



Microplastic pollution in water and fish samples around Nanxun Reef in Nansha Islands, South China Sea

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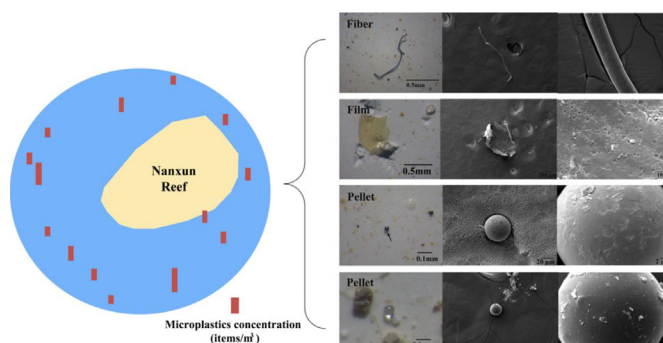
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HIGHLIGHTS

- The first report on microplastics pollution in waters and fish from Nansha Islands
- Blue microbeads were the dominant type in surface water from Nanxun Reef.
- Fibers were the main shape in fish samples from Nansha Islands.
- PVC was the major polymer type in surface water from Nanxun Reef.

GRAPHICAL ABSTRACT



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ABSTRACT

Nanxun Reef is one of the typical reefs in Nansha Islands, South China Sea. As the Nansha Islands are surrounded by certain developing countries, the economic and population growth have resulted in increased surface runoff of persistent organic pollutants in offshore areas. Microplastic has been found in many freshwaters and sea areas in recent years. However, the levels of microplastics contamination in Nansha Islands are still uncharted. In this study, 15 water and 35 fish samples were collected around the Nanxun Reef. The average concentration of microplastics was 1733 items/m³ for surface water samples and 3.1 items per individual for fish samples. The majority of ingested microplastics by fish were fibers, mostly transparent or blue. In surface water samples, blue microbeads were the main types of microplastics, accounting for 76.5% of all the detected particles. The main size of microplastics was <0.5 mm both in water and fish samples. Our results demonstrated that fishery activities and human domestic sewage might be the dominant sources of microplastic pollution in the Nansha Island, South China Sea.

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1. Introduction

Plastic is widely used in industrial production and human everyday life. In biomedicine, plastics act as the packaging material for many medicinal products. And some polymers are promising candidates for wound healing applications (Ma et al., 2019). Cui (Cui et al., 2018) reports a new way of using Polydimethylsiloxane-Titania Nanocomposite

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Coating to protect metals from corrosion. Various polymeric materials exhibit potential applications in electromagnetic interference shielding (Jiang et al., 2019), such as polyacetylene and polyaniline (Hu et al., 2019). Some natural polymers and products also strengthen the plastics pollution in the environments (Shi et al., 2019; Yang et al., 2019). Though some recycled polymers have been reported (Kong et al., 2018a; Kong et al., 2018b; Li et al., 2017), traditional plastics are still used extensively. Plastic residues can be transported by external forces such as rivers, ocean currents and wind power in the natural environment (Cole et al., 2011). Based on previous studies, plastic debris was found even in remote corners of the earth (Kuhn et al., 2018; Munari et al., 2017; Reed et al., 2018; Trevail et al., 2015). Due to their stability, plastic debris are normally removal-resistant (Cole et al., 2011). These debris will be degraded into tiny particles when it was subjected to ultraviolet radiation, wave impact or weathering factors (Barnes et al., 2009; Gewert et al., 2015). Tiny granules with size <5 mm are termed microplastics. Microplastics particles derived from degradation of large plastic debris, as stated, are defined as secondary microplastics (Ryan et al., 2009). Besides, plastic particles manufactured purposefully in diameter <5 mm are called primary microplastics, such as microbeads from cleansers and toothpastes (Cole et al., 2011). When decomposed into small particles, physical degradation and photodegradation of plastic debris can be greatly weakened, resulting in the enrichment of microplastics in soil or sediment for a long time. Therefore, as an emerging contaminant, microplastic pollution has become a hot issue of common concern in the world (Halden, 2015).

Microplastic pollution exists in most parts of the whole world, which is widely detected in sediments (Alomar et al., 2016; Peng et al., 2017), soils (Liu et al., 2018; Zhang and Liu, 2018), surface water of oceans, estuary (Lin et al., 2018; Mani et al., 2015; Xu et al., 2018) and lakes (Anderson et al., 2017; Su et al., 2016). Microplastics could absorb other pollutants from the surrounding environment, such as polychlorobiphenyl (PCBs), dichloro diphenyltrichloroethane (DDT) and heavy metals, acting as vectors to accumulate and transfer pollutants into organisms (Teuten et al., 2009; Ziccardi et al., 2016). Microplastics could also release toxic additives like plasticizers (Teuten et al., 2009). In addition, microplastic particles could be eaten mistakenly as food by shellfish, fish (Digka et al., 2018; Wright et al., 2013), seabirds (Amelineau et al., 2016), sea turtles (Duncan et al., 2019; Pham et al., 2017) and even mammals (Hernandez-Gonzalez et al., 2018). Ingestion of microplastic particles has been reported to negatively affect the organisms, including organ damage, gastrointestinal obstruction and growth restriction (Wright et al., 2013). The cumulation and transport of microplastics in food web pose a potential threat to human health (Teuten et al., 2009).

As an advanced and productive marine ecological system in the world, coral reefs support high marine biodiversity and absorb carbon dioxide, thus alleviate greenhouse effect and provide ecosystem services for humans (Hughes et al., 2017; Ko et al., 2014). However, this specific ecosystem has degraded seriously because of human influence and destruction in recent years. Persistent organic pollutants (POPs) contamination caused by anthropogenic activities has been detected in coral reef, including polychlorinated biphenyls and dichlorodiphenyltrichloroethane (Li et al., 2019). Studies have found that microplastics may compose a main threat for reef by restraining coral growth and reduce calcification (Chapron et al., 2018). Tang J reported that the immune system and anti-stress capacity of *Scleractinian coral P. damicornis* would be damaged when exposed to microplastics (Tang et al., 2018).

The South China Sea (SCS) is a semi-closed sea in the Pacific Ocean. Marine environment is deeply affected by anthropogenic activities through all sorts of pollution (Wang et al., 2017). As the natural fishery for surrounding countries, pollution in SCS is getting worse and worse, causing by the fishing activities and anthropogenic activities from several developing countries (Li et al., 2019). Lately, a large number of microplastics were found in the SCS (Cai et al., 2018). Nanxun reef is

one of the coral reef atolls in Nansha Islands, microplastics may also be found here under the ocean currents of the SCS, contributed by the industrial development and rapid population growth in the surrounding countries. However, studies about microplastic pollution in the SCS is still deficient.

In this study, we investigated the abundances, distribution, and morphological specificity of microplastics in the surface water and fish samples from Nanxun Reef. This research could offer basic data for monitoring microplastic pollution in the South China Sea.

2. Materials and methods

2.1. Sample collection

Surface water was sampled from 15 points (Supplementary Table S1) in the Nanxun Reef in May 2018. Twenty liters of water was collected with a sampler and then filtered with a 48 μm steel sieve. Pure water was used to rinse the residue on the sieve and transferred to a glass bottle. Two replicates were sampled at each site. Samples were stored at 4 $^{\circ}\text{C}$ before laboratory processing. A total of 35 fish samples were captured by artisanal fishing in areas surrounding the Nanxun Reef in May 2018. The weight, body width and length of each fish sample were recorded (Table 1).

2.2. Microplastic extraction

Before laboratory treatment, 30% H_2O_2 was added to water samples to dissolve the organic substances (Hidalgo-Ruz et al., 2012; Nuelle et al., 2014). And then the water was filtered pass a 0.45 μm filter papers under vacuum. The filter papers were stored in a clean glass dish.

For the treatment of fish samples, the stomach, esophagus and intestine of each sample were put into a clean glass bottle. 150 mL of KOH (10%, V/V) was used for dissolving the organismal tissue. Bottles were deposited in a constant temperature incubator at 60 $^{\circ}\text{C}$ for 7 days (Ghosal et al., 2018). To increase the density of the solution, 500 mL of zinc chloride solution was added to each bottle (Jabeen et al., 2017). Hydrochloric acid solution was used to neutralize precipitations. After standing for 24 h, the supernatant was filtered with a 0.45 μm filter papers under vacuum and the filter papers were stored as mentioned above.

2.3. Microscope inspection

A stereomicroscope (Optec SZ680, China) was used to scan the particles on the filter paper. The observed microplastics were classified as four types (Fig. 1) based on their shape: fiber, pellet, fragment and film (Cole et al., 2011; Hidalgo-Ruz et al., 2012). A fiber is a long and thin line with a cylindrical or slender shape. A fragment was characterized as a piece of debris. A film appears in the shape of slice. A pellet was considered to a three-dimensional sphere. Hemispherical transparent particles are mostly algae (Yan et al., 2019). Microplastics were divided into five classes based on their size: <0.5 mm, 0.5–1 mm, 1–2 mm, 2–3 mm, 3–5 mm. The number, size, color and type of all microplastic granules on each filter paper were recorded.

2.4. Microplastic identification

Microplastics cannot be precisely discerned just by visual method. Raman spectroscopy can accurately identify the chemical ingredient of plastic particles (Di and Wang, 2018). In this research, 95 particles selected in the microscope observation were detected by micro-Raman spectroscopy (Thermo Fisher Scientific DXR2, USA). The spectrums acquired by analysis were contrasted with the spectral library to identify its composition. Selected typical microplastic particles for electron microscopic observation and photographing (SEM; Hitachi S-4800, Japan).

Table 1

Species, maximum (max) sizes, body weight and abundances of microplastics in fishes from Nanxun Reef.

Species	Max length (cm)	Body weight (g)	Microplastics (items/individual)	Microplastics (items/g)
<i>Cephalopholis urodeta</i>	11.0–11.8	20.26–25.16	1–8	0.04–0.39
<i>Melichthys vidua</i>	11.5–16.2	44.16–158.01	0–14	0–0.10
<i>Balistapus undulatus</i>	10.5–11.0	30.60–61.76	0–10	0–0.16
<i>Balistes capistratus</i>	8.0–15.0	103.40–110.49	1–6	0.01–0.06
<i>Odonus niger</i>	11.8–13.9	49.52–88.22	1–2	0.01–0.04
<i>Xanthichthys caeruleolineatus</i>	17.0–21.0	120.67–254.26	0	0
<i>Pseudobalistes fuscus</i>	17.2	22.50	2	0.09
<i>Acanthurus pyroferus</i>	9.8–11.5	38.59–73.48	0–4	0–0.10
<i>Acanthurus japonicus</i>	7.0–10.0	22.50–49.06	1–6	0.04–0.12
<i>Naso lituratus</i>	16.8–20	147.72–196.35	0	0
<i>Naso brevirostris</i>	20.0	212.48	6	0.03
<i>Acanthurus lineatus</i>	16.0	175.49	1	0.01
<i>Pomacanthus imperator</i>	7.1	224.64	0	0
<i>Dasyatis kuhlii</i>	60.0	2300	6	0
<i>Monocentrus japonica</i>	13.0	54.96	1	0.02
<i>Zebrasoma veliferum</i>	13.0	122.41	0	0

2.5. Quality assurance and quality control

The glass bottles used in this study were rinsed with distilled water in advance. The scalpels and tweezers for dissecting were disinfected.

Sieves were rinsed with distilled water before use. To protect the laboratory environment from plastic contamination, all bottles and dishes were covered with glass cap. The experimenters should wear lab coats during the experiment.

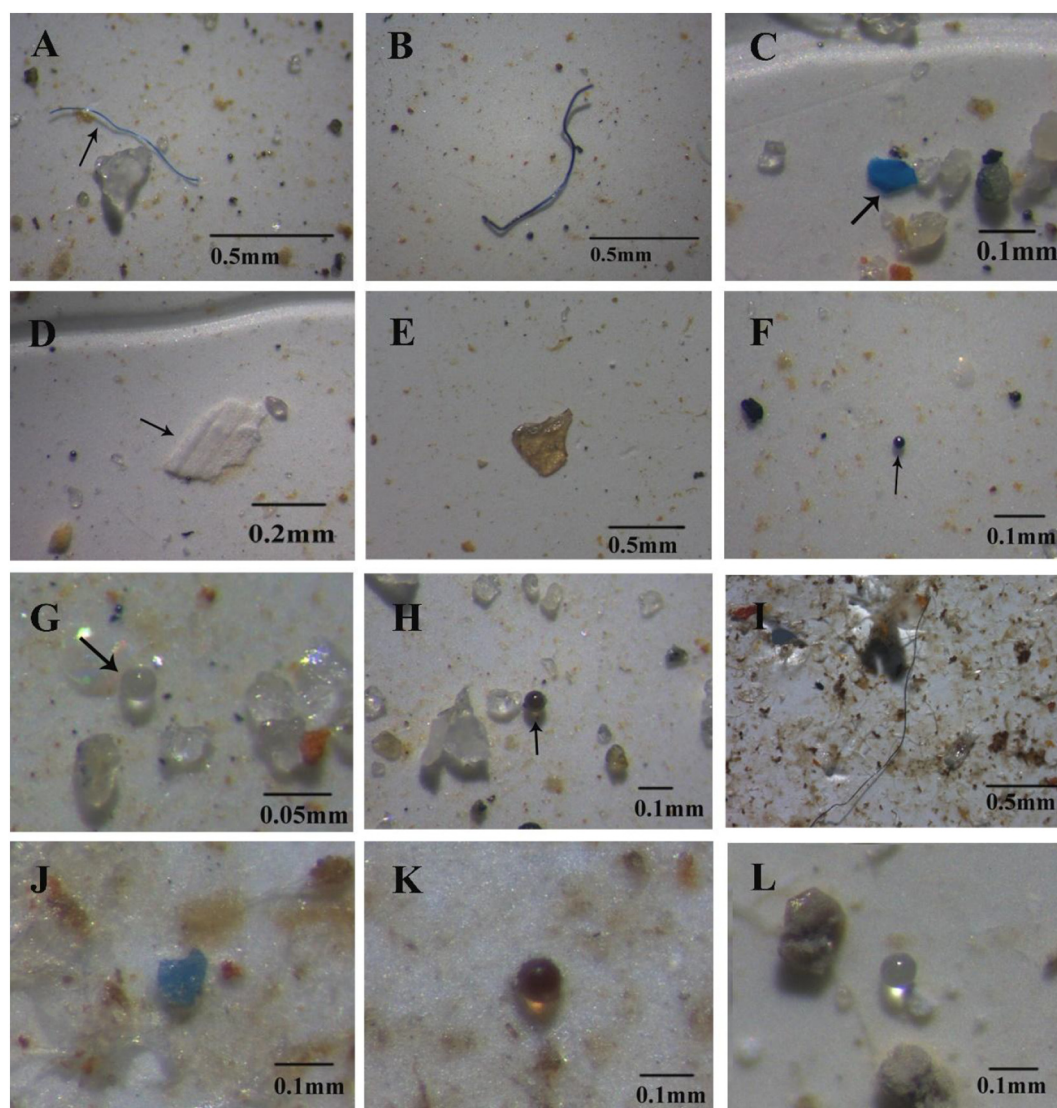


Fig. 1. Types of microplastics in the surface water (A–H) and fish samples (I–L) from Nanxun reef: (A, B, I) fibers, (C, D) films, (E, J) fragments, (F, G, H, K, L) pellets.

2.6. Date analysis

Data analysis was performed by SPSS ver. 20.0 (SPSS, Inc., Chicago, IL, USA). The differences in ingestion microplastics between the two groups (herbivorous and carnivorous species) were analyzed using Student's *t*-test. *P* < 0.05 was considered to indicate statistical significance.

3. Results and discussion

3.1. Microplastics pollution in surface water

3.1.1. Abundance of the microplastics

Microplastic particles were found in all surface water samples assembled from 15 locations in the Nanxun Reef, ranging from 1250 items/m³ to 3200 items/m³ in the investigated areas, with an average of 1773 items/m³ (Fig. 2). Among them, the highest abundance of microplastics was detected in S15 (3200 items/m³), while the lowest abundance was found in S1 and S8 (1250 items/m³). Microplastics abundance was affected by many elements, such as anthropogenic (Beer et al., 2018), surrounding environment and weather (Kim et al., 2015). The development of the surrounding countries may pose a potential risk to the microplastics pollution in the SCS. The fishery activities could be a nonnegligible source of microplastics. The anthropogenic activities in Nansha Islands could also increase the microplastics pollution. Nansha Islands are affected by the southwest wind in summer and the northeast wind in winter (Shen et al., 2010). The currents impact the contaminant distribution in marine, such as microplastics and POPs. Because of the physiographic incidence of the islands, the southwest wind displays a clockwise direction of revolution above the Nansha Islands (Shen et al., 2010). Therefore, the uneven distribution of microplastics in Nanxun reef may be affected by currents and monsoons.

In previous studies, microplastics in surface water were collected by trawl, samplers or sieves. As different sampling methods might lead to different results, we compared our results with the microplastic

densities reported by the same sampling method. The abundance of microplastics in Nanxun reef is higher than that in North Yellow Sea (545 ± 282 items/m³) (Zhu et al., 2018), lower than that in the South China Sea (2569 ± 1770 particles/m³) (Cai et al., 2018), Maowei Sea (4.5 ± 0.1 particles/L) and Yangtze Estuary (4137.3 ± 2461.5 n/m³). Compared with Nansha islands, estuaries and coastal areas are more closed to residential and industrial areas, higher microplastics in which might be caused by frequent human activities in these areas. Mariculture zones are also significant contributors to microplastics pollution, leading to higher abundance of microplastics in the coast. In addition, waves and winds in sea areas, and different sampling time could affect the distribution of microplastics as well (Zhu et al., 2018).

3.1.2. Types, colors, and sizes of microplastics in surface water

In Nanxun Reef, the most abundant microplastics in surface water were pellets, followed by fragments, fibers and films (Fig. 3). The main color of pellets was blue (Fig. 4). The blue spherical pellets were usually termed microbeads. Microplastics with a size of <0.5 mm were major in surface water samples (Fig. 5). Typical microplastic granule were scanned by SEM (Fig. S1). Probably due to different materials, the surface morphology of the pellets with the same shape and color are different when scanned by SEM. It has been reported that microbeads often used for exfoliating scrubs by young people (McDevitt et al., 2017). Pui Kwan Cheung (Cheung and Fok, 2017) reported that 10,000 to 100,000 beads could be released using a facial scrub. The microbeads in the care products showed a colorless granular or blue standard sphere. In this study, blue microbeads accounted for around 76.5% of the microplastics in seawater. It has been reported that 11 blue spherical microbeads were found on the nets of fishing boats down the sea-coast of Hong Kong. The microbeads could pass through the sewage filter easily due to their little size (Cheung and Fok, 2016). Our results suggested that human domestic sewage could be a main provenience of microplastics in the SCS. Besides, fibers were widely detected in our study as well. A previous study indicated that plastic fibers in the ocean environment might come from human textile washing (Browne et al., 2011). Fibers are extensively used in fisheries activities such as

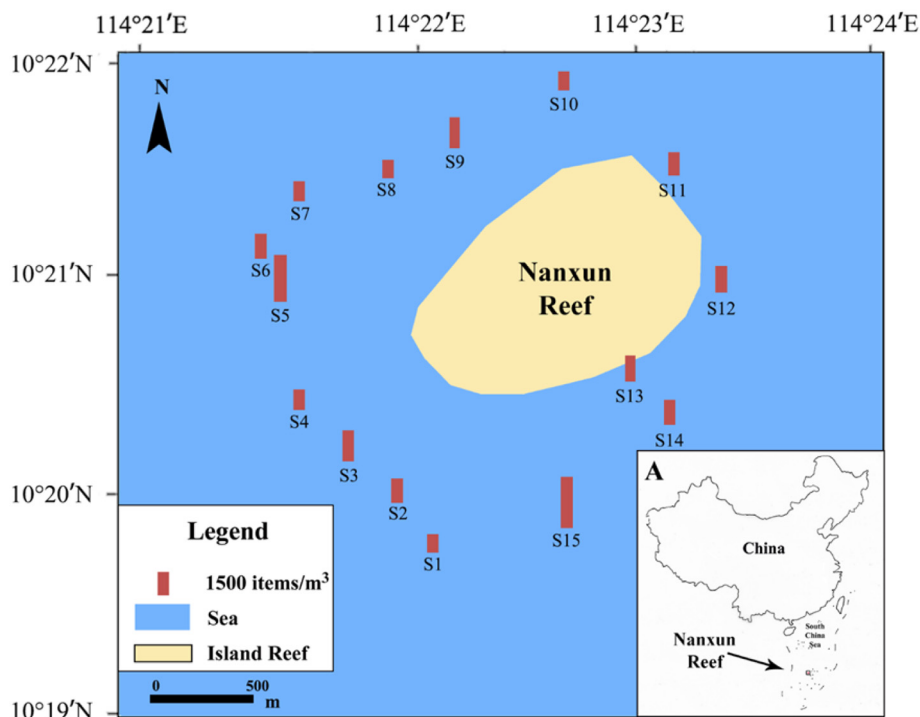


Fig. 2. The concentration of microplastics in the Nanxun reef. Red pillar shows the concentration of microplastics in the surface water. Insert A reveals the locations of sampling situations in South China Sea. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

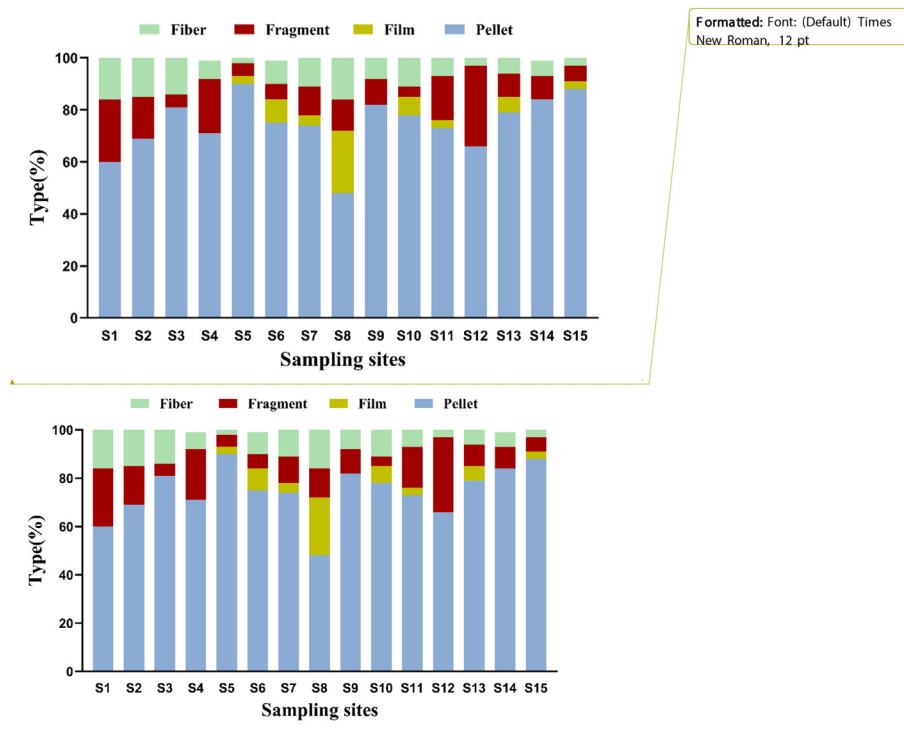


Fig. 3. Proportion of microplastic type in the water samples from Nanxun reef.

the manufacture of fishing ropes and fishing nets. Thus, our results suggested that fisheries activities might also be the source of microplastics pollution in South China Sea.

3.1.3. Polymer type

In this study, an amount of 95 granules were randomly selected for Raman spectroscopy. As indicated in Table S2, polyvinylchloride (PVC) were the most common polymers in the analyzed particles, which were commonly used in the building industry, accounting for 37.9% and followed by polyamide (PA, 21%) and polyethylene (PE, 21%). Besides, there were a few polypropylene (PP, 10.9%) detected here. PVC is the most frequently detected polymer forms in microplastics from Changjiang Estuary (Xu et al., 2018). Possible sources of PA include clothing, packaging and fishing (Yan et al., 2019). PE is widely applied in food packaging bags and agricultural film (Zhu et al., 2018). PP is generally used in fishing tools. The results suggested that anthropogenic wastes might be a significant source of microplastics in the SCS.

3.2. Microplastics pollution in fishes

3.2.1. Abundances of microplastics in fishes

A total of 35 marine fishes were observed here, and 21 of them were polluted by microplastics. The density of microplastics in the polluted individuals ranged from 1.0 to 14.0 (average: 3.1) particles per fish, which was much higher than that in previous reports. A level ranging from 0.5 to 1.4 particles per fish was observed in six seawater fish species sampled from the Gulf Coast of Texas in the United States (Peters et al., 2017). In 178 fish collected along the Saudi Arabian coast of the Red Sea, only a total of 26 microplastic granules were detected in all the samples (Baalkhuyur et al., 2018). However, the density of microplastics in fish here was quite similar with that from the Maowei Sea (7.4 ± 0.4 particles per fish) (Zhu et al., 2019) and that from Poyang Lake in China (0–18 items per individual) (Yuan et al., 2019).

Among 16 detected species, the concentration of microplastics was highest in individuals from *Melichthys vidua*, with 9 pellets and 5 fibers. Of all observed microplastics in fish samples, the major type was fiber

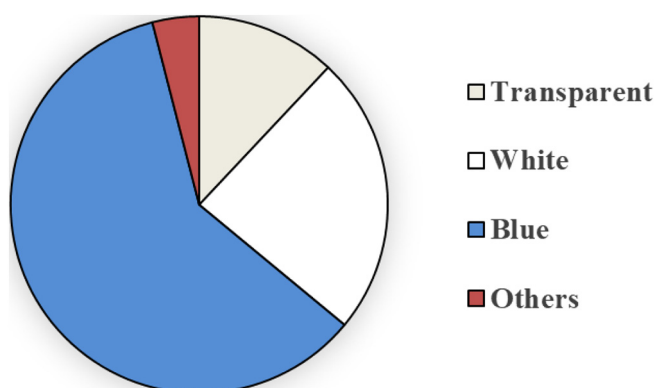


Fig. 4. Proportion of microplastic color in the water samples from Nanxun reef.

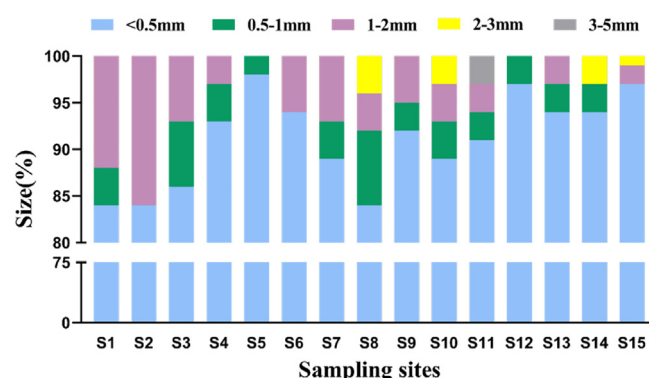


Fig. 5. Proportion of microplastic size in the water samples from Nanxun reef.

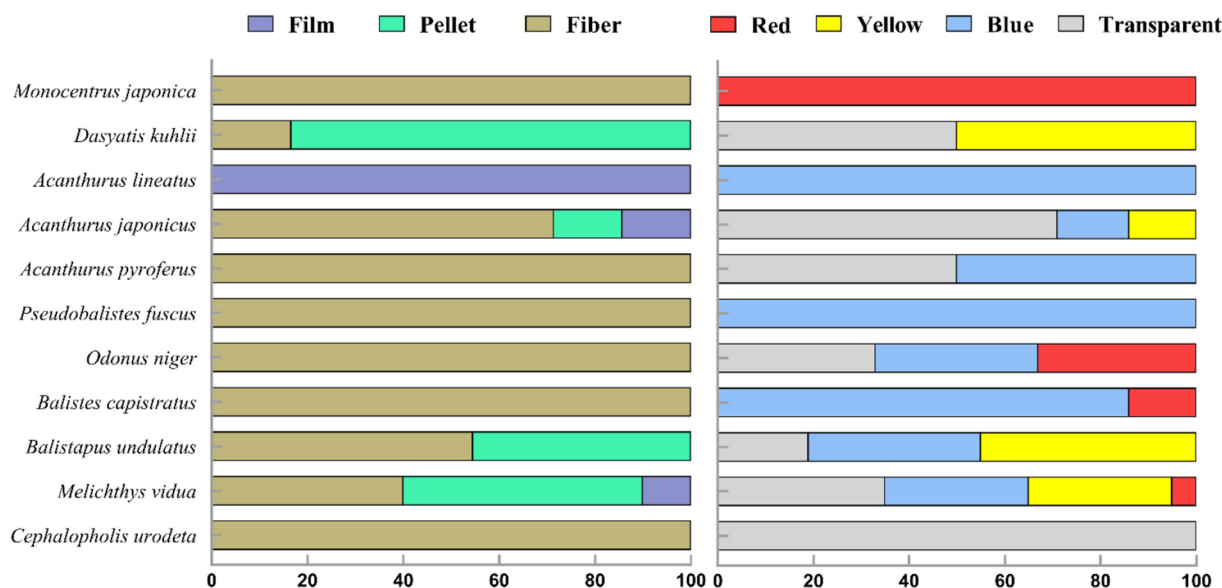


Fig. 6. Proportion of ingestion microplastics type and color in the gastrointestinal tract and gills tract of fishes from Nanxun reef.

and the most common color was transparent. The length of fibers ranged from 3 mm to 5 mm, with a diameter <0.5 mm. Likewise, the size of most pellets detected here was <0.5 mm.

It has been reported that tiny size microplastics are more easily ingested and accumulated into organs (Su et al., 2019). In this research, higher concentration of microplastics was observed in the carnivorous species including *Balistes capistratus*, *Melichthys vidua*, *Cephalopholis urodeta* and *Odonus niger*. Herbivorous fish which feed on algae exhibited lowest microplastics ingestion, such as *Zebrafish*, *Acanthurus pyroferus* and *Acanthurus lineatus*. However, there was no significant difference in the microplastic ingestion between herbivorous species and carnivorous species ($P > 0.05$). Microplastic ingestion is reported to be correlated with diverse feeding habits and microplastics concentrations in the seabed (Battaglia et al., 2016; Romeo et al., 2015). The sites of collected fish samples are close to each other in this survey, thus the microplastic concentrations in these samples were mainly affected by the eating habits. Fish could mistake microplastics for plankton and eat them, or they may ingest prey attached to microplastics, or ingest microplastics during feed activities unconsciously. A narrow food source for those herbivorous species may decrease the risk for microplastic ingestion (Mizraji et al., 2017).

3.2.2. Types, and colors of microplastics in fishes

As shown in Fig. 6, the primary type of microplastics among the investigated fishes was fiber. The fiber with slender shape is similar to zooplankton and leads to easy ingestion by mistake. Fibers may easily enrich in organs. Large numbers of fibers were observed in the gastrointestinal tract and gills tract of fish from Maowei Sea (Zhu et al., 2019). Fiber was also the major type of microplastics in the Khalida Jabeen's study (Jabeen et al., 2017). Blue and transparent were the main color of microplastics observed here, which was similar to that reported in a previous study (Zhu et al., 2019). Blue particles might be more attractive than other colors to marine organisms and more easily to be ingested by mistake. Besides, the transparent particles are quite similar to little jelly fish or zooplankton, which might result in being eaten commonly by fish.

4. Conclusion

This research revealed the microplastic pollution in surface water and fishes collected from Nanxun Reef. Microplastics were found in all surface water samples and most fish samples. The major type of

microplastics was microbeads in surface water. Blue is the major color of microplastics. A total of 35 fishes was detected, representing 16 species. Microplastics were observed in 21 individuals. Ingested microplastic consisted primarily of fibers, mostly blue or transparent. The size of microplastics which observed in this study were almost <0.5 mm. Our results suggested that human domestic sewage might be a main provenience of microplastics in the Nanxun Reef. This research could offer useful data for monitoring microplastic pollution in the South China Sea.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2019.134022>.

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