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Microplastic accumulation in a lizard species: Observations from the terrestrial environments☆

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ABSTRACT

Microplastics are a global environmental problem, polluting both aquatic and terrestrial environments. Terrestrial lizards are suitable model organisms to study human-induced pollution in these areas, as they can live in urbanized areas where microplastics are most abundant. Therefore, we analyzed the prevalence of microplastics (MPs) in a common Lacertid lizard, the snake-eyed lizard, *Ophisops elegans*. We detected MPs in the gastrointestinal tract (GIT) of 33 of 152 specimens from 18 populations. The detected MPs had six distinct polymer compositions, namely Polyethylene terephthalate, Polyacrylonitrile, Polypropylene, Polyethylene, Poly methyl methacrylate and Polyamide. The majority of these MPs were fiber-type and the dominant color was navy blue. The lengths of MPs varied from 37 to 563 μm, with an average length of 175 μm. MPs were detected in the GITs of 43% of juveniles (n = 7), 30% of males (n = 105), and 18% of females (n = 40), with a mean of 0.27 per specimen. Furthermore, we found that microplastic densities varied with habitat distance from human settlements, supporting the theory that high levels of microplastic contamination are associated with extensive anthropogenic activity.

1. Introduction

Since the 1950s, plastics have played pivotal roles in various industries and daily necessities due to their enduring nature and versatility. Annual plastics production currently stands at around 390 million tonnes, and this is expected to double by 2050 (PlasticsEurope, 2022). The environmental and biological implications of commonly used plastics are manifested through microplastics (MPs). These are water-insoluble particles between 1 μm and 5 mm, made up of synthetic or semi-synthetic polymers (Frias and Nash, 2019). Over the past decade, there has been a notable surge in research dedicated to understanding the effects of MPs on various organisms and evaluating pollution levels in both terrestrial and aquatic environments (Akdogan et al., 2023; Almas et al., 2022; Cao et al., 2022; Dissanayake et al., 2022; Kee et al., 2020; Sönmez et al., 2023). Extensive literature has unequivocally demonstrated that a wide range of animal species ingest microplastics (Gündoğdu et al., 2020; Aydın et al., 2023; Kankılıç et al., 2023; Porretti et al., 2023; Szkudlarek et al., 2023; Terzi, 2023; Burger et al., 2024). Ingesting microplastics can cause significant adverse effects, including reduced growth and reproduction and altered behaviour and physiology (Franzellitti et al., 2019; Araújo and Malafaia, 2020; Burgos-Aceves et al., 2022; Doyle et al., 2022). These microplastics, which accumulate in animal tissues, have the potential to be transmitted to human beings via food chain (Wu et al., 2019; Dong et al., 2023).

Although microplastics have received attention as an important pollutant in the scientific community over the last decade, insufficient attention has been paid to microplastic uptake in terrestrial environments (Hou and Rao, 2022). This represents a gap in the scientific understanding of microplastics, as it is highly likely that various terrestrial organisms ingest them (de Souza Machado et al., 2018; Mackenzie and Vladimirova, 2023). In addition, microplastics can migrate into the atmosphere and terrestrial environments through uncontrolled disposal sites, landfills, and various anthropogenic activities, resulting in significant soil and airborne contamination (Allen et al., 2022; Nizzetto et al., 2016; Wright et al., 2020). It has been reported that these accumulated microplastics in the soil can be transported by animals such as earthworms to other habitats (Rillig et al., 2017). These plastic-contaminated soils are accidently ingested by some terrestrial invertebrates at the bottom of the food chain, and can subsequently move up the food chain (De Souza Machado et al., 2018). Furthermore, these plastics can be

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ingested by species that would not consume them directly (Da Costa Araújo and Malafaia, 2021). Soil-associated invertebrates of taxa Formicidae, Coleoptera, and Orthoptera are among the prey of lizards, and varying levels of microplastics have been found in the stomach contents of these lizards, suggesting that they consume microplastics directly or through their diet (Mackenzie and Vladimirova, 2023).

Microplastic studies in reptiles were mostly focused on sea turtles (do Sol et al., 2011; Beckwith and Fuentes, 2018; Jung et al., 2018; Wilcox et al., 2018; Gündoğdu et al., 2019). Eastman et al. (2020) reported that 39 (92.86%) of the 42 juvenile sea turtles they analyzed had ingested plastic from the gastrointestinal tract and that plastic ingestion is a critical problem for sea turtles from the earliest stages of life. On the other hand, snakes, one of the main groups of reptiles, have been documented to be entangled in plastic bags (Sindha et al., 2020) and swallow plastic bottles (Vasaruchapong and Chanhome, 2013), bags (Deshmukh et al., 2017; Strine et al., 2014) and bottle caps (Lettoof and Orton, 2020). Lizards provide an excellent model to study microplastic uptake, as they can live in urbanized areas where microplastics are most abundant, often feeding on invertebrates (Tok et al., 2016; Silva et al., 2020). Knowledge of microplastic pollution levels in lizards is limited (Mackenzie and Vladimirova, 2023). Therefore, our study focused on the snake-eyed lizard, which has a wide distribution area including European, Asian, and African continents. The snake-eyed lizard, *Ophisops elegans* is the most common lizard in Türkiye (Akkaya and Uğurtaş, 2006). It is distributed in the Balkan countries, Aegean and Mediterranean islands, and southwest Asia with a vertical distribution up to 2000 m (Aghasyan et al., 2021).

The main objective of this study was to investigate the relationship between microplastic prevalence and human population density using a lizard species with a wide distribution in terrestrial habitats as a model organism. A further aim of this study was to ascertain the prevalence, types, shapes, and sizes of microplastics in the gastrointestinal tract of a lizard species. Hence, we investigated the prevalence of microplastics in the snake-eyed lizards living in densely populated and unpopulated areas in Türkiye. We hypothesized that lizards living near densely populated areas would have higher exposure to contaminants and that quantifying this would contribute to the understanding of anthropogenic plastic pollution. This knowledge contributes to developing of biomonitoring tools and paves the way for using of lizards as biosamples for microplastic pollution.

2. Materials and methods

2.1. Sampling and microplastic extraction

A total of 152 specimens of *Ophisops elegans* (105 males, 40 females, 7 juveniles) were used which were preserved at the Zoology Museum of Adıyaman University (ZMADYU) (Fig. 1, Table S1). In order to reveal the relationship between microplastic pollution and population density, the most common lizard species in Türkiye was selected as a model organism from the populated and un-populated regions. Sampling was performed with the permission of the local ethics committee (Dollvet-Hadyek, decision number: 2014/02) for animal experiments. *O.elegans* is a small lizard species with a maximum life span of seven years for females and eigth years for males (Altunışık et al., 2017). The majority of the food composition of the species is composed of insects (67%) and

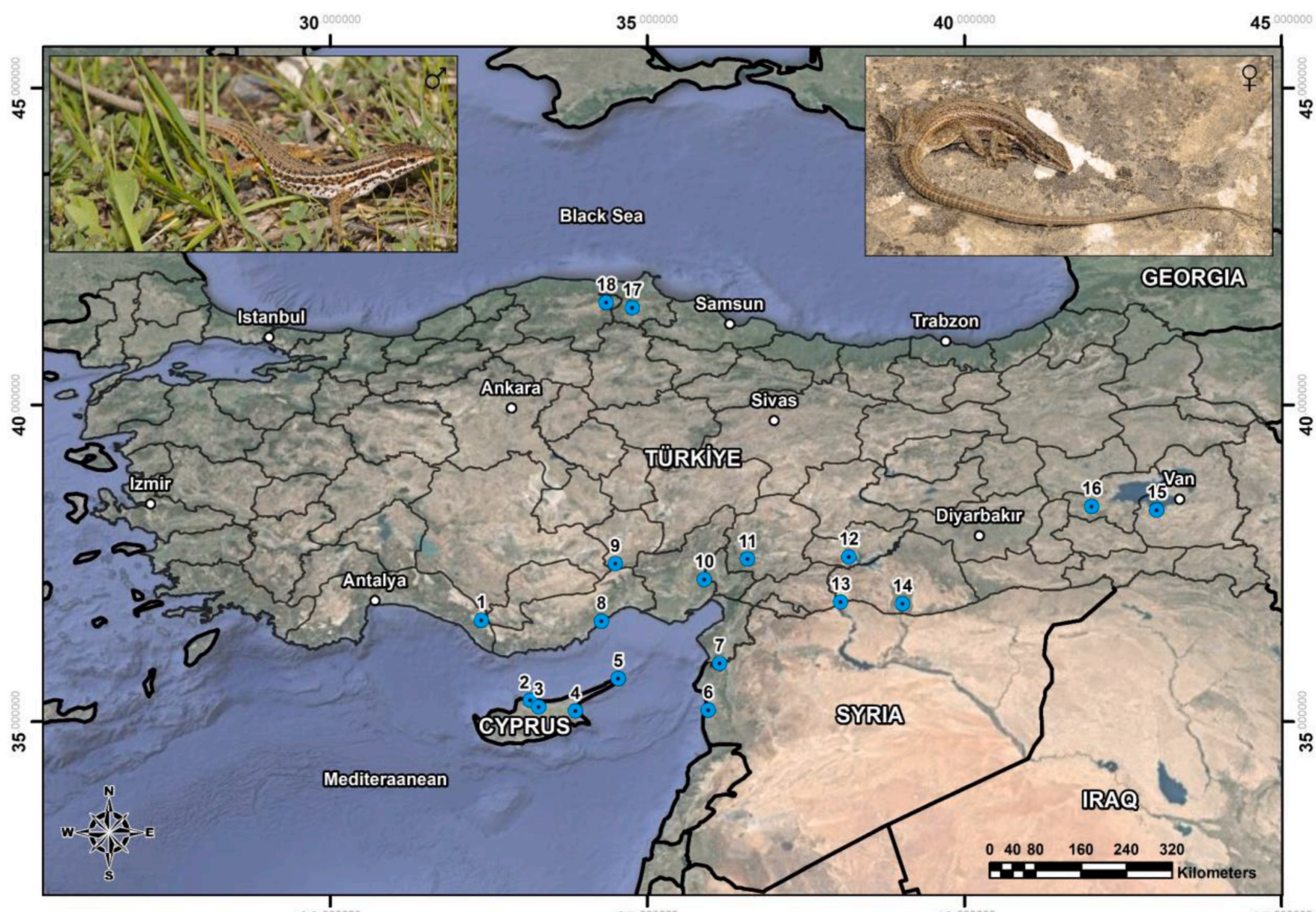

Fig. 1. Sampling locations in this study. The location numbers are the same as in Table 1.

Aranea (13%) (Tok et al., 2016). Sex was determined based on the presence or absence of a hemipenis. If a specimen had enlarged testes and/or epididymides in a male, or enlarged vitellogenic follicles and/or oviducts and/or elongated oviducts in a female, it was considered an adult (Altunışık et al., 2024).

Specimens preserved in glass jars filled with 96% ethanol underwent a first rinsing with distilled water before recording snout-vent length (SVL, mm) and weight (g) (Table S2). The gastrointestinal tract (GIT) of the specimens was then extracted using a stainless steel dissection tool and the weight of the GIT was measured. GITs were then transferred to separate flasks, and 150 ml of H_2O_2 (30%) was added. The flasks were covered with aluminum foil and left for 3 day at 65 °C with 80 rpm shaking. Two flasks from each batch received just H_2O_2 without the addition of GIT (referred to as "blank" samples), and the digestion process was carried out at the designated temperature and time in order to assure contamination control during the procedure. After the flasks were allowed to cool to room conditions (RC), the contents were filtered using a vacuum pump and a Whatman GF/C filter with a pore size of 1.2 μm and a diameter of 47 mm. Following the filtration process, the filters were placed on glass Petri plates and kept in RC (Tatlı et al., 2022).

2.2. Contamination control of microplastics

A robust set of precautions was implemented to ensure the utmost precision and mitigate the risk of MP contamination during the experimental phase. Of paramount importance among these precautions was the scrupulous upkeep of laboratory hygiene. Throughout the experiments, researchers consistently wore cotton lab coats and gloves devoid of any polymer materials. Furthermore, all liquid substances, including distilled water, H_2O_2, and ethanol, underwent meticulous filtration using Whatman GF/C filter paper with a pore size of 1.2 μm and a diameter of 47 mm. Moreover, lab equipment such as beakers, glass jars, aspirator bottles, flasks, and petri plates, crucial for the sampling process, were diligently cleansed with filtered water and safeguarded with aluminum foil before utilization (Tatlı et al., 2022).

2.3. Microscopic examination

A thorough examination of the filter papers was carried out to determine their classification as MP using a Leica S6D microscope. This determination was made on the basis of the physical properties as described in a previous study by Hidalgo-Ruz et al. (2012). We acknowledged that there may be errors associated with these visual assessments. Following a meticulous visual assessment, particles suspected to be MP were delicately transferred onto fresh filter papers using a needle. Subsequently, these particles were documented via photography using a digital camera (Leica S6D, Switzerland) affixed to the microscope, and their morphologies were utilized to categorize them into fiber or fragments. The application ImageJ (http://imagej.nih.gov/ij/) was employed to compute the length of the particles. To monitor for possible air contamination during microscopy, a separate clean Petri plate filled with appropriately filtered water was kept near the microscope throughout observations. Subsequent to the comprehensive analysis of all filter papers, the Petri plate, maintained as a blank control to detect MP contamination, underwent microscopic examination as well. The number of MPs (0.10 item filter^{-1}) detected in negative blank samples was subtracted from the total MP amount. The recovery efficiency (%) of MP extraction was recorded using three different polymers and the spiking approach using PE, PET, and PP particles. The MPs were introduced in blank glass flasks (containing 200 mL H_2O_2) and the blank process was followed. After filtration, the recoveries (%) for PE, PET and were 95%, 93%, and 96%, respectively.

2.4. Polymer verification

A PerkinElmer Spectrum 100 FT/IR (Fourier Transform Infrared) spectrophotometer equipped with an Attenuated Total Reflectance (ATR) instrument was used for sample characterization. The FTIR data collection involved 32 scans for each reading, spanning the range of 4000–650 cm^{-1}, with a resolution set at 2.0 cm^{-1} (Fig. 2). Subsequently, the acquired spectra were compared against FT–IR spectra from reference polymer materials and subjected to library searches using Spectrum Search Plus Software. Only particles showing a match of over 70% were deemed to be microplastics, provided the data aligned with available spectral information in the library (Terzi et al., 2022).

2.5. Data analysis

The presentation of microplastic data was in the form of the number of microplastics per individual across all stations. ANOVA was used for morphometric differences between locations and chi-square and t-tests were conducted to examine MP prevalence between locations. The Pearson correlation was used to demonstrate the association between the size/weight and the prevalence of microplastics in the GIT of the snake-eyed lizards. In order to investigate the relationship between the prevalence of microplastics and proximity to settlements (PTS), the locations were divided into two groups as near and far. Among these locations, those close to the settlement up to 2 km were classified as A, and those more than 2 km away were classified as B, as given in Table 1. The analyses were conducted using R Programming Language v4.3.1 (R Core Team, 2023) and data were visualized using the ggplot2 package (Wickham, 2016).

3. Results

3.1. General characterization of microplastics

129 particles suspected of being MP were analyzed by ATR-FTIR device and 41 particles (31%) were identified as synthetic polymers (Fig. 2). MPs were categorized into two separate groups based on their physical features: fiber or fragment (Fig. 3) The vast majority of these MPs were in the form of fiber (90%, n = 37), while a small number were in the form of fragments (10%, n = 4). Six different polymer compositions were evident in the identified MPs (Fig. 4). The materials are listed in order of prevalence as follows: Polyethylene terephthalate (PET) at 80.4%, Polyacrylonitrile (PAN) at 7.3%, Polypropylene (PP) at 4.9%, and Polyethylene (PE), Poly methyl methacrylate (PMMA) and Polyamide (PA) at 2.4%. The color (n = 6) of MPs found in the GIT of the lizards are as follows; navy blue (76%) > red (10%) > transparent (5%) = turquoise (5%) > brown (2%) = pink (2%) (Fig. 5). The lengths of MPs varied from 37 to 563 μm, with an average length of 175 ± 140 μm (Table 2).

3.2. MP distribution in populations

MPs were found in 17 out of 18 populations (94%) (Table 1). In these populations, 33 out of 152 GIT samples (21%) were found to contain MPs. The diagram also depicted relationships between different locations and using nodes to symbolize categorical factors (Fig. 3). The number of MP per individual in the populations varied between 0 and 0.71 (mean: 0.27) (Table 1). The Anavarza population had the highest prevalence of MPs at 5.71%, while the Taşköprü population had the lowest prevalence. Among the populations in which MP was detected, MP was found in 4 out of 7 lizards analyzed in Çiçekalan and it was recorded as the highest value with a rate of 57%. The Çiçekalan habitat is a location with a high human population density and a notable accumulation of plastic waste. On the other hand, Harran, where MP was detected in only one of 12 individuals among the populations where MP was detected, was the population with the lowest prevalence (n = 12, 8%) (Table 1). The habitat is located in an area away from the city center and human settlement. There is little evidence of plastic waste in the area. The studied populations (n = 18) differed from each other in

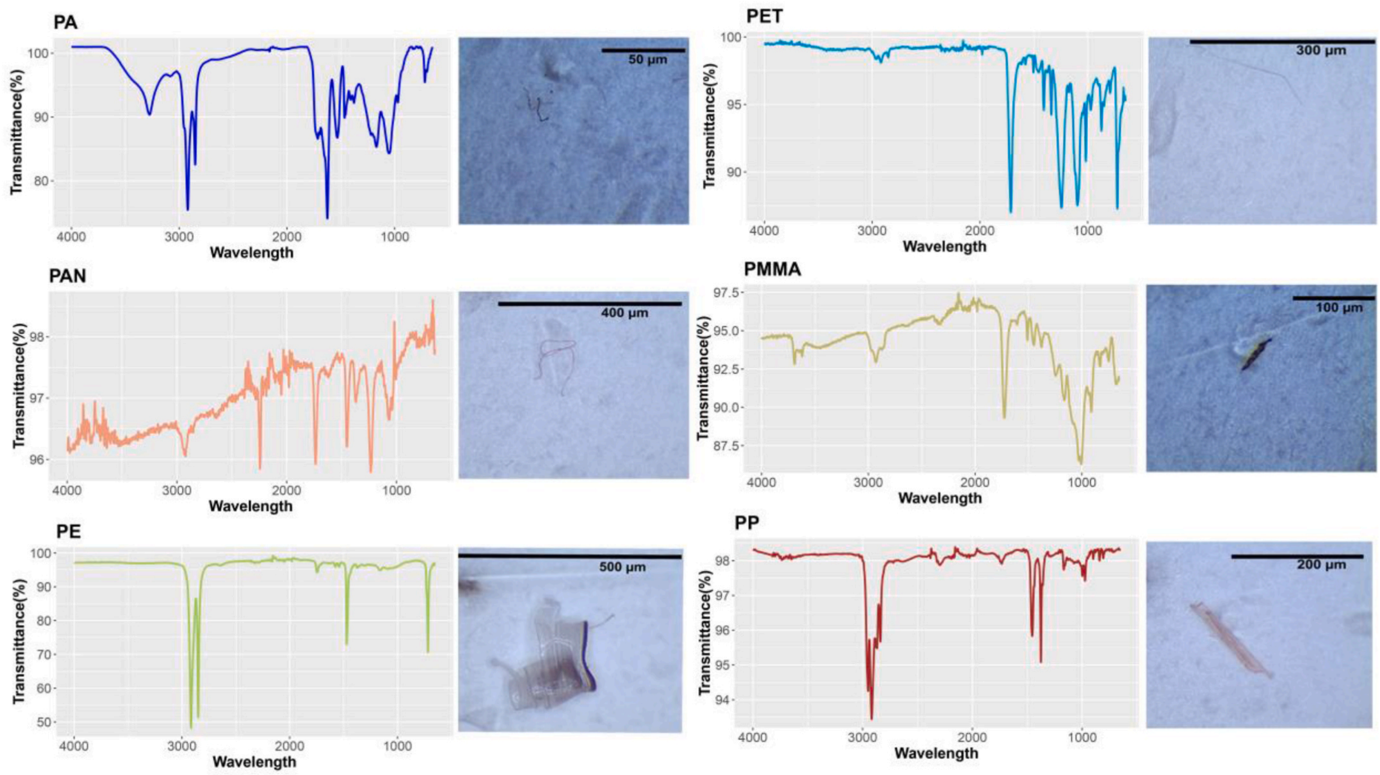

Fig. 2. FTIR spectrums and stereomicroscope photographs of microplastics detected in the sampled lizards.

Table 1
Data on lizards sampled from different locations in Cyprus, Türkiye and Syria and microplastics detected in their gastrointestinal tracts.

Locations' ID and PTS	Locations' Country	Locations' Name	Lizard (n)	Length (cm) Mean	Weight (g) Mean	[a]Lizard-MP	[b]FO (%)	[c]MP	[d]MP ind.	[e]MP ind·
1-B	Antalya/Türkiye	Kuşyuvası	3M-3F	41.70	1.93	1	17	1	1.0	0.17
2-B	Northern Cyprus	Lapta	7M-2F	45.99	2.85	1	11	1	1.0	0.11
3-B	Northern Cyprus	Gönyeli	5M-7F	41.69	1.80	2	16	2	1.0	0.16
4-B	Northern Cyprus	Magosa	9M-1F-2J	36.52	1.64	2	16	2	1.0	0.16
5-A	Northern Cyprus	Dipkarpaz	7M	44.90	2.69	2	28	2	1.0	0.28
6-A	Tartus/Syria	Tartus	6M-1F-1J	43.73	2.12	3	38	4	1.3	0.50
7-B	Hatay/Türkiye	Görentaş	4M-2F-1J	38.27	1.92	1	14	2	2.0	0.28
8-A	Mersin/Türkiye	Erdemli	7M-2F	44.29	2.65	3	33	3	1.0	0.33
9-A	Niğde/Türkiye	Ulukışla	8M-6F	40.43	1.73	4	28	4	1.0	0.28
10-A	Adana/Türkiye	Anavarza	7M	43.44	1.96	2	28	5	**2.5**	**0.71**
11-B	K.Maraş/Türkiye	Başkonuş	6M-2F	44.64	2.46	1	13	1	1.0	0.13
12-A	Şanlıurfa/Türkiye	Çiçekalan	4M-2F-1J	43.06	1.96	4	**57**	4	1.0	0.57
13-A	Adıyaman/Türkiye	Atakent	8M-1F-1J	45.33	2.13	3	30	**6**	2.0	0.60
14-B	Şanlıurfa/Türkiye	Harran	7M-5F	48.16	3.09	1	**8**	1	1.0	0.08
15-B	Bitlis/Türkiye	Konuksayar	7M-1J	42.48	1.94	1	13	1	1.0	0.13
16-B	Van/Türkiye	Akdamar	3M-1F	49.91	2.56	1	25	1	1.0	0.25
17-B	Sinop/Türkiye	Bektaş	3M-1F	47.46	2.87	1	25	1	1.0	0.25
18-B	Kastamonu/Türkiye	Taşköprü	4M-4F	46.01	2.41	0	0	0	0.0	**0.00**
		Total	**152**	**[e]43.77**	**[e]2.26**	**33**	**22**	**41**	**1.2**	**0.27**

PTS: Proximity to settlements; A: near, B: far; M: male, F: female, J: juvenile [a] The number of lizard MP detected; [b] Frequency of occurrence; [c] Detected total microplastic; [d] Per individual for lizard containing microplastic; [e] General average.

terms of MP per individual (Chi-square test: 25.333; df = 14; $p < 0.01$) and lizards residing near human settlements exhibited increased accumulation of microplastics (Independent sample *t*-test; t = 5.205; df = 16; $p < 0.001$).

In total, 41 identified microplastics were distributed across 8 diverse polymer types. Among the populations studied, the Atakent population stood out with the highest polymer diversity, as 4 different polymer types (PET, PMMA, PP and PA) were observed (Fig. 4). Among the lizards with MP presence, more than 80% were identified as PET, and this polymer type was identified in all 17 locations (Fig. 4). Upon analyzing the distribution of MPs in the samples, it is evident from Table 2 that each station where MPs were detected visibly contains at least one fiber. While fiber was identified as the predominant shape in 17 stations, fragments were also observed in the three stations. In 94% of the locations, at least one navy blue MP was found (Fig. 5). Besides, Anavarza, Atakent, Dipkarpaz, Gönyeli, Tartus, Ulukışla, and Çiçekalan are locations that also contain MP of non-navy blue colors such as brown, pink, red, transparent, and turquoise (Fig. 5). MP was detected in the GITs of 43% of juvenile individuals, 30% of male individuals, and 18% of female individuals, with mean 0.27 per specimen. MP was detected in male individuals in all populations except for Harran and Lapta (Fig. 5). In these two populations, all identified MPs were found in the GITs of

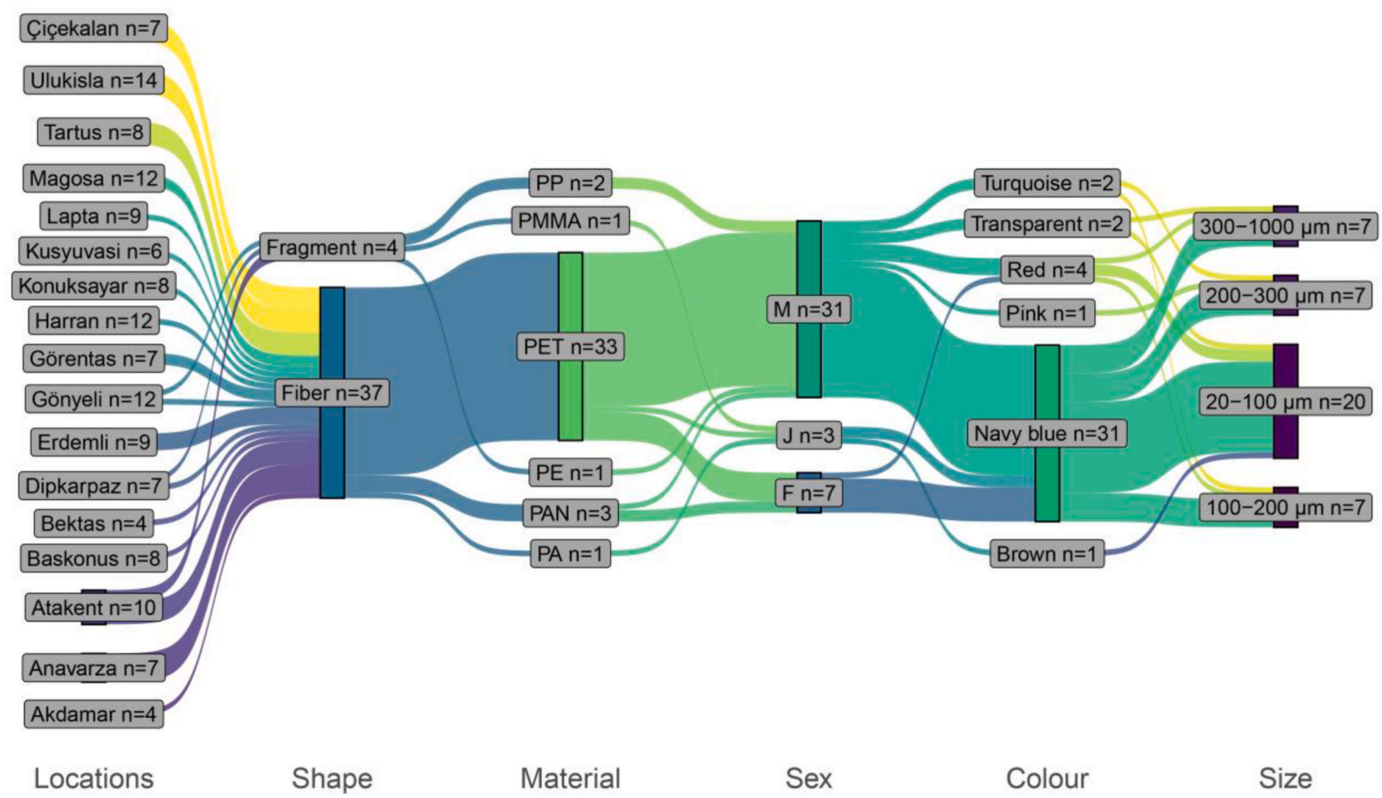

Fig. 3. A Sankey diagram illustrating the intricate relationships among the locations, shape, material, sex, colour and size. Nodes in flow were shaped by microplastic patterns. M: male, F: female, J: juvenile. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

female individuals. Besides, Atakent and Magosa were the populations where MP was detected in juvenile individuals (Fig. 5).

There were statistically significant differences in the average size and weight of specimens among populations (ANOVA, size: $F = 4.355_{17,142}$, $p < 0.05$; weight: $= 3.607_{17,142}$, $p < 0.05$). However, in analyzing the relationship between the morphological parameters (length-weight) of the lizard specimens and the number of MPs present in their gastrointestinal tracts, no statistically significant correlation was identified (Pearson correlation, size: $r = -0.218$; $df = 18$; $p = 0.385$; weight: $r = -0.297$; $df = 18$; $p = 0.231$). Additionally, the size of MPs and the length/weight of lizards were not significantly correlated (Pearson Correlation Coefficient; SVL: $r = 0.17$, $df = 41$, $p = 0.291$; weight: $r = 0.05$, $df = 41$, $p = 0.779$).

4. Discussion

The release of MPs into the environment is strongly influenced by human activity levels (Browne et al., 2011; Jambeck et al., 2015), and it is possible to observe variations in MP concentrations across different geographical regions (Ghosh et al., 2023). In this study, the significant difference in the prevalence of MPs between the sites sampled was one finding that confirmed this situation. The study found that the Anavarza population had the highest MP per individual, while no MP was detected in the Taşköprü population, which had the lowest. As Anavarza is a residential area and a popular tourist destination, it is recognized that people leave a lot of plastic waste there. In contrast, MP was not detected in any of the lizards inhabiting the Taşköprü population, which is located in a natural environment far from settlements. In general, MPs were detected in 33 out of 152 specimens analyzed, accounting for approximately 22% of the snake-eyed lizards. On the other hand, in a preliminary study of microplastic pollution in lizards, researchers found MPs in 12% of GIT samples from *Hemidactylus mabouia* and 6% from *Tropidurus torquatus* living in southwestern Paraguay (Mackenzie and Vladimirova, 2023). The researchers also analyzed the stomach contents of an amphibian (*Rhinella diptycha*) in the region and found that a higher proportion of specimens (48%) were exposed to microplastics in amphibians than in lizards. Additionally, dried lizard samples used as traditional medicinal material in China were found to contain high levels of MP (Lu et al., 2020). In a study on water frogs, the presence of microplastics was reported in 82% of the 176 specimens analyzed (Tatlı et al., 2022). Furthermore, MP was detected in all five (100%) European common frogs analyzed in the Cottian Alps (Pastorino et al., 2022). In light of the above studies, amphibians and lizards have different prevalences of microplastics. Many factors can influence this situation, such as the pollution of the area, the type of diet of the animal, or the size of the animal.

Several studies have underscored the link between intense anthropogenic activities and increased levels of microplastic pollution in various geographical regions (Jang et al., 2020; Kee et al., 2020; Lin et al., 2021; Liu et al., 2021). Accordingly, we have observed that there is a difference between locations in terms of MP per specimen, more in areas where human activities are intensive. Our findings demonstrate significant distinctions in the mean length and weight of specimens across different populations. Nevertheless, no association was established between the morphological parameters of lizards and the presence of MPs in their gastrointestinal tract.

In this study, we also evaluated the impact of age on MP levels in the gastrointestinal tracts of young and adult specimens. It was observed that juveniles with smaller stature had more MPs in their GITs than adults. A similar result was reported for a frog species, *Rhinella diptycha*, where juvenile *R. diptycha* were found to be 1.34 times more likely to ingest microplastics than adult *R. diptycha* (Mackenzie and Vladimirova, 2023). In contrast, the adult *Hemidactylus mabouia* lizard ingested 6.23 times more microplastics than the juvenile *H. mabouia.* On the other hand, a critical gap in the existing scientific literature on the effect of sex on microplastic exposure in animals was highlighted by Prokić et al.

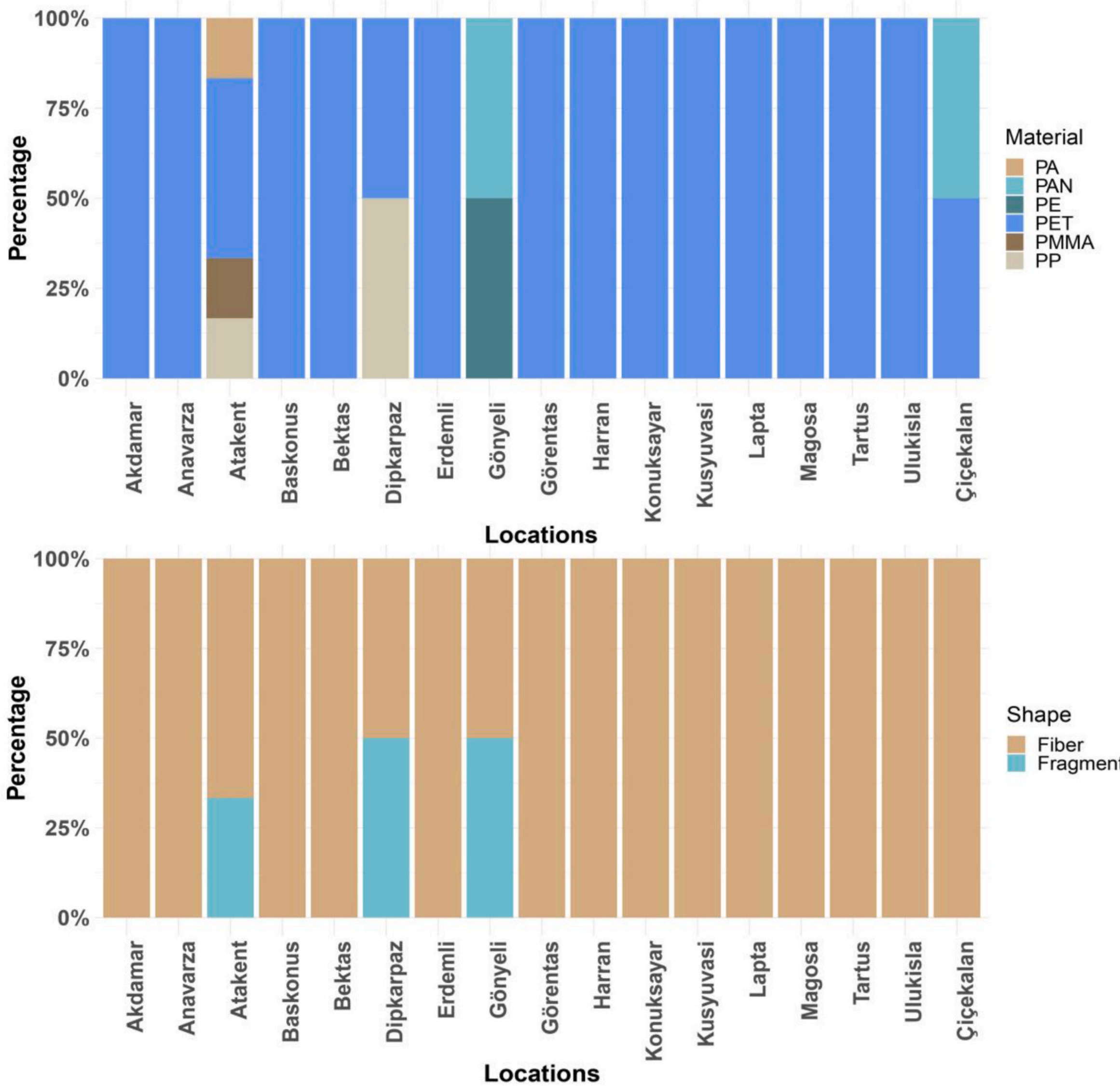

Fig. 4. A stacked bar graph illustrating the proportional distribution of material and shape within microplastics in the snake-eyed lizards.

(2021). For example, research on sharks (Alomar and Deudero, 2017; Bernardini et al., 2018), Norway lobster (Murray and Cowie, 2011), and common periwinkle (Doyle et al., 2019) has not revealed any sex-related differences in microplastic intake. However, investigations into wild fish and crustacean populations suggest that females tend to accumulate higher levels of MP compared to males (Bordbar et al., 2018; Mcgoran et al., 2018; Su et al., 2019; Welden and Cowie, 2016). In those studies, researchers speculated that the higher percentage of plastic ingestion found in females was probably related to higher feeding activity and energy demands prior to the reproductive period (Cartes et al., 2008). In addition, the fact that females are generally larger than males in both size and weight and have different feeding behaviors (e.g. females foraging at greater depths in fish) has been reported as another possible explanation (Prokić et al., 2021). In sex-based assessment, the number of microplastics per specimen detected in the gastrointestinal tract of the snake-eyed was higher in males than in females in this study. This difference may be due to the higher number of male specimens (105 vs. 40 individuals) in our study compared to females.

It is proposed that the presence of smaller MP particles may result in the potential for accumulation within lower trophic levels of the food chain (Wang et al., 2021), thereby influencing their retention time within organisms (Yu et al., 2021). The dimensions of microplastics carry significant implications, particularly regarding their capacity, the smaller variants, to amass within lower trophic levels, thereby influencing their retention time within organisms The previous study on the lizards *H. mabouia* and *T. torquatus* did not provide any information on the size of microplastics (Mackenzie and Vladimirova, 2023). However, in the snake-eyed lizard, the average size of the observed microplastics was 175 μm.

In accordance with previous studies (Hu et al., 2022; Wayman et al., 2024; Wong et al., 2020), 83% of microplastics exhibited a length below 300 μm, with the most prevalent length range being 20–100 μm (49%). This was lower than the range of 100–150 μm found in previous studies on adult frogs by Pastorino et al. (2023) and Tatlı et al. (2022). Lima et al. (2000) examined three frog and three lizard species living in the same region in terms of food composition and found that frogs eat smaller prey than lizards with the same gape size. It was also reported that the prey of frogs and lizards differed and frogs ate more amphipods, mites, and ants than lizards, while lizards ate more termites, centipedes, isopods, and orthopterans than frogs (Lima et al., 2000). A prior research found that most of the snake-eyed lizard's food was insects, with sizes ranging from 2 to 15 mm (Tok et al., 2016). In this context, the widespread detection of small microplastics in the GIT samples examined in our study can be explained by the lizard's lifestyle habits and feeding

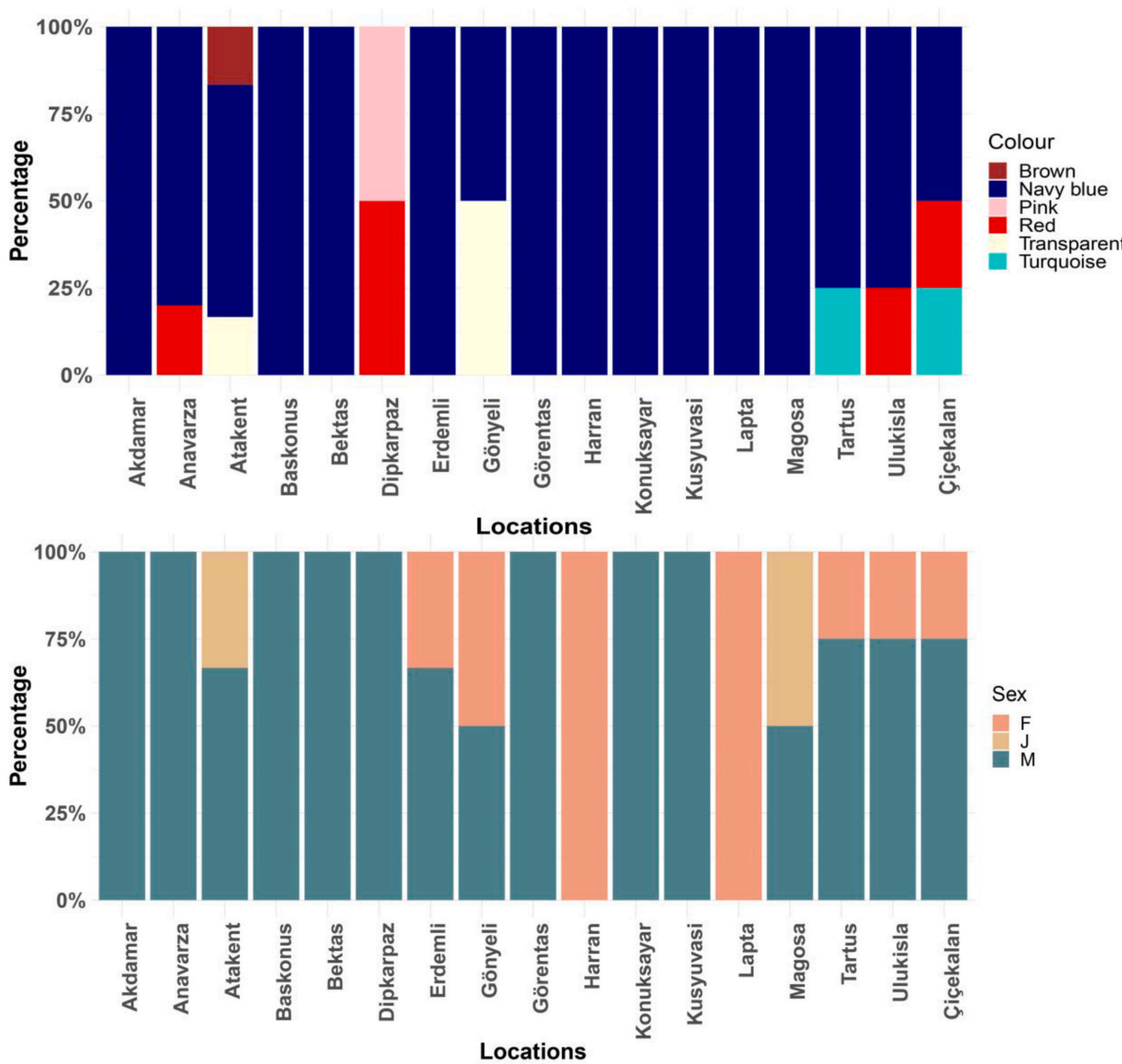

Fig. 5. A stacked bar graph illustrating the proportional distribution of colour and sex within microplastics in the snake-eyed lizards. M: male, F: female, J: juvenile. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

Table 2
Size and shape distribution of microplastics detected in the gastrointestinal tract of the snake-eyed lizards.

Location Number	Location Name	Mean MP Size (μm)	MP Shape	
			Fiber	Fragment
1	Kuşyuvası	88	+	–
2	Lapta	563	+	–
3	Gönyeli	383	+	+
4	Magosa	67	+	–
5	Dipkarpaz	140	+	+
6	Tartus	210	+	–
7	Görentaş	62	+	–
8	Erdemli	244	+	–
9	Ulukışla	126	+	–
10	Anavarza	160	+	–
11	Başkonuş	179	+	–
12	Çiçekalan	252	+	–
13	Atakent	122	+	+
14	Harran	74	+	–
15	Konuksayar	179	+	–
16	Akdamar	74	+	–
17	Bektaş	84	+	–
18	Taşköprü	–	–	–
Total		175 μm		

behavior characteristics.

The predominant microplastic form among the analyzed samples was fiber, with fragments constituting a smaller proportion (90% vs. 10%). The fiber was the primary component observed at all locations and was consistent with findings from previous studies on lizards (Mackenzie and Vladimirova, 2023), adult frogs (Pastorino et al., 2022; Shetu et al., 2023; Tatlı et al., 2022) and tadpoles (Hu et al., 2018; Kolenda et al., 2020). It is well-documented that fiber is the most common microplastic form in the environment (Altunışık, 2023; Browne et al., 2011; Çevik et al., 2022). It is known that these fibers originate from different products such as textiles, fabrics, and carpets, which are commonly used products and are shed both during use and washing (De Falco et al., 2018; Hernandez et al., 2017). Considering the aforementioned details, it is presumed that the fibers identified at the sampling sites could potentially originate from synthetic clothing. A possible explanation for this is that snake-eyed lizards can also dwell in populated regions. This hypothesis is corroborated by the observation that the number of microplastics identified in lizards residing in proximity to human settlements was greater than in lizards inhabiting more isolated regions.

In our study, we detected six different types of polymer, the most common being PET at 80% (Fig. 3). PET is a widely utilized polymer in the production of water and soft drink bottles, as well as in some cleaners and fabrics (PlasticsEurope, 2022). Furthermore, PET and PE were often the most common polymer types reported in Europe (Cera et al., 2020). The presence of PET-based plastics in 16 of the 18 locations examined in this study (Fig. 4) is indicative of the frequent use of this polymer in these regions and its subsequent release into the environment. We could not conduct a comparison of these results, as the prior study on lizards solely focused on characterizing microplastics in terms of their shape and color. However, there are records in the literature reporting the common polymer type as polyamide (PA) (Pastorino et al., 2022), nylon (Kolenda et al., 2020), Polystyrene (PS) (Shetu et al., 2023) or Polyester (PES) (Wayman et al., 2024) in studies on amphibians and avians. The common aspect of these studies was that they had a lower polymer diversity compared to our study. The presence of microplastic pollution in terrestrial ecosystems is intricately tied to multiple factors, such as the widespread usage of plastic products, the efficacy of waste management practices, and the deposition of plastic waste carried by precipitation and wind. The concentration of microplastics in terrestrial environments sees a notable surge due to intensive plastic consumption and inadequate waste management systems, particularly prominent in urban settings, as highlighted by Wong et al. (2020). This hypothesis is supported by the fact that the habitats of the lizards analyzed in this study are generally located in the vicinity of human settlements.

GITs from different sites varied in MP intensity and six different MP colors were identified (Fig. 5). It is noteworthy that the dominant color observed in these MPs was navy blue. On the other hand, in the lizard *H. mabouia*, the dominant microplastic colors were reported to be colorless and blue. Furthermore, the dominant color of microplastics in some fish (Pereira et al., 2023), frogs (Kolenda et al., 2020; Tatlı et al., 2022), and birds (Carlin, 2019) species was shown to be navy blue. Although the prevalence of blue-colored microplastics ingested by marine organisms can be attributed to the resemblance of plastics to their prey (Ory et al., 2017; Rios-Fuster et al., 2019), this cannot be extrapolated to lizards, which are terrestrial organisms. The greater visibility of colored plastics, compared to transparent ones, may have contributed to their higher percentages. Frogs and lizards have different life histories and feeding habits. Generally, lizards can swallow larger foods, while frogs use their tongues to hunt insects (Huo and Rao, 2022). The ingestion of microplastics by animals can take various forms, either directly through primary consumption or indirectly through prey consumption (e.g. trophic transfer from tadpoles to fish and from them to mice) or through the gills if aquatic (De Souza Machado et al., 2018; Da Costa Araújo et al., 2020). In this respect, we do not know whether the microplastics found in our samples were ingested by the lizards through food or accidently. Further experimental and field studies are needed to confirm this.

5. Conclusions

In this study, microplastic accumulation in a lizard species has been evaluated in terms of shape, size, polymer type, and color. In addition, the question of whether microplastic intake varies depending on age was also answered. Juveniles had a higher concentration of microplastics in their gastrointestinal tracts compared to both males and females. Of the total populations analyzed, 94% were found to contain microplastics. Small-sized microplastics were the most common type of plastic found in the gastrointestinal tract of lizards, with fiber and navy blue being the most common shapes and colors, respectively. It is also assessed that the density of microplastics varies with the distance of habitats from human settlements, contributing to the hypothesis that there is a correlation between intensive anthropogenic activities and increased levels of microplastic pollution.

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Competing interest declaration

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.

CRediT authorship contribution statement

Abdullah Altunışık: Writing – review & editing, Writing – original draft, Visualization, Supervision, Methodology, Investigation, Formal analysis. **Mehmet Zülfü Yıldız:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Investigation, Funding acquisition, Data curation, Conceptualization. **Hatice Hale Tatlı:** Software, Methodology, Investigation, Formal analysis, Conceptualization.

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.

Data availability

No data was used for the research described in the article.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2024.124754.

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