Optimized microplastic analysis based on size fractionation, density separation and μ-FTIR

Olga Konechnaya, Sabine Lüchtrath, Larissa Dsikowitzky and Jan Schwarzbauer

ABSTRACT

Microplastic particles have been recognized as global hazardous pollutants in the last few decades pointing to the importance of analyzing and monitoring microplastics, especially in soils and sediments. This study focused on a multi-step approach for microplastic analysis combining grain size fractionation, density separation and identification by μ-FTIR-spectroscopy. Eight widely used polymers (polyethylene (PE), polypropylene (PP), polyvinylchloride (PVC), polystyrol (PS), polyethylenterephthalate (PET), polymethylmethacrylate (PMMA), polyurethane (PU) and polyamide (PA)) were fractionated into four groups of grain sizes (0.1–5 mm). Thereafter, sea sand was spiked with these particles to test a $ZnCl_2$-based density separation for the polymer types and the various grain sizes. The obtained recovery rates were close to 100% showing that $ZnCl_2$-based density separation is suitable to separate the polymer particles from a sandy matrix. This approach was extended on three further environmental matrices and recovery rates for two of them (sandy-silty and fine-grained sediment) also provided reliable values (94–106%). Lastly, the developed multi-step approach was verified by analyzing an environmental sample (sediment from river Tiranë, Albania) characterized by smaller grain size and moderate organic matter content. Identification of two polymer types in different grain size classes verified the suitability of the developed approach for microplastic analyses on particulate matter such as soils and sediments.

Key words | density separation, microplastics, multi-step approach, recovery rates, μ-FTIR

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INTRODUCTION

First reports on plastic deposited in environmental settings were published in the early 1970s with little attention on the hazardous consequences (Carpenter & Smith 1972; Colton *et al.* 1974). Almost 30 years later, more attention was drawn on this topic again at the beginning of the 21st century due to increasing plastic amounts in the ocean ecosystems. Nowadays, plastic materials are used in many industry branches and daily life; it is difficult to renounce plastic. The consumption and production of plastic material is increasing annually and reached up to 335 million tons in 2016 (AISBL 2018).

The environmental effects are not related only to plastic itself, but to their very small pieces called microplastics. Per definition, microplastics are plastic particles smaller than 5 mm diameter (Thompson *et al.* 2004; Arthur *et al.* 2009). Microplastics can be subdivided according to Hidalgo-Ruz *et al.* (2012) into small microplastics (1 μm–0.5 mm) and large microplastics (0.5–5 mm). Moreover, microplastics can be categorized into primary and secondary microplastics (Cole *et al.* 2011). Primary microplastics are originally manufactured plastics with a size less than 5 mm including textiles, medicines or personal care products (Sharma & Chatterjee 2017). Secondary microplastics are formed by fragmentation of litter through ultraviolet (UV) – or photodegradation – but also by other physical, chemical and biological processes (Thompson *et al.* 2009; Galgani *et al.* 2013). Degradation processes of plastic particles are very slow, thus the particles persist in the environment for a very long time in the range of up to hundreds of years (Barnes *et al.* 2009; Li *et al.* 2018).

Microplastics are considered as hazardous pollutants (Cole *et al.* 2011) due to their effects in the environment

and on human health. Especially aquatic organisms are highly affected by microplastics, since microplastics can accumulate in cells and tissues (Thompson et al. 2004, 2009). The ingestion of microplastics in humans can cause changes in chromosomes, which may lead to infertility or cancer (Sharma & Chatterjee 2017). Moreover, microplastics can act as a vehicle for transporting toxic chemicals through trophic levels (Browne et al. 2008; Bakir et al. 2012; Wessel et al. 2016). These particles absorb not only persistent organic pollutants (POPs) such as polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs), but also heavy metals such as copper and nickel (Brennecke et al. 2016). The reported negative effects of microplastics in depositional environments clarify that it is important to investigate and monitor microplastic pollutions and to identify and quantify microparticles in aquatic and terrestrial compartments, living organisms or even food products (Thompson et al. 2009; Wegner et al. 2012; Imhof et al. 2013; Lusher et al. 2013, 2014; Ivar do Sul & Costa 2014).

Common analytical procedures for the determination of microplastics in environmental samples comprise extraction, separation, identification and quantification (Shim et al. 2017). One preliminary method to characterize large microplastics (1–5 mm) is based on visual sorting by the naked eye, which is not applicable for smaller microplastic particles. It is difficult to identify microplastics of different size, shape and polymer types from complex environmental samples using a single analytical method. Therefore, a combination of more than one analytical tool will be valid to use.

Soils are composed of mineral and organic components together with water, air and organisms (Liu et al. 2019). The presence of microplastic particles in soils makes it more difficult to extract them from the complex mixture. Therefore, it is a challenge to separate microplastics (MPs) from soil samples. In order to separate MP from environmental solid samples, two procedures are essential. The first one is to remove the organic part, and the second one is to separate MP from the mineral matrix. Hence, pretreatments of the environmental samples are necessary. In general, H_2O_2 or Fenton's reagent are commonly used to remove the organic matter from environmental matrices (Sujathan et al. 2017). The effectiveness of these procedures has been documented in recent studies (Avio et al. 2015; Hurley et al. 2018) and was therefore applied in our study.

There are different kinds of separation techniques known for the extraction of microplastics from sediments or environmental mineral matrices depending on size, shape or density of both the matrix and the microplastics (Crawford & Quinn 2017). Elutriation column (Claessens et al. 2013), sediment-microplastic isolation (SMI) (Coppock et al. 2017), pressurized fluid extraction (Fuller & Gautam 2016) and density separation using different kinds of salt solutions have been studied. However, all possibilities have drawbacks and cannot completely or easily separate all types of microplastics from sediments.

Sodium chloride (NaCl), calcium chloride ($CaCl_2$), sodium iodide (NaI), zinc chloride ($ZnCl_2$) or sodium polytungstate ($Na_6(H_2W_{12}O_{40})$) were suggested in previous studies (Hidalgo-Ruz et al. 2012). However, salt solutions are either expensive, toxic to the environment (NaI, (Crichton et al. 2017)), may react with the organic matter ($CaCl_2$, (Scheurer & Bigalke 2018)), or their density is not sufficient for the whole range of different polymers (NaCl, (Liu et al. 2018)). Hence, it is more difficult to separate heavier polymers like PET or PVC.

For identification purposes of microplastic particles, Fourier transform infrared spectroscopy (FTIR), Raman spectroscopy or scanning electron microscopy (SEM-EDS) are used (Vianello et al. 2013; Käppler et al. 2016). μ-FTIR spectroscopy provides a simple and efficient method to detect and identify microplastics due to its non-invasive and non-destructive character, simple sample preparation procedure and the possibility to correlate with IR radiation for thick samples (Ojeda et al. 2009). FTIR enables detection and qualitative identification of microplastics as different chemical functional groups absorb infrared radiation at specific wave numbers and deliver individual fingerprint spectra. The obtained spectra are commonly confirmed by a polymer spectrum library. Reflectance, transmission and attenuated total reflectance (ATR) modes are available for the identification of small microplastic fibers down to a size of 10–20 μm. The lower limit is given by the diameter of the IR beam (Shim et al. 2017).

However, detection of complex microplastic contamination especially in environmental samples, with lower concentration levels within complex matrices, requires the application of multi-step analytical methods. Therefore, the main object of the present research is to test a newly designed multistep approach, which combines the methods (i) size fractionalization, (ii) density separation and filtration and (iii) micro-FTIR spectroscopy to unambiguously identify microplastic particles in sediment and soil matrices. We focused in particular on the detailed analysis of microplastics at different grain size classes. In addition, it is to test if μ-FTIR technique is applicable for an unambiguous and simultaneous detection of the eight most common polymers and to test the suitability of this approach for environmental samples.

MATERIALS AND METHODS

Preparation of material

The eight most commonly used polymers polyethylene (PE), polypropylene (PP), polyvinylchloride (PVC), polystyrol (PS), polyethylenterephthalate (PET), polymethylmethacrylate (PMMA), polyurethane (PU) and polyamide (PA) (Nylon 6) in variable colors were chosen (purchased in a hardware store) for our study. The selection was based on their high domestic and industrial usage, their economic relevance and production rates. These polymers were ground and homogenized by hand using a metal file (Emil Lux GmbH) to obtain plastic pieces in various sizes and shapes (Table 1). The densities of the selected polymers ranged from $0.25 \ \text{g·cm}^{-3}$ (PU) to $1.39 \ \text{g·cm}^{-3}$ (PET).

Size fractionation of polymers

Reference material of polymer product samples was separated into four groups of grain size according to DIN4188. Grain size fractions of $>5 \ \text{mm}$, $1–5 \ \text{mm}$, $400–1,000 \ \mu\text{m}$, $200–400 \ \mu\text{m}$ and $100–200 \ \mu\text{m}$ were obtained using sieve stack AS 200 from Retsch under water flow ($5–10 \ \text{L}$ water) for 15 min with an amplitude of 40. Thereafter, all sieved fractions were dried at $40 \ ^\circ\text{C}$ in an oven for two days (48 h).

Density separation

According to Semensatto & Dias-Brito (2007), a $ZnCl_2$-solution with a density of $1.7 \ \text{g cm}^{-3}$ was prepared by dissolving $72.07 \ \text{g} \ ZnCl_2$ in $46.84 \ \text{mL}$ distilled water. As the reaction is exothermic, it needed to be cooled with an ice bath during addition of $ZnCl_2$. After dissolving the total $ZnCl_2$ and filtrating of the solution, a density of $1.63 \ \text{g cm}^{-3}$ with a pH value of 3 were achieved for the initial solution.

As presented in Figure 1 in our study, six steps for density separation were used. Approximately 0.5 g of each grain size fraction of each polymer particles were mixed with 10 g of pre-cleaned calcinated sea sand representing a sediment matrix (Th. Geyer GmbH & Co. KG) and added into 250 mL of the prepared $ZnCl_2$ solution (density $1.6–1.7 \ \text{g cm}^{-3}$, density increase with recycling circles). After stirring three times for around 2 min, sedimentation was allowed for a time period of 10 mins. Noteworthy, with decreasing size of microplastics, the stirring time while density separation with $ZnCl_2$ was increased from 30 s to 10 min. Thereafter, the lighter plastic particles were separated via overflow from the solution. After filtration and washing with deionized water the separated particles were collected on a filter before they were dried in the oven at $70 \ ^\circ\text{C}$ for 1 h (Figure 1).

For quality assurance, this procedure was performed in triplicate for each polymer type and grain size fraction for determining average values and standard deviation.

Recovery experiments

For determining recovery rates of the density procedure, 10 g of sea sand were spiked with microplastic particles at the four different grain sizes, respectively, and subjected to the whole procedure described above. The absolute yields were obtained by gravimetric determination. The recovery rates were determined in triplicate and average values with standard deviations were calculated.

Testing on real environmental samples

Three natural samples consisting of the beach sand from Scotland (TOC = 0.24%), of fine-grained river sediments from Teltow Canal (Berlin, Germany) (TOC = 9.11%), and of a soil sample from flood plains of the Wurm river (North Rhine Westphalia, Germany) (TOC = 7.67%) were

Table 1 | Color, origin and density of the used plastic pieces

| Synthetic substance	Color	Precursor product	Density (g·cm^{-3})
Polyurethane (PU)	beige	PUR- foam	0.25
Polyvinylchloride (PVC)	orange	PVC pipe	1.16–1.3
Polystyrol (PS)	red/yellow	document box	1.04–1.09
Polyamide (PA) (Nylon 6)	pink	kitchen spoon	1.13–1.15
Polyethylenterephthalate (PET)	blue	bottle neck	1.34–1.39
Polypropylene (PP)	black	bottle caps	0.9–0.92
Polyethylene (PE)	green	bottle caps	0.91–0.95
Polymethylmethacrylate (PMMA)	white	rear reflector	1.19
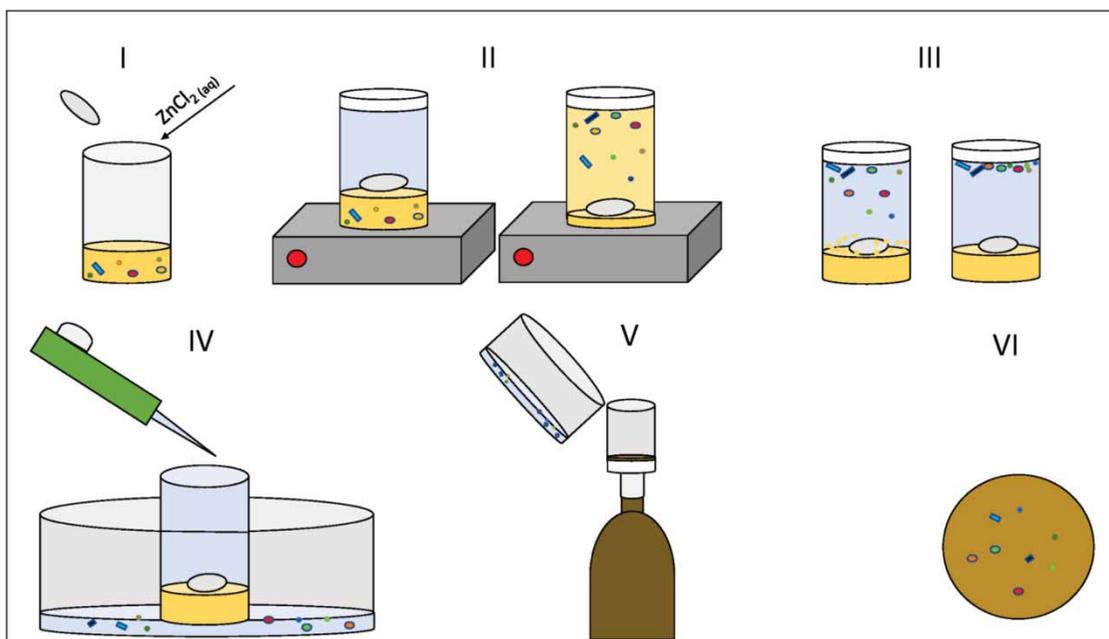
Figure 1 │ Scheme of density separation steps: I adding agitator and zinc chloride solution with density >1.6 g cm^{-3}; II stirring suspension for three times; III allow sediment to settle down and microplastic particles to float; IV making an overflow by adding slowly ZnCl$_2$-solution; V filtration and washing filter with deionized water afterwards; VI collected and ZnCl$_2$ free microplastic particles.

spiked with microplastic particles of five common polymers. For testing purposes, only PP, PE, PET, PVC and PS were chosen as they are the most commonly and widely used polymers. 0.025 g of each polymer type in each of four grain sizes (1–5 mm, 400–1,000 μm, 200–400 μm and 100–200 μm) was mixed together with the beach sample material. Hence, in total 0.1 g of each from five plastic particle types and 50 g of the sediment sand were mixed together. Grain size fractionation was executed on the whole sample as described in the section 'Size fractionation of polymers' (sieve mesh size of 5 mm, 1 mm, 400 μm, 200 μm and 100 μm). All subsamples were dried at 40 °C for three to four days. The density separation by ZnCl$_2$ with density of 1.63 g/cm^3 was accomplished for all four samples of polymer particles with different grain sizes.

Also, an organic-rich soil sample was taken from a sediment core of a floodplain of the Wurm river near Aachen, at a depth of 170–180 cm dated to avoid any cross-contaminations. However, the rich organic fraction interfered with the density separation by floating partly together with the plastic particles on the top of the separation solution. Therefore, after an extended period of two days, microplastics were separated from the fine-grain matrix and recovery factors were calculated by gravimetrical determination.

As a proof-of-concept, the Albanian riverine sediment (TOC = 0.65%) sample has been subjected to a modified procedure. 100 g of the soil sample was sieved (without spiking) into five grain sizes adjusted to smaller sieve meshes (>1 mm, 500–1,000 μm, 100–500 μm, 50–100 μm and 20–50 μm) and proceeded as above. After density separation by ZnCl$_2$ solution, the subsamples were treated at 60 °C with 35% H$_2$O$_2$ for the reaction time of 12 h. The subsamples were filtered and determined at μ-FTIR using attenuated total reflection (ATR)-imaging mode.

Preparation and μ-FTIR measurements

Spectroscopic analyses were carried out with an FTIR spectroscope Spotlight 400 Series from Perkin Elmer. Each microplastic particle containing subsample was placed on prepared KBr pellet to ensure smooth surfaces and pressed for 30 s with 10–12 tons. This KBr pellet together with plastic particles on its surface was brought in close contact with the germanium crystal of the ATR objective. IR spectra were measured using a Mercury Cadmium Telluride (MCT) detector in a wavenumber range of 4,000–750 cm^{-1} with the spectral resolution of 16 cm^{-1}. For every spectrum, 8–16 scans were co-added. Background as well as crystal measurements were performed with the same settings. Identification of polymers was done with our own database recorded with the Spectrum program.

To build a specific database for rapid identification, polymer samples from different sources were measured via ATR-FTIR spectroscopy on a Spotlight 400 Series from Perkin Elmer with a germanium ATR-unit. The spectra were recorded in reflectance mode within a range from 4,000 to 750 cm^{-1} and resolution of 16 and 8–16 scans were co-added. Each measurement for all polymers was performed five times.

RESULTS AND DISCUSSION

Identification of different plastics in terms of based polymer type in complex environmental samples at low levels of amount requires optimized analytical approaches comprising different complementary sample treatment and measuring steps. In this study the combination of grain size distribution, density separation and μ-FTIR detection is applied and optimized for soil and sediment samples from the riverine aquatic environment. The optimization included an assessment of suitable reproducibility and sensitivity, the test on natural matrices by spiking experiments, the strategy of μ-FTIR based detection and finally the verification by applying the method on a real environmental sample.

Reproducibility and effectiveness

In order to optimize microplastic isolation and to assess this method in terms of reproducibility, a combined two-step approach was developed and tested. Grain size fractionation was combined with following density separation by ZnCl$_2$ solutions. Recovery experiments of spiked sand samples in different grain size were processed separately for the eight polymers (PE, PP, PVC, PS, PET, PA, PMMA and PU) (Table 2) which were used to determine recovery rates for density separation. Since all procedures were performed in triplicate, average values and corresponding standard deviations were calculated. The applied procedure was modified from Imhof *et al.* (2012). They used ZnCl$_2$ solution for separating microplastics from sediment by Munich Plastic Sediment Separator (MPSS) for two grain size fractions. However, our study extended the approach to four grain size fractions (even smaller particles) and by simplifying the final step by using a simple overflow approach.

Grain size separation reveals fractions with differing physical properties. Not only density itself but also shape and flexibility are parameters affecting the separation. However, since grain size classification might have an effect on density

Table 2 │ Recovery rates together with standard deviation for microplastic particles in sizes of 100–200, 200–400, 400–1,000 and 1,000–5,000 μm

1,000–5,000 μm			400–1,000 μm		
	Recovery rate	Standard deviation		Recovery rate	Standard deviation
PE	100	1.3	PE	100	0.3
PP	100	0.5	PP	99	1.2
PVC	98	1.5	PVC	99	1.8
PET	102	2.9	PET	99	0.8
PA	97	0.6	PA	97	1.6
PS	101	0.4	PS	99	1.4
PU	104	6.4	PU	98	1.7
PMMA	93	2.5	PMMA	98	0.6
200–400 μm			100–200 μm		
PE	97	0.8	PE	95	1.6
PP	98	1.6	PP	98	0.8
PVC	97	1.1	PVC	94	1.9
PET	100	1.1	PET	98	0.1
PA	96	2.6	PA	96	3.4
PS	98	0.4	PS	96	3.3
PU	96	3.5	PU	95	3.1
PMMA	99	1.1	PMMA	99	3.0

separation, it is mandatory to evaluate the analytical reproducibility and effectiveness for each grain size fraction separately.

All obtained recovery rates are given in Table 2. The values cover a narrow range between 94% and 103%.

The distribution of recovery rates with respect to grain size of plastic particles is illustrated in Figure 2 and points to a general decreasing trend of recovery rates for most polymers with decreasing grain sizes. For PE, PP, PET, PA, PS and PU their recovery rates decrease slightly from 98–103% to 94–97% (with the standard deviation between 0.3% and 3.3% with decreasing particle sizes). The highest recovery rate (103 ± 6%) was achieved by sieving PU >1 mm. PU particles are very light with the smallest density (Table 1). This might lead to a redistribution of PU particles with a moderate standard deviation up to 6% and therefore the largest recovery rate of 103% for the grain size of 1–5 mm.

Interestingly, PMMA shows a reversed trend as compared to the other polymers. This observation might be related to parameters like shape and flexibility of PMMA particles that probably differ as compared to the other polymer particles and affect the separation and, consequently, the recovery rates. However, the standard deviation for
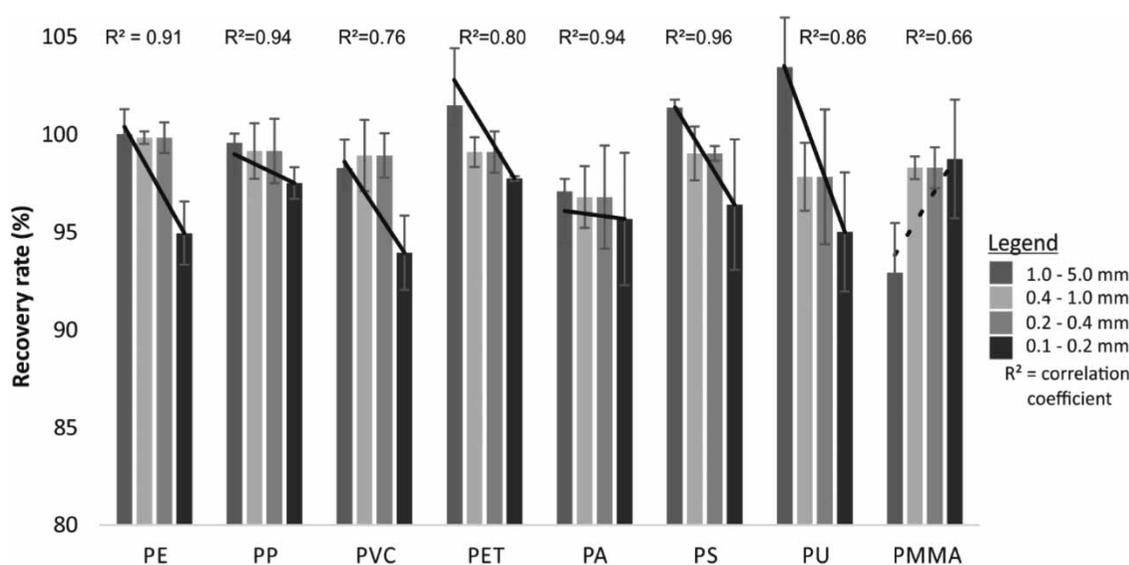
Figure 2 │ Distribution of recovery rates (%) depending on particle size for eight polymers. General trends are indicated pointing to the inverse behavior of PMMA particles.

recovery rates for PMMA is between 0.6% for the middle fraction and 3.0% for the smaller grain size and still does not show a big difference.

Our results are comparable to already published data. Previous studies (Nuelle *et al.* 2014; Coppock *et al.* 2017; Quinn *et al.* 2017) reported similar recovery factors for the most common polymers between 90% and 99% by using NaCl, NaI, $NaBr_2$ or $ZnCl_2$ as the floating media.

Nuelle *et al.* (2014) investigated seven polymers with grain sizes >500 μm and obtained recovery factors between 91% and 99% by using NaCl and NaI solutions. Coppock *et al.* (2017) used a different technique (sediment-microplastic isolation (SMI)) with various density solutions (NaCl, NaI and $ZnCl_2$) and obtained recovery factors from 92% to 98% for PE, PVC and nylon. There are some differences in the applied approaches as compared to the experiments performed in this study. Many of the aforementioned studies were performed on polymers slightly different from our study. Furthermore, particles smaller than 500 μm, even <100–200 μm, were included in our study but not considered in the mentioned studies (Nuelle *et al.* 2014; Coppock *et al.* 2017; Quinn *et al.* 2017). Since those smaller particles exhibit partly elevated effects on human and animal health, also these small size fractions need to be included in analyses.

Quinn *et al.* (2017) validated a variety of density separations by using NaCl, NaBr, NaI and $ZnBr_2$ solutions. Noteworthy, $ZnBr_2$ was reported as the most suitable brine solution for density separation, but with a disadvantage of being environmentally harmful and expensive. However, $ZnCl_2$ solution was not considered in their investigations,

whereas our study demonstrated that the environmentally less harmful $ZnCl_2$ is easy in solution preparation, recyclable and suitable for all eight polymers with recovery rates higher than 94%. This was supported by Imhof *et al.*'s (2012) investigation of $ZnCl_2$ solution for separating microplastics from sediment using MPSS with recovery rates of up to 100% for large and small microplastic particles, respectively.

Spiking experiments with natural samples

Since a real sample matrix influences the separation of microplastics, the recovery experiments as described above have been applied to three real samples. These samples exhibit differences in grain size distribution (sandy vs fine-grained material) and organic matter content.

The recovery rates of the samples with sandy matrix (Scotland beach sand) (Table 3(a)) vary between 70% and 133% with an average near 100%. The overall recovery rate was 106%.

The second spiking experiments used a fine-grained sediment sample from the Teltow Canal, Berlin. The obtained recovery rates are similar to those from sand from Scotland and vary between 70% and 109% (Table 3(b)). The average recovery rate for all grain sizes polymers is 96%.

The third sample used for spiking experiments derived from a sediment core taken at a riparian wetland of the Wurm river, North Rhine Westphalia. This sample exhibited a higher organic content which causes difficulties in applying the described two-step approach leading to recovery rates in this case up to 250% (Table 3(c)). This was obviously

Table 3 | Recovery rates for spiked natural samples

Particle size [µm]	Plastic load [g]	Yield of plastic [g]	Recovery rate [%]
>1 mm	0.1347	0.0943	70.0
400–1,000 µm	0.1272	0.1685	132.5
200–400 µm	0.1382	0.1472	106.5
100–200 µm	0.1222	0.1423	116.4
Sum	0.5223	0.5523	105.7
(a) Scotland beach sand			
>1 mm	0.1497	0.1022	68.3
400–1,000 µm	0.1200	0.1309	109.0
200–400 µm	0.1214	0.1256	103.5
100–200 µm	0.1219	0.1256	103.0
Sum	0.5130	0.4843	95.9
(b) fine-grained sediment from Teltow canal			
>1 mm	0.1277	0.3428	268.4
400–1,000 µm	0.1247	0.5656	453.7
200–400 µm	0.2620	0.3693	138.5
Sum	0.51440	1.2718	247.2
(c) organic rich sample from Wurm river			

due to an insufficient separation of microplastic particles from organic matter, which exhibit similar densities, respectively. Additionally, more than half of the organic part was lost during sieving due very fine sample grain size distribution.

For two of three spiked samples the recovery rates are between 96% and 106%. This result shows that even here $ZnCl_2$ solution is suitable for five common polymers and sandy samples as well as for fine grained samples. Only for the organic-rich sample from the Wurm river was it difficult to separate microplastics from the spiked matrix. The sample was taken from the depth of 170–180 cm to avoid cross contamination. High organic content with similar density to microplastic aggravates the condition of finding microplastics. Nevertheless, all polymers were correctly identified by µ-FTIR examination.

## Identification of microplastics using µ-FTIR technique

Reflectance µ-FTIR spectroscopy is an efficient, simple and non-destructive method for the detection of microplastics (Jung *et al.* 2018). FTIR spectroscopy allows us to identify correctly most plastic polymers based on their specific infrared absorption bands.

One crucial point is the sample preparation prior to µ-FTIR analyses. Mostly, filters containing microplastics are directly placed under crystal using ATR-FTIR

measurements and analyzing all particles in a chosen filter area step by step (Imhof *et al.* 2013; Käppler *et al.* 2016). In this study, a KBr pellet preparation has been used for fixing the separated particles on its surface to enable close contact to the surface of the germanium crystal using ATR mode of µ-FTIR measurements. This approach has been applied to all samples derived from both spiking experiments and real sample investigations.

Prior to all FTIR measurements in ATR reflectance mode, a microscopic analysis from the measured area of $100\,\mu m \times 100\,\mu m$ (or $200\,\mu m \times 200\,\mu m$) was performed (Figure 3). Based on the different colors of microplastic particles used in this study, identification of individual polymers by optical analyses was enabled (as illustrated in Figure 3). This allowed, in numerous cases, a cross check with IR derived data.

However, analytical identification was based on the characteristic IR absorption representing the molecular structures of the individual polymers. In principal, two approaches for identification can be followed: *ab initio* identification by spectral interpretation or identification based on comparison with reference spectra. Regarding the first approach, systematic changes in molecular structure of the eight polymers included in this study can be linked with three dominant structural moieties, the aliphatic and aromatic carbon backbone as well as carbonyl functional groups (as amides, urethanes, etc.). A systematic interpretation of the corresponding IR bands is illustrated as decision tree in Figure 4. The presence of the characteristic
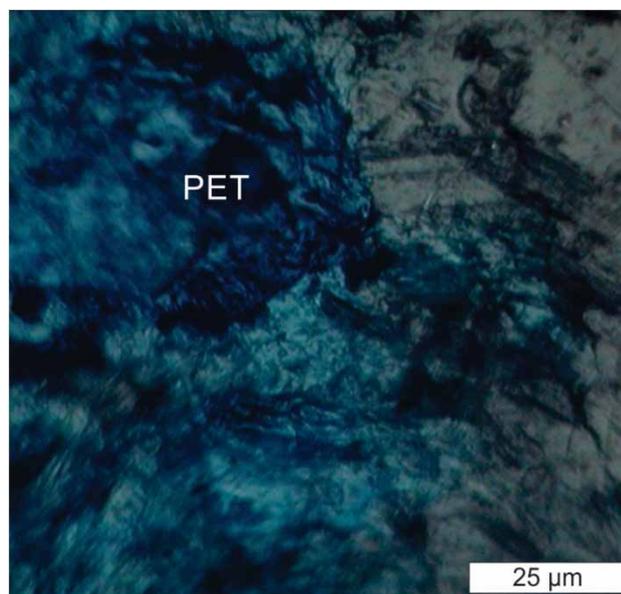
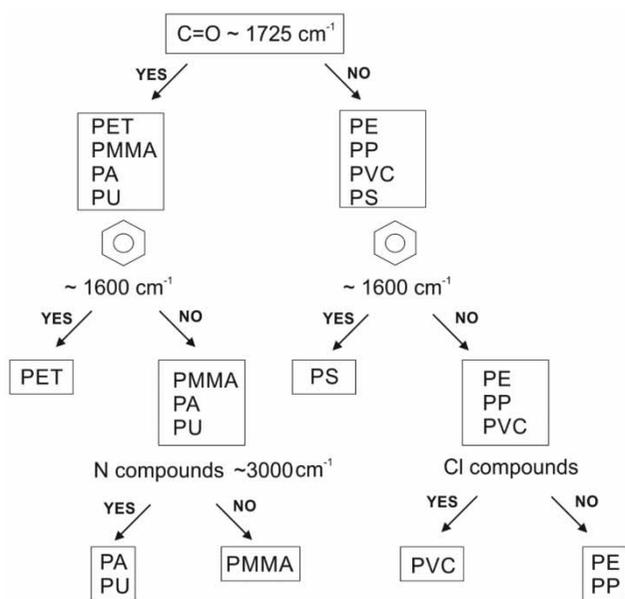**Figure 3** | Microscopic picture of PET pressed on KBr pellet in ATR-micro mode.

Figure 4 │ Decision tree for polymer identification.

Here, the aromatic moieties and the presence or absence of C-Cl absorption can differentiate PS, PE, PP and PVC. This approach can easily be followed by the corresponding spectra as illustrated in Figure 5.

However, in complex matrices such as sediments or soils where, in particular, the natural organic material cannot fully be removed, this approach might fail. Alternatively, a comparison or matching with reference spectra can be used. This approach needs a verification of similarity for spectra obtained by different IR approaches. For μ-FTIR it has been revealed in this study that spectra obtained by ATR reflectance and ATR transmission are directly comparable. However, also for this approach, a superimposition with signals of the natural organic matter needs to be considered. Both the optical approach as well as the μ-FTIR analyses have been applied successfully for all recovery experiments, spiking experiments and the natural sample.

Application on real sample from Albania

As a proof-of-concept, the developed multi-step approach was applied on one environmental sample from Albania. The soil sample was taken in May 2018 at a riparian wetland of the river Tiranë and was expected to contain microplastics contamination due to a waste deposit nearby. The approach covered all steps (grain size fractionation, density separation, μ-FTIR analyses in ATR mode) but was modified to the specific needs of the sample. Two finer classes, a

C=O subdivides the eight polymers into two groups. The first group contains the carbonyl containing polymers PET, PMMA, PA and PU. PET can further be differentiated from the others by the presence of aromatic signals pointing to its phenyl moiety. The remaining polymer types can be separated by the N-H signal that is visible for PA and PU but is not observed for PMMA. The second non-carbonylic group of polymers can be similarly subdivided (Figure 4).
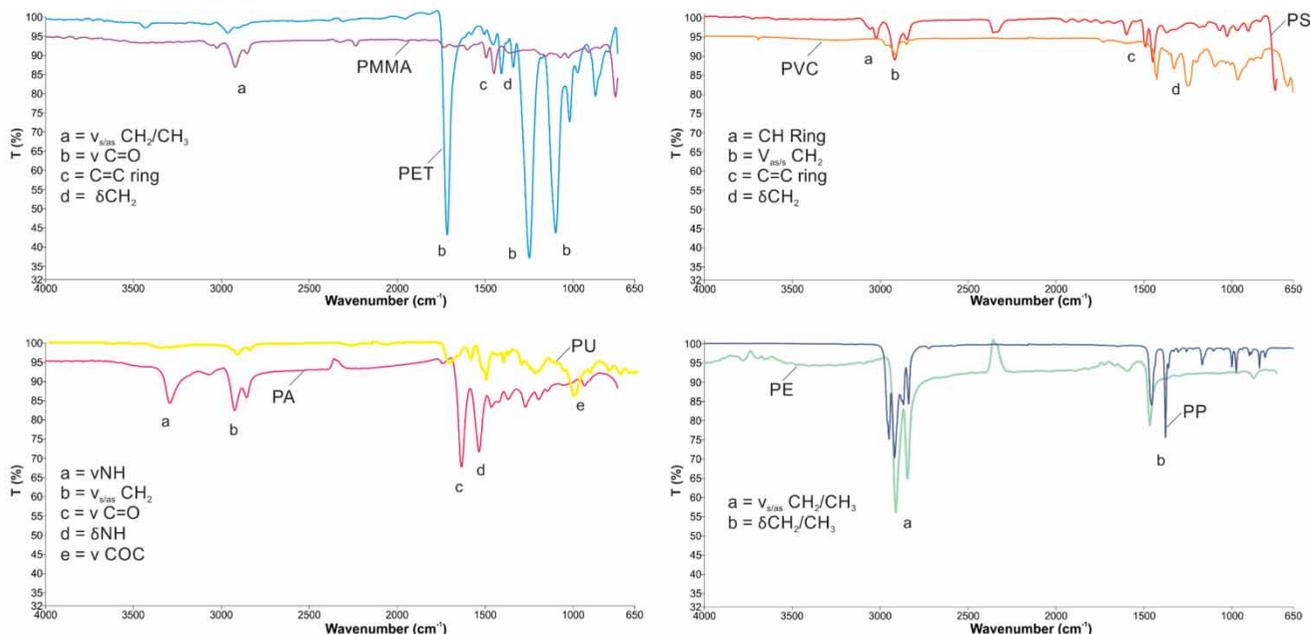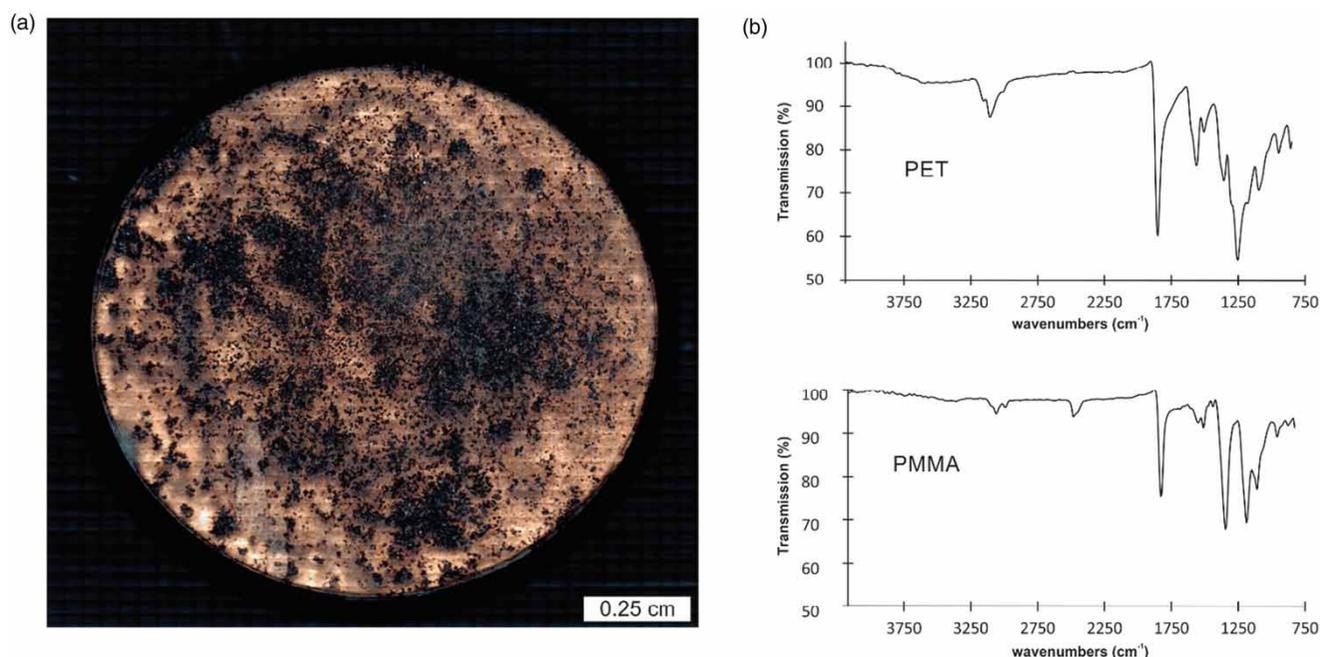

Figure 5 │ Spectra of all eight polymers measured in ATR-μ-FTIR mode. The most characteristic signals used for the decision tree in Figure 4 are marked.

Figure 6 | (a) Close picture of KBr pellet of the smallest size fraction of 20–50 μm; (b) PET and PMMA Spectra detected in the Albanian sample in A.

50–100 μm fraction and a 20–50 μm, as result of the very muddy property of the sample, have extended the sample size fractionation. Due to the experiences with the organic rich Teltow Canal sediments, one further sample treatment step was added: the removal of organic matter by 35% H_2O_2 under stirring and heating up to 60 °C (Cole *et al.* 2011; Sujathan *et al.* 2017).

An applied technique for removing organic material from environmental samples with H_2O_2 was not possible to completely detach the organic matter in various grain size fractions. Analyses of the KBr pellets of each sub fractions by microscopy visualized the organic matter residues as depicted in Figure 6 for the finest grain size class.

Systematic investigations of all grain size fractions by μ-FTIR using ATR mode revealed the occurrence of two out of the eight preselected polymers in different grain size fractions. PET was found in the grain size fraction 50–100 μm at the elevated amount, whereas PMMA was detected in the fraction 0.5–1 mm sporadically.

combining grain size fractionation, density separation with $ZnCl_2$ and final μ-FTIR based detection. The advantage of this method is the fast and simple sample preparation without need for special equipment and an easy application in any laboratory. High recovery rates for all tested polymers and all tested grain sizes in various types of particulate matter were achieved considering interferences by different sample matrices. The identification was accomplished by non-destructive μ-FTIR spectroscopy method using specifically prepared KBr pellets.

The suitability of the multi-step approach was verified by application on an environmental sediment sample from river Tiranë (Albania) which was expected to contain microplastics due to a nearby open waste dump. The analyses revealed two types of polymers that were found in different grain sizes.

Further potential of this multi-step approach is expected to be obtained by expanding the range of considered polymers and especially by applying pyrolysis-based detection methods for quantitation purposes.

CONCLUSIONS

The present study developed a simple and cost-effective multi-step approach to analyse microplastics in environmental particulate samples, such as soils and sediments, by

SUPPLEMENTARY MATERIAL

The Supplementary Material for this paper is available online at https://dx.doi.org/10.2166/wst.2020.173.

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