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Microplastic prevalence in marine fish from onshore Beibu Gulf, South China Sea

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In recent years, microplastics have been widely detected in marine fish and may pose potential risks to fish and human health. Even though microplastic pollution is a critical issue, it represents as yet non-quantified threat for some context in the marine environment. In this study, 271 individuals comprising 32 species of marine fish collected from Beibu Gulf were examined for microplastic prevalence, with an aim to provide data on the abundance, physical classification, and chemical characterization of microplastics in the great bay along South China Sea. The results showed that the occurrence rate of microplastics was 93.7%, and the average abundance of microplastics was 1.02 ± 0.18 items per individual (ranging from 0.03 to 4.00 items per individual). Microplastic accumulation was observed with a great variation in different species, body sizes, living habitats, and feeding habits of marine fishes. The dominant polymers identified by μ -FTIR were polyethylene terephthalate and polypropylene. Fibers, smaller sizes (<1 mm), and transparent color were the major characteristics of the detected microplastics, which might be important factors affecting the bioaccumulation. The present study revealed that microplastics in marine fish is a widespread issue in onshore Beibu Gulf.

KEYWORDS

microplastics, marine fish, contamination, abundance, fibers

Introduction

The durability of plastics which make them particularly useful in the modern world can also be a lethal disaster to marine wildlife (Hammer et al., 2012). Microplastics (<5 mm) derived from the fragments or degradation of plastics is a widespread occurrence in aquatic environments and has become a tremendous concern worldwide (Auta et al., 2017). Microplastics with a high density that exceeds that of seawater (>1.02 g/cm³) probably sink through the water column and accumulate in the sediment, while low-density particles tend to float on the sea surface and in the water column. Given the abundance and the small sizes of microplastics, it is not surprising that microplastics now appear to be a ubiquitous pollutant for various marine organisms in the oceans (Claessens et al., 2011; Rezania et al., 2018).

Microplastics are more likely in the same size range as planktons, and the possibility for uptake by many marine organisms are high (Browne et al., 2008). According to a recent review (Savoca et al., 2021), 386 marine fish species-including 210 commercially important species-have ingested microplastics, and the incidence rate of plastic ingestion has been increasing by 2.4 \pm 0.4% per year during the last decade. Microplastics were frequently found in the gastrointestinal tracts and gills of marine fishes and even detected in the skin, liver, and muscles (Savoca et al., 2021; Ugwu et al., 2021). Once microplastics ingested by organisms end up in the digestive tract, contaminants may desorb from the plastic material and accumulate in the tissue or blood of the organism. It is particularly concerning that microplastics and their associated pollutants (e.g., PCBs and PBDEs) can be accumulated in marine fishes and transferred to other animals at higher trophic levels (Antao Barboza et al., 2018). Accordingly, it is predictable that humans are exposed to microplastics at different levels due to high seafood consumption worldwide (Antao Barboza et al., 2018). Humans occupy a high trophic level in the marine food chain and can potentially be exposed to micro- and nanoplastics. Therefore, microplastic contamination in marine fish needs to be closely monitored for their potential health risks on food safety.

In recent years, microplastic research in China has directed increased attention towards microplastic pollution in aquatic environments. Beibu Gulf is located in the northwestern part of the South China Sea. The total area of Beibu Gulf is approximately 130,000 km² and rich in oil, gas, and biological resources (Gao et al., 2017). It is directed west to Vietnam, and two Chinese provinces, Guangxi and Hainan, lie to the north and east, respectively. It is also rich in fish resources and represents as a traditional fishing ground of China and the China-Indo Peninsula. There are several evidence from recently published articles about the emerging microplastic contamination in water, sediment, and biota from Beibu Gulf, such as in Maowei Sea (Zhu et al., 2019; Zhu et al., 2021), Qin River (Zhang et al., 2020a), fishery areas, and mangrove wetlands

(Li et al., 2019; Xue et al., 2020; Zhang et al., 2020b; Zhang et al., 2021). According to Zhu et al. (2022), the abundances of microplastics in seawater and sediment in Beibu Gulf were 0.67 items/m³ and 4.33 items/kg of dry weight, which give a hint on the possibility of microplastic transportation to fish and other marine organisms. Even though there are few studies regarding microplastic pollution in the sediments and water around Beibu Gulf, there still remains a gap on data on the occurrence of microplastics in fishes from the gulf. Therefore, it is important to have some statistics on the accumulation level of microplastics in fish from Beibu Gulf. Furthermore, Beibu Gulf is a typical area to study the co-influence of social behaviors and the fishery industry on microplastic pollution, and recognizing the status of the pollution and their ecological impacts in research regions would be helpful in taking mitigation measures and policies to reduce microplastics in the oceans.

Materials and methods

In this study, marine fishes were collected by trawling in 30 sampling sites from onshore Beibu Gulf area (21.04-22° N, 108.21-109.45° E) (Figure 1). Trawling was performed within 30 min in each site under the speed of 3 to 4 knots per hour. Fishes were collected and classified, and the body size and weight were measured on board. A list of the measured data with the name of all species is provided in Supplementary Table S1. Then, the fish surfaces were cleaned with ultrapure water and dissected carefully with sterilized tools. The gastrointestinal tracts (GITs, including the stomach and intestine) and gills were extracted for microplastic detection. The collected tissues were digested with 10% KOH solution in glass conical flasks and incubated in an oscillation incubator at 60°C with a rotation speed of 80 rpm for at least 24 h to remove organic matters. After digestion, saturated NaCl solution (1.2 g/ml in density) was added into the flask, stirred using a glass rod, and kept for 2 h to separate the microplastics via density separation. The overlying water was directly filtered with membrane filter (Millipore NY20, pore size 20 µm) using a vacuum pump. The flotation and filtering were repeated several times to reduce the loss of microplastics as much as possible. The filter paper was placed into a cleaned glass petri dish with a cover for further analysis. To avoid contamination, all the liquids, including ultrapure water and NaCl and KOH solutions, were filtered with glass microfiber filters (Whatman GF/F, pore size = $0.7 \,\mu$ m) prior to use. All apparatuses used for microplastic analysis were rinsed three times with filtered water and were immediately covered when not in use. A procedural blank without sample was performed during microplastic extraction and inspection to analyze the airborne contamination. The blanks were in triplicate, and data were corrected for procedural contamination. Microplastic identification was detected under a stereo light microscope (Olympus SZX10, Tokyo, Japan). Images of the suspected plastic items were taken with a digital camera



(Olympus DP80, Tokyo, Japan). A subsample (>40%) of suspected microplastics were selected and detected by FTIR (Nicolet iN 10, Thermo Fisher, USA) coupled with MCT detector to identify the polymer composition. The range of 4,000-400 cm⁻¹ under the infrared spectrum was measured, and 16 acquisitions on each particle were identified. OMNIC software (Thermo Fisher, USA) was used as the reference of standard FTIR spectrum databases for the comparison of the obtained spectra. Microplastics were confirmed based on the matching degree with the standard spectrum at >80%. There were some fibers collected from the blank controls which have been confirmed by µ-FTIR as cotton cellulose, which were not included in the calculation of microplastic data. Statistical analyses such as one-way ANOVA, t-test, or nonparametric tests were carried out using IBM SPSS Statistics (version 20.0) at a significance level of p < 0.05. Furthermore, linear regression analyses were conducted between the number of fish traits (i.e., body size, living and feeding habits) and microplastic abundances of fishes with linear regression models. All fish trait regression models were conducted with pooled and individual species data.

Results

Microplastics were found in 23 out of 32 fish species with an occurrence rate of 93.7% within 254 of the total 271 individuals. Of the accumulated microplastic fish, the microplastic abundance ranged from 0.03 to 4.00 items per individual, with

an average of 1.02 ± 0.18 items per individual (Figure 2A). The highest abundance of microplastics was observed in Sillago sihama (4.00 items per individual), Centrobergx lineatus (2.46 items per individual), and Scatophagus argus (2.00 items per individual). Therefore, the present study confirmed that fish from onshore Beibu Gulf were widely contaminated with microplastics. Microplastic accumulation was observed in both gills and GITs. There was a slight difference of microplastic occurrence rate in GIT (54.4%) and gills (45.6%). However, there was no statistically significant correlation between microplastic abundance and body length (p = 0.287, *t*-test), but a positive relationship (p = 0.000, t-test) was distinguished between the microplastic abundance and the wet weight of fish samples. Demersal fishes contained lower microplastic abundance (0.95 ± 0.17 items per individual) than the pelagic fishes (1.27 \pm 0.46 items per individual), but no significant difference was found (p = 0.59, t-test).

According to the physical characteristics, fibers (98%) were the dominant microplastic shape, followed by fragments (1%) and films (1%) (Figure 2B). Besides the three species, the microplastics collected from 20 other species were all fibers. A small fraction of fragments was found in *Chorinemus* sp. (CH) and *S. sihama*, while fibers, fragments, and films were obtained from *Cathorops steindachneri* (Figure 2B). The dominant size of microplastics was in the range of 0.02–1 mm, accounting for 66% of microplastics (Figure 2C). According to the colors, a comparative variation was found within different fish species. Transparent (27%), black (25%), and blue (24%) colors were the





dominant colors, while red (10%), green (7%), yellow (4%), and white (3%) were less in amount (Figure 2D). Polyethylene terephthalate (PET) showed the highest percentage (32%), and then polypropylene (PP) (21%) was followed as the second dominant polymer. Polyvinyl chloride (PVC) (16%), polyester (PES) (11%), polyacrylic (PAC) (11%), polysulfide (PSU) (5%), and poly methyl propenyl ether (PMPE) (5%) were also detected in the samples.

Discussion

Microplastic abundance

It is well known that microplastics are accumulated in some organs of the fish body. Fish may consume microplastics by mistake since the sizes of microplastics are similar to the food particles. Previous field studies have revealed microplastic ingestion by many commercial fish species from the Yellow Sea (Sun et al., 2019), the Bohai Sea (Wang et al., 2021), the North Sea (Kühn et al., 2020), the East China Sea (Wu et al., 2020), and the North America (Baechler et al., 2020). However, the quantities of microplastics observed in fishes are generally small in amount, in the range <2 particles per individual (Savoca et al., 2021). Zhang et al. (2021) KK found that the microplastic abundance in fish from the mangrove wetland of Beibu Gulf was 0.72–5.39 items per individual, and the count was also high in GITs than in gills. Of the 80 fish studied, only three had no microplastics, and the average quantity was 6.6

items per individual in fish bodies from central and western coastal areas of Guangdong Province (Pan et al., 2021). Moreover, the average abundance of 584 fish individuals was 2.14 items per individual from Bohai Sea, China (Wang et al., 2021). Similarly, in this study, the microplastic abundance ranged from 0.027 to 4.000 items per individual in collected marine fishes. A previous study also confirmed that fish samples captured from offshore Beibu Gulf were contaminated with microplastics (Koongolla et al., 2020). According to that, the microplastic abundance in offshore Beibu Gulf ranged from 0.027 to 1.000 items per individual, with an average of 0.228 ± 0.080 items per individual within 12 fish species. We can predict the reason for the higher abundance of microplastics accumulation in onshore fish as to the direct contamination of land-based microplastics. Due to the wave reactions and current patterns of the seawater, microplastics can be present in high amounts in deep water areas. Therefore, onshore and offshore results may reveal the microplastic pollution variation with respect to the impacts of human activities by the distance from the coastline. A study from Dafeng River revealed that the microplastic pollution level in the water and sediment during the dry season was approximately two to three times higher than that during the rainy season (Liu et al., 2021). Moreover, Liu et al. (2021) found the microplastic pollution levels in the water, sediment, and fish of Dafeng River to decrease in the following order: fish > sediment > surface water in terms of items/kg. Therefore, it is obvious that environmental factors directly influence the microplastic prevalence in fish.

Recently, several studies have been conducted on the ingestion of microplastics by commercially important marine species throughout the world, where microplastics were detected frequently (Savoca et al., 2021; Ugwu et al., 2021). The direct consumption of microplastics and the incidental accumulation by a contaminated prey at lower trophic levels are reasons for the presence of microplastics in the intestine and stomach of fish (Jovanovic et al., 2018). According to Lam et al. (2022), microplastics were detected in cultured fish with an average abundance of 35.36 items per individual. The fish intestine contained more microplastics (23.91 items per individual) than the stomach (12.80 items per individual). Another study from Dafeng River showed the contents of microplastics in the digestive tracts and gills of fish, which ranged from 0.3-6.7 items per individual and 0.1-3.0 items per individual, respectively (Liu et al., 2021). In this study, microplastics were observed in both gills and GITs. The highest abundance of microplastics was found inside the GITs and accounted for 54.4% of the total microplastics, while those in the gills accounted for 45.6%. The percentage of fish with microplastics in the GIT is variably reported in the literatures, such as 65% of 178 individuals of fish from the Red Sea (Baalkhuyur et al., 2018), 58% of 1,337 individuals of fish from the Mediterranean Sea (Guven et al., 2017), 38% of 120 individuals of fish from the Mondego River estuary in Portugal (Bessa et al., 2018), and 19.8% of 263 individuals of fish from Portuguese coastal waters (Neves et al., 2015). According to these reports, microplastic accumulation in fish may be depend on the region and the number of fish samples collected.

Characteristics of microplastics

Based on their geometry, microplastics were classified into the following classes in this study: pellets, fragments, film, and fibers. In this study, 98% of collected microplastics were fibers, 1% fragments, and 1% films, respectively. Fibers can be aggregated in the marine environment due to the fragmentation of fishing nets, and hence there is a high probability for this to be ingested mistakenly by fish. According to the literature survey, the main sources of fibers are generated by human activities, including ship traffic, fisheries, sewage discharges, and wastewater from coastal areas (Cesa et al., 2017; Gago et al., 2018). Importantly, the aggregation of microplastics in the environment can directly influence the possibility of accumulation of microplastics inside fish. Several studies showed higher frequencies of fibers compared with other forms of microplastics within a variety of marine environments (Zhao et al., 2014; Koongolla et al., 2018). However, ingestion of plastic fibers can get tangled and form agglomerates that can potentially block the GITs, resulting in the accumulation of plastic fibers inside the fish body (Neves et al., 2015; Lin et al., 2020).

Moreover, the size variation of microplastics plays an important role, which leads to a high impact on microplastic pollution. According to this study, the dominant size range of the collected microplastics inside fish was between 0.02 and 1 mm, but there was not any relationship with microplastic abundance in benthic and pelagic fishes with reference to the size of microplastics (p = 0.5664). Interestingly, large-sized fishes such as Grammoplites scaber (30 cm) and C. lineatus (40 cm) accumulated only 2- to 3-mm range and 4- to 5-mm range of microplastics, respectively. It may imply that large-sized fishes which have a large mouth gape tend to consume large-sized microplastics, while small fishes ingested small microplastics. This effect was already noticed in 1994 by Shaw and Day, who recognized the preferential removal of smaller-sized particles by marine organisms (Shaw and Day, 1994). Several colors of microplastics were noticed, such as transparent (27%), black, (25%) and blue (24%) as the dominant colors, while red, yellow, green, and white were lesser in amount. The dominant colors (transparent, black, and blue) are hardly visible under the seawater environment. Therefore, it may create a higher possibility for fish ingestion by mistake.

The fingerprint-like molecular composition of polymers with a repeat unit structure allows for a clear assignment of microplastic samples. According to this study, we found PET and PP as the dominant polymers in the collected microplastics of fish from onshore Beibu Gulf. However, we also obtained few other polymer types, such as PVC, PSU, PAC, PES, and PMPE (Figure 3). The polymer composition diversity of microplastics in fish might be derived from different sources of plastic pollution in the marine environments. In fact, most fibers (e.g., PET and PA fibers) reported in this study can be denser than water and are expected to sink and therefore become available to benthic feeders. Pelagic fish are usually visual predators and are more likely to confound particles and prey items (de Sa et al., 2015). However, polyethylene, PP, and PES are the most prevalent in the aquatic environment (Rezania et al., 2018). These materials have also been identified as the most abundant in previous assessments in biota (de Sa et al., 2018). However, it is essential to conduct long-time monitoring projects on microplastic relevant to the associated contaminants in marine organisms. Thus, one-time fish collection only provides a snapshot of microplastic accumulation in fish. Furthermore, the presence of microplastics found in stomachs of several commercially important fish species may present a potential risk to human health due to the transfer of these small plastic items and/or associated contaminants to edible fish tissues (Gallo et al., 2018). Regions where fish consumption is especially high were reported to be contaminated with a large number of microplastics (Antao Barboza et al., 2018). However, these findings reveal significant data for global microplastic pollution with reference to microplastics in marine fish body.

Conclusion

This study was conducted with the objective to reveal the current state of microplastic contamination in commercial fish species from Beibu Gulf with reference to distribution, morphology, and chemical characteristics. Among the collected fish species, the highest abundance of microplastics was observed in S. sihama, C. lineatus, and S. argus. Overall, the average microplastic abundance was 1.02 ± 0.18 items per individual from 23 fish species. According to habitat, a higher microplastic abundance was found in benthic species than that in pelagic species. There is also a positive relationship between microplastic occurrence and the wet weight of 254 fish samples. The microplastic occurrence rate was divided within two organs such as GIT (54.4%) and gills (45.6%). Fibers were the dominant form accounting for more than 98% from each station, and 0.02to 1.00-mm size range was prominent. However, a wide variety of colors could be seen in the collected microplastics, while transparent, black, and blue were common. Based on FTIR results, the majority of microplastics were identified as PET (32%) and PP (21%). These exposed risks need to be assessed through further investigation considering the environmental realistic concentrations of microplastics and the potential transfer of pollutants to human.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding authors.

Author contributions

JK: formal analysis, visualization, writing—original draft, and writing—review and editing. LL: methodology, formal analysis, and visualization. C-PY: investigation and sample analysis. SL: methodology. Y-FP: data curation. X-RX: review and editing and supervision. H-XL: conceptualization and

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writing and review. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/ fmars.2022.964461/full#supplementary-material

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