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Research Article

Microplastic ingestion by *Psettodes erumei* in the northern Oman Sea

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Abstract

Recently, the contamination of microplastics (MPs) has emerged as a globally significant issue, raising concerns across all environmental matrices, particularly in marine environments. This study investigates the ingestion of MPs by *Psettodes erumei*, a commercially important demersal fish in the northern Oman Sea, Iran, with the aim of assessing the presence and characteristics of MPs in gills, guts, and skin. A total of 154 microplastic particles were identified across all tissues examined. These MPs were characterized based on their physical (size, shape, and color) and chemical (polymer types) attributes using a stereomicroscope and Raman spectroscopy respectively. The results showed that fibers were the most prevalent shape and blue and black being the dominant colors. Predominant MPs ranged in size from 50-200 µm. Polyethylene (PE) and polypropylene (PP) were the most common polymers detected. These findings highlight the pervasive nature of MP contamination in marine environments and raise concerns about potential health risks to humans consuming contaminated seafood.

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Introduction

Plastics are manufactured in vast quantities worldwide due to their exceptionally low cost, versatility, durability, and waterproofing properties (Nejat *et al*., 2024). In 2022, global plastic production reached 400.3 million tons (Europe Plastics, 2024), making it the most serious threat to environmental health following climate change (Dick-Vethaak and Martínez-Gómez, 2022). Plastic waste is commonly classified by size into macro (>25 mm), meso (25-5 mm), micro (<5 mm), and nano (<1000 nm) particles (Cozzolino *et al*., 2020). Microplastic pollution refers to solid plastic particles that are ≤5 mm and come in various forms such as pellets, fibers, fragments, films, and granules (John *et al*., 2021). Microplastics (MPs) are categorized into primary and secondary types (Du and Wang, 2021). Primary MPs are produced as plastic pellets (microbeads), fibers, films, granules, and powders used in products like cosmetics, sunscreens, detergents, and pharmaceutical vectors (Carr *et al*., 2016). Secondary MPs are generated by the physical, chemical, and biological degradation of larger plastic pieces through mechanical abrasion, ultraviolet radiation, sunlight degradation, and biological breakdown in the environment (Auta *et al*., 2017). Due to their persistence in the natural environment and potential adverse effects on living organisms through the emission of toxic chemicals, microplastics have gained increased research attention in recent years (Wagner *et al*., 2024) and have been identified as a major issue for ecosystem health. Therefore, the growing accumulation of microplastics in the environment is now considered one of the most significant environmental challenges of these years (Jakobs *et al*., 2023). Microplastic particles are ubiquitous, especially in the marine environment. The marine ecosystem is a vast reservoir of water that covers much of the Earth's surface and supports significant biodiversity through global primary production (Alimba and Faggio, 2019). Improper management of plastic waste has resulted more than 8 million tons of plastic debris entering the ocean annually (Dhiman and Marques, 2016). Meanwhile approximately 60 to 80 percent of all marine debris is composed of plastic waste and in some areas this figure reaches 90 to 95 percent. Given that marine products are a primary food source for humans, there are growing global concerns about microplastic pollution in marine ecosystems (Klingelhöfer *et al*., 2020).

Due to their small size and low density, MPs are accessible to a wide range of organisms, including species widely consumed in the human diet (Barboza *et al*., 2020). Among various marine species, fish consumed as seafood have drawn significant attention regarding MPs. While it is known that trophic transfer of MPs occurs, researchers still do not fully understand the fate of MPs in food webs (Covernton *et al*., 2021). Therefore, it is expected that the accumulation and distribution of MPs in commercially important aquatic organisms will lead to higher exposure risks for the human population, with potential adverse effects over time (Makhdoumi *et al*., 2021). Numerous researchers have studied the accumulation of MPs in various organs of marine fish (e.g., Zakeri *et al*., 2020; Yagi *et al*., 2022; Saemi-Komsari *et al*., 2023; Nejat *et al*., 2024). According to studies, MPs have been found in the digestive systems of 390 fish species (Savoca *et al*., 2021). Information on which types of fish

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ingest MPs and where these MPs accumulate enhances our understanding of MP pollution in fish.

The occurrence and abundance of MPs ingested by fish are believed to be influenced by habitat type (Baalkhuyur *et al*., 2018), trophic transfer (Nikki *et al*., 2021), and feeding strategy (Wootton *et al*., 2021). Therefore, we hypothesized that the abundance and presence of MPs might be influenced by water depth as well as habitat type. Accordingly, in the present study the fish species were selected from the benthic habitat level. Despite numerous studies on various commercial fish (e.g., Parker *et al*., 2020; Nematollahi *et al*., 2021; Valinasab, 2021; Eryasar *et al*., 2022), there is limited information on the occurrence of MPs in commercial fish of the northern Oman Sea. The fisheries and aquaculture sector provides a significant portion of the country's protein and plays an important role in the food chain, fisheries economy, and the consumption of various seafood, particularly among the local coastal residents. It is crucial to ensure the safety of seafood consumption due to the inadvertent introduction of MPs pollutants into the human body can pose serious and potentially irreversible health risks, particularly for high-dose consumers. The aim of the present study was the ingestion of MPs in terms of abundance, morphotypes and polymer types from the Oman Sea in the gills (Gl), gut (GT) and skin (Sk) of an important commercial fish (*Psettodes erumei*) that are consumed by local people. Additionally, it enhances our understanding of the mechanisms contributing to pollution according to demersal habitat.

Materials and methods

Study area

The present study was conducted along the north coastline of the Oman Sea extending from the Chabahar Bay in southeast Iran, Sistan and Baluchestan Province (60° 53' N, 25° 32' E). This bay, the largest on the Oman Sea, spans approximately 20 km by 17 km (Hosseini *et al*., 2020) and encompasses the area from Konarak to Chabahar. This region is at high risk for MPs contamination due to several industrial activities, including shipbuilding, desalination plants, tourism, residential zones, fishery operations, shipping and berths. Also, wastewater discharge from water-balancing vessels, and buoys significantly contributes to MPs pollution. Additionally, municipal effluents and frequent petrochemical releases further degrade these waters. Consequently, this active coastal region has a high potential for pollution (Fig. 1).

Fish species

Psettodes erumei, a commercially important species is the only member of the *Psettodidae* family found in the Persian Gulf and the Oman Sea (Eagderi *et al*., 2019). This demersal marine flatfish resides on sandy and muddy substrates (Ghanbarzadeh *et al*., 2019) at depths ranging from 1 - 100 meters, typically between 20 - 50 meters. During the day, it is usually deeply buried in the substrate but emerges to hunt at night and primarily feeds on small fish (Hensley, 1997). Recently, it was evaluated for the IUCN Red List of Threatened Species in 2019 (Munroe *et al*., 2020).

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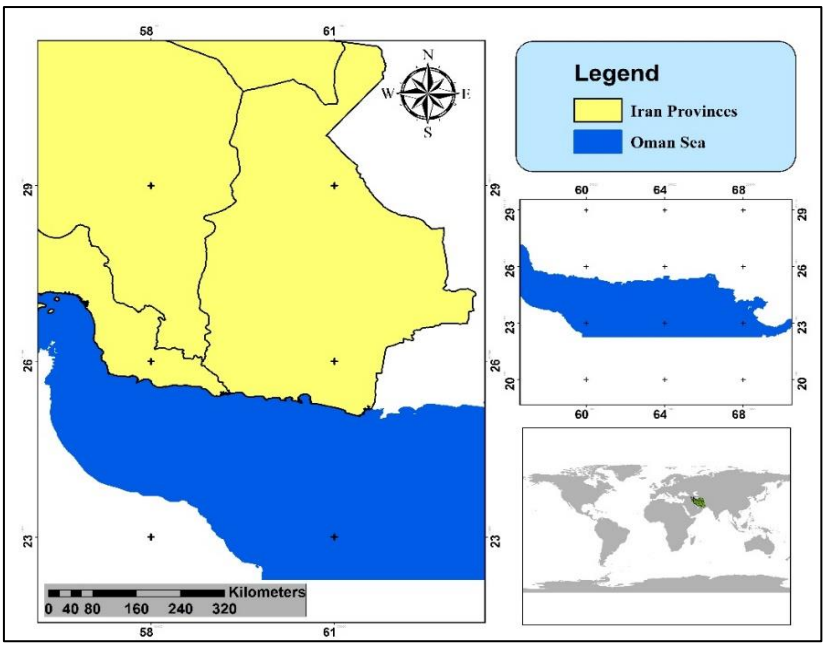

Figure 1: Geographical location of study area in the northern Sea of Oman.

Fish collection

A total of 10 samples of highly consumed demersal commercial fish (*Psettodes erumei*) were directly purchased immediately after being caught by local fishermen in June 2022 at the commercial piers of Konarak. After collection, the fish were wrapped in aluminum foil on ice packs. Then, all collected fish were immediately frozen at -20 ◦C and transported in a travel fridge to the Laboratory of Fisheries at the University of Tehran, Iran, for further analysis.

Sample preparation and treatment

Prior to dissection, the samples were defrosted at room temperature. The fish were measured for their total length (cm) and weight (g). Each fish was then dissected using a scalpel, scissors, and forceps. The gut, gills, and 10 g of skin specimens were extracted, weighed, and placed into labeled pre-cleaned 100-ml glass beakers. To extract MPs from the fish tissues, KOH 10% solution (Merck, Germany) prepared with distilled water, was added in sufficient volume to immerse the specimens and digest the organic tissue. The beakers were covered with aluminum foil to prevent contamination and placed in an oven at 60°C for 48 to 72 hours to ensure complete digestion. After chemical digestion, the beaker contents were filtered through Whatman 42 filter papers (S&S blue band, grade 589/3, and 2 µm pore size) using a vacuum pump and a Buchner funnel. The filters were dried in pre-cleaned and sterilized cabinets. The filters were then stored in clean, covered petri dishes at room temperature until further analysis (Nematollahi *et al*., 2021). These processes were carried out at the Laboratory of Fisheries, Department of Fisheries and Environment, University of Tehran, Iran.

Identification of MPs

Reliable extraction and identification methods for microplastics (MPs) are crucial. These methods include assessing identical length, fiber thickness, color

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homogeneity, lack of organic or cellular structures, and ensuring plastic fibers exhibit three-dimensional bending (Lusher *et al*., 2017). After chemical digestion, all filters obtained from the previous steps were observed, and MPs were counted based on their physical characteristics, including color, shape, and size, under a stereomicroscope (Motic, SMZ-171) with up to 100x magnification at the Faculty of Chemistry and Chemical Technology of the University of Ljubljana (UL FKKT), Slovenia. Additionally, images were taken with a Nikon digital camera mounted on the microscope. Image-processing software "Nikon NIS Elements" was used to measure the maximum length (L) of the particles in µm. Particle size classification was divided into several subgroups: $0 \leq L < 50$ µm, $50 \leq L < 200$ µm, $200 \leq L < 500$ µm, $500 \leq L < 1000$ µm, and $1000 \leq L < 5000$ µm. The color of MP items was classified into seven categories (blue, black, red, brown, white and green). The shape was classified into six types (fiber, fragment, flake, film, and foam). Moreover, to identify the chemical characteristics of the selected MPs, a Raman spectrometer (Renishaw, New Mills Wotton-under-Edge, Gloucestershire, UK) equipped with a 785 nm red diode laser was used at the Laser Lab, Department of Physics and Astronomy, Vrije University Amsterdam, The Netherlands. For detecting the polymer type of MPs, the obtained spectra via Raman were background corrected and normalized using MATLAB (version: R2023b (23.2.0.2365128)) with the 'backcor' function.

Quality control

A series of control measures were implemented to ensure and confirm the accuracy of the data. Initially, all external air inputs, including doors, windows, and cooler channel valves, were closed. To prevent environmental microplastic contamination, laboratory benches and work surfaces were cleaned using pre-filtered ethanol (Merck, Germany). Additionally, no plastic containers or synthetic clothing were used, and cotton lab coats were worn during sample analysis in the lab. The 10% KOH solution was prepared using distilled water and fish skin surfaces were cleaned with pre-filtered ethanol (Merck, Germany). All laboratory glassware was washed with phosphate-free soap, rinsed three times with pre-filtered deionized water, stored in 10% nitric acid for at least one day, then rinsed three times with pre-filtered deionized water, and dried under a fume hood. Furthermore, all reagents and solutions were filtered through S&S filter paper (2 µm pore size grade 589/3 blue band). To assess whether airborne MP particle contamination occurred in the laboratory, an empty control container was placed on the laboratory bench throughout the experiment. The analysis did not confirm any MP contamination in the control dish.

Software and data analysis

Statistical analyses of the data were conducted using GraphPad Prism (version 10.2.3). The normality of the data was assessed using the Shapiro-Wilk test. Since the data distribution was not normal ($p \leq 0.05$), the Kruskal-Wallis test was applied with a confidence level greater than 95%. This test is used to compare more than two

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independent groups and is the non-parametric equivalent of one-way ANOVA. It is worth noting that, given the sample size of 10 per tissues (gill, gut and skin), using non-parametric test is a suitable option because it does not require the assumption of normality and are particularly more reliable for small sample sizes. Graphs were drawn using Excel Version 2016. Furthermore, ArcGIS version 10 was employed to generate the geographical map of the study area.

Result

Abundance and distribution of MPs in the fish gill, gut and skin

To understand the level of contamination, the extent and frequency of microplastics present in fish tissues were analyzed. Totally, 10 fish specimens for each of three tissues (gill, gut, and skin) were examined throughout the study. A total of 154 particles were detected in all desired tissues. Specifically, 61 particles were found in the gill, 55 particles in the gut, and 38 particles on the skin (Fig. 2). Since the gut and skin data did not pass the normality test, a non-parametric Kruskal-Wallis test was applied to assess significant differences in the frequency of microplastic (MP) items across the gills, gut, and skin. The results indicated no statistically significant differences between the mean of the three groups (p=0.2522, Kruskal-Wallis statistic = 2.755). A total of 30 values across three groups were analyzed, and no significant variation was found between the tissues at a significance level of (p>0.05).

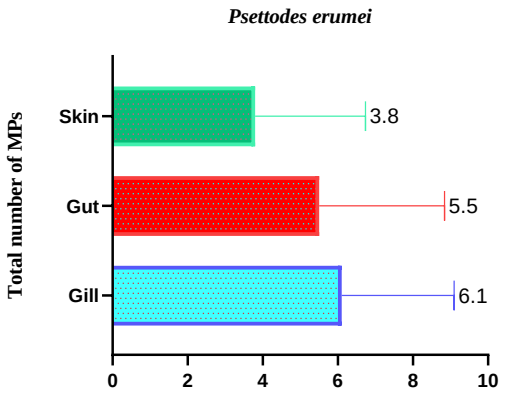

Figure 2: Comparison of MPs abundance and distribution in the Gill, Gut and Skin of *Psettodes erumei.*

Physical characteristics of MPs

The main physical characteristics of MPs are size, shape, and color. So, Particle types in terms of shape, color and size in three examined organs (gill, gut and skin) of *Psettodes erumei* are shown in Figure 3. With respect to the morphology, MPs were categorized into fibers, fragments, foams, films and flakes. The abundance of MPs in all desired tissues followed the order of (82%) fiber>(8%) fragment>(6%) flake>(3%) foam>(1%) film (Fig. 3a). The observed colors in various tissues were blue, black, red, brown, white and green respectively (Fig. 3b). Particles size ranges were into 5 classified groups followed by 0-50> 50–200> 200–500> 500–1000> 1000–5000 µm in all tissues (Fig. 3c). Additionally, various shapes of MPs found in the tissues are displayed in Figure 4.

Chemical characteristics of MPs

Raman spectroscopy was used to effectively analyze the polymeric compounds of MPs in the gill, gut and skin of the *Psettodes erumei* (Fig. 5). Of the 39 suspected selected particles extracted from the samples, 30 were verified as MPs.

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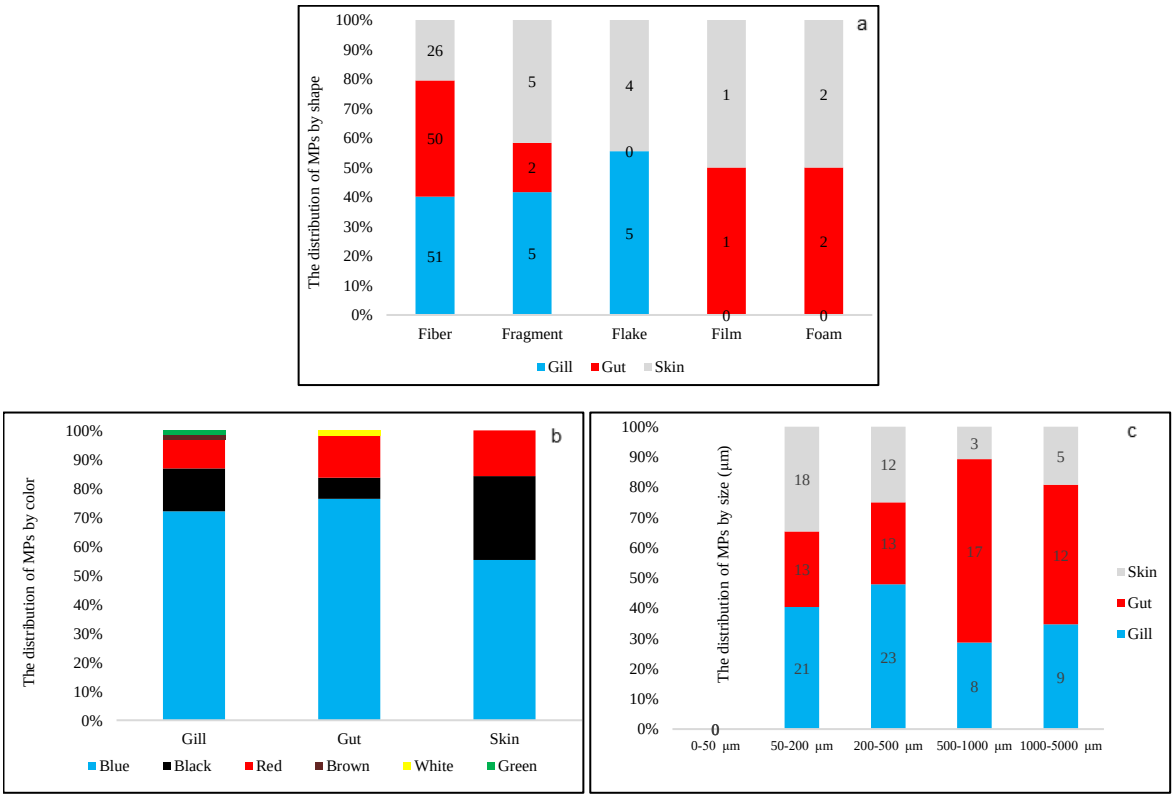

Figure 3: Comparison of MPs based on shape (a), color (b) and size (c) in the Gill, Gut and Skin of *Psettodes erumei.*

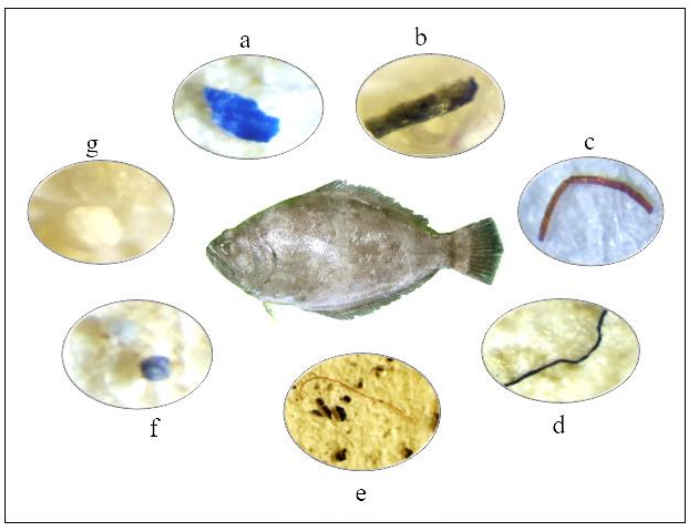

Figure 4: Various shapes of MPs found in the Gill, Gut and Skin of *Psettodes erumei*, (a) fragment, (b) film, (c-e) fibers, (f-g) foam.

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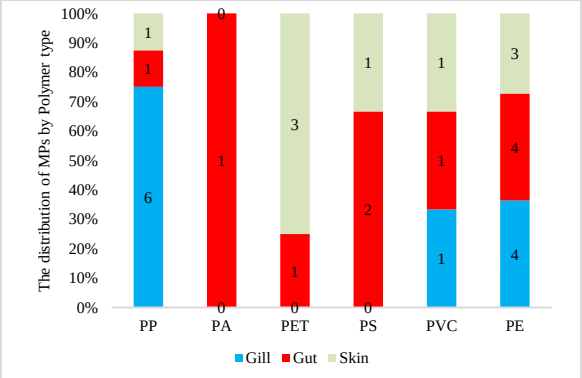

Figure 5: Comparison of MPs based on polymer type in the Gill, Gut and Skin of *Psettodes erumei.*

The detected polymers included polystyrene (PS), polypropylene (PP), and polyethylene terephthalate (PET), Polyethylene (PE), Polyamide (PA) and Polyvinylchloride (PVC). The most dominant polymer was PE (36.67%) followed the order of PP (26.67%) > PET (13.33%) > PS (10%) > PVC (10%)> PA (3.33%).

Discussion

This study provides the microplastic assessment of a demersal commercial fish species of *Psettodes erumei* in the northern Oman Sea first time. The number of microplastics (MPs) in the examined tissues varied due to their functional roles. The results revealed that all 10 fish specimens contained microplastics, consistent with Abbasi *et al*. (2023), who reported a 100% recovery of MPs in all fish samples from this region. Also, Ghosh *et al*. (2021) investigated MPs pollution in some commercial fish from the Bay of Bengal that reported MPs were found in all species. On the other side, a study by Karbalaei *et al*. (2019) reported that 9 out of 11 fish species in Seri Kembangan, Malaysia contained MPs.

In the present study, a total of 154 MPs were detected across all tissue (gill, gut and skin) samples highlighting the pervasive nature of MP contamination in marine environments. Several studies have reported MPs contamination in different fish tissues (Abbasi *et al*., 2018; Barboza *et al*., 2020; Rasta *et al*., 2023). Although, the highest number of microplastics was recorded in the gills, with 61 particles (39%). This can be attributed to the gills' primary function in respiration, where water is filtered, potentially trapping MPs more effectively than other tissues. Then fish gut contained 55 particles (36%), reflecting the ingestion of MPs through feeding activities. The presence of MPs in the gut suggests these particles are ingested along with food, contaminated prey, mistaking plastic particles for food and through sediment intake (Barboza *et al*. 2020). Additionally, trophic transfer where smaller species are preyed upon by higher trophic organisms, may contribute to MP presence in the gut (Farrell and Nelson, 2013). In parallel, Jaafar *et al*. (2021) found that Malaysian commercial marine fish had approximately 92% of MPs in the gills and 86% in the digestive system. In contrast, Rasta *et al*. (2023) investigated fish species

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from the Caspian Sea in Iran and recorded 63 MPs in the gills and 91 MPs in the gastrointestinal tract (GIT) of the analyzed samples. In comparison, the skin showed the lowest number of MPs than gill and gut with 38 particles (25%). This could be due to the skin's direct exposure to marine water, indicating significant contamination through environmental contact. A comprehensive study by Abbasi *et al*. (2018) also analyzed the skin alongside other targeted tissues (gut, muscle, gills and liver), highlighting the varied distribution of MPs across different fish tissues.

As there is limited information on the incidence of MPs in commercial fishes in the Oman Sea, especially in the deep-sea area, we hypothesized that the occurrence of MPs may be affected by water depth as well as habitat type. We assumed that fishes residing in demersal habitats have a higher ingestion rate of MPs. This hypothesis is supported by studies such as Yagi *et al*. (2022) which examined five commercially important demersal species (n = 235) in habitats near Japan (the East China Sea). Moreover, other studies (e.g., Jabeen *et al*., 2017; Gosh *et al*., 2021) are in line with this finding which highlighted that these demersal species exhibited a higher ingestion rate of MPs compared to pelagic species. Although some studies illustrated differences in the distribution of MPs between demersal, neritic and pelagic fish species in some marine environments (e.g., Adika *et al*., 2020; Rasta *et al*., 2023). The MPs found in the examined tissues varied in shape, color, and size. With respect to morphology, fibers were the most prevalent shape similar the finding (e.g., Wang *et al*., 2021; Piyawardhana *et al*., 2022). The predominance of synthetic fibers in the marine environment is attributable to their widespread origins, primarily from textiles (Napper *et al*., 2016), sewage, and river runoff. Additionally, intense fishing activities in the northern Oman Sea, along with the disposal of fishing gear and ropes, may also serve as significant sources of these fibers (Bessa *et al*., 2018).

Based on observation, six different colors of MPs were identified in all analyzed tissues. The color variation indicates the diverse sources of MPs, originating from different consumer products and industrial processes. Consistent with a study by De-La-Torre *et al*. (2019), the most dominant colors in different tissues were blue and black. Among the five categorized MPs size, the most common size range was 50–200μm. The dominance of smaller particles suggests that MPs are subject to fragmentation and degradation in the environment, leading to a higher prevalence of smaller particles. Moreover, some studies have indicated that plastic particles smaller than 150 μm can be absorbed into fish internal tissues (Barboza *et al*., 2020). It is worth mentioning that the detection of particles smaller than 200 μm in this study heightens concerns about the potential long-term accumulation of microplastic pollution in fish.

PE and PP were the most common types of microplastics identified in our study, corroborating the findings reported by Abbasi *et al*. (2023) for the same area. This distribution reflects the widespread use of these polymers in various products and their entry into the marine environment. PP and PE, being widely used in packaging and

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consumer goods, are particularly pervasive (Plastics Europe, 2021). The prevalence of these polymers in fish tissues underscores the extensive pollution of marine ecosystems by plastic materials. Additionally, single-use plastics, primarily polypropylene (PP), along with rope and fishing net materials, are the most abundant types of microplastics found in the marine environment (Nelms *et al*., 2019).

It is suspected that these microplastics originate from the weathering and degradation of plastics discarded by local populations, tourism and industrial activities, fishing operations and marine transport on the northern Oman Sea. The presence of microplastics in fish tissues poses significant health risks to marine life and raises concerns about their adverse toxicological effects (de Bruin *et al*., 2022). Additionally, the bioaccumulation and biomagnification of MPs through the food chain have implications for human health (Alfonso *et al*., 2024). Hence, this study provides a comprehensive analysis of the abundance, physical characteristics, and chemical composition of MPs in the gill, gut, and skin of *Psettodes erumei*. The distribution patterns of microplastics in the various tissues did not significantly differ from one another, but the results indicated a high degree of pollution in each of them.

According to the finding of this study, offers valuable insights into the sources and pathways of MP pollution in the Oman Sea. These results highlight the urgent need for measures to reduce plastic pollution and mitigate its impact on marine life and ecosystems. This can be achieved through policy interventions aimed at restricting plastic production and consumption, advancements in waste management technologies, public awareness initiatives promoting sustainable lifestyles and ongoing research.

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Conflicts of interest

The authors declare no conflict of interest.

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