





Introduction

After three years of dedicated microplastics research, four projects (BASEMAN, EPHEMARE, PLASTOX and WEATHER-MIC) funded under the framework of JPI Oceans have presented their results. The projects focused on defining baselines and standards for microplastics analysis, understanding the ecotoxicological effects of microplastics and their associated pollutants, and investigating the weathering of plastics in marine waters.



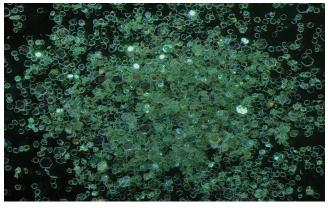






What are microplastics?

Microplastics are defined as plastic particles that are smaller than 5 mm in size, including particles in the nano-size range^{1,2}. The smallest particles studied by the JPI Oceans BASEMAN and WEATHER-MIC projects were a few micrometres in size (equivalent to less than half the thickness of a single human hair). EPHEMARE and PLASTOX tested the effects of particles down to the nano-scale as these are readily ingested by zooplankton and filter feeders. In terms of numbers, 92% of all plastic items found at sea fall into the microplastics category3. Although lower in total number of items, macroplastics (>25 mm) account for the largest mass of plastics and can break down into micro- and nanoplastics. Microplastics classify as "primary" when purposefully manufactured to small size, such as microbeads in cosmetics, and "secondary" when resulting from breakdown of any larger plastic object4.



MICROPLASTIC PARTICLES FOUND IN COSMETIC EYELINER.PHOTO CREDIT: FILIPA BESSA

The mass production of plastics dates back to the end of the 1940s. Plastics are synthetic materials manufactured from a wide range of organic polymers and have countless applications owing to their broad range of physical and chemical properties. For example, polystyrene used to make disposable cups is very different from nylon, used to make fishing nets. The top five plastic polymers contributing most to total plastics, with regard to their current production volumes are polyethylene, polypropylene, polyvinylchloride, polyurethane and polyethylene terephthalate⁵.

Furthermore, plastic products contain a wide range of chemical additives including colourants, plasticisers, antioxidants and flame retardants to give them their desired properties. The JPI Oceans microplastics projects studied more than ten types of plastics⁶ to understand how theyinteract with the marine environment and its inhabitants.

Microplastics distribution and abundance

The first report on plastic pollution in the marine environment appeared in the early 1970s7. Today we know that microplastics are found in environmental compartments all over the globe, from the tropics to the poles⁸ and from the Swiss Alps⁹ to the Mariana Trench¹⁰. The BASEMAN project provides evidence of high microplastics concentrations in various studied areas like the Fram Strait^{11,12}, Arctic Sea⁸, North Sea¹³, Rhine river¹⁴, Bergen Fjord¹⁵ and the Mediterranean Sea^{16,17,18,19}. Particularly high concentrations were found in closed systems like the Mediterranean Sea and the Bergen Fjord. A remarkable finding was that the microplastics contamination in Arctic Sea ice is among the highest in the world. Additionally, the researchers highlighted that smaller sized microplastics ((100 µm) play a major role in microplastics contamination^{8,9,13,14,15,19,20,21,22}.



The focus of the collaborative BASEMAN project was the standardisation of methods to describe the abundance and distribution of microplastics in European waters, aiming to facilitate Member State compliance with policy obligations. To achieve this goal, different analytical techniques to identify and quantify microplastics (such as Fourier-transform infrared spectroscopy, Raman spectroscopy and pyrolysisgas chromatography/mass spectrometry) were compared^{20,23}, showing the complementary nature of the investigated techniques.

BASEMAN developed new applications, data analysis tools and databases during the project. For example, the researchers generated multi-spectroscopic databases^{21,24}, to harmonise the different analytical techniques. The development of automated analysis pipelines significantly reduced the working time and personnel demand of the data analysis, also allowing the identification and quantification of microplastics independent from human bias²⁵.

A dedicated database²⁴ enabled a standardised analysis of microplastics, adopted by a series of studies in various environmental compartments^{8,9,12,13,14,15,20,21,22}. The latest version of this analysis pipeline also facilitates the separation of microplastic particles and fibres within a dataset²¹. To allow the application of these tools for various instruments from all manufacturers, BASEMAN provided the software sIMPle (Systematic Identification of MicroPLastics in the Environment)²⁶. It is available free of charge via the website www.simple-plastics.eu and has already been applied to different analytical techniques²⁷.

Inter-lab comparisons among participating BASEMAN partners, using their new microplastics reference kit, showed that additional method development (including strict quality assurance/quality control criteria) is needed to quantify the particle fraction smaller than 100 µm with meaningful reproducibility. To optimise the density separation of microplastics from sediments, scientists investigated different systems. In addition, different purification methods (such as the use of alkaline digestion, enzymatic digestion²⁸ and wet oxidation – including Fenton's reagent^{11,14}) were developed and evaluated. One major outcome was the development of a purification reactor¹³, which reduced contamination risks and improved the ease of use.

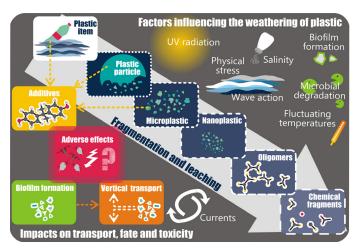
Researchers from the BASEMAN partnership embarked on joint scientific cruises in Galway Bay (Ireland) and Ria de Vigo (NW Spain) to facilitate the comparison, evaluation and standardisation of sampling methodologies for microplastics

in sediment and water²⁹. Additionally, they proposed various marine species to include in biomonitoring of microplastics in European waters. Finally, BASEMAN developed three white papers on standardisation/harmonisation protocols for monitoring microplastics in seawater, sediments and biota (in collaboration with the JPI Oceans project EPHEMARE)^{30,31,32}.

researchers highlighted possible issues biodegradable plastics, which are promoted as less harmful to the environment. During a research campaign, scientists found polycaprolactone (which is considered biodegradable) in nearly 10% of their net tows¹⁹. Under laboratory conditions, it breaks down in around 6-12 days¹⁹. However, the BASEMAN findings suggest that this type of plastic does not readily degrade in natural sea conditions and may therefore not be a viable solution to reduce marine litter¹⁹. The project further suggests that future research should focus on the sinks and sources, fates and residence times of various polymers, especially concerning the smaller-sized microplastics 8,9,13,14,15,19,20,21,22

Weathering

When plastic is introduced into the ocean, its properties change and it breaks down yielding smaller particles due to exposure to oxygen, sunlight, waves, biofilms, etc^{33,34}. These different factors influencing plastic weathering must be considered when assessing the environmental fate and impacts of microplastics. The WEATHER-MIC project focused on the effect of weathering on the physicochemical properties, surface characteristics, environmental fate and potential toxic effects of microplastics.



SUMMARY OF THE FACTORS INFLUENCING THE WEATHERING OF PLASTIC IN THE MARINE ENVIRONMENT WITH RESULTING IMPACTS ON TRANSPORT AND FATE PROCESSES AND POSSIBLE ADVERSE EFFECTS. FIGURE CREDIT: JAHNKE ET AL.³³



The research led to a novel mass spectrometry "fingerprinting" methodtoidentify plastic polymers following artificial weathering experiments³⁵. The development of advanced models, combined with monitoring data, enabled further investigation of the transport of microplastics in the water column and coastal environment. The researchers also adapted OECD guidelines for the ecotoxicological testing of chemical substances to microplastic particles. This allowed them to screen the effects of virgin and weathered microplastics and their leachates in a standardised way. Their newly developed standard testing procedure enables researchers to evaluate the fitness-related consequences of microplastics on consumers³⁶.

WEATHER-MIC also studied the effects of microbial community attachment (biofilms) on microplastics¹⁵. These biofilms can have an effect on the swimming behaviour of grazers exposed to microplastics³⁷.

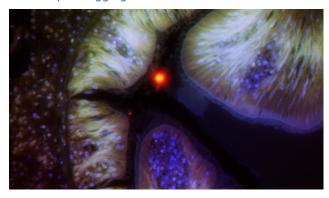
Pre-production polymers with low additive contents were shown to elicit only limited effects³⁸, indicating that additives may account for a major share of observed effects. Hence, consumer products containing WEATHERadditives demand greater attention. MIC thus highlights the importance of including weathering effects in risk assessments microplastics pollution in real world environments.

Ingestion and accumulation

Microplastics can be taken up by a wide range of marine organisms and, for example, clog their filter apparatus or gastrointestinal tracts. The EPHEMARE and PLASTOX projects both conducted field and laboratory studies investigating microplastics ingestion and potential for accumulation. Their research output included the development of methods for extracting microplastic from biological samples^{39,40}. For example, the researchers found microplastics in mussels and in the guts of crabs and wild-caught fish, highlighting ingestion is widespread in the marine environment across species with different feeding strategies and from different trophic levels and habitats^{41,42,43,44,45}. Microplastics abundance varied significantly among different species and organisms of the same species.

Filter feeders (e.g. mussels)^{46,47}, deposit feeders (e.g. beach hoppers)⁴⁸ and predators (e.g. fish)⁴⁹ being fed in laboratory conditions actively ingest, but also egest

microplastics within the size range of their common food. In most organisms, larger microplastic particles (\rangle 20 µm) do not accumulate and transit quickly through the digestive tract before being expelled. Subsequently, these faeces or pseudo-faeces accumulate in bio-deposit aggregates.



SECTION ACROSS THE DIGESTIVE TRACT OF A MUSSEL AFTER EXPOSURE TO SEAWATER CONTAINING MICROPLASTICS. THE MICROPLASTIC PARTICLE IS VISIBLE IN BRIGHT RED. PHOTO CREDIT: CATIA GONÇALVES

In mussels, particles smaller than 2 µm translocated from the digestive gland into the gills and were eventually excreted. Current models show no accumulation of microplastics inside mussels exposed to environmental concentrations. Field samples confirmed this prediction.

Laboratory studies demonstrated the transfer of microplastics from prey to predator in experimental food chains⁵⁰. However, bioaccumulation generally does not occur as the particles are excreted at a similar rate to which they are taken up. It is necessary, however, to consider that current experimental studies and field campaigns rarely detect nanoplastics due to technical limitations and costly or time-consuming analytical methods.

Effects of microplastics on marine organisms

The EPHEMARE and PLASTOX projects also studied the toxicological effects of microplastics at different trophic levels in the marine ecosystem. The leaching of toxic additives and the sorption and transport of persistent organic and metal pollutants were additional impacts investigated in the studies. EPHEMARE and PLASTOX set up collaborative experiments among partner laboratories to study the cellular, molecular, physiological and biological effects of microplastics in the marine environment.



The experiments used virgin microplastic reference materials, environmentally relevant reference materials⁵¹ and microplastic particles with added organic and metal pollutants to investigate the impacts on a wide array of species, from bacteria to fish. Toxicity to organisms was assessed using ecologically relevant endpoints, including behaviour (affecting the ability to find food and partners), reproduction, embryogenesis and mortality. To stimulate the industry to produce more environmentally friendly materials, and regulatory agencies to set environmental standards for plastics, EPHEMARE developed toxicity tests specifically suitable for microplastics⁵².

The projects tested microplastics at environmentally relevant concentrations and did not detect lethality or acute toxicity in marine organisms. Microplastics generally did not seem to induce mortality, nor significantly affect growth, nor cause mutagenic effects. Results showed only subtle effects on the organisms' health status, such as cellular and oxidative stress and a range of species-specific nonlethal effects^{53,54,55,56,57,58}.

However, in bivalves (a class of molluscs) microplastics led to a decrease of energy reserves⁵⁹. Larger (125-500 µm) particles negatively affected the efficiency of digestion due to stress, toxicity and lower nutrient uptake⁵⁹. Lugworms exposed to microplastics showed decreased feeding activity over time. Predatory fish showed a significant decrease in female growth and reproductive outcomes (i.e. a decreased larval hatching rate) when exposed to microplastics, with and without sorbed pollutants⁶⁰. In phytoplankton, a reduced growth rate and reduced chlorophyll production may occur, although standard endpoints are not very sensitive⁶¹.

Recent scientific publications have thus revealed low toxicity of virgin microplastics on marine biota^{33,35,36,37,38,39,40,60,62,63,64}. However, these studies did not focus on the additive chemicals that are present in microplastics in the marine environment. A wide range of organic and metal additives are used in the production of plastic materials and some types of plastic can contain up to 20-30% in weight of chemical additives. Studies conducted in PLASTOX and EPHEMARE have shown that chemical additives can leach out into marine^{65,66,67,68} and fresh^{50,69} waters, where they can exert toxic effects^{70,71}. PLASTOX also developed a novel method for the extraction and analysis of a wide range of polar and nonpolar organic chemicals in seawater⁷².

To fill knowledge gaps, the researchers highlighted the need to assess hotspots of microplastics pollution,

the long-term environmental effects of microplastics and to consider the full scope and cumulative impact of multiple pressures on the ocean.

Effects of sorbed organic pollutants

While microplastics release additive chemicals, they can also sorb and transport hydrophobic chemicals and metals from their surrounding media, prior to ingestion by biota. As a result, microplastics may represent an alternative exposure route for such pollutants. In addition to assessing the direct effects of microplastics, the PLASTOX and EPHEMARE projects also studied sorption of different persistent organic pollutants (POPs) to a range of polymer types and investigated the ecotoxicological impact of POPs and metals associated with microplastics on key European marine species.

In recent years, a number of studies have attempted to investigate the bioavailability of microplastics-sorbed contaminants, mostly focusing on ingestion of contaminated microplastics by aquatic species, and reporting transfer and accumulation in tissues and toxicological effects⁷³. A previous study has highlighted that under environmentally relevant exposure scenarios, the microplastics exposure route for organic contaminants is negligible with respect to the combined intake from food and water⁷⁴.

The studies conducted within the framework of the PLASTOX and EPHEMARE projects highlighted the complexity of this issue. The degree of pollutant bioavailability was partially related to the specific combination of plastic and pollutant and partially related to the experimental design and exposure conditions. The bioavailability of sorbed POPs and metals differed between species and was strongly dependent on factors such as lipid content, gut residence time of microplastics, as well as the presence of dissolved pollutants in the surrounding environment. As a result, a range of different outcomes were observed, including increased bioavailability³⁷, no significant change in bioavailability^{75,7677} and even a clear reduction in bioavailability⁷⁶. Furthermore, detailed sorption studies conducted within PLASTOX showed that POPs accumulate more in polyethylene than in polypropylene, polystyrene and polyethylene terephthalate, suggesting that both sorbent (e.g. polymer type) and sorbate properties (e.g. molecular size and polarity), as well as extrinsic environmental conditions influence the sorption process.

PLASTOX, among other recent studies, indicated that the



current knowledge on smaller microplastic particles ($\langle 10 \mu m \rangle$) and nanoplastics ($\langle 1\mu m \rangle$) is still limited. The smaller the plastic particles, the more environmental and human health risks they may pose. Additional research on their current environmental concentrations is thus of utmost importance. Since knowledge gaps remain and new research questions have arisen, JPI Oceans launched a second joint call for transnational research projects which led to the selection of six cutting-edge research projects on microplastics in the ocean.

The six selected projects (ANDROMEDA, HOTMIC, FACTS, microplastiX, i-plastic, RESPONSE) will perform research on sources of microplastics, methods for identifying smaller micro- and (nano-) plastics, monitoring their circulation in marine systems and their effects on marine organisms.

The previous JPI Oceans microplastics projects also put special emphasis on public outreach via social media, workshops, stakeholder meetings, popularized publications and awareness raising campaigns. One example is the EPHEMARE photo competition - IMPACT 2017. A complete overview is available in their project reports, which you can access through the following links:

http://www.jpi-oceans.eu/baseman

https://www.jpi-oceans.eu/ephemare

https://www.jpi-oceans.eu/plastox

http://jpi-oceans.eu/weather-mic



Background and policy context



'ZOO PLASTICS' - PHOTO CREDIT: FILIPA BESSA

Plastics in the marine environment have become a major concern because of their persistence in the environment including the oceans, and adverse consequences to marine life. According to estimates from Eunomia (2016) between 27–67 million tons of plastic can be found in the world's ocean as of 2016.

In recent years many governmental initiatives have been launched to reduce the input of plastics into the (marine) environment. A ban on microbeads in cosmetics has come into force in many countries. In January 2018, the EU published its plastics strategy that aims to transform the way products are designed, produced, used and recycled in the EU so that the current 30% recycling rate can be increased dramatically.

However, plastic pollution of the ocean is not only a European problem, but a global problem. The necessity to tackle the plastics issue was recognised by both the G7 and G20 countries. In particular, the groups of states acknowledge that marine litter, in particular plastic litter, poses a global challenge, directly affecting marine and coastal life and ecosystems and potentially also human health. Accordingly, increased effectiveness and intensity of work is required to reduce the input of plastic litter into the environment. Under the Agenda 2030 for Sustainable Development of the United Nations countries worldwide also pledge to tackle the marine litter issue.

The EU Marine Strategy Framework Directive (MSFD – 2008/56/EC) requires the maintenance or achievement of Good Environmental Status of the Seas and Oceans in order to protect marine species and habitats from human activity. The 2017 Commission Decision 2017/848/EU on the Common Understanding of the Directive replacing Decision 2010/477/EU sets out requirements to develop new criteria and specifications and standardised methods for monitoring and assessment. The Decision requires the development of new mandatory criteria under Descriptor 10 for litter and micro litter in the environment and the impact of the litter on marine species through ingestion and entanglement.

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- THE DIFFERENT TYPES OF PLASTICS STUDIED/ENCOUNTERED INCLUDE:
 POLYETHYLENE (PE) INCLUDING LOW-DENSITY POLYETHYLENE (LDPE) AND HIGH-DENSITY POLYETHYLENE (HDPE), POLYPROPYLENE (PP), POLYSTYRENE (PS), POLYETHYLENE TEREPHTHALATE (PET), POLYCAPROLACTONE (PCL), POLYVINYLCHLORIDE (PVC), POLYAMIDE (PA), POLYESTER, POLYURETHANE (PU), ETHYLENEVINYL ACETATE (EVA), POLYACRYLIC, POLYISOPRENE, EPOXYDIC RESIN, POLYBUTHYLENE TEREPHTHALATE, POLYTERPENIC RUBBER, POLYVINYL ACETATE (PVA), SILICONE AND COPOLYMERS.
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