

# Microplastics in the environment and the food chain

## 1. Summary

Plastics are used, due to their excellent properties, as materials in a growing number of applications. Recently, the subject of microplastics in the environment and the food chain has been discussed extensively. Several studies show the magnitude of the pollution of microplastics in sewage treatment plants, waters (rivers and lakes), oceans and shore sections, as well as fishes, mussels and invertebrates. Plastics decompose under the influence of various environmental factors. Generally, plastic particles with a size of less than five millimeters are referred to as microplastic particles.

This article gives an overview of the term “microplastics”. It describes the definition, occurrences, sources and analytical testing approaches in general, and at WESSLING in particular. Also, additional investigations that are required to develop validated methods for sampling and analysis are discussed, after clarification of the potential risk to various organisms.

## 2. Introduction

Global plastic production is still growing, reaching 311 million tons in 2014. In Europe, it seems to be stable over the last ten years, with an amount of around 60 million tons annually. 39.5% of these plastics are used for packaging and as these are single-use products (that will turn into trash in the same year), this sector alone generates around 24 million tons of plastic waste in Europe each year. In total, 25.8 million tons of post-consumer plastic waste was reported and treated in 2014 in the EU (8 million tons were landfilled, 7.7 million tons were recycled and 10.1 million tons were recovered) [1] which is only a little bit higher than the 24 M tons yearly packaging material production. Other plastic markets (e.g. automotive, building & construction, electric & electronic equipments) could contribute to the reported waste quantity, even though short-term obsolescence is not general here, but used products from previous years will continuously appear in the waste stream (and presumably with values much higher than 1.8 M tons per year). This gap between production and waste data confirms that remarkable amounts of plastic waste end up in the environment. Supposedly, the primary source of plastic waste is the littering of packaging material.

Commonly used plastics are very resistant to environmental damages, causing persistence. Plastic debris has accumulated both in the terrestrial and marine environment [2]. Table 1 shows the typical types of polymers which have been identified as microplastics in the environment.

The term “microplastics” classifies a group of plastics according to their size. The size of the particles of microplastics is defined by a general, but not regulatory, terminology as being between 1 and 5 mm. Microplastic particles found in the environment can be classified as primary or secondary microplastics. Primary microplastics are industry produced particles with determined sizes and shapes. These kinds of particles are either used as granules for the manufacture of further plastic products or directly, for example, as abrasive materials in consumer products. However, secondary microplastics are disintegrated fractions of plastic litter in the environment. Fragmentation of plastics in the environment is carried out by mechanical, chemical, physical and biological stress and leads to microplastic particles. One significant factor is UV light – UV-B (~295-315 nm) and UV-A (~315-400 nm) [4] – which provides the activation energy required to initiate the incorporation of oxygen atoms into the plastics [5]. This process causes

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chain cleavages in the polymer structure and results in smaller and smaller fragments (photodegradation). Even if disintegration occurs, it is not followed directly by biological degradation in the case of conventional plastics, because the fragments' molecular weight might be still too high to be available for microbes. This is the main reason for one of the most important problems of the ecosystem: the presence of (micro) plastics in the environment.

The increasing number of studies worldwide, identifying microplastics in different environmental matrices, such as water and sediments, is an indication of a global contamination made by humans. Monitoring of this huge pollution is still in its early stage. A problem in terms of the observation, identification and elimination of microplastic particles is that even though their number highly exceeds that of larger plastic items in the marine environment, their mass still comprises only a small proportion of the total [6].

The following sections describe microplastics in the environment and the food chain, their sources and potential risks to different organisms.

### 2.1. Microplastics in the environment

Small plastic pieces were first observed in the marine environment in the 1970s. In the past years, plastic fragments smaller than 5 mm have been referred to as microplastics (MPs). In some cases, further differentiation is made for particles smaller than 1  $\mu\text{m}$  (nanoplastics) [7]. A harmonised terminology in the European Union is still missing. The Technical Subgroup on Marine Litter (TSG ML) proposed size classes and terms related to the typical dimensions of the affected organisms and the industrial applications of plastics (Table 2) [8].

Microplastics can be divided into two groups. Primary microplastics were originally designed to be in this small size range, these are usually used in cosmetics (e.g., facial or hand cleanser). Secondary microplastics come from the disintegration of larger plastic debris [9]. While secondary microplastics are slowly generated from waste which has been released into the environment, primary microplastics are directly emitted through wastewater systems. Statistics show that the plastic content of cosmetics products is minor. Around 6% of the ingredients in liquid soaps and 0.6% of skin cleansing products contain microplastics [10]. Furthermore, microplastics will be replaced by the industry by 2020 in Germany [11].

The proportion of secondary microplastics in the environment is much higher than that of primary microplastics. However, a detailed quantification and statistics are still missing. One reason for this is that validated methods to monitor and measure the particles in different matrices like water or sediments are missing.

Most of the studies investigate microplastics in the marine environment, but in the last years, studies of limnic systems, e.g., surface water of lakes and rivers have been published. Pollution in lakes on different continents has been detected: Lake Garda in Europe [12] and the Laurentian Great Lakes [13] surrounded by more developed environment showed microplastics contamination, as well as such remote locations as a mountain lake in Mongolia [14].

Rivers can be the major pathways of plastics. Based on measurements on the Austrian Danube, more than 1500 tons of plastics smaller than 5 cm enter the Black Sea annually [15]. It is a very good example, that the Austrian Environmental Protection Agency pays attention to the topic and actively conducting measurements [16]. This attitude promotes discussion between scientists and the authorities, contributing to the future legislation needed. The pollution occurring in Austria should affect the lower region of the river as well, but it has not been examined yet. Throughout the Rhine River, microplastics between 300  $\mu\text{m}$  and 5 mm have been also identified at all of the 11 sampling points, showing a peak concentration in the Rhein-Ruhr area (15-20 particles/ $\text{m}^3$ ) [17]. In addition, freshwater systems, which are related to drinking water quality, are also objects of interest to be analysed in terms of microplastics. Instead of microplastics having been detected in water bases widely, only one occurrence has been reported in drinking water. This value of seven particles/ $\text{m}^3$  was also supposed to have originated from an abraded seal or pipe and not from groundwater [18].

Microplastics also have been detected in wastewater treatment plants (influent, effluent) [19] and in different sediments (beach, deep sea, freshwater lake) [20]. In Germany, 12 wastewater treatment plants were studied; plastic particles in the effluent ranged from 100/ $\text{m}^3$  to 1500/ $\text{m}^3$ , predominantly in the size range of 50-100  $\mu\text{m}$ . A final filtration system installed in one of the plants reduced the number of particles from 1131/ $\text{m}^3$  to 29/ $\text{m}^3$  [21].

Unfortunately, the results of all of the studies cannot be compared properly, because of the different sampling and identification methods, as well as the different dimensions used in quantification. Table 3 shows the different units of 43 studies, known by us.

Most of the studies examined matrices such as fluids or sediments. However, air is another potential environmental element that could contain microplastics, as was suggested by a study. Indoor and outdoor air samples taken in France contained microplastic particles (50-80% of them in the range between 100-500  $\mu\text{m}$ ) [22]. Their presence could cause direct human health hazards, because small particles can enter the lungs easily. Moreover, PM2.5 size range microplastics have been identified from the wear of tyres [23], and these small size particles can persist in the respiratory system.

## 2.2. Relevant sources and potential pathways of microplastics

During everyday life, several plastic items are used. Our society is dependent on plastic products and is using this material nearly everywhere from plastic packaging materials to textiles and tyres of vehicles. To analyse the sources of microplastics in the environment, a differentiation of primary and secondary microplastics is necessary. Beyond their origin, it is important to investigate their distribution, but in the end, entry into the environment has to be prevented.

Primary microplastics are used in cosmetics as abrasive detergents, but also for other applications such as lubricants, carriers of pigments, additives or water softeners. Additional applications in medicine are also possible as vectors for active pharmaceutical ingredients. In some technical processes, well-defined granules, so called micronized synthetic waxes are used. These are usually applied in coatings, pigments/masterbatches, adhesion promoters, inks, paints or food coating [8].

Most of the studies reported that the main problem of the pollution was the generation of secondary microplastics due to the fragmentation of the debris. Littering (i.e., discarded plastic items) is the main issue, especially in countries with no waste management systems. Furthermore, plastics are persistent in the environment and degradation, depending on the type of plastics, takes decades or centuries. The discharge of synthetic fibres of textiles are another source of secondary microplastics [8]. During the washing and, potentially, the drying process, huge quantities of fibres end up in the sewage system [24]. Moreover, sources such as abrasion of tyres and loss of pellets in the manufacturing process can also be mentioned, however, more studies monitoring plastics in air are needed. A discrimination of land-based and sea-based sources could provide another view of possible pathways (see Table 4.)

The overview (Table 4) shows that, in terms of land-based sources, besides individual littering, the input of factories and sewage plants contaminate the environment. At the end of this route, parts of the plastic waste end up in freshwaters or marine environment.

## 2.3. Sorption of harmful substances

In recent years, more and more studies highlight the threat of microplastics to the ecosystem, not only physical injury and ingestion (causing, e.g., inflammation of the stomach/gut), but also chemical exposure through the uptake of pollutant-loaded microplastic fragments (pollution transport, co-pollution). One way is the leaching of additives from polymers. Many of them are classified as toxic or are endocrine disruptors, e.g., bisphenol A (BPA), phthalates and flame retardants, such as polybrominated diphenyl ethers (PBDEs) [26].

On the other hand, persistent organic pollutants (POPs) have very large water-polymer distribution coefficients, in the favour of plastics, so they are effectively adsorbed on microplastics from the surrounding water [27], [28]. Analytical methods were developed to extract, concentrate and identify POPs that may have accumulated on plastic fragments and plastic pellets. The results of this study confirm that plastic debris is a trap for POPs [29]. This sorption enhances their bioaccumulation properties, making them more easily introduced to the fauna in a concentrated initial dose.

The most investigated pollutants relating to sorption on MP surfaces are polycyclic aromatic hydrocarbons (PAHs, predominantly phenanthrene), dichlorodiphenyltrichloroethane (DDT) and its breakdown products, and polychlorinated biphenyls (PCBs). The variation in the concentrations of adsorbed contaminants can be very high. PCBs and PAHs were detected in all of the samples taken in the open ocean and on beaches. Concentrations show high variability in different fragments (PCBs: 1-436 ng/g; PAHs: 1-9300 ng/g), but a trend has been established, showing that fragments from urban beaches have higher concentrations than those taken on remote beaches and in the marine environment. Concentrations of DDT and its breakdown products ranged from 0.2 to 124 ng/g [30]. Adsorption of two components, that are currently in the centre of interest, on MPs was also investigated by a study: di-2-ethylhexyl phthalate (DEHP) and perfluorooctanoic acid (PFOA). PFOA showed little affinity to be adsorbed either on polyethylene (PE) or polyvinyl-chloride (PVC), and the same was true for DEHP and PVC. However, DEHP was as effectively adsorbed on PE as DDT both on PE and PVC under simulated physiological conditions [31]. It has already been reported that these adsorbed pollutants can migrate to different species: pyrene contaminated MP localisation in tissues occurred in mussels and there a marked accumulation was shown [32]. Regarding higher trophic levels, a bird feeding experiment confirmed the transfer of PCBs from ingested plastics [33].

The effect of microplastics on the transport of potentially toxic elements has not been studied many times yet. The effect of chromium(VI) was tested in fish (*Pomatoschistus microps*); levels of lipid peroxidation significantly increased in individuals which were introduced to Cr(VI) and 1-5  $\mu\text{m}$  polyethylene (PE) spheres. This did not occur under Cr(VI) or MP exposure alone [34].

Another study highlights that the presence of microplastics had no effect on the bioavailability of silver in fish (*Danio rerio*). 10-106  $\mu\text{m}$  PE beads incubated with silver before exposition significantly reduced Ag uptake during the test [35]. This emphasizes that the investigation of inorganic pollutants cannot be omitted, because these can also be adsorbed on MPs. Release after ingestion needs further studies.



## 2.4. Microplastics in the food chain

The effect of plastic waste on the ecosystem has become obvious over the past years: many sea turtle, seabird and marine mammal species are affected. The ingestion of plastic debris leads to mechanical or physical stress, and the inner organs of the organisms are injured. Secondly, entanglement in packaging bands, synthetic ropes and lines, or unleashed drift nets (ghost nets) can cause bodily harm which could also result in the passing away of the animals [36].

Microplastics in waters are introduced into the food chain by mistaken identity or indiscriminate ingestion [28]. Because of their size and presence both in benthic and pelagic zones, main consumers besides zooplankton are invertebrates and fish species.

Based on laboratory feeding tests, MPs in the size range of 1-30  $\mu\text{m}$  are ingested by zooplankton [37]. Trophic level transport of 10  $\mu\text{m}$  polystyrene spheres from mesozooplankton to a higher level (macrozooplankton) has been observed [38].

Plastic ingestion has already been described widely in low trophic fauna such as in sea cucumbers [39], lugworms [40], brown shrimps [41], seaweeds [42] and crabs [43]. Mussels in five European countries have been investigated for microplastics: only the Danish sample showed negative results. The numbers of detected MPs in samples from France, Spain, Italy and the Netherlands ranged from 0.04 to 0.34 pieces per gram of tissue [44]. Microplastics uptake by mussels (*Mytilus edulis*) was described in other studies as well [40], [45], [46], it seems the most popular low trophic test organism currently. Further, microplastics are able to translocate to the circulatory system [46] and tissues in invertebrates (mussels, crabs) and transfer to higher trophic levels (crabs) [45].

In case of Vertebrates, the digestive tracts of fish species have been investigated at different points of the European coastline. In the North and Baltic Seas, three demersal fish species (cod [*Gadus morhua*], dab [*Limanda limanda*], flounder [*Platichthys flesus*]) and two pelagic species (herring [*Clupea harengus*] and mackerel [*Scomber scombrus*]) have been sampled. MPs have been detected in all fish species in the demersal zone, in an average of 3.4% of the individuals (with a mean number of  $0.03 \pm 0.18$  plastic items per fish). If we compare this result with the average ingestion rate observed in pelagic species, we would see that MP ingestion is more probable in the pelagic zone (10.7%, mean number of  $0.19 \pm 0.61$  items per fish). Here it is very important to make a distinction: no microplastics have been detected in herring individuals, and mackerels show the highest ingestion rate (17.7%). It is assumed that herrings were in a stage of maturity where they showed a reduced feeding activity, because most of the individuals had empty digestive tracts. Considering only the contaminated individuals, 44% were related to the

demersal zone and 56% to the pelagic zone. Almost 40% of the ingested particles were polyethylene (PE) [47]. The distribution of ingested microplastics was reversed at the Portuguese coasts: 36.5% of contaminated individuals were pelagic feeders and 63.5% of them were benthic. MPs were detected in 17 species, with an ingestion rate of over 30%. The mean number of ingested microplastics was  $0.27 \pm 0.63$  per fish regarding the whole sample. Considering only the affected individuals this value is  $1.40 \pm 0.66$  per fish [48]. In the Adriatic Sea MPs were detected in 28% of the 125 individuals. The average number of MPs extracted in positive fish ranged from 1 item/individual to  $1.78 \pm 0.97$  items/individual. 65% of the particles were identified as PE and 19% as polyethylene-terephthalate (PET). PE and polystyrene (PS) particles were also extracted from the liver tissue [49]. A French study – sampling 11 water streams for common gudgeons (*Gobio gobio*), a fish which is common throughout Europe – found microplastics in 12% of the investigated digestive tracts [50]. It is important to note, that investigation of micron sized MPs are underrepresented in field collected organisms (especially in fish stomach and gut), because of the detection methods. As laboratory feeding experiments confirm the easy uptake of  $<100 \mu\text{m}$  particles (usually fluorescent polystyrene particles,  $\sim 10 \mu\text{m}$ ) [37], [42], and the translocation of these small particles through the food chain can occur easily, so it is highly recommended that they are monitored in field collected organisms as well.

As foods are not tested widely for MPs, data in this field are very limited. The “effects on the ecosystem” approach provides data showing that, through the food chain, human food is also affected. MPs contamination in lower trophic level organisms are described several times, some of them are subject to direct human consumption (e.g., the mussel *Mytilus edulis*). Microplastics in fish were mostly present in the digestive tracts, which are usually not consumed. However, since translocation was proven in Invertebrates, it can also be expected in fish (one result already showed translocation to the liver).

All areas of life over the last centuries have been penetrated by plastics, so human ingestion of MPs can be presumed through other channels, i.e., food contact materials. Co-pollution through microplastics can also contribute to the contamination of food, but – unlike microplastics – most of these components are nowadays investigated before distribution or during authority inspection. The presence of microplastics in the Invertebrate and Vertebrate biota, the negative effects of co-pollution warrants for inspection in the most affected fields (i.e., fish) to maintain the high quality food chain safety.

In conclusion, data are very limited and very diverse in terms of geographical locations, habitats and organisms. In the case of environmental samples, biota of freshwater is less investigated. In general, pub-

lished data are based partially on different experimental methods and different quantity dimensions. To get representative results or to get an overview of the presence of MP in the food chain, scientific results are needed.

### 3. Risk potential - Impact of microplastics on organisms

A high potential of adverse effects by microplastics on organisms in oceans, rivers and seas, as well as creatures living on land is assumed. The first studies in recent years showed physical effects on organisms, for instance injuries due to the morphology, density or size of the particles. Furthermore, factors like accumulation and translocation also influence physical impact [51]. The behaviour of a 1 µm particle is different from that of a 5 µm particle. This means that toxicity could be totally different. For example, a particle greater than 1 µm which is ingested with food might be released by the gastrointestinal organs. However, particles smaller than 1 µm could be transferred through cell membranes. Additionally, consideration of two other aspects is necessary to estimate risk potential. One is leaching of additives that are incorporated into plastic, and then released over time into the environment, and the other one is the sorption of harmful substances (see chapter 2.3) [16].

Figure 1 shows an overview of the potential pathways for the transport of microplastics (especially in seas or rivers) and their biological interactions.

The process starts by the fragmentation of primary or/and secondary microplastics due to UV radiation, followed by mechanical and microbial degradation. Plastics are present in different areas and compartments. Because of their density, particles can swim at the water surface or sediment to the bottom. Sinking due to biofouling is also feasible. Hence different organisms living in different habitats like fishes and zooplankton, mussels or crustacean or other invertebrates can ingest microplastics. Furthermore, the ingestion of plastics depends on particle sizes and shapes. An overview of organisms which are susceptible to ingestion of microplastics in the context of exposure pathways is shown in Table 5.

Impacts on organisms due to the ingestion of macro- and microplastics are possible and have been observed several times. For example, blockages in the digestive system or abrasions from sharp objects, blockage of enzyme production, diminished feeding stimulus, nutrient dilution, reduced growth rates, lowered steroid hormone levels, delayed ovulation and reproductive failure or embedding of small particles in tissues [51].

Direct health hazards (i.e., direct consumption) on birds, reptiles and mammals (such as humans) have not been specified yet, but a very serious indirect hazard is that microplastics are able to translocate to

the circulatory system [46]. This predicts their presence in animal tissues and supposes the transfer to mammals and also the translocation in their bodies.

### 4. Identification of microplastics in different matrices

Currently, a general method for the analysis, identification and quantification of microplastics, and to determine the source of entry in the environment is not present and developed. The strategy to identify microplastics – including sampling, sample preparation and analytics – depends on different factors, such as the matrix (water, sediment, organisms, etc.), or the location of the plastics or the possible contamination. In this chapter, actual strategies are demonstrated. Additionally, the experience of WESSLING will also be presented.

#### 4.1. Sampling

The basic differences in the sampling methods used are related to the nature of the matrices sampled, water and sediments. During the sampling of water, volume reduction is the principle technique. An advantage of volume reduction is that only the solid material found in a huge volume (m<sup>3</sup>) will be collected and this way further handling in the laboratory is very convenient. For the sampling of rivers or seas, a neuston or plankton net is used with a mesh size of >300 µm (Figure 2). This method is useful for the sampling the surface of water bodies, but sediment plastics cannot be captured.

For the sampling of sediments, a bulk technique is applied. In this case, the volume of the material (in general, solid inorganic material (sand)) is not reduced. Bulk samples are most appropriate when microplastics cannot be easily identified visually, because they are covered with sediment particles and their abundance is low, and would require the sorting of a large amount of sediment [3]. Direct sorting by sieving prior to carrying out compositional analysis is practical.

Additionally, it is important to record the amount of the sample taken, in volume or mass, to be able to reference the identified plastics. Both techniques require further sample preparation in the laboratory.

Sampling for environmental analysis at WESSLING, shown here at a wastewater plant, was carried out by collecting up to 1 m<sup>3</sup> of wastewater through a pump system equipped with a filter (concentrating a high volume of sample). Figure 3 presents the sampling.

#### 4.2. Sample preparation

The collection of samples in the environment was described in chapter 4.1. In the next step, the separation of plastic particles or fibers from the sediments and organics (e.g., plant matter), which would have



an effect on the analytics, is necessary. For this purpose, different strategies can be used. Due to the different densities of water, sediments and plastics, density separation and sedimentation in solutions, such as sodium chloride or zinc chloride, are useful. Imhof et al. developed a device called the Munich Plastic Sediment Separator (MPSS), which they used to prepare sediment samples and to separate inorganics from plastics by floating [53]. The principle is shown in **Figure 4**.

In general, organics have to be kept out of the sample. This can be achieved by bases, acids, oxidizing agent or enzymes. These procedures, especially degradation via enzyme treatment, can also be used for the sample preparation of organisms and foods.

In the last step of sample preparation, plastic particles are collected on aluminium oxide or gold filters. Figure 5 describes the principle of separation and purification of plastics in a collected water sample at WESSLING GmbH. The concentrated residue on the filter (15 µm mesh size) was removed and then chemically treated, followed by density separation. In the last step, particles were separated from the liquid surface and filtered.

### 4.3. Identification of microplastics

The identification of microplastics is carried out by Fourier transform infrared (FTIR) [3], [54] or Raman spectroscopy [12], [53]. Both the FTIR and Raman methods determine polymers through the energy absorptions of specific functional groups. The methods, in combination with an optical microscope, are suitable for the analysis of different particles in terms of size, structure and polymer type. FTIR microscopy is able to analyse particles which are larger than 10 µm and Raman microscopy has a resolution of 1 µm. A limitation of these optical techniques is that impurities like biofilms or organics could influence the spectra. Also, exact quantification is impossible. Alternatives for the quantification of polymers could be Pyrolysis-GC-MS or TDS-GC-MS [55], but the disadvantages are that these techniques are time consuming, not completely developed and costly.

The most commonly used technique to analyse microplastics is FTIR microscopy, at WESSLING as well. The analysis can be carried out by using either transmission mode or ATR (attenuated total reflection) mode. The transmission mode is used, when the particles are concentrated on an aluminium oxide filter. The ATR mode is applied, when the particles are concentrated on a gold filter. The FTIR method is able to analyse the sample particle by particle or it can scan a whole area to detect a huge number of particles.

Differences in the measurement methods are mentioned below. Figure 6 shows the FTIR analysis of an aluminum oxide filter in transmission mode. In this case, a particle by particle analysis was chosen and

a spectrum was recorded. In general, the evaluation of the spectrum can be done by comparison with a database. Figure 7 shows the scan of an area, also performed in transmission mode. The advantage of this technique is that an area with a high amount of measurement points could be analyzed and visualized in an image based on the characteristic signals of plastics at 1480-1430 cm<sup>-1</sup> and 1790-1700 cm<sup>-1</sup>. The image is created with a color scale reflecting signal intensity.

In the following section, results of the analysis of sea salt at WESSLING is shown. The sample was prepared as described in chapter 3.2, by dissolving the salt in H<sub>2</sub>O, density separation, chemical treatment and concentration on gold filter. Analysis was carried out by FTIR microscopy (ATR-mode). Figure 8 and Figure 9 show the microscopic images of the measurement points for FTIR spectroscopy and the spectra. The analysis of sea salt showed different fibers and particles. Part of the impurities could be identified as microplastics. According to the microscopic images, organic impurities were also present. This proved that sample preparation is the most important step, and that elimination of organics is difficult.

In comparison to FTIR analysis, results of Raman spectroscopy are shown in Figure 10. The Raman technique is able to analyse objects down to a size of 1 µm. Identification of the particles is achieved by a reference or a database.

### 5. Conclusions and outlook

In recent studies, microplastics have been identified in several environmental matrices and in biota as well. Microplastics in different environmental matrices have been investigated using different sampling and analysis methods recently. WESSLING has demonstrated the possibility to identify microplastics in environmental compartments, from sampling, through sample preparation, to analysis. For comparable results, a uniform definition of microplastics have to be established first (e.g., whether a lower size limit should be introduced beyond the upper size limit of 5 mm, in parallel with the term of nanoplastics or not). This differentiation would not be necessary if sampling and recovery methods would be standardised. Also, analysis methods to identify plastics are varied as well (FTIR microscopy, Raman spectroscopy, pyrolysis GC-MS, etc). For quantification, also in terms of size distribution, a suitable technique has to be developed. Standard methods would make validated results available worldwide. Sampling and analysis processes are not uniform yet, and many further monitoring programs are needed to gain comparable and validated results.

In terms of the field of freshwater and based on the current state of the science, the following research is needed to be done [56]:

1. Monitoring the presence of microplastics in freshwater systems.
2. Investigating the sources and fate of freshwater microplastics.
3. Assessing the exposure to microplastics.
4. Evaluating the biological effects of microplastics exposure.
5. Understanding the interaction between microplastics and other freshwater contaminants.
6. Developing a novel framework for the risk assessment of microplastics.

To realize these goals, cooperation between stakeholders (science, authorities, water works, WWTPs, NGOs) has to be enforced to gain up-to-date data on the state of the environment. Monitoring of microplastics as a descriptor of the environment should be integrated into the Water Framework Directive (20/60/EC). This initiative is also supported by the fact, that the issue of MPs is already addressed by the European Marine Strategy Framework Directive (2008/56/EC). Moreover, pollutant transport (especially regarding WFD priority substances like di(ethylhexyl) phthalate, nonylphenol, octylphenol, and PAHs) makes them promising candidates to be included in the directive [56].

Beyond the monitoring of water bodies – until limit values are not defined legally – analysis based on self-declaration of WWTPs would be recommended, as this is a significant pathway of microplastics pollution. Water works should also analyze MPs during their processes to ensure 100% quality and safety, and to ensure universal access to clean water as a basic human right.

Currently, the potential risk to organisms in the ecosystem and to humans is not determined and clarified. Some of the negative effects on the ecosystem due to microplastics have already been demonstrated (discrepancies of the digestive tract, pollution transport, translocation to tissues) but there are several more, presumably harmful ones which are not known yet, and so further research is needed. In this context, areas of further research projects should address [51]:

- Destination of ingested microplastics within organisms and their physical effects.
- Impact of different shapes and plastic types on organisms.
- Bioavailability of sorbed persistent organic pollutants (POP) and other contaminants.
- Transfer of microplastics to higher trophic levels.

Additionally, the potential hazard on food safety and human health have to be investigated. Presently, only minor activities are ongoing.

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