Standardised protocol for monitoring microplastics in sediments

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Standardised protocol for monitoring microplastics in sediments

BASEMAN

MICROPLASTICS ANALYSES IN EUROPEAN WATERS

2018

WP4 Sampling methodologies for microplastics in the marine environment: standardisation, suitability and intercomparison

Deliverable 4.2
Standardised protocol for monitoring microplastics in sediments
31st May 2018

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Disclaimer: This document has been developed through a collaborative approach among partners of the Work Package 4 of the JPI-Oceans BASEMAN project. The document should be regarded as presenting an informal consensus position on best practice agreed by all partners. Nonetheless, the document does not necessarily represent the views of the wider JPI-Oceans consortium and/or the European Commission.

Executive Summary

Marine anthropogenic litter has long been recognised as an emerging pollutant of global concern. Its ubiquitous distribution and its direct and indirect impacts on aquatic ecosystems, marine fauna and local economies have been recently highlighted by several studies and international organisations around the world.

Although comprised of different materials, plastic constitutes the most abundant fraction reported in worldwide surveys, with percentages that are variable from region to region.

Among plastic materials, *microplastics* (herein MPs), represent a huge concern due to their impacts resulting from fragmentation under weathering conditions (e.g. solar radiation, water temperature and abrasion processes) and from their ability to adsorb persistent, bioaccumulative and toxic chemicals (PBTC) (e.g. polychlorinated biphenyls - PCBs, polycyclic aromatic hydrocarbons – PAHs) and trace elements (e.g. Cu, Zn, etc.).

In addition to these impacts, recent studies have also reported the potential for MPs to be easily mistaken as food particles and subsequently ingested by a wide range of organisms throughout the different environmental compartments (e.g. sediment, water, air).

Under the scope of the JPI-Oceans, BASEMAN is an international and interdisciplinary collaborative research project that aims to overcome the lack of standardised methodologies through a profound and detailed comparison and evaluation of all approaches from sampling to identification of MPs. The two overall goals of the project are firstly "The validation and harmonisation of analytical methods" which is indispensable for the second goal of "Identification and quantification of MPs". Based on these goals and with the overall aim of creating a standardised methodology to allow microplastics harmonised long-term monitoring in Europe, the BASEMAN project provides a set of recommended protocols to allow comparisons among studies. With this in mind, the protocols will focus on sampling, processing and analysis of MPs in samples from different environmental compartments, specifically addressing MPs from intertidal and subtidal sediments.

This protocol is aimed at improving sampling, processing and MPs data collection quality while also allowing comparison amongst different studies throughout Europe.

João Frias
31st May 2018

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Standardised protocol for monitoring microplastics in sediments

Introduction

Rationale

This protocol serves as a guide for sampling, processing, analysing and monitoring of microplastics within both intertidal and subtidal sediments, through proposing standardised methodologies.

Four main aspects are covered in this document which include:

(1) sample collection;

(2) laboratory processing;

(3) identification of microplastics; and

(4) recommended environmental variables to be recorded while sampling sediments.

Each section includes a list of the material and reagents needed to conduct each task, followed by the current best practice methodologies available (May 2018) which have been carried out for microplastic collection, processing and/or identification. The layout of the document is such that each section can be printed out separately, along with the respective forms (available in the appendix). It is envisaged that the methodologies shared for field and laboratory work will result in improved data comparability amongst surveyors with different levels of technical and/or knowledge backgrounds in this field.

Note: This document does not include any health and safety considerations in relation to sampling and laboratory procedures. Please carry out a risk assessment for all field and lab work activities and adhere to the health and safety guidelines at your institute or organisation.

Concepts and definitions

Marine anthropogenic litter

According to the Marine Strategy Framework Directive (MSFD – 2008/56/CE), marine anthropogenic litter is "*any persistent, manufactured or processed solid material discarded, disposed of or abandoned in the marine and coastal environment*". This definition *"consists of items that have been made or used by people and deliberately discarded or unintentionally lost into the sea or coastline including such materials transported into the marine environment from land by rivers, drainage or sewage systems, wind. For example, marine litter consists of plastics, wood, metals, glass, rubber, clothing, paper etc., but by far the most abundant and problematic are buoyant and persistent plastics.*"

Also, the MSFD definition does not take into consideration the range of sizes of marine anthropogenic litter, which authors such as Van Cauwenberghe *et al.,* 2015, have addressed. In this paper, size ranges are defined for microplastics (≤5mm), mesoplastics (>5mm and <2.5cm) and macroplastics (≥ 2.5cm) (see Table 1, pg. 7).

In addition to size, it is also important to consider how polymers are affected by degradation processes. Because plastics are formed by long polymeric chains, they experience slow rates of degradation, which allows them to persist in the environment for prolonged periods of time without substantial structural alterations. Nonetheless, plastic items can suffer degradation processes fostered by photo-, thermic-, chemical and mechanical degradation, resulting ultimately in the fragmentation of plastic litter into smaller pieces described as *microplastics*.

Microplastics

Despite the fact that the term *microplastics* has been in use since 2004 when Thompson *et al.* used it for the first time, there is still no clear definition that is broad enough to encompass all particles/items that might fit into this definition. This term was initially used to describe the accumulation of microscopic pieces of plastic in marine sediments and in the water column of European waters. In 2009, Arthur *et al.,* added an upper size limit to *microplastics*, with their definition of "*plastic particles smaller than 5mm*".

In 2011, the definition of *microplastics* was further refined by Cole *et al.,* which distinguished them according to origin into primary (produced to be of microscopic dimensions) or secondary (resulting from fragmentation and degradation processes at the environment).

Since then, several attempts have been made to create an all-inclusive definition that considers and includes physicochemical properties such as particle size, chemical composition and solubility in water, without much success (Verschoor, A.J., 2015).

In this report we define *microplastics* as any synthetic, solid particle[1] or polymeric matrix[2] which is insoluble in water, with a size range from 1 μm to 5 mm[3], of either primary or secondary origin.

Notwithstanding this definition, and based on the currently available methodologies to sample, characterise and identify microplastics, the recommended lower size limit for monitoring purposes, to sample microplastics in sediments, is **100 μm**.

[1] Synthetic particles of regular (e.g. beads, pellets) or irregular shape (e.g. fragments).
[2] Synthetic fibres either in their individual or composite forms.
[3] The size range hereby described takes into consideration the paper by Arthur *et al.,* 2009 for the upper size limit and the paper by Gigault *et al.,* 2018 for the lower size limit, where nanoplastics are defined as having a size range from 1nm – 1 μm.

#1 Collection

Material

Intertidal

1. Measuring tape
2. Pencils, Datasheet, Labels
3. Camera
4. GPS
5. Quadrats (30x30cm) (optional)
6. Metal shovel/spade
7. Glass jars or grip- or zip- lock plastic bags
8. Aluminium tray (large)
9. Permanent marker

Subtidal

10. Metal ruler
11. Camera
12. GPS
13. Glass jars or grip- or zip- lock plastic bags
14. Metal spoon
15. Metal corer (optional)
16. Stainless steel benthic grab with metal collection trays
17. Pencils, Labels, Datasheets
18. Metal forceps
19. Glass microfiber filters
20. Glass petri dishes
21. Syringe and suction tube

A - Intertidal sediment

Sampling

Beaches are dynamic systems with ever-changing conditions and sampling for microplastics should take into consideration that high tide lines are highly variable over relatively short periods of time. In order to account for this, monitoring surveys should be held, whenever possibly, once per season (spring, summer, autumn and winter).

To define the sampling area, mark out a 100m transect in width, parallel to the water edge (sea), using a measuring tape or similar (Fig.1) and take note of the GPS coordinates on each side of the transect (Fig 1., A and B). This transect will define the sampling area i.e. from the shoreline (low tide, light grey, AC1) to the above the strand line (accumulation zone, dark grey, AC2).

In Figure 1 the dark grey line represents the mean height of the spring tide line and the light grey line represents the low tide line. Please note that in many beaches this second tide line might not be always visible on the shore.

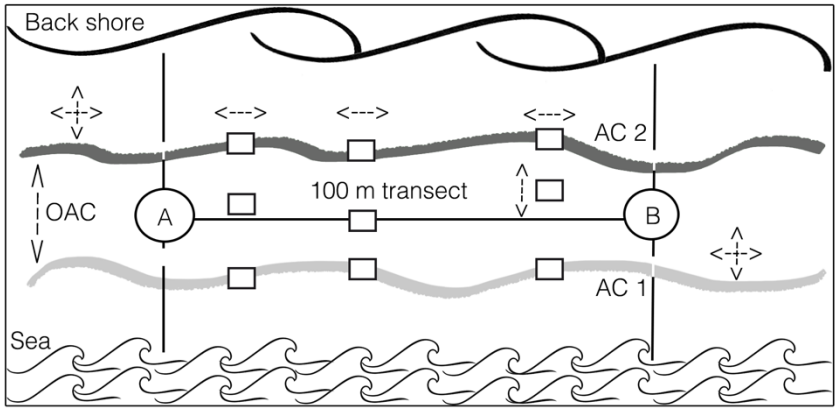


Figure 1 is a visual representative example of what could be draw in your datasheet.
The datasheet in the appendix provides an area for the surveyor to draw a representation of the beach on the day of the survey.

Figure 1 – Example of 100m transect (adapted from OSPAR, 2010 and NOAA, 2013)
(AC – accumulation area, OAC – outside accumulation area)

The sampling design is stratified random so collect a minimum of 3 samples (represented by square areas in Fig. 1) along a transect in each high tide line (AC1 and AC2 in Fig. 1). Make sure you also survey the area between the two high tide lines (OAC). Mark your sampling unit (**30 x 30 cm**) using the measuring tape or a quadrat and record the GPS coordinates of each unit.

Collect the top **5 cm** (total volume of approximately **4,500 cm³ = 4.5 L**) of sediment using a metal shovel or similar and ideally store the sample in labelled glass jars, previously decontaminated. Alternatively, they can be stored in labelled grip- or zip-lock bags. This method will allow to estimate concentration of microplastics both horizontally and vertically, allowing collected data to be compared with a wide range of studies. Datasheets for this task can be found in the appendix (Form 1).

B - Subtidal sediment

Sampling tools

There are four main types of subtidal sediment collection tools:

1. Grab samplers
2. Core samplers
3. Dredge samplers
4. Remotely Operated Vehicles

The two most common benthic samplers are Grabs and Corers, and the most common tools are, respectively the van Veen grab and the Box Corer. Either of these are good options, however, we recommend using a **box corer sampler** (e.g. Reineck box corer), as this sampling tool has minimal impact in surface deformation and maintains the sediment integrity which allow for the investigation of historical inputs of contaminants such as microplastics and will contribute to two descriptors (6 – Sea floor integrity and 10 – Marine litter) of the MSFD. In addition, this method allows an easier calculation of the volume of sediment collected.

Reduction of cross-contamination risks

Although it is extremely difficult to minimise contamination risks while sampling at sea, there are precautionary measures that can be taken into consideration:

- **Clean the area** – before assembling working areas, make sure that those areas are clean;
- **Daily controls** – insert glass microfiber filters (e.g. Whatman GF/C or similar) in petri dishes in the wet and dry laboratories of the research vessel to monitor airborne particles;
- **Controlled environment** – have a reduced number of people working on each task and record on your datasheet of the colour of the clothes each person is wearing underneath their protective gear and/or lab coat;
- **Procedural blanks** – run procedure blanks in parallel to sample processing. The procedural blanks will follow the same steps of the sample treatment, being the main difference the fact that they are run without the sample itself. Whenever possible, run at least a minimum of three (n=3) procedure blanks to validate data. Conduct this procedure every time you run a battery of samples.

Sediment collection and storage

The sampling design should follow a stratified random approach and being able to collect at least 6 samples per site (see datasheet in the appendix). Samples should be collected from different depths and sediment types[4], to allow comparison among studies. Please follow carefully all instructions to deploy and retrieve the sampling tool without compromising the sample, the tool or any crew member.

On retrieval of the benthic sampler you have the option to subsample your van Veen grab or box core using a metal corer (e.g. round metal pipe marked at 5 cm) or you can retrieve the surface sediments to 5 cm depth. Place the sample/subsample into labelled glass jars or grip- or zip- lock plastic bags, double bagged. If the core has water on the surface, there is also the option to carefully remove the supernatant water into a glass jar by using the syringe and suction tube. This should allow its collection without compromising the integrity of the surface of the sample.

If samples are not being processed immediately it is suggested that they are frozen and stored at -20ºC until further processing[5].

It is advised to collect associated environmental data (see #4) to the sediment sampling, in order to allow comparability between studies. Datasheets for this task can be found in the appendix (Form 2).

[4] Sediment type (see table A1 in the appendix) can highly influence the microplastic retention. In some cases, sediment requires to be pre-treated to remove organic matter (e.g. H_2O_2 at 6-10% solution).

[5] The goal of this monitoring is to assess microplastics, therefore please do not add any chemical reagent to the sample.

#2 Processing

Material needed

1. Acid-resistant plastic box with protective lid
2. Graduated cylinder
3. Petri dishes (glass)
4. Beakers (glass)
5. Metal forceps
6. Metal sieve 63 µm
7. Metal spoon
8. Aluminium tray (medium)
9. Filtration kit including vacuum pump
10. Glass microfiber filters
11. Desiccator

Reagents needed

Glass decontamination - Ensure all glass used is microplastic-free by using:
Nitric acid HNO_3 1% solution, CAS no. 7697-37-2 | Alternatively: intensive rinsing with filtered water and 1 µm filtered denatured alcohol, CAS no. 64-17-5. Glass needs to be rinsed with filtered or ultrapure water before use. Ideally, it should dry up-side-down in order for airborne microplastics not to accumulate in it.

Sediment Pre-treatment – to remove organic matter
Hydrogen peroxide (H_2O_2) 6-10% solution, CAS no. 7722-84-1

Density separation solutions
A list of density separation solutions is provided in **Table A2**, page 11 in the appendix.

Washing and rinsing
Ultrapure water | Alternatively: 1 µm filtered tap water or 1 µm filtered distilled water

Note: All solutions and rinsing liquids need to be filtered (1 µm) prior to use, apart from the ultrapure water, to reduce potential contamination. Please take extra care while preparing all the solutions and follow the health and safety guidelines according to your institute or organisation.

Density separation

From the wide range of polymers and density separation solutions described in literature, tables A2 and A3, page 11, compile the most common density separation solutions and the polymer densities, respectively. Table A2 is of particular relevance as it provides a safety-price index based on 3 major categories: 1) health hazard; 2) toxicity and 3) average price (€ per 250g). Health hazard details were retrieved from the Hazardous Materials Identification System (HMIS®) and from the National Fire Protection Association (NFPA) data forms; and toxicity details were retrieved from the Material Safety Data Sheet (MSDS) for each reagent. Average prices are the result individual quotes requested to different suppliers in Ireland, in March 2018. There is a wide range of prices within European countries, so please bear in mind that a specific reagent might actually be cheaper in your country.

The combination of these two tables will provide enough information to conduct a safe, accurate and precise decision-making process to asses microplastic contamination levels in sediment samples for both monitoring and scientific research purposes. Therefore, it is recommended to use either a saturated sodium chloride (NaCl – density: 1.2 g cm^{-3}) or sodium tungstate dihydrate ($Na_2WO_4 \cdot 2H_2O$ – density: 1.4 g cm^{-3}), as they are economical methods that allow polymers to float thus facilitating heavy density separation. For more details, particularly on the types of polymers that each will be able to suspend, please consult table A3. Please note that sodium chloride will not facilitate the separation of denser polymers such as polycarbonates (PC), polyurethanes (PU), polyethylene terephthalate (PET), polyvinyl chloride (PVC) or polytetrafluoroethylene (PTFE) (see Tables A2, A3 and A4).

In Table A3 the green line represents sodium chloride and the purple line represents sodium tungstate dihydrate. Theoretically this means that polymers above the lines are potentially positively buoyant in those density separation solutions. For more details check Table A4.

Cross-contamination risk reduction measures

- **No synthetics** - use a 100% cotton lab coat and avoid wearing synthetic clothes underneath the lab coat. Also, whenever possible, record the colour of the clothes worn underneath the lab coat as a precaution;
- **Daily controls** – insert glass microfiber filters (e.g. Whatman GF/C or similar) in a labelled open petri dish to monitor airborne particles. Air movement in the laboratory should be minimised by closing all windows and doors and by moving a slow pace;
- **Decontamination of glass material** – pre-clean all glassware before use;
- **Procedural blanks** – run procedure blanks in parallel to sample processing. The procedural blanks will follow the same steps of the sample treatment, being the main difference the fact that they are run without the sample itself. Whenever possible, run at least a minimum of three (n=3) procedure blanks to validate data. Conduct this procedure every time you run a battery of samples.

Intertidal and Subtidal sediments

Sediment pre-treatment

allows the recovery and reuse of density separation solutions

In order to recover the more expensive density separation solutions, sediments should follow a pre-treatment prior to density separation. For this, it is recommended to add a volume of 100 ml of a 6-10% H_2O_2 solution to the sediment, mixed with a metal spoon or a glass rod for 1 minute and allow to sit for 18h in a fume hood, covered with aluminium foil. The reaction will degrade organic matter, creating bubbles and foam. When the reaction stops, the sediment needs to be thoroughly washed with ultrapure water until it has no more bubbles, and then rinsed through a metal sieve with a mesh size of 63 μm[6].

Microplastic separation

If samples are stored in an aluminium tray, transfer each sample to individual decontaminated labelled 1L glass beaker and cover the beaker with aluminium foil. Make sure you run the procedure blanks alongside with sediment sample processing. Add the density separation solution, about three times the volume of sediment. Use the density separation method of your choice from table A2.

For monitoring purposes use either the sodium chloride or the sodium tungstate dihydrate solution. Stir and mix the sediment and the density separation solution using a metal spoon and allow it to settle for 1 hour. Filter the solution using a filtration kit with either inorganic membranes (e.g. Anodisc) or fibre glass filters (e.g. GF/C filters). After placing the solution into the filtration funnel, cover the top with aluminium foil. After introducing all the liquid from your jar, wash and rinse the walls of the filtration device with ultrapure water to ensure that all particles are recovered on the filter. Insert the filter into labelled petri dishes and dry in a closed glass desiccator, using silica gel or copper sulphate pentahydrate to enhance the drying process.

When fully dried observe under a stereomicroscope, a micro-FTIR, a micro-RAMAN or if microplastics are of considerable size, using directly an ATR-FTIR.

Datasheets for filter observation under a stereomicroscope can be found in the appendix (Form 3).

Note: for large sediment volumes (> 1L) another method that could be taken into consideration is the Munich Plastic Sediment Separator (MPSS) described by Imhof *et al.,* 2012 (for more details please see reference 7).

[6] After this step, you can either leave the sample in the hoven at 40ºC overnight, until is dry, or you can go directly to the microplastic separation procedure. This will depend on the number of samples which have to be processed.

#3 Identification

Relevant criteria to take in consideration during identification include physical properties (size, type, colour) and chemical properties. These criteria were established during a WP4 BASEMAN workshop held in Lisbon, Portugal, in September 2017.

Physical properties

Size classification

Despite the on-going debate regarding microplastics size classification, this criterion was based on a discussion held at the workshop and recent peer-reviewed publications. Table 1 describes the most common size categories for marine anthropogenic litter, including microplastics.

Table 1 – Size ranges of marine anthropogenic litter.
Adapted from Van Cauwenberghe *et al.,*2015 and Gigault *et al.,* 2018

Terminology	Size range
i. Macroplastics	>2.5 cm
ii. Mesoplastics	0.5 – ≤ 2.5 cm
iii. Large microplastics	1 – ≤5 mm
iv. Small microplastics	1 µm – ≤ 1000 µm
v. Nano plastics	1nm – ≤ 1 µm

At this moment, it is globally accepted that the lower size value for small microplastics (iv) is 1 µm (van Cauwenberghe *et al.,* 2015 and Gigault *et al.,* 2018). The current processing technology allows the user to identify particles smaller than 10 µm (µ-FTIR with an FPA detector) or down to 1-2 µm (µ-RAMAN). However, Micro-RAMAN is not be a recommendation here due to the increased amount of time needed to invest in sample processing.

Nonetheless, for monitoring purposes of microplastics in intertidal and subtidal sediment samples, the recommended lower size limit by BASEMAN WP4 is **100 µm**.

Type

This criterion is focuses on to the most common microplastic types described in peer-reviewed publications and the categories suggested are the following:

1. Pellet
2. Fragment
3. Fibre
4. Film
5. Rope and filaments
6. Microbeads
7. Sponge/foam
8. Rubber

Figure 2 illustrates six of the eight categories of microplastics commonly identified in visual identification. This figure does not include microbeads and rubber, as microplastics of these types are rarely recorded from environmental samples, despite being retrieved from zooplankton samples. The photographs below are simply illustrative of the different types of microplastics that can be found in environmental samples.

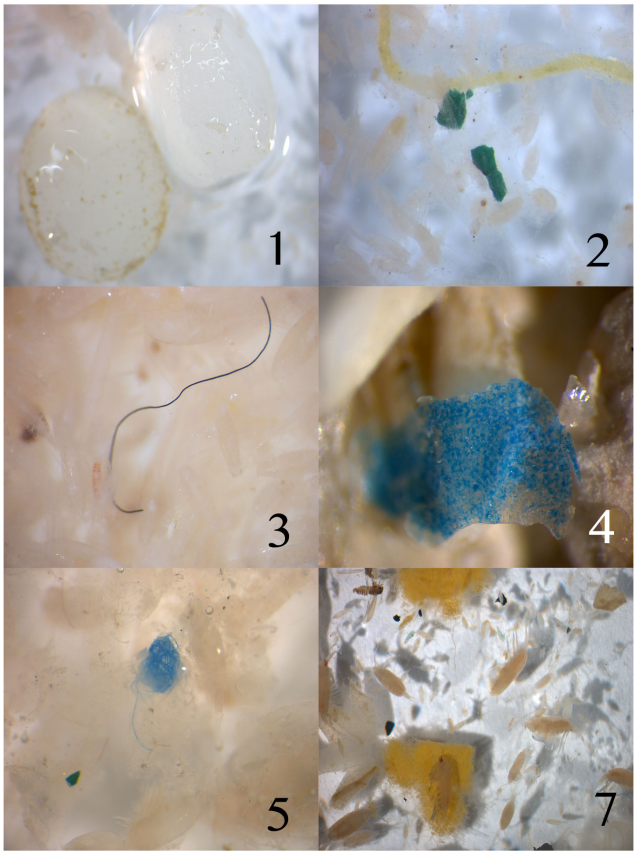


Figure 2 – Examples of microplastic types. Numbers correspond to previously mentioned categories, i.e. 1. Pellets; 2 Fragments; 3. Fibre; 4. Film; 5. Rope and filaments; 7. Sponge/foam. (credits: João Frias)

Colour

This criterion might be relevant to specific projects aiming to identify factors of geographical influence and/or impact on local marine species. In the workshop, this criterion was selected to be included in monitoring for its importance in local studies. The most common colours identified are described below:

1. Black ■
2. Blue ■
3. White □
4. Transparent □
5. Red ■
6. Green ■
7. Multicolour ■
8. Others ■ ■ ■ ■ ■

In this criterion, a separate class was attributed to multicolour, as it represents microplastics that have one colour on one side and another colour on the other side (e.g. neoprene particles), or rope or filaments that might contain more than one colour. The difference between white and transparent is the opacity - white is opaque and transparent is translucent. Colours such as purple, pink, grey, yellow or brown should be included in the category "Others", unless they have relevance for a specific project. Colour can cause some controversy, so in order to correctly assess this criterion, it is important to go beyond visual identification and use other spectroscopy techniques.

Note: while sampling on a research vessel, make sure to collect paint scrapings or other relevant samples from the vessel to serve as a colour control measure.

Chemical properties

Identification of polymer type and its elements can be done through the following techniques 1) micro-Fourier Transformed Infrared spectroscopy (μ-FTIR), 2) Attenuated Total Reflection Fourier Transformed Infrared spectroscopy (ATR-FTIR)[7], 3) micro-Raman spectroscopy (μ-RAMAN), and 4) Pyrolysis-gas chromatography-mass spectrometry (Py-GCMS), to name a few of the most common methodologies.

From the above-mentioned techniques, it is recommended to use micro-FTIR and ATR-FTIR[7], to clearly identify the polymer type.

It is important to note that Py-GCMS provides results in mass and not in number of particles, besides permanently destroying the sample. Therefore, it should be used as a complementary technique to microplastic processing. We recommend the use of micro-FTIR before using Py-GCMS, but in case where only one of these two techniques will be able to be conducted on the samples, we highly recommend the use of the micro-FTIR. In addition, the micro-RAMAN can also be a destructive technique, unless the excitement energy is low. This quantitative technique is highly time consuming and therefore not recommended for monitoring purposes.

Reporting results

Microplastics reporting units

Reporting units are the extremely important to allow comparison among studies. The proposed reporting units for microplastics retrieved from sediment samples are:

1. no. MPs per area (# particles km^{-2} | # particles m^{-2})
2. no. MPs per volume (# particles m^{-3})
3. no. MPs per mass (# particles kg^{-1} dry sediment)
4. mass of MP per area (g MP km^{-2} | g MP m^{-2})
5. mass of MP per volume (g MP L^{-1} | g MP cm^{-3})

Note: It is suggested and highly encouraged that authors, whenever possible, report results in all mentioned units.

Please also note that visual identification by itself **is not enough** and it **does not replace** ATR- or micro-FTIR. In fact, as state before, it is recommended to use these techniques in order to correctly identify polymer type and colour (potentially).

[7] For large microplastics and above (see table 1).

#4 Environmental variables

This section reflects on other environmental variables that might have an influence on the presence and microplastic densities in intertidal and subtidal sediments. We recommend data collection for the following environmental variables:

A. Intertidal Sediments

- Type of sediment, determined by Granulometry and %TOC
- Wind speed and direction
- Beach slope
- Approximate redox-potential discontinuity (aRPD) layer
- Amount of macro- and meso- marine anthropogenic litter
- Proximity to urban and/or industrial areas
- Proximity to river streams and/or estuaries
- Proximity to wastewater treatment plants
- Proximity to beach infrastructures (e.g. cafes, restaurants, nightclubs) and
- Behaviour of beach goers (e.g. dumping)

B. Subtidal Sediments

- Type of sediment, determined by Granulometry and %TOC
- Sea State; Wave height
- Collection depth, temperature and salinity
- Floating macro and mesoplastics
- Proximity to urban and/or industrial areas
- Proximity to river streams and/or estuaries
- Proximity to wastewater treatment plants (marine outfall)

Please note that surveyors do not have to collect data for all these environmental variables, as some of them depend on the specific goals of projects. The authors have provided in the appendix three forms[8] to collect data while sampling intertidal and subtidal sediments, as well as filter observation datasheets for the lab.

[8] This document provides an excel datasheet with the metadata proposed to collect while sampling.

#5 Appendix

Tables

Table A1 - Sediment grain sizes according to the Wentworth scale (Wentworth, 1922)

Aggregate name	Grain diameter
Boulder	> 256 mm
Cobble	64 – 256 mm
Very coarse gravel	32 – 64 mm
Coarse gravel	16 – 32 mm
Medium gravel	8 – 16 mm
Fine gravel	4 – 8 mm
Very fine gravel	2 – 4 mm
Very coarse sand	1 – 2 mm
Coarse sand	500 µm – 1 mm
Medium sand	250 –500 µm
Fine sand	125 – 250 µm
Very fine sand	62.5 – 125 µm
Silt	3.9 – 62.5 µm
Clay	0.98 – 3.9 µm
Colloid	< 0.98 µm

Table A2 – Common density separation solutions

Chemical formula	Reagent name	CAS no.	Density solution (g cm^{-3})	Health Hazard (Toxicity)*	Average price (€ per 250g) †	Safety-Price Index
NaCl	Sodium chloride	7647-14-5	1.0 – 1.2	1 (low)	€ (3)	■
$Na_2WO_4 \cdot 2H_2O$	Sodium tungstate dihydrate	10213-10-2	1.40	2 (low)	€ (70)	■
NaBr	Sodium bromide	7647-15-6	1.37-1.40	2 (low)	€ (3-5)§ €€€€ (430)§	■ ■
$3Na_2WO_4 \cdot 9WO_3 \cdot H_2O$	Sodium polytungstate	12141-67-2	1.40	2 (low)	€€€€ (276)	■
$Li_6(H_2W_{12}O_{40})$	Lithium metatungstate	127463-01-8	1.6	1 (moderate)	€€€€ (360)‡	■
$ZnCl_2$	Zinc chloride	7646-85-7	1.6 – 1.8	3 (high)	€ (45)	■
$ZnBr_2$	Zinc bromide	7699-45-8	1.71	2 (high)	€€€ (200)	■
NaI	Sodium iodide	7681-82-5	1.80	2 (moderate)	€€€ (130)	■

*Health hazard retrieved from NFPA/HMIS forms and toxicity values retrieved from MSDS forms; † quotes for Ireland dated from March 2018, **please note that price values may vary in other countries;** § The cost of Sodium bromide (NaBr) is one example of the price fluctuation between countries – in Germany is very cheap (■) and in Ireland is extremely expensive (■) which would drastically affect the Safety-Price index ‡Lithium metatungstate quotes only available for a volume of 250 ml.

Table A3 - Densities of common polymers (adapted from Enders et al., 2015)
Density limit using: ■ Sodium chloride and ■ Sodium tungstate dihydrate and all above 1.40 g cm^{-3}

Abbreviation	Polymer	CAS no.	Density (g cm^{-3})
PS	Polystyrene	9003-53-6	0.01 – 1.06
PP	Polypropylene	9003-07-0	0.85 – 0.92
LDPE	Low-density polyethylene	9002-88-4	0.89 – 0.93
EVA	Ethylene Vinyl Acetate	24937-78-8	0.94 – 0.95
HPDE	High-density polyethylene	9002-88-4	0.94 – 0.98
PA	Polyamide	63428-84-2	1.12 – 1.15
PA 6,6	Nylon 6,6	32131-17-2	1.13 – 1.15
PMMA	Poly methyl methacrylate	9011-14-7	1.16 – 1.20
PC	Polycarbonate	25037-45-0	1.20 – 1.22
PU	Polyurethane	9009-54-5	1.20 – 1.26
PET	Polyethylene terephthalate	25038-59-9	1.38 – 1.41
PVC	Polyvinyl chloride	9002-86-2	1.38 – 1.41
PTFE	Polytetrafluoroethylene	9002-84-0	2.10 – 2.30

Polymers until the marked lines are retained by the solutions. Please note that this is a theoretical model and some polymers with higher densities could potentially be found in sediments even using a solution with density lower to 1.40 g cm^{-3}.

Table A4 – Buoyancy of common polymers (adapted from Crawford and Quinn, 2017)

Abbreviation	Polymer	Density (g cm^{-3})	Buoyancy
PS	Polystyrene	0.01 – 1.06	Positive (↑)
PP	Polypropylene	0.85 – 0.92	Positive (↑)
LDPE	Low-density polyethylene	0.89 – 0.93	Positive (↑)
HPDE	High-density polyethylene	0.94 – 0.98	Positive (↑)
Seawater		**1.025**	
PA	Polyamide	1.12 – 1.15	Negative (↓)
PA 6,6	Nylon 6,6	1.13 – 1.15	Negative (↓)
PMMA	Poly methyl methacrylate	1.16 – 1.20	Negative (↓)
PC	Polycarbonate	1.20 – 1.22	Negative (↓)
PU	Polyurethane	1.20 – 1.26	Negative (↓)
PET	Polyethylene terephthalate	1.38 – 1.41	Negative (↓)
PVC	Polyvinyl chloride	1.38 – 1.41	Negative (↓)
PTFE	Polytetrafluoroethylene	2.10 – 2.30	Negative (↓)

Polymer density might vary with additives added during production, and therefore this table is a theoretical model.

Forms

1. Intertidal sediment sampling datasheet

Country_____ **Location area** _____

Sampling site code _____ **Beach name**_____

Date: _____ **/** _____ **/** _____ (dd/mm/yyyy) **Sampling season:**_____

Start time: _____:_____ AM | PM **End time:** _____:_____ AM | PM

Beach Characteristics

Slope: _____(degrees) **Beach length**: _____ (m)

Beach substrate: ☐ Sand ☐ Gravel ☐ Boulder. ☐ Other_____

Dune Substrate: ☐ Sand ☐ Gravel ☐ Boulder. ☐ Other_____

Atmospheric conditions: ☐ Wind ☐ Rain ☐ Waves (strong, moderate, low): _____

Did any of the following atmospheric conditions affect OR prevent the sampling on this day?

☐ Storm | hurricane ☐ Ice | Frost ☐ Fog | Smog ☐ Dust- or sand-storm ☐ Waves exceptionally high

GPS coordinates:

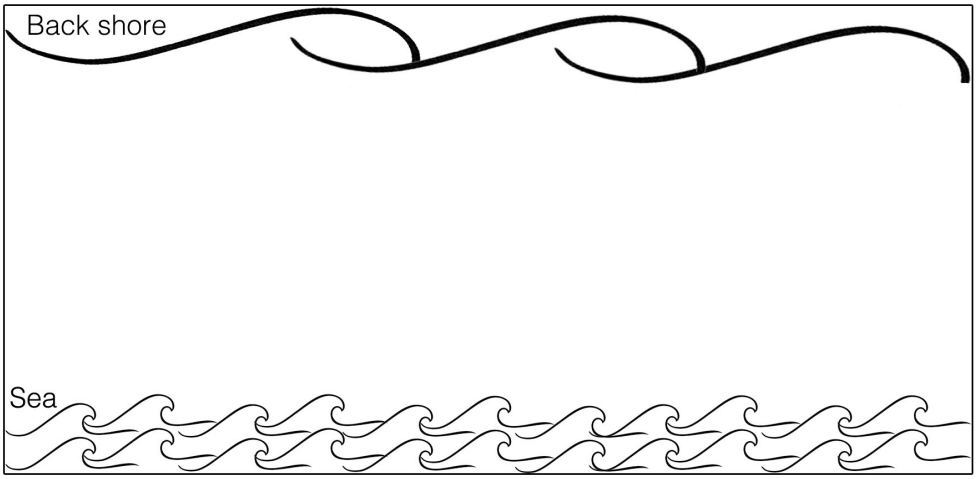
A _____ **B**_____

AC1 1. _____ 2. _____ 3. _____

AC2 1. _____ 2. _____ 3. _____

OAC 1. _____ 2. _____ 3. _____

SAMPLING



1. Draw the high tide lines representing the main accumulation areas (AC1 and AC2);

2. Mark starting point A and finish point B. These should have 100m of distance between them;

3. Draw the squares where sampling was conducted. (For example see Fig. 1).

Comments/Notes:_____

_____

_____

_____

_____

_____

_____

1. Intertidal sediment sampling datasheet

Beach use: ☐ Urban ☐ Rural ☐ Other: _____

Proximity to | Presence of ☐ Industry ☐ Tourist Attractions (e.g. surf schools) ☐ Rivers ☐ Sewage inputs ☐ Harbours/Ports ☐ Fishing facilities ☐ Marina

Factors that might influence the presence of marine anthropogenic litter:

☐ Beach Clean-up activity: Event locally organised by ☐ municipality ☐ civic movement/NGO.

How frequently is this beach cleaned? ☐ Annually ☐ Quarterly ☐ Bimonthly ☐ Monthly ☐ Weekly ☐ Daily

☐ Recent storms or extreme events. Which? _____

☐ Festivals or other events. Which? _____

Impacts on marine fauna

Did you find dead animals? ☐ Yes ☐ No. How many: _____

Which species?

#1 _____ #3 _____

#2 _____ #4 _____

Were the dead animals entangled in marine litter? ☐ Yes ☐ No. How many: _____

Please provide more details on the entanglement:

#1 _____ #3 _____

#2 _____ #4 _____

Name of surveyor _____

Contact (e-mail): _____@_____

Was marine litter collected in this activity? ☐ Yes ☐ No

Notes:_____

_____

_____

_____

_____

_____

2. Subtidal sediment sampling datasheet

Country/Campaign_____ **R/V Vessel** _____

Station name _____ **Sampling site code** _____

Date: _____ **/** _____ **/** _____ (dd/mm/yyyy) **Sampling season:**_____

_

Start time: _____**:**_____ AM | PM **End time:** _____**:**_____ AM | PM

Latitude _____ **Longitude** _____

Sampling tool _____ **brand and/or specifications** _____

Wind Speed_____ (knots) **Wind Direction**_____ (degrees)

Sea State (Douglas scale) _____ (0-9) or **Sea State (Beaufort scale)** _____ (0-12)

Station characteristics

Depth: _____(m)

Bottom Temperature: _____(ºC) **pH**: _____

Bottom Oxygen: _____ **Bottom Salinity**: _____

Was a CTD profile conducted in this station? ☐ Yes ☐ No

Was surface water collected from the core | grab? ☐ Yes ☐ No

If yes, take note the sample code(s) _____

	Sample code	**Thickness** (cm)	**Sediment description** (colour, smell, surface texture)	**Photo**
Replicate 1				
Replicate 2				
Replicate 3				
Replicate 4				
Replicate 5				
Replicate 6				

Comments:_____

_____

_____

_____

2. Subtidal sediment sampling datasheet

Name of surveyor _____

Contact (e-mail): _____@_____

Notes_____

_____

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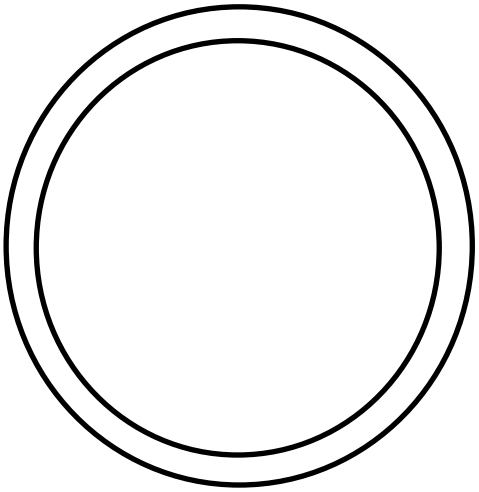
3. Filter observation datasheet

Date: ____/____/20___

Sample code _____ Filter no. _____

Date of collection: ____/____/20___ Magnification _____ x



_____

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_____

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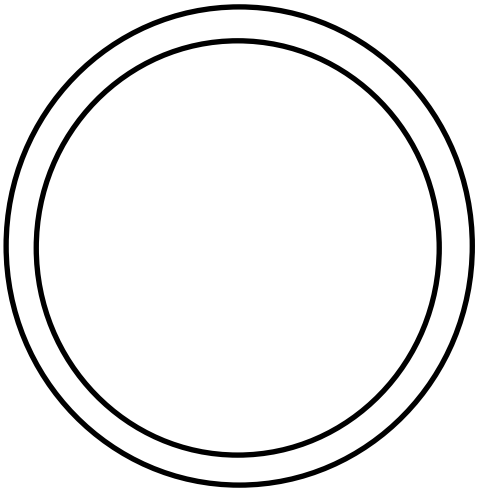
_____

Notes: _____

_____

Sample code _____ Filter no. _____

Date of collection: ____/____/20___ Magnification _____ x



_____

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Notes: _____

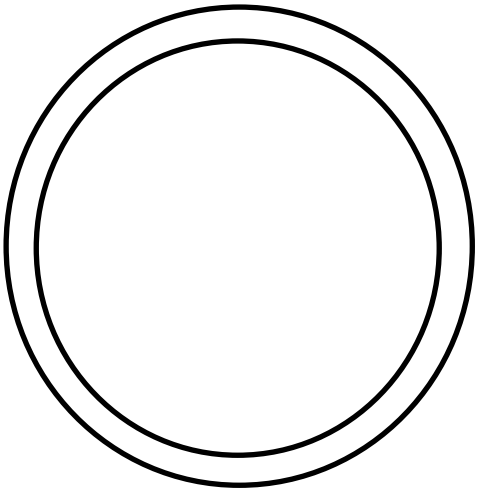
_____

3. Filter observation datasheet

Date: ____/____/20___

Sample code _____ Filter no. _____

Date of collection: ____/____/20___ Magnification _____ x

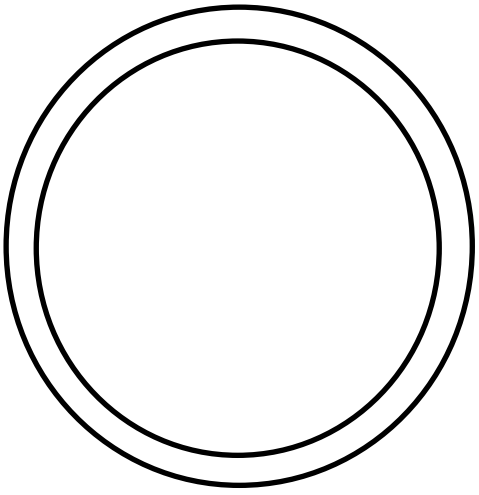


Notes: _____

_____

Sample code _____ Filter no. _____

Date of collection: ____/____/20___ Magnification _____ x



Notes: _____

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