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Review Article Toxicity of micro/nanoplastics in the environment: Roles of plastisphere and eco-corona



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HIGHLIGHTS

Microplastics shape microbial commu-

- nity and metabolisms in the plastisphere. • The nano-bio interface mediates the
- formation of eco-corona.
- Eco-corona changes micro/nanoplastics' properties and increases environmental risks.
- Extrinsic and intrinsic factors contribute to environmental toxicity of micro/ nanoplastics.

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ABSTRACT

Micro/nanoplastics (MPs/NPs) are a growing threat to environmental health as these particles are dispersed to remote locations. However, the migration process of NPs differs from MPs due to their differences in sizes and physicochemical properties, thereby inducing different environmental behaviours and fates. While MPs provide surfaces to host microorganisms to form a plastisphere, NPs are smaller than microorganisms, which are often encapsulated by protein or organic matter to form unique eco-corona. Both plastisphere and eco-corona alter the physiochemical property of MPs/NPs, thereby changing their environmental toxicity. To fully understand the toxicity of MPs/NPs after forming plastisphere or eco-corona, this review aims to evaluate the roles and toxicities of MPs/NPs in the environment. Specifically, this review discusses the formation of plastisphere on MPs and eco-corona on NPs, summarizes the biochemical mechanisms of toxicity of MPs/NPs, and assesses their potential health threats to humans. Finally, perspectives are provided to better manage plastic pollution to protect the environment and human health.

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Abbreviations		NPs	Nanoplastics
		OTU	Operational taxonomic units
ARGs	Antibiotic resistance genes	PCB	Polychlorinated biphenyls
CAT	Catalase	PE	Polyethylene
c-di-GMP Bis (3'-5')-cyclic dimeric guanosine monophosphate		PET	Polyethylene terephthalate
DLVO theory Derjaguin–Landau–Verwey–Overbeek theory		PP	Polypropylene
DOM	Dissolved organic matter	PS	Polystyrene
EPS	extracellular polymeric substances	PVC	Polyvinyl chloride
GST	Glutathione S-transferase	RNA	Ribonucleic acid
HGT	Horizontal gene transfer	ROS	Reactive oxygen species
MPs	Microplastics	rRNA	Ribosomal RNA
MDA	Malondialdehyde	NADPH	Nicotinamide adenine dinucleotide phosphate
NOM	Natural organic matter		

1. Introduction

Plastics provide many benefits to modern society by bringing convenience to our daily lives. Compared to the production of 2 million tons in 1950, plastic production has risen to 359 million tons in 2018 (Wang et al., 2021b). However, over 40% of these plastics are being used only once, with 9–40% being recycled (Wright and Kelly, 2017). As a result, many spent-plastics are indiscriminately discharged into the environment. It is estimated that over 250 million tons will be accumulated in the environment by 2025 (Jambeck et al., 2015). Due to their environmental stability, plastic pollution is a considerable public-health threat.

Plastic particles are defined by their size as microplastics (MPs) (<5 mm) and nanoplastics (NPs) (<0.1–1 μ m) (Gigault et al., 2021; Amaral-Zettler et al., 2020). The polymer surface of plastic provides a durable substrate to allow microorganisms to attach, which can be transported over long distances (Wright et al., 2020). Recently, Zettler et al. (2013) introduced the term "plastisphere" to describe this mini ecosystem. Though the biomass of the plastisphere accounts for only <0.2% of the total biomass in the ocean, the plastisphere biomass might be considerable as <1% of plastics are detected in the marine environment (Van Sebille et al., 2015). The diverse microorganisms in the biofilm community of the plastisphere can alter their functions and metabolisms in the environment and potentially spread pathogenic bacteria to endanger public health. Therefore, it is crucial to understand the ecology of the plastisphere to better manage adverse ramifications of plastic pollution.

In comparison to MPs, NPs possess smaller dimensions and are unable to form biofilms with microorganisms. Despite making up a minimal portion of the total particle mass, which includes both plastics and their associated environmental corona, NPs possess unique biophysical and chemical properties that confer a higher level of toxicity when compared to MPs (Sharma et al., 2022; Wheeler et al., 2021). The interactions between NPs and environmental components, including protein and natural organic matter (NOM), have attracted much recent attention (Junaid and Wang, 2021). NOM can be adsorbed onto NPs in the environment to form a biomolecular-coated layer known as eco-corona (Kihara et al., 2020). The interactions between NPs and environmental components depend on both intrinsic (properties of NPs and environmental components) and extrinsic factors (pH, ionic strength and temperature) (Ali et al., 2022). The formation of eco-corona on NPs is driven by various forces, including ligand exchange, steric hindrance and hydrogen bonding (Philippe and Schaumann, 2014). The eco-corona can further regulate the colloidal behaviours of NPs via enhanced bridging behaviours by connecting particles via surface charge to form morphologically-different aggregates (Cao et al., 2022; Hakim and Kobayashi, 2021).

Both plastisphere and eco-corona can modify the physicochemical properties of MPs/NPs, thereby influencing their environmental behaviours and toxicity. For example, Nasser et al. (2020) reported that eco-corona can alter contaminant bioavailability and plastic sinking/floating behaviour. Additionally, eco-corona can also decrease the toxicity of polystyrene (PS) NPs by 15% to onion *Allium Cepa* due to lowered oxidative stress (Giri and Mukherjee, 2022). However, there is a paucity of comprehensive understanding concerning the overall effects of plastisphere and eco-corona on the toxicity of MPs/NPs.

Besides the effects of plastisphere and eco-corona layers on MPs/NPs, plastic particles themselves can induce adverse effects on organisms, including oxidative stress, reduced growth rate and inflammation (Prata et al., 2020). In addition, MPs/NPs may lead to unknown toxic effects due to the adsorbed toxic chemicals, including drugs and heavy metals (Wang et al., 2021). Enhanced antibiotic gene transfer due to the close contacts of microbial members in plastisphere might also cause a public threat to human health once they transferred into pathogenesis (Hodson et al., 2017; Wu et al., 2019).

Most reviews focus on the toxicity of MPs/NPs due to the inherent properties of the plastics instead of the interactive effects between particles and environmental components (Huang et al., 2021; Wang et al., 2021a). Considering that plastisphere and eco-corona can alter the bioavailability and environmental behaviour of MPs/NPs, it is important to understand their altered toxicity to reveal their environmental risks. To this end, the objectives of this review are to: 1) discuss the formation and composition of the plastisphere, 2) introduce eco-corona and its ecological features, 3) summarize the biochemical mechanisms of MPs/NPs toxicity, and 4) evaluate the potential health threats of MPs/NPs.

2. Plastisphere - a miniature biofilm ecosystem on microplastics

2.1. Plastisphere formation due to microbial colonization

MPs provide a substrate and unique niche for various microbial cultures to attach and colonize, forming biofilms in the environment. Such biofilms are termed as plastisphere, which consists of a complex community, including bacterial, archaeal, eukaryotic microorganisms and microscopic animals, thereby forming a new mini-ecosystem (Zettler et al., 2013). Similar to other biofilms, its development involves four stages, including initial attachment, irreversible attachment, biofilm maturation (including cluster and microcolony formation) and dispersion (Fig. 1) (Sauer et al., 2002), with corresponding unique protein production and gene expression in each stage (Petrova and Sauer, 2009; Sauer et al., 2022). For example, the second messenger bis (3'–5')-cyclic dimeric guanosine monophosphate (c-di-GMP) participates in biofilm creation. It has been observed that plastisphere has a higher concentration of c-di-GMP than that in its surroundings (13.0–19.0 μ g/g) (Su et al., 2022).

Upon interaction with the environment, MPs tend to adsorb organic and/or inorganic substances, forming a layer or film on their surface (Rummel et al., 2017), which creates a conducive environment for the survival and reproduction of microorganisms. The attachment process of bacteria is influenced by various factors such as hydrophobicity, particle shape, polymer type, and roughness (Du et al., 2022). Generally, the growth



Fig. 1. The formation of plastisphere on the microplastic surface.

differences of biofilms on different polymer types are the main cause of the floating/sinking behavior of microplastic-induced plastisphere (Liu et al., 2022a). However, it should be noted that this initial attachment is reversible, so bacteria are often observed returning to the surrounding environment. Over time, the surface hydrophobicity of MPs decreases (Tu et al., 2020) and the attachment of microorganisms can also modify the density of the plastic-biofilm particles (Liu et al., 2022a). As a result, this initial biofilm alters the vertical transport of MPs, creating a new ecological niche for other bacterial cells (Cai et al., 2019; Kooi et al., 2017).

Once cell clusters commence their development, the biofilm develops to the second stage, i.e., irreversible attachment. In this stage, a usual pattern is that multiple bacteria are in contact with one another, which are tightly attached to the microplastic surface. It is reported that irreversible attachment initiates a cascade of alteration in bacteria. Typical changes are cessation of flagella-mediated motility and initiation of some biosynthesis gene activation, including lipids and polysaccharides, to cement themselves to the surface or each other (Kumar et al., 2020; Rumbaugh and Sauer, 2020). In addition, biofilm development is associated with tolerance to antimicrobial agents (Gupta et al., 2013), such as β -lactamase and phenazine. The antimicrobial tolerance is recognized as a hallmark characteristic of biofilms. Further recruitment of bacteria on the biofilm to form a mature and three-dimensional structure (Lorite et al., 2011). There are a variety of well-established biofilm structures, including unstructured, overall flat, mushroom-like or pillar-like interspersed with fluid-filled channels (Wood et al., 2000). During this stage, the bacteria near the microplastic surface are gradually separated from the surroundings, including essential energy or nutrient sources. The bacteria in biofilms experience constantly changing conditions induced by cellular crowding and chemical gradients, ultimately resulting in stratification within the biofilm (Serra and Hengge, 2014). Therefore, microorganisms at different locations in the biofilms undergo concentration gradients of nutrients and oxygen, which shape the microbial structure and drive physiological differentiation within biofilms (Serra and Hengge, 2014).

Finally, bacteria can leave biofilms and return to the surroundings at the dispersion stage (Rumbaugh and Sauer, 2020). During the dispersion process, the solid and matrix-encapsulated biofilm cells escape from biofilms, thereby leaving biofilms with a central void (Petrova and Sauer, 2016; Steinberg et al., 2020). Not surprisingly, these dispersion events disseminate microorganisms and allows them to colonize at new locations. The driving forces of dispersion are chemical gradients in the biofilms, which were induced by increased nutritional competition as the biofilm grows in size (Heacock-Kang et al., 2017). In addition, various environmental factors influence the formation of plastisphere, such as salinity, nutrients and energy (Wright et al., 2020).

2.2. Microplastics shape a unique microbial community distinct from their surroundings

The microbial community in plastisphere is not significantly different from that formed on the surface of other materials, as the material surface only affects the initial attachment (Wright et al., 2020). Amaral-Zettler et al. (2020) revealed a high diversity of microorganisms in the early biofilm of plastisphere. The biofilms are aggregates of various microorganisms surrounded by extracellular polymeric substances (Flemming et al., 2016). Plastisphere is a micro-ecosystem consisting of primary producers, predators, symbionts and decomposers (Amaral-Zettler et al., 2020). The typical species within the plastisphere are summarised by Du et al. (2022). However, a considerable proportion of unique species are observed in the plastisphere compared with the surroundings (Li et al., 2021b). MPs can adsorb pollutants from their surrounding environment (Wang et al., 2020b). These chemicals, as well as additives in the plastics, act as selectors to shape the microbial structure (Li et al., 2021a). Moreover, the discharged MPs undergo different environments during the drifting process and carry microorganisms from the original environment, thereby leading to different microbial communities between the plastisphere and the environment. In this situation, it may be possible to trace the MPs due to their unique microbial footprints.

However, the microbial composition in both surroundings and plastisphere is changed with microbial colonization, which is different in their life cycle (Yang et al., 2020). In the soil environment, the microbial communities in plastisphere also exhibit significant differences from those in bulk soil (Huang et al., 2019), with the differences narrowing over time. For example, Puglisi et al. (2019) indicated that the plastisphere on aged-MPs showed a similar microbial structure to the surrounding landfill environment. Some external factors including soil properties and microplastic characteristics (e.g., types, size and hydrophobicity) can affect the plastisphere microbiome (Zhu et al., 2022). Compared to conventional plastics, biodegradable plastics (polylactic acid, polyhydroxybutyrate and polybutylene adipate terephthalate) can produce MPs in soil during their biodegradation process. Rüthi et al. (2020) found that MPs with high biodegradability harbour more unique species than non-biodegradability ones. In addition, the colonization process also depends on the chemical composition of MPs. For instance, the genus Alcanivorax prefers to attach to low-density polyethylene (PE) MPs because they can utilize the alkanes as a carbon source, while the carbonyl group of polyamide 6 is responsible for the high abundance of the genus Erythrobacter (Xie et al., 2021).

Apart from microplastic properties, some soil chemical characteristics can also influence the composition of microbial communities in plastisphere. Soil pH is central to nutrient bioavailability, enzyme activity and interactions between contaminants and MPs. Liu et al. (2021b) found that soil pH not only directly influenced the microbial community in plastisphere, but also indirectly affected it via changing the soil bacterial diversity. Moreover, soil pH can significantly influence the surface features especially the hydrophobicity, thereby affecting adsorption efficiency of MPs. For example, the concentration and speciation of arsenic on MPs varied with soil pH (Li et al., 2021c), which influence the microbial structure in plastisphere due to its selective pressure (Rousk et al., 2010). Given that MPs can also change soil properties, it is necessary to provide more information to evaluate the role of MPs in soils.

MPs provide a new niche by selectively harbouring microorganisms from the surrounding environment, which occurs as early as two days after plastics discharge into the environment. Pioneering bacteria are responsible for cell adhesion in the initial attachment stage. For example, Comamonadaceae, Gammaproteobacteria, and Alphaproteobacteria are the most widespread species found in this stage to form early biofilms since they can generate extracellular polymeric substances (EPS) to facilitate cell adhesion and successive colonizers (Andersson et al., 2009). In addition, Alteromonas, Thalassobius, Neptuniibacter and Poseobacter are also present in the early stage (Zhang et al., 2022). Further, the microbial diversity of plastisphere on MPs increases with exposure time (Yang et al., 2020), but with lower alpha diversity after long-term incubation (Sun et al., 2022). The underlying reason is that valuable metabolic products are continuously released and can be utilized by other community members (Polz and Cordero, 2016). This creates an interactive biofilm network and results in a high diversity and richness in microorganisms.

2.3. Unique functions and metabolisms of plastisphere in soils

The metabolic potential of microbial members within the plastisphere is not well understood. Studies have shown that microorganisms on MPs exhibit greater metabolic activity and possess distinct genetic profiles compared to those in the surrounding environment. For example, Bryant et al. (2016) and Zhu et al. (2005) found that a significant proportion of RNA reads on MPs were mapped to eukaryotic rRNAs, suggesting the presence of microbial eukaryotes in the plastisphere community. These microorganisms are thought to be involved in various metabolic processes, such as chemotaxis and adhesion, chemical metabolism, and polymer degradation (Amaral-Zettler et al., 2020).

The discrepancy in microbial communities between the plastisphere and surroundings also results in different functions and metabolisms, including elemental biogeochemical cycling, chemical compound degradation and pathogenic ability. Zhu et al. (2022) revealed that the soil plastisphere community possesses diverse metabolic pathways such as C, N and S metabolic pathways, which differ from those found in the soil environment. Stratification within the biofilm creates a micro-environment for oxygen and nutrient gradients during plastisphere development. Such conditions can lead to an anoxic condition in the interior spaces due to the gradient of dissolved oxygen (Virdis et al., 2011). This results in higher abundance of denitrifiers and denitrifying genes on MPs than those in the surroundings (Su et al., 2022). This difference also exists in the cycling of C and S via methylotrophy and methanol oxidation (Li et al., 2021b). Research indicates that the content of hydrophobic compounds in the MPs is 10⁶-times higher than that in the surrounding environment (Mato et al., 2001). In addition, PE and polypropylene (PP) release methane, ethane and ethylene (Royer et al., 2018). These compounds can lead to the clustering of specific microorganisms due to microbial migration towards hydrocarbons or chemotaxis (Pandey and Jain, 2002). This microbial chemotaxis plays a critical role in the initial attachment stage of plastisphere formation. These findings suggest that plastisphere communities take part in several important ecological processes in soils.

Compared with the soil environment, some specific metabolic genes in plastisphere are more abundant. For example, Li et al. (2023) found that human disease pathways (e.g., antibiotic resistance and infectious diseases) were overrepresented in the soil plastisphere. In this case, the soil plastisphere may pose higher risks to human health than the original environment. In addition, xenobiotic biodegradation and metabolism pathways associated with cofactors, amino acids and terpenoids are more activated in plastisphere (Miao et al., 2019). These pathways are responsible for some artificial substrates or related to organic degradation, indicating that microbial members might utilize MPs or additives as their C sources (e.g., enrichment of *anyA*, *nplT*, and endoglucanase-encoding gene), while N metabolism pathways are similar to biotransformation of phosphonate and some carbohydrates (Luo et al., 2022).

Further, MPs can act as "special microbial accumulators" to stimulate microbial growth related to their own degradation (Allen et al., 2019). An example of this metabolism is actinobacteria, which can decrease in the soil environment, but become enriched within the plastisphere after exposing to MPs (Huang et al., 2019; Yi et al., 2021). This selective enrichment is observed under nutrient-poor conditions, and some specific species related to hydrocarbon degradation such as Sphingomonadaceae are present (Oberbeckmann et al., 2018). Plastic polymers are energy-rich compounds, which could provide potential energy and substrate for microbial growth. Theoretically, PE contains -425 to -422 kJ/(mole O_2) useable energy, which is similar to glucose at -479 kJ/mole O2 (Oberbeckmann and Labrenz, 2020). The energy usage for microorganisms depends on their hydrolysability because plastics must be decomposed into small biomolecules before entering the cells (Gewert et al., 2015). Therefore, compared to some nonhydrolyzable plastics (e.g., PE and PP), hydrolyzable plastics can be more easily utilized by the cells through destroying the amide or ester bonds via hydrolysis or enzyme interactions. For example, hydrolases including lipases and cutinases are able to depolymerize polyethylene terephthalate (PET) (Gewert et al., 2015; Krueger et al., 2015). Similarly, the bacterium Ideonella sakaiensis isolated from a plastic recycling facility can degrade PET to monomers by hydrolases (Yoshida et al., 2016). Mature plastisphere harbours polymer degraders, but they live near the microplastic surface. Therefore, additional research is warranted on the inner biofilm communities to examine novel plastic-degrading organisms instead of the current focus on the upper biofilm.

In addition to influencing soil microbial metabolism, MPs also affect the plant microbiome through the formation of plastisphere. The activity of C and N related degradation enzymes is increased 2-10 times after adding poly (3-hydroxybutyrate-co-3-hydroxyvalerate) to soil (Zhou et al., 2021). MPs can also alter symbiotic relationship between plants and their roots. For example, polyethersulfone and PP MPs enhance root colonization by 1.4-8 times, while poly (ethylene terephthalate) decreases root colonization by 50% (de Souza Machado et al., 2019). These changes in microbial structure can influence the nitrogen cycle, particularly nitrogen fixation, in soil. Specifically, low dosages of MPs impact N fixation genes (nifD, nifH and nifK) in soil (Feng et al., 2022). However, a 7% concentration (w/w) of low-density PE increases gene nifH and other N fixation genus abundance (Fei et al., 2020; Rong et al., 2021). It is important to note that these studies were conducted under laboratory conditions and further research in field settings is needed to gain a more comprehensive understanding of the effects of MPs on soil nitrogen cycling processes.

3. Nano-bio interfaces mediate the formation of eco-corona

3.1. Interactions between nanoplastics and environmental components

In biomedical applications, bio–nano interfaces are formed by physicochemical interactions, kinetics, and thermodynamics when nanoparticles are in contact with biological systems, which results in the formation of protein coronas (Zhu et al., 2020). Similarly, organic matter in the environment can encapsulate nanoparticles to form an eco-corona layer (Fig. 2A) (Baalousha et al., 2018), which is recently converted from protein coronas (Wheeler et al., 2021). Different from protein coronas, eco-corona contains biomolecules including natural organic matter and pollutants instead of just proteins (Junaid and Wang, 2021). Specifically, eco-corona is formed on nanoparticles when biomolecules or metabolic products of surrounding microorganisms are exoproteomes (Nasser and Lynch, 2016), with bio-corona being attributed to adsorption of endogenous proteins (Canesi et al., 2017).

The interfaces between protein and NPs have been well documented, so protein corona might provide a reference to clarify the formation of



Fig. 2. (A) The formation of various corona layers via the interactions between nanoplastics and environmental chemicals (NOM, PPCPs and metabolites) or proteins. The Protein Data Bank (PDB) created with BioRender.com. (B) The proportion changes of the plastic core with decreasing the particle size to the nanoscale (Song et al., 2022). Copyright © 2022, American Chemical Society. (C) Eco-corona coated plastic particles changing their mechanical behaviour (Witzmann et al., 2022). Copyright © 2022, American Chemical Society.

eco-corona due to similar fundamental principles (Wheeler et al., 2021). Generally, various noncovalent interactions and forces are responsible for the interactions between proteins and NPs such as hydrophobic interactions, van der Waals forces, electrostatic forces and hydrogen bonding (Nel et al., 2009). During interactions, protein coating on the NPs may perform conformational changes, resulting in altered functions and/or avidity effects (Cabaleiro-Lago and Lundqvist, 2020; Yu et al., 2022).

The kinetics of corona formation on NPs are influenced by: 1) type and quantity of adsorbed biomolecules, 2) physicochemical properties of nanoplastic surface, and 3) binding affinity (Liu et al., 2022b). Generally, the binding affinity with NPs varies among components, with the affinity of proteins being lower than humic acids (Ramsperger et al., 2018). Consequently, the components with a high binding affinity strongly attach to nanoplastic surfaces to form a hard layer, whereas a soft layer can be further formed above the hard layer (Fig. 2A) (Pulido-Reyes et al., 2017). A hard layer generally undergoes higher conformational changes, slower exchange duration, stronger binding affinity and longer retention time than a soft layer (Yang et al., 2013), but they may occasionally resemble each other (Kihara et al., 2021). The formation of hard corona above NPs by reaction with human serum albumin has been reported in the human plasma system (Monopoli et al., 2012). However, identifying soft corona is a challenging aspect of future research because it is difficult to distinguish between unbound components and soft corona.

NPs can be heterografted with various natural components such as NOM, biomolecules, contaminants and clays (Gigault et al., 2021). Macromolecules with similar particle size to NPs influence the freedom degrees of macromolecules and further alter their efficiency of attachment. This indicates that macromolecule size effects endow NPs with unique environmental behaviours (Fig. 2C). Further, pH and ionic strength disproportionately affect the attachment efficiency of NPs. As mentioned above, MPs are large enough to provide attachment surfaces for microorganisms, which further develop into complex biofilms. Although the biomass of these organisms is high enough, the organisms still account for a small part of the overall mass (Isaacson et al., 2009). In contrast, the eco-colloidal layer can evolve to be a primary part of the overall particles due to the small scale of NPs. Research verified that the

proportion of NOM in the overall masses increases as plastic size decreases (Fig. 2B) (Gigault et al., 2021). Therefore, NPs harbour unique transport, uptake and accumulation pathways compared to MPs, with these characteristics highly dependent on eco-corona properties.

3.2. Eco-corona controls nanoplastic aggregation and deposition

When NPs interact with biomolecules, the biomolecules attach to the hydrophobic surfaces of NPs to form an eco-corona on the attached molecules (Zeng et al., 2019). These biomolecules are mainly produced from the metabolic activities of organisms, such as EPS or exoproteome (Nasser and Lynch, 2016). Hydrophobic and electrostatic interactions are the driving force to form eco-corona, which eventually alter the surface morphology (Rummel et al., 2021), charge (Saavedra et al., 2019), chemistry (Ramsperger et al., 2020) and potential mechanics of NPs (Hakim and Kobayashi, 2021). Among these interactions, the aggregation behaviour of NPs is the crucial change because it influences nanoplastic colloidal stability (Philippe and Schaumann, 2014).

Aggregation and deposition occur when the attractive van der Waals force outweighs opposing Coulomb forces between nanoplastic particles (Petosa et al., 2010). However, surface modifications mediated by eco-corona shift the balance between these two forces (Grasso et al., 2002). Various colloidal theories have been applied to describe the behaviours of nanoplastic particles (Nel et al., 2009). For example, attractive or repulsive interactions between nanoplastic particles can be predicted by Derjaguin–Landau–Verwey–Overbeek (DLVO) theory (Min et al., 2008). Based on the DLVO theory, the sum of van der Waals and electrical double–layer interactions determines the stability of nanoparticles (Petosa et al., 2010). Besides traditional DLVO theory, non-DLVO forces including steric interactions and hydration forces also influence the stability of nanoplastic-aggregates/deposits (Hu et al., 2010; Philippe and Schaumann, 2014).

Generally, NOM provides more negative charge to NPs because of its intrinsic negative charges. Therefore, this adsorbed-NOM can reduce Coulomb forces by neutralizing the positive charge to induce aggregation (Reynaud et al., 2022). On the contrary, nanoparticles are electrostatically stabilized if the initial charge is negative, or sufficient NOM reverses

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the surface charge (Borgnino, 2013). In terms of NOM with large molecules, the steric hindrance needs to be considered with electrostatic effects. For example, NPs covered by sodium alginate are stabilised by steric hindrance (Pradel et al., 2021).

Another mechanism of aggregation is molecular bridging, which is known as flocculation (Cao et al., 2022; Hakim and Kobayashi, 2021; Li et al., 2022; Rummel et al., 2021). Two dissolved organic matter (DOM) molecules can connect via hydrogen bonding based on observations from atomic force microscopy (Ghosh et al., 2008). However, cation bridging, especially calcium bridging, is more common and has been observed in various humic substances (Huangfu et al., 2013; Liu et al., 2011). However, the effectiveness of this bridging mostly depends on the content of carboxyl groups in the polymeric chains (Labille et al., 2005).

4. Biochemical mechanisms of micro/nanoplastic toxicity

4.1. Eco-corona relieves nanoplastic toxicity to biota

Monopoli et al. (2012) reported the formation of eco-corona by incubating nanoparticles in human blood. The proteins interact with nanoparticles to undergo a conformational change, thereby leading to the alteration of biological functions or even initiating an inflammatory signalling (Deng et al., 2011; Ding et al., 2013). Nanoplastic-induced complex formation with biological molecules in biological fluids has been demonstrated (Kihara et al., 2020). These results indicate that such corona confers a "self" identity to the NPs, thereby eliciting less of an immune response and consequently increasing the uptake of NPs. For example, eco-corona reduces the toxicity of PS-NPs to onion Allium Cepa due to lower oxidative stress (Giri and Mukherjee, 2022). In addition, the serum corona coated on the NPs takes part in the interaction with cells (Kik et al., 2020), thereby influencing their cellular uptake. In this situation, NPs carrying contaminants can easily contact cells within living organisms. However, the interactions of NPs with a biological surface (cell membrane or cell wall) and how NPs enter bacterial or living organism cells are complicated. Considering the diversity of lipopolysaccharides in cell membranes, their contact modes vary with the composition of bacterial surfaces (Fig. 3) (Pulido-Reves et al., 2017).

After forming an eco-corona, the influence of NPs on cellar responses of organisms are different due to its new biological identity, which relies on the type of adsorbed biomolecules and the suspended medium. The



Fig. 3. The pathway for entering cell induced by corona coated nanoparticles (Zhu et al., 2013a). Copyright © 2012 American Chemical Society.

biological identity of NPs is attributed to the interactions between biomolecular corona and biological systems. This corona is conveyed when NPs move from one biological environment to another (Docter et al., 2015). However, NPs can retain a fingerprint that indicates its history in the final corona (Wheeler et al., 2021). Given that bacterial cells continuously excrete metabolites to the environment, this change in microenvironment might also alter corona components. For example, a previous investigation observed the aggregation of nanoparticles and changes in protein corona due to secretion of different molecules, thereby influencing the uptake rate by human and carcinoma cells (Albanese et al., 2014).

Proteins or biomolecules in the environment can encapsulate NPs, which further alter their physicochemical properties, bioreactivity and transport due to formation of an eco-corona (Pulido-Reyes et al., 2017). NPs in the environment present unique reactivity and bioavailability, which further affect their environmental fate and behaviours, resulting in toxic effects on various organisms (Table 1). The mitigated toxicity is observed after exposure to nano-PS owing to the buffering effects of the eco-corona, which hinders nanoplastic surface reactivity by providing a barrier to prevent direct contact between NPs and bacteria cells (Fadare et al., 2020). Most research has focused on toxicity changes induced by eco-corona development. A study compared the toxicity of PS NPs with different surface charges (e.g., plain, aminated, and carboxylate) and nanoplastic concentration on the toxicity of PS to the condiment plant Allium cepa. The results showed reduced toxicity of eco-corona coated NPs due to agglomeration effects (Giri and Mukherjee, 2022). Similarly, another study highlights the eco-corona of plastic debris in soil, which can retain humic-like compounds, thereby elevating the overall bacterial abundance and microbial metabolic potential (Liu et al., 2022c). These results indicate that the eco-corona reduces the oxidative stress and subsequent toxic effects of NPs in soil. Albeit NOM decreases the toxicity of NPs, some endocrine-related genes exhibit elevated expressions, such as glutathione S-transferase (GST) and catalase (CAT) (Fadare et al., 2020). However, some studies show adverse effects (i.e., eco-corona increasing the toxicity of NPs), including growth inhibition, altered feeding behaviour and increased oxidative stress (Nasser and Lynch, 2016; Shiu et al., 2020).

In sum, the presence of eco-corona on the surface of NPs, has been found to reduce the toxicity of NPs in most studies. This reduction in toxicity is primarily attributed to the decreased reactive oxygen species (ROS) production through a barrier process, which is induced by adsorbed biomolecules on NPs. However, some studies have observed increased toxicity with the presence of an eco-corona, highlighting the need for further research to fully understand the factors that regulate the toxicity of

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NPs in the environment. Additional research is needed to determine the impact of adsorbed protein and EPS on NPs and their effect on reactivity and fate, as well as ingestion and organism toxicity. Additionally, while previous research has primarily focused on the biomedical applications of eco-corona, it is important to investigate its development in terrestrial environments to better understand the biological impacts of NPs over time.

4.2. Micro/nanoplastics induce oxidative stress of bacteria and living organisms

MPs/NPs induce genomic instability and mutation via production of ROS (Fig. 4) (Alimba and Faggio, 2019). Microplastic-induced ROS can come from three sources: 1) produced by MPs/NPs during the aging process, 2) bacterial responses after contact with additives and adsorbed chemicals on MPs/NPs, and 3) oxidative stress due to MPs/NPs (Dev et al., 2022; Shi et al., 2022a). Increased free radicals and a lack of corresponding protection lead to oxidative stress in bacteria, mainly due to reduced antioxidant enzymes (Sangkham et al., 2022). The histological changes and induction of oxidative stress occur when microorganisms are exposed to MPs/NPs. In this situation, cytotoxicity may be an important mechanism inducing a toxic response. Furthermore, short-term exposure to MPs may cause a decrease in the activity of antioxidant enzymes because of the energy consumed in combating oxidative stress (Hamed et al., 2020), whereas long-term experiments observed lipid peroxidation indicating an accumulative effect (Varó et al., 2019). The specific oxidative stress pattern induced by MPs/NPs is difficult to speculate, but its roles in affecting oxidative homeostasis are well-documented based on cellular research (Zheng et al., 2019).

Oxidative stress has been observed in various organisms, including earthworms (Jiang et al., 2020), nematodes (Yu et al., 2020) and bacteria cells (Zhang et al., 2022). Changes in their antioxidant systems reveal oxidative stress induced by MPs/NPs. For example, the activity of CAT and malondialdehyde (MDA) increased by 120–270% in earthworms (*Eisenia fetida*) after exposure to low-density PE MPs (0.1–1.5 g/kg) (Chen et al., 2020). Apart from oxidative stress, MPs can cause cell toxicity due to the microplastic itself or adsorbed pollutants (Barboza et al., 2018; Rochman et al., 2014). These chemicals should be eliminated to prevent accumulation in the cells of living organisms. Two phases are responsible for their hazardous metabolisms. Firstly, oxygen atoms are added to these toxic compounds by the cytochrome P450 system. Then, the oxygen-added materials are converted into more hydrophilic compounds by conjugating with glutathione, glucuronic acid, sulphate and other endogenous substances (Falfushynska et al., 2019). GST is the main

Table 1

Toxicity	changes	due to	o the	formation	of	eco-corona.
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Type of plastics	Test Organism	Concentration	Time	Role of eco-corona	Reference				
PE	Oryza sativa	0–6.0 g/kg	3 months	Increasing operational taxonomic units (OUT), decreasing diversity and evenness, reducing the biomass and production of rice	Liu et al. (2022b)				
Amidine-PS and carboxyl-PS	Daphnia magna, Brachionus calyciflorus, and Thamnocephalus platyurus	10-400 mg/L	NR	Significantly reducing their acute toxicity	Saavedra et al. (2019)				
PS	Solanum lycopersicum	5 mg/L	0–72 h	Enhancing plant growth, seed germination, and chlorophyll content	Lakshmikanthan and Chandrasekaran (2022)				
PS	Allium cepa	12.5–50 mg/L	96 h	Stimulating agglomerate of nanoplastics to reduce uptake, decreasing the oxidative stress	Giri and Mukherjee (2022)				
PS	Chlorella sp.	100 mg/L	12–48 h	Decreasing generation of ROS	Natarajan et al. (2020)				
PS	Chlorella vulgaris	25-400 mg/L	72 h	Decreasing toxicity	Hanachi et al. (2022)				
PS	Dunaliella tertiolecta, Thalassiosira pseudonana, Skeletonema grethae, and Phaeodactylum tricornutum	10 ⁻⁴ -250 mg/ L	48 h	Reducing survival of phytoplankton	Shiu et al. (2020)				
PS	Crassostrea gigas	0.1–100 mg/L	1–5 h	Increasing production of ROS	González-Fernández et al. (2018)				
PS	Mytilus galloprovincialis	1–50 mg/L	1 h	Decreasing toxicity	Canesi et al. (2017)				

Note: NR, not reported; PE, polyethylene; PS, polystyrene.



Fig. 4. The diagram of cytotoxicity induced by micro/nanoplastics. The micro/nanoplastics can be endocytosed into lysosomes and then induce lysosomal membrane permeabilization resulting in cathepsin release (Wang et al., 2018). The released plastic particles and cathepsins lead to caspase-independent cell death and damage mitochondrial to generate ROS and further induce DNA damage (Yong et al., 2020).

enzyme in detoxifying these chemicals, especially these electrophilic xenobiotics (Kim and Kang, 2015), which plays an important role in the second phase of detoxification in plants. Pflugmacher et al. (2021) evaluated the phytotoxicity of new and aged polycarbonate MPs to garden cress *Lepidium sativum*. Results showed that the toxicity of polycarbonate decreases with increasing aging time due to a reduction of leached toxic chemicals.

The toxicity of MPs/NPs has a strong dependence on particle size. The size of plastics is a crucial factor because smaller size facilitates internalization and enhances accumulation in the cells of living organisms. Furthermore, a higher specific surface area means a greater adsorption ability for contaminants. For example, NPs with a 20 nm size induce more transgenerational toxicity than those with a 100 nm size by producing more ROS in the nematode *Caenorhabditis elegans* (Liu et al., 2021a). However, Le et al. (2018) found that MPs with a 1.0 µm size exhibited the highest toxicity to the nematode *C. elegans* due to oxidative stress, but less toxicity was observed when reducing the size to 0.1 µm. Therefore, the toxicity of MPs/NPs to biota is highly dependent on the size of the plastics and organisms.

Overall, MPs/NPs with a high surface area lead to oxidative stress due to ROS production induced by adsorbed chemicals, such as bisphenol-A (Wei et al., 2019) and phthalates (Zhang et al., 2020), or inflammatory reactions. In addition, exposure to MPs/NPs disturbs glutathione and its dependent response cycles, which are responsible for antioxidant responses in organisms, thereby disturbing antioxidant balance and causing oxidative damage.

5. Threats of micro/nanoplastics to environmental health

5.1. Release of microplastic leachates in terrestrial ecosystems

Generally, plastic products contain different types of chemical additives, including plasticizers, fillers and flame retardants (Kwon et al., 2017). The weight fraction of additives breaks down as ~70% plasticizers, 25% flame retardants and 0.1-3% stabilizers (Hahladakis et al., 2018). Micro/nanoplastic additives can easily penetrate cell membranes and induce toxic effects on bacteria and plant cells due to the lipophilic characteristics of most plastic additives, ultimately bioaccumulating along the food chain. The release of additives from MPs mainly relies on microplastic properties, including the microplastic composition and leachate types, microplastic size, and external conditions (Shen et al., 2019; Sheng et al., 2021). It was reported that lactic acid released during the aging process of poly (lactic acid) inhibited the germination and growth of Lolium perenne (perennial ryegrass) (Boots et al., 2019). Another study found that benzothiazole released from aged shoe-sole fragments have a negative effect on plant growth and photosynthesis in mung bean (Vigna radiata), while similar fragments without benzothiazole had less inhibition (Lee et al., 2022). In aquatic environments, exposure to bisphenol A for 60 days significantly changed the sex ratio of medaka (Oryzias latipes) indicating endocrine disruption (Yokota et al., 2000). Similarly, it was reported that long-term exposure to microplastic leachates, such as bisphenol A, led to serious harm on the soil microbiome, especially their enzymatic activity (Zaborowska et al., 2022).

The toxicity of plastics contains two primary aspects, the plastic itself and the release of toxic chemicals (Wright et al., 2013). Such additives are easily released into the surrounding environment to induce toxicity because most additives are physically rather than chemically bound to the polymer (Hamlin et al., 2015). The discrepancy of biological responses to complicated chemicals in the leachate varies with chemical additives. For example, polyvinyl chloride (PVC) is the most toxic polymer to barnacle larvae (Amphibalanus amphitrite) compared to other recyclable polymer types (Li et al., 2016). These studies suggested that plastic leachates, especially PVC, led to organism toxicity, while such adverse effects were not attributable to specific chemical compounds in the environment. Additives of particular concern are brominated flame-retardants, bisphenol A and phthalates (Banerjee and Shelver, 2021; Campanale et al., 2020; Toni et al., 2017). Moreover, surfactants can be disruptive to cellular surface structures, such as proteoglycans, and hinder cellular signalling processes at a moderate level, whereas a high level of these chemicals is able to lyse the lipid bilayer of the plasma membrane of human cells (Yong et al., 2020). Therefore, further research is necessary on microplastic leachates to better inform and manage plastic pollution and potential toxicities.

5.2. Micro/nanoplastics as vectors of pollutants and pathogenic bacteria in soils

The hydrophobic characteristics and high specific surface area endow MPs/NPs to accumulate organic pollutants, such as polyaromatic hydrocarbons and pharmaceuticals (Li et al., 2018; Wang et al., 2021c). These microplastic-borne pollutants can be transported across great distances, thereby contaminating multiple environments and exposing biota across several trophic levels. Desorption of contaminants from transported MPs may harm organisms in surrounding ecosystems. For instance, Ma et al. (2020) found that the combination of MPs with tetracycline enhanced the development of antibiotic resistance in the *Enchytraeus crypticus* (Oligochaeta). Similarly, Wang et al. (2020a) investigated the toxicity of MPs adsorbed with polychlorinated biphenyls (PCB) and observed a high bioaccumulation of PCB in the earthworm *E. fetida*.

Adsorbed nutrients from the environment can attract microorganisms and viruses to adhere to their surface (Frère et al., 2018). Many MPs have low density prior to pollutant contamination, allowing for greater transport through winds or bioturbation (Rezaei et al., 2019; Zhu et al., 2018), with terrestrial environment being a major sink of MPs and pathogenic microorganisms (Van Wijnen et al., 2019). Given MPs originating from soil often carry pathogenic species to downstream receiving environments, MPs can serve as vectors for transport of pathogenic microorganisms. For example, Gkoutselis et al. (2021) found that MPs acted as microhabitats, which not only draw in specific fungal communities but also gather certain pathogens that are harmful to humans, such as Cryptococcus and Phoma-like species. As the transport function of MPs in aquatic environment has been well documented, it still unknown whether the plastisphere is maintained and can function in the sharply contrasting conditions of soil environment. What we know is that the geographic location and environmental conditions play an important role in shaping the microbial community of plastisphere. This means that such communities might change with changing environmental conditions (Oberbeckmann and Labrenz, 2020). While, recent studies report that potential pathogens colonise in MPs, additionally research is required to address the fate and transport of microplastic-attached pathogenic microorganisms under real-world environmental conditions.

5.3. Enhancing the spread of antibiotic resistance genes in soils

MPs and antibiotic resistance genes (ARGs) are widely present in soils (Wang et al., 2021c) and it is posited that the distribution pattern of ARGs is strongly influenced by microplastic exposure. The plastisphere on MPs contains a myriad of antibiotic-resistant bacteria, indicating its role as an important sink of ARGs (Sills et al., 2020). For example, Yang et al. (2022) found that the total abundance of ARGs in the plastisphere is increased by 0.26–1.4 times compared with surrounding soil. ARGs in plastisphere vary pending on microplastic types and sizes. Large MPs harbours higher ARG abundance than smaller ones, being 98–154% greater (Lu et al., 2020). This is ascribed to larger MPs containing more antibiotics and heavy metals, thereby providing more selective stress



Fig. 5. Microplastics and nanoplastics induced HGT of ARGs including conjugation and transformation.

(Zhu et al., 2013b). In addition, the selective enrichment efficiency of ARGs on MPs is influenced by the aging process. ARG abundance on weathered-MPs exceeded that of less weathered-MPs by 45.7% (Lu et al., 2020). These results indicate that MPs may be hazardous as they serve as a hotspot to spread ARGs.

Research shows the promotional effects of MPs on ARG spread, with Fig. 5 illustrating the detailed mechanisms. Specifically, the enhanced spread of ARGs is attributed to: 1) facilitated growth of antibiotic resistance bacteria, 2) changed microbial structure of ecosystems, 3) enhanced horizontal gene transfer of ARGs due to increased ROS production and high-density microorganisms, and 4) adsorbed pollutants (heavy metals and drugs), which are able to provide selective pressure for microbes in the plastisphere. MPs increase the abundance of complete ammonia oxidation (Comammox) Nitrospira inopinata and the diversity of microbial communities in sludge systems (Dai et al., 2020), which can act as a host of antibiotic resistance. Added poly (vinyl alcohol) was shown to promote the proliferation of antibiotic resistance bacteria (Wang et al., 2021c). Further, the enrichment of microbial members is generally accompanied by the growth of bacteria carrying ARGs, thereby increasing the abundance of ARGs (Aminov, 2011; Oiu et al., 2012; Sills et al., 2020).

The high density of microorganisms in the plastisphere enhances bacterial conjugation and ultimately accelerates gene transfer of antibiotic resistance (Amaral-Zettler et al., 2020). MPs provide a persistent surface not only for microorganism attachment but also for developing intense interactions to gene exchange (i.e., horizontal gene transfer, HGT). The HGT of ARGs is mainly due to the transport of mobile gene elements to bacterial recipients. Among various mobile gene elements, integrons and plasmids are recognized as indicators of HGT (Ma et al., 2011). In landfill leachate, class 1 and 2 integron-integrase genes (*intl1* and *intl2*) are associated with most of the ARGs, with all ARGs being correlated with mobile gene elements in long-term experiments (Shi et al., 2020).

Increased cell permeability induced by ROS production and leached chemicals is also responsible for enhanced HGT of ARGs. ROS production is a complicated process, which is produced either from the interaction between the microplastic and the receptors on the surface of the cell membrane or via nicotinamide adenine dinucleotide phosphate (NADPH) oxidase generated by the endocytosis process of MPs (Banerjee and Shelver, 2021). Moreover, some studies observed high levels of ROS due to the mitochondria response induced by internalized MPs (Wang et al., 2018). The promoted ROS production is the most important factor for enhancing conjugation by microplastic exposure. ROS production is size-dependent and is associated with HGT efficiency. However, a high concentration of ROS may induce cell death, thereby inhibiting HGT of ARGs. For example, the transfer efficiency of ARGs increased by 2.5-fold at a 10 nm microplastic dosage at 10 mg/L, whereas it is decreased by 2-fold when microplastic dosage increased to 100 mg/L compared to control groups (Zha et al., 2022).

Overall, MPs accelerate the spread of ARGs, indicating an overlapped transmission route of ARGs with MPs (Shi et al., 2022b). In this case, pathogens living in the MPs are able to acquire ARGs and can ultimately reach the human body via ingestion. Therefore, public health actions are warranted to alleviate potential human-health risks posed by microplastic-transported ARGs.

6. Conclusions and perspectives

MPs provide a surface for microbial colonization. Similar to biofilms developing on other surfaces, the formation of the plastisphere involves four stages: initial attachment, irreversible attachment, biofilm maturation and dispersion. Both microbial composition and functional metabolisms are changed along the progression of colonization, and they differ throughout their entire life cycle. Current research indicates that the plastisphere differs from the surrounding community, while it is similar to that formed on other inert surfaces (Li et al., 2021b). The community

differences are attributed to 1) MPs releasing toxic chemicals to select the growth of specific microorganisms, and 2) the initial colonization of microorganisms. Functional metabolism research demonstrates that the microorganisms within the plastisphere are more metabolically active and diverse (Bryan et al., 2016), including chemotaxis and adhesion, chemical metabolism and polymer degradation.

In contrast, NPs can be encapsulated by an eco-corona comprised of proteins, metabolic molecules of microorganisms, DNA and allochthonous compounds. The formation of eco-corona depends on the nano-bio interface and various noncovalent interactions and forces (e.g., hydrophobic interactions, van der Waals forces electrostatic forces and hydrogen bonding) that are responsible for the interaction between biomolecular compounds and NPs. The eco-corona not only alters the environmental behaviour (especially the aggregation and deposition) of NPs, but also endows NPs with different biophysicochemical properties.

The biochemical toxicity induced by MPs/NPs involves three aspects: 1) the release of toxic additives, 2) inducing oxidative stress and cellular toxicity, and 3) property alterations by eco-corona changing its toxicity. MPs/NPs are a passive dosing source for toxic additives due to their ubiquitous occurrence in plastics and weak binding features. Such chemicals can be released during the lifetime of MPs/NPs in the environment, thereby resulting in persistent adverse impacts on the environment. The oxidative stress is mainly attributed to enhanced ROS production originating during the polymer aging process, and the bacteria response due to their contact with molecular or microplastic toxicity compounds. The eco-corona composition reflects the location and history of its formation, which provides the possibility of modelling the nano-plastic transport pathway and their environmental fate.

MPs/NPs with high sorption ability and rich attachment sites for microorganisms make them potential vectors for transport of harmful pollutants and bacterial pathogens in the environment. The sorption behaviours of MPs/NPs depend on the type of plastics and pollutants, biofilm compositions and environmental conditions. Some pathogenic species proliferate in the plastisphere, which warrants more attention because of its public health threats. Further, the close proximity and density of microbial communities in the plastisphere enhance ARG transfer among microorganisms. Once transferred to pathogens, it threatens human health consequences from bacterial treatment.

Several research gaps were identified regarding the adverse effects of MPs/NPs:

- Given the stable and diverse community structure, it is recommended to further elucidate the role of inter-kingdom interactions among various microbial organisms in the plastisphere.
- 2) The eco-corona and plastisphere layers impart highly altered physiochemical properties to MPs/NPs. However, current toxicity studies mostly focus on virgin MPs/NPs, which results in limited information on their environmental risk and ecological behaviours. Therefore, the role of eco-corona and plastisphere layers in subsequent studies should become a prerequisite for toxicity research.
- 3) Additional research is needed to evaluate the effects of environmental factors on the formation of eco-corona and plastisphere layers.
- 4) Future research should pay more attention to the differences in toxicity between MPs and NPs, and the effects of environmental weathering on toxicity metrics.
- 5) Currently, toxicity research has mainly focused on PS. However, given the different functional groups present in various types of plastics, it is important to gain a more comprehensive understanding of the changes in toxicity that occur after the formation of eco-corona by other types of NPs.
- 6) The roles of MPs/NPs in transporting pollutants and pathogenic bacteria is best understood for aquatic systems, but few studies concentrate on soils. Considering the diversity of organisms in soils, rigorous studies of soil systems are required to reveal how micro/ nanoplastic transport in the soil environment ultimately influences soil, environmental and human health.

- 7) Although the vector role of MPs/NPs is well documented, it still lacks direct evidence and specific health implication. More efforts are required to clarify whether plastic pollution is a "potential risk" or a "real threat" to soil, environmental and human health.
- 8) Given that some chemicals can enhance the spread of ARGs, there is a lack of information about the effects of additives and leachates from MPs on the dissemination of ARGs in soils and the larger environment.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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