particles down to the diameter of the infrared beam aperture, more precisely 10 µm (Shim et al., 2016). Following the recommendation under the European Union's Marine Strategy Framework Directive (MSFD), at least 10 % of the total number of particles (Galgani et al., 2023) retrieved from the samples were analysed through ATR-FTIR. The ATR-FTIR spectra of samples were determined after four scans at a wavelength range of 4000–400 cm⁻¹. The obtained spectra were compared with reference libraries, and results with confidence levels >75 % were accepted. Although rayon is a semi-synthetic nature-based polymer, in this study it was considered as an MP and used for the assessment of MPs contamination of canned seafood samples, following similar studies (Pereira et al., 2023).

2.2.3. Procedural control

To prevent MPs contamination, sampling processing was performed inside a laminar flow cabinet, with the working surface pre-cleaned with 70 % ethanol. All glassware and dissection materials were rinsed with 70 % ethanol and filtered deionized water prior to use. All H₂O₂ solutions used for the digestions were previously filtered through 0.45 µm

cellulose membrane filters. Contamination blanks consisting of open glass petri dishes with deionized water, were used during all processing stages to examine for airborne contamination. Gray 100 % cotton laboratory coats were worn during laboratory processing to control potential cross-contamination from investigators' clothing.

2.3. Data analysis

MPs contamination was expressed by the average number of MPs per seafood can (extrapolated for the entire quantity in the can), and by the average number of MPs per weight (g) of seafood tissue, and volume (mL) of immersion liquid. Nested 2-way ANOVA, with seafood type and sample component as fixed factors, was used to investigate if the number of MPs per canned seafood type (sardine in tomato sauce, sardine in sunflower oil, octopus in tomato sauce, mussels in escabeche sauce, tuna in olive oil, and chub mackerel in sunflower oil) and/or per sample component (immersion liquids and seafood tissues) varied significantly. Data normality and homogeneity of variances were verified with Shapiro-Wilk and Levene's test, respectively. Pearson correlation coefficient was performed to determine a possible relationship between the number of ingredients and the number of MPs in the cans. Differences were considered significant at $p < 0.05$. TIBCO Statistica™ (version 13) analytics software was used for data analysis.

3. Results

3.1. Microplastics in canned seafood

The presence of MPs was non-detected in the procedural blanks used to evaluate possible contaminations during analytical procedures.

A total of 52 suspected particles were recovered from the six types of canned seafood. From these, 12 particles were excluded as they were identified as organic bone structures. From the remaining 40, with sizes ranging from 50 μm to 3000 μm , all particles were analysed with ATR-FTIR, and 23 (58 %) were successfully confirmed as plastic polymer. The MPs not considered in the present study results ($n = 17$) either obtained a spectrum with less than 75 % of confidence level, or were not

suitable to be properly placed on the crystal surface of the ATR-FTIR. From the 60 cans examined in the present study, 45 % ($n = 27$) presented contamination by MPs, and from the MPs recovered from the cans, 65 % were collected from the subsamples of seafood tissues, while the remaining 35 % came from the immersion liquids (Fig. 1).

For mussel in escabeche sauce and chub mackerel in sunflower oil, MPs were detected only in the tissues, while for sardine in sunflower oil MPs presence was observed only in the immersion liquids (Fig. 2). For all the samples, an average of 0.043 ± 0.070 MPs/g of seafood tissue, 0.006 ± 0.015 MPs/mL of immersion liquid, and 3.5 ± 5.2 MPs/can was obtained (Fig. 2). The highest average of MPs/can was registered for octopus in tomato sauce (5.2 ± 7.5 MPs/can) and tuna in olive oil (5.2 ± 5.1 MPs/can). Regarding the two different components from the cans, the highest average of MPs in the seafood tissues occurred in octopus in tomato sauce (0.070 ± 0.106 MPs/g), while the highest average of MPs in the immersion liquids were registered for sardine in tomato sauce (0.012 ± 0.020 MPs/mL), and tuna in olive oil (0.012 ± 0.026 MPs/mL) (Fig. 2).

No variations between the number of MPs from the six different types of canned seafood ($F = 1.256$; $p = 0.288$) were observed. Nevertheless, the total content of MPs in the seafood tissues was significantly higher than in the immersion liquids ($F = 3.862$; $p < 0.05$). Regarding the relationship between the number of ingredients in the cans and respective MPs content, no significant correlation was obtained ($R = 0.1554$; $p \geq 0.236$).

3.2. Characteristics of the microplastics

The types of MPs recovered from the samples were fibers (75 %), fragments (18 %), and films (8 %). As described in Galgani et al., 2023, fragments are irregularly shaped hard particles with broken-off edges, films are particles derived from sheets or thin films, and fibers are slender thread-like particles. Three photographic examples of the different types of MPs recovered from the canned seafood samples are presented in Fig. 3.

In Fig. 4, the characteristics of the MPs recovered from each seafood type studied are presented.

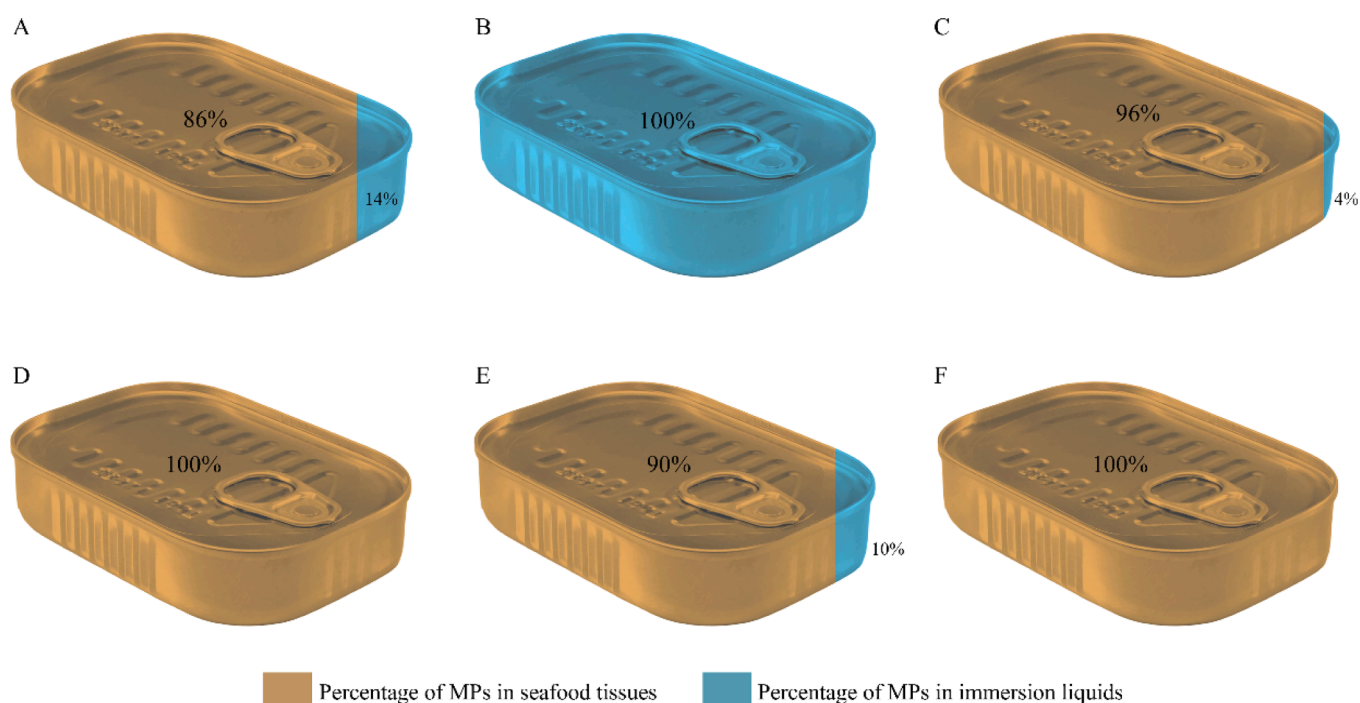


Fig. 1. Percentage of MPs occurrence in the seafood tissues and immersion liquids from each seafood type (A – sardine in tomato sauce; B – sardine in sunflower oil; C – octopus in tomato sauce; D – mussel in escabeche sauce; E – tuna in olive oil; F – chub mackerel in sunflower oil).

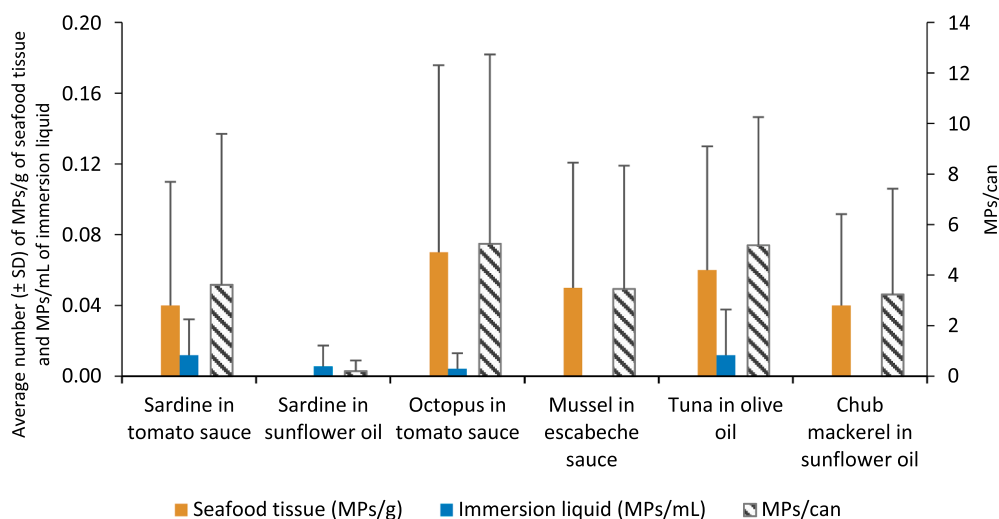


Fig. 2. Average number ($\pm$ SD) of MPs/g of seafood tissue, MPs/mL of immersion liquid, and MPs/can for each seafood type studied.

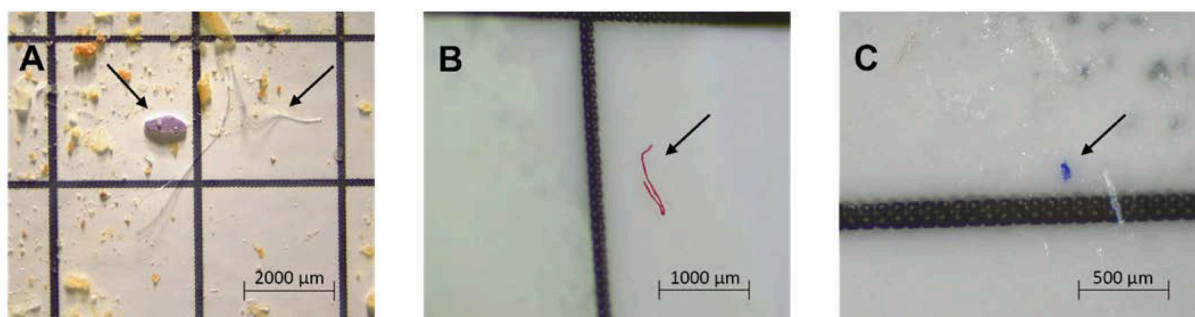


Fig. 3. Examples of MPs (black arrows) recovered from different canned seafood samples. A – Purple fragment and transparent fiber recovered from the tissue of a mussels in escabeche sauce sample; B – Red fiber recovered from the tissue of a chub mackerel in sunflower oil sample; C – Blue film recovered from the immersion liquid of a sardine in sunflower oil sample. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

MPs from eight different colours were observed: blue was the most common (37.5 %) and present in all types of canned seafood, followed by transparent (30 %), black (10 %), red (7.5 %), yellow (5 %), white (2.5 %), and green (2.5 %). The MPs size ranged between 50 μ m and 3000 μ m, with an average of 789 ± 858 μ m. Most of the observed MPs presented a size <500 μ m (45 %), followed by 38 % in the size range of 500–1000 μ m, and 18 % with size >1000 μ m. The most common plastic polymers analysed and identified in ATR-FTIR were polyester (PES, 26 %), followed by polypropylene (PP, 17 %), polyethylene (PE, 17 %), rayon (17 %), nylon (13 %), and polyvinyl (9 %).

Fibers were identified across the six types of canned seafood studied, exhibiting five distinct colours, predominantly blue (40 %) and transparent (37 %). These fibers presented an average length of 882 ± 937 μ m. Most of the examined fibers originated from PES (40 %), followed by rayon (27 %), nylon (20 %), and PE (13 %). Fragments were present in four types of canned seafood. They presented four different colours, with black (29 %) and yellow (29 %) being the most prevalent. An average length of 643 ± 535 μ m was obtained for these MPs, and the recovered fragments were mostly derived from PP (67 %), followed by PE (16.5 %) and polyvinyl (16.5 %). Films were the less common MPs type recovered from the canned seafood, being present in only three types of samples. They were mostly blue (67 %), with an average length of 200 ± 173 μ m. These film particles were primarily composed of polyvinyl (50 %) and PE (50 %).

Sardine in tomato sauce presented MPs contamination in 40 % of the cans, with 3.6 ± 6.0 MPs/can in average. Sardine tissues presented an

average of 0.040 ± 0.070 MPs/g, while the tomato sauce exhibited an average of 0.012 ± 0.020 MPs/mL. Fibers were the predominant type of MPs (89 %) (Table 2). The other studied seafood type with sardine, immersed in sunflower oil, recorded the lowest occurrence of contaminated cans (20 %), along with the lowest average number of MPs/can (0.2 ± 0.4). Also, this seafood type was the only where no MPs were recovered from the solid tissues. The immersion oil presented an average of 0.005 ± 0.010 MPs/mL, and an equal number of fibers and films (50 %) were recovered (Table 2). Octopus in tomato sauce presented 60 % of contaminated cans, with an average of 5.2 ± 7.5 MPs/can. This was the seafood type with more MPs extracted from the solid tissues, with an average of 0.070 ± 0.106 MPs/g of octopus. Fibers, fragments and films were recorded (Table 2). Mussels in escabeche sauce presented 40 % of contaminated cans, with an average of 3.5 ± 4.9 MPs/can. No MPs were recovered from the immersion sauces, and fibers (80 %) were the predominant type of MPs extracted from the mussels' tissues (Table 2). Tuna in olive oil exhibited the highest incidence of contaminated cans with MPs (70 %). Together with the previously mentioned octopus in tomato sauce, this seafood type presented the highest average of MPs/can (5.2 ± 5.1), and contained fibers, fragments and films (Table 2). Chub mackerel in sunflower oil presented 40 % of contaminated cans, and no MPs were recovered from the immersion oil. Fibers were the only type of MPs present (Table 2).

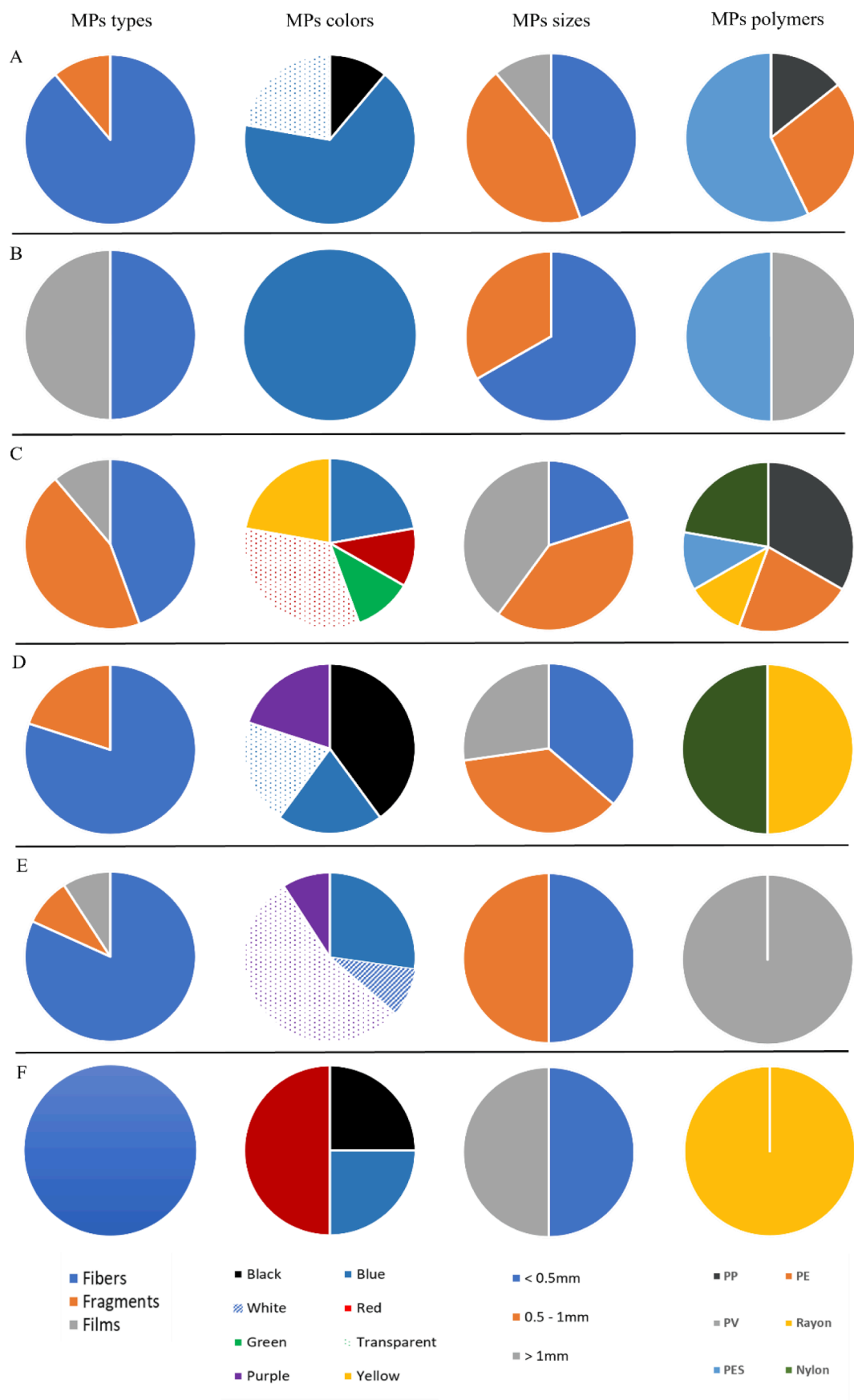


Fig. 4. Characteristics of the MPs (type, colour, size, and polymers) recovered from each seafood type (A – sardine in tomato sauce; B – sardine in sunflower oil; C – octopus in tomato sauce; D – mussel in escabeche sauce; E – tuna in olive oil; F – chub mackerel in sunflower oil).

Table 2

Microplastics in the seafood cans: percentage of contaminated cans, number of MPs in solid and liquid components, MPs/g of tissue, MPs/mL of liquid, percentage of fibers, fragments, and films, and extrapolated MPs/can.

Seafood type	Contaminated cans (%)	MPs tissues (n)	MPs/g tissue (average $\pm$ SD)	MPs liquids (n)	MPs/mL liquid (average $\pm$ SD)	Fibers (%)	Fragments (%)	Films (%)	MPs/can (average $\pm$ SD)
Sardine in tomato sauce	40 %	4	0.040 $\pm$ 0.070	5	0.012 $\pm$ 0.020	89 %	11 %	0 %	3.6 $\pm$ 6.0
Sardine in sunflower oil	20 %	0	0	2	0.005 $\pm$ 0.010	50 %	0 %	50 %	0.2 $\pm$ 0.4
Octopus in tomato sauce	60 %	7	0.070 $\pm$ 0.106	2	0.005 $\pm$ 0.010	44.4 %	44.4 %	11.1 %	5.2 $\pm$ 7.5
Mussels in escabeche	40 %	5	0.050 $\pm$ 0.071	0	0	80 %	20 %	0 %	3.5 $\pm$ 4.9
Tuna in olive oil	70 %	6	0.060 $\pm$ 0.070	5	0.012 $\pm$ 0.026	82 %	9 %	9 %	5.2 $\pm$ 5.1
Chub mackerel in sunflower oil	40 %	4	0.040 $\pm$ 0.052	0	0	100 %	0 %	0 %	3.2 $\pm$ 4.2
Total	45 %	26	0.043 $\pm$ 0.070	14	0.006 $\pm$ 0.015	75 %	18 %	8 %	3.5 $\pm$ 5.2

4. Discussion

In the present study, all types of canned seafood revealed contamination with MPs, although with no statistically significant differences identified between them ($F = 1.256$; $p = 0.288$). Nevertheless, an important finding resulted from our research, particularly concerning the distinction between the solid and liquid components of canned products: the number of MPs in the seafood tissues was significantly higher than in the immersion liquids ($F = 3.862$; $p < 0.05$).

The sample size selected for the investigation ($n = 60$ cans) aligns with similar studies investigating the presence of MPs in canned seafood, notably: Karami et al. (2018) examined 80 cans, Hussien et al. (2021) studied 21 cans, Akhbarizadeh et al. (2020) investigated 50 cans, and Diaz-Basantes et al. (2022) analysed 32 cans. Our objective was to explore a diverse range of seafood types, hence a sample size of 10 can per seafood type was selected. In parallel, the sample size was also selected based on specific characteristics of canned seafood samples, where the existence of complex matrices with high level of fats can pose challenges to the efficiency of MPs extraction, particularly during sample digestion and filtration, as detailed in the methodology optimization from Silva et al. (2022).

Having into account our findings (an average MPs contamination of 0.043 ± 0.070 MPs/g of canned seafood) and the annual per capita consumption of 2 kg of canned seafood in the USA (NOAA, 2022), the estimated potential human intake can reach a maximum of 226 MPs per year per capita. The estimated intake of MPs per capita will vary in other countries depending on their consumption of canned seafood, but this example helps understanding the quantity of MPs ingested by a consumer of canned seafood throughout a year. Such estimates can significantly contribute for the establishment of concentration limits for MPs, not only in seafood but also in other food sources. This is crucial considering the documented physical and chemical toxic effects of MPs observed in animals, including oxidative stress, genotoxicity, reproductive disorders, and mortality, (Qiao et al., 2019), and the potential risks for human health as a consumer.

Although there is limited information regarding MPs contamination in canned seafood, previous studies have documented the presence of MPs in fresh samples of the organisms studied in the present research. For example, MPs were previously reported for sardine stomachs (da Silva et al., 2022), *K. pelamis* tunas' gastrointestinal tracts (GITs) (Sathish et al., 2020), octopus GITs (Pedà et al., 2022), *M. galloprovincialis* mussels (Expósito et al., 2022), and chub mackerel stomachs, gills, GITs, and dorsal muscle (Barboza et al., 2020; da Silva et al., 2022). The contamination of seafood with MPs can result from two possible paths: i) direct and continuous contact between marine organisms and the surrounding contaminated environments and preys (e. g., water filtration through gills, interaction with marine litter, accidental ingestion of MPs, or ingestion of contaminated preys) (Scacco et al., 2022); and ii) human handling and industrial process (Karami

et al., 2018).

Concerning path i), and focusing on the organisms studied in the present study, sardines and chub mackerel have some similarities. They are pelagic organisms inhabiting coastal waters, where the problem of MPs presence is particularly severe due to both inland and marine pollution (Bao et al., 2023). Plus, both species are predominantly planktivorous (chub mackerel also feed on larger organisms such as small fish), consuming plankton over a wide size range available in the environment (Herrera et al., 2019). Several studies already shown that MPs are ingested by several zooplankton taxa, entering the food web of secondary consumers, and being transferred to the following trophic levels (Botterell et al., 2019). Furthermore, and due to the overlap size range of MPs and plankton, planktivorous species also potentially ingest, accidentally, MPs from the surrounding environment (Galloway et al., 2017). MPs recovered from our samples of canned chub mackerel are similar to the ones reported by Herrera et al. (2019) for fresh organisms ($n = 120$, sample digestion: 10 % KOH, polymer identification: FTIR): blue, black and red fibers as the most abundant MPs.

Mussels are filter feeders capable of filter large volumes of water through their gills, feeding on small, suspended particles (Cole et al., 2023). Due to an efficient filtering capability, mussels increase their susceptibility to contaminants present in surrounding environment, MPs included, bioaccumulating them in their bodies. For this reason, mussels are considered good bioindicators of environmental disturbances (Cole et al., 2023). Furthermore, mussels subjected to processing methods (like canned mussels) might suffer the translocation of MPs to the edible tissues, suggesting that these particles may be introduced via de-shelling or insufficient cleaning procedures rather than entirely sourced from the surrounding environment (Li et al., 2018). In the present study, fibers were the dominant type of MPs obtained from canned mussels, a finding consistent with most of the studies performed with fresh mussels (e.g. Li et al., 2018; $n = 162$, sample digestion: 30 % H_2O_2 , polymer identification: micro-FTIR), since there is no available data regarding canned mussels.

MPs occurrence in cephalopods is, in general, poorly studied. In 2020, Oliveira et al. reported MPs occurrence in the digestive tracts of cuttlefish, while in 2023, Wang et al. demonstrated the presence of MPs in the stomachs of pelagic squids. The present study assumes greater importance given the lack of research on the MPs occurrence in cephalopods, more precisely octopus, a relevant and widely exploited species. As suggested by Pedà et al. (2022), common octopus is an organism capable of using marine litter, mostly composed by plastic debris, as shelter, mainly on soft substrates where rocks are scarce. Furthermore, bottom traps used in the artisanal fishing of octopus often accumulate marine litter together with the organisms, enhancing interactions between the organisms and plastic debris (Pedà et al., 2022). Our results for canned octopus revealed fibers as the most abundant type of MPs, and transparent the most common colour, both results in accordance with Pedà et al., (2022; $n = 6$, sample digestion: 10 % KOH, polymer

identification: FTIR), where MPs were recovered from the GITs of common octopus captured in the western Mediterranean Sea.

In the case of tuna, these animals are large migratory and top predator organisms, indicators of ocean health since they are exposed to several anthropogenic pressures and MPs pollution among different oceanic regions (Pereira et al., 2020; Pereira et al., 2023). Tuna from canned samples belong to the species *K. pelamis* (skipjack tuna), a pelagic-oceanic top predator known for having a selective predatory activity, feeding on large fish and cephalopods. This feeding behaviour with preference for large dimension preys, together with the size of MPs found in these organisms, suggest that it is unlikely that the skipjack tuna misidentifies plastic items as food, but rather ingests them through contaminated preys (Pereira et al., 2020). Negative impacts on the feeding behaviours, growth, development, reproduction, and lifespan, as well as physical injuries, were already reported for several marine organisms exposed to MPs. Additionally, MPs may not affect species only at the organism level but, over time, could represent a harmful effect at the population structure (Barboza et al., 2020; Botterell et al., 2019). Fibers were the most common type of MPs recovered from our canned tuna samples, and a similar result was obtained for fresh skipjack tunas studied in Hadibarata et al., (2021; n = 10, sample digestion: 15 % KOH, polymer identification: Gas Chromatography-Mass Spectrometry).

Regarding path ii), the presence of MPs in canned seafood can result from contaminations occurred since the seafood capture, where organisms contacts with some synthetic inputs from fishing and storage equipment, until the preparing and packaging process in the canneries (Diaz-Basantes et al., 2022; Karami et al., 2018). The canning of seafood is a complex process divided into several steps until the final supply to human consumers. Raw organisms are primarily submitted to cleaning, prior to viscera removing. Inadequate cleaning practices and materials could be a source of MPs contamination (Akhbarizadeh et al., 2020), as well as the improper seafood evisceration can cause the relocation of MPs from the digestive tracts to the edible tissues. In some canning methods, after a precooking step, seafood is left air cooling, allowing possible airborne contamination with MPs (Akhbarizadeh et al., 2020). The process of filling cans with immersion liquids may be performed either by machines or hand, with hand filling being very common in several countries, leading to more contaminations with plastic particles, metal fragments and other debris (Warne, 1988). The addition of external ingredients to the cans (e.g., salt, water, spices and herbs) can be an extra source of MPs contamination in the final canned products, as these ingredients can also be contaminated with MPs (Akhbarizadeh et al., 2020).

Results from the present study demonstrate MPs contamination in different types of canned seafood, which was also observed in the few studies recently reported. Samples from tuna, salmon, sardines, sprats, and mackerel, canned in several immersion liquids, were shown to be contaminated with MPs (Karami et al., 2018; Hussien et al., 2021; Akhbarizadeh et al., 2020; Diaz-Basantes et al., 2022). In the present study, an average of 0.043 ± 0.070 MPs/g of seafood tissue was obtained for all the seafood types. Similar results were published in Akhbarizadeh et al. (2020) where, for example, tuna in olive oil samples presented an average of 0.09 ± 0.01 MPs/g, a similar result to our tuna in olive oil samples (0.060 ± 0.070 MPs/g). In contrast, Diaz-Basantes et al. (2022) registered higher amounts of MPs for the same organism in different immersion liquids, with 6.92 ± 1.20 MPs/g for tuna in water, and 4.42 ± 0.84 MPs/g for tuna in oil. The application of different analytical methodologies for MPs quantification can also contribute to the differences in the results among the limited number of studies concerning MPs in canned seafood, hindering comparisons between the available data. The uniformization of MPs quantification protocols would be important for a better assessment of MPs presence in food sources.

Canned seafood is typically ready to be eaten right out of the container, without any additional cleaning. Since most of the filling liquids are not discarded but, instead, consumed along with the seafood,

it is crucial to investigate and compare the potential occurrence of MPs in both the seafood tissues and the immersion liquids. In the present study, most of the MPs were recovered from the canned seafood tissues (average of 0.043 ± 0.070 MPs/g), presenting a MPs contamination significantly higher than the one obtained for the immersion liquids (average of 0.006 ± 0.015 MPs/mL). Despite the lower incidence of MPs in the liquid component of the cans, only mussel in escabeche and chub mackerel in sunflower oil registered total absence of MPs from the immersion liquids; furthermore, samples from sardine in sunflower oil only presented MPs in the analysed liquid. These results, together with the fact that consumers have the option of eating the entire content of a can or discard the liquid component (such as oil or sauce), may be important to enhance our understanding and regulation of human exposure to MPs through the consumption of canned seafood products. Diaz-Basantes et al. (2022) also detected MPs in both tissues and immersion liquids of tuna canned in water and oil, while Karami et al. (2018) did not observed MPs in the filling liquids of canned sardines and sprats. On the other hand, Akhbarizadeh et al. (2020) and Hussien et al. (2021) did not distinguished between the MPs contamination in the solid and liquid fraction of the canned seafood.

In the current study, only sardine in sunflower oil showed no MPs contamination in the tissues, while all other types of seafood analysed exhibited MPs in their solid content. Regarding MPs in the immersion liquids, no MPs were recovered from the mussels escabeche sauce or chub mackerel sunflower oil. Although no significant differences existed between the samples, the two types of seafood canned in tomato sauce exhibited a higher occurrence of MPs in both the seafood tissues (octopus in tomato sauce), and the immersion liquids (sardine in tomato sauce). This is a potentially important finding since these two types of seafood canned in tomato had a higher number of ingredients in their composition, and most consumers ingests the tomato sauce together with the seafood, not discarding it. Despite the existence of a slight positive relationship between the number of ingredients and MPs content in the cans, Pearson correlation coefficient did not exhibit significant differences. In upcoming investigations, it would be interesting to study cans with a broader range of number of ingredients for a better understanding of this potential correlation. For example, in Akhbarizadeh et al. (2020), results from canned tuna and mackerel demonstrated a significant positive correlation between MPs and the salt content of samples, considering salt as a potential source of MPs contamination. An important finding, considering that salt is a commonly used ingredient in human diets, and is present in the ingredients of all the seafood types sampled in the present study. The potential increase in the number of MPs with the addition of external ingredients is a very interesting an important issue that needs a more detailed investigation in upcoming studies.

Fibers were the most abundant type of MPs, corresponding to 75 % of the total items, more than 4 times higher than fragments, and 10 times higher than films. Previous works already reported fibers as the major type of isolated MPs from seafood tissues (e.g. Barboza et al., 2020), and canned seafood samples (Akhbarizadeh et al., 2020). Additionally, fibers are among the most abundant types of MPs occurring in the marine and aquatic environment (Barboza et al., 2020). Once in the environment, fibers can reach concentrations up to thousands of particles per cubic meter, being available for ingestion by a wide range of organisms, like fish and mussels (Cesa et al., 2017). These synthetic items are mostly manufactured from polyamide (PA), polyethylene terephthalate (PET, from the family of PES), or PP (Andrady, 2011), and their presence is commonly attributed to the release of synthetic fibers from clothing during washing, degradation of cigarette butts leading to the release of cellulose acetate fibers, and fragmentation of fishing equipment such as ropes, lines, or nets (Cole, 2016; Wright et al., 2015).

The most abundant identified polymers were PES, corresponding to 26 % of the total plastic items, followed by PP (17 %), PE (17 %), rayon (17 %), nylon (13 %), and polyvinyl (9 %). In general, PES is between the three most common polymers found in aquatic organisms and

processed seafood, together with PP, and polystyrene (PS) (Karami et al., 2018). In addition, previous studies with canned seafood are reported PES and PP as the most identified polymers (Karami et al., 2018; Akhbarizadeh et al., 2020). Worldwide, PES is a polymer frequently correlated with the textile industry, often used in fibers for clothing, but also for food packaging; regarding PP, and due to its low cost and flexibility, is one of the most used plastic polymers, from fishing nets to plastic packaging and textiles; PE is applied in a wide range of inexpensive applications, including supermarket bags or plastic bottles; rayon, a silk man-made fiber manufactured from naturally regenerated cellulose, is mainly used in the textile industry, surgical materials, or hygiene products; polyvinyl is widely used in plumbing pipes, shower curtains, or windows frames, and nylons are primarily used in clothing and fishing gear applications (Andrady, 2011).

Blue was the predominant colour of the MPs found in the present study (37.5 %), a colour frequently observed in other works with fish (Barboza et al., 2020; Scacco et al., 2022; Pereira et al., 2023). MPs colours can increase the possibilities of ingestion, especially in visual predators such as fish and larvae (Ríos et al., 2022). Also, the predominance of blue may be due to a higher number of MPs of this colour in seawater (Lusher et al., 2014). Being more abundant, blue MPs have higher probabilities of being ingested by organisms than MPs of other less common colours. For instance, some species appear to preferentially ingest blue MPs due to the resemblance, both in colour and size, with their copepod prey (Ory et al., 2017). This unintentional ingestion of MPs is a problem since the first stages of fish life, as demonstrated in Steer et al. (2017), reporting blue fibers as the main ingested MPs in a variety of fish larvae. Further, Renzi et al. (2019) revealed that the European anchovy's feeding habits, preferentially ingesting dark preys, could be responsible for the mostly black and blue MPs identified in their gastrointestinal tracts. In Ríos et al. (2022), a wild freshwater fish species showed ingestion preference for blue (and yellow) MPs fragments, in a laboratory trial conducted with six different colours. Therefore, most of detected MPs in the current study in seafood tissues can in fact be originating from direct and continuous contact between marine organisms and the surrounding contaminated environments and preys, but more research is still needed, namely regarding seafood handling and processing in canning industry, to identify all possible steps of MPs contamination in order to avoid them.

5. Conclusions

The obtained results provide conclusive evidence of MPs contamination in a wide range of commercially available canned seafood products. To the best of our knowledge, this represents the first investigation into the occurrence of MPs in samples of canned octopus, chub mackerel, and mussels, significantly contributing to a better understanding on MPs contamination in such relevant seafood organisms. For all the types of canned seafood analysed, 45 % of the cans contained MPs, with an average of 3.5 ± 5.2 MPs/can. Among the several types of MPs identified, blue PES fibers were the most prevalent type. A better understanding on the possible MPs inputs from the capture of seafood organisms, through all industrial canning stages (including the incorporation of external ingredients potentially contaminated with MPs) until the final product available to consumers, is crucial. This knowledge is essential for insights into the presence of MPs in canned seafood, and for the implementation of effective prevention measures. The current findings, indicating an estimated maximum intake of 226 MPs per capita per year (based on available data from USA consumers), provides valuable insights into the quantity of MPs that an individual potentially ingests through canned seafood ingestion. Until now, no food products are regulated concerning the presence and concentration of MPs, and further studies on the possible risks for human health due to MPs ingestion are needed.

6. Environmental implication

Studying microplastics in canned seafood is vital for understanding their environmental impact. Microplastics are plastic particles already manufactured in small sizes (primary) or originated from the fragmentation and degradation of larger plastics (secondary), that contaminate oceans and inhabiting organisms, present and relevant for the human food chain. Investigating their presence in canned seafood helps assess human exposure and health risks. The present study raises awareness about plastic pollution, urging plastic waste reduction and management for sustainable production and consumption practices, benefiting both the environment and public health.

CRedit authorship contribution statement

Diogo M. Silva: Writing – original draft, Methodology, Investigation, Data curation, Conceptualization. **C. Marisa R. Almeida:** Writing – review & editing, Supervision, Conceptualization. **Francisco A. Guar-diola:** Writing – review & editing, Supervision, Conceptualization. **Rúben Pereira:** Writing – review & editing, Methodology. **Sabrina M. Rodrigues:** Methodology, Writing – review & editing. **Sandra Ramos:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization.

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.

Data availability

Data will be made available on request.

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References

- Akhbarizadeh, R., Dobaradaran, S., Nabipour, I., Tajbakhsh, S., Darabi, A. H., & Spitz, J. (2020). Abundance, composition, and potential intake of microplastics in canned fish. *Marine Pollution Bulletin*, 160, Article 111633. <https://doi.org/10.1016/j.marpolbul.2020.111633>
- Andrady, A. L. (2011). Microplastics in the marine environment. *Marine Pollution Bulletin*, 62, 1596–1605. <https://doi.org/10.1016/j.marpolbul.2011.05.030>
- Bao, M., Xiang, X., Huang, J., Kong, L., Wu, J., & Cheng, S. (2023). Microplastics in the atmosphere and water bodies of coastal agglomerations: A mini-review. *International Journal of Environmental Research and Public Health*, 20(3), 2466. <https://doi.org/10.3390/ijerph20032466>
- Barboza, L. G. A., Lopes, C., Oliveira, P., Bessa, F., Otero, V., Henriques, B., Raimundo, J., Caetano, M., Vale, C., & Guilhermino, L. (2020). Microplastics in wild fish from North East Atlantic Ocean and its potential for causing neurotoxic effects, lipid oxidative damage, and human health risks associated with ingestion exposure. *Marine Pollution Bulletin*, 717, Article 134625. <https://doi.org/10.1016/j.scitotenv.2019.134625>

- Botterell, Z. L. R., Beaumont, N., Dorrington, T., Steinke, M., Thompson, R. C., & Lindeque, P. K. (2019). Bioavailability and effects of microplastics on marine zooplankton: A review. *Environmental Pollution*, 245, 98–110. <https://doi.org/10.1016/j.envpol.2018.10.065>
- Cesa, F. S., Turra, A., & Baruque-Ramos, J. (2017). Synthetic fibers as microplastics in the marine environment: A review from textile perspective with a focus on domestic washings. *Science of The Total Environment*, 598, 1116–1129. <https://doi.org/10.1016/j.scitotenv.2017.04.172>
- Cole, M. (2016). A novel method for preparing microplastic fibers. *Scientific Reports*, 6, 34519. <https://doi.org/10.1038/srep34519>
- Cole, M., Artioli, Y., Coppock, R., Galli, G., Saad, R., Torres, R., Vance, T., Yunnice, A., & Lindeque, P. K. (2023). Mussel power: Scoping a nature-based solution to microplastic debris. *Journal of Hazardous Materials*, 453, Article 131392. <https://doi.org/10.1016/j.jhazmat.2023.131392>
- Da Silva, J. M., Alves, L. M. F., Laranjeiro, M. I., Bessa, F., Silva, A. V., Norte, A. C., Lemos, M. F. L., Ramos, J. A., Novais, S. C., & Ceia, F. R. (2022). Accumulation of chemical elements and occurrence of microplastics in small pelagic fish from a neritic environment. *Environmental Pollution*, 292(Part B), 118451. Doi: [10.1016/j.envpol.2021.118451](https://doi.org/10.1016/j.envpol.2021.118451)
- Diaz-Basantes, M. F., Nacimba-Aguirre, D., Conesa, J. A., & Fullana, A. (2022). Presence of microplastics in commercial canned tuna. *Food Chemistry*, 385, Article 132721. <https://doi.org/10.1016/j.foodchem.2022.132721>
- Exposito, N., Rovira, J., Sierra, J., Gimenez, G., Domingo, J. L., & Schuhmacher, M. (2022). Levels of microplastics and their characteristics in molluscs from North-West Mediterranean Sea: Human intake. *Marine Pollution Bulletin*, 181, Article 113843. <https://doi.org/10.1016/j.marpolbul.2022.113843>
- FAO (2021). FAO Yearbook. Fishery and Aquaculture Statistics 2019/FAO annuaire. Statistiques des pêches et de l'aquaculture 2019/FAO anuario. Estadísticas de pesca y acuicultura 2019. Rome/Roma. Doi: [10.4060/cb78741](https://doi.org/10.4060/cb78741)
- Galgani, F., Ruiz Orejon Sanchez Pastor, L., Ronchi, F., Tallec, K., Fischer, E., Matiddi, M., Anastasopoulou, A., Andresmaa, E., Angiolillo, M., Bakker Paiva, M., Booth, A. M., Buhalko, N., Cadiou, B., Claro, F., Consoli, P., Darmon, G., Deudero, S., Fleet, D., Fortibuoni, T., Fossi, M.C., Gago, J., Gerigny, O., Giorgetti, A., Gonzalez Fernandez, D., Guse, N., Haseler, M., Ioakeimidis, C., Kammann, U., Kühn, S., Lacroix, C., Lips, I., Loza, A.L., Molina Jack, M.E., Noren, K., Papadoyannakis, M., Pragnell-Raasch, H., Rindorf, A., Ruiz, M., Setälä, O., Schulz, M., Schultze, M., Silvestri, C., Soederberg, L., Stoica, E., Storr-Paulsen, M., Strand, J., Valente, T., Van Franeker, J.A., Van Loon, W., Vighi, M., Vinci, M., Vlachogianni, T., Volckaert, A., Weiel, S., Wenneker, B., Werner, S., Zeri, C., Zorzo, P. & Hanke, G. (2023). Guidance on the monitoring of marine litter in European seas – an update to improve the harmonized monitoring of marine litter under the Marine Strategy Framework Directive, EUR 31539 EN, Publications Office of the European Union, Luxembourg, doi: [10.2760/59137](https://doi.org/10.2760/59137), JRC133594.
- Galloway, S., Cole, M., & Lewis, C. (2017). Interactions of microplastic debris throughout the marine ecosystem. *Nature Ecology & Evolution*, 1, 0116. <https://doi.org/10.1038/s41559-017-0116>
- GESAMP (2016). Sources, fate and effects of microplastics in the marine environment: part two of a global assessment. In P. J. Kershaw, & C. M. Rochman (Eds.), (IMO/FAO/UNESCO-IOC/UNIDO/WMO/IAEA/UN/ UNEP/UNDP Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection), Reports and Studies GESAMP No. 93 (220 p). <http://www.gesamp.org/site/assets/files/1275/sources-fate-and-effects-of-microplastics-in-the-marine-environment-part-2-of-a-global-assessment-en.pdf> Accessed September 6, 2023.
- Hadibarata, T., Sathishkumar, P., Prasatia, H., Hikmat, Pushtasari, E. D., Tasfiyati, A. N., Muzdalifah, D., Waluyo, J., Randy, A., Ramadhaningtyas, D. P., Zuas, O., & Sari, A. A. (2021). Microplastic contamination in the Skipjack Tuna (*Euthynnus affinis*) collected from Southern Coast of Java, Indonesia. *Chemosphere*, 276, 130185. doi: [10.1016/j.chemosphere.2021.130185](https://doi.org/10.1016/j.chemosphere.2021.130185)
- Herrera, A., Štindlová, A., Martínez, I., Rapp, J., Romero-Kutzner, V., Samper, M. D., Montoto, T., Aguiar-González, B., Packard, T., & Gómez, M. (2019). *Marine Pollution Bulletin*, 139, 127–135. <https://doi.org/10.1016/j.marpolbul.2018.12.022>
- Hussien, N. A., Mohammadein, A., Tantawy, E. M., Khattab, Y., & Malki, J. S. A. (2021). Investigating microplastics and potentially toxic elements contamination in canned Tuna, Salmon, and Sardine fishes from Taif markets, KSA. *Open Life Sciences*, 16, 827–837. <https://doi.org/10.1515/biol-2021-0086>
- Karami, A., Golieskardi, A., Choo, C. K., Larat, V., Karbalaie, S., & Salamatinia, B. (2018). Microplastic and mesoplastic contamination in canned sardines and sprats. *Marine Pollution Bulletin*, 612, 1380–1386. <https://doi.org/10.1016/j.scitotenv.2017.09.005>
- Li, J., Green, C., Reynolds, A., Shi, H., & Rotchell, J. M. (2018). Microplastics in mussels sampled from coastal waters and supermarkets in the United Kingdom. *Environmental Pollution*, 241, 35–44. <https://doi.org/10.1016/j.envpol.2018.05.038>
- Lusher, A. L., Burke, A., O'Connor, I., & Officer, R. (2014). Microplastic pollution in the Northeast Atlantic Ocean: Validated and opportunistic sampling. *Marine Pollution Bulletin*, 88(1–2), 325–333. <https://doi.org/10.1016/j.marpolbul.2014.08.023>
- Oliveira, A. R., Sardinha-Silva, A., Andrews, P. L. R., Green, D., Cooke, G. M., Hall, S., Blackburn, K., & Sykes, A. V. (2020). Microplastics presence in cultured and wild-caught cuttlefish. *Sepia officinalis*. *Marine Pollution Bulletin*, 160, Article 111553. <https://doi.org/10.1016/j.marpolbul.2020.111553>
- Ory, N. C., Sobral, P., Ferreira, J. L., & Thiel, M. (2017). Amberstripe scad *Decapterus muroadsi* (Carangidae) fish ingest blue microplastics resembling their copepod prey along the coast of Rapa Nui (Easter Island) in the South Pacific subtropical gyre. *Science of The Total Environment*, 586, 430–437. <https://doi.org/10.1016/j.scitotenv.2017.01.175>
- Pedà, C., Longo, F., Berti, C., Laface, F., De Domenico, F., Consoli, P., Battaglia, P., Greco, S., & Romeo, T. (2022). The waste collector: Information from a pilot study on the interaction between the common octopus (*Octopus vulgaris*, Cuvier, 1797) and marine litter in bottom traps fishing and first evidence of plastic ingestion. *Marine Pollution Bulletin*, 174, Article 113185. <https://doi.org/10.1016/j.marpolbul.2021.113185>
- NOAA, National Marine Fisheries Service (2022). Fisheries of the United States, 2020. U. S. Department of Commerce, NOAA Current Fishery Statistics, No. 2020. <https://www.fisheries.noaa.gov/national/sustainable-fisheries/fisheries-united-states>. Accessed September 12, 2023.
- Pereira, J. M., Rodríguez, Y., Blasco-Monleon, S., Porter, A., Lewis, C., & Pham, C. K. (2020). Microplastic in the stomachs of open-ocean and deep-sea fishes of the North-East Atlantic. *Environmental Pollution*, 265(Part A), 115060. Doi: [10.1016/j.envpol.2020.115060](https://doi.org/10.1016/j.envpol.2020.115060)
- Pereira, R., Rodrigues, S. M., Silva, D., Freitas, V., Almeida, C. M. R., & Ramos, S. (2023). Microplastic contamination in large migratory fishes collected in the open Atlantic Ocean. *Marine Pollution Bulletin*, 186, Article 114454. <https://doi.org/10.1016/j.marpolbul.2022.114454>
- Qiao, R., Sheng, C., Lu, Y., Zhang, Y., Ren, H., & Lemos, B. (2019). Microplastics induce intestinal inflammation, oxidative stress, and disorders of metabolome and microbiome in zebrafish. *Science of Total Environment*, 662, 246–253. <https://doi.org/10.1016/j.scitotenv.2019.01.245>
- Renzi, M., Specchiulli, A., Blasković, A., Manzo, C., Mancinelli, G., & Cilenti, L. (2019). Marine litter in stomach content of small pelagic fishes from the Adriatic Sea: Sardines (*Sardina pilchardus*) and anchovies (*Engraulis encrasicolus*). *Environmental Science and Pollution Research*, 26, 2771–2781. <https://doi.org/10.1007/s11356-018-3762-8>
- Ríos, J. M., Tesitore, G., & de Mello, F. T. (2022). Does color play a predominant role in the intake of microplastics fragments by freshwater fish: An experimental approach with *Psilododon eigenmanniorum*. *Environmental Science and Pollution Research International*, 29(32), 49457–49464. <https://doi.org/10.1007/s11356-022-20913-8>
- Sathish, M. N., Jeyasanta, I., & Patterson, J. (2020). Occurrence of microplastics in epipelagic and mesopelagic fishes from Tuticorin, Southeast coast of India. *Science of The Total Environment*, 720, Article 137614. <https://doi.org/10.1016/j.scitotenv.2020.137614>
- Scacco, U., Mancini, E., Marcucci, F., & Tiralongo, F. (2022). Microplastics in the deep: Comparing dietary & plastic ingestion data between two mediterranean bathyal opportunistic feeder species, *Galeus melastomus* (Rafinesque, 1810) and *Coelorhynchus caelorhynchus* (Risso, 1810), through stomach content analysis. *Journal of Marine Science and Engineering*, 10, 624. <https://doi.org/10.3390/jmse10050624>
- Shim, W. J., Hong, S. H., & Eo, S. (2016). Identification methods in microplastic analysis: A review. *Analytical Methods*, 9, 1384–1391. <https://doi.org/10.1039/C6AY02558G>
- Silva, D. M., Almeida, C. M. R., Guardiola, F., Rodrigues, S. M., & Ramos, S. (2022). Optimization of an analytical protocol for the extraction of microplastics from seafood samples with different levels of fat. *Molecules*, 27, 5172. <https://doi.org/10.3390/molecules27165172>
- Smith, M., Love, D. C., Rochman, C. M., & Neff, R. A. (2018). Microplastics in seafood and the implications for human health. *Current Environmental Health Reports*, 5, 375–386. <https://doi.org/10.1007/s40572-018-0206-z>
- Steer, M., Cole, M., Thompson, R. C., & Lindeque, P. K. (2017). Microplastic ingestion in fish larvae in the western English Channel. *Environmental Pollution*, 226, 250–259. <https://doi.org/10.1016/j.envpol.2017.03.062>
- United Nations Environment Programme (2021). Drowning in Plastics – Marine Litter and Plastic Waste Vital Graphics. *United Nations Environment Programme (UNEP), Secretariats of the Basel, Rotterdam and Stockholm Conventions (BRS) and GRID-Arendal*. DOI: [10.13140/RG.2.2.13444.65928](https://doi.org/10.13140/RG.2.2.13444.65928)
- Wang, Y., & Chen, X. (2023). Microplastics in pelagic squid (*Dosidicus gigas*) from the eastern tropical Pacific Ocean: Characteristics, spatial variation, and preliminary risk assessment. *Frontiers in Environmental Sciences*, 11, 1069124. <https://doi.org/10.3389/fenvs.2023.1069124>
- Warne, D. (1988). Manual on fish canning. *FAO Fisheries Technical Paper*. No. 285. Rome, FAO, 71p. <https://www.fao.org/3/t0007e/t0007e.pdf> August 28, 2023.
- Wright, S. L., Rowe, D., Reid, M. J., Thomas, K. V., & Galloway, T. S. (2015). Bioaccumulation and biological effects of cigarette litter in marine worms. *Scientific Reports*, 5, 14119. <https://doi.org/10.1038/srep14119>