

1 **Sorting microplastics from other materials in water samples by ultra-high-definition imaging**

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11

12 **Abstract**

13 In this study a commercial particle analyzer was used to image and help sorting microplastic particles
14 (MPs) dispersed in filtrated and de-aerated tap water. The device provides a relatively easy and fast
15 procedure for obtaining ultra-high-definition imaging, allowing the determination of shape, size, and
16 number of 2D-projections of solid particles. The image analysis revealed clear differences among the
17 studied different MPs originating from the grinding of five common grades of plastic sheets as they
18 affect the image rendering differently, principally due to the light scattering either at the surface or in
19 the volume of the microplastics. The high-quality imaging of the device also allows the discrimination
20 of the microplastics from air bubbles with well-defined spherical shapes as well as to obtain an
21 estimate of the size of MPs in a snapshot. We associate the differences among the shapes of the
22 identified MPs in this study depending on the plastic type with known physical properties, such as
23 brittleness, crystallinity, or softness. Furthermore, as a novel method we exploit a parameter based
24 on the light intensity map from moving particles in cuvette flow to sort MPs from other particles,
25 such as, wood fiber, human hair, and air bubbles. Using the light intensity map, which is related to
26 the plastic-water refractive index ratio, the presence of microplastics in water can be revealed among
27 other particles, but not their specific plastic type.

28 **1. Introduction**

29 Microplastics (MP) in open water is currently a global problem affecting health, environment, and
30 economy [1-3]. The main obstacle to a solution to counter the spread of MPs is the lack of appropriate

31 devices to detect these particles in aquatic environment and in situ. The main reason comes from the
32 fact that MPs undergo different interactions with the environment. The complex shape and small size
33 of MPs makes their properties different from pristine ones, which leads to an inappropriateness of the
34 international standards for the definition of quality of macroscopic plastic products. A current
35 limitation on the identification and analysis of MPs is the need for sample preparation, which is also
36 required for some spectral measurement techniques, such as Fourier-transform infrared (FTIR) and
37 Raman spectroscopy [4-6]. Currently, emerging method to detect MPs utilize hyperspectral imaging
38 of dry MPs [7]. However, other methods such as digital holographic microscopy [8,9], laser speckle
39 pattern [10] and smartphone-enabled method [11] requiring neither sample treatment nor sorting have
40 shown promises for MP detection directly from water. Nevertheless, the water volume probed at once
41 by these devices remains rather small and not convenient for large field study. Similarly, other flow
42 field applications for microparticles such as flow cytometer [12,13] are impaired in the use of
43 unfiltered samples due to strict limitations in the flow cavity dimensions that are prone to blockages
44 in case the sample has a wide particle size distribution or even a few particles beyond or close to the
45 flow tube dimensions. There is an inevitable need for devices with larger volume for MP detection in
46 water and real-time and in situ applications.

47 Besides the shape and size, another factor contributing to the challenges in MP detection in water is
48 their appearance. Plastics can be transparent or hazy in water [14] and, by extension, MPs can exhibit
49 similar characteristics. Haziness from plastics can be due to surface roughening of originally
50 transparent (clear) plastic by mechanical wear, by chemical reaction, or due to refractive index
51 inhomogeneity in the volume of the plastic originating from external stress. In all cases, light is
52 scattered from the plastic, which causes translucent or opaque appearance of the sample. Both
53 transparency (ISO 13468-2, ASTM D1746-09) and haziness (ISO 14782: 2021 (en), ASTM D1003-
54 21) of plastics are commonly detected using a spectrophotometer. Information on ageing of
55 transparent or white plastic caused by an exposure to Sun light, moisture, temperature, and chemical
56 reactions, is also obtained by the so-called yellowness index (YI, ISO 17234:2014), which is extracted
57 from the spectrum of the plastic sample in the visible range of light. The concept of yellowness index
58 has been exploited, e.g., in the investigation of marine pellets [15].

59 However, the above figures of merit are based on optical measurements of plastics in air. In principle,
60 similar definitions and standards of plastic quality as above can be extended to describe macroscopic
61 plastics or MPs in aquatic environments, but the probed area of the sample has to be flat and there is
62 the issue of water absorption especially in the spectral region of near-infrared radiation. Measurement
63 conditions can be much more complex in real water systems, e.g., open, industrial, or waste waters,

64 than in laboratory-prepared samples due to the presence of other organic and inorganic materials than
65 plastics, and MPs having irregular and non-flat structures. Finally, temporal changes in the
66 measurement conditions and the lack of suitable measurement devices can often make difficult the
67 interpretation of the results.

68 Subjective visual inspection [16-18] has been one of the methods considered for the identifications
69 of MPs, despite a reduced reliability of MP detection due to possible human interference. Moreover,
70 for smaller MP sizes, in micron range, visual inspection becomes challenging [19,16]. Therefore, we
71 suggest, in this paper, the use of an imaging device, primarily developed for wood fibers and fines
72 analysis [20], as a promising tool to read various properties of MPs from a water sample volume
73 without sample pretreatment other than diluting microplastics containing water sample by tap water.
74 Using a novel concept of light intensity bitmap in the field of MP studies, it is possible to distinguish
75 unknown particles from other unknown inorganic or organic particles, that are simultaneously present
76 in the water sample to belong to the material group of plastics. The light intensity bitmap depends on
77 the refractive index ratio between plastics and water and depends on the wavelength of visible light
78 [21]. We demonstrate the viability of light intensity bitmap using homemade MPs in a water volume.
79 We note that other microscope-based techniques, such as binocular dissecting microscopes [22] and
80 near-infrared hyperspectral imaging [23], are alternative MP identification solutions, although they
81 are usually used on dry samples after pretreatment of samples [24].

82

83 **2. Materials and Methods**

84 The plastic samples used for the preparation of the artificial MPs are commercial sheets from
85 Goodfellow Ltd (UK). Studied MP samples include low-density polyethylene (LDPE), polystyrene
86 (PS), polypropylene (PP), polyamide (PA) and unplasticized polyvinyl chloride (UPVC). The MPs
87 were obtained by grinding plastic pieces with Retsch Ultra Centrifugal Mill ZM 200. Before grinding,
88 the plastic sheets were cut into small pieces under 1 cm² and hardened in liquid nitrogen for an easier
89 grinding. These pieces were then inserted into the mill in small amounts together with a spoonful of
90 liquid nitrogen to prevent the plastic from melting during grinding. Sieves with 1 mm- and 0.25 mm-
91 mesh sizes were used in sieving the resulting MPs during the grinding. The grinding speed was 10,000
92 rpm in the case of the 1 mm sieve and 6,000 rpm in the case of the 0.25 mm sieve, to avoid overheating
93 of the plastics. The grinding yields nano- and microplastics in the size range 100 nm- 250 μm and
94 these have different morphologies, which was confirmed by SEM analysis.

95 Valmet Fiber Image Analyzer (Valmet FS5) is an offline fiber and particle image analyzer for daily
96 laboratory or research use. Equipped, in this version, with an Ultra-High-Definition imaging unit
97 (UHD), it can be used for very tiny particle detection such as MPs. It was developed primarily for
98 fiber and particle analyses in pulp and paper industry. The device consists mainly of a sample
99 handling unit, an imaging unit and a touch screen user interface unit, all connected via computer. The
100 sample is diluted in tap water and is being pumped through the sample circulation loop. Consistency
101 of the sample can be set by hand or let the device adjust the consistency to the value set by user. The
102 sample flows through a flow cell in which particles are being illuminated and imaged with industrial
103 area scan monochrome camera. Imaging area is broad enough to measure large (10 mm) particles and
104 accurate for the observation of the smaller ones (minimum detectable particle dimension is in the
105 range of 1 μm). We exploit in this study snapshot images but it is also possible to record video from
106 the moving objects, e.g., MPs, and observe their changing appearance as a function of time as the
107 water flow propagates. Such a method is not the focus of this article; however, similar to
108 stereomicroscopic view, it would lead to three-dimensional representation of the objects within the
109 water sample and yield a better description of the immersed objects. The measurement of one 0.5 L
110 sample, volume chosen for this study, is based on the circulation measurement mode of the device
111 and it takes about 4 min depending on the sample type and settings chosen by the user. There is also
112 the option of non-circulation measurement. In such a case the sample volume (in principle infinite)
113 is not limited but it goes to waste. We preferred the circulation mode to preserve the MPs for future
114 use. One measurement typically can include hundreds of thousands of particles. In the case there were
115 only a single MP, of a size from 1 μm or larger in the water sample, it would be detected by the
116 imaging device once it passes the detection window. As a proof of concept each plastic type was
117 analyzed separately. The software calculates the morphological properties of the objects. Particle
118 length, width, area, and count are the most common properties reported by the device. In our case,
119 the analysis is done partly independently on the device's software since it must be taught beforehand
120 to recognize the different particle/object types. This could be realized with machine learning that has
121 been suggested, e.g., to automatize data handling in holographic imaging of MPs [25]. Yet, machine
122 learning is efficient also in such a case to monitor, e.g., 3D structures of transparent microscopic
123 objects [26] but would require some changes in the case of the detection setup to obtain interesting
124 3D appearance of transparent MPs. Note that the method described by Horisaki et al. can be used also
125 for opaque objects by detecting reflected light from an object.

126 The light intensity distribution of the imaged objects is obtained as a bitmap that is used in further
127 analysis of the MPs, as a novel method to screen MPs from other objects in flowing water. We

128 developed an additional analysis method enabling a better rendering of the captured images and an
129 improved contrast. This methodology described together with the results, for better clarity, is based
130 on the calculation of a spatial intensity quantity, M , defined as $M = \frac{I(x,y)}{I_0(x,y)}$. In this equation, $I(x,y)$ is
131 the intensity of the (x,y) pixel of one image and $I_0(x,y)$ the intensity at the same image pixel of a
132 reference (background) image, captured with no particle inside the measuring chamber. The
133 illumination and imaging system of the used device is certified to be highly stable, which allow us
134 this data processing. By definition, $0 \leq M \leq 1$. However, M may be greater than 1, depending on the
135 chemical structure of the particle and its inherent optical properties. Later, results are presented as 1
136 $- M$, for an even better visibility of the particles. The light intensity bitmap M depends on the
137 refractive index of the object and ambient medium, such as water. Wavelength-dependent refractive
138 index of a medium is an intrinsic material property that can be used for the identification of the object
139 similar to spectral fingerprints of a medium. Artificial MPs tend to have a rather homogenous spatial
140 refractive index, and hence this property can be used for sorting them from other particles in water.

141

142 **3. Results**

143 The pristine plastic samples of this study are transparent or translucent in air. Scattering of light from
144 such plastics immersed in water is much weaker than in the air. The scattering of light is strongest
145 when the size of the scatterer or its features, such as the surface roughness, is of the same order as
146 magnitude as the wavelength of the incident light. However, it is important to distinguish between
147 surface and volume scattering of an MP. While MP is embedded in water, the scattering from surface
148 roughness can be weak when the average surface roughness of MP is smaller than the effective
149 wavelength (in water) of the probing light. The strength of light scattering also depends on the
150 magnitude of the refractive index difference between the plastic sample and the ambient water. The
151 smaller the refractive index difference, the weaker the surface scattering resulting from
152 microroughness. In the case of volume scattering, due to volume inhomogeneities inside the MP, the
153 situation is different because water is not in contact with the internal parts of the MP. Therefore,
154 volume scattering is an intrinsic property of a particular MP. However, if the plastic is porous, then
155 water can penetrate the pores and hence influence the strength of volume scattering of light. Ageing
156 of plastic and the mechanical stress such as induced by our fabrication method, can affect both the
157 extrinsic and intrinsic properties of plastics, e.g., light scattering, which can similarly influence the
158 optical properties of the MPs.

159 We first deal with irregular sizes and shapes of MPs detected by the imaging tool and related peculiar
160 optical properties, such as, a lens formation or hole inside a MP. Figure 1 shows images of MPs in
161 water obtained for different types of MP. Figure 1a is an example of image obtained for PA MPs.
162 These MPs appear as dark and irregular contrary to observations in classical bright-field imaging. In
163 typical bright-field microscopy, transparent objects usually have low image contrast with its
164 background. Here, on the contrary, one can observe that the MPs, within this size range, show high
165 image contrast. This is certainly due to light scattering since PA is translucent, as stated above.

166 It is to be noted, in Fig. 1a, that many well-defined circular objects of various sizes are also present.
167 These are air bubbles formed by the stirring of water in the volume compartment with a propeller.
168 The biggest one is about 1.87 mm in diameter, while some smaller ones have diameters of 225 μm or
169 60 μm . The air bubbles appear dark because light is incident from denser medium, i.e., water
170 (refractive index, RI: $n = 1.33$), to air ($n = 1$) and the light rays can experience reflection at the
171 boundary, refraction across the boundaries, or total reflection. Additionally, the observed darkness
172 of the air bubbles can also be explained using the refractive index difference between the various
173 constituents. For example, the RI difference between water and air is larger than of the one between
174 water and the MPs. The theory of the influence of the RI difference on the appearance of the air
175 bubbles (dark), their radius-dependent optical properties, and other experiments on air bubbles in
176 water are explained in [27]. In principle, air bubbles can be used as reference particles with well-
177 known optical properties. Similar air bubbles can also be identified in the other studied images. More
178 detailed studies on the behavior of air bubbles in water can be found for instance in these two articles
179 [28,29].

180 The device's software has already an algorithm to neglect the air bubbles in the counting of the
181 identified particles in the analysis of an image. Note that this function was disabled for this current
182 study. In the practical case of MP imaging, omitting air bubbles simplifies the monitoring of particles
183 and their abundance in a water sample. It is also possible that a MP acts as a nucleation unit of air
184 bubbles. An example of it is shown in Fig. 1e, where several bubbles (half spheres) are on a smooth
185 (average surface roughness (measured with Mitutoyo SJ-210 profilometer, Japan) on both sides ca.
186 0.06 μm) hand-cut MP-film type of PET. As a side remark, bubbles can affect strongly the
187 conclusions drawn after optical detection of hydrophobic MPs by modifying the lift of the particles.
188 This may yield a confusion between a low- or high-density plastic if the air bubbles are not identified.
189 Since the setup uses a vertical flow it has practical importance regarding monitoring of air bubble
190 attachment on MPs, a method that has been suggested for the enrichment of microplastics in vertical
191 water flow [30]. We remark that according to the theory of optics, primary spherical MPs will yield

192 different light intensity bitmap from air bubbles because the refractive index contrast of water-air
193 bubble interface is much higher than that of water-primary MP interface, and there is no total
194 reflection present in the case of water-primary MP.

195 In Fig. 1b, several fragments of PP can be observed (circled in green and brown), which also appears
196 as a dark pattern due to light scattering like the other samples. Fig. 1c shows identified fragments of
197 LDPE. The larger fragment (highlighted in a red rectangle) is rather irregular, and a quantity such as
198 “aspect ratio” has little meaning in this case. Fig. 1d shows a zoomed-in image of the bigger LDPE
199 fragment. The interesting feature in the enlarged image is that, in the false color presentation of the
200 light intensity distribution, one can distinguish both dark (blueish) and bright (yellowish) locations in
201 the MP area. Very bright patterns correspond to transparent locations. After a detailed analysis of the
202 bitmap intensity of this MP, it turned out that some of the bright spots are much brighter than the
203 same spots without the MP, i.e., the background illumination in water only. Such bright spots (shown
204 by red arrows in Fig. 1d correspond to “lensing effect” due to a very thin portion of the MP from
205 these smaller irregular areas the MP that focus incident light. Such an effect can exist only if the front
206 and back of a part of the MP is optically smooth. This property is hardly present with organic particles.
207 The lensing effect is therefore an additional feature for the discrimination of MP against other
208 particles.

209 There are other less bright yellowish spots showing transparency or translucency, but darker areas
210 dominate over the whole MP surface in Fig. 1d that are due to light scattering and complex shape of
211 this MP. A narrow dark bridge is located in the bottom part of the MP, and it encloses a relatively
212 large bright area that is actually water (shown by the black arrow in Fig. 1d), i.e., a hole in the MP.
213 This MP is relatively large, and phenomena of geometrical optics apply for light interaction with the
214 MP. Obtaining MPs with very complex shape is one advantage of the grinding method used to
215 generate them. It creates MPs similar to the ones formed in natural environment with no predefined
216 shape, unlike other means, such as cutting leading to MP with high geometrical shapes, see Fig. 1e.

217 **Fig. 1f is the description of the entire procedure allowing the imaging and recognition of MP particles**
218 **in a water sample using the commercial device.**

219 Fig. 2 shows images of MPs obtained from originally transparent pristine plastics (PS and UPVC)
220 that have undergone similar preparation process as the translucent ones. Fig. 2a shows the image
221 (false color) for PS MP. Obviously, these scatter light like the MPs from the translucent MPs resulting
222 in a dark appearance in the image. Unlike in the previous samples, here, the grinding process of the
223 plastic sheet is the main reason for scattering of the incident light from roughened surface and possibly
224 also a volume inhomogeneity of the MPs. On the contrary, the MP obtained from UPVC, in Fig. 2b

225 (false color), shows a platy form with rather high transparency close to the middle. This large particle
226 is enlarged in Figs. 2c and d for closer examination. Its edges appear darker since light rays are
227 reflected and refracted and do not reach the detector. The contour of the MP (Fig. 2d) shows that
228 UPVC particle is an irregular and truncated pyramid-like block.

229 From images acquired by the ultra-high definition analyzer, one can easily isolate MP particle and
230 perform different analyses on the extracted image. The process is shown in Fig. 3 for the case of
231 LDPE sample. Fig. 3b is an isolated MP obtained by simply cropping the central part of the original
232 image (Fig. 3a) and removing the background (Fig. 3c). The image of the particle (the large LDPE
233 MP highlighted by a red rectangle in Fig. 1c) is a projection onto a plane and allow to estimate the
234 surface area of the considered MP by counting the number of black pixels (Fig. 3d). One can remark
235 that for high change in shape within the depth direction, such a data processing has a low significance,
236 and an estimation of the volume would be more relevant. However, for tiny particle as in most of
237 other presented cases, such a fast study, easily integrable into the software of the measurement device,
238 gives a realistic evaluation of the shape and size of the plastic particles.

239 Similar processing and image projection was applied to PA, PP, and LDPE MPs and are summarized
240 in Table 1. Concerning the number of particles, we counted the same amount (around 25) of MPs
241 made of PP, PS, and LDPE in the 0.5 L of water. We counted about twice less (10) for the MP made
242 of PA and only 3 particles for UPVC. These values are given only for illustration since the samples
243 were homemade and the amount of MP/L is not known. The main aim of this study was to
244 demonstrate the feasibility of imaging MP analysis by imaging them in tap water samples. The
245 number of counted particles is different in each sample because the value comes from the analysis of
246 one snapshot at a fixed time. However, such a figure indicates the possibilities offered by such a
247 technique in examining a relatively large volume of water. It is to be noted that the analysis of larger
248 sample volumes is also possible by using a continuous flow mode. Recognition of plastic particles
249 can be implemented directly in the software through machine learning processes to automate the
250 detection and counting process.

251 The shape of the MPs depends primarily on the plastic type. It is clear from Table 1 that MP from PP
252 and PA are quite similar in shape whereas the ones from UPVC, PS, and LDPE are completely
253 different. PS particles are more rounded than UPVC ones that are presenting clear facets and angles.
254 LDPE MPs are the one presenting the most random shape including fibers, as shown in Fig. 4,
255 apertures, and lens-type areas. The overall size distribution is relatively different also from a plastic
256 type to another. For instance, LDPE present both large and small MP types. PA MPs have areas
257 around 0.2 mm^2 , which corresponds to the grinding tool while PP MPs are about twice smaller. It is

258 to be noted that the projection area of particles is more descriptive but exact quantity would be the
259 volume of the MP which is not easy to access. This analysis can be used to anchored in the memory
260 of the measurement device the characteristic shapes of the different MPs and help the teaching
261 necessary for machine learning-based recognition.

262 Next, we consider the concept of light intensity bitmap analysis in more details. Although many MPs
263 illustrated in this study have a dark appearance, they actually transmit light. This is a property that
264 can be used in screening of MPs from wood fibers, human hair (quite often present in waste waters)
265 and air bubbles. We start with the concept of light intensity ratio (M). It is defined as the ratio of the
266 intensity of the transmitted light through an object in water to the intensity of light through water only
267 (background). This ratio depends usually on the location of image pixel over the object. We have
268 observed that a quantity $1 - M$ map is sensitive to screen between different objects in laminar water
269 flow system.

270 Before demonstrating the full power of $1 - M$ map, we wish to remark that human hair and wood
271 fibers (soft or hard) differ very much from the optical properties of a MP. Indeed, wood fibers have
272 complex optical properties because it consists mostly of cellulose (refractive index ca. 1.53,
273 crystalline cellulose is birefringent), hemicellulose (ca. 1.53) and lignin (ca. 1.61) [31], hence wood
274 fibers present volume light scattering. Similarly, human hair can be considered as an organic material
275 that has refractive index ca. 1.54.

276 In Fig. 5 is the $1 - M$ map of one snapshot captured by the high-resolution imaging device. By arrows,
277 we highlight a human hair, a PP MP, a wood fiber with a kink and an air bubble captured in the image.
278 The hair was introduced in sample of PP MPs and the wood fiber is due to previous measurement of
279 wood pulp sample in the device. These four particles are analyzed in detail in Fig. 6.

280 On the left hand-side are shown the $1 - M$ maps, in the middle and on the right hand-side panels are
281 $1 - M$ values taken from the locations shown by the colored arrows and lines on the $1 - M$ maps. It
282 is also to be noted that representing $1 - M$ in figures allows a better contrast, but the discussion is
283 done directly on the M -values. In the case of human hair, Fig. 6 a, M is oscillating along the length
284 of the hair, with a mean value $\bar{M} \approx 0.7$ and a standard deviation $\sigma \approx 0.1$. This oscillation in intensity
285 is due to light interference. On the contrary, M varies a lot obtained in the cross-direction but has
286 rather regular bell-shaped function following the shape of the hair itself. In Fig. 6 b is shown $1 - M$
287 for a soft wood fiber. In the location of the fiber between the two blue arrows M is fluctuating in an
288 irregular manner and relatively big difference appear in the magnitude of the M ($\bar{M} \approx 0.6$ and $\sigma \approx$
289 0.08). In the cross direction the M is changing strongly, and the distribution is not regular like in the

290 case of the human hair. The next case, Fig. 6c is the PP MP. Both in x - and y - directions $1 - M$ has
291 pretty much a constant value ($\bar{M} \approx 0.9$ and $\sigma \approx 1.5 \cdot 10^{-4}$) different from the cases of wood fiber
292 and human hair. In the inset are shown $1 - M$ for zoomed portions of this MP. Finally, in Fig. 6d, are
293 $1 - M$ data for an air bubble ($\bar{M} > 0.9$ and $\sigma \approx 1 \cdot 10^{-5}$). The data of air bubble show higher
294 symmetry and regularity than the MP. Surely the circular appearance of air bubble discriminates it
295 from this MP. However, one might think that a spherical primary MP could be mixed with an air
296 bubble. However, optical properties of an air bubble and primary spherical MPs are quite different.
297 Firstly, the refractive index ratio of water- air bubble is much higher than that of water -primary
298 spherical MP which has consequences both in light reflection and refraction in such objects.
299 Furthermore, a flowing air bubble may not be perfect sphere but can be flattened. Detection of primary
300 MPs is a topic of our future studies. This is enabled by the refractive index difference between the
301 media leading to variations in the light intensity bitmap.

302 In the case of a mixture of different plastic type MPs in water their presence can be revealed by the
303 light intensity map, but their specific plastic type cannot be identified. We think this sorting of
304 different MPs directly from water is at least a partial solution to avoid laborious laboratory studies
305 and would be a practical concept to investigate industrial and urban wastewaters taken, e.g., from
306 onsite pipelines. The device is portable and either single or double devices could be used before a
307 filter to pre-screen any presence of MPs and their images and characterization like done above, and
308 after the filter to monitor again presence of possible remaining MPs in water.

309

310 **4. Discussion**

311 This study reports on imaging of homemade MPs that have irregular shapes and structures. Irregular
312 MPs, such as fragments, are found in aquatic environments. Typical properties of interest of MPs are
313 their size, shape, abundance, and the type of plastic [32]. This information can be obtained by using
314 a several imaging techniques based on microscopies namely light, fluorescent, polarization, scanning
315 or transmission electron, atomic force microscopies [8,33], and hyperspectral imaging [34]. However,
316 these conventional devices require extensive pretreatment methods and are also time consuming as
317 well as limitation imposed the resolution of some of these devices. In most cases, visual inspection,
318 with it challenges, may precede these methods. For sorting of MPs from other materials, the
319 commercial imaging device is showing promising features for pre-screening of MPs directly from a
320 water sample. However, the data are not obtained continuously as a function of time because the
321 recorded video consists of image frames after snapshot illumination. Hence, there is a deadtime

322 between successive illumination and image capturing, and MPs can be flow pass without detection.
323 Nevertheless, each image frame can be studied later, like in this paper for observation of MPs and
324 their abundance. Such an abundance gives minimum estimate of MPs in a water sample. This would
325 give statistical results useful in industry, for instance. Improved data processing can be implemented
326 directly in the device's software and combined with automatic recognition of plastics particles. This
327 last step requires machine learning process to teach the device since plastics have some characteristic
328 shape, size, and optical features to be compared to other organic or inorganic particles.

329 The light intensity ratio is suggested as a measure to screen presence of a MP by using the regularity
330 of $1 - M$ obtained over a MP. Hence, it serves as a parameter that a MP has been found but it cannot
331 identify the plastic type. This finding of a MP is already important step regarding the laborious current
332 procedures in laboratories. Extensive identification of MPs would require measurement of spectral
333 properties from MPs, but there is currently no such measurement mode in the device. However, this
334 can be integrated in future. Alternatively, one can attach other modes to filter the measured water
335 volume to detect dry MPs using, e.g., portable reflectometer [35] or Raman spectrometer [36] from a
336 mixture of MPs. This would make identification of MPs possible in field conditions such as municipal
337 or industrial wastewater treatment plants. In addition, reliable and fast methods for MP analysis are
338 urgently needed for environmental monitoring. Driving forces include, e.g., international agreements
339 or policies such as EU Marine Strategy Framework Directive [37]. Currently, samples for monitoring
340 MPs are collected infrequently and results are obtained after laborious analysis in research
341 laboratories, and thus, relatively fast on-site analysis would be appreciated. Using demonstrated
342 imaging methods during a research cruise, large-scale monitoring campaigns would reveal local and
343 temporal changes rather than indicate only the abundance of MPs.

344 The results presented here based on UHD imaging brings the goal of field measurements of MPs
345 directly from water samples nearer. After further development, imaging methods for MP analysis
346 may be rather easily integrated to industrial process water or wastewater monitoring, because similar
347 techniques are already used, e.g., for fiber analysis. This requires further development of spectral
348 measurements to identify the MPs as well as image processing, e.g., to estimate real projection area
349 of a MP as we have demonstrated by bitmap study of present samples. Image data from MPs show
350 that it is possible to screen between transparent and translucent MPs.

351 The intensity ratio M , and therefore $1 - M$, varies significantly with the refractive index of the particle
352 and its overall optical properties. We also showed that these values can vary along preferential
353 directions with a relatively high contrast (proportional to the standard deviation of M) but with a quite
354 high mean value, this is the case of a hair. M values can be randomly fluctuating showing low mean

355 value and similar standard deviation than the hair, this is the case of a wood fiber. M values can be
356 very high and nearly constant for an air bubble. Finally, for a microplastic, M present high values,
357 still lower that for an air bubble and wood fiber, with low but not negligible fluctuations. Combining
358 conclusions on the statistics of M values and their distribution on several key-directions of the
359 particles, is a clear indicator of the particle type.

360 In case of a biofilm attached over MP it is usually an issue and typically requires removal in sample
361 preparation prior to MP characterization. In case of transparent biofilm, that consists mostly of water,
362 identification of MP by $1 - M$ should not be problematic, but this requires further experiments. In
363 case of a heterogenous opaque biofilm there are more challenges but the intensity ratio, if non- zero,
364 is a convolution of transmittance of MP and the biofilm. This too requires future experiments in trying
365 to identification of a MP beneath the opaque biofilm.

366 **5. Conclusion**

367 We demonstrate in this research that a commercially available image analyzer primarily designed for
368 the detection of wood fibers in aquatic samples can be efficiently used for MP detection. Using this
369 device, we have been able to determine the shape, size (projected surface), analyzed microplastics
370 prepared by grinding of five common plastic sheets namely, PA, PP, LDPE, PS, and UPVC. The
371 results show that the microplastics exhibit clear difference in terms of shapes and transparency, after
372 grinding, which arise from the mechanical properties of the plastic type and can, therefore, be used
373 as an indirect way to classify plastic types. The size of the particles differs also from one plastic to
374 another, despite using the same settings for the grinding tool. Combining the ultra-high-definition
375 imaging along with the light bitmap obtained from the device with automated machine learning would
376 lead to a complete device for MP recognition in wastewater treatment environment as the device can
377 already distinguish MPs, wood fibers, human hair, and air bubbles.

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380 research and innovation” (PREIN, 320166) and of Business Finland through the project on
381 “Circulating economy of water in industrial processes” (CEIWA, #541/31/2021).

382

383 **FIGURES AND TABLES**

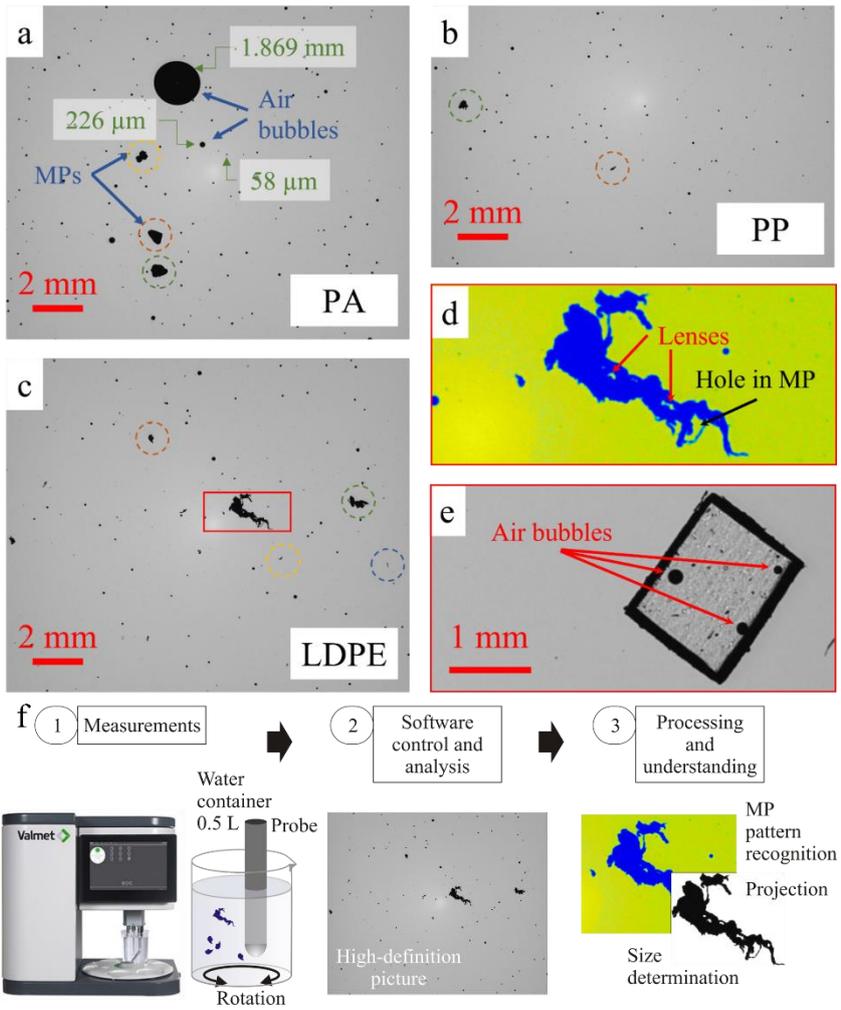
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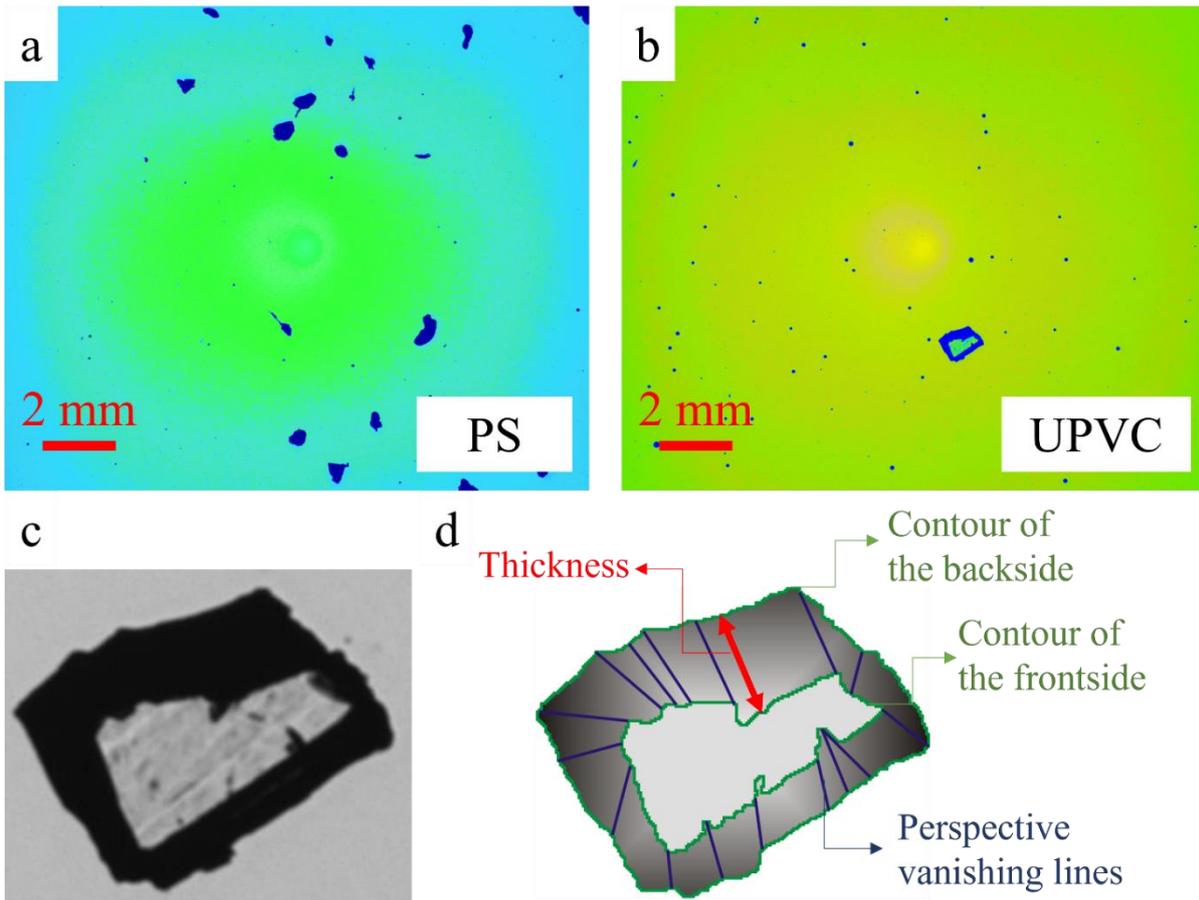
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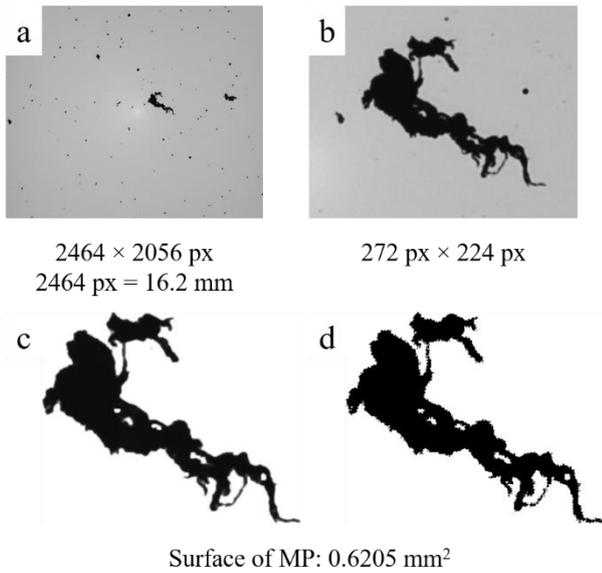
389
 390 *Figure 1: Images acquired with Valmet FS5 UHD device for different samples in water including a)*
 391 *PA particles, b) PP particles, and c) LDPE particles. d) zoomed-in and enhanced image of a*
 392 *microplastic particle from c) (red contour). Red arrows in d) highlight bright “lensing” areas in the*
 393 *microplastic particle. Black arrows in d) highlights a hole in the MP. e) air bubbles on a PET MP. f)*
 394 *Procedure to image and recognize MPs in a water sample.*

395
 396
 397



398
 399 *Figure 2: Processed images in order to highlight the different plastic microparticles a) PS and b)*
 400 *UPVC plastic particles. Highlight on UPVC particle: c) Original cropped image and d)*
 401 *contoured image showing an irregular-cut pyramid due to the grinding.*

402



403

404 *Figure 3: Isolating MP from the acquired images and extracting an estimation of the projected*
 405 *surface area of the particle. a) original image, b) cropped image, c) isolated MP, and d) projected*
 406 *image of the MP in black and white allowing an estimation of the size.*

407

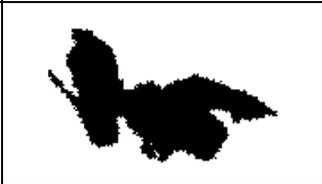
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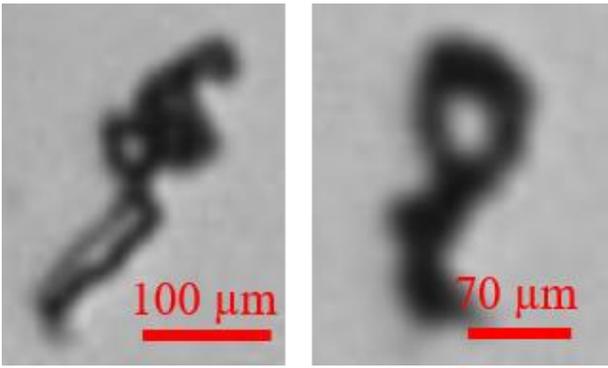
410

411 *Table 1: Selected MP extracted from Fig. 1 to estimate the surface area: Plastic types and*
 412 *corresponding figures from which the MP images have been extracted; isolated MPs without*
 413 *background; projected images for pixels counting; surface area of the MP.*

Plastic type / color of the dashed circle on Fig. 2a	Isolated image of MPs	Projected image	Surface area of the MP [mm ²]
PA / Green in Fig. 2a			0.252
PA / Brown in Fig. 2a			0.219
PA / Yellow in Fig. 2a			0.134
PP / Green in Fig. 2b			0.092
PP / Brown in Fig. 2b			0.016
LDPE / Red rectangle in Fig. 2c / Fig. 2d			0.621
LDPE / Yellow in Fig. 2c			0.007

LDPE / Blue in Fig. 2c			0.004
LDPE / Green in Fig. 2c			0.176
LDPE / Brown in Fig. 2c			0.048

414



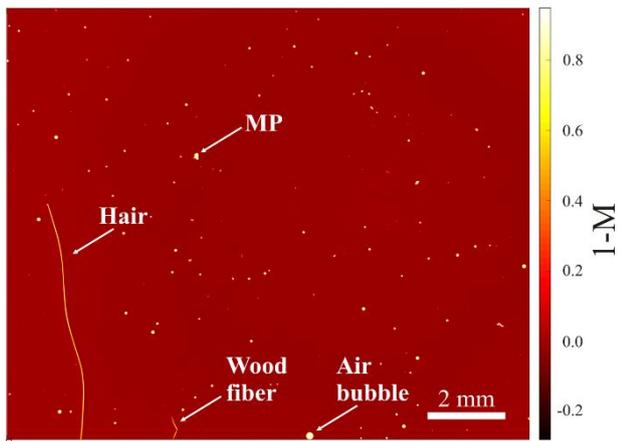
415

416 *Figure 4: Image of MP extracted from Fig. 2c (LDPE samples) showing opening, facets, or*
417 *"lenses" in the particles.*

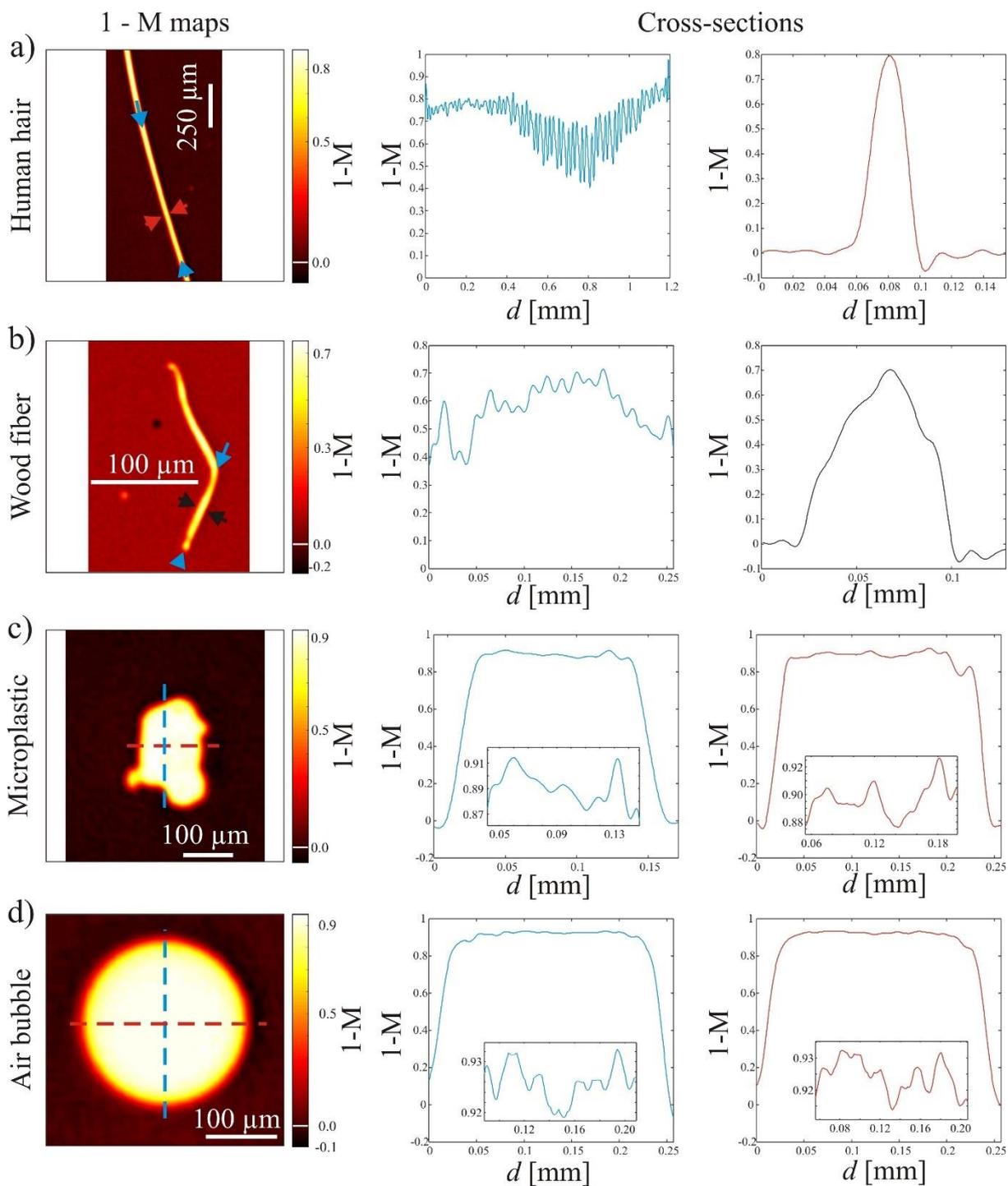
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421 *Figure 5: Example of a 1 – M map obtained from one snapshot captured by UHD FS5 device. It*
422 *shows a human hair, a kinked soft wood fiber, a PP microplastic and an air bubble. Each particle is*
423 *further analyzed in Fig. 6.*



424

425 *Figure 6: Intensity distribution analysis through $1 - M$ values for a) a human hair, b) a soft wood*
 426 *fiber, c) a microplastic, and d) an air bubble. Left: $1 - M$ maps. Middle and right: cross-section*
 427 *along the colored arrows or dashed lines shown on the $1 - M$ maps. Insets in c) and d) are zoom-in*
 428 *curves on the central part of the $1 - M$ cross-sections.*

429

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