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Review

Microplastics in the Aquatic Environment: Occurrence, Persistence, Analysis, and Human Exposure

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Abstract: Microplastics (MP) have recently been considered as emerging contaminants in the water environment. In the last number of years, the number of studies on MP has grown quickly due to the increasing consciousness of the potential risks for human health related to MP exposure. The present review article discusses scientific literature regarding MP occurrence and accumulation on the aquatic compartment (river, lake, wastewater, seafood), the analytical methods used to assess their concentration, their fate and transport to humans, and delineates the urgent areas for future research. To better analogize literature data regarding MP occurrence in the aquatic compartment we subdivided papers based on sampling, analytical methods, and concentration units with the aim to help the reader identify the similarities and differences of the considered research papers, thus making the comparison of literature data easier and the individuation of the most relevant articles for the reader's interests faster. Furthermore, we argued about several ways for MP transport to humans, highlighting some gaps in analytical methods based on the reviewed publications. We suggest improving studies on developing standardized protocols to collect, process, and analyze samples.

Keywords: emerging contaminants; microplastics pollution; aquatic environment; analytical methods

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1. Introduction

In recent years, several efforts have been devoted to the detection and removal of those contaminants that are referred to as contaminants of emerging concern (CECs), i.e., any chemical present in water or the environment at very low concentration levels, or only recently detected [1–8]. Microplastics (MP) have all the characteristics to belong to this category especially for their presence in the water environment [9].

Plastic materials play an important role in everyday life and the improvement of human health, referring to disposable medical equipment, food packaging, technology, and so on. However, the greatest exposure to microplastics contamination of the environment and the human body could have awful effects over time. During the COVID-19 pandemic, increasing consumption of single-use plastic, especially personal protective equipment (PPE) such as face masks, gloves, and gowns, that represent one of the safety methods being used in virus prevention, but also plastic used by households to wrap and take foods from supermarkets and restaurants, has been observed [10]. Since the use of PPE is indispensable to the current pandemic scenario, it is important to identify proper ways to dispose of the resulting plastic wastes [11]. In fact, according to a World Wide Fund for

Nature (WWF) report of 2020, the improper disposal of just 1% of the masks would result in the dispersion of more than 40,000 kg of plastic in the environment (based on a weight of 4 g/mask and about 10 million face masks per month) [12]. This phenomenon is leading to an increase in plastic pollution and a consequential higher amount of microplastics dispersed in the environment in the future.

As stated by the National Oceanic and Atmospheric Administration (NOAA), plastic particles with a diameter lower than 5 mm are defined as microplastics (MP). Regarding nanoplastics (NP) dimensions, there is still no consensus in the literature [13] and some authors state that NP have a diameter lower than 1 µm [14–18], while others assert that NP range is below 100 nm [19,20]. Depending on their origin, microplastics can be classified as primary or secondary microplastics. Primary microplastics are intentionally manufactured particles with sizes <5 mm including microbeads in personal care products, cleaning agents, coatings, paints, industrial abrasives for delicate surfaces, and feedstocks used in the manufacturing of plastic materials [21]. Secondary microplastics, much more abundant than primary, are those derived from the deterioration of larger plastics (macro- and meso plastics) during utilization (fragmentation of synthetic fibers in the process of clothes washing, the release of particles from tires) or after discarding (bottles and shopping bags) [14,22,23].

Plastic fragmentation can be induced by chemical and physical aging, especially on land due to higher ambient temperatures, frictional forces, and UV exposure [24], and also via (bio)degradation, the most common process in the aquatic compartment [25,26]. The addition of pro-oxidant compounds (transition metals added in the form of stearates of Fe^{3+}, Mn^{2+}, or Co^{2+}) leads to the so-called oxo-biodegradable plastics, very attractive for packaging employment thanks to the possibility, claimed by the supplying companies, to have environmentally friendly plastic articles. Even though such additives accelerate polymer oxidation and abiotic (photo-induced) degradation without the influence of light, no improvement of environmental impact with respect to conventional polymers has been observed [27]. On the contrary, they might have a negative effect on the environment due to the release of metals and chemicals such as microplastics, generated by incomplete biodegradation [28]. Such MP particles (derived from waste fragmentation) are found in marine species as a consequence of passive (gill water filtration) or active (ingestion) intake [29]. Furthermore, as particle size decreases and surface area increases, the rate of polymer biodegradation enhances, varying by polymer type and ambient conditions with half-lives ranging from days to centuries [30]. Together with microplastics and nanoplastics, also organic and inorganic additives, unreacted monomers, and other compounds used in the formulation of plastic materials are released into the environment [31]. This represents a risk for human health due to the fact that the majority of additives and plasticizers, such as phthalic acid esters or phthalates (PAEs), alkylphenols (in particular 4-tert-octylphenol and nonylphenol), bisphenol A, di(2-ethylhexyl)adipate (DEHA), and brominated flame retardants (BFRs), have endocrine disruption and carcinogenic properties [32]. Moreover, MP can easily adsorb some organic contaminants such as persistent organic pollutants (POPs), mainly polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and dichlorodiphenyltrichloroethane (DDT), but the role of MP in POPs increment to marine organisms is still not clear and much debated in the literature [33]. Depending on the original form of plastics, the residence time in the environment, and the degradation processes that occurred on the surface, microplastic may have the shapes of fibres, fragments, spheres, beads, films, flakes, pellets, and foam [34]. Size and shape affect the microplastics' potential to cause physical harm to organisms, being taken up by cells or transported in soil [35]. Large microplastics are unlikely to be taken up by most plants and soil organisms, while nanoplastics can be taken into cells, posing an environmental risk [36,37].

Consequently, the complete characterization and the multidisciplinary overview of microplastics contamination in terms of chemical composition, abundance, distribution, uptake, and transport to the human body are of paramount importance to evaluate their

impacts and to enable the critical and efficient procedure to avoid clinical repercussions. This review covers microplastics contaminated water environments, describing the analytical methods used for their characterization and quantification, thus discussing the most recent studies on their occurrence, fate, and behavior.

2. Materials and Methods

In this review, the authors attempt to provide a general overview of several implications associated with microplastics occurrence in the aquatic compartment. The methodology of the present work is briefly outlined in the following.

2.1. The Methodological Approach of the Review

Based on the recent scientific literature, this review had three main objectives: (1) to outline and discuss the presence of different analytical methods used to assess MP concentration, technical considerations and limitations; (2) to summarize contamination and accumulation of MP in the aquatic compartments and propose future research directions according to the existing literature; (3) to define the urgency and seriousness of microplastics in water by emphasizing the various ways for microplastics transport to the human body.

2.2. Data Sources Assimilation and Analysis

The keywords "emerging contaminants", "microplastics pollution", "aquatic environment", "analytical methods" were selected individually or jointly to search for relevant information on Web of Science, Scopus, and Google Scholar. Key literature published between 2004 and 2020 (up to December) was assimilated and analyzed.

3. Microplastics in the Aquatic Environment

3.1. Occurrence and Abundance of Microplastics in Water

Microplastics research is mainly focused on the water compartment [38], firstly marine environment [22,39–42], and recently moved also to wastewater [43–46], rivers, and lakes [47–50]. The first evidence of microplastics particles in the marine environment date back to the 1970s, when pellets (2.5–5.0 mm) on the Sargasso Sea Surface [51], spherules (0.1–2.0 mm) in the coastal waters of southern New England [52], spherules and disks (0.2–4.9 mm) in the surface waters of the Atlantic Ocean [53] and pellets (1–5 mm) in the surface waters of the Pacific Ocean [54] were detected.

Since 2004, when Thompson quantified the abundance of microplastics by collecting sediment from beaches, estuarine and subtidal sediments in the UK [16], an increasing amount of researchers have investigated microplastics occurrence, fate, and transport in the marine environment covering the shorelines of all continents [55,56], big and small islands [57–59], passing from Atlantic [60–63], Pacific [64,65], and Arctic ocean [66], and North [67,68], Adriatic [69,70], Baltic [71,72], and Mediterranean sea [73–80] and even in deep-sea habitats [81].

A small fraction of microplastics found in the ocean environment is related to marine activities such as the fishing industry (employing plastic equipment), while the majority of them (around 80%), has at its source, plastic litter derived from land [39,40]. The indiscriminate plastics disposal on beaches and coastal areas [82,83], terrestrial river input to the sea [21,84], stormwater runoff [85], passive uptake in marine species [86–90], wastewater discharge, and deposition of atmospheric microplastics constitute the main contribution to the microplastic occurrence in the marine compartment. Microplastic debris is transported widespread across large distances by ocean currents [22,57,91], winds, river outflow, and drift [92,93] with a consequent spatial and temporal mutability of their abundance [83,94] as highlighted by the occurrence of this litter in remote and uncontaminated areas [95] such as the poles [96], the ocean depths, and mid-ocean islands [59].

Different sampling techniques have been employed to assess microplastics abundance in water compartments such as sediment recovery from the seafloor, beaches and estuaries,

beachcombing to analyze plastic litter on the shoreline, observation of marine visible plastic debris performed by divers, use of marine trawls to collect particles within the water column, and examination of plastic fragments ingested by marine organisms [22,34].

In Table 1 are summarized the most relevant works regarding microplastics abundance in the marine environment subdividing them by matrix sampled (costal beach and deep sediments, surface, and sub-surface water and water column), sampling, and analytical methods, emphasizing both MP concentration, size range, and polymer type. Works that employed recovery of coastal beach and deep sediments principally expressed microplastics concentration as particles/kg of dry sediment (average of 5×10^2, range 0.6–6.6 $\times$ 10^3 particles/kg) and particles/m^2 (average of 3×10^2 and range 1.5–6.3 $\times$ 10^4 particles/m^2).

Table 1. Microplastics abundance in the marine environment.

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
Sediment	Recovery from beach	FTIR	particles	(0–4)	>1.6 µm	PE, PP, PS, ABS, PVA, NY	[97]
		MO	particles/m^2	(30–800)	-	-	[98]
				1.51	-	-	[99]
				(12–1300)	-	-	[100]
			particles/kg	(1.3–36.3)	-	PS	[101]
		MO and FTIR	particles/m^2	(36–228)	1–5 mm	PE, PP, PET	[102]
				(0–62,800)	1–5 mm	PS, PE, PP	[103]
				46.6 (2–178)	0.3–4.75 mm	PE, PP, PS, NY	[104]
				(16.67–489.7)	1–4.75 mm	PS, PP, PE	[105]
				39 (25–53)	0.1–5 mm	PES, EPM	[106]
				284 (2–1258)	-	PE, PET, NY	[107]
			particles/kg	12.1 (5.99–21.6)	-	PE, PP, TPU, NY, PS, PET, PVC	[80]
				(0.58–2116)	-	PE, PP, PET, PA, PS, PES	[108]
				(60–300)	-	PET, RY	[109]
				(33–439)	0.5–3 (average 1.96) mm	PE, PP, NY, PS, PES	[110]
				160 (76–295)	0.20–0.94 (average 0.51) mm	PP, PE, PS	[111]
		MO and RMS	particles/kg	236 (72–1512)	<1 mm (55%)	PP, PE, PES	[112]
		MO and TDS-Pyr-GC-MS	particles/kg	1.8 (1.3–2.3)	0.100–1.000 mm	PP, PE, PET, PVC, PS, PA	[67]
	Van Veen Grab	MO and FTIR	particles/kg	(2.5–87.5)	-	PE, PP, NY, TPU, EVOH, LLDP/Oct	[70]
				119 (49–390)	0.038–1 mm	PE, PP, PS, PVA, NY	[113]
				15 (0–27)	0.1–4 mm	PES, PVA	[106]
				15 (7–25)	average 1.29 mm	PET, PP, PE, PA, CPH, PVC, PS	[114]
		MO and RMS	particles/kg	0.98	0.2–5 mm	PE, PP, PS	[115]
				(54–506)	-	PE, PP, NY, PVC	[116]
			particles	62	0.25–5 mm	PA, PET, PP, acrylic	[117]
	Box corer	MO and FTIR	particles/kg	4356 (42–6595)	<0.150 mm	PTFE, CPE, PA, PP, NR	[118]
				(672–2175)	0.030–0.500 mm	PE, PP, PS, PVA, PVC, PA, PAN, PES	[119]
				134 (60–240)	0.06–1 mm	PE, PET, PES, CPH, cellulose, acrylic	[120]
				(560–4205)	0.05–5 mm	PP, PE, PS, PET, NY	[121]
				(5.30–68.88)	0.10–4.76 mm (average 1.63)	PP, PET, RY	[122]
	Recovery by divers	MO	particles/kg	900	-	-	[123]
		MO and FTIR	particles/kg	10	-	RY, PP	[124]
	Recovery with a ring	MO and FTIR	particles/kg	129.7 (102.9–163.3)	-	PE, PEVA, PP, PET, PS, ALK	[125]

Table 1. *Cont.*

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
Surface Water	Rotating drum	FTIR	particles	(0–2)	>1.6 µm	PE, PP, PS	[97]
	PE bottles	MO	particles/m^3	152.000–2,420,000	<1 mm	-	[68]
	Metal screen	MO and FTIR	particles/m^3	88,000	0.2–1 mm	ALK, PR, PS, PP, PE, PAS, PES, SR	[126]
	Steel sampler	RMS	particles/m^3	5000–34,000	-	PE, PVC, PP, NY	[116]
	Stainless-steel hydrophore	MO and FTIR	particles/m^3	46 (20–120)	average 1.29 mm	PET, PP, PE, PA, PVAC	[114]
	Manta Trawl or other nets	MO	particles/m^2	0.13 (0.01–0.42)	-	-	[74]
				(0.011–0.285)	-	-	[127]
				0.406 (0.012–2.66)	average 2.69 mm	-	[69]
				0.112 (0.006–1)	0.4–5.2 mm (average 1.48)	-	[77]
		MO and RMS	particles/m^2	0.006–0.095	0.3–2.5 mm	PP, PE, PS, PET, PMMA	[128]
				0.0006–0.042	0.5–5 mm	PE, PP, NY	[129]
		MO and FTIR	particles/m^2	(0.021–0.578)	average 1.95 mm	PE, PP, PS, PA	[76]
				(0.0156–0.618)	average 1.3 mm	PP, PE, PS	[71]
				(0.043–1.46)	0.5–5 mm	PE, PP, PET	[102]
		MO	particles/m^2	(0.021–6.553)	-	-	[130]
			particles/m^3	0.01–0.35	-	-	[73]
				7.68 (0.94–69.6)	0.3–5 mm	-	[131]
				(600–1200)	0.2–5 mm	-	[132]
	Manta Trawl or other nets equipped with a flowmeter	MO and FTIR	particles/m^3	(0.03–491.0)	>0.3 mm	-	[133]
				0.34 (0–1.13)	0.25–7.71 mm (average 1.93)	PES, PA, PE, PVC, acrylic, cellulose	[66]
				0.01–0.09	0.3–5 mm	-	[134]
				1.00	0.2–20 mm	PE, PP, PA, NY, EP, PVC, PS, PVA, PET	[78]
				0.33 (0.01–1.23)	0.3–5 mm	PE, PP, PS, PET	[135]
				0.06	0.3–5 mm	PE, PP	[136]
				0.71 (0–3)	0.125–15.98 mm	PP, PE, PS, PA, CPH	[137]
		MO and RMS	particles/m^3	0.24	0.335–5 mm	PE, PP, PS	[115]
Sub-surface Water	Water intake system equipped with a flow-meter /pump	MO	particles/m^3	(8–9180)	0.068–5.810 mm (average 0.606)	-	[64]
		MO and RMS	particles/m^3	2.46	1.25–2.5 mm (90%)	-	[63]
		MO and FTIR	particles/m^3	2.68 (0–11.5)	0.25–7.71 mm (average 1.93)	PES, PA, PE, PVC, acrylic, cellulose	[66]
Water Column	Multi-level trawl	MO and FTIR	particles/m^3	0.68	0.5–5 mm		[138]
	Sea-bird CDT		particles/m^3	70.8	0.4–8.3 mm	PES, PET	[139]
	Bongo net equipped with a flow-meter /pump		particles/m^3	4.8–18.4	0.335–5 mm	CPH, ALK, PVA, PE	[140]
				(0.045–2569)	0.02–5 mm	PES, PE, PP, ALK, PCL, PS PEA, PU	[141]

Abbreviations: FTIR: Fourier-Transform Infrared Spectroscopy; RMS: Raman Spectroscopy; MO: microscope; PE: polyethylene; PP: polypropylene; PS: polystyrene; PET: polyethylene terephthalate; ABS: acrylonitrile butadiene styrene; PVA: polyvinyl alcohol; PVAC: polyvinyl acetate; PVC: polyvinyl chloride; PES: polyester; PAN: polyacrylonitrile; PA: polyamide; PTFE: polytetrafluoroethylene; PU: polyurethane; NY: Nylon; RY: Rayon; ALK: alkyd resin; CPE: chlorinated polyethylene; NR: nitrile rubber; LLDP/Oct: linear low-density polyethylene-octene copolymer; EVOH: ethylene vinyl alcohol copolymer; TPU: thermoplastic polyurethane; PVS: polyvinyl sulfate; PAS: polyacrylate-styrene; SR: synthetic rubber; PR: phenoxy resin; PCL: polycaprolactone; PEA: polyethylacrylate; PEVA: polyethylene-vinyl acetate; EPM: polyethylene-propylene; EP: epoxy resin; CPH: cellophane.

Together with the simple recovery of sediment from the beach, more elaborate sampling methods regard the use of Van Veen Grab [113] or Box Corer [119] to collect deeper sediment. Van Veen Grab is a clamshell bucket that collects sample sediments in water environments and has a sampling surface of 0.1 m^2 and has a depth of 20 cm. The Box Corer is a marine sampling for soft sediments in lakes or oceans. It is designed for the minimum disturbance of the surface and the penetration depth is 0.5 m. For surface, sub-surface and deep water (along the water column) matrix, the concentration unit depends on the sampling method. In particular, particles/m^3 (average of 6.3×10^4 particles/m^3 ranging

from 0.03 to 2.4 × 10^6 particles/m^3) is employed when a known water volume is sampled using a pump [63,141] or a net equipped with a digital flow meter [132], while particles/m^2 (average of 0.55 ranging from 0.0006 to 6.6 particles/m^2) is preferred as unit when the net surface was considered [42,142,143]. The number of particles recovered strongly depends on the trawl type used, as reported by Di Mauro et al. [140] that discover total microplastic concentrations 10,000 times higher using Niskin bottles, that allow collecting smaller plastic particles, with respect to bongo and neuston nets (110,000 in respect to 9.1 particles/m^3). Moreover, as shown by Isobe [133,144] and Cózar [145] microplastics amount exponentially increases as their particle size decreases due to the degradation of a large plastic particle to multiple small pieces, while keeping its original volume and weight. Consequently, particle amount and microplastic concentration in all the measurement units used are affected by particle sizes considered. Thus, in some cases, an apparent low contamination could be reported if only high particle sizes are detected, while on the other hand, bigger concentrations might be disclosed when also lower particle sizes are sampled and analyzed.

Regarding microplastics abundance during the time, Law [146] observed no relevant difference in over 10 years in the North Atlantic gyre, with a concentration of 0.02 particles/m^2, while Claessens [113] revealed an increase of microplastic amount (from 55 to 156 particles/kg of dry sediment) in sediment sampled along the Belgian coast in 15 years. Notwithstanding several studies have demonstrated that low-density microplastics are mainly found in the sea-surface microlayer [82,147], their position in the water column can differ such as in estuarine habitats and they can sink if marine organisms stick to them [16,82,92,148]. For instance, on average of 0.68 particles/m^3 at the ocean surface compared to 0.02 particles/m^3 in the bottom was detected by Kooi [138], while Oztekin [149] discovered a microparticle density of 2.7 particles/m^3 for sea surface and 24 particles/m^3 for the water column. Moreover, microplastics can be redistributed throughout the water column after a storm that can re-suspend litter on the seabed [150].

More recently, microplastic particles have been documented in estuaries [21,38,151] and freshwater systems [47–50,152–155] such as lakes [156] and rivers [157]. In Table 2 are summarized almost all works concerning microplastics abundance in the freshwater environment and estuary subdividing them by matrix sampled (sediments and water), sampling, and analytical methods, highlighting both MP concentration, size range, and polymer type. As in the case of the marine environment, works that employed recovery of shoreline and deep sediments mainly expressed microplastic concentration as particles/kg of dry sediment (range 0.5–75 × 10^3 particles/kg) and particles/m^2 (range 0.2–26 × 10^4 particles/m^2) while, for water matrix sampled, the concentration units mostly used are particles/m^3 (ranging from 0.0005 to 1.1 × 10^6 particles/m^3) when the water volume is known and particles/m^2 (ranging from 0.01 to 1.4 × 10^4 particles/m^2) when the water surface was considered.

Table 2. Microplastics abundance in freshwater and estuary.

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
Rivers							
Sediment	Recovery from shoreline	MO and FTIR	particles/kg	(786–1368)	0.06–5 mm	PE, PP, EPDM, PS, PET, PVC	[158]
				(178–544)	-	PE, PP	[159]
				802 (5.3–160)	0.1–5 mm	PP, PES, RY, PR	[160]
				210 (99–410)	0.06–5 mm	PET, PE, PP, PS	[161]
				422		PA, PE, PP, PMMA, PES	[162]
				(7–1029)	0.06–5	PES, PA, PE, PP, PS	[163]
		MO and RMS	particles/kg	350 (185–660)	1–4 mm	PP, PE, PMMA, PET, PVC, PS,	[164]
				118 (50–195)	0.045–5 mm	PET, PE, PP, PS, PA	[165]
		MO, SEM and RMS	particles/kg	414 (307–580)	0.5–5 mm	PS, PE, PET, PP, PA, PVC	[166]

Table 2. *Cont.*

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
		MO and SEM	particles/kg	(161–448)	0.06–2.8 mm	-	[167]
		Visual sorting	particles/m^2	5595 (16–258,408)	0.3–5 mm	PS	[168]
		MO and LC-MS/MS	particles/kg	136–2060	0.01–5 mm	PET, PC	[169]
		MO and FTIR	particles/m^3	(58–1265) particles/m^3	0.055–5 mm	PE, PP, PET, PS, PVA, EVA, PTFE, PMMA	[170]
				847 (100–3600)	0.01–5 mm	-	[171]
	Van Veen Grab		particles/kg	1669 (80–9597)	0.02–5 mm	PE, PP	[172]
				1971	0.05–5 mm	PP, PES, PE, PA, PVC, ALK, PS, PU, acrylic	[173]
				0.81 (0.46–1.62)	-	PE, PP, PES PS, PTFE, PAC	[174]
				2052	-	PA, PE, PP, PMMA, PES	[162]
		MO and RMS	particles/kg	82 (25–30)	<5 mm	PS, PP, PE, PC, PVC	[175]
		MO, FTIR, Py-GC-MS	particles/kg	2080	0.02–5 mm	PE, PP, PS	[176]
		MO	particles/m^2	13,832	0.04–2mm	PE	[177]
		Fluorescence MO, FT-IR	particles/kg	32,947 (18,690–74,800)	0.02–5 mm	PE, PP, PES, PVC PS, NY, PU, PET	[178]
	Other grabs	MO and RMS	particles/kg	30	0.05–2 mm	NY, PES	[179]
		MO and FTIR	particles/kg	(10–520)	1–4 mm	PE, PA, PP, PET	[180]
				2071 (68–10,500)	0.01–5 mm	-	[171]
				(0–97)	1–5 mm	PE, PP, PET, PS	[181]
		MO and SEM	particles/kg	(360–1320)	<5mm	-	[182]
		Fluorescence MO	particles/kg	832 (65–7562)	95%<0.4 mm	-	[157]
	Wooden ruler and frame	MO and FTIR	particles/kg	(260–11,070)	0.01–0.5 mm	APV, CPE, EPDM, PES, PP	[183]
			particles/m^2	3408–13,618	0.1–5 mm	PE, PP, PS	[184]
				100,000 (48,000–187,000)	0.01–5 mm	-	[171]
		MO and FTIR	particles/m^3	1200 (0–67,500)	0.1–9.6 mm	Cellulose, PET, PES, PVA	[185]
	Bulk sample collected in containers of different type			2724 (379–7924)	0.02–2 mm	PP, PE, PET	[172]
				1650 (293–4760)	0.05–5 mm	PP, PES, PE, PA, PS, PU, PVC, ALK, acrylic	[173]
				164,790 (86,000–106,1000)	0.05–1 mm	PE, PET, PP, PS, PAN, PVA, NY	[186]
				6186 (1770–14,330)	0.05–1.5 mm	PE, PS, PP, PVC	[187]
		MO and RMS	particles/m^3	910 (0–5400)	0.1–0.5 mm	PP, PE, PET, PES	[188]
			particles/m^3	5850	0.05–2 mm	NY, PES	[179]
		Fluorescence MO	particles/m^3	120 (0–300)	-	-	[157]
				(2516–2933)	0.05–5 mm	PET, PP, PE, NY, PS	[189]
		MO and FTIR	particles/m^3	13.79 (3.52–32.05)	-	PE, PP, PS, PES, PTFE, PAC	[174]
Surface Water	Water intake with a pump			(0.1–5.6)	1–5 mm	PE, PP, PET, PS	[181]
				(67–11,532)	0.02–0.3 mm	PE, PP, EPDM	[190]
				67.5	0.075–5 mm	PE, PP, PB, PA, PVC, PS, PET	[191]
		MO and RMS	particles/m^3	4703 (1597–12,611)	<5 mm	PS, PP, PE, PC, PVC	[175]
				694 (483–967)	0.045–5 mm	PET, PE, PP, PS, PA	[165]
		MO and SEM	particles/m^3	(3670–10,700)	<5mm	-	[182]

Table 2. *Cont.*

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
	Manta Trawl or other nets	MO	particles/m^3	(4–108)	0.1–5 mmm	-	[192]
		MO and FTIR	particles/m^3	3.5	-	PA, PE, PP, PMMA, PES	[162]
				1.8 (0.1–4.7)	0.5–5 mm	PE, PP, PS	[193]
				(1.47–43.11)	1–5 mm	PE, PP, PS, PET	[194]
				0.6	0.075–5 mm	PE, PP, PB, PA, PVC, PS, PET	[191]
		MO and RMS	particles/m^3	2.57 (0–13.7)	0.1–0.5 mm	PP, PE, PET, PES	[188]
			particles/m^2	(3–31)	0.1–5 mm	PP, PE, PS, PET	[195]
		MO and Py-GC-MS	particles/m^3	(0.9–13)	0.5–5 mm	PE, PP, PVC, PS, PU	[196]
		MO and SEM	particles/m^3	(1.94–17.93)	0.330−2 mm	-	[197]
	Manta Trawl or other nets equipped with a flowmeter		particles/m^2	893	0.3–1 mm	PS, PP, acrylate, PES, PVC	[198]
		MO and FTIR		(58–1265)	0.055–5 mm	PE, PP, PET, PS, PVA, EVA, PTFE, PMMA	[170]
			particles/m^3	450 (140–1960)	0.25–5 mm	PP, PE, PET, PS, PVC, PA, PVA, EVA	[199]
				1.6 (0–12)	-	PE, PP, PS	[200]
				11.6 (0.3–58.9)	0.3–5 mm	PES, PA, PE, PP, PS	[163]
				(0.1–4.6)	1–5 mm	PE, PP, PET, PS	[181]
				0.15 (0–3.4)	0.7–5 mm	PE, PS, PP	[201]
		MO, FTIR and Py-GC-MS	particles/m^3	5.57 (0.88–13.24)	0.15–5 mm	PE, PP, PS	[176]
Water Column	Water intake with a pump	MO and FTIR	particles/m^3	846 (360–1273)	0.05–5 mm	PP, PES, PE, PA, PS, PU, PVC, ALK, acrylic	[173]
	Manta trawl	MO and FTIR	particles/m^3	(0.76–12.56)	1–5 mm	PE, PP, PS, PET	[194]
Bottom Water	Manta trawl	MO and FTIR	particles/m^3	(1.43–34.63)	1–5 mm	PE, PP, PS, PET	[194]
Lakes							
Sediment	Recovery from shoreline	SEM and RMS	particles/m^2	483–1108	-	PS, PE, PP, PA, PVC	[202]
		RMS	particles	6172	-	PE, PP	[203]
		SEM and FTIR	particles	3209	-	PE, PP, PET	[204]
			particles/m^2	1.72 (0.18–8.34)	5-20 mm	PE, PP	[205]
		UV-MO and SEM	particles/kg	174 (109–266)	0.3–5 mm	-	[206]
		MO and RMS	particles/m^2	563	<5 mm	PE, PP, PS, PET, PVC	[207]
				(50–1292)	0.1–5 mm	PP, PE, EVA, PVC, NY, PC	[195]
			particles/kg	560 (270–866)	0.5–5 mm	PS, PE, PET, PP, PA, PVC	[166]
				403 (180–693)	0.05–5 mm	PET, PA, PE, PP PVC, PS, PMMA	[208]
	Box corer	MO, FTIR and RMS	particles/m^2	400	-	PES, PET	[156]
		Mo and FTIR	particles	35	0.5–3 mm	PE, PP, nitrocellulose	[203]
		MO	particles/kg	300	0.5–5 mm	-	[209]
		MO and RMS	particles/kg	539	0.5–1 mm	PS, PAN, PVC	[210]
	Van Veen grab	MO and RMS	particles/m^2	253 (96–496)	<5 mm	PE, PS, PP	[211]
			particles/kg	(54–506)	0.05–5 mm	PP, PE, NY, PVC	[116]
		MO, SEM and FTIR	particles/kg	(14–24)	<5 mm	PE, PET, PP, PVC	[212]
		MO and FTIR	particles/kg	0.81 (0.46–1.62)	-	PE, PP, PS, PES, PTFE, PAC	[174]
				309 (92–604)	0.3–2 mm	NY, PE, PS, PP, PVC	[213]

Table 2. *Cont.*

Matrix Sampled	Sampling Method	Analytical Method	Abundance Average (Range)		Size Range	Polymer Type	Ref.
	Other grabs	MO, FTIR, SEM-EDS	particles/kg	(11–235)	0.005–5 mm	CPH, PET, PES, PP	[214]
		MO and FTIR	particles/kg	395.8 372 (8–1070)	<5mm >0.125 mm	PA, PS, PU, PET PS, PE, acryl, NY	[215] [216]
Surface Water	Bulk sample collected in containers of different type	MO, FTIR, SEM-EDS	particles/m^3	(3400–25,800)	0.005–5 mm	CPH, PET, PES, PP	[214]
		MO and RMS	particles/m^3	(5000–34000)	0.05–5 mm	PP, PE, NY, PVC	[116]
		MO and FTIR	particles/m^3	250 (0–710)	>0.125 mm	PE, PP, PS, PU, PVC, PA, PES	[216]
	Water intake with a pump	MO and FTIR	particles/m^3	1660–8925	0.05–5 mm	PET, PP, PE, NY, PS	[189]
				13.79 (3.52–32.05)	-	PE, PP, PS, PES, PTFE, PAC,	[174]
	Manta Trawl or other nets	MO, SEM and RMS	particles/m^3	(900–4650)	0.05–5 mm	PE, PP, PS, PVC	[217]
		SEM	particles/m^2	43	0.355–5 mm	-	[218]
		MO	particles/m^2	20 (1–44)	0.355–5 mm	-	[219]
			particles/m^3	0.13 (0.01–0.42)	-	-	[74]
		UV-MO and SEM	particles/m^3	4.2 (0.05–32)	0.355–4.75 mm	-	[220]
		MO and SEM-EDS	particles/m^3	(0.82–4.42)	0.3–5 mm	-	[206]
		MO and RMS	particles/m^2	(53–748)	-	-	[221]
		MO and FTIR	particles/m^2	5–758	0.1–5 mm	PP, PE, PS, PET	[195]
			particles/m^3	0.40 (0.372–1.29)	>0.125 mm	PE, PP, PS, PU, PVC, PA, PES	[216]
				(11.9–61.2)	0.005–5 mm	PP, PE	[222]
			particles/m^2	0.028 (0.013–0.054)	0.3–2 mm	Ny, PE, PS, PP, PVC	[213]
	Manta Trawl equipped with a flowmeter	MO, FTIR and Py-GC-MS	particles/m^2	(0–110)	-	PE, PP, PS, PVC, PET	[223]
Other freshwater							
Sediments of Lagoon	Box corer	MO and FTIR	particles/kg	(672–2175)	0.03–0.5 mm	PE, PP, PES, PS, PAN, PVC, NY	[119]
	Recovery from shoreline	MO and FTIR	particles/kg	(2340–6920)	0.2–5 mm	PP, PE	[224]
				(2.9–36.3)	0.025–5 mm	PE, PP, PES, PS, PVC, PTFE, PA	[225]
Surface Water of Antarctic	Net sampler	MO and FTIR	particles/m^3	0.00095 (0.00047–0.00143)	0.01–3.5 mm	Acrylic, PES, PTFE	[156]
Estuary							
Sediment	Box corer	MO and FTIR	particles/kg	121(20–340)	0.05–5 mm	RY, PES, acrylic	[226]
	Van Veen grab	MO and SEM	particles/kg	(0–126)	<5 mm	-	[227]
	Recovery from shoreline	MO, SEM and FTIR	particles/kg	351 (46–916)	0.3–4.75 mm	PE, PET, PS, PVC, PP	[228]
Surface Water	Water intake with a pump	MO	particles/m^3	4137 (50–10,200)	0.5–5 mm	-	[229]
	Bulk sample	MO and FTIR	particles/m^2	(192.5–11,890)	0.1–5 mm	PE, PP, PS	[184]

Abbreviations: FTIR: Fourier-Transform Infrared Spectroscopy; RMS: Raman Spectroscopy; MO: microscope; PE: polyethylene; PP: polypropylene; PS: polystyrene; PET: polyethylene terephthalate; PVA: polyvinyl alcohol; PVAC: polyvinyl acetate; PVC: polyvinyl chloride; PES: polyester; PAN: polyacrylonitrile; PA: polyamide; PTFE: polytetrafluoroethylene; PU: polyurethane; NY: Nylon; RY: Rayon; ALK: alkyd resin; CPE: chlorinated polyethylene; PR: phenoxy resin; PEVA: polyethylene-vinyl acetate; CPH: cellophane; EPDM: ethylene propylene diene rubber; PMMA: polymethyl methacrylate; APV: acrylates/polyurethane/ varnish cluster; PB: polybutylene. Py-GC-MS: Pyrolysis gas chromatography-mass spectrometry; SEM-EDS: Scanning electron microscopy-Energy-dispersive X-ray Spectroscopy; LC-MS/MS: Liquid Chromatography tandem-Mass.

The occurrence of microplastics in estuaries is higher than that in sea samples (4137 respect to 0.167 particles/m^3 [229,230]) due to the input of anthropogenic debris from fresh-

water systems and beaches and wash-back by marine surface currents [151,231]. Several studies regarding estuaries aim to demonstrate the presence of microplastics in surface waters [21,232,233] and sediments [234–236] mainly coastal beach ones [38]. This acute presence of microplastic pollution in estuaries indicates that terrestrial river input is an important source of microplastics to coastal and marine environments [9,148,151,237,238]. Less than one microplastic particles/m^2 at the surface of the Rhine, one of the largest European rivers [198], and 7 particles/m^3 along the Ofanto river in Southeast Italy [196] were detected, while higher concentrations were determined along the Snake and Lower Columbia rivers [188] and in the surface waters of the Wei River Basin, in northwestern China [182]. One of the most studied lakes is the Great Lake Basin of North America [203,205,223], where the average abundance of microplastics floating on the surface was as high as 0.043 particles/m^2 [218]. A similar amount (0.048 particles/m^2) was detected in Europe, in Lake Geneva, Switzerland [239].

Finally, the occurrence of microplastics in wastewater was recently reported and reviewed [44,45,55]. Sources of microplastics in wastewater are fibers from synthetic textiles [240] industries and the domestic ones released when washing clothes [241], glitter [242], detergents for contact lens, plastic particles used in air blasting [147], and dust formed during plastic items production. The majority of microplastics (>80%) are removed from the wastewater during the primary treatment [243] being captured by sedimentation and skimming processes [244,245] and thus depositing in the sludge fraction. The resulting effluents have a low microplastic concentration (0–447·10^3 particles/m^3), but the high volume discharged daily results in significant pollution of the water compartment [44,243,245–247] with an estimated median value of 2×10^6 released particles/day [45].

Regarding microplastic form in the water environment, the dominant shapes are fibers, mainly derived from laundry activities and conveyed over wide distances by the marine stream, followed by fragments with polyethylene, polypropylene, and polystyrene as the most abundant polymers.

Although all the works reported suggest a high contamination of the water environment, a full comparison among the various studies is still difficult and often impossible due to the lack of a standardized procedure for water sampling, particle counting, and polymer matrix individuations.

Based on the above reported microplastic contamination all over the world, in Figure 1 we show a map of the distribution of microplastics in the aquatic environment, from sea to freshwaters, according to nations in order to indicate the areas where microplastic research is actually performed, highlighting those of serious pollution and, on the other hand, places where a lack of microplastic monitoring is observed. Indeed, studies on the occurrence and distribution of microplastics in aquatic environments are mainly focused in the more industrialized areas, such as south-east Asia, especially China, Europe and North America, while Africa, South America and North Asia remain poorly investigated.

The map was created using all the concentrations reported in Tables 1 and 2, with units of particles/m^2, particles/m^3, and particles/kg distinguished by different shapes and colour from light yellow to dark red according to microparticle densities as shown in Figure 1.

As we can see from this map, the more polluted areas are in China and some European and North American countries, however too many areas remain uninvestigated, so further studies should be performed in these countries in order to give a wider view of microplastic contamination all over the world.

Therefore, microplastic pollution in water environments is a severe problem and MP, as other contaminants, could be assessed with the systematic and quantifiable approach in life cycle assessment (LCA). Recently, first methodological approaches for including impacts of the marine litter of microplastics to LCA were tested [248]. The assessment of marine litter impact (derived from the microparticle abundance in the marine environment and their effects on different organisms) in LCA was strongly dependent on the number of microplastic particles produced from the original litter over time. So, the model used

for impact assessment within LCA includes the relationship between fragmentation and degradation of microplastics [248]. With this approach, the authors were able to develop an applicable characterization model for LCA that can be refined through further research activities and with the help of additional data in order to reach a better understanding of this issue.

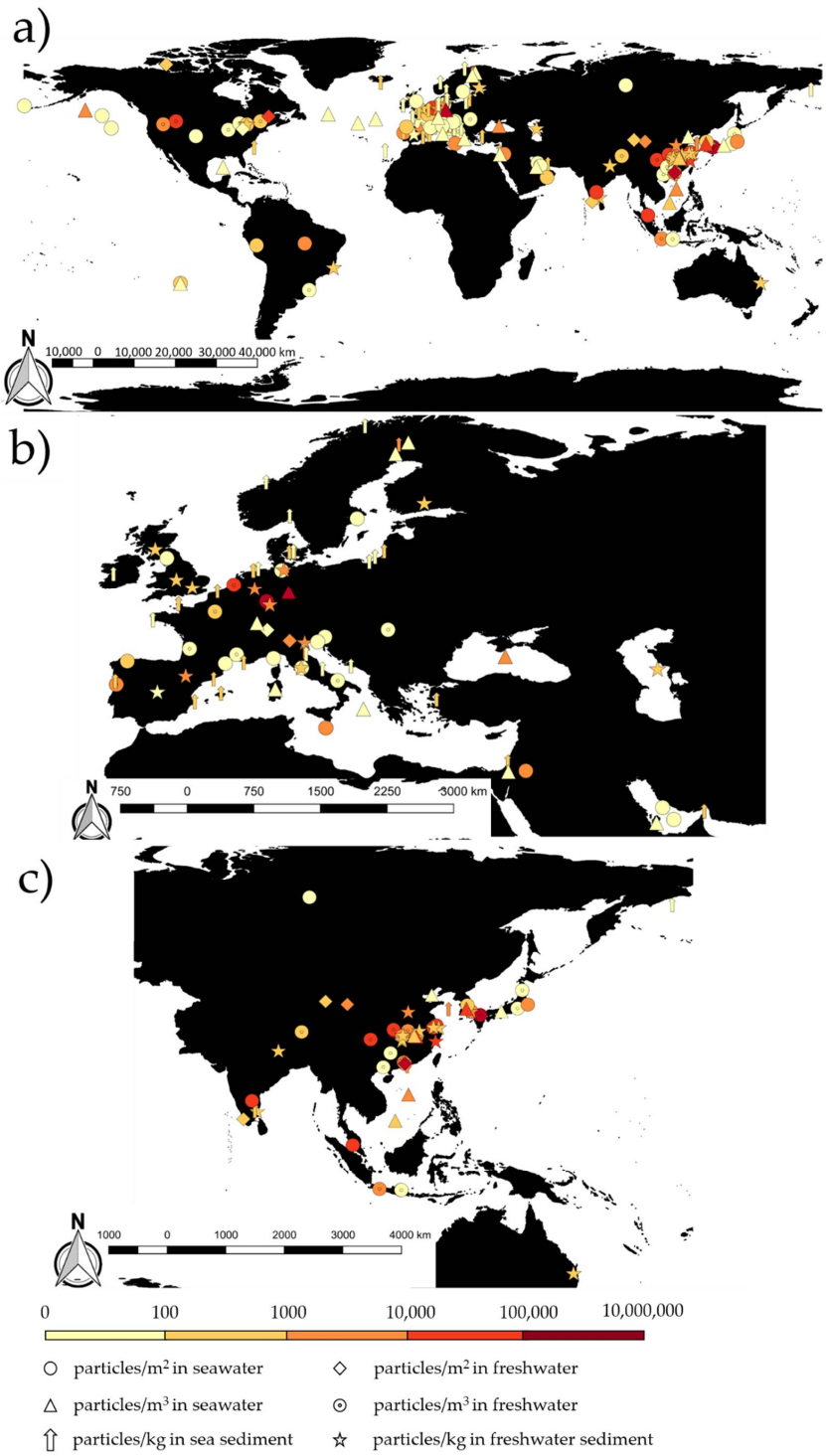


Figure 1. (**a**) Map of distribution of microplastics in aquatic environments (based on data in Tables 1 and 2); (**b**) magnification of Europe; (**c**) magnification of south-east Asia.

3.2. Analytical Approaches for MP Separation, Identification, and Quantification

Several analytical methods have been developed to measure MP in water and the most important procedures include separation, identification, and quantification.

Separation of microplastics is usually achieved by sieves with mesh sizes from 0.038 to 4.75 mm, used singly or in a series. Moreover, filters with small mesh sizes (0.02–5 µm) are also used to separate small microplastics or nanoplastics [50,249]. For plastic particles <1 µm, chromatographic techniques, active, and passive separation, are typically used. Active separations, such as the field flow fractionation (FFF) technique [250], apply external fields into microfluidic environments for the separation of dispersed particles, while passive separations, such as hydrodynamic chromatography (HDC) [251], utilize hydrodynamic and surface forces to separate particles in liquid [252]. The extraction process is usually based on the different densities of the samples. When MPs are less dense than water, they can be directly separated using a net or filtered by filters or sieves [67]. Flotation is preferred with sediments. Low density plastics have a density range from 0.9 to 2.3 g/cm^3, while the density of sediments and soil particles is higher (2.6–2.7 g/cm^3). High density salt solutions such as $ZnCl_2$, $ZnBr_2$, NaCl, NaI, and $CaCl_2$ allow MPs to float and be separated from sediments that sink [253,254]. While this method is efficient for low-density microplastics, high-density microplastics such as polyvinyl chloride and polytetrafluoroethylene cannot be easily recovered. In order to separate plastics and organic matter from the minerals in the mixture solution after centrifugation, a rubber disc is inserted into the middle of a centrifuge tube. This expedient can prevent mineral particles from being resuspended, substantially reducing separation time [255]. Moreover, a high amount of organic matter could affect separation from MPs; for this reason, acid solvents, alkali solvents, or oxidation agents, including KClO (30%), NaOH (56% or 52.5 M), H_2SO_4 (96%), HNO_3 (65% or 22.5 M), and H_2O_2 (30% or 32.6 M), have been used to digest and remove organic matter from flotation [255]. However, a recent study demonstrated that many of these agents decompose, disintegrate, or modify the weight, number, and shapes of MP particles [256]. Polyamide (PA), polyester (PET), and polycarbonate (PC) have a low resistance to strong acids. PA, polyethylene (PE), and polypropylene (PP) are resistant to 10% KOH, but polycarbonate (PC) and PET are degraded. Treatment with NaOH 40% at T = 60 °C caused deformation of PA fibers, yellowing of PVC granules, and melding of polyethylene particles. Enzymatic digestion is a biological means to hydrolyze proteins and break tissues. Enzymes ensure no loss or degradation of plastics; however, enzymatic digestion can be long and expensive [257].

The quantification of MPs is still a major challenge, due to their special chemical and physical properties (very high molecular weights and poor solubility in most solvents). MPs cannot be analyzed by classical analytical methods such as Gas Chromatography-Mass Spectrometry (GC-MS) and Liquid Chromatography tandem-Mass Spectrometry (LC-MS/MS). Table 3 outlines the main analytical techniques employed for MP detection, advanced visualization, and counting, focusing on the operation principles, advantages, and disadvantages of each method.

Table 3. Comparison of main analytical methods for MP detection and visualization.

Analytical Techniques	Advantages	Limitations
Methods for MP detection		
Fourier Transform Infrared Spectroscopy (FTIR) Spectra collected in Transmittance, Reflectance or Attenuated Total-Reflectance (ATR) mode	- Identification (non-destructive) of polymer matrix - Selective and reproducible - Small sample amounts - Limited sample preparation - Quantification of different types of particles (with sizes $\leq$2 µm) - Localization of debris in living organisms	- Not totally reliable on weathered samples, degraded samples or mixed polymer samples. - Combination with microscope is expensive
Raman spectroscopy	- Identification (non-destructive and non-contact) of polymer matrix - Selective and reproducible - Small sample amounts - Limited sample preparation - Identification and quantification of individual sub-micron particles - Localization of debris in living organisms	- Not totally reliable on weathered, colored, and degraded samples or mixed polymer samples - Auto-fluorescence mask the signal - Samples may be damaged - Time consuming for wide area imaging - Combination with microscope is expensive
Pyrolysis gas chromatography-mass spectrometry (Py-GC-MS) MS analysis of microplastics thermal degradation products	- Identification of polymer matrix and organic additives - Quantification of small amounts in mass concentration (<0.5 mg) - Limited sample pre-treatment - Suitable for biological matrices and environmental screening	- Require an expert operator and a sampler equipped with a thermal desorption system - Time-consuming - Particles size major than fractions of mm - No data on particle number, size, and shape
Time-of-Flight Secondary Ion Mass Spectrometry (ToFSIMS)	- Identification of polymer matrix - High spatial resolution imaging - Suitable for mixtures of particles - Particles size in the low µm range - Information on inorganic and organic chemical contaminants	- Complex to use and expensive - Relative quantification of different polymers is usually not possible - Identification complicated by weathering or by surface contaminants - Sample must be vacuum compatible
Advanced methods for MP visualization		
Fluorescent tagging with Nile Red (labelling of microplastics when irradiated with blue light)	- Detects and quantifies particles (20 µm–1 mm) - Low cost and semi-automated	- Require sample purification - Not reliable with weathered samples - Organic matter in the matrix may produce false positives - Not able to identify polymer types - Dye fluorescence may prevent identification by Raman spectroscopy
Scanning electron microscopy (SEM)	- Provides high-resolution image	- Require coating for working at high vacuum - Destructive - Charge effects
Scanning electron microscopy-Energy-dispersive X-ray Spectroscopy (SEM-EDS)	- Chemical and morphological characterization of particles - No requirement of coating due to work in low vacuum - Environmental-SEM-EDS gives elemental composition and surface morphology of microplastics with no charge effects	- Complex to use - Expensive

Basically, two different analytical strategies have been discussed in recent publications [258–261]. The first one is based on the isolation of MP particles by density separation and spectroscopic identification [82,262,263] through Fourier-Transform Infrared Spectroscopy (FTIR, available in transmission, reflectance, and attenuated total reflectance,

ATR) or Raman Spectroscopy. The reflectance and ATR modes do not require sample preparation steps for thick and opaque microplastics [264]. These methods deliver information about polymer species, particle numbers, and size distribution [265], important parameters used to estimate the microplastics' toxicological potential. Raman method spectroscopy (<125 mm and >5 mass%) [266], micro-Raman spectroscopy (>100 µm), micro-FTIR spectroscopy (>100 µm) [267], m-Raman spectroscopy (>1 mm), m-FTIR spectroscopy (>10 mm) [268], macroscopic dimensioned near-infrared (NIR) in combination with chemometrics (>10 mm and 1 mass%) [266], and hyperspectral imaging technology (0.5–5 mm) can be used to identify plastics type, but these techniques are time-consuming, and they can be expensive and result in an uncertain extrapolation of MP quantity [269]. For example, the ATR mode produces stable spectra even from irregular microplastic surfaces; however, a lot of microplastics are weathered or made of complex materials, thus an experienced operator is necessary for the acquisition and interpretation of the spectra. Moreover, the probe used in ATR is made of germanium and can be easily damaged by hard and sharp particles remaining on filter papers [264]. The Raman signal is suitable for particles below 20 µm, but the spectra are heavily influenced by dyes, additives, and organic and inorganic substances. In addition, some materials exhibit fluorescence, masking the vibrational information [270].

The second strategy is based on thermal decomposition coupled to mass spectrometry, which is more suitable for MP quantification. Unice et al. [253] developed a method for polymers of tires (sample amount of 0.5 mg) using pyrolysis-gas chromatography mass spectrometry (pyr-GC-MS). Quantification was based on specific pyrolysis products and deuterated internal standards. The method needed only minor sample preparations and yielded a limit of detection of 14 µg/g of tire tread in soil and sediments. For the quantification of thermoplastics, Duemichen et al. [254] described a combination of thermal extraction with thermogravimetric analysis and subsequent analysis with thermal desorption gas chromatography mass spectrometry (TDS-GC-MS) that required a sample quantity of more than 20 mg. After pyrolysis at 600 °C and enrichment of the degradation products on solid-phase absorbers, plastic species were quantified by GC-MS analysis detecting polymer-specific decomposition products [271]. The deficiencies of these methods are the expense of the equipment and the complicated operation process.

Identification of microplastics passes through visualization analysis to characterize the color, shape, and light transmission, using naked eyes or analytical methodologies like optical microscopy [34,272,273]. Different microscopy techniques can be used according to the dimension and the physical properties of the MP samples. A stereomicroscope, thanks to two separated optical paths, allows for obtaining three-dimensional images of a specimen. Polarized light microscopy permits the acquisition of images at different heights [274]. Electron microscopy is useful for ambiguous particles [275,276]. In particular, electron microscopy coupled with X-ray (SEM-EDS) is a useful analytical technique to obtain high-resolution and chemical information and can identify plastic particles in complex matrices. Despite the efficiency of this technique for the chemical and morphological characterization of particles, there are some disadvantages associated with SEM-EDS analysis, such as a long time for the sample preparation and analysis, instability of some polymers, complex protocols for the analysis, and high costs of instrumentation [252].

Recently, new advanced techniques have been proposed for the characterization of microplastics, and sometimes, microplastics were produced in the laboratory to obtain standard reference materials. Monteleone et al. [277] used fluorescence lifetime imaging microscopy (FLIM) to characterize microplastics. Several kinds of microplastics produced with a cryogenic swilling mill, with and without heat treatment, were subjected to FLIM with excitation wavelengths of 470 nm and 440 nm. The fluorescence lifetimes of each microplastic were evaluated and compared and no significant differences emerged in the fluorescence lifetimes of PET samples coming from different countries; therefore, the lifetimes measured did not depend on the origin of the microplastics.

Berto et al. [278] reported a preliminary characterization of carbon stable isotopes ($^{13}\delta$) of different plastic polymers. Stable isotope analysis is a technique that measures the relative abundance of stable isotopes giving an isotopic ratio that can be used as a research tool. It is widely used in food analysis [279,280], in forensic cases [281], and in medical diagnostics [282] for the monitoring of indoor and outdoor air quality [283,284] and the characterization of commercial cleaning products [285]. This technique has been applied only rarely to assess the presence of microplastics. A proof-of-concept study was performed with fully labeled ^{13}C polyethylene to follow its fate across the aquatic microbial–animal interface. It emerged that the biodegradation of PE-MPs (<100 μm) occurred in natural waters, becoming part of nutritionally valuable biomolecules for aquatic organisms [286].

In [278,287], standard petroleum and plant-derived polymers were analyzed to estimate the carbon isotope ratio. The $^{13}\delta$ values of several polymers spanned over a wide range: PTFE, silicon, and ABS showed $^{13}\delta$ values between −41% and −35%, petroleum polymers had 13 values between −34% and −24%, and PLA (a biodegradable polyester derived from the fermentation of starch and condensation of lactic acid) had a value of −14%. Thus, this technique showed an interesting perspective for the discrimination between petroleum and plant-derived polymers in aquatic environments. Furthermore, unlike vibrational spectroscopy techniques such as Raman, it is not sensitive to variables such as colors.

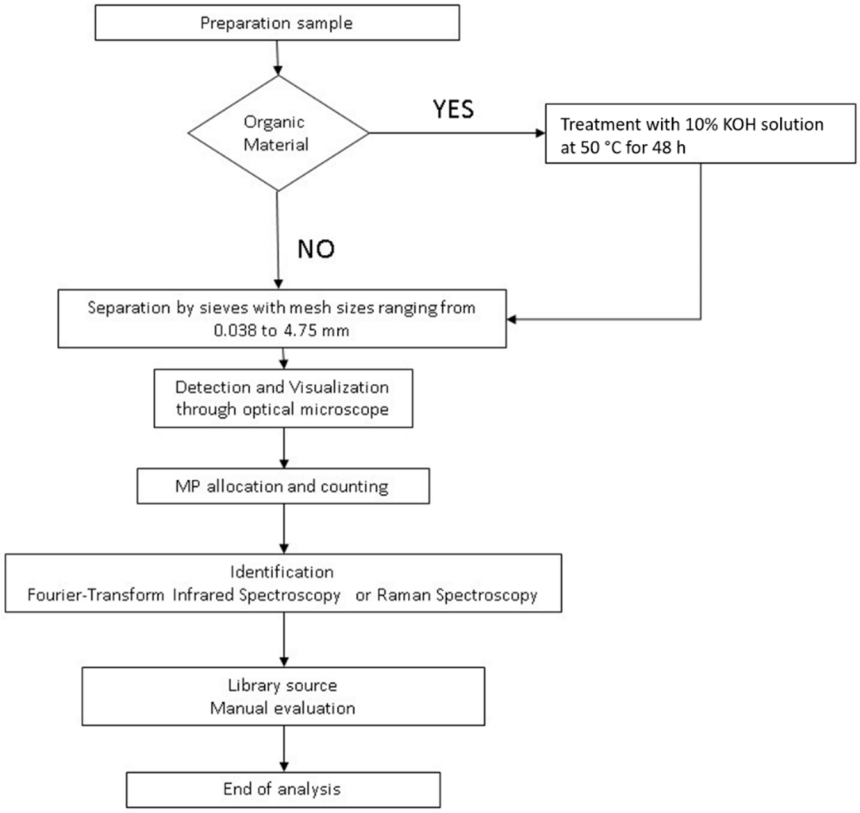
Another analytical method includes high temperature gel-permeation chromatography (HT-GPC) coupled with an IR detector. HT-GPC-IR can assess polyolefin microbeads in aqueous environments and more complex matrices such as personal care products [288] and also gives the distribution of molecular masses.

In conclusion, FTIR and Raman spectroscopy are commonly used for identifying the polymeric composition of microplastics, whereas mass spectrometry-based methodologies, even though they require a lower amount of sample, are more expensive and trickier to operate. The same can be said by comparing optical microscopy, the most used technique for microplastic visualization, with the most advanced techniques such as SEM-EDS.

Consequently, interlaboratory analyses should be performed to provide comparability of data regarding the chemical characterization and quantification of microplastics. In fact, the use of one technique can only lead to the overestimation or underestimation of microplastics. Fragmented microplastics were underestimated and fibers were overestimated using stereomicroscopy compared to FTIR in samples coming from sea surface microlayers and beach sand. The total abundance by FT-IR was higher than by microscopy both in the sea surface microlayers and beach sand samples. Stereomicroscopy mainly allowed for the identification of 50–100 μm fragmented microplastics, while FTIR also identified smaller sizes [289].

Therefore, in Scheme 1, we reported a possible simple protocol for the identification of microplastics in aqueous samples that can include a first preparation step of the sample (identification of organic matter presence with the preliminary treatment of degradation), separation with sieves with different mesh sizes, a visualization step with a microscope to count and estimate the sizes of the MPs, and in the end, a screening with FT-IR or Raman to identify the chemical composition of the MPs.

Although new accurate and sophisticated analytical methods have been reported for microplastic detection and characterization, FT-IR and Raman spectroscopy remain those commonly used for identifying the polymeric composition of microplastics due to their inexpensiveness and user-friendliness.



Scheme 1. Flowchart for the procedure of MP sample data analysis for full detection, identification and quantification.

3.3. Transport of MP to Humans

Microplastics detected in the water environment can reach the human body through ingestion [290,291] of sea products and also drinking water [292] and related beverages. Several papers considered the occurrence of microplastics in marine organisms and their ingestion, sorption, bioaccumulation, and translocation, identifying the presence of plastics in the gastrointestinal tract of marine animals [293,294], causing concern worldwide to MP transport to humans [87–90]. Microplastic particle uptake by fish can be passive (e.g., gill water filtration) and active through ingestion both of particles themselves or contaminated prey [295].

The principal works that gave microplastics concentration as the number of particles/g or particles/individual are reported in Table 4, with a focus on marine species, analytical method used, size range, and polymer type.

Table 4. Occurrence and quantification of microplastics in marine species.

Marine Species	Analytical Method	Abundance Average (Range)	Size Range	Polymer Type	Ref.
Whales	MO and FTIR	-	0.3–7 mm	RY, PES, acrylic, PP, PE	[296]
Atlantic Herring, Sprat, Common Dab, and Whiting	MO and FTIR	-	0.300–0.400 mm	PMMA	[297]
European anchovies	MO and RMS	-	0.124–0.438 mm	PE, styrene/acrylonitrile	[298]
Bivalves (*Mytilus edulis* and *Crassostrea gigas)*	MO and RMS	0.36–0.47 particles/g	>0.005 mm	-	[290]

Table 4. *Cont.*

Marine Species	Analytical Method	Abundance Average (Range)	Size Range	Polymer Type	Ref.
Mussels (*Mytilus edulis*) and lugworms (*Arenicola marina*)	MO and RMS	0.2–1.2 particles/g	0.015–1 mm	-	[293]
Dogfish, hake, red mullet	MO	1.56 particles/individual	0.38–3.1 mm	-	[299]
Semipelagic fish	MO	3.75 (2.47–4.89) particles/individual	0.5 mm	-	[300]
Pelagic and demersal fish	MO and FTIR	1.90 particles/individual	0.13–14.3 mm	PA, cellulose, RY	[301]
Benthic and pelagic fish	MO and FTIR	0.27 particles/individual	0.217–4.81 (average 2.11) mm	PP, PE, ALK, RY, PES, NY	[302]
Different fish species	MO and FTIR	2.36 particles/individual	average 0.656 mm	polystyrene: isoprene, PE, PP	[75]
Red mullet (*Mullus surmuletus*)	MO and FTIR	(0.32–0.68) particles/individual	-	PET, CPH, Polyacrylate, PAN	[303]
Deep benthic invertebrates	MO and FTIR	1.582 particles/g	0.023–6.25 (average 1.191) mm	ALK, PES	[139]
Benthic organisms	MO and FTIR	(1.7–47.0) particles/g	0.05–5 mm	PP, PE, PS, PET, NY	[121]
Mussels (*Mytilus edulis*)	MO and FTIR	0.9–4.6 particles/individual 1.5–7.6 particles/g	0.033–4.7 mm	CPH, PET, PES	[304]
Thamnaconus septentrional	MO and FTIR	1.1–7.2 particles/individual 0.2–17.2 particles/g	0.04–5 mm	CPH, PET, PES	[305]
Different fish species	MO and FTIR	2.5 particles/individual	0.2–5 mm	PE, PP	[136]
Bivalve (oyster, mussel, Manila clam and scallop)	MO and FTIR	0.97 (0–2.8) particles/individual 0.15 (0–1.8) particles/g	0.1–0.2 mm	PE, PP, PS, PES, PEVA, PET, PU	[306]
Deep-sea fish	MO and FTIR	Stomach: 1.96 particles/individual and 1.56 particles/g; Intestine: 1.77 particles/individual and 4.89 particles/g	<1mm	CPH, PA, PET,	[307]
Indian white shrimps	MO and FTIR	0.39 particles/individual 0.04 particles/g	0.157–2.785 mm	PA, PES, PE, PP	[308]

Abbreviations: FTIR: Fourier-Transform Infrared Spectroscopy; RMS: Raman Spectroscopy; MO: optical microscope; PE: polyethylene; PP: polypropylene; PS: polystyrene; PET: polyethylene terephthalate; ABS: acrylonitrile butadiene styrene; PVA: polyvinyl alcohol; PVAC: polyvinyl acetate; PVC: polyvinyl chloride; PES: polyester; PAN: polyacrylonitrile; PA: polyamide; PTFE: polytetrafluoroethylene; PU: polyurethane; NY: Nylon; RY: Rayon; ALK: alkyd resin; CPE: chlorinated polyethylene; NR: nitrile rubber; LLDP/Oct: linear low-density polyethylene-octene copolymer; EVOH: ethylene vinyl alcohol copolymer; TPU: thermoplastic polyurethane; PVS: polyvinyl sulfate; PAS: polyacrylate-styrene; SR: synthetic rubber; PR: phenoxy resin; PCL: polycaprolactone; PEA: polyethylacrylate; PEVA: polyethylene-vinyl acetate; EPM: polyethylene-propylene; EP: epoxy resin; CPH: cellophane.

Researches have shown that mussels [304], shellfish (including crustaceans and bivalves), commercial fish [302], and also cultured organisms [309] are often contaminated with microplastics. Moreover, several papers identify potable water, bottled water and beverages [310–314] as potential sources of MP. Recently, annual microplastics consumption, expressed as particles/person, was estimated to be 11,000 from shellfish [290], 37–1000 from sea salt [315]. In addition, Cox et al. [292] evaluated the total annual microplastics consumption based on microplastic presence in commonly eaten food and their daily intake in the American diet. The annual estimated intake of microplastics depends on age and sex, ranging from 39,000 to 52,000particles/person. Moreover, a great increase in the amount of

microplastics intake is observed in individuals who drink water only from plastic bottles with respect to those who consume only tap water (additional 90,000 annual microplastics particles compared to 4000).

Notwithstanding the above discussed works assert an elevated microplastics intake, a deeper investigation on the translocation and accumulation of MP in the human body is needed to better characterize their potential to harm humans. In fact, MP occurrence and detection in fluids of the human body remains a poorly investigated field, but it could be very useful to assess the interaction of plastic particles with human apparatus.

4. Conclusions

Plastic microparticles accumulated over the environment increase stress for the marine, freshwater, and terrestrial ecosystems. In this review, we analyzed the most recent literature related to microplastics in the aquatic environment, highlighting contamination of water (river, lake, wastewater, seafood), and the strength and limitations of current analytical methods employed in microplastics detection and their transport to humans.

Based on our investigation of literature data concerning microplastic pollution in the water environment, some conclusions could be made:

- As inferred from the data reported above, a standard procedure for sampling, counting and confirmming the presence of microplastic is still not present in the literature. These aspects result in concentrations expressed in different units and also diverse data obtained from the same authors depending on the sampling method. Consequently, the comparison between MP abundance values reported in various works regarding the water compartment is sometimes difficult or impossible. We suggest improving studies in developing standardized protocols to collect, process, and analyze samples in order to obtain data useful for assessing a real contamination issue.
- Although FT-IR and Raman Spectroscopy are useful techniques for identifying the polymeric composition of MP and Pyr-GC-MS and other MS-based methodologies may be applied to monitor microplastics and to indicate the type of polymer, we think that interlaboratory analyses should be performed to ensure comparability of chemical characterization and quantification data and suggest the more proper analytical method based on the matrix sampled and particle sizes.
- Most of the studies showed evidence of nano and microplastics accumulation in sea products, which are part of the human diet, and recently also in drinking water and related beverages. However, in our opinion, a deeper investigation of the transport and accumulation of MP in humans, and the consequent effects on human health, is needed.

In conclusion, we believe that in this field future research efforts should focus on developing standardized and reproducible methodologies and analytical strategies capable of counting and characterizing MP in all environmental matrices, especially fluids of the human body, which are poorly investigated, to better characterize their risk for humans.

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