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REVIEW ARTICLE

Microplastics as an emerging threat to human health: Challenges and advancements in their detection

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ABSTRACT

Microplastic pollution has emerged as a significant environmental concern, with potential direct and indirect impacts on ecosystems. Microplastics are pervasive, found in water, food, and even the air we breathe. While their influence on human health is still unclear, microplastics are known to possess endocrine-disrupting properties and can accumulate persistent organic pollutants. Accurate measurement and categorization of microplastics are crucial to understanding their prevalence and impact on contamination. Fortunately, there are several methods available, such as visual analysis, fluorescence techniques, vibrational spectroscopy, and electron microscopy, that offer optimal accuracy in detecting and quantifying microplastics. The increasing presence of microplastics in the food chain has prompted global research efforts to assess potential risks to human health. However, despite ongoing advancements, challenges remain in standardizing analytical procedures and developing methods capable of detecting microplastics as small as nanometers. Visual classification-based methods, though limited in detecting smaller microplastics, show promise for improvement through integration with advanced technologies. This study primarily focuses on microplastic sampling strategies, detection methods, and their respective advantages and disadvantages, shedding light on the advancements and challenges in the field.

Keywords: microplastics; microplastics classification; microplastics persistence; sampling methods; detection methods; FTIR; Raman; biosensors

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1. Introduction

Polymer science has undergone revolutions through the years, since the discovery of the synthetic plastic and has undergone various changes related to its composition, shape and size[1]. These formulations are a part of our modern lifestyle. Plastic is a versatile material because of its low density, low electrical and thermal conductivity and corrosion resistance. These properties allow it to act as an oxygen and water barrier, and it reduces its cost and increases its manufacturing in a wide range of formulations and applications. The global market making it an advantageous discovery, but at the same time it is a global pollutant[2]. Microplastics which are one of the formulations of this material also add up to the above-mentioned fates of plastic in today's world[3].

Microplastics (MPs) are generally described as solid particles or polymeric matrices which are insoluble in water with regular or irregular form. Microplastics are generally of a size less than 5 mm (0.2 inch) and could even be visualized by the naked eye (when seen present in the sediments).

Microplastics have the properties of that of plastics with the difference being their size and the areas in which these are present due to their microsize, which allows them to penetrate through any kind of barrier[4]. Microplastics are non-biodegradable making them accumulate and persist after being introduced in the environment. These can be introduced into the environment in two ways, in one way these are just manufactured in the industries and in the second way these are a result of the breakdown of the larger plastic items and these are named as primary microplastics and secondary microplastics respectively.

The key concern with microplastic is its impact on the ecosystem and human health. The microplastics itself contains the hazardous chemicals as added during their production and vector for the transportation of other toxic chemicals in the ecosystem[5].

Micro and nano plastics have three main entry points to the human system, through contaminated food, through inhalation and penetration through the skin from water[5].

The studies have found micro and nano plastics in human food (salt, sugar), beverages (alcohol), vegetables and fruits (via contaminated soil), and marine species (fishes). According to a recent World Health Organization report, the contaminated food web is the primary entry point for micro and nano plastics into the human system[6].

Detection of microplastics and their classifications is crucial to understand their level of contamination and their spreading across the human habitat. Microplastic contamination is spreading throughout the environment, including seawater, sediments, rivers, soil, and even the air we breathe[7]. It will not be enormous to say that we are currently living in a plastic world. The detection methods are not precise and efficient enough to classify the microplastics and even enable us to work with the samples with more thickness and irregular shapes. The sampling strategy is also a crucial step in the analysis of microplastics and played an important role in its detection, classification, and other studies.

The current review focused on the general introduction and classification of microplastics, with a focus on sampling and detection strategies. The study also focused on the limitations and progress of the analytical methods utilized for microplastic detection.

2. Classification of microplastics

The classification of microplastics can be varied based on various terms. These could be categorized based on the shape they possess, their colour or the form in which they are present in varied environments. However, the general categorization is based on the source through which they enter the environment; these are primary microplastics and secondary microplastics. **Figure 1** shows the classification of microplastics according to their shape, source, colours, and composition.

2.1. Classification based on their mode of entrance in the environment

2.1.1. Primary microplastics

Primary microplastics are the manufactured plastics of a specific shape and size, designed for a specific commercial or medicinal purpose[8]. These are the plastic pieces or particles that are already 5.0 mm or less in size before entering the environment. These are used in cosmetics, air blasting technology and facial cleansers[9]. Microfibers are a class of primary microplastics which are usually shed from clothing and other kinds of textiles while washing or any wear, the shed includes polyester, acrylic or nylon-based materials. In cleansers, they are used as scrubbers for exfoliating purposes. The majority of primary microplastics are created as a result of regular plastic product use. These polymers sometimes referred to as “micro-beads” or “micro-exfoliates”, can vary in form, size, and composition depending on the product.

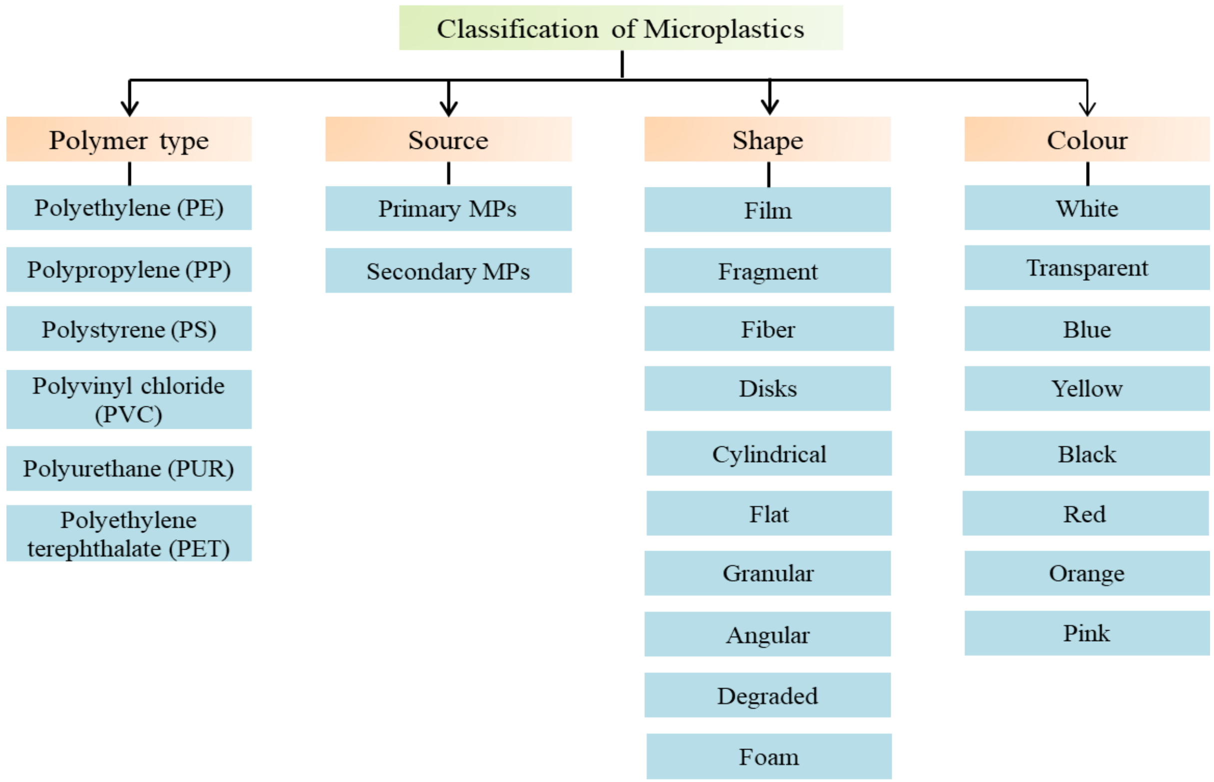

Figure 1. Classification of microplastics based on shape, colour, source, and composition.

2.1.2. Secondary microplastics

These microplastics are a result of the breakdown of the already existing larger plastic items in the environment[8]. The fragmentation is the cause of the microplastic formation, after their disposal in the environment. Plastic being non-biodegradable and insoluble in water just persists in the environment, whether on land or any water body. But over a course of time, it undergoes changes due to other environmental factors which lead to the natural weathering process, breaking down these polymers, acting as a pollutant to break into micro sizes. The plastic debris which leads to this fragmentation process includes items like water or soda bottles, plastic bags, fishing nets, plastic containers and tire wear. These are already manufactured and produced in the macro sizes, but their irresponsible disposal in the environment has led them to become one of the major pollutants imposing risk to the nature. The processes which convert them to the micro size are sun’s radiation, oceanic waves, UV radiation and biological degradation. When exposed to sunlight over a prolonged period, the photo-degradation of the plastic occurs. It occurs on the absorption of photons of the wavelengths generally present in the sunlight like the visible light, infrared radiation and ultraviolet light. The ultraviolet radiation from the sunlight induces the process of oxidation of the polymer matrix, which leads to bond cleavage. This polymer degradation changes the properties of the plastic like tensile strength, colour and shape.

2.2. Classification based on their shape

Information concerning the chemical and physical characteristics of MPs is crucial for determining their original source as well as for evaluating any possible hazardous and environmental impacts they have in a detailed manner. In addition, the form of microplastics is thought to be a crucial signal for determining their origin and rate of disintegration. As a result, it may be used to determine if they have a primary or a secondary origin. These shapes can be film, fragment, fiber[10], disks, cylindrical, flat, granular, foam, angular or degraded. These shapes depend on whether the microplastic sample is in the form of a pellet or a fragment and their concentration varies in sediments and water[11].

2.3. Polymer form

The various types of plastics are manufactured in the industries depending on their demand in the market, as the microplastics are their degraded forms, so they are persistently present in the environment having the same constituent properties. The predominant polymer types are polyethylene (PE), polypropylene (PP) and polystyrene (PS). Polyvinyl chloride (PVC), polyurethane (PUR) and polyethylene terephthalate (PET) are also the major polymer types governing the chemical and physical properties of the microplastics. Among these, polyethylene is the most common due to its higher demand.

2.4. Colour

For varied manufacturing purposes, microplastics are available in different colours, for this, the industry adds various pigments to the polymers. Additionally, environmental weathering causes colour fading or change their original colour to appear yellowish over the course of time. Microplastics are present in various colours from transparent, white, red, orange, blue, brown, tan, and yellow to pink, including other variations in colours.

3. Presence of microplastic in ecosystem

Microplastics can be found in various environments worldwide, including oceans, rivers, lakes, soil, and air. The abundance of microplastics in these environments is a growing concern due to their potential impacts on human health and the environment[12].

The abundance of microplastics worldwide is staggering. It is estimated that there are trillions of microplastic particles in the oceans alone, with more than 14 million tons of plastic entering the oceans each year. Microplastics have been found in marine organisms, including fish and shellfish, which can then enter the human food chain.

The abundance of microplastics in the soil is also another growing concern. Microplastics can be introduced to the soil by using plastic mulch in agriculture or applying sewage sludge. These microplastics can have negative impacts on soil health and can also be taken up by plants, potentially entering the human food chain.

The abundance of microplastics in the air is also a growing concern. Microplastics can be released into the air through the breakdown of larger plastic particles or the wear and tear of synthetic clothing. These microplastics can be inhaled by humans and animals and may have negative health impacts. **Figure 2** illustrates the distribution of microplastic levels across various ecosystems in different regions around the world.

3.1. Marine ecosystem

The abundance of microplastics in marine water habitats has become a significant environmental concern in recent years. Studies have shown that microplastics are present in oceans, seas, and even in some freshwater bodies worldwide. These particles can be ingested by marine organisms, leading to potential harm to their health and ecosystem function.

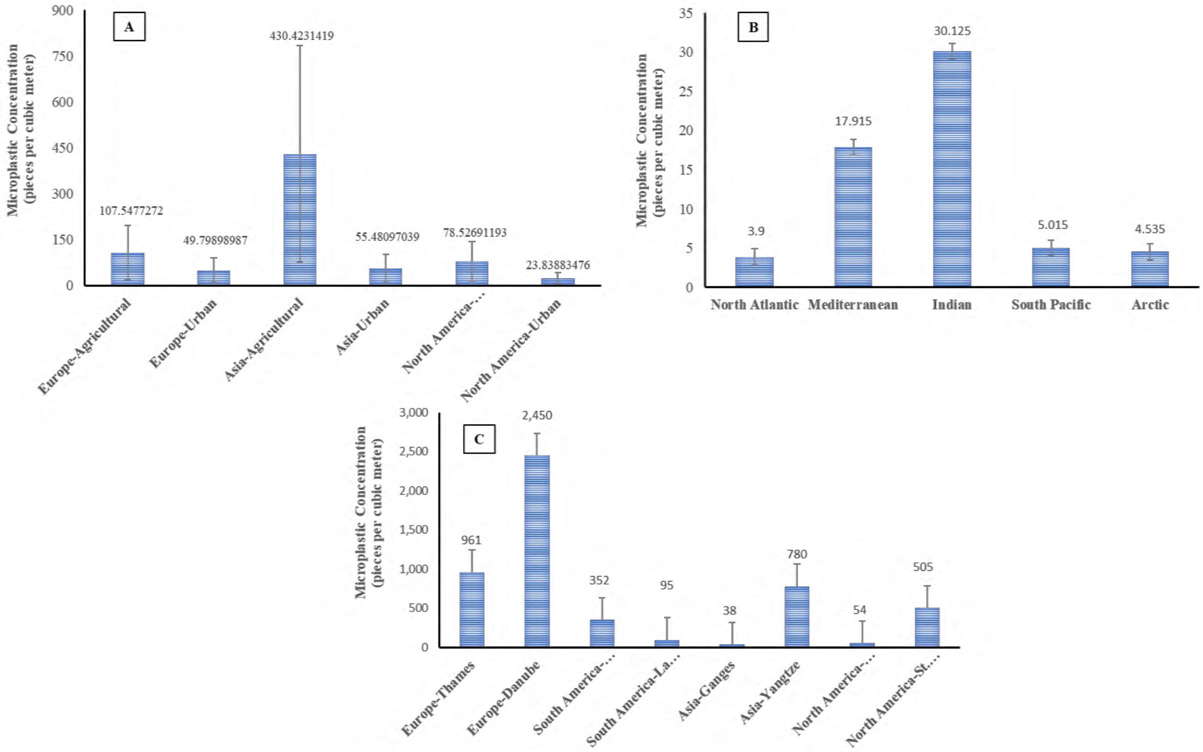

Figure 2. Comparative analysis of microplastic abundance in global ecosystems. Graphs A, B, and C represent the levels of microplastics in terrestrial, marine, and freshwater environments, respectively.

Research has shown that microplastics can accumulate in high concentrations in certain areas of the ocean, such as gyres and coastal regions. They can also be found in the sediment on the ocean floor. In some cases, the abundance of microplastics in these areas has reached alarming levels that pose a risk to marine life. A study conducted in the North Atlantic Ocean found that the concentration of microplastics ranged from 0 to 7.8 items per cubic meter[13]. The Mediterranean Sea varies greatly depending on location. A study in the western Mediterranean found concentrations ranging from 0.22 to 35.61 items per cubic meter[14]. Microplastics have been found in various locations throughout the Indian Ocean. One study in the Bay of Bengal found concentrations ranging from 0.02 to 60.23 items per cubic meter[15]. A study in the South Pacific Gyre found concentrations ranging from 0.02 to 10.01 items per cubic meter. Microplastics have been found in the Arctic Ocean, with concentrations ranging from 0.07 to 9.00 items per cubic meter[16].

Microplastics in marine habitats can cause physical harm, such as blockages in the digestive system, as well as chemical harm due to the toxic substances that can be found on the surface of the particles. Microplastics have also been shown to affect the behavior of marine animals, including their feeding patterns and reproductive success.

It is important to note that microplastic concentrations can vary widely depending on location and the methodology used to sample and analyze them. However, the presence of microplastics in every corner of the world’s oceans highlights the urgent need for global action to reduce plastic pollution and prevent further harm to marine ecosystems.

3.2. Terrestrial ecosystem

Studies have shown that microplastics can be found in soils, sediments, and even in the air we breathe. Sources of microplastics in terrestrial habitats include the breakdown of larger plastic debris, the release of microfibers from clothing and textiles during washing, and the use of plastic mulch in agriculture. Additionally, microplastics can also be transported from other environments such as rivers and oceans via wind and other mechanisms.

Some studies have reported high concentrations of microplastics in urban areas, while others have found them in natural environments such as forests and parks. The size and shape of microplastics can also affect their abundance, as smaller particles are more likely to be transported by wind and water[17]. A study published in 2021 reported that the abundance of microplastics in agricultural soils in Europe ranged from 0 to 126 items per cubic meter[18]. Another study published in the same journal in 2020 found that microplastics were present in urban soils in Europe, with concentrations ranging from 4 to 60 items per cubic meter[19]. In the journal Environmental Science and Pollution Research 2021, it is reported that microplastics were present in agricultural soils in China, with concentrations ranging from 9 to 508 items per cubic meter. Another study published in the same journal in 2020 found that microplastics were present in soils in South Korea, with concentrations ranging from 0 to 65 items per cubic meter[20]. A study published in 2021 reported that microplastics were present in soils in Canada, with concentrations ranging from 0 to 92 items per cubic meter. Another study published in the same journal in 2021 found that microplastics were present in urban soils in the United States, with concentrations ranging from 5 to 30 items per cubic meter[21]. The impacts of microplastics on terrestrial ecosystems are not fully understood, but they may pose a threat to soil organisms and plant growth. Microplastics can also act as a carrier for pollutants and other contaminants, which can accumulate in the food chain.

Efforts are being made to reduce the abundance of microplastics in terrestrial habitats. These include reducing plastic waste, improving waste management practices, and developing alternative materials to replace plastics. Additionally, more investigation is required to better comprehend the consequences of microplastics on terrestrial ecosystems and to create efficient mitigation solutions.

3.3. Freshwater ecosystem

The broad availability of microplastic in freshwater habitats is affected by various factors such as the level of pollution, the type of plastic waste, and the water flow rate. A study conducted in the United States found that urban streams had higher concentrations of microplastics than rural streams. The researchers suggested that this was due to the higher levels of human activity in urban areas, leading to increased plastic waste and pollution[22].

Another study conducted in Canada found that the broad availability of microplastic in freshwater habitats was higher in areas with high population densities and urbanization. The amount of microplastics in freshwater ecosystems was found to be significantly influenced by the type of plastic trash, such as garment fiber and packaging materials[22].

In addition to the composition of plastic waste, the rate of water flow significantly influences the abundance of microplastics in freshwater habitats. Extensive research conducted worldwide has provided valuable insights into the concentrations of microplastics in various rivers and regions. In Europe, studies conducted in the United Kingdom's River Thames and Austria's Danube River reported microplastic concentrations ranging from 517 to 1405 pieces per cubic meter and 700 to 4200 pieces per cubic meter, respectively[22]. Moving to Asia, research on China's Yangtze River found microplastic concentrations ranging from 94 to 1466 pieces per cubic meter[23]. Similarly, in the Ganges River in India, concentrations varied from 5 to 71 items per cubic meter[24]. In North America, investigations carried out in the Great Lakes region of the United States revealed microplastic concentrations ranging from 43 to 65 items per cubic meter[25]. Similarly, a study in Canada's St. Lawrence River reported concentrations between 10 and 1000 items per cubic meter[22]. Shifting to South America, research conducted in Brazil's Amazon River found microplastic concentrations ranging from 4 to 700 items per cubic meter[26]. In the La Plata River in Argentina, concentrations varied from

18 to 172 items per cubic meter[27]. These findings highlight the diverse range of microplastic concentrations observed in freshwater habitats across different continents, emphasizing the global significance of this issue. Overall, the prevalence of microplastic in freshwater habitats is a complex issue influenced by various factors, necessitating a comprehensive approach for effective mitigation. Implementing proper waste management practices and promoting waste reduction strategies are crucial steps in reducing the abundance of microplastics in freshwater habitats. Furthermore, increasing awareness and providing education about the environmental impact of plastic waste is vital for fostering behavioral changes and minimizing the introduction of microplastics into these ecosystems. By adopting a multifaceted approach, we can work towards safeguarding the health of freshwater ecosystems and mitigating the detrimental effects of microplastic contamination.

3.4. Air

Microplastic pollution in the air is a growing concern worldwide. While research on airborne microplastics is still limited compared to marine microplastics, studies have identified several regions with varying levels of abundance. Urban areas, due to human activities and the presence of plastic waste, exhibit higher concentrations of microplastics in the air[28]. Coastal regions, particularly those near densely populated and industrial areas, also experience significant microplastic pollution as these particles are transported by wind and waves[29]. Even remote Arctic regions are not immune, as microplastics have been found in Arctic snow and ice, suggesting long-range atmospheric transport. Agricultural areas with practices involving plastic materials and fertilizers can release microplastics into the air through wind erosion[30]. Furthermore, industrial zones contribute to localized high concentrations of airborne microplastics through processes such as combustion and fragmentation of plastic materials. It is crucial to note that the abundance of microplastics in the air can vary depending on local factors such as population density, pollution sources, wind patterns, and climate conditions. Further research is needed to fully understand the distribution of microplastics in different parts of the world and their potential impacts on human health and ecosystems[31]. Based on the data obtained from the aforementioned studies, an abundance graph representing various ecosystems was constructed (**Figure 2**).

The increasing concentration of microplastic can serve as an additional transmission vector for the coronavirus. Recent research has demonstrated that SARS-CoV-2 can remain viable in aerosol droplets for approximately three hours and on plastic surfaces for up to 72 hours under specific conditions (room temperature of 20 °C and a relative humidity of 40%). Therefore, it is imperative that future efforts prioritize the appropriate management of plastic waste that may be contaminated with SARS-CoV-2. This includes the proper handling and disposal of used medical face masks and gloves. By implementing effective waste management strategies, we can significantly reduce the risk of novel coronavirus transmission through microplastics[32].

4. Sampling of microplastics

The escalating problem of microplastic pollution is causing growing concern, given its pervasive nature and potential for harm. Developing optimal techniques to accelerate the identification process and detect minuscule plastic particles is imperative. Shovels, trowels, spades, scoops, and spatulas are the tools most commonly used in microplastic analyses of sediments. Meanwhile, manta trawls are the primary equipment used for extracting microplastics from surface water samples[33].

The initial phase of the sampling procedure involves the collection of representative samples from the aquatic environment, comprising both the aqueous and solid phases, as a prelude to microplastic investigations (**Figure 3**).

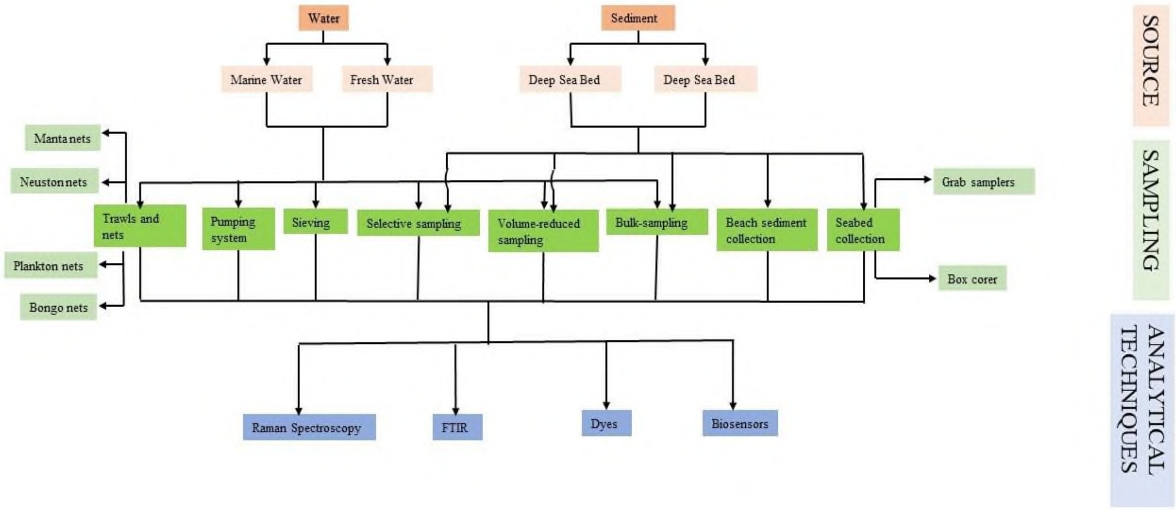

Figure 3. A schematic flow of microplastic sampling and detection methods.

4.1. Sampling of microplastics from water

The most commonly used technique for extracting microplastics from water samples and the supernatant containing plastics from sediment samples is filtration or screening, where the size of the pores in the filters or sieve mesh can vary significantly. The size of the detected microplastics depends on the size of the mesh or pore used[7].

4.1.1. Trawls, nets and mesh

Microplastic particles (MPs) are abundant in seawater, with fibrous MPs (microfibers) being the most prevalent shape observed. However, the number of microfibers present may be underestimated as towed nets with mesh sizes of 300–350 µm are commonly utilized to collect samples for estimating MP concentrations in seawater, allowing microfibers to pass through due to their narrow width[34].

The direct filtration method is better suited for analyzing very small microplastics or samples with limited volume, as it can capture particles as small as possible. The sieve pre-concentration method is a simple and effective approach for analyzing saltwater samples with volumes ranging from hundreds to thousands of litres. Although the trawling method may miss most microplastics smaller than 300 µm, it remains the most suitable technique for collecting a large volume of water over a wide area, making it the current standard for parallel comparison[35].

4.1.2. Pumping system

Pump sampling entails manually or mechanically pushing water through an inline filter. Pumps or bulk samplers are utilized to draw water from different depths and volumes. This non-discrete sample collection method enables the detection of smaller microplastics while minimizing fiber loss. The well-established system offers precise control over the volume of filtered water, allowing for sampling standardization. However, transporting and applying samples can be challenging, and it only permits sampling of a single point, with a risk of contamination[33].

4.2. Sampling of microplastics from sediments

Direct forceps sampling, sieving, and sediment sample collecting are all methods used to gather microplastics from beaches. To collect samples from the seabed, specialized equipment and a vessel are required, such as grab samplers and box corers[7]. Grab samplers provide a rapid and straightforward method of acquiring a seabed sample that includes microplastics.

Another method is the density flotation technique. Density-based techniques are commonly used to separate low-density plastics from high-density minerals in environmental matrices, making it easier to detect polymers[36]. The microplastic removal procedure from soil and sediment samples involves three phases: the sample is mixed thoroughly with the flotation solution, allowed to rest for flotation and settling, and the supernatant is filtered or sieved[37].

4.3. Sampling of microplastic from biota

Due to their small size, microplastics can easily be inhaled or ingested by biological organisms, which are then consumed by humans through the food chain. To collect organisms for analysis, grasps, traps, creels, or bottom crawling can be used for benthic invertebrates, while manta or bongo nets can be used for planktonic and nektonic invertebrates and trawls at various depths can be used for fish. Depending on the study subject, hand collection can be used for bivalves or crustaceans, or electrofishing can be used. Gill nets are commonly employed in freshwater areas[38].

When it comes to the build-up of plastic waste in fishes, the gastrointestinal system is the most commonly researched organ because it may be used to quantify the number of microplastic pieces present in a species' body or to pinpoint the presence, distribution, and kind of waste in particular areas[39].

Depending on the size of the microplastic which needs to be sampled, its sampling method is chosen. For smaller microplastic, mesh nets with smaller pore sizes are also preferred irrespective of the biota in which the microplastic is found.

5. Detection of microplastics

Microplastics are minute synthetic polymer fragments created by human activities. They typically measure less than 5 mm in size but can vary in size, colour, and composition. Analyzing microplastics requires specific methods and techniques, commonly referred to as analytical techniques[40]. Most micro and nano plastics are generated through the degradation of larger plastic items, making their detection process challenging. This has significant implications for human health. Microplastics can be found in various environments, including water (both fresh water and marine), sediments, soil, the atmosphere, biota, fish digestive systems, humans (blood and intestine), and food, among others. To classify microplastics based on their origin or presence in different environments, various types of analytical techniques can be employed. These techniques can be used individually or in combination, as a single method may not be effective for analyzing microplastics in all environmental contexts[41].

To achieve a comprehensive understanding of microplastics, it is essential to systematically collect and review the latest research and advancements in the field. This will allow for the identification of the most effective strategies compared to others[42]. Visual inspection is a widely employed technique by researchers as the initial step in characterizing microplastics. While the human eye can detect larger and more visibly colored microplastics, scientific methods such as spectroscopy, microscopy, and thermal analysis are necessary to identify translucent microplastics measuring 1 mm or smaller[43]. These scientific procedures provide a more accurate and detailed analysis of microplastic characteristics, aiding in their precise identification and classification.

In addition to physical methods, chemical analytical techniques can also be employed to study microplastics, utilizing various dyes and fluorescent methods. However, in many studies and research, light microscopy, Fourier-Transform Infrared Spectroscopy (FTIR), Raman spectroscopy, and chromatography are predominantly utilized based on the size and specific characteristics of the targeted microplastics. For example, light microscopy is effective in detecting larger microplastic fragments but does not provide information about the polymer type. Another microscopy method, Scanning Electron Microscopy (SEM), is frequently employed to differentiate between different types of microplastics. **Figure 4** in the literature depicts various microplastic detection methods, including visual inspection, chemical analysis, spectroscopy, and microscopy, along with their respective limitations and advantages.

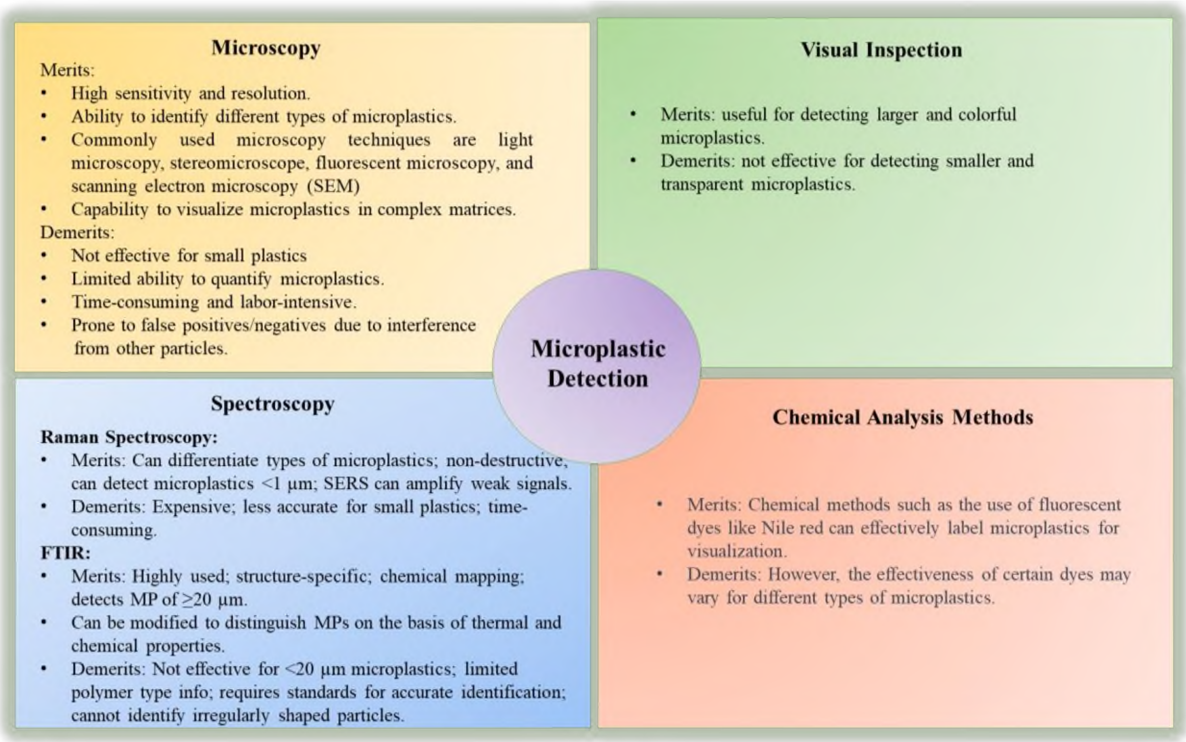

Figure 4. Schematic illustration of various microplastic detection methods, including visual inspection, chemical analytical methods, spectroscopy, and microscopy, highlighting their limitations and advantages based on the literature review.

5.1. Chemical methods

Dyes can be advantageous for visualizing microplastics in specific cases. Fluorescent dye, such as Nile red, is used to label microplastics, allowing for enhanced visibility. Nile red is dissolved in an appropriate organic solvent based on the type of microplastic being detected. It attaches to the polymer of the plastic through Van der Waals forces, occasionally accompanied by dipole interactions for polar particles[44]. However, Nile red and similar dyes have demonstrated limited effectiveness in detecting certain microplastics, like those composed of polyethylene and polyvinyl materials[45]. Efforts have been made to overcome this limitation by introducing various functional groups to Nile red, yielding derivatives with improved binding efficiency. The efficiency of microplastic labeling with Nile red dye is influenced by the polarity of the solvent used[44]. Additionally, other hydrophobic dyes with thermal expansion and contraction capabilities, such as safranine T and fluorescein isophosphate, have been found useful for microplastic identification. Thermal expansion involves applying heat treatment during the staining process to prolong fluorescence intensity, allowing for longer observation periods[46].

5.2. Raman spectroscopy

Raman spectroscopy is one of the emerging techniques utilized for the identification of distinct forms of microplastics based on their spectra, enabling analysis of their vibrational and rotational modes[47]. It is a non-destructive analytical method that utilizes scattered light to assess the vibrational energy of a sample. Raman spectroscopy is capable of recognizing microplastics with sizes up to <1 μm. Raman spectroscopy is typically considered more expensive compared to FTIR (Fourier Transform Infrared Spectroscopy)[48]. However, both techniques can be used as complementary tools in microplastic detection. Each technique has its strengths and limitations, and combining their capabilities can provide more comprehensive and reliable results in confirming the presence of microplastics. Fluorescence poses a common challenge in Raman spectroscopy when detecting microplastics, as it can disrupt Raman signals and hinder accurate identification. To overcome this issue, Surface-Enhanced Raman spectroscopy (SERS) is employed. SERS utilizes a laser light source and nano-scaled roughened metal surfaces to enhance weak Raman spectra signals. This technique resonates the surface charge and amplifies Raman signals, improving the detection of microplastics[49].

However, it is essential to acknowledge that the procedure of microplastic characterization using Raman spectroscopy can be time-consuming and prone to inaccuracies, particularly as the size of the plastic approaches the detection limit. In recent years, algorithmic approaches have been developed to visualize and improve the outcomes of microplastic characterization via Raman spectroscopy. During this process, the sample undergoes Raman mapping, which involves the acquisition of a pixel array forming Raman spectra. Subsequently, these spectra are decoded to generate precise Raman images[50].

Two methods have been identified for decoding these spectra. The first method involves a logic-based algorithm that merges multiple Raman images with different peaks into a single peak, resulting in an increased signal-to-noise ratio of the image. The second method utilizes principal component analysis (PCA) to decode the Raman spectra, even in the absence of standard spectra, by capturing their key features[50].

To enhance the effectiveness of Raman spectroscopy, various substrates have been utilized in combination with SERS. In a study, klarite was employed as a substrate, effectively intensifying Raman signals[51]. Additionally, substrates like gold (Au) and silver (Ag) have been utilized. Among these, Au particles offer advantages over Ag particles as a substrate due to their resistance to oxidation and high efficiency in detecting polystyrene (PS) microplastics[52].

5.3. Fourier-Transform Infrared Spectroscopy (FTIR)

Fourier-Transform Infrared Spectroscopy (FTIR) is a widely employed technique for detecting and characterizing microplastics. In FTIR, the sample is exposed to infrared light of a specific wavelength, and the equipment detects the infrared absorbance based on the microplastic's structure. Changes in the dipole moment of chemical bonds, particularly in polar molecules like the carbonyl functional group, are crucial for efficient FTIR analysis[53].

However, FTIR alone has limitations in identifying irregular-shaped microplastics. To address this challenge, attenuated total reflection FTIR (ATR-FTIR) is used, which provides more information on irregular microplastics compared to transmission FTIR. ATR-FTIR is particularly suitable for thick or opaque samples. Additionally, micro-FTIR is utilized to detect smaller particles by employing membrane filters and generating high-resolution sample maps[54].

Thermal gravimetric analysis-Fourier Transform Infrared Spectroscopy-gas chromatography-mass spectrometry (TGA-FTIR-GC/MS) is widely used to differentiate microplastics based on their thermal properties and obtain information on their thermal degradation[55].

Chemical imaging of unselected areas can also be accomplished using FTIR microscopes equipped with focal plane array (FPA) detectors. Each piece in the FPA functions as an independent infrared sensor, enabling convenient measurement of large fields. However, determining and quantifying microplastic particles with FPA-based FTIR can be time-consuming and involve multiple manual stages[56–58].

Despite their effectiveness, these analytical tools have technological constraints, such as generating large amounts of data that can be challenging to store, as well as being time-consuming, costly, and inefficient. Consequently, researchers are increasingly exploring the use of biosensors for microplastic characterization due to their superior properties, including increased specificity, sensitivity, lower detection limits, and rapid response. **Table 1** provides an overview of various techniques used for microplastic detection, including their respective size limits of detection, as well as recent advancements aimed at improving sensitivity and accuracy.

Table 1. Microplastic detection techniques with limit of detection and technological advancements.

Analytical technique	Size of microplastic	Advancements	Description	References
Raman spectroscopy	<1 µm	SERS using klarite as a substrate.	Surface-enhanced Raman spectroscopy (SERS) combined with appropriate substrate has great potential in the detection of microplastic. It can get highly sensitive and detect microplastic up to the size <1 µm.	[51], [52], [59]
		SERS with gold (Au) nanoparticles as a substrate.	Gold has several advantages over silver nanoparticles such as resistance to oxidation. In a study done by Jung et al.[47] it was found that gold was sensitive enough to detect polystyrene (PS) microplastic in an aqueous solution.	
Fourier Transform Infrared Spectroscopy (FTIR)	≥20 µm	Thermal gravimetric analysis-Fourier Transform Infrared Spectroscopy-gas chromatography-mass spectrometry (TGA-FTIR-GC/MS)	FTIR coupled with TGA is highly sensitive and provides high resolution of microplastic detection since it can distinguish plastics on the basis of their thermal properties and thermal degradation.	[55], [48]
		Attenuated total reflection FTIR (ATR-FTIR)	ATR-FTIR can even gather information on thick and opaque microplastic making it more reliable for future advancements in it.	
Dyes	≥20 µm	Hydrophobic dye (Nile red)	It is a lipophilic dye that allows in-situ staining. Polarity of the solvent plays a major role in the intensity of the fluorescent signal. Nile red mixed with its other derivative in water at pH 2.5 was found to be more precise in detection.	[44], [46]
		Hydrophilic dye (Safranine T and Fluorescein is phosphate using thermal expansion and contraction property)	Heat treatment done during the staining of dye to microplastic can maintain the intensity of the fluorescence for months and make the staining more sensitive for detection.	
Biosensors	100 ± 10 µm	Peptide biosensors	It depends on the detection of a particular type of microplastic (for hydrophobic microplastic, hydrophobic peptides are used. It is limited to the size range.	[60], [61]
		Surface Plasmon Resonance (SPR) biosensors	In an experimental study done by Iri et al.[61] estrogen receptors were used and it was found that they can be used for real-time detection of polystyrene in SPR.	

5.4. Terahertz-based detection of microplastic

Microplastics are difficult to detect and characterize due to their small size, which is typically on the order of micrometers or even smaller. The wavelength of the incident light used for detection and characterization typically ranges from infrared light, which has a wavelength of a few micrometers, to terahertz waves, which have wavelengths on the order of millimeters[62,63]. The selection of a specific wavelength depends on the size of the microplastic being investigated[64,65]. Fourier-Transformed Infrared Spectroscopy and Raman spectroscopy have been used to identify and characterize microplastics due to the similarity between their wavelength and the size of the microplastic. Terahertz waves, on the other hand, are more relevant for macroscopic scale investigations[64], allowing for fast detection and imaging of microplastics embedded in larger media. Terahertz waves have been utilized for imaging, sensing, and estimating desired materials, and the time-domain experiments provide amplitude and phase information, enabling the extraction of the complex refractive index of the target materials[66–69]. The refractive index and absorption coefficient can then be employed to probe the desired materials. However, to effectively apply terahertz spectroscopy to microplastic investigations, it is necessary to consider the optical properties of heterogeneous dielectric mixtures and the structure of the sample.

A terahertz-based approach has been developed for the detection of high-density polyethylene (HDPE) microplastic of sizes from 150 to 400 µm mixed with the salt. The detection has been done by time-domain spectroscopy using a collimated terahertz beam with a size of a few centimeters in the transmission mode[70].

The effective medium theory was used to determine the refractive index and absorption coefficient of the samples. The observation shows that the refractive index changes monotonically with the volume ratio, while the absorption coefficient is more sensitive to specific conditions due to the scattering effect in the powder-type sample. Therefore, terahertz spectroscopy focusing on the refractive index can be a more feasible and efficient method for microplastic detection in complex media. This methodology and analysis process can be extended to other materials embedded in complex media.

While the study demonstrated the potential of terahertz time-domain spectroscopy for monitoring microplastics in table salts, there are some limitations and potential drawbacks that need to be considered. One of the main limitations is that the study was conducted on a small sample size and with a limited range of microplastic particles. Further studies on a larger scale and with a wider range of microplastic particles are necessary to validate the effectiveness and reliability of the proposed methodology. Additionally, the practical applicability of the proposed method needs to be evaluated on real-world samples, as the properties of the embedded microplastics in such samples can vary significantly from those in laboratory-prepared samples.

5.5. Biosensor-based approach of microplastic detection

Biosensors are becoming increasingly popular and are receiving a lot of attention these days. The benefits of biosensors include real-time analysis and increased sensitivity.

A great deal of progress has been made in the detection of microplastics utilizing biosensing matrices. Surface plasmon resonance (SPR) biosensing with estrogen receptors, cyanobacterial extracellular polymeric substances (EPS), and peptide-based biosensors have all been investigated recently for the observation of nano and microplastics.

Microplastic was identified in the SPR biosensor utilizing estrogen receptors (ER) by constructing a low-concentration real-time system utilizing estrogen receptors. The plastic sample was pulverized before being filtered with filter paper. The interaction between the ER and the microplastic was studied using chromatography. ER was immobilized on the biosensors using plasma, and the microplastic was detected in SPR utilizing this biosensor[60].

Cyanobacterial extracellular polymeric substances, (EPS) based biosensors are extensively used by researchers due to their distinctive and diversified properties. Due to their structural physical and chemical divergence, they can be used in bioleaching and bioremediation. EPS is often used in biofilm production, can bind metals, and forms bonds/forces like London forces, electrostatic interactions, and hydrogen bonds to bind with microplastics. EPS has highly negatively charged acids that bind to microplastic. These interactions are observed using Electrochemical Impedance Spectroscopy (EIS)[71].

In peptide-based biosensors, peptides are used which can bind with oxidized as well as unoxidized microplastic. It can be very helpful to detect microplastics extracted from oceans because most of them get oxidized due to being underwater for a long time.

In the experimental study titled “Sensitive and specific capture of polystyrene and polypropylene microplastics using engineered peptide biosensors”[61], peptides were used to detect hydrophobic microplastics, specifically polypropylene (PP) and polystyrene (PS). PP and PS were crushed to bead size and separated using size sieves. Since the microplastics are present in an oxidized form in nature, these PP and PS were also oxidized and bound with the hydrophobic peptide. This binding was further analyzed using FTIR. However, to ensure the binding of the peptide a fluorescein isothiocyanate (FIT) as a tracer was affixed at the end of the peptide.

5.6. Detection of microplastic using a machine learning approach

Machine learning approaches have the potential to monitor the pollutants in the environment and the extent it affects human health. Microplastic detection in biological ecosystems is essential because of its detrimental impact on human and animal health via the food web. The current detection methods are not reliable or sufficient for quantifying and classifying microplastics, which is required to understand the extent and impact of microplastic pollution.

One of the promising tools for identifying microplastic is the use of machine learning approaches for analyzing hyperspectral imaging data. The imaging system aids in the location of an object like a microplastic in an image, which is then processed for classification by a neural network for accurate identification[72]. The neural networks used supervised or unsupervised learning approaches to process the input data. The supervised learning approaches used a labeled data set as a sample to train the neural network, whereas the unsupervised learning approaches rely on detecting the hidden pattern from an unlabeled dataset[73]. According to one study, the supervised learning-based approach is only feasible in this scenario because it is unrealistic to have a model that accurately simulates how plastic reacts with light in the real world. The study made use of spectral images of 66 textile samples with varying texture patterns for training the neural network model. The evaluation resulted in a 95% accuracy in microplastic classification after a thousand iterations of training[72].

Machine learning (ML) could help in eradicating microplastic contamination from biological ecosystems by accurately detecting it and giving an image of how they are dispersed. ML could be one of the best solutions for accurately detecting microplastic particles.

5.7. Advancements in the detection of microplastic using nanomaterials

Nanotechnology has received a lot of interest recently. Many advances are being made in this industry to make it more efficient and environmentally friendly. Many studies have been conducted to investigate the use of nanoparticles such as gold and silver to improve the findings of Surface Enhanced Raman Spectroscopy (SERS). In addition, experiments such as integrated carbocatalytic oxidation and hydrothermal (HT) hydrolysis of microplastics over magnetic spring-like carbon nanotubes are carried out to degrade the microplastics found in the wastewater.

Initially, in the nanoparticle-doped SERS, silver (Ag) was used to detect microplastics. In an experiment done, a ratio of 1:1 of PS and silver colloids gave the best results. The silver colloid can be combined with a sample solution in any proportion to achieve varied enhancement efficiencies as a liquid SERS substrate. Achieving the optimal SERS (Surface-enhanced Raman spectroscopy) signal requires maintaining the appropriate ratio between the sample and silver nanoparticles. However, it's important to note that silver nanoparticles have certain limitations that should be considered[74].

Gold (Au) nanoparticles were utilized as a substrate to overcome this constraint. Gold has various advantages over silver nanoparticles, including oxidation resistance. Gold was discovered to be sensitive enough to identify polystyrene (PS) microplastics in an aqueous solution[52].

However, methods like integrated carbocatalytic oxidation and hydrothermal (HT) hydrolysis of microplastics over magnetic spring-like carbon nanotubes were used. The hot environment is beneficial to both catalytic and heat-driven Peroxymonosulfate (PMS) activation in terms of producing more Reactive Oxygen Species (ROS) for the oxidation of MPs and intermediates, particularly because hydrophilic species are more sensitive to ROS. Carbon nanotubes are employed in the Advanced Oxidation Process (AOP) as a metal catalyst. In an experimental study on "Degradation of cosmetic microplastics via functionalized carbon nanosprings"[75], manganese oxides were found as high-performance Fenton-like catalysts with the advantages of abundance on earth and little toxicity to the environment. Nanocomposites were created by encapsulating manganese carbide nanoparticles in helically N-doped carbon nanotubes using one-pot pyrolysis, which was inspired by the properties of both metal and carbon catalysts. Under hydrothermal (HT) conditions, the hybrids were used for PMS activation and MPs degradation. It was discovered that both the HT environment and carbon-driven Surface Radical-Advanced Oxidation Process (SR-AOPs) played critical roles in MPs mineralization.

6. Conclusion

In conclusion, the detection and monitoring of microplastics are essential for understanding their prevalence and impact on human health. Microplastics are pervasive in the environment and can be found in the water we drink, the food we consume, and even the air we breathe. They pose a serious threat to human health due to their endocrine-disrupting properties and ability to accumulate other persistent organic pollutants. Despite their potential risks, the existing detection technologies face several challenges, such as standardizing the analytical procedure and detecting microplastics as small as nanometers. To overcome these challenges, researchers are constantly developing new and innovative methods for detecting and analyzing microplastics. These methods include Fourier-Transform Infrared Spectroscopy (FTIR), Raman spectroscopy, and micro-FTIR imaging, among others.

FTIR spectroscopy is a widely used technique that allows for the identification of microplastics based on their unique spectra. Raman spectroscopy, on the other hand, provides valuable information on the chemical structure of microplastics and can be complementary to FTIR analysis. However, it is important to note that both techniques have their strengths and limitations. While FTIR is highly effective in identifying microplastics, Raman spectroscopy can offer more detailed structural information. It is worth mentioning that Raman spectroscopy may encounter fluorescence issues during measurements, which can affect the accuracy of the results. Furthermore, advancements in modified FTIR techniques, such as TGA-FTIR-GC/MS, ATR-FTIR, μFTIR, and FPA-FTIR, have improved the detection capabilities for microplastics. Similarly, modified Raman spectroscopy techniques like Surface-Enhanced Raman Spectroscopy (SERS), utilizing substrates such as klarite, Gold (Au), and Silver (Ag), have enhanced the detection sensitivity. Nevertheless, ongoing research efforts are necessary to enhance the accuracy and precision of these techniques.

In addition to developing new detection methods, it is crucial to standardize the sampling procedure to ensure consistent and reproducible results. Currently, there is no standardized method for sampling microplastics, leading to variations in the outcomes of different studies. Therefore, researchers are actively working on developing standardized protocols for sampling microplastics in diverse environmental matrices, including water, sediment, and soil.

In conclusion, addressing the issue of microplastic pollution requires collaborative efforts from scientists, policymakers, industries, and individuals. It is imperative to develop effective strategies to reduce microplastic pollution at its source by promoting the use of biodegradable alternatives to traditional plastics. Simultaneously, continuous efforts must be made to improve the detection and monitoring of microplastics, considering the complementary roles of FTIR and Raman spectroscopy, while addressing the fluorescence challenges associated with Raman measurements. By doing so, we can deepen our understanding of the impact of microplastics on human health and the environment, leading to informed mitigation strategies.

Author contributions

Conceptualization, DK, BS and GS; methodology, DK; formal analysis, DK, AK and SG; data curation, BS, GS and MG; writing—original draft preparation, BS, GS, DK and MG; writing—review & editing, DK; supervision, AK and SG.

Conflict of interest

The authors declare no conflict of interest.

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