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RESEARCH ARTICLE

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Review on the ecotoxicological impacts of plastic pollution on the freshwater invertebrate Daphnia

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Abstract

The environmental impacts of plastic pollution have recently attracted universal attention, especially in the aquatic environment. However, research has mostly been focused on marine ecosystems, even though freshwater ecosystems are equally if not more polluted by plastics. In addition, the mechanism and extent to which plastic pollution affects aquatic biota and the rates of transfer to organisms through food webs eventually reaching humans are poorly understood, especially considering leaching hazardous chemicals. Several studies have demonstrated extreme toxicity in freshwater organisms such Daphnia. When such keystone species are affected by ambient pollution, entire food webs are destabilized and biodiversity is threatened. The unremitting increase in plastic contaminants in freshwater environments would cause impairments in ecosystem functions and structure, leading to various kinds of negative ecological consequences. As various studies have reported the effects on daphnids, a consolidation of this literature is critical to discuss the limitations and knowledge gaps and to evaluate the risk posed to the aquatic environment. This review was undertaken due to the evident need to evaluate this threat. The aims were to provide a meaningful overview of the literature relevant to the potential impact of plastic pollution and associated contaminants on freshwater daphnids as primary consumers. A critical evaluation of research gaps and perspectives is conducted to provide a comprehensive risk assessment of microplastic as a hazard to aquatic environments. We outlined the challenges and limitations to microplastic research in hampering better-focused investigations that could support the development of new plastic materials and/or establishment of new regulations.

KEYWORDS

ecosystem, freshwater Daphnia, microplastics, nanoplastics, pollution

1 | INTRODUCTION

The initial studies regarding the possible ecotoxicological impact of microplastics (MPs) on freshwater organisms were performed in the latter half of the 2000s.[1] There has been growing societal and scientific concern about the effects of plastic particles on marine and freshwater organisms, and currently, this area is one of the most intensely researched environmental topics.[2,3] For decades, plastic pollution has been part of the freshwater environment, and it is expected to expand exponentially in the coming years.[4] MPs have been discovered in isolated and protected areas and found in all freshwater ecosystems.[5] MP exposure has a variety of negative consequences on freshwater biota, from primary consumers such as members of the genus Daphnia (daphnids) to top predators and even humans.[2,6–8]

Plastics were developed over 200 years ago—thus, before the twentieth century—by using natural compounds such as tree-sap latex, insect secretion shellac, celluloids, and rubber.[9] Scientific and technological advancements regarding new synthetic substances have greatly increased the production of plastics, making them an important commodity in the near term.[10] Plastics are versatile polymers with multiple applications and have become essential in human lives. Indeed, between 1950 and 2015, an estimated 8.3 billion tons of plastic were manufactured.[11,12] The demand for plastics continues to increase due to their inherit versatility and resistant properties; for example, plastics can be strong, lightweight, and corrosion and heat resistant. With the global population expected to increase to 9.7 billion by 2050 and the likely increase in affluence in developing nations, the demand for plastics will undeniably rise as well. In 2019, the annual universal manufacturing of plastics reached 370 million tons.[13] The one-time-use approach towards plastic items—together with the current levels of production, improper disposal, poor waste management, and low recovery rate—leads to hazardous plastic waste being thrown out into the environment, thus contributing to the great risks posed to ecosystems.[14–16]

Apart from the intentional production of MPs for use in personal care products, plastic, under environmental stimuli and natural conditions, such as physical abrasion and ultraviolet lights, degrades to smaller fragments, namely MP and, eventually, nanoplastic (NP) particles.[17] Plastic degradation occurs via the following mechanisms; photo-oxidation due to ultraviolet (UV) light exposure, thermal degradation due to heat, hydrolysis of ester bonds, and microbial degradation.[18,19] During the manufacturing process, some toxic residual monomers remain unpolymerized within the plastics. As the plastics decompose, the residual monomers are released into the environment.[20] Phthalates, benzene, bisphenol A and phenol, among other toxicants, have been documented to be released by ester bond hydrolysis.[21] Of particular concern are MPs and NPs identified in almost all ecosystems across the world.[22,23] Zhang et al.[24] noted that MP pollution is widespread in marine, freshwater, terrestrial, and increasingly atmospheric environments. With a diverse network of source-pathway-sink linkages, these habitats are interconnected, a factor that could impact the flow and retention of MPs in environmental matrices.[24]

The size definitions of MPs and NPs differ among publications; however, the most commonly used definition for MPs is 1–5 mm, while NPs are often defined as <20 nm or those between 1 and 100 nm (the latter according to strict nanomaterials definitions).[25] Due to their small size, MP and NP particles are progressively ingested by organisms at a variety of trophic levels and developmental stages, particularly at the lower end of the food chain; therefore, they are transferred to top consumers through feeding.[26,27] Various additives, such as primary polymers as well as a few other chemicals, are used in plastic products to improve the properties and persistence of plastics.[28] These additives consist of inorganic materials such as silica and carbon, which contribute to flexibility and stability.[15,29] The list of chemical additives and their potential leachates is presented in Table 1.[29,30] Polyethylene (PE), polypropylene (PP), polystyrene (PS), polyethylene terephthalate (PET), polyvinyl chloride (PVC), and polyamide (PA) are the most prevalent polymers.[31] PE and PP float in water due to their low density, while higher density polymers, such as PS, PVC, PA, and PET, are deposited via inclination through the water column. Consequently, MP contaminants are found in every layer of aquatic ecosystems.[32]

Until now, ecotoxicological research on MPs has primarily used marine animals (77%) rather than freshwater organisms (23%).[2] Many studies addressing the ecotoxicity of plastic pollution have focused on marine environments (Figure 1). However, recent studies have revealed that freshwater ecosystems are also subject to plastic pollution.[33–35] These plastics impact freshwater species and the ecosystem as well as food meant for human consumption, posing a potential food safety risk. The abundant distribution of MPs and NPs in freshwater results in a wide spectrum of aquatic biota being exposed to them, typically through ingestion, which has lasting effects throughout the food cycle, even without direct exposure.[36] In addition to the risk of negative consequences from plastic ingestion, toxic reactions and responses could result from other contaminants released from plastics.[37] Anthropogenic sources are continuously and unprecedentedly accumulating plastic pollutants in every aquatic environment and cause ecological processes and structures to be disrupted both directly and indirectly. Aquatic ecosystems are interconnected with the terrestrial environment; consequently, changes in one system impact the other.[38]

Studies have indicated that plastic particle uptake is a common phenomenon among marine zooplankton of different taxa.[28,39] For example, fluorescent carboxylated polystyrene MPs (polymethyl methacrylate) of 20 nm–1 mm were detected in the gut of freshwater zooplanktons.[40,41] The ingestion of plastic particles, especially MPs and NPs, has the potential to impair the behavior and metabolism of aquatic fauna and induce oxidative stress, tissue damage, and death.[42–44] Assessing the impact of plastic pollution in aquatic ecosystems is quite challenging. Despite the ubiquity of freshwater plastic pollution and associated chemicals, few studies have investigated the toxicological effects of macroplastics (>5 mm) or MPs in freshwater ecosystems.[34] Freshwater daphnids have been used as a study species for plastic pollution in various ecotoxicological investigations.[45–48] Daphnids, especially *Daphnia magna*, are a typical and standard

TABLE 1 The types of plastics and common chemical additives in use and their potential leachates.

Additive type	Example substance	In which plastics is it used?
Plasticizers	Short, medium, and long chain chlorinate paraffins. Phthalates: Bis (2-ethylhexyl)phthalate (DEHP), dibutylphthalate (DBP), dipehnylphthalate (DPP). Adipates: diheptyl adipate (DHA), heptyl adipate (HAD), heptyl octyl adipate (HOA).	Mostly used in PVC and cellulose based polymers where they can make up to 75% wt/wt of the final product.
Flame retardants	Brominated flame retardants; polybrominated diphenylethers (PBDEs),decabromodiphenylethane. Phosphorous flame retardants; tris(2-chloroethyl)phosphate (TCEP), tris(2-chlorisopropyl) phosphate (TCPP).	Brominated compounds can reach 25% wt/wt of the final polymer.
Stabilizers, ultraviolet stabilizers, antioxidants	Bisphenol A (BPA), cadmium and lead compounds nonylphenols, octylphenols butylated hydroxytoluene	Up to 3% wt/wt; phenolics generally added at lower amounts.
Slip agents	Fatty acid amides fatty acid esters zinc stearate	Added at up to 3% wt/wt depending on the polymer type
Biocides	Organotins, arsenic compounds triclosan	Added primarily to soft PVC and polyurethane foams
Inorganic pigments	Cadmium, chromium and lead compounds, zinc oxide, iron oxide titanium dioxide lead carbonate aluminium and copper powders	Non-fluorescing substances show lower migration rates.
Organic pigments	Cobalt(II) diacetate	Insoluble, low migration tendencies.
Fillers	Calcium carbonate, zinc oxide barium sulphate glass microspheres nanomaterials clays	Can make up to 50% wt/wt of the polymers

Abbreviations: PVC, polyvinylchloride; wt/wt, weight to weight.

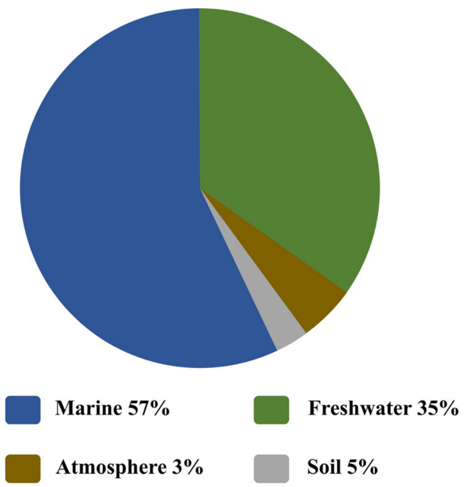

FIGURE 1 Distribution of publications related to microplastics in the Web of Science database in different ecological systems (retrieved in 2021).

ecotoxicity model and show high sensitivity to toxicants.[49] They are filter feeders located on the lower end of the food chain, and they are essential components of freshwater food webs as well as food sources for many aquatic organisms.[50] Daphnia, commonly known as the water flea (Crustacea; Branchiopoda), are planktonic filter-feeder crustaceans that belong to Phyllopoda.[51] Daphnia belongs to the Cladocera family of branchiopods, whose bodies are encased in an uncalcified shell known as the carapace.[50,52] Cladocera range in size from 0.5 to 5.0 mm in length; however, members from one genus, Leptodora, reach up to 18 mm in length.[53,54] Males have larger antennules, a modified post-abdomen and first legs with a clasping hook, allowing them to be distinguished from females.[50] The genus Daphnia contains more than 100 species of freshwater plankton that can be found all over the world.[50,55] The species can be found in a wide range of freshwater habitats, from small ephemeral pools to large lakes and seasonally flooded depressions, except for extreme habitats such as hot springs, summer droughts or severe winters. Extreme environmental conditions alter the daphnid life cycle.[52] In lakes and ponds, they are the dominant zooplankton and an important element of the food chain. Daphnids are the most common food of planktivorous fish in many lakes, although this dominance may vary from time to time.[50] The ecology of daphnids has been investigated in terms of their role as a primary consumer in aquatic food chains, toxicity, phenotypic plasticity and behavior and the evolution of sexual and asexual reproduction.[52] Because daphnids are filter-feeders, they collect food with the help of a filtration apparatus comprising the phyllopods, which have flattened leaf-like appendages that generate a stream of water.[50] Even bacteria can be captured with the feeding apparatus. Green algae are reported to be the best food source for daphnids.[50,52]

Pollution of the aquatic environment by plastic particles and its possible consequences has recently been identified as a major global concern that impacts ecosystem functioning. MPs have a wide spectrum of physicochemical features and are widely distributed in aquatic habitats. They disperse diversely in different area of the aquatic environment, and thus, a wide range of aquatic species are potentially susceptible to these contaminants.[2,56] The plastic pollution is most acute in marine environments, but it is now recognized that the problem also applies to freshwater environments. There are numerous knowledge gaps, not least in terms of fundamental knowledge about their distribution and existence in freshwater environments and the relevance and extent of their impacts on members of Daphnia. Freshwater daphnids provide critical pathways for MP pollution, and although current studies have revealed the susceptibility of freshwater biota to translocation, ingestion, and trophic transfer, specific challenges concerning methodological standardization remain largely unresolved.[57] The published reviews on the impacts of MPs on freshwater ecosystems have mostly emphasized the source fate, occurrence, and abundance of MP particles, instrumental detection and analytical methods, and studies with little importance on its effects on freshwater biota.[58–63] Although ecotoxicological studies have been concerned about the environmental impact of MPs, relatively limited knowledge exists about assessment of freshwater organisms. Thus far, most studies have been conducted on fish, and more studies about the effects of MPs on other groups of organisms, especially freshwater invertebrates, are needed. Furthermore, there are considerable differences between the forms of MP most typically detected in the environment, those reported in field investigations and those used in laboratory experiments, and some of the reported results are contradictory. Nevertheless, toxic effects strongly depend on MP type, size and shape. There is a need for a baseline understanding of current data concerning the topic. Therefore, this review aims (1) to discuss and summarize the existing research trends and results in freshwater environments in the context of plastic pollution, focusing on the role of *D. magna* as an ecotoxicological model and (2) to highlight the impact of plastic pollution exposure on freshwater daphnids. It also provides remarks on some consistencies and inconsistencies among studies with respect to different ecologically relevant endpoints of daphnids, such as food uptake, growth, development, reproductive performance, mortality and survival associated with MP polymer shape, type, and size. The majority of available data that have been published in the last several years are highlighted. Finally, perspectives and research gaps are examined, future research priorities, promising areas, and major issues to be addressed are presented.

(A)

Number of Literature*	Daphnia	Freshwater	Plastic pollution	Ecology
Directly relevant (N = 203)	✓	✓	✓	✓
Indirectly relevant (N = 53)	✓	✓	✓	✓

*Examination of titles and abstract to detect relevancy

Data extracted and analyzed for review under topics

- Plastic pollution effect on daphnids
- Ecotoxicological effects of microplastic on daphniids

Discussion under topics

- Plastic pollution in marine and soil ecosystem
- Global treads and regulation of plastic pollution
- Ecology of freshwater regarding daphnids

(B)

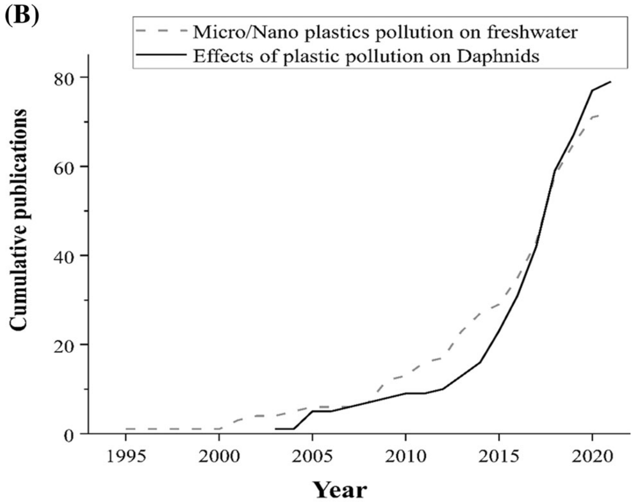

FIGURE 2 A table displaying the methodology for the systematic review of plastic pollution and its impact on freshwater daphnids (A); The gray broken line represents the cumulative number of papers published on micro/nanoplastic pollution and *Daphnia magna*, and the black line denotes all papers published on the effects of plastic on freshwater daphnids. The lines represent the sum of all articles when the last literature review on each topic was completed (B)

2 | METHODOLOGY

2.1 | Findings

This paper reviews a variety of studies on plastic pollution in freshwater ecosystems and the impact on freshwater daphnids. It should be noted that this review provides a cross-sample of research in this area. A widespread literature review was conducted by using the ISI Web of Science Core Collection (http://apps.webofknowledge.com), Scopus (https://www.scopus.com), ScienceDirect (https://www.sciencedirect.com), Wiley Online Library (http://onlinelibrary.wiley.com), SpringerLink (http://link.springer.com), ACS Publications (http://pubs.acs.org), Taylor & Francis Online (http://www.tandfonline.com), and RSC Publishing (http://pubs.rsc.org) databases for studies up to February 2022. Only peer-reviewed literature was included. The keywords for article searches were "microplastics", "plastic waste" in combination with "organisms/biota/freshwater/Daphnia/crustaceans", "ingestion/uptake/transfer", "toxicological/effect/impacts". Articles with the keywords "plastic pollution" and "freshwater Daphnia/crustaceans" were divided into four categories from the database, including "freshwater biology", "environmental sciences", "ecotoxicology", and "environmental studies". Additional studies that were not found in the initial literature search but were deemed relevant by the authors were also added. The 254 publications found were then individually reviewed, and duplications and irrelevant publications were removed. Each paper's abstract and title were evaluated to determine whether it was appropriate for the review. The papers were further divided into categories according to the issues investigated, and finally, the notable study areas were discussed.

2.2 | Results

In total, 205 publications were selected and summarized based on the criteria environmental compartments, biological groups of examined organisms, and ecotoxicological effects of MPs on freshwater daphnids. Figure 2A presents a flow diagram defining this method. In terms

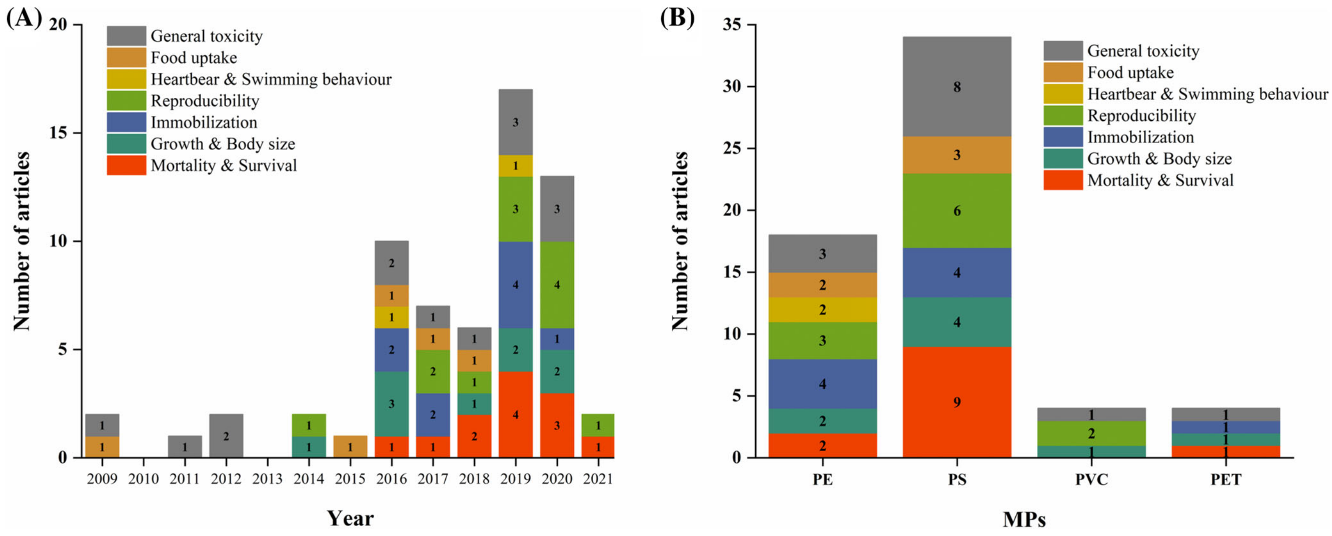

FIGURE 3 Ecotoxicological effects of microplastics on daphnids (from 2009 to 2021). The total number of studies is shown above each bar. (A) Studies were defined according to the years and ecotoxicological effects; (B) Plastic pollution and the freshwater environment. Ecotoxicological effects of microplastics on freshwater daphnids. The total number of studies is presented by a number within each colored section of a bar. The types of microplastics and ecotoxicological effects were used to categorize the studies. PE, polyethylene; PET, polyethylene terephthalate; PS, polystyrene; PVC, polyvinyl chloride

of polymers, the following list was used to categorize MP materials reported in the literature: PS, PE, PP, polyester, PVC, and polyether. The PE family includes both low- and high-density PE. The selected plastic types include the principal groups of plastic particles reported in plastic Europe 2019.[13] The effects of MPs on the freshwater environment and biota, especially daphnids, have been an emerging topic in research for a long time, especially in the last decade with the rapid growth in the number of publications (Figure 2B).

This increasing trend has been especially notable between 2015 and 2021. In 2015, the number of publications on the effect of MPs and NPs on daphnids doubled, and there has been a large increase since that year. Of the 205 articles identified in our first literature search, 171 were classified as related to plastic pollution in freshwater or the effect of MPs on daphnids and were thus included in our systematic review. The ecotoxicological effects enumerated included food uptake, immobilization, development, mortality, reproductive impairment, mortality and survival, physical effects, behavioral effects and oxidative stress. From 205 articles, only 79 were relevant to the effects of plastic pollution on daphnids.

Of all relevant references ($n = 205$) included in our publication, only 26 are reviews, mostly related to marine pollution; two focus on the effects of MPs on human health; and three are related to the occurrence, sources, and detection of MPs or NPs. Only one of the eight scientific reports and only two of the 26 reviews concern the effect of MPs on freshwater. MP-specific studies on freshwater daphnids have been classified into seven main categories – (1) mortality and survival, (2) development-growth rate and body size, (3) immobility, (4) reproduction, (5) heartbeat and swimming behavior, (6) food uptake and (7) toxicity – and different ecotoxicological effects of diverse MP types have been documented. Many studies have focused on the ecotoxicological effects of MPs on reproduction, growth rate, and mortality (Figure 3A,B). In the following sections, the type of experiment and duration or size of MPs and concentrations are reported for each study.

Figure 3A represents the number of publications by ecotoxicological effects on daphnids for each year. Most of the papers from 2009 to 2015 evaluated how MPs produce toxicity and alter the growth rate and feeding of daphnids. In 2016, there was a nearly 10-fold increase in publications, and six out of the seven of these topics mentioned above were covered in these papers. There was a further increase in 2019, and since 2019, the most frequently studied topics have been the effects of MPs on daphnid mortality and survival and the reproduction rate.

Many researchers have investigated the impacts of MPs and NPs in marine environments.[64–67] However, little information exists on this problem in freshwater ecosystems.[17,61,68] Plastic materials and their accumulation in freshwater environments are a source of concern due to rising global consumption and natural resilience to the degradation of plastics. The past 10 years have seen a marked increase in studies identifying the presence of MPs in various freshwater environments, including rivers, lakes, ponds, and reservoirs.[4,69] However, the particle concentrations reported in freshwater investigations are inconsistent, especially because researchers have used different units and sampling methods for quantification.[62] Plastic pollution in freshwater environments has been reported to be the source of numerous hazardous and ecologically harmful effects.[70,71] Diverse pollutant types released in a watershed primarily end up in reservoirs or downstream of a river because they are naturally located at lower elevations, namely terrains and valleys.[72] MPs are abundantly spread in freshwater streams and sediments, and their presence and effect on the local environment have gotten more attention during the last few years.[61,73,74]

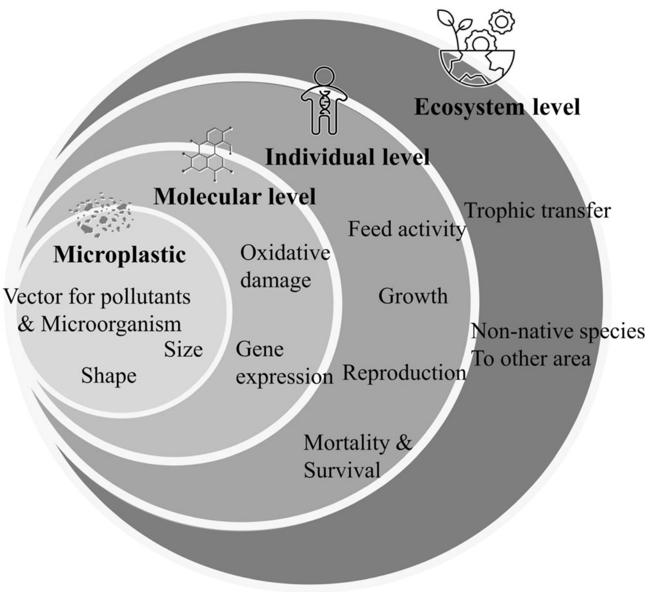

FIGURE 4 A summary of the reported impacts of microplastics on aquatic biota

Plastic hazardous waste is well known to constitute a direct threat to freshwater ecosystems, and numerous species have been identified as very susceptible to damage caused by plastics and plastic debris.[70,75–77] Adding to the direct effects, plastic particles in freshwater have been shown to contain relatively high levels of different organic pollutants. Poisonous chemicals, such as nonylphenol (NP), polychlorinated biphenyls (PCBs), organic pesticides such as polycyclic aromatic hydrocarbons (PAHs), dichlorodiphenyltrichloroethane (DDT), polybrominated diphenyl ethers (PBDEs), and bisphenol A (BPA) have been found consistently bound to plastic debris.[78,79] The presence of these chemicals further increases the risks associated with biota ingesting plastic debris, and many of these chemicals can be biomagnified to a great extent and could pose a direct risk to human health.[78,80] These toxic agents have been linked to a variety of issues, such as developmental impairment and growth abnormalities.[81–83] Because researchers have recently begun to emphasize the importance of MP pollution in freshwater systems, even in drinking water, research about this issue has been deemed mandatory.[84] Furthermore, researchers have called for the incorporation of strategies and plans to decrease the effect of MPs in freshwater environments near urban areas.[85] Moreover, the role of the freshwater system as the main source of MPs to the marine environment should not be neglected.[86] Investigation and research on watersheds and freshwater ecosystems could provide crucial knowledge to those who aim to devise methods that could solve MP pollution in freshwater.[87] Some authors have attempted to assess the presence of synthetic polymers in freshwater ecosystems.[88,89] Currently, plastic pollution has been reported in the freshwater of some countries in different areas of the planet.[88] The findings have revealed alarming quantities of MPs in these ecosystems. Therefore, there is a growing need for new studies on the adverse effects of their presence. A summary of selected studies determining microplastic concentrations in freshwater sampling studies are presented in the supplementary table (Table S1).

In mid-2014, the first research on MP consumption in freshwater animals was published.[4] Most alarmingly, the latest study on this topic demonstrated MP levels of up to 0.14 MPs per mg tissue in 50% of invertebrate samples collected from a riverine valley in South Wales.[90] With their large specific surface area and good adsorption, MPs are important carriers of chemicals and microorganisms, and they may pose a significant threat to the aquatic biota and ecosystem (Figure 4).

3 | *DAPHNIA MAGNA* AS ECOTOXICOLOGICAL MODEL AND BIOINDICATOR

Daphnia (Crustacea; Branchiopoda) are planktonic filter-feeder crustaceans with flattened leaf-like legs that belong to the Phyllopoda family.[51] Daphnia belongs to the Cladocera family of branchiopods, whose bodies are encased in an uncalcified shell known as the carapace.[91] Males are distinguished from females by their smaller size, larger antennules, modified post-abdomen, and first legs, which are armed with a hook used in clasping. The genus Daphnia includes more than 100 identified species of freshwater plankton organisms found around the world.[91] The life cycle of Daphnia during the growth season is characterized by its asexual mode of reproduction which is known cyclical parthenogens. A female produces a clutch of parthenogenetic (amictic) eggs after every adult molt.[92] Under favorable conditions, they reproduce by amictic parthenogenesis, producing genetically identical offspring that build up a population consisting of only females. This can be continued for several generations, resulting in an exponential growth of clonal lineages.[93] When unfavorable conditions arise (e.g., overcrowding, presence of predators, food shortage, change in day-length or temperature), the animals switch to sexual reproduction. Males are produced parthenogenetically, and females switch to the production of sexual eggs. A single female may first produce diploid amictic eggs and subsequently produce two meiotic haploid eggs that need to be fertilized.[94] The dynamics of food uptake in Daphnia follow a functional response type 1. Below a certain food concentration (the incipient limiting level), the food uptake from the water (feeding rate) is proportional to the food concentration, and the filtering rate (amount of water filtered per unit time) is maximal.[91]

MPs have a large surface area and can gather large amounts of hydrophobic organic contaminants (HOCs).[45] MP ingestion has been found in a variety of freshwater daphnids, causing a variety of physical consequences, thereby introducing MPs into the aquatic food web.[40,95,96] *D. magna* is a filter feeder that forages non-selectively on particles with diameters ranging from <1 μm to around 70 μm and is a major zooplankton species that may consume MPs in the aquatic environment.[97] However, MP ingestion by freshwater invertebrates is poorly understood.[98] Exposed species with different feeding strategies to 1, 10, and 90 μm fluorescently labeled polystyrene spheres (3–3000 particles ml^{-1}). They investigated how exposure to natural particles and developmental stages modulate MP ingestion. *D. magna* did not ingest MPs with a diameter of 90 μm, whereas the other freshwater invertebrates preferred MPs >1 μm in diameter. Size preference in *D. magna* was determined by life stage, with larger individuals consuming more and larger MPs.[98] The apparent gut residence period and gut clearance were longer for irregularly shaped MPs, and acute inhibitory effects were more pronounced as compared to regularly shaped MPs.[45] As a result, MP morphology should be considered while conducting studies with filter feeders and MPs because the majority of MPs found in the environment are likely to be irregularly shaped.

Daphnids are able to ingest both irregularly and regularly shaped PE MPs, but egestion of regularly shaped MPs occurs more quickly.[45] The size range of particles collected is determined by the filter combs on their thoracopod exopodites.[99] Consumption of MP particles that fill the intestine might impede the digestive processes, and even when there is enough food, the animal's fitness and energy resources are affected by the subsequent uptake of nutrients.[99] It should also be noted that the combination of (1) the MP morphology as well as the surface features of polymers such as shape and size and (2) an organism's physiology could have a significant impact on MP particle uptake and residence.[100] Despite the limited likelihood of chemicals leaching out into the gut lumen of freshwater organisms exposed to MPs, very few studies have discriminated between the impacts of the plastic and the additives that are added into the same synthetic polymer. Thus, risk assessment studies are required.

D. magna is well-established as an important species in certain freshwater environments.[101] This freshwater crustacean is used extensively to monitor pollution environments all around the world and plays a key part in the establishment of regulatory standards by government bodies.[101] Daphnids are unable to differentiate between particle characteristics such as size and quality, which suggests a lack of selection during feeding and, as a result, likely MP intake. Accumulating MP due to unselective feeding is particularly important because the species is a primary food source for fish. Moreover, carboxylated PS MPs around 20 and 1000 nm can pass through the intestinal epithelium of daphnids.[41] The bioaccumulation of these PS NPs has been linked to negative impacts on daphnid growth, mortality, and reproduction and, therefore, may have lasting effects throughout the food chain.

Some of the many desirable properties of daphnids are a short reproduction time (approximately 1 week at 20°C, making it possible to monitor ontogeny response), a small body size, ease of culture in a simple environment, and susceptibility to a wide range of toxic substances.[102] Food uptake patterns in *D. magna* follow a type 1 functional response. This means that the food intake from the water is proportional to the food concentration below a particular level, and the filtration rate is maximum because the filtering rate decreases as the food concentration in the water increases, and the feeding level remains constant above this point.[50] Moreover, when exposed to low levels of food, *D. magna* can reduce its feeding rate as a result of the decrease in food and potentially reduced survival, growth, and reproduction dynamics.[103] Any unwanted food, which might be indigestible or toxic, could lead to deficiencies in crucial nutrients, possibly causing a decrease in growth or death. However, research has shown that *D. magna* can limit the filtering rate by narrowing their valves in the presence of toxic cyanobacteria, which are larger blue-green algae, to reduce the movement of thoracic appendages and exclude large particles.[104]

D. magna has been the most studied freshwater invertebrate in laboratory experiments and has been used as an ecotoxicological model to evaluate the chronic and acute toxic effects of MPs at different concentrations and sizes.[2,41,105] Due to favorable properties and their role in the ecosystem, some daphnid species, such as *D. magna*, *Daphnia cucullata*, *Daphnia longispina*, and *Daphnia pulex*, have been used extensively in ecotoxicological studies for plastic pollution and chemical substances. This is because they are easy to culture in the laboratory, have high phenotypic adaptability and clonal reproduction, and demonstrate rich testing output and short generation time with several life-history traits.[52,106] It is well established that testing in *D. magna* is relatively inexpensive, rapid, sensitive, and, compared with other methods, requires small sample volumes (e.g., mesocosm tests).[105] Indeed, daphnids are crucial to the balance of aquatic ecosystems, and a reduction in their α or β diversity would seriously disrupt these ecosystems. Hence, daphnids are a desirable freshwater aquatic bioindicator species – and they have been the most widely used species for ecotoxicological laboratory studies to examine chronic and acute toxic effects of MPs of different concentrations and sizes. These studies are discussed in the following sections.

4 | SOURCE, DISTRIBUTION AND TRANSFER OF MICROPLASTICS IN FRESHWATER DAPHNIDS

The distribution of MPs in the environment occurs via various transport media: sewage sludge, industrial and municipal wastewater, urban runoff, and dust. Accordingly, MPs can be found in almost all environmental components such as freshwater, seawater, soils, sediments, and the atmosphere.[58,62,84,107,108] Major sources of MPs are textiles made of synthetic materials (34%), tire abrasions (29%), city dust (24%), marine coatings (4%), road signs/dust (7%), microbeads (2%), and plastic pellets.[107] Specifically, the most common source of MPs for dahpnids in the freshwater environment is from daily supplies such as bottles and bags.[99] The abundance, distribution, and occurrence of MPs in the environment are affected by various factors: the type of environment; characteristics such as the type, shape, density, and size of MPs; climatic zones (such as wind or waves); industrialization; urbanization (proportional to the concentrations of MPs); waste management; general development; and the living standards of the society taken into account.[62]

MPs are resistant to deterioration and do not break down in water, so they stay in the environment for a long time. MPs have great bioaccumulation potential.[109] There are two sources of MPs in the environment, namely primary and secondary. Primary sources include intentionally produced MPs for some products: textiles (washing synthetic garments that provide ~35% of primary MPs), cosmetics (for example, microbeads in facial brushes [~2%] as MPs intentionally added in personal care products), and electronic equipment.[110–112] In addition, tire abrasions during driving (28%), city dust (24%), road construction (7%), marine pavement (3.7%), and plastic pellets (0.3%) are important sources of primary MPs. Most of these enter the environment via wastewater produced throughout their usage or production processes. Secondary MPs are produced by the effects of various processes such as biological (bacteria, fungi, and algae), physical (weather, temperature, and mechanical forces), chemical degradation by oxidation, and finally, photo degradation (UV light). The secondary sources include the breakdown of larger plastic objects such as bottles, plastic bags, or fishing nets that are released into the environment; degradation is caused by chemical oxidation, physical corrosion, UV radiation, and probably biodegradation.[113–116] Other degradation processes alter the physicochemical properties of the polymer, rendering it brittle, and then mechanical degradation, most important for plastic in the aquatic environment, breaks them down into smaller pieces: MPs are 1-mm.[117,118] In other circumstances, these MPs are reported to degrade further to yield NPs.[119] The source of MPs and their relative abundance varies by region, depending on the capability and effectiveness of waste management. Primary MPs are more problematic, and the majority of these plastics come from land-based sources that have access to marine environments through rivers and other ways.[120,121]

Agriculture is one of the most important points of entry for MPs in the environment.[122] The main sources are sewage sludge applications, soil conditioners, fertilizers/compost, and vinyl coatings. Low-density PE films, which are widely used to protect agricultural products, suppress weeds, increase temperatures, and retain irrigation water in the soil, can reach soil or water resources by breaking into small pieces through irrigation channels. Organic fertilizers used in agriculture and horticulture around the world tend to be a neglected source of MPs, and fertilizers pre-treated by composting and fermentation are also included in this category.[123] However, significant amounts of MPs come from various industries (through wastewater or disposal of plastic residues) and households (from washing linen fibers, personal care products use). MPs can be carried by the wind, washed from land to surface waters during precipitation if released into the environment (especially rainwater runoff), and transported in freshwater and seawater.[58,84,124,125] Industrial and domestic wastewaters are also one of the main sources of MPs containing synthetic fibers (e.g., polyester, PES).[108]

MPs have become a matter of concern in the ocean and the aquatic environment in general.[126] They primarily enter the aquatic environment as microbeads (<1 mm) from cleaning agents, cosmetics, and broken pieces of plastic. These have resulted from either the washing process or represent degraded plastic trash and debris.[127] Wastewater treatment plants are reported to be the source of massive and widely scattered volumes of MPs in freshwater – denoted by an increased concentration of MPs downstream, adding to the contribution of MPs from other sources as well as from wastewater treatment plants.[128] Humans uptake MPs primarily through food and beverages, such as fish, mollusks, sea salt, beer, sugar, and even tap and bottled water.[129–131]

MPs are dispersed in both seawater and freshwater.[84] MPs have been found in deep-sea sediment environments with depths ranging from 1100 to 5000 m.[132] They have also been discovered in the deepest parts of the seawater.[133] Freshwater has a similar amount and distribution of MPs as seawater.[114] MPs from various sources enter waterways, often end up in freshwater, and eventually reach seas and oceans; however, this process relies on the proximity of the source to seawater or freshwater. As a result, MPs have been found all over the world in aquatic ecosystems. PE, PP, and PS have been the most commonly identified materials, with the majority of them being <5 mm in size. Hence, MPs are ubiquitous in water bodies, where marine organisms can easily ingest and transmit them, putting food security and safety at risk.[121]

MPs are known to influence approximately 700 species of aquatic organisms, and they can be found at various trophic levels. MP accumulation in species at low trophic levels has a food web–linking effect.[134] The physical and chemical properties of microplastics facilitate the sorption of contaminants to the particle surface, serving as a vector of contaminants to organisms following ingestion. Bioaccumulation factors for higher trophic organisms and impacts on wider marine food webs remain unknown.[134] Much of the concern surrounding microplastics is due to the chemical additives and sorbed contaminants having the capacity to desorb into an organism. Chemicals including phthalates, bisphenol A, flame retardants, PCBs, pesticides, fertilizers, and heavy metals are known endocrine disruptors, carcinogens, and mutagens.[135] The leaching of additives from plastic combined with the sorption of chemicals to plastic renders microplastics a cocktail of toxic contaminants. The biomagnification of organic pollutants from lower trophic levels to higher trophic levels has been demonstrated as has the capacity of microplastics to act as a vector of these contaminants to aquatic biota.[21] Microplastics are hydrophobic particles that behave like DDT and PCBs, acting as chemical inhibitors within living organisms and bioaccumulate within the food web.[136] Microplastics can absorb a wide range of pollutants and are able to leach out chemicals such as phthalate and BPA which can enter the tissues of aquatic species and pose a threat to humans.[137] As aquatic species prey on MPs, they migrate through the food web.[27] MPs are transferred tropically in vertebrates as well as crustaceans and plankton. Aquatic invertebrates frequently consume MPs that are the size of several planktons, which tend to be transferred to vertebrates at the upper end of the food chain. Some researchers, however, believe that organisms can quickly and effectively eliminate MPs, and hence the high MP level would be unlikely to have a major impact on organisms.[138] Setälä et al.[27] discovered that nutrient movement happened after 3 h of contact between macroplankton and medium-sized plankton ingesting polystyrene MPs. Another study demonstrated that although the number of MPs transferred from daphnids to the fathead minnow (*Pimephales promelas*) was small, trophic transfer occurred between these two species.[139]

How long MPs stay and how they accumulate in biota are crucial factors that influence trophic transmission. The shape and size of MPs are also significant factors.[140] Individual differences in aquatic organisms may also influence residency time. The longer the residence time in biota, the easier the transfer of MPs along trophic levels.[141]

5 | ECOTOXICOLOGICAL EFFECTS OF MICROPLASTICS ON DAPHNIDS

5.1 | Physical toxicity

Daphnia magna is a keystone species in freshwater habitats and the most commonly used freshwater aquatic indicator species for ecotoxicological studies.[101] *D. magna* has been subjected to a variety of MPs in numerous studies.[46,97,142,143] MPs had various ecotoxicological effects, including decreased reproductive and survival rates to no effect at all, depending on the MP exposure rate and concentration.[6] However, many researchers have found that exposure to high levels of MPs increases mortality and decreases growth rate. A study, expressed the levels as total biomass, exposure to 10^5 microplastics ml^{-1} resulted in a 21% reduction in total biomass compared to control.[81] Another study investigated three 21-day laboratory bioassays with model MPs (1–5 μm diameter). In each bioassay, one control (no MPs) and three MP concentrations (0.04, 0.09, 0.19 mg/L) were tested. In all the bioassays, MPs caused parental and juvenile mortality, and reduced the somatic growth, reproduction, and population growth rate.[144] As mentioned in Section 2, there has been a marked increase in studies that have evaluated the ecotoxicological effects of MPs (Figure 3A). Freshwater crustaceans are vulnerable to the adverse effects of different types of plastics in their environment.[72] Besides, longitudinal analyses have shown that *D. magna* needed several generations to recover from the adverse effects.[145] Plastic ingestion by nine Crustacea species has been discussed: two in natural and semi-natural habitats and the others under controlled conditions.[146] Several studies have reported the adverse effects of MPs on *D. magna* as lethal, non-lethal, or both.[72] Of the 21 studies, 5 (24%) have emphasized the lethal effects on daphnids, 12 (57%) the sublethal effects, and 4 (19%) both lethal and sublethal effects. These studies are summarized in (Table S2).

MPs' ecotoxicological effects on reproduction, growth rate, and death have been explored by many studies (Figure 3B). They found that the effects are more related to food availability rather than MP toxicity.[147] Nevertheless, researchers have determined that of the sizes and types of MPs, atypical shapes are more toxic to *D. magna* than spheres, which could clog the gut system, and smaller MPs increase immobilization, induce the stress response, and decrease the growth rate of *D. pulex*.[46,97,148] Likewise, according to several studies, the toxicological damage to a daphnid depends on the particle concentration, duration of exposure, shape, chemical composition, and size, with stronger effects for smaller particles.[29,32] Besides, some studies have found that MP uptake by daphnids is time and concentration-dependent.[46] For example, researchers have evaluated the consequences of PS on cladocerans under laboratory conditions. The effects ranged from decreased survival to changes in the ability to reproduce. The mere presence of certain types of plastics in the environment could have sublethal to lethal consequences.[146,149] The use of fluorescently labeled MP beads in animals is a standard methodology. This approach allows researchers to follow their accumulation easily by using fluorescence microscopy. To date, researchers have explored various types of plastics and various sizes using mostly *D. magna* as well as other invertebrates.[40] The list of freshwater daphnids that have been exposed to various microplastics is presented in Table 2.

To provide a thorough explanation of the presence of MPs in organisms, MP digestion and egestion quantities must be established. There are many laboratory studies on invertebrate species; few studies, however, have examined whether MP ingestion impacts egestion rates, especially at concentrations present in the environment.[7,150,151] Despite the fact that multiple studies suggest that MP egestion is substantial, they have been reports of MPs moving out of the digestive tract and into other body tissues. For example, in *D. magna*, 1 μm MPs could translocate across the gut epithelial barrier.[41] When daphnids were 1 week old, they were given 2 μm and 100 nm fluorescently labeled PS beads (1 mg l^{-1}) for 24 h; egestion in a clean environment was then examined 24 h later.[105] During both phases, the fluorescence intensity in homogenized tissues was measured to detect particle loads in the body. Egestion and ingestion rates were evaluated in the presence and absence of food. Both particle sizes were easily swallowed, although the mass of the ingested 2 μm particles was five times higher than for 100 nm particles. Complete egestion did not occur within 24 h; however, larger quantities of the 2 μm particles were frequently ingested. Particulate loads in the animal body strongly decreased in the presence of food. In the presence of 100 nm particles, the feeding rates were reduced by 21%, but after 21 days of exposure, no effect on reproduction was detected despite the high body load of the particles. It is difficult to draw clear conclusions due to the variety of methodologies that have been used to examine the vulnerability of freshwater organisms to MP ingestion and the effects derived from this ingestion.[152,153] Therefore, laboratory studies should focus on assessing intake and egestion rates during extended periods of exposure.[154] Algal biofilm growth on MPs over time should also be considered; this parameter could impact the ingestion rate of an aquatic organism and makes tests more ecologically relevant.[155]

MP uptake is expected to have a variety of effects on an organism based on its form, size, concentrations, exposure time, and feeding method.[39] Ingestion is also affected by the size, shape, type, and concentration of MPs.[46,108] *D. magna* has been found to ingest long PET fibers (<1400 μm).[156,163] MP accumulation would decrease the rate of intake at higher MPs concentrations. The MP shape is particularly relevant to residence time. Fibers 1–5 mm long, for example, have been found to collect in the guts of crustaceans.[164] However, some studies did not report the MP burden in the context of concentrations or abundances.[107,165,166] MP concentrations reported in field investigations and those employed in experimental studies differ significantly. The utilization of a broad range of environmentally realistic concentrations as well as higher concentrations could provide

TABLE 2 Summary of selected freshwater daphnids that have been subjected to various microplastics.

Species	MP type	MP size	MP shape	Tested concentrations	Exposure time	Endpoint examination	Observations	References
Daphnia pulex	PS NPs	75 nm	Monodispersed microspheres	2 mg l^{-1} (1.06 × 10^9 particles ml^{-1})	48 h to 21 days	Survival, growth, reproduction, oxidative stress, and heat shock proteins	LC_{50} = 76.69 mg l^{-1}; decreased reproduction rate; decreased growth rate; induced stress response	148
D. pulex	Primary fluorescently labeled plastic microspheres Secondary irregularly shaped plastics Three temperatures (18, 22, and 26°C)	1–5 and 1–10 μm	Suspension	1 × 10^3, 1 × 10^4, 1 × 10^5, 1 × 10^6 and 1 × 10^7 particles ml^{-1}	96 h	Acute toxicity test by increasing the temperature	Increased mortality as the temperature increased	156
D. magna	Carboxylated PS MPs	20 and 100 nm	PS beads	2 mg l^{-1}	30 min to 24 h	Uptake, accumulation, and depuration	Accumulation in gut epithelial layer with faster depuration for larger beads	41
D. magna	Primary fluorescently labeled plastic microspheres Secondary irregularly shaped particles Three temperatures (18, 22, and 26°C)	1–5 and 1–10 μm	Beads	1 × 10^3, 1 × 10^4, 1 × 10^5, 1 × 10^6, and 1 × 10^7 particles ml^{-1}	96 h	Acute toxicity test	Increased mortality as the temperature increased (both primary and secondary MPs)	156
D. magna	PET microfibres with and without algal (*Desmodesmus subspicatus*) pre-feeding	300 μm	Fibers	12.5–100 mg l^{-1} algae (density 5 × 10^4 cells)	48 h	Uptake, feeding performance, and immobilization	No lethality in daphnids fed with algae; unable to recover from microfibre-induced stress after an additional 24-h incubation period	46
D. magna	MPs and NPs combined with Phe (chemical stressor)	50 nm and 10 μm	Suspension	50 and 500 mg l^{-1}	14 days	Uptake vector property of MPs	Significant Phe bioaccumulation, dissipation and transformation in daphnids that received 50 nm compared with 10 μm particles, demonstrating the higher adsorption rate of hydrophobic contaminants on smaller particles	157
D. magna	Primary and secondary MPs	1–5 μm	Beads	1 × 10^2–3 × 10^4 MPs ml^{-1}	12 min to 21 days	Uptake and depuration	Increased gut passage time with aggregates after secondary MP exposure; reduced feeding and reproduction at high MP levels; secondary MPs are more harmful than primary uptake	143

TABLE 2 (Continued)

Species	MP type	MP size	MP shape	Tested concentrations	Exposure time	Endpoint examination	Observations	References
D. magna	PE MPs	1 and 100 μm	Dry powder	12.5–400 mg l^{-1}	96 h	Uptake and immobilization	1 μm MP ingested and caused immobilization; 96-h EC_{50} for 1 μm MPs is 57.43 mg l^{-1}	97
D. magna	Fluorescently labeled PS beads with and without the alga *Raphidocelis subcapitata*	2 μm and 100 nm	Fluorescently labeled beads	1.4×10^5 and 3.1×10^8 particles ml^{-1} 6.7×10^5 cells ml^{-1}	21 days	Feeding rate assessment	Five times higher ingestion rate for 2 μm; 21% decreased feeding rate with no significant effects on reproduction	105
Daphnia galeata	PS NPs	52 nm	Beads	5 mg l^{-1}	5 days	Survival, reproduction, and growth rate	Decrease in survival; decrease in reproduction; abnormal embryonic development	146
D. magna	PC, PET and PVC	~ 40	Beads	2.9×10^5 MP l^{-1}	2 days	GST	- -	158
D. magna	PS	1–10 μm	Beads	0.1 mg l^{-1}	2 days	CAT GP_x GST MDA SOD	↑↑ ↓- ↑↑ – –	158
D. magna	PMMA	29.5 ± 26 μm	Not available	Not available	48 h	Uptake	100% ingestion of MPs	40
D. magna	PS	1 μm	Not available	Not available	Not available	Uptake	Ingestion of PS particles	159
D. magna	PS PS + Phe	50 and 500 nm, 5, 10, and 15 mm	Suspension	PS: 2.5–50 mg l^{-1} Phe: 0.05–1..2 mg l^{-1}	48 h–14 days	Uptake	Significant dose–response effect for 50 nm PS ($EC_{50} = 15.13 \pm 3.34$ mg l^{-1}); 50 nm PS act as a vector for Phe by absorbing and concentrating them on the surface; physical damage to *D. magna*	157
D. magna	PET	62–1400 μm	Fibers	0.02 g l^{-1}	24–48 h	Uptake	Increased mortality	46
D. magna	PS	50 nm to 10 μm	Beads	2.5–50 mg l^{-1}	14 days	Uptake	Long-term Phe accumulation; effects on dissipation and degradation of Phe	157
D. magna	PS	1–5 μm	Fluorescently labeled beads	1×10^2–1×10^5 particles ml^{-1}	21 days	Population and total biomass	Decreased population	81
D. magna	PS	2 μm	Beads	2.5 mg ml^{-1}	21 days	Uptake and retention	Increased mortality	160
D. magna	PS	15 μm	Beads	5×10^6 MPs ml^{-1}	28 days	Survival	Decreased population	161
D. magna	PVC	95 μm	Beads	10 mg l^{-1}	21 days	Reproduction	Decreased reproduction rate	83

(Continues)

TABLE 2 (Continued)

Species	MP type	MP size	MP shape	Tested concentrations	Exposure time	Endpoint examination	Observations	References
D. magna	PS	53 nm	Beads	0.32 mg l^{-1}	103 days	Reproduction	Decreased reproductive rate; decreasing trend in the number of offspring	82
D. magna	PS	1–10 μm	Beads	0.125, 1.25, 12.5 μg ml^{-1}	21 days	Uptake	Decreased reproduction, growth, and swimming rates	162

Abbreviations: CAT, catalase; EC_{50}, half-maximal effective concentration; GP_x, glutathione peroxidase; GST, glutathione S-transferase; LC_{50}, median lethal concentration; MDA, malondialdehyde; MPs, microplastics; NPs, nanoplastics; PE, polyethylene; PET, polyethylene terephthalate; Phe, phenanthrene; PMMA, polymethyl methacrylate; PS, polystyrene; PVC, polyvinyl chloride; SOD, superoxide dismutase.

information on dose-dependent effects and ingestion.[153] The surface characteristics of MPs, together with an organism's physiology, may substantially affect the uptake and residence of MP particles.[100] It should also be acknowledged that as soon as a plastic is released into the ecosystem, biomolecules interact with them, resulting in ecocorona formation on surfaces. This phenomenon changes the plastic properties and affects the ingestion rates among biota.[167,168]

A total of 63 studies reporting ecotoxicological effects of MPs on freshwater *D. magna* were identified (Figure 3A). Frydkjær et al.[45] reported that high concentrations of PE particles reduced the movement of *D. magna*, whereas irregularly shaped fragments (10–75 μm) affected *D. magna* more intensely than regularly shaped beads (10–106 μm). They discovered that *D. magna* quickly ingested both irregularly and regularly shaped PE MP particles, but egestion of regularly shaped MP was faster than egestion of irregularly shaped MP. Additional studies are needed to determine the long-term implications of various irregular MP morphologies. Rehse et al.[97] showed that limnic *D. magna* was immobilized by digestion of 1 μm PE particles as the concentration and exposure time increased, but 100 μm particles were not ingested due to the large size and did not lead to physical effects.

Canniff and Hoang[163] reported that *D. magna* survival and reproduction were unaffected by exposure to PE microbeads from 63 to 75 μm despite causing intestinal blockage and promoting the growth of the alga *Raphidocelis subcapitata* during the 21-day experiment. In this study, the trout chow (YCT) and algae were co-cultured. *D. magna* fed on green algae because its primary feeding strategy is to filter suspended particles to extract nutrients such as green algae. Their results revealed that MPs could serve as substrates for algal growth. According to the findings, when particle concentration and exposure time increased, the number of ingested beads also increased. Algae have been observed to grow and form biofilms on plastic substrates. Scherer et al.[7,98] proposed that MPs with algae considerably reduced MP intake by *D. magna*. This finding is similar to a previous conclusion drawn by Ayukai et al.,[169] that is, in their study, *Acartia clausi* showed preferential feeding when exposed to MP spheres and algae.

According to Frydkjær et al.,[45] microbeads were swallowed by *D. magna*, resulting in body concentrations of 30–50 MPs per organism. These body concentrations are higher than those discovered by Canniff and Hoang (0.44–15.06 MPs per organism).[163] Frydkjær et al.[45] used a broader range of particle sizes (10–106 μm) and exposure concentrations (10–5000 mg l^{-1}), which explained the difference in ingestion. The difference in the concentrations and the methods used makes it difficult to draw conclusions. Hence, uniformity of MP properties, such as the reported particle size, will improve study comparisons.

Rehse et al.[97] reported size-dependent changes in the inhibitory effects of MPs. After short-term exposure, smaller PE particles are more toxic to *D. magna* than larger ones. The authors demonstrated that absorption of 1 μm particles increased immobilization in a dose- and time-dependent manner with a half-maximal effective concentration (EC_{50}) of 57.43 mg l^{-1} after 96 h. However, the daphnids were unable to ingest 100 µm particles, and no effect was observed. All experiments were conducted in sterile conditions. Similar results were obtained by exposing *D. magna* to PS.[157] The researchers found that 50 nm PS NPs substantially increased the bioaccumulation of the plastic additive phenanthrene in *D. magna* and both enhanced the toxic effects, while 10 µm PS MPs did not show significant effects. Phenanthrene was used as a model compound; it has significant mutagenic and carcinogenic toxicity to organisms.[170] In a similar study, when MPs of 50 nm and 10 µm were combined with phenanthrene and exposed to *D. magna*, the smaller MPs absorbed more hydrophobic contaminants.[157] Therefore, nanometer-sized particles should receive more attention, including considering their interactions with hydrophobic contaminants in the environment. The effects of NPs on the food web in freshwater organisms, as well as their chronic toxicity, should be a study priority.

5.2 | Chemical and cellular toxicity

Few studies have investigated the effect of MPs combined with environmental stressors or other chemicals. The bioavailable sizes could be corrected by determining the upper and lower particle size limits.[39] Therefore, the size and other morphologies of MPs should be considered because they affect MP bioavailability. Furthermore, while the absorption of MPs might occur quickly, full gut depuration can often take more than 24 h.[171,172] Laboratory investigations should therefore seek to quantify egestion and ingestion rates over prolonged exposure times.[154] Zhang et al.[173] reported that the co-exposure of PS MPs (1 and 10 µm) and the antibiotic roxithromycin (ROX) led to acute toxicity, significant biological reactions, and oxidative stress in *D. magna*. Co-exposure to PS and ROX amplified the activity of superoxide dismutase (SOD) in daphnids more than when treated with ROX alone. The results may be attributed to ROX being adsorbed to PS, a phenomenon that might lead to greater accumulation of ROX in *D. magna* than when ROX is present alone. Similarly, Qu et al.[174] found that the SOD activity induced by co-exposure of MPs and venlafaxine was three times higher than that induced by venlafaxine alone. Felten et al.[175] focused on the combined effects of the insecticide deltamethrin and PE particles (1–4 m) on *D. magna* for 21 days. They discovered that the unfavorable impacts on survival, fertility, and brood quantity were synergic. Lin et al.[176] first reported that the bioaccumulation of polyaromatic hydrocarbon (PAHs) mixtures mainly related to the dermal uptake of *D. magna*, while mixtures of NPs (100 nm) increased the mass transfer of PAHs to lipids in the gut. Kim et al.[177] suggested that the immobilization of *D. magna* subjected to nickel and PS particles (194 nm) was lower than those subjected to nickel and PS-COOH MPs (182.7 nm). According to their study, PS MPs had a minor antagonistic effect on nickel-mediated toxicity, and PS-COOH had a minor synergic effect when combined with nickel. Their research revealed a combination of toxic effects, most likely due to the unique features of MP surface functional groups and related contaminants. Kalčíková et al.[178] reported that biofouling (aging) behavior supported Ag adsorption onto PE microbeads from cosmetic products and affected its subsequent leaching. Subsequently, aged microbeads with absorbed Ag significantly increased the combined toxicity to aquatic organisms, reduced growth rates and root length of duckweed *Lemna minor*, and completely inhibited daphnid motility.

Biofouling could be a major factor influencing MP characteristics, pollutant adsorption and release into the environment, and toxicity. Smaller MPs appear to affect *D. magna* by adhering to the inner and outer cuticle layer of the carapace, lowering filtering activity, compromising gut integrity, and entering tissues and cells.[179] Also, MP's biofouling affect the molting of daphnids[180]. The life cycle of *D. magna* is distinguished, and ecotoxic acute tests are conducted at the offspring phase. Therefore, molting is a vital aspect of daphnids because it is closely related to their growth. Castro et al.[179] show significant differences between the number of molts according to the MP's exposure time.[180] The combined mechanism of toxicity, interaction of MPs with co-existing pollutants in freshwater biota, and biofouling-related toxicity remains unclear. Hence, future studies should focus on these areas to address this gap.

In addition to ingestion, MPs also affect *D. magna* egestion. This process is critical for organisms; insufficient egestion, or even complete absence, results in reduced food intake and, consequently, starvation.[46,181] A large portion of *D. magna* (83%) fed regularly shaped MPs emptied their gut during the initial depuration, while none of the organisms fed with irregularly shaped MPs were able to clear their guts within 90 min.[45,163] This is a major concern because irregularly shaped MPs are widespread in aquatic ecosystems.[45] Small MPs could pass through the intestinal epithelium of *D. magna* and accumulate in lipid storage droplets.[41] Although the majority of findings have contradicted this assertion.[4] Some studies with *D. galeata* exposed to MPs have revealed the transfer of particles from the external body to the internal organs, ovaries, brood chamber, caudal appendices, and thoracic appendices, as well as storage in lipid droplets.[146] It should also be noted that time allows for biofilm growth, which could affect the ingestion rates of aquatic biota and requires more ecologically appropriate tests.[155] A positive linear association has been discovered in several investigations between the amount of MPs swallowed and the amount present in the surrounding medium.[39,98] This link could be measured by means of trophic transfer factors, which could be used to compare MPs, species, and exposure conditions.[57,182] In addition, ingestion and egestion rates of MPs by other aquatic invertebrates with similar autecology should be investigated to gain a complete picture.

Jaikumar et al.[156] compared the influence of MPs at a lower temperature and revealed that exposure to a high temperature and MPs had a negative effect on the survival rate. They also claimed that by raising the temperature, primary MPs had a more toxic effect on *Ceriodaphnia dubia* than secondary MPs. The authors exposed *D. pulex*, *D. magna*, and *C. dubia* to MPs at different temperatures and discovered that sensitivity to various MPs seems to vary from species to species, while the acute sensitivity of daphnids suddenly increases with the temperature.[156] Kokalj et al.[183] simulated real environmental conditions to evaluate whether pre-feeding daphnids with algae has any effect and found that pre-feeding did not affect MP uptake on

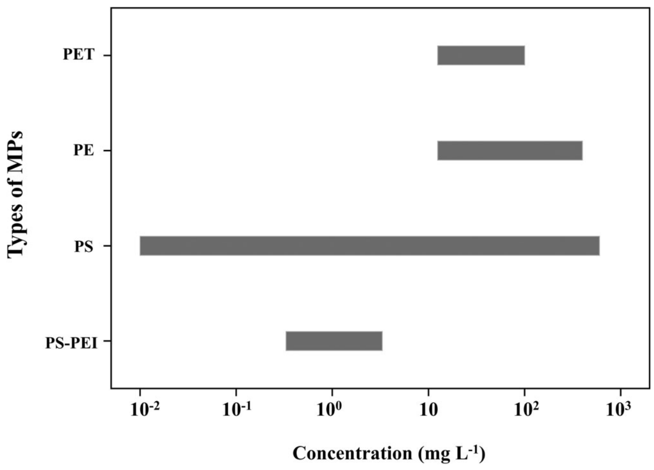

FIGURE 5 The ecotoxicological range of microplastic concentrations in *Daphnia magna*[219]

daphnid growth and there was no increase in mortality in pre-fed daphnids.

PS is the most common type of MP consumed in the species of Crustacea (Figure 5). Small PS particles have a negative impact on daphnid reproduction and body size: 67.7% of malformed offspring were observed.[92,108,184] *D. magna* took up PS beads ranging in size from 50 nm to 15 μm, and Ma et al.[157] found a high correlation between particle size and toxicological effects and interactions with hydrophobic contaminants in the environment. Significant toxicity and physical damage to *D. magna* were observed for 50 nm PS particles, with an EC_{50} of 15.13 ± 3.34 mg l^{-1}.

Given the environmental occurrence of small plastic particles, the lack of studies examining nanoscale particles represents a discontinuity with their environmental relevance and risk. Apart from plastic morphology and type, there have been studies related to the physicochemical characterization of MPs. PE stands out as the second most common MP species studied in daphnids (Figure 5). Researchers have investigated the ecotoxicological effects of PE on daphnids. For PE, PS and PVC, the most frequently examined topics for each type of MP have been: 21% of studies with PE have examined immobility; 28% of studies with PS have examined mortality and survival, followed by toxicity (25%); and 50% of the studies with PVC on the reproduction rate.

6 | DISCUSSION

Microplastic research in the aquatic environment has evolved over the last decade, with remarkable growth in the number of publications.[185] The number of laboratory and field studies on MPs describing their interactions and impacts on freshwater species, particularly daphnids, has increased dramatically. However, there is a gap in ecotoxicological knowledge on the behavior of MPs in freshwater biota (Figure 6). This review has presented the current state of knowledge on the impact of MPs on freshwater daphnids. The available data and research gaps have been noted, and the need for proper risk assessments has been emphasized. The majority of the studies conducted on freshwater invertebrates have been conducted in laboratory conditions, answering questions related to depuration, uptake, digestion, and ecotoxicological effects of MPs.[6] However, there is still a lack of sufficient knowledge about MPs in freshwater, including their fast monitoring and health effects on biota. Many researchers have commented on the lack of ecotoxicological data on the behavior of MPs in freshwater species.[17,61,186] Some researchers have raised concerns over the quality of some of the research, and modest efforts have been made to put the results from diverse research on the effects of MPs on aquatic biota into a risk context – but this endeavor is hindered by the inconsistent study conditions.[152,186] The majority of MP pollution ecotoxicity investigations in the literature have focused on apical endpoints such as decreased growth, mortality, body size, feeding, immobilization, and reproduction failure. To the best of our knowledge, the study by Trestrail et al.[187] is the first to provide insights into toxicity mechanisms. The authors investigated the reactions to phenol-formaldehyde (PF) MPs produced by two types of PF foams, and they compared the biological effects to those of MP leachates derived from foam MPs that had been freshly crushed (50 mg MP ml^{-1}) after shaking for 24 h at 25°C. After 24-h exposure, the MPs were acutely toxic to *D. magna*, with a median lethal concentration (LC_{50}) ranging from 15 to 27 mg ml^{-1}. Hence, to better understand the toxicity mechanisms, it is advisable to conduct additional research at the cellular and molecular levels.

In most laboratory-based studies, parameters differ extensively across studies, including the properties of the media, the level of agitation, the solid-to-liquid ratio, time and temperature. Some reviews have highlighted that consideration should be given to performing studies at environmentally relevant plastic concentrations.[2,62] For example, Li et al.[188] used 1000–5000 cm^2 l^{-1}, equivalent to 100–500 g l^{-1} plastics, for their toxicity test. Lithner et al.[189] used a liquid-to-solid ratio of 10:1 (equivalent to 100 g l^{-1}) and 4:1 (equivalent to 250 g l^{-1}) for leaching plastic. Bejgarn et al.[190] used a liquid-to-solid ratio of 10 (equivalent to 100 g l^{-1}) to prepare plastic leachate for their toxicity test.

Some plastic particles have the potential to adversely affect this organism when exposed at very high concentrations (e.g., EC_{50} of 8.6×10^7 particles l^{-1}).[143] Moreover, environmental MPs exist as a mixture, and this should be reflected in ecotoxicological studies – for example, testing fragments, beads, and fibers at the same time in the proper concentrations and proportions would be useful. There are also inconsistencies in the reporting of particle concentrations within field studies and those applied in experimental studies, as well as the units employed. Concentrations from field records are described as the number of particles per surface area or volume, but in most cases, laboratory results are stated as mass per volume, which makes a comparison of concentrations problematic. Particle concentrations in laboratory experiments vary from high (108 particles l^{-1}) to ecologically relevant (such as 400 particles l^{-1}).[57] However, some researchers focusing on identifying effect-based thresholds and have already addressed the incompatibility in the units and employed

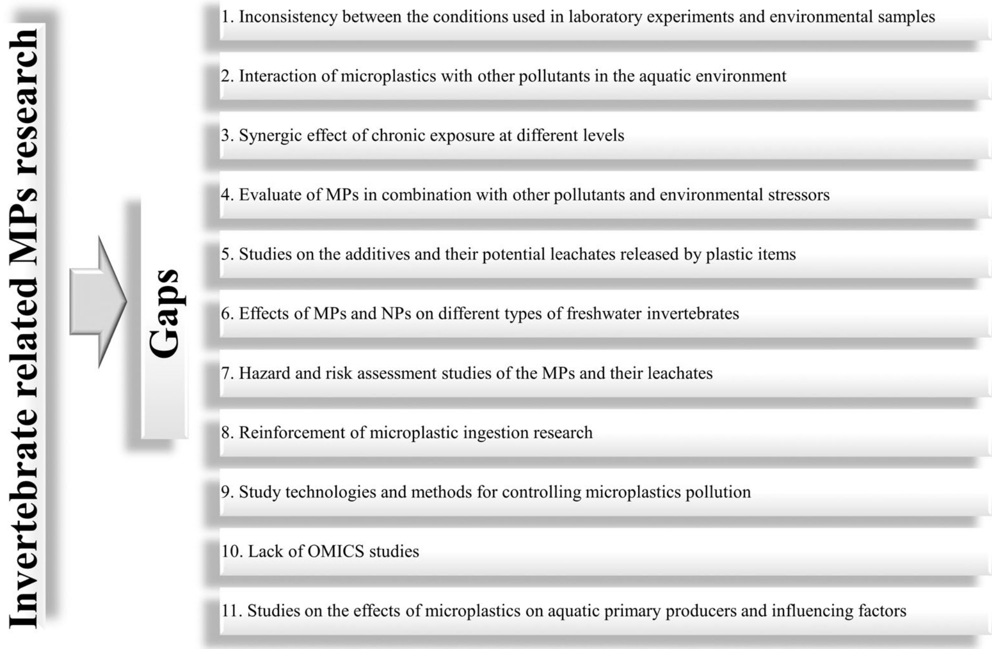

FIGURE 6 Significant gaps in invertebrate related microplastic studies

concentrations relative to environmentally actual values.[150,191,192] Given the environmental prevalence of small plastic particles, the lack of studies examining particles <100 μm represents a discontinuity with environmental relevance and risk.

Apart from plastic morphology and type, few studies have been related to the physicochemical characterization of MPs. According to estimates, > 90% of the aquatic plastic waste had been in the environment for more than 2 years, and the majority of plastic debris within the environment is highly weathered.[193] Most studies evaluating MP pollution in freshwater daphnids have used new plastic products. Only a few studies have reported using plastics that had been weathered before experiments. In many instances, the extent and method of weathering have been poorly described or are insufficient to simulate actual weathering.[194–196] Hence, laboratory studies considering plastic degradation due to weathering and a long period of time are essential to understand the broader risks of MP pollution on freshwater biota. Environmental conditions of freshwater systems fluctuate extensively, seasonally, or daily.[197]

Turbulence is another important factor that has mostly been neglected in studies. There is strong evidence that turbulence increases the leaching of additives of polymers and thus impacts aquatic biota. There is only one study in which the researchers assessed the impact of turbulence. Suhrhoff et al.[198] suggested that a comparably high proportion of BPA is leached into the water when subjected to turbulence. They indicate that the leached PVC additives were between 20 and 79 times stronger under turbulent conditions. Hence, this factor should be considered in future experiments. This is primarily evident in ponds, streams and lakes where low water renewal rates and frequent physicochemical variations might affect additive leaching from weathered plastics.

Thaysen et al.[199] performed one of the few studies providing evidence on physicochemical variations of MPs. They evaluated the chemical composition and toxicity of expanded polystyrene (EPS). They observed a reduction in reproductive output and 40% mortality in *D. dubia*. Studies on the negative impacts of plastic leachate on freshwater biota have recently been reported, reflecting the growing global concern about plastic pollution in the aquatic environment. Leachate from plastics has previously been shown to cause acute poisoning in *D. magna*. In these experiments, treatments were frequently indicated as a percent dilution of generated leachates. However, the concentration of plastic particles tested for leaching varied greatly between investigations, ranging from 5 $g\,l^{-1}$ (200:1 liquid-to-solid ratio)[200] to 250 $g\,l^{-1}$ (4:1 liquid-to-solid ratio).[200,201] As a result, comparing data from different tests, polymers, and species was difficult.

Researchers have evaluated the leaching of plastics over 24–48 h according to the United States Environmental Protection Agency and European Union standard guidelines.[202] While the majority of studies have used long-term leaching experiments (3–28 days), one study examined exposure of *C. dubia* to EPS leachates for 30 min at a relatively high temperature.[199] In addition, just one study looked into the impact of pH variations.[203] As a result, it is evident that a set of standardized rules for experimental preparation is required. This might be used to imitate realistic environmental conditions while meeting standard requirements for ecotoxicity testing. To provide a useful risk assessment, ecotoxicological investigations require consistent treatments. Additional studies are necessary in this regard to elucidate the extent of the influence of physicochemical variations and leachate toxicity on freshwater organisms.

Despite the fact that PE is the most abundant polymer in environmental samples, PS has been the most popular test material in published studies.[154] MPs found in the environment are primarily distinguished by morphological characteristics such as shape, color, and size.[204] Different size classifications have been employed, but there are inconsistencies in reporting particle size ranges across freshwater organisms, as well as a general lack of consistency in categorizing macroplastics, MPS, and NPs. Nearly all MP investigations have used particle sizes that are smaller than those that can be reliably identified in the environment (<131 μm), and they have mostly focused on spherical particles, with fragments or only a small number of fibers being tested.[154,205] MPs of smaller sizes had particle per liter counts orders of magnitude greater than the MPs of larger sizes tested.[44,206,207] In such instances, determining whether smaller particles are more hazardous than larger ones are unlikely. Reporting in particles per liter is preferred because it is directly equivalent to environmental occurrence records.

Despite the prevalence of fibers and fragments in environmental samples, a recent review on the subject identified a concern.[186] The majority of the experiments have employed spherical particles received directly from the manufacturer, while only a few studies have tested the impact of exposure to weathered fragments and fibers.[143,208] Compared with microbeads, plastic fragments with sharp edges have a harmful impact on freshwater biota.[209] They increase the possibility of physical damaging the gastro-intestinal wall. It is worth noting that plastic particles in freshwater environments have different shapes and are often found in the form of fragments, fibers, foams, and films. To better understand the potential adverse impacts of MP and NP, studies must take into account different shapes and different realistic exposure times on different freshwater model organisms.

Several studies have investigated a variety of particle sizes, but the exposure has always been based on mass per liter. Furthermore, experimental exposure doses were often at least two orders of magnitude higher than those seen in the environment. In terms of environmental relevance, the ecotoxicity evaluation methods used in much of this research have been questioned.[186] There are numerous limitations in the experimental designs, including the lack of environmental relevance related to the exposure time; the lack of details regarding the shape, size, and concentration of the tested MPs; and the lack of detailed MP characterization such as density, size distribution and evaluation of compounds and chemicals potentially already adsorbed to MPs before exposure to daphnids.[152] Moreover, when looking at the morphology of plastic particles and the polymer types utilized in experimental investigations, there is an apparent discrepancy between what has been found in field studies and what has been employed in the experiments. The most common morphological types employed in experimental research have been spheres and beads, followed by fragments and fibers. However, field studies examining MP burden in freshwater species have revealed that fibers are the most common particle type ingested (ranging from 46.6% to 100%).[210–212] Besides, some researchers suggest that films, fibers, and fragments could be more harmful than beads.[213–215] Explanations for the use of spherical beads in most experimental studies are diverse, though the fact that they are more widely available compared with other morphological types may be a principal factor. As Ogonowski et al.[143] indicated, because the effects caused by MPs could be attributed to plastic morphology, it is recommended that future experimental studies include polymer shapes found in higher concentrations in the environment to allow for greater ecological relevance.

Most investigations using high concentrations of commercially available spherical plastic particles and short exposure durations have revealed significant toxicity. However, irregularly shaped MPs are predominant in the environment. Nonetheless, little is known concerning the multigenerational effects of irregularly shaped MPs on *D. magna*. Thus far, only one study has investigated the impact of irregularly shaped PS MPs (<63 mm) and kaolin as a natural reference particle on the survival, reproduction, and growth rate of *D. magna* over four generations under food-limited conditions. The authors suggested that exposure to high MPs concentrations reduced daphnid survival, reproduction, and growth, resulting in extinction within one to four generations.[216] Notably, kaolin exposure at identical amounts had no adverse effects. Thus, more studies with more realistic exposure scenarios are needed in this regard.

In terms of MP ingestion in freshwater organisms, fragments are thought to be more likely to induce internal abrasion. However, there is limited experimental evidence to confirm this hypothesis. So far, only one study has reported a fragment EC_{50}, namely 8.6×10^7 particles l^{-1} for *D. magna*.[143] To clarify the existence of MPs in organisms, the intake of MPs must be examined with egestion rates.[154] When calculating MP ingestion and egestion rates in freshwater daphnids, test conditions must be taken into account because, in addition to the life stage of the test species and feeding mode, the presence of food or the type of food could influence the results.[152] There are plenty of laboratory-based MP exposure experiments on invertebrate species; however, few studies have assessed whether MP consumption impacts egestion rates, especially at levels equivalent to those present in the environment[98,150,217]. Even though multiple studies suggest that MP egestion is significant, there have been a few reports of particles translocating from the digestive tract to other body tissues. Specifically, Rosenkranz et al.[41] showed in *D. magna* that 1 μm spheres translocated across the gut epithelial barrier. There are also limited studies in terms of combined contamination of MPs and other pollutants such as aromatic hydrocarbons, polybrominated diphenyl ethers, and polychlorinated biphenyls on freshwater daphnids. There are only two studies thus far that have reported co-exposure of *D. magna* to MPs and ROX. According to the studies, the effect of combined toxicity was principally dependent on the experimental method.[157,173] Ma et al.[157] exposed *D. magna* to 50 nm and 10 μm combined with phenanthrene, which is a model polycyclic aromatic hydrocarbon known to have mutagenic and carcinogenic impacts on aquatic organisms; they found greater adsorption of this hydrophobic contaminant on smaller particles. Additional studies are required to untangle the interactions between MPs and environmental contaminants.

Taken together, this review has demonstrated that the morphology, size, concentration, ingestion rate, and egestion rate of MPs have not been standardized to investigate the effects in freshwater biota. Given the technical difficulties and challenges in MP research, it is particularly vital that future studies develop standardized techniques by performing high-quality studies that examine more environmentally realistic effects to allow comparability of data. Recent omics studies have discovered that pathways associated with stress-related defense, energy metabolism, and cytoskeletal dynamics are changed in response to MPs, resulting in ingestion/ feeding and reproduction disruption.[218] For additives, omics data revealed that multiple biological processes such as oxidative/detoxification stress, energy homeostasis, lipid mechanism, skeletal development, and signal transduction are affected, which may result in reproductive toxicity, developmental abnormalities, neurotoxicity and hepatotoxicity in aquatic invertebrates. Additionally, future omics studies are necessary to evaluate the proteomic response of chemical additives and their potential leachates and address the knowledge gap at different biological levels (i.e., mechanism, transcription, protein, organism) and expand the understanding of the underlying molecular mechanisms of plastic additives toxicity and establish the linkage to adverse outcomes at the organism/population level.

7 | CONCLUSIONS

To the best of our knowledge, this is the first comprehensive review highlighting the studies of the possible ecotoxicological impacts of plastic particles on freshwater daphnids. The review, discuss and summarize the existing research trends and results in freshwater environments in the context of plastic pollution, focusing on the role of *D. magna* as an ecotoxicological model and highlights the impact of plastic pollution exposure on freshwater daphnids. In addition to reviewing the literature, we have outlined some scientific gaps in this field. The ubiquitous distribution of microplastics in global waters makes a vast range of aquatic biota susceptible to microplastics exposure. Both field and laboratory studies have demonstrated the adverse effects of microplastics on aquatic biota. However, there is an inconsistency between the conditions used in laboratory experiments and the MP characteristics that have been found in environmental samples. In most cases, researchers have only evaluated one type of plastic particle with limited comparisons with another MP type. How mixtures of different material types and size groups interact with freshwater biota remains unknown. Moreover, there is incompatibility in environmentally relevant concentrations in microplastics exposure studies. Besides, the synergic effect of prolonged and chronic exposure at different levels of biological communities has not been investigated. Hence, plastic characterization is critical to better understand the impact of plastic pollution on aquatic biota. Moreover, research on the ecotoxicological effects on freshwater daphnids has been at the individual level, and more studies are needed to examine the tissue, cell, and gene levels, including population dynamics, to elucidate the mechanisms of toxicity fully.

More studies are needed to evaluate MPs in combination with other pollutants and environmental stressors, as well as the combined effects of plastics with heavy metals and their impact on freshwater organisms. Information on the additives and their respective leachates released by plastic items is also critical to relate the overall effect to specific compounds in the mixtures and integrate/compare data. In many toxicity investigations, model organisms are subjected to leachates whose composition is unknown or only partially known. Information on additives used for the production of a given plastic article are generally not accessible. Plastics contain a wide array of known substances, but also a number of non-intentionally added substances that represent compounds not yet completely identified. Nevertheless, they are important when evaluating strategies for recycling or substitution for safer items. It is essential to assess the chemical composition of plastics items as well as their potential leachates; however, methodologies for plastic preparation greatly differ. Consequently, results from different laboratories cannot be compared. Toxicological evidence indicates that the exposure to mixtures of chemicals leached from plastics induced adverse effects. Overall, numerous gaps hinder a comprehensive hazard and risk assessment of the MPs and their leachates.

Moreover, there have been only a few papers on the effects of NPs on daphnids, mainly on the growth and reproduction of daphnids. Hence, additional ecotoxicology investigations are necessary. Thus far, the majority of microplastics toxicity studies are mainly focused on the possible harmful effects of ingested microplastics (including the associated toxicants) to aquatic fauna, especially on fish. However, knowledge about impacts of MPs and NPs exposure on other types of species, particularly aquatic primary producers, which represent a significant trophic level in the food chain, is much less known.

The form and types of MP most typically found in the environment or reported in field investigations differ significantly from those employed in laboratory experiments. Daphnids are the most studied organism group in the laboratory, and there have been comparatively few studies of other groups of organisms. This limitation is a major shortcoming in compiling standardized data considering plastic pollution in the aquatic environment.

8 | PERSPECTIVES AND RECOMMENDATIONS FOR FUTURE STUDIES

Based on the evidence presented in this review paper, it is clear that there is a fundamental gap in our knowledge of the ecotoxicology of microplastics in freshwater ecosystems. Hence, future researches should focus on the standardization and implementation of non-target methods for the chemical screening of plastic leachates, and to identify all components and eventual relationships with the effects observed in aquatic biota. Studies exploring the mechanism of action and ecotoxicological effects of environmentally relevant MP concentrations on freshwater organism health are needed. Despite some acute toxicity studies indicating negative effects on freshwater zooplankton, nothing is known about the long-term biological and ecological implications of NPs. Therefore, there is a need for further studies in this regard.

To understand how the potential future environmental problem of plastic particles may affect freshwater biota, supplementary investigations and long-term studies that employ environmental conditions should be performed. This endeavor would provide more detailed information for the potential adverse effects of MPs and NPs and could forecast future scenarios in the aquatic environment. Future studies should also examine lower plastic particle concentrations in the presence of dissolved organic matter, natural particles, and environmental microorganisms. Moreover, plastic weathering processes should be evaluated to quantify the release of plastic particles and to assess their toxic effects under more natural conditions. Future studies on daphnids in freshwater are needed, in which MPs are kept similar in terms of weathering status, type, size, and shape (by standardizing with the same groupings in different studies), as well as the exposure time. In other words, new studies are needed in which all these parameters are standardized. When there are more studies matched according to the experimental environment; MP type, size and shape, and weathering status, there would be a greater chance of supporting the findings with a meta-analysis. Collecting metadata on ecotoxicological effects in the future would help fill gaps. Studies of chemical dynamics within the gut of organisms are also needed in order to better understand the processes that govern bioaccumulation of plasticizers and co-transported chemicals.

A more comprehensive assessment of the toxicity and environmental concerns of plastic particles on aquatic biota necessitates a broader methodological approach that considers emissions into the air as well as potential consequences other than acute toxicity. Thus, setting standard measures and procedures is crucial. Education, awareness, and communication involving industries, consumers, governments, and non-governmental organizations should be commenced to allow for cooperative action and informed choices to reduce and control MP pollution in the freshwater environment. The plastic pollution problem is a complicated issue and will require organized actions, such as correct disposal, strict legislation, recycling strategies, regular evaluation, replacement of synthetic polymers with alternative materials, and ecological restoration.

AUTHOR CONTRIBUTIONS

Conceptualization; Maranda Esterhuizen, Young Jun Kim. Methodology; Afshin Samadi. Writing-Original draft preparation; Afshin Samadi. Supervision; Young Jun Kim, Maranda Esterhuizen. Writing-Reviewing and Editing; Afshin Samadi, Maranda Esterhuizen, Young Jun Kim, Youngsam Kim, Sang-Ah Lee. Visualization, Investigation; Afshin Samadi, Youngsam Kim, Maranda Esterhuizen, Sang-Ah Lee. Project administration: Young Jun Kim, Maranda Esterhuizen. Funding acquisition; Young Jun Kim, Maranda Esterhuizen. Software, Validation; Afshin Samadi, Youngsam Kim.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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