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Microplastic contamination and characteristics spatially vary in the southern Black Sea beach sediment and sea surface water

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ABSTRACT

In this study, the abundance, and characteristics of the microplastics on the southern coast of the Black Sea were assessed. More than 70% of the detected microplastics were smaller than 2.5 mm and mostly consisted of fibers and fragments. The average microplastic abundance in the beach sediment and seawater were 64.06 ± 8.95 particles/kg and 18.68 ± 3.01 particles/m³, respectively. The western coast of the study area (Marmara region) was the most polluted area, and a spatially significant difference was determined in terms of abundance. The composition in the beach sediment (particles/kg) was dominated by styrene acrylonitrile copolymer (SAC) (40.53%), polyethylene terephthalate (PET) (38.75%), and polyethylene (PE) (6.91%), whereas the seawater (particles/m³) was dominated by PET (57.26%), PE (13.52%), and polypropylene PP (11.24%). The results of our study can be a baseline for environmental modeling studies and experimental studies on the marine organisms inhabiting the Black Sea.

1. Introduction

A vast array of synthetic organic polymers, commonly known as plastics, were developed to answer the specialized needs of different industries and consumers. Today, plastics are essential materials in our daily lives. Global production has increased rapidly parallel to the demand, and approximately 7000 million tons of plastics were produced between 1950 and 2018 (Geyer et al., 2017; PlasticsEurope, 2020). Plastics are recyclable materials; however, only 9% is being recycled, on the other hand, 12% is being incinerated and the remainders end up in landfills or the natural environment (Gever et al., 2017). Due to its durability, it is more likely to accumulate in the environment. Seas and oceans are sinks of plastic waste generated from different sources (Li et al., 2016). Among these, microplastics (MPs) are a form of plastic defined as particles less than 5 mm in size (Van Cauwenberghe et al., 2015b). MPs can be produced in microscopic size to be used in personal care products, detergents, etc. (Napper et al., 2015) or originate from the fragmentation of macro plastic items in the marine environment by environmental factors (Zhang et al., 2021). Plastic particles in the marine environment have been an object of research since the 1970s (Carpenter and Smith, 1972; Colton et al., 1974; Morris and Hamilton,

1974; Wong et al., 1974) and became a major area of interest within the field of marine pollution. In recent years, there has been an increasing interest in the spatiotemporal distribution of plastics and their impact on marine fauna and flora (Ozturk and Altinok, 2020). As a result of increased research efforts, the magnitude of the problem was revealed. Today, to the best of our knowledge, MPs can be found in all parts of the marine environment, including the poles (Lusher et al., 2015), remote beaches (Edo et al., 2019; Herrera et al., 2018), and even the deepest parts of the ocean (Bergmann et al., 2017). Besides, plastic accumulation in the marine environment endangers the marine environment in a variety of ways. Ingestion of MPs by marine organisms is the most common impact, which results in transfer along with the food web (Ozturk and Altinok, 2020; Setälä et al., 2014). Several studies revealed that ingested MPs cause weight loss (Besseling et al., 2013), movement restriction (Morgana et al., 2018), decreased food intake (Wright et al., 2013a), stress (Rochman et al., 2013), increased mortality rates (Jemec et al., 2016), growth deceleration (Gandara e Silva et al., 2016), and accumulation in soft tissues (Van Cauwenberghe and Janssen, 2014) in aquatic organisms.

The Black Sea is an almost enclosed sea connected to the Mediterranean with the narrow Turkish Straits System (Bosporus and

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Dardanelles) and Sea of Marmara. Annually, approximately 370 km³ freshwater including the largest rivers of Europe (Danube, Dniester, and Dnieper rivers) discharged into the Black Sea from the drainage basin which is about 2 million km² and covers almost all of the European countries (BSC, 2008). The coastal area of the southern Black Sea is a primary spawning ground for small pelagic fish species; European anchovy (Engraulis encrasicolus), and European sprat (Sprattus sprattus) (Gucu et al., 2016; Sahin and Düzgünes, 2019). The mentioned species are planktivorous fish feeding on a diverse group of zooplanktons, which are known to ingest microplastics accidentally during their feeding activity (Botterell et al., 2019). Microplastic ingestion by European anchovy was reported by Mazlum et al. (2017); however, their findings are at a preliminary level. The mentioned pelagic fish species constitute more than 80% of the total catch in the Black Sea (FAO, 2020). However, they are being fished at biologically unsustainable levels (FAO, 2018). Nevertheless, there is no comprehensive study on the impacts and ingestion of MPs by marine fish species in the Black Sea. Moreover, the levels of microplastic pollution on the spawning ground are not well understood. Considering the highly populated coastal zone, large drainage basin, high durability of the plastic particles in the marine ecosystem, and uncontrolled discharges, the Black Sea is at the risk of accumulation.

Synthetic polymers expose to a wide variety of physical, chemical, and mechanical effects upon entering the marine environment resulting in different shapes, sizes, and even densities (Zhang et al., 2021). Thanks to the mentioned effects, the composition and abundance of the microplastic particles collected from different parts of the marine environment vary. The particles float and transported by winds and currents, suspended in the water column, or sink to the bottom (Zhang, 2017). However, microplastic particles' physical properties and behaviors have not been extensively understood (Wang et al., 2016). Understanding these properties by field studies is crucial for simulating the fate in the marine environment (Critchell and Lambrechts, 2016). Plastic pollution has become a major area of interest in the Black Sea. Several studies reported a high share of plastic waste on the Black Sea coast on the macro scale (Aytan et al., 2020b; Oztekin et al., 2020; Paiu et al., 2017; Simeonova et al., 2017; Terzi et al., 2020; Terzi and Seyhan, 2017; Topçu et al., 2013). However, the number of studies on microplastic pollution in the Black Sea environment is scarce. The first known study on microplastic pollution in the Black Sea was conducted by Aytan et al. (2016) on southeastern coastal waters. Several studies reported microplastic pollution in the deep-sea sediment (Cincinelli et al., 2021), seawater (Berov and Klayn, 2020; Eryasar et al., 2021), and mussels (Gedik and Eryaşar, 2020). To our best knowledge, this is the first study evaluating the microplastic pollution on the beach sediments and adjacent waters of the Black Sea considering geography and climate. This study aims to collect data on the abundance, characteristics, and spatial distribution of MPs on the southern Black Sea coast (Turkish coast). The results of this study will be a valuable baseline for simulation studies to predict the transportation patterns and hot spots. The concentrations and synthetic polymers presented in our study can also be a baseline for experimental studies on commercially and ecologically important fish species in the Black Sea.

2. Materials and methods

2.1. Study area and sampling

The southern coast of the Black Sea is highly populated, with approximately 24 million inhabitants (TURKSTAT, 2020). The western section of the coast is considerably more populated due to the high number of industrial facilities. Beach sediment and seawater samples were collected from 23 stations (Fig. 1) in March 2021 (stations 1–7) and April 2021 (stations 8–22). The coordinates and local names of the stations were given in Supplementary Table 1. The Turkish coast of the Black Sea was divided into four different subregions (eastern Black Sea

(EB), middle Black Sea (MB), western Black Sea (WB), and Marmara (M) regions) based on geography and climate. Each station was considered as a sampling unit in those regions, which was also previously adopted by Hidalgo-Ruz and Thiel (2013). The first eight stations (1–8) were located on the EB, and the rest were on MB (9–12), WB (13–19), and M (20–23) (Fig. 1).

Three replicates of beach sediment were collected at the high tide line. The sediment samples were collected from the top 5 cm layer within a 30×30 cm area using a metal shovel. Sediment was transferred into aluminum foil containers and wrapped with aluminum foil to prevent contamination before analysis. Three replicates of seawater samples were collected with a 25 μm plankton net in the surf zone (breaker zone). The net was fully submerged to seawater near the surface and towed horizontally parallel to the coast along 20–50 m depending on the water condition. The net was washed with seawater from the outside and the sample in the collector was transferred to a glass jar. The volume of filtered seawater was measured using a flow meter (Hydro-Bios).

2.2. Pre-experimental set-up and contamination control

All the experimental processes were carried out in a controlled and clean lab environment. Polymer-free cotton aprons and gloves were used throughout the work. The laboratory equipment (beaker, filter, bottles, and petri dish, $\it etc.$) were rinsed with filtered ultrapure water before use. The solutions were filtered through the Whatman GF/C filter (47 mm diameter, 1.2 μm pore size) and stored in glass containers. To detect airborne contamination during the visual inspection, a petri dish filled with filtered ultrapure water was placed near the microscope as a blank. The blank was checked after processing each sample and any particle found was transferred to a new filter. The collected particles were evaluated using Frontier Fourier-Transform Infrared Spectrometer (FTIR), and in case non synthetic polymers were detected the data was corrected by subtracting from the total MP count.

2.3. Extraction of microplastics

An extraction method that was adopted from Hidalgo-Ruz et al. (2012) was used to extract MPs from the beach sediment and seawater samples. Sediment samples were dried at 60 $^{\circ}$ C for 48 h. A total of 100 g subsamples were transferred to the beakers and 500 ml of supersaturated ZnCl₂ solution (1.65 g/cm³) was added. The content was mixed for 2 min using a glass stirring rod. The samples were left to settle for approximately 4 h. The supernatant was collected using a vacuum pump with a glass pipe and filtered through a 25 µm filter. This procedure was repeated three times for each subsample to maximize the extraction. Simultaneously, seawater samples were transferred into beakers. To digest the organic material in the beach sediment and seawater samples, 50 ml of H₂O₂ solution (30%) was added, and the beakers were covered with aluminum foil to prevent airborne contamination. After three days of digestion at 65 °C, the content of the beakers was filtered through Whatman GF/C filter. In some cases, due to the high number of particles in the sample, multiple filters were used to expedite visual observation under the microscope. The filters were kept in the glass petri dishes until visual inspection.

2.4. Inspection and verification of MPs

The filters were inspected under a stereomicroscope. All the particles suspected to be MP were transferred to a new filter using a needle. Each particle was photographed using a digital camera, counted, measured, and categorized by shape (fiber, foam, fragment, film, or pellet). To verify and identify the particles, FTIR (PerkinElmer) was used. Polymer identification was conducted under 4000–650 cm $^{-1}$ spectral range, with 18 repetition scans (n) at 4 cm $^{-1}$ resolutions. The obtained data was compared with the instrument library data and the particles with $>\!70\%$ match were accepted as MPs.

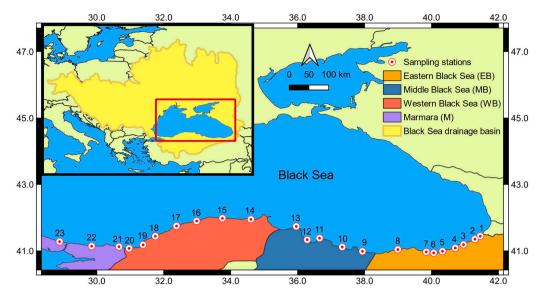


Fig. 1. Map of the study area and the sampling stations.

2.5. Data analysis

MP abundance of beach sediment and seawater samples were estimated as particles/kg and particles/m³, respectively, and given as mean \pm SE. The normal distribution and equal variances of the data were tested using Shapiro-Wilk and Levene's tests, respectively. In case the assumption of one-way ANOVA is met, the data were tested using oneway ANOVA followed by a Tukey HSD post hoc; otherwise, Kruskal-Wallis test followed by a pairwise Wilcoxon test with Bonferroni correction was used. The relationship between the MP abundance in beach sediment and seawater samples, as well as MP abundance and population were assessed using Spearman's correlation. The composition of the microplastics was visually compared using nonmetric multidimensional scaling (NMDS). The difference between the regions was tested using analysis of similarities (ANOSIM) with Bray-Curtis distance and 999 permutations. When significant differences were detected using ANOSIM, the similarity percentage breakdown (SIMPER) procedure with Bray-Curtis distance was applied to determine which region(s) differ and to identify the major contributors. Shannon diversity index was calculated to compare the microplastic diversity between the regions. The data was analyzed using R ver. 4.1.0 (R Core Team, 2021) using vegan (ver. 2.5-7) (Oksanen et al., 2020) and rstatix (ver. 0.7.0) (Kassambara, 2021) packages. The data was visualized using ggplot2 (Wickham, 2016) package and QGIS 3.16 (QGIS Development Team, 2021).

3. Results

3.1. Characteristics of the collected microplastics

The collected polymers were verified using FTIR. A total of 15 synthetic polymer types were determined from the beach sediment and seawater samples (Fig. 2).

3.1.1. Beach sediment samples

A total of 327 suspected MPs were extracted from the beach sediment samples. FTIR analysis revealed that 308 (94.19%) of the particles were synthetic polymers, of which 128 (39.14%) particles were determined as styrene acrylonitrile copolymer (SAC), followed by 114 (34.86%) polyethylene terephthalate (PET), 22 (6.73%) polyethylene (PE), and 19 (5.81%) polystyrene (PS) (Fig. 3A). Among the suspected microparticles, 17 (5.20%) of the particles were cellulose (C) followed by 2 (0.61%) calcite (Ca), which were subtracted from the data for further

analysis. The most common shape was fragment (46.75%), followed by fiber (41.23%), film (10.06%), and pellet (1.95%) (Fig. 3B). The determined MPs' size ranged between 0.15 and 4.99 mm, and the average size was 1.60 ± 0.06 mm (Fig. 3C).

3.1.2. Surface seawater samples

The number of the suspected MPs from seawater samples was 1041. After an FTIR analysis of the suspected particles, 33 (3.17%) cellulose (C), 9 (0.86%) starch (S), and 1 (0.10%) calcite (Ca) particles were identified and removed from the data. The composition of the collected MPs was dominated by PET (54.90%) followed by PE (13.46%), and PP (11.06%) (Fig. 3D). More than half of the MPs were in fiber shape (55.27%); on the other hand, very few pellets were observed (0.2%) (Fig. 3E). The size of the MPs was ranged between 0.23 and 4.98 mm and the average size was estimated as 2.07 ± 0.04 mm (Fig. 3F).

3.2. The abundance of the MPs

MPs were detected in all samples of both beach sediment and surface seawater. The MP abundance in the beach sediment was ranged between 9.35 and 172.90 particles/kg with a mean abundance of 64.06 \pm 8.95 particles/kg. The MP abundance significantly varied between the stations (one-way ANOVA, F(22,46) = 9.54, p < 0.05). The highest abundances were recorded on station 4 (172.90 \pm 8.09 particles/kg), 1 $(140.19 \pm 32.37 \text{ particles/kg})$ and $11 (121.50 \pm 5.40 \text{ particles/kg})$, on the other hand, lowest abundances were recorded on station 19 (9.35 \pm 3.06 particles/kg), 18 (11.54 \pm 2.00 particles/kg), and 12 (18.69 \pm 5.40 particles/kg) (Fig. 4A). Statistically, a significant difference was detected between the regions as well (Kruskal-Wallis test, $\chi^2(3) = 16.95$, p <0.05). The MP abundance in WB was found to be significantly lower than the other regions (Wilcoxon test, p < 0.05) (Fig. 4C). MP abundance in seawater samples were differed between the stations (one-way ANOVA, F (22,46) = 11.31, p < 0.05). The highest, lowest, and mean abundances were 4.42 \pm 1.34, 55.67 \pm 9.72, and 18.68 \pm 3.01 particles/m³, respectively. Stations 19 (55.67 \pm 9.72 particles/m³), 20 (44.35 \pm 14.45 particles/m³), and 21 (43.41 \pm 17.13 particles/m³) were found to be the most polluted, whereas station 12 (4.42 \pm 1.34 particles/m³), 11 (4.95 \pm 1.29 particles/m³), and 7 (5.66 \pm 1.24 particles/m³) were found as less polluted (Fig. 4B). The MP abundance in the seawater samples in the M was significantly higher than other regions (Kruskal-Wallis test, $\chi^2(3)$ = 27.29, p < 0.05, Wilcoxon test, p < 0.05) (Fig. 4D). A Spearman's correlation test was applied to determine the relationship between the beach sediment and seawater MP abundance. A negative correlation was

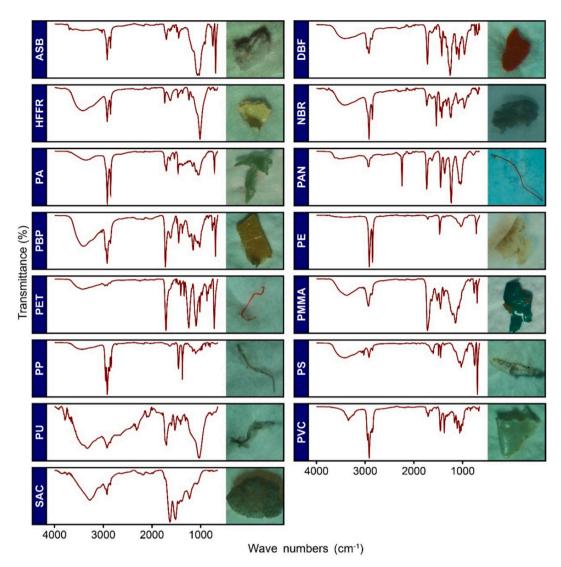


Fig. 2. Stereomicroscope photos and FTIR spectra of microplastics collected from the southern Black Sea coast. ASB: acrylonitrile butadiene styrene, HFFR: halogen free and flame retardant, PA: Polyamide, PBP: poly (BMA-co-PEGMA), PET: polyethylene terephthalate, PP: polypropylene, PU: polyurethane, SAC: styrene acrylonitrile copolymer, DBF: dibenzofulvene, NBR: nitrile butadiene rubber, PAN: polyacrylonitrile, PE: polyethylene, PMMA: poly (methyl methacrylate), PS: polystyrene, PVC: polyvinyl chloride.

determined; however, the correlation was weak and statistically insignificant (${\rm R}^2=-0.12, p=0.34$). We also tested the relationship between the population and microplastic abundance, but no significance was noticed for both beach sediment (${\rm R}^2=0.57, p=0.43$) and seawater (${\rm R}^2=0.59, p=0.40$).

3.3. The composition of the MPs

The composition of the MPs highly varied between the stations and regions (Fig. 5). The NMDS exhibited some degree of separation on MP composition between the regions on both beach sediment (Fig. 6A) and seawater samples (Fig. 6B). Thus, ANOSIM was implemented to test the difference of which results revealed that the composition was significantly different for beach sediment (ANOSIM, p < 0.05, R = 0.17), and seawater (ANOSIM, p < 0.05, R = 0.22). The composition of beach sediment and seawater were different within the regions as well (Supplementary Fig. 1).

The average Shannon diversity index (H) in beach sediment samples were estimated as 1.06 \pm 0.36, 0.50 \pm 0.19, 0.28 \pm 0.16, and 0.78 \pm 0.15 on the other hand, in seawater samples it was 0.93 \pm 0.22, 0.94 \pm 0.12, 1.20 \pm 0.04, and 1.31 \pm 0.11 in M, WB, MB, and EB, respectively.

Although the Shannon diversity index on M was relatively higher in the beach sediment (Fig. 6C) and lowest in the seawater (Fig. 6D), no statistical significance was determined between the regions beach sediment (ANOVA, F(3,19) = 2.17, p > 0.05) and seawater (ANOVA, F(3,19) = 2.34, p > 0.05).

The average dissimilarity ranged from 57.22 to 73.82% for the beach sediment and SAC, PET, PS, and PE were the highest contributors of dissimilarities between the regions. On the other hand, in the seawater samples, the average dissimilarity ranged between 43.17 and 73.92%, and PET, PE, PP, and PE were identified as the highest contributors (Table 1).

4. Discussion

The results of this study showed that microplastics are ubiquitous in the beach sediment and seawater of the southern coast of the Black Sea. A total of 1368 suspected microplastic particles were extracted from beach sediment and seawater samples collected from 23 stations along the southern coast of the Black Sea. A high percentage of the suspected microplastic particles were determined as polymers (94.19% from beach sediment, and 95.87 from seawater) which indicates the digestion

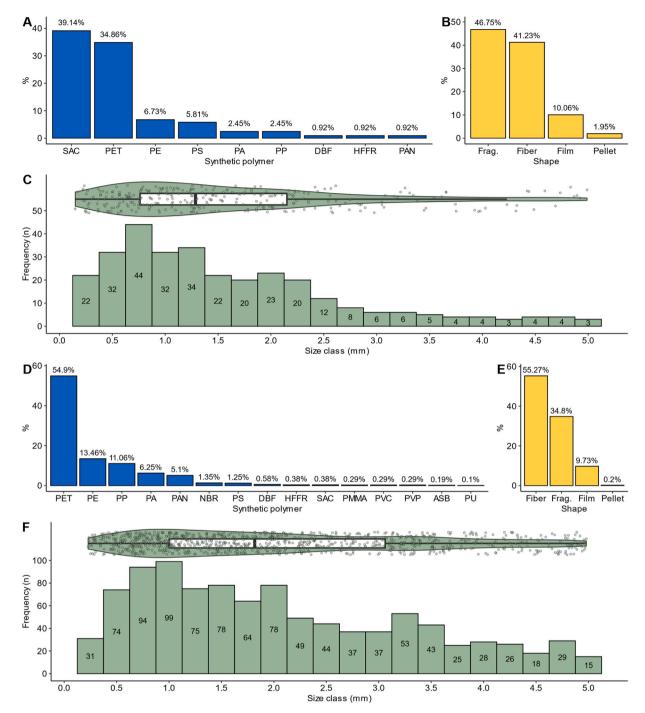


Fig. 3. Distribution of polymer type, shape, and size of the MPs collected from beach sediment (A, B, C) and seawater (D, E, F).

protocol is highly effective in eliminating organic particles. The variety of determined polymers was higher in seawater compared to beach sediment. The particles were dominantly by MPs less than 2.5 mm in size (>70%). The small sized microplastics have more bioavailability on lower levels of food chain (Wright et al., 2013b) which cause increased bioaccumulation and biomagnification on higher trophic levels (Farrell and Nelson, 2013). These organisms may threat human health on consumption (Seltenrich, 2015). The determined microplastics mostly consisted of fiber and fragment shape (>80%) which are predicted to be secondary microplastics. The fibers were suggested to be a result of wastewater discharges from washing machines (Browne et al., 2011); on the other hand, the fragments are more likely the result of the breakdown of larger plastic items in the marine environment.

The abundance of microplastics was significantly different between the stations and regions. A high abundance in the Marmara region (M) was observed as expected since it is Turkey's most populated and industrialized region. However, our results indicated no significant relationship between the abundance and population in the study area. Similar results were reported in different studies (Blumenröder et al., 2017; Nel et al., 2017; Nel and Froneman, 2015); on the contrary, several studies suggested a relationship between population and microplastic abundance (Browne et al., 2011; Cordova et al., 2019). Thus, the abundance in the region may rather be the result of high industrial activities. The eastern Black Sea (EB) is the second most polluted region. The population and industrial activities are lower compared to M and MB; however, the region receives much higher precipitation during

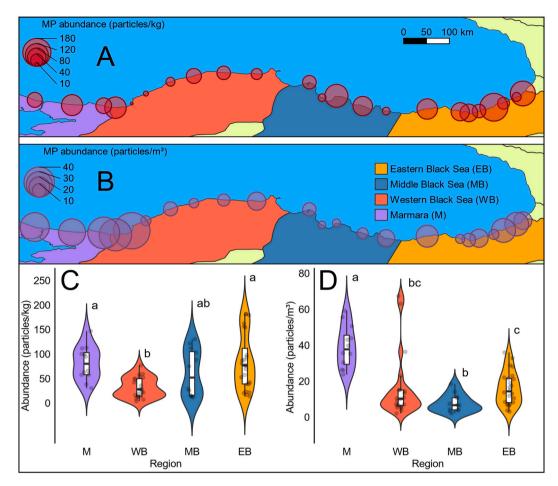


Fig. 4. The abundance of MPs in beach sediment (A) and seawater (B) on each station. Comparison of MP abundance in beach sediment (C) and seawater (D) between the regions. Different letters on C and D indicates statistically significant difference (Wilcoxon test, p < 0.05).

the year. High percentage of marine litter abundance in the region is attributed to rivers (Terzi et al., 2020) which may be the case for microplastics as well. The production and release of microplastics are unarguably dependent on the human population, socio-economic status, and sewage treatment facilities; however, the distribution in the marine environment highly depends on several environmental and physical processes and characteristics such as particle shape, particle density, seawater density, biofouling, surface currents, wind and wave-induced drift, turbulent vertical mixing (Critchell and Lambrechts, 2016; Wang et al., 2016; Zhang, 2017). The behavior of the microplastic particles in the marine environment is still a matter of debate (Wang et al., 2016). Miladinova et al. (2020) conducted a simulation study to predict the fate of microplastic particles released from five different locations in the Black Sea. Independent from the source, the highest accumulation rate was predicted on the Turkish coast (our study area) attributed to having the longest coastline among the surrounding countries. The spatial accumulation rates were highly variable and the coastal area of the Marmara region (M) was the most polluted area which is in line with our results. However, the given numbers in the mentioned study were much higher than our *in-situ* results ($>5 \times 10^3$ particles/m³), which may result from limitations of input level data for the simulations. Only a few studies of which result in high variability due to different methodologies and locations, report the input from Danube River to the Black Sea (Lechner et al., 2014; Liedermann et al., 2018; Van der Wal et al., 2015). Thus, more comprehensive studies with standardized methods are necessary for understanding the input from the sources.

Coastal sediments are a sink for microplastics in the marine environment (Browne et al., 2011). The substrate on the coastal area acts as a

filter and accumulates the particles transported by wind and waves. A very limited number of the studies focused on the microplastic pollution in sea sediments of the Black Sea (Aytan et al., 2020c; Cincinelli et al., 2021), on the other hand, to our knowledge, this is the first study evaluating microplastic pollution in beach sediment. Aytan et al. (2020c) reported abundance varying between 74.1 and 1778.8 particles/m² in sediment samples collected from 5 and 100 m depth from the southeastern Black Sea. The study conducted by Cincinelli et al. (2021) evaluated a comparatively larger area and reported abundance varying between 0 and 390 particles/kg sediments collected between the depths of 22 to 2131 m depth. Both studies revealed high spatial variance in MP $\,$ abundance. The abundance on the beach sediment is relatively lower compared to deep-sea sediments. The coastal area is more open to effects such as wind, beach cleaning, etc. which may cause a decrease in the microplastic abundance over time. Our study confirmed the high spatial variation in abundance as well as composition in the beach sediment on the southern Black Sea coast. Similar abundances and variance between stations were reported from beaches in France (Doyen et al., 2019), Poland (Graca et al., 2017), Iran (Naji et al., 2016, 2017), Slovenia (Laglbauer et al., 2014), and Belgium (Claessens et al., 2011).

There are a few field studies on microplastic pollution in seawater off the Black Sea coasts. Aytan et al. (2016) reported an average MP concentration of $1.2\pm1.1\times10^3$ particles/m³ in November and $0.6\pm0.55\times10^3$ particles/m³ in February in the southeastern Black Sea. The another study conducted on the Turkish coast (Sinop) determined the average microplastic particles on the sea surface and water column as 2.667 ± 2.325 particles/m³ for and 24.475 ± 26.153 particles/m³, respectively (Oztekin et al., 2017). Another study reported that

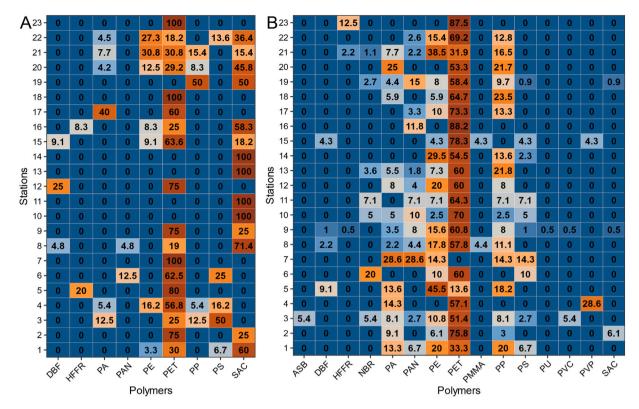


Fig. 5. Heatmap of the percentage of polymer type of microplastics in beach sediment (A) and seawater (B) by the stations.

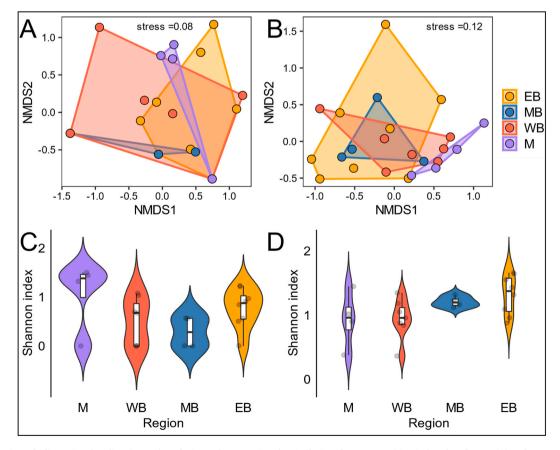


Fig. 6. Non-metric multidimensional scaling (NMDS) analysis result comparing the similarity of MP composition in beach sediment (A) and seawater (B) among the regions. The distribution of Shannon diversity index (H) on beach sediment (C) and seawater (D) among the regions.

Table 1The results of SIMPER analysis based on the Bray-Curtis dissimilarity.

Beach sediment samples				Seawater samples			
EB vs MB	Average dis. = 73.28%	Contrib. %	Cum. %	EB vs MB	Average dis. = 55.7%	Contrib. %	Cum. %
	SAC	45.05	45.05		PET	40.25	40.25
	PET	33.78	78.83		PE	16.03	56.28
	PS	9.515	88.34		PA	10.22	66.5
EB vs WB	Average dis $= 73.82\%$	Contrib. %	Cum. %	EB vs WB	Average dis. $= 53.77\%$	Contrib. %	Cum. %
	PET	41.26	41.26		PET	42.51	42.51
	SAC	31.74	72.99		PE	15.42	57.93
	PS	11.13	84.12		PP	10.28	68.21
EB vs M	Average dis. $= 57.22\%$	Contrib. %	Cum. %	EB vs M	Average dis. $= 62.39\%$	Contrib. %	Cum. %
	SAC	31.29	31.29		PET	49.04	49.04
	PET	29.13	60.42		PE	15.99	65.03
	PE	16.23	76.65		PP	12.87	77.9
MB vs WB	Average dis. $= 68.06\%$	Contrib. %	Cum. %	MB vs WB	Average dis. $= 43.17\%$	Contrib. %	Cum. %
	SAC	68.71	68.71		PET	48.16	48.16
	PET	18.96	87.68		PP	14.86	63.02
	DBF	4.121	91.8		PE	14.23	77.25
MB vs M	Average dis. $= 71.79\%$	Contrib. %	Cum. %	MB vs M	Average dis. $= 73.92\%$	Contrib. %	Cum. %
	SAC	44.96	44.96		PET	40.11	54.25
	PET	26.14	71.09		PE	11.26	15.24
	PE	15.42	86.51		PP	10.26	13.87
WB vs M	Average dis. $= 66.02\%$	Contrib. %	Cum. %	WB vs M	Average dis. $= 62.28\%$	Contrib. %	Cum. %
	PET	35.34	35.34		PET	50.78	50.78
	SAC	28.6	63.95		PE	16.18	66.96
	PE	18.95	82.9		PP	13.42	80.38

microplastic concentrations on the Bulgarian coast of the Black Sea were varied between 1.14×10^4 and 1.91×10^5 particles/km² (Berov and Klayn, 2020). However, the mentioned studies lack a chemical digestion process and identification of the synthetic polymers using FTIR or RAMAN spectroscopy techniques, which may cause overestimation by including the microparticles out of interest. Thus, standardized methods including these processes should be used in the future studies to obtain for accurate, reliable, and comparable results. In addition to these, in their recent study with updated methods, Aytan et al. (2020c) reported that microplastic particles varied between 1.78 and 40.03 particles/m³ in seawater off the southeastern coast of the Black Sea. Relatively, similar abundances in our study were estimated for the seawater sampled from the same region (EB and MB). Comparatively higher (Huang et al., 2019; Zhang et al., 2020) and lower (Baini et al., 2018; Robin et al., 2020) abundances were reported from different marine environments as well.

The composition of the microplastics collected from the southern Black Sea was evaluated. The overall composition in the beach sediment was dominated by SAC (40.53%), PET (38.75%), PE (6.91%), and PS (6.03%), whereas the seawater was dominated by PET (57.26%), PE (13.52%), PP (11.24%), and PA (7.17%). The predominance of PE, PP, PS, and PET could be attributed to their high global productions rates and common usage (PlasticsEurope, 2020). Interestingly, SAC is an uncommon polymer that was widely found in the beach sediment. Gündoğdu et al. (2017) investigated fouling assemblage on benthic plastic in the eastern Mediterranean Sea and reported SAC as uncommon in the study area. In our study, 71% of the SAC were fragments which are more likely secondary microplastics resulting from breakdown of larger items. PET was the most common synthetic polymer which was encountered in beach sediment at 18 stations and seawater at 22 stations out of 23 stations, whereas SAC (14 stations) and PE (19 stations) were the second most common synthetic polymers in beach sediment and seawater (Fig. 5). SAC was commonly detected in the beach sediment samples however, encountered only one station in seawater. Higher density than the seawater and fouling are considered as the main drivers of the difference. The Shannon diversity index (H) in M, WB, MB, and EB follow an order from high to low. The polymer complexity decreased from west to east, however, ANOVA results suggested no difference in synthetic polymers between the regions. Relatively higher complexity in M and WB can be linked to intensive industrialization in those regions. The microplastic composition of the regions visually exhibited some

degree of separation on the NMDS plot (Fig. 6A, B). Moreover, the composition of the synthetic polymers between the regions was significantly different according to ANOSIM test results. The highest dissimilarities in beach sediment were found between EB-WB (73.82%), EB-MB (73.28%), and MB-M (71.79%) and explained with SAC, PET, PS, and PE, whereas M exhibited the higher dissimilarity in seawater and highest dissimilarities were found between MB-M (73.92%), EB-M (62.39%), and WB-M (62.28%); driven by PET, PE, and PP.

The Black Sea fisheries are highly dependent on small pelagic fish species. The stocks are under the risk of overexploitation (FAO, 2018; Tsikliras et al., 2015) due to different fisheries management strategies of bordering countries (Duzgunes and Erdogan, 2008). On the other side, pollutants from many various sources threaten the Black sea ecosystem (Bat et al., 2018). Although it varies over the years, ~70% of the total catch consists of European anchovy (E. encrasicolus) on the Turkish coast. European anchovy is a planktivorous fish that primarily feed on zooplanktons and especially copepod (Calanus sp.) constitutes a high share of their diet in the Black Sea (Mazlum et al., 2017). The ingestion of microplastic beads is a long known fact from experimental studies (Ayukai, 1987). Recent studies reported decreased reproduction (Cole et al., 2015), modulation in antioxidant-related gene expression, changes in antioxidant enzyme activities (Choi et al., 2020), and altered feeding selectivity (Coppock et al., 2019). A study conducted by Shore et al. (2021) reported reduced population growth in a marine copepod and attributed to fecundity. A low number of microplastic particles were found in zooplankton samples collected from the southeastern Black Sea (Aytan et al., 2020a). However, taking account of the fact that increased microplastic abundance in the Black Sea may result in the same scenario, resulting in reduced energy transmission between trophic levels and reduced fish population. Our study area is an important spawning ground for fish species inhabiting the Black Sea (Gucu et al., 2016; Sahin and Düzgünes, 2019). Fish larvae primarily feed based on prey size (Takasuka et al., 2003). Thus, ingestion of microplastics by fish in early life stages is a common phenomenon (McGregor and Strydom, 2020; Steer et al., 2017). There are wide varieties of studies showing the effects of microplastics on the marine organisms, however, most of them were conducted using very high experimental concentrations compared to marine environment (Van Cauwenberghe et al., 2015a). On the contrary, Lönnstedt (Lönnstedt and Eklöv, 2016) suggested concentrations in the marine environment alter hatching, growth, and feeding preferences of fish larvae. Thus, along with fisheries management strategies,

reduction of non-fisheries-related impacts such as MPs in the marine environment should be considered as a tool for sustainable fisheries. Apart from direct effects on the marine organisms, species consumed as whole such as mussels result in microplastic uptake by humans (Gedik and Eryaşar, 2020; Gündoğdu et al., 2020) which creates food safety risk.

5. Conclusion

Microplastic pollution and composition in beach sediment and adjacent seawater on the southern Black Sea are unknown. This study was aimed to determine the spatial variance in abundance and characteristics of the microplastic particles distributed in the southern coast of the Black Sea. Microplastics were encountered in all the samples and more than 90% of the suspected microplastics were validated as synthetic polymers. The southwestern coast of the Black Sea was more polluted and complex in terms of composition compared to other regions. The area is more industrialized and populated compared to other regions of the study area; however, our results indicated an insignificant correlation between the microplastic abundance and population. Thus, it is concluded that facilities, winds, and currents are more likely the main drivers of distribution of microplastics in the study area. Considering the Black Sea is an almost enclosed sea and microplastics as a durable pollutant, the abundance will increase over time unless reduction at source strategies is implemented. However, the accumulation is ubiquitous and inevitable with current practices and measures of the countries in the Black Sea drainage basin.

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CRediT authorship contribution statement

Yahya Terzi: Conceptualization, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Software, Visualization, Project administration, Funding acquisition. Kenan Gedik: Conceptualization, Investigation, Methodology, Data curation, Writing – review & editing. Ahmet Raif Eryaşar: Investigation, Writing – review & editing. Rafet Çağrı Öztürk: Conceptualization, Investigation, Writing – review & editing. Ahmet Şahin: Investigation. Fatih Yılmaz: Investigation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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