

Article

The Migration and Pollution Risk of Microplastics in Water, Soil, Sediments, and Aquatic Organisms in the Caohai Watershed, Southwest China

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Abstract: The migration and driving factors of microplastics (MPs), as an emerging pollutant, have been reported in plateau lakes. However, whether MPs can accumulate to an extreme degree in the local aquatic organisms of plateau lakes remains unclear. Therefore, the present study mainly aims to investigate the MPs accumulated in tissues of grass carp as well as reveal their migration processes and driving factors in the Caohai watershed, a typical plateau lake in southwest China. Density flotation (saturated NaCl solution) and laser direct infrared imaging spectrometry were used to analyze the relative abundance and morphological characteristics of MPs, respectively. The results showed that the MPs' abundance in soil, water, and sediments ranged from 1.20×10^3 to 1.87×10^4 n/kg, from 9 to 223 n/L, and from 5.00×10^2 to 1.02×10^4 n/kg, respectively. The contents and composition of MPs in forestland soils were more plentiful in comparison with cultivated land soils and marshy grassland soils. Polyethylene (PE), polyvinylchloride (PVC), PA from caprolactam (PA6), and PA from hexamethylene diamine and adipic acid (PA66) were detected in grass carp, and PE was detected in all organs of grass carp. MP concentrations in the stomach, intestines, tissue, skin, and gills of grass carp ranged from 54.94 to 178.59 mg/kg. MP pollution probably mainly originated from anthropogenic factors (road traffic, farming activities, the habits of residents scattered around the study area, etc.) due to the Caohai watershed's considerable proximity to Weining city. In addition, wind, land runoff, rivers, and atmospheric deposition in the locality directly and indirectly promoted MP migration. Our results suggested that although there is moderate MP pollution in soil, water, sediment, and grass carp in comparison with other areas, it is necessary to pay attention to PE and PVC migration via the various environmental media and the risks associated with consuming the local grass carp. The local government can make several policies to reuse and recycle agricultural film to alleviate local PE and PVC pollution.

Keywords: microplastics; grass carp; tissues; Caohai watershed; plateau lake



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1. Introduction

Owing to their extraordinary properties, such as durability, flexibility, light weight, low thermal conductivity, and low cost, plastics and their byproducts are widely used in industries such as raw plastic manufacturing, laundry services, and textile manufacturing,

as well as in home activities and daily lives, with applications in automobiles, electronics, packaging, agricultural products, construction materials, and medical treatments [1,2]. The cumulative value of global plastic production exceeded 5 billion tons in 2016, and this value is expected to reach 33 billion tons by 2050 [3,4]. Due to inefficient disposal and long decomposition times, a large amount of plastic debris is accumulating in the environment [5]. Owing to its persistent nature, plastic debris may travel long distances and is even found in remote locations far from human activity [3]. In the natural environment, plastic debris undergoes further degradation and disintegration caused by various physical, chemical, and biological forces, leading to the formation of millions of microplastics (MPs) [6]. The degradation of MPs is affected by a variety of environmental factors, including light, temperature, humidity, oxygen, etc. Additionally, a high temperature (above 120 °C), ultraviolet light, and an acid-base environment are all conducive to the degradation of MPs. Some specific microorganisms can also effectively degrade MPs under suitable temperature and pH conditions [7].

MPs are pieces of plastic debris that are smaller than 5 mm in diameter [8]. However, some scientists have also defined MPs as plastic debris <1 mm [9]. MPs comprise small fragments of plastics with a variety of morphologies, such as ropes, nets, fibers, microbeads, and styrofoam [10]. They are usually formed via natural environmental degradation of larger pieces of plastic trash, such as plastic bottles, personal care products, tire wear, plastic bags, microwave containers, and fishing nets, as well as via the breakdown of polyester-, acrylic-, and nylon-based clothes and other plastic wares in the washing process [11]. In the environment, MPs can be of primary or secondary origin. Compared with primary MPs, secondary MPs are much more common [12]. In recent decades, many studies concerning plastic debris pollution have been reported; MPs have been found in habitats from the poles to the equator, in soils/sediments from inland to the deep sea, and in organisms from polychaete worms to humans [13]. MPs are persistent pollutants that deteriorate gradually in the environment, usually taking hundreds to thousands of years or more [14]. Therefore, the relative abundance of MPs (RA-MPs) on land and in water bodies has increased annually over the past several decades [15]. MPs are small enough to be taken up by biota and can enter the food chain, causing bioconcentration and magnification [16]. Evidence suggests that MPs can be taken up and stored by tissues and cells, with probable negative consequences for health [9]. It has been reported that MPs have negative effects on the digestion, growth, and reproduction of aquatic organisms [17]. In addition, MPs can adsorb different chemicals, such as heavy metals, algal toxins, and antibiotics, and can be adsorbed by organisms such as bacteria and fungi, causing enrichment and potential enhanced toxicity [18]. Due to their persistence, accumulation in the environment, and potential ecological and human health risks, MPs have become an increasing concern [19].

Previous studies related to MPs have focused mainly on their pollution levels, distribution characteristics, original sources, migration, degradation, fate, interactions with other pollutants [20], bioconcentration, biomagnification, and potential ecological and human risks [21]. Rasmussen et al. [22] reported that 88% of the MPs (on average) in stormwater were retained in ponds and that 95% of the plastic debris (>500 µm) in stormwater was removed through the outlets of ponds. In a review of a global MP dataset comprising 862 water samples and 445 sediment samples from 63 articles, Tan et al. [19] reported that the MP abundance in water and sediment samples spans 10 and 4 orders of magnitude, respectively. Gong et al. [23] collected water samples from rivers and reservoirs in eleven typical nature reserves and scenic districts in the Sichuan, Chongqing, Hunan, Guizhou, and Guangxi Provinces and studied the relative abundance and distribution characteristics of MPs, finding that the number of MPs in nature reserves and scenic districts ranged from 542 to 5500 items/m³. In aquatic ecosystems, MPs may be mistaken for food by

fish, invertebrates, and microorganisms, leading to physical damage, digestive system blockage, and reduced nutrient absorption, affecting overall health and reproductive capacity [24,25]. However, it is also claimed that the terrestrial environment is a much larger sink of plastic pollution.

The Caohai watershed, located on the Yunnan–Guizhou Plateau, is 2504 m above sea level, and its average annual temperature is only 10–12 °C. Plastic films are widely used in agricultural activities to improve soil hydrothermal conditions, leading to plastic pollution. To our knowledge, few studies have systematically investigated the source and migration of MPs at the watershed scale. Zhang et al. [2] studied the RA-MPs and MP particle size in cultivated soils, estuarine sediments, lake sediments, estuarine water, and lake water in the Caohai watershed. Their studies suggested that the MPs in farmland soils were mainly derived from plastic film and domestic sewage and that estuarine water is the first temporary carrier of MPs after they enter the water environment. Wu et al. [8] studied the RA-MPs and its influencing factors in cultivated soil, water, and sediment in Caohai Lake during dry and wet periods, and they suggested that the relevant government department should take measures to reduce the MP pollution in Caohai Lake, especially from the source. Both of these studies focused on the pollution level, particle size, and distribution characteristics of MPs in/around Caohai Lake, while the sources, migration, and ecological risk of MPs in various media and aquatic organisms were ignored. This study proposes the following two hypotheses: (1) the various media in Caohai Lake have been contaminated by MPs; and (2) the aquatic organisms have probably been endangered by MPs. Therefore, the specific objectives of this study are as follows: (a) to investigate the RA-MPs and MP composition in soils associated with different land uses (farmland, forestland, and marshy grassland) and in water and sediment samples from rivers and Caohai Lake, revealing the mechanisms influencing MPs at the watershed scale; and (b) to study the MP levels and composition in the organs of grass carp (*Ctenopharyngodon idella*) from Caohai Lake to explore the risk and exposure pathways.

2. Materials and Methods

2.1. Study Region

The Caohai watershed (26°31′ N–27°27′ N, 103°36′ E–104°36′ E) is in the southwest corner of Weining County on the northwest edge of Guizhou Province. Located in the hinterland of Wumeng Mountain at the top of the central part of the Yunnan–Guizhou Plateau, Caohai Lake is the largest natural freshwater plateau lake in Guizhou Province [26]. The terrain in Caohai town is undulating and flat, with minor variations in topography. It is a wide and gently sloped hilly basin and ridge plain. This area has been referred to as a “highland pearl”, bird paradise, underwater forest, species gene bank, and open-air museum [27]. The Caohai wetland is an important habitat for many rare birds, such as the black-necked crane, and is an important node in the western passage, which is one of the three major bird migration routes in China. The highest point in the town is located 2504 m above sea level on Tashan Mountain in the east of the city, and the lowest point is at 2172 m above sea level on Yangguan Mountain in the west of the city. The average elevation of the watershed is 2267 m. The Caohai watershed has an average annual temperature of 10 to 12 °C, an average annual rainfall of 800 to 1200 mm, an annual sunshine duration of 1300 to 1700 h, and a frost-free period of 195 to 227 d. It is in a typical subtropical plateau monsoon climate zone with dry winters and springs and humid summers and autumns.

2.2. Sampling

In July 2023, soil samples were collected from 9 plots (Figure 1): 3 in cultivated land (CS-1, CS-2, and CS-3), 3 in forestland (FS-1, FS-2, and FS-3), and 3 in marshy grassland

(MS-1, MS-2, and MS-3). In each plot, soil (0–20 cm) was sampled at 5 sites (200 m from each other) with a stainless-steel shovel, the soils were mixed, and 1 kg of mixed soil was collected as the final sample. Nine water samples (WR-1~WR-9) and nine superficial sediment (0–10 cm) samples (SR-1~SR-9) were taken from the four rivers flowing into Caohai Lake, including the Zhonghe River, Baima River, Dongshan River, and Wanxia River, and one water sample (WR-10) and one sediment sample (SR-10) were taken from the lake outlet river. Five water (10 cm below the water surface) samples (WL-1~WL-5) and five sediment (0–20 cm) samples (SL-1~SL-5) were taken from Caohai Lake. The dissolved oxygen (DO), pH, and oxidation-reduction potential (ORP) were determined on site using a portable water quality analyzer (HQ30d, HACH, Ames, IA, USA). The grass carp (*Ctenopharyngodon idella*) were obtained from the staff responsible for fish regulation during the time.

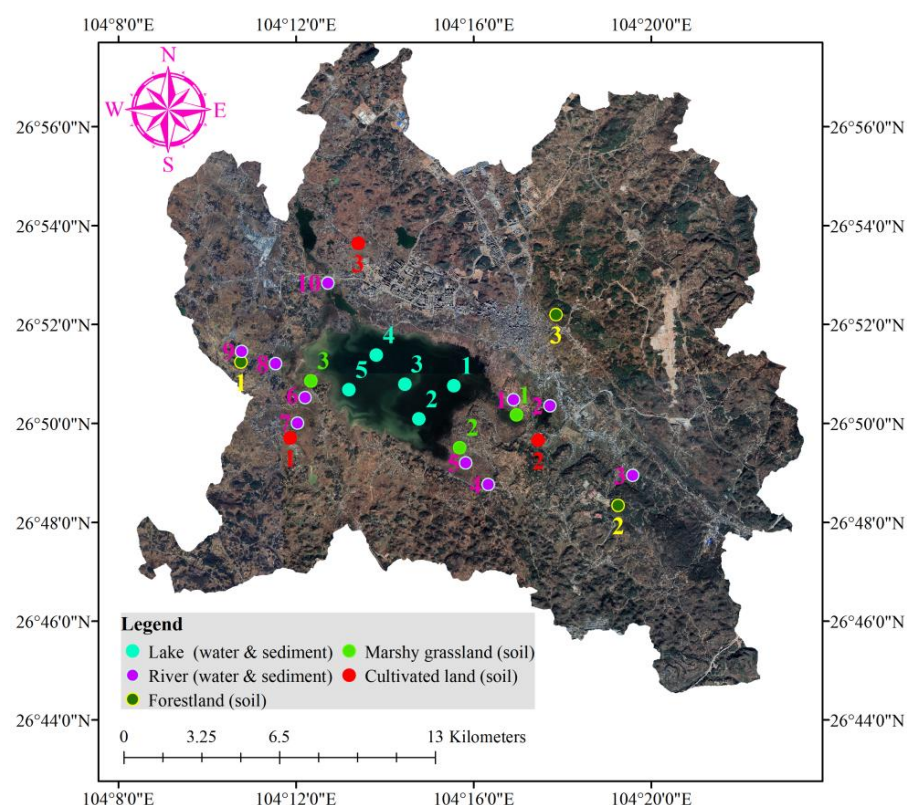


Figure 1. Location and sampling points of the study area.

2.3. Analytical Procedures

2.3.1. Soil and Sediment Samples

Each soil or sediment sample was dried to constant weight in an oven at 75 °C for 48 h. One hundred grams (accurate to 0.001 g) of the dried soil/sediment sample was weighed into a 500 mL triangular beaker, and then 250 mL of 1.2 g mL^{−1} saturated NaCl solution was added to the bottle for MP flotation. The mixture was fully stirred with a glass rod for 5 min and then allowed to stand at room temperature for 24 h. Then, the supernatant was extracted, collected, and immediately filtered through a 20 µm stainless-steel filter [9]. The particles on the membrane were washed into a 150 mL glass reagent bottle with 30% H₂O₂. The flotation step was repeated 3 times for each sample, and all collected supernatants were placed into the same beaker. Then, 0.01 g of FeCl₃ was added, and the beaker was sealed. Subsequently, the bottles were heated in a water bath at 60 °C for 72 h to remove organic matter [28]. After standing for 24 h, the liquids in the beakers were filtered through 0.45 µm mixed-cellulose filter membranes (diameter of 50 mm), and

the filter membranes were stored in Petri dishes away from light before observation [6]. Before testing, the membranes were washed with ethanol, the samples were concentrated and dripped on highly reflective glass, and then the morphological characteristics of MPs were analyzed via laser direct infrared imaging spectrometry (Agilent 8700LDIR, Agilent Technologies Co., Ltd., Santa Clara, CA, USA). Meanwhile, a blank control was created to exclude contamination in processing the samples. Due to the large number of samples and the cumbersome counting of MPs in the digestion process, the classification of particle size was simplified in this study. Although there may be some deviation in the evaluation of particle size (20 to 100 and 200 to 500 μm), it also provided a certain reference.

2.3.2. Water Samples

Water samples were passed through a cellulose filter membrane (diameter 50 mm) with a pore diameter of 20 μm . All the substances on the membrane were washed off with 30% H_2O_2 into a small 150 mL beaker, 0.01 g of FeCl_3 was added, and the beaker was sealed. The following steps (digestion and subsequent flotation) were consistent with the soil/sediment sample treatment process.

2.3.3. Organs of Grass Carp

The organs of grass carp (stomach, intestine, skin, gill, and fish tissue) were separated and washed, in accordance with Liu et al. [29]. Step I: Samples were weighed (accurate to 0.0001 g), put into a 50 mL beaker (beaker A), and dried to constant weight in an oven at 60 °C. Then, 30 mL ethanol was added, the beakers were heated in a water bath at 60 °C for 10 min, and the filtrate was poured out, which was repeated three times. Step II: 10 g of chloroform was added, and the mixture was extracted under ultrasonication at 40 kHz for 10 min. The supernatant was collected in a new beaker (beaker B), and the process was repeated three times. Subsequently, the extractant (chloroform) was replaced with hexafluoroisopropanol, and step II was repeated. The extractant (hexafluoroisopropanol) was then replaced with dimethylbenzene, and step II was repeated once more. Step III: All the supernatants were collected in beaker B. The extract (approximately 90 g) was concentrated on a heating plate at 80 °C to approximately 3 g, transferred into a 20 mL transparent glass sample bottle, and further concentrated to within 1 g. Then, a glass pipette was used to drop the sample into the pyrolysis gas chromatography-mass spectrometry (Py GCMS) sample holder, which was heated on a heating plate at 80 °C, and the MPs in different organs of grass carp were analyzed via Py GCMS (GCMC-QP2020, Shimadzu Global Laboratory Consumables Co., Ltd., Kyoto, Japan) after the solvent in the crucible had completely evaporated. The testing conditions are listed in Table 1.

Table 1. Testing conditions for MPs in organs of grass carp.

Items	Conditions
Pyrolyzer	PY-3030D Frontier (Frontier Lab., Kyoto, Japan)
Pyrolysis temperature	550 °C
Split ratio	5:1
Chromatographic column	Rtx-5MS (30 m $\times$ 0.25 mm $\times$ 0.25 μm)
Chromatogram temperature program	Maintain 40 °C for 2 min, heat to 320 °C at a rate of 20 °C/min, maintain for 14 min; total time—30 min.
Ionization temperature	230 °C
m/z scanning range	19–600

2.4. Statistical Analysis

In this study, the RA-MP values in soil and sediment samples are reported in n/kg (n/m , dw), those in water samples are reported in n/L (n/v), and those in the organs of grass

carp are reported in mg/kg (m/m) [29]. All data were managed, processed, and analyzed using Excel 2024 and Origin 2021, and the plots of sampling information were designed with ArcGIS 10.3. The difference in the RA-MPs in the various media was examined using analysis of variance (ANOVA) in IBM SPSS Statistics Version 20 [30].

3. Results

3.1. Relative Abundance of MPs in Soils from Different Land Uses in the Caohai Watershed

The RA-MPs in soils from cultivated land ranged from 1.20×10^3 to 4.73×10^3 n/kg, with a mean value of 2.76×10^3 n/kg (Figure 2); those in soils from forestland ranged from 7.93×10^3 to 1.87×10^4 n/kg, with a mean value of 1.19×10^4 n/kg (Figure 2); and those in soils from marshy grassland ranged from 1.80×10^3 to 4.33×10^3 n/kg, with a mean value of 2.73×10^3 n/kg (Figure 2). These results were attributed to the widespread use of plastic film in agricultural activities in the Caohai watershed and the fact that plastic film is almost never recycled. Before our research began, we predicted that the MP content in the soil would be the highest, but the results were unexpected. In terms of the mean values, the RA-MP values in soil samples from forestland were much greater than those in soil samples from cultivated land, followed by those in soil samples from marshy grassland. The size ranges of MPs were uniformly divided into two categories: 20–100 and 100–500 μm (Figure 3). The percentages of MPs in different particle size ranges were different in soil samples from different land uses and from different sampling sites under the same land use. In cultivated land, MPs with a size range of 20–100 μm account for 89 to 98%, and MPs with a size range of 100–500 μm account for 2 to 11% (Figure 3a). In forestland, MPs with a size range of 20–100 μm account for 94 to 100%, and MPs with a size range of 100–500 μm account for 0 to 6% (Figure 3b). In marshy grassland, MPs with a size range of 20–100 μm account for 70 to 81%, and MPs with a size range of 100–500 μm account for 19 to 30% (Figure 3c). Generally, the percentage of MPs with a size range of 20–100 μm is much greater than that of MPs with a size range of 100–500 μm in soil samples from the Caohai watershed.

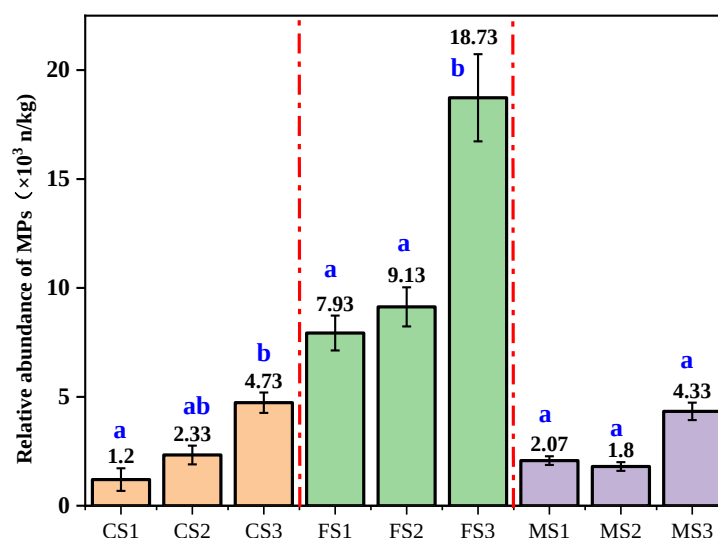


Figure 2. RA-MPs in soil samples from different land uses in the Caohai watershed (note: CS, FS, and MS represent cultivated land, forestland, and marshy grassland). Mean $\pm$ SD; $n = 3$. Different superscript letters in each row represent significant differences between different treatments (ANOVA, $p < 0.05$).

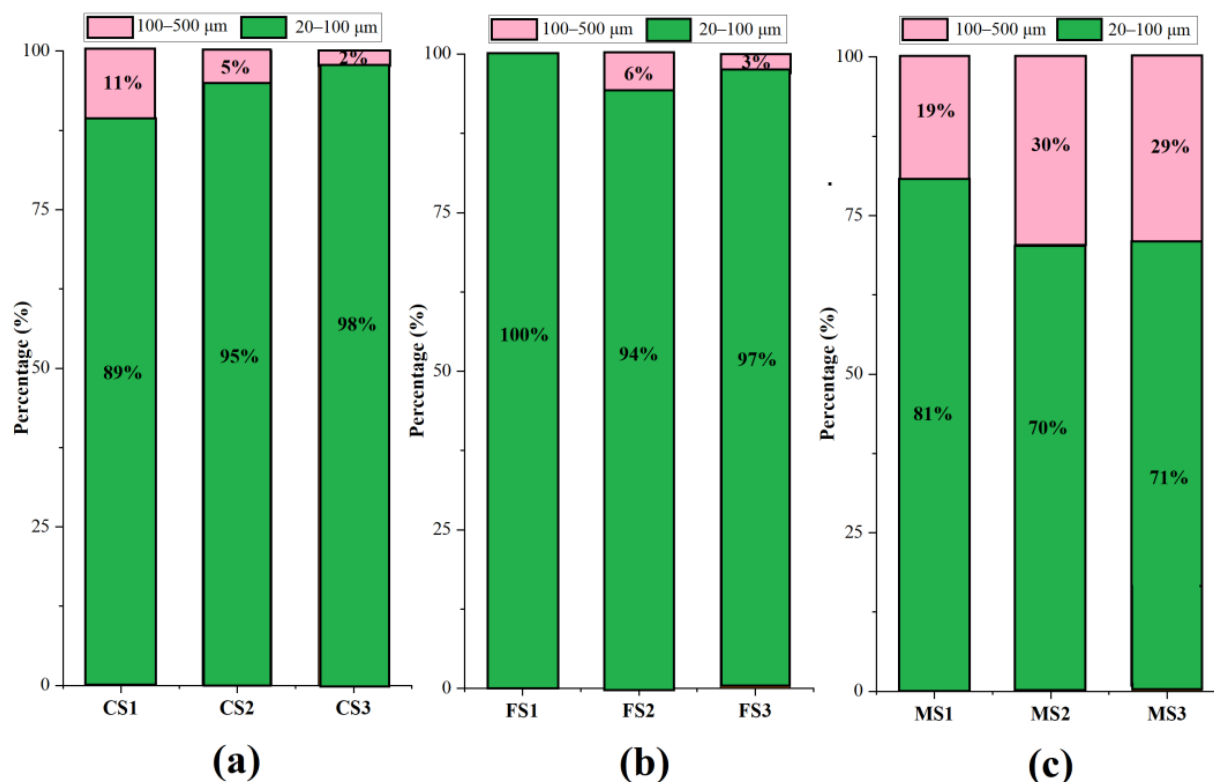


Figure 3. Percentages of MPs with different sizes in soils from different land uses in the Caohai watershed. ((a). Cultivated land; (b). forestland; and (c). marshy grassland). (Note: the average values of each group are used in this figure).

3.2. Relative Abundance and Particle Size of MPs in Water and Sediment from Rivers and Caohai Lake

The RA-MPs in river water samples ranged from 18 to 213 n/L, with a mean value of 59.90 n/L. Among the four main rivers, the RA-MPs from sites adjacent to Caohai Lake decreased in the following order: Baima River (WR-5, 213 n/L) > Dongshan River (WR-6, 107 n/L) > Wanxia River (WR-8, 46 n/L) > Zhonghe River (WR-1, 25 n/L) (Figure 4a). The RA-MPs in WR-10, which is the outlet river of Caohai Lake, was 47 n/L. The RA-MPs in the overlying water of Caohai Lake ranged from 9 to 223 n/L, with a mean value of 58.80 n/L (Figure 4b). The RA-MPs in sediment samples from rivers ranged from 5.33×10^2 to 1.02×10^4 n/kg, with a mean value of 3.72×10^3 n/kg (Figure 4c), and that in sediment samples from Caohai Lake ranged from 5.33×10^2 to 2.27×10^3 n/kg, with a mean value of 1.35×10^3 n/kg (Figure 4d). Based on the mean values, the RA-MPs in soil samples was much greater than that in sediment samples.

MPs with a size range of 20–100 µm in river water samples accounted for 70 to 97%, and those with a size range of 100–500 µm accounted for 3 to 30% (Figure 5a). MPs with a size range of 20–100 µm in lake water samples accounted for 78 to 100% (Figure 5b), and those with a size range of 100–500 µm accounted for 3 to 30%. MPs with a size range of 20–100 µm in river sediment samples accounted for 83 to 100% (Figure 5c), and those with a size range of 100–500 µm accounted for 0 to 17% (Figure 5d). MPs with a size range of 20–100 µm in lake sediment samples accounted for 85 to 100%, and those with a size range of 100–500 µm accounted for 0 to 15%. In water/sediment samples from both rivers and Caohai Lake, the portion of MPs with a size range of 20–100 µm was much greater than that of MPs with a size range of 100–500 µm.

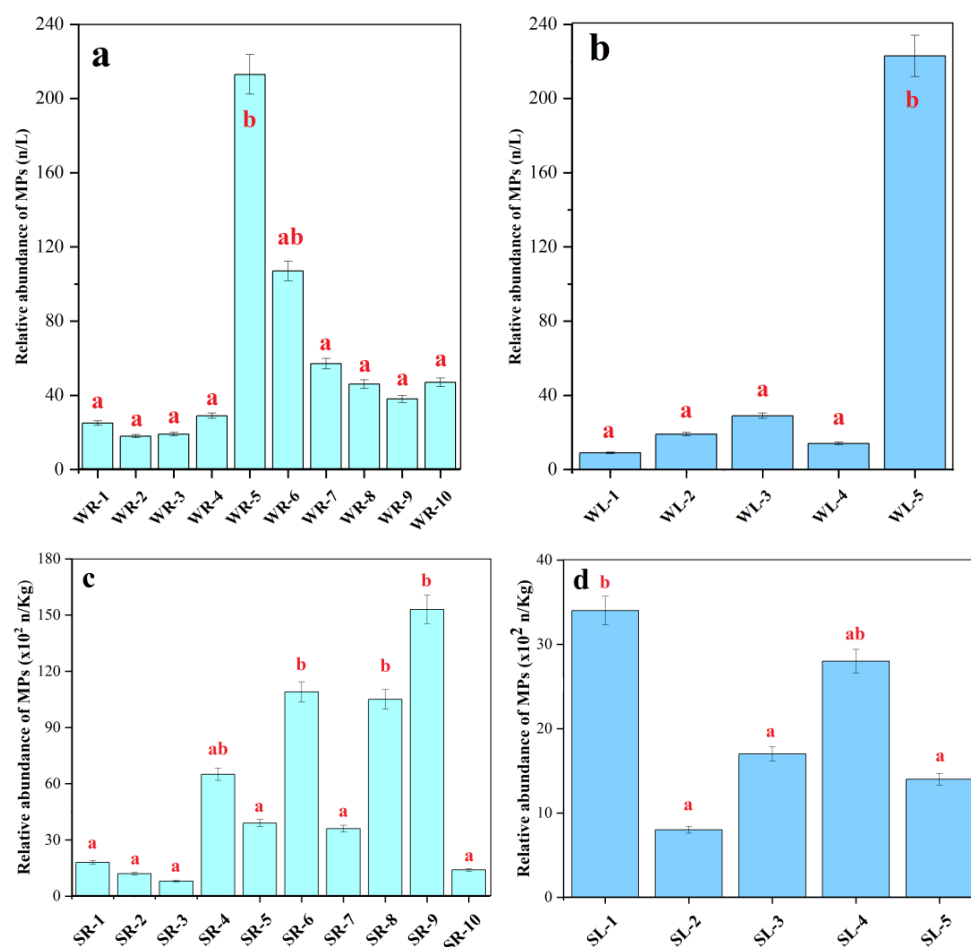


Figure 4. RA-MPs in water from rivers (a), water from Caohai Lake (b), sediment from rivers (c), and sediment from Caohai Lake (d). (Note: the average values of each group are used in this figure). Mean $\pm$ SD; $n = 3$. Different superscript letters in each row represent significant differences between different treatments (ANOVA, $p < 0.05$).

3.3. MP Components in the Caohai Watershed

Nineteen kinds of MPs were identified from soils in the Caohai watershed, including polyurethane (PU), acrylate copolymer (ACR), ethylene vinyl acetate (EVA), polyvinylchloride (PVC), polymethylmethacrylate (PMMA), polyamide (PA), polypropylene (PP), polyimide (PI), polytetrafluoroethylene (PTEE), polyvinyl butyral (PVB), polyethylene terephthalate (PET), epoxy phenol novolac (EPN), polysulfone (PSF), ethylene acrylic acid (EAA), fluororubber (FKM), polycarbonate (PC), polyethylene (PE), polystyrene (PS), and fluorosilicone rubber (FMQ). The main polymeric components of MPs (proportion greater than 1%) in soils from different land uses substantially differed (Figure 6). The predominant polymeric form in cultivated land soil was PC with an overall percentage of 34.7%, followed by PVC (28.7%), PET (7.9%), PSF (6.9%), PU (4.0%), and ACR (4.0%); that for forest soil was PU with an overall percentage of 41.2%, followed by ACR (15.4%), PMMA (11.4%), EVA (10.7%), PA (8.7%), PAC (6.9%), and PE (4.0%); and that for marshy grassland soil was PA with an overall percentage of 54.5%, followed by PVC (40.2%) and PP (2.3).

Fifteen kinds of MPs, including PU, ACR, EVA, PVC, PMMA, PA, PP, PI, PTEE, PVB, PET, EPN, PSF, EAA, and FKM, were identified from the water samples. The predominant polymeric form in Zhonghe River water was PA with an overall percentage of 34.8%, followed by PI (24.2%), PP (13.6%), EPN (7.6%), EAA (4.5%), and PU (4.5%); for Baima River water, the predominant polymeric form was PI with an overall percentage of 82.8%, followed by PP (6.9%) and PVC (6.9%); for Dongshan River water, the predominant polymeric

form was PI with an overall percentage of 56.8%, followed by PVC (25.0%), PA (11.4%), PP (2.3%), and PMMA (1.1%); and for Wanxia River water, the predominant polymeric form was PVC with an overall percentage of 43.1%, followed by PA (34.1%), EVA (8.9%), pu (6.5%), and ARC (3.3%). The predominant polymeric form for outlet river water was PA, with an overall percentage of 71.8%, followed by PVC (10.3%), PP (5.1%), and ACR (5.1%). The predominant polymeric form for lake water was PA, with an overall percentage of 27.4%, followed by PI (12.8%), PET (12.0%), PU (12.0%), PP (11.0%), and FKM (6.8%).

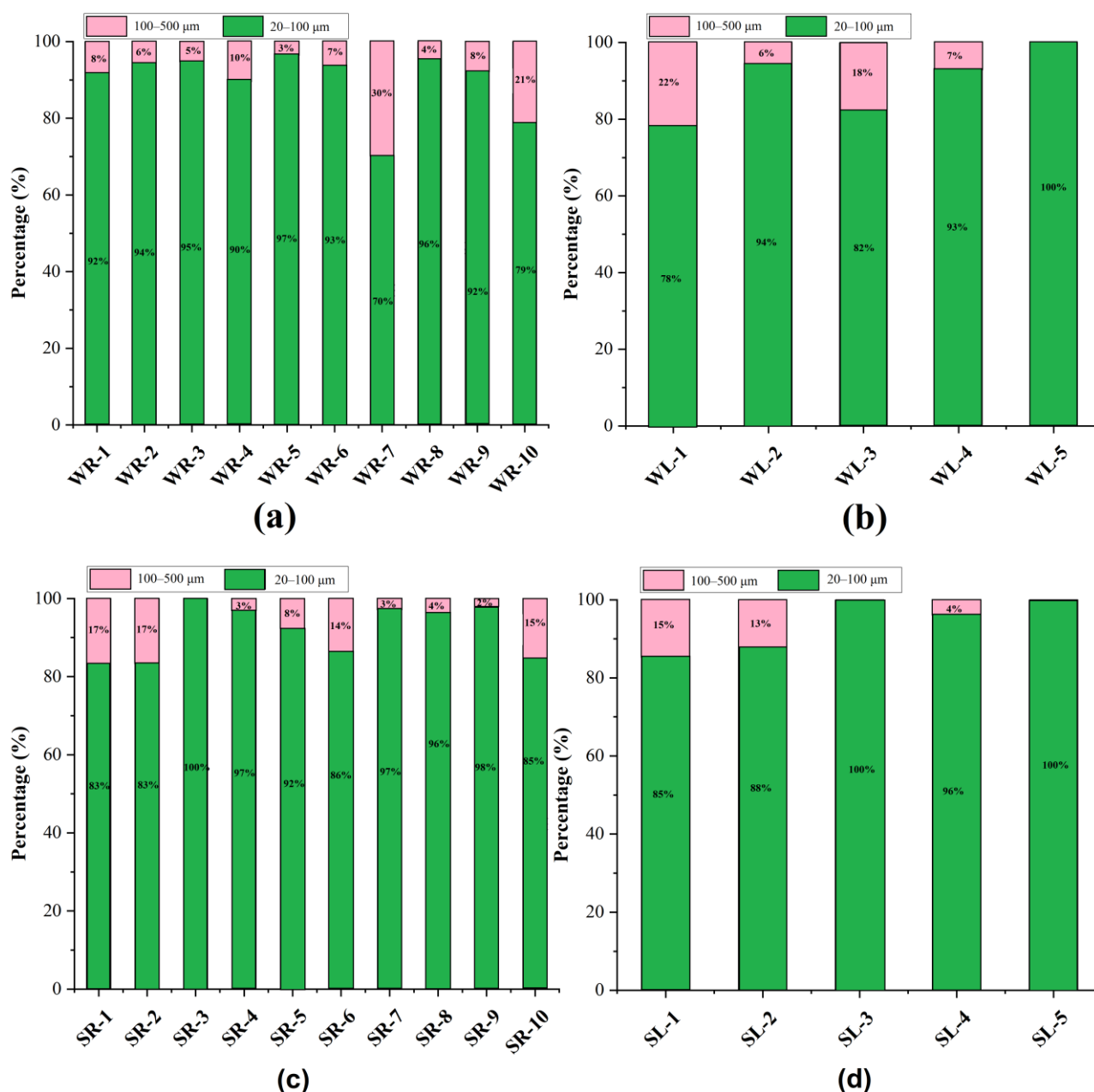


Figure 5. Percentages of MPs of different sizes in water from rivers (a), water from Caohai Lake (b), sediment from rivers (c), and sediment from Caohai Lake (d). (Note: the average values of each group are used in this figure).

Nineteen kinds of MPs were identified from the sediment samples, including PU, ACR, EVA, PVC, PMMA, PA, PE, PP, PI, PS, PTEE, PVB, PC, PET, EPN, EAA, FKM, FMQ, and polyoxymethylene (POM). The predominant polymeric form for Zhonghe River sediment was PA, with an overall percentage of 72.0%, followed by PVC (13.0%), PU (5.0%), ACR

(2.5%), and PP (2.5%); for Baima River sediment, it was PI, with an overall percentage of 60.0%, followed by PET (11.1%), PA (6.7%) and PU (6.7%), and PS (4.4%) and EAA (4.4%); for Dongshan River sediment, it was PET, with an overall percentage of 64.2%, followed by PA (8.8%), PMMA (5.4%), PU (4.1%), EAA (2.7%), and PI (2.0%); and for Wanxia River sediment, it was PU, with an overall percentage of 50.7%, followed by PMMA (15.3%), PA (12.0%), PVC (9.1%), ACR (7.3%), and EVA (2.9%). The predominant polymeric form for outlet river sediment was PI, with an overall percentage of 66.7%, followed by PA (12.5%), PET (8.3%), and PMMA (4.2%). The predominant polymeric form for outlet river sediment was PA, with an overall percentage of 59.0%, followed by PVC (13.0%), ACR (5.0%), EAA (4.0%), EPN (4.0%), and PP (3.0%).

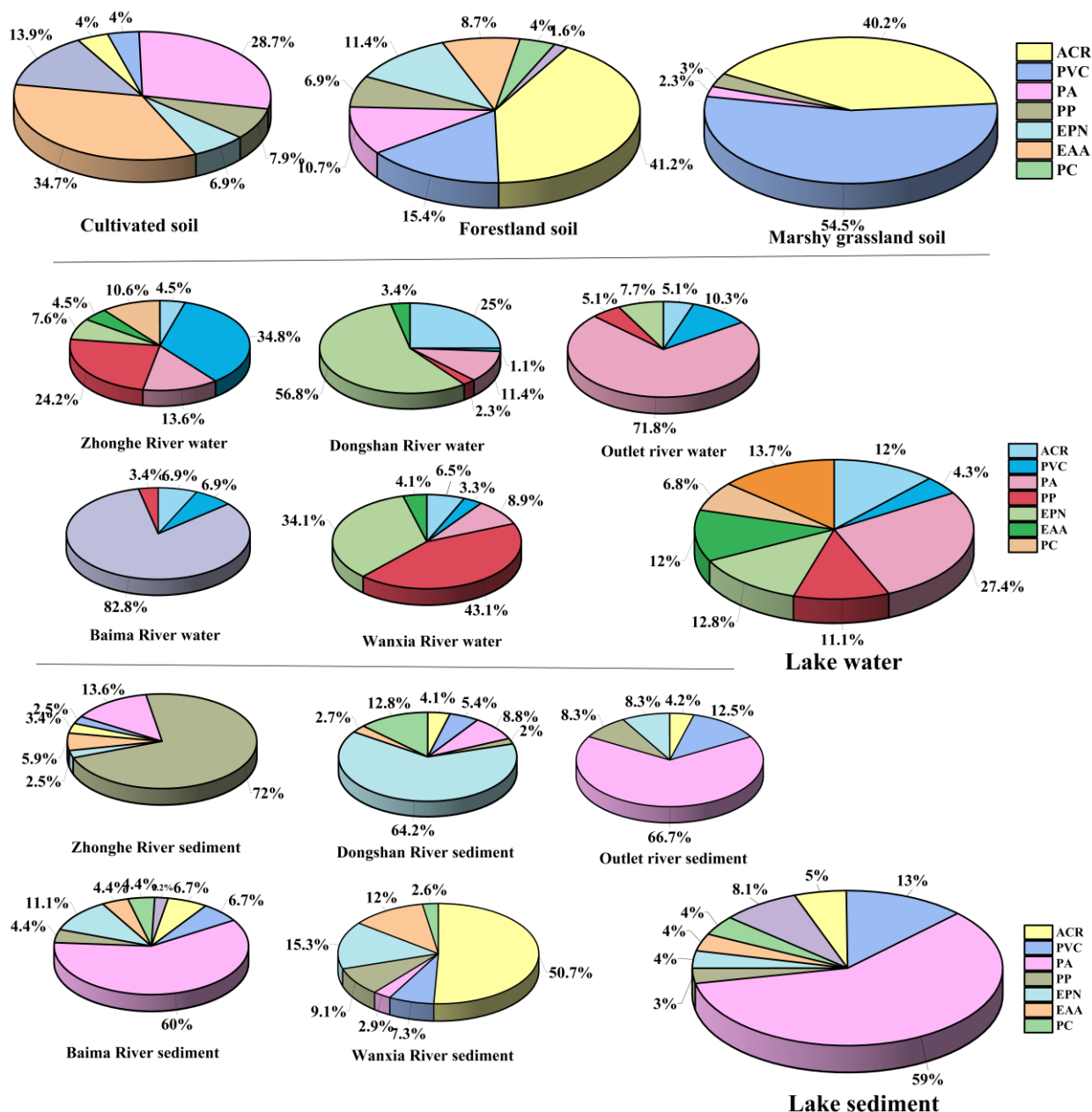


Figure 6. Predominant polymers in MPs in the Caohai watershed.

3.4. MPs in the Organs of Grass Carp

To investigate MP pollution in grass carp from Caohai Lake, we measured 11 kinds of common MPs (PS, PE, PP, PMMA, PVC, PET, PC, polyamide 6 (PA6), polyamide 66 (PA66),

PLA, and PBAT) in the organs of grass carp. As listed in Table 2, four kinds of MPs were detected in grass carp. PE was detected in all organs, and its concentrations in the stomach, intestine, skin, gill, and fish tissue were 54.94 mg/kg, 51.69 mg/kg, 27.53 mg/kg, 34.16 mg/kg, and 20.45 mg/kg, respectively. PVC was detected in the intestine, skin, gill, and fish tissue at concentrations of 126.90 mg/kg, 78.42 mg/kg, 98.07 mg/kg, and 33.96 mg/kg, respectively. PA6 and PA66 were detected only in the skin and gills, respectively. Pollutants can pose a risk to the health of macro aquatic animals through inhalation, ingestion, or dermal contact [31]. Based on the characteristics of MPs in different organs of grass carp, it is believed that PE and PVC were taken up mainly through ingestion or dermal contact. Polyamide 6 and PA66 were taken up mainly through dermal contact and inhalation, respectively.

Table 2. Concentrations of MPs in the organs of grass carp.

	PE (mg/kg)	PVC (mg/kg)	PA6 (mg/kg)	PA66 (mg/kg)
Stomach	54.94	ND *	ND	ND
Intestines	51.69	126.90	ND	ND
Tissue	27.53	78.42	ND	ND
Skin	34.16	98.07	27.27	ND
Gills	20.45	33.96	ND	34.61

Note: “*” ND means not detected, and the detection limit for all studied MPs in the organs of grass carp was 0.13 mg/kg.

4. Discussion

4.1. MPs in the Caohai Watershed

Compared with previous studies on the Caohai watershed [2,8], MPs in cultivated soil were a little lower in quantity in the present study, and the mean value has decreased from 4783 ± 1892 n/kg to 2756 ± 1041 n/kg. However, MP pollution in both water and sediments has become more severe (Table 3). The MP contents in cultivated soils from the Caohai watershed were lower than those in samples from Shanghai, Beijing, and France but much greater than those in samples from the Tibetan Plateau. The MP contents in the water of Caohai Lake were greater than those in many other lakes/rivers, such as Poyang Lake, Taihu Lake, Dongting Lake, Nainital Lake, etc., and slightly lower than those in lakes in the NW Himalayas (India). The MP contents in the sediments of Caohai Lake were also much greater than those in the sediments of many other lakes and like those in Nainital Lake in India. MPs have been detected in fishes from many other water bodies (Table 3), such as Lake Ontario, the Northeast Atlantic, and Poyang Lake. In the present study, the total MPs in different organs of grass carp ranged from ND to 178.59 mg/kg. However, the unit used in the present study was n/fish, making it difficult to compare pollution levels. However, several studies have shown that MPs can lead to changes in the antioxidant activity of enzymes such as glutathione and superoxide dismutase, as well as an increase in oxidative stress indicators such as malondialdehyde [32,33]. MPs mainly accumulate in the intestines of fish, causing physical damage, inflammatory reactions, and changes in the microbiome. MP particles rubbed against the intestinal wall, causing the thinning of the intestinal muscle layer and triggering intestinal inflammation. Meanwhile, MPs altered the composition of the gut microbiome, increasing the relative abundance of opportunistic pathogens [34].

Table 3. Comparison with the results of previous studies in the Caohai watershed and other regions.

Location (Region)	Date	Abundance			Main Particle Size	Reference
		Min.	Max.	Mean		
Soils						
Caohai (China) (n/kg)	2023	1200	18,733	5807 ± 1806	20–100 μm	Present study
Caohai (China) (n/kg)	2021	3000 *	8640 *	4783 ± 1892 *	0–0.5 mm	[2]
		—	—	4410 ± 1635 #		[8]
Shanghai (China) (n/kg)	2023	63,400	328,000	—	20–30 μm	[35]
Beijing (China) (n/kg)	2023	14,420	38,820	24,763	<0.5 mm	[9]
Tibetan Plateau (China) (n/kg)	2018	5	340	47.12	50–500 μm	[36]
French territory (France) (n/kg)	2020	258	3096	597 ± 895	1–315 μm	[37]
Water						
Caohai (China) (n/L)	2023	9	223	59 ± 17	20–100 μm	Present study
Caohai (China) (n/L)	2021	4.6 *	10.1 *	6.5 ± 3.3 *	0–0.5 mm	[2]
		2.7 #	10.5 #	5.6 ± 2.0 #		[8]
Poyang Lake (China) (n/L)	2018	5	34	—	<0.5 mm	[38]
Taihu Lake (China) (n/L)	2015	3.4	25.8	—	100–1000 μm	[39]
Dongting Lake (China) (n/L)	2018	0.32	0.48	0.97 ± 0.42	<0.5 mm	[40]
Chao Lake (China) (n/L)	2019	0.33 *	0.62 *	2.13 ± 1.53 *	<1 mm	[6]
	2020	0.24 #	0.49 #	1.68 ± 1.58 #		
Nainital Lake (India) (n/L)	2023	8.6	56.0	—	0.02–1 mm	[41]
Antuã River (Portugal) (n/L)	2016	0.058	0.1265	—	-	[42]
Lake in NW Himalayas (India) (n/L)	2022	13	238	130	0.9–0.333 mm	[43]
Siberian lakes (Russia) (n/L)	2020	4 n/L	26 n/L	11 ± 7 n/L	<1.0 mm	[17]
Sediment						
Caohai (China) (n/kg)	2023	533	10,200	2933 ± 763	20–100 μm	Present study
Caohai (China) (n/kg)	2021	1320 *	4260 *	2094 ± 923 *	0–0.5 mm	[2]
		—	—	1872 ± 1107 #		[8]
Poyang Lake (China) (n/kg)	2018	54	506	—	<0.5 mm	[38]
Taihu Lake (China) (n/kg)	2015	11	234.6	—	100–1000 μm	[39]
Dongting Lake (China) (n/kg)	2018	210	520	385 ± 696	<0.1 mm	[44]
Chao Lake (China) (n/kg)	2019	60	1064	308 ± 231	<1 mm	[6]
Nainital Lake (India) (n/kg)	2023	400	10,600	—	0.02–1 mm	[41]
Antuã River (Portugal) (n/kg)	2016	18	629	—	-	[42]
Fish						
Caohai (China) (mg/kg)	2023	ND	126.90	—	20–100 μm	Present study
Lake Ontario (Canada) (n/fish)	2015	3	915	59 ± 104	-	[45]
Northeast Atlantic (n/fish)	2013	0	2	—	1.0–2.0 mm	[46]
Poyang Lake (China) (n/fish)	2018	0	18	—	<0.5 mm	[40]

Note: “*” indicates the dry season; “#” indicates the wet season.

The RA-MP values in forest soils were more than twice those in cultivated soils in the Caohai watershed. In both CS-1 and CS-3, PC was the main component of MPs, accounting for 72% and 36%, respectively, while it accounted for only 11% of the MPs in CS-2. PC is widely used in construction, automotive applications, medical equipment, and electronic devices [47]. Sites CS-1 and CS-3 are both adjacent to residential gathering points, with CS-1 being especially close. Therefore, it is believed that PC pollution mainly derives from daily residential activities. The PC pollution near CS-2 was probably caused by the application of farmyard manure. At CS-2 and CS-3, PVC and EPN were the main MP components; we believe that this may be closely associated with the application of plastic film. The MP composition in all forestland soils was more complex than that in cultivated soils and marshy grassland soils. The level of human activity in forestland is relatively low, and the possibility of direct introduction from human activities is also low. Therefore, MP pollution in the forestland soils mainly originated from atmospheric

deposition. Atmospheric deposition and road traffic are important pathways for MPs to enter soils. For the first time, Dris et al. [48] revealed the presence of MPs in atmospheric fallout in Paris. They used a funnel in a glass bottle on the rooftop of a building to collect fallout samples for 3 months, and their research indicated that MP atmospheric fallout ranged from 29 to 280 particles per m^2/day . Klein and Fischer [49] reported that the median abundance of MPs in the Hamburg metropolitan area, which has a population of 1.8 million, reached 136.5 and 512.0 particles per m^2/day . Due to the coverage of dead branches and leaves on the forestland surface, surface runoff does not occur easily, which facilitates the accumulation of MPs in forest soil. In the present study, FS-3 is adjacent to Weining city, to the north (the wind direction in the Caohai watershed is mainly southerly). The RA-MPs in the soil sample from FS-3 was the highest among the forest samples. FS-1 is far from Weining city and major highways, and the RA-MPs in the soil sample from FS-1 was the lowest among the forest samples. Therefore, we speculate that MP pollution in forest soil is closely related to atmospheric deposition and is strongly influenced by wind direction. The main MP components in soil samples from marshy grasslands were PVC and PA, and those in the organs of grass carp were PE and PVC. These lands were once cultivated, and the PVC pollution was probably caused by the application of plastic film. PA is used mainly to manufacture components such as gears, bearings, and seals, and as a synthetic fiber, it is also used to make clothing, tents, and other textiles [50]. PE is often used to manufacture plastic bags, food packaging, etc., which may be part of domestic waste produced by residents [51]. Therefore, we speculate that MP pollution in the marshy grassland soils may mainly come from residual plastic film.

The MPs in sediment samples were more abundant and complex than those in water samples. Among the four inflow rivers, the Zhonghe River is the largest. Its flow is 3–5 times greater than that of the other three rivers. Therefore, the content and type of MPs in the Zhonghe River are closely related to the level of MP pollution in Caohai Lake (overlying water and sediment). In terms of the composition of MPs in the overlying water, the Zhonghe River (WR-1) has a considerable impact on the eastern, southeastern, and central parts of Caohai Lake (WL-1, WL-2, WL-3, and WL-4, respectively). The Baima River (WR-5) affects mainly the southeastern, southern, and central parts of Caohai Lake (WL-2 and WL-3), and its impact is extremely limited due to its low flow. The Dongshan River (WR-6) and Wanxia River (WR-8) affect mainly the western and northwestern parts (WL-5 and WL-4) of Caohai Lake. A comparison of the MP composition in water and sediment samples from each sampling site revealed that some MP types, such as PU, ACR, PVB, and PTEE, are prone to enrichment in sediments, and some MPs, such as PVC, PA, and PI, tend to be suspended in the overlying water and migrate with water flow. Additionally, an increase in water flow will shorten the residence time of MPs on the surface of the medium and reduce deposition. Small particle size media have a large specific surface area and provide more deposition sites. Rough surfaces enhance the physical interception effect [52]. When the medium saturation decreases, the capillary force generated at the water–air interface will capture microplastics and promote deposition. In soil detection, PET and PVC MPs were often misjudged as mineral impurities because their density was higher than that of the flotation solution, resulting in the abundance being underestimated by 30–50%. If high-density MPs are ignored in an ecological risk model, their potential impact on benthos and soil microorganisms may be underestimated [53]. Currently, the most used extraction solution is a cheap and easily available saturated NaCl solution. Some studies also used saturated NaI, sodium polytungstate, and ZnCl_2 solutions to extract MPs with a density of more than 1.2 g/cm^3 , and the extraction efficiency was higher. However, the unit price of these chemicals is high, and improper treatment of waste liquid can cause a burden on the environment. Through optimizing the flotation liquid formula, coupling it with

auxiliary technology, and establishing a standardized process, the separation efficiency can be significantly improved [54]. In the future, an intelligent flotation system combining artificial intelligence and automation technology is expected to represent a more accurate solution for evaluating the abundance of MPs.

4.2. Pollution Sources and Migration of MPs in the Caohai Watershed

Based on the present study and previous reports, there are probably five main sources contributing to MP pollution in the Caohai watershed (Figure 7). (a) Road traffic. MPs are emitted in large amounts in high-density traffic areas, and road runoff contributes significantly to deteriorated water quality in receiving waters [55,56]. The total length of highways in the Caohai watershed is 92.05 km. The MP pollution caused by road traffic is non-negligible. (b) Scattered residents. There are approximately 61,282 scattered residents in the watershed. MPs originate from their home activities, including the daily usage of plastic items, chemical products, etc. (c) Cultivated land. This is one of the main sources of MP pollution in the Caohai watershed. Due to the high altitude and low temperature, plastic film is widely used in agricultural activities. (d) Weining city. Municipal sewage contains many MPs, which mainly originate from plastic microbeads in daily chemical products, fibers in laundry wastewater, car tire wear debris, and other fragmented plastic waste. More than 90% of MPs can be removed in wastewater treatment plants. Even if the concentrations of MPs in effluent are low, the large volume of effluent discharged into the lake makes them non-negligible. Weining city has a population of approximately 320,000 and an urban area of approximately 36.47 km². It is also a main source of MP pollution in the Caohai watershed. In Gullbergsvass in the central part of Gothenburg (Sweden), the RA-MP concentration is 1500 n/L in stormwater, 5.10×10^4 n/L in washwater, and 2.60×10^6 n/kg dw in swept sand [57], and MPs ≥ 20 μ m were found to be dominated by tire wear particles, accounting for 38%, 83%, and 78% of stormwater, washwater, and swept sand, respectively. (e) Atmospheric deposition. MPs originating from urban areas (Weining city), scattered residents, cultivated soils, and road traffic can be driven into the atmosphere by wind. In addition, some MPs in the atmosphere originate from other areas and are carried by atmospheric flow. Most MPs in the atmosphere return to the ground (roads, buildings, cultivated land, marshy grassland, forestland, streets, water, etc.) through atmospheric deposition, and some of them are transported to other areas via atmospheric flow. As shown in Figure 7, most of the MPs from the above sources are transferred into Caohai Lake via river inflow, land runoff, and groundwater supply. In the water body (Caohai Lake), MPs in overlying water can be transferred into sediment via sedimentation, and MPs in sediments can be released into overlying water via suspension, uptake by submerged plants, intake by zoobenthos, etc. MPs in overlying water can also be adsorbed, concentrated, and magnified through the food chain. Additionally, some MPs migrate out of the water body along with outflow.

Notably, the RA-MP values in forestland soils were much greater than those in cultivated land soils and marshy grassland soils in the Caohai watershed. It is believed that MPs in forestland soils derive mainly from atmospheric deposition. This may be because the forest surface is covered with dead branches, fallen leaves, weeds, and other debris, forming a unique network with a loose structure that hinders surface runoff, leading to the infiltration of MPs into the soil. In addition, the RA-MPs in soils from site FS3, which is the closest to Weining city, was the greatest among all forestland soil samples, followed by that in soils from site FS2, which is the next closest site to Weining city. We believe that Weining city is the primary source of MP pollution in the Caohai watershed.

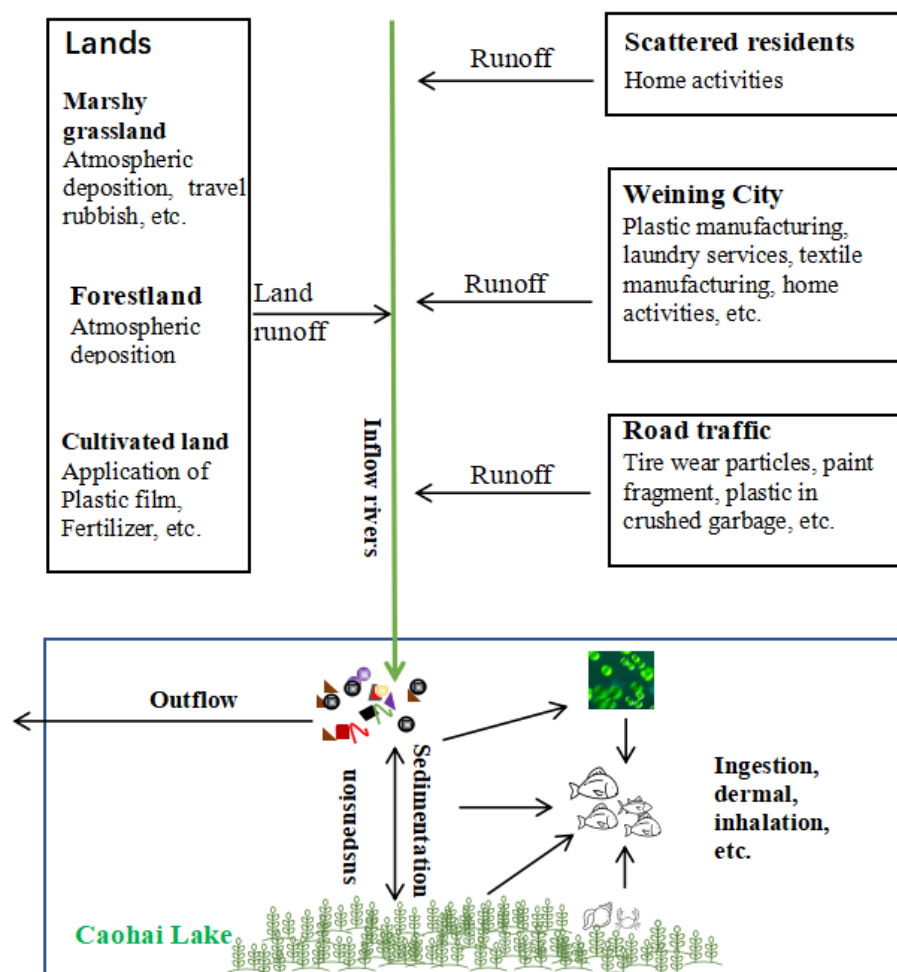


Figure 7. Schematic diagram of MP migration in the Caohai watershed.

5. Conclusions

In the present study, 20 kinds of MPs were detected in soil, water, and sediment samples in the Caohai watershed. Special attention should be given to PA, as it is widely distributed and abundant in soil, sediment, and water in the watershed. The RA-MP values in forestland soils were much greater than those in cultivated land soils and marshy grassland soils and much greater than those in sediments at some sites. The abundance of small MPs (20–100 μm) was much greater than that of larger MPs (100–500 μm). MPs may have originated from road traffic, Weining city, cultivated land, and scattered residents. Wind, land runoff, rivers, and atmospheric deposition are the main factors driving MP migration in the Caohai watershed. Most MPs accumulated in forestland soils and sediments, and some of the MPs moved out of the watershed via river and atmospheric transport. Four kinds of MPs, PE, PVC, PA6, and PA66, were detected in grass carp. PE was detected in all organs (stomach, intestine, skin, gill, and fish tissue), and PVC was detected in the intestine, skin, gill, and fish tissue. Our results suggested that although there is moderate MP pollution in soil, water, sediment, and grass carp in comparison with the other areas, it is necessary to pay attention to PE and PVC migration via the various environmental media and the risks associated with consuming the local grass carp, which is in accordance with our hypothesis and objectives. The local government can make several policies to reuse and recycle agricultural film to alleviate local PE and PVC pollution.

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Abbreviations

Polyurethane (PU), acrylate copolymer (ACR), ethylene vinyl acetate (EVA), polyvinylchloride (PVC), polymethylmethacrylate (PMMA), polyamide (PA), polypropylene (PP), polyimide (PI), polytetrafluoroethylene (PTEE), polyvinyl butyral (PVB), polyethylene terephthalate (PET), epoxy phenol novolac (EPN), polysulfone (PSF), ethylene acrylic acid (EAA), fluororubber (FKM), polycarbonate (PC), polyethylene (PE), polystyrene (PS), and fluorosilicone rubber (FMQ).

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