



Final Draft **of the original manuscript**

Lechthaler, S.; Hildebrandt, L.; Stauch, G.; Schüttrumpf, H.:
**Canola Oil Extraction in Conjunction with a Plastic Free
Separation Unit Optimises Microplastics Monitoring in Water
and Sediment.**

In: Analytical Methods. Vol. 12 (2020) 42, 5128 – 5139.

First published online by RSC: 24.09.2020

<https://dx.doi.org/10.1039/D0AY01574A>

1
2
3
4
5
6
7 1 Canola Oil Extraction in Conjunction with a Plastic
8
9
10
11 2 Free Separation Unit Optimises Microplastics
12
13
14
15 3 Monitoring in Water and Sediment
16
17
18
19

20 4 *Simone Lechthaler^{*12}, Lars Hildebrandt³⁴, Georg Stauch², Holger Schüttrumpf¹*

23 5 ¹Institute of Hydraulic Engineering and Water Resource Management, RWTH Aachen University,
24
25 6 Mies-van-der-Rohe-Straße 17, 52056 Aachen, Germany
26
27

28
29 7 ²Chair of Physical Geography and Geoecology, RWTH Aachen University, Wüllnerstraße 5b, 52062
30
31 8 Aachen, Germany
32
33

34 9 ³Helmholtz-Zentrum Geesthacht, Centre for Materials and Coastal Research, Institute of Coastal
35
36 10 Research, Marine Bioanalytical Chemistry, Max-Planck-Str. 1, 21502, Geesthacht, Germany
37
38

39 11 ⁴Universität Hamburg, Department of Chemistry, Inorganic and Applied Chemistry, Martin-Luther-
40
41 12 King-Platz 6, 20146, Hamburg, Germany
42
43

44 13 *Corresponding Author:

46
47 14 Simone Lechthaler
48
49

50
51 15 Phone: +49 (0) 241 8025266; Fax: +49 (0) 241 8025750
52
53

54 16 E-Mail: lechthaler@iww.rwth-aachen.de
55
56
57
58
59
60

1
2
3 17 KEYWORDS: Microplastics; Density-independent; Oil Extraction; Environmental Samples;
4
5 18 Separation Method; Method Validation, Density separation
6
7

8 19 ABSTRACT
9

10 20 Microplastics are widely distributed in the environment and to define contamination hot spots,
11
12 21 environmental samples have to be analysed by means of cost- as well as time-efficient and reliable
13
14 22 standardised protocols. Due to the lipophilic characteristics of plastic, oil extraction as a fast and
15
16 23 density-independent separation is beneficial for the crucial extraction step. It was extensively
17
18 24 validated (480 experiments) in two test setups by using canola oil and a cost-effective, plastic-free
19
20 25 separation unit with spiked microplastic (19 different polymer types) in the density range from
21
22 26 $\rho = 11 - 1,760 \text{ kg/m}^3$ and in the size range from 0.02 mm - 4.4 mm. Thus, an innovative, new
23
24 27 method combination was developed and profoundly validated for water and sediment samples
25
26 28 using only a short settling time of 15 minutes. Some experiments were also carried out with zinc
27
28 29 chloride to obtain additional reference data (particles $\leq 359 \mu\text{m}$). The total mean recovery rate was
29
30 30 89.3%, 91.7% within the larger microplastic fraction and 85.7% for the small fraction. Compared to
31
32 31 zinc chloride (87.6%), recovery rates differed not significantly with oil (87.1%). Furthermore, size
33
34 32 limits were set, since the method works best with particles $0.02 \text{ mm} \geq d \leq 3 \text{ mm}$. The proposed
35
36 33 method exhibits higher efficiency (84.8% for 20 - 63 μm) for the potentially most harmful
37
38 34 microplastic size fraction than the classic setup using brine solution. As a result, oil is a comparably
39
40 35 effective separation medium and offers further advantages for separating water and sediment
41
42 36 samples due to its density independence, simple and fast application and environmental
43
44 37 friendliness. Based on this, a new extraction protocol is presented here that confirms oil separation
45
46 38 as a sound and effective separation in microplastic analysis and identifies previously missing
47
48 39 information.
49
50
51
52
53
54
55
56
57
58
59
60

40 INTRODUCTION

41 In 1972, the detection of plastics in the environment was published for the first time.¹ Since then,
42 research has focused more and more on detection and analysis of plastic debris and especially
43 smaller plastic fragments, so-called microplastics.² Microplastics (MP) are defined as plastic
44 particles smaller 5 mm.^{3 4 5 6} Up to now, MP have been detected not only in the marine
45 environment,^{7 8} but also in fluvial systems^{9 10 11 12} that are determined as main transport paths for
46 MP into marine waters.¹³ According to estimations, 1.15 to 2.41 million tons of plastic waste are
47 brought into the oceans by rivers¹⁰ whereby larger plastic fragments can degrade into MP.
48 Additionally, lakes,¹⁴ shorelines,¹⁵ urban areas,¹⁶ wastewater treatment plants^{17 18 19} and sewage
49 sludge^{20 21} have been identified as areas where MP can be transported or accumulated. Road dust
50 and consequently tire abrasion constitute a large fraction, that is transported via street runoff into
51 sewers, surface waters and soils.^{22 23 24}

52 Besides the pollution of the aquatic environment, soils and sediments have come into focus playing
53 a key role as a sink for MP although the densities of MP, ranging from 11 kg/m³ for expanded
54 polystyrene (EPS)²⁵ up to 2,300 kg/m³ for polytetrafluoroethylene (PTFE),²⁶ suggest otherwise. MP
55 have not only been found in soils²⁷ but also in fluvial sediments,²⁸ floodplain sediments,²⁹ lake
56 sediments,³⁰ marine sediments,^{31 32 33 34 35} coastal sediments^{36 37 38 39 40} and also in deep-sea
57 sediments.^{41 42 43 44 45}

58 Since plastic is a persistent material, the consequence of MP entering the environment is an
59 accumulation in different areas, such as sediments, soils and finally deep sea sediments.¹³ As a
60 result, the ingestion by organisms has far-reaching impacts concerning harmful effects by co-
61 contaminants like pollutants or chemical additives of MP.⁴⁶ Thus, identification and further
62 understanding of accumulation hot-spots in the environment are of fundamental importance for
63 which sampling and evaluation is an essential step.

64 **Separation Methods and Materials.** Although samples have been taken in every environmental
 65 compartment and some protocols and guidelines for MP analysis have already been published,^{47 48}
 66 ^{49 50} there is no standardised separation method to extract MP from different matrices in
 67 environmental samples, like water and sediment samples (cf. Table 1).

68 **Table 1. Separation methods for extracting MP from environmental matrices.**

Method / Material	Material Characteristic	Recovery Rate [%]	Reference
Zinc Chloride (ZnCl ₂)	$\rho = 1,600 - 1,700 \text{ kg/m}^3$	95.5	51
Sodium Iodide (NaI)	$\rho = 1,800 \text{ kg/m}^3$	83.3	52
Calcium Chloride (CaCl ₂)	$\rho = 1,350 - 1,400 \text{ kg/m}^3$	49.0 - 62.0	38
Pressurized Fluid Extraction	-	85.0 - 111.0	53
Elutration + NaI Solution	Acid (HCl) 32.3 M; $\rho = 1,190 \text{ kg/m}^3$; Alkaline (NaOH) 52.5 M; $\rho = 2,130 \text{ kg/m}^3$; Nitric Acid (HNO ₃) 22.5 M; $\rho = 1,520 \text{ kg/m}^3$; Hydrogen peroxide (H ₂ O ₂) 32.6 M; $\rho = 1,110 \text{ kg/m}^3$ (30%); NaI $\rho = 1,600 \text{ kg/m}^3$	98.0 - 100.0	54
Enzymatic Purification	5% SDS; 1,100 U/mL Protease; >30 U/mL Cellulase; 35% H ₂ O ₂ ; >50 U/mL Chitinase; $\rho = 1,700 \text{ kg/m}^3 \text{ ZnCl}_2$	84.5	55
Electrostatic Separation	-	99.0	56
Froth Flotation with Oil	Wetting Agent (Pine Oil); Froth Conditioner	55.0	51
NaCl with Oil	$\rho = 1,200 \text{ kg/m}^3$, Oil	82.0	57
Canola Oil	$\eta < 200 \text{ cP}$	96.2 (91.7)	52 (²⁵)
Castor Oil	$\eta > 300 \text{ cP}$	99.9	58
Olive Oil	-	93.5	59

1
2
3 69 Regarding sediment separation, all methods using brine solutions, like NaCl, ZnCl₂, NaI, CaCl₂, are
4
5 70 based on the specific density of the solution and thus exclude certain polymer types with higher
6
7 71 densities such as PTFE ($\rho = 2,300 \text{ kg/m}^3$). Even though different densities are used concerning
8
9 72 these separation mediums, for example $1,200 \text{ kg/m}^3$ and $1,400 \text{ kg/m}^3$,⁶⁰ not the entire range is
10
11 73 covered, leading to discrimination against specific MP particles.

12
13
14
15 74 Imhof *et al.* (2012)⁵¹ developed a separation unit (Munich Plastic Sediment Separator, MPSS) that is
16
17 75 used with ZnCl₂-solution ($\rho = 1,600 - 1,700 \text{ kg/m}^3$) and reached a recovery rate from 95.5% for
18
19 76 small MP up to 100% for large MP. However, huge amounts of ZnCl₂ solution which is considered
20
21 77 corrosive and hazardous must be used for this separation. In a separation including acid and
22
23 78 alkaline digestion, dissolution or degradation of polymers can occur.⁶¹ Besides the selection of a
24
25 79 suitable separation medium, the choice of the separator is also important and influences the
26
27 80 achievable recovery rates. Electrostatic separation works without any chemical treatment but is
28
29 81 mainly used for large samples and the loss of sediment is indicated as disadvantage.⁵⁶ Regarding
30
31 82 separatory funnels, the outlet limits for example the size of separated MP particles in addition to
32
33 83 the chosen density of the separation medium.⁶² Thus, both aspects (separator and medium) must
34
35 84 be considered within the separation process in order to achieve the maximum recovery rates due to
36
37 85 the lack of standardisation. Furthermore, the application time per each sample is an important
38
39 86 factor with regard to high throughput to enable representative sampling of larger catchments. A
40
41 87 relatively new separation method is the extraction with oil which is hardly used at present, but has
42
43 88 many advantages over conventional methods.

44
45
46
47
48 89 **Oil Separation.** Only few studies were published regarding oil separation.⁶¹ Firstly, Crichton *et al.*
49
50 90 (2017)⁵² compared the oil method using canola oil with the use of a CaCl₂ and NaI solution for
51
52 91 extraction of post-consumer plastic fibres (polyester (PES), nylon), particles (acrylonitrile
53
54 92 butadiene styrene (ABS), vinyl, EPS) and PVC with densities between $15 - 1,450 \text{ kg/m}^3$, diameters

1
2
3 93 from 0.01 to 3.86 mm and length from 0.7 to 20.81 mm. To separate the sample material, sediment
4
5 94 and oil were mixed in an Erlenmeyer flask and then decanted into a separatory funnel. Resulting
6
7 95 recovery rates for oil separation ($96.2 \pm 2.2\%$) were significantly higher than for NaI ($83.3 \pm 5.8\%$)
8
9 96 or CaCl_2 ($69.0 \pm 3.6\%$). Their method validation focused on sediment and seawater samples.

11
12 97 Similar results were presented by Mani *et al.* (2019)⁵⁸ who employed castor oil and investigated the
13
14 98 following virgin polymers: polypropylene (PP), polystyrene (PS), polymethyl methacrylate (PMMA)
15
16 99 and polyethylene terephthalate glycol-modified (PET-G) in a size range from 0.3 - 1 mm. For
17
18 100 separation, they used a separation funnel from which oil and MP were filtrated while sediment was
19
20 101 drained. The achieved mean recovery rate was $99.0 \pm 4.0\%$ and $74.0 \pm 13\%$, respectively, based on
21
22 102 subjecting environmental samples from marine and fluvial surface waters as well as beach
23
24 103 sediments and agricultural soil to the treatment. Additionally, Karlsson *et al.* (2017)⁵⁷ also obtained
25
26 104 significantly higher recovery rates with oil throughout extraction of three different polymers (low
27
28 105 density polyethylene LDPE, high density polyethylene HDPE, PP).

29
30
31
32 106 Scopetani *et al.* (2020)⁵⁹ developed a new separation approach by using olive oil and
33
34 107 polytetrafluoroethylene (PTFE) cylinders to freeze the samples after homogenisation and remove
35
36 108 the frozen oil layer with MP afterwards. Consequently, only PTFE MP particles are inseparable with
37
38 109 this method. They investigated low density polymers PE and polyurethane (PU) (group 1), medium
39
40 110 density polymers PS and polycarbonate (PC) (group 2) and high density polymers PVC and PET
41
42 111 (group 3) in sizes ranging from 0.2 up to 2 mm and densities between 50 and $1,450 \text{ kg/m}^3$ in
43
44 112 compost and agricultural soil. The results differed for compost (90.5%) and soil (96.5%) and were
45
46 113 affected by the polymers density: 89.1% (group 1), 95.9% (group 2) and 95.5% (group 3).

47
48
49
50 114 Based on these results and the mentioned disadvantages of using electrostatic separation or a
51
52 115 density-dependent separation as well as possible inconsistencies (choice of oil and separation
53
54 116 medium) and missing investigations (post-processing and post-consumer materials) in oil
55
56
57
58
59
60

1
2
3 117 separation, this study presents an extensive validation of MP extraction by using canola oil as
4
5 118 separation medium. The proposed method is density-independent, confirmed by the numerous
6
7 119 different MP types, which is the decisive advantage. MP researchers should consider that, according
8
9 120 to the Stokes' law, small particles ($\sim 10 \mu\text{m}$) with a low density difference compared to a
10
11 121 surrounding medium bearing a high dynamic viscosity can need > 1 day to rise only by several
12
13 122 centimetre, for instance in a separatory funnel. One main difference of this study compared to the
14
15 123 validation of previous working methods is also based on the use of MP particles down to a few
16
17 124 micrometers in size which show the lowest velocity in the density separation setup.^{63 64}


18
19
20
21 125 For the separation, canola oil was used as well as different polymer types and shapes because of
22
23 126 missing results on polymer types concerning oil extraction. The investigations were performed
24
25 127 with oil and MP particles to reproduce water samples and additionally with a sediment-MP mixture
26
27 128 to simulate sediment samples. With these investigations, all polymers and different shapes typically
28
29 129 found in the environment ranging from 0.02 mm up to 4.4 mm with densities between
30
31 130 $11 \geq \rho \leq 1,760 \text{ kg/m}^3$ were analysed to determine recovery rates. In addition, the smaller particle
32
33 131 fraction (20 - 359 μm) was also tested with sediment in ZnCl_2 solution since a method validation for
34
35 132 these small sizes is still missing. Furthermore, this study applies the oil separation with an easy
36
37 133 applicable, plastic free separation unit that prevents material loss as well as size limits, simplifies
38
39 134 the treatment procedure and allows the separation of a high sample number in relatively short
40
41 135 time. Therefore, the results go beyond the previous outcomes and existing studies and provides the
42
43 136 necessary information and results so that this methodology can be established within research in
44
45 137 the future. The experiments verify and validate the method with numerous investigations and in
46
47 138 more detail regarding oil as extraction medium and a plastic free separation unit as separator.
48
49
50
51
52
53
54
55
56
57
58
59
60

1
2
3 139 MATERIAL AND METHODS
4

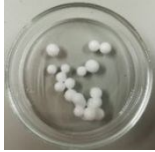








5 140 Here, two experimental series were conducted to validate the oil method and to separate MP from
6
7 141 water and sediment samples. Within the first setup, the size limit was 0.1 - 4.4 mm while the second
8
9 142 one focused on very small MP particles from 20 - 359 μm . Furthermore, we used only a very short
10
11 143 settling time of 15 minutes to develop a method that is suitable for a high throughput. Each type of
12
13 144 polymer (n=19) was investigated as water and sediment samples and each experiment was
14
15 145 repeated 10 times.
16
17
18

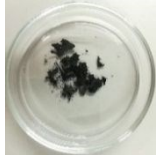
19 146 **First Experimental Setup.** In the first experimental setup, we used eight different polymers and nine
20
21 147 different types, describing shape or consistency, for the investigations resulting in eleven polymer
22
23 148 types (cf. Table 2). The selection was based on the polymers commonly found in the environment
24
25 149 that are described in most of peer-reviewed publications analysed by Frias *et al.* (2018)⁴⁹. The used
26
27 150 polymers types (n = 11) had different densities, colours, shapes and sizes. They were chosen to be
28
29 151 easily recognisable for the subsequent evaluation by counting to define the respective recovery
30
31 152 rate. Concerning fibres and filaments, the diameter was equal to the indicated size. Therefore, fibre
32
33 153 agglomerates were used concerning PVC and carbon fibres to facilitate the evaluation. The size of
34
35 154 the polymers ranged from 0.1 - 4.4 mm and the densities from 11 - 1,760 kg/m^3 . Concerning the
36
37 155 polymer types, we investigated post-processing products (microbead, fibre, filament, foam, film,
38
39 156 sphere, pellet) as well as post-consumer products (fragment, rubber). The PET fragments were
40
41 157 recovered from PET bottles and the synthetic rubber was originated from a pre-owned tyre.
42
43
44
45

46 158 **Table 2. Used polymer types (size limit: 0.1-4.4 mm) to validate the oil separation method.**
47

Polymer	Type	Density [kg/m^3]	Color	Size [mm]	Visual Image
Carbon	Filament	1,760	Black	0.1	

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

EPS Expanded Polystyrene	Foam	11	White	2.0	
PA (CoPA) Polyamide	Fibre	1,101	Blue	0.5	
PA (PA6) Polyamide	Pellet	1,110	Orange	0.5	
PE Polyethylene	Microbead	913	Blue	0.1	
PE Polyethylene	Film	917	Transpare nt	3.0	
PET Polyethylene terephthalate	Fragment	1,289	Colorful	1.0	
PP Polypropylene	Pellet	838	Black	3.0	
PS Polystyrene	Sphere	1,021	Orange	4.4	
PVC Polyvinylchlorid e	Fibre	1,315	White	2.5	

Synthetic Rubber	Rubber (Fragment)	1,051	Black	3.0	
------------------	--------------------------	-------	-------	-----	-------------------------------------------------------------------------------------

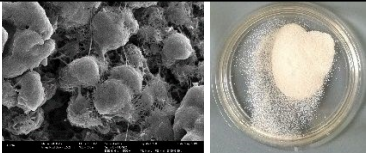
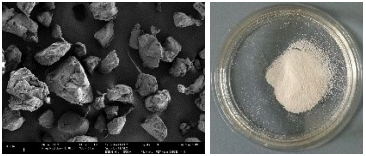
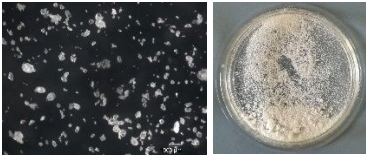

159 **Second Experimental Setup.** The second experimental setup focused on small, manually
160 fragmented, post-processing MP (cf. Table 3). MP in this size category are predominantly found in
161 the environment in fragmented shape as larger particles degrade and fragment there through the
162 various environmental influences.⁶⁵ The size range was 20 - 359 μm concerning the polymers PE,
163 PET, PP and polyvinylidene chloride (PVDC). To evaluate the influence of particle size on extraction
164 efficiency for exactly the same chemical matrix, we fractionated the PE and PET particles (20 -
165 63 μm , 63 - 125 μm , 125 - 250 μm) resulting in eight polymer types in total for all investigations. A
166 gravimetric analysis was applied because of the small sizes of the particles. To validate the results,
167 blank samples were produced, ten each for water and sediment and included in the analysis.

168 To obtain respective in-house reference particles, PE, PET and PVDC were purchased as powders
169 from Goodfellow (Hamburg, Germany). The chemical identity of all materials was confirmed by
170 attenuated total reflection Fourier transform infrared (FTIR) spectroscopy using an ALPHA I in
171 conjunction with a germanium or a diamond ATR crystal (Bruker Optik GmbH, Ettlingen, Germany).
172 The PVDC was sieved over a 1 mm stainless-steel sieve to remove particles > 1 mm. PE and PET
173 were subjected to a cascade of 250 μm , 125 μm , 63 μm and 20 μm stainless-steel sieves (Retsch,
174 Haan, Germany) in conjunction with an AS 300 sieving machine (Retsch, Germany). PP MP
175 reference particles were generated by milling PP pellets (isotactic, $M_w = 12,000$ g/mol,
176 $M_n = 5,000$ g/mol; Sigma-Aldrich, Muenchen, Germany). The grinding process by means of a ball
177 mill (Planetary ball mill PM400, Retsch, Hahn, Germany) is described in previous work.⁶³ Particle
178 sizes of the MP (cf. Table 3) were determined microscopically (PALM MicroBeam Microscope 192;
179 Carl Zeiss AG, Oberkochen, Germany) and by static light scattering using a red and a green laser

180 (Analysette 22 Wet Dispersion Unit and MicroTec plus; Fritsch, Idar-Oberstein, Germany). To check
 181 for surface irregularities, PE and PET were further investigated by confocal laser microscopy and
 182 scanning electron microscopy (cf. Table 3).⁶³

183 Since the amount of MP for each separation was very little (0.14 ± 0.04 g) and low recovery rates
 184 were expected, because material loss can quickly occur in these size ranges, the whole experimental
 185 set up for sediment samples was repeated by using $ZnCl_2$ as the most common separation
 186 medium.^{51 66 61} Thus, we are the first to also provide missing information on separation efficiency
 187 for most commonly applied density separation up for small MP particles down to $20 \mu m$ and
 188 validate the whole methodical procedure.

189 **Table 3: Used polymer types (size limit: 20 - 359 μm) to validate the oil separation method.**

Polymer	Type	Density [kg/m ³]	Color	Size [μm]	SEM / Microscope Image
PE Polyethylene	Fragment	960	White	20 - 63 63 - 125 125 - 250	
PET Polyethylene terephthalate	Fragment	1,300 - 1,400	White	20 - 63 63 - 125 125 - 250	
PP Polypropylene	Fragment	910	White	51 ± 11	
PVDC Polyvinylidene chloride	Fragment	1,630	White	359 ± 124	

190 **Experimental Procedure.** For both experimental setups, we used a Sediment-Microplastic-Isolator
 191 (SMI) to isolate MP from water and sediment samples (see Figure 1). The construction of this SMI

1
2
3 192 was provided by Coppock *et al.* (2017)⁶⁶ and reproduced for this method validation without plastic.
4

5 193 It consists of an aluminium tube with a total height of 479 mm. By using aluminium instead of
6

7 194 plastic, both cross-contamination and the accumulation of MP through hydrophobic interactions on
8

9 195 the surface of the separation equipment can be excluded. In the middle of the SMI, an aluminium
10

11 196 gate valve is installed to separate the upper and lower part of the tube. We decided to use a gate
12

13 197 valve instead of the ball valve, because preliminary tests have shown that closing the ball valve
14

15 198 partly creates turbulence which possibly pulls floating MP into the lower part of the separation
16

17 199 unit.
18
19
20

21 200 First, the device is loaded and after a settling time, during which a cover is used to prevent cross-
22

23 201 contamination from surrounding air, the gate valve is closed to separate sediment and MP in the oil
24

25 202 layer. In addition, cotton coats are used during separation processes to minimise self-
26

27 203 contamination from clothing. The whole apparatus is detachable to be cleaned and made of
28

29 204 aluminium as this material is resistant against oil and water and does not release plastic particle
30

31 205 during usage. Especially in regard to the gravimetric evaluation of the second setup, the SMI was
32

33 206 cleaned after each use with the same amount of distilled water and ethanol. With only one
34

35 207 treatment step and direct vacuum filtration from the separation unit, additional material loss and
36

37 208 possible size limits are prevented.
38
39
40

41 209 During the first setup, the SMI's aluminium surface was reactive due to mechanical roughening by
42

43 210 the stainless steel stirrer that was used for homogenisation of MP and oil. However, aluminium is
44

45 211 obviously susceptible to corrosion by acidic ZnCl_2 . Therefore, a completely identical SMI made of
46

47 212 PVC had to be used for all experiments with ZnCl_2 . To take possible cross-contamination from PVC
48

49 213 into account, blank samples were prepared.
50
51
52
53
54
55
56
57
58
59
60

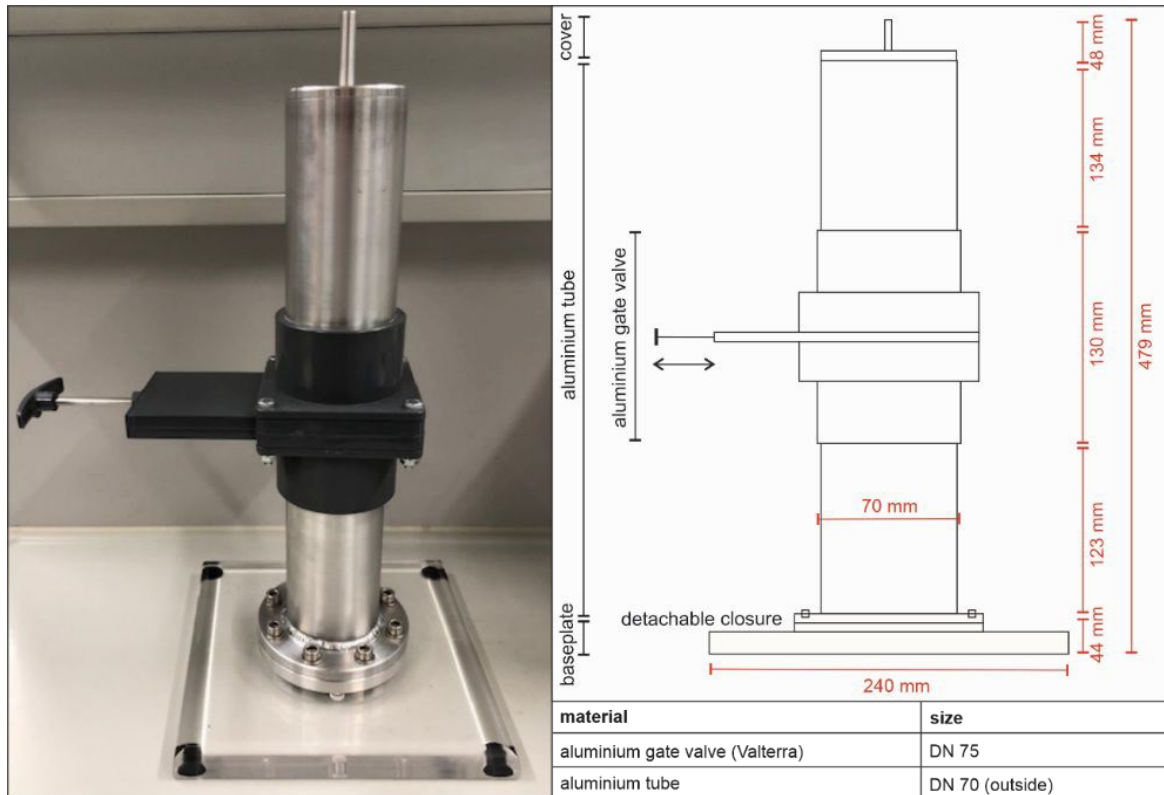


Figure 1. Photo (left) and drawing (right) of the plastic free SMI.

All experiments were performed firstly with canola oil and distilled water or ZnCl_2 ($\geq 97\%$, krist., $\rho = 2.92 \text{ g/cm}^3$; Carl Roth) and secondly in addition with sediment either from a natural river catchment (1st setup) or consisting of artificial quartz sand composed of equal parts of the grain fractions 0.3 - 0.6 mm, 0.71 - 1.25 mm and 2.0 - 4.0 mm (2nd setup). The fluvial sediment contained hardly any organic matter, therefore the method validation was carried out for the matrices water and sediment but not for organic matrix. The total volume for each separation in the SMI needs to be over 1,000 mL in order to fill the tube high enough to use the gate valve. It separates the lower part with distilled water (and if applicable sediments) and the upper part with distilled water and the oil layer with possible MP particles.

For each test implementation 1,000 mL distilled water, 10 mL canola oil and ten particles / 0.14 g of the investigated polymer were used and afterwards 10 g of fine-grained fluvial sediment / artificial

1
2
3 227 quartz sand were added. Additionally, within the second experimental setup, after defining the
4
5 228 recovery rates for oil extraction, the same experiments were conducted with ZnCl₂ solution of
6
7 229 density 1,650 kg/m³ and quartz sand to compare the results for such small MP size ranges. Since
8
9 230 the evaluation was conducted on a gravimetric basis, artificial sediment was used for the method
10
11 231 validation instead of natural, fluvial sediment to be sure of no organic content.

12
13
14
15 232 First, oil, distilled water and the polymer particles were filled into the SMI so that all particles came
16
17 233 directly into contact with the oil layer. Subsequently, the content was homogenised manually with a
18
19 234 stainless steel stirrer for 30 seconds which was then cleaned with distilled water over the unit to
20
21 235 avoid losing particles. After a settling time of 15 minutes, during which the SMI was covered up, the
22
23 236 gate valve was closed to separate the lower part containing water and, if used, sediments and the
24
25 237 upper part with oil / ZnCl₂ layer and MP particles. In the first setup, the upper part was decanted
26
27 238 into a beaker directly from the SMI which was additionally cleaned afterwards. Thus, it was easily
28
29 239 possible to recognise and count the spiked MP particles in the beaker because of sizes, colours and
30
31 240 limited number of ten particles per setup. As each polymer type was analysed in a separate
32
33 241 experimental procedure, there was no misidentification. The recovery rate for each polymer type
34
35 242 was determined with this particle count. This evaluation-method based on visual counting was
36
37 243 applied after Crichton *et al.* (2018)⁵². In the second setup, the upper part of the SMI was directly
38
39 244 vacuum filtrated on a glasmicrofibre filter (round filter MN GF-6 $d = 5.5$ cm). Additionally, ethanol
40
41 245 (96%) was used to clean the tube after oil experiments and rinse out all small MP particles from the
42
43 246 aluminium surface of the apparatus. After drying for 24 h at 40 °C, the filters were weighed and
44
45 247 recovery rates determined by a gravimetric analysis.

46
47
48
49
50 248 The evaluation resulted in recovery rates for water and sediment samples with oil extraction. Since
51
52 249 water samples are taken with nets to collect buoyant MP,⁶⁷ the filter residues are backwashed.
53
54 250 Therefore, the MP particles in both experimental setups without sediment are comparable to MP
55
56
57
58
59
60

251 from environmental water samples by neglecting organic and other environmental influences (salt
 252 water, UV light) on the particles (Figure 2 (1) and (3)). Furthermore, recovery rates for each
 253 polymer type and size for sediment samples were determined by using oil and spiked fluvial (1st
 254 setup) and artificial (2nd setup) sediment (Figure 2 (2) and (4)). Additionally, the extraction for
 255 sediment samples and ZnCl₂ ($\rho = 1,650 \text{ kg/m}^3$) was investigated (Figure 2 (5)). The evaluation of
 256 both experimental setups differs by visual analysis and gravimetric analysis. However, the results
 257 show recovery rates for the different polymer types.

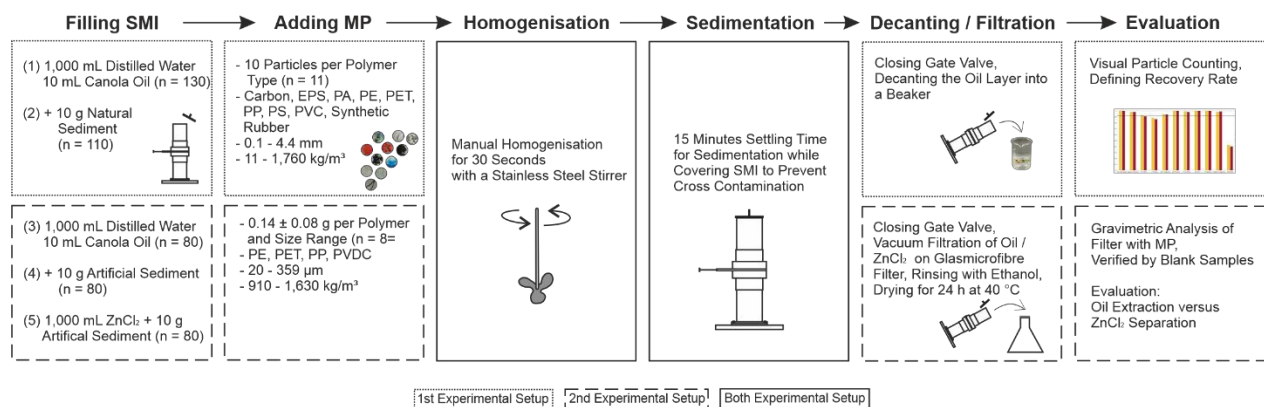


Figure 2. Flow chart of the experimental procedure.

RESULTS

261 As a first concise evaluation, we compared the recovery rates with other methods using oil or ZnCl₂
 262 (cf. Figure 3). It becomes evident, that pure oil extraction generates significantly higher recovery
 263 rates than these, where oil is only used additionally as wetting agent for froth flotation (recovery
 264 rates < 82.0%). Regarding the different oils, castor oil achieved the highest recovery rate (99.9%),
 265 followed by canola oil (96.2%), olive oil (93.5%) and again canola oil with our, slightly lower
 266 recovery rates (85.8% / 91.7%). Regarding small MP particles ($\leq 359 \mu\text{m}$), the recovery rates of
 267 canola oil (85.8%) and ZnCl₂ (87.6%) were similar, whereas previous studies reached significantly
 268 higher recovery rates with ZnCl₂ (95.5%⁵¹, 95.8%⁶⁶).

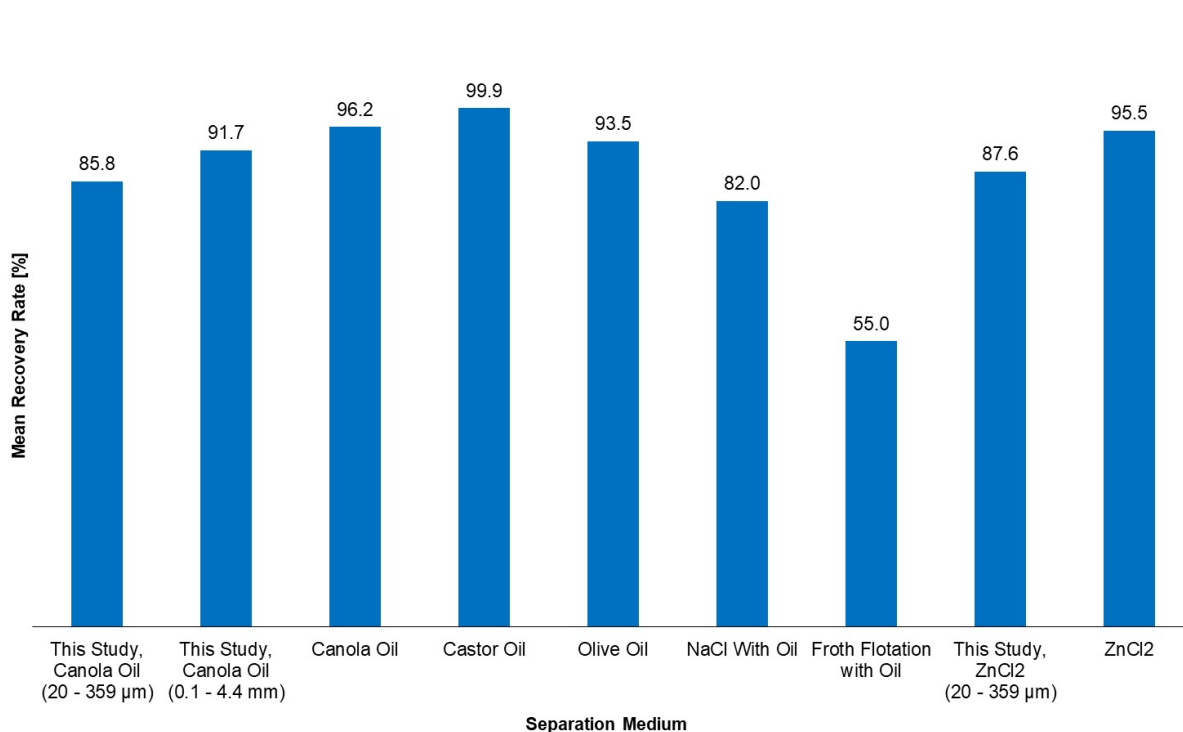


Figure 3: Recovery rates of separation methods using oil or ZnCl₂ (data from ^{52 58 59 57 51}).

The stable attraction between oil and synthetic polymers can be explained by the lipophilic characteristics of molecules from canola oil and the synthetic polymer's hydrocarbon chains which are lipophilic, too.⁵⁸ Crichton *et al.* (2017)⁵² referred to this attraction as oleophilic interaction. All polymer types from the first experimental setup regardless of density and size accumulate in the canola oil layer while sediment accumulates on the bottom of the beaker.

A correlation analysis was performed for all recovery rates determined with oil extraction. Therefore, Pearson's t-test was run assuming normally distributed data and it was analysed if size or density influenced the recovery rates because of the great variability there. Within the size fractions, the median was used for descriptive statistical analysis. Regarding all particle sizes (0.02 mm - 4.4 mm), there was no statistically significant correlation with the recovery rates (Pearson's t-test, $p > 0.05$, $n = 38$). However, it was assumed that the wettability influenced by the size of the particle surface is responsible for the oleophilic interaction which is why possible size limits were also defined based on the two experimental setups. The same results were obtained

284 regarding the density (Pearson's t-test, $p > 0.05$, $n = 38$) which emphasises oil extraction as a
 285 density-independent separation method. Nevertheless, the density within the two test setups was
 286 considered in the evaluation.

287 **First Experimental Setup.** 240 experiments were conducted and the individual results of each
 288 polymer type with recovery rates for water and sediment samples are shown in Table 4 sorted in
 289 ascending order. The lowest recovery rates are 42% (water) and 40% (sediment) for PS sphere.
 290 The highest recovery rate with 100% in both test implementations are achieved for PE microbeads,
 291 PE film, PP pellets and EPS foam. The results differ for all polymers concerning a lower recovery
 292 rate in the experiments with sediment, except for rubber fragments, but these differences are not
 293 significant as being within the standard deviation (Table 4). However, the mean recovery rate for
 294 water samples is 91.9% and for sediment 91.5% with an average difference of $\Delta = 0.5\%$. The
 295 median is 98% for both test implementations and underpins the high efficiency of the separation
 296 method. The standard deviation is relatively high ($\sigma_{\text{Water}} = 16.2\%$ and $\sigma_{\text{Sediment}} = 16.8\%$) for both
 297 matrices. This results from the low recovery rates of PS spheres as a statistical outlier (Dean Dixon
 298 Q-Test, $n = 11$, $\alpha = 0.001$) what can be attributed to the particle size which is clearly the largest and
 299 thus provides the greatest surface wettability. With 42% and 40%, the recovery rates strongly
 300 deviate from mean value and median and are significantly lower than all other rates. If PS sphere
 301 would not be considered in the calculation, the mean recovery and precision would be significantly
 302 higher: $96.9 \pm 4\%$ for water and $96.6\% \pm 4.5\%$ for sediment.

303 **Table 4. Recovery rates for each polymer type and statistical evaluation including mean value, median and standard**
 304 **deviation.**

Polymer Type	Recovery Rate Water [%]	Recovery Rate Sediment [%]
PS Sphere	42	40
PA Fibre	88	86
PA Pellet	92	91

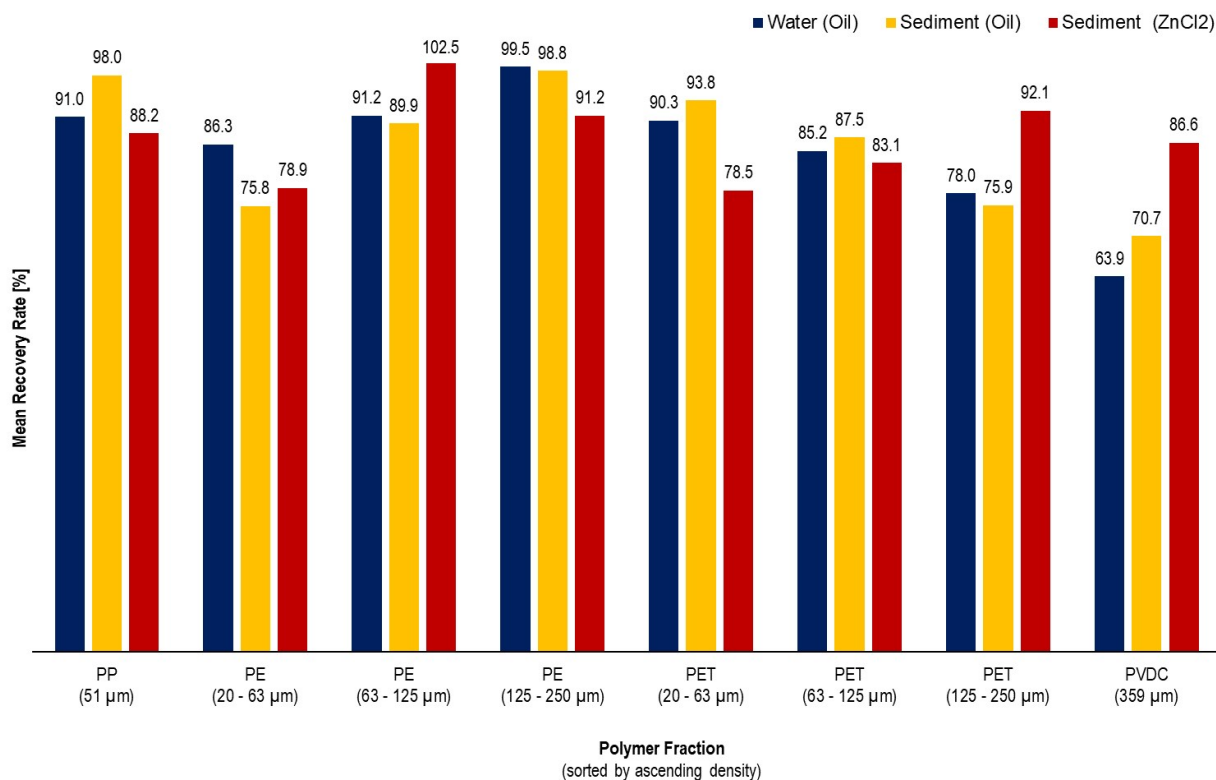
PET Fragment	94	94
Carbon Filament	98	98
Rubber Fragment	98	99
PVC Fibre	99	98
PE Microbead	100	100
PE Film	100	100
PP Pellet	100	100
EPS Foam	100	100
Statistical Parameter	Recovery Rate Water	Recovery Rate Sediment
Mean Value	91.9	91.5
Median	98.0	98.0
σ (Standard Deviation)	16.2	16.8

305 Although, there is no statistically significant correlation between the polymers' density and the
 306 related recovery, it seems that the density supports the recovery rate additionally, since the types
 307 with the lowest densities (EPS, PP, PE) have reached a recovery rate of 100% within all
 308 investigations. As the density does not seem to be responsible for the low recovery rate of PS
 309 sphere, an influence of the particles' size is conceivable. Even if the statistical evaluation showed no
 310 correlation, PS spheres as largest polymer type showed the lowest recovery rates. Future analysis
 311 of different types of PS might help to find an explanation. Therefore, the size and the particle's
 312 surface of this particle need to be considered. The spherical shape offers a large, plain surface,
 313 which can influence the stable attraction between PS sphere and oil and consequently the load
 314 capacity. To determine size ranges for this method, additionally preliminary tests were conducted
 315 for the fragments, where the sizes could be variated. PET fragments got the highest recovery rate
 316 with a size of 1 mm instead of 2 or 3 mm and rubber fragments with 3 mm than 4 or 5 mm. Since
 317 the PS sphere has a size of 4.4 mm it is suggested to optically sort out particles > 3 mm in
 318 environmental samples and to identify them with an (ATR) FTIR to avoid possible undetected

1
2
3 319 particles. All in all, recovery rates $> 90\%$ can be achieved mainly per particles ≤ 3 mm what must
4
5 320 be considered as method limitation. It is already proven that a post-treatment step with ethanol
6
7 321 after oil separation prevents a possible interference in case of FTIR measurements.⁵² As this
8
9 322 method validation worked with visual identification of the MP particles, no cleaning with ethanol
10
11 323 has been applied.

12
13
14
15 324 **Second Experimental Setup.** 240 experiments were performed to define recovery rates for eight
16
17 325 different polymer types. The recovery rates for water and sediment samples are similar per
18
19 326 polymer type and show a high precision ($\Delta = 0.2\%$) (cf. Figure 4). The highest recovery rates are
20
21 327 99.5% (water) / 98.8% (sediment) for PE ($125 - 250 \mu\text{m}$), the lowest recovery rates are detected
22
23 328 for PVDC with 63.9% (water) / 70.7% (sediment). Since gravimetric analysis was done, blank
24
25 329 samples were included in the evaluation of each experiment's result and in addition recovery rates
26
27 330 for MP in sediments where determined by using density-dependent separation with ZnCl_2 . Even if
28
29 331 correlations could not be found with density or size and recovery rates, the results show for oil
30
31 332 extraction lower recovery rates with ascending density, while ZnCl_2 seems to work better with
32
33 333 higher densities, like PVDC with $\rho = 1,630 \text{ kg/m}^3$. Furthermore, oil extraction compared to ZnCl_2
34
35 334 achieves better results for all small MP fraction, including PP ($51 \mu\text{m}$), PE ($20 - 63 \mu\text{m}$), PET ($20 -$
36
37 335 $63 \mu\text{m}$). Thus, the lower size limit for oil extraction can be set to $20 \mu\text{m}$.

38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60



336

337 **Figure 4: Recovery rates for all eight polymer types within oil extraction with water and sediment and ZnCl₂ separation of**
 338 **sediment samples.**

339 With regard to statistical evaluation, the overall mean value, median and standard deviation σ were
 340 determined (cf. Table 5). The mean values indicate a slightly higher recovery rate for ZnCl₂
 341 separation with sediment compared to oil extraction, while the median is slightly lower. The
 342 relatively high standard deviation for oil extraction can be explained with the low recovery rates for
 343 PVDC and PE (125 - 250 µm). Since PVDC has the largest particle size and the highest density, this
 344 polymer could be considered an outlier. Therefore, the recovery rates would be 88.8% (water) /
 345 88.0% (sediment).

346 **Table 5: Statistical evaluation including mean value, median and standard deviation for oil separation of water and**
 347 **sediment samples and ZnCl₂ separation of sediment samples.**

Statistical Parameter	Oil Separation: Recovery Rate Water	Oil Separation: Recovery Rate Sediment	ZnCl ₂ Separation Recovery Rate Sediment
Mean Value	85.7	85.9	87.6

Median	88.3	88.7	87.4
σ (Standard Deviation)	10.1	9.9	7.4

348 DISCUSSION

349 Compared to the results of Crichton *et al.* (2017)⁵², Mani *et al.* (2019)⁵⁸ and Scopetani *et al.*
350 (2020)⁵⁹, the oil separation method was extended to previously unexamined polymer types as well
351 as particle sizes and a promising separation device (cf. Table 6). Crichton *et al.* (2017)⁵²
352 investigated post-consumer polymers of EPS, PVC, PA, PES and ABS and concerning the polymer
353 type fibres and particles. Mani *et al.* (2019)⁵⁸ analysed post-processing (virgin) polymers of PP, PS,
354 PMMA and PET-G in size ranges from 0.3 - 1 mm with castor oil while all polymers had a fragment
355 shape. Scopetani *et al.* (2020)⁵⁹ examined post-processing polymers of PE, PU, PS, PC, PVC and PET.
356 In our comprehensive study, we evaluated both, post-processing (virgin) and post-consumer
357 polymers within 19 different polymer types considering all common polymers, shapes and
358 consistencies. Thus, the range of MP particles investigated has been significantly extended, which
359 demonstrates the previously lacking applicability and efficiency of oil separation in MP analysis.
360 Regarding the used types of oil, castor oil definitely has a higher dynamic viscosity η than canola
361 and olive oil (cf. Table 6) as well as a higher surface tension γ , what is probably responsible for a
362 high stable attraction. Furthermore, the use of separatory funnels can cause a limiting effectiveness
363 in terms of size,⁶² while the SMI works with a single separation step and without size limits, similar
364 to the cylinder used by Scopetani *et al.* (2020)⁵⁹. In contrast to separatory funnels, it enables
365 stirring and herewith mitigates burial of particles in sediment particles. In addition, compared to
366 castor oil the acquisition costs for canola oil are considerably lower. The SMI's design has thus been
367 improved to reduce possible influences and convinces by the single treatment step which, together
368 with the short settling time, significantly minimises the preparation time of each sediment sample
369 and is additionally more cost-effective due to the use of canola oil.

370 **Table 6: Comparison of published oil-extraction protocols with characteristics of separation medium,^{68 69 70 71} separator,**
 371 **polymers (post-processing = P-P, post-consumer = P-C) with shapes, densities and sizes, test matrices and mean**
 372 **recovery rates.**

Characteristic	This Study	Crichton <i>et al.</i> (2017) ⁵²	Mani <i>et al.</i> (2019) ⁵⁸	Scopetani <i>et al.</i> (2020) ⁵⁹
Medium	Canola Oil	Canola Oil	Castor Oil	Olive Oil
- Density (ρ)	- 916 kg/m ³	- 916 kg/m ³	- 959 kg/m ³	- 911 kg/m ³
- Dynamic Viscosity (η)	- 71.63 mPa s	- 71.63 mPa s	- 852.84 mPa s	- 74.1 mPa s
- Surface Tension (γ)	- 31.1 mN/n	- 31.1 mN/n	- 39.0 mN/n	- 33.0 mN/n
Separator	SMI	Separatory Funnel	Separatory Funnel	Cylinder
Polymers	P-P, P-C: EPS, CoPA, PA6, PE, PET, PP, PS, PVC, PVDC, Synthetic Rubber, Carbon	P-C: EPS, PVC, PA, PES, ABS	P-P: PP, PS, PMMA, PET-G	P-P: PE, PU, PS, PC, PVC, PET
Shapes	Pellets, Fibres, Spheres, Microbeads, Films, Fragments, Filaments, Foams	Fibres, Particles	Fragments	-
Polymer Densities (ρ)	11 - 1,760 kg/m ³	15 - 1,450 kg/m ³	840 - 1,270 kg/m ³	50 - 1,450 kg/m ³
Particle Size Range	0.02 mm - 4.4 mm	0.01 - 3.98 mm (width)	0.3 - 1.0 mm	0.2 - 2.0 mm
Test Matrices	Fluvial / artificial sediments	Beach Sediments	Marine / fluvial suspended surface solids, beach sediments, agricultural soil	Soil, Compost
Mean Recovery Rate	85.8%	96.2%	99.9%	93.5%

1
2
3 373 In comparison to density-dependent separation methods, the results indicate even higher efficiency
4
5 374 for the smallest MP fraction in sediment samples. This underpins the high potential of this method
6
7 375 for researcher applying any advanced spectroscopic imaging with size detection limits down a few
8
9 376 micrometers. Even the most costly instrument cannot compensate for bias which was introduced at
10
11 377 the sampling and extraction state. Oil separation works for all MP particles separately from the
12
13 378 density although the influence of density and also of particle size cannot entirely be excluded (as a
14
15 379 cause of limited statistical power). Thus, other factors need to be considered, too. Compared to
16
17 380 heavy metal solutions, like $ZnCl_2$, oil extraction bears higher environmental friendliness and low
18
19 381 costs.⁶¹ The costs per sample are negligible compared to $NaCl$, $CaCl_2$, NaI and $ZnCl_2$.⁵² With regard to
20
21 382 the highest efficiency, separation methods can therefore also be used in complementary ways.
22
23
24

25 383 All in all, the method validation improves the MP separation due to the following aspects: the
26
27 384 design of the SMI is easy to apply (filling, homogenising, covering, sealing and vacuum filtering) and
28
29 385 has been improved by previous possible influences (cross-contamination, accumulation by
30
31 386 hydrophobic interactions, gate valve instead of ball valve). The high throughput is mainly due to the
32
33 387 short settling time of 15 min and the one-step treatment, which nevertheless show comparatively
34
35 388 high recovery rates. Furthermore, vacuum filtration is carried out directly from the separation unit,
36
37 389 where otherwise a separating funnel is often used, which again has limitations due to the outlet. In
38
39 390 comparison to previous studies, the wide variety of different polymers in terms of density, shape
40
41 391 and origin also confirms the density independence of this methodology in a much more
42
43 392 comprehensive and intensive way.
44
45
46
47

48 393 To our best knowledge, our method validation provides the highest degree of comprehensiveness
49
50 394 considering the variability of polymer types, densities ($11 \leq \rho \leq 1,760 \text{ kg/m}^3$) and sizes (0.02 mm -
51
52 395 4.4 mm). Moreover, different shapes and stages of the product life cycle were considered (post-
53
54 396 processing / consumer). Summing, it was aimed for covering a representative cross-section of
55
56
57
58
59
60

1
2
3 397 environmental particulate plastic. On the other hand, this variability could be responsible for the minor
4
5 398 decreases in efficiency. However, the benefits are an extensive and robust validation of oil extraction in
6
7 399 combination with the SMI for environmental MP analysis
8
9

10 400 CONCLUSION

11
12
13 401 The experimental results show that the proposed oil extraction protocol in conjunction with the
14
15 402 SMI poses an innovative and new method combination and is easily applicable for all types of
16
17 403 polymer regardless of density, shape or consistency. Only the particle size is a not negligible factor
18
19 404 and the highest recovery rates are achieved for particles $0.02 \text{ mm} \geq d \leq 3 \text{ mm}$. Therefore, the
20
21 405 results show in addition to the high effectiveness also the method's limitations. In comparison with
22
23 406 other separation methods, oil separation with canola oil is a cost-effective and environmentally
24
25 407 friendly method. No hazardous chemicals are needed and per separation only 10 mL canola oil are
26
27 408 used. For future studies, the methods (oil / ZnCl_2) can be considered complementary, since both
28
29 409 seem to work better for different size classes.
30
31
32
33

34 410 A limiting factor regarding the upcoming separation of sediment samples with this method is the
35
36 411 small samples size of sediment (10 g) that can be analysed per investigation hampering
37
38 412 representativeness. The uncertainty increases with extrapolation of determined MP concentrations
39
40 413 to a larger volume. Furthermore, the separation efficiency with light sediment particles and the
41
42 414 influence of organic matter need to be considered. However, these questions have to be answered
43
44 415 in future studies by application of different sediment types and organic matrix as well as a
45
46 416 corresponding sampling design and subsequent uncertainty evaluation to assess the homogeneity
47
48 417 of MP distribution in different compartments. Additional information can be generated by
49
50 418 numerical simulation, where first assumptions of MP in sediments can be based on settling and rise
51
52 419 velocities, erosion and infiltration behaviour.^{72 73 74} This study has thus made a major contribution
53
54
55
56
57
58
59
60

1
2
3 420 to the validation of oil separation in all size, density and shape ranges of MP in water and sediment
4
5 421 samples.

6
7
8 422 For the generation of a larger picture of MP contamination of environmental systems, validated
9
10 423 methods that guarantee high throughput, cost-efficiency, low risk potential for the operator and
11
12 424 environmental compatibility are mandatory. Oil separation in conjunction with the SMI fulfils all
13
14
15 425 these criteria.

16
17
18 426 AUTHOR INFORMATION

19
20
21 427 **Acknowledgments**

22
23
24 428 The authors gratefully acknowledge the support in the experimental procedure by Tabea Quante
25
26 429 and Tom Lennart Heid and the corresponding authors thanks the funding of RWTH Scholarships for
27
28 430 Doctoral Students.

29
30
31 431 **Notes**

32
33
34 432 The authors declare no competing financial interest.

35
36
37
38 433

434 REFERENCES

- 435 1 E. Carpenter and K. L. Smith Jr., *Science*, 1972, **175**, 1240–1241.
- 436 2 R. C. Thompson, Y. Olsen, R. P. Mitchell, A. Davis, S. J. Rowland, A. W. G. John, D.
437 McGonigle and A. E. Russell, *Science*, 2004, **304**, 838.
- 438 3 R. C. Thompson, C. J. Moore, F. S. Vom Saal and S. H. Swan, *Philosophical transactions of*
439 *the Royal Society of London. Series B, Biological sciences*, 2009, **364**, 2153–2166.
- 440 4 A. Andrady, *Marine Pollution Bulletin*, 2011, 1596–1605.
- 441 5 M. Cole, P. Lindeque, C. Halsband and T. Galloway, *Marine Pollution Bulletin*, 2011, **12**,
442 2588–2597.
- 443 6 C. Arthur, J. Baker and H. Bamford, *Proceedings of the International Research Workshop*
444 *on the Occurrence, Effects and Fate of Microplastic Marine Debris. Sept. 9-11, 2008*,
445 NOAA Technical Memorandum NOS-OR&R-30, 2009.
- 446 7 H. S. Auta, C. Emenike and S. H. Fauziah, *Environment International*, 2017, 165–176.
- 447 8 J. G. B. Derraik, *Marine Pollution Bulletin*, 2002, 842–852.
- 448 9 A. McCormick, T. Hoellein, M. London, J. Hittie, J. Scott and J. Kelly, *Ecosphere*, 2016, **7**,
449 1–22.
- 450 10 L. C. M. Lebreton, J. van der Zwet, J.-W. Damsteeg, B. Slat, A. Andrady and Reisser Julia,
451 *Nature Communications*, 2017.
- 452 11 M. Wagner, C. Scherer, D. Alvarez-Munoz, N. Brennholt, X. Bourrain, S. Buchinger, E.
453 Fries, C. Grosbois, J. Klasmeier, T. Marti, S. Rodriguez-Mozaz, R. Urbatzka, A. D. Vethaak,
454 M. Winter-Nielsen and G. Reifferscheid, *Environmental Science Europe*, 2014, **26**, 1–9.
- 455 12 D. Eerkes-Medrano, R. C. Thompson and D. C. Aldridge, *Water Research*, 2015, **75**, 63–
456 82.
- 457 13 K. Waldschläger, S. Lechthaler, G. Stauch and H. Schüttrumpf, *Science of the Total*
458 *Environment*, 2020, 136584.
- 459 14 A. Baldwin, S. Corsi and S. Mason, *Environmental Science and Technology*, 2016, 10377–
460 10385.
- 461 15 M. A. Browne, P. Crump, S. J. Niven, E. Teuten, A. Tonkin, T. Galloway and R. Thompson,
462 *Environmental Science and Technology*, 2011, 9175–9179.
- 463 16 R. Dris, J. Gasperi, V. Rocher, M. Saad, N. Renault and B. Tassin, *Environmental*
464 *Chemistry*, 2015, **12**, 592.
- 465 17 G. Kalcikova, B. Alic, T. Skalar, M. Bundschuh and A. Zgajnar Gotvajn, *Chemosphere*,
466 2017, 25–31.
- 467 18 S. M. Mintenig, I. Int-Veen, M. Löder, S. Primpke and G. Gerdt, *Water Research*, 2017,
468 365–372.
- 469 19 P. Kay, R. Hiscoe, I. Moberley, L. Bajic and N. McKenna, *Environmental science and*
470 *pollution research international*, 2018, **25**, 20264–20267.

- 1
2
3 471 20 K. A. V. Zubris and B. K. Richards, *Environmental Pollution*, 2005, **138**, 201–211.
4
5 472 21 D. Habib, D. C. Locke and L. J. Cannone, *Water, Air, and Soil Pollution*, 1996, 1–8.
6
7 473 22 P. J. Kole, A. J. Löhr, van Belleghem, Frank G. A. J. and a. M. J. Ragas, *International Journal*
8 474 *of Environmental Research and Public Health*, 2017, **14**, 1–31.
9
10 475 23 C. Vogelsang, A. Lusher, M. E. Dadkhah, I. Sundvor, M. Umar, S. B. Ranneklev, D. Eidsvoll
11 476 and S. Meland, *Microplastics in road dust - characteristics, pathways and measures*.
12 477 Technical Report M-959, 2018.
13
14 478 24 B. Baensch-Baltruschat, B. Kocher, F. Stock and G. Reifferscheid, *The Science of the total*
15 479 *environment*, 2020, **733**, 137823.
16
17 480 25 S. Lechthaler, G. Stauch and H. Schüttrumpf, in *Proceedings of the 2nd International*
18 481 *Conference on Microplastic Pollution in the Mediterranean Sea*, ed. M. Cocca, E. Di Pace,
19 482 M. E. Errico, G. Gentile, A. Montarsolo, R. Mossotti and M. Avella, Springer International
20 483 Publishing, Cham, 2020, pp. 282–286.
21
22 484 26 K. Enders, R. Lenz, C. A. Stedmon and T. G. Nielsen, *Marine Pollution Bulletin*, 2015, **100**,
23 485 70–81.
24
25 486 27 M. Bläsing and W. Amelung, *Science of the Total Environment*, 2018, 422–435.
26
27 487 28 S. Klein, E. Worch and T. P. Knepper, *Environmental Science and Technology*, 2015,
28 488 6070–6076.
29
30 489 29 M. Scheurer and M. Bigalke, *Environmental Science and Technology*, 2018, **52**, 3591–
31 490 3598.
32
33 491 30 A. Ballent, P. L. Corcoran, O. Madden, P. A. Helm and F. J. Longstaffe, *Marine Pollution*
34 492 *Bulletin*, 2016, **110**, 383–395.
35
36 493 31 P. Näkki, O. Setälä and M. Lehtiniemi, *Environmental pollution (Barking, Essex : 1987)*,
37 494 2019, **249**, 74–81.
38
39 495 32 E. K. Fischer, *Distribution of microplastics in marine species of the Wadden Sea along*
40 496 *the coastline of Schleswig-Holstein, Germany*, Hamburg, 2019.
41
42 497 33 C. Lorenz, L. Roscher, M. S. Meyer, L. Hildebrandt, J. Prume, M. G.J. Löder, S. Primpke and
43 498 G. Gerdts, *Environmental Pollution*, 2019.
44
45 499 34 Y. Y. Tsang, C. W. Mak, C. Liebich, S. W. Lam, E. T.-P. Sze and K. M. Chan, *Marine Pollution*
46 500 *Bulletin*, 2017, **115**, 20–28.
47
48 501 35 M. Zobkov and E. Esiukova, *Marine Pollution Bulletin*, 2017, **114**, 724–732.
49
50 502 36 C. Alomar, F. Estarellas and S. Deudero, *Marine Environmental Research*, 2016, 1–10.
51
52 503 37 A. Turra, A. B. Manzano, R. J. S. Dias, M. M. Mahiques, L. Barbosa, D. Balthazar-Silva and
53 504 F. T. Moreira, *Scientific Reports*, 2014, **4**, 4435.
54
55 505 38 A. Stolte, S. Forster, G. Gerdts and H. Schubert, *Marine Pollution Bulletin*, 2015, 216–229.
56
57 506 39 O. Piperagkas, N. Papageorgiou and I. Karakassis, *Estuarine, Coastal and Shelf Science*,
58 507 2019, **219**, 169–175.
59
60

- 1
2
3 508 40 M. Gregory, *N.Z. Journal of Marine and Freshwater Research*, 1978, **12**.
4
5 509 41 L. van Cauwenberghe, A. Vanreusel, J. Mees and C. R. Janssen, *Environmental Pollution*,
6 510 2013, **182**, 495–499.
7
8 511 42 L. C. Woodall, A. Sanchez-Vidal, M. Canals, G. L. J. Paterson, R. Coppock, V. Sleight, A.
9 512 Calafat, A. D. Rogers, B. E. Narayanaswamy and R. C. Thompson, *Royal Society open*
10 513 *science*, 2014, **1**, 140317.
11
12 514 43 M. Bergmann, V. Wirzberger, T. Krumpfen, C. Lorenz, S. Primpke, M. B. Tekman and G.
13 515 Gerdt, *Environmental Science and Technology*, 2017, **51**, 11000–11010.
14
15 516 44 D. K. Kanhai, C. Johansson, J.P.G.L. Frias, K. Gardfeldt, R. C. Thompson and I. O'Connor,
16 517 *Deep Sea Research Part I: Oceanographic Research Papers*, 2019, **145**, 137–142.
17
18 518 45 X. Peng, M. Chen, S. Chen, S. Dasgupta, H. Xu, K. Ta, M. Du, J. Li, Z. Guo and S. Bai,
19 519 *Geochem. Persp. Lett.*, 2018, 1–5.
20
21 520 46 R. C. Thompson and I. E. Napper, in *Plastics and the Environment*, ed. R. E. Hester and R.
22 521 M. Harrison, Royal Society of Chemistry, Cambridge, 2018, pp. 60–81.
23
24 522 47 European Commission, *Guidance on Monitoring of Marine Litter in European Seas. A*
25 523 *guidance document within the Common Implementation Strategy for the Marine*
26 524 *Strategy Framework Directive*. MSFD Technical Subgroup on Marine Litter, Publications
27 525 Office, Luxembourg, 2013.
28
29 526 48 J. Masura, J. Baker, G. Foster and C. Arthur, *Laboratory Methods for the Analysis of*
30 527 *Microplastics in the Marine Environment. Recommendations for quantifying synthetic*
31 528 *particles in waters and sediments*, U.S. Department of Commerce, NOAA Technical
32 529 Memorandum NOS-OR&R-48, 2015.
33
34 530 49 J. Frias, E. Pagter, R. Nash, I. O'Connor, O. Carretero, A. Filgueiras, L. Viñas, J. Gago, J.
35 531 Antunes, F. Bessa, P. Sobral, A. Goruppi, V. Tirelli, M. L. Pedrotti, G. Suaria, S. Aliani, C.
36 532 Lopes, J. Raimundo, M. Caetano, L. Palazzo, G. A. de Lucia, A. Camedda, S. Muniategui, G.
37 533 Grueiro, V. Fernandez, J. Andrade, R. Dris, C. Laforsch, B. M. Scholz-Böttcher and G.
38 534 Gerdt, *Standardised protocol for monitoring microplastics in sediments*, JPI Oceans,
39 535 2018.
40
41 536 50 GESAMP, *Guidelines for the monitoring and assessment of plastic litter in the ocean. A*
42 537 *report to inform the Second United Nations Environment Assembly*, Reports and
43 538 Studies 99, 2019.
44
45 539 51 H. Imhof, J. Schmid, R. Niessner, N. Ivleva and C. Laforsch, *Limnology and Oceanography:*
46 540 *Methods*, 2012, 524–537.
47
48 541 52 E. M. Crichton, M. Noël, E. A. Gies and P. S. Ross, *Analytical Methods*, 2017, **9**, 1419–
49 542 1428.
50
51 543 53 S. Fuller and A. Gautam, *Environmental Science and Technology*, 2016, 5774–5780.
52
53 544 54 M. Claessens, L. van Cauwenberghe, M. B. Vandegehuchte and C. R. Janssen, *Marine*
54 545 *Pollution Bulletin*, 2013, **70**, 227–233.
55
56
57
58
59
60

- 1
2
3 546 55 M. G. J. Löder, H. K. Imhof, M. Ladehoff, L. A. Löschel, C. Lorenz, S. Mintenig, S. Piehl, S.
4 547 Primpke, I. Schrank, C. Laforsch and G. Gerdt, *Environmental Science and Technology*,
5 548 2017, 14283–14292.
- 7 549 56 S. Felsing, C. Kochleus, S. Buchinger, N. Brennholt, F. Stock and G. Reifferscheid,
8 550 *Environmental Pollution*, 2018, 20–28.
- 10 551 57 T. M. Karlsson, A. D. Vethaak, B. C. Almroth, F. Ariese, M. van Velzen, M. Hassellöv and H.
11 552 A. Leslie, *Marine Pollution Bulletin*, 2017, **122**, 403–408.
- 13 553 58 T. Mani, S. Frehland, A. Kalberer and P. Burkhardt-Holm, *Analytical Methods*, 2019.
- 14 554 59 C. Scopetani, D. Chelazzi, J. Mikola, V. Leiniö, R. Heikkinen, A. Cincinelli and J. Pellinen,
15 555 *The Science of the total environment*, 2020, **733**, 139338.
- 17 556 60 V. Hidalgo-Ruz, L. Gutow, R. C. Thompson and M. Thiel, *Environ. Sci. Technol.*, 2012, **46**,
18 557 3060–3075.
- 20 558 61 F. Stock, C. Kochleus, B. Bänisch-Baltrusch, N. Brennholt and G. Reifferscheid, *Trends in*
21 559 *Analytical Chemistry*, 2019, **113**, 84–92.
- 23 560 62 M. Lares, M. C. Ncibi, M. Sillanpää and M. Sillanpää, *Environmental science and pollution*
24 561 *research international*, 2019, **26**, 12109–12122.
- 26 562 63 L. Hildebrandt, N. Voigt, T. Zimmermann, A. Reese and D. Proefrock, *Marine*
27 563 *Environmental Research*, 2019, 104768.
- 29 564 64 L. Hildebrandt, D. M. Mitrano, T. Zimmermann and D. Proefrock, *Front. Environ. Sci.*,
30 565 2020, **8**, 305.
- 31 566 65 T. Bond, V. Ferrandiz-Mas, M. Felipe-Sotelo and E. Van Sebille, *Critical Reviews in*
32 567 *Environmental Science and Technology*, 2018, **48**, 685–722.
- 34 568 66 R. Coppock, M. Cole, P. Lindeque, A. Queiros and T. Galloway, *Environmental Pollution*,
35 569 2017, 829–837.
- 37 570 67 J. Frias, E. Pagter, R. Nash, I. O'Connor, O. Carretero, A. Filgueiras, L. Viñas, J. Gago, J.
38 571 Antunes, F. Bessa, P. Sobral, A. Goruppi, V. Tirelli, M. L. Pedrotti, G. Suaria, S. Aliani, C.
39 572 Lopes, J. Raimundo, M. Caetano, L. Palazzo, G. A. de Lucia, A. Camedda, S. Muniategui, G.
40 573 Grueiro, V. Fernandez, J. Andrade, R. Dris, C. Laforsch, B. M. Scholz-Böttcher and G.
41 574 Gerdt, *Standardised protocol for monitoring microplastics in seawater*, JPI Oceans,
42 575 2019.
- 44 576 68 E. A. Melo-Espinosa, Y. Sánchez-Borroto, M. Errasti, R. Piloto-Rodríguez, R. Sierens, J.
45 577 Roger-Riba and A. Christopher-Hansen, *Energy Procedia*, 2014, **57**, 886–895.
- 47 578 69 V. R. Patel, G. G. Dumancas, L. C. Kasi Viswanath, R. Maples and B. J. J. Subong, *Lipid*
48 579 *insights*, 2016, **9**, 1–12.
- 50 580 70 R. Przybylski, C. G. Biliaderis and N. A. M. Eskin, *J Am Oil Chem Soc*, 1993, **70**, 1009–
51 581 1015.
- 53 582 71 S. N. Sahasrabudhe, V. Rodriguez-Martinez, M. O'Meara and B. E. Farkas, *International*
54 583 *Journal of Food Properties*, 2017, **25**, 1–17.

1
2
3 584 72 K. Waldschläger and H. Schüttrumpf, *Environmental Science and Technology*, 2019, **53**,
4 585 1958–1966.

5
6 586 73 K. Waldschläger and H. Schüttrumpf, *Environmental Science and Technology*, 2019, **53**,
7 587 13219–13227.

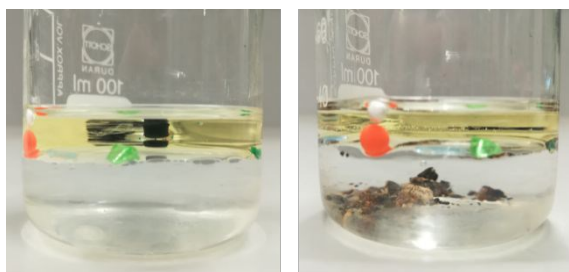
8
9 588 74 K. Waldschläger and H. Schüttrumpf, *Environ. Sci. Technol.*, 2020.

10 589

11
12 590

13
14 591

15
16
17 592 GRAPHICAL ABSTRACT



27
28 594

29

30

31

32

33

34

35

36

37

38

39

40

41

42

43

44

45

46

47

48

49

50