Scientific contribution

Insights into Microplastics: from Physical and Chemical Characterisation to its Potential as a Vector.

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Abstract:

Particles with the largest dimension of less than 5 mm, also termed as microplastics (MPs), gained a lot of scientific and media attention in the last decade. MPs in the environment are of importance because of their potential for further fragmentation, accumulation, and impact on biota in the terrestrial and water environments. MPs research is challenging due to their diversity in size, shape, and chemical structure. For research purposes, MP particles can be purchased, but in terms of chemical structure, they properties might not correspond to the ones of MPs, found in the environment. Compared to purchased MPs, plastic products in the environment can contain different additives, despite being the same polymer type as purchased MPs. Therefore, for environmental studies preparation of MP particles from plastic products is preferable. In this contribution two methods for laboratory preparation of MP particles, different sizes and polymer types, are presented. Method using ultrasound probe was found to be suitable for obtaining polyester fibres from thin sewing thread, while cryogenic milling was found to be preferable method for MPs preparation from larger and thicker plastic particles. In this way, MPs of other types of plastic (polyethylene terephthalate, polystyrene, polyvinyl chloride, polypropylene) were prepared from beforehand manually cut plastic particles, originating from everyday plastic products.

Keywords: Miroplastics; Cryomilling; Separation; Challenges of preparation; Surface topology

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1. Introduction

Industrial development and increased usage of single-use plastic products in everyday life consequently result in increased plastics production. In 2018, the global plastics production was reported to be 359 million tonnes and it increased to 381 million tonnes in 2019, with China contributing the highest share (PlasticsEurope, 2020). In Europe, however, a slight decrease is observed in yearly plastics production from 2017. Data show that plastics production in Europe decreased from 61.8 million tonnes in 2018 to 57.9 million tonnes in 2019 (PlasticsEurope, 2020). Lately, due to the pandemic, a sharp rise in use of disposable plastics, for convenience or hygiene (e.g., pandemic) is observed all over the world. Inevitable issue of extensive plastics production and its use is the occurrence and formation of microplastics particles (MPs) in the environment.

MPs have gained a lot of attention in the past years, especially due to their ubiquitous presence around the world and in various environments, such as oceans (e.g. Great Pacific Garbage Patch), freshwater, wastewater, soil and air (Van A, et al. 2012; Hoellein TJ, et al. 2017; Magni S, et al. 2019; Huang Y, et al. 2020; Chen G, et al. 2020). Their presence has been linked to various risks and impacts on biota in the environment and indirectly to human health due to its fragmentation and leaching of additives, added during plastic production process, and other contaminants (Horodytska O, et al. 2020). In the future, even more attention could be given to the MPs, since their further fragmentation can result in nanoplastics, which are potentially one of the most hazardous type of debris in marine environment due to their possible uptake by marine animals into their bodies (Koelmans AA, et al. 2019).

MPs research is challenging due to their four characteristics: origin, shape, size, and polymer type. Origin of MPs is interconnected with the shape of MPs. Namely, based on their origin, MPs can be divided into primary MPs and secondary MPs. Primary MPs are intentionally produced in such small size for its use in e.g. personal hygiene products, pharmaceutical industry, and are mainly found in the shape of spheres and cylindrical pellets (Hartmann NB, et al. 2019; Cole M, et al. 2011; Hernandez LM, et al. 2017). Secondary MPs, however, are formed by fragmentation of larger plastic debris under the influence of mechanical, physicochemical, and biological factors, such as ultraviolet light, temperature, pH, abrasion forces and microbial degradation. Fragmented particles can be found in the shape of films, fibres and fragments (Hartmann NB, et al. 2019; Cole M, et al. 2011; Ngo PL, et al. 2019).

MPs are, as derived from marine MPs research, commonly known as particles or fragments with the size of less than 5 mm in diameter (GESAMP, 2016). Despite its widespread use in research studies, the uniform definition of MPs among scientific community is not yet established. Scientist have proposed different classifications, that differ on upper and/or lower size limits of particles to be termed as MPs. Some propose the lower size limit for MPs should be 1 mm (Ivleva NP, et al. 2017), while other suggest 100 nm (Rios Mendoza LM, et al. 2018). The latest scientific discussion from European researchers on the topic of terminology used in plastic debris research (Hartmann NB, et al. 2019) recommends classification of plastic debris to be based on the chemical composition, solid state, solubility, size, shape, colour and origin. According to the size, Hartmann et al. (2019) propose classification of particles into four groups: nanoplastics (1 to <1000 nm), microplastic (1 to < 1000 μm), mesoplastics (1 to < 10 mm) and macroplastics (1 cm and larger).

MPs size is challenging from the analytical point of view, especially for quantification. In fact, for MPs identification, the most developed and verified methods so far, are the non-destructive infrared spectroscopy methods and the destructive ones, named thermochemical methods. In the group of non-destructive infrared spectroscopy methods belong the attenuated total reflectance Fourier-transform infrared spectroscopy (ATR-FTIR), micro-FTIR (μ-FTIR) and Raman spectroscopy. Whereas in the group of destructive methods, the two main important are pyrolysis gas chromatography mass spectrometry (Py-GC-MS) and thermogravimetry with additional thermal desorption gas chromatography mass spectrometry analyses (TED-GC-MS) (Pico Y, et al. 2019; Krauskopf L-M, et al. 2020; Dümichen E, et al. 2017). With these methods polymer type of MPs can be determined.

Plastic products are made out of numerous polymer types as well as copolymers, to which many additives can be added (Hartmann NB, et al. 2019; Kusch P and Knupp G, 2004). The most abundant

polymer types, determined in plastic debris in the environment, are polyethylene (PE), polystyrene (PS), polyvinyl chloride (PVC), polyethylene terephthalate (PET), polyester (PES) and polypropylene (PP), and polyamides (PA) (Gatidou G, et al. 2018; Sang W, et al. 2021; Díaz-Jaramillo M, et al. 2021).

The overall issue regarding MPs is not only their presence as environmental micropollutants, rather their vector potential. Since their physical presence as particles and their mostly hydrophobic surface nature, they can act as an adsorption site for organic contaminants, such as pharmaceutical residues, pesticides, polychlorinated biphenyls and polycyclic aromatic hydrocarbons, as well as for inorganic contaminants, like heavy metals (Zhang H, et al. 2018; Dong Y, et al. 2020; Wang F, et al. 2020; Šunta U, et al. 2020; Bhattacharya A, et al. 2019).

To perform environmentally applicable adsorption studies for contaminants in laboratory settings, MPs of defined size and polymer type have to be purchased or prepared. Since purchasing of MP particles is not possible at all needed sizes and types, here it is a representation of two manners in which MPs of desired sizes can be prepared in laboratory, using ultrasound treatment and cryogenic milling.

2. Methods

MP particles (1 mm-100 μm) for laboratory research purposes were prepared with cryogenic milling (fragments) and ultrasound treatment (fibres). Cryogenic milling with liquid nitrogen (LN, Messer, Germany) and ball mill (MillMix 20, Tehtnica, Slovenia) was used to prepare samples of PET, PS, PVC, PE and PP. 3 g of MPs of individual polymer type (size range 1 -5 mm) was placed in the grinding jar containing a stainless-steel milling ball. Before milling, grinding jar was submerged in LN for 6 min and afterwards milled in 4 series for 2 min at 35 Hz (PET) and at 25 Hz (PS, PVC, PP, PE) with intermediate cooling in LN for 1 min. Obtained particles were additionally sieved through 1 mm (Retsch, Germany) and 100 μm (Fipis, Slovenia) stainless steel sieve to obtain particles of desired size (1 mm - 100 μm). Scanning electron microscopy (SEM) was used to examine the morphology of particles. Original particles (5 mm - 1 mm) and cryogenically milled particles (1 mm - 100 μm) were coated with Au/Pd (PECS Gatan 682), placed on the double-sided adhesive carbon tape on the aluminum stubs, and analysed by JEOL JSM-6500F Field-Emission Scanning Electron Microscope (JEOL LTd, Japan) operated at 15.0 kV (Gniadek M and Dąbrowska A,2019; Božič D, et al., 2021).

Ultrasound separation. The ultrasound probe (Labsonic M, Sartorius, Germany) was used to disperse the sewing thread into individual fibres. 50 mg of PES sewing thread (Moon, UK), that was beforehand manually cut into 1 mm pieces, was placed into a beaker, containing 25 mL of dichloromethane (Sigma Aldrich, USA). Dichloromethane was used because no attachment to the probe or to the glass walls of the beaker was observed, contrary to other media. The probe (Ø 3 mm) was then placed into the dichloromethane mixture and operated in continuous mode for 5 min (0.8 cycle, 60 % amplitude). Following the separation, mixture was filtered through 0.45 μm cellulose acetate (CA) filter (Sartorius, Germany). Obtained fibres were transferred from the CA filter into a glass Petri dish, half covered with glass lid, and dried in the oven at 65°C for 30 min. The dispersion of thread to fibres was examined under optical microscope Olympus CX21 (Japan).

3. Results and Discussion

Commercially available MPs and environmental plastics might seem similar in shape or/and size, however, they differ in the composition regarding additives and surfactant content, as well as morphology and density. Therefore, purchased MPs are not necessarily comparable to the MPs found in the environment (Eitzen L, et al. 2019). Laboratory pre-treatment enabling the preparation of MPs of defined size rely on cryomilling or cryogenic milling. This is a milling process performed near LN temperature (- 180 °C). It is achieved using cryomill or the combination of the immersion of grinding jar, containing the sample, in LN and high-energy milling (e.g., ball mill). Subsequent sieving can be used to obtain particles of the desired size range (Eitzen L, et al. 2019; Lagarde F, et al. 2016; Bai C, et al. 2000). Cryomilling has been used in numerous studies regarding MPs, either as a preparation technique for obtaining particles of specific size ranges (below 1 mm) (Eitzen L, et al.

2019; Bannick CG, et al. 2019; Capolupo M, et al. 2020) or as a technique for samples homogenisation before MPs determination (Dümichen E, et al. 2017).

In this study the cryogenic ball grinding process was used to obtain a fine fraction with PP, PS and PET particles in the size between 100 μm and 1 mm. However, there were few difficulties regarding the PVC and PE cryogenically milled particles. Since PVC fragments originated from tablet pharmaceutical packaging partially covered with aluminium, it was difficult to distinguish PVC particles from the aluminium residue. In case of PE grinding, the glass transition temperature (Tg) was crucial. PE has the lowest glass transition temperature (Tg) from all polymers, ranging from -100°C to -70 °C (Eitzen L, et al. 2019). PE particles were not milled into finer particles (1 mm - 100 μm), rather a colour change was noticed, due to their heat-oxidation (Rugg FM, et al. 1954). For cryogenic milling of PE particles, therefore, the cryomilling procedure needs to be improved, implementing longer precooling time and/or shorter grinding cycle.

An insight in the topological diversities in MPs' surfaces after cryogenic milling, was achieved with SEM microscopy (Fig. 1). PET cryogenically grinded particles (1 mm – 100 μm) had more ragged surface compared to the manually cut particles from PET bottle (5 mm – 1 mm). This indicates that MPs particles of smaller sizes have more potential for interactions with organic pollutants, as well as for microorganisms to adhere and form biofilm. The effect of MPs size on arsenic adsorption onto PS MPs was confirmed in study by Dong et al. (2020), where the amount of adsorbed arsenic was decreasing with increasing MPs size.

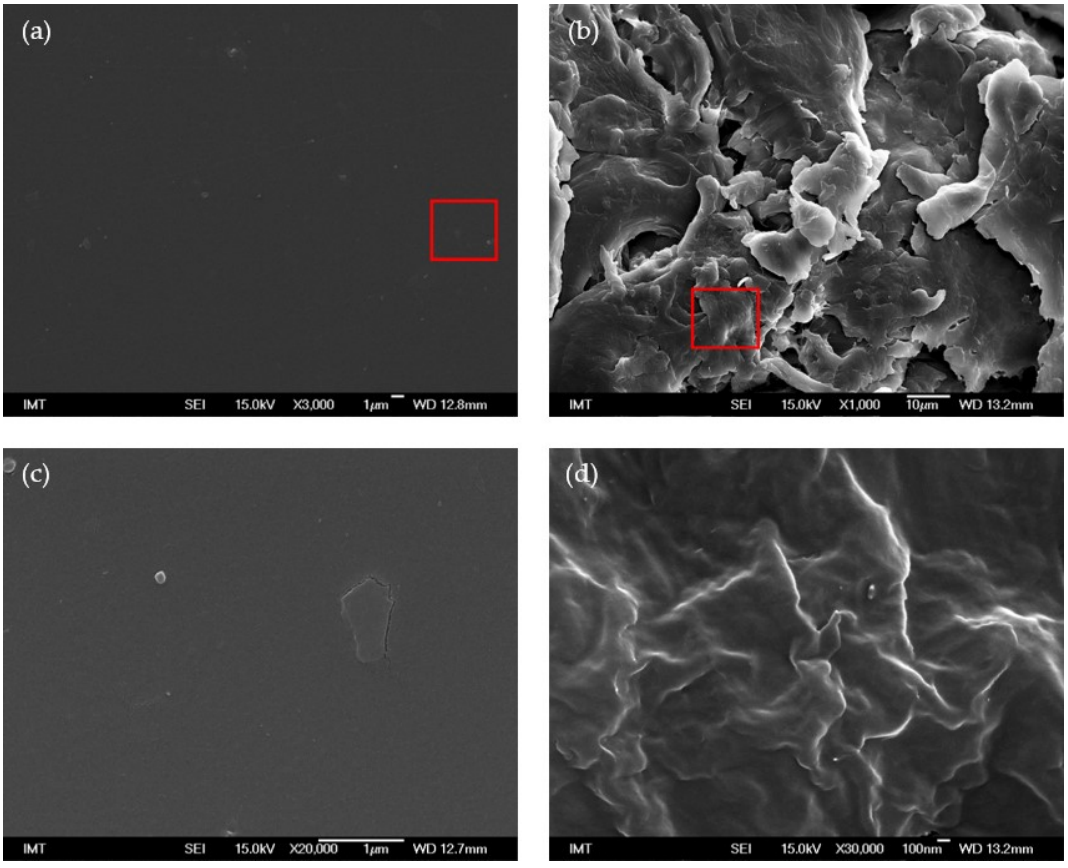

Figure 1: Pre-cut fragment (5 mm - 1 mm) and cryomilled particle (1 mm - 100 μm) of PET under SEM microscope at 3 000 x magnification (a), 1 000x magnification (b), 20 000 x magnification (c) and 30 000 x magnification (d). Red rectangles in (a) and (b) indicate the areas containing the structures shown in (c) and (d).

The cryogenic milling has beside roughness also some other physical consequences, which influence MPs and the course of the experimental studies and its results. Density and surface charge are two of those. Eitzen et al. (2019) observed that some cryomilled PS particles did not completely disperse in water and were accumulating along the glass walls. Obtained cryomilled particles can become

charged and consequently float in the selected medium when according to their density they should sink or be submerged (von der Esch E, et al. 2020).

Therefore, von der Esch et al. (2020) developed and specified a method for secondary MPs generation in laboratory setting using ultrasonic bath. In this study fragmentation of PS, PET, PE, PP, PVC, PA and polylactic acid polymer particles was tested. Just before that study, we have tested the preparation of PES fibres from a sewing thread using ultrasound probe. Sewing thread was successfully dispersed to individual PES fibres (Fig. 2), however, due to the chosen medium (dichloromethane), leaching of the colour from coloured sewing thread was observed. This indicates that the solvent might be too strong for PES fibres and damages of fibre's surface and structure can occur. Selection of medium for dispersion has to be taken into account when preparing the material for the experiment. In the method proposed by von der Esch et al. (2020), 0.25 M potassium hydroxide was used, which supposedly does not dissolve MPs.

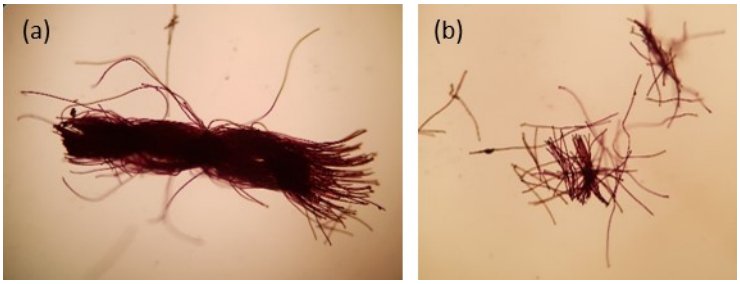

Figure 2: Polyester sewing thread before (a) and fibres after (b) dispersion with ultrasound probe treatment under 40x magnification.

4. Conclusions

MPs have gained a lot of scientific attention in the last decade in various scientific disciplines. For studying the effects of MPs in the environment, however, the purchased particles might not be representative material to use in research studies. They can differ in chemical structure and additive content, compared to the MP particles found in the environment. In this contribution two manners for successful MPs preparation in laboratory setting from plastic products we use in everyday life are presented, one using cryogenic milling and the other using ultrasound probe.

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Conflicts of Interest: The authors declare no conflict of interest.

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