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# **Gathering at the top? Environmental controls of microplastic uptake and biomagnification in freshwater food webs**

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## **Abstract**

Microplastics are ubiquitous in the environment, with high concentrations being detected now also in river corridors and sediments globally. Whilst there has been increasing field evidence of microplastics accumulation in the guts and tissues of freshwater and marine aquatic species, the uptake mechanisms of microplastics into freshwater food webs, and the physical and geological controls on pathway-specific exposures to microplastics, are not well understood. This knowledge gap is hampering the assessment of exposure risks, and potential ecotoxicological and public health impacts from microplastics.

This review provides a comprehensive synthesis of key research challenges in analysing the environmental fate and transport of microplastics in freshwater ecosystems, including the identification of hydrological, sedimentological and particle property controls on microplastic accumulation in aquatic ecosystems. This mechanistic analysis outlines the dominant pathways for exposure to microplastics in freshwater ecosystems and identifies potentially critical uptake mechanisms and entry pathways for microplastics and associated contaminants into aquatic food webs as well as their risk to accumulate and biomagnify.

We identify seven key research challenges that, if overcome, will permit the advancement beyond current conceptual limitations and provide the mechanistic process understanding required to assess microplastic exposure, uptake, hazard, and overall risk to aquatic systems and humans, and provide key insights into the priority impact pathways in freshwater ecosystems to support environmental management decision making.

## **1. Introduction**

Besides the many technological benefits of living in the “plastic age” (Thompson, 2009), the sheer plethora of plastic products, their unsustainable use and disposal combined with their high durability in the environment cause pollution risks with widespread environmental and public health concerns (Andrady, 2009; Koelmans, 2015). A substantial increase in the worldwide production of plastics, from 2 million tons per year in 1950 to more than 335 million tons in 2016 have led to high global levels of plastic pollution (Bergmann, 2015; Dris, 2015; Eriksen, 2014). There is growing concern that waste management is largely incapable of handling the volumes of plastics being produced among the myriad disposal pathways (Koelmans, 2015), resulting in the release of large amounts of plastic waste into the environment. Up to 12.7 million tons of mismanaged plastic entered the world’s oceans in 2010

alone, and is predicted to increase by an order of magnitude by 2025 if no action is taken (Jambeck, 2015). Given that these estimates have arisen from consideration of a 50 km coastal margin only and thus, excluded riverine plastic contributions from the wider river basins, the real numbers of plastic inputs into the oceans are likely to be even higher.

While larger plastic items present a visible environmental risk, and a concern following ingestion by aquatic animals and birds (Besseling, 2015; Foekema, 2013), there is an increasing awareness and concern about the environmental and public health impacts of smaller microplastic (MP) particles of less than 5 mm size, originating from a variety of sources with different chemical properties (Avio, 2015; Koelmans, 2015). MPs, and their even smaller counterparts nanoplastics (i.e. particles < 100 nm) (Mattsson, 2015), are composed of a wide variety of polymer types resulting in distinct physical properties (i.e., in their shape, size, buoyancy). The variations in their physical properties ultimately influence particle transport and retention dynamics, their deposition, and bioavailability in the environment (Dris, 2015). The chemical and physical properties of MPs found in the environment are, in the first instance, controlled by their source. These can be classified as primary (manufactured) particles such as microbeads used in cosmetics and human care products, or secondary MPs, which result from the breakdown of larger plastic items (Figure 1). Fragmentation commonly results from industrial abrasive processes (GESAMP, 2016), or degradation via micro-cracking and embrittlement as a result of exposure to salt, wave action (Zbyszewski, 2014) and/or UV light and freezing and thawing cycles (Da Costa, 2018).

MP pollution is recognised as a global environmental threat with MPs being widely abundant in marine ecosystems, e.g. in the 5 ocean gyres, and stretching to the deep sea and remote arctic areas (Foley, 2018). While the transport, fate and behaviour of MPs have been studied predominantly in marine ecosystems, significant knowledge gaps remain in freshwater and terrestrial ecosystems (Hurley, 2018; Klein, 2015; Wagner, 2014; Windsor, 2019). MPs have been detected at alarming concentrations in river corridors and sediments (Horton, 2017; Rochman, 2018; Tibbetts, 2018). There is growing consensus that rivers are retention sites for MPs and serve as conduits for their downstream transport to the oceans (Fig. 1) (Ballent, 2016; Brennecke, 2016; Duis, 2016; Hoellein, 2019; Hurley, 2018; Klein, 2015; Lebreton, 2017; Thompson, 2004; Wolff, 2019).

Due to their unavoidable interactions with food webs they have been found in a wide range of marine organisms from phytoplankton to fishes, sea birds and even filtrating cetaceans (Fossi, 2016; Setälä, 2014). Recent meta-analyses point towards highly heterogeneous responses to

MP ingestion in different groups of aquatic organisms. For some taxa negative effects were found in terms of impacts on consumption (and feeding), growth, reproduction, and survival of fish and aquatic invertebrates (Foley, 2018; Wright, 2013). Besides the physical impacts on animals (e.g. blocking of digestive pathways, choking), MPs may be potentially toxic to animals, either through the leaching of additives (Dris, 2015; Eerkes-Medrano, 2015; Wagner, 2014), or by acting as vectors for pathogens, bacteria, viruses (Lamb, 2018) and contaminants that adsorb to their surface (Hartmann, 2017; Lamb, 2018). Ingested MPs may cause numerous adverse impacts on aquatic organisms. Even pristine plastics can cause acute immobilization, depletion of energy reserves (Wright, 2013), a decrease in predatory performance or hepatic inflammation, while MPs associated with persistent organic pollutants have been shown to cause acute or chronic toxic effects (Rehse, 2016; Rochman, 2013; Van Cauwenberghe, 2014). Critically, additives (e.g. Bisphenol A – BPA, an endocrine disrupting substance) or flame retardants such as Poly-brominated Diphenyl Ethers (PBDEs) have been shown to have increased toxicity effects on organisms when associated with MPs than when exposed in solution (Planelló, 2008; Wardrop, 2016). The reason for this is ‘Trojan horse vector-effects’ whereby MPs carry toxic additives directly into the animal’s bodies after their ingestion (de Sá, 2018; Luís, 2015; Oliveira, 2013). With recent evidence of MPs entering the human food chain (Deng, 2017; Li, 2018; Wright, 2017), and nanoplastics having the potential to penetrate cell walls of organisms (Kashiwada, 2006; Rosenkranz, 2009), regulators, water and chemical industries globally are concerned about the environmental and public health impacts of additives such as BPA and other endocrine disrupting substances such as Nonylphenol (NP) leaching from MPs (Dris, 2015; Eerkes-Medrano, 2015; Wagner, 2014). However, some studies have found that MP presence reduced the short-term effects of BPA on freshwater zooplankton (Rehse, 2018), with long-term effects still being unknown.

Despite some recent studies of MP occurrence in rivers and lakes (Ballent, 2016; Besseling, 2015; Estahbanati, 2016; Foley, 2018; Klein, 2015), and their potential effects on a wide variety of organisms (Rehse, 2016; Rochman, 2013; Van Cauwenberghe, 2015), there remains a critical lack of understanding regarding the fate and transport mechanisms of MPs and how these affect their uptake and propagation in freshwater food webs (Eerkes-Medrano, 2015; Foley, 2018). While there is initial evidence of trophic transfer in simple food chains (Chae, 2018), major knowledge gaps remain on the direct and indirect uptake and transfer of MPs in the environment, including their capacity for biomagnification while moving through complex freshwater food webs.

Assessing the risk of MP uptake and propagation in aquatic food webs requires the development of detailed understanding of potential direct and indirect MP entry points and mechanistic pathways in food webs. In addition, understanding of how MP fate and transport processes control their accumulation in freshwater habitats, and of the magnitude of site-specific exposures, is needed to enable prediction of MP bioaccumulation and biomagnification and develop appropriate mitigation measures. Here, we present here a comprehensive review and synthesis of key research challenges in:

- (i) Characterising particle properties and their ageing, along with freshwater properties such as hydrodynamic and sediment conditions that control the environmental fate, transport and development of accumulation hotspots of MPs in freshwater ecosystems;
- (ii) Analysing how physical controls on MP presence in freshwater environments determine the environmental exposures and accessibility of MPs in freshwater ecosystems
- (iii) Identifying potential uptake mechanisms and entry pathways of MPs and associated contaminants into aquatic food webs, and their capacity to accumulate and biomagnify following initial entry.

We identify 7 key research challenges (Table 1) that represent milestones in understanding the controls of exposure, uptake and propagation of MPs in aquatic food webs, and discuss these in further detail below.

## **2. Fate and transport mechanisms control microplastic exposure**

The exposure and potential entry points of MPs into aquatic food webs are determined by the spatial distributions and time scales of mechanisms that control the input, transport and transformation as well as the potential accumulation of different types of MPs in the environment. An assessment of MP uptake and potential propagation through aquatic food webs therefore requires detailed understanding of MP sources and their activation, as well as of the environmental fate and transport mechanisms of MPs (Fig 1).

MPs enter aquatic environments through a wide range of pathways, including runoff from land surfaces and roads, river bank waste disposal, atmospheric deposition and outfall of wastewater treatment plants (Fig 1) (Dris, 2016; GESAMP, 2016; Horton, 2017; Horton, 2018b; Wagner, 2014). While it has been shown that wastewater treatment plants can retain significant amounts

of MPs and are efficient to filter out some types of primary MP particles (Murphy, 2016; Prata, 2018; Ziajahromi, 2016), there is evidence that they are less effective in the retention of synthetic microfibers and nanoplastics (Rochman, 2015). Other MP entry pathways into terrestrial and aquatic ecosystems include agricultural sources including organic fertilizer products (Weithmann, 2018), plastic in potting soil, such as tree ties in forestry and plastic tarps on agricultural fields (Fig 1). Recognition of the specific entry pathways and related differences in transport mechanisms is crucial since the environmental fate of highly concentrated point source inputs (e.g. from wastewater treatment plants) is likely to differ significantly from lower concentration diffuse inputs (e.g. atmospheric fallout, road runoff or agriculture) (Fig 1).

Understanding of the chemical and physical properties of MPs, as well as how the hydrodynamic and substrate conditions at the sediment-water interface control MP transport in lakes and rivers is still very limited. Recent studies have hinted that MPs are omnipresent in freshwater ecosystems, and in particular at sediment-water interfaces (Besseling, 2015, 2018; Foley, 2018; Hurley, 2018), indicating that benthic and hyporheic zones of rivers may represent MP accumulation hotspots (Drummond et al., 2020; Frei et al., 2019) (Fig 1). Depending on their density and buoyancy, MPs of different origin are likely to differ in their transport properties, their probability of burial in riverbed sediments and thus, the types of interactions with different trophic levels of aquatic organisms. For instance, in contrast to the high-density polymers found in benthic and hyporheic accumulation zones in many rivers (Besseling, 2018; Foley, 2018; Hurley, 2018), most of the MPs polluting the water column of the Canadian Great Lakes were found to be low density synthetic polymers (polyethylene, polyester) (Ballent, 2016). Similar results were found within large river systems such as the Danube and Rhine rivers (Lechner, 2014; Mani, 2015). Yet, the fact that low density MPs have also been found in streambed sediments highlights the fact that the mechanisms which control deposition and entrapment (Hoellein, 2019), including potential interactions of the bed substrate and its associated benthic flora, remain to be quantified. While there is increasing understanding and insight into the relevance of the specific mechanisms controlling MP transport in freshwater ecosystems such as hyporheic exchanges (Drummond et al., 2020; Frei et al., 2019) or gravitational settling, including governing equations and quantitative models (Box 1), identifying the required parameters across the relevant spatial and temporal scales remains challenging.

Furthermore, the spatial patterns and temporal dynamics of proportional contributions of different types and sources of MPs still need to be determined, in order to parameterise transport and exposure models. The vast majority of freshwater MP studies to date have been conducted under steady flow conditions, and little attention has been paid to analysing the impact of hydrodynamic conditions at sampling sites (Fig 2). We have only just started to explore how the temporal dynamics of MP fragmentation, deposition, degradation and eventually removal are controlled by hydrodynamic variability (Drummond et al., 2020; Hurley, 2018), with the balance between these processes being defined by the local exposure time. It will be critical to identify how the hydrodynamic forcing and the variability in physical and chemical properties which affect MPs (Box 1) may change with dynamic flow conditions (e.g. temperature, nutrient content, UV exposure). It has yet to be established how this variation in flow may affect the deposition (Tibbetts, 2018) and transformation of MPs, such as their breakdown and decomposition or release of additives such as BPA and NP (Fig 2). This includes the distribution of MPs within the floodplains of rivers during high flow events as well as their behaviour under drastically different environmental conditions (i.e., when exposed to air during dry conditions in ephemeral streams or on elevated river banks (Fig 2)). Indeed, with globally increasing frequency and severity of droughts (Hoellein, 2019; Naumann, 2018), intermittency and flashiness of streams, understanding of the impact of stream drying and increased UV exposure on MP degradation will become increasingly important, as will understanding the disturbances to sediment-entrained MPs due to turbulent mixing as streams re-wet.

The transport mechanisms and accumulation of MPs (Box 1) are furthermore affected by biological processes, including MP particle ageing and decay as well as biomolecule adsorption and biofilm colonization (i.e. accumulation of micro-organisms on wetted MP surfaces) (Harrison, 2018; Nasser, 2016), and entrapment in biofilms at aquatic-terrestrial interfaces (Fig 2) (de Sá, 2015; Duis, 2016; Rummel, 2017). Biofilms growing on the surfaces of ageing MPs can change their hydrodynamic and buoyancy properties, potentially affecting their transport, fate, accumulation and uptake by organisms (Corcoran, 2015). The magnitude of such biological interactions and their dependency on environmental conditions such as temperature, light exposure and nutrient concentrations have yet to be established. Additionally, the formation of homo-aggregations (microfiber clumps) and hetero-aggregations (similar to marine snow) may also have an effect on the fate and transport of MPs (Alimi, 2018). Important lessons can be learned here for instance from recent advances in understanding MP settling

velocity in marine environments (Khatmullina, 2017), or using mechanistic behaviour of naturally occurring allochthonous particles as analogues (Hoellein, 2019). However, the stability of MPs and their likelihood to form aggregates may be different in freshwater versus saline sea water and will depend on the polymer type (Lagarde, 2016; Michels, 2018).

Addressing the interdisciplinary challenges of establishing how the fate and transport of MPs control their entry into freshwater food webs will require:

- The characterisation of spatially heterogeneous MP sources of different plastic types and how dynamic source activation controls the way MPs are released into the environment [*Challenge 1*] as well as,
- The identification of how MP properties, and freshwater hydrodynamic and biological processes interact in affecting MP fate and transport, including their degradation, ageing, break down and the release of toxins [*Challenge 2*] (Table 1).

For these challenges to be addressed successfully, scientific advances across traditional disciplinary boundaries are necessary that require to leave our current disciplinary ‘comfort zones’ in the material sciences and analytical chemistry, ecohydrology, environmental chemistry and biology (Table 1).

### **3. Microplastics entry points into freshwater food webs**

Once they have entered freshwater environments, there is a risk that MPs may be taken up into freshwater food webs. The location and mechanism for MP entry into freshwater food webs is determined by site- and material-specific conditions. Recent investigations have focussed on the discrimination between various routes of entry according to whether the MP particles were primary or secondary MPs (Ballent, 2016; Dris, 2015; Duis, 2016; Weithmann, 2018).

Potential MP entry points through direct uptake into aquatic food webs exist at almost every trophic level and may involve suspended MPs that are filtered from the open water column or MPs accumulated in the benthos at the sediment-water interface (Fig 3). Phytoplankton can directly adsorb nanoplastics through cell-walls and zooplankton such as daphnia and fish can ingest nanoplastics and MPs directly from the water column (Costa, 2016). Lacustrine invertebrates such as oligochaete worms, amphipods, and freshwater snails can ingest artificially manufactured plastic microspheres under laboratory conditions (Imhof, 2013). However, so far only limited empirical evidence of MP uptake mechanisms and trophic transfer by freshwater fauna exists, as only isolated cases of direct ingestion under laboratory

conditions, or detection in the wild (without knowledge of the specific plastic sources) have been reported (Imhof, 2013; Pazos, 2017).

It is plausible that benthic fauna, especially filter feeders, like zebra and quagga mussels (*Dreissena sp.*) will ingest MPs suspended in the water column, with the risk of transferring them to the benthic food chain via coating in faeces or pseudofaeces (Lederer, 2006) (Fig 3). Pseudofaeces of *Dreissena* and other common freshwater bivalves (*Corbicula sp.* and Unionidae mussels) are rejected organic and inorganic particles covered by mucus in the mantle cavity of the mussel, and expelled outside (Dermott, 2005). It is well established that many benthic animals prefer to feed on *Dreissena* pseudofaeces because of their high nutritional values (Stewart, 1998). Due to their high biomass, filter feeding benthic macroinvertebrates such as bivalves may therefore be responsible for significant transfer of MP from the water column to benthic food webs.

A further preferential pathway of MPs to enter freshwater food webs is through their entrapment in benthic or hyporheic biofilms (Sgier, 2016), where they accumulate and are potentially made more readily available to other organisms. Here they can pose a critical risk for preferential direct MP uptake and subsequent ingestion by biofilm grazers (Fig 3) (McCormick, 2016). In marine environments, organic aggregates (so called marine snow) can trap MPs and transport the normally buoyant plastics to the deep ocean, ultimately increasing their bioavailability to deep-sea fauna (Porter, 2018).

So-called Trojan horse vector-effects may potentially increase the indirect uptake of xenobiotics from MPs, for instance when additives such as Bisphenol-A (BPA) leach from the biofilm after ingestion by biofilm grazers (Fig 3). Furthermore, biofilms on MP surfaces may create shortcuts for MPs to enter food webs at several trophic levels (Rummel, 2017). In addition to Trojan horse effects, MPs may affect the structure and metabolic functionality of biofilms (Harrison, 2018; Rummel, 2017). Since biofilms carry out a number of crucial ecosystem services in rivers, such as respiration, nutrient uptake and attenuation of pollutants, any MP induced damage to biofilms bears the risk of impacting ecosystem functioning at the river scale (Besseling, 2018; Harrison, 2018).

Beyond physical controls on MP fate and transport, it is important to advance the understanding of how animal behaviour and feeding types affect MP uptake and ingestion, which may vary by development stage and morphological characteristics (Scherer, 2017). For example, De Sá (2015), showed that the common goby (*Pomatoschistus microps*) ingests MP particles (420–

500 µm) of different colours together with natural prey (*Artemia salina*) (de Sá, 2015). Here, the selectivity of MP uptake was dependent on the particle colour, with similar behaviour found for sunfish (Peters, 2016). Similarly, benthic sea turtles showed strong selectivity for soft, clear plastic, suggesting that sea turtles ingested plastic because it resembled natural prey such as jellyfish (Schuyler, 2012). In contrast, pelagic turtles were found to be less selective in their feeding, though they showed a trend towards selectivity for rubber items such as balloons (Schuyler, 2012).

In order to assess the entry point of specific risks of MP uptake and their dependence on environmental exposure conditions, MP exposure hotspots (e.g. through filter feeding or biofilm entrapment) need to be identified and mechanisms of potential uptake pathways characterised and their relative contributions to total organism body-burdens quantified [**Challenge 3**] (Table 1). This includes uptake by phytoplankton as well as MP entrapment in aquatic biofilms, presenting a potential pathway for increased MP uptake by biofilm grazers and entry into freshwater food webs. The quantification of potential environmental and health consequences will require:

- Determination of the potential for Trojan Horse effects or shortcuts for xenobiotics into aquatic food webs [**Challenge 4**] and
- Quantification of the importance of ecological behavioural aspects (from colour preferences to bioturbation) that affect MP abundance, distribution and uptake [**Challenge 5**] (Table 1).

#### **4. Migration and potential biomagnification of microplastics in freshwater food webs**

Once MPs and nanoplastics enter freshwater food webs, there is a risk of biomagnification into higher trophic levels (Sanchez, 2014). The potential for biomagnification is generally not yet well established. Previous studies on MPs in freshwater food webs have been either limited to a single species (Rehse, 2016; Van Cauwenberghe, 2015), or trophic transfer in simple food chains (Chae, 2018; Rochman, 2017). Possible transfers across more complex aquatic food webs have not yet been assessed.

Direct ingestion and trophic transfer of MPs have been confirmed in several field and laboratory experiments (Kim, 2018; Ziccardi, 2016) and repeated occurrence of MP particles in fish have been explained by both direct ingestion and trophic transfer (Phillips, 2015). Indeed, fish have been most widely studied to date, with findings ranging from an absence of

MPs (Lake Geneva) (Faure, 2012), to 8% of the freshwater fish and 10% of marine fish in the Gulf of Mexico (Phillips, 2015), 12% of adult gudgeons (*Gobio gobio*) in 11 French streams (Sanchez, 2014), 45% of all sunfish (*Lepomis gibbosus*) specimens in the Brazos river (Texas, USA) (Peters, 2016) and 55% of Nile perch (*Lates niloticus*) and Nile tilapia (*Oreochromis niloticus*) from Lake Victoria (Biginagwa, 2016). Despite the growing number of case studies evidencing MP presence in fish (Collard, 2018; Horton, 2018a), there is little understanding of the mechanisms and drivers of their uptake and propagation in aquatic food webs. It has been suggested that the feeding behaviour of different species appears to significantly affect their risk of MPs ingestion. Given high benthic concentrations of MPs relative to water column concentrations, benthivore fish may have a greater MP exposure than planktivorous fish (Fig 3) (McNeish, 2018). The absence of MPs in the fish studied in Lake Geneva as compared to gudgeons in French streams may be explained by a difference in the feeding preferences of the different species. Gudgeons are primarily benthivores, thus might be exposed to a higher risk of MP ingestion than carnivorous, planktivorous or herbivorous fish (Sanchez, 2014).

Simplified food chain studies have demonstrated that when MPs are abundant, they can be readily transferred along trophic levels (Ivar do Sul, 2014). Trophic transfer of fluorescent plastic microspheres from *Artemia nauplii* to zebrafish (*Danio rerio*) (Batel, 2016) and Spined Stickleback (*Gasterosteus aculeatus*) was shown in laboratory studies (Katzenberger, 2015) where fish were fed with MP-exposed Nauplius (crustaceous larvae). Both, MPs and persistent organic pollutants associated with MPs such as BPA, have been shown to migrate to fish tissue (Batel, 2016), highlighting the risks of Trojan Horse effects. Furthermore, direct trophic transfer from mussels (*Mytilus edulis*) to crabs (*Carcinus maenas*) has been observed under laboratory conditions, with plastic microspheres being transferred into the crab's haemolymph, stomach, hepatopancreas, ovary and gills (Farrell, 2013).

Despite the mounting evidence of MPs within all levels of freshwater food webs described above, evidence for the transfer among different components of food webs is limited. It will be crucial to identify and understand the mechanisms of MP uptake and pathways in more complex food webs than previously studied in order to understand what risk potential biomagnification presents for human consumption [**Challenge 6**] (Table 1). In this respect it will be essential to understand how MPs are retained in tissues over long time scales, or if they are largely expelled during egestion, growth, or metamorphosis. For example, Al-Jaibachi et al. (2018) showed that fluorescently stained MP beads taken up by mosquito larvae remained after metamorphosis in the winged adults. This suggests that long term retention of MPs in

tissues is possible, and that insects with aquatic larval stages and terrestrial adult stages may represent an underappreciated flux of MPs among aquatic and terrestrial ecosystems (Al-Jaibachi, 2018).

## **5. The ecological and public health impacts - Microplastic effects on aquatic ecosystem functioning**

The effects of MPs on freshwater ecosystem functioning remains largely unknown (Eerkes-Medrano, 2015; Lambert, 2016). Laboratory studies confirmed that water fleas (*Daphnia magna*), freshwater invertebrates and several fish species actively feed on MP particles (1-100 µm), and that uptake of MP particles caused acute immobilisation of these animals (Besseling, 2018; Oliveira, 2013; Rehse, 2016), and affected predator–prey relationships (Rochman, 2017). However, some studies also found MP impacts on ecosystem processes to be negligible (Canniff, 2018; Malinich, 2018; Redondo-Hasselerharm, 2018; Weber, 2018), complicating predictions of ecosystem level implications. Furthermore, transgenerational effects on *Daphnia magna* indicated only negligible effects on the first generation but when neonates were exposed to same concentration of MPs they went extinct after 2 generations (Martins, 2018). This highlights the need to expand testing further than the first generation. While it is reasonable to expect that some ecosystem effects occur at longer time scales or larger spatial scales, their specific relevance has yet to be established.

Many benthic consumers in freshwaters (e.g., Chironomidae larvae, Oligochaeta worms, gammaridae, amphipods) act as ecosystem engineers in sediments and are highly exposed to MPs, their chemical additives, sorbed contaminants, and potential microbial pathogens (Frère, 2018; McCormick, 2014), resulting in a critical risk of wide ranging impacts in particular on benthic ecosystem functioning (Izvekova, 1972; Ward, 2007). For example, lugworms (*Arenicola marina*) that ingested MPs showed decreased bioturbation (Wright, 2013), which lowered the primary productivity of bioturbated sediment and altered lugworm respiration (Green, 2016). Similar impacts on key bioturbators of freshwater ecosystems could have significant consequences at the ecosystem level. In shallow lakes, filter-feeding benthic species such as *Chironomus plumosus* larvae filter large volumes of water, importing oxygen and particulate matter down to 20 cm into the sediment and regulate lake's phosphorus concentrations and trophic status (Hölker, 2015; Lewandowski, 2007; Roskosch, 2011). Thus,

any reduction of the activity of *C. plumosus* larvae due to MPs might severely impact on the entire lake ecosystem.

In addition to identifying pathways and mechanisms of how MPs enter and propagate within aquatic food webs, future research should establish how freshwater MPs impact the behaviour and performance of organisms and key ecosystem processes. We suggest new research to move beyond the assessment of individual species interactions with MPs. The next frontier in this research will measure MP effects on food webs and ecosystem functioning, as well as quantifying the potential combined impacts of MPs and their associated additives, sorbed contaminants and pathogens [**Challenge 7**] (Table 1).

## **6. Concluding remarks and a look forward**

Increasing evidence and rising public awareness of the omnipresence of MPs in understudied freshwater ecosystems and within humans at the end of the food chain (e.g. identification of MPs in human stool samples (Parker, 2018)), highlight that scientific interactions and knowledge exchange across disciplinary boundaries are critical for evaluating the environmental and public health risks of MPs. There are increasing concerns of MP uptake through food and bottled drinking water or by inhalation of microfibers from clothing or soft furnishings (Catarino, 2018), in particular with the proven transport of microplastics in the atmosphere along large distances (Allen, 2019). The challenges we present are interlinked and require interdisciplinary collaboration among aquatic ecologists, (eco)hydrologists, fluvial geomorphologists, biogeochemists, analytical chemists and microbiologists, cell biologists and physicians. The seven major challenges outlined here will help interdisciplinary researchers to facilitate collective goals and direct future systematic analyses of MP fate and transport controls on potential entry points and uptake mechanisms into aquatic food webs. The resulting knowledge will enable confirmation of the major propagation and biomagnification routes and subsequent organismal and ecosystem impacts of MPs in freshwater ecosystems and determination of the relative risks to human health, allowing targeted mitigation strategies with maximum impact to be developed.

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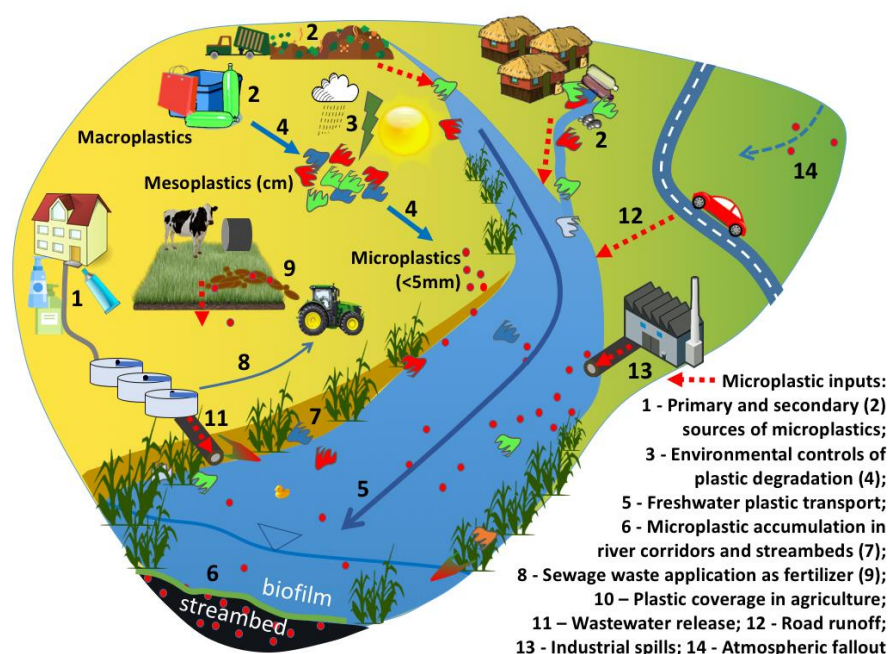
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### Author contributions

All co-authors contributed to the drafting of the manuscript, and all have approved the final version.

### Figures:

Figure 1 Mechanisms of plastic fate and transport processes across the river basin continuum, highlighting sources, input pathways and accumulation and breakdown mechanisms.



**Box 1 Quantification of key microplastic transport processes in freshwater systems:** While many of the processes that control the dynamics of small, light, organic particles in freshwater systems are reasonably well understood, understanding of which specific processes and system conditions dominate the fate of different types of microplastics is still lacking. Consequently, microplastic transport and ecological uptake can only be partially quantified at the present time. The current approaches used in modeling microplastic transport processes are illustrated in Figure 2. Models for *microplastic transport and retention in streams* currently assume that Stokes' settling velocity describes the transport of microplastics to the sediment bed. Stokes' settling velocity depends on particle density, size, and shape (Chubarenko et al., 2016; Dietrich, 1982; Kooi et al., 2017). Hydrodynamic transport – specifically *hyporheic exchange* – also delivers microplastic to the streambed, and this process is generally more important than settling of microplastic, especially for smaller particles (Drummond et al., 2020). Hyporheic exchange is caused by flow interactions with sediment beds and benthic algal mats, and is generally proportional to the square of in-stream velocity (Arnon et al., 2010; Boano et al., 2014; Packman et al., 2000). *Remobilization* of microplastics occurs during both baseflow and stormflow (Drummond et al., 2014; Drummond et al., 2017; Phillips et al., 2019), and remobilization has been modeled as a function of stream power, bed friction factor and particle size (Nizzetto et al., 2016). *Filtration, aggregation and other immobilization processes* within the sediments have been observed in laboratory columns and the processes parameterized in ways that can be incorporated into larger-scale models (Alimi, 2018). *Bioturbation* mixes particles in sediment beds, and models are available to describe both the burial of microplastic and their remobilization to the sediment-water interface (Roche et al., 2016). *Photodegradation* of microplastic has been measured in laboratory experiments and been shown to be both polymer-specific and flow-dependent (Hebner and Maurer-Jones, 2020). Note that episodic burial/remobilization will definitely influence the amount of time microplastics are exposed to light vs. benthic/hyporheic biodegradation; while this is unlikely to have been observed directly *in situ*, the significance of this process is known (Ward et al., 2015). Biochemical pathways of polymer biodegradation by bacteria and fungi are highly complex, and include polymer hydrolysis and oxidative degradation, which can transform both hydrolyzable and nonhydrolyzable polymers (Yuan et al., 2020). *Uptake and bioaccumulation* of microplastics and nanomaterials in organisms is dependent on particle size and species-specific uptake rates that vary based on their developmental stage (Fueser et al., 2019; McNeish et al., 2018; Scherer, 2017; van den Brink et al., 2019). The role of atmospheric transport and disposition as routes of environmental exposure and transport are just beginning to be explored (Lim et al., 2020). The parameterization of the governing equations in aforementioned models for quantifying MP fate, transport, degradation and uptake processes remains a challenge, with further experimental evidence needed to evaluate the range in polymer types and environmental conditions which alter each of the rates quantified for each process.

Figure 2 Physical (A), and biological (B) controls of microplastic fate and transport in freshwater ecosystems, including (C) impacts of flow variability and increased temperature during low-flow conditions and flow intermittence on microplastic accumulation and breakdown

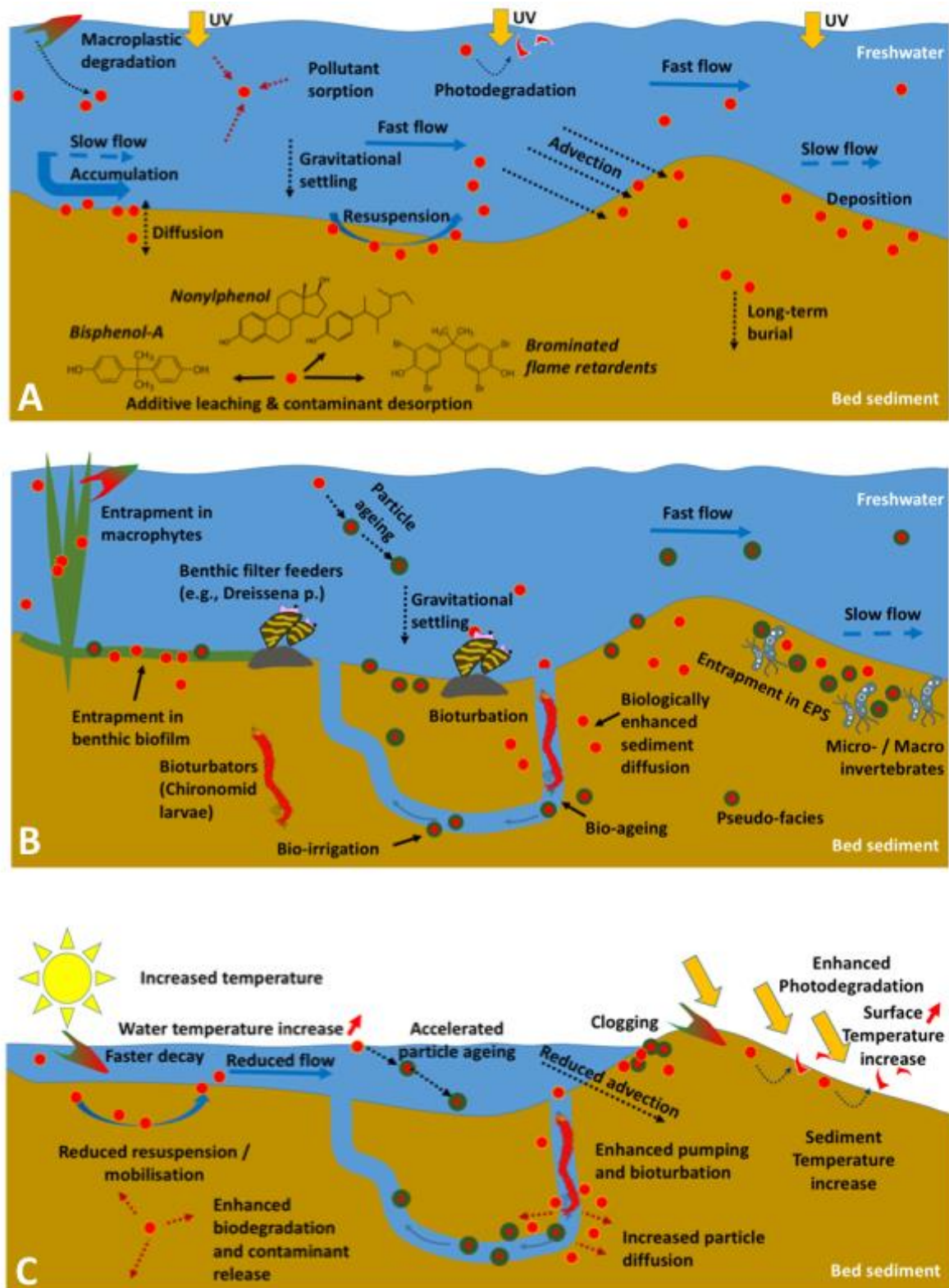
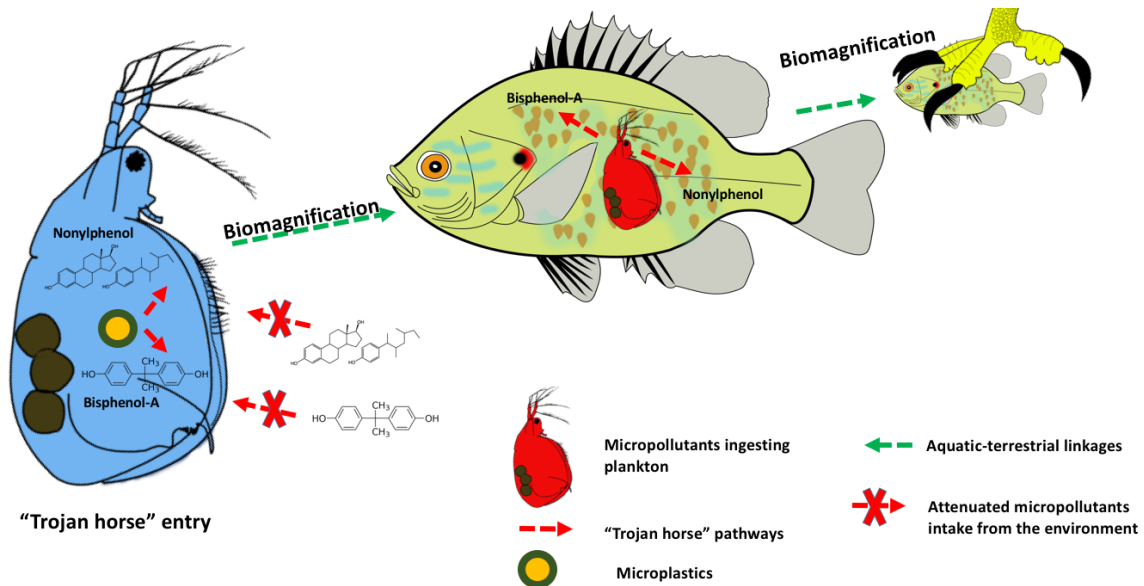


Figure 3 Microplastic uptake and propagation in an example aquatic food web, including leaching of additives and sorbed contaminants as part of Trojan horse effects, and potential biomagnification pathways



	Research need	Scientific advances needed to achieve this
<b>Challenge 1:</b>	Characterize the spatially heterogeneous sources of different plastic types and how their activation and contribution to river corridor plastic pollution vary over time.	<p>Agreed and validated methods and protocols for sampling, identification and quantification of MPs of different compositions and shapes</p> <p>Global baseline of MP abundance in freshwater ecosystems and characterisation of MP properties.</p> <p>Temporally and spatially dynamic sampling strategies that support identification of event-based (drought, flooding and re-wetting) inputs of MPs from point versus diffuse sources</p>
<b>Challenge 2:</b>	Establish how microplastic properties, hydrodynamics, and biological processes affect the fate and transport of microplastics, their degradation, ageing, breakdown, and the release of toxins	<p>Analytical tools for assessment of MP ageing, degradation, and determination of the relative contribution of different processes (e.g. abrasion, UV-irradiation) on degradation as a function of MP composition (e.g. shape, density)</p> <p>Lab-assays for assessment of release of additives from MPs under ecologically and biologically relevant conditions (ionic strengths, pH, natural organic matter and abundance of other biomolecules)</p> <p>Flume and field-scale approaches for assessing the impacts of hydrodynamics, geological setting on accumulation of MPs in sediment; agreed sampling methods will be needed here also to allow comparisons of different hydrological conditions</p> <p>Full characterisation of MP ageing of different polymer products under the influence of variable environmental conditions, and re-evaluation using aged MPs where relevant</p>
<b>Challenge 3:</b>	Quantify microplastic exposure hotspots and mechanisms of uptake pathways into food webs	<p>Based on knowledge from Challenges 1 and 2 confirm hotspots of MP accumulation, and characterise conditions for deposition and accumulation; and for MP degradation</p> <p>From these determine organisms at specific risk, and undertake targeted lab and mesocosm studies to determine if these organisms do preferentially accumulate MPs (pristine versus variously aged and bio-fouled)</p> <p>Laboratory experiments of food webs of increasing realism to assess role of amongst others, food, water, natural organic</p>

		matter, pseudofaeces on MP uptake and accumulation in target organisms. Again, comparison of pristine and variously aged and bio-fouled MPs is needed.
<b>Challenge 4:</b>	Identify potential Trojan Horse effects and other shortcuts for xenobiotics into aquatic food webs	<p>Lab-based experiments under realistic exposure conditions (e.g. presence of natural organic matter and other competitive binders) to assess affinity of pollutants for MP surfaces and residence times – comparison of pristine and variously aged and bio-fouled MPs binding affinities, loading capacities and retention times</p> <p>Studies mimicking gut pH of target organisms to assess release of additives from the MPs before / during / following uptake into target organisms, including biofilms again considering pristine and aged/bio-fouled variants</p> <p>Investigation of the role of natural organic matter as a sink / source for additives / xenobiotics and how this influences MP Trojan Horse effects</p>
<b>Challenge 5:</b>	Determine the importance of ecological behavioural aspects (from colour preferences to bioturbation) that affects microplastic abundance and uptake.	<p>Grouping of recognition mechanisms and food preferences of different organisms and determination of correlations with different types / sources of plastics and their resulting MP degradation products</p> <p>Targeted lab-based experiments to confirm if preferential uptake occurs under realistic exposure conditions, i.e. in the presence of equal or larger quantities of test organisms preferred food sources and whether aged or bio-fouled MPs are taken up preferentially</p> <p>Analysis of MP abundance effects and thresholds relative to natural food sources to determine if tipping point effects are likely</p>
<b>Challenge 6:</b>	Identify microplastic propagation pathways in	Mesocosm experiments with complex food-webs and realistic environmental and ecological conditions, including also realistic

	complex food webs and assess risks of potential biomagnification for human health	hydrological conditions (e.g. using recirculating streams) and aged / bio-fouled MPs  Comparison / quantification of various uptake routes – direct exposure, indirect exposure via food / pseudofaeces – and their relative contributions to total uptake under food abundant and food scarcity scenarios
<b>Challenge 7:</b>	Assess the impacts of freshwater microplastic accumulation and associated additives, sorbed contaminants, and pathogens on the behaviour and performance of host organisms and key ecosystem functioning	Ecotoxicity tests related to target organisms and under the realistic and competitive exposure conditions outlined above with appropriate controls for the various additives that can potentially be released  Spiking experiments for additives to assess co-exposure effects, again under realistic / competitive conditions since the target xenobiotic will not be the only species that binds to the MP surface under environmental conditions  Co-investigation of MPs and flow conditions (flooding/drought / riffles for deposition etc.) on bioavailability, uptake and retention of MPs.  Assessment of wider ecosystems services, such as water purification by biofilms, bioturbation by benthic species etc.

**Table 1** The key research challenges whose resolution will represent milestones in understanding the controls of exposure, uptake and propagation of microplastics in aquatic food webs and selection of scientific advances required to address those key challenges.

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