

Advances in Research of Collection and Detection Methods of Microplastics in Environment

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Abstract As a new type of environmental pollutants, microplastics have gradually attracted people's attention. A large number of plastics discharged into the environment by human beings are constantly aging and breaking, and finally become microplastics. Microplastics can adsorb pollutants in the environment, and their components have certain toxicity, which can cause different degrees of harm to organisms. Due to the structural characteristics of microplastic particles, such as small particle size, large specific surface area, and their distribution in different environmental media, it is very difficult to accurately detect microplastics. Reliable collection and detection methods are the key to the study of environmental behavior of microplastics. In this study, the collection and detection methods of microplastics in the environment were reviewed, and the development direction of microplastics detection technology in the future was prospected. This study has a certain reference value for the related research and the prevention and treatment of micro-plastic pollution.

Key words Microplastics, Collection method, Detection method

1 Introduction

Microplastics are a kind of tiny plastic particles with particle size less than 5 mm, composed of acrylonitrile-butadiene-styrene (ABS), polyethylene terephthalate (PET), polystyrene (PS), polycarbonate (PC), polypropylene (PP), polyethylene (PE) and polyvinyl chloride (PVC)^[1–2]. Microplastics are generally divided into primary and secondary microplastics according to their sources^[3]. Primary microplastics refer to microplastics with micron-sized particles prepared at the beginning of the industrial production process, such as plastic particles added to daily facial and body cleansing products, personal care products such as exfoliating and body scrubs, or cosmetics^[4]. Secondary microplastics refer to the microplastics formed by the disintegration or degradation of large plastics in the environment through physical, chemical and biological processes, usually in the form of fragments or fibers^[5–6].

Since plastic products have the characteristics of low cost, good plasticity and strong durability, the number of plastic products used by human beings is also increasing year by year. According to statistics, the world produces more than 3 million t of plastics every year, and it is expected to reach 33 billion t by 2050, which also leads to an increase in the production of microplastics^[7–8]. Microplastics have the characteristics of strong chemical stability and difficult degradation^[9], leading to the con-

tinuous accumulation of microplastics in the ocean, soil and other ecological environments, and the abundance increases year by year, affecting the normal operation of the entire ecosystem. Microplastics also have large specific surface area and adsorption characteristics, and can adsorb heavy metals and other pollutants in the environment on their surface and become carriers of various pollutants, causing secondary pollution to the environment. In addition, after being eaten by marine organisms and terrestrial animals, microplastics will be transmitted along the biological chain, which will eventually harm human health and pose a threat to the ecosystem^[10].

As a new pollutant, microplastics have aroused widespread concern and research^[11]. With the deepening of people's understanding of microplastics, the detection technology of microplastics is also developing. In this study, we reviewed the existing detection methods of microplastics to provide theoretical support for the prevention and control of microplastics pollution.

2 Collection methods of microplastics

For the detection of microplastics, it is usually necessary to obtain the corresponding morphological size, environmental abundance and composition information at low concentrations, and have a good particle size detection limit, but there is still a lack of quantitative standardized sampling and analysis methods for the collection and detection of microplastics.

2.1 Collection in water environment Microplastics are widely distributed in oceans, rivers, lakes, reservoirs, ponds and other water environments, and their abundance is low. When studying the distribution of microplastics in a water body, trawls with different pore sizes are usually used to collect or enrich microplastics in the water body^[12]. The trawl covers a large sampling area, which can collect a large volume of microplastics in water and reduce the sampling volume. However, the mesh size of the trawl af-

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fects the size of the collected microplastics. The trawl with small-sized mesh can collect small-sized microplastics that have a greater impact on the water environment, but the mesh is easily blocked by suspended sediment, algae or plankton^[13]. Therefore, when using the trawl to collect microplastics in the water environment, the trawl mesh of appropriate size should be carefully selected. For the same water environment, the abundance of microplastics observed by trawls with different mesh sizes is quite different^[14].

2.2 Collection in soil The distribution of microplastics in the soil is modular and uneven, and some farmlands where plastic films have been used and land plots where plastic waste has been landfilled usually have higher content of microplastics than other areas^[15–16]. Therefore, in order to obtain reliable data, the selected sampling method should reflect the representativeness of the sample. According to the actual situation, the sampling scheme of soil medium generally includes simple random sampling, quadrat sampling, cross-sectional sampling and stratified random sampling^[17]. The purpose of simple random sampling is to explore the distribution of microplastics in a small plot, and the purpose of quadrat sampling is to explore the distribution of microplastics in a large plot. Cross-sectional sampling is to determine the degree of contamination along linear features (such as roads, drains, *etc.*), define the concentration gradient, and determine the degree of regional contamination. Stratified random sampling is mainly used to determine the degree of contamination of sub-areas in the whole sampling area^[14]. The sampling depth of the soil is usually 10 cm below the soil surface, and the sampling tools selected are different according to different research purposes, usually including stainless steel spoon, stainless steel shovel, box sampler, cutting ring, soil drill, *etc.*^[18].

2.3 Collection from ambient air Due to the small volume and relatively low density of microplastics, they are easily transported by the wind and can remain in the atmosphere for a long time, so it is also necessary to explore the distribution and concentration of microplastics in the ambient air^[19]. At present, the collection methods of microplastics in the atmosphere reported at home and abroad are mainly passive and active sampling technologies.

Passive sampling technology uses gravity or weather conditions to continuously collect microplastics deposited in the atmosphere. The passive sampling device is mainly composed of three parts, namely the collection column, the connecting pipe and the terminal collection bottle. During operation, the collection column made of stainless steel or glass on the top collects microplastics, dust, precipitation and other settling substances in the atmosphere, and the settling substances reach the terminal collection bottle through the connecting pipe^[20]. Passive sampling results are greatly affected by weather conditions such as wind direction and wind speed, so it is necessary to accurately record the weather conditions at the time of sampling to analyze the correlation between the detection results of air microplastics and weather conditions^[21–22]. Since there is no unified technical standard for pas-

sive sampling of microplastics in ambient air, and it is difficult to obtain microplastic particles suspended in the air by passive sampling, the detection results of microplastics in ambient air may deviate from the actual concentration under different sampling conditions^[14].

Active sampling technology is a technology to collect microplastics in the atmosphere with a certain volume within a certain period of time through vacuum negative pressure and other methods. The active sampling device consists of a pump, replaceable tubing, and a collection container with a filter^[23]. The pore size of the collection container filter will have an impact on the test results of microplastics. A small pore size is easy to be blocked, and a large pore size will not collect small-sized microplastics^[24]. Commonly used filters include quartz fiber GF/A Whatman filter, glass microfiber Whatman GF/A filter, and Teflon ECHO PM environmental filter^[25–26].

2.4 Collection from organism Because of the small size, microplastics are easily eaten by organisms, especially aquatic organisms, and accumulate in their bodies, eventually causing damage to their digestive tract and intestines and stomach. At present, there are two main methods for collecting microplastics in organisms: plankton sampling and macrobiology sampling. The plankton sampling method is to use vertical trawl to intercept plankton, and then carry out biological extraction, microplastic separation and other operations; the macrobiology sampling method is to directly dissect the organism, and analyze the microplastic content of the obtained tissue sections. This method is applicable to shellfish, fish, waterfowl and other macrobiology^[27].

3 Detection methods of microplastics

Although the elements that constitute plastics usually include only a few elements such as carbon, hydrogen, oxygen and nitrogen, plastics have large molecular weights and complex structures, and usually contain different kinds of plastic additives. At present, the detection methods used in the research of microplastics mainly include microscopic counting, Raman spectroscopy, Fourier transform infrared spectroscopy, scanning electron microscopy, thermal analysis technology and so on.

3.1 Microscopic counting method The microscopic counting method uses optical microscope and stereomicroscope to observe and identify the color, shape and other physical characteristics of microplastics, and classify and count them. This method has the advantages of simple operation, short detection time and low detection cost. Song Young Kyoung used stereomicroscope to identify the filtered microplastic samples, and divided them into four categories according to their shapes and measures their size ranges^[28]. Although visual observation can identify microplastics with particle size as low as several hundred μm , the results are usually affected by the subjective choice of the operator, the quality of the microscope and the shape, color and size of the microplastics. When the size of the microplastics is less than 100 μm , it is difficult to identify and identify them by visual observation, and the

error rate is high, with a misjudgment rate of more than 20%^[29]. DEKIFF *et al.*^[30] found that the error rate of colorless and transparent microplastics was more than 70% compared with the results of optical microscopy and micro-Raman spectroscopy. The smaller the particle size, the higher the error rate. Therefore, the microscopic counting method is more used in the preliminary identification of microplastics.

3.2 Raman spectroscopy Raman spectroscopy is a commonly used method to identify the chemical composition of microplastics, and can identify microplastics with a particle size of 1–20 μm ^[31–32]. The method has the advantages of non-destructiveness, low sample size, high throughput, high reliability and *etc.* Wang Sai *et al.*^[33] used the confocal micro-Raman imaging system to detect the content of microplastics in water by quantitative analysis of three-point sampling method using the pretreatment method of primary filtration, digestion, elution and secondary filtration. The method is easy to operate, high in recovery rate and suitable in time and cost, but the accuracy of the method needs to be improved. Surface-enhanced Raman spectroscopy, as a new analytical technology with great potential, has the characteristics of high sensitivity, small water interference, short detection time, no need to separate samples in advance and can provide the fingerprint spectrum of the analyte, and has been used for the identification and detection of microplastics and nanoplastics in the environment^[34]. Because the conventional surface-enhanced Raman spectroscopy (SERS) substrate is easy to produce fluorescence interference under laser excitation, thus reducing the detection sensitivity, some researchers have integrated plasma nanomaterials into the preparation of surface-enhanced Raman spectroscopy substrate, effectively improving the detection sensitivity of microplastics^[35]. Yin *et al.*^[36] used a sponge-supported gold nanoparticle layer as the SERS substrate to reduce the fluorescence interference generated by the conventional substrate under laser excitation, improve the sensitivity of the detection method, obtain the relationship between the peak intensity of SERS and the concentration of microplastics, and realize the quantitative detection of microplastics in seawater^[37]. Lê *et al.*^[38] successfully detected 0.4 μm microplastic particles from water by inserting silver-gold nanostars into anodized aluminum nanopores as a substrate for surface-enhanced Raman spectroscopy. SERS method makes up for the shortcomings of traditional microplastics detection methods, such as complicated sample pretreatment procedures and low sensitivity, and has a huge development space in the detection of microplastics.

3.3 Fourier transform infrared spectroscopy Fourier transform infrared spectroscopy can provide a unique infrared spectrum for a particular chemical bond. Different materials have different bonding components, which makes it possible to identify unknown substances by comparing the spectra of known materials^[39]. Because of its high reliability, Fourier transform infrared spectroscopy is also the most commonly used technique for chemical characterization of microplastics, and it can identify microplastics with particle sizes ranging from 10 to 20 μm . Fourier transform infrared

spectroscopy has been extended and optimized, and now there are micro-infrared spectroscopy methods, attenuated total reflection infrared spectroscopy and focal plane array infrared spectroscopy, which improves the identification range of microplastic particle size. Tang Qingfeng *et al.*^[40] used microscope-Fourier transform infrared spectroscopy ATR technology to detect microplastics in landscape water samples and soil in Beijing. The method successfully realizes the analysis of various characteristics of microplastics in environmental water samples, such as material, shape, size, color and the like, and has the advantages of simplicity, accuracy and reliability. Huang Kaisheng *et al.*^[41] analyzed the existence of microplastics in 43 batches of typical daily chemical products sold in the market by Fourier transform infrared spectroscopy, and realized the morphology and composition analysis of plastic beads in daily chemical products.

3.4 Scanning electron microscopy (SEM) Scanning electron microscopy can observe the characteristic surface texture of microplastics, such as cracks and pits, and analyze the weathering process of plastics with high resolution. Through combining of scanning electron microscopy and energy dispersive X-ray spectroscopy, the elemental composition of microplastics and the relevant information of inorganic additives they contain can be obtained, thus further distinguishing between natural materials and microplastics. Scanning electron microscopy and energy dispersive spectroscopy (SEM-EDS) can characterize the surface morphology of microplastics and determine the elemental composition of surface polymers through imaging and elemental analysis, which can quickly and effectively screen a large number of microplastic particles and reduce the possibility of identification errors, with much higher accuracy than visual and microscopic observation. The main disadvantage of this method is the relatively high cost of detection and the need for pretreatment before detection, which is not conducive to the treatment of a large number of samples. Xu Zhouying *et al.*^[42] successfully observed the local surface microstructure of microplastic particles by means of scanning electron microscopy. Yu Qingxin *et al.*^[43] observed four types of microplastics in soil by scanning electron microscopy, including fibrous, fragmentary, film-like and bead-like, and further observed tiny features such as wear marks and cracks on the surface of microplastics. Because the scanning electron microscope can not distinguish the color of the sample, it can only get the morphological characteristics of the tested sample and can not know the element composition, so it can be combined with the energy spectrometer to detect the microplastics, and can get the morphology and element composition of the microplastics. Dehghani *et al.*^[44] used the SEM-EDS method to distinguish microplastics of different sizes and shapes from traces of Al, Na, Ca, Mg and Si particles, and detected the presence of plastic additives and other adsorbed fragments on the surface of microplastics. However, the SEM-EDS method also has some limitations, such as the laborious and expensive sample preparation steps, the time-consuming full inspection of all samples, which limits the number of microplastics that

can be analyzed in a certain period of time, resulting in low efficiency. In addition, the color of the sample cannot be used as a marker in SEM-EDS analysis, so the technique is mostly used to analyze specific microplastics.

3.5 Thermal analysis technique Thermal analysis refers to the decomposition of polymers into small molecules at high temperatures, and then the identification of the components and types of microplastics by means of characteristic spectra, with the sample mass limited to 0.1 to 0.5 mg. This method is suitable for the preliminary screening of microplastics, with high selectivity, and can quickly determine the type of microplastics^[45–46]. Under the condition of high temperature, this method is destructive to a certain extent, resulting in the size, shape and color of the sample can not be distinguished, and the sample needs to be put into the pyrolysis tube, which limits the size of the microplastic. Because this method has some defects in the detection of microplastics, it is usually used to analyze microplastics by combining thermal analysis with other analytical methods. At present, the thermal analysis techniques used in the detection of microplastics are usually pyrolysis gas chromatography-mass spectrometry and thermal extraction-pyrolysis gas chromatography-mass spectrometry, in which pyrolysis gas chromatography-mass spectrometry (Py-GC-MS) requires the particle size of the detected object to be greater than 100 μm . This technology is mainly used to rapidly crack the sample through a high-temperature cracking furnace, then separate the sample through a chromatographic column, and identify the polymer type and quality of the 0.1–0.5 mg sample through mass spectrometry, with high sensitivity^[47], but the standard chromatogram needs to be determined with a standard substance before use. This technique is destructive, expensive, complex and time-consuming, and therefore it is not suitable for large sample size analysis^[48]. Fischer *et al.*^[49] performed reliable qualitative and semi-quantitative analysis on 8 kinds of microplastics (PE, PP, PS, PET, PVC, PMMA, PA6, PC) at the same time of using Curie point pyrolysis-gas chromatography-mass spectrometry (CP-Py-GC-MS). DÜMICHEN *et al.*^[50–51] developed a thermal extraction-pyrolysis gas chromatography-mass spectrometry (TED-GC-MS) analysis method for microplastics smaller than 100 μm , and can qualitatively and semi-quantitatively analyze samples containing PE, PP, PS, PET and PA within 100 mg within 2–3 h. The advantage of this method is that it has high analysis efficiency and is superior to pyrolysis gas chromatography-mass spectrometry in performance, while the disadvantage is that it is difficult to distinguish compounds with similar sample mass and degradation temperature.

3 Conclusions and prospects

As plastic products have been widely distributed in all aspects of human society, micro-plastic pollution has spread all over the ocean, fresh water, soil and atmosphere. Researchers have done extensive research on the types, distribution, migration, detection and hazards of microplastics. As an important means of re-

search and evaluation, the existing detection methods of microplastics have their own advantages and disadvantages, and have not yet formed a set of mature, unified and reliable detection methods. Therefore, it is recommended that the research on microplastics detection technology should be strengthened and improved from the following aspects:

(i) The development and application of new treatment methods and detection technologies for microplastics. The enrichment and pretreatment of new microplastics have not formed a unified standard method, and the detection technologies involved in this study have their own limitations. Therefore, it is still urgent to develop other simple and fast methods to ensure the effectiveness and reliability of the results. It is recommended to establish a fast, efficient and convenient detection method for microplastics, which is conducive to the detection of large batches of microplastics samples and saves time and cost. (ii) It is recommended to establish a widely applicable standard method for the detection of microplastics, including the requirements of sampling scheme, sample pretreatment, sample detection and analysis of test results, so as to provide reliable and comparative data for scientific research. (iii) It is necessary to collect microplastic graphic results of various chemical components and establish and improve the microplastic characteristic parameter information base, to improve the efficiency of microplastic identification and analysis, shorten the identification cycle, reduce the workload, and reflect the characteristics of microplastics more comprehensively. Besides, we should continue to study the source of microplastics, clarify the rules of migration and transformation of microplastics in different media, and reduce the harm of microplastics pollution to human body.

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KM and its mechanism of action. The findings revealed that KM markedly elevated the levels of pregnenolone and allopregnenolone in the prefrontal cortex, hippocampus, and amygdala, while concurrently reducing the blood concentrations of ACTH and corticosterone in rats exhibiting the natural enemy sound-induced anxiety-like model. The aforementioned results indicate that KM exerts anxiolytic effects by modulating the levels of progesterone and allopregnenolone in the brain and regulating the activity of the hypothalamic-pituitary-adrenal (HPA) axis.

Huang Huihui *et al.*^[13] investigated the anxiolytic properties of KM by utilizing an elevated plus maze (EPM) in rats and mice. Their findings revealed that, following acute administration of mice and continuous administration of rats in EPM, KM markedly elevated the percentage of open arm entries (OE%) and open arm time (OT%) in the EPM. However, KM did not influence the locomotor activity of rats and mice in the EPM. Furthermore, solid-phase extraction in conjunction with high-performance liquid chromatography-mass spectrometry (HPLC-MS) was employed to ascertain the concentration of neurosteroids within the hippocampus of rats, with the objective of investigating the anxiolytic impact of KM and its influence on the neurosteroid level within the hippocampus. The findings revealed that KM elevated the levels of the neurosteroids pregnenolone and allopregnenolone within the hippocampus. The aforementioned results indicate that KM exerts anxiolytic effects by increasing the levels of the neurosteroids pregnenolone and allopregnenolone in the hippocampus. Notably, the effective dose is considerably lower than the median lethal dose (LD_{50}), exhibiting a high degree of safety. Consequently, KM is expected to be developed into a novel type of high-efficiency and low-toxicity anxiolytic drug.

6 Prospects

G. elegans is documented in the *Compendium of Materia Medica* and numerous other pharmacological monographs. KM represents the primary bioactive component of *G. elegans*. KM exhibits a multitude of pharmacological activities, including anticancer, anti-inflammatory, analgesic, and anxiolytic effects. It boasts several advantageous properties, such as low toxicity, high efficiency, a vast range of sources, and can be utilized as a starting raw material for the synthesis of a plethora of products with medicinal value and crucial pharmaceutical intermediates. Nevertheless, the study of the pharmacological effects of KM is still in its

infancy. It is imperative to continue to comprehensively and systematically study the chemical composition and molecular mechanism of the pharmacological effects of KM, and to further explore potential pharmacological effects, in order to provide a theoretical basis for the further development and utilization of KM.

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