

Detection and public health risk assessment of microplastics in disposable (PET) bottled water produced and sold locally in the Aegean Region

Ege Bölgesinde üretilen ve yerel olarak satılan tek kullanımlık (PET) şişelerdeki sularda mikroplastik tespiti ve halk sağlığı risk değerlendirmesi

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Abstract: Intensive use of plastic has led to the accumulation of plastics in all ecosystems and inevitable environmental pollution. Plastic wastes have undergone structural degradation with the effect of environmental factors and have been disintegrated into nano and microparticles; thus, might accumulate in living organisms and reach unpredictable levels in the food chain. In recent years, the impacts of these particles called "microplastics" (MP's) have become one of the most important issues in the scientific world. The aim of this study is to evaluate the possible presence of MP's in drinking water, that represents the most important nutrition element for human beings. For this purpose, samples of 6 different brands of disposable (PET) bottles produced and sold locally were examined. A total of 36 samples in bottles with 2 different volumes were analyzed in accordance with international standards and the results were evaluated. As a result of the study, the presence of MP's was detected in all samples analyzed. A total of 207 MP's were found in 36 samples. As a result of the analysis, a mean of 7.35 ± 9.66 MP L⁻¹ particles was detected. It was determined that the most dominant type in terms of shape was fiber (91%), the most dominant type in terms of color was blue (57%) and the most dominant type in terms of size was 0.1-1 mm (71%). When Estimated Daily Intake (EDI) rates were calculated for public health risk assessment, it was determined that the most affected group is the 3-6 age group (EDI_{(avg)}} = 0.42). The importance of making recycling more widespread, raising awareness of consumers and making the necessary legal regulations on the issue was emphasized in order to reduce the problem at its source.

Keywords: Plastic, microplastic, environmental pollution, public health

Öz: Plastiğin yoğun kullanımı nedeniyle tüm ekosistemlerde plastik atıkların birikmesi kaçınılmaz bir çevre kirliliğine yol açmıştır. Plastik atıklar çevresel faktörlerin etkisiyle yapısal bozulmaya uğrayarak nano ve mikro partiküllere ayrılmakta; böylece canlı organizmalarda birikebilmekte ve besin zincirinde öngörülemeyen seviyelere ulaşabilmektedir. Son yıllarda "mikroplastik" olarak adlandırılan bu parçacıkların etkileri bilim dünyasının en önemli konularından biri haline gelmiştir. Bu çalışmanın amacı, insanlar için en önemli besin ögesi olan içme suyundaki olası mikroplastik varlığının tespit edilmesidir. Bu amaç doğrultusunda, yerel olarak üretilen ve satılan 6 farklı markanın tek kullanımlık (PET) şişelerindeki numuneler incelenmiştir. İki farklı hacme sahip şişelerde toplam 36 adet numune uluslararası standartlara uygun olarak analiz edilmiş ve sonuçlar değerlendirilmiştir. Çalışma sonucunda analiz edilen tüm numunelerde mikroplastik varlığı tespit edilmiştir. Analizler sonucunda, 36 örnekte toplam 207 mikroplastik bulunmuştur. Ortalama olarak ise $7,5 \pm 9,66$ MP L⁻¹ tespit edilmiştir. Şekil açısından en baskın türün fibril (%91), renk açısından en baskın türün mavi (%57) ve boyut açısından en baskın türün 0,1-1 mm (%71) olduğu belirlenmiştir. Halk sağlığı risk değerlendirmesi için hesaplanan günlük alım (EDI) oranları göre en fazla etkilenen grubun 3-6 yaş arası grup olduğu (EDI_{(avg)}} = 0,42) tespit edilmiştir. Çalışma sonucunda, sorunu kaynağında azaltmak için geri dönüşümün yaygınlaştırılması, tüketicilerin bilinçlendirilmesi ve konuyla ilgili gerekli yasal düzenlemelerin yapılmasının önemi vurgulanmıştır.

Anahtar kelimeler: Plastik, mikroplastik, çevre kirliliği, halk sağlığı

INTRODUCTION

In 1972, the world became aware of the presence of micro-sized plastic particles in the aquatic environment for the first time, when it was reported that large numbers of small floating plastic particles were found in the Sargasso sea surface water (Carpenter and Smith, 1972). In 2004, these small particles were defined as microplastics (Thompson et al., 2004). Then, plastics smaller than 5 mm were accepted as microplastics by the Steering Committee of the National Oceanic and Atmospheric Administration (NOAA) Marine Debris Program. According to this definition, plastics were

specified as Macroplastic ≥ 25 mm, Mesoplastic 25 – 5 mm, Microplastic ≤ 5 mm – 1 μ m and Nanoplastic < 1 μ m.

As microplastics are highly absorbent, they attract pollutants such as heavy metals, endocrine disrupting chemicals and durable organic pollutants (Yurtsever, 2015; GESAMP, 2015). These harmful substances cause harmful effects such as birth defects, cognitive development disorders, reproductive problems and cancer. Although studies on microplastic pollution have increased today, there are not enough scientific research on the determination of

microplastic pollution in food and its effects on human health (Li et al., 2018). Although microplastics do not have a direct negative effect on human health, it is necessary to increase the number of scientific studies that may adversely affect human health due to the dangerous substances in the structure of microplastics or adhere to the surface. It has been suggested that nanoplastics rather than microplastic particles adversely affect human health (Rist and Hartmann, 2018). In a study on microplastics ingested by rodents, it was determined that microplastics were very difficult to absorb in the blood. In the same study, it was observed that a very small part of the microplastics could reach the blood circulation via lymph, but it was also determined that this amount could not penetrate the organs and was probably eliminated via the spleen (Bouwmeester et al., 2015). In another study, it was determined that most of the ingested microplastics and nanoplastics were removed from the human body (Smith et al., 2018). Ragusa et al. (2021) even reported its presence in the human placenta. Conti et al. (2020) demonstrated the presence of microplastics in vegetables and fruits for the first time in the world. Zuccarello et al. (2019) detected microplastics in drinking water bottled in Italy. Microplastics cannot pass through cell membranes due to their size. However, the effects of microplastics such as inflammation in the intestines are possible and thus can affect the immune system. In addition, microplastics found in stool samples taken from people living in different countries showed that microplastics negatively affected the digestive system (Schwabl et al., 2019).

The main purpose of this study was to evaluate the possible amounts of microplastics (MP) in drinking water brands, which are produced and distributed locally. Also, it was investigated whether the different volumes of the drinking water bottles could have an effect on the amount of microplastic. In this way, it is aimed to increase the awareness of both consumers, producers and local authorities.

MATERIAL and METHODS

Collection of Samples

In this study, 0.5 and 1.5 L samples were taken from 6 different brands of drinking water produced locally in Izmir, Aydın, Muğla and Denizli provinces and sold in markets in Muğla Province, and the possible presence of microplastics was analyzed. Three samples were taken from different volume sub-samples of each brand of water. The samples were immediately placed in a portable freezer and brought to Muğla University Faculty of Fisheries Water Quality Laboratory at a constant temperature of 4 °C and stored in the dark, under appropriate conditions until the analysis.

Analysis of samples

Various chemical and/or physical pretreatments were applied to detect the presence of microplastics in samples

(sediment, water and/or living tissue) taken from different compartments of the natural ecosystem. No chemical treatment was required to detect the presence of MP in water samples. For this reason, the standard method used to detect the presence of MP in waters all over the world was also used to detect the possible presence of MP in the sampled drinking waters in this study. The samples were filtered using GF/F Whatman® brand glass fiber filter papers with a diameter of 47 mm and a mesh size of 0.7 µm, using a vacuum pump (approximately 100 mbar under a maximum pressure of 2 bar).

Each of the filter papers on which the samples were filtered were taken into glass petri dishes separately, placed in a closed fume hood and left to dry at room temperature (23 °C). Then, each dried filter paper was examined and photographed twice by different researchers under two identical (twin) BOECO 55 MST 606 stereo microscopes, and the microplastics on it were counted and grouped according to their size, color and shape. The sizes of MP's were divided into two subgroups as 0.1–1.0 mm and 1.0-5.0 mm. The detection limit of stereomicroscopes of the particles was ~100 µm. As the stereomicroscopes may not accurately separate natural and synthetic particles, it has been suggested to use both microscopy and other advanced techniques (eg. FTIR, RAMAN etc.) to identify the possible MP's particles. Also, the results can be influenced by the quality of the microscope, the researcher, and the particle sizes which may lead to an underestimation or overestimation of amount of MP's (Song et al., 2015; Loder and Gerdt, 2015). For smaller size MP's (<500 µm), visual identification may not reliable at all times. As the sizes of microplastics decrease, the amount increases, and the accuracy of identification is reduced, which may cause considerable challenges in microplastic identification. To visually determine whether a particle is of plastic, the criteria of Hidalgo-Ruz et al. was met (Hidalgo-Ruz et al., 2012; Baltic, 2017), even though, a subset of samples was confirmed by Raman-spectroscopy. Microplastics were classified as red, blue, green, yellow, white and other in terms of color and as fragments, fibers and film particles in terms of shape, and counting was carried out in line with these groupings (Nuelle et al., 2014; Lots et al., 2017; Yabancı et al., 2019).

Prevention of possible microplastic interferences during laboratory studies

All equipment (washing bottle, volumetric flask, petri dish, beaker etc.) used during the analyzes carried out in the laboratory were cleaned with pre-filtered (GF/F glass fiber filter paper used in this study /0.7 µm- 47 mm diameter) distilled water in order to remove possible particles in it and were preserved in a fume hood, carefully covered with blotting paper. Nitrile gloves were used during the analyzes and all the doors and windows of the laboratory were always closed during the procedures in order to cut off the air flow (Torre et al., 2016; Crawford and Quinn, 2016). In addition, in order to detect any contamination that may occur by air, some filter

papers (GF/F glass fiber) (Field blanks) were left for control at 4 different points of the laboratory throughout the entire study. These results were deduced from all the results obtained by counting the possible plastics on these filter papers and it was tried to minimize the errors caused by the interferences that might arise from the environment. During the laboratory study, it was calculated that the time taken for each sample to be ready for analysis after it was brought from the natural environment was approximately 30 minutes (± 3 minutes). It was calculated how much interference the microplastics detected on the filters placed in different parts of the laboratory for control purposes could do, according to the 30-minute time frame and based on this number, the environment-based interventions found were subtracted from all the results obtained, and the most accurate findings were tried to be reached (Lusher et al., 2014; Catarino et al., 2017; La Daana et al., 2017). As a result of the study, $0.056 < 1$ piece in a 30-minute period for all MP's in the form of white fibers were detected in the field blanks.

Identification of polymers by micro-Raman

In order to capture the MP's image accumulated on the filters in the clearest way, the surface of the entire filter was scanned in 20-fold magnification. For the scanning area, the closest dimensions to the surface area of the filter, $4.4 \text{ mm} \times 4.4 \text{ mm}$ ($\cong 69 \text{ mm}^2$), were chosen (The diameter of the filter is 47 mm and its area is $\cong 74 \text{ mm}^2$ including the hollow partition at the edges). Thanks to the special computer software of the device, MP's quantity, size and morphology of the particles (image analysis) were determined. For scanning in the device, 532 nm excitation laser, 5 s integration time, laser intensity of 12%, laser spot size about 0.7 mm, spectral resolution of 5 cm^{-1} and spectral range between 500-4000 cm^{-1} were set. In order to define the results, they were compared with the sample data used for the identification of plastic-derived substances in the reference library within the device itself. Library matches with a rank ≥ 550 to ≤ 700 was analyzed and interpreted separately (Woodall et al., 2014). The particles suspected to be MP's were identified by scanning all surface areas of the filters. The particles suspected to be plastic with a similarity rate of more than 51% as a result of the scan were evaluated. The mean of similarity was 57.8% for all samples. Only those with a plastic-based structure were evaluated in the results of the study.

Estimation of microplastic intake by humans

The estimated daily intake (EDI) of MPs in bottled water was calculated using the following formula:

$$EDI = \frac{C \times IR}{bw}$$

EDI; Estimated Daily Intake ($\text{MP kg}^{-1} \text{ d}^{-1}$),

IR; Ingestion Rate (L d^{-1}),

C; Concentration of Mps (particles L^{-1}),

bw; body weight (kg).

Estimated Daily intake (EDI) of MP's is calculated based on the Daily average consumption of drinking water (IR), concentration of MP's in drinking water (C) and body weight (*bw*). The data of water consumption and body weight were estimated based on the 4 different target groups (infants, children, teenager and adults). The average water consumption rates (IR) in infants (0–2 years old), children (3–6 years old), teenagers (7–16 years old) and adults (≥ 17 years old) were 0.08, 0.85, 2 and 2.5 L day^{-1} , respectively. Body weight of target groups were considered 10, 15, 50 and 78 kg, respectively (Ghoochani et al., 2017; Yousefi et al., 2018). In addition to this IR, for the year 2022, an average of 63 L of water in plastic and glass bottles per adult person in Türkiye per year (31.5 L on average for a pet bottle year^{-1} person $^{-1}$) was consumed (SUDER, 2022).

Statistical Analyses

STATISTICA STAT 10.0 program was used in the statistical evaluation of the obtained data. A comparison was made between sample groups with 2 different volumes (0.5 L and 1.5 L) using Analysis of Variance (ANOVA), and it was revealed whether there was any statistically significant difference. In cases where there was a difference between groups, Tukey post-hoc test was used after ANOVA to determine which group the difference originated from. Obtained results were evaluated at $p < 0.05$ confidence level.

RESULTS

Presence of MP's in the samples

In the study, a total of 36 samples of drinking water in two different volumes (0.5 and 1.5 L) belonging to 6 different brands, which were bottled in the Aegean region and sold locally within the borders of Muğla Province, in disposable bottles were used. As a result of the examination of all samples, a total of 207 microplastic particles were detected (Figure 1).

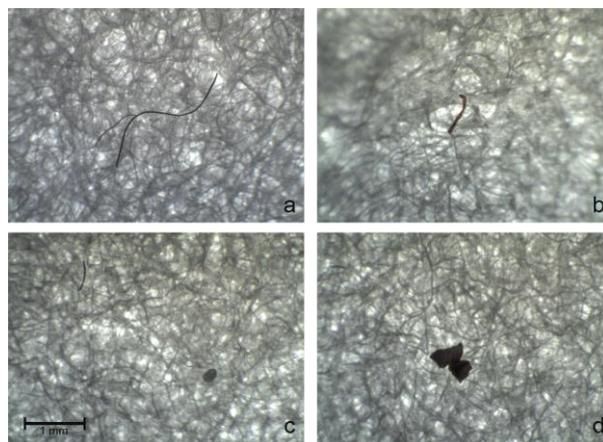


Figure 1. Microplastic samples detected on filters during the study. (a) blue fiber longer than 1 mm, (b) red fiber smaller than 1 mm, (c) black fragment and white fiber smaller than 1 mm, (d) black film smaller than 1 mm

The microplastics accumulated on the filter papers placed at 4 different points of the laboratory throughout the study in order to prevent microplastic contamination that might come from the air during laboratory studies, were counted under the microscope. As a result of the countings, it was determined that the amount of microplastics that could have interfered with the air was a very low value for each type or color per 30 minutes, which was the average processing time, and it was ignored ($0.056 < 1$ piece in a 30-minute period for all).

In the light of the findings, a total of 95 microplastics were detected in bottles with a volume of 0.5 L (mean 5.27 ± 5.35 particle bottle⁻¹) and a total of 112 microplastics in bottles with a volume of 1.5 L (mean 6.22 ± 11.21 particle bottle⁻¹). As a result of the study, a minimum of 1 particle bottle⁻¹ and a maximum 22 particle bottle⁻¹ in bottles with a volume of 0.5 L, and the presence of a minimum of 1 particle bottle⁻¹ and a maximum of 50 particles in a bottle⁻¹ in bottles with a volume of 1.5 L were found.

Concentration of MP's per liter

All results were calculated according to 1 L volume in order to compare the MP accumulation amounts in drinking water samples in bottles of different volumes. According to the results of these calculations, a total of 265 microplastics (average 7.35 ± 9.66 MP L⁻¹) were detected in all samples (n=36). A total of 190 particles (mean 10.56 ± 10.71 MP L⁻¹) were detected in 0.5 L volume bottles and 74.67 particles (mean 4.15 ± 7.47 MP L⁻¹) in 1.5 L volume bottles. As a result of the analysis, when all samples (n=36) were taken into account, a mean of 7.35 ± 9.66 MP L⁻¹ particles were detected in the drinking water used in the study.

According to the ANOVA results applied to the sample group, when comparing the amount of MP in water bottles of different volumes, it was determined that there was a statistically significant difference between the mean amount of MP in 0.5 L water bottles and the mean MP amount determined from 1.5 L water bottles in 1 L volume ($F= 4.33$; $p < 0.045$).

Types of MP's in the samples

The MP found in the filters examined during the study was classified into 3 subgroups (Fiber, fragment and film) in terms of type. In line with the findings, a total of 188 fibers, 17 fragments and 2 film-shaped microplastics were detected in all filters. In all samples, fiber-shaped microplastics with 47 pieces, fragment-shaped microplastics with 2 pieces and film-shaped microplastics with 2 pieces were counted at the highest value in a single filter.

As a result of the countings made by grouping the samples according to the types of microplastics, it was determined that fiber-shaped MP was more dominant (90.8%) in both 1.5 L and 0.5 L water bottles (Figure 2).

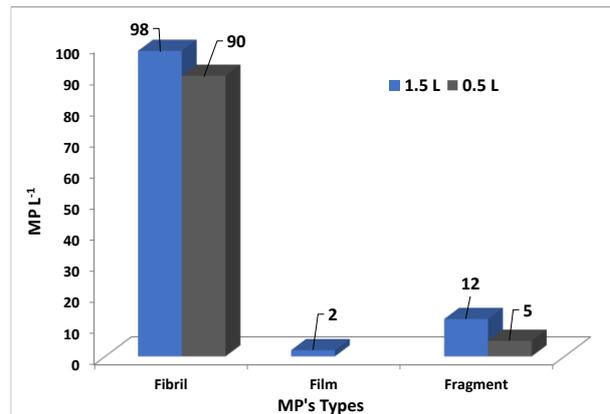


Figure 2. Dominant microplastic accumulations in 0.5 and 1.5 L water bottles in terms of shape

According to the results of the ANOVA test, which was carried out in line with the findings obtained according to the countings made by grouping in terms of shape, a statistically significant difference was determined between the shape of MP's in the drinking water samples sold in disposable bottles ($F= 5.98$; $p < 0.004$). Fiber-shaped MP's were statistically more abundant than other shaped microplastics (Figure 3).

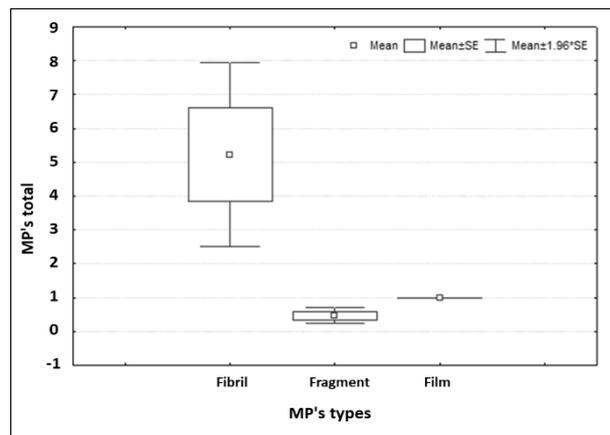


Figure 3. ANOVA results show that there is a difference between microplastic accumulation in terms of shape

In terms of MP colors in the filters examined during the study, they were classified into 6 subgroups as white, red, blue, yellow, green and other. In accordance with the visual countings, it was determined that the blue colored MP was the most dominant microplastics in terms of colors with 118 particles and other colored MPs with 62 units. In all samples, blue colored microplastics were counted with 42 pieces and other colored microplastics were counted as the highest value in a single filter with 5 pieces.

By grouping the MP colors in the samples and the volumes of the bottles (0.5 and 1.5 L), it was determined that the blue colored MP was dominant in both water bottle types (51.58% at 0.5 L volume and 61.61% at 1.5 L volume), followed by the other colored MP (32.63% at 0.5 L volume and 27.68% at 1.5 L volume). (Figure 4).

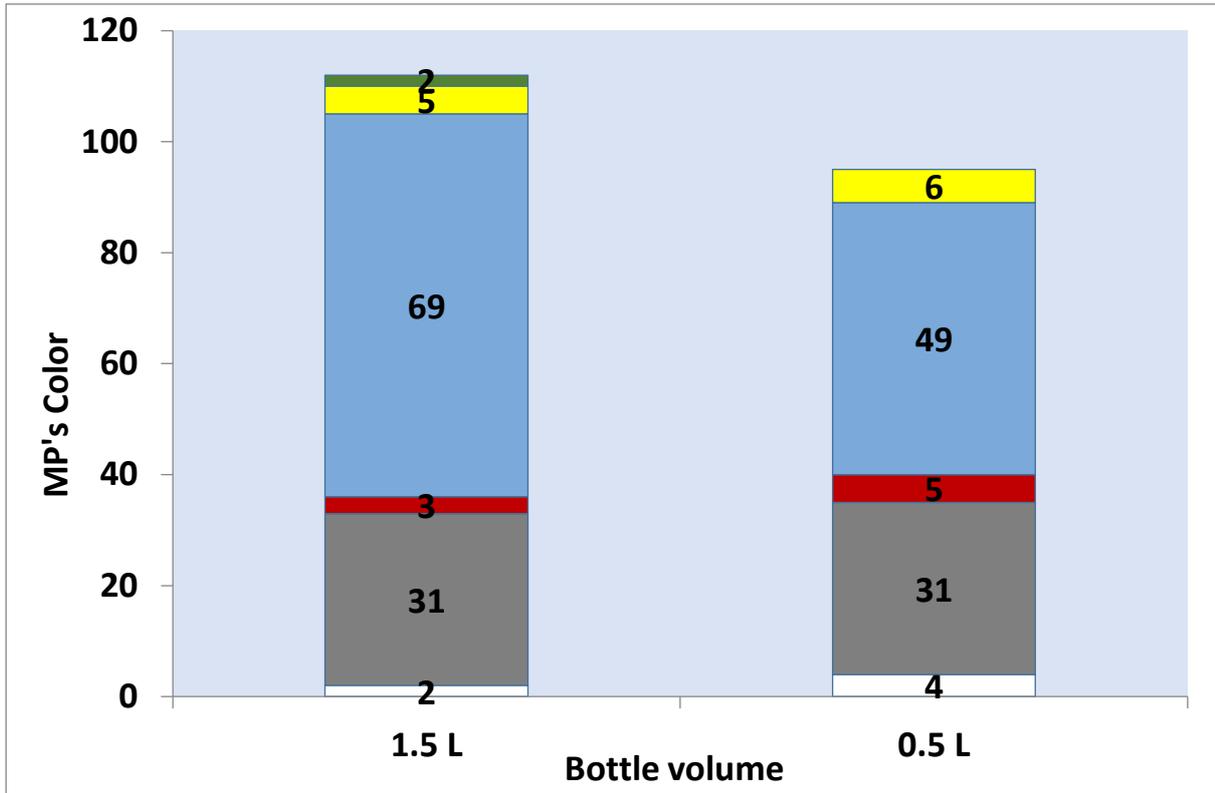


Figure 4. Microplastic accumulation dominant in terms of color groups in 0.5 and 1.5 L water bottles

In terms of color in all samples, it was determined that the MP in the blue and other colored group was more dominant (57% and 30% respectively), but according to the ANOVA results, it was determined that the amount of MP in different colors in the disposable water bottles did not have any statistical significance between the color groups. ($F= 2.330$; $p= 0.051$) (Figure 5).

The lengths of microplastics detected during visual counting of the filters were determined in two subgroups (0.1-1.0 mm and 1-5 mm) using scaled oculars. The analysis showed that 146 of the total microplastics detected were in the size range of 0.1-1 mm and 61 of them were in the size range of 1-5 mm (Figure 6). As a result of the statistical analysis, no significant difference was found between the groups.

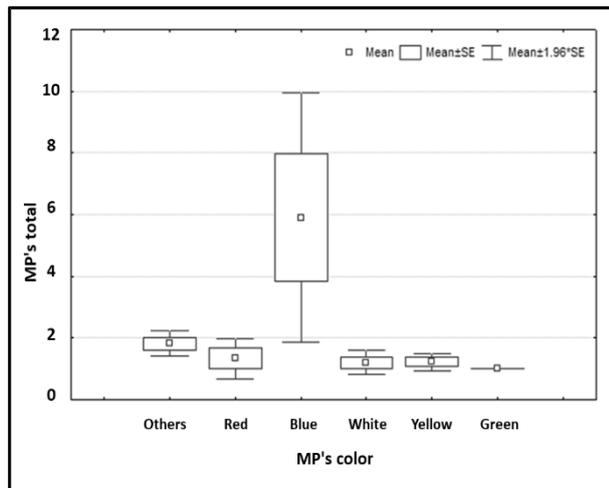


Figure 5. ANOVA results showing that there was no difference between microplastic accumulation in terms of color groups

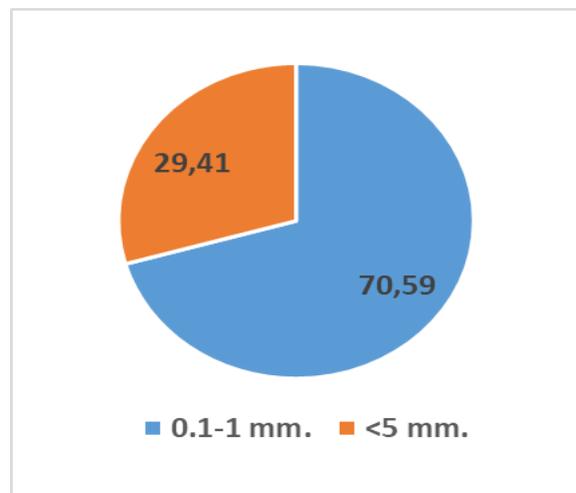


Figure 6. Distribution of MP's in terms of size groups (mm)

Qualitative assessment of MP's in the samples

According to the results of Raman spectroscopy, it was determined that MPs in the samples examined had Polyethylene (PET), polypropylene (PP) and Polyamid-Nylon (PA) type polymer structures. The most dominant of these plastics is PET with 78%, while the rarest is PA (Nylon) with 6%. The average identification rate of Raman spectroscopy was 57.8% (Figure 7).

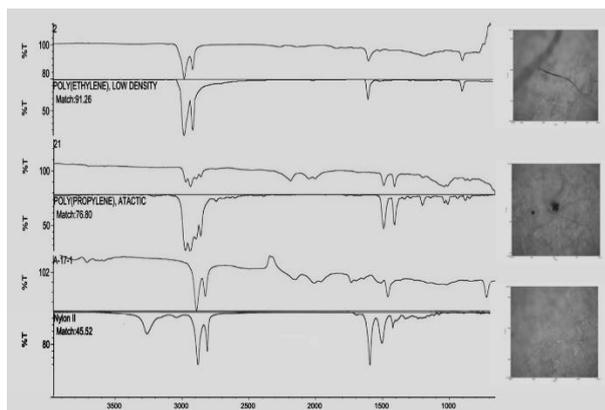


Figure 7. According to the Raman spectroscopy results, the spectrums determined with the highest detection ratio

Public health risk assessment

As a result of EDI calculations, it was determined that MPs in drinking water in 0.5 L and 1.5 L plastic bottles exposed the 3-6 age group the most (Table 1). In the light of the findings, the results of EDI_(0.5 L), EDI_(1.5 L) and EDI_(avg) were close to each other in each age group. Calculated by taking the mean of all results with 0.5 and 1.5 L bottles and using the annual average water consumption value in adults, the values of Annual Ingestion Rate (AIR) were found as 2.25, 2.52 and 2.32 kg person⁻¹ year⁻¹ respectively.

Table 1. EDI (kg d⁻¹) values were calculated in terms of water consumption according to each age group in relation to the results obtained from 0.5 and 1.5 L bottles

Ages	EDI _(0.5 L)	EDI _(1.5 L)	EDI _(avg)	IR (L day ⁻¹)	Bw (kg)
0-2	0.04	0.05	0.05	0.08	10
3-6	0.32	0.35	0.33	0.85	15
7-16	0.22	0.25	0.23	2	50
>17	0.18	0.20	0.18	2.5	78

According to the EDI calculation based on MP's accumulation in 0.5 and 1.5 L bottles compared to 1 L, it was determined that the most exposed group was again the 3-6 age group (Table 2). However, in this calculation, the EDI 1 L (0.5 L) value was found to be significantly higher than the other groups. Considering the annual average water consumption value in adults, the AIR 1 L (0.5 L), AIR 1 L (1.5 L) and AIR 1 L (avg) values for each 1 L of water

consumption were calculated as 4.26, 1.68 and 2.97 kg person⁻¹ year⁻¹, respectively.

Table 2. EDI (kg d⁻¹) values were calculated in terms of the results obtained from the comparison of 0.5 and 1.5 L bottles to 1 L according to each age group

Ages	EDI _{1 L (0.5 L)}	EDI _{1 L (1.5 L)}	EDI _{1 L (avg)}	IR (L day ⁻¹)	Bw (kg)
0-2	0.08	0.03	0.06	0.08	10
3-6	0.60	0.24	0.42	0.85	15
7-16	0.42	0.17	0.29	2	50
>17	0.34	0.13	0.24	2.5	78

DISCUSSION

As a result of this study, MP presence was detected in all 36 samples obtained from drinking water of 6 different brands sold in disposable bottles with two different volumes (0.5 and 1.5 L). One of the most important needs for the continuity of life is drinking water and these waters are suitable for consumption in terms of human health. Research on the presence and amount of MP in food products and the impacts of these pollutants on foods such as directly consumed drinking water is a recently emerged gray area. International standards regarding the risk, hazard and extent of exposure associated with MP entry into the human metabolism are not yet fully clarified.

There are some studies in the world whose subject is similar to this study. In a study conducted in 2018, the presence of MP in disposable (PET) bottles of 11 different brands was investigated and a mean of 14 ± 14 MP L⁻¹ particles (varying in the range of 2-44 MP L⁻¹) were detected (Schymanski et al., 2018). In 2018, Mason et al. investigated the presence of MP in 279 drinking water samples belonging to 11 different brands that were stored in 500-600 mL bottles and sold to 5 different continents, and the presence of MP was detected in 93% of all samples and an average of 10.4 MP L⁻¹ (> 100 µm) was found. In the study of Cox et al., in which they investigated the presence of MP in drinking water samples taken from the tap and bottled water in Canada in 2019, an average of 94.37 MP L⁻¹ particles were detected in bottled water and an average of 4.23 MP L⁻¹ particles in tap water. In a study carried out to detect the presence of MP in the drinking water of 10 different brands sold in single-use bottles in the markets in Thailand, the presence of MP was detected in all of the samples and the average was found to be 12 ± 1 MP L⁻¹ (> 50 µm) (Kankanige and Babel, 2020). In another study investigating the presence of MP in drinking water, which focused on distribution networks of drinking water and as a result, the possible pollution risk in 5 different water supplies was revealed. A mean of 174 ± 405 MP m⁻³ particles was determined in all drinking water distribution systems examined (Kirstein et al., 2021).

Although the results of this study show some similarities when compared with previous studies conducted in the world, there are also studies with very different findings. There may be some obvious reasons for these differences. The most important of these are methodological differences. Most of the studies (including this study) use the counted particles for which they could fully confirm the polymeric nature using Raman spectroscopy method. On the other hand, in some studies, sample filters are painted with dyes with different luminescence properties (eg. Nile red) and assuming that these dyes highlight the plastic particles, counting and identification processes are performed only on particles that glow. Some studies use bottled drinking water, while others have sampled tap water directly. The number of samples used in the studies also varies widely. As a result, the subject of standardization in studies related to MP remains blurry. There is an urgent need for the organizations leading such studies in the world to explain more clear and universal methodologies that may be applicable by all scientists in the world.

The results of this study are compatible with the results of studies similar to this subject in the world. (Table 3).

Table 3. Comparison of study results with similar studies conducted in the world

Container	Liquid type	Sample number (n)	Mean microplastic number (MP L ⁻¹)	Study
Single use (PET bottle)	Drinking water	295	10*	Mason et al., 2018
Single use (PET bottle)	Drinking water	32	16	Oßmann et al., 2018
Single use (PET bottle)	Drinking water	38	14*	Schymanski et al., 2018
Single use (PET bottle)	Drinking water	3	3	Kosuth et al., 2018
Single use (PET bottle)	Drinking water	18	8	Winkler et al., 2019
Single use (PET bottle)	Drinking water	65	12*	Kankanige and Babel, 2020
Single use (PET bottle)	Drinking water	69	3	Zhou et al., 2021
MEAN		74	9	
Single use (PET bottle)	Drinking water	18	7*	Current study

* > 50 - 100 µm

It was determined that the most dominant polymer chain detected by Raman spectroscopy in the study was PET. The predominance of PET type MPs might be due to the material from which the bottles were made. Although the rules to be followed in facilities where drinking water is stored and bottled in containers of different volumes are very strict, the hygiene rules taken to prevent factors that may endanger human health might be taken into consideration more. Considering the process in which drinking water is put into bottles, the method used is the technique of filling the bottles as soon as possible with pressure. Thus, during this process, due to pressure, fractures and ruptures may have occurred on the inner surfaces of the bottle. In addition, pressurized water may create stress fractures on the inner surface of the bottle, and these fractures may cause ruptures in later processes (transportation, storage, etc.) (Kankanige and Babel, 2020). In addition, if the bottles are kept in conditions where the thermal impact may occur, such as sunlight or hot temperatures, where water is stored, these stress fractures may cause degradation. Therefore, the MP release from the packaging material is an outcome of its natural degradation and other causes in the production process, distribution and storage, and consumer behavior.

According to the results of Raman spectroscopy, the rarest polymer was found as PA (Nylon). PA is the most common type of MP in airborne particles in a laboratory environment. Studies have shown that atmospheric microfiber wastes in urban areas, for 2–355 microfibers m⁻² particles, 50% are cotton/ wool (which are PA nylon) with 17% referring to other plastics (Dris et al., 2016). It is thought that white colored fibril-shaped plastic cotton/wool PA nylon particles may originate from the white coats worn by the laboratory team. As a result, the small number of PA detected in the samples may be due to air circulation in the laboratory environment.

The leading organizations of the world (FAO, ATSDR, WHO, GESAMP etc.) have not yet made valid and accepted regulations about the possible negative impacts of MP's detected in foods on human health. There are legal restrictions, the validity of which is open to question, only for certain foods that have the possibility of direct contact with the contaminant and can be consumed directly with very little pre-treatment (such as mussels). Potential risks posed by MP's for drinking water have just started to be studied, and there is no legal limit accepted yet. As a result, it should be noted that many parameters have an impact on the EDI, including the consumption behavior and quality of the bottle material. Consequently, further research is necessary to determine the permissible daily intake of MPs in drinking water.

CONCLUSION

Plastic has become one of the basic materials that human beings cannot give up using due to its ease of production and availability, and its high reuse rate. However, due to its intensive and unconscious production and/or use, its

uncontrolled presence in nature in recent years has led to undeniable environmental problems. So much so that these residual wastes of plastics, which are divided into particles too small to be seen by environmental factors and called "microplastics", have been heavily involved in the food chain and even started to be detected in the structure of basic nutrients that are directly consumed as human food. Current study and similar studies in the world have shown that even in drinking water, which is in the most basic food group and bottled untouched, MP is present. Although microplastic has been recognized as an environmental problem in the first quarter of the 21st century, it is difficult to say that there is sufficient research on this issue. Although human exposure to microplastic is accepted as a global problem, it is still an open question what precautions should be taken related to many different parameters such as uncertainty, variability and lifetime accumulation. Currently, available information on the potential adverse effects of MP on human health is insufficient, scattered and yet far from the desired level. There are no regulations regarding microplastics as a food pollutant in the world and in our country. Although there is a general tendency to draw a legal framework regarding MP found in foods and natural ecosystems, various information gaps complicate the issue at this point. As a result of future studies, ensuring the unity in analytical methods, determining the toxic effects of MP and eliminating the deficiencies in monitoring data will also guide the legal regulations to be made in this regard.

In order to reduce the problem at its source, making recycling more widespread, raising awareness of consumers and initiating the necessary legal actions on the subject are important for both food safety, human and animal health and ecological balance.

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AUTHORSHIP CONTRIBUTIONS

All phases of the study, such as conceptualization, methodology, research, verification, software, etc. were carried out by Aykut Yozukmaz.

CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest or competing interests.

ETHICS APPROVAL

No specific ethical approval was necessary for this study.

DATA AVAILABILITY

For questions regarding datasets, the corresponding author should be contacted.

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