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Microplastic pollution in the rivers of the Tibet Plateau☆

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ABSTRACT

The Tibet Plateau, the so-called Third Pole of the world, is home to the headstreams of many great rivers. The levels of microplastic pollution in those rivers, however, are unknown. In this study, surface water and sediment samples were collected from six sampling sites along five different rivers. The surface water and sediment samples were collected with a large flow sampler and a stainless steel shovel, respectively. The abundance of microplastics ranged from 483 to 967 items/m^3 in the surface water and from 50 to 195 items/kg in the sediment. A large amount of small, fibrous, transparent microplastics were found in this study. Five types of microplastics with different chemical compositions were identified using micro-Raman spectroscopy: polyethylene terephthalate (PET), polyethylene (PE), polypropylene (PP), polystyrene (PS), and polyamide (PA). These results demonstrate that rivers in the Tibet Plateau have been contaminated by microplastics, not only in developed areas with intense human activity but also in remote areas, where microplastic pollution requires further attention.

1. Introduction

Plastic production has risen from 0.5 million tons per year in 1950 to 330 million tons in 2016 (PlasticsEurope, 2017). White pollution caused by large single use items such as plastic bags, has received extensive attention, while the smaller, more harmful microplastics have only been the subject of research since the beginning of this century (Browne et al., 2007). Traces of microplastics exist not only in surface water, but also in deep water, sediment, soil, and organisms (Bergmann et al., 2017; Chae and An, 2018; Zhang et al., 2018a,b). Microplastics, with their particular sizes and stable properties, are the breeding places for microorganisms and carriers of pollutants. Plasticizers, flame retardants, and other chemicals are added to plastic products to improve their performance. These chemicals can be released by microplastics into the environment, creating complex ecological risks (Liu et al., 2019). Microplastics can also be swallowed by aquatic animals, seabirds, or mammals; once accumulated in the food chain, they eventually enter the bodies of the top predators, including humans (Barboza et al., 2018; Nelms et al., 2018).

With respect to the presence of microplastics in water, the importance of marine microplastic pollution research is self-evident. However, freshwater areas are of great significance. Most microplastics in the ocean come from land. Freshwater ecosystems play an important role in microplastic transportation. The investigation of about the locations and characteristics of microplastic pollution in freshwater areas is helpful for conducting a source analysis of marine microplastics and the subsequent formulation of the relevant policies (Choi and Lee, 2018; Iacovidou et al., 2019). Moreover, the widespread presence of microplastics in water sediment and soil in freshwater areas threatens the environment, as microplastics can be distributed and have an impact on organisms (Fossi et al., 2016; Jin et al., 2018). Freshwater areas are in more direct and frequent contact with humans than oceans. Upstream input, domestic sewage and solid waste are potential sources of microplastics in freshwater areas (Li et al., 2018a,b,c; Xiong et al.,

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2018); The increase in microplastic pollution, and its impact on humans is beginning to receive more attention (Zhang et al., 2018a,b). In recent years, many studies on microplastics in freshwater areas have been published, with most concentrating on developed areas with intense human activity (Giovacchini et al., 2018; Wang et al., 2017a,b; Wen et al., 2018a,b). It is reasonable for the majority of research to focus on populated areas, since microplastics, as artificial products, must be derived from human activities. Remote areas tend to be sparsely populated, with microplastics originating from fewer sources than in populated areas. However, due to the lack of effective management, microplastics have also been found in remote areas, such as Lake Hovsgol, Mongolia and Siling Co, China (Free et al., 2014; Zhang et al., 2016). Some light microplastics can also reach remote areas via atmospheric transport (Zhang et al., 2016). Although microplastic concentrations in remote areas may be relatively low, they should not be ignored. In this study, we investigated the abundance, distribution, and sources of microplastics in the rivers of the Tibet Plateau. Our results can be used to augment the current database of microplastic pollution and provide useful references for further research.

2. Materials and methods

2.1. Study area

The Tibet Plateau has an average elevation of more than 4000 m with the largest horizontal and vertical extent in the world. The fragile ecosystem of the Tibet Plateau faces unprecedented challenges due to global climate change and human activities (Ma et al., 2018; Sheng et al., 2019). The melting of glaciers, desertification of land, erosion of soil, and pollution of water all threaten the area's ecology. The Tibet Plateau is home to the headstreams of many important rivers in eastern Asia, including the Yangtze River, Yellow River, Nujiang River, and Brahmaputra River. These rivers are of particular significance to many East Asian countries. Water quality of these rivers is critical to the lives of billions of people. Although some researchers have investigated heavy metals and other pollutants on the Tibet Plateau (Bing et al., 2018; Li et al., 2018a,b,c), microplastics, as an emerging pollutant, have only been examined in the sediments of four lakes in the area (Zhang et al., 2016).

In this study, six river sampling sites along National Roads 109 and 318 were selected. We collected surface water and shoreline sediment. A Global Positioning System (GPS, GEOXT, Trimble, Sunnyvale, USA) was used to determine the location of the sampling sites, which were from the Buqu River (the source of the Yangtze River), Naqu River (the upper part of the Nujiang River), Lhasa River (a tributary of the Brahmaputra River), Brahmaputra River, and Nyang River (another tributary of the Brahmaputra River). The data from the sampling sites are presented in Fig. 1 and Table S1. Basic information for the cities and villages in the study area is presented in Table S2.

2.2. Sampling

All sampling was completed within 4 days (9–12 July 2018). The collection of surface water samples was performed using a large flow sampler (KLL-S4, SEBA, Germany) (Jiang et al., 2018; Wang et al., 2017a,b; Wang et al., 2018a,b). The sampling methods employed were from published studies, with some modification (Baldwin et al., 2016; Su et al., 2018). During the collection of each sample, 30 L of surface water was filtered with a 0.045 mm stainless steel sieve. Three replicate samples were collected using the same method at each sampling site. All solids on the sieve were flushed into a 500 mL sampling jar using a pressure spray pot containing deionized water. We then added 5% formalin solution to the sampling jar and stored it in a sampling box (Wang et al., 2017a,b; Zhang et al., 2017). The top 2 cm of the sediment in a 0.04 m^2 range area between the shoreline and water edge was collected using a stainless steel shovel. Five randomly selected sites more than 20 m apart were selected for sample collection, with a sample quantity of approximately 200 g. All sediment samples were then mixed, wrapped in aluminum foil, and stored in a sample box. The samples were then separated and sealed to avoid contamination before the laboratory process.

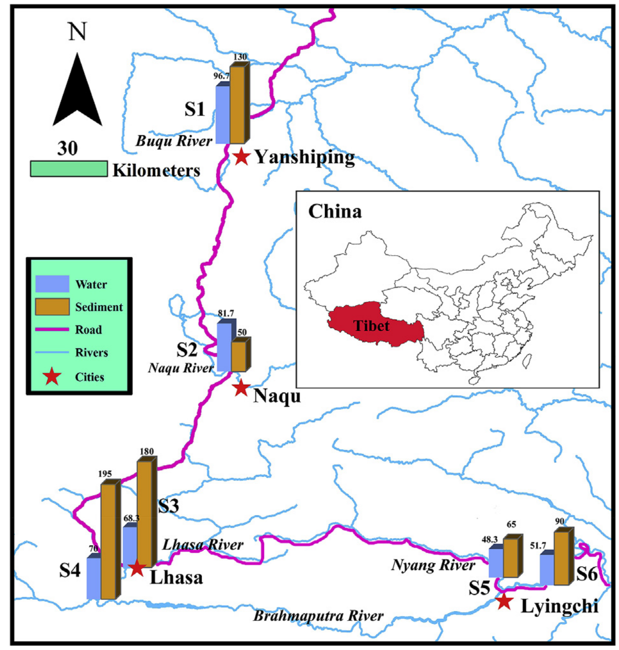

Fig. 1. Sampling sites and abundance of microplastics
(The unit of abundance is 10 items/m^3 in surface water and items/kg in sediment).

2.3. Sample analysis

Microplastics were separated from the surface water and sediment samples for observation. Wet peroxide oxidation (WPO) was used to remove organic matters (Baldwin et al., 2016; NOAA, 2015). We used 20 mL ferrous sulfate solution as a catalyst, 40 mL hydrogen peroxide solution (30%) was used as an oxidant to decompose organic matters. Sand and minerals were then removed by density separation. The water samples were flushed into a precleaned 1 L beaker and oven dried at 70 °C before zinc chloride solution (1.5 g/cm^3) was added. After settling for 24 h, the supernatant was filtered through a 0.22-μm pore size GF/C filter (Membrane Solutions LLC., Kent, WA, USA).

For the treatment of the sediment samples, sediment samples were dried in an oven at 70 °C. The dried sediment samples were put through a 2 mm stainless steel sieve to remove impurities. At least 300 g of the sediment sample from each site remained after the samples had been dried and sieved. For each site, 50 g (dw) of the sediment sample was randomly selected and weighed in a precleaned glass beaker. A zinc chloride solution was added to disaggregate the dried sediment, and this mixture was stirred with a magnetic mixer. The supernatant was then extracted and treated in the same manner as the surface water samples described above. After digestion and density separation, microplastics from the sediment samples were filtered onto GF/C filters.

These filters were then photographed, and the shapes, sizes, and colors of the particles were recorded using a stereoscopic microscope at a 100-fold magnification (SZX7, Olympus, Japan) (Blumenröder et al., 2017; Pan et al., 2019; Yu et al., 2018). The microplastics fell into three categories based on the length of each particle's longest edge: <0.5, 0.5–1, or 1–5 mm. The collected particles were then divided into three categories based on their shape: fiber, fragment, or pellet (Gray et al., 2018; Wang et al., 2018a,b). Suspected microplastics were identified using the criteria proposed in published studies (Free et al., 2014; Mani et al., 2016). A fiber was considered to be a long, thin line with a slender shape, a fragment was a piece of debris from a larger plastic item, and a pellet was a microplastic having a spherical or cylindrical shape. Many published studies tested only one sample at each sampling site; to obtain more convincing results, we analyzed two samples at each sampling site. (Di and Wang, 2018; Kataoka et al., 2019). A Renishaw inVia Raman spectroscope (Wotton-under-Edge, Gloucestershire, UK) was used for further identification. The wavelength of the incident laser was set to 532 nm and the Raman spectra ranged from 50 to 3500 cm^{-1}. The suspected particles were transferred using tweezers to a silicon wafer for Raman spectroscopy testing with the exception of those that were too small to move (Wen et al., 2018a,b; Xiong et al., 2018; Zhang et al., 2017; Zhao et al., 2015).

2.4. Quality assurance and quality control

All equipment was carefully cleaned with deionized water 3 times before use and wrapped in foil when not in use. In order to avoid potential pollution from the laboratory, all processes were performed in a clean room with an intelligent air shower system (Fig. S1). During sampling and laboratory analysis, all researchers wore nitrile gloves and cotton coats. Blank tests were used to evaluate potential contamination from the laboratory. In each group of the blank test, 30 L of deionized water was filtered through the GF/C filters instead of the samples in the laboratory. The experimental table and experimental instruments of the blank tests were consistent with those of the official test. Six blank tests were conducted and six filters were obtained. These six filters were placed uncovered on the experimental table for 48 h and then observed using a stereoscopic microscope (100x). No suspected microplastics were found in these filters. Thus, the results of the blank test showed that the potential pollution from the laboratory could be ignored.

2.5. Data analysis

The abundance of microplastics in the surface water is expressed in items/m^3; in the sediment, it is expressed in items/kg. All statistical analyses were performed using SPSS Statistics (22.0, IBM, USA). A one-way ANOVA was used to determine the differences in the abundance of the microplastics among the different sampling sites. Tests with $p < 0.05$ were considered to be statistically significant. ArcGIS (10.3, Esri, USA) was used to describe the geographic location.

3. Results

The microplastic abundance of each sampling site is shown in Table 1. The abundance of microplastics varied from 483 to 967 items/m^3 in the surface water. The highest microplastic abundance in surface water was found at the Buqu River (S1), located at the border between Tibet Province and Qinghai Province. The lowest concentration of microplastics was found at upstream Nyingchi (S5). In the sediment samples, the abundance of microplastics varied from 50 to 195 items/kg, with the highest concentration found at downstream Lhasa (S4) and the lowest at Site S2. Site S2 is located in the Naqu River, which is near a rural area.

Table 1
Abundance of microplastics collected from the surface water and sediment.

Sites	Surface water	Sediment
S1	967 ± 141 items/m^3	130 ± 71 items/kg
S2	817 ± 589 items/m^3	50 ± 7 items/kg
S3	683 ± 354 items/m^3	180 ± 42 items/kg
S4	700 ± 94 items/m^3	195 ± 64 items/kg
S5	483 ± 118 items/m^3	65 ± 21 items/kg
S6	517 ± 24 items/m^3	90 ± 14 items/kg

In keeping with the suggestions of many published research, the microplastics in this study were classified by shape, color, and size. Fig. 2 presents photographs of typical microplastics that were taken with a digital camera attached to a stereoscopic microscope. Fig. 3a shows the proportion of different shapes of microplastics collected at each sampling site. Fiber was the most common shape among the collected microplastics in both the surface water and sediment samples, accounting for 69.0%–92.7% in the surface water and 53.8%–80.6% in the sediment. The rest of the particles were either pellets or fragments. The proportions of the collected microplastics of different colors are shown in Fig. 3b. While transparent microplastics were the most common, blue, black, white, red and green microplastics were also in this study. Fig. 3c shows the different sizes of the collected microplastics. In this investigation, microplastics smaller than 1 mm accounted for more than 70% of the total microplastics found in the surface water and sediment.

Following the preliminary visual identification, suspected microplastic particles collected were further identified using micro Raman spectroscopy. The results are shown in Fig. 4. Five kinds of microplastics with different chemical compositions were discovered. Polyethylene terephthalate (PET) was the most common microplastic type found in the sediments and polyethylene (PE) was dominant in the surface water samples. The polymer types of other microplastic particles were identified as polypropylene (PP), polystyrene (PS), and polyamide (PA).

4. Discussion

4.1. Levels of microplastic pollution

Our results indicated that all the rivers represented in this study were contaminated by microplastics that were found in all the surface water and sediment samples. Comparison of the results from other rivers and the rivers in this study are presented in Table 2 and Table 3. Since there is no established standard for evaluating microplastics, the unit of microplastic abundance has not been clarified in related studies. Some researchers use items/m^3 for water and items/kg for sediment, and others use items/m^2 for both water and sediment. Since abundance levels with different units cannot be compared, the levels of microplastic pollution in this study were only compared with studies that used the same units of abundance.

The differences in the abundance of microplastics in rivers around the world are significant. According to published studies, reasons for these differences include source loading, hydrodynamic conditions and geographical conditions (Bordós et al., 2019; Gray et al., 2018; Kataoka et al., 2019). In surface water, the abundance levels of microplastics in the Amsterdam Canal and the Hanjiang River are several to 10 times higher than those of the rivers in this study. It is worth noting that the Buqu River (S1), Three Gorges Reservoir, and Yangtze River estuary are different parts of the same river, and the microplastic pollution level in the surface water of the

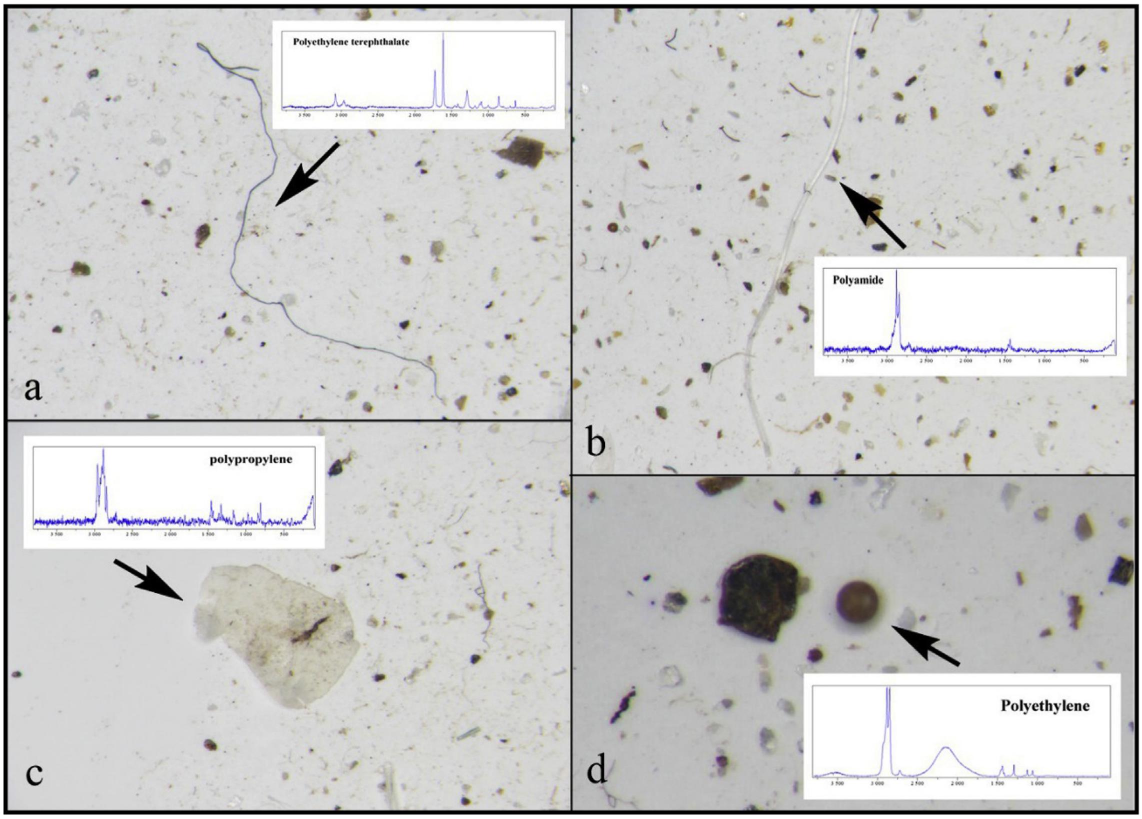

Fig. 2. Photographs of typical microplastics from rivers in the Tibet Plateau, colorful fiber (a), transparent fiber (b), fragment (c), and pellet (d).

Buqu River is significantly lower than those of the two downstream sections ($p < 0.05$). In the sediment samples, the concentration of microplastics was also relatively low, but the difference was not as obvious as that in the surface water; a similar situation is reported with studies on the Danube River (Lechner et al., 2014). With the exception of the Rhine River, Meuse River, and Huangpu River, all of which are associated with intense human activity, the levels of microplastics in the sediments in this study did not differ in the order of magnitude from those of other rivers.

Given the limited human impact in the study area, the fact that lower levels of microplastic pollution were discovered in this investigation is a reasonable result. Published studies suggest that the level of microplastic pollution is related to population density and urbanization. Relatively intense human activities, therefore, have led to high concentrations of microplastics (Mani et al., 2016; Wang et al., 2017a,b; Wen et al., 2018a,b). However, that even remote rivers in the Tibet Plateau are not immune to microplastic contamination. Microplastic pollution still needs to be considered when studying ecologically fragile areas such as the Tibet Plateau (Fan et al., 2019; Li et al., 2018a,b,c).

4.2. Sources of microplastics

The rivers in the Tibet Plateau have fewer sources of microplastics than developed areas, and many common sources can be ignored. In general, upstream input, fisheries and ship navigation are common sources of microplastics in rivers (Kiessling et al., 2019; Peng et al., 2018). The sampling sites in this study were located in the source area of the rivers. There were no fisheries and ships near the six sampling sites. Therefore, we suggest that damaged fishing nets and paint peeling off ships were not sources of microplastics. In addition, there is very little industrial activity along the rivers. Therefore, we suggest that the microplastics originated primarily from the daily activities of residents and tourists. On the Tibet Plateau, the way of life and standard of living of its inhabitants are approaching those of more developed areas. Plastic products have become indispensable. Plastics are used in food packaging, household goods, clothing and furniture. Daily activities, such as bathing and oral hygiene, all lead to the production of microplastics. The shedding of fibers from clothes and plastic pellets in pharmaceuticals and personal care products (PPCPs) may also be sources of the microplastics discovered in rivers (Kalčíková et al., 2017). Since plastic fibers have a small mass and can be transported over long distances by the wind; in other regions, the urban areas themselves are sources of microplastics. Moreover, garbage discarded by residents and tourists is an important source of plastic waste. This plastic waste can be fragmented into secondary microplastics under a variety of conditions, with some studies suggesting that the intense UV radiation on the Tibet Plateau can accelerate the process (Nel et al., 2018).

The polymer composition of the microplastics collected in this study is compatible with the above hypothesis. In the Raman spectroscopy identification, the most common chemical compositions were PET and PE. PET is a common ingredient for clothing and water bottles, most colored fibers and some transparent fragments (Wang et al., 2017a,b). Numerous disposable products, such as disposable cutlery and bags, are used in the research area, most of which are made of cheap and lightweight PE (Zhang et al., 2017). PE, a common additive of in PPCPs, is also used for plastic pellets. The rest of the plastic pellets in the samples were identified as PS (Su et al., 2016). PS is used to make rubber and disposable tableware, which are commonly seen in the study area. A small number of microplastic particles were identified as PP or PA, which are used in the manufacture of various fabrics and bags (Kapp and Yeatman,

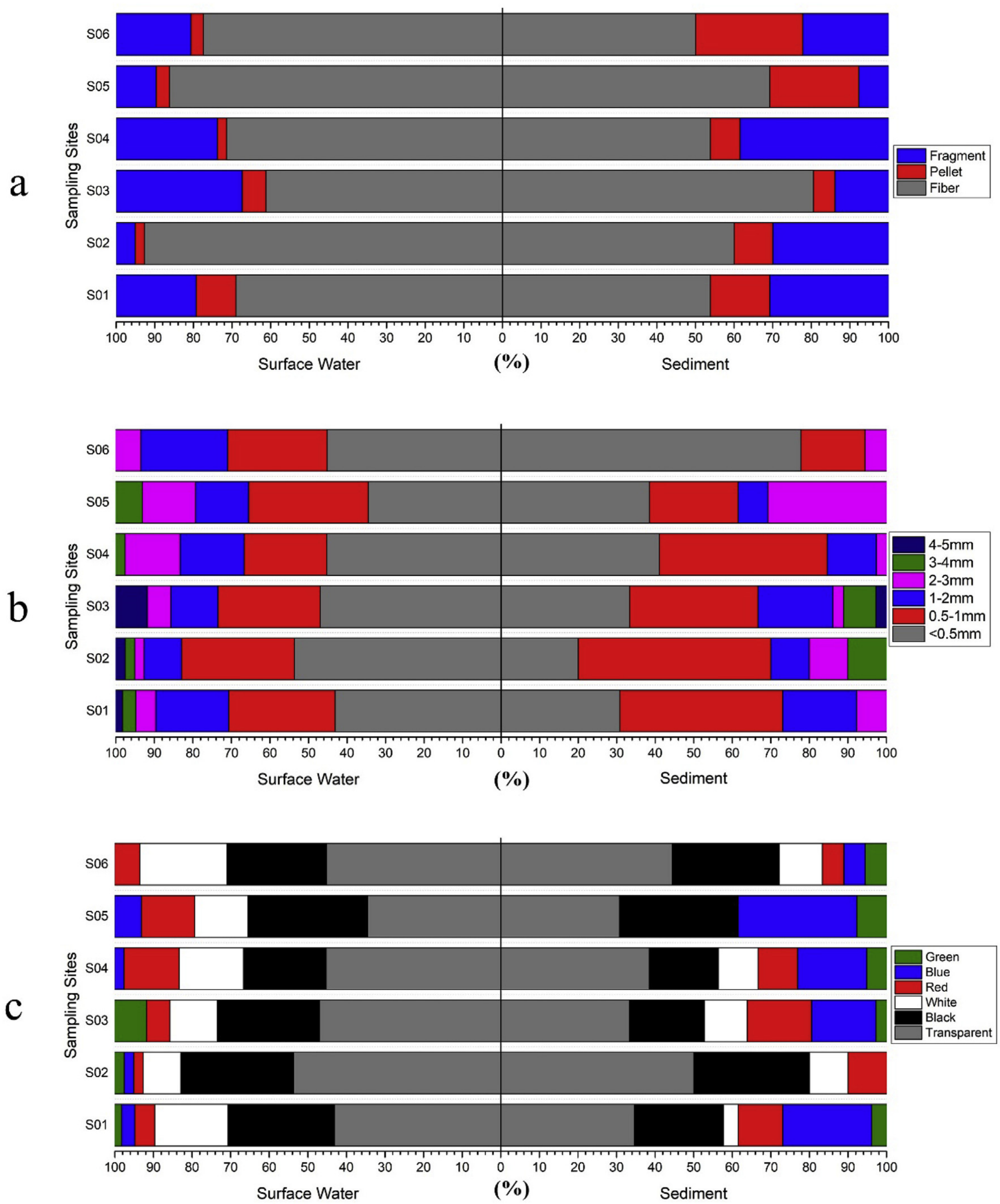

Fig. 3. Proportion of microplastics' shape (a), color (b), and size (c). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

2018; Klein et al., 2015).

The morphology of the collected microplastics is indicative of their origin. Transparent microplastics were found in large amounts. In general, inferior disposable products—such as disposable tableware and bags—are made of transparent plastic (Su et al., 2016). The digestion protocol used in this study, however, did tend to bleach colored particles, possibly leading to an overestimation of the number of transparent particles (Wen et al., 2018a,b). Inferior disposable products, which are large and easily broken or shredded, comprise a major source of microplastics. At present, many countries, including China, restrict the production and use of these products (Peng et al., 2018). Although the proportion of transparent plastics produced is larger than that of other colors, most of the microplastics collected in this study were colored. Some products need to be colored in order to be used effectively, such as certain kinds of containers, machinery, and equipment. In addition, since the sampling sites were all located near roads, some black particles may have come from car tires, while the white particles may have originated from road paint (Turner, 2018).

Solid waste and domestic wastewater are considered sources of fragments, while the pellets and fibers come from decomposed

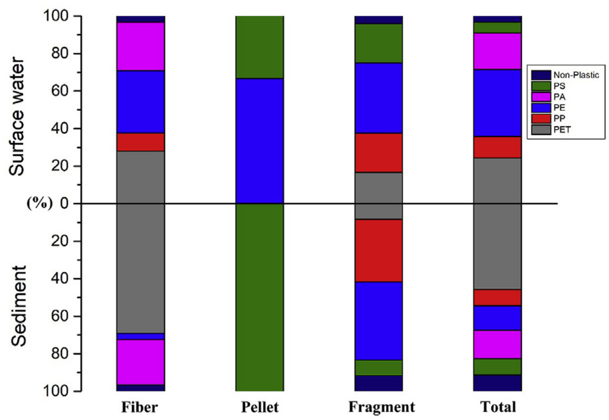

Fig. 4. Results of the micro-Raman spectroscopy identification.

fabrics. Fiber was the most commonly found shape in this study. With the development of the national economy, almost all Chinese households now have washing machines. These appliances may clean clothes effectively, but they produce a large amount of microplastics in the process (Hernandez et al., 2017; Napper and Thompson, 2016). In addition to the sources of microplastics mentioned above, microplastic sources specific to the Tibet Plateau should not be ignored. Lungtas and tents, which are ubiquitous on the Tibet Plateau, are typical examples (Fig. S2). Most Lungtas, which are small flags used for religious blessings, are now made of artificial fabrics, such as PET, PE, and PA. Some of these flags are burned, while others are discarded religious activities. Tents are simple forms of shelter for which plastics have replaced traditional animal skins and natural fibers as raw construction materials. Plastic tents are also sources of fragment and fiber microplastics.

4.3. Distribution of microplastics

Although all the sampling sites in this study had lower microplastic pollution levels than those of other rivers, some variability in the distribution of microplastics was observed. In the sediment, relatively high concentrations of microplastics were found in the sediment at sampling sites near Lhasa ($p < 0.05$). Lhasa, the capital of China's Tibet Autonomous Region, is the political, economic, cultural, scientific, and educational center of Tibet; it is also a holy site of in Tibetan Buddhism and a famous tourist attraction. In addition to its nearly one million permanent residents, Lhasa hosts 30 million visitors per year. In this study, there were two sampling sites in the Lhasa area, of which S3 was located in the urban area and S4 was located downstream of Lhasa. Residents and visitors contributed a large amount of microplastics to this part of the study area. Human activity at the remaining sampling sites was relatively sparse. Therefore, it was reasonable to find lower concentrations of microplastics in the sediments of these rural sites.

Published studies have suggested that the complete scientific management of wastewater and solid waste is the key to solve the problem of microplastic pollution. Related environmental protection measures are particularly important in ecologically fragile areas, such as the Tibet Plateau (Free et al., 2014; Sheng et al., 2019; Zhang et al., 2016). As the main source of microplastics, the proper treatment of wastewater and solid waste is of fundamental importance in the control of microplastic pollution. The Lhasa sewage treatment plant went online in 2001, while the Lhasa municipal solid waste landfill commenced operation in 2002. These facilities have helped to reduce microplastic pollution from wastewater and solid waste. The local government officially began to construct the second phase of the Lhasa Municipal Wastewater Treatment Plant and the second phase of the Lhasa Municipal Landfill Project in 2014; construction was based on the original sewage treatment plant and landfill site. These projects will provide improved help for the prevention and control of microplastic pollution in the future (Eckert et al., 2018; Li et al., 2018a,b,c). However, despite the low concentration of microplastics in the sediment samples, the overall level of microplastic pollution in the surface water of the rural areas was no better than that in the Lhasa area ($p > 0.05$). Although the rural areas have a lower population density than Lhasa, these regions lack facilities to properly dispose of solid waste and wastewater (Free et al., 2014; Xiong et al., 2018). Thus, a large amount of wastewater and solid waste is not properly treated before being discharged into the river. In addition, the

Table 2
Abundance comparison with other rivers (surface water).

Surface water	Location	Abundance (items/m^3)	Methodology	Lower limit (μm)	Reference
Yangtze River Estuary	China	500- 10,200	Pump 12–20 L	500	Zhao et al., (2014)
Amsterdam canal	Netherland	4800-18,700	Bulk water 2 L	10	Leslie et al., (2017)
Hangjiang River (Wuhan)	China	2600–3200	Pump 20 L	50	Wang et al., (2017a,b)
Yangtze River (Wuhan)	China	1450–4000	Pump 20 L	50	Wang et al. (2017a,b)
Antua River	Portugal	58–1265	Pump 1200 L	55	Rodrigues et al., (2018)
Three Gorges Reservoir	China	1594- 12,611	Pump 25 L	48	Di and Wang, (2018)
Gallatin River	USA	1- 67,500	Bulk water 1 L	100	Barrows et al., (2018)
Rivers in Tibet plateau	China	483–967	Bulk water 30 L	45	This study

Table 3
Abundance comparison with other rivers (sediment).

Sediment	Location	Abundance (items/kg)	Methodology	Reference
Rhine River	Germany	228–3763	Shovel	Klein et al., (2015)
Rhine & Meuse River	Netherlands & Germany	1400–4900	Grab Sampler	Leslie et al., (2017)
Yangtze River Estuary	China	20–340	Box corer	Peng et al., (2017)
Beijiang River	China	178–544	Shovel	Wang et al. (2017a,b)
Xiangjiang River	China	27–866	Shovel	Wen et al., (2018a,b)
Antua River	Portugal	18–629	Grab Sampler	Rodrigues et al., (2018)
Huangpu River	China	53–1600	Shovel	Peng et al., (2018)
Three Gorges Reservoir	China	25–300	Grab Sampler	Di and Wang, (2018)
Rivers in Tibet plateau	China	65–195	Shovel	This study

scattered and frequent migration of herdsmen using plastic tents has made it difficult to prevent and control microplastic pollution. The vast area of the Tibet Plateau, the long journeys of many of its inhabitants, and generally inconvenient transportation have all proven to be challenges in the prevention and control of microplastic pollution in this area, especially in rural areas. In addition to the expansion of existing environmental protection facilities, much effort should be expended to prevent and control microplastic pollution in the Tibet Plateau.

5. Conclusion

In this study, we have investigated and discussed the abundance, morphology and chemical composition of microplastics in the rivers of the Tibet Plateau. Fiber-shaped and small-sized microplastics dominated the samples. The amount of PET and PE microplastics was greatest in sediments and surface water, respectively. The abundance of microplastics in the sediments of urban areas was higher than that in rural areas. There was no statistical difference between the abundance of microplastics in the surface water of urban and rural areas. We suggest that the main source of microplastics is daily human activities. Improving the facilities used to treat solid waste and wastewater is the key to dealing with the microplastic of microplastics. This research fills a gap in the study of microplastic pollution in the Tibet Plateau. Nonetheless, the occurrence of microplastic pollution at different sites and in different seasons still requires additional research.

Declarations of interest

None.

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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.2019.03.022.

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