Micro- and nanoplastics in wastewater treatment systems and receiving water

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Acronyms

COD	Chemical oxygen demand
FT-IR	Fourier-transform infrared spectroscopy
MNP	Micro- and nanoplastics
MRI	Magnetic resonance imaging
p.e.	Population equivalent
PE	Polyethylene
Pest	Polyester
PET	Polyethylene terephthalate
PMMA	Polymethyl methacrylate
PP	Polypropylene
PS	Polystyrene
PTFE	Teflon
PVC	Polivynil chloride
SEM	Scanning electron microscopy
TEM	Transmission electron microscopy
WWT	Wastewater treatment
WWTP	Wastewater treatment plant

1 Introduction and Literature Review

1.1 Summary

Plastic waste is a widespread and persistent global challenge with negative implications for the environment, economy, human health and aesthetics (Jeftic et al., 2009). Plastic pollution has been a focus of much environmental research over the past few decades, particularly in relation to macroplastics that are easily visible by the naked eye. However, there has been greater concern in recent years for smaller debris at the micro and nano scales. Although current studies have contributed to the advancement of knowledge on the source, distribution, fate and impact of microplastics, most have focussed in the marine environment. Scarce knowledge is available for freshwater systems, especially free-flowing waters that are known to serve as important transport vectors of land-based pollutants to oceans. While microplastics research is still in its emerging stage and various knowledge gaps still remain, as a pollutant, management of inputs is essential to avoid negative impacts. Thus, the role of wastewater treatment systems and natural fluvial vectors in delivering these emerging contaminants to the environment should be considered. Understanding fundamental aspects pertaining to the sources, distribution, degradation, transport and removal of microplastics in these systems is essential to develop effective strategies to mitigate the discharge of these particles to the sea.

1.2 Background and Rationale

Plastic waste is pervasive and increasing in land and water environments globally. In 2013, global plastics production was estimated at 299 million tonnes, a 3.9 % increase from 2012 (Plastics Europe, 2015). In the EU, Germany and the UK are the two highest producers of plastic waste, recovering 80 % and 26 % of it, respectively (Hartl et al., 2015). Most of this plastic is non-biodegradable and remains as waste in the environment for a long time (European Commission DG Environment, 2011), with approximately 10 % ending up in the oceans (Thompson, 2006). Plastics are lightweight and buoyant, and easily transported long distances across a wide range of environments (Coe and Rogers, 1997), rendering them ubiquitous contaminants. Previous research from shoreline and beach surveys across all continents indicate that plastic waste commonly accounts for 50-90 % of all marine litter (Derraik, 2002), of which 80 % originates from land-based sources (GESAMP, 1991; Coe and Rogers, 1997; Andrady, 2011), highlighting the role of fluvial systems as important transport routes of these contaminants to the sea. However, compared to marine systems, data for freshwaters remains limited, and the magnitude of their impact is yet to be assessed (Eerkes-Medrano et al., 2015).

The emphasis on plastic pollution research in oceans may be because, until recently, its accumulation and impacts appeared to be more evident in these environments (Ryan et al., 2009). For example, "patches" of accumulated floating macroplastic debris were observed in gyres and convergence areas in oceans over a decade ago (e.g. Pacific garbage patch; Moore et al., 2001; Ryan et al., 2009; European Commission, 2011), garnering widespread attention of the media, policymakers, and the scientific community (European Commission, 2011). Furthermore, the marked mechanical effects of plastic litter on marine biota due to entanglement and ingestion raised concerns of its potential harms to biodiversity and ecosystems (Derraik, 2002; Ryan et al., 2009; Thompson et al., 2009). While oceans have been used as waste dumps for years (despite global efforts to prevent this; Gordon, 2006), the majority of plastic litter is produced inland, thus examining their transport to marine environments by rivers can allow for identification and regulation of its main sources (Ryan et al., 2009; Dris et al., 2015).

At present, the increased awareness of the growing production and accumulation of plastic pollution in the environment has brought greater focus to the need for development of policies and management strategies. The United Nations Environment Programme (UNEP), for instance, called for an urgent need to address plastic pollution of oceans through implementation and enforcement of coordinated strategies, effective policies and regulations, campaigns, and other incentives at national, regional and global levels (Jeftic et al., 2009). The European Marine Strategy Framework Directive (MSFD) 2008/56/EC emphasised the need for more data on the amount, distribution, and composition of plastic debris (Galgani et al., 2011; Sadri and Thompson, 2014). But despite the extensive research devoted to monitoring plastic debris over the past decades, the full extent of its quantity, distribution, and impact remains widely unknown. Thus, the problem of controlling plastic waste may be confounded by lack of measurement of the extent and thus understanding of source and impact, rather than strategy (Coe and Rogers, 1997). For example, the importance of plastic fragments at the micro and nano scales has only recently been recognised, and method development to define and measure them is still under way. Microplastics and nanomaterials have been classified by Scotland's Centre of Expertise for Waters (CREW) as emerging contaminants, or alternatively, as "contaminants of emerging concern" (CEC) for Scottish watercourses, due to the lack of adequate data for reliable risk assessment and their toxic characteristics (Hartl et al., 2015). Therefore it is essential to refine the initial estimates of plastic debris in oceans and inland waters to include these smaller and "invisible" fractions and identify their main sources before further actions or regulations can be implemented.

The concepts of micro- and nano-sized plastics as emerging contaminants, and the role of wastewater and freshwater systems as sinks or sources of these

materials to the environment provide the underlying motivation of this study. The purpose of this report is to revise and summarise theory and literature relevant to the topic of microplastics and nanoplastics pollution in freshwaters and wastewater systems, the selection of methodology, and the definition of research objectives. The first section explores the concepts of micro- and nanoplastics, as defined by various authors, leading into a discussion of the current knowledge relating to these plastic debris categories in freshwater systems in the following section. Next, a brief overview of wastewater treatment systems in the UK is included to set the conceptual framework to explore their potential role as transport routes of plastic debris to rivers, and conversely, the impact of these materials on treatment plant efficacy. The following section focuses on the subject of method development by providing a quick overview of techniques and approaches used by various authors, focusing on those relevant to the expected experimental plan for this study. The introductory chapter ends with the statement of purpose and specific objectives that will be addressed in this PhD research.

1.3 Microplastics

Plastic litter can occur in a wide range of sizes, characterised differently by different authors. The literature commonly distinguishes between two broad classes of plastics: macroplastics (>5 mm) and microplastics (< 5 mm) (Arthur et al., 2009; Thompson et al., 2009; Faure et al., 2012; GESAMP, 2015), but different terms and size ranges have been used across studies (**Table 1**). Furthermore, there does not seem to be a unified lower limit for measurement of microplastics, although for practical purpose, 333 µm (~0.3 mm) is often used when sampling with neuston nets (Arthur et al., 2009; Roex et al., 2013). In more recent papers, the term "nanoplastics" has been introduced (Besseling et al., 2014; Wagner et al., 2014; Koelmans et al., 2015). This size class has been defined as particles smaller than 0.2 mm based on the WG-GES size classification (Wagner et al., 2014); and, smaller than 100 nm according to the general definition used for nanomaterials (Koelmans et al., 2015). However, nanoplastics have been mostly overlooked in the literature, evidenced by a lack of discussion of its definition and quantification. But despite being the least-studied, it has been suggested that this category may be the most hazardous (Koelmans et al., 2015), requiring further investigation. For purposes of this project, the generalised definition of microplastics will be used, referring to particles or fibres in the range of <100-5 mm in size, while nanoplastics will be defined as fragments between 1-100 nm across. The rest of the discussion will focus on micro- and nanoplastics (MNP) jointly as a single size class for ease of reference.

Table 1 Definitions of plastic size classes in the literature

Preffix	Size Class	Size Range	Source
nano	nano, micro, millimetre (NMM)	not available	Besseling et al., 2014 (abstract)
	nanoplastic	< 0.2 mm	Wagner et al., 2014
		< 100 nm	Koelmans et al., 2015
micro	microlitter	~0.06 - 0.5 mm	Gregory and Andrady, 2003
	microplastic	< 0.5 mm	Thompson et al., 2004
			Fendall and Sewell, 2009
			Hoellein et al., 2014 (abstract)
			Sanchez et al., 2014
			Corcoran et al., 2015
		0.333 - 5 mm	Arthur et al., 2009
	micro debris	< 2 mm	Lechner et al., 2014
	small microplastic	< 1 mm	Vianello et al., 2013
		0.2 - 1 mm	MSFD Technical Subgroup on Marine Litter, 2013
		> 0.3 mm (< 1 mm)	Faure et al., 2015
	large microplastic	1 - 5 mm	MSFD Technical Subgroup on Marine Litter, 2013
			Faure et al., 2015
meso	mesolitter	> 0.5 mm	Gregory and Andrady, 2003
	meso debris	5 - 25 mm	MSFD Technical Subgroup on Marine Litter, 2013
		2 - 20 mm	Lechner et al., 2014
		> 5 mm	Sanchez et al., 2014
macro	macroplastic/ macro debris	> 25 mm	MSFD Technical Subgroup on Marine Litter, 2013
		20 mm	Sanchez et al., 2014
		> 5 mm	Faure et al., 2015
mega	mega debris	100 mm	Sanchez et al., 2014

Microplastics can originate from primary and secondary sources, and this determines their shape and composition. Primary microplastics are manufactured in small sizes for different applications, including use of personal care products, synthetic fibres for clothes, and pre-production pellets for fabrication of other plastic products (Thompson et al., 2004; Arthur et al., 2009; Ryan at al., 2009; Roex et al., 2013; Storck et al., 2015). Also, researchers suggest that the manufacture of nanoplastics may increase with their use in electronic devices, medicines, cars, and airplanes (Roex et al., 2013), which may be of growing concern as effective detection methods have yet to be developed. These MNP will be collected mostly intact in industrial and household sewage, and go through wastewater treatment (WWT) facilities before being discharged into the aquatic environment (Roex et al., 2015). In addition to primary sources, secondary MNP originate from larger pieces due to weathering by UV radiation and physical defragmentation by mechanical forces, and their production rates

depend on environmental characteristics and type of plastics (Williams and Simmons, 1996; Thompson et al., 2004; Arthur et al., 2009; Ryan at al., 2009; Cole et al., 2011; Dubaish and Liebezeit, 2013), thus their direct input to oceans may be harder to trace and quantify.

Chemical composition, size and surface features of MNPs can provide insight to its origins. For example, microplastics found in personal care products tend to be smaller than 0.3 mm, contain additives (e.g. plasticisers), and are composed mainly of polyethylene (PE), but also commonly contain polypropylene (PP), polyethylene terephthalate (PET), polymethyl methacrylate (PMMA) and Teflon (PTFE) (Roex et al., 2015; Storck et al., 2015). Pre-production pellets will be mainly spherical or cylindrical around 5 mm in size (GESAMP, 2015). In addition, PE, PP, and polystyrene (PS) are often used in packaging and thus are indicative of urban origins; while denser polymers like polyvinyl chloride (PVC) and polyester (Pest) are commonly used in construction and textiles, respectively (GESAMP, 2015), and will be introduced likely as fragments and fibres from sewage effluent (Sadri and Thompson, 2014). To my best knowledge, there is no data in the literature on the relative abundances of primary to secondary MNP, and only few studies examine the correlation between larger and smaller fragments for secondary MNP. Thus, there is a need to address these knowledge gaps for accurate quantification of MNP fractions, assessment of the relationship among abundances of different size classes, and application of precise source characterisation approaches for understanding the potential contributions of different urban and industrial sources (Lee et al., 2013). This information is crucial from a management and policy standpoint, since it is predicted that even if land-based inputs are controlled, plastic debris densities in oceans will continue to increase from secondary sources (Eerkes-Medrano et al., 2015).

1.4 Microplastics in Freshwater

1.4.1 Freshwater

Microplastics were described as early as the 1960s and 1970s (GESAMP, 2015), but it was not until 2004 that the term became widely used (Thompson et al., 2004). Although plastic litter is not a new problem, only recently have MNP become a focus of the scientific community with publications on the topic increasing rapidly (Faure et al., 2012; GESAMP 2015), particularly in marine systems (see reviews by Andrady et al., 2011 and Cole et al., 2011). Existing data on MNP pollution of freshwater and terrestrial habitats are less abundant than those for marine systems (**Figure 1**; Thompson et al., 2009; Wagner et al., 2014, Eerkes-Medrano et al., 2015), but the number of publications are also increasing, most of them since 2014. The research published between 2011 and 2014 on microplastics in freshwaters in Asia, Europe, and North America has recently been reviewed (Eerkes-Medrano et al., 2015); all reviewed papers

report the presence of different size classes of plastics in freshwaters, as well as high relative abundances of MNP compared to macroplastics, in both sediment and surface waters (**Table 2**).

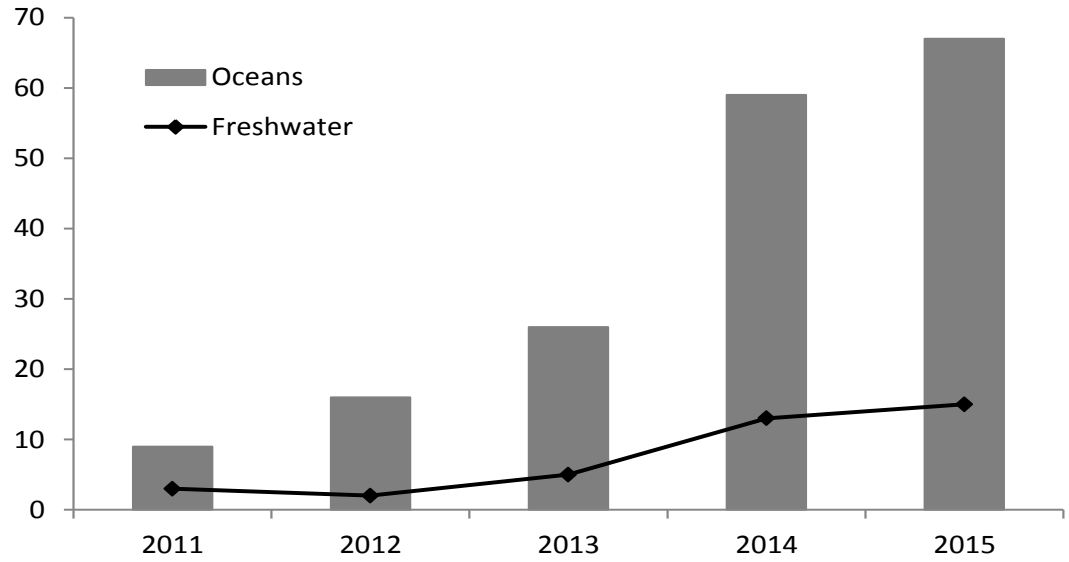

Figure 1 Comparison of marine vs freshwater studies of microplastics pollution published between 2011 – 2015, based on Web of Knowledge search engine accessed 25/11/2015.

In America, most research has concentrated in the North American region, with only one study providing MNP data from Chile in South America (Browne et al., 2011). Furthermore, the North American studies have focussed mainly in the Great Lakes area (Zbysewski and Corcoran, 2011; Eriksen et al., 2013; Rios-Mendoza and Evans, 2013; Zbysewski et al., 2014; Corcoran et al., 2015), including the St. Lawrence River watershed (Castañeda et al., 2014). Of these, only two studies collected data from freshwater bottom sediments (Castañeda et al., 2014; Corcoran et al., 2015), and one considered open-water survey (the Laurentian Great Lakes system; Eriksen et al., 2013). The rest of the studies focussed on beach surveys. Across these studies, MNP were present in both sediment and surface waters, with higher MNP densities compared to macroplastic densities, and with high predominance of pellets and fragments, indicative of primary sources. In addition, microbeads found in the St. Lawrence River were comparable in size, shape and composition to those found in the Laurentian Great Lakes, indicating a possible transport of MNP sources from the municipalities along the river to the lakes (Castañeda et al., 2014).

Table 2 Studies on microplastics pollution in freshwaters published since 2011

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
America	USA	Los Angeles River, Coyote Creek/San Gabriel River system	Moore et al. 2011	surface, mid, and near-bottom water	manta trawl with mesh size 1 mm for surface samples; manta trawl with streambed sampler for mid and bottom samples; initial visual sorting with naked eye and dissecting microscope; sieving through mesh sizes 4.75, 2.8, 1.0 mm of smaller pieces;	1-4.75 mm, >4.75 mm	Greatest abundances and densities observed during wet periods (i.e. rain event on November 22), with total counts of 74 items per m^3 in Coyote Creek, 337 items per m^3 in San Gabriel, and 12,932 items per m^3 in L.A. River, for pieces 1-4.75 mm in size. Microplastics <5 mm were 16x more abundant than macroplastics (3x more by weight). Most abundant debris type were foamed polysterene, followed by pellets, hard plastic fragments, thin films, line, and whole items. Extrapolation of data using flow rates estimated a yield of 2.33×10^9 plastic objects and particles for all sampling devices over 72-hr period.
America	Canada/USA	Lake Huron	Zbyszewski and Corcoran 2011	sediment	beach surveying for collection of visible debris with stainless steel trowel; FT-IR; SEM	<5 mm plastic pellets, >5 mm broken plastic, polystyrene	In Lake Huron, a total of 3,209 pieces were found, including 2984 pellets, 108 fragments, and 117 pieces of styrofoam.
America	Canada/USA	Lakes Superior, Huron, and Erie	Eriksen et al. 2013	surface water	manta trawl with mesh size 333 µm; size fraction sieving (0.355-0.999 mm, 1.00-4.749 mm, >4.75 mm); SEM; energy dispersive x-ray spectroscopy (EDS)	0.355-0.999 mm, 1.00-4.749 mm, >4.75 mm	Spatial variability was observed across samples, ranging from ~450 to >450,000 items per km^2, and Lake Erie (the most populated) had the highest abundances. An average abundance of 43,157 items per km^2 was calculated for all samples. Pellets and fragments were more abundant, and the smallest size class accounted for 81% of the total count. Most pieces are suspected to originate from consumer products, likely introduced by nearby urban effluent.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
America	USA	Great Lakes	Rios Mendoza and Evans 2013 (abstract)	n/a	n/a	n/a	n/a
America	Canada/USA	St. Lawrence River	Castañeda et al. 2014	sediment	benthic grab with mesh size 500 µm; visual manual separation and identification under dissecting microscope; differential scanning calorimetry for chemical composition	0.40-2.16 mm (range of microbeads collected)	Microbeads were found at 8 of 10 sites. Mean density was 13,759 $\pm$ 13,685 items per m^2 across all sites. They were a variety of colours and sizes and their melting point suggested PE composition.
America	USA	Dunkirk, Fredonia, and Plattsburg WWTP in New York	Chaskey et al. 2014 (poster)	WWTP effluent	volume reduced sampling and array sieving with mesh sizes 1 mm, 355 µm, 125µm; H_2SO_4 and H_2O_2 digestion and filtration with mesh size 125 µm; inspection under dissecting microscope	<1 mm	Suspect plastic-like particles were present in all WWTP effluent and discharged at rates of 109,556, 81,911, and 1,061,953 particles per day from Plattsburgh, Fredonia, and Dunkirk, respectively. Particle colour ranged from bright red and blue to opaque, and signs of erosion and UV-degradation were observed.
America	USA	North Shore Channel	Hoellein et al. 2014 (abstract)	n/a	SEM; rRNA sequencing (in progress)	0.3-5 mm (micro)	Higher microplastic concentrations were observed downstream of WWTP relative to upstream, mainly consisting of fragments and plastic fibers.
America	USA	North Shore Channel	McCormick et al. 2014	surface water	neuston nets with mesh size 333 µm; sieving through mesh sizes 2 mm and 330 µm; H_2O_2 digestion; density separation with NaCl; direct count with stereo microscope; SEM; bacterial measurements (DNA extraction and sequencing); dissolved nutrients (SRP, NH_4,NOx)	2 mm-330 µm	Microplastics were found in all samples, and concentrations were higher downstream of WWTP effluent discharge. Mean concentrations were 1.94 (0.81) m^3 upstream and 17.93 (11.05) m^3 downstream. Foams and pellets were found downstream, in lower concentrations than fragments and fibers. Extensive colonisation of microplastics was observed, mainly consisting of prokaryotic cells. Higher constituent concentrations were also measured downstream of WWTP.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
America	USA	Chesapeake Bay estuarine rivers	Yonkos et al. 2014	surface water	manta trawl with mesh size 0.33 mm; fraction sieving through 5.0 mm and 0.3 mm; H_2O_2 digestion of oven-dried samples; visual pre-selection with dissecting microscope; Raman microspectroscopy	0.3-2 mm, 2-5 mm	Microplastics were collected in 59 out of 60 samples. Concentrations were low and variable across replicates, locations and sampling period, ranging over 3 orders of magnitude between <1.0 g/km^2 and 563 g/km^2. Concentrations peaked in September at all sites, and microplastics concentrations increased with proximity to more densely urban areas. Sizes 0.3-2.0 mm and flexible sheets were more abundant, followed by synthetic fibers, extruded polystyrene, and larger fragments (2.0-5.0 mm)
America	Canada/USA	Lakes Erie and St. Clair	Zbyszewski et al. 2014	sediment	beach surveying for collection of visible debris with stainless steel trowel; FT-IR; SEM	<2 cm (styrofoam, pellets, plastic fragments), intact or near-intact debris	In Lake Erie, a total of 1,576 pieces were collected, including 603 pellets, 934 fragments, and 39 pieces of styrofoam. In Lake St. Clair, a total of 817 pieces were collected, including 110 pellets, 192 fragments, 234 pieces of styrofoam, and 281 intact or near-intact debris
America	Canada	Lake Ontario	Corcoran et al. 2015	sediment	beach surveying for collection of visible debris; Raman; box corer for lake bottom sediment samples; size fraction sieving (<0.5, 0.5-0.71, 0.71-0.85, 0.85-1, >1 mm), density separation with sodium polytungstate (SPT); microscopy; FTIR	<1 cm, 1-5 cm, >5 cm for visible samples; <5 mm (micro)	A total of 6,172 pieces were collected from beach sites, including pellets, fragments, intact items, and polysterene. Pellets showed composition of PE and PP. Pieces in the 1-5 cm range and white/translucent colours were predominant. Microplastics ranging from 0.5-3 mm in size were found in bottom sediments at depths <8 cm, with PE accounting for the majority of microplastics. Higher abundances were observed in sediment collected at a site near the centre of the lake as compared to a site near the outlet.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
Asia	Mongolia	Lake Hovsgol	Free et al. 2014	shoreline debris; surface water	shoreline surveying for collection of visible pieces; manta trawl with mesh size 333 µm; Tyler sieves, H_2O_2 digestion and density separation, direct count under a light microscope	0.355-0.999 mm, 1.00-4.749 mm, >4.75 mm	A total of 409 items were collected as shoreline debris, of which 77% were macroplastics. Pelagic microplastics averaged 20,264 items per km^2, ranging from 997-44,435 items per km^2, mostly consisting of fragments, films, and lines/fibers.
Europe	Switzerland	Lake Geneva	Faure et al. 2012	surface water; sediment; fish and birds	sand sieving and beach surveying for collection of coarse fragments; manta trawl with mesh size 300 µm; collection of gut content from fauna; direct count under stereo microscope	<2 mm, 2-5 mm (sediment); <5 mm, > 5mm (water)	A total of 1-7 fragments were retrieved per sand sample, mostly including polystyrene. In lake water samples, densities were 7,649 items per km^2 and 48,146 items pe km^2 for macro- and microplastics, respectively (data from only one sample). No ingested plastics were observed in guts of fauna.
Europe	Germany	Jade System, Southern North Sea	Dubaish and Liebezeit 2013	surface waters; effluent (paper recycling plant)	grab sampling at 20 cm depth, 1-6 L, sieving with mesh sizes 80 and 450 µm for sampling; vacuum filtration through 1.2 µm cellulose nitrate filter	n/a	Mean abundances for microplastics in the Jade System were 64 $\pm$ 194 and 88 $\pm$ 82 items per litre for granules and fibres, respectively. Microplastics numbers in sewage effluent were comparable to those in Jade system ranging across sampling dates from 23-52 items per litre for granular pieces, 23-25 items per litre for fragments, and 12-41 items per litre for fibres. Based on these numbers, an average annual input of $9x10^8$ particles from the wastewater treatment plant was estimated.
Europe	Italy	Lake Garda	Imhof et al. 2013	sediment	random grid sample; density separation; Raman; SEM	9-500 µm, 500 µm-1 mm, 1-5 mm, >5mm	More particles were found in the north shore in concentrations of 483 $\pm$ 236 and 1,108 $\pm$ 983 particles per m^2 for macro- and microplastics, respectively. In the south shore mean abundances were 8.3 and 108 $\pm$ 55 particles per m^2 for macro- and microplastics, respectively.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
Europe	Italy	Lagoon of Venice	Vianello et al. 2013	sediment	box corer; density separation with NaCl and filtration through 32 µm steel-wire sieve and 0.7 µm fiberglass filter; micro-FT-IR; SEM	<1 mm (small micro)	Total abundance of small microplastics (S-MPPs) ranged from 2,175 to 672 particles per kg^{1}. Polymer types identified included: PE, PP, PEP, Pest, PAN, PS, Alkyd, PVC, PVOH, ad Polyamide, with PE and PP accounting for more than 82% of total S-MPPs. Irregular fragments, fibers, films, and pellets/granules, respectively, were observed in 87%, 10%, 2%, and 1% of all stations, and 93% of S-MPPs were size 30-500 µm. There was high correlation between total S-MPPs concentrations with the finer fraction of sediment and metal pollution index. Signs of degradation were observed.
Europe	Netherlands	River Dommel	Besseling et al. 2014 (abstract)	n/a	spatially and temporally explicit model based on advective transport, homo- and hetero-aggregation, sedimentation-resuspension, polymer degradation and burial.	nano-, micro-, and millimetre (NMM) sized particles; size range for each class not specified	Particle size, biofilm formation and water turbulence affect fate and retention of NMM sized polymer particles and the positioning of the accumulation hot spots along the river.
Europe	Austria/Slovakia	Danube river	Lechner et al. 2014	surface water	volume-reduced sampling with driftnets with mesh size 500 µm; density separation; visual sorting and direct count with naked eye	<2 mm (micro), 2-20mm (meso)	Average plastic load of the river was 316.8 $\pm$ 4,664.6 items per $1000m^3$ (79.4% industrial, 20.6% others, by density), equivalent to 4.8 $\pm$ 24.2 g per $1000m^3$ (29.7% industrial, 70.3% others, by mass).

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
Europe	Sweden	Langeviksverket WWTP in Lysekil	Magnusson and Nóren 2014	incoming and effluent water; sludge; recipient water	Ruttner sampler for effluent water filtered through mesh size 300 µm; towing of zooplankton net with mesh size 300 µm at 20, 50, and 200 m downstream of effluent tube; visual examination under stereo microscope; FT-IR	≥300 µm	Incoming water had mean concentration of 15,000 particles per m^3, equivalent to inflow of 3,200,000 particles per hour. More than 99% were retained in sludge, and effluent water discharged 1,770 particles per hour. Shape affected retention, with fibres being retained to a higher degree. Concentrations of 1.1-1.8 particles per m^3 were found in recipient water compared to 0.45 m^3 in reference site, and concentrations were higher near the effluent tube compared to 200 m downstream.
Europe	UK	Thames river	Morritt et al. 2014	surface water	GPS tracked fyke nets (mesh size not defined); direct counts	n/a	A total of 8,490 plastic items were collected between 17 September and 13 December 2012, and were grouped into 7 main contributory categories. General plastics made up 20-25% of total litter in all sites, while wrappers and containers accounted for 21-28%. No major trends were observed moving from upstream to downstream sites, but higher number of items observed near sewage treatment outflows.
Europe	UK	Tamar estuary	Sadri and Thompson 2014	surface water	manta net with mesh size 300 µm; size fraction sieving (3 mm, 1 mm, and 270 µm); FTIR	>5 mm, 3-5 mm, 1-3 mm, <1 mm	Overall mean concentrations were 0.028 items per m^3. Microplastics accounted for 81% of all plastic debris, and the 1-3 mm size class was most abundant. Polyethylene was most abundant (40%), followed by PS (25%),and PP (19%).
Europe	Germany	Elbe, Mosel, Neckar, and Rhine rivers	Wagner et al. 2014	sediment	density separation; visual inspection	<5 mm	Concentrations of 34-64 items per kg^1 dy weight were measured, and River Rhine had the highest load. Fragments accounted for 60% of total microplastics, and the remainder were fibers.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
Europe	Switzerland	Lakes Geneva (including inlets and outlets), Constance, Neuchâtel, Maggiore, Zurich, and Brienz	Faure et al. 2015	surface water; sediment; fish and birds	spatial sample collection for beach sediments; density separation with NaCl and filtration through mesh size 300 µm; manta trawl with mesh size 300 µm for water samples; collection of gut content from fauna; visual sorting under dissecting microscope for larger fragments; H_2O_2 digestion; FTIR; mass spectrometry (gas and liquid chromatography)	>5 mm (macro), >1 mm (large micro), > 300 µm (small micro)	Microplastics were found in all beach sediments and surface water samples. Across all lakes, beach average densities were 1,300 $\pm$ 2,000 and ranged from 20-7,200 items per m^2 for microplastics; and 90 $\pm$ 250 and ranged from 0-150,000 items per m^2 for macroplastics. Measured densities at the surface of all swiss lakes averaged 91,000 $\pm$ 120,000 and 1,800 $\pm$ 3,100 particles per km^2 for micro- and macroplastics, respectively. In rivers, average densities for all samples were 790 $\pm$ 1,600 and 1.9 $\pm$ 3.5 items per h^1 for micro- and macroplastics, respectively. Pellets were less abundant but had a higher mass proportion than foams and fibers. In terms of composition, of all particles analysed, 62% were PE, 15% PP, and 12% PS. Only 7.5% of fish analysed showed signs of ingested plastics, while plastics were found in 8 of 9 dissected birds, with a mean of 4.3 $\pm$ 2.6 items per bird.
Europe	Germany	Rivers Rhine and Main	Klein et al. 2015	sediment	composite bulk sample of wet sediment sieved through mesh size >10 mm; dry sieveing with mesh size 63, 200, 630 µm; density separation with NaCl and vacuum filtration through 45 mm filter; H_2O_2 and H_2SO_4 digestion with vacuum filtration; direct count with naked eye and under binocular microscope for pieces 63-630 µm; FTIR	630-5000, 200-630, 63-200 µm (discarded particles <63 µm)	Microplastics in the Rhine and Main shore sediments accounted for 228-3,763 and 786-1,368 items per kg^1 , respectively. The 630-5,000 µm category was most abundant by weight, but numerically, the 63-200 µm pieces were predominant. Spheres and fibers were more abundant in the lower size categories, and fragments dominated the 630-5,000 µm size class. Over 50% of total plastic weight was attributed to PE and PP, while numerically, PS particles were more abundant.

Continent	Country	Water Body	Authors	Samples Collected	Techniques Used	Size Classes	Main Findings
Worldwide	Australia (Port Douglas), Japan (Kyushu), Oman, United Arab Emirates (Dubai), Chile (Viña del Mar, Punta Arenas), Philippines (Malapascua Island), Portugal (Faro), Azores (Ponta Delgado), USA (Virginia, California), South Africa (Western Cape), Mozambique (Pemba), UK (Sennon Cove, Plymouth, Tyne)		Browne et al. 2011	sediment; effluent	van Veen grabs for sampling of effluent; density separation with NaCl; FTIR;	n/a	Abundance of microplastics ranged from 2 to 31 fibres per 250 mL of sediment, mostly consisting of Pest (56%), followed by acrylic (23%), PP (7%), PE (6%), and polyamide fibres (3%). Microplastics abundances were positively correlated with population density, and disposal sites contained >250% more microplastics than reference sites. Effluent contained at least 1 particle of microplastic per litre, and again Pest was predominant.

In Asia, a study in Lake Hovsgol, Mongolia, reported average pelagic microplastics densities of 20,264 items per km^2, despite its remoteness and low population density (Free et al., 2014). As microplastic abundance would be expected to be relatively lower in such areas, this was attributed to the lake's long residence time, small surface area, and lack of proper waste management (Free et al., 2014), indicating strong need for effective control measures. Although no other studies were found for MNP in freshwaters in Asia, the continent contributes considerably to the global plastic production (Plastics Europe, 2015); and marine data indicate that over the last decade, plastic litter increased by a factor of 10 every 2-3 year in the Japanese coast (Haruo, 2000). More recently, MNP pollution has been reported in coastlines of Japan (Browne et al., 2011) and Korea (Lee et al., 2013; Kang et al., 2015). In this context, the region may present useful opportunities for studying these plastic particles in freshwaters under highly populated and industrialised conditions.

The rest of the available literature presents studies across Europe, where work has been conducted in Switzerland (Faure et al., 2012; Faure et al., 2015), Italy (Imhof et al., 2013; Vianello et al., 2013), Germany (Dubaish and Liebezeit, 2013; Wagner et al., 2014; Klein et al., 2015), , Netherlands (Besseling et al., 2014), Austria (Lechner et al., 2014), and the UK (Morritt et al., 2014; Sadri and Thompson, 2014). In addition, a single study collected and summarised global data from coastlines across all continents (Browne et al., 2011). In Switzerland and Italy, for example, the research studies focussed on lake systems, where microplastics were observed in Lake Geneva (Faure et al. 2012; 2015), Lake Garda (Imhof et al., 2013), and the Lagoon in Venice (Vianello et al., 2013), usually at higher concentrations than macroplastics. In Germany, microplastics commonly in the form of fragments, granules, and fibres were reported in sediments in various rivers, including the Rhine, Elbe, Mosel, Neckar, and Main rivers (Wagner et al., 2014; Klein et al., 2015), and the Jade system of the southern North Sea (Dubaish and Liebezeit, 2013). Globally, microplastics abundances generally were positively correlated with population density, and proximity to disposal sites (Browne et al., 2011). Nevertheless, the spatial coverage of the MNP studies in freshwater systems remains limited.

Most of the earlier freshwater research appears to have focussed on lentic systems, but rivers and wastewater treatment systems are gaining more attention as important vectors not only to sea, but also to lakes and estuaries. Rivers are dynamic systems with less water volume for dilution relative to lakes and oceans, so they can concentrate MNP, particularly urban rivers receiving direct sewage discharges (McCormick et al., 2014; Rech et al., 2014). Also, riverine systems can act as temporary sinks, while transport can quickly increase during rain events, as influenced by factors such as flow rate and bottom currents (Galgani et al., 2000; McCormick et al., 2014; Rech et al., 2014). For example, beach studies in Brazil (Araujo and Costa, 2007; Ivar and Costa, 2013)

attributed the presence of solid waste, including plastics, to domestic sources along the river basin, proximity of river sources, and increased river flow during high rain events (Rech et al., 2014; Sadri and Thompson, 2014). Similarly, a study identified the Danube River as an important transport route of plastics from production sites in Germany and Austria to the Black Sea, and proposed that inter- and intra-annual variations in MNP drift densities were linked to differences in the release of plastics from processing facilities (Lechner et al., 2014). In Chicago, higher MNP densities were observed after rain events during wet periods for two urban rivers, with evidence of higher abundances of primary MNP that are not regulated by total maximum daily loads (TMDLs), and are being discharged into oceans (McCormick et al., 2014). However, contradicting data also emerged. A study of larger plastic pieces (size categories not defined) in the Thames river did not find major trends from up- to downstream sites, although generally higher abundances were observed in sites near sewerage discharge (Morritt et al., 2014). In the Tamar River estuary in the UK, the authors also did not find evidence that the river acted as a sink or source of plastic fragments present in the system (Sadri and Thompson; 2014), although this is believed to be influenced largely by its location in an unpopulated catchment, while fluvial systems would be expected to act as sources under more populated or industrialized conditions (Eerkes-Medrano et al., 2015), emphasizing the need for further evaluation of freshwater systems in order to be conclusive.

1.4.2 Wastewater

The relationship between population density, as well as urban and industrial activities with MNP concentrations can be explored via analysis of wastewater effluent in treatment facilities and receiving waters. A handful of studies are available providing data on effluent discharge of MNP from wastewater treatment plants (WWTP) in the US (Chaskey et al., 2014; Hoellein et al., 2014: McCormick et al., 2014), UK (Browne et al., 2011), Gemany (Dubaish and Liebezeit, 2013) and Sweden (Magnusson and Nóren, 2014). In the North Shore Channel in Chicago, higher microplastic concentrations, consisting mostly of fragments and plastic fibres, were observed downstream of the WWTP relative to upstream (Hoellein et al., 2014: McCormick et al., 2014). Likewise, a global study found that disposal sites contained >250% more microplastics than reference sites upstream (Browne et al., 2011). In New York, discharge rates of 109,556, 81,911, and 1,061,953 particles per day were reported from three different WWTP (Chaskey et al., 2014), while an average annual input of 9×10^8 particles was estimated from a WWTP in Germany (Dubaish and Liebezeit, 2013). A more recent study in Europe was conducted in a relatively small plant in Langeviksverket serving ~12,000 population equivalents (p.e) (Magnusson and Nóren, 2014). Here, although most of the microplastics entering the WWTP were retained in the sludge, the plant continued to discharge MNP, as evidenced by higher concentrations in the recipient water compared to the reference site

upstream (Magnusson and Nóren, 2014). It is possible that larger WWTP will contribute larger MNP loads, and thus an additional filtration step before discharging effluent to receiving waters may help reduce its MNP concentrations (based on comparable data collected in two other larger WWTP in Sweden, Magnusson and Nóren, 2014). However, the general absence of quantitative studies in WWT systems does not allow for a proper assessment of their contribution, making this an area of high priority for further MNP research.

1.4.3 Ecological Impacts

A few studies on freshwater systems have also considered the ecological effects of MNP (see review by Eerkes-Medrano et al., 2015), although little remains known. Owing to their small size, MNP are ingested directly and indirectly by aquatic species more readily than larger particles, sometimes when mistaken for food, and can lead to harmful physical effects (Derraik, 2002). Evidence from marine studies for example, indicate that MNP ingestion may lead to choking, blocked digestive tracts, damage to organs, debilitation, and ultimately death (see review by Derraik, 2002). Some available studies in freshwater have observed similarities in MNP ingestion by freshwater and marine organisms (Imhof et al., 2013; Rochman et al., 2013; Sanchez et al., 2014; see review by Eerkes-Medrano et al., 2015), while others found little evidence of uptake by fish and bird species in lakes (Faure et al., 2012; Faure et al., 2015). In addition, microplastic pieces can adsorb persistent organic pollutants (POPs), potentially introducing toxicity throughout the food web (Mato et al., 2001; Endo et al., 2005; Bakir et al., 2012; Rios-Mendoza and Evans, 2013), which could eventually reach humans by bioaccumulation (Farrel and Nelson, 2013). Desorption of POPs and other manufacturing additives can increase pollutant concentrations in waters and increase the susceptibility of the larger pieces to degradation (Dubaish and Liebezeit, 2013). Nevertheless, information on sorption and leaching of POPs from microplastics is scarce (Arthur et al., 2009), and most of the knowledge on toxicity derives from marine and laboratory experiments (Eerkes-Medrano et al., 2015), while data from freshwaters remains limited.

It has also been suggested that certain MNP, such as synthetic fibres can be airborne and as inhaled by humans, make their way to lung tissue and potentially contribute to the formation of tumours (Pauly et al., 1998). Conversely, MNP surfaces (plastisphere) provide habitats for microbial colonisation and biofilm formation, allowing for migration of opportunistic pathogens and invasive species (Zettler et al., 2013, McCormick et al., 2014). The latter may be relevant for WWTP as it could affect the functioning of the treatment processes, as well as increase the transport of WWT bacteria from these facilities to receiving waters (Zettler et al., 2013; Tagg et al., 2015).

1.5 WWT Systems in the UK

Wastewater treatment systems may be potential point sources for MNP pollution to aquatic ecosystems. Considerable amounts of MNP collected by sewerage systems can be discharged into the waterways via WWTP effluent, particularly near urban and industrial centres receiving large quantities of sewage every year (Browne et al., 2011; Eriksen et al., 2013; Eerkes-Medrano, 2015). The WWTP may not remove MNP completely and an estimated 160 particles per inhabitant per hour are discharged from these facilities into receiving waters (Storck et al., 2015), and subsequently transported to oceans. Therefore, the role of WWTP in the degradation, transport, and removal of MNP, particularly those originating from primary sources, should be considered. Additionally, as the systems are expected to function properly in order to minimize treatment costs and ensure adequate water quality standards, the impact of MNP in the treatment process should also be evaluated.

In the UK, over 11 billion litres of wastewater are collected daily, most commonly via combined sewerage systems, although separate collection systems for surface water and foul drainage also exist (Defra, 2012). Treatment of the collected sewage in the country is regulated by the Urban Waste Water Treatment Directive (91/271/EEC) and, widely, involves: preliminary treatment in which screening traps are used to remove large solids; primary treatment in large tanks for sedimentation of suspended solids; secondary or biological treatment for breakdown and reduction of residual dissolved organic matter; and tertiary treatment tailored for removal of specific pollutants such as phosphorus or nitrogen (Defra, 2012; Scottish Water, 2015: The World Bank Group, 2015). In Scotland, wastewater collection, transport and treatment is largely designed and operated as a 5-stage process by Scottish Water (**Figure 2**), and is regulated by the Scottish Environment Protection Agency (SEPA). However, depending on the size of the community (measured in p.e. units), and the specific water quality requirements, treatment stages can be added or modified as needed and allowed by available resources (Defra, 2012; The World Bank Group, 2015). For example, between the 1990s and 2000s, “less sensitive areas” were identified across the UK, allowing for primary treatment to be minimised according to the Directive, but currently this designation is no longer used in the UK (Defra, 2012). Furthermore, the Directive mandates secondary treatment for agglomerations of >2000 p.e., and advanced or tertiary treatment for agglomerations greater than 10,000 p.e in sensitive areas. The UK generally is compliant with these provisions (Defra, 2012).

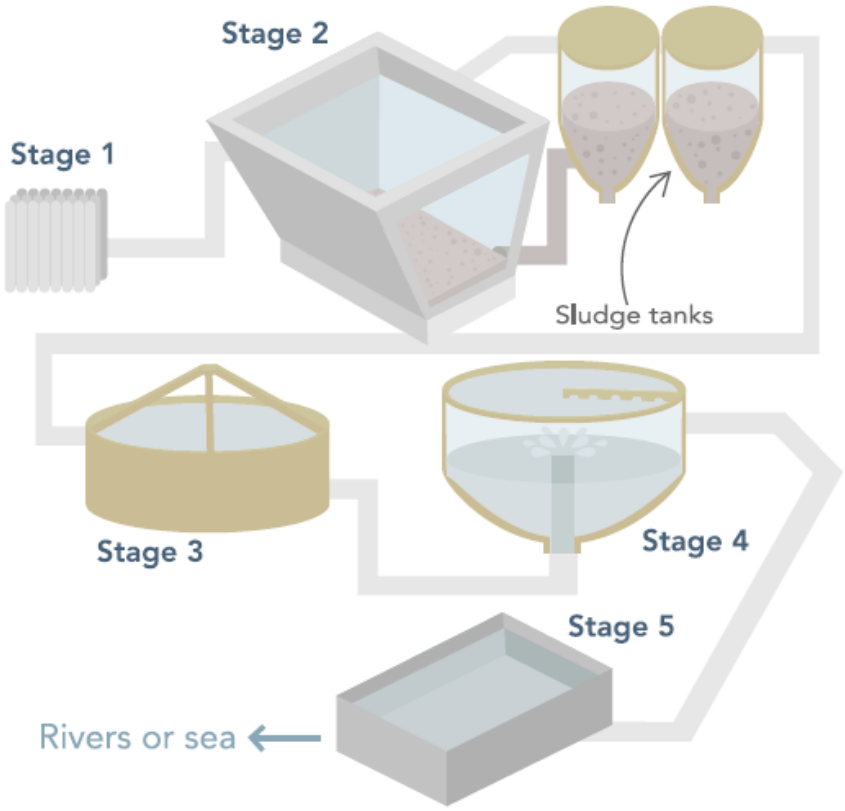

Figure 2 Scottish Water's 5-stage wastewater treatment process (Source: Scottish Water)

The Directive also requires that both collection systems and WWTP have the capability to deal with seasonal changes, especially peak wet weather flows, and the UK has made improvements to both, as described in the Defra report (2012). These include upgrading the capacity of sewer networks across the country (e.g. London, Ayrshire, Belfast) between 2010 and 2015, and ensuring implementation of secondary treatment in all required WWTP to achieve 100% compliance (Defra, 2012). In Scotland, under storm conditions, flooding is prevented by using combined sewage overflows (CSO) that are monitored and improved if considered to be Unsatisfactory Intermittent Discharges (UID) under the agreed Quality & Standards (Q&S), partly determined by aesthetic requirements (Scottish Water, 2015).

1.6 Methods for Studying MNP

Because MNP research is still a developing field, there are no standardised procedures for their study, and method advancement is still in its early stages (Eerkes-Medrano et al., 2015; Tagg et al., 2015). The different size class distinctions and methods used may reduce comparability of results across studies, highlighting the need to unify size class definition and develop simple, low-cost, and precise methods for their detection and monitoring (Galgani et al., 2013; Eerkes-Medrano et al., 2015). However, it may still be too early to do so, as we have yet to identify the spectrum, sizes, and types of MNP that require

greater attention; thus for now, standardised procedures may prove useful only in situations that call for regular site-specific monitoring or have limited budgets (Free et al., 2014; Eerkes-Medrano et al., 2015). Nevertheless, a review of methods for identification and quantification of MNP in marine environments is available (Hidalgo-Ruz et al., 2012), and more recently, the NOAA Marine Debris Program published a manual on recommended laboratory methods for quantifying plastics in oceanic waters and sediments (Masura et al., 2015). The methods used for freshwater systems are similar to those implemented in marine studies.

The review of methods presented here is based on the anticipated pathway for the study and includes the forensic techniques predominantly mentioned in the literature, tailored to gather information for quantification and characterisation of MNP, as well as describe their behaviour and fate in WWT and fluvial systems.

1.6.1 Sampling and Sorting

Traditional sampling techniques for both surface water and sediments are common. Surface waters are often sampled through volume-reduced methods using manta trawls and neuston nets (Hidalgo-Ruz et al., 2012), while bulk grab sampling has been described for effluent discharge collection (Browne et al., 2011; Chaskey et al., 2014; Magnusson and Nóren, 2014). For lake sediments, selective sampling of visible pieces from beach transects was a frequent practice, and bulk sampling equipment has been used for collection of lake bottom sediments (Hidalgo-Ruz et al., 2012; Castañeda et al., 2014; Corcoran et al., 2015).

Sample processing usually involved a combination of approaches including visual pre-selection, size fraction sieving, flotation and density separation, filtration, and acid, alkaline or enzymatic digestion (Hidalgo-Ruz et al., 2012; Cole et al., 2014; Tagg et al., 2015). Sieve analysis is useful for separation of particles into different size ranges. A wide range of sieve sizes have been used across studies, and will largely determine the minimum sizes of plastic debris that are collected and quantified (Hidalgo-Ruz et al., 2012). For example, higher MNP abundances are usually reported where smaller mesh sizes were used in sieving and filtration (Hidalgo-Ruz et al., 2012; Storck et al.,2015). This is important as it may reduce the comparability or accuracy of results, possibly underestimating abundances in some cases from loss of material that is not retained in sieves and gets discarded.

Further sorting is performed with the use of flotation and density separation, and studies also differ in the solutions used for this method. Most authors reported the use of sodium chloride (NaCl) for separation of low density particles from sediment as the lower plastic density encourages flotation

(Hidalgo-Ruz et al., 2012). A few studies have also employed sodium iodide (NaI) and sodium polytungstate (SPT) for polymers with higher densities, although this tends to be more costly (Claessens et al., 2013). However, the basic method is the same across studies. Briefly, the sample is mixed with the solution, shaken for a certain amount of time, and left to settle so that the low-density particles rise to the surface. The floating pieces can be manually picked out, and the smaller ones can be extracted under vacuum filtration of the supernatant through membrane filter (Hidalgo-Ruz et al., 2012). The filtered samples can then be kept under sterile conditions until further analysis.

Sample digestion was also employed by most studies for isolation of MNP from confounding organic material. Similar to density separation, different solvents may be utilised although hydrogen peroxide (H_2O_2) treatment is more common. The effectiveness of this approach was tested (Nuelle et al., 2014; Tagg et al., 2015), recommending that a 7-day sample treatment with 30-35% of H_2O_2 is more effective in removing organic material from the filter, improving filtration time without damaging the polymer.

After initial sorting, suspected MNP particles are characterised according to different categories (e.g. type, shape, colour), and quantified for assessment of spatial and temporal distributions (Hidalgo-Ruz et al., 2012). Quantities of MNP are often expressed as abundance, mass, or both, and the units used to report results differ among studies. Abundances of MNP are commonly expressed as particles per unit area for sediment samples (e.g. km^2 or m^2), and particles per unit volume for water samples (e.g. m^3), although total pieces counted were also reported in shoreline surveys (**Table 3;** see reviews by Hidalgo-Ruz et al., 2002 and Eerkes-Medrano et al., 2015). Mass was occasionally used in grams per km^2, and yield in items per unit time were also reported in studies considering discharge from WWTP and river flow. Larger pieces are often counted with the naked eye or under a stereo microscope with further identification of smaller pieces with the use of forensic techniques.

Table 2 Units commonly used by freshwaters studies on microplastics

Sample Type	Unit	Studies
sediment/ shoreline debris	total pieces	Zbyszewski and Corcoran 2011
		Faure et al. 2012
		Free et al. 2014
		Zbyszewski et al. 2014
		Corcoran et al. 2015
	items/particles per m^2	Imhof et al. 2013
		Castañeda et al. 2014
		Faure et al. 2015
	items/particles per kg^1	Vianello et al. 2013
		Wagner et al. 2014
		Klein et al. 2015
	items/particles per 250 mL of sediment	Browne et al. 2011
water/effluent water	items/particles per m^3	Moore et al. 2011
		Magnusson and Nóren 2014
		McCormick et al. 2014
		Sadri and Thompson 2014
	items/particles per km^2	Faure et al. 2012
		Eriksen et al. 2013
		Free et al. 2014
		Faure et al. 2015
	items/particles per litre	Browne et al. 2011
		Dubaish and Liebezeit 2013
	items/particles per 1000 m^3	Lechner et al. 2014
	grams per 1000 m^3	Lechner et al. 2014
	grams per km^2	Yonkos et al. 2014
yield/discharge (rivers/WWTP)	items/particles per 72 hrs	Moore et al. 2011
	items/particles per year	Dubaish and Liebezeit 2013
	items/particles per day	Chaskey et al. 2014
	items/particles per hour	Magnusson and Nóren 2014

1.6.2 Analytical Techniques for Identification

Source characterisation and composition of MNP can be performed with the use of electron microscopy and spectroscopic techniques. Electron microscopy can be employed when wavelength becomes a limiting factor in light microscopy, since electrons have shorter wavelengths, which enables better resolution (Nanoscience Instruments) and thus provides further insight on the chemical and morphological characteristics of the plastic particles. There are two types of

electron microscopy: scanning electron microscopy (SEM) and transmission electron microscopy (TEM), with the main difference being that in SEM, electrons are bounced off the sample, while in TEM electrons go through the sample creating a shadow. While SEM appears to be employed often (Zbyszewski and Corcoran, 2011; Eriksen et al., 2013; Imhoff et al., 2013; Vianello et al., 2013; Hoellein et al., 2014; McCormick et al., 2014; Zbyszewski et al., 2014) no studies reported using TEM. Application of TEM may be particularly useful for the rarely-identified plastics in the nano-sized category.

Similarly, spectroscopic tools can be used for added analysis of individual particles, with Raman and Fourier Transform-Infra Red (FT-IR) spectroscopy used more frequently. These techniques are applied to gather information on the chemical composition of polymers and the crystalline structure of the particles that can provide insight into the sorption behaviour for persitent, bioaccumulating, and toxic substances, as well as the degradation of MNP based on bond distances (Gerrard and Madams, 1986). Here, the basic principle is that infrared radiation is passed through a sample, where it is absorbed, reflected or transmitted. For example, Raman spectroscopy measures light scattered whereas FT-IR measures absorbed light. Although there are few differences between techniques, the end result is a molecular fingerprint represented by absorption and transmission, and similar to a fingerprint no two will be alike since different materials will generate different spectra based on their unique molecular structures, which allows for identification of the compounds (Das and Agrawal, 2011). This information can be used to trace sample important in understanding site-specific loadings. By comparison, FT-IR is being implemented more recently and broadly than Raman, perhaps owing its popularity to being non-destructive, less costly and easier to use, and involving less sample preparation (Tagg et al., 2015).

Spectroscopy methods can be combined with microscopy to improve accuracy. For example, a couple of studies reported the use of micro-FT-IR and molecular mapping by focal plane array (FPA), proposing this approach can help to reduce scanning time, and facilitate the analysis of entire membrane filters and smaller pieces without affecting spatial resolution (Vianello et al., 2013; Tagg et al., 2015).

While the presence of microplastics in effluent indicates that current WWTP processes are not removing MNP from wastewater, there is an absence of discussion of approaches to examine the system and the impact that MNP may have on WWT efficiency, such as bacterial treatment or blockages. Bacterial growth, for example, can be measured a number of ways, including direct count techniques or measuring of chemical oxygen demand (COD; APHA, 1992), in which case an increase in COD would indicate a reduction or poisoning of the bacterial population. Otherwise, with a healthy bacterial population, COD should

be removed faster. Additionally, techniques to test for blockages can be potentially employed. For example, assessing whether biofilms are trapping or removing some of the MNP fragments in trickling systems, using magnetic resonance imaging (MRI) methods to visualise if flow paths (pore spaces) are blocked by MNP. The latter could be done by using laboratory sand columns to mimic WWTP processes, and employing standardised protocols to measure hydraulic conductivity and water quality parameters.

1.6.3 Modelling of Transport

Higher abundances may be expected in habitats that accumulate smaller particles of sediment (Browne et al., 2011), and their distribution may be influenced by sediment transport and deposition processes (Castañeda et al., 2014; Vianello et al., 2013; Klein et al., 2015). Hydrodynamic effects can have greater influence in MNP distribution than population density, industrial activities, or sewage discharge and MNP concentrations in river shores in Germany (Klein et al., 2015). In this regard, transport models can be useful tools to model MNP behaviour in riverine systems and evaluate the factors that control their transport and distribution. Sources and discharge can be used as with other contaminants to predict MNP loading and build models for MNP transport and identification of areas of high deposition, although this approach was rarely discussed in the MNP literature. In the Danube, plastic load at mean flow in the Danube, and a correction factor for population density were used to calculate plastic inputs to the Black Sea (Lechner et al., 2014). Flow rate data from two California rivers were also used to estimate yields of >2 billion particles over a 72 hour period (Moore et al., 2011). In Venice, high correlations were observed between small microplastics and fine grain size, indicating both follow similar sinking and accumulation processes, with higher accumulation of MNP in low energy sites (Vianelllo et al., 2013). Fundamentally, if plastics behave in the same way as sediment, available hydraulic models can be easily applied to MNP load studies, and if they behave differently, the models can be fine-tuned to get their behaviour in properly. One possibility for this is the use of Delft hydraulics model (Delft 3D suite) for rivers and estuaries. This model allows particle tracking and has a morphology module that predicts sediment movement (Deltares, https://oss.deltares.nl/web/delft3d/about). Correlation analysis can be performed on grain and MNP size classes, and incorporated into the transport model to project loading of MNP from freshwaters systems, including WWTP.

1.7 Research Objectives

This PhD study aims to describe and model the behaviour of MNPs in wastewater treatment and fluvial systems.

The specific objectives of this project are:

1. Detect and quantify MNP in incoming and effluent water in a WWTP and recipient water in an urban catchment
2. Identify the main sources and categories of MNP entering these systems
3. Evaluate the impact of MNP on WWTP efficacy at different treatment stages
4. Assess the ability of WWTP to process MNP, and predict loading of MNP to the environment
5. Model transport and distribution of MNP in receiving waters to determine whether these systems are acting as sinks or sources

The premise of this research project is to fill in gaps of knowledge about MNP debris in freshwaters and wastewater, and generate incisive understanding of the distribution and behaviour of MNPs in these environments. Therefore, we expect that the results of this project will inform stakeholders (e.g. legislators, manufacturers, industry) and aid in the identification of priority areas for further research, monitoring and regulation of MNP, and the development of effective programmes and mitigation strategies. This project is also relevant to the Scottish Government's Hydro Nation goals of connecting research and policy, developing the economic, environmental and social values of Scotland's water resources, and raising Scotland's international profile and knowledge exchange.

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