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Freshwater microplastics in Norway

A first look at sediment, biota and historical plankton samples from Lake Mjøsa and Lake Femunden

Photo: NIVA

Norwegian Institute for Water Research

REPORT

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Summary

The Norwegian Environment Agency (Miljødirektoratet) tasked NIVA to investigate the presence of microplastics in one of Norway's largest freshwater ecosystems, Lake Mjøsa, using monitoring methods which have been recently optimised for the marine environment. Presented here is a baseline description of microplastic distribution in Lake Mjøsa and Lake Femunden, as well as NIVAs recommendations for future monitoring of microplastics in the Norwegian freshwater environment. Microplastics were identified in sediment across all sites in Lake Mjøsa. Core slices from a known volume of sediment are well suited to investigate both geographical and historical distribution of microplastics. Bivalves, such as the duck mussel appear less useful as a test medium but might be useful for comparative analyses against the marine environment. Samples of historical plankton could be a useful way to study temporal changes in a specific area. However, fibres need to be excluded from historical samples as contamination during past sampling campaigns cannot be accounted for. Contamination mitigation measures should be considered in ongoing plankton sampling to facilitate microplastic monitoring in the future. It is not likely that choosing to investigate only one matrix (i.e. just sediments or just plankton samples) will provide a robust assessment of microplastic contamination for a whole ecosystem. A combination of water, sediment and biota samples are recommended. Microplastic monitoring could also be introduced to already established monitoring programs (e.g. Milfersk and/or ØKOSTOR), albeit gradually, to get a better understanding of microplastic distribution both geographically and between different matrixes (such as water, sediment and different biota). Long-term, continuous monitoring will eventually generate a knowledge base for assessing both the fate of microplastics and the effects they may have on organisms.

Four keywords	Fire emneord
1. Microplastic	1. Mikroplast
2. Environmental contamination	2. Forurensning
3. Sediment	3. Sediment
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Freshwater microplastics in Norway:

A first look at sediment, biota and historical plankton samples from Lake Mjøsa and Lake Femunden

Preface

NIVA has, on behalf of the Norwegian Environment Agency (Miljødirektoratet), carried out a study of microplastics in the freshwater environment of Lakes Mjøsa and Femunden in 2018. This report lists the major findings of microplastics in sediments from both lakes, and in duck mussels (*Anodonta anatina*) and historical plankton samples from Lake Mjøsa.

Sediment samples were collected using a catamaran vessel from Trolling Adventure AS. David P. Eidsvoll, Jan-Erik Thrane and Morten Jartun from NIVA performed sediment sampling with help from Stein Kristian Nordsveen and Atle Rustadbakken (Trolling Adventure AS). Asle Økelsrud from NIVA and Atle Rustadbakken collected the sediment samples from Lake Femunden and duck mussels (*Anodonta anatina*) from Lake Mjøsa. Finally, samples of historical zooplankton were selected from the archives of NIVA Region East (Ottestad) with help from Øyvind Garmo and Jarl-Eivind Løvik. Nina T. Buenaventura, David P. Eidsvoll, Anna Luise Ribeiro, and Amy Lusher from NIVA performed the laboratory analyses, including sample processing, visual analysis and chemical analysis using FT-IR. Pyrolysis-GC-MS analysis was performed on a selection of sediment sample extracts by Joakim Skovly at Eurofins Bergen. Amy Lusher and Morten Jartun have been responsible for writing the report which was quality assured by Bert van Bavel and Marianne Olsen (NIVA).

Oslo, 29.11.2018

Morten Jartun

Table of contents

Summary

The Norwegian Environment Agency (Miljødirektoratet) tasked NIVA to investigate the presence of microplastics in one of Norway's largest freshwater ecosystems, Lake Mjøsa, using monitoring methods which have been recently optimised for the marine environment. Mjøsa has previously been well described for other environmental impacts, including water chemistry, sediment chemistry, ecosystem effects and climatic assessments. Knowledge of microplastics in the Norwegian freshwater environment is limited. Presented here is a baseline description of microplastic distribution in Lake Mjøsa and Lake Femunden, as well as NIVAs recommendations for future monitoring in Norwegian freshwater environments.

In this survey, microplastics (>36 µm) have been analysed in sediments from 20 different sites in Lake Mjøsa using well-established methods from the marine environment. Sampling locations were pre-selected based on possible sources of plastic and microplastic to the lake. This includes effluents from wastewater treatment plants, road drainage, urban areas, rivers and agricultural drainage. Sediment accumulation areas representing sites with fewer potential sources of microplastic input were also identified.

Sediment sampling in Lake Mjøsa was carried out in August 2018 using a core sampler mounted on a hydraulic line. Duck mussels (*Anodonta anatina*) were collected from 3 – 6 m depth outside Brumunddal harbour. Historical plankton samples from NIVA's archive dated between 1973 – 2017 were also analysed. Lake Femunden was chosen as a reference location and ten sediment cores were sampled in September 2018 using the same methodology as in Lake Mjøsa.

Microplastics were identified in sediment across all sites in Lake Mjøsa. There was variation in the number of particles between sites and within sites. The highest microplastic values were reported at the urban area of Hamar (Site 13, 7.31 MPs g^{-1}) and Mjøsabrua (Site 4, 3.89 MPs g^{-1}) situated close to the road. Lowest microplastic values were reported within the sediment accumulation areas at Skreia (Site 17, 0.04 MPs g^{-1}). All sediment accumulation areas had similar microplastic values compared to the reference locations in Lake Femunden. Sites influenced by rivers, urban areas (including roads) and WWTPs showed comparatively higher numbers of microplastics than all sites in Lake Femunden and the sediment accumulation areas within Lake Mjøsa. Only one duck mussel from Lake Mjøsa contained a single microplastic. Nine out of twelve historical plankton samples contained plastic fragments and numbers of microplastics ranged from 0 to 14 particles per sample, indicating microplastic input already in the 1970s.

Sediment slices (~1 cm) from a sediment core were shown to be well suited to investigate both geographical and historical distribution of microplastics. Bivalves, such as the duck mussel appear less useful as a test medium but might be useful for comparative analyses to the marine environment. Samples of historical plankton could be a useful way to study temporal changes in a specific area. However, fibres were excluded from historical samples as contamination during past sampling campaigns cannot be accounted for. Contamination mitigation measures should be considered in ongoing plankton sampling to facilitate combined microplastic and neuston monitoring in the future. Mass based analysis of microplastics were also used in this study, although this method resulted in several chemical analytical challenges. Mass based determination might be a useful way to determine mass fluxes of microplastics to and from the environment

It is not likely that choosing to investigate only one matrix (i.e. just sediments or just plankton samples) will provide a robust assessment of microplastic contamination for the whole ecosystem. A

combination of water, sediment and biota samples are recommended. Microplastic monitoring could also be introduced to already established monitoring programs (e.g. Milfersk and/or ØKOSTOR), to get a better understanding of microplastic distribution both geographically and between different matrixes (such as water, sediment and different biota). Long-term, continuous monitoring will eventually generate a knowledge base for assessing both the fate of microplastics and the effects they may have on organisms.

Sammendrag

Tittel: Mikroplast i ferskvannssystemer i Norge. Et første blikk på sediment, biota og historiske planktonprøver fra Mjøsa og Femunden.
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Forfatter(e): Amy L. Lusher, Nina T. Buenaventura, David P. Eidsvoll, Jan-Erik Thrane, Asle Økelsrud og Morten Jartun.
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NIVA har på oppdrag fra Miljødirektoratet utført en undersøkelse av mikroplast i Mjøsa, Norges største innsjø. Dette er gjort ved å benytte metoder utviklet og etablert for marint miljø. Mjøsa med sitt store nedbørsfelt er tidligere godt beskrevet for ulike miljøpåvirkninger, inkludert vannkjemi, sedimentkjemi, påvirkninger på økosystem og klimavurderinger. Kunnskapen om mikroplast i ferskvannssystemer i Norge er begrenset. I denne rapporten presenterer vi resultater for fordelingen av mikroplast i Mjøsa og Femunden. Til slutt foreslår vi også hvordan studier av mikroplast kan inngå i framtidige overvåkingsprogram for ferskvann.

I denne undersøkelsen har vi analysert innholdet av mikroplastpartikler (>36 µm) i sedimenter fra 20 ulike stasjoner i Mjøsa basert på veletablerte metoder fra marine undersøkelser. Prøvetakingslokalitetene ble valgt basert på potensielle kilder til plast og mikroplast i Mjøsa, inkludert utløp fra kommunale renseanlegg, vegavrenning, urbane områder, elvetilførsler og avrenning fra landbruk. Dype områder i Mjøsa, såkalte akkumulasjonsbasseng, ble valgt for å representere områder med antatt begrensede tilførsler av mikroplast.

Prøvetakingen av sedimenter ble foretatt i august 2018 ved hjelp av en kjerneprøvetaker montert på en hydraulisk linehaler. I tillegg ble det samlet inn andemusling (*Anodonta anatina*) fra 3-6 meters dyp utenfor Brumunddal havn. Vi har også analysert historiske planktonprøver fra NIVAs rikholdige arkiv fra 1973-2017. Femunden ble valgt som referansesjø, og mikroplastinnholdet i 10 sedimentkjerner ble bestemt på samme måte som for prøvene fra Mjøsa.

Mikroplast ble påvist i sedimenter fra alle stasjoner i Mjøsa. Antall partikler varierte både mellom ulike stasjoner og i dypet. Høyest antall partikler (MP) ble funnet utenfor Hamar (lokalitet 13, 7.31 MP g^{-1}) og ved Mjøsbrua (lokalitet 4, 3.89 MP g^{-1}). Laveste innhold av mikroplastpartikler ble påvist i det dype akkumulasjonsbassenget utafor Skreia (lokalitet 17, 0.04 MP g^{-1}). Prøvene fra akkumulasjonsbasseng i Mjøsa hadde tilsvarende verdier som lokalitetene i referansesjøen Femunden. Lokaliteter påvirket av elver, urbane områder (også veger) og renseanlegg hadde høyere antall mikroplastpartikler enn alle lokalitetene i Femunden og akkumulasjonsbassengene i Mjøsa. Det ble kun påvist mikroplast (én enkelt partikkel) i én prøve av andemusling. Ni av tolv historiske planktonprøver inneholdt mikroplastfragmenter fra 0-14 partikler per prøve, noe som antyder tilførsler av mikroplast fra 70-tallet

Resultatene viser at analyse av tynne (ca. 1 cm) sjikt fra en sedimentkjerne er en velegnet måte å se på både geografisk og historisk fordeling av plastpartikler. Andemuslinger viser seg å være et mindre godt egnet prøvemateriale i ferskvann, men kan lett opparbeides for mikroplastanalyser, og er sammenlignbart med marine undersøkelser. Prøver av historisk plankton kan være en velegnet måte å studere tidstrender for et spesifikt område, men fiber er nødt til å ekskluderes fra analysene av de historiske prøvene pga. kontamineringsfare under selve prøvetakingen. Dagens prøvetaking av plankton kan med enkle tiltak begrense faren for kontaminering med fibre slik at plankton kan være et mulig prøvemateriale for mikroplast i framtida. Massebaserte analyser av mikroplast ble også utført

I denne undersøkelsen. Denne metoden ga noen analytiske utfordringer, men massebasert bestemmelse kan være en nyttig måte å studere masseflyten av mikroplast til og fra naturmiljøet.

Det er lite hensiktsmessig å studere kun ett prøvemedium (f.eks. kun sedimenter eller kun planktonprøver) for å overvåke mikroplast i et stort ferskvannsøkosystem som Mjøsa. En kombinasjon av vann-, sediment- og biotaprøver vil være å anbefale. Det anbefales også å innføre overvåkning av mikroplast i allerede etablerte overvåkingsprogram (f.eks. Milfersk og/eller ØKOSTOR), om enn gradvis, for å få en bedre forståelse av både den geografiske fordelingen og mellom ulike prøvematerialer (som vann, sediment og ulike organismer). En langsiktig, kontinuerlig overvåkning vil etter hvert frembringe en kunnskapsbase og database som gir et mye bedre grunnlag for å vurdere både skjebne til partikler og effektene de eventuelt måtte øve på organismene.

1 Introduction

Plastic is a ubiquitous environmental contaminant found around the globe. This includes microplastics (<1 mm) which have been identified in remote environments including polar regions, the deep sea and isolated mountain lakes. This widespread environmental presence highlights a need to understand sources and consequences of these small particles. Microplastic contamination is expected to increase in years to come, especially in view of increasing global plastic production and the subsequent degradation and fragmentation of larger plastics in nature.

Monitoring surveys of microplastics have been primarily focused towards the marine environment; these have identified microplastics in surface waters, the water column and deposited in sediments (reviewed in Lusher 2015). However, surveys of microplastic deposition to sediments are sparser with limited repetition; coastal and intertidal locations have been investigated in more detail (Blumenröder et al., 2017; Martin et al., 2017; Møskeland, 2018). Worryingly, microplastic presence in environmental samples suggests that biota can interact with this form of pollution (*e.g.,* Rezania et al., 2018). Research into consequences of biota interacting with microplastic is ongoing; currently, there is sufficient information demonstrating that uptake, including ingestion, is occurring in nature. Further research into associated consequences is still required.

Freshwater systems including lakes, rivers and streams are not free of microplastic contamination. In fact, freshwater systems, namely rivers, have been identified as an important source of microplastic pollution to marine ecosystems (Rech et al., 2014). Research into microplastics in freshwater systems began much later than marine systems and the number of peer-reviewed studies are still outweighed almost 9:1. A recent review of publications from 1980 to May 2018 found that only 13% of peer-reviewed publications were freshwater-orientated (Blettler et al., 2018). Concurrently, there are still major knowledge gaps surrounding microplastics in freshwater environments (Lambert and Wagner, 2018; Scherer et al., 2018).

Freshwater systems can act as receivers, transport routes (from terrestrial to aquatic, freshwater then marine) and sinks (isolated lakes) of microplastics. Some examples of microplastics input from terrestrial sources entering freshwater systems include: waste water treatment plants (WWTPs) and sewage systems or storm overflow systems, terrestrial runoff and application of biosolids from WWTPs, incidental release from plastics use of land (*e.g.,* tyre wear), release from industrial products and processes, emissions from manufacturing and construction sites, and atmospheric deposition of airborne particles (Lambert and Wagner, 2018). Level of input is likely to be highly influenced by land use, with isolated water bodies expected to have low levels of input compared to areas with high populations (Eriksen et al., 2013). River transport will be dependent on river hydrology (*e.g.,* flow conditions, daily discharge, flooding) and river morphology (*e.g.,* vegetation pattern). Particles may break down because of river conditions, which could also affect microplastic bioavailability in freshwater systems (Besseling et al., 2017; Hurley et al., 2017).

Microplastics have been found in freshwater surface waters and sediments although there is disparity between numbers of microplastics identified. For more information on microplastic occurrence in freshwater systems refer to extensive reviews conducted by Wu et al., (2018), Li et al. (2018) and Khan et al., (2018). Differences in presented results may be related to inherent natural conditions, human activity and sources of input, as well as different sampling approaches in each study (Eerkes-Mendrano et al., 2015).

Impacts from microplastic contamination of rivers and lakes are far from being fully realised (Scherer et al., 2018) and investigating interactions between biota and microplastics in freshwater systems has been identified as a high priority research need (Besseling et al., 2017). However, microplastic ingestion by freshwater invertebrates is rarely reported outside laboratory studies. Understanding microplastic uptake and effects is important in terms of performing risk assessments to evaluate exposure risk. This is especially important for freshwater benthic organisms, which may be at a higher risk due to microplastics depositing onto sediments (Redondo-Hasselerharm et al., 2018).

More than 200 different marine species have been found to ingest microplastics, while less than 70 freshwater species have been found to ingest micro- or nanoplastics (Lusher, 2015; Scherer et al., 2018). Current level of knowledge related to freshwater biota is highly biased towards laboratory exposure experiments. Studies investigating potential adverse effects on freshwater biota are scarce. Potential physical impacts of micro- and nanoplastic exposure include internal blockages, reduced dietary intake and internal injury (Scherer et al., 2018). However, controlled exposures exhibited no overt toxicity for environmentally relevant concentrations of microplastics (Redondo-Hasselerharm et al., 2018). There does not appear to be any information available regarding benthic dwelling organisms in freshwater ecosystems and knowledge on biological impact of microplastics is limited.

Unfortunately, current understanding of microplastics in freshwater environments is fragmented due to inherent differences between freshwater studies. This includes the amount and type of data generated, habitats, study species, geographical locations, social and economic context of individual studies. Knowledge of microplastics in Nordic freshwater environments is even more restricted to ministerial reports and MSc thesis reports (*e.g.,* Bottolfsen, 2017; Buenaventura, 2017; Magnussen et al., 2016). Prior to the current study, no freshwater studies investigating microplastic impact on Nordic lake ecosystems were available. Therefore, the purpose of this report was to create a baseline of knowledge of microplastics in Norwegian lakes.

1.1 Aims and deliverables

This report aims to contribute to increasing knowledge of microplastic contamination in freshwater environments. This will be accomplished by:

1. Mapping microplastic occurrence in Lake Mjøsa to provide a baseline of knowledge
2. Quantifying different plastic types (shape, size, polymer) found within Lake Mjøsa influenced by potentially different levels of anthropogenic inputs
3. Describing potential accumulation of microplastics within sediment core samples
4. Inferring, where possible, potential sources of microplastics detected in samples
5. Identifying knowledge gaps and method development requirements that may be useful in future limnetic investigations
6. Providing recommendations for future investigations in Lake Mjøsa and other freshwater ecosystems

To do so, an assessment of microplastic contamination in two of Norway's lakes was carried out by investigating sediments, biota and historical water samples. Mjøsa was identified to represent a moderately influenced lake. NIVA also chose to investigate Lake Femunden as a reference lake as it is remote from sources of contamination (Table 1). Results obtained from this study will provide a foundation for recommended further microplastics monitoring in Norway, and in freshwater in general.

1.2 Choice of freshwater lakes

Mjøsa is the largest lake in Norway, and one of the deepest lakes in Europe (453 m, Table 1). The catchment area (~17 000 km^2) includes large mountain areas in the north, and forest and agricultural areas east and west of the lake. There are five urban areas including the three large cities of Hamar, Gjøvik and Lillehammer. Mjøsa is a drinking water source for approximately 80,000 people through municipal and private water supply, as well as industry. Mjøsa supports a rich diversity of fish equating to about 20 different species including trout (*Salmo trutta*), perch (*Perca fluviatilis*) and roach (*Rutilus rutilus*). This attracts many recreational fishers to the region. With such a large catchment area and many user interests, it is no surprise that Mjøsa may be affected by several sources of pollution including road drainage, industry, decommissioned landfills, active landfills, agriculture and municipal waste-water treatment plants (WWTPs). Furthermore, the water surface is subject to the deposit of long-range air pollution. Recent environmental monitoring includes detailed studies of, among other things, eutrophication and organic and inorganic contaminants in the food chain (Løvik et al., 2017; Jartun et al., 2018).

Femunden is the second largest natural lake in Norway (Table 1). It covers a catchment area of 1800 km^2, mostly consisting of mountains and forests. The area around Lake Femunden has relatively few known sources of potential contamination draining into the lake, although there may be some sources relating to boat traffic, small local sewage treatment facilities at Elgå and some outdoor activities (hiking, camping, fishing, canoeing and kayaking). Lake Femunden was chosen as a reference lake because it is a large, deep fjord lake similar to Mjøsa, but is located in a strictly rural area, which is, to a limited degree, influenced by few diffusive sources of contamination from anthropogenic activities.

Table 1. Descriptive overview of Lake Mjøsa and Lake Femunden investigated for the presence of microplastics in the present study.

	Mjøsa	Femunden
East	10.994	11.897
North	60.791	62.185
Water volume (km^3)	65	6
Surface area (km^2)	369	203
Maximum depth (m)	453	1
Catchment (km^2)	17 251	1 790
Sewage, discharge from WWTP (person equivalent)	200 000	200
Theoretical residence time (years)	4.9 years	7.6 years
Water input from Gudbrandsdalslågen (m^3 s^{-1})	256	-
Lake outlet runoff (m^3 s^{-1})	321	24

1.3 Definition of microplastics

NIVA uses particle size definitions of microplastics in accordance with international standards (1 mm, GESAMP 2015) but appreciates that 5 mm is widely used from a regulatory perspective. Therefore, in this report we utilise two size classes: small microplastics, <1 mm, and large microplastics, 1 – 5 mm. These definitions are to conform to the reporting standards of scientists, monitoring programs and international advisory bodies. The lower limit of microplastics in this study is defined by sampling method (36 µm for sediments; 50 µm for biota and historical plankton samples).

2 Methods

Three different types of samples were collected and analysed for microplastics: sediment cores, historical plankton and duck mussels. Sediment cores were collected to represent sediment deposition. Historical plankton samples were analysed to represent temporal trends. Duck mussels were collected to investigate uptake of microplastics by freshwater biota. All samples were analysed using visual and chemical analytical techniques to allow a thorough investigation into the types of microplastics found in Norwegian freshwater environments.

2.1 Site selection

Twenty locations within Lake Mjøsa and ten locations within Lake Femunden were identified for sediment sampling (Figure 1). These sites were chosen to represent the various potential sources of microplastics into the lakes, as well as reference points in Lake Mjøsa's natural sediment accumulation areas. Several of the selected stations have been previously described regarding location, depth, sediment quality and content of certain pollutants (Table 2). Ten locations within Lake Femunden were chosen as reference sites as they are far from sources of anthropogenic input.

Table 2. Location of stations in Lake Mjøsa with a description of the assumed main source of anthropogenic influence. Five sites within Lake Mjøsa's natural sediment accumulation areas (SAA). These were chosen to represent sites with low levels of anthropogenic influence.

ID	Station	Depth (m)	Influence	Comment	Latitude (N)	Longitude (E)
1	Nes, Tingnes	95	SAA	Accumulation basin	60.7710	10.9871
2	Lillehammer WWTP	30	WWTP	South of WWTP	61.0949	10.4513
3	Lillehammer	4	Urban	South of Vingnes bridge	61.1037	10.4503
4	Mjøsbrua	5	Road	South of Mjøsa bridge	60.9236	10.6797
5	Moelv	25	River	Some industry	60.9208	10.6814
6	Redalen	54	River	Agricultural drainage	60.8952	10.6897
7	Gjøvik	13	Urban	Skibladner pier	60.7986	10.6998
8	Hunnselva	20	River	Industry and agriculture	60.7930	10.7037
9	Rambekk	18	WWTP	Close to Rambekk WWTP	60.7790	10.7121
10	Breili	88	SAA	Some agricultural drainage	60.7520	10.7826
11	Brumunddal	43	Urban	Some river input, industrial	60.8673	10.9272
12	Helgøya	94	SAA	Accumulation basin	60.7024	11.0402
13	Hamar	8	Urban	Industry, marina	60.7899	11.0691
14	Åkersvika	29	River	Svartelva, some farming some road drainage	60.7756	11.0638
15	HIAS, Hamar	29	WWTP	Close to WWTP outlet	60.7667	11.0653
16	Lenaelva	40	River	Agricultural drainage	60.6638	10.9872
17	Skreia	450	SAA	Accumulation basin	60.5831	11.2653
18	Espa	42	Road	Emissions from E6	60.6339	11.1142
19	Morskogen	317	SAA	Accumulation basin, E6	60.4742	11.2139
20	Minnesund	46	SAA	Accumulation basin	60.4059	11.2377

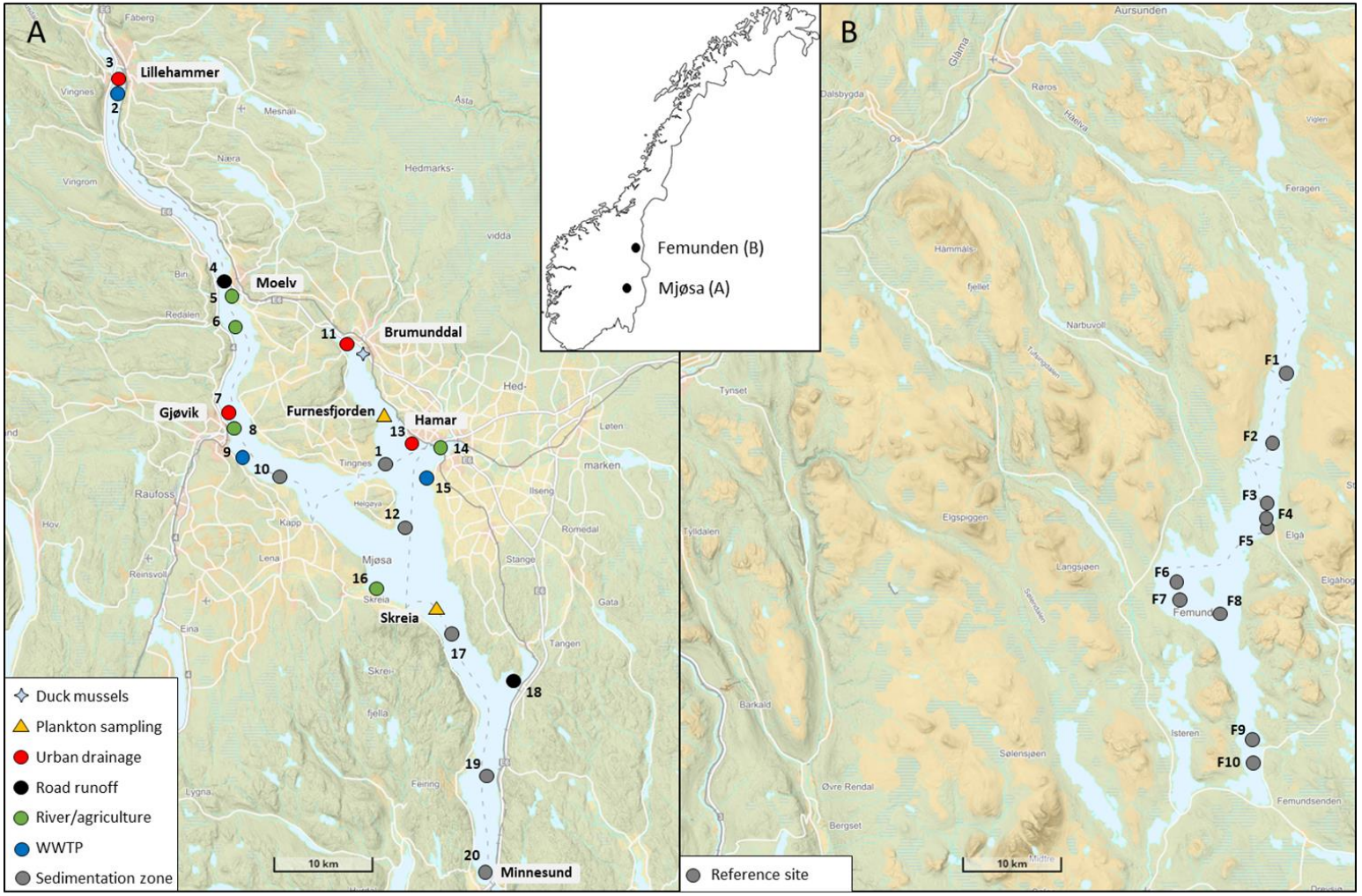

Figure 1. Map of Lake Mjøsa (A) and Lake Femunden (B) including sites for duck mussel collection (cross), historical plankton sampling stations (triangles) and sediment sampling (circles). Sediment sites are coloured to represent the different potential sources of contamination. These include, urban drainage (red), road run-off (black), river and agricultural input (green), wastewater treatment plants, WWTPs (blue) and sites in Lake Mjøsa's natural sediment accumulation areas (grey). Basemaps acquired from www.norgeskart.no

Table 3. Location of stations in Lake Femunden. All sites are assumed to be far from sources of anthropogenic influence

ID	Station	Depth (m)	Latitude (N)	Longitude (E)
F1	North 1	31	62.2869	11.9369
F2	North 2	32	62.2434	11.9344
F3	Elgå 1	36	62.1843	11.9285
F4	Elgå 2	27	62.1724	11.9235
F5	Elgå 3	12	62.1670	11.9282
F6	Mid 1	13	62.1225	11.7513
F7	Mid 2	19	62.1047	11.7603
F8	Mid 3	31	62.0927	11.8306
F9	South 1	14	61.9790	11.9213
F10	South 2	29	61.9649	11.9285

2.2 Sample collection

Sediment sample collection was carried out in Lake Mjøsa and Lake Femunden between August 6th – 9th and September 25th – 26th 2018. Sediment stations were chosen prior to field work based on the influence from potential anthropogenic sources (Table 2; Table 3). Duck mussels (*Anodonta anatina*) were collected close to Brumunddal marina, a small boat harbour (N 60.878, E 10.926) by a local diver from a depth of 6 – 8 meters on August 25th 2018.

2.2.1 Sediment sampling

Both lakes were sampled following the same procedure. At each location a Kajak-Brinkhurst sediment corer with an acid-proof test tube (Ø 8.5 cm), was deployed from the stern of the vessel. On contact with the substrate a closing mechanism was automatically triggered, keeping the sediment core of approx. 30 cm within the tube. Each core was retrieved to the back deck by help of a hydraulic hauling mechanism. On deck, the weight on top of the corer was removed, and a pressing device was used to gently push the sediment core out from the bottom of the corer making the top layer available at the top. Sections of 1 cm were sliced carefully and transferred to a glass jar. For some of the sites, high water content or larger grain sizes made it difficult to cut out even slices. In those cases, a steel ring of 1 cm in height was used to visualize the required depth, and carefully scooped the sediment amount within this ring into the glass jar.

A van Veen grab had to be used in the deepest parts of Lake Mjøsa (Sites 17 and 19, Figure 1). This was due to length of the wire and challenges in getting the corer to enter the substrate vertically. In addition, Sites 2 and 3 (Figure 1), had to be sampled with the van Veen grab. At these four sites the grab was cautiously lowered to the substrate, monitored by a sounder, to limit impact on the top layer sediment. This approach secured an undisturbed sample substrate to 10 – 15 cm depth within the grab. Samples were collected by opening the top lid of the grab once aboard the vessel. A steel spoon was used to transfer each centimetre slice into the glass jars.

A replicate core was collected from 10 locations in Lake Mjøsa to provide samples for grain size analysis.

Figure 2. Deployment of Kajak-Brinkhurst sediment corer during sampling in Lake Mjøsa and Lake Femunden. Images show core being deployed (A) and retrieved (B) at the stern of the vessel. Images: David P. Eidsvoll, NIVA.

2.2.2 Water sampling – samples of historical plankton

It was not possible to collect bulk water samples in Lake Mjøsa as planned. This was due to technical issues with equipment. As a substitution for high-volume water samples, we decided to analyse samples of plankton. This allowed us to perform analyses on historical samples from the 1970s up to present day. NIVA has performed studies and monitoring of freshwater parameters such as phyto- and zooplankton communities since the late 1960s and thus had access to these stored samples. Twelve samples from NIVA's archive were selected for microplastic analysis (Table 4). All but one sample were collected from one of NIVAs main sampling stations outside Skreia (Figure 1). This is a particularly deep part of the lake within the main basin. A single sample (HP_05.1973) was collected in Furnesfjorden in the north-eastern part of Lake Mjøsa (Figure 1). Samples were collected using landing nets with a 30 cm Ø opening and 60 – 90 µm mesh size or large bulk water samples (approx. 225 litres) for quantitative analyses. Nets were lowered to an assigned depth and slowly pulled back up to the surface, which means that for a depth of 80 or 120 m a total water amount of approx. 5500 or 8500 litres were filtered, respectively. Samples were persevered in formalin for storage.

Table 4. Historical plankton samples from Lake Mjøsa. * (Q): collected as quantitative samples using a Schindler trap of 25 L and a 60 µm mesh. These samples are a collection of 9 subsamples 25 litres, a total amount of 225 litres.

Sample code	Location	Depth (m)	Mesh (µm)	Sampling volume (l)	Date
HP_05.1973	Furnesfjorden (Q)	0.5 – 80	60	225*	03.05.1973
HP_09.1973	Skreia	0 – 80	95	5652	13.09.1973
HP_09.1978a	Skreia (Q)	20	60	225*	14.09.1978
HP_09.1978b	Skreia (Q)	0.5	60	225*	14.09.1978
HP_07.1981	Skreia (Q)	0.5 – 50	60	225*	17.07.1981
HP_06.1989a	Skreia	0 – 120	60	8478	06.06.1989
HP_06.1989b	Skreia	0 – 120	60	8478	21.06.1989
HP_06.1991	Skreia	0 – 120	60	8478	07.06.1991
HP_09.1998	Skreia (Q)	0 – 50	60	225*	21.09.1998
HP_10.2002	Skreia (Q)	0.5 – 50	60	225*	24.10.2002
HP_10.2011	Skreia	0 – 120	60	8478	28.10.2011
HP_06.2017	Skreia (Q)	0 – 50	60	225*	13.06.2017

2.2.3 Biota sampling

A sample of 10 individual freshwater duck mussels (*Anodonta anatina*) were collected from Brumunddal marina by a local diver from depths ranging between 3 and 6 metres. Only living individuals with no visible signs of damage were collected. Individuals were frozen (– 20 °C) in their shells as soon as possible after collection.

2.3 Sample preparation

2.3.1 Sediment

Each core slice was processed following NIVA protocol (17654) for density separation of sediments. First, each core slice was freeze dried and sample weight (g, d.w.) recorded. Density separation using sodium Iodide (NaI) was carried out using two extractions. There were some minor alterations to the protocol when core samples contained high levels of organic matter which hindered the effectiveness of density extraction. This followed NIVA protocol (17655) for organic matter removal.

Density separation

Firstly, a saturated NaI density solution (1.8 g cm^{-3}) was added to each sample in a Falcon tube and each was filled to the top. The Falcon tubes were closed and agitated vigorously for 1 minute. Samples were left for 24 hours to allow fine particulate matter to settle out of suspension. Floating material was filtered through two nested sieved to separate size classes. Material remaining in the larger sieve (75 µm) was decanted into a vacuum filter and passed through glass microfibre filter papers (GF/D, pore size 2.7 µm). Filter papers were left to dry at room temperature in a closed petri dish. Material which passed through the 75 µm sieve but retained on the 36 µm sieve was retained for later analysis. For the second density extraction, additional NaI solution was added to the remaining material in the Falcon tube. Samples were agitated again and after 24 hours settling time the extraction was repeated. Each sample produced two filter papers (extract 1 and extract 2). All filter papers were stored in a sealed petri dish prior to analysis.

Organic matter removal (OMR)

Thirty-seven core extracts (37/200) required further processing due to large quantities of organic matter. Samples were deemed not suitable for visual analysis when organic matter formed a layer (>1 mm) on the surface of the filter paper. Fenton's solution (30% H_2O_2 with Fe catalyst) was employed as an additional processing step. Samples were pre-treated with hydrogen peroxide (H_2O_2) overnight to kick-start the reaction. The iron (Fe) catalyst was added followed by H_2O_2 in a ratio of 1:1 until reactions were complete. Reactions were carried out in an ice bath to keep temperatures to a minimum and preserve potential microplastics. Processing time varied depending on organic matter composition. Of the 37 samples that required OMR, 18 (32%) had to undergo the procedure twice due to incomplete OMR during the first round. All samples were suitable for visual analysis once OMR was complete.

2.3.2 Historical plankton samples

NIVA obtained 12 historical plankton samples dating back to 1971 (Table 4). Each historical plankton sample was first rinsed to remove formalin in a fume hood onto a 50 µm metal sieve. Material retained on the sieve was resuspended in a Falcon tube before adding 100 ml of KOH (10%). Each sample was left at room temperature for 24 hours before filtering onto Whatman filter papers and stored in a sealed petri dish prior to analysis (GF/D, pore size 2.7 µm).

2.3.3 Biota – Duck mussels (*Anodonta anatina*)

Ten duck mussels were analysed for microplastics using a digestion protocol which removes organic material. First, the maximum length of individual mussels was measured (mm) using callipers. Individuals were opened by cutting the abductor mussels the soft tissue was rinsed with pre-filtered water and excess water discharged before the soft tissue was carefully dissected out with a scalpel. Individual mussels were weighed (g w.w.), added to individual glass conical flasks and covered with aluminium foil. NIVA has optimised the use of potassium hydroxide when working with biota soft tissue (Bråte et al., 2018). A solution of 10% KOH (250 ml) was added to each conical flask before they were placed in an incubator (New Brunswick™Innova® 44/44R) at 60°C and agitated at 125 rpm for 24 hours.

Duck mussels are sedimentary and there was a large amount of sediment in the digestate which required further processing. To compensate for this each sample was passed through a 50 µm metal sieve and resuspended into a pre-rinsed Falcon tube. Next, each sample was extracted based on density, first by a freshwater extraction by filling the falcon tube with additional filtered water, shaken gently and left to stand for 24 hours. The over laying water was filtered off (e.g., extract 1) and the remaining material resuspended in a high-density solution (NaI, 1. 7 g cm^{-3}) and left to settle for a further 24 hours. Finally, the remaining overlaying water was filtered off (e.g., extract 2). All filtered extracts (n= 20, on Whatman GF/D, pore size 2.7 µm) were stored in a sealed petri dish prior to analysis.

2.4 Sample analysis

After preparation, all samples were analysed by visual identification followed by chemical confirmation of the polymer material. Visual analysis followed standard NIVA protocols where potential plastics were isolated, photographed, described in terms of shape and colour, and measured along the longest and shorted length (mm). This was carried out using a stereomicroscope with an Infinity 1-3C mounted camera and INFINITY ANALYZE and CAPTURE software. All particles found were marked on the filter paper for easy identification prior to chemical characterization. Visual identification of microplastics, especially in the smaller size range (<1 mm), should always be supported by secondary analyses to confirm the polymeric material (Lusher et al. 2017). Therefore, all particles identified during visual ID were further characterised using chemical analytical techniques.

Historical plankton samples were collected prior to the introduction of sampling steps to prevent procedural contamination from plastic fibres (i.e. several decades prior to the onset of microplastic research). Therefore, fibrous particle presence was recorded but not quantified for historic plankton samples in this report.

2.4.1 FT-IR

Chemical characterization of potential plastic particles was performed using a combination of single point measurement Attenuated Total Reflectance - Fourier Transformed Infra-Red Spectrometry (ATR-FT-IR; for particles > 300 µm) and single-point µFT-IR. NIVA conducted ATR-FT-IR on all extracted particles. This exceeds the recommendation for reporting under European Union's Marine Strategy Framework Directive (MSFD) where it is recommended that a proportion (5 – 10%) of all samples should be routinely checked to confirm the accuracy of visual examination (Gago et al., 2016). All FT-IR results, regardless of measurement techniques, were compared to an extensive library of polymers to identify the polymer type of each particle.

2.4.2 Pyrolysis GC-MS

Ten composited samples from the top (0 – 1 cm, 36 – 75 µm) core slice were processed using Pyrolysis GC-MS to determine the weight-based polymer concentration of particles in the 36 – 75 µm size range by NIVA's partner Eurofins. The original idea was to analyse the same sample twice: once using visual and chemical characterisation per sample, and secondly using Pyrolysis GC-MS to obtain the mass of the different polymers in the samples. For several technical reasons this approach was not feasible for Lake Mjøsa sediment samples. Sediment samples for Pyrolysis G-MS need to be relatively free from interferences including fine grains and high organic matter content. Therefore, ten fractions were selected, 36 – 75 µm, from the top of slice (0-1- cm) of 10 sediment cores from Lake Mjøsa for independent analysis at Eurofins. Each extract (n=2) of each sample (n=10) was combined by filtering onto a 25 µm stainless steel metal mesh according to the procedure of Fischer and Scholz-Böttcher (2017). Two of the sample extracts were cloudy-coloured in appearance and these samples were not analysed. Two additional samples showed large 'sedimentation' on the 25 µm stainless steel filters are were also not further processed or analysed by Pyrolysis GC-MS. The six remaining filters were carefully placed in a Pyrolysis cup and placed in the Pyrolysis auto sampler unit. Pyrolysis was performed at 590 ˚C for 15 seconds under helium and transferred to the GC injector and subsequently to a 30-meter DB-5 column. Full scan spectra were obtained from 50 – 650 mass units at 70 eV at a rate of 2.5 scans per second. From the full scan spectra specific quantifier and qualifier masses were chosen and extracted. The samples were quantified against eight calibration curves for eight common types of plastics including polyethylene (PE), polypropylene (PP), polystyrene (PS), polyvinyl chloride (PVC), polyethylene terephthalate (PET), polycarbonate (PC), polymethylmethacrylate (PMMA), polyamide-6 (PA6) with a linear range from 5 – 20 µg to 200 – 400 µg depending on the polymer type. Although the method is relatively specific interference have been documented, for example chitin and organic material on the benzene fragment ion of benzene of PVC (Fischer and Scholz-Böttcher, 2017). For this reason, PVC data was removed from the results.

2.4.3 Sediment grain size analysis

Grain size analysis was performed on 10 selected samples of top-layer sediments from Lake Mjøsa. The method determined the finer (<63 µm) fraction consisting of silt and clay. Analysis was performed on freeze dried material using a 63 µm sieve. Results are given as fraction (%) of sample below 63 µm. For quality assurance, two duplicates were performed in parallel to the main batch of samples.

2.5 Data analysis

Analysis of data was conducted on sediments, biota and historical plankton separately. For sediment samples, both density extracts per core slice were compiled together for the calculation of results and are reported as number of microplastics per gram (MPs g^{-1} d.w) calculated from visual and chemical analysis. Where Pyrolysis GC-MS was carried out on a subsample of the top core slices the results are reported in µg per sample. To compare both data sets, number of microplastics and mass per sample are used. Finally, sediment results are discussed per core slice. Site locations have been removed to prevent bias in the data analysis. Site characteristics are included in the latter analysis. Biota results are presented as number of microplastics per individual (MPs $indi^{-1}$) as well as per gram (MPs g^{-1} d.w). Historical plankton results are reported as number of microplastic per sample and standardised based on original sample volume to microplastics per litre (MPs l^{-1}).

2.6 Contamination controls

The research team identified possible sources of procedural contamination prior to the start of the project and therefore carried out procedural blanks throughout. This was to avoid contamination at all stages of the project, thus ensuring comparable results. Previous methods testing found it not necessary to include procedural blanks during field work as each core slice was open to air for a maximum of 30 seconds. Opening and closing blank bottles for air samples takes more time than transferring the core slice to the jar itself. In addition, samplers wore cotton clothing and rinsed all equipment between samples. Procedural blanks during processing include controls for density separation and exposure to air during preparation and sieving. All equipment was cleaned with pre-filtered water and the use of plastic laboratory equipment was kept to a minimum. Blank information is reported alongside the results.

3 Results

Microplastics were identified in sediment across all sites in Lake Mjøsa. There was variation in the number of particles between sites and within sites. The highest microplastic values were reported at Hamar (Site 13, 7.31 MPs g^{-1}) and Mjøsbrua (Site 4, 3.89 MPs g^{-1}) which were identified as urban and situated close to the road. Lowest microplastic values were reported within the sediment accumulation areas at Skreia (Site 17, 0.04 MPs g^{-1}). All sediment accumulation areas had similar microplastic values to the reference locations in Lake Femunden. Sites influenced by rivers, urban areas (including roads) and WWTPs had comparatively higher numbers of microplastics than all sites in Lake Femunden as well as the sediment accumulation areas in Lake Mjøsa. Only one duck mussel from Lake Mjøsa contained a single microplastic. Nine out of twelve historical plankton samples contained plastic fragments and numbers of microplastics ranged from 0 to 14 particles per sample, indicating microplastic input in the 1970s.

3.1 Correction from contamination in blanks

Up to six blanks were performed alongside each density extraction for sediment (Table 5). Chemical analysis of particles found in blanks showed that they consisted of cotton and cellulose fibres and a single polypropylene (PP) fragment. These particles were deemed procedural contamination. Cellulose and cotton are not included in our definition of microplastics and were subsequently not included in the results. The single pink PP fragment was observed in a blank corresponding to core section 7 – 8 cm. No pink fragments were observed in the corresponding core slices. Subsequently, there was no blank correction required for sediment analysis. Three procedural blanks were carried out alongside the processing of biota soft tissue as well as historical plankton. No particles were observed therefore no blank correction was required.

Table 5. Results from blanks carried out during sediment processing for both high density, sodium iodide (NaI) extractions (A, B). Blank correction was required for one core slice (7 – 8 cm).

	Number of blanks	Blanks with particles	Confirmed plastic	Blank correction
0 – 1 cm A	6	2	0	n.a.
0 – 1 cm B	6	0	0	n.a.
1 – 2 cm A	5	0	0	n.a.
1 – 2 cm B	5	1	0	n.a.
2 – 3 cm A	5	1	0	n.a.
2 – 3 cm B	5	1	0	n.a.
3 – 4 cm A	5	1	0	n.a.
3 – 4 cm B	5	0	0	n.a.
7 – 8 cm A	5	1	1	yes
7 – 8 cm B	5	0	0	n.a.
Total	**62**	**7/62 (11.3%)**	**1/62 (1.6%)**	

3.2 Lake Mjøsa sediments

The method chosen to process freshwater sediment samples was successful for 81% of cores using density separation. The remaining 37 extracts needed additional steps to remove organic matter. Total sediment volume processed from each core slice varied per site. This was heavily dependent water content and the presence of organic material. Core slice weight ranged, on average, from 8.00 to 25.03 g^{-1} (d.w.) per site.

3.2.1 Sediment dating

It was beyond the scope of this current project to date the sediment cores taken in 2018. Dating was previously performed on sediment cores from Vingrom (Sites 4 – 6), Gjøvik (Sites 9 – 10), Skreia (Site 17) and Brumunddal (Site 11) in 2005 – 2006 using analyses of ^{210}Pb, ^{226}Ra and ^{137}Cs activity via gamma spectrometry (Fjeld et al., 2006). Assuming annual sedimentation rates of 0.25 –1 cm it was estimated that 1 cm equals 1 to 4 years. Based on the sedimentation previously reported for Lake Mjøsa using ^{137}Cs activity, the records that we present here probably go back to ca. 1980s. Due to variations across the lake which will likely influence sedimentation depth, it is not possible to provide more precise dating estimates for this report. Our cores from 2018 representing 0 – 8 cm are likely to extend back between 20 to 30 years.

3.2.2 Grain size analysis

Results for grain size analysis are presented in Table 6. Fine-grained sediments (<63 µm; silt and clay) dominate at the majority of the sites investigated. This includes samples outside of WWTPs (e.g., Sites 2 and 15), urban areas (e.g., Sites 11 and 13) and downstream of potential road runoff (e.g., Site 17). The batch of samples processed here (top core slice, 0 – 1 cm) does not allow for any deep statistical evaluation.

Table 6. Grain size distribution (< 63 µm) in 10 samples of top-layer (0 – 1 cm) sediments in Lake Mjøsa Site ID given in Figure 1.

ID site	Dry matter (%)	<63 µm (%)
2	44	84
4	18	67
8	21	52
9	27	56
11	15	70
12	21	53
13	23	80
15	13	69
17	23	66
18	32	49

3.2.3 Microplastics in Lake Mjøsa sediments

Microplastics were identified in sediment across all sites. There was variation in the number of particles both between sites and within sites (Figure 3). Highest microplastics values were reported at Site 13 and Site 4 (7.31 and 3.89 MPs g^{-1} respectively). Site 13 is located close to the urban area of Hamar where there is a high level of industry, road and boat traffic and Site 4 is located close to the Mjøsa bridge and main road (E6). The lowest microplastic value was reported at Site 17 (0.04 MPs g^{-1}) which was identified to represent an area in Lake Mjøsa with low levels of anthropogenic influence as it is in a natural sediment accumulation area. Interestingly, sediment from the urban area by Lillehammer (Site 3) contained fewer levels of microplastics. This is not surprising as it is situated close to the main river inlet of Gudbrandsdalslågen, depositing large amounts of glacial sediments in this area. Repeated and high-resolution samples would be required in this specific location to further investigate the deposition related to urban runoff.

When standardised to MPs g^{-1} (d.w) it is possible to compare sites within Lake Mjøsa at different core depths. The upper most sediment layer, 0 – 1 cm, generally appear to contain higher levels microplastics per site than the deepest core slice, 7 – 8 cm (Figure 4, more detail in Supplementary Information).

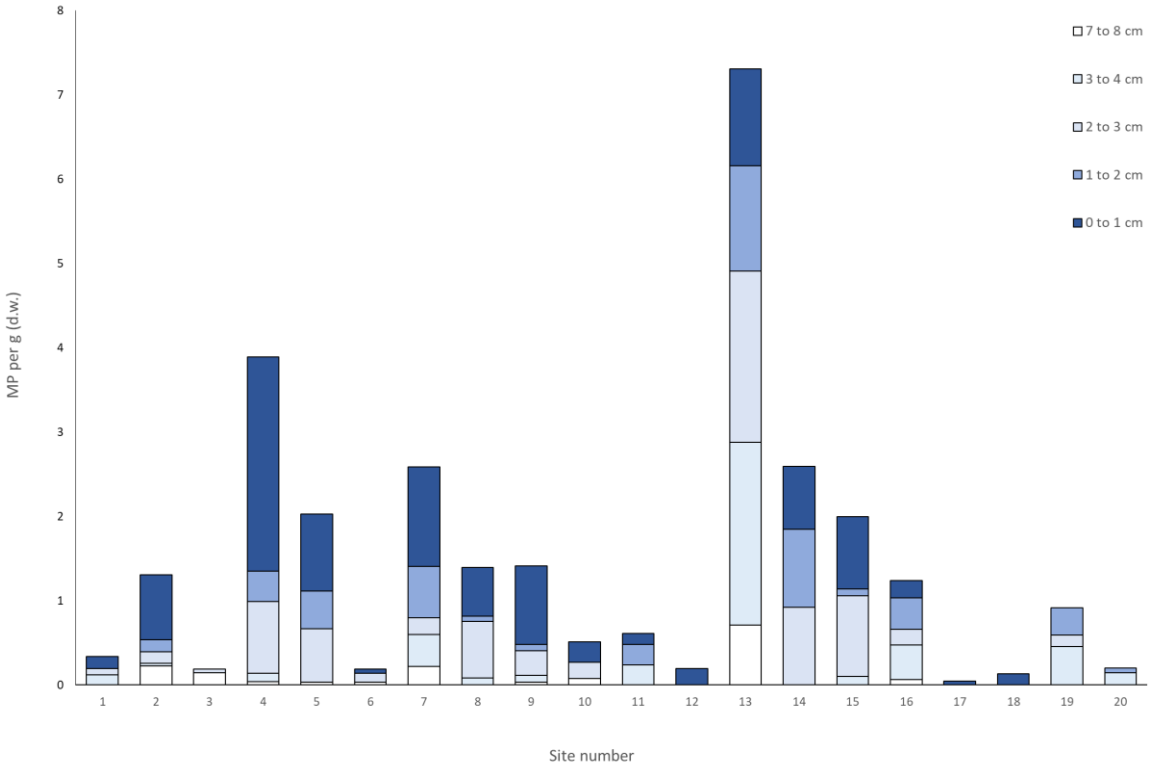

Figure 3. Distribution of microplastics (>36 µm) extracted from twenty sites within Lake Mjøsa. Data is presented as microplastics per gram (MPs g^{-1} d.w.). Bars are divided by core slice depth.

Below, each core slice is discussed in more detail in terms of particles found before and after polymeric verification (Table 7).

Core slice 0 – 1

The top 0 – 1 cm of all 20 sites were assessed for the presence of microplastics. Visual analysis suggested that all sites contained potential microplastics. Three sites were found to be free of microplastics after correction using FT-IR. These were Lillehammer (Site 3), Åkersvika (Site 14) and Minnesund (Site 20). Initially 161 particles were isolated during visual analysis. Particle composition consisted of fragments (53%), fibres (46%), beads (1%). Subsequent FT-IR analysis confirmed 101 particles as plastic and 1 particle as semi-synthetic.

Core slice 1 – 2

Slice 1 – 2 cm of all 20 sites were assessed for the presence of microplastics. Visual analysis suggested that 12 of the sites contained potential microplastics which were all confirmed following FT-IR analysis. Initially 114 particles were isolated during visual analysis. Particle composition consisted of fragments (64%), fibres (35%), bead (1%). Following FT-IR characterization, 79 particles were confirmed plastic and 2 particles were confirmed semi-synthetic.

Core slice 2 – 3

Slice 2 – 3 cm of all 20 sites were assessed for the presence of microplastics. Visual analysis suggested all but one of the sites contained potential microplastics. Following correction for FT-IR analysis, 15 sites contained microplastics. Initially 164 particles were isolated during visual analysis and particle composition included fragments (38%) and fibres (62%). Following FT-IR characterization, 96 particles were confirmed plastic and 7 particles were confirmed semi-synthetic.

Core slice 3 – 4 cm

Slice 3 – 4 cm of all 20 sites were assessed for the presence of microplastics. Visual analysis suggested 14 of the sites contained potential microplastics. Following correction for FT-IR analysis, 12 sites contained microplastics. Initially 131 particles were isolated during visual analysis and particle composition included fibres (56%) and fragments (43%). Following FT-IR characterization, 83 particles were confirmed plastic and 8 particles were confirmed semi-synthetic.

Core slice 7 – 8 cm

Slice 7 – 8 cm of all 20 sites were assessed for the presence of microplastics. Visual analysis suggested 11 of the sites contained potential microplastics. Following correction for FT-IR analysis, 10 sites contained microplastics. Initially 62 particles were isolated during visual analysis and particle composition included fibres (52%) and fragments (48%). Following FT-IR characterization, 35 particles were confirmed plastic and 4 particles were confirmed semi-synthetic.

Table 7. Identification of plastic particles following verification of visually accepted particles.

Core slice	Visual	Reject poor match	Reject organic	Lost during analysis	Plastic	Semi-synthetic
0 – 1 cm	161	14	29	16	101	1
1 – 2 cm	114	24	3	6	79	2
2 – 3 cm	164	21	34	6	96	7
3 – 4 cm	131	31	8	1	83	8
7 – 8 cm	62	11	6	6	35	4

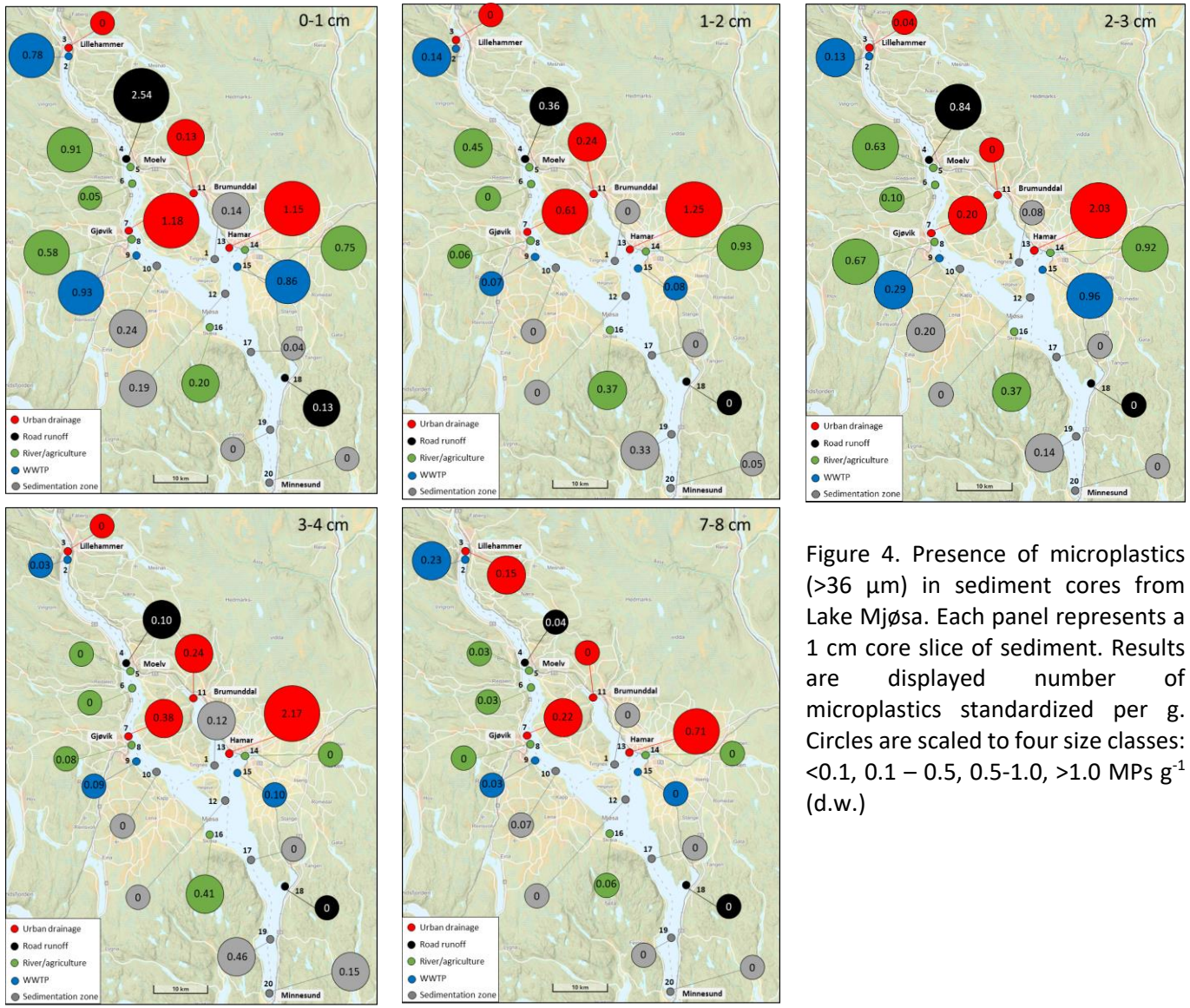

Figure 4. Presence of microplastics (>36 µm) in sediment cores from Lake Mjøsa. Each panel represents a 1 cm core slice of sediment. Results are displayed number of microplastics standardized per g. Circles are scaled to four size classes: <0.1, 0.1 – 0.5, 0.5-1.0, >1.0 MPs g^{-1} (d.w.)

3.3 Lake Femunden sediments

Ten sites within Lake Femunden were analysed for the presence of microplastics. When all core slices were summed together only sites 1, 6, 9, 10 from Lake Femunden did not contain microplastics (Figure 5). The number of particles per slice ranged from 0 to 2. Most particles were found in the top core slices (0 – 1 and 1 – 2 cm). Five particles were extracted from the top core slices, four were identified as viscose and polyamide (PA) fibres and one was identified as a PP fragment. Five particles were extracted from the middle core slices which contained four viscose fibres and one polyester fibre. A single viscose fibre was extracted from the third core slice from Lillehammer WWTP (Site 2). No other microplastics were found at this depth across all Lake Femunden sites.

3.4 Comparison between Lake Mjøsa and Lake Femunden

There was some uncertainty in the dry weight values for Lake Femunden. When results are corrected there were significantly fewer particles within Lake Femunden compared to Lake Mjøsa. (Figure 5). For example, the number of particles in Lake Mjøsa ranged from 0 to 7.31 MPs g^{-1} (d.w.) whereas number of particles in Lake Femunden ranged from 0 to 0.69 MPs g^{-1} (d.w.) Interestingly, all but one of the sediment accumulation areas within Lake Mjøsa showed similar values to the reference locations in Lake Femunden (Site 19, Morskogen). Furthermore, sites influenced by rivers, urban areas (incl. roads) and WWTPs had comparatively higher numbers of microplastics than all sites in Lake Femunden as well as sediment accumulation areas in Lake Mjøsa (Figure 5). The number of replicates per category did not allow further statistical testing of the differences between the lakes.

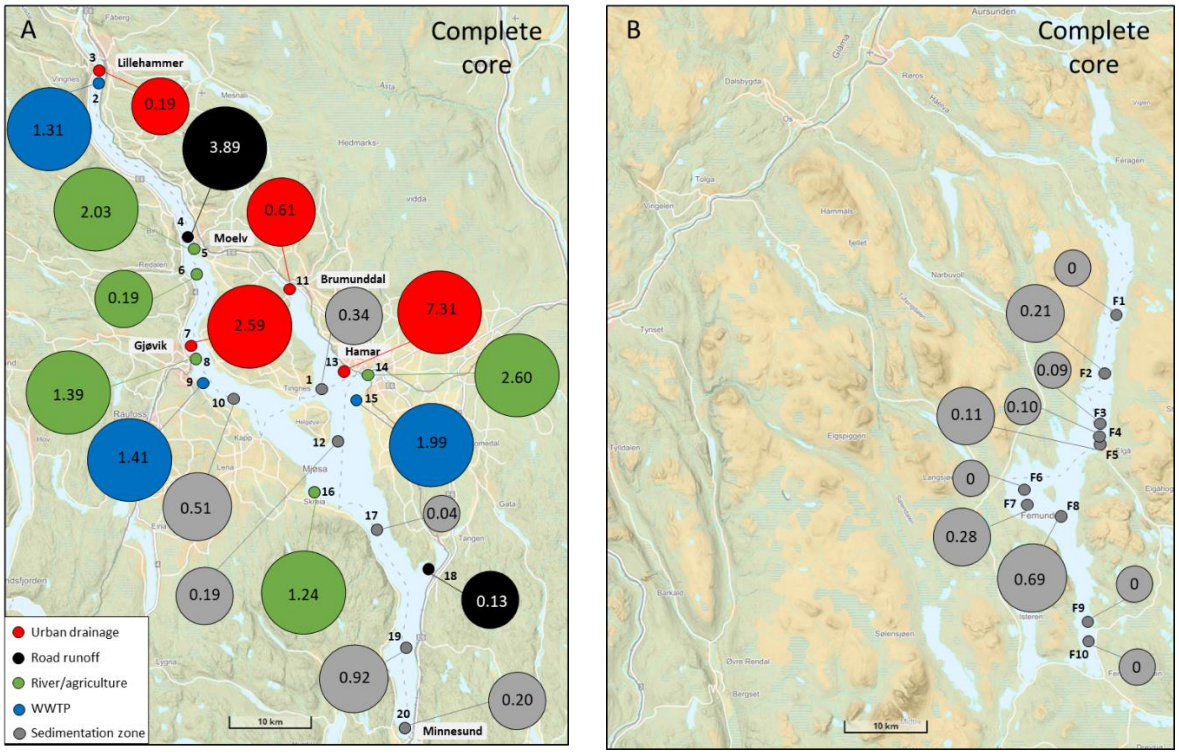

Figure 5. Microplastic (>36 µm) abundance in sediment from Lake Mjøsa (A) and Lake Femunden (B). Results are displayed as per complete core (0 – 8 cm) where each core slice was corrected based on sediment dry weight and the microplastic concentrations from each core slice added together. Circles are scaled to four size classes: <0.1, 0.1 – 0.5, 0.5-1.0, >1.0 MPs g^{-1} (d.w.) (MPs g^{-1} d.w.)

3.4.1 Summary of particle characteristics from Mjøsa and Femunden sediments

SHAPE: Particle shape was consistently dominated by either fragments (0 – 1 cm, 1 – 2 cm, 7 – 8 cm) or fibres (2 – 3 cm, 3 – 4 cm) in Lake Mjøsa sediments. In total, fibres accounted for 50%, fragments 49% and beads 1%. Conversely, Lake Femunden sediments contained mostly fibres (91%).

SIZE: Size distribution of plastics extracted from sediments within Lake Mjøsa show that 60% of plastics were less than 1 mm in size, 36% of plastics were between 1 – 5 mm in size and 4% of plastics were greater than 5 mm in size. There seems to be an equal variance across core depth as well (Figure 6). All particles extracted from Lake Femunden sediments were <350 µm in size.

COLOUR: Blue was the most prominent particle colour found in Lake Mjøsa (41%). Red was the second most common (21%) followed by green (17%) (Figure 7). Most of the particles in Lake Femunden were blue (58%) and black (33%).

POLYMER: The types of polymers identified varied across sites (Figure 8). Although classification by site, as well as category, is highly uncertain based on the scale of the project as well as local, and daily fluxes in water currents. However, there are some polymers to note. The most abundant polymers across all core depths were acrylic (31%) and polyester (incl. PET, 31%). Most acrylics and polyesters have high densities (e.g. PET, 1.38 g cm^{-3}) and their presence in sediment samples is not unsurprising. PA and PS were less abundant in deeper core slices. PVC was only found at Rambeck (Site 9). Two sites had a large variety of polymers identified, these were Gjovik (Sites 7) and Hamar (Site 13) which were categorised being influenced by locality to urban areas. On the other hand, Breili (Site 10) had a limited number of polymer types. This was categorised as being within a sedimentation accumulation area. This suggests that polymers identified could be related to the level of pollution in a given area. Further investigations are required.

POLYMER MASS: Pyrolysis GC-MS provided additional information on the mass of each polymer in the sample fraction from 36 – 75 µm (Table 8). Eight polymers are used in the initial analysis however results for only seven polymers are presented here. PVC was removed due to interference in the data set. The lower size fractions 36 – 75 µm across the six sites contained PET, PS, PE and a small amount of PC. It is not possible to compare the data obtained from Pyrolysis GC-MS to those obtained through FT-IR. When looking at the microplastic data based on particle number and mass it is hard to compare these numbers directly as different size fractions of particles were analysed.

Table 8. Results of Pyrolysis GC-MS from the size fraction 36 – 75 µm (µg $sample^{-1}$)

		Site 2	Site 4	Site 6	Site 12	Site 14	Site 20
Polyethylene	(PE)	240	1740	48	56	526	817
Polypropylene	(PP)	<1	<1	<1	<1	<1	<1
Polystyrene	(PS)	<1	48	<1	<1	<1	<1
Polyethylene terephthalate	(PET)	44	10	<1	22	144	24
Polyamide 6	(PA6)	<1	<1	<1	<1	<1	<1
Polymethylmethacrylate	(PMMA)	14	24	<1	<1	28	40
Polycarbonate	(PC)	1	<1	<1	<1	<1	<1
	Total	**301**	**1820**	**49**	**78**	**700**	**880**

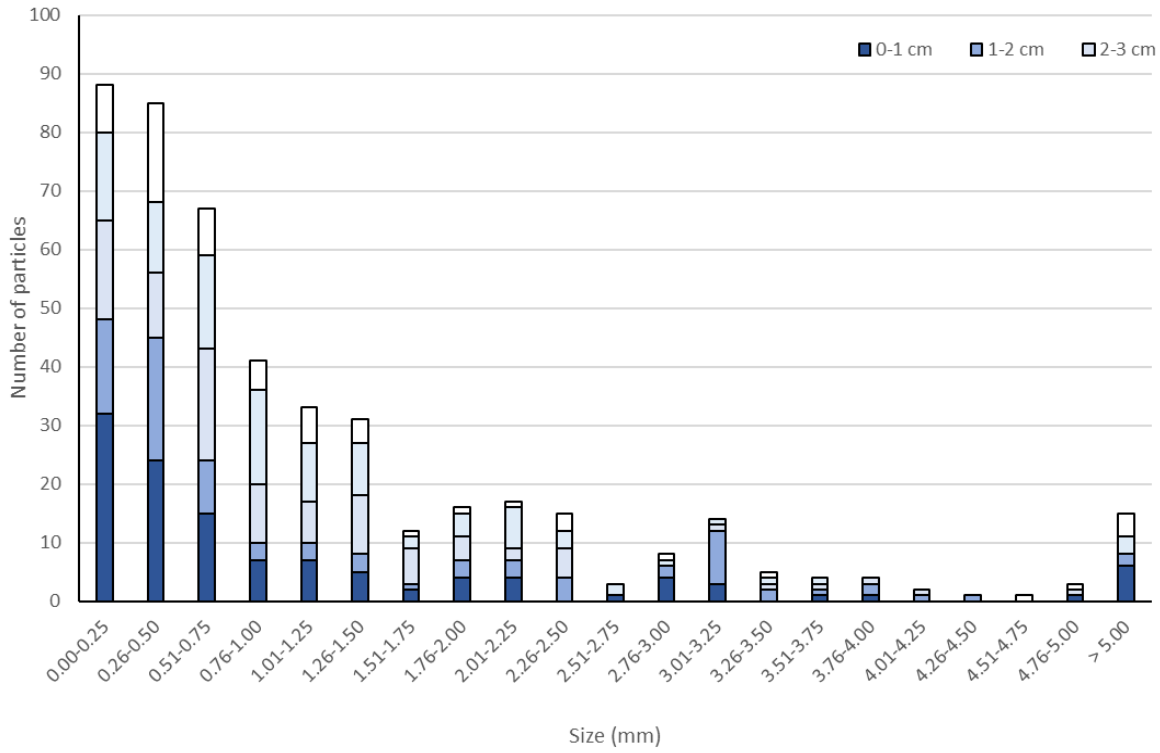

Figure 6. Size distribution of plastic particles >36 µm extracted from core slices (0 – 1, 1 – 2, 2 – 3 cm, 7 – 8 cm) from 20 sites within Lake Mjøsa.

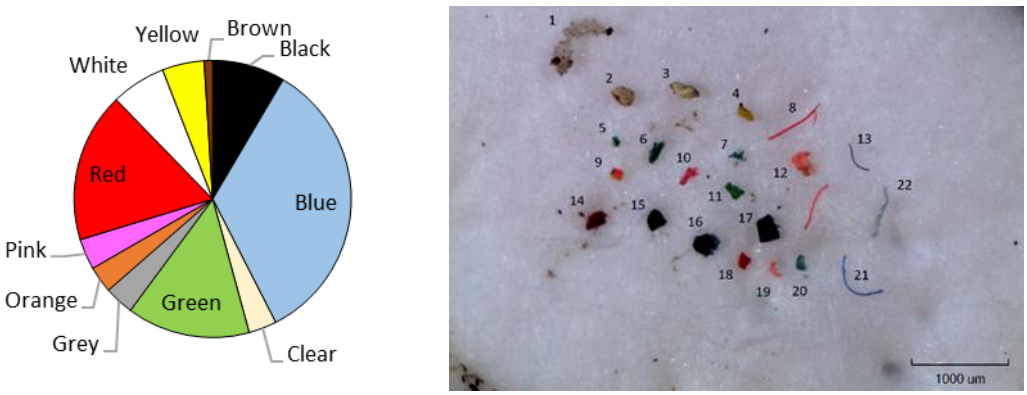

Figure 7. Colours of microplastics (>36 µm) found at all sites within Lake Mjøsa and example of microplastics extracted from a sediment core.

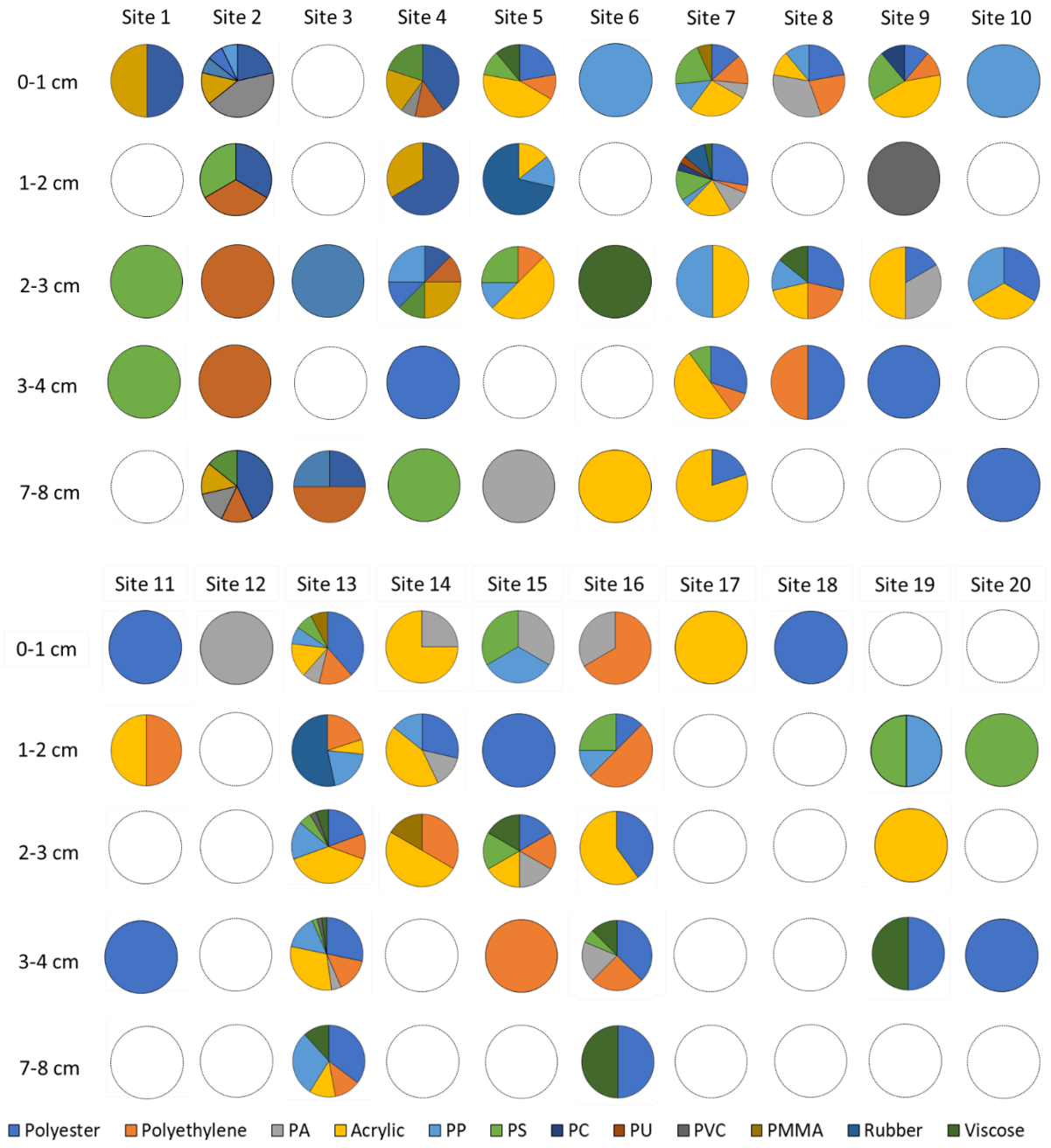

Figure 8. Polymer composition within each core slice from Lake Mjøsa.

3.5 Historical plankton samples

Twelve historical plankton samples were analysed for the presence of microplastics (Table 9). Fibres were observed in all samples, but they were not included in the analysis due to unknown levels of procedural contamination. Nine of the historical plankton samples contained plastic fragments ranging from 0 to 14 particle per sample. Fragments ranged in size from 81 µm to 1985 µm (mean: 294 ± 317 s.d.) in longest dimension and 36 µm to 420 µm (mean: 153 ± 89 s.d.) in the shortest dimension. Based on the longest dimension 97% would be classified as small microplastics <1 mm. Of the original 82 fragments isolated from the historical plankton samples, 61 were analysed with FT-IR (75%). The most abundant type of material isolated from plankton samples was rubber (inc. high-cis polybutadiene rubber, HBR). Other polymers included PVC, PE, polystyrene (PS) and PP (Figure 9).

Table 9. Historical plankton samples from Lake Mjøsa. Results are displayed based on the number of particles extracted from each sample and standardised to microplastics per litre (MPs l^{-1}). * Many small black fragments but below the detection limit

Code	Volume (l)	Fragments	Polymers	Fibres	MPs l^{-1}
HP_05.1973	225	14	Rubber, PVC, acrylic	Present	0.062
HP_09.1973	5652	0		Present	0
HP_09.1978a	225	1	Rubber	Present	0.004
HP_09.1978b	225	3	PE, PS, PVC	Present	0.013
HP_07.1981	225	1	PE	Present	0.004
HP_06.1989a	8478	10	Rubber	Present	0.001
HP_06.1989b	8478	12	Rubber	Present	0.001
HP_06.1991	8478	0		Present	0
HP_09.1998	225	6	PE, PVC, PP	Present	0.026
HP_10.2002	225	7	PE	Present	0.031
HP_10.2011	8478	0*		Present	0
HP_06.2017	225	7	PE, PP, PU, PS, PVC, Rubber	Present	0.031

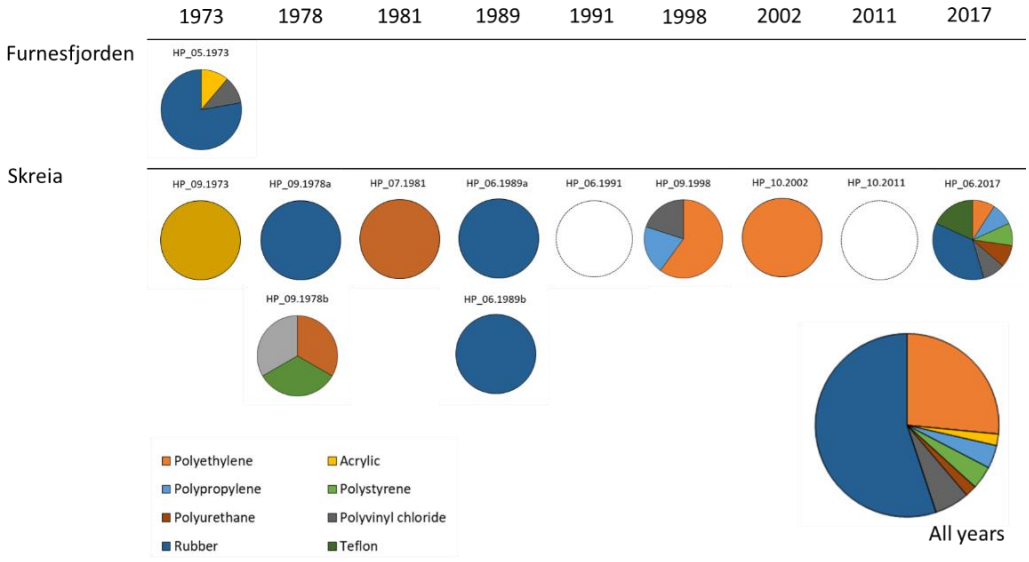

Figure 9. Polymer variation in historical plankton samples from Lake Mjøsa in the years from 1973 to 2017.

3.6 Duck mussels

Ten individual duck mussels were analysed for the presence of microplastics. Only three individuals contained a single potential plastic particle. Each particle was tested on FT-IR. An orange fragment (339 x 233 µm) was identified as rubber (0.69 match to HBR). The remaining two blue fibres were identified as cellulose (>0.68 match). Based on this information only the single mussel containing the fragment was included in the results.

4 Discussion

This is the first time that a freshwater system in Norway has been investigated through a large scale, coordinated survey for the presence of microplastics. Sediment samples from across Lake Mjøsa contained microplastics throughout different sediment depths. In general, the upper most core slices contained more microplastics. Sites which were categorized as potentially polluted due to locality to urban areas, WWTPs and riverine input contained greater numbers of plastics than sites which were identified as sediment accumulation areas. Sediment accumulation areas contained comparatively low numbers of microplastics when compared to Lake Femunden, which was chosen as a reference location due to its rural location away from anthropogenic inputs. Focusing on a specific form of anthropogenic input with repeated sampling will allow a more detailed analysis of the particle characteristics in future monitoring programs. Along with sediment accumulation, historical plankton samples appear to be a promising additional sample matrix to understand the temporal changes microplastics found in lake water columns. Based on current data, freshwater mussels appear to be promising as a sampling matrix, but it is recommended that repeated samples with an increased number of individuals from additional locations are investigated further. Other biota should be tested for suitability as a monitoring tool. Freshwater fish, including trout and perch may be an option as they are already being routinely sampled for other monitoring programs.

4.1 Suitability of methods

4.1.1 Sediments

Two methods of collection were used in this study, a sediment core (16/20 sites in Lake Mjøsa, all 10 sites in Lake Femunden) and a Van Veen grab (4/20 sites in Lake Mjøsa). Following careful equipment deployment and retrieval it was possible to obtained intact stratified sediment samples from all stations. The sectioning for sediment depth profile is a method that has been used for monitoring hazardous substances in sediment samples (*e.g.,* Olsen et al. 2018) as well as for monitoring microplastics in sediments (*e.g.,* Martin et al., 2017). This study showed that this is an appropriate approach for monitoring in freshwater sediments. It was not possible to retain the water-surface interface at some of the sites due to less compact sediments and larger grain sizes which affect the adhesive forces within the corer. Future methods should consider retaining this layer to look at potential settling and release of particles.

The sediment processing method was effective but did require alteration to the originally proposed methods. Twenty-eight of the core slices (28/100) required further processing due to large quantities of organic matter. Fenton's solution (30% H_2O_2 with Fe catalyst) was employed as an additional processing step and had to be repeated a second time for a some of the samples (Table 10). After OMR all core slices could be analysed for the presence of microplastics.

4.1.2 Water samples

NIVA wanted to include samples of the water column in this project using a high-volume freshwater pump which would allow us to filter 10,000 litres over a short period of time. However, due to technical issues with this pump at the time of sampling we were not able to collect samples. This method should still be considered for further investigations of microplastics.

Table 10. The effectiveness of using density separation and organic matter removal for sediments from the freshwater environment. n = total filter papers

Core slice	OK after Density	OK after OMR x1	OK after OMR x2
0 – 1 cm (n=40)	34/40	2/6	4/4
1 – 2 cm (n=40)	31/40	6/9	3/3
2 – 3 cm (n=40)	35/40	1/5	4/4
3 – 4 cm (n=40)	28/40	7/12	5/5
7 – 8 cm (n=40)	35/40	3/5	2/2

4.1.3 Historical plankton samples

It was effective to process historical plankton samples and further archived samples should be investigated to look after additional sites and years. Plankton is being collected in several ongoing monitoring projects such as MILFERSK (zooplankton, Lake Mjøsa) and ØKOSTOR (phyto- and zooplankton in 26 different lakes) and microplastic analyses may easily be included in these programs in the coming years.

4.1.4 Biota

Previous research has shown that mussels are a promising bioindicator for the smallest sized microplastic (<1 mm) in the marine environment (Bråte et al. 2018; Li et al., 2019). This is due to their ecology, ease of sampling, the standardised sample processing and further analysis. However, not enough replicate mussels were used in this study to make the same assessment of their use in freshwater ecosystems. Observations during sample processing included a large amount of organic and inorganic matter present in many of the individuals. This is likely due to them being collected from a submerged position from benthic sediments. The choice of sampling sites for mussels should be carefully considered. Furthermore, duck mussels (*Anodonta anatina*) are a motile species and an understanding of their population movements should be taken into account. It might be necessary to consider additional species of biota. Originally, we had tried to obtain sediment dwelling worms, but these were not present in the samples. Fish might also be a suitable monitoring species for freshwater ecosystems. Fish have been shown to ingest microplastics in the marine environment (Lusher et al., 2013) and several publications have emerged describing microplastics in fish from European streams and rivers (Eerkes-Medrano and Thompson, 2018). Furthermore, data from Oslo rivers shows that the same methods can be applied (Garmo et al., 2018). Lake Mjøsa supports a rich diversity of fish including trout, perch and roach. Stomach and/or intestine samples of these species should be considered for future investigations. It is easy to collect both benthic feeding and pelagic fish within ongoing monitoring program such as MILFERSK (Jartun et al., 2018).

4.1.5 Sample analysis

As mentioned in a previous NIVA report: **"***Classifying plastics based on their shape, size and polymer type is appropriate for monitoring surveys, but colour should not be used as the primary identification parameter due to the subjectivity of visual colour. If used, more robust colour identification should be implemented*" (Lusher et al., 2017).

In future monitoring programs sample collection and processing should be modified to target smaller particles, depending on the aim of individual projects. This project was able to work with a lower limit of 75 µm with µFT-IR and with the addition of Pyrolysis CG-MS we could work with smaller size fractions >36 µm. Depending on the aims of the project particle size and/or mass should be considered.

Currently optimised methods favour descriptions of particles based on size, shape and polymer although the use of Pyrolysis GC-MS to get mass balances is promising. It is not possible to ascertain how many particles results from Pyrolysis represents, or the shape and size. This information may be of interest when investigating the fate of plastics and the potential uptake by organism which is heavily reliant on particle shape characteristics. In cases where impact on the environment is investigated, it will be preferable to focus on individual particles rather than total mass.

4.2 Possible sources of microplastics

It is important to note that there are multiple diffuse sources of input to freshwater environment ranging from agricultural to urban areas. This makes inferring microplastic sources extremely difficult. There were numerous polymers identified in this study. It is not possible to identify the exact sources of these microplastics, but it is possible to discuss possible sources based on polymer classification. Many of the common polymers have specific applications. For example, polyesters are commonly used in textiles. Sources of polyesters could be related to release of fibres from WWTPs plants (*e.g.,* Kay et al., 2018), but may also be related to atmospheric deposition to the lake surface (*e.g.,* Dris et al., 2013). Acrylic, most commonly PMMA, is used in textiles and paint. Some potential sources may be the WWTPs as well as release from urban environments, or marinas and vessels operating in the area. Polyesters and acrylic have high densities and their presence in sediments in not surprising as they sink in freshwater.

It is not surprising that PP and PE were found in this study as they are two of the most common polymers in terms of production and those found in the marine environment. These polymers are used for many different applications from general use plastics (plastic bags and bottles, clothing) to primary microplastics incorporated into consumer products. PP and PE have low densities and float in sea water although biofouling facilitates sinking. There are a number of different rubber polymers including polybutadiene rubber (HBR), styrene butadiene rubber (SBR) and neoprene. Again, the uses and therefore sources are highly varied and may include road and tyre wear, industrial and recreational activities along the lake.

4.3 Comparison to other freshwater investigations

It is difficult to compare sediment microplastics loads from this investigation to previous studies. No previous studies have used sediment cores to investigate microplastic distribution within freshwater sediment. Furthermore, previous riverine and lake investigations have been focused on surface waters and WWTP effluents (Li et al. 2018). Nevertheless, previous lake investigations have looked at variables influencing microplastic presence. These include surface area, catchment areas, elevation, remoteness and lake use (Eerkes-Medrano and Thompson, 2018). North American research has focused heavily on the Great Lakes which are larger than Norwegian and European lakes.

This appears to be the first study to investigate historical plankton samples from a freshwater lake. Previous studies have found microplastics inside freshwater fish around the world, but the number of investigations is still limited compared to the marine environment (Eerkes-Medrano and Thompson, 2018).

4.4 Recommendations for monitoring in freshwater ecosystems

4.4.1 Investigate temporal trends microplastics

Sediment cores can provide a description of microplastics incorporated into sediments. Cores can be dated to investigate settling and may show changes in microplastic content in relation to plastic use and release over time. It is recommended that a study which includes replicate samples with sediment dating and microplastic analysis is carried out.

Historical plankton samples can facilitate an investigation into the presence of microplastics in samples which were collected before the onset of microplastic investigations. This is a valuable tool to monitor whether there has been a change in the types of particles over time. Furthermore, ongoing plankton monitoring programmes could incorporate microplastic monitoring during future sampling campaigns.

Long-term or continuous monitoring will generate a database that will provide a better understanding of distribution and fate in the environment

4.4.2 Investigate spatial trends in microplastics

Utilising a single matrix alone is not suitable for monitoring microplastics. A combination of water, sediment and biota are recommended. Twenty sites with varying levels of potential anthropogenic input were investigated in this study. It is recommended that future monitoring is focused on a specific form of anthropogenic input with repeated sampling. This will allow a more detailed analysis of particle characteristics. For example, focusing on the urban areas of Hamar and Gjøvik where greater number of microplastics were observed.

4.4.3 Investigate interactions with biota

Biota are a suitable monitoring tool when studies aim to investigate interaction between biota and microplastics. Sessile organisms are recommended for this purpose as they can represent a site-specific impact. Sediment dwelling organisms or fish in lakes can provide information of lake populations. As they are motile, they would not be suitable to compare different locations within the same water body. Fish may be a promising monitoring tool as they are already collected for ongoing monitoring programs. Furthermore, their stomachs and intestines are not currently utilised, and data collected will be complementary to analysis of other pollutants.

4.4.4 Investigate influence of environmental variables

It is very likely that environmental variables influence the distribution of microplastics within freshwater ecosystems. Variables include hydro-chemical, hydro-physical conditions, atmospheric condition and atmospheric deposition. In the current study these environmental variables were not investigated and should be considered in the future.

5 Conclusion

In this baseline study, the presence of microplastics in two of Norway's largest lakes has been confirmed. Microplastics (>36 µm) were found throughout sediment cores in Lake Mjøsa and Lake Femunden and there appeared to be differences related to the source of anthropogenic input. Sites identified as sedimentation accumulation areas in Lake Mjøsa had lower levels of microplastics than sites identified in close to urban locations, rivers and WWTPs. In addition, archived historical plankton samples proved a useful tool to investigate temporal trend in microplastic abundance. The methods and results presented here will provide a foundation for future microplastic monitoring in the freshwater environment of Norway. A combination of sediment, water and biota monitoring is recommended to be incorporated into already established monitoring programs, albeit gradually, to get a better understanding of microplastic distribution both geographically and between different matrixes (such as water, sediment and different biota). Long-term, continuous monitoring will eventually generate a knowledge base and database that provides a much better basis for assessing both the fate of microplastics and the effects they may have on the organisms

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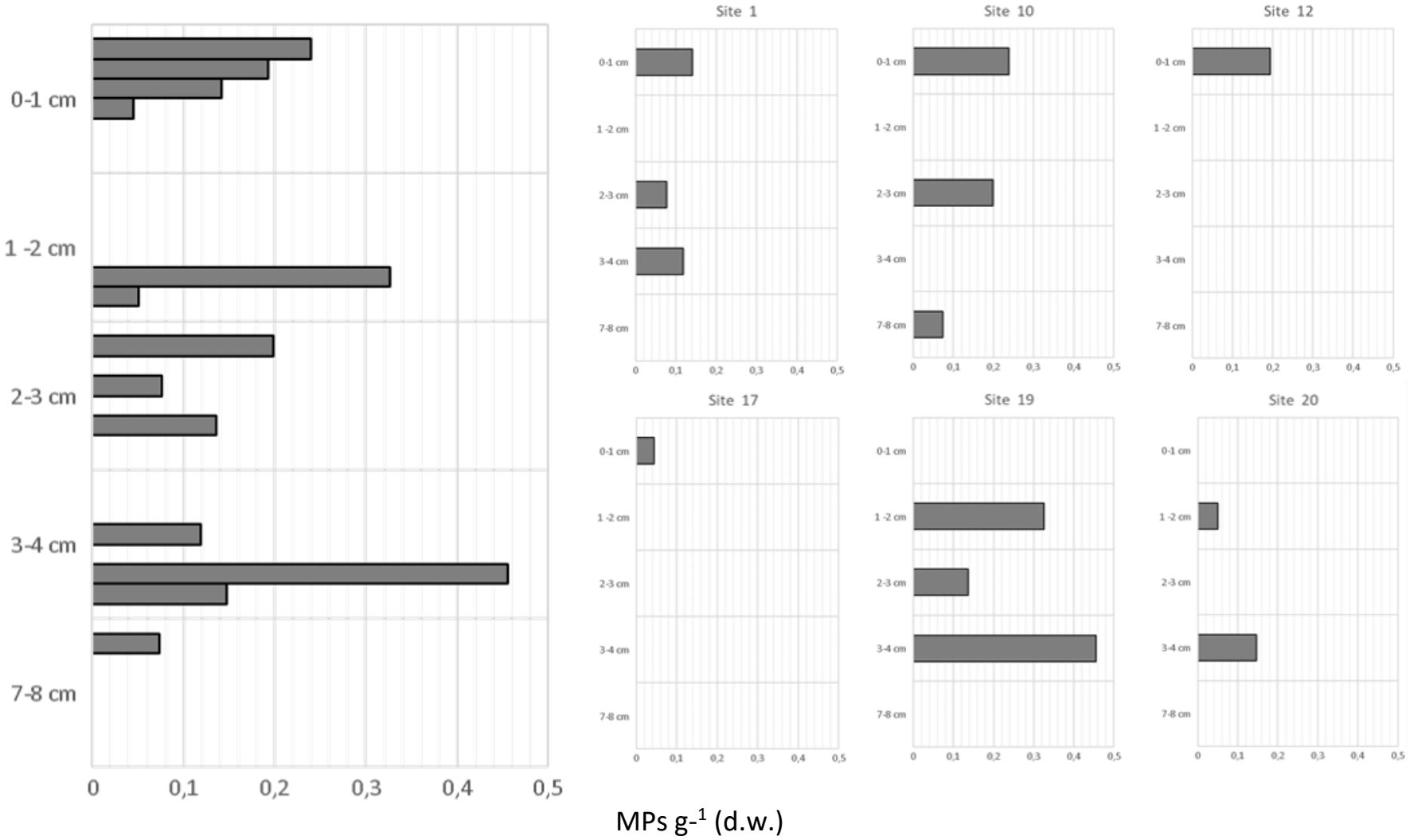
7 Supplementary information

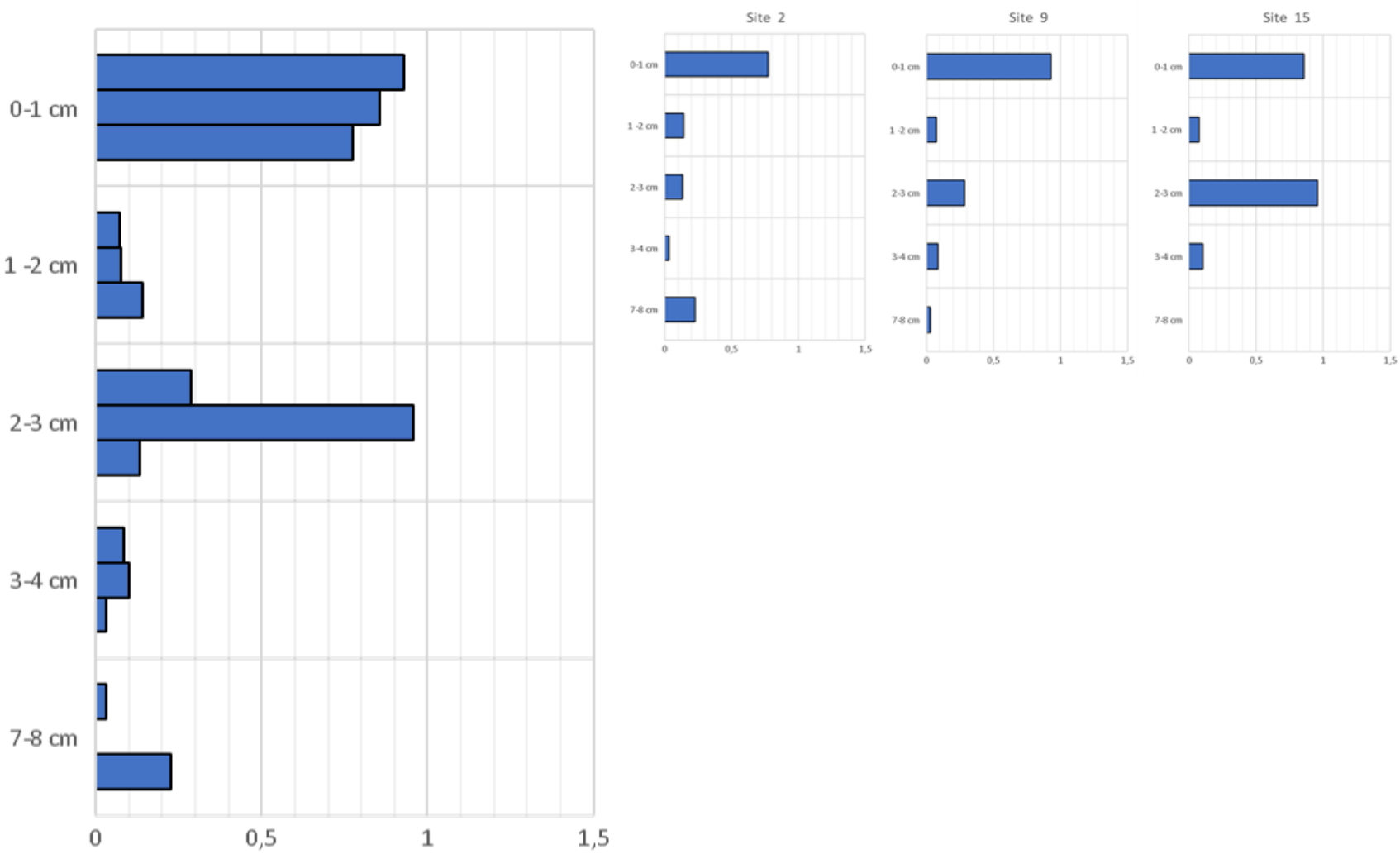
Comparison of data from sites divided into potential influence:

A) Sediment accumulation area
B) Waste water treatment plants (WWTPs)
C) River drainage
D) Urban drainage
E) Roads

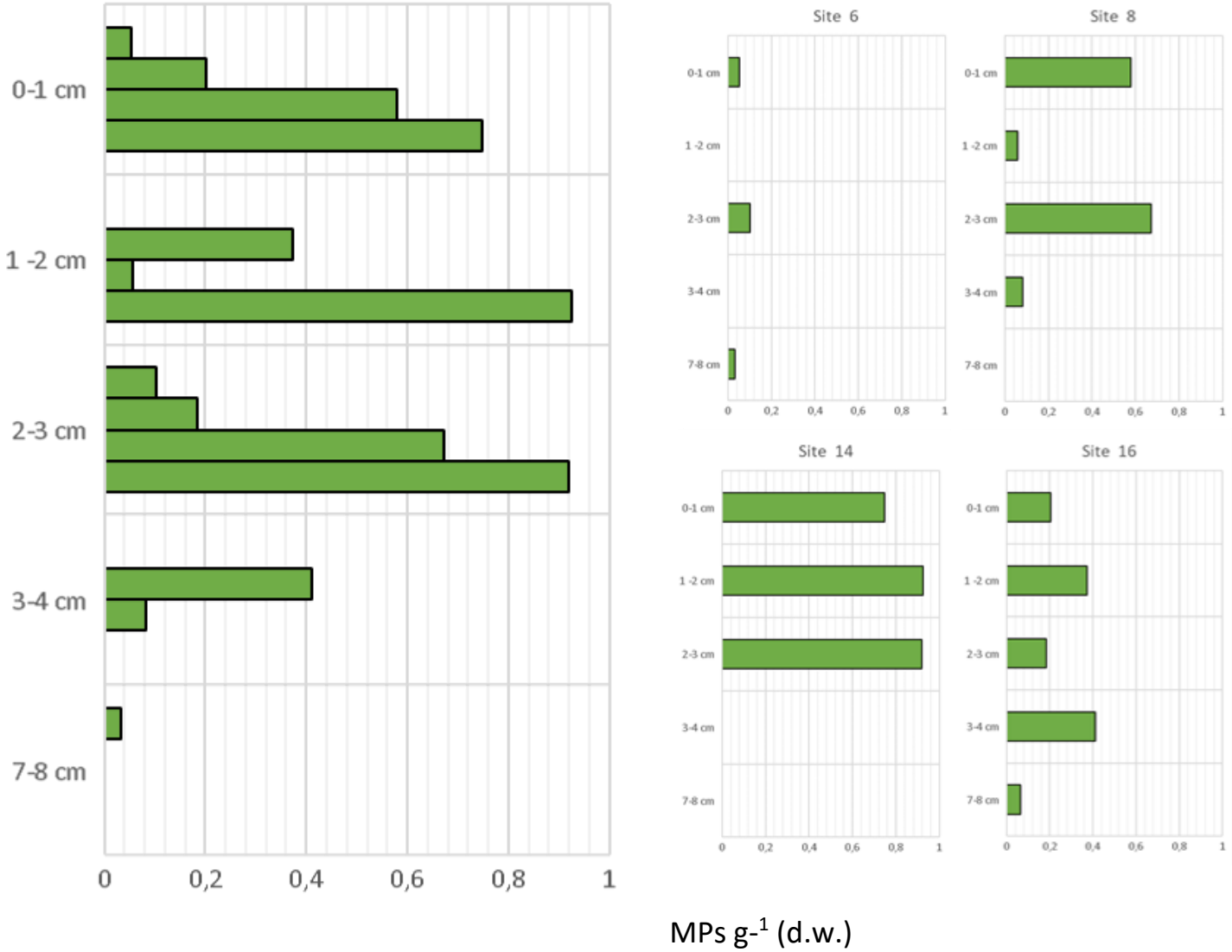
A) Sediment accumulation area



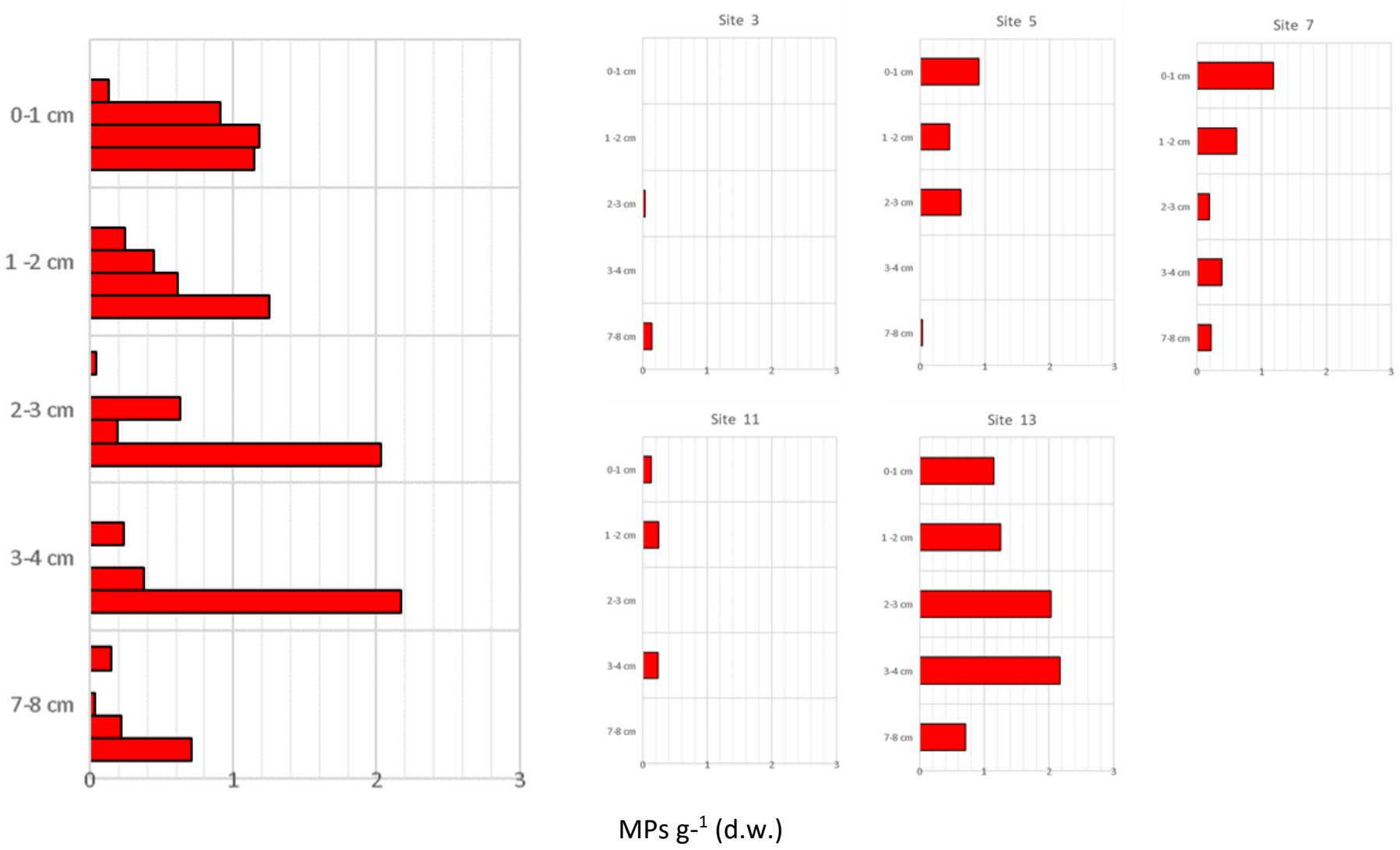
B) Waste water treatment plants (WWTPs)



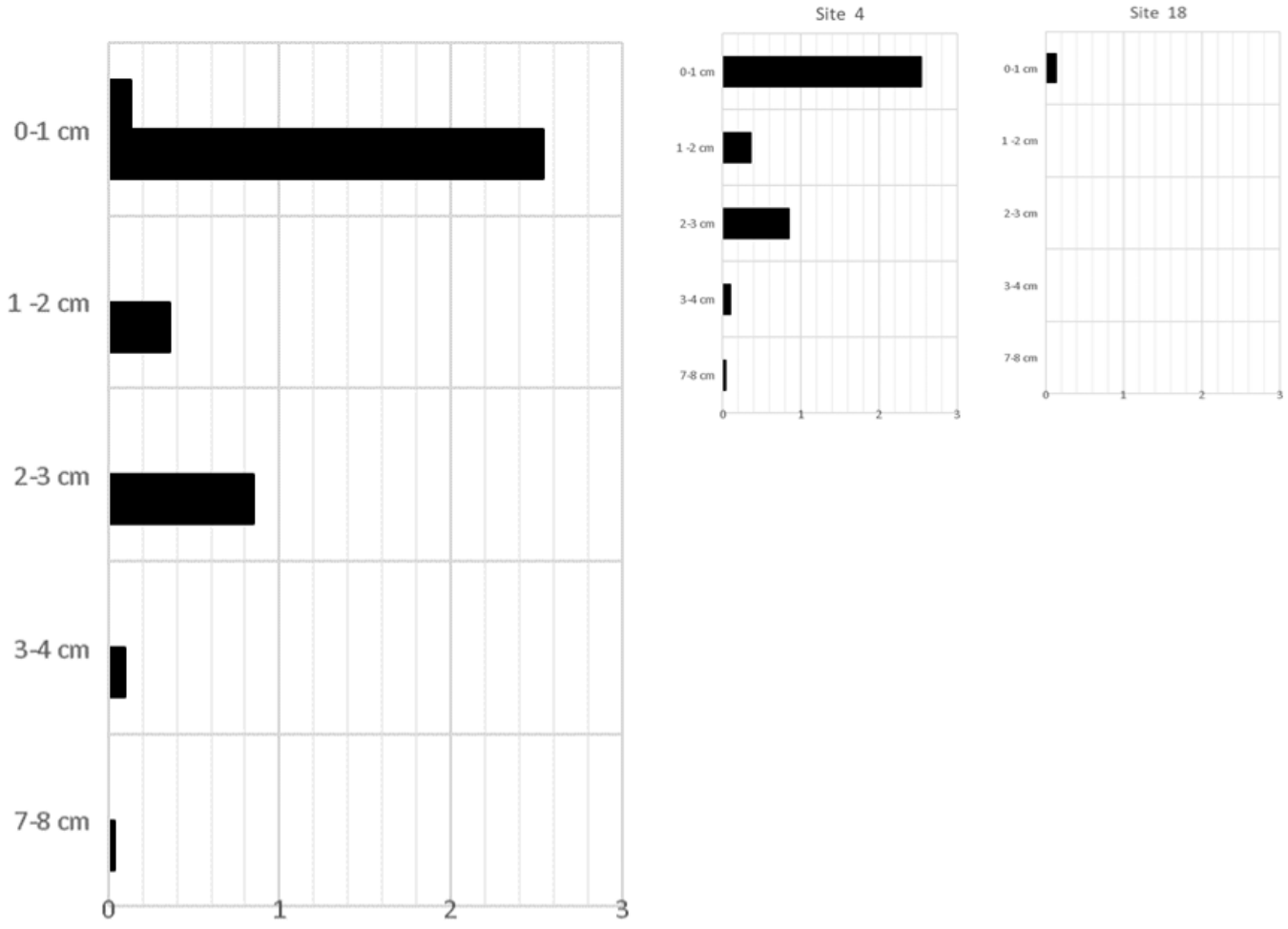
C) River drainage



D) Urban drainage



E) Roads



MPs g^{-1} (d.w.)

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