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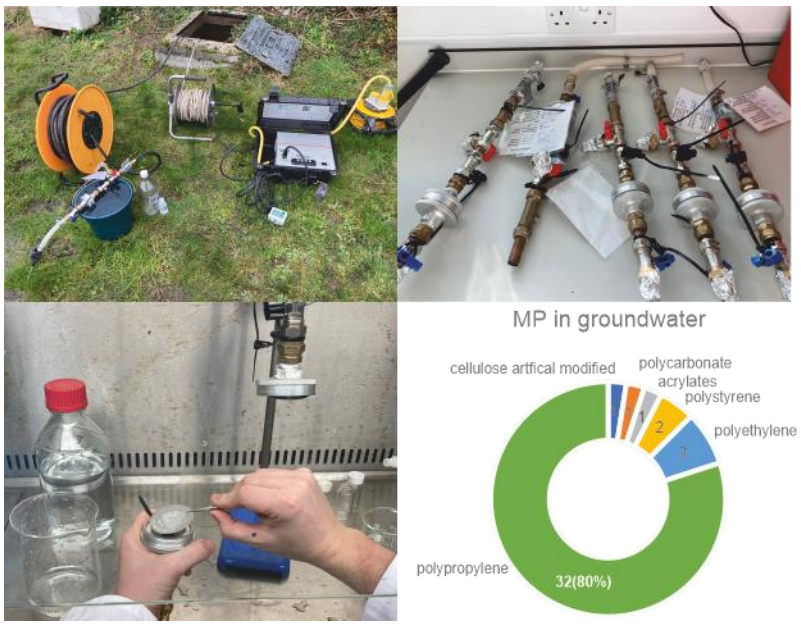
Microplastics in UK groundwater and stygobites: protocols for sampling, analysis and pilot study results

Environmental Change, Adaptation & Resilience Programme

Open Report OR/22/015



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Summary

A protocol for groundwater and stygobite microplastic (MP) sampling and analysis was developed and tested based on adapting established methods for drinking water sources. This study was undertaken to develop a protocol for groundwater that could be used for wider assessments of MP in groundwater sources in the UK and provide early evidence on the type and numbers of MP in groundwater sources and recommendations for future work on this topic. A total of 11 groundwater samples from 8 groundwater sources and a method blank were collected, processed and analysed for MP by Fourier Transform Infrared Spectroscopy (FTIR). Two stygobite samples and a method blank were also sampled and processed by FTIR. A total of nine MP compositions were analysed for by FTIR. Overall low numbers of MPs were detected in groundwaters, with the highest numbers being detected from pumped sources within the Thames gravels (up to 18 MPs) compared to the pumped Chalk (up to 5 MPs). After blank correction, a total of 40 MP particles were found across all samples using FTIR. A single polypropylene (PP) particle was found in the method blank. Overall, a very small proportion of the particles detected by FTIR were microplastics. Of those detected as MP the largest detected was 183 μm, 28% of particles were <50 μm, 78% <100 μm and 88% <150 μm. PP dominated the polymer composition of MPs found in samples collected from boreholes (80%), four other polymers were also detected including polyethylene (PE - 8%), polystyrene (PS - 5%) and acrylate, polycarbonates and artificial cellulose all ≤ 3%. No MP were detected above the method blank for stygobites samples. Two PP MPs were detected in the stygobite method blank and two PP MPs detected in one of the stygobite samples. The methodologies developed and tested are described in detail in this report and were adapted from existing methods previously used to sample treated drinking water sources and are highly suited to sampling pumped groundwater sources with low turbidity for MPs. The method was suitable for collecting large groundwaters samples and it was possible to filter up to 100 L in the field from the majority of sources within a relatively short time period (i.e. 1h). It would be possible to filter 2 or 3 times this volume in the field from many sites without the filter clogging, based on the small sub-sample used in this pilot study.

1 Introduction

There is currently no systematic monitoring of microplastics (MP) in UK groundwater or in sentinel groundwater ecology (e.g. stygobites), and no documented/standardised protocols that have been field-tested on untreated groundwater sources. The specific aim of this project was to develop standardised sampling and analysis protocols for microplastic (MP) particles in groundwater and groundwater stygobites (i.e. Niphargids). These protocols were then developed and tested at a small number of sampling sites in England as part of a pilot study, with a focus on sites used for drinking water supply. A separate BGS Open Report (Shockley and Lapworth (2022), OR-22-014), comprised a short global literature review on microplastics (MP) in groundwater, another output from this project. This review highlighted the limited work currently published on MPs in groundwater and the urgent need to develop standardised sampling and analytical protocols to assess MP in groundwater.

Prior to the start of the project a knowledge exchange workshop was held on 2 December 2021, with the purpose of bringing together and developing an informal network of colleagues working on MP research across CEH/BGS/Roehampton University/Birmingham University/Royal Holloway EA/Defra/DWI. The aim was to link together current projects on MPs, determine what has already been done to i) avoid duplication, ii) gather information to help develop a single protocol for groundwater sampling and analysis and iii) assess opportunities for co-location of research sites where possible and inform and feed into the outcomes of this project.

2 Methods

This section of the report covers considerations for the development of a sampling and analytical protocol for assessing MP contamination in groundwater and dependent ecology, specifically Niphargids, which are found in many karst groundwater systems in the UK and worldwide (Roberson et al., 2009). This was informed by the current literature, summarised by Shockley and Lapworth (2022) as well as by selected protocols that are currently used for surface water and drinking water MP assessments by UKCEH and others which analyse MP using Fourier Transform Infrared spec. (FTIR). We took the opportunity of the Groundwater MP workshop in December 2021 to ask a range of researchers about the different MP sampling and analytical methods currently being employed in the UK for assessing MPs in surface waters.

2.1 DEVELOPMENT OF A SAMPLING PROTOCOL FOR MP IN GROUNDWATER SYSTEMS

Based on a review of the literature (Shockley and Lapworth 2022) and discussions as part of the workshop in December 2021 two principal methods have been identified for isolating groundwater MPs. The first method, for example reported in Johnson et al. (2020) uses in-line woven steel filters to isolate MP from waters. These can be of varying sizes and surface areas depending on the application and anticipated turbidity of the waters in question. For low turbidity waters, such as processed drinking waters (Johnson et al. 2020), or perhaps groundwaters, lower surface area (45 mm diameter) filters may be a suitable option. These in-line filter systems are potentially highly suited for pumped groundwater samples, where the pressure from the pump allows large volumes of water to be filtered in relatively short timescale.

The second method, currently being trialled by colleagues at University of Roehampton and University of Birmingham, as part of the *PlaStyx* project involves collecting groundwater in a suitable storage container and then passing this through larger diameter steel mesh sieves (c.100 mm diameter, using 123 and 25 micron mesh) by sequential gravity filtration. Figure 1 shows examples of these two methods. The pros and cons of these two methods are summarised below in Table 1.

Figure 1 In-line sampler (A) and sieve mesh set-up (B). Filtering a groundwater sample using the in-line filter system and preparing a field blank putting MP filtered DI water through the stacked sieve system.

Table 1 Comparison of the pros and cons of the two different sampling methods

Method 1: In-line filtration using a sealed filter holder and rig (45 mm diameter)	
Pros	Cons
• Filter/filter holder preparation prior to sampling can be highly controlled • There is minimal risk of contamination from airborne MP prior to/during sampling • Post sampling handling can also be highly controlled minimising MP contamination • Smaller pore size cut-offs can be used due to additional backpressure applied by the pump • Highly suited for sampling sources where there is an in-situ pump/tap to connect filter rig to • Filtering time/volumes can be modified by flow and filter size • Rigs are re-usable • Sampling potentially easily compatible with other sampling protocols that require a pumped sample • Sequencing filters is possible	• Cost in assembling the filter rig • Considerations needed to minimise contamination from the rig itself • Need for a pump to draw water from the groundwater source • Weight/cost of the rig potentially limits the number of samples that can be collected using this technique • For highly turbid groundwaters small inline filters (45 mm) may become blocked quickly in the field reducing the volume of water that can be filtered
Method 2: Open filtration using large diameter sieves	
Pros	Cons
• Cost for collecting samples are lower due to simple sampling rig design • Does not rely on the use of a pump • Less involved preparation of rig needed prior to sampling • Easier to use and can be easily explained to non-experts • Sequencing filters is easy to do using this method • Can be easily deployed to a larger number of samplers at the same time e.g. for a large campaign • Larger volumes can be filtered within a reasonable timeframe	• Much higher risk of contamination prior and during sampling – e.g. from clothing • Requirement for filter handling in non-sterile conditions after sampling • Need for large MP free sample container and lids to minimise MP contamination in the field • The requirement for transfer of water sample to and from containers again risks introducing MP contamination in the field

There are a number of considerations when designing a protocol for sampling MP in groundwater and MP in stygobites. Some of these are listed below:

- How to minimise contamination from clothing and local environment when sampling?
- The need to ideally sample directly from the well and minimise contact with fixed tubing/storage containers used at some sites (e.g. private/public sources)
- What pump to use to minimise contamination by MP of interest?
- What connectors to use to minimise MP contamination?

- What type of filter holders and mesh/filters to use?
- What tubing to use to minimise contamination by MP of interest?
- Do we sample in duplicate to look at reproducibility?
- What materials to use for the sampling water - rig, filter holder etc?
- What materials to use for stygobite sampling?
- What methods to use to get a representative sample of stygobites?
- How much water can be filtered within a reasonable timeframe to obtain a representative sample?
- Can the method be scaled up easily for larger studies in the future?
- How comparable is the method to others being used in the UK?
- How to prepare a realistic field/full method blank?

Directed by the considerations listed above and based on an initial assessment of the risks of contamination from the sieve filtration method (Table 1) and the availability of sampling rigs for the in-line filter method we decided to initially trial the in-line filter method as part of this groundwater pilot study. We followed a similar approach to that described by Johnson et al. (2020) in terms of the method used to filter and isolate MPs and used the same filter holders and filters. There are clearly some limitations to using this method compared to the sieve method (see Table 1), but overall this was considered to be the current best available method, particularly with regard to minimising contamination risks. This is a major consideration for groundwaters due to the relatively low background level of contamination anticipated in groundwaters compared to rivers, for example. The use of these rigs on other DEFRA and UKWIR funded projects for surface water and drinking water sources also allows for better direct comparison with results from other studies looking at MP occurrence in UK waters. We also plan to trial the sieve method and compare results through paired sampling at a subset of sites, but this is beyond the scope of this project and is not covered in this report.

For consistency, we decided to standardise the stygobite sampling method for MP with the standard net methods for collecting stygobites (e.g. McInerney et al., 2014). This gives a representative stygobite sample, but the drawback is that the method does use a small nylon net to trawl for stygobites and also disturbs the sediment at the bottom of the well prior to trawling. Both of these factors are potential sources of MP contamination and the latter may also affect turbidity. However, these sources of contamination were mitigated by making sure in the subsequent sample processing step that stygobites were thoroughly rinsed with MP free water and ethanol prior to digestion for MPs (details in section 2.3). A method blank was also prepared alongside the samples to quantify any contamination from reagents and handling steps. In the future it might be possible to use natural fibre nets or metal nets which would reduce contamination during sampling. However, contamination during sample processing is probably the larger source of contamination for assessing MP in stygobites.

Duplicate sampling is a good practice to understand the error in the method used to assess MP in samples. Due to time limitations for sample processing and analysis not all samples were taken in duplicate for the pilot study. A subset of sites (n=3) were sampled in duplicate for this pilot study to assess reproducibility.

2.2 SAMPLING FOR MP IN GROUNDWATERS AND STYGOBITES

The generalised workflow for sampling for both MP in groundwater and MP in stygobites are presented in Figure 2.

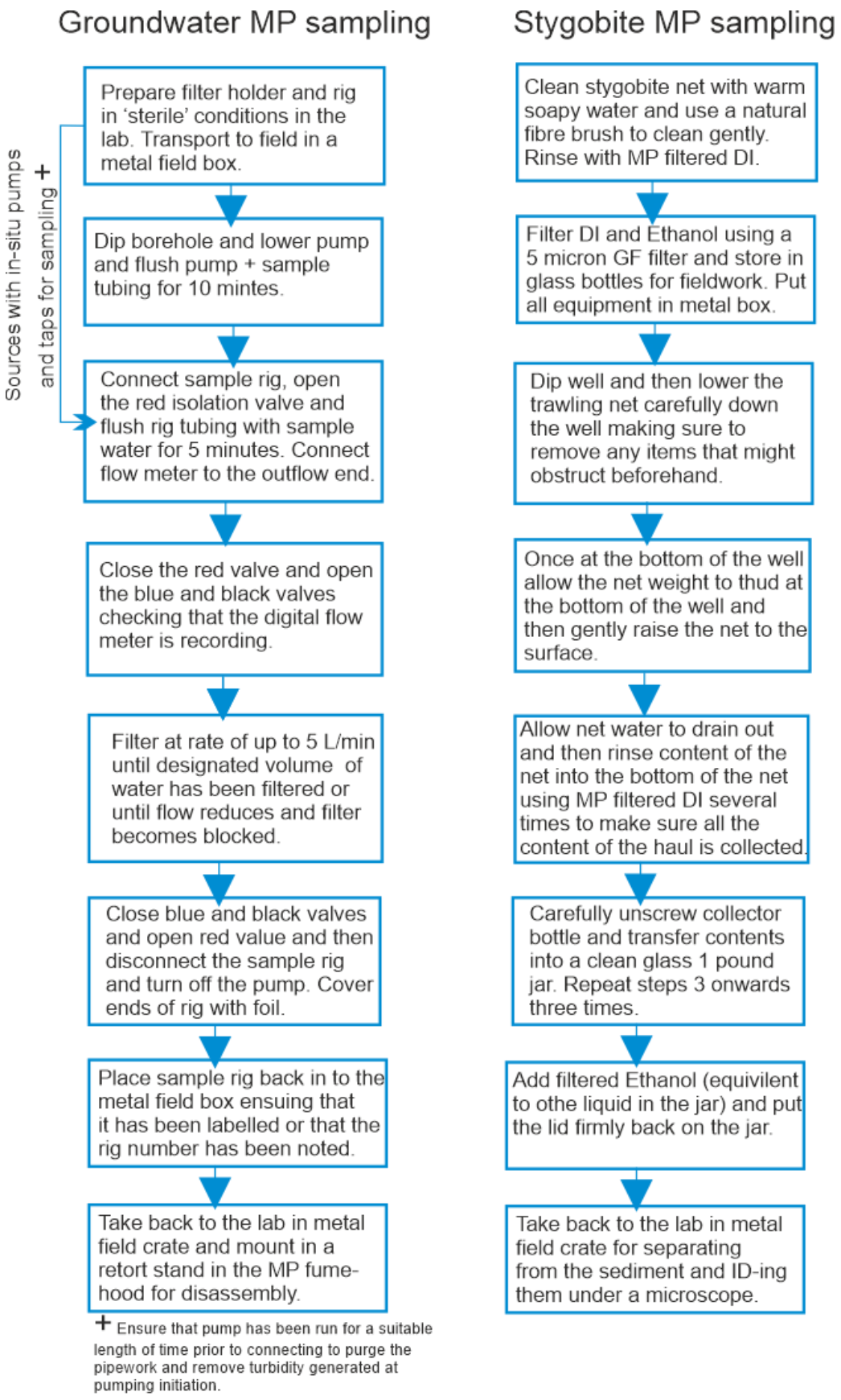

Figure 2 Generalised workflow for sampling for MP in groundwater and MP in stygobites.

2.2.1 Sampling MPs in groundwater

Steel filter holders in a 5 micron steel mesh (Wolftechnik, Germany) were used (see Fig 3 and 4) in a purpose built sampling rig. These were designed to be isolated in the field using isolation valves either side of the filter holder. A third valve was located prior to the filter to allow this section of the sample rig to be flushed with sample water prior to sampling. The rig was made using copper and brass connectors (Figure 3). These were connected to the outflow of the pump tubing using an approved hose (SILEX platinum cured silicone braided hose). The rig had a WRAS approved brass double non-return value. Brass connectors and galvanised jubilee clips were used between the nitrile rubber pump tubing and the rig. A Geotech Geosub 2 pump was used to pump each well (where this was possible, i.e. there was no in-situ pump), this had three key benefits, i) it is a pump made without plastic components, ii) the flow rate can be varied using a controller and samples can be pumped from a depth of up to 60 m below ground level, and iii) the pump is more portable and easier to handle compared to larger Grundfos pumps we have available at BGS (Figure 4). The sampling rig had a small number of plastic components which are unavoidable – i) a rubber O ring to seal the filter in the filter holder, and ii) the isolation valves also contain a small HDPE component. For duplicate sampling we also used a small Y piece to split the flow which was made of plastic. These were the items that were available to us at the time for undertaking this work, some of these components could be manufactured in steel or brass but this was beyond the scope of this pilot study. Regardless, we did collect a field blank to test for contamination so these potential sources of contamination are accounted for when reporting results from groundwater sources.

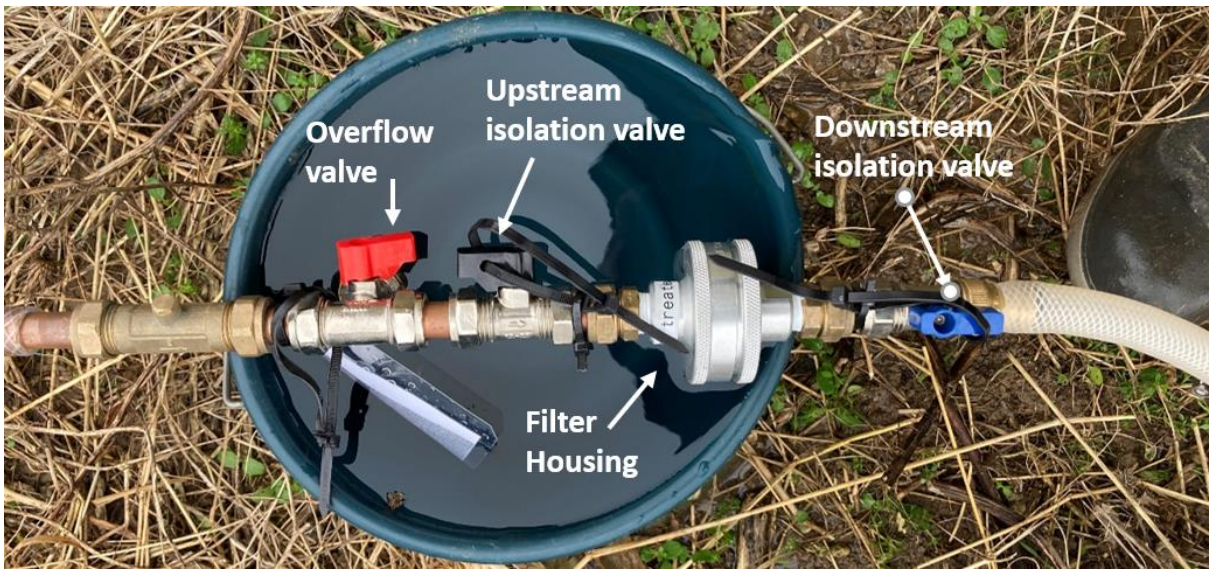

Figure 3. Sampling rig for MP filtering in the field.

Figure 4. Sampling equipment used at sources which did not have a pump installed. A=well, B=Geotech pump and controller, C=pump tubing, D=steel in-line filter and filter holder, E=digital flow meter.

Prior to sampling for MP the sampling tubing was thoroughly flushed (for 10 min) with groundwater to remove any contamination from previous sites or the tubing. The section of the rig prior to the filter was then flushed for 5 min at a flow rate of 5 L/min. The section to the filter was then opened and the flush valve closed and c. 100 L of sample passed through the filter at a flow rate of between 4—5.5 L/min. Flow and volumes were measured at the outflow point using a digital flow-meter (DigiFLow 6710M). For the majority of sites (9/12) filtering 100 L of sample was quite straightforward using the small in-line rig. At a small number of sites (3/12) a smaller volume of water was filtered through the sample rig. At 2/12 of these this was due to the fact that sampling for stygobites was undertaken prior to pumping for MP in groundwater which disturbed the sediment at the bottom of the well and led to significant turbidity in the pumped water samples. Table 2 summarises the site and sampling details.

Table 2. Site details

Site	**Volume filtered (L)**	**Source type**	**Geology**
1‡*	3	EA water level monitoring well	Chalk
2‡*	3	EA water level monitoring well	Chalk
3	100	Private borehole	Chalk
4	25	Private borehole	Chalk
5*	100	Private well	Chalk
6	100	Public borehole	Thames Gravels
7	100	Public borehole	Thames Gravels
8	100	Public borehole	Thames Gravels

‡ Site was trawled for stygobites prior to sampling,* Geotech Geosub 2 pump used to sample source, at other sites in-situ pumps were used.

2.2.2 Sampling for stygobites (*Niphargus Kochianus*)

A standard modified Cvetkov net sampler made of nylon was used to trawl for stygobites at two sites which are known to contain significant numbers of Niphargids (*N. Kochianus*), for example see Reiss et al. (2019) for further details. Three trawls of the borehole were done to collect stygobites and the method is briefly described below. The net was thoroughly washed prior to use and rinsed with MP filtered DI water. The net and related equipment was stored in a metal box. At site the net was lowered to the bottom of the well and the weight allowed to drop on to the bottom by jerking the rope securing the net. The net was then raised steadily until it was able to be taken out of the well. At the surface the inside of net was then rinsed with filtered DI and the Niphargids, sediment and other material collected at the bottom of the net and allowed to drain through the net. When the fluid had drained out of the net and filled the bottle collector (LDPE) the small nylon mesh cap at the bottom was then unthreaded carefully (Figure 5) and all the sample was collected in a pre-washed glass jar. Any material from the collector was then washed into the jar again using ethanol poured from a glass bottle and the lid secured on the jar. This was repeated 3 times to collect a 'representative' sample. Samples were taken back to the Wallingford laboratory and the stygobites ID'd using a Zeiss STEMI 2000 microscope. Glassware was used to store the stygobites and metal tweezers used to handle them. Care was taken to remove as much sediment as possible from the Niphargids by swishing then in ethanol. All the stygobites collected were stored in a cleaned glass beaker in 50% ethanol with a foil cover temporarily prior to processing.

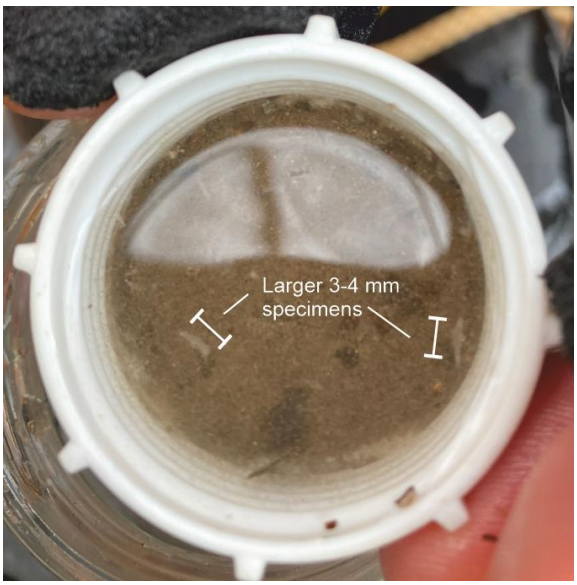

Figure 5. Sampling for stygobites – close up of the nylon filter mesh with white stygobites and brown sediment from the trawling. The diameter of the cap is c. 30 mm for an approximate scale.

2.3 SAMPLE PROCESSING FOR MP

Sample processing for groundwaters is described below step-by-step, this is based on an adapted standard operating procedure (unpublished) developed by UKCEH and used by Horton et al. (2021). All DI, HCl, peroxide and Fenton's regent was filtered (0.7 micron) to remove MPs. The majority of the sample handling and processing was undertaken in a dedicated fume hood.

2.3.1 Dismantling the rig and transfer of MP from steel filter to beaker for acid treatment.

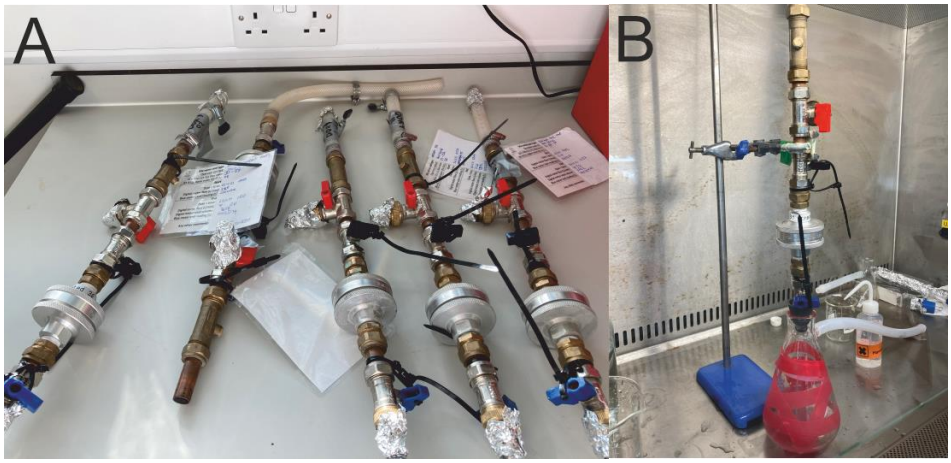

Figure 6. Sampling rig disassembly in the fume hood, (A) sampling rigs back from the field, (B) sampling rigs mounted in the retort stand in the fume hood (step 1).

1. In a fume hood mount and secure the sampling rig in a retort stand and connect to a vacuum suctioning unit.
2. Open all three taps and suck surplus water from the bottom using a vacuum pump.
3. Open the filter holder while the rig is still secured in the retort stand.
4. Remove the steel filter by using a small glass pipette to gently push out the steel filter.

Figure 7. Steel filter being removed for MP processing from a filtered sample in the fume hood, opened rig on high hand side.

5. Rinse the particles from the steel filter into a glass beaker using filtered (0.7 micron) DI.
6. Scrape the filter mesh carefully with the small glass pipette to dislodge MPs and rise with (0.7 micron) filter DI. Repeat this several times to make sure as many MP are transferred to the beaker as possible.
7. Add HCl acid to the beaker to bring it up to 2% HCl, leave for 24 hours covered with foil.
8. Wrap the steel filter in foil and label.

2.3.2 Preparing the rig for sampling again:

9. Clean the sample holder and rig with detergent and natural fibre brush, rinse with filtered DI and return to the retort stand in the fume hood ensuring all the pipe sections are well rinsed with DI.
10. Rinse the new steel filter with filtered DI, place in the filter holder and reassemble in retort stand, tighten to hand tight only.
11. With all 3 taps open suck out the excess water from the bottom. Opening and closing the blue or black valves.
12. Close the red overflow/bypass tap and secure the top black tap closed.
13. Cover ends with foil and store in a safe place before it is reused.

2.3.3 Particle deposition for FTIR analysis

14. In the fume hood set up a small vacuum filtration unit and carefully place the 25 mm silver filter on to the top of the vacuum frit with the vacuum on using steel tweezers.
15. Place the blue rubber bund on to the centre of the silver filter using the same tweezers.
16. Pipette all the 2% HCl solution and particles into the centre of the bund ensuring that the liquid does not top the level of the bund.
17. Rinse the beaker with filtered DI and repeat step 16 two more times to transfer all the particles to the silver filter. Rinse the blue bund into the beaker and transfer carefully to the centre of the silver filter using the large glass pipette.
18. Stop the vacuum pump and very carefully transfer the silver filter to the labelled glass FTIR slide
19. Secure the silver filter with 2-3 small stickers at the very edge of the silver filter and put in the slide holder and cover (Figure 8, below).
20. The slide is now ready for FTIR analysis.

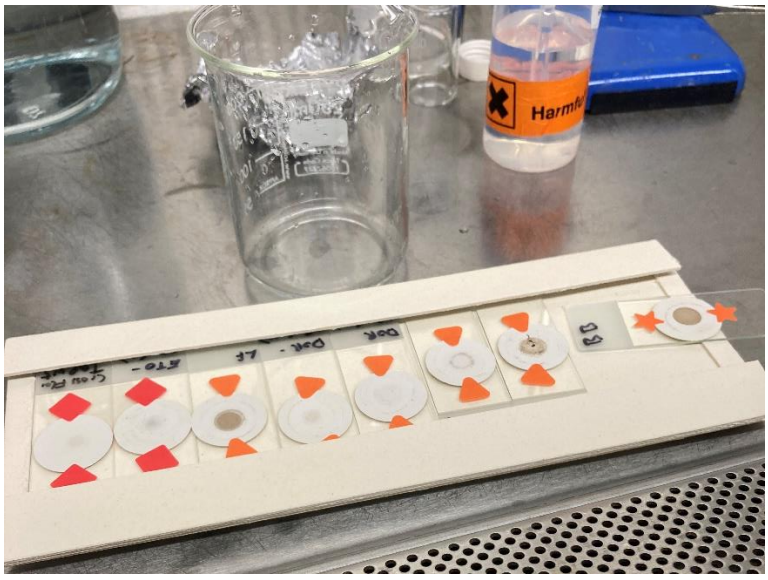

Figure 8. Securing samples on silver filters for FTIR.

2.3.4 Preparing water blanks

21. Put 10 L of 0.7 micron glass fibre filtered DI through an assembled sample rig and hosing assembly and go through steps 1-20.

2.3.5 Preparing and digesting Niphargids

All individual Niphargids per sampling were pooled together before weighing them with a Sartorius (6 decimal place) balance to obtain the dry weight for each sampling per location.

22. Weigh a clean 50 mL beaker and put in the fume hood.
23. Take the beaker with Niphargids in 50% ethanol to the fume hood.
24. Using a rinsed large 40 mL pipette take off all the ethanol making sure not to pipette any Niphargids out.
25. Add DI and transfer all Niphargids to the 50 mL beaker.
26. Rinse beaker with 20 mL DI and repeat step 24 three times rinsing each time with DI.
27. Cover with foil and dry in clean oven at 50C° ~48h to remove any excess moisture.
28. Weigh once cooled and dry and note the total weight.
29. Add 20 mL 30% H_2O_2 using a clean glass volumetric cylinder. The beakers were left in the clean oven at 50C° for 48h, which after they were taken to room temperature to cool down for 24h. This allowed the H_2O_2 to cool down before Fenton reactant was added.
30. 2 mL of Fenton's reactant was added in fume hood and left there to break down any remaining carcass pieces and left for further 24h.
31. Filtered DI was added to the sample to dissolve iron precipitates.
32. The particles were then deposited on silver filters using steps 14-20 above.

2.4 SAMPLE ANALYSIS FOR MP

2.4.1 FTIR analysis

All microplastics within a selected filter area were identified and quantified with an imaging mFTIR spectrometer (PerkinElmer Spotlight 400, Figure 9) set to collect spectra in the range between 4000 and 700 cm^{-1} wave numbers. Full details of the analytical protocol are given in Horton et al (2021). Each sample scan took c. 90 minutes to complete. We used the open source siMPle software (Simple-plastics.eu.,2022) to undertake spectral analysis of particles. We reported on the following nine polymers: polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET), poly(methyl methacrylate) (PMMA), polyamide (PA), polystyrene (PS), poly- urethane (PU), polycarbonate (PC), polyvinylchloride (PVC), acrylonitrile butadiene styrene (ABS) and cellulose artificial modified (CAM). These polymers are among the most commonly manufactured and are often reported in environmental samples (Liu et al., 2019 a,b). However, it is important to note that this is a very limited database and that there is a paucity of environmentally modified polymer spectra with which to assess environmental MPs.

Figure 9. FTIR analysis of samples using a Perkin Elmer Spotlight 400 system.

2.5 ANALYTICAL QA/QC

A full method blank, i.e. using MP filtered DI water, was run through both the groundwater MP and stygobite sampling and processing protocol for comparison with results obtained from groundwater samples. MP results from groundwater and stygobite samples were blank corrected prior to reporting MP occurrence.

All reagents used for processing water and stygobite samples were filtered (<5 micron) prior to use to remove MP contamination and also no plastic materials were used in the processing of samples that are quantified using the FTIR method. Glass containers were used for storing reagents and samples and a PFE water bottle was used for rinsing filters and glassware. All filter handling and processing was undertaken in a fume hood dedicated for MP sample handling and filter rigs were also assembled in the same fume hood. All water used for blanks and for cleaning the rig etc was filtered (<5 micron).

2.6 PILOT STUDY SITE SELECTION

Boreholes at 8 sites within Berkshire and Oxfordshire were sampled for MP in January 2022 as part of the pilot study to assess whether microplastics could be detected in groundwater samples. The sites selected contained a mix of Chalk boreholes and surface water influenced gravel boreholes, the latter selected for contrast. Six sites were pumped groundwater sources, two were EA monitoring borehole sites (Site 1 and Site 2) which were also sampled for Niphargids. The stygobite sampling was undertaken on 6th January 2022, groundwater MP sampling was undertaken after the net sampling so as not to affect the net sampling results due to pumping/disturbance. This had the drawback of increasing the turbidity of the water in the well column which meant that significantly lower volumes of water could be filtered from these sites.

Three of the pumped sites were private groundwater sources in the Chalk and 3 other pumped sites were public supply sources in the Thames gravels. The sources in the Thames gravels were sampled in duplicate. Potential sources of MP in the pumped Chalk sites are from the regular application of sludge to agricultural land surrounding each site. Sources from the Thames gravels are in hydraulic continuity with the River Thames and therefore this is likely to be the main source of MP, as well as potentially local surface sources.

All of the sources sampled had steel casing, however, we cannot rule out the potential for in-situ sources of MP from debris that has fallen into the wells, particularly those which are the EA monitoring wells and are not actively pumped. At the pumped sites the impact of in-situ sources is likely to be limited and there is good well head protection at each site.

3 Results and Discussion

3.1 FTIR ANALYSIS OF GROUNDWATERS

After blank correction, a total of 40 MP particles were found across all sites using FTIR. A single PP particle was found in the method blank. Overall, a very small proportion of the particles detected by FTIR were MPs. Of those detected as MP the largest detected was 183 μm, 28% of particles were <50 μm, 78% <100 μm and 88% <150 μm (Table 3). Polypropylene (PP) dominated the polymer composition of MPs found in samples collected from boreholes (80%), four other polymers were detected including polyethylene (PE - 8%), polystyrene (PS - 5%) and acrylate, polycarbonates and artificial cellulose all ≤ 3% (Figure 10). Table 4 details the blank corrected detections made at each site, including a breakdown of the duplicate results.

No MPs were found in numbers exceeding the blanks at either of the two EA groundwater monitoring wells that were sampled as part of this study (Table 4). However, only very low volumes were filtered at these sites due to the fact that the groundwaters were highly turbid following the net trawling, so this could at least partly explain why this was the case. The high TDS in these samples also led to significant quartz MPs in these samples which could have interfered with the MP detection using FTIR. One option to avoid overlapping with quartz would

be to process groundwater samples with high turbidity using an additional density separation step to improve MP detection if these types of samples are of future interest. However, this would be more time consuming and introduces another step and additional sources of contamination to the methodology. It is also worth noting that these are not typical of the sort of groundwater samples that would be encountered (certainly not from pumped sites), and it is not recommend to disturb the sediment at the bottom of the well prior to sampling groundwater using this low-capacity in-line method in the future. It was only done on this occasion due to the need to undertake stygobite sampling before sampling the groundwater for MPs. However, it does at least show that the sediment at the bottom of the well is not highly contaminated with MPs.

Between 1-5 MPs were found at the three sites what were pumped private sources abstracting from the Chalk (Table 4). In contrast between 0-18 MPs were detected in public supply sources that abstracted from the Thames gravels. Polyethylene was detected at 4 of the 6 public supply sites where MPs were detected. Two of the repeat samples gave good consistency of results for MP (Table 4). However, site 8, which had the highest number of detections gave very poor reproducibility (18 and 0 MPs respectively). The reason for this is uncertain, but it certainly shows that there is a high level of uncertainty in MP detections at this low level and with low numbers of samples. There is also uncertainty due to the limited database used for the spectra matching methods in the siMPle software. These results suggests that repeated sampling is needed to establish baseline conditions in groundwater.

The low level of MP occurrence (e.g. a single PP MP for this batch of samples) in the full method blank was a reassuring result, however this may also be due to the limitations of the SiMPle software. It shows the importance of undertaking method blanks, and also that with care low blank contamination can be achieved. Several groundwater sources (n=4) had MP detections that were comparable to the blank and were therefore reported as zero once blank correction had been undertaken (Table 4).

Table 3. Particle size characteristics of MP detected in groundwater samples.

Total no. MPs detected	40
Largest MP detected (µm)	183
% MPs < 150 µm	88%
% MPs < 100 µm	78%
% MPs <50 µm	28%
Average MP size (µm)	80

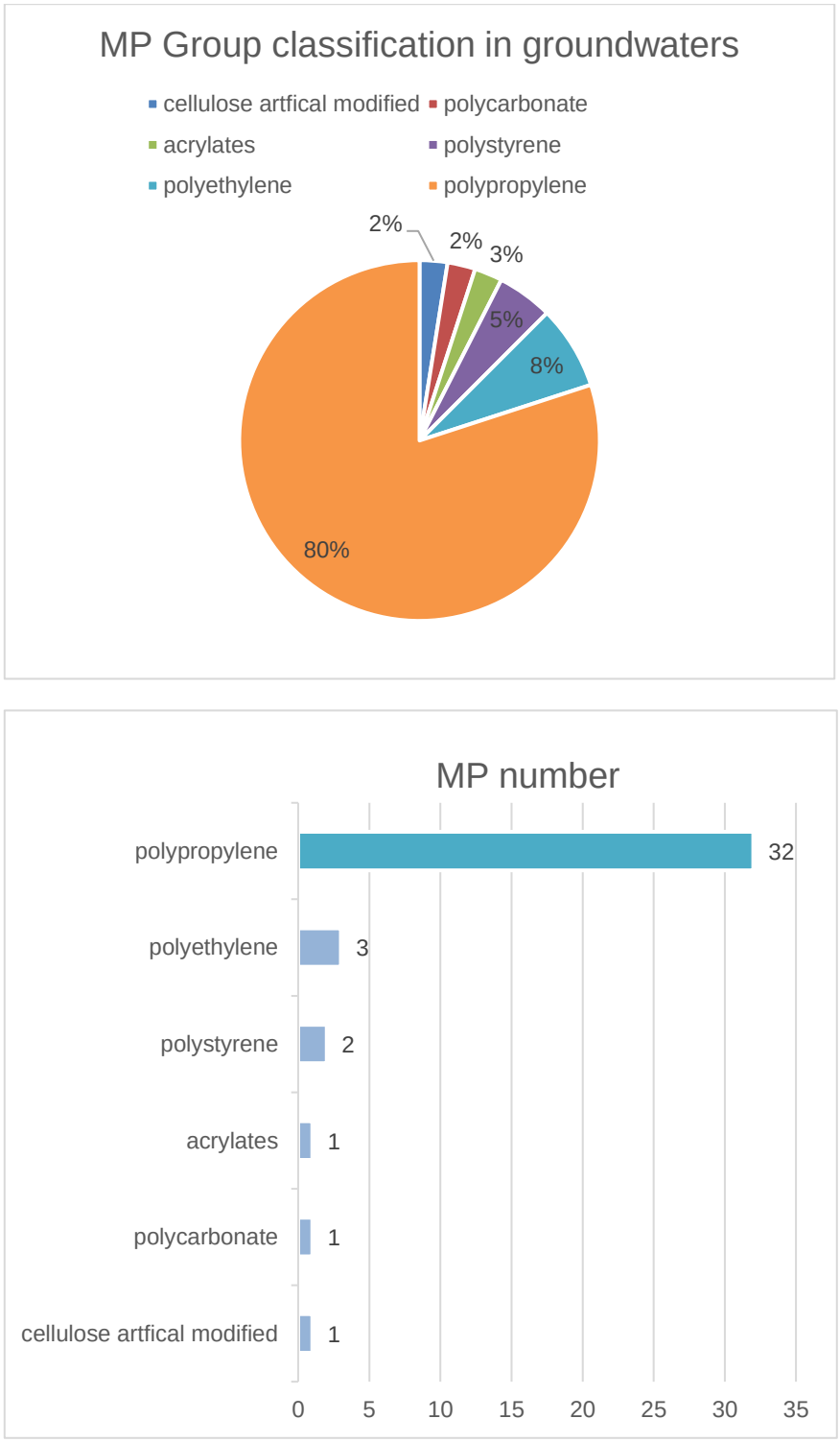

Figure 10. MP polymer composition of boreholes sampled in January 2022, top - % of MP by different composition, bottom - number of polymers by composition.

Table 4 MP results for boreholes, number of MPs, total volume filtered and MPs/L. Orange = EA water level monitoring sites, Blue = Private groundwater sources in the Chalk, Purple = Public supply sources in Thames Gravels

Site	No. MPs*	Total volume filtered (L)	MPs/L*
EA water level monitoring sites in chalk			
1	0	3	-
2	0	3	-
Private groundwater sources in the Chalk			
3	2	100	
cellulose artificial modified	1		0.01
polystyrene	1		0.01
4	1	25	0.04
polypropylene	1		0.04
5	5	100	0.05
polyethylene	1		0.01
polypropylene	4		0.04
Public groundwater sources in the Thames Gravels			
6 (1)	1	100	0.01
polyethylene	1		0.01
6 (2)	0	100	<0.01
7 (1)	8	100	0.08
acrylates/polyurethanes/varnish	1		0.01
polypropylene	6		0.06
polystyrene	1		0.01
7 (2)	0	100	<0.01
8 (1)	18	105	0.17
polycarbonate	1		0.01
polyethylene	1		0.01
polypropylene	16		0.15
8 (2)	6	105	0.06
polyethylene	1		0.01
polypropylene	5		0.05

*These values are blank corrected. The method blank had a single PP MP.

The dominance of PP within samples is concurrent with other studies looking at MP contamination in raw groundwater sources for potable use in the UK (Johnson et al., 2020). However, the small proportion of PE and lack of detection of PET in samples is divergent from this earlier work. An average particle size of 80 μm detected is concurrent with other studies looking at MPs in groundwater sources (e.g. Panno et al., 2019; Johnson et al., 2020; Samandra et al., 2022) and with 88% of particles identified being below 150 μm suggests that this size range may be typical of MP found in groundwaters. Average particle sizes and particle size distribution will however be affected by the particle size LOD 25 μm for FTIR. This is the current practical analytical cut-off for FTIR, without increasing the scan times and file sizes significantly as to make these unwieldy and unstable to use. The siMPle software used may also have underestimated the presence of MP particles in some samples, such as the two EA sites where excess silica and organic material was deposited to the silver membrane filter. Additionally, polymers such as PA (polyamide) were not detected. Whether this is due to an

actual lack of presence of these polymers or the inability of siMPle to detect them is unclear. Filters were stored as part of this project and may be re-scanned at a later date if better spectroscopic techniques or software become available.

3.2 FTIR ANALYSIS OF STYGOBITE SAMPLES

All Niphargids were identified as *Niphargus Kochianus.* The results from the identification, sample processing and FTIR analysis are summarised in Table 5.

Table 5. MP in Stygobite samples.

Site	N° of *N. Kochianus*	Dry mass (mg)	N° of MP*	Types of MP	Size range (μm)
Site 1-STYG	80	9	0	NA	NA
Site 2-STYG	35	7.7	2	PP	100-417
Blank -STYG	NA	NA	2	PP	104-177

*The numbers reported here are not blank corrected.

The low numbers of MP detected in *N. Kochianus* are consistent with results from the paired groundwater samples at these two sites (see Table 4). The sample from site 1 had zero MP detected and the sample from site 2 had 2 PP MP detected. These were all below DL once censored using the method blank. In the case of the stygobite blank only 2 PP MPs were detected (Table 5).

What was assumed to be iron precipitation was visible on the FTIR scanning region. It could be that this is either an artifact of iron contamination from the casing, or more likely it could be due to precipitation and incomplete dissolution of the Fenton's reagent. This might have the effect of covering and obscuring MPs from the stygobite samples during the deposition step and subsequent FTIR. The solutions were quite clear and transparent when deposition took place, and were washed with c. 100 mL filtered DI, so it is hoped this would have removed any remaining iron precipitation. One option to check results could be to re-float the FTIR sample and redeposit it on to a new silver filter slide to see if any other MPs are then subsequently detected by FTIR.

4 Recommendations for Future Work on MP in Groundwaters

Based on the initial findings from this pilot study there are a number of recommendations which we have made regarding the next potential steps for undertaking future work on MP in UK groundwaters. We recommend that:

1. For pumped, low turbidity, groundwater sources the in-line filter system trialled in this pilot study is continued to be used for sampling groundwater for MP.
2. Five-micron filters continue to be used, as they did not give major issues in the initial field trial, but that 25 μm filters are also trialled for comparison. With the larger pore size filter clogging and backpressure may be reduced and as the current limit of the FTIR is 25 μm this will not compromise the MP analysis, at least not until analytical methods improve significantly.
3. In future larger filter volumes are trialled, e.g. 200 L where this is practical. This will increase the ability to assess MP contamination given the low background levels typically found in groundwaters. This could involve the use of sequential sampling rigs, or the use of a splitter placed upstream of duplicate rigs, dependent on the properties of the sampled borehole (i.e. if turbidity is sufficient to only allow for <100 L of groundwater to be passed through a single filter).
4. The sieve filter system is trialled alongside the in-line method on a small subset of sites to compare results and assess levels of contamination in samples and full method blanks.
5. Gas chromatography pyrolysis analysis methods are trialled alongside FTIR methods in the future to explore this as an option for high level screening for MPs in environmental waters.
6. The FTIR data processing is undertaken using both the siMPle and pMPf software as results from siMPle could be conservative compared to pMPf.
7. Baseline groundwater MP assessments are undertaken sampling from a significantly larger number of sites as an initial phase of assessment to gathering evidence on MP occurrence in UK groundwaters. We recommend that both monitoring sites and pumped sites (e.g. public and/or private sources) with steel and PVC casing are incorporated in the sampling design as part of a larger study.
8. Groundwater sources within the key hydrogeological units used for drinking water sources are targeted as part of this initial large assessment (e.g. gravels, Chalk, sandstone and limestone).
9. Further work on groundwater stygobites is recommended now that a method has been trialled. However, it is worth noting that sites with abundant stygobites may be a biased towards less contaminated groundwaters.
10. We also propose that targeted sampling is undertaken in settings with known potential sources of MP which have higher risk of contaminating groundwater, including agricultural land with high sewage sludge application rates (or to soils with low OM content), surface water influenced boreholes, sites with shallow groundwater tables and also sites down gradient of historical unlined landfills which have received plastic waste.

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British Geological Survey holds most of the references listed below, and copies may be obtained via the library service subject to copyright legislation (contact libuser@bgs.ac.uk for details). The library catalogue is available at: https://of-ukrinerc.olib.oclc.org/folio/.

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