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REVIEW

Impacts of Biofilm Formation on the Physicochemical Properties and Toxicity of Microplastics: A Concise Review

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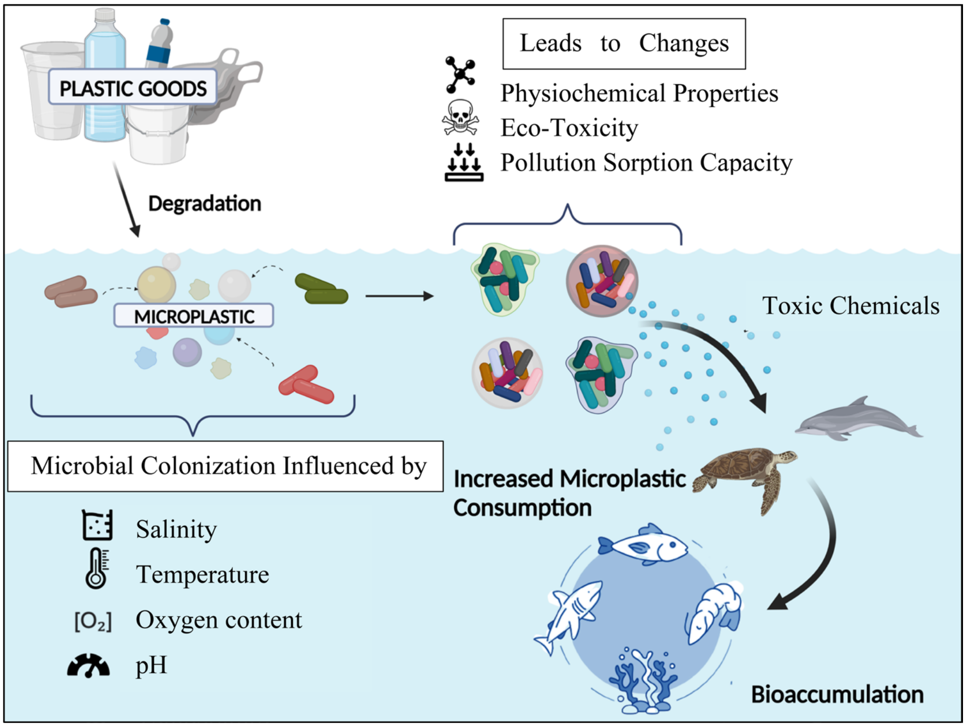
Abstract

Microplastics (MPs) are of global concern due to their slow degradation in the environment and the potential of inducing adverse effects on organisms. In aquatic ecosystems, microbes routinely colonize MPs and develop biofilms on their surfaces. Biofilms are assemblages of surface-associated microbial cells that are enclosed in an extracellular polymeric substance. Emerging evidence has suggested that the development of biofilm can alter the physicochemical properties and the pollutant adsorption capability on MPs. In this article, we review the impacts of biofilm formation on MP properties, ecotoxicity, and fate. First, we summarize the environmental factors that modulate biofilm formation, as well as the unique components of biofilm on MP. Next, we review current understanding on the influence of biofilm formation on the physical and chemical properties of MPs and discuss how these changes affect their pollutant adsorption capacity. Finally, we discuss how biofilm formation on MPs affects their ingestion by organisms and nutrient cycling in aquatic environments.

Graphical Abstract



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Abbreviations

BPA	Bisphenol A
DOM	Dissolved organic matter
EPS	Extracellular polymeric substance
HDPE	High-density polyethylene
MP	Microplastic
PA	Polyamide
PAH	Polycyclic aromatic hydrocarbons
PCB	Polychlorinated biphenyl
PE	Polyethylene
PET	Polyethylene terephthalate
PFOS	Perfluorooctanesulfonic acid
POP	Persistent organic pollutant
PP	Polypropylene
PS	Polystyrene
PSF	Polyester
PVC	Polyvinyl chloride
UV	Ultraviolet
WWTP	Wastewater treatment plant

Introduction

The use of plastics in our everyday life is increasing because of their widespread incorporation into many consumer goods (Thompson et al. 2009). Plastic is one of the most desirable synthetic materials because it is highly durable, light weight, and cost-effective (Sangroniz et al. 2019). Despite US governmental action in 2015 to limit the production of plastic microbeads, worldwide production of plastics continues to increase annually, with approximately 370 million tons produced in 2019 (Bhagwat et al. 2021a; McDevitt et al. 2017). The 2020 coronavirus (COVID-19) pandemic has exacerbated an already dire situation, with plastic waste expected to double by 2030 (Patrício Silva et al. 2021; Peng et al. 2021).

Of the millions of tons of plastic generated each year, only a small fraction is either recycled or incinerated, with the remainder entering landfills or the natural environment (Geyer et al. 2017). Once in landfills, plastic debris can undergo a variety of weathering processes that are abiotic or biotic in nature (Amelia et al. 2021; McGivney et al. 2020). These weathering processes can produce microplastics (MPs), which are typically defined as fragments of any type of plastic with a size ranging from 1 µm to 5 mm (Frias and Nash 2019). Other sizes of plastics include nanoplastics (< 1 µm), mesoplastics (> 5 mm), and macroplastics (> 5 or > 25 mm) (Weber et al. 2022). Some main sources of MPs to the aquatic environment include plastic waste washed by wind and rain, plastic litter from ships, and fishing (Li et al. 2021). The sources of MPs can be broadly categorized into primary or secondary. In general, primary sources of MPs are those that are released into the environment directly. Some of them are purposely created on the micron scale and are often incorporated into consumer products, such as toothpaste, cosmetics, and biomedical products. Primary MPs can also release from the abrasion of larger plastic materials, such as the shredding of synthetic textiles during washing and erosion of tires and brakes during driving. Secondary sources of MPs are from the breakdown of larger plastics in the environment due to weathering (e.g., physical and biological weathering) (Thushari and Senevirathna 2020). A previous study has suggested that up to ~ 30% of plastic in aquatic environments are coming from primary sources, while 66–88% are likely from secondary sources (Boucher and Friot 2017; Estahbanati and Fahrenfeld 2016). The various ways in which MPs are produced and can enter the aquatic environment is illustrated in Fig. 1.

MPs have been found in oceans and freshwater systems worldwide, including deep sea, marshes, lakes, and streams (D'Avignon et al. 2022; Eerkes-Medrano et al. 2015; Krause et al. 2020; McCormick et al. 2014; Połeć et al. 2018). MP abundance is influenced by both environmental and anthropogenic factors, including wave currents, wind conditions, and level of human activity and industrialization (Shahul Hamid et al. 2018; Talbot and Chang 2022). In aquatic environments near WWTPs, MP abundance can be as high as 8,766 particles/m^3 of water (Thushari and Senevirathna 2020). In less-polluted areas, MP concentration typically ranges between 1 and 10,000 particles/m^3 (Koelmans et al. 2019). A few studies have also reported that the amount of

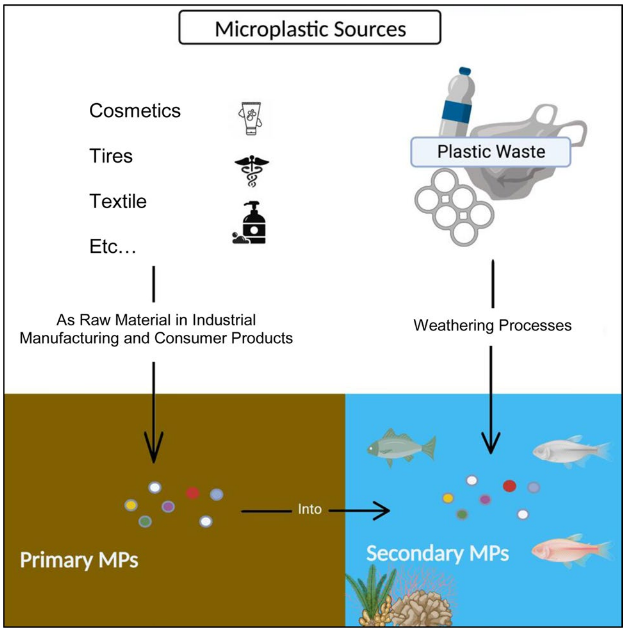

Fig. 1 Possible sources and routes of entry of MPs into aquatic environments

MPs released from landfill leachates is generally in the range of 0 to ~ 300 particles/L (He et al. 2019; Su et al. 2019; Xu et al. 2020). Among various MPs, approximately 55% of all MPs in the aquatic environment are polyethylene (PE) and roughly 17% are polypropylene (PP) (Issac and Kandasubramanian 2021). Other MPs in aquatic environments include polystyrene (PS), polyamides (PA), polyethylene terephthalate (PET), polyester (PSF), and polyvinyl chloride (PVC) (Issac and Kandasubramanian 2021).

MPs can interact with nutrients, organic matter, and other pollutants in the surrounding water. When microorganisms adhere to MPs, they can divide and reproduce on the surface of MPs to form biofilm (He et al. 2022; Wang et al. 2021b). Biofilm is composed of a large consortium of microorganisms and an extracellular polymeric substance (EPS) comprising polysaccharides, proteins, and DNA (di Martino 2018; Lopez et al. 2010). Microbes obtain several benefits from biofilm, including protection against the exposure to ultraviolet (UV) radiation, antimicrobial compounds, and environmental changes (Costa et al. 2018; Erni-Cassola et al. 2020; Yin et al. 2019). The EPS matrix also acts as a nutrient trap to support microbial growth and development (Costa et al. 2018; Flemming and Wingender 2010; Toyofuku et al. 2016; Vu et al. 2009).

Many studies have shown how environmental factors, such as water pH, temperature, and salinity, affect biofilm formation on MPs (e.g., Kesy et al. 2019; Nguyen et al. 2022; Oberbeckmann et al. 2018). Emerging evidence has also demonstrated that the interactions between microorganisms and MPs can affect the physiochemical properties of MPs (Chen et al. 2020; Kaiser et al. 2017; Weinstein et al. 2016). Nevertheless, the impact of these changes on the environmental fate and ecological risks of MPs is yet to be fully explained. The environmental behaviors and toxicity of MPs in aquatic environments, and the influence of biofilm on aging MPs, have been reviewed previously (e.g., Issac and Kandasubramanian 2021; Luo et al. 2022; Ma et al. 2020; Wu et al. 2021). In this review, we focus on the current state of knowledge on the microorganisms–MPs interactions to address the following three major questions: (i) How do microorganisms proliferate on MPs to form biofilm and what are the key factors influencing their formation? (ii) How do microorganisms affect the physiochemical properties of MPs and what are the underlying mechanisms? (iii) How does the formation of biofilm affect the bioaccumulation and toxicity of additives and adsorbed pollutants? Finally, we identify key research gaps and suggest possible future research directions to better understand the health implications of biofilms and MPs.

Development, Composition, and Mediating Factors for Biofilm–MP formation

To determine the behavior and environmental fate of biofilm-coated MPs, it is necessary to first understand how biofilm is developed. The general formation of biofilm is proposed to occur in several steps (1) reversible adhesion, (2) irreversible adhesion, (3) biofilm formation, and (4) dispersal (Fig. 2) (Boakye et al. 2019; Muhammad et al. 2020; Wang et al. 2021b). In reversible adhesion, free-floating microorganisms make initial contact with a surface through electrostatic forces, hydrophobic interactions, and/or Van der

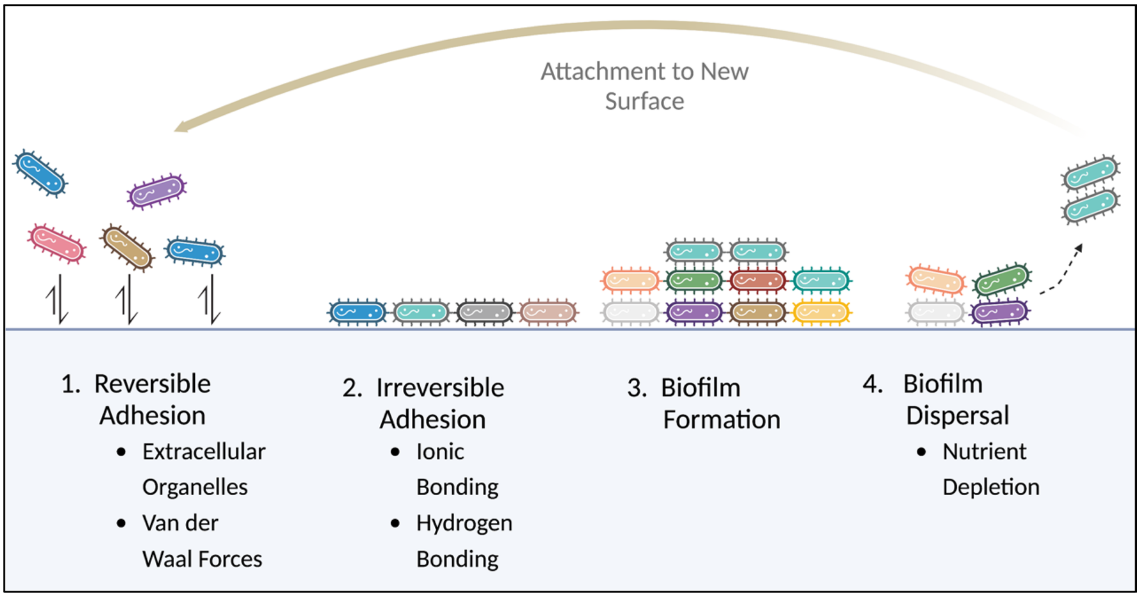

Fig. 2 A proposed process of biofilm formation on the surface of MPs

Waals forces (Kumar and Anand 1998). During this stage, microbes can proceed to form biofilm or return back to the surrounding environment. Microbial extracellular organelles including pili and flagella may be used at this stage (Boakye et al. 2019; Ma et al. 2022; Toyofuku et al. 2016). The shift from reversible adhesion to irreversible adhesion involves the introduction of covalent, ionic, and hydrogen bonding which affix microorganisms into place (Kumar and Anand 1998; Muhammad et al. 2020). Surface proteins and adhesins such as fimbriae and lipopolysaccharides may also be used by bacteria for attachment (Muhammad et al. 2020; Toyofuku et al. 2016). Next, adhered microorganisms may divide and secrete EPS, forming microcolonies (Kumar and Anand 1998; Toyofuku et al. 2016; Zhao et al. 2013). The EPS plays a key role in microbial structure, attachment, water retention, and resistance from environmental stressors, such as UV radiation and biocides (Flemming and Wingender 2010; Toyofuku et al. 2016; Vu et al. 2009). The EPS can also serve as a nutrient source and electron donor/acceptor for microorganisms (Flemming and Wingender 2010). The EPS can account for over 90% of the dry mass of biofilm (Flemming and Wingender 2010). Between the firmly compacted cells, a network of hollow channels forms, which allows for the exchange of oxygen, nutrients, and waste (Geisel et al. 2022; Quan et al. 2022). The final stage of biofilm formation is environment dependent and involves the detachment of microorganisms from the biofilm. The detachment process can be stimulated by various factors, such as depletion in nutrients and reduction in environmental oxygen level. These changes can ultimately lead to modulation in the expression of matrix-synthesizing genes (Hunt et al. 2004; Karatan and Watnick 2009).

MPs in marine environments can be colonized by a variety of bacteria and fungi, and the specific types of bacteria vary with environmental conditions, such as light intensity, pH, and temperature (Rummel et al. 2017). Common bacteria phyla identified on MPs collected from marine environments include Proteobacteria, Bacteroidetes, and Firmicutes (Table 1). De Tender et al. (2017) used next-generation sequencing to analyze the biofilm communities on PE MPs incubated in marine environments. Their results revealed the presence of several species from the Ascomycota and Basidiomycota phyla and to a lesser extent Zygomycota (de Tender et al. 2017). On the other hand, Kettner et al. (2019) and Wang et al. (2021c) demonstrated that exposure of PE and PS MPs to urban river or downstream of wastewater treatment plant (WWTP) results in the colonization of these MPs by microorganisms, such as Blastocladiomycota, Mucoromycota, Rhinosporideacae, and Rhizidiomyces. These results indicate that the composition of microorganisms found on MPs is influenced by sampling locations.

Biofilm composition also appear to differ during different stages of biofilm formation. For example, Gammaproteobacteria, particularly from the genus Oleinacter, was found to comprise up to 59% of the microbial community on PVC during early colonization stages (Pollet et al. 2018; Stabnikova et al. 2021). However, these microbes were quickly outnumbered by Alphaproteobacteria (predominantly from the Rhodobacteraceae and Flavobacteriia families) shortly thereafter (Pollet et al. 2018; Stabnikova et al. 2021). Other microbial families (e.g., *Phyllobacteriaceae,*

Table 1 The predominant microorganisms found in biofilm on MP or plastic debris in aquatic environments

Microorganism	Phylum/class	Abundance	Type and location[a]	References
Bacteria	Gammaproteobacteria	Comprised up to 59% of biofilm during the first hours of biofilm development	PET in the North Sea PS in artificial seawater	Oberbeckmann et al. (2016) Ye et al. (2021)
	Alphaproteobacteria	Comprised the majority of biofilm during late stages of biofilm development	PVC and PP in the Yellow Sea and the South China Sea PE in the Baltic Sea	Xu et al. (2019) Kesy et al. (2019)
	Bacteroidetes	Comprised up to 96% of biofilm during latest stages of biofilm development	PET in the North Sea PS in seawater microcosms PVC in the Haihe River	Oberbeckmann et al. (2016) Ye et al. (2021) Wu et al. (2019)
	Firmicutes	Comprised 11.7% of biofilm during late stages of biofilm development	PE in lake water from Jinan, China PET in the Chicago River, USA	Gong et al. (2019) Hoellein et al. (2014)
Algae	Microalgae	> 240 microalgal species developed in biofilm	PET and HDPE in freshwater microcosms	Nava et al. (2022)
	Diatoms	No Data	PET in the North Sea, PS in seawater microcosms	Oberbeckmann et al. (2016)
Fungi	Ascomycota	Comprised 0.6% of total biofilm after 22 weeks of incubation	Plastic debris in Osted Harbor, Belgium	de Tender et al. (2017)

[a] Note that some studies are incubation experiments with water collected from the fields

Planctomycetaceae) are most abundant at the later stages of biofilm development (Pinto et al. 2019). Additionally, it has been shown that MP-coated biofilms possess greater species diversity compared to biofilm on natural substrates (Mughini-Gras et al. 2021). The Shannon–Wiener diversity index of MP–biofilm is found to be 40% and 25% higher than that of leaves and surrounding water, respectively (Wu et al. 2019). As a result of the uniqueness of biofilm-coated MPs, they form an ecological niche typically referred to as a "plastisphere" (Zettler et al. 2013; Amaral-Zettler, 2020). In some studies, it has also been reported that biofilm microbial diversity on MPs peaks after 1 week of microorganism colonization in marine environments (Delacuvellerie et al. 2019; Yang et al. 2020; Stabnikova et al. 2021).

Compared to marine MP–biofilms, freshwater MP–biofilms are characterized by lower values on the Shannon–Wiener Diversity index, suggesting they possess lower species richness and diversity (McCormick et al. 2014; Miao et al. 2019b; Yang et al. 2020; Fang et al. 2021). Hoellein et al. (2014) analyzed the biofilm on PET MPs obtained from three Chicago freshwater settings: a river, pond, and an artificial stream. They found that the predominant taxa of microorganisms in all samples was identical: *Proteobacteria*, *Verrucomicrobia*, *Firmicutes*, and *Bacteroidetes*. Similar to marine environments, microbial communities found on MPs collected from freshwater is distinct from that in the surrounding waters (Harrison et al. 2018; McCormick et al. 2014). This is likely due to the unique substrate that the MPs provided for microbial colonization.

Interestingly, Miao et al. (2021b) reported that bacterial networks formed on PVC were more complex than on natural substrates. Compared to natural substrates, freshwater MP–biofilms appear to possess lower alpha diversity (Miao et al. 2019b; Mughini-Gras et al. 2021). Nguyen et al. (2023) studied biofilm composition on biodegradeable plastics from a freshwater reservoir and found temporal changes in biofilm composition. Overtime, early colonizers (e.g., Rhodobacteraceae) decreased in abundance, while several other groups increased by up to four times thereafter (Nguyen et al. 2023). Therefore, bacterial colonization and biofilm formation on MPs are a very dynamic process and can vary spatiotemporally.

Bio-based plastics (i.e., made with biodegradable or renewable materials, such as cellulose) have received a lot of attention these days; however, there are issues associated with their use, such as the requirements of specific conditions for their biodegradation and the high environmental impact during manufacturing (Wang et al. 2021a; Tabone et al. 2010). Biofilm can also form on biodegradable plastics (Napper and Thompson 2019). Kirstein et al. (2018) found that there were differences between the microbes that colonized biodegradable plastics (e.g., made of polylactic acid; PLA) and conventional plastics. Morohoshi et al. (2018) also reported that some microbes could degrade a biodegradable plastic called poly(3-hydroxybutyrate-co-3-hydroxyhexanoate). Importantly, a study showed that biodegradable plastics that are made of polylactic acid (PLA) may produce more MPs than PS during degradation (Lambert and Wagner 2016). Zuo et al. (2019) also showed that biodegradable plastics such as those made with poly(butylene adipate co-terephthalate) can adsorb more phenanthrene (an organic pollutant) when compared to the conventional MPs. Clearly, there is an urgent need to further understand the risk of biodegradable plastics and their interactions with biofilm before fully adopting the use of biodegradable plastics.

Pathogenic Microorganisms in MP–Biofilm

In marine environments, a few studies have suggested that potentially harmful microorganisms can be enriched on MPs, including *Vibrio and Arcobacter spp.* (McCormick et al. 2014; Harrison et al. 2018). Zettler et al. (2013) found that a member of the genus *Vibrio,* which is pathogenic to humans and various aquatic animals, constituted nearly 24% of PP biofilm in samples collected from the North Atlantic Ocean. This finding is alarming because most members of this genus of bacteria are rarely found in abundance greater than 1% in the community (Zettler et al. 2013). *Vibrio sp.* is also highly pathogenic to fish and invertebrates, and it can induce gastroenteritis, muscle necrosis, and eye lesions (Zhang et al. 2020b). Furthermore, Viršek et al. (2017) observed high levels of *Aeromonas salmonella* on PP and PE MPs sampled from the Slovenian Coast. *Aeromonas salmonella* is the causative agent of salmonella disease and is linked to widespread mortality in fish, reptiles, crustaceans, and several other animals. On the other hand, Curren and Leong (2019) identified *Arcobacter* and *Photobacterium rosenbergii* on MPs, and these species have been associated with gastrointestinal disease and coral bleaching, respectively. Opportunistic pathogens such as *Pseudoalteromonas, Burkholderia, Alteromonas*, and *Tenacibaculum* have also been observed on MPs (Hou et al. 2021; Ogonowski et al. 2018; Sun et al. 2020). Notably, Hou et al. (2021) reported that there was no enrichment of pathogenic bacteria on MPs when compared to the surrounding environment. On the other hand, there are currently no reports of harmful fungi being found on MPs. However, harmful algae such as *Coolia* and *Ostreopsis* spp. have been observed on plastic debris collected from the Catalan coast of Spain (Masó et al. 2003).

Pathogens are also associated with MP–biofilms collected from freshwater environments. *Flavobacterium*, which can cause columnaris in fish, has been identified on the surface of MPs (Szabó et al. 2021). Szabó et al. (2021) have also found members of the *Mycobacterium* genus on PP MPs. Many species of mycobacteria (e.g., *M. marinum*) are pathogenic to fish and their exposure could lead to aberrant swimming behavior, weight loss, swelling, and liver granulomas (Hashish et al. 2018). Other pathogenic species such as *Helicobacter* spp., *Enterobacter* spp., and *Escherichia* spp. have also been found on MPs (Murphy et al. 2020). Notably, Shen et al. (2021) demonstrated that MPs could reduce the effectiveness of UV light disinfection treatment in conditions mimicking wastewater treatment, likely owing to the blockage of UV ray penetration by MPs. Collectively, these findings suggest that biofilm-coated MPs may harbor potentially harmful pathogens which could pose a serious threat to aquatic health.

Biofilm found on the surface of MPs has been proposed to promote gene exchange and can harbor antibiotic resistance (Arias-Andres et al. 2018; Wu et al. 2019). For example, Zhang et al. (2020a) found that the abundance of antibiotic-resistant bacteria on MPs collected from a pond was 100–5000 times higher than in the surrounding water. Sucato et al. (2021) collected PE MPs from a stream and seawater off the coast of Sicily, and they observed that the occurrence of antibiotic resistance genes on PE was higher than in the surrounding water. Similarly, Pham et al. (2021) observed an increase in the abundance of antibiotic-resistant genes on PS and PE MPs following their incubation with WWTP sludge. Considering the high mobility and persistence of MPs, biofilm-coated MPs could be long-term sources of antibiotic resistance.

MP Properties that Influence Biofilm Development

The type and the physical–chemical properties of MPs greatly influence the abundance and composition of biofilms on MPs. One important property is the density of MPs, which can affect the vertical distribution and mobility of MP in the water column, subsequently affecting the process of microbial colonization (Stabnikova et al. 2021; Shamskhany et al. 2021). MPs with a lower density than water (e.g., PP, PE) tend to float along the water surface while denser plastics (PVC, PET) sink to the substratum. This is an important consideration because different microbial communities exist at different water layers. For example, Jones et al. (1991) studied bacterial community structure in several freshwater lakes in Michigan, USA, and they observed significantly higher numbers of gram-negative heterotrophic bacteria in the shallowest regions of the lake compared to deeper layers of water. Conversely, anaerobic microorganisms are more abundant in deeper layers of water (Niu et al. 2021). Notably, Niu et al. (2021) observed a higher abundance of MP-degrading bacteria on MPs collected from deeper layers of water. Hence, the density of MPs can influence microbial colonization and their environmental mobility and fate.

A few studies have suggested that the functional groups of certain types of MPs can attract or repel microorganisms. For example, the cyclohexyl functional group, which is found in a very limited number of MPs, repels certain bacterial species (Sanni et al. 2015; Wang et al. 2021b). In contrast, the presence of aromatic groups in MPs has been shown to promote the attachment of some bacteria (Wang et al. 2021b; Sanni et al. 2015). Several studies have also demonstrated that the surface properties of MPs affect microbial colonization. Miao et al. (2021a) observed that the biomass and microbial diversity on PVC MPs are highly different from that on PET MPs. Nava et al. (2022) found that microalgae biomass is higher on PET MPs when compared to high-density PE (HDPE) MPs. An increase in surface roughness of MPs has also been shown to enhance microbial attachment, likely owing to an increased surface area (Hossain et al. 2019). Foulon et al. (2016) observed that the colonization time of *Vibrio* bacteria is longer on rough PS MPs than on smooth PS MPs. Interestingly, Parrish and Fahrenfeld (2019) observed that Betaproteobacteria are more abundant on smooth PS MPs, while Gammaproteobacteria are more on rough PE. On the other hand, Frère et al. (2018) found that MP size has no effect on bacterial community structure; however, they observed that PS MPs exhibit distinct bacterial assemblages when compared to PE and PP MPs. These results suggest that differences in surface roughness and MP types can contribute to differences in microbial colonization.

It is important to also note here that many plastics contain additives, such as colorants, flame retardants, stabilizers, and plasticizers (Hahladakis et al. 2018). Increasing evidence has shown that these additives can affect microbial colonization on MPs. For example, additives leached from PVC and HDPE can inhibit the growth and proliferation of *Prochlorococcus spp.* (Tetu et al. 2019), a cyanobacterial species commonly observed on plastics (Rogers et al. 2020; Lear et al. 2021). Similarly, brominated flame retardants and bisphenol compounds (e.g., BPA), which are commonly incorporated into MPs, are toxic to microalgae and bacteria (Debenest et al. 2010; Kousaiti et al. 2020; Li et al. 2009). In contrast, additives such as phthalate esters have been found to increase *Pseudomonas* biofilm formation under certain conditions (Wang et al. 2022). On the other hand, the color of plastic debris also appears to affect biofilm composition and abundance on their surfaces. De Tender et al. (2015) collected resin pellets from a beach in Oostende, Belgium, and they observed that yellow- and blue-colored resin pellets have higher abundance of *Mycobacterium frederiksbergense* when compared to other color of pellets. Overall, these findings suggest that certain additives or coloring chemicals found in MPs can modulate the type and abundance of microorganisms colonized onto MPs.

Environmental Factors that Influence Biofilm Community and Structure

Environmental factors, such as temperature, salinity, water flow, and nutrient concentration, can have an impact on the development and composition of biofilm on MPs. Using bioreactors, Nguyen et al. (2022) demonstrated that biofilm communities on MPs are highly influenced by organic content in the water, followed by salinity and dissolved oxygen content. Notably, Oberbeckmann et al. (2018) reported that the more nutrients available, the faster primary and secondary biofilm (i.e., late colonizers) can develop on MP. Another important factor that can influence biofilm formation is water temperature (de Tender et al. 2015; Aryal et al. 2019). Chen et al. (2019) found that biofilm formation on plastic is higher in the summer than winter when temperature and nutrient levels are optimal. Oberbeckmann et al. (2014) also found that biofilm thickness on PET increases from winter to summer. In another study by Oberbeckmann et al. (2018), they observed that temperature is an important factor influencing microbial community structure on PE and PS. Additionally, Zeraik and Nitschke (2012) found that temperature influences the adhesion of *Pseudomonas aeruginosa* to PS surfaces in certain conditions. The increased biofilm formation at elevated temperature is probably associated with increased enzymatic activities and metabolic rates, which in turn promoting the development of microbes (Chen et al. 2019; Garrett et al. 2008). Additionally, the functioning of extracellular attachment organelles such as flagella can be influenced by temperature (Alotaibi 2021; Humphries 2013).

Salinity also appears to affect biofilm formation. For example, Li et al. (2019a) observed that salinity is the primary determinant of bacterial diversity on plastic debris. Similarly, Oberbeckmann et al. (2018) observed that salinity helps shape microbial community structure on PE and PS. Kesy et al. (2019) also suggested that salinity is the major factor affecting microbial assemblages on MPs. On the other hand, Suhrhoff and Scholz-Böttcher (2016) reported that salinity has very low impact on the leaching of additives from MPs.

Most microbes grow best at pH 5 to 8.5, and this optimum pH allows microbes to efficiently pump out protons (H^+) from their cell to maintain a relatively neutral cytoplasm (Garrett et al. 2008). Unfavorable pH levels can therefore disrupt homeostatic function and in turn adversely affect biofilm formation. It has been suggested that changes in pH may also impair the secretion of exopolysaccharides in microorganisms, which is a key component of EPS (Ju et al. 2022; Mahto et al. 2022). Once biofilm formed, however, the microorganisms within the biofilm would become less sensitive to extreme pH changes than the planktonic state (Yin et al. 2019). On the other hand, some studies have shown that the surface of PE MPs become negatively charged in marine environments, which may hinder attachment of bacteria onto MPs (Fotopoulou and Karapanagioti 2012).

Currently, there is limited information on the influence of fluid dynamics on biofilm formation. Some simulation studies have suggested that a high water flow may lead to higher rates of erosion and detachment of microbes (Gomes et al. 2014; Picioreanu et al. 2001). Other study has reported that high water flow decreases the density but increases the diversity of biofilm (Han et al. 2018). However, it has also been shown that high water velocity can sometimes increase biofilm formation. For example, Lehtola et al. (2006) found that the formation of biofilm on PE pipes increases as water velocity increases. The increase in biofilm formation at high water velocity is likely due to an increase in water mixing, thereby increasing nutrient exchange and dissolved oxygen content.

Physicochemical Changes of MPs Induced by Biofilm Formation

Hydrophilicity and Density

Biofilm formation has been shown to alter several chemical and physical properties of MPs. One of the properties that can be altered by biofilm development is hydrophilicity. Specifically, the development of biofilm can increase the hydrophilicity of plastics, including those composed of PS and PE (Ganesan et al. 2022; Liu et al. 2022b; Lobelle and Cunliffe 2011; Nauendorf et al. 2016). For example, Lobelle and Cunliffe (2011) showed that submersion of PE plastic bags in seawater results in a decrease in their hydrophobicity. This observation was likely due to the formation of biofilm on the plastic (Lobelle and Cunliffe 2011). Importantly, Chavant et al. (2002) suggested that biofilm formation is generally faster on hydrophilic surfaces than on hydrophobic surfaces. In contrast, Pompilio et al. (2008) found that the hydrophobicity of PS increases following its adhesion by *Stenotrophomonas maltophilia*, suggesting changes in hydrophobicity can be influenced by the type of plastic material and microorganisms.

Apart from hydrophilicity, multiples studies have reported that biofilm formation can increase the density of MPs, leading to changes in their environmental transport (Kaiser et al. 2017; Int-Veen et al. 2021; Semcesen and Wells 2021). For example, using an ex situ experiment, Semcesen and Wells (2021) demonstrated that biofilm formation can increase particle density, leading to increased sinking behavior in aquatic environments. Similarly, Int-Veen et al. (2021) reported that biofouling plays an important role in driving vertical transportation of MPs in the water column. Kaiser et al. (2017) also demonstrated that the sinking velocity of PS MPs is enhanced after biofilm formation (81% and 16% increase in marine water and estuarine water, respectively). However, Kaiser et al. (2017) noted that biofilm formation alone is not sufficient to increase sinking of PE MPs; the attachment of macro-fouling organisms such as mussels is necessary for enhancing plastic sinking. Minerals can also incorporate into the biofilm matrix, leading to increases in volume (Flemming and Wingender 2010; He et al. 2022). Kalčíková and Bundschuh (2022) suggested that biofilm may reduce the bioavailability of MPs and adsorbed pollutants by increasing their sedimentation rate. Therefore, a change in density and sinking velocity of MPs can have an important influence on the environmental fate and transport, thereby affecting their exposure to organisms.

MP Degradation by Microorganisms

In recent years, an increasing number of studies have shown that biofilm formation can alter surface texture and degrade MPs. Some microbes within biofilms can secrete enzymes to degrade MPs, such as esterases, lipases, and cutinases (Kaushal et al. 2021). To date, over 400 species of bacteria and fungi have been found to possess plastic-degrading properties (Lear et al. 2021). Mouafo Tamnou et al. (2021) observed that in an acidic aquatic microcosm, the highest weight loss (6.25%) of PE is achieved following its incubation with *Pseudomonas aeruginosa* for 30 days at 44 °C (Mouafo Tamnou et al. 2021). Under in vitro condition, Qi et al. (2021b) recorded a weight loss of 13.6% in 7 days using various engineered microbes. On the other hand, Yoshida et al. (2016) report that *Ideonella sakaiensis* 201-F6 can secrete two enzymes capable of hydrolyzing PET completely within 6 weeks. The first enzyme, PETase, transforms PET to mono-(2-hydroxyethyl) terephthalate (MHET). The second enzyme, MHETase, converts MHET to ethylene glycol and terephthalic acid (Palm et al. 2019; Yoshida et al. 2016). Notably, a study has shown that addition of MPs in activated sludge can increase denitrification rate by providing additional anaerobic atmosphere on the inner part of the MPs (Li et al. 2020). However, some studies have reported that biofilm formation decreases MP degradation by blocking UV radiation and increasing MP sinking (Andrady 2015; Weinstein et al. 2016). The general process of plastic breakdown is illustrated in Fig. 3.

A few studies have shown that the MP surface becomes rougher following biofilm formation. For example, Özdemir et al. (2022) reported holes, grooves, and cracks in PE MPs following 10 days of incubation with thermophilic bacteria. Auta et al. (2017) also showed changes in surface roughness in PE, PET, PP, and PS after biofilm formation. Notably, MP surface roughness may affect the sorption ability of pollutants (Gao et al. 2021; Joo et al. 2021), a topic discussed later. Identification of bacteria and fungi species in plastic degradation has led to the development of novel MPs mitigation strategies. However, there are several reasons why this mitigation strategy has not materialized. First, degradation process can be highly dependent on treatment condition. Second, different MPs appear to require different microorganism for degradation, which means multiple strains of bacteria may have to be utilized (Kaushal et al. 2021; Yuan et al. 2020). Lastly, as previously discussed, toxic additives can release into the surrounding water during weathering/degradation, leading to changes in exposure risk and toxicity (Luo et al. 2020). Selected examples of microorganisms that can degrade plastic are summarized in Table 2.

Alterations in the Pollutant Adsorption Capacity by Biofilm

MPs have been reported to influence the mobility and toxicity of metals (Liu et al. 2021). There is also accumulating evidence that biofilm-coated MPs can adsorb more heavy metals than virgin MPs (Guan et al. 2020; Liu et al. 2022b; Richard et al. 2019; Qi et al. 2021a; Qiongjie et al. 2022). For example, Qiongjie et al. (2022) reported that the adsorption capacity for copper and lead is higher in biofilm-coated PS MPs than in virgin or UV-treated PS MPs. Liu et al. (2022b) incubated PE and PET plastic debris in different freshwater environments, and they also observed higher

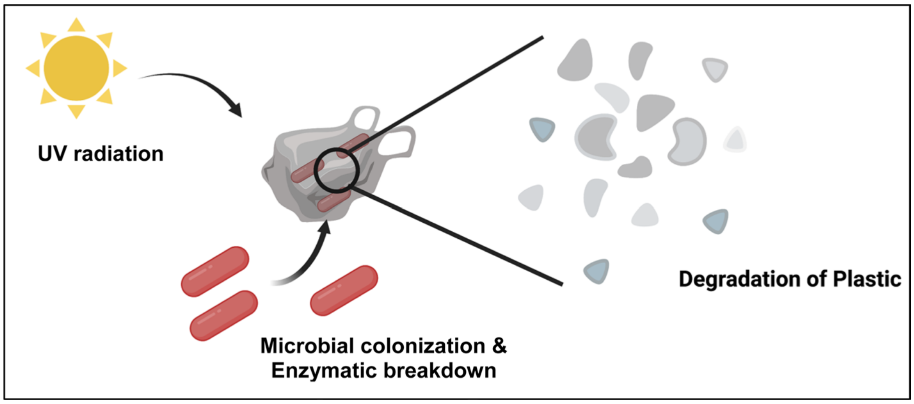

Fig. 3 A simplified illustration of plastic breakdown by UV radiation and microbial action

Table 2 Selected examples of microorganism that can degrade plastic

Plastic type	Microorganism	Degradation time (days)	Weight loss (%)	References
PE	Complex mixture	182.5	19	Syranidou et al. (2017)
PE	*Bacillus* sp. strain YP1	60	10.7	Yang et al. (2014)
PE	*Pseudomonas aeruginosa*	30	6.25	Mouafo Tamnou et al. (2021)
PP	Complex mixture	60	14.7	Park and Kim (2019)
Low density PE	*Anabaena spiroides*	7	8.18	Vimal Kumar et al. (2017)
PVC	*Pseudomonas citronellolis*	30	13.07–18.58	Giacomucci et al. (2019)
PET	Enzyme from *Saccharomonospora viridis* cloned in *Escherichia coli*	3	13.5 for PET-GF and 27 for PET-S	Kawai et al. (2014)

levels of lead, cadmium, and zinc on biofilm-coated MPs. The authors suggested that the formation of anionic functional groups in biofilm (e.g., carboxyl) may attract positively charged metals, thereby increasing their adsorption. A few studies have also demonstrated that biofilm-coated MPs can adsorb radioactive caesium and chromium onto their surfaces. (Johansen et al. 2018, 2019). Currently, it is still unclear whether the valency of metals (e.g., divalent metals vs trivalent metals) affects their adsorption affinity into MP–biofilm. Richard et al. (2019) determined the adsorption of various metals (e.g., aluminum, manganese, nickel, zinc) into biofilm–MP and did not observe the influence of valency on their adsorption. They also reported that plastic type and biofilm formation are more important in modulating metal accumulation onto MPs (Richard et al. 2019). On the other hand, Liu et al. (2022b) showed that the adsorption capacities for lead (II) is higher than that for cadmium (II) and zinc (II). The authors proposed that the differences in the adsorption capacities into MP–biofilm are governed by film diffusion kinetics (Liu et al. 2022b). On the other hand, a study by Tu et al. (2020) observed that biofilm-coated MPs can contain new functional groups, such as the carbonyl group, ketone, amino, phenyl-OH, and aromatic groups, which were not present in virgin MPs. Other ionizable functional groups in EPS of biofilm include phosphoryl, amino, and hydroxyl group (Liu et al. 2022b). Because these functional groups are negatively charged, it has been suggested that formation of these groups can enhance electrostatic interactions and complexation between metals and biofilm-coated MPs (Bhagwat et al. 2021b; Guan et al. 2020; Liu et al. 2022b; Qiongjie et al. 2022). Additionally, Li et al. (2019b) found that sewage sludge treatment with MPs results in the formation of new functional groups on the surface of MPs, which appears to contribute to a higher adsorption capacity for cadmium. On the other hand, biofilm formation may increase the formation of hydrous oxides that can scavenge metals (Bhagwat et al. 2021b; Richard et al. 2019). Interestingly, biofilm formation may also enhance desorption of certain metals from MPs. For example, Kalčíková et al. (2020) reported that biofilm-coated PE MPs releases 71% of the adsorbed silver while virgin MPs only released 29%.

Several environmental factors may influence metal adsorption onto MPs. Holmes et al. (2014) found that adsorption of cadmium, cobalt, and nickel to aged and virgin MPs increases with increasing water pH, whereas adsorption of chromium decreases with increasing pH. Lin et al. (2021) found that lead adsorption onto MPs increases when pH increases. On the other hand, Oz et al. (2019) observed that temperature is positively correlated with lead and aluminum adsorption onto virgin MPs, although the influence appear to be relatively weak. Additionally, a few studies have reported that salinity (i.e., ionic strength) of the surrounding water can influence metal adsorption on both biofilm-coated and virgin MPs (Qi et al. 2021a). Lin et al. (2021) found that higher salinity reduces the adsorption of lead onto MPs. Liu et al. (2022b) also reported that the addition of calcium salt decreases the adsorption of lead onto MPs. Overall, these studies demonstrated that both the introduction of new functional groups by biofilm and the physical–chemical condition of the water are important factors influencing the adsorption capacity of MPs for metals.

There is accumulating evidence suggesting that biofilm can enhance the adsorption of persistent organic pollutants (POPs) onto MPs. POPs are a group of toxic chemicals that are resistant to degradation and therefore can persist in the environment for extended periods of time (D'Agostino et al. 2020). For example, Cui et al. (2023) observed that the formation of biofilm onto HDPE MPs increases the adsorption of various POPs, including polybrominated diphenyl ethers, polychlorinated biphenyls, and α-hexabromocyclododecane. They also reported that biofilm formation increases the surface area of HDPE MPs, potentially contributing to the increased chemical adsorption. Bhagwat et al. (2021b) observed that biofilm formation on PE MPs increases the adsorption of perfluorooctanesulfonic acid (PFOS) by over 75%. Similarly, Wu et al. (2017) and Wang et al. (2020a) demonstrated that the adsorption of polycyclic aromatic hydrocarbons (PAHs) and antibiotics such as tetracycline is higher in biofilm-coated MPs (Wu et al. 2017). Interestingly, Jin et al. (2020) found that some microbes in MP–biofilms can degrade PAHs, thus having a potential to modulate the toxicity of PAH-adsorbed MPs.

The adsorption of organic pollutants in MPs can be influenced by environmental factors. For example, Fu et al. (2021) reported that the adsorption capacity of the antibiotic sulfamethoxazole in PA MPs is higher in acidic conditions ($\text{pH} \leq 6.7$) than in alkaline conditions ($8 \leq \text{pH} \leq 9$). Kong et al. (2021) found similar findings with artificially aged PET and PP MPs. They found that at higher water pH, sulfamethoxazole became negatively charged and therefore created repulsion with negatively charged MPs (Kong et al. 2021). On the other hand, Kong et al. (2021) observed that as salinity increases, sulfamethoxazole adsorption capacity onto some artificially aged MPs decreases due to its competition with Na^+ ions. However, Joo et al. (2021) reported that higher water salinity increases PFOS adsorption onto PE and PS MPs. Research has shown that dissolved organic matter (DOM) can also reduce the adsorption of certain pharmaceuticals and personal care products onto PE, likely owing to complexation of DOM with organic pollutants or competition for sorption sites on MPs (Wu et al. 2016).

Risk of biofilm–MPs in aquatic environments

Biofilm-coated MPs which have a similar appearance, taste, and smell to nutrient-dense foods may be more likely to be consumed by organisms when compared to virgin MPs (Botterell et al. 2020; Stabnikova et al. 2021). For example, filter feeders such as bivalves were found to ingest biofilm-coated MPs over virgin MPs (Fabra et al. 2021). Vroom et al. (2017) found that some zooplankton consumed aged MPs at higher amounts than virgin MPs. Hodgson et al. (2018) found that biofilm increased the amount of shredding of plastic carrier bags performed by an amphipod. Sandy anemone (*Bunodactis reynaudi*) also showed a preference to ingest aged plastics, suggesting biofilm may influence its food selection (Weideman et al. 2020). Fabra et al. (2021) reported that uptake of biofilm-coated MPs in oysters was significantly higher than that of virgin MPs. Interestingly, Allen et al. (2017) found that biofilm reduced the ingestion of plastic by coral. Overall, these findings suggest that biofilm formation on MPs can alter their ingestion by organisms. Notably, some MP-coated biofilms are known to release noxious signals called infochemicals into water which encourage nearby organisms to ingest them (Botterell et al. 2020). For example, certain species of phytoplankton can release dimethyl sulfide (DMS) and dimethylsulfoniopropionate (DMSP) which prompt nearby foragers to consume them (Botterell et al. 2020).

Several recent studies have suggested that MPs may contribute to the formation of algal blooms (Nava and Leoni 2021; Stauffer et al. 2019; Wang et al. 2021d). Liu et al. (2022a) observed that MP abundance was positively correlated with algal density in early stages of algal bloom but negatively correlated in later stages. Wang et al. (2021d) observed that the co-occurrence of MPs and lead (II) promoted the growth of *Microcystis aeruginosa*, which is a species of cyanobacteria that can form blooms. Additionally, Cole et al. (2015) found that MP exposure reduced the ingestion of *Thalassiosira weissflogii* (i.e., a species that is typically associated with the formation of red tide) by a marine copepod (*Calanus helgolandicus*). Likewise, MPs may replace food in the diet of zooplankton, which reduces their grazing on primary producers, like algae (du Plooy et al. 2017; Kvale et al. 2021). Therefore, MPs appear to have the potential to promote algal blooms which could lead to various adverse consequences in aquatic environments, such as their production of harmful toxins and depletion of dissolved oxygen.

A few studies have demonstrated that biofilm-coated MPs could affect nutrient cycling in aquatic environments (Shen et al. 2022). For example, biofilm-coated MPs are found to affect nitrogen cycling by increasing denitrification in artificial freshwaters (Chen et al. 2020). A study also showed that when biofilm disintegrates, nitrogen and phosphorous are released back to the environment. Miao et al. (2019a) observed that PS MPs reduce the enzymatic activity of β-glucosidase and leucine aminopeptidase, which are important enzymes for carbon and nitrogen cycling, respectively. Additionally, Cluzard et al. (2015) demonstrated that MP pollution may have the potential to modulate ammonium fluxes in intertidal sediments. Furthermore, the degradation of plastics has been suggested to be a major source of dissolved organic carbon (Fauvelle et al. 2021; Romera-Castillo et al. 2018), thus having a potential to influence carbon cycle. These changes in nutrient cycling could impact the growth and community dynamics of phytoplankton.

Conclusions and Future Directions

Biofilm formation onto MPs involves the attachment of microorganisms on their surface, the secretion of EPS, and the replication and colonization of microorganisms. Biofilms on MPs can harbor potentially pathogenic microorganisms, facilitating their spread across aquatic communities. The formation of biofilm on MPs can be influenced by various environmental factors, including salinity, dissolved oxygen content, and water flow velocity. Biofilm has also been shown to alter the sinking, hydrophobicity, and functional groups of MPs. These changes ultimately alter the adsorption capacity for other pollutants. Notably, biofilm-coated MPs can also release chemical signals into water which encourage their ingestion by organisms. This may have the potential to increase the uptake and biomagnification of pollutants along the food chain.

Over the past decades, significant effort has been made to understand the interactions between microorganisms and MPs and their implications on the environmental fate and toxicity in aquatic environments. Nevertheless, there are key research gaps that need to be addressed in future studies. First, there are few studies examining the behavior and fate of biofilm-coated MPs in freshwater settings. The influence of hydrogeological factors and biofilm on the mobility and deposition of MPs in freshwater environments is a critical research gap. Second, future studies on biofilm–MP interactions should focus on field experiments (e.g., types of colonizing species, modification of MP structure and composition, physical–chemical effects on bacterial attachment and detachment) to account for a myriad of complex biotic and abiotic factors that likely cannot be replicated in the laboratory. Third, although ingestion of MPs is well documented in a diverse range of aquatic organisms, it is still unclear how the development of biofilm on MPs modifies their uptake and toxicity (e.g., transmission of pathogens and adsorbed pollutants). Advancing our understanding of i) the fundamental mechanisms in the formation of biofilm on MPs and their composition and ii) the changes in the environmental transport, fate, and toxicity of MPs by biofilm would be essential steps toward the development of relevant mitigation strategies to tackle MP pollution.

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Declarations

Competing interests The authors declare no competing or financial interests.

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