

Plastic Particles in the Gastrointestinal Tract of Some Commercial Fish Species Inhabiting in the Gulf of Bejaia, Algeria

Z. Zeghdani^{1,*}, S. Mehdioui², Y. Mehdioui², R. Gherbi¹ and Z. Ramdane¹

¹ Laboratoire de Zoologie Appliquée et d'Ecophysiologie Animale, Faculté des Sciences de la Nature et de la Vie, Université de Bejaia, Bejaia 06000, Algérie ; ² Département des sciences biologiques de l'environnement, Faculté des Sciences de la Nature et de la Vie, Université de Bejaia, Bejaia 06000, Algérie

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Abstract

In the present study we report for the first time, the presence of plastic particles on the gastrointestinal tracts of the examined commercial fish species: *Sardinella aurita* (n=60), *Sardina pilchardus* (n=60), *Pagellus acarne* (n=45), *Trachurus trachurus* (n=38), *Boops boops* (n=40), *Sparus aurata* (n=6). The sampled fish species (n=249) inhabiting in demersal, benthopelagic and pelagic marine ecosystems (Gulf of Bejaia) were examined to test eventual differences in micro and macro plastic particles. Overall, results showed that 58.63% of gastrointestinal tracts were contained plastic particles. An average 8.45 ± 14.69 items per fish were recorded. The most common morphotype particles were fiber (85%) whereas dominant colors of plastic debris were the blue and the red among three types of particle colors. Our results shows that there are no significant differences between females and males specimens in terms of ingested plastic particles. The highest abundance of plastic particles in fish gastrointestinal tracts was recorded in wet season.

Keywords: Plastic particles, Microplastics, Commercial fish species, Gulf of Bejaia.

1. Introduction

Plastics are the most widely used material (Millet *et al.*, 2018), inexpensive, lightweight, strong, durable, corrosion-resistant materials, with high thermal and electrical insulation properties (Thompson *et al.*, 2009), and this is why this material persists in the environment (Teuten *et al.*, 2009).

Sunlight, wind, and waves are the main natural factors transforming plastics into small particles (Matjasic *et al.*, 2021). When the size of these particles is less than 5 mm they are called microplastics (Hartmann *et al.*, 2019), some researchers use 0.5 or 1 mm as a maximum size for microplastics (Andrady, 2011; Cole *et al.*, 2011).

Microplastic particles can have either been manufactured purposely with that size (primary microplastic) or proceeding from fragmentation by different physical, chemical, and biological degradation (secondary microplastic) (Wright *et al.*, 2013; Rainieri *et al.*, 2018).

Plastics are emergent pollutants and have been found almost in every part of the planet (Mishra *et al.*, 2021) and their risk (macroplastics and microplastics) on the marine environment have been addressed by reporters as an emerging global problem that detrimentally affects marine organisms (Derraik, 2002). Regarding their size and form (fibers, fragments, etc.), this particle can be unfortunately ingested by biota (Galgani *et al.*, 2010; Andrady, 2011).

For these reasons many scientific works were conducted on microplastic ingestion by fish species (Takarina *et al.*, 2022; Piyawardhana *et al.*, 2022; Thiele *et al.*, 2021; Yin *et al.*, 2019), especially, in the Mediterranean Sea (Bellas *et al.*, 2016; Guven *et al.*, 2017; Pennino *et al.*, 2020), where researchers reported the occurrence of microplastic particles in the gastrointestinal tract of fish (Jabeen *et al.*, 2017; Jaafar *et al.*, 2021; Parvin *et al.*, 2021; Akhter and Panhwar, 2022).

The Mediterranean Sea recently classed as the most impacted regions of the world by plastic debris, demersal species from coastal zones ingest more plastic particles than other species (Murphy *et al.*, 2017), polymers can act as vector and adsorb heavy metals from the water column (Holmes *et al.*, 2014; Boucher *et al.*, 2016) which have a tendency to flow (Lagarde *et al.*, 2016).

Low-density microplastics are found in surface waters (Thompson *et al.*, 2004), they held some heavy metals (Brennecke *et al.*, 2016), then ingested by several species of zooplankton (Cole *et al.*, 2013) than by larvae and adults of fish (Browne *et al.*, 2013; Lusher *et al.*, 2013; Rochman *et al.*, 2013).

Researchers have reported that microplastics can enter the human body through the food chain and human exposure to microplastics could lead to harmful health (Couture, 2017), microplastics enriched bacterial pathogens (Junaid *et al.*, 2022) and can serve as carriers of antibiotic-resistant bacteria (Pham *et al.*, 2021).

The implications of the complex microplastics-heavy metals-bacterial pathogens to the human health are

* Corresponding author. e-mail: zeghdani1993@gmail.com; zouhir.zeghdani@univ-bejaia.dz.

significant (Wang et al., 2021). In effect, this complex seems to amplify the health risks for humans and animal.

In Algerian coasts, no studies were conducted on plastic particles containing in the gastrointestinal tracts of commercial fish species. The aim of the present study is to examine some commercial fish species (from Algerian coasts) for the presence of plastic debris (microplastics and macroplastics) in the gastrointestinal tract, to characterize the isolated plastic debris (form, color, size, etc.), and finally to analyze the variation of ingested plastic particles according to fish parameters, fish habitat and seasonal variation. These results maybe give us a clear idea on the presence of these toxic pollutants in the Algerian marine ecosystems, and the health risks that can induce to consumers of fish.

2. Materials and methods

A total of 249 specimens of fish belonging to six species (with high commercial value) were sampled in the port of Bejaia, Algeria (Figure.1) or from fish markets.

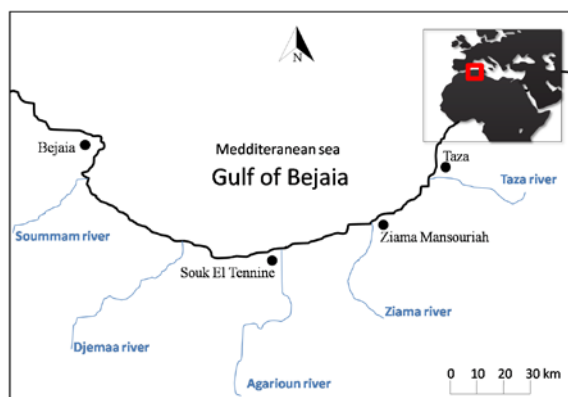


Figure 1. Location of the study area (Gulf of Bejaia)

In our sampling, we focused on fish species occupying various habitats (demersal, benthopelagic and pelagic) (Table.1).

In the laboratory, we worked in sterile space, in order to reduce the risk of air contamination using clean materials (disinfect with ethanol 70%), after identification of sampled fish specimens, total length (cm) and total weight (g) were measured for each specimen.

Table 1. Habitat and Biological parameters of examined fish species in the study (n = 249).

Fish species	(n)	Habitat	Mean weight (g) ± SD	Mean length (cm) ± SD
<i>Sardinella aurita</i>	60	Pelagic	26.95 ± 23.1	16.43 ± 4.1
<i>Sardina pilchardus</i>	60	Pelagic	25.43 ± 11.5	12.49 ± 3.9
<i>Pagellus acarne</i>	45	Benthopelagic	48.75 ± 18.4	14.61 ± 3.6
<i>Trachurus trachurus</i>	38	Pelagic	44.08 ± 12.31	16.84 ± 5.1
<i>Boops boops</i>	40	Demersal	56.91 ± 32.4	17.83 ± 2.7
<i>Sparus aurata</i>	6	Demersal	304.3 ± 11.2	25.67 ± 2.6

The protocol adapted by Baalkhuyur *et al.* (2018) was applied regarding its simplicity and feasibility. The different stapes devoted to detect the plastic particles in the examined fish individuals are: -Fish specimens were dissected and sexed (visual observations). After that, the gastrointestinal tract (GIT) was removed (from esophagus to the opening) and weighted;-The specimen's gut were rinsed with distilled water, and then carefully moved into cleaned Petri dishes. For each sample a code number was given; -After removal, the samples of GIT were placed in an oven for 1 hour 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). NaOH (1 M and 10 M), has been successfully applied to remove biogenic material; -30ml of a 1 M NaOH solution were added to reinforce the digestion (remove the remaining biological material and non-digestible residue) (Cole *et al.*, 2014; Catarino *et al.*, 2017); -Samples were manually shaken intermittently during the incubation period in order to facilitate complete digestion, and after the incubation, samples were inspected under binocular stereoscope visually (Hidalgo-Ruz *et al.*, 2012; Free *et al.*, 2014).

After identification, the plastic particles were counted, photographed and measured using the software "imagej" (ver: 1.4.3; <https://imagej.nih.gov/>), color and shape were also determined for each plastic particle (Jabeen *et al.*, 2017).

The occurrence of Frequency (FO %) of the collected microplastics in the digestive tracts was calculated using the following formula:

FO (%) = (Ni/N) × 100, where

FO% = frequency of occurrence of plastic particles;

Ni = number of gastrointestinal tracts that contained plastic particles;

N = total number of gastrointestinal tracts examined.

Data were analyzed using SPSS 14.0 software and EXCEL 2010. Independent t-test was performed to determine if there are differences on the abundance of plastic particles between wet season and dry season, and between males and females (95% confidence level). A significant difference in the abundance of plastics among individuals was tested applying one-way ANOVA. The Tukey test's HSD test was set at * = p < 0.05 and ** = p < 0.01 values.

3. Results

3.1. Biological parameters of fishes

A total of 249 specimens attached to 6 species were analyzed (Table.1). The body weight of fish specimens varies from 10.30 g to 328.20 g (31.74 g ± 20.42), and the total length varies from 10.90 cm to 24.20 cm (15.11 cm ± 2.5)

3.2. Intensity of plastic consumed by fishes

Evidence of plastic particles was appeared to be in 146 specimens from the total 249 (58.63%) examined in this study. These potential contaminants were found in the 6 species (Table.2), and therefore in pelagic, benthopelagic, and demersal species. The average number of particles

ingested was (8.45 ± 14.69 particles per specimen) ranging from 0 to 69 particles per gastrointestinal tract, the highest

average was recorded in *Boops boops* L. (21.03 ± 23.49 particles per specimen).

Table 2. Frequency of plastic particles ingestion by fish species.

Fish species	n of examined GIT	n of contaminated GIT	Rate (%)
<i>Sardinella aurita</i>	60	35	58.33
<i>Sardina pilchardus</i>	60	26	43.33
<i>Pagellus acarne</i>	45	33	73.32
<i>Trachurus trachurus</i>	38	24	63.16
<i>Boops boops</i>	40	22	55
<i>Sparus aurata</i>	6	6	100

GIT: Gastrointestinal Tract

Collected microplastics and mesoplastics from the examined fish specimens (2347 particles) recovered various morphotypes and colors of plastic particles.

3.3. Morphotype, size and color of the plastic particles retrieved

Two morphotypes of plastic particles were extracted (Figure.2), where the most observed were fibers with 85% ($p < 0.01$). In *Sardinella aurita*, *Sardina pilchardus* and *Trachurus trachurus* fibers reached 100%.

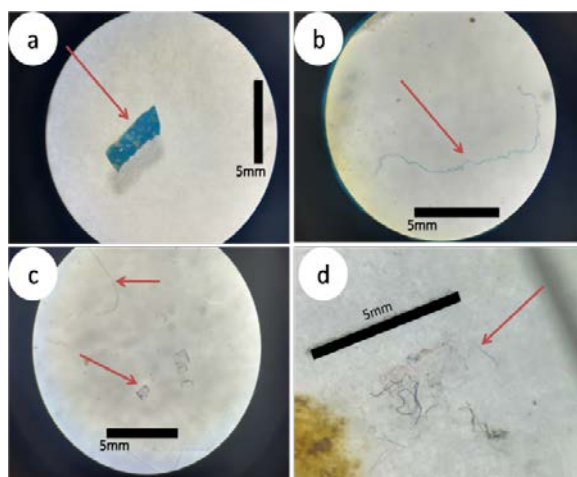


Figure 2. The morphotypes (a) fragment ingested by examined *Sparus aurata*; (b) mesoplastic fiber; (c) transparent fragments and a red fiber observed in the gut of *Boops boops*; (d) various fibers ingested by examined *Sardina pilchardus* (see red arrows).

Fragment morphotypes was exclusively extracted from *Boops boops*, *Sparus aurata* and *Pagellus acarne* with 15% of the total number of plastics (Figure.3a). The size of plastic particles varied from 0.1 mm to 5 mm for microplastics and the highest size of a mesoplastic was 18 mm, microplastics were the most observed with 94.58% of the total number of particles ($p < 0.01$) (Figure.3b).

Microplastics lower than 2 mm (size $< 2\text{mm}$) represent 71% of the number of analyzed plastics.

In the study region, the extracted plastics represent numerous colors (especially red, blue and transparent). The dominant color was the blue plastic particles with 56.4% followed by the red color with 41.3%. Transparent and other colors were not highly observed (Figure.3c).

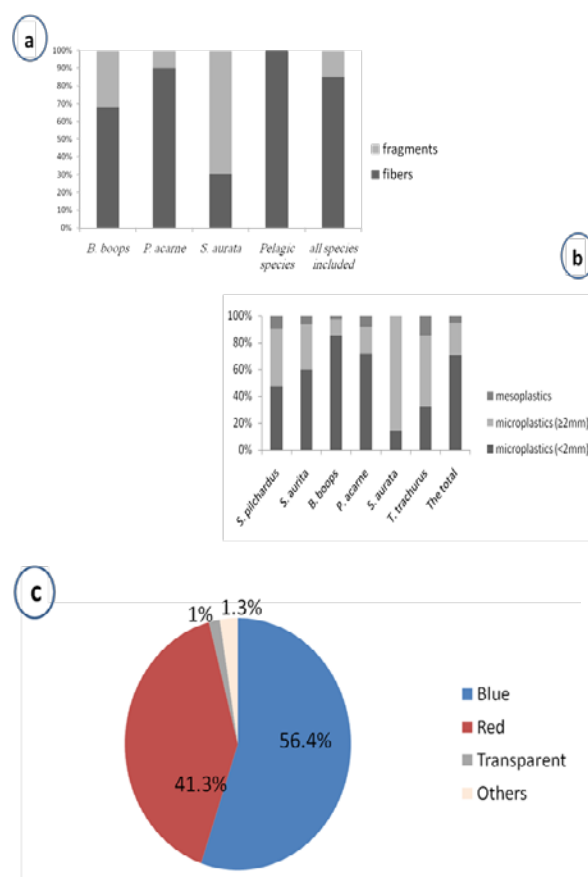


Figure 3. morphotypes (a), size (b) and color (c) of the ingested plastics by the contaminated fish species.

3.4. Occurrence and mean number of collected microplastics according to sex.

From the total of samples, 123 were females, 96 were males and 30 were undetermined individuals. The percentage of females with infected gastrointestinal tract reached 74.1% followed by males (FO reached 21%). When comparing the number of ingested plastics particles we found that females ingest slightly more than males (1055 and 1022 particles respectively) and for undetermined gender, a number of 270 particles was noted. According to sex, the mean number of plastic particles (Figure.4) shows no significant differences between the 3 genders (ANOVA, Tukey HSD $p\text{-value} > 0.05$).

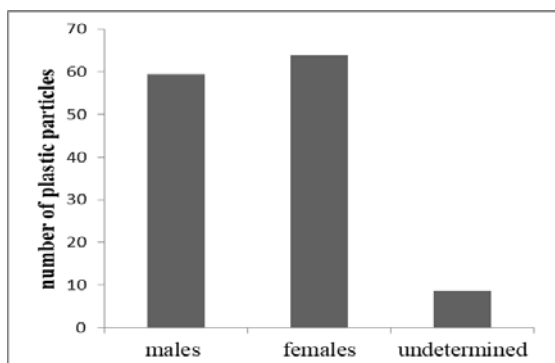


Figure 4. mean number of plastic particles ingested by sex (global examined fish species).

3.5. seasonal variation and plastic ingestion

Contaminated fish specimens by plastic particles were high in the wet season (Figure.5a), with a mean of 26.71 ± 4.2 individuals contain plastic. In the dry season the mean number of contaminated fish specimens was lower (8.71 ± 1.7). A one-way ANOVA test between the two seasons (wet and dry) confirms that the collected plastic debris was significantly higher in wet season than the dry season ($p < 0.01$).

Our results indicate a significant differences in the mean number of ingested plastics among the two seasons (one-way ANOVA; $F=6.99$; $p < 0.05$). The total number of plastic particles ingested by fish in wet season was 2092 particles (418.4 per species), in contrast, the dry season counted only 129 (25.8 per species) (Figure.5b).

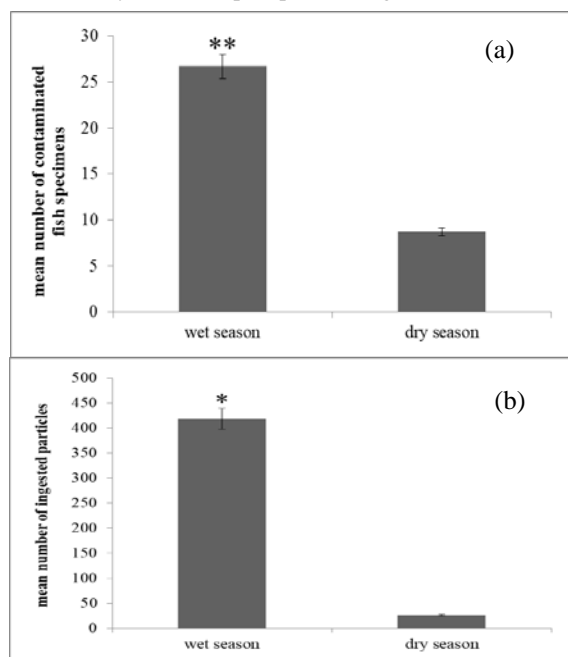


Figure 5 the mean number of contaminated fishes (a) and the mean number of ingested plastic particles (b) in the wet and the dry season. **: $p < 0.01$; *: $p < 0.05$

4. Discussion

In the present study, fishes from three regimes e.g. pelagic, benthopelagic and demersal were gathered to investigate the presence of plastic particles in the gastrointestinal tract. Our results revealed higher abundance of these potential contaminants in all examined

fishes especially in demersal fish species. These results confirm that plastic particles are present throughout the water column, and probably, a part of these plastic particles make complex with other organic or chemical elements (metallic contaminants) enhancing their sedimentation and their availability for benthopelagic and demersal fishes in deeper marine water. Lagarde *et al.* (2016) reported that these plastic particles adsorb heavy metals from the water column, which have a tendency to flow. In this context, a recent study conducted by Akhter and Panhwar (2022) revealed the presence of microplastic materials in crabs and fishes from pelagic and mesopelagic area in Pakistan. The authors stated that microplastics materials are present in pelagic regime than demersal and validate that low weight of plastic particles floats on ocean surfaces.

All examined fish species (*Sardinella aurita*, *Sardina pilchardus*, *Pagellus acarne*, *Trachurus trachurus*, *Boops boops* and *Sparusaurata*) consumed plastic particles. Our results showed that more than a half of the sampled fish specimens contain marine plastics. The recorded frequency of occurrence reaches 58.63%. Our results corroborate with the previous studies were high value were reported. For example, Ferreira *et al.* (2016) reported in *Cynoscion acoupa* from the Goiana Estuary 51%, in the Northwest Atlantic, Wieczorek *et al.* (2018) assessed the presence of microplastics in the stomachs of nearly three out of every four mesopelagic fish. Karbalaei *et al.* (2019) highlighted that 9 of 11 examined fish species from Malaysia (*Megalaspis cordyla*, *Epinephelus coioides*, *Rastrelliger kanagurta*, *Thunnus tonggol*, *Eleutheronema tridactylum*, *Clarias gariepinus*, *Colossoma macropomum*, *Nemipterus bipunctatus* and *Ctenopharyngodon idella*) were contaminated.

Neves *et al.* (2015) measured microplastics in stomach contents of 17 fish species from Portuguese coast (comprising *B.boops*, *P.acarne*, *T.trachurus* and *S.pilchardus*). In this study region, the authors reported relatively low rate (19.8% of 263 specimens) of fish contaminated with plastic particles regarding our results.

In addition, the mean number of plastic items ingested by specimens of *B. boops* was higher (21.03 ± 23.49) than those reported by Nadal *et al.* (2016) in the same fish species around the Balearic Islands (3.75 ± 0.25 microplastic items per fish). Many previous studies (Romeo *et al.*, 2015; Anastasopoulou *et al.*, 2013) have related the rate of marine debris or particles ingestion by fish to the variability of feeding habits.

Only two morphotypes of plastics were recorded (fibers with 85% and fragments with 15%) in the sampled specimens of all examined species. Our results corroborate with those of Neves *et al.* (2015), Ferreira *et al.* (2018), Sathish *et al.* (2020) and Merga *et al.* (2020) who reported clearly, the predominance of fibers as the most common particles ingested by fishes. These results maybe explained by the abundance of fibers in their ecosystems. After Valente *et al.* (2019) fibers are the most common in the environment. Fishing nets and textiles are possible sources of most fibers (Kane and Clare, 2019). They are similar in shape to the fish feed, which promote the ingestion by specimens (Walkinshaw *et al.*, 2020).

Fragments were extracted only from the gastrointestinal tracts of *B. boops*, *S. aurata* and *P. acarne*. *B. boops* which are semipelagic to demersal fishes and *P. acarne* a

benthopelagic species so they contact various plastic morphotypes than pelagic fishes when they are in movement (Sbrana *et al.*, 2020).

From the total of marine debris extracted, 94.58% were microplastics while mesoplastics represent a low percentage, all specimens contaminated ingest microplastics more than mesoplastics, the same results were reported in previous studies (Romeo *et al.*, 2015; Murphy *et al.*, 2017; Jabeen *et al.*, 2017). The obtained results confirm that the abundance of microplastics was higher than mesoplastics in all investigated fish species. Ingestion of small particles is maybe easier by small size of marine organisms (Neves *et al.*, 2015).

Our findings highlight that the blue particles were by far the most frequent plastics in the gastrointestinal tract contents; this is a result that was reported worldwide (Barboza *et al.*, 2019, Merga *et al.*, 2020). Some authors relate that (abundance blue particles) to their availability in the environment (Ferreira *et al.*, 2018) or because fishes mistake them more as they are like food (Barboza *et al.*, 2019). The red particles came second representing 41.3% of the total plastic debris, the source of red fibers is the fishing industry mainly fishing nets (Cole *et al.*, 2011; Lusher *et al.*, 2013; Nelms *et al.*, 2018).

The results show no significant differences in the ingestion of plastic debris between males, females and undetermined specimens ($p > 0.05$). Maybe the availability of these pollutants particles in the ecosystems (pelagic and benthic) offers the same chance for all these categories, although they have different feeding habits.

Contaminated fish specimens by plastic debris in gastrointestinal tract was significantly higher in wet season as compared to dry season ($p < 0.01$). The wet season showed a high contamination (2092 plastic particles) compared to dry season (129 plastic particles). Precipitation and strong winds during the wet season (63.7 mm in our study) stimulate the movement and degradation of plastic debris (Cheung *et al.*, 2016). In addition, rivers are important sources of plastic contamination (Rowley *et al.*, 2020; Xu *et al.*, 2020), many microplastics and mesoplastics are maybe introduced to the sea by many rivers connected to the gulf of Bejaia. This high input of marine debris into the coastal waters depends on the seasonal activity making therefore plastic particles available to fish species then to consumers of fish.

5. Conclusion

This study shows high ingestion of plastic particles by commercial fishes from the gulf of Bejaia (Algeria). Extracted plastic debris were analyzed for the first time in the gastrointestinal tract of examined fish species. These pollutants exhibit a variability in colors, morphotypes and size, their abundance is higher in the wet season and in fish species from different ecosystems (pelagic, benthopelagic and demersal), and particularly those having a benthopelagic and demersal behavior. The obtained results maybe give an insight on the state of the marine ecosystem health of Algerian coast (especially in the studied region).

We highly recommend using the digestion process by sex in the future investigations on plastic particles ingested by fish species. We also recommend substantial investigations on the impact of these potential toxic

pollutants on the health of consumers, and on depollution techniques reducing this kind of pollutants in the environment.

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