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# Occurrence and spatial distribution of microplastics in sediment and fish along the Persian Gulf—a case study: Bushehr Province, Iran

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**Abstract** Microplastics (MPs) contamination in the marine environment is a global threat. The present study is the first to comprehensively investigate the MPs contamination in the marine environment in Bushehr province along the Persian Gulf. For this purpose, 16 stations were selected along the coast and 10 fish samples were collected. The results obtained from MPs in sediment samples indicate the mean abundance of MPs in different sediment samples was 57.19 Particles/Kg. The dominant MPs color in sediment samples was black, accounting for 47.54%, followed by white (36.07%). As for MPs in fish, the highest MPs digested in different fish samples were

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Department of Environmental Health Engineering, Faculty of Health and Nutrition, Bushehr University of Medical Sciences, Bushehr, Iran 9. In addition, over 83.3% of MPs observed in fishes were black followed by red and blue (6.67%). Overall, the presence of MPs in fish and sediment can be attributed to improper disposal of industrial effluents; an efficient measurement is required in order to improve the quality of the marine environment.

**Keywords** Microplastic · Marine pollution · Coastal sediment · *Platycephidae indicus* 

# Introduction

Microplastics (MPs) are extensively known as a major threat to marine life (Hernandez-Gonzalez et al., 2018; Phuong et al., 2016). The ubiquitous presence of MPs due to the increasing demands of plastics all over the world exacerbates this issue; it is considered a hot topic for aquatic media (Lithner et al., 2011; Thiel et al., 2018). The global production of plastics has experienced significant incremental trends over the past 50 years; it raised from 1.7 to 335 million tones between 1950 and 2016 (Karbalaei et al., 2018). The mismanagement of plastic waste leads to ending up this hazardous waste with xenobiotic substances to marine, threatening the aquatic organisms (Bagheri et al., 2020). It is estimated that annually approximately 28 million tons of plastic debris could find their way to water bodies (Onoja et al., 2022). The effluents from the terrestrial and marine-based activities including the local industrial drainage, agricultural runoff, band wastewater treatment plants have the most contribution to marine pollution (Karbalaei et al., 2018). When the plastics enter the marine environment, they are exposed to physical fragmentation (e.g., physical forces, UV-radiation, and chemical and biological processes; the plastics are converted to different shapes and specific densities with diameters less than 5 mm Chinfak et al., 2021; Cincinelli et al., 2021). Depending on their densities, floating properties, and size, the MPs ending upon the oceans can either be suspended on the water column or deposited on the sediment following sea tidal (Chinfak et al., 2021; Thiel et al., 2018). MPs due to their inherent composition and large surface volume ratio can act as dispersal vectors of chemical additives and organic and toxic materials present in ocean media; they are considered as a "cocktail of contaminants," For instance, Wang and Wang (2018) reported the higher capacity of phenanthrene on MPs in sediment samples (Wang & Wang, 2018). Being the same size as planktonic organisms and high bioavailability, the ingestion of MPs is widely reported in the evidence focused on the marine environment (Cole et al., 2011; Suaria et al., 2016). Ingestion of the MPs by the fishes can introduce toxic materials into the bodies and subsequently the human food chain (Abbasi, 2021; Akhbarizadeh et al., 2018). Therefore, the presence of MPs in fishes' body can be considered as marine pollution bioindicators and the presence of hazardous substances in the marine environment. In addition, the MPs can introduce both direct (mistakenly consumed as prey) and indirect (if fishes consumed the contaminated prey) into the cells and tissues of the fish body (Baalkhuyur et al., 2018; Horton et al., 2017) Furthermore, the presence of MPs has been widely reported in fish bodies, especially the muscles which are highly edible and of concern for the human body (Hernandez-Gonzalez et al., 2018; Lusher et al., 2013; Vandermeersch et al., 2015). Moreover, tidal currents can transport the MPs in a coastal area; the density of MPs increases due to attachments of their surfaces with clay mineral and biota and leads to sinking in the sediments (Chinfak et al., 2021; Tien et al., 2020). Here we present the comprehensive results of a large-scale survey of MPs in muscle fishes and sediment on the different coastline of the Persian Gulf in Bushehr province.

The additional aims of the present study were to determine the geographical distribution of MPs and extensive characterization of polymer composition in both sediment and fish samples.

# Materials and methods

# Study area

The present study was performed along the coastline of the Persian Gulf and different cities in Bushehr Province. This area with tropical and subtropical climate and average annual rainfall of 155 mm is located between 50°06' to 52°58'E longitude and 27°14' to 30°16'N latitude in the south of Iran. This area is one of the most rapidly growing industrial provinces in Iran; Asalouye is globally known as the capital of energy in Iran. Rapid development in industrialization and urbanization have caused severe water pollution due to the discharge of direct untreated wastewater from several industrial and petrochemical companies into the Persian Gulf. In this area, a variety of fish species are observed in the sea which is the main source of meat with beneficial advantages for people living in the vicinity. Therefore, proper preservation from the effluents discharged from different anthropogenic activities should be considered in priority by the authorities. The sediment and fish samples were withdrawn to investigate the MPs pollution in the marine environment in Bushehr province, facing different industrial and domestic pollution. Sampling was carried out in November 2020 and a total of 16 sampling points were selected for sediment collection, as illustrated in Fig. 1.

# Sediment sampling and microplastic extraction

In this study, 16 sediment samples throughout Bushehr province were taken based on potential contamination points. The location of different sampling points is illustrated in Fig. 1. At 16 sampling points, triplicate sediment samples were collected using a five-point sampling approach with a length of 10 m. Briefly, at each sampling point, five sediment samples were taken from corners and the center of the square with a depth of 5 cm. In general, 3 kg of sediment core subsamples representative of the depth of 0–5 cm collected from each quadrant location were immediately placed in a glass **Fig.1** The map of the study area, Bushehr province along the Persian Gulf coastline



container pre-washed with n-hexane and transported to the laboratory. After thorough mixing, 1 kg sediment samples from each sampling point were weighed and placed in aluminum containers to dry at 60° C for 48 h in the oven, then passed through a sieve with pore size 2 mm. Then 200 g well-mixed sediment sample was weight and transferred to a 1-L beaker with 200 mL hydrogen peroxide (30%) to remove the organic matters present in the sediment (Masura et al., 2015). It is important to note that blanks were used to prevent the error in the sampling process. The samples were placed on the incubator shaker and agitated for 2 days. After completely shaking the sediment samples and disappearance the bubbles, the samples were placed in an oven at 70 °C to dry. Then 300 mL salt-water containing NaI with a density of 1.6 mg/cm3 was added to the sediment samples to separate the microplastic based on density separation theory (Lots et al., 2017; Patchaiyappan et al., 2020). The resulting solutions were agitated gently for 5 min. The samples were then allowed to settle for 24 h. The floating part of the solution was then transferred on filter paper with the size of 2 µm (5893, S&S) and filtered in a vacuum with pressure. The filter papers were dried 24 h at 50 °C and the MPs were characterized in terms of abundance, color, shape, and polymer composition.

### MPs identification

#### Microscopic observation

The characteristics of MPs were identified using a binocular microscope (HUND (Wetzlar) Stereo microscope with a magnification of  $\times 200$ ) in terms of count and color (Hidalgo-Ruz et al., 2012). The size range of plastics observed in the sediment samples was lower than 2 mm. The abundance of MPs in sediment samples was expressed based on the number of MPs per dry weight of sediment (n/kg).

### Microplastic identification and analysis

The surface morphology and polymeric properties of microplastics were characterized with scanning electron microscopy (SEM) and Raman micro-spectroscopy, respectively. The surface structure and morphology of microplastics were analyzed using scanning electron microscopy (SEM) with 25 kW (model: TESCAN, MiRA3). It is important to note that the samples were coated with gold to avoid charging during the analysis. In addition, the chemical compositional analysis of microplastic particles was characterized with Confocal Raman Spectrophotometry (Lab Ram HR, Horiba, Japan). The types of polymers present in the microplastic particles were matched with the reference spectra from the library.

Fish sampling and MPs separation from its gut

This fish samples with triplicate were taken from 2 local markets in each 5 counties (totally 10 local markets), commercial fishes (*Platycephidae Indicus*) with average size and weight of 38–42 cm and 368–504 g, respectively. After samples collection, fishes were kept in cold boxes and immediately transferred to the laboratory. The fishes were washed with

distilled water and prepared for removing their guts. The extraction of microplastics from fish's gut was performed using the improved method of (Karami et al., 2017). In order for digestion, the fish's gut sample was removed and placed in a 100-mL Erlenmeyer flask containing 20-50 mL of 10% KOH. The content was digested in the Erlenmeyer in an oven at 40 °C for 48 to 72 h. It is important to note that the samples not fully digested were again added to the samples with 10% potassium hydroxide and placed in an oven at 40 °C. After the digestion process is completed, the digested samples were diluted with warm water in order to preserve the integrity of microplastic. Then the samples were filtered with S&S filter paper with apertures smaller than  $2 \mu m$ , the filter paper was washed with distilled water to remove the remaining KOH on its surface. Finally, the filter paper containing the microplastic was dried at room temperature. In order to prevent external contamination of the samples, the surface of the samples was covered with paper at all stages of separation.

# Data analysis

All statistical analyses were performed using SPSS (v.22). Prior to statistical analysis, all data were analyzed for normality and homogeneity of variance. The normality of data was tested and found to be non-normal; the statistical analysis was performed based on nonparametric analysis. One Way analysis of variance (ANOVA), Tukey and Dunnett tests were performed to compare several groups with the control sample.

#### Quality assurance and quality control

During the different sampling steps and experiments, the blanks were considered to prevent the error in all process; no MPs were found in the blanks. In addition, all the surfaces and the equipment were cleaned with ethanol and distilled water before the use, tinfoil paper was used to avoid the contamination of laboratory containers during the experiments due to probable airborne and artificial plastic pollution.

In order to determine the recovery of microplastics, the commercial polyethylene and polypropylene plastic products were ground to obtain MPs particles in range of 200  $\mu$ m–2 mm. Then, the products were mixed with field-cleaned sands the mixture was used for flotation method. The recovery rate for the present study was obtained to be 95%.

# **Results and discussion**

Abundance, color, shape composition and spatial distribution of MPs in sediment samples

A total of 16 sediment samples were collected from different coaches of Bushehr Province (See Fig. 1). MPs with diameters lower than 2 mm were found in all sediment samples. Table 1 provides the abundance of MPs in different sampling points. As summarized in Table 1, the mean abundance of MPs in different sediment samples was found to be 57.19 Particles/ Kg; the highest abundance was observed in S3, while the lowest abundance was observed in S7. The spatial distribution of MPs concentration in different sediment sampling points is illustrated in Fig. 2. The presence of the highest concentration of MPs in the north and northwestern Bushehr can be attributed to the high urbanization and industrialization of this area. Bushehr city is the capital of Bushehr province with a higher density of population compared to other areas. Bushehr is an ancient city in the south of Iran; annually, a numerous amount of tourists visit this city. Another most possible and major pathway of the entrance of MPS into beach sediments is attributed to roads near the beach and the city storm water runoff (Tiwari et al., 2019; Veerasingam et al., 2016). A comparative investigation on the occurrence of MPs throughout the world is presented in Table 2. As shown in Table 2, the abundance of MPs in Bushehr province sediment samples is lower compared to other beaches all over the world.

The color distribution of MPs identified from all sediment samples is summarized in Table 1 and Fig. 3. According to Table 1, among 915 MPs observed in sediment samples, the dominant MPs color in sediment samples was black, accounting for 47.54%, followed by white (36.07%), blue (12.57%), red (3.28%), and brown (0.55%). The dominance of black MPs is also reported in sediment samples in wetlands located in Sweden and Australia (Abbasi, 2021; Ziajahromi et al., 2020). Generally, the presence of a wide range of colors for MPs is attributed to various anthropogenic and industrial activities in the area. The direct entrance of municipal and industrial

Table 1 The total number (Particles/Kg) and characteristics of MPs in different sediment sampling sites (Zone UTM 39 N)

No	County	Longitude (UTM)	Latitude (UTM)	Total MPs	Color			Shape			
					White	Brown	Black	Blue	Red	Fiber	Fragment
<b>S</b> 1	Bushehr	483,394	3,207,447	65	30	5	15	15	0	55	10
S2	Bushehr	482,954	3,205,084	80	25	0	40	15	0	75	5
<b>S</b> 3	Bushehr	481,676	3,198,638	85	35	0	45	5	0	80	5
<b>S</b> 4	Bushehr	483,458	3,196,780	65	30	0	30	5	0	65	0
S5	Bushehr	485,978	3,192,002	60	20	0	30	10	0	55	5
S6	Tangestan	503,433	3,183,272	40	20	0	15	5	0	40	0
<b>S</b> 7	Tangestan	507,368	3,168,988	30	10	0	15	5	0	30	0
<b>S</b> 8	Tangestan	513,829	3,142,984	35	10	0	20	5	0	35	0
S9	Dayer	536,747	3,098,039	45	10	0	20	10	5	45	0
S10	Dayer	578,578	3,079,802	50	20	0	25	0	5	50	0
S11	Dayer	593,664	3,079,502	60	20	0	25	15	0	45	15
S12	Kangan	607,037	3,075,887	55	15	0	30	0	10	55	0
S13	Kangan	634,975	3,059,459	65	20	0	35	5	5	65	0
S14	Kangan	645,511	3,056,394	60	20	0	25	10	5	50	10
S15	Asalouyeh	656,357	3,041,937	55	20	0	30	5	0	55	0
S16	Asalouyeh	665,697	3,038,063	65	25	0	35	5	0	55	10



Fig. 2 The spatial distribution of MPs in different sediment samples

including petrochemical effluents and fishing activities into the marine environment is the most possible reason for the presence of a spectrum of color for MP in sediment samples (Manbohi et al., 2021; Rasta et al., 2020).

The shape composition of MPs in sediment samples withdrawn from different coast areas is summarized in Table 1. Generally, the identified MPs were classified in two categories: Fiber and fragment. Out of 915 MPs found in sediment samples, 855 MPs belonging to the fiber category accounted for 93.44% of all MPs, while 6.56% of MPs had the fragment shape. Figure 4 shows the fiber and fragment MPs identified in sediment samples. In line with other similar studies focused on MPs in sediment samples (Abbasi, 2021; Choong et al., 2021; Ghayebzadeh et al., 2020; Mataji et al., 2020; Mehdinia et al., 2020; Zakeri et al., 2020), in the present study, the fiber MPs dominate in all sediment samples. The shapes of MPs can provide valuable information on the origin of MPs and the industries and anthropogenic activities producing the MPs (Kowalski et al., 2016). The most probable sources of fiber MPs in marine environments and sediment are fishing lines, nets, rope, and fabrics (Sang et al., 2021). In addition, fibers released from clothes washing can introduce the marine environment through municipal sewage and runoff (Zhao et al., 2015). The high abundance of fibrous MPs in the sediment samples in Bushehr province is attributed to improper discharge of municipal and industrial wastewater into marine environments.

Table 2         A Comparative
study of MPs abundance in
various beach sediments

References	Country	MPs abundance (n/Kg)	Size range
Sathish et al. (2019)	Tamil Nadu, India	$309 \pm 184$	0.3–4.75 mm
Tiwari et al. (2019)	Girgaon Mumbai, India	$220 \pm 50$	36 µm–5 mm
Patchaiyappan et al. (2020)	Andaman, India	$220 \pm 50$	0.3–5 mm
Vianello et al. (2013)	Venice, Italy	672—2175	<1 mm
Yu et al. (2016)	Bijianshan, China	$102.9 \pm 39.9$	NA
Qiu et al. (2015)	Beibu Gulf, China	5020—8720	<5 mm
Yabanlı et al. (2019)	Datça Peninsula, Turkey	$1154.4 \pm 700.3$	<5 mm
Wang et al. (2018)	Eastern coastline, China	$32,947 \pm 15,342$	< 300 µm
Abbasi (2021)	Hashilan, Iran	3–8	2 mm
This study	Iran	45-80	<2 mm



Fig. 3 The colors of MPs observed in sediment samples

Raman spectroscopy analysis

In the present study, epifluorescence microscopy and DXR Raman microscope, as an efficient and well-established technique was used to identify the polymer composition of MPs in different sediment samples. The identification of the polymer composition provides valuable information on the origin and sources of MPs (Yaranal et al., 2021). To this end, the polymer composition of MPs in 16 different sediment samples throughout the Bushehr province was examined with Confocal Raman Spectrophotometry (Lab Ram HR, Horiba, Japan). Table 3 provides the information on the polymer composition of MPs in different sediment samples. As summarized in Table 3, Polypropylene (PP), polystyrene (PS), polyethylene terephthalate (PET), Nylon are the major polymers in the structures of MPs in different sediment samples. In addition, the comparison of measured Raman spectra with Raman spectral library showed that 72.22%





 Table 3
 The polymer composition observed in MPs sediment samples

Sampling no	PP	PS	PET	Nylon
1			*	
2			*	
3		*		
4				*
5			*	
6		*		
7			*	*
8	*		*	
9			*	
10			*	
11			*	
12			*	
13			*	
14			*	
15			*	
16			*	

Polypropylene (PP), polystyrene (PS), and polyethylene-tere-phthalate (PET)

of MPs match with PET, followed by PS (11.11%), Nylon (11.11%), and PP (5.56%). In addition, the results of Raman spectroscopy of the various MPs are shown in Fig. 5. The dominance of PET in MPs samples withdrawn from sediment and coach line is reported in sediment samples in the Caspian sea in Iran (Abbasi, 2021) and Ireland (Marques Mendes et al., 2021). As this study site is located in close vicinity to one of the most urbanized and industrialized provinces in Iran, the disposal of plastic particles through discharge of industrial wastewater and fishing activities is the most possible source of PET in sediment samples.

#### MPs in fishes (abundance, color, shape)

A total of 10 local fish samples from five different cities along the Persian Gulf were taken to evaluate the number of MPs, their shape, and color in fish gutes. Table 4 summarizes the detailed information on MPs in fish samples. Generally, the MPs with diameters lower than 2 mm were observed in all local fish samples. According to Table 4, a total of 60 MPs were observed in 10 different local fish samples taken from seas near five cities of Bushehr province. The MPs abundance in different fish samples was not homogenous; the highest (9) and lowest (2) numbers of MPs ingested by the fish belonged to fish samples taken from seas in Bushehr and Tangestan, respectively. The establishment of more industries and visitors in Bushehr is the most possible reason for the discharge of different MPs to the marine environment and finally, more MPs digested by the Fish. Dongdong Zahang et al. (2019) surveyed the presence of MPs in Fish in Shengsi, China, and reported that the microplastics items observed in the fish sample were in the range of  $2.3 \pm 1.5 - 7.3 \pm 3.5$  items which are inconsistent

with the results obtained from the present research

(Zhang et al., 2020).

The color distribution of MPs indigested by the fishes samples is summarized in Table 4. According to Table 4, over 83.33% of MPs observed in fishes were black followed by red and blue (6.67%), and white (3.33%). It is important to note that none of the MPs indigested by the fish was in brown. In a similar study, Abbasi et al. (2018) reported that MPs in black color (71%) were the predominant MPs in fish muscles sampled from Musa Estuary, Persian Gulf (Abbasi et al., 2018). In addition, Lusher et al. (2013) found black particles as most prevalent in stomachs of fish samples from the English Channel (Lusher et al., 2013). However, Zhang et al. (2020) found that blue MPs are the predominant MPs in fishes in artificial reefs around the Ma'an Archipelago, Shengsi, China (Zhang et al., 2020). Furthermore, black particles were most prevalent in the stomachs of fish from the English Channel (Lusher et al., 2013).

Table 4 summarizes the shape composition of MPs observed in the fish gut samples. As seen in Table 4, Fiber MPs composite 96.67% of all observed MPs in fish samples, while only 3.33% was attributed to Fragment MPs. Figure 6 shows the fiber and fragment MPS identified in fish guts samples. As mentioned earlier, the most possible reason for the entrance of fiber MPs into the marine environment is the improper discharge of industrial and municipal effluents. The dominance of Fiber MPs in fish gutes is reported in other similar studies. For instance, Zhang (2020) reported that fiber MPs contain 91.1% of total MPs in fish muscles (Zhang et al., 2020). In addition, Keshavarzifard et al. (2021) found that 78.6% of MPs observed in white shrimp (*Metapenaeus affinis*) samples from the northwest Persian Gulf are fibrous (Keshavarzifard et al., 2021).



Fig.5 Raman spectra of different MPs in sediment samples: Nylon a, PET b, PP c, PS d

The polymer composition of MPs in fish

In the present study, the polymer composition of MPs in fish samples gutes was analyzed using DXR Raman microscope. Figure 7 provides the information on the polymer composition of MPs in different fish gutes samples. Generally, Polypropylene (PP), polyethylene terephthalate (PET), and Nylon were the major polymers in the structures of MPs in ten

different fish samples. Of the ten fish samples analyzed in the present study, the polymers including PET, PP, and Nylon were 4,4, and 2, respectively. Due to non-selective feeding habits, the fish ingest the MPs in a mistake (Naidu et al., 2018). Maghsodyan et al. (2020) reported that PS, PP, PET, LDPE, and nylon were the most MPs polymers *Periophthalmus waltoni* fish in mangrove forests in southern Iran (Keshavarzifard et al., 2021).

Table 4 The abundance and characteristics of MPs in different fish samples

No	County	Local dock	Mean MPs	Color					Shape	
				White	Brown	Black	Blue	Red	Fiber	Fragment
<b>S</b> 1	Bushehr	Jofreh	9	1	0	7	1	0	8	1
S2	Bushehr	Reshehr	7	0	0	7	0	0	7	0
<b>S</b> 3	Tangestan	Delvar	2	0	0	2	0	0	2	0
<b>S</b> 4	Tangestan	Karri		0	0	5	0	0	5	0
S5	Dayer	Bord Khun	7	0	0	5	0	2	6	1
<b>S</b> 6	Dayer	Dayer	5	0	0	5	0	0	5	0
<b>S</b> 7	Kangan	Kangan	8	0	0	5	2	1	8	0
<b>S</b> 8	Kangan	Siraf	5	1	0	3	0	1	5	0
S9	Asalouyeh	Nakhel Taghi	7	0	0	6	1	0	7	0
S10	Asalouyeh	Asalouyeh	5	0	0	5	0	0	5	0

# **Fig. 6** Identification of MPs in fish samples: **a** Fiber **b** Fragment



#### Conclusion

This study surveyed the status of MPs in sediment and fish samples in one of the most industrious provinces in Iran. Although the improper discharge of industrial and municipal effluent into the marine environment is extensively observed in this province, the mean number of MPs in sediment was lower than the beaches over the world. However, the highest abundance of MPs was observed in Bushehr city, the capital of Bushehr province, due to more industrial and anthropogenic activities and tourism which contaminant the marine environment. The results on MPs in sediment and fish samples showed that PP, PS, PET, and Nylon were the dominant polymer composition in MPs. Overall, the abundance of MPs in fish and sediment can arise some health effects; therefore, the authorities are recommended to plan promising and effective measurements to improve the quality of the marine environment.



Fig.7 Raman spectra of different MPs in fish samples: PP (c), PET (b), Nylon (c)

Author contributions TT and SEH: conceptualization, methodology, writing-reviewing and editing. MRB: sample preparation and chemical analysis. FA and ARP: writingreviewing, and editing.

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**Data availability** Some or all data models that support the findings of this study are available from the corresponding author upon reasonable request.

#### Declarations

**Conflict of interest** The authors declare no conflicts of interest in place and comply with international, national, and/or institutional standards on research involving Human Participants and/or Animals and Informed Consent.

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