Exploring microplastic pollution in the pristine Ghar-e-Tangi Cave: first evidence from Pakistan's subterranean ecosystem

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Abstract: Microplastics (MPs) are a pervasive environmental pollutant, yet their presence in subterranean environments, particularly in remote locations, remains understudied. This study quantifies the abundance, distribution, and characteristics of MPs in the sediments of Ghar-e-Tangi, an isolated cave in Balochistan, Pakistan, and explores their potential input pathways. Triplicate sediment samples were collected from three distinct sites along a gradient from entrance to deeper sections. MPs were extracted and analyzed for their number, color, size, and shape using microscopy, while MPs ≥ 1 mm were characterized via Fourier transform infrared spectroscopy (FTIR). Significant differences were observed in the distribution of MP by shape (P=0.004), color (P=0.002), and size (P=0.005), as well as across the sites (P=0.001–0.041). MP abundance decreased significantly from the entrance to deeper sections (Site A=99 MPs/kg, Site B=49 MPs/kg, Site C=37 MPs/kg, P=0.001). Transparent MPs were predominant (38.4%, 23.67±10.97), along with eight identified colors. Of the five shapes identified, fragments were the most prevalent (36.8%, 22.67±11.72). MPs measuring >3-5 mm constituted the largest proportion (45.4%, 28.0±14.42). The observed gradient suggests surface runoff and atmospheric deposition as primary input pathways. These findings align with global studies, highlighting the pervasive nature of MPs, even in remote environments such as the Ghar-e-Tangi cave.

Keywords: microplastic pollution, cave environment, remote ecosystems, input pathways

INTRODUCTION

Natural geological processes and human-induced physicochemical alterations play a comparable role in regulating ecosystem functioning [1]. Anthropogenic drivers of change pose a major threat to ecosystem stability and have devastating effects on biodiversity [2]. The ubiquitous presence of synthetic polymer plastics is one of the major indicators of human activity worldwide [3]. These plastics are chemically diverse and are widely used. One of the significant characteristics of synthetic polymers is their high malleability throughout different stages of production, including raw materials, the manufacturing process, and product stages. The diverse polymers are further processed and modified physically (e.g., pellet formation, extrusion, blowing, melting) as well as chemically (e.g., mixed with dyes, plasticizers, clarifiers, bleaches, co-polymers, and other chemicals such as polycarbonates) [4]. These modifications produce advanced materials that offer numerous benefits, such as exceptional durability, affordability, malleability, and flexibility. Such benefits have driven a steady increase in plastic production with annual production reaching 400.3 million tons in 2022, 1.6% higher than in 2021 [5].

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Plastics are a major environmental and health concern because they typically do not biodegrade but instead fragment into smaller particles, forming microplastics (<5 mm) [6]. The minimum size of MPs is not strictly defined. Mesh sizes of 0.33 mm or 0.18 mm are usually used for sample collection [7]. Sources of MP pollution include industrial effluents, surface runoff, wastewater, and improper waste disposal [8]. The small size of MPs makes them a common source of marine, freshwater, and terrestrial contamination. MPs serve as vectors for toxic chemical transport into oceans, air, and soils because of their adsorptive nature. These tiny particles are ingested by biota, causing reduced food intake, developmental disorders, and behavioral changes [9]. Recent research indicates that MPs can disrupt microbial communities in soil and water, potentially affecting ecosystem functions [10,11]. Additionally, studies show that MPs accumulate in the food chain, posing risks to human health [12,13].

Most MPs in the marine ecosystem are manufactured, used, and discarded on land [14-16] where they undergo various physical and chemical changes, eventually fragmenting through environmental processes [15]. Consequently, the first interaction of MPs with biota often occurs in terrestrial environments, where they contribute to environmental toxicity. However, scientific investigations on MPs in the terrestrial environment lag behind studies on aquatic environments. MP pollution on land is estimated to be 4-23 times higher than in oceans [17]. Agricultural lands alone might hold more MPs than ocean basins [16]. A considerable portion of the 8.3 billion metric tons of plastic produced since the 1950s [18] is likely present as MPs in various terrestrial locations, including caves. Recent assessments have found significant MP contamination in soils, particularly in agricultural fields heavily affected by wastewater irrigation and plastic mulch films [19,20]. Studies have identified MPs in groundwater, raising concerns about their impact on drinking water resources [21,22].

Microplastics are ubiquitous and can infiltrate virtually every environment. Recent studies have detected MPs in the Arctic and Antarctic, underscoring their global reach and scale of contamination [23,24]. Additionally, the presence of MPs in the atmosphere shows their potential for long-distance transport through air currents [25]. Microplastics have even been detected in human blood and lungs, raising significant public health concerns [26,27].

Studies on MPs in subterranean ecosystems and pristine caves have revealed notable contamination levels. For instance, water and sediment samples from the Karst Region (Kras) in Italy showed MP contamination ranging from 37 to 86 MPs/L in water and 776 to 2064 MPs/kg in sediments [28]. Karst systems near tourist routes exhibited higher contamination levels than the less accessible speleological areas [29]. Fibers were the most common type of MP, with polyesters and polyolefins being the primary polymers. Most MPs were smaller than 1 mm [28-30]. These findings highlight the importance of comprehensive monitoring and investigating input pathways to implement conservation strategies in karst systems to safeguard biodiversity, preserve habitats, and protect water resources.

Ghar-e-Tangi, located in the remote area of Balochistan, Pakistan, is a rarely explored cave with a difficult, narrow, and dangerous passage. This study aims to quantify MP pollution in the sediments of Ghar-e-Tangi Cave, marking the first report of its kind from Pakistan. Given the increasing evidence of MPs in various environments, it is crucial to investigate their presence in secluded and less disturbed areas to understand the full extent of this pollution. Moreover, understanding MP contamination in such remote and rarely studied areas can provide valuable insights into the spread and impact of these pollutants, which is essential for developing effective mitigation strategies.

MATERIALS AND METHODS

Ethics statement

This study was conducted in accordance with ethical research guidelines, ensuring minimal environmental impact on the Ghar-e-Tangi cave ecosystem. Sediment samples were collected using non-invasive techniques, ensuring that only a minimum amount of material necessary for analysis was taken to preserve the natural state of the site. No flora, fauna, or other ecological components were harmed or disturbed during the fieldwork. Given the cave's isolation, formal permissions for site access were not required. The collected data is solely intended to advance environmental science and support the development of effective environmental protection policies.

Description of Ghar-e-Tangi Cave

Location

Ghar-e-Tangi Cave is located about 900 km southwest of Islamabad, Pakistan, in the Khuzdar district of Balochistan Province. It is situated in the mountains of Sasol Valley, east of Khuzdar, at 27.930299°N, 66.761773°E (Supplementary Fig. S1). The cave lies in a remote area at a high elevation, with no established access routes. Sasol Valley is known for its rugged terrain and sparse population.

Physical characteristics

Ghar-e-Tangi is a natural limestone cave formed in the rugged mountains of the valley. It extends several hundred meters deep into the mountain, with hazardous conditions that require personal protective equipment (PPE). The cave floor sediments and walls near the entrance are dry; however, conditions gradually become wetter deeper into the cave, culminating in muddy sediments in the innermost sections. The weather at the entrance is moderate and windy.

Accessibility

Accessing the cave is difficult, involving navigating through rugged terrain and climbing cliffs. Only a few local hikers and tourists, often with the help of a local guide, reach the site. The cave entrance is narrow, and the only entry path involves crawling, which limits human visitation (Supplementary Fig. S1). The challenging landscape and lack of formal paths contribute to limited human presence.

Fauna

The cave hosts various species, including bats, millipedes, cockroaches, and snakes. Due to the limited human activity, these species are relatively undisturbed.

Sampling locations

Sediment samples for microplastic pollution were collected from three sites within the cave (Supplementary Fig. S1). Site A was located at the entrance to the cave. Site B was about 200 m into the cave, accessible only through a narrow, crawling passage. Site C was about 400 m into the cave, where the walls were damp and the floor sediments muddy.

Sampling and sample preparation

Sediment samples were collected from the upper 2.5 cm of the cave floor within a 30 cm² quadrat using a small shovel and stored in sterilized glass jars. Three replicate samples were taken from each site. Four hundred g of sediments were placed into an 800-mL beaker and oven-dried at 60°C overnight. The samples were then sieved through a 5-mm mesh, and the resulting material underwent density separation.

Density separation and sieving

An aqueous lithium meta-tungstate solution (300 mL) was added to the dried sediments in the beaker, and the mixture was vigorously stirred with a spatula for several minutes until microplastics (MPs) floated to the surface. The floating material was passed through a 0.18-mm custom sieve, and the retained solids were transferred to a 500-mL beaker and oven-dried at 60°C. The dried solids were then weighed.

Digestion and density separation

To digest organic matter, 20 mL of 0.05 M Fe solution and 20 mL of 30% H_2O_2 were added to the solids in the beaker and heated to 60°C on a hotplate with a magnetic stirrer. The reaction was carefully monitored to prevent overflow, with 10-20 mL of additional H_2O_2 added as needed until the organic material was completely oxidized. To float the MPs, 6 g of NaCl per 20 mL of the solution was added and the mixture was heated to 75°C until the salt dissolved. The solution was transferred to a density separator and allowed to settle overnight. The floating solids were collected with a 0.18-mm sieve, and the separator was rinsed with distilled water to ensure all solids were transferred to a 0.3-mm sieve. The sieve was air-dried before microscopic analysis.

Quality control

To prevent contamination with plastic, all sampling and analytical steps were carried out using non-plastic equipment. Reagents and distilled water were prefiltered using a 1-μm glass filter. The apparatus was thoroughly cleaned with prefiltered distilled water and covered with aluminum foil when not in use. A dedicated, contamination-free laboratory area was used for processing the samples. Personnel involved in the study were prohibited from using plastic or synthetic fiber items (e.g., clothing, cosmetics, and accessories). To detect potential contamination, three procedural blanks were run in parallel with the sediment samples from each site. These blanks consisted of wet filter papers sealed in aluminum foil that were exposed to the cave environment during sampling and processed alongside the samples. Any false-positive MPs identified in the blanks were subtracted from the sample counts.

Microscopic and FTIR analysis

The MPs retained on the 0.18-sieve were identified based on shape, color, and size using a light microscope at 40× magnification. MPs >1 mm were extracted using forceps and transferred to a vial for further analysis. The polymer type of the MP was identified using Fourier transform infrared spectroscopy (FTIR) with an Agilent Cary 630 instrument, operating in the 4000-500 cm^{-1} wavelength range. The spectra were compared with the Agilent Polymer Handheld ATR Library, and a minimum recognition threshold of 70% was applied for polymer identification.

Statistical analysis

Statistical analyses were performed using SPSS to compare MP abundance, shape, size, and color across the three sampling sites (Sites A, B, and C). For normally distributed data, one-way analysis of variance (ANOVA) was used to compare means, with Tukey's post hoc test for pairwise comparisons. The Kruskal-Wallis's test was used for non-normally distributed data, and Dunn's test and Bonferroni correction for post hoc pairwise comparison. A two-way ANOVA was also conducted to examine the interaction between sampling sites and MP categories. The significance level (α) was set at 0.05.

RESULTS

MP abundance and distribution

A total of 185 MP particles were recorded across the three sampling sites, ranging from 37 to 99 particles per site. There was a clear contamination gradient with particle abundance decreasing deeper into the cave. The highest concentration was observed at site A (the cave entrance, 1 m depth), where 99 MPs/kg of sediment were found, accounting for 53.5% of the total MP count. This high concentration is likely due to plastic pollution carried by the wind and trapped at the entrance. At site B (200 m depth), the average concentration was 49 MPs/kg, representing 26.5% of the total count, and at site C (400 m depth), the average was 37 MPs/kg, comprising 20% of the total. This distribution indicates a clear reduction in MP concentration with increasing depth. Pairwise comparisons of MP characteristics (shape, color, and size) across the three sites showed statistically significant differences (Table 1). For shape, ANOVA (Tukey's Test) revealed significant differences between sites A and C (P=0.001), site A and site B (P=0.012), and site B and site C (P=0.034). For color, Kruskal-Wallis (Dunn's Test) revealed significant differences between sites A and C (P=0.008), sites A and B (P=0.025), and sites B and C (P=0.041). For size, ANOVA (Tukey's Test) indicated significant differences between sites A and C (P=0.003), sites A and B (P=0.017), and sites B and C (P=0.028).

Table 1. Pairwise comparisons of microplastic parameters (shapes, colors, and sizes) across three sampling sites (Site A, Site B, and Site C)

Parameter	Comparison	Test Used	P	Effect Size
Shapes	Site A vs. Site B	ANOVA (Tukey's Test)	0.012	0.45
	Site B vs. Site C	ANOVA (Tukey's Test)	0.034	0.38
	Site A vs. Site C	ANOVA (Tukey's Test)	0.001	0.62
Colors	Site A vs. Site B	Kruskal-Wallis (Dunn's Test)	0.025	0.50
	Site B vs. Site C	Kruskal-Wallis (Dunn's Test)	0.041	0.42
	Site A vs. Site C	Kruskal-Wallis (Dunn's Test)	0.008	0.58
Sizes	Site A vs. Site B	ANOVA (Tukey's Test)	0.017	0.40
	Site B vs. Site C	ANOVA (Tukey's Test)	0.028	0.35
	Site A vs. Site C	ANOVA (Tukey's Test)	0.003	0.57

Shapes

A total of five types of MP shapes were identified (Table 2). Fragments were the most common (mean 22.67±11.72, 36.8%), followed by globular particles (mean 14.0±8.89, 22.7%), with smaller proportions of fibrils, sheets, and pellets (Supplementary Fig. S2). The distribution of MP shapes was statistically significant (P=0.004), and significant differences

Table 2. Shapes of MPs (number/kg of sediment) found in Ghar-e-Tangi Cave and their variance among the sites and shapes

Parameters		Stations			Total	%age	Descriptive Statistics		
		Site A	Site B	Site C			Mean	SD	SE
Shapes	Fragments	36	18	14	68	36.7	22.67	11.72	6.77
	Sheets	19	7	4	30	16.2	10.00	7.94	4.58
	Fibrils	14	11	9	34	18.4	11.33	2.52	1.45
	Pellets	6	2	3	11	5.9	3.67	2.08	1.20
	Globular	24	11	7	42	22.7	14.00	8.89	5.13
Descriptive Statistics	Total	99	49	37	185	-	-	-	-
	Mean	19.8	9.8	7.4					
	SD	11.23	5.89	4.39	-	-	-	-	-
	SE	5.02	2.63	1.96	-	-	-	-	-
TWO-WAY-ANOVA	**Source**	**DF**	**SS**		**MS**		**F Statistic**		**P value**
	Factor A-rows	4	573.3333		143.3333		7.7758 (4,8)		0.007326
	Factor-B-columns	2	432.5333		216.2667		11.7324 (2,8)		0.004179
	Error	8	147.4667		18.4333		-		-
	Total	14	1153.3333		82.381		-		-

SD – standard deviation, SE – standard error, DF – degree of freedom, SS – sum of squares, MS – mean square

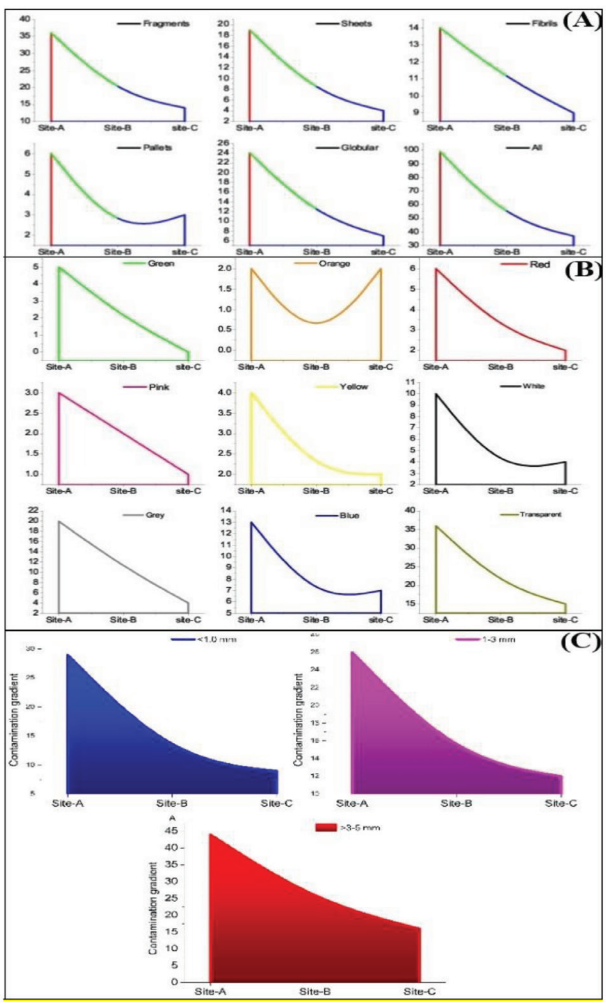

Fig. 1. Contamination gradient of microplastics in Ghar-e-Tangi Cave from 1 m to 400 m depth. **A** – Concentration gradients of different shapes as numbers/kg of sediments. **B** – Concentration gradient of different colors as numbers/kg of sediments. **C** – Concentration gradient of various size fractions as numbers/kg of sediments.

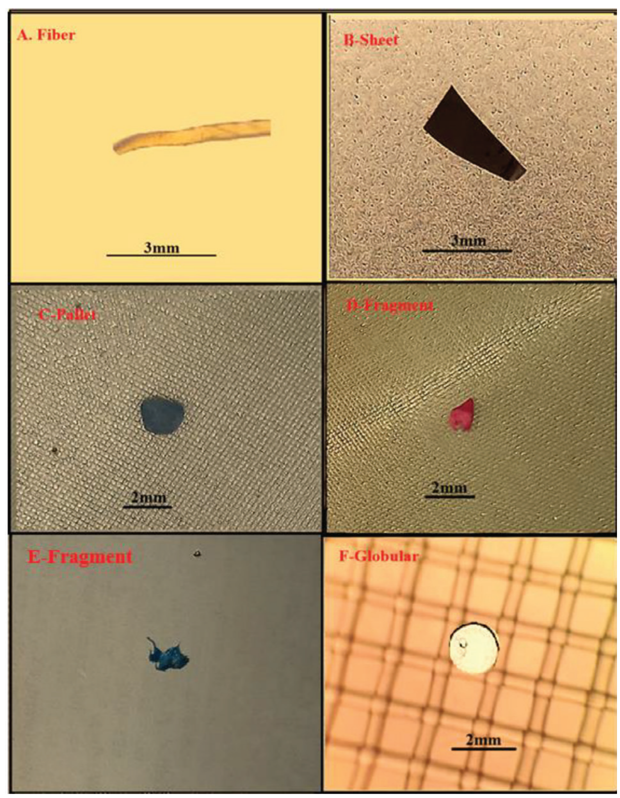

Fig. 2. Micrograph of some MP shapes found in Ghar-e-Tangi Cave.

were noted in the distribution of shapes between sites (P=0.007). Fibrils accounted for 18.4%, sheets for 16.2%, and pellets for 5.9%. A clear concentration gradient from site A to C was observed for all shapes (Figs. 1 and 2).

Table 3. Color diversity of MPs (numbers/kg of sediment) found in Ghar-e-Tangi Cave and their variance among the sites and colors

Parameters		**Stations**			**Total**	**%age**	**Descriptive Statistics**		
		Site-A	Site-B	Site-C			Mean	SD	SE
Colors	Green	5	2	0	7	3.8	2.33	2.52	1.45
	Orange	2	0	2	4	2.1	1.33	1.15	0.67
	Red	6	3	2	11	5.9	3.67	2.08	1.20
	Pink	3	2	1	6	3.2	2.00	1.00	0.58
	Yellow	4	2	2	8	4.3	2.67	1.15	0.67
	White	10	3	4	17	9.2	5.67	3.79	2.19
	Grey	20	11	4	35	18.9	11.67	8.02	4.63
	Blue	13	6	7	26	14.1	8.67	3.79	2.19
	Transparent	36	20	15	71	38.4	23.67	10.97	6.33
Descriptive Statistics	Total	99	49	37	185	-	-	-	-
	Mean	11	5.44	4.22					
	SD	10.98	5.73	4.05	-	-	-	-	-
	SE	3.66	1.91	1.35	-	-	-	-	-
TWO-WAY-ANOVA	**Source**	**DF**	**SS**		**MS**		**F Statistic**		**P**
	Factor A-rows	8	1238.0741		154.7593		11.515 (8,16)		0.00002488
	Factor B-columns	2	240.2963		120.1481		8.9397 (2,16)		0.002474
	Error	16	215.037		13.4398		-		-
	Total	26	1693.4074		65.1311		-		-

SD – standard deviation, SE – standard error, DF – degree of freedom, SS – sum of squares, MS – mean square

Table 4. Size variations of MPs (Numbers/kg of sediments) found in Ghar-e-Tangi Cave and their variance among the sites and sizes

Parameters		**Stations**			**Total**	**%age**	**Descriptive Statistics**		
		Site-A	Site-B	Site-C			Mean	SD	SE
Size Range	<1.0 mm	29	11	9	49	26.5	16.33	11.02	6.36
	1-3mm	26	14	12	52	28.1	17.33	7.57	4.37
	>3-5mm	44	24	16	84	45.4	28.00	14.42	8.33
Descriptive Statistics	total	99	49	37	185	-	-	-	-
	Mean	33.0	16.33	12.33	-	-	-	-	-
	SD	9.24	6.11	3.06	-	-	-	-	-
	SE	5.34	3.54	1.77	-	-	-	-	-
TWO-WAY-ANOVA	Source	DF	SS		MS		F Statistic		P
	Factor A-rows	2	250.8889		125.4444		9.5678 (2,4)		0.02989
	Factor B-columns	2	720.8889		360.4444		27.4915 (2,4)		0.004599
	Error	4	52.4444		13.1111		-		-
	Total	8	1024.2222		128.0278		-		-

SD – standard deviation, SE – standard error, DF – degree of freedom, SS – sum of squares, MS – mean square

Color diversity

Eight different colors of MPs were identified in addition to transparent particles (Table 3, Supplementary Fig. S3). The distribution of MP colors was statistically significant (P=0.002), with significant differences between sites (P=0.000). The most common MPs were transparent (23.67±10.97, 38.4% of all), followed by grey (11.67±8.02, 18.9%), and blue (8.67±3.79, 14.1%), while other colors, white, yellow, red, and pink, were less frequent. The less common colors provide valuable insight into the potential input pathways. The concentration of most colors decreased from the entrance to the deeper parts of the cave (Fig. 1).

Size variation

The MP size distribution showed that the largest size range (>3-5 mm) accounted for the highest percentage (45.4%), followed by the 1-3 mm range (28.1%) and the <1 mm range (26.5%). (Table 4, Supplementary Fig. S3). The concentration gradient across the size ranges followed a similar gradient, decreasing from the entrance to the deeper sections of the cave (Fig. 1). The distribution of MPs by size was statistically significant (P=0.004), with differences in size distribution observed across sites (P=0.029). The largest size range (>3-5 mm) accounted for 45.4% (mean 28.0±14.42 at each site) of the total particles, followed by the 1-3 mm range (28.1%, mean 17.33±7.57 at each site) and the <1 mm range (26.5%, mean 16.33 ±11.02).

Polymer composition of MPs

A total of 136 particles were analyzed for polymer composition (sites A=70, B=38, C=28) using FTIR (Table 5). Eight polymer types were identified (Fig. 3). Polyethylene (56.6%) was the most prevalent polymer, followed by polyester (14%), polypropylene (8.8%), and polyvinyl chloride (8.8); smaller amounts of polystyrene, polyethylene terephthalate, acrylic, and polyamide were also identified.

Table 5. Composition analysis of MPs (>1mm size)

Parameters	Stations			Total	%age
	Site A	B	C		
Polyethylene (PE)	40	22	15	77	56.6
Polypropylene (PP)	6	2	4	12	8.8
Polyvinyl-chloride (PVC)	4	3	5	12	8.8
Polystyrene (PS)	1	0	0	1	0.7
Polyethylene terephthalate (PET)	6	2	1	8	5.9
Polyester (PES)	8	7	4	19	14.0
Acrylic (AC)	2	1	1	3	2.2
Polyamide (PA)	3	1	0	4	2.9

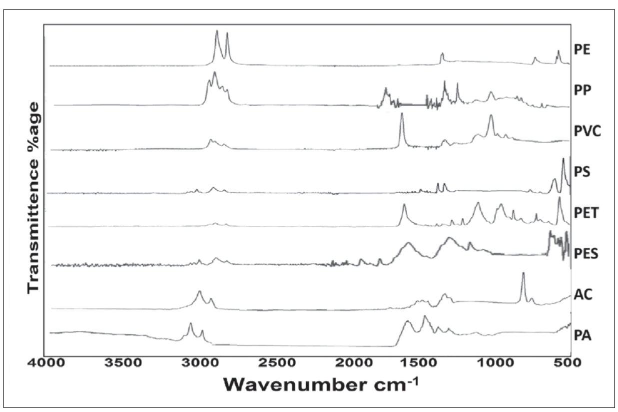

Fig. 3. FTIR spectra for composition analysis of MPs. Polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC), polystyrene (PS), polyethylene terephthalate (PET), polyester (PES), acrylic (AC), polyamide (PA).

DISCUSSION

The current findings on microplastic pollution in Ghar-e-Tangi Cave add to the growing body of evidence showing that caves, due to their isolated nature, act as a sink for MP pollution. This reflects broader issues related to the mismanagement of plastic waste in the environment. The contamination levels recorded in Ghar-e-Tangi Cave ranged from 37-99/kg of sediments, depending on the location. These concentrations are comparable to those found in other cave environments, though some studies report higher or lower levels. In 27 natural caves worldwide, the MP concentration ranges from 7.8 to 4777 particles per kilogram of sediment [31]. In Bue Marino Cave, Sardinia, Italy, only 7.8 MPs/kg were found, at much lower concentrations than observed in Ghar-e-Tangi Cave [32]. In contrast, the Classical Karst Region recorded 4,777 MPs/kg [33], and in Cliff Cave, Missouri, USA, 843 MPs/kg were reported [34]. These concentrations are higher than those found in Ghar-e-Tangi Cave. Meanwhile, in Postojna Cave in Slovenia, and a cave in India, MP concentrations were as low as 9 particles/kg [35], which is significantly lower than in Ghar-e-Tangi. In more pristine environments such as the Antarctic and Arctic regions, MP concentrations in sea ice samples were reported as just 1 to 20 particles/kg, indicating minimal human interference [36,37]. Overall, MP contamination in Ghar-e-Tangi Cave can be considered moderate to high compared to global cave studies.

The dominant presence of fragments and fibers in Ghar-e-Tangi Cave is consistent with findings from other regions [31-37]. However, a unique feature of this study is the significant presence of globular-shaped MPs. This finding mirrors a study in Slovenia's Postojna Cave, where plastic microbeads and fragments were also found in large concentrations [35]. This suggests that caves are repositories for plastic pollution, with atmospheric deposition, runoff, industrial activities, and transport spills being the primary input pathways.

The study identified eight distinct colors of MPs, with transparent particles being the most common (38.4%), followed by grey (18.9%), and blue (14.1%). These colors provide important clues about the potential sources of pollution. Transparent MPs are often associated with consumer waste, such as bottles and packaging, while grey and blue MPs are typically linked to industrial sources, particularly synthetic textiles.

Other colors, including white, yellow, red, pink, orange, and green, accounted for less than 10% of the total, reflecting sporadic input from various products like household items and plastic toys. The size distribution of MPs showed that larger particles predominated. The concentration gradient across different size ranges followed a similar decreasing trend in shapes and colors from the cave entrance to its deeper parts.

The predominance of polyethylene (PE), polyester (PES), and polypropylene (PP) in the cave is consistent with findings from cave systems around the world [31]. These polymers are widely used in the manufacturing industry. The higher concentration of polyester fibers in the cave suggests that textile-related input is significant. The detection of polyvinyl chloride (PVC) is noteworthy, as it has not so far been reported in similar isolated environments.

The results from Ghar-e-Tangi Cave align with studies conducted in other remote environments. For example, research in the Swiss Alps found similar MP shapes, such as fragments and fibers in mountain soils, highlighting the role of atmospheric transport in spreading MPs over long distances [38]. Similarly, MP contamination has been reported in snow samples from remote regions, underscoring the efficiency of transport mechanisms and emphasizing that even isolated areas are not immune to MP pollution [39]. The concentration gradient in Ghar-e-Tangi Cave, with the highest MP concentrations at Site A (the entrance) and the lowest at Site C (the deepest part), can be attributed to sediment trapping and reduced human activity in the deeper sections. A similar pattern has been observed in China, where MP pollution in deeper environments was reduced by sedimentation and limited human impact [40].

The diversity of MP shapes, colors, and sizes in Ghar-e-Tangi provides insights into the sources and pathways of pollution. Fragments likely result from the breakdown of larger plastic items through mechanical degradation [22,41], while fibers are associated with synthetic textiles, pointing to domestic and industrial inputs from washing [42,43]. Films are typically linked to plastic bags and wraps transported by wind [44,15], and globular and pellet-shaped MPs are often associated with spills, which can be carried by surface runoff [45,46]. The variety of MP shapes at Site A (the cave entrance) indicates multiple input pathways, including wind and surface runoff. At the deeper sites (B and C), the increased proportion of finer particles, particularly fibers and fragments, suggests that MPs may be transported by animals or water flow. These insights into input pathways and transport mechanisms are essential for developing targeted mitigation strategies for this unique habitat [16,47,48]. Wind, water runoff, and wildlife movements can all contribute to MP transport over large distances and their deposition in pristine environments [49]. Despite the difficult accessibility of Ghar-e-Tangi Cave, the diversity of MPs found indicates the need for further research to identify specific input pathways.

The findings from Ghar-e-Tangi Cave are consistent with global trends in cave MP pollution. They support the idea that caves are microcosms of broader environmental plastic contamination. The variety of MP shapes, colors, and polymer types observed in Ghar-e-Tangi mirrors those found in other isolated environments, indicating the widespread and persistent nature of MP pollution. These similarities suggest that comparable processes and sources of MP contamination are at play across different geographical and ecological settings, highlighting the global scope of this issue.

CONCLUSIONS

This study found a moderate but significant concentration of microplastics in Ghar-e-Tangi Cave, with a diverse range of shapes, colors, and sizes. The primary input pathways for these pollutants appear to be surface runoff, atmospheric deposition, and, to a lesser extent, human activity. The variety of microplastic colors and shapes suggests multiple pollution sources, including consumer products, synthetic textiles, and industrial activities. These findings provide a baseline for future research on the long-term ecological impacts of microplastics, particularly on cave fauna, and highlight the need for effective pollution mitigation strategies. The study also underscores the urgent necessity for improved plastic waste management policies in the region.

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Data availability statement: The dataset underlying the reported findings is available here: https://www.serbiosoc.org.rs/NewUploads/Uploads/Luqman%20et%20al_Dataset.pdf

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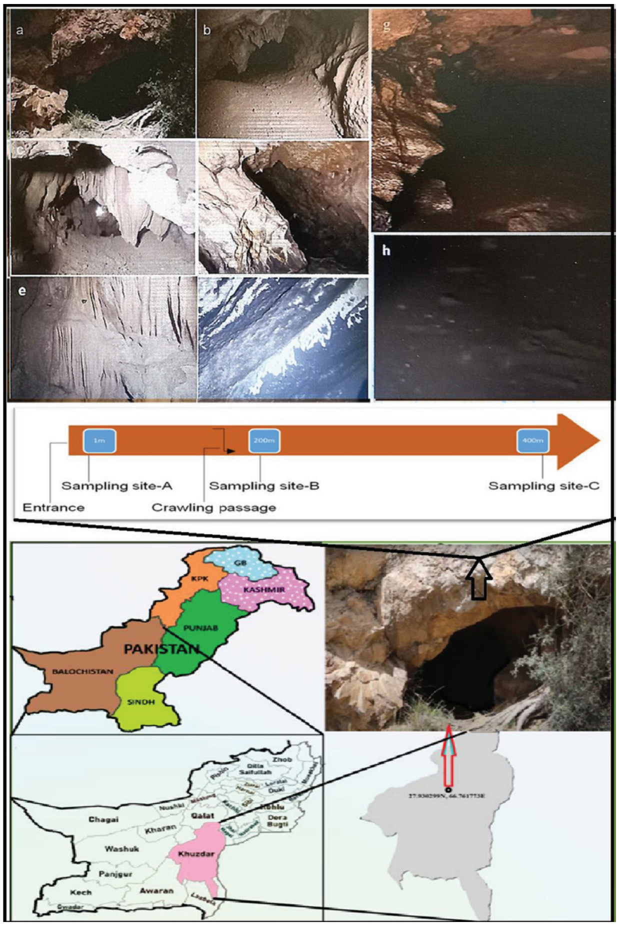
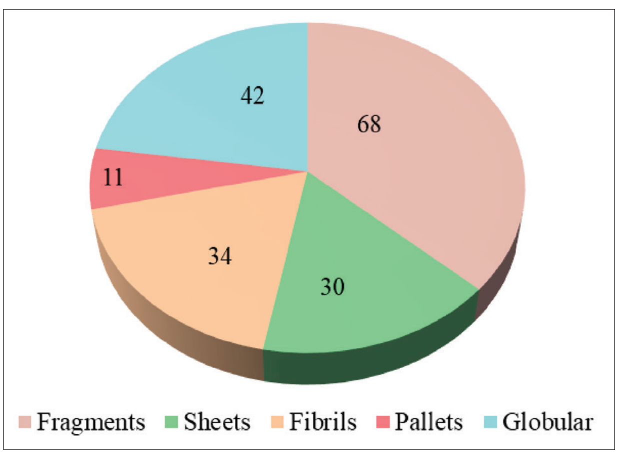
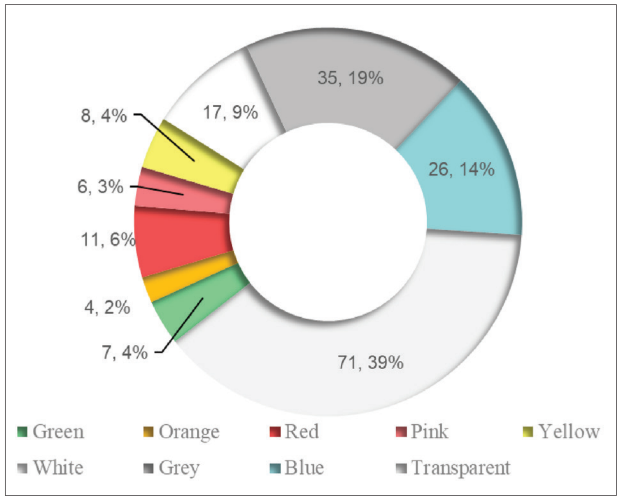
SUPPLEMENTARY MATERIAL



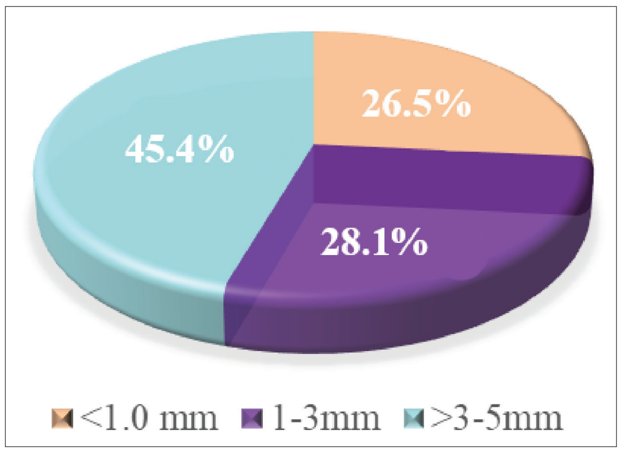
Supplementary Fig. S1. (a-h) Details of the inside area of Ghar-e-Tangi Cave including sampling sites.



Supplementary Fig. S2. The total number of MPs of various shapes found in Ghar-e-Tangi Cave.



Supplementary Fig. S3. The overall distribution (percentage) of various colors of MPs in Ghar-e-Tangi Cave.



Supplementary Fig. S4. Size percentage of MPs found in Ghar-e-Tangi Cave.