Microplastic in the gastrointestinal tract of fishes along the Saudi Arabian Red Sea coast

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Abstract

This study assesses the presence of microplastic litter in the contents of the gastrointestinal tract of 26 commercial and non-commercial fish species from four difference habitats sampled along the Saudi Arabian coast of the Red Sea. A total of 178 individual were examined for microplastics. In total, 26 microplastic fragments were found. Of these, 16 being films (61.5%) and 10 being fishing thread (38.5%). FTIR analysis revealed that the most abundant polymers were polypropylene and polyethylene. The grouper (*Epinephelus* spp.) sampled at Jazan registered the highest number of ingested microplastics. This fish species is benthic and feeds on benthic invertebrates. Although differences in the abundance of microplastic ingestion among species were not statistically significant, a significant change was observed when the level of ingestion of microplastics particles was compared among the habitats. The higher abundance of microplastics particles may be related to the habitats of fish and the presence of microplastics debris near the seabed. The results of this study represent a first evidence that microplastic pollution represents an emerging threat to Red Sea fishes, their food web and human consumers.

Keywords: Stomach content; FT-IR; Polymer; Commercial fish; Grouper; Mesopelagic fish

1.1 Introduction

Plastic has become one of the most common manufacturing materials in the world because it is reusable, durable, cheap, and lightweight (Andrady and Neal, 2009). However, the properties that make plastic so useful also make it a significant threat in the environment, where it lasts for decades (Sigler, 2014). Their low density leads to low weight but also renders much of plastic material positively buoyant, allowing for long-range transport in the ocean (Ryan et al., 2009). As a consequence of their global spread across the ocean, marine plastic litter has become a global pollutant, present across all oceans, including the most remote areas of the planet (Cózar et al., 2014, 2015, 2017).

It has been estimated that around 80% of marine debris originates from land-based activities, including litter derived from agriculture, industry, dumping of waste, and discharge with land run-off and rivers. The remaining 20% is derived from ocean based sources, including plastic materials released by commercial shipping, fishing activity (e.g. fishing lines and nets), and recreational boats (Li et al., 2016). Once entering the ocean, heavier plastic materials sink to the seafloor, while lighter, buoyant pieces are dispersed by currents or might sink after being ballasted by biofouling, entering oceanic circulation to accumulate in ocean gyres and semi-enclosed seas (Cózar et al., 2014, 2015, 2017).

Marine plastic litter is slowly broken up by mechanical, chemical, and photolytic degradation processes, resulting in a continuous decline in size, with the modal size of offshore fragments of floating plastic debris being smaller than 1 cm in diameter (Cózar et al., 2014). In general, the term (MP) refers to pieces of plastic smaller than 5 mm, either because of design, such as small rounded microbeads produced as resin pellets and powders in cosmetics and scrubs (Zitko and Hanlon, 1991) or as the outcome of fragmentation processes (Ryan et al., 2009). The size of microplastics, from 10's of microns to a few mm, overlaps with the prey size of a broad range of marine organisms (Lusher et al., 2015), creating a risk of micro plastic ingestion by marine organisms. Indeed, plastic particles have been found across the marine consumer food web, including zooplankton (e.g. salps and copepods), benthic filter feeders (e.g. bivalves and corals), as well as vertebrates (e.g. fish, marine mammals, and seabirds;) (Sul et al., 2014). Plastic ingestion may cause internal blockages and injury to the digestive tract of fish (Cannon et al., 2016; Nadal et al., 2016), which can lead to starvation or malnutrition (Gregory, 2009). An additional and potentially harmful aspect of plastic ingestion by animals is the possibility that hazardous chemicals in the plastics may leach out and be absorbed into the animal's body. This could potentially cause toxic effects to the animal (Rochman et al., 2015).

The Saudi Arabian coast of the Red Sea has been recently characterized as supporting a much lower load of floating microplastic fragments than expected based on its nature as a semi-enclosed sea with an inverted estuarine circulation (Marti et al., 2017). Although Marti et al. (2017) found relatively low concentration of microplastic in this region, abundant fibers were also observed and the ingestion of microplastic by marine organisms that inhabit this area may occur. Synthetic ~~fibres~~fibers, mainly derived from degradation of plastic debris (e.g. rope, packaging materials, and washing of synthetic clothing), are the most abundant type of microplastics in the Red Sea (Marti et al., 2017), and maybe derived from land inputs with sewage and waste water or atmospheric deposition (Marti et al., 2017). Yet, the ingestion of marine plastic debris by marine organisms in the Red Sea has not yet been assessed.

Here we assess the abundant of marine plastic litter in the gastrointestinal tract of Red Sea fishes sampled along the Saudi Arabian coast of the Red Sea. The primary aim of this study was to describe and compare the types of microplastic ingested by fish across different habitats, while testing for possible differences in the frequency of microplastics ingestion among these habitats. Specifically, we examined individuals from 26 fish species from four habitat types: demersal, seagrass, coral reef, and mesopelagic habitats.

~~2.~~2 Materials and methods

~~2.1.~~2.1 Sampling

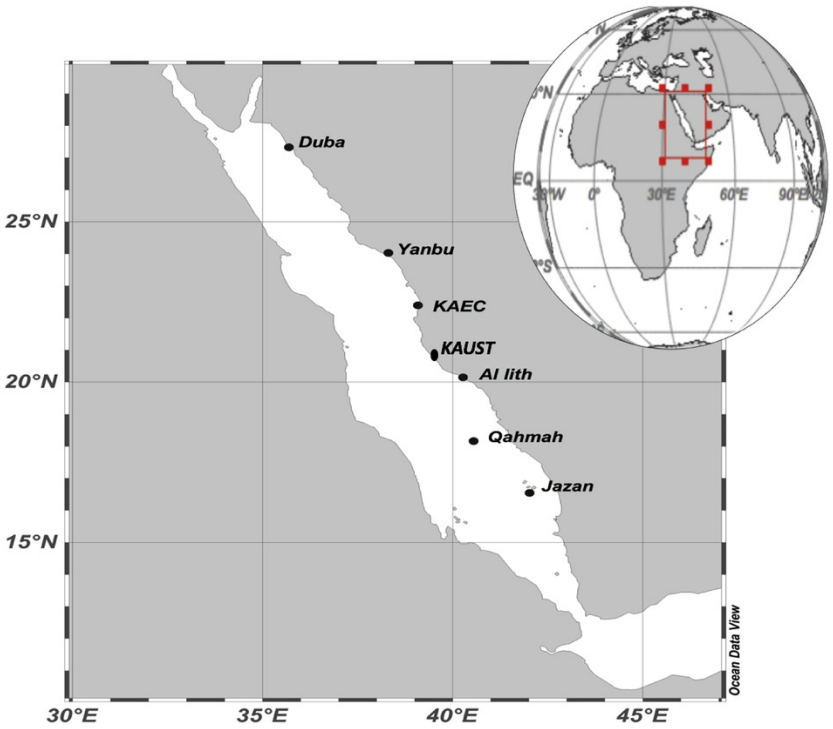
Red Sea fish, including 178 individuals from 26 species from 4 different habitats were collected. The number of individuals of each species depended on availability and was, therefore, not under our control, leading to different numbers of individuals for each species. A total of 89 individuals of commercially important fish were sampled. From these, 38 individuals were from demersal species, 43 individuals from coral reef species, and 8 individuals from seagrass habitats (Table 1). The remaining 89 individuals were from non-commercial species, including 17 individuals from demersal species, 42 individuals from coral reef species and 30 individuals of mesopelagic species; (Table 1). All individuals, from commercial and non-commercial species, were sampled from seven locations along the Saudi Arabian Red Sea coast (Fig. 1, Table 1).

Table 1 Mean values and range of fish length, weight, and stomach weight for all species, (n) = number of fish collected.

alt-text: Table 1

Species name	Species common name	Habitat	Location	Commercial Yes/No	Sample (n)	Mean length (cm) ± SD	Length range (cm)	Mean weight (g) ± SD	Weight range (g)	Mean stomach weight (g) ± SD
Acanthurus gahhm	Black surgeonfish	Demersal	Jizan	Yes	10	33.82 ± 3.72	40–28.1	535.1 ± 187.97	848–308	21.27 ± 6.71
Pristipomoides typus	White snapper	Demersal	Jizan	NO	5	28.46 ± 2.46	32.1–25.7	252.2 ± 70.08	368–188	3.71 ± 0.48
Epinephelus areolatus	Areolate grouper	Seagrass	Jizan	Yes	5	28.42 ± 4.24	33.7–23.8	281.6 ± 127.85	447–175	9.43 ± 1.29
Pristipomoides multidens	Goldbanded jobfish	Demersal	Jizan/Qahmah	Yes	10	28.2 ± 2.66	33.2–25.5	236.7 ± 66.26	364–185	5.47 ± 2.19
Lutjanus kasmira	Bluestripe snapper	Coral reef	Jizan	Yes	12	24.45 ± 3.77	34.9–20.3	233.17 ± 143.5	665–121	4.22 ± 3.52

Lethrinus microdon	Smalltooth emperor	Coral reef	Jizan	Yes	10	29.53 ± 5.2	38.5–22.7	316.5 ± 146.22	605–147	6.66 ± 2.52
Epinephelus chlorostigma	Brownspotted grouper	Seagrass	Jizan	Yes	3	36.33 ± 9.92	42.7–24.9	700.33 ± 443.36	1019–194	12.27 ± 5.87
Gymnocranius grandoculis	Bluelined large-eye bream	Coral reef	Jizan	No	10	28.23 ± 2.31	33.1–26.1	344 ± 110.16	609–244	5.21 ± 2.32
Parascolopsis eriomma	Rosy dwarf monocle bream	Demersal	Jizan	Yes	5	23.3 ± 1.13	24.8–22.2	171 ± 21.37	200–147	4.13 ± 2.41
Sargocentron spiniferum	Sabre squirrelfish	Coral reef	Qahmah	NO	5	30.68 ± 0.89	32–30.1	427 ± 45.39	505–394	11.8 ± 3.11
Epinephelus radiatus	Oblique-banded grouper	Demersal	Qahmah	NO	7	29.34 ± 3.33	34.6–25.2	359.29 ± 129.06	582–217	9.14 ± 3.13
Lipocheilus carnolabrum	Tang's snapper	Demersal	Qahmah	Yes	7	24.39 ± 3.9	31.6–20.7	214 ± 120.32	444–117	4.29 ± 2.56
Plectorhinchus gaterinus	Blackspotted rubberlip	Demersal	Qahmah	Yes	6	26.53 ± 1.96	29.5–24.2	235.17 ± 46.23	298–181	6.33 ± 3.44
Epinephelus epistictus	Dotted grouper	Demersal	Jizan	NO	5	31.4 ± 6.9	38–21.5	424.4 ± 231.23	716–148	9.2 ± 3.7
Pygoplites diacanthus	Royal angelfish	Coral reef	Offshore KAUST	No	5	14.06 ± 2.55	17–10	74 ± 21.82	99–47	7.6 ± 2.07
Cephalopholis argus	Peacock hind	Coral reef	Yanbu	Yes	4	23.63 ± 1.3	25.5–22.5	201 ± 43.64	266–172	7.25 ± 5.85
Abudefduf sexfasciatus	Scissortail sergeant	Coral reef	Al-Lith	No	5	14.63 ± 0.63	15.5–14	60.8 ± 5.4	67–55	1.35 ± 0.43
Acanthurus sohal	Red Sea surgeonfish	Coral reef	Al-Lith	Yes	3	18.9 ± 3.29	21.5–15.2	92 ± 37.04	128–54	3.67 ± 1.15
Dascyllus trimaculatus	Threespot dascyllus	Coral reef	Al-Lith	No	2	10.5 ± 0.71	11–10	32.5 ± 0.71	33–32	1 ± 0
Chaetodon austriacus	Blacktail butterflyfish	Coral reef	Duba	No	10	10.82 ± 0.44	11.5–10	34.8 ± 3.94	39–26	1.1 ± 0.57
Neoniphon sammara	Sammara squirrelfish	Coral reef	Al-Lith	No	5	15.62 ± 1.64	18.2–13.8	31.2 ± 6.65	37–23	1.6 ± 0.55
Naso unicornis	Bluespine unicornfish	Coral reef	Offshore KAUST	Yes	2	40 ± 2.83	42–38	901 ± 70.71	951–851	130 ± 7.07
Thalassoma rueppellii	Klunzinger's wrasse	Coral reef	Al-Lith	Yes	12	16.12 ± 1.75	19.5–14	49.25 ± 17.7	85–23	1.33 ± 0.65
Benthosema pterotum	Skinnycheek lanternfish	Mesopelagic	KAEC	No	10	0 ± 0	2.5–1.9	0 ± 0	0.194–0.112	2.24 ± 0.2
Maurolicus mucronatus	Dragonfishes	Mesopelagic	KAEC	No	10	0 ± 0	2.9–2	0 ± 0	0.188–0.11	2.34 ± 0.25
Vinciguerria mabahiss	Panama lightfish	Mesopelagic	KAEC	No	10	0 ± 0	1.9–1.5	0 ± 0	0.027–0.014	1.72 ± 0.13



~~Figure 1~~Fig. 1 Map of the Red Sea coastal area showing the location of all sampling sites used in this study.

alt-text: Fig. 1

These included 100 individuals captured by artisanal fishermen using traps at depths between 50 m and 100 m, during 2016 and 2017 in coastal areas near Jazan and al-Qahmah. These fish corresponded to 9 commercial species and 5 non-commercial species (Table 1). A total of 43 individuals of coral reef fish were captured in 2011 and 2012, 10 at Duba, 4 individuals from Yanbu, 27 individuals at Al-Lith, and 7 individuals in offshore reefs from KAUST, including 4 species of commercial species and 5 non-commercial species (Table 1). A total of 10 individuals from three abundant species of non-commercial mesopelagic fish (Dalpadado and Gjøsaeter, 1987) were collected in 2014 using Tucker nets at a depth of 700 m offshore from King Abdullah Economic City (Table 1). We examined all of the Red Sea fish individuals we could obtain. However, the number of individuals obtained was not the same across species, because of their different abundance, commercial interest and difficulty to catch (e.g. mesopelagic fish are notoriously difficult to catch).

~~2.2.~~2.2 Sample preparation

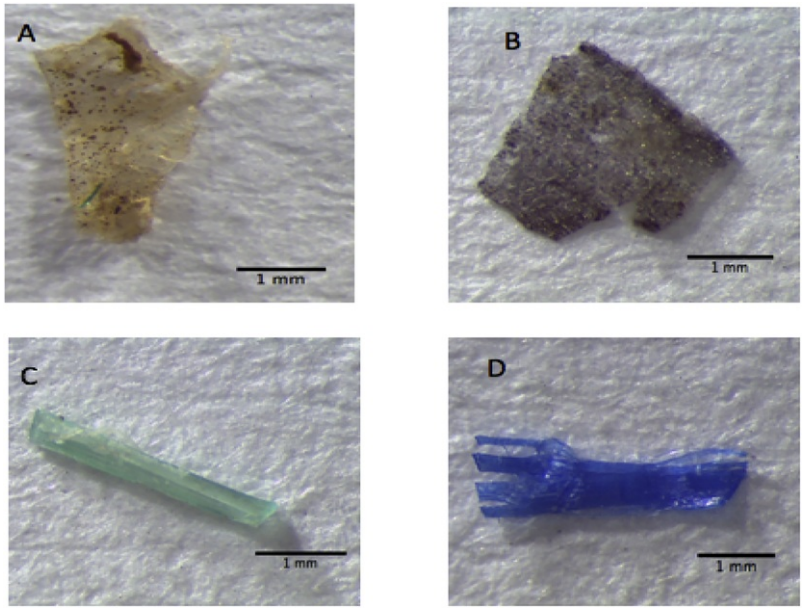
In the laboratory, fish were allowed to thaw at room temperature before examination, and the species was subsequently identified. At the start of the study, sample blanks were placed alongside the sample. For each fish, the total length (TL) was taken (the length (cm), from the tip of the snout to the tip of the longer lobe of the caudal fin), and total wet weight (g) were measured prior to dissection.

Fish were dissected using scissors and forceps to remove the digestive tract from the top of the oesophagus to the anus (Lusher et al., 2013). In order to prevent external contamination entering the gastrointestinal tract during the preparation, the individual (GTI) fish samples were transferred into 50 ml falcon tubes after the dissection and kept capped until analysis. After removal, the samples were placed in an oven for 1 ~~hour~~h at 60 °C. To increase the efficacy of the extraction of plastic from the tissue, a digestion protocol was adapted from the procedure given by Cole et al., 2014 to increase the efficiency of the extraction of plastic from the tissue. NaOH (1 M and 10 M), has been successfully applied to remove biogenic material (e.g. zooplankton). We chose this method because it is simple, inexpensive, involves low chemical hazard, and allows the sampled to be analysed by FTIR following separation. Thirty~~ml~~ milliliter of a 1 M NaOH solution (Sigma Aldrich, Steinheim, Germany) were added to remove the biological material present in the samples (Cole et al., 2014; Catarino et al., 2017), with non-digestible residue (e.g. shells and plant) remaining, in addition to microplastic materials, following the chemical digestion.

Samples were manually shaken intermittently for about 40 s, each time, during the incubation period in order to facilitate complete digestion. The digested samples were then filtered through a 200 µm stainless steel sieve, and the residue retained on the sieve was backwashed into a Petri dish with distilled water. The microplastic loads reported here should be considered conservative estimates, as we cannot ensure that recovery was 100%.

~~2.3.~~2.3 Detection of microplastic

The samples were visually inspected for the presence of microplastic under a binocular stereoscope (Stemi 2000 Zeiss with PI 10×/23 maximum magnification), using distilled water to rinse the GIT contents and help identify plastic particles. The samples were carefully inspected for the presence of plastic particles, including the edge of the Petri dish, where micro-plastic particles usually attach. The particles were counted and photographed (Fig. 2), and image-processing software "ImageJ" (v.1.50i; http://imagej.nih.gov) was used to measure the maximum length of each particle (units mm).



~~Figure 2~~Fig. 2 The morphotypes included (A, B) films (C, D) fishing threads.

alt-text: Fig. 2

~~2.4.~~2.4 FTIR polymer identification

Fourier-transform infrared spectroscopy (FTIR) is a fingerprinting technique used extensively for the characterization of plastic polymer particles. Carbon-based polymers can be described easily from different bond compositions by yielding a unique spectrum that discriminates plastic particles from other organic and inorganic particles (Löder et al., 2015). The FTIR spectra of the samples were obtained with a Nicolet 6700 µFT-IR spectrometer (Thermo~~TM~~™) equipped with a DTSG-KBr detector coupled with a microscope, collected in the transmittance mode according to Yang et al. (2015). The measurement resolution was set at 4 cm^{-1} in the range of 4000–650 cm^{-1} with 32 scans. All spectra were post-processed under an automatic baseline correction mode via the OMNIC library software. To confirm the polymer type, all spectra were compared with Hummel Polymer (Thermo Fisher Scientific, USA) as a reference. When interpreting FTIR output, only readings with confidence levels of 50% or greater (Lusher et al., 2013) and those considered to have reliable spectra matches (after visual inspection) were accepted. Only these particles were included for further analysis. The FTIR test was performed to confirm the identity of each putative plastic item found during the inspection of the gastrointestinal tract, representing 2% (26 particles) that had been correctly identified as plastics using visual microscopy, and the colour was determined visually. Fibers were the most common morphotypes of plastics in the present study (98%), consistent with previous reports (Lusher et al., 2013; Rochman et al., 2015; Neves et al., 2015; Nadal et al., 2016) and the majority of them were black. Unfortunately, the FTIR test could not be applied to those items identified as possible fibers, as the FT-IR procedure could not produce spectra for fibers of such small width, so FTIR examination is inconclusive. Hence, a hot point test (hot needle held with forceps, Devriese et al., 2015) was applied on a sub-sample of ~~fibres~~fibers (n = 48) to determine the plastic nature of these fibers. Moreover, it is likely that these fibers belong to the category microplastics since non-synthetic fibers are more likely to be discarded in the extraction and clean up procedure described for biota. In this paper, we report

microplastic particle counts including these particles.

~~2.5.~~2.5 Contamination prevention and blanks

In order to reduce the risk of contamination, especially airborne contaminants such as fibers, special attention was taken to prevent sample contamination during the dissection, extraction, sorting, and the visual identification. Clean dissection tools were wiped with 70% ethanol and used for every individual fish. Cotton clothing and cotton lab coats, as well as gloves (nitrile) were worn when working to reduce contamination along the study.

Blanks were taken during every dissection and inspection sessions. For each sample, a new petri dish filled with distilled water was placed alongside the sample during the dissection and the visual inspection as a contamination control. After the inspection of the sample, the control dish was checked for any contamination using a binocular stereoscope. A mean of two fibers per blank measurement (range 0 to 3 fibers per blank sample) were found. Hence, we subtracted the mean blank value (i.e. 2 fibers) from the value obtained for each fish, to avoid biasing the results by including fibers derived from airborne contamination. Sample blanks revealed negligible levels of contamination, thus airborne contamination was not a risk in accordance with the corresponding fish sample.

~~2.6.~~2.6 Statistical analyses

Prior to data analyses, we grouped all fish individuals by habitat, without considering their species, which was then used in fixed factor for statistical analysis. One-way ANOVA was performed, after checking for homogeneity of variances and for normality with Shapiro-Wilk test, to test for differences in the abundance of microplastics particles in fish from four habitats: coral reef, demersal, mesopelagic and seagrass, followed by Tukey's HSD test (assuming homogeneous variances). Significant differences were recorded at $p < 0.05$. Data were analysed using RStudio (v1.1.419) software. A chi-square test ($\chi 2$) of independence was utilized to test for differences between microplastic particle ingestion between fish species (commercial vs. non-commercial), with a significance level of $p < 0.05$, conducted in SPSS v 1.0.0.800 (http://www-03.ibm.com/software).

~~3.~~3 Results

The 178 individuals examined spanned a range of sizes (length: 1.2 to 42.9 cm, weight: 0.014 to 1019 g), and habitats (demersal, coral reef, seagrass, and mesopelagic, Table 1). Microplastic fragments were found in a total of 26 of the fish examined (14.6% of the sampled fish) (Table 2). Eighteen out of the 26 fish species examined contained plastic fragments, resulting in an average prevalence of plastic fragments within each species of 14.4 (±0.3 SE) %. The highest number of ingested microplastics per individual was observed in *Parascolopsis eriomma*, a species feeding on benthic invertebrates in muddy and sandy offshore sediments. One of the samples from Jazan had ingested 3 particles consisting of 2 films (e.g. bags, wrapper, or part of them), and 1 fishing thread (including those released from nets). Within a species, the highest prevalence of microplastic ingestion (>20% of individuals) was found in the groupers (*Epinephelus* spp.) and the blackspotted rubberlip (*Plectorhinchus gaterinus*). Although differences in the prevalence of microplastics ingestion between fish species (commercial vs. non-commercial) were not statistically significant (Chi-square test; $\chi 23 = 6.04$, $p = 0.109$).

Table 2 Frequency of microplastic ingestion per fish species.

alt-text: Table 2

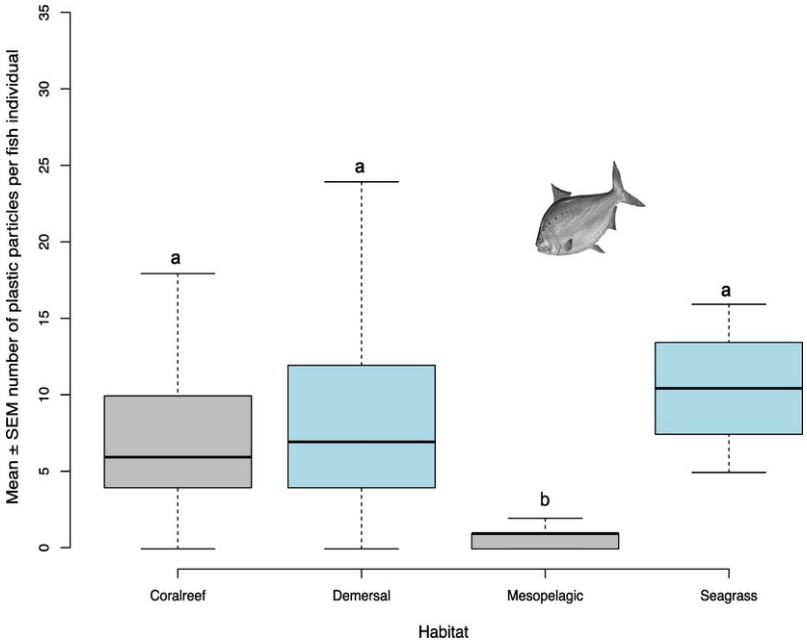
Species name	Habitat	Location	Commercial- Yes/No	Number of stomachs examined	Number of microplastic found in stomach per species	Total number of microplastic found in stomach per species	Average microplastic size per species (mm)	% Ingestion
Acanthurus gahhm	Demersal	Jizan	Yes	10	1	1	2.7	100
Pristipomoides typus	Demersal	Jizan	NO	5	0	0	0	0
Epinephelus areolatus	Seagrass	Jizan	Yes	5	1	1	1.8	20
Pristipomoides multidens	Demersal	Jizan/Qahmah	Yes	10	2	2	3.8	20
Lutjanus kasmira	Coral reef	Jizan	Yes	12	2	2	2.16	16.67
Lethrinus microdon	Coral reef	Jizan	Yes	10	2	2	1.48	20
Gymnocranius grandoculis	Coral reef	Jizan	No	10	2	2	2.35	20
Epinephelus chlorostigma	Seagrass	Jizan	Yes	3	1	1	1.9	33.33

Parascolopsis eriomma	Demersal	Jizan	Yes	5	3	3	1.38	60
Sargocentron spiniferum	Coral reef	Qahmah	NO	5	0	0	0	0
Epinephelus radiatus	Demersal	Qahmah	NO	7	1	1	2.14	14.29
Lipocheilus carnolabrum	Demersal	Qahmah	Yes	7	2	2	1.87	28.57
Plectorhinchus gaterinus	Demersal	Qahmah	Yes	6	2	2	3.31	33.33
Epinephelus epistictus	Demersal	Jizan	No	5	1	1	2.71	20
Pygoplites diacanthus	Coral reef	Offshore KAUST	No	5	0	0	0	0
Cephalopholis argus	Coral reef	Yanbu	Yes	4	0	0	0	0
Abudefduf sexfasciatus	Coral reef	Al-Lith	No	5	1	1	1.2	20
Acanthurus sohal	Coral reef	Al-Lith	Yes	3	0	0	0	0
Dascyllus trimaculatus	Coral reef	Al-Lith	No	2	0	0	0	0
Chaetodon austriacus	Coral reef	Duba	No	10	1	1	4.68	100
Neoniphon sammara	Coral reef	Al-Lith	No	5	1	1	1.51	20
Naso unicornis	Coral reef	Offshore KAUST	Yes	2	0	0	0	0
Thalassoma rueppellii	Coral reef	Al-Lith	Yes	12	1	1	1.93	8.33
Benthosema pterotum	Mesopelagic	KAEC	No	10	1	1	2.58	100
Maurolicus mucronatus	Mesopelagic	KAEC	No	10	1	1	1.42	100
Vinciguerria mabahiss	Mesopelagic	KAEC	No	10	0	0	0	0
Total				178	26			
Average of stomachs with microplastic per species					1.00			
Average by the total number of fishes by species					0.146			

The size of microplastic particles ingested by the fish varied from 1 to 3 mm, and the average size for all particles collected was 2.39 ± 0.28 (SE) mm. Microscopic examination revealed that the dominant type of microplastic fragment was microplastic fibers, followed by film, and fishing thread (Fig. 4a), with the microplastic materials presenting a diversity of colours (blue, black, green, white, and red) (Fig. 4b). Characterization of microplastic fragments using FTIR spectroscopy showed that most of the particles were polypropylene, polyethylene, polystyrene, polyvinyl chloride, and polyacrylonitrile (Fig. 4c).

The plastic nature of fibers could not be verified using FTIR spectroscopy due to their small width. However, a hot needle test (Devriese et al., 2015), was consistent with 77% of the fibers (37 out of 48 fibers tested) being of plastic materials.

A one-way ANOVA test between ~~subject~~subjects was performed to compare the ingestion of microplastics level across habitats. This analysis indicated that the mean microplastics particles (per individual fish) from the demersal, seagrass, and coral reef habitats were significantly higher than in the mesopelagic habitat (one-way ANOVA; $p < 0.001$; $F = 13.83$; Fig. 3).



~~Figure 3~~Fig. 3 -Data represents mean ± SEM number of microplastics particles per fish individual among habitat of fish. Letters above error bars indicate similarities (e.g., a) or differences (e.g., b) among habitats (ANOVA with Tukey post-hoc analysis, $p < 0.05$).

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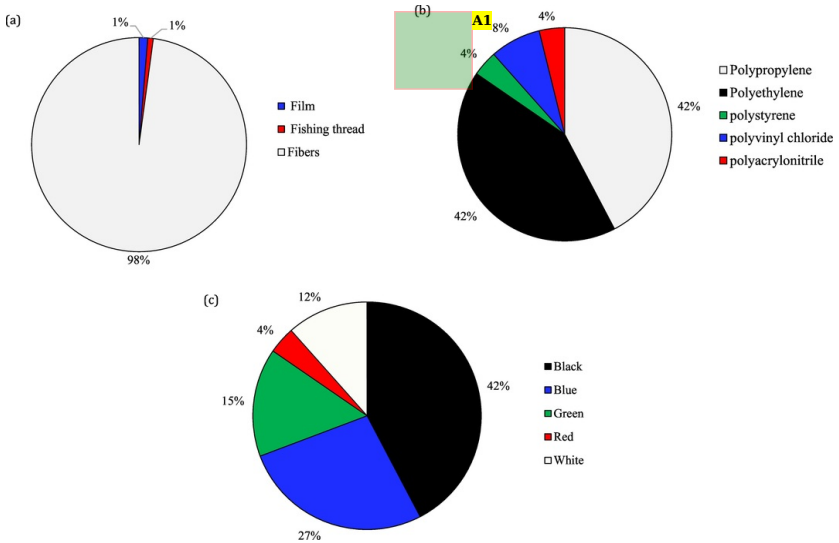

~~Figure 4~~Fig. 4 -Microplastic particles extracted from fishes varied by form, polymer type and colour. (a) Suspect debris was categorized as microplastic film-, fishing thread, or ~~fibrs~~ fibers (>5 mm) (n = 26 particles), (b) FT-IR analysis was used to determine the constituent polymer (n = 26 particles) of the suspected microplastic litter. Identification of polymers was performed by comparison with a library of standard spectra and only polymers matching reference spectra for ~~more than~~ ≥50-% were accepted. Values are expressed in percentages, (c) colour categories of plastic fragments in the stomach contents. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

alt-text: Fig. 4

Annotations:

A1. the (b) and (a) need to be adjust as one show far (a) in the corner and (b)is up

This analysis indicated that the mean microplastic particles (per individual fish) from demersal, seagrass, and coral reef habitats were significantly higher than that in the mesopelagic habitat (one-way ANOVA; $p < 0.0001$; $F = 13.83$ Fig. 3). Indeed, mesopelagic fish were significantly smaller than fish sampled from other habitats.

The number of microplastics particles per individual fish increasing as the 2/3 power of fish length ($R^2 = 0.36$, $p < 0.0001$). The results of Chi-square test of independence showed that the commercial and non-commercial fish did not differ in the likelihood of containing microplastics items ($\chi 23 = 6.04$, $p = 0.109$), nor did the frequency of plastic abundance differ with trophic mode of the species (ANOVA, $p > 0.05$).

~~4.~~4 Discussion

The present report provides a first assessment of plastic debris contained in fishes along the Saudi Arabian Red Sea coast, involving a broad range of fishes (in terms of size, taxonomy, and habitat). The study showed that about one-seventh of the fish examined in this study had ingested small plastic pieces, with 65% (10 of 13) of the commercial species examined containing microplastic debris. The proportion of non-commercial fish that contained microplastic debris is 35% (8 of 13 species). The overall prevalence of fish with ingested plastic debris was 14.60%, within the range of that found in studies elsewhere (Table 3), despite the very low loads of floating microplastic in the Red Sea (Marti et al., 2017). The absence of microplastics in 30.7% (n = 8 spp.) of the species examined here may be due to the small sample size for those species (mean n = 0.5, range 2 to 10 individuals, see Table 2), so we cannot exclude the possibility that they would show a similar prevalence of marine microplastic in their guts if a larger sample size would have been obtained.

Table 3 Summary of the prevalence of plastic items found in fish in previous studies and the results reported here for Saudi Red Sea coast.

alt-text: Table 3

Area	Type of fish	Sample	% Ingestion	Size of MP particles (mm)	Ref
1- North Pacific Subtropical Gyre	Mesopelagic	141	9.20%	2.2 ± 1.9	Davison and Asch (2011)
2- Portuguese coast	Commercial	263	32.70%	2.11 ± 1.67	Neves et al. (2015)
3- English Channel	Pelagic and demersal	504	36.50%	0.13 ± 14.3	Lusher et al. (2013)
4- Mediterranean Sea	Pelagic fish	121	18.2%	1.51 ± 16.50	Romeo et al. (2015)
5-Swedish west coast	Demersal fish	62	68%	--	Karlsson et al. (2017)
6- Northwest Atlantic	~~Mesolelagic~~ Mesopelagic fish	280	73%	969 ± 1048	Wieczorek et al. (2018)
7- French rivers	Wild fish	186	12%	--	Sanchez et al. (2014)
8- Adriatic Sea	Commercial	125	28%	1.78 ± 0.97	Avio et al. (2015)
9- Red Sea	Commercial and non-commercial	178	14.60%	2.39 ± 0.28	This study

The demersal species studied were generally carnivorous and omnivorous, feeding on a variety of food of both plant and animal origin (e.g. benthic fish, crustaceans, mollusks and algae). About 1/3 (38.5%) of the demersal species examined had at least one individual with ingested microplastics.

Coral reef and seagrass fish species showed a prevalence of 46.2% and 7.7% of individuals containing microplastics debris, respectively. This suggests that species associated with reef habitats are more likely to ingest microplastic particles than those in seagrass habitats. However, we found no significant difference in the prevalence of microplastic ingestion across different habitats, possibly a consequence of low power in our analysis due to limited sampling size for some feeding habits. Feeding habits and habitat can influence the likelihood of ingestion of plastic debris, consistent with evidence that microplastic ingestion depends on feeding strategies (Jabeen et al., 2017). This suggests that ingestion of microplastic might occur depending on the feeding habits of fish regardless of prey type.

Mesopelagic species had ingested plastic debris, but showed a low prevalence, with only 7.7% of the individuals containing plastic debris, comparable to results of mesopelagic fish sampled in the North Pacific Subtropical Gyre

(Davison and Asch, 2011), (Table 3). The Red Sea mesopelagic fish examined were captured at depth (700 m) and yet contained floating plastic debris. This may be attributable to the daily vertical migrations that characterizes these animals (Klevjer et al., 2012)-, which swim to feed on the surface at night and then return to the mesopelagic layer to seek refuge from predators during the day (Klevjer et al., 2016). Although mesopelagic fish are not currently of commercial interest, they play an important role in the marine food web (Gjøsaeter et al., 1980) and comprise the largest stock of fish in the ocean (Irigoien et al., 2014). Therefore, even if the prevalence of microplastic ingestion by mesopelagic fish is low. This is likely attributable to the small size (mean ± SE length = 2.1 ± 1.1 cm) of mesopelagic fish compared to those in other habitats (mean length > 20 cm, Tukey HSD, $p < 0.001$). They may represent a major link in the transfer of microplastic debris up the food web, as mesopelagic fishes are important prey for tuna (Marchal and Lebourges, 1996), squid (Koz, 1995), and marine mammals (Naito et al., 2013), among others.

The mean size of all particle fragments retrieved from the guts of fish in this study (2.39 ± 0.28 mm) is similar to the mean size of floating plastic items in the Red Sea (2.08 ± 2.74 mm) (Marti et al., 2017) and is similar to the size range of plastic fragments retrieved from fish guts elsewhere (Table 3). Moreover, (Cózar et al., 2014) reported a size-dependent loss of floating microplastic during oceanic transport, with the size class where losses occur concentrated around sizes of 2.2 mm. This matches the mean size of microplastic ingested by Red Sea fishes, as well as those found in some other locations (e.g. 2.2 ± 1.9 in the North Pacific Subtropical Gyre, Davison and Asch, 2011, Table 3). This might suggest that ingestion by fish may play a major factor in the removal of floating microplastic from ocean waters. Hence, the results presented could suggest that Red Sea fishes are likely to play a major role as sinks of floating microplastics, which would lead to the transference of microplastic along the food web, to which human are connected.

This might suggest that ingestion by fish it can play a major factor in the removal of floating microplastic from ocean waters. The identified plastic particles in this study were composed mainly by the polymer polypropylene, consistent with the prevalence of these materials among plastic debris floating in the Red Sea (Marti et al., 2017). In nature, these polymers have been reported to contain adsorbed persistent organic pollutants that potentially impact marine organisms (Teuten et al., 2009). In addition, additives added into plastic polymers during the manufacturing processes, such as phthalates and bisphenol A, are hazardous to marine biota through their role as endocrine-disrupting chemicals that can mimic, compete with, or disrupt the synthesis of endogenous hormones (Talsness et al., 2009). Hence, fish containing microplastics could also be affecting by associated hazardous chemical compounds, with possible impacts on fish health transferred along the food web.

Microplastic were found in the GIT of one in each six individuals of Red Sea fish examined here, regardless of habitat or feeding habit, with the number of putative synthetic fibers in the fish increasing significantly with body size. The prevalence of microplastic ingested by Red Sea fishes was comparable to that reported in other marine ecosystems, despite the Red Sea supporting the lowest load of floating microplastic so far reported (~~Martin~~Marti et al., 2017). Hence, we suggest that Red Sea fishes are likely to play a major role as sinks of floating microplastic, which would lead to the transference of microplastic along their food web, to which human may be connected. Indeed, the prevalence of microplastics on fish was comparable for commercial species (i.e., those consumed by humans) and non-commercial species.- Managing plastic litter is, therefore, not only essential to maintain good ecological health in the Red Sea and elsewhere, but also to protect consumers from ingesting the plastic we dispose in the environment (Koelmans et al., 2017).

~~5.~~5 Conclusion

In the present study, we reported plastic pollution in commercial and non-commercial fishes from Red Sea for the first time. This study provides an important contribution to the knowledge and understanding of plastic occurrence in these commercial and non-commercial fish, given also their importance in Red Sea catches and human consumption.

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Highlights

- Microplastics pose a risk to organisms.
- Plastic litter in fishes from different habitat types (coral reef, demersal, mesopelagic and seagrass) were assessed.
- ~~**The characterization of microplastics in the stomach of commercial and non-commercial species** are~~ **is** ~~**investigated.**~~ Microplastic were found in the guts of one in each six indivduals of Red Sea fish examined here, regardless of habitat or feeding.
- The majority of plastics were fibers, a limited number of particles has scored.

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