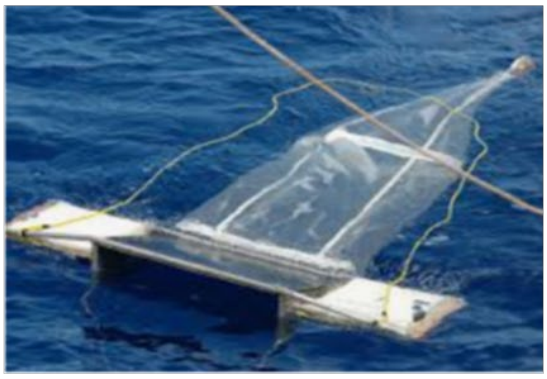
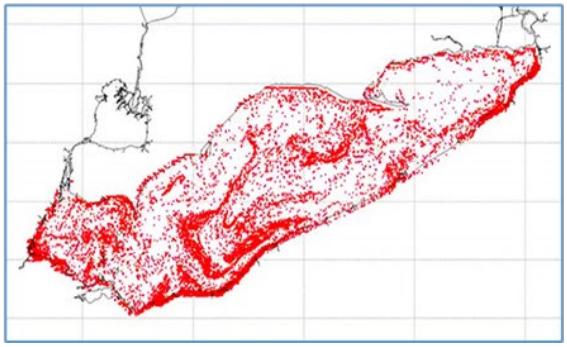
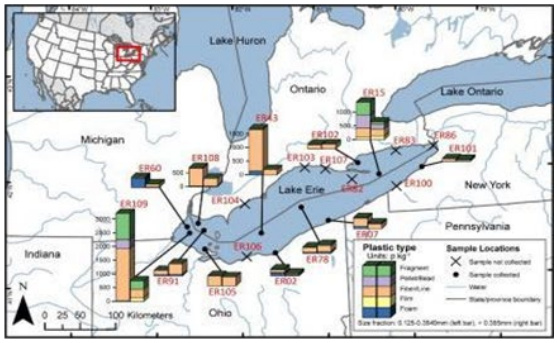
Great Lakes Microplastics Monitoring and Risk Assessment

Literature Review and Synthesis

September 5th, 2023 (Revised June 5th, 2024)







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Cover Images

Top left: A collection of microplastic particles. Source: https://www.flickr.com/photos/oregonstateuniversity/21282786668 (Image by 5Gyres, courtesy of Oregon State University, License: Attribution-ShareAlike (CC BY-SA 2.0))

Top right: Researchers using a manta tow, a net that collects sea surface samples, during SEAPLEX. Taken on August 16, 2009. Source: https://schmidtocean.org/wp-content/uploads/NOAA-manta_net_600.jpg
NOAA Okeanos Explorer Program https://www.flickr.com/photos/scrippsocean/3856797572
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Lower left: Matthew Hoffman, (https://www.rit.edu/directory/mjhsma-matthew-hoffman) assistant professor in RIT's School of Mathematical Sciences, used computer simulations to follow the volume of plastic debris moving across state and international boundaries—from Illinois to Michigan and from Canada to the United States (https://www.rit.edu/news/researchers-study-plastic-pollution-great-lakes).
Video version: https://phys.org/news/2018-08-tons-plastic-trash-great-lakes.html
https://www.youtube.com/watch?v=VrySaUqbtV4 More videos here, including a 3D version (shallow and deep in different colors) for Lake Erie: https://www.youtube.com/@mattyhoff14/videos

Lower right: Figure 2 from: Lenaker, P.L., Corsi, S.R. and Mason, S.A., 2020. Spatial distribution of microplastics in surficial benthic sediment of Lake Michigan and Lake Erie. *Environmental Science & Technology*, *55*(1), pp.373-384.

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List of Acronyms and Abbreviations

ATR – attenuated total reflectance
°C – degrees Celsius
CA SWRCB – California State Water Resources Control Board
CASRN – Chemical Abstract Service Registration Number
CMC – contaminant of mutual concern
CPE – chlorinated polyethylene
ECCC – Environment and Climate Change Canada
ECHA – European Chemicals Agency
EGLE – Michigan Department of Environment, Great Lakes, and Energy
EPS – expanded polystyrene
EVA – ethylene vinyl acetate
FTIR – Fourier transform infrared spectroscopy
g/cm^3 – gram per cubic centimeter
GLNPO – Great Lakes National Program Office
GC/MS – gas chromatography/mass spectrometry
GLWQA – Great Lakes Water Quality Agreement
HDPE – high-density polyethylene
IJC – International Joint Commission
kg – kilogram
km^2 – square kilometer
L – liter
LAMP – Lakewide Action and Management Plan
LDPE – low-density polyethylene
m^2 – square meter
m^3 – cubic meter
mm – millimeters
MT – metric ton
nm – nanometer
NOAA – National Oceanographic and Atmospheric Administration
OMECP – Ontario Ministry of Environment, Conservation and Parks
PTFE – polytetrafluoroethylene
PVC – polyvinyl chloride
PYR – pyrolysis
QA/QC – quality assurance/quality control
SBR – styrene-butadiene rubber

SCCWRP – Southern California Coastal Water Resources Project
SEM/EDS – scanning electron microscopy with energy dispersive X-ray spectroscopy
SOGL – State of the Great Lakes
SPT – sodium polytungstenate
ToMEx – Toxicity of Microplastics Explorer
µm – micrometers
UNEP – United Nations Environment Programme
USEPA – United States Environmental Protection Agency
USGS – United States Geological Survey
UV – ultraviolet
WPO – wet peroxide oxidation
WWTP – wastewater treatment plant
XPS – extruded polystyrene
XRF – x-ray fluorescence

1. Introduction

Plastics are a ubiquitous material in modern life due to their versatility, durability, and low cost. Global plastic production and consumption has increased dramatically since the mid-20th century and is expected to continue increasing (UNEP, 2021). According to the United Nations Environment Programme (UNEP), approximately 9.2 billion metric tons (MT) of plastic have been produced between 1950 and 2017, with more than half of this amount produced since 2004. Over 400 million MT of plastic were produced in 2020, and annual production is expected to increase to over 1,100 million MT by 2050. Only around 10% of these plastics are recycled and 14% incinerated, with the remaining 76% ending up in landfills or discarded into the environment (UNEP, 2021; Geyer et al., 2017). The inherent durability of plastics coupled with rising production rates and improper disposal methods is causing substantial accumulation of plastic in the environment.

Microplastics, which generally include plastic particles less than 5 millimeters (mm) in size, have gained increased attention due to their prevalence in the environment. Due to their small size, microplastic particles are easily transported by wind, ocean currents, stormwater, and biota and have been found all over the world in marine, freshwater, terrestrial environments, biota, and even in the atmosphere (ITRC, 2023).

Depending on the source, microplastic particles may be classified as primary or secondary microplastics (ITRC, 2023). Primary microplastics are tiny plastic particles that were designed specifically for their use in industrial and commercial products. Secondary microplastics are generated through the chemical and physical degradation of larger plastic products, such as water bottles, plastic bags, cigarette butts, paints, plastic sheeting, synthetic textiles, and tires. Plastic debris in the environment breaks down into numerous smaller secondary microplastic particles due to a combination of mechanical weathering, chemical reaction, ultraviolet (UV) radiation, and biodegradation (PlasticsEurope, 2019). There are also numerous pathways by which microplastics enter the environment (Rochman et al., 2019; ITRC, 2023). Urban stormwater runoff can transport plastic litter, abrasion dust from car tires, and road paint. Wastewater effluent may include microbeads from personal care products and microfibers from textiles. Agricultural runoff may incorporate microplastics degraded from greenhouse films, plastic mulch, irrigation systems, and planters. Other sources of microplastics include spills during manufacturing and shipping, runoff from recycling facilities and landfills, and discarded fishing gear.

Our understanding of the impacts of microplastic contamination on humans and wildlife increases every year. Today, scientists generally agree that microplastics can have adverse effects on organisms (Bucci et al., 2020). The mechanisms of effects remain less clear. The effects of microplastics on aquatic ecosystems can vary since plastics come in many different shapes and sizes, and can have diverse chemical makeups, which include their base polymer, microstructure, and chemical additives (Thornton-Hampton et al., 2022a; Rochman et al., 2019). Several laboratory studies have detected effects on organisms including tissue inflammation, changes to gene expression, reduced growth and feeding, decreased reproductive output, and increased mortality, while others did not detect any effects (reviewed in McIlwraith and Rochman, 2020 and Mehinto et al., 2022). Plastics can also leach additives or sorb contaminants present in the environment and act as vectors for other potentially toxic compounds.

The Great Lakes ecosystem contains 84% of the available freshwater in North America; is home to 3,500 plant and animal species; and supports sectors such as fisheries, industry, tourism, and recreation in

both Canada and the United States (USEPA, 2023a; GLC, 2023). The Great Lakes Water Quality Agreement (GLWQA) (IJC, 2012) is an agreement between the governments of the United States and Canada, which was amended in 2012 to better manage current environmental issues and prevent emerging issues from threatening ecosystem health and water quality within the Great Lakes. Mandated under the GLWQA, the State of the Great Lakes (SOGL) reports are published every 3 years and use 9 indicators, supported by 45 sub-indicators, to evaluate progress towards the general objectives of the GLWQA and report on status and trends.

In 2016, the bi-national International Joint Commission (IJC) recognized the importance of the microplastics problem, held a workshop with 33 experts, and issued a report (IJC, 2017). The IJC report provided recommendations on science, pollution prevention, and education and outreach, including the following:

> *"The Parties should jointly undertake monitoring, science and research initiatives for a binational assessment of microplastics in the Great Lakes to inform decision-making by (1) developing and/or adopting standardized sampling and analytical methods (2) developing a transport model to determine the sources and fate of microplastics (3) assessing potential ecological and human health impacts and (4) investing in research for source reduction, improved recycling, and reduced release of plastic pollution."*

The IJC's Great Lakes Water Quality Board also maintains a watching brief on the topic of microplastics. This watching brief, last updated in May 2022, is a living document summarizing the current state of the microplastics issue and developments related to the management of microplastics within the Great Lakes basin. IJC makes this watching brief publicly available online here: https://ijc.org/sites/default/files/WQB_MicroplasticsWatchingBrief_May2022.pdf.

McIlwraith et al. (2023; based on the 2020 report by McIlwraith and Rochman for ECCC) proposed that plastic debris be used as a "Toxic Chemicals" sub-indicator in the SOGL reports to aid evaluation of General Objective #4 of the GLWQA: *"Be free from pollutants in quantities or concentrations that could be harmful to human health, wildlife or organisms, through direct exposure or indirect exposure through the food chain."* Another alternative to establishing microplastics "Toxic Chemicals" as a sub-indicator would be to add microplastics as a Contaminant of Mutual Concern (CMC) under Annex 3 of the GLWQA and report on microplastic levels in various media, similar to reporting on mercury levels in water, sediment, fish, and herring gull eggs. Note that all CMCs are included as Toxic Chemical sub-indicators, but not all Toxic Chemicals sub-indicators are designated as CMCs. Additionally, the McIlwraith et al. (2023) report characterized the status of plastic pollution in the Great Lakes and suggested strategies and metrics for tracking and reporting plastic pollution.

This IJC report builds on McIlwraith and Rochman (2020) and Earn et al. (2021) to synthesize recent advances and knowledge of plastics pollution relevant to the Great Lakes. The specific objectives of this report are to:

1. Provide background on microplastics and propose a standard definition for microplastics in the Great Lakes basin.
2. Review sampling and analytical methods for microplastics and make recommendations on harmonized methods to improve the quality and comparability of results.
3. Synthesize available microplastics monitoring data across the Great Lakes basin.

4. Synthesize research on the ecological effects of microplastics.
5. Review risk assessment methodologies and suggest strategies for development of a risk assessment framework for microplastics.
6. Review existing Great Lakes monitoring policies and programs to assess how they may be modified to include microplastics.

1.1 Methods

This literature review for microplastics monitoring data in the Great Lakes builds on the work of McIlwraith and Rochman (2020), who reviewed and cataloged 34 journal articles and 12 government reports and extracted and entered data from 28 selected papers into a database. McIlwraith and Rochman (2020) cataloged all papers published through November 2020 by searching for the terms "Great Lakes" and "plastic" and "Great Lakes" and "microplastic" in Web of Science (all databases) and Google. Following their initial review, the authors updated their database by reviewing additional papers published from December 2020 through November 2022 which added 12 new papers with extractable data (McIlwraith et al., 2023). The current study builds on their efforts by including more recent studies on microplastics monitoring in water, sediments, and aquatic and riparian species; by including additional information on field and laboratory methods and quality assurance/quality control (QA/QC) procedures; and by including a review of literature on the ecotoxicology of microplastics to freshwater aquatic species.

For this study, a literature search was conducted by searching for keywords "microplastics" and "Great Lakes" in Google Scholar and ScienceDirect. The search was conducted in March 2023, and articles published after November 2022 that were not included in the McIlwraith et al. (2023) study were added to the database. Relevance was assessed by reviewing the title and abstract and scanning the body of the paper, if necessary. References cited in these articles were also reviewed and accessed based on their relevance to the Great Lakes; non-Great Lakes articles were reviewed and included if they focused on broadly applicable monitoring and sampling methods or recommendations, or discussed other topics that were not covered by Great Lakes-specific studies.

Five relevant studies (from November 2022-March 2023) were identified through our literature review and added to the reviews from the work of McIlwraith and Rochman (2020) and McIlwraith et al. (2023); three of the five new studies reported monitoring data and were added to the database, for a total of 43 papers that performed monitoring in the Great Lakes. Of these 43 papers, 39 had data that were extractable (see Appendix A.1 for full list of papers). Four papers (Damien and Frasier, 2020; Holland et al., 2016; Lewis et al., 2000; Zbyszewski & Corcoran, 2011) were determined either to not be applicable to this project during review or their data was included in other papers that were already extracted. The extracted microplastics monitoring data included sampling locations and methods, concentrations, size distributions, and material types. Microplastics monitoring data were classified by matrix as either water, sediment, biota, or shoreline debris (e.g., beaches). Data were collected through a combination of summarization of raw data and estimations from figures and tables presented in published reports where raw data were not provided. Studies that reported surface water concentrations were not consistent in the units used, some reported in particles per square kilometer (km^2), while others used particles per cubic meter (m^3) or particles per liter (L). To compare between these studies, we converted all units to particles/m^3. For trawling results provided as particles/km^2, particle counts were converted to a volumetric unit (particles per m^3) using the following equation: Np/(h*w*d), where Np is the number of particles in a sample, h is the height of the manta trawl (in meters), w is the width of the

manta trawl (in meters), and d is the distance traveled (in meters). Sediment samples reported as particles/kg dry sediment weight were not converted due to lack of soil property data needed to convert to a volumetric unit. Other information extracted from the studies included details on field and laboratory methods, and whether any specific QA/QC procedures (e.g., contamination controls, spike recoveries, etc.) were followed and reported. The extracted data from the studies reviewed in this report can be accessed as a dataset on the data repository website Borealis under the Rochman Lab Dataverse.

A separate literature review on ecotoxicological effects of microplastics was also conducted. Studies were selected in coordination with a parallel effort to update the Southern California Coastal Water Resources Project (SCCWRP) Toxicity of Microplastics Explorer (ToMEx) database. ToMEx is a publicly accessible database on the ecotoxicity of microplastics in marine and freshwater environments that was updated in 2023 to ToMEx 2.0. In this latest update, SCCWRP identified over 350 microplastics ecotoxicity studies conducted between January 2021 and January 2023. Of this list, approximately 190 studies were focused on freshwater environments. These 190 studies were reviewed as part of our literature review for relevance to the Great Lakes. Since the majority of these studies were laboratory experiments, the primary focus was on identifying studies that focused on organisms commonly found in and around the Great Lakes. 62 studies relevant to the Great Lakes were identified, and reviewed for information such as the organisms studied, toxicity endpoints, and effects observed. The full list of papers is included in Appendix A.3.

2. Characteristics of Microplastics

Microplastics are generally considered to include plastic particles smaller than 5 mm in size by published literature. Microplastics exhibit wide variation across characteristics such as size, shape, density, polymer type, additives, and color. These characteristics are discussed in more detail in the following sections. Each of these characteristics may influence the ecotoxicological effects and fate and transport of microplastics in the environment as well as inform particle sources (Thornton Hampton et al., 2022a). Particle morphology may indicate the microplastics' source (Helm, 2017), and polymer type, density, size, and shape can also influence their fate and movement through the Great Lakes basin.

There currently is no consensus on an all-inclusive definition of microplastics due to the wide range of particle sizes, shapes, materials, and behaviors, and as a result there are challenges with consistent detection, identification, and reporting of these particles (Frias and Nash, 2019). Efforts have been made for standardization, such as the United States Environmental Protection Agency (USEPA) Draft National Strategy to Prevent Plastic Pollution which includes recommendations on developing definitions for micro- and nanoplastics and standardized methods for their collection, extraction, quantification, and characterization, but there is currently no generally accepted definition or standardized sampling method for microplastics. Because of this, data on microplastics are not currently harmonized (USEPA, 2023). This makes it difficult to accurately compare studies across different regions, matrices, and sampling and analysis methods, and makes it impossible to implement an effective monitoring and risk management framework to protect the health of the Great Lakes, wildlife, and the public. Thus, a clear definition of microplastics is needed.

While various definitions of microplastics have been proposed, this review found wide variation in how microplastics were defined among academic literature and government reports. Of the approximately 90 publications reviewed, 50 included a definition of microplastics based on size. All but three studies defined the upper size limit of microplastics as 5 mm; the remaining publications used an upper size limit of 1 mm, 10 mm, and 20 mm. Few studies defined a lower size limit, but those that did appeared to use sizes based on common sampling methods (e.g., standard mesh sizes of 330 micrometers [µm] or 45 µm). Three publications mentioned a lower size limit of 330-350 µm, one mentioned 45 µm, and one study defined microplastics as particles in the 0.1 µm – 5mm size range, with particles smaller than 0.1 µm being classified as nanoplastics in this particular study. This review proposes a standard definition based on size to inform future microplastics monitoring and risk assessment efforts across the Great Lakes. Our recommendation is harmonized with the definition recently formalized in the State of California (see section 2.7.2).

2.1 Size

Plastic particles are present in the environment in a range of sizes. Microplastics, which are larger than nanoplastics but smaller than meso- and macroplastics, are generally considered to include particles smaller than 5 mm along their longest dimension.

The toxicity of microplastics is influenced by size, particularly for the food dilution and tissue translocation biological endpoints (Bucci et al., 2022; Thornton Hampton et al., 2022a). Food dilution takes place when macro or microplastics are ingested, either directly or via transfer from prey. These particles decrease the overall nutritional value of an organism's diet while contributing to a sense of false satiation. Long term impacts connected to decreased energy reserves include altered swimming behavior, decreased growth, altered reproduction, lowered fecundity, altered respiration, and in limited

cases, reduced survival. Particle size is a key microplastic characteristic when determining if ingestion is physically possible (Thornton Hampton et al., 2022a). Small microplastics are of particular concern with respect to tissue translocation as they can more easily transfer between tissues and cells of organisms (Rochman et al., 2019; Thornton Hampton et al., 2022a). For example, a fish study showed that microplastic particles smaller than 130 µm may translocate from the gut to muscle and liver, but the exact mechanisms are not well understood (McIlwraith et al., 2021). More recently, Mehinto et al. (2022) ran a binomial logistic regression model using 27 studies of 19 species and found that particles shorter than 83 µm were most likely to translocate.

Finally, particle size may also affect transport through the atmospheric and aquatic environments. Smaller particles are less likely to be entrained into the atmosphere due to a smaller cross-sectional area that is exposed to wind but may have longer residence times when suspended (Brahney et al., 2021). In aquatic media, larger particles are more likely to settle and are less likely to be entrained in the ambient flow than smaller particles. In fact, smaller particles (100 - 200 µm) were observed to be entrained in turbulent flow regardless of particle density, which may help explain their presence in remote regions (Shamskhany and Karimpour, 2022).

2.2 Primary and Secondary Microplastics

Microplastic particles may originate from a variety of sources (Helm, 2020) and are commonly classified as either primary or secondary microplastics (Rochman et al., 2019; ITRC, 2023). Strategies to mitigate microplastic pollution may also vary by source and their relative contributions to the Great Lakes and its tributaries.

- **Primary microplastics** are materials that are manufactured as micro-sized plastic particles. Examples of primary sources or products containing primary microplastics include toothpaste or facial cleansers where microbeads are used as abrasives; makeup containing glitter; and coatings on seeds, fertilizers, or pesticides. Resin beads (or pre-production plastic pellets, commonly known as nurdles) used in plastic manufacturing are another source of primary microplastics and may be accidentally released during manufacturing or transport.
- **Secondary microplastics** result from the breakdown of larger (macro) debris or plastic products, and their composition reflects the parent material. These are thought to be more common in the environment. Secondary microplastics can be further classified as use-based or degradation-based (OECD, 2021). Use-based secondary microplastics are generated unintentionally due to abrasion occurring during the use of products containing synthetic polymers. Common examples are microfibers released from synthetic textiles during washing; tire and road wear particles emitted during road transport activity; and paint flakes worn off from the surface of buildings, roads, and ships. Degradation-based secondary microplastics are those originating from the fragmentation of larger plastic items discarded in the environment. Plastic products left outside or disposed of in landfills can break down through chemical and physical processes resulting in the formation of microplastics.

Both primary and secondary microplastics can be transported to waterbodies, including the Great Lakes, by a variety of pathways including shipping, road-wear, beach litter, tributaries, urban and industrial stormwater discharge and runoff, and wastewater treatment plants (WWTPs) (Earn et al., 2021; ITRC, 2023). Typically, microfibers originate from clothing during washing and drying and are likely to be transported via wastewater discharge or ambient air. Other sources of fibers include direct wear and

tear on fishing nets, ropes, and other marine equipment and debris. Rubbery particles are typically associated with road tire wear and are likely transported via stormwater runoff. Pathways are discussed in greater detail in Section 4.6.

2.3 Morphology

Microplastics are encountered in a variety of shapes, which are determined by the manufacturing process and subsequent weathering and breakage (Helm, 2017; Yu et al., 2023). Figure 2-1 presents examples of common microplastic morphologies.

Figure 2-1. Microplastic Particle Morphologies Include (a) Pellets, (b) Foams, (c) Film, (d) Fibers, (e) Fragments, (f) Fiber bundles, and (g) Spheres (Image courtesy of Martindale et al. (2020)).

Common microplastic shapes include the following (Rochman et al., 2019; McIlwraith and Rochman, 2020; Yu et al., 2023):

- **Fibers** are flexible, with equal thickness throughout and ends that are clean-cut, pointed, or fraying. Typically, they are tensile and resistant to breakage. Fibers are present in a range of colors, which may be inconsistent across one particle due to bleaching. Fishing lines are sometimes classified as a distinct shape from fibers (Yu et al., 2023).

- **Fiber bundles** comprise 20 or more individual fibers tightly wound in a mass that cannot be untangled. Fibers present in bundles should be consistent in appearance (i.e., color, thickness, surface texture).
- **Fragments** have a rigid structure and sometimes irregular shape. They can be round, subround, angular, or subangular. They are not always equally thick throughout and can appear twisted or curled. Shavings, droplets, and seams from plastic manufacturing fit within this category. Paint flakes may also fall under this category. Fragments can be any color or combination of colors.
- **Spheres** are round with smooth surfaces. Spheres may also be present as hemispheres, following breakage during manufacturing, use, or weathering. They typically measure 2 mm or larger.
- **Pellets** (sometimes called "nurdles") are similar to spheres but tend to be larger, generally ranging between 3 mm and 5 mm. Pellets are often rounded or cylindrical in shape. Both spheres and pellets can be any color.
- **Films** are flat, thin, and malleable. Films can fold or crease but do not break apart easily. Films are typically partially or fully transparent and are found in a range of colors.
- **Foams** are soft, compressible, and cloud-like. They are usually white and/or opaque but can be any color.
- **Rubbery particles**, sometimes referred to as tire dust, are stretchy and resistant to breakage. The pieces are often cone shaped or S-shaped and are typically black in color.

Yu et al. (2023) defines a "fit for purpose" system of tiered classification of microplastics with three levels. The coarsest classification level ("abundance") focuses solely on particle size (e.g., nanoplastics, microplastics) and is appropriate for studies that only require particle counts but not information on particle characteristics. The medium level ("coarse morphology") includes fibers, fragments, and spheroids. These categories can be used for purposes where microplastics shape is of interest for, for example, field and laboratory ecotoxicological studies, and modeling particle behavior and transport. The most resolved level ("source specific") may be used where detailed morphology is desired and includes primary and secondary microplastics with further classes (e.g., microbead, pellet, fragment, film, foam, fiber) and sub-classes (e.g., spherical bead, irregular bead, commercial fragment, tire/road wear, paint, fiber bundle, line, polystyrene [PS] foam, polyurethane [PUR] foam, extruded polystyrene [XPS], expanded polystyrene [EPS], synthetic fibers, and semi-synthetic fibers). Some of these more resolved categories require additional lines of evidence (e.g., polymer composition, chemical additives, density) to complement the morphological characterization of particles.

Microplastic morphology can be indicative of source type and pathways and may be used to inform mitigation (Rochman et al., 2019). The shapes of microplastics can also affect their interactions within biological systems as summarized in ITRC (2023). Microplastic particles with more irregular shapes or fibers may attach more readily to internal and external surfaces of organisms. Microplastics with spherical shapes may cause less injury and gut inflammatory reaction than irregular shapes. For example, polypropylene fibers were found to have a higher toxicity to *Hyalella azteca* (an amphipod) than polypropylene beads. Sharp-edged and rough microplastics may cause more mechanical injuries to the gut epithelium in organisms than smooth particles (ITRC, 2023).

2.4 Chemical Composition

2.4.1 Polymer Type

Microplastics are composed of a diverse suite of polymer types. All plastic polymers consist of repeating monomers, which form the backbone of the polymer. This backbone structure is the fundamental difference between polymer types, informing a plastic's physical and chemical properties (Rochman et al., 2019). The toxicity of microplastics depends on their polymer type, as well as on their size and shape. It is important to distinguish between the toxic potential of plastic constituents (i.e., polymer, monomer units, and additives) and the potential for microplastics to release those constituents into the environment. Release of a constituent is based on the chemical properties of the polymer, the properties of the constituent, and the media into which it is being released (ITRC, 2023).

Polymers can be classified based on several different criteria (ITRC, 2023). One such criterion is whether the polymer is synthetic or semi-synthetic. Natural polymers (e.g., wool, cotton) are typically excluded from the definition of microplastics, unless they have been treated or chemically modified.

- **Synthetic plastics** are typically made from petroleum products. They start as a hydrocarbon molecule, or monomer, that is then repeatedly linked to itself to form the polymer backbone. Depending on the polymerization process and the monomer used, the plastic may be a thermoplastic or a thermoset plastic.
    - **Thermoplastics** soften on heating and harden on cooling and can be reshaped even after they are initially formed.
    - **Thermoset plastics** do not soften on heating, and once manufactured, cannot be melted and reshaped.
- **Bio-based plastics**, which are less common than petroleum-based products, may include materials that are made from biological materials, are biodegradable, or both. Biodegradable plastics can break down into organic matter, carbon dioxide, and water but typically only under certain environmental conditions (i.e., specific ranges in temperature, humidity, etc.), and the presence of microorganisms. These conditions are normally met only at industrial composting facilities (ITRC, 2023; Moshood et al., 2022). Outside such facilities, biodegradable plastics may persist in the environment similar to synthetic plastic materials.
- **Semi-synthetic plastics** include fibers such as rayon, which has a cellulose base material that is dissolved from the original plant material and reprocessed into semi-synthetic fibers.

Table 2-1 below lists common synthetic polymers along with the corresponding resin code and examples of common products. Resin codes are stamped onto plastic products and are often used to identify which plastics can be recycled in a given community. Resin codes 1 through 6 correspond to the six most widely used plastic polymers found in a wide range of packaging and other products. These six polymers along with polyurethane were the most commonly reported polymer types by the Great Lakes monitoring studies reviewed for this report (see Section 4.5). Resin code 7 corresponds to a broad category of hundreds of polymer types, only some of which are listed here. While these may be the most commonly occurring polymer types, there are hundreds of plastic polymers in use with different microstructure configurations. Polymer chains can be straight, branched, cross-linked, or generally amorphous which can give the plastic different properties such as density, melting point, and color. Plastics can even include multiple types of monomers that can be configured as a copolymer chain or as layered composites (Science History Institute, 2023).

Table 2-1. Plastic Polymers

Resin Code	Plastic Type Abbreviation	Plastic Type Name	Product Examples	Density (g/cm^3)	Melting Point (°C)
1	PET	Polyethylene terephthalate	Water and soft drink bottles, salad dressing/peanut butter containers, rope, carpet, polyester fibers used in textiles	1.38 – 1.41	265
2	HDPE	High-density polyethylene	Milk jugs, juice bottles, freezer bags, trash bags, shampoo/detergent bottles	0.94 – 0.99	130
3	PVC	Polyvinyl chloride	Plumbing and construction materials, pipes, liners, cosmetic containers, commercial cling wrap, siding	1.40 – 1.42	227
4	LDPE	Low-density polyethylene	Squeeze bottles, regular cling wrap, trash bags, shopping bags, furniture	0.89 – 0.93	110
5	PP	Polypropylene	Microwave dishes, medicine bottles, straws, ice cream tubs, yogurt containers, detergent bottle caps	0.87 – 0.92	176
6	PS EPS	Polystyrene Expanded polystyrene	PS—CD cases, disposable cups, egg cartons, cutlery, video cases EPS—Foam polystyrene, hot drink cups, food takeaway trays, protective packaging pellets	1.04 – 1.08 --	240 --
7	POM	Acetal (polyoxymethylene)	Fan wheels, gears, screws	1.3 – 1.6	172
	PMMA	Acrylic (polymethyl methacrylate)	Aquariums, fiber optics, paint	1.09 – 1.18	200
	ABS	Acrylonitrile butadiene styrene	Car parts, Lego, wheel covers	1.04 – 1.06	--
	PA	Nylon (polyamide)	Air bags, clothing, thread	1.07 - 1.16	179 – 265
	PES	Polyester	Fibers, rope	--	--
	PBT	Polybutylene terephthalate	Keyboards, relays, switches	1.47 – 1.49	--
	PC	Polycarbonate	Eyewear, safety helmets	1.20 – 1.22	--
	PEEK	Polyetheretherketone	Bearings, pump, pistons	1.26 – 1.54	--
	PLA	Polylactic acid (bioplastic)	Packaging, syringes, textiles	1.23 – 1.26	--

Resin Code	Plastic Type Abbreviation	Plastic Type Name	Product Examples	Density (g/cm^3)	Melting Point (°C)
	PSU	Polysulfone	Appliance parts, filters	1.24 – 1.6	--
	PTFE	Polytetrafluoroethylene	Teflon	2.1 – 2.3	327
	PUR, PU	Polyurethane	Adhesives, coatings, foams	--	--
	SAN	Styrene acrylonitrile	Brushes, hangers, printers	1.06 – 1.4	--

Source: ITRC, 2023; Grigorescu et al., 2019; SpecialChem, 2023; Sigma Aldrich, 2023

Different polymer types have different residual monomers that differ in toxicity. Some polymers, such as polyvinyl chloride (PVC) and polyurethane, contain variable amounts of residual monomers that are carcinogenic or mutagenic at high concentrations, whereas the monomers from other polymers, such as polyethylene and polypropylene, are considered to be less hazardous (Rochman et al., 2019).

2.4.2 Additives

During the plastic manufacturing process, additive(s) may be introduced to modify the properties of the plastic material. There are many different types of resins and polymers used to create plastics and each one has unique properties. Common plastic additives can generally be grouped into four categories based on the effect to the resin: colorants, fillers, reinforcements, and functional additives (ITRC, 2023).

- **Colorants**, such as dyes or pigments, are chemical compounds that are used to alter the color of a polymer. Dyes are soluble colorants used to add color to polycarbonates and polystyrene, whereas pigments, which are insoluble, are used to add color to polyolefins (ITRC, 2023).
- **Fillers** are inert material that add bulk to plastics, coatings, adhesives, and sealants. In addition to providing a cost benefit by lowering manufacturing cost, fillers can also improve moldability and stability of the polymer, reduce thermal expansion, and increase the heat-deflection temperature. Alumina trihydrate, barium sulfate, carbon black, calcium carbonate, calcium sulfate, clay, glass beads and fibers, kaolin, mica, and wollastonite are common fillers (ITRC, 2023).
- **Reinforcements** used in plastics include carbon, glass, mica, aramids, and other materials in the form of particulates, fibers, mats, or fabrics that are added to plastics to increase strength or provide other beneficial physical traits (ITRC, 2023).
- **Functional additives** are compounds that are added to plastics to enhance or alter existing properties of plastics or add new properties. These compounds are classified by the desired effect that the compound will have on the polymer. Examples include flame retardants, antimicrobials, and plasticizers (ITRC, 2023). Some additives modify final properties of the plastic, while others are used as processing aids during plastic manufacture (Barrick et al., 2021).

Common additives include brominated flame retardants, phthalates, nonylphenols, bisphenol A, and antioxidants. As microplastics degrade over time due to chemical and physical action, additives leach out and there is a risk that they may contribute to adverse effects on ecosystems. The ecotoxicity of several common plastic chemical additives has been characterized with results suggesting that additives can lead to neurotoxicity, inflammation, and alteration to lipid metabolism, and can have carcinogenic effects (Barrick et al., 2021). A global survey of plastic additives found 10,547 chemicals currently in use, with information availability varying considerably across chemicals and regions. Most substances have

information on their use or registration status in specific regions (>90%), followed by production volumes (70%), functions (69%), and any reported hazard classifications (61%) (Weisinger et al., 2021). For the rest, substantial information gaps persist: industrial sectors of use (40%), regulator harmonized hazard classifications (22%), and compatible polymer types (16%). Around 3% of the substances lack any information other than their chemical names and Chemical Abstract Service Registration Numbers (CASRNs). Moreover, it is not easy to get information about the additive chemicals and their amounts within different plastic products. This creates a barrier to understanding sources, fate, and effects of plastic additives.

2.5 Other Characteristics

2.5.1 Density

The density of microplastic particles ranges from less than 0.05 gram per cubic centimeter (g/cm^3) for polystyrene foam to 2.3 g/cm^3 for polytetrafluoroethylene (PTFE) and this affects their fate and transport in the environment. Low-density microplastics float on water surfaces and are exposed to winds, waves, and currents, with wind being the most important factor for transport in aquatic ecosystems. As such, low-density microplastics may travel rapidly in the environment and may be transported long distances. High-density microplastics tend to be less mobile in the environment and are more likely to sink and accumulate in sediments (ITRC, 2023; Helm, 2020).

Density also affects how microplastics interact with organisms in the environment. Many high-density microplastics are likely to sink and are therefore more likely to impact benthic biota. Low-density microplastics are more likely to be present in surface waters, washed up on shorelines, or throughout the water column (ITRC, 2023). However, low-density microplastics are also found in sediments, suggesting that biofouling (i.e., the colonization of submerged microplastics by bacteria and other microorganisms) causes an increase in density over time through biofilm development and contributes to a loss of buoyancy and sinking (Semcesen and Wells, 2021).

Finally, the density of plastic polymers is an important consideration in methods to separate microplastic particles from sediment and other sample materials (see Section 3.2.3 and Table 3-11).

2.5.2 Melting Point

Plastics that can be softened by heating and hardened by cooling and are called thermoplastics. Certain other plastics retain their shape after forming, and after hardening, they cannot be remelted. These plastics are known as thermoset plastics (ITRC, 2023).

Thermoplastics have a wide range of melting points, depending on the type and extent of cross-linking and chemical bonds that occur. Polymers can also have no defined molecular structure, called amorphous polymers, which are generally transparent and have a lower melting point. Polymers that are highly structured, called crystalline, are generally translucent and have a higher melting point (Science History Institute, 2023). The six most common plastic polymers (see Table 2-1) are all thermoplastics. Of those six, polyethylene has the lowest melting point, with low-density polyethylene (LDPE) and high-density polyethylene (HDPE) melting at about 110 degrees Celsius (°C) and 130 °C, respectively. In contrast, polyester has the highest melting point at about 265 °C.

2.5.3 Color

Plastics come in a wide range of colors, based on the pigments and other colorants added to the plastic polymer during the manufacturing process (ITRC, 2023). While relatively few studies have examined the

effect of color on the fate and environmental effects of microplastics, Zhao et al. (2022) reviewed available studies and noted that color may be a relevant property in this regard. For example, color affects light absorption, including light in the UV spectrum, and may affect the rate of plastic degradation. Some studies have also found that color may affect the diversity of microbial colonies that form on microplastics. Finally, color may affect the rate of ingestion of microplastics, especially for species that are visual predators prone to ingesting microplastics that resemble their prey.

2.6 Effects of Weathering on Microplastics

Weathering processes may affect a range of plastic properties. For example, low-density polymer microplastic particles have been found in sediments, suggesting that biofouling causes an increase in density and contributes to a loss of buoyancy and sinking (ITRC, 2023; Semcesen and Wells, 2021). A review of studies on the weathering of microplastics found that weathering can affect a range of physical and chemical properties of plastic particles, including an increase in the number of oxygen-containing functional groups on the particle surface; changes in particle color; decreased particle size and increased surface roughness; an increase in crystallinity; and an increased potential for leaching of chemicals including additives and fillers, monomers, and oxygenated intermediates. The review also noted that weathered microplastics may exhibit an increased potential for sorption of organic and inorganic contaminants, and for adhering to and forming aggregates with other microplastic particles as well as other solid particles present in the environment (Duan et al., 2021).

2.7 Definition of Microplastics for the Great Lakes

2.7.1 Legal and Regulatory Definitions

Some definitions of microplastics from current and proposed legislative and regulatory actions in North America and Europe are listed below:

- The California State Water Resources Control Board (CA SWRCB) recently adopted the following definition of microplastics (CA SWRCB, 2020):
  "'*Microplastics in Drinking Water' are defined as solid polymeric materials to which chemical additives or other substances may have been added, which are particles which have at least three dimensions that are greater than 1 nm and less than 5,000 micrometers (µm). Polymers that are derived in nature that have not been chemically modified (other than by hydrolysis) are excluded."* The CA SWRCB further breaks down "microplastics in drinking water" into the following size-based subcategories:
  - Nanoplastics: 1 nanometer (nm) to <100 nm
  - Sub-micron plastics: 100 nm to <1 µm
  - Small microplastics: 1 µm to <100 µm
  - Large microplastics: 100 µm to <5 mm
  - Mesoplastics: 5 mm to <2.5 cm; and
  - Macroplastics: >2.5 cm

  However, the state's monitoring and risk assessment framework only considers particles greater than 1 µm.
- The National Oceanographic and Atmospheric Administration (NOAA) does not have a formal definition of microplastics but considers microplastics to be particles smaller than 5 mm in size (NOAA, 2009).

- The Microbead-Free Waters Act of 2015 (U.S. Public Law 114-114) does not address secondary microplastics, but included the following definition for primary microbeads:
  "The term 'plastic microbead' means any solid plastic particle that is less than five millimeters in size and is intended to be used to exfoliate or cleanse the human body or any part thereof."
- Environment and Climate Change Canada's (ECCC) Draft Science Assessment of Plastic Pollution (ECCC, 2020) defines microplastics as "*plastic particles less than or equal to 5 mm in size*." The report further defines nanoplastics as "*a subset of microplastics. They are primary or secondary microplastics that range from 1 to 100 nm in size in at least one dimension.*" In addition, the Canadian "Microbeads in Toiletries Regulation" defines plastic microbeads as "*any plastic particle equal to or less than 5 mm in size*" (Health Canada, 2018).
- The European Chemicals Agency (ECHA), in its draft proposal to restrict the use of intentionally added primary microplastics, defines microplastics as follows (ECHA, 2020):
  "*'microplastic' means particles containing solid polymer, to which additives or other substances may have been added, and where ≥ 1% w/w of particles have (i) all dimensions 0.1 µm ≤ x ≤ 5 mm, or (ii), a length of 0.3 µm ≤ x ≤ 15 mm and length to diameter ratio of >3.*" Note that the second part of the definition applies specifically to microfibers.

Note that other size-based definitions for microplastics have also been proposed, including in the academic literature. Bermúdez and Swarzenski (2021), for example, proposed a size classification scheme aligned with prey or food particle sizes for a range of marine organisms. The proposed size ranges include nano-sized plastics (2 µm – 20 µm), micro-sized plastics (20 µm – 200 µm), and meso-sized plastics (200 µm – 2000 µm) (see Table 2-2 below).

Table 2-2. Microplastic Size Classification Scheme Based on Marine Plankton

Current size categories	Size range	Proposed size categories	Size Range	Organism of equivalent size
Nanoplastic	0.001-1 µm	Femto-size plastics	0.02-0.2 µm	Virus
Microplastic	1-1000 µm	Pico-size plastics	0.2-2 µm	Bacteria
		Nano-size plastics	2-20 µm	Flagellates
		Micro-size plastics	20-200 µm	Diatoms, dinoflagellates, ciliates, daphnids
		Meso-size plastics	200-2000 µm	Amphipods, appendicularians, chetognatos, copepods, thaliaceans
Mesoplastic	1-10 mm			
		Macro-size plastic	0.2-20 cm	Euphausiids, heteropods, jellyfish, larval fish, mysids, pteropods, solitary salps
Macroplastic	> 1 cm			
		Mega-size plastic	20-200 cm	Jellyfish, colonial salps

Source: Bermúdez and Swarzenski, 2021

2.7.2 Proposed Definition of Microplastics for the Great Lakes

To harmonize microplastics monitoring and reporting across the Great Lakes, the following size-based definition is proposed, based on the regulatory definition adopted by the CA SWRCB but modified to focus on particles greater than 1 µm, which reflects the current focus of CA microplastic monitoring and risk assessment programs:

"*Microplastics are defined as solid polymeric materials to which chemical additives or other substances may have been added, which are particles greater than 1 µm and less than 5,000 µm in all three dimensions. Polymers that are derived in nature that have not been chemically modified (other than by hydrolysis) are excluded."*

In addition to total counts or concentrations, the following parameters should be reported to facilitate comparison across studies:

- Morphology, as % of total
- Polymer type (if identified), as % of total
- Size range assessed, based on field and lab methods
- Size fractions (if distinguished), as % of total

Other parameters may be of interest depending on research or monitoring questions, e.g., color.

3. Monitoring and Analytical Methods

Microplastics are collected and analyzed from a variety of sample matrices, including water, sediments, and biota, using a range of methods. A standard procedure for microplastic analysis involves sample collection, followed by drying, debris removal, density separation, digestion, enumeration, and chemical identification. Though most studies follow this general order, some steps may be performed before or after others depending on the sample properties and study requirements (ITRC, 2023; Fuschi et al., 2022; Prata et al., 2019).

This section discusses the methods for the collection, processing, and analysis of environmental samples for microplastics used by the studies included in this review that measured microplastics concentrations in the Great Lakes basin. It also incorporates methodological findings and recommendations from recent published literature (including non-Great Lakes studies) on the sampling and analysis of microplastics. In addition, we refer readers to a series of Standard Operating Procedures in Appendix C of the report titled "Final Report of the IJC Science Advisory Board Work Group on Microplastics: Monitoring, Risk Assessment, and Management of Microplastics in the Laurentian Great Lakes."

3.1 Sample Collection Methods

Sample collection methods for microplastics vary depending on the matrix or compartment being studied. For this report, matrices were split up into the following categories:

- **Water** samples are taken from the body of a lake or river to measure concentrations in a volume of water. Historically, these samples have generally been taken from the surface. Measurements from these samples are usually reported as particles/L or particles/m^3, although with trawl sampling it can also be reported as the number of particles per km^2 covered. Water column samples are similar to surface water samples except they are taken from below the surface of the water, somewhere in the water column above the sediment and are typically reported as particles/m^3.
- **Sediment** samples are taken from the bottom of a lake or river or terrestrial area to measure concentrations in a volume, mass, or fixed surface area of sediment. Due to the differences in porosity and water content of sediment, measurements from these samples are typically reported as particles/kilogram (kg) of dry sediment but can also be reported as particles/m^3 or particles/L of dry or wet sediment.
- **Shoreline debris** samples measure the concentration of particles that exist on or within a couple centimeters of the land surface in a fixed surface area. These samples typically involve collection of large visible debris within a designated beach site or area. This matrix was considered distinct from sediment samples due to different collection, analysis, and reporting methods in the reviewed literature which are discussed in more detail in Section 3.1.3. Measurements from these samples are typically reported as particles/square meter (m^2).
- **Biota** samples measure the number of particles that exist within an organism. Historically, these samples generally have measured the number of particles in the gastrointestinal tracts of fish or whole organisms. These studies typically report measurements as particles/individual or particles/g tissue.

No Great Lakes studies were identified that analyzed microplastic concentrations in the air, or that measured rates of aerial deposition of microplastic particles. However, this is an emerging area of research and is recommended for consideration in future Great Lakes research.

3.1.1 Water

Water samples can be collected via trawl, grab, or pump composite sampling methods. Brander et al. (2020) and Miller et al. (2021) provide a summary of these methods:

- **Trawls** are conducted using manta nets or neuston nets, which are nets that can be towed behind a vessel to pass through a relatively large volume of water to collect particles in an end piece of the net. The volume of water sampled is typically measured using flowmeters, or calculated based on the boat speed, sampling duration, and net opening size. Manta nets sample a layer of water at the surface, while neuston nets can be towed at a specified depth, typically around 16 cm below the surface.
- **Grab samples** can be collected by submerging and opening a container at a desired location in the water column. Shallow grab samples can be collected with a simple glass jar, while deeper water column samples require devices such as a Van Dorn sampler or depth integrated sampler.
- **Composite sampling** usually involves pumping or flowing a volume of water through a filter and collecting the entrained particles. The sample shows the composite or average concentrations of the volume of water or distribution of concentration over the time period that it was collected. These methods can also be used to collect grab samples if the sample volume or period is small enough.

Table 3-1 summarizes the water sampling methods used by Great Lakes studies that were included in this review, most of which are for surface water. Most studies (12 of 17) used manta trawls to collect samples from the water surface. A smaller number of studies (6 of 17) collected grab samples, which are easier to collect than trawl samples but typically collect a smaller volume that may not be representative of the area, especially when heterogeneity in particle count and characteristics is high. Composite samples such as with pumps can collect large volumes of water at greater depths, but this method requires specialized equipment and may run into similar issues as trawl samples based on filter size.

Table 3-1. Surface Water Collection Methods Reviewed (N=17*)

Collection Method	Number of Articles	Journal Articles
Trawl	12	Baldwin et al., 2016; Cable et al., 2017; Cox et al., 2021; Eriksen et al., 2013; Fox et al., 2022; Hendrickson et al., 2018; Lenaker et al., 2019; Mason et al., 2016; Mason et al., 2020; McCormick et al., 2014; Minor et al., 2020; Vincent and Hoellein, 2021
Grab	6	Crew et al., 2020; Grbic et al., 2020; Hou et al., 2021; Hollein et al., 2021; McNeish et al., 2018; Vincent and Hollein, 2021
Composite	2	Fox et al., 2022; Grbic et al., 2020

*Note some studies may have employed multiple methods.

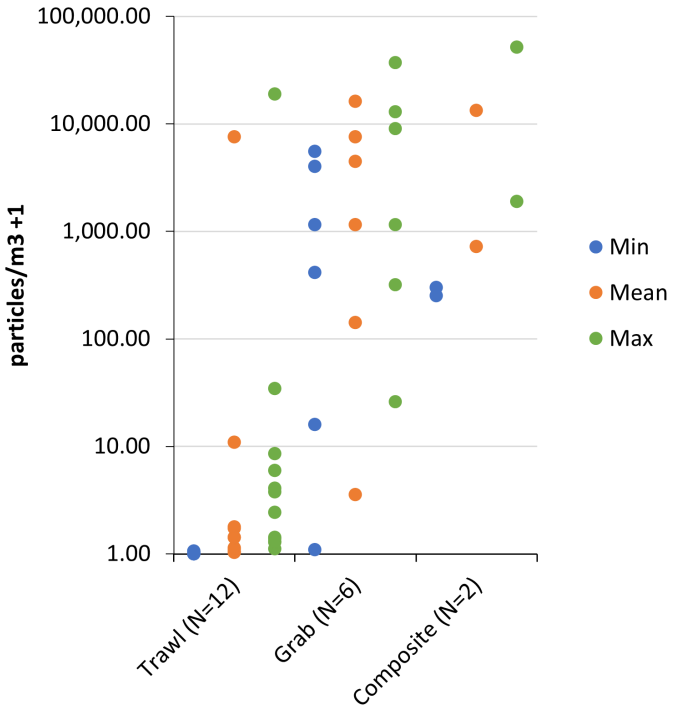
A summary of the water sample collection parameters is provided in Table 3-2. Studies that used trawl sampling techniques typically used manta trawls and neuston nets to cover large areas of lakes. Most studies sampled surface waters within 16 cm of the surface, but two studies (Baldwin et al., 2016; Lenaker et al., 2019) used submerged neuston nets to collect samples at depths up to 6 m. Grab samples were primarily taken with glass jars from the water surface, with the exception of Hou et al. (2021) who used a Van Dorn sampler to collect a sample 30 cm deep. The most common mesh size used for manta trawls was 330 µm, but mesh sizes may range from 100 - 500 µm (Pasquier et al., 2022). Fox et al. (2022) used McLane pumps to sample depths up to 240 m deep. The pumps were equipped with sequential

300-µm and 100-µm mesh filters to collect different sized debris. Grbic et al. (2020) used an autosampler to collect a grab sample every hour for 3 days; these grab samples were combined into a composite sample for the site.

Table 3-2. Surface Water Collection Parameters of Studies Reviewed

Collection Method	Sample Depths	Sample Volume	Filter/Mesh Size Used in the Field	Type of Equipment
Trawl	0 – 6 m	45,000 – 200,000 L	100 - 500 µm	Manta Trawl, Neuston Net
Grab	0 – 0.3 m	1 – 100 L	N/A	Glass Jar, Steel Bucket, Van Dorn Sampler
Composite	0 - 240 m	4 – 570 L	100 - 300 µm	Autosampler, McLane Pump

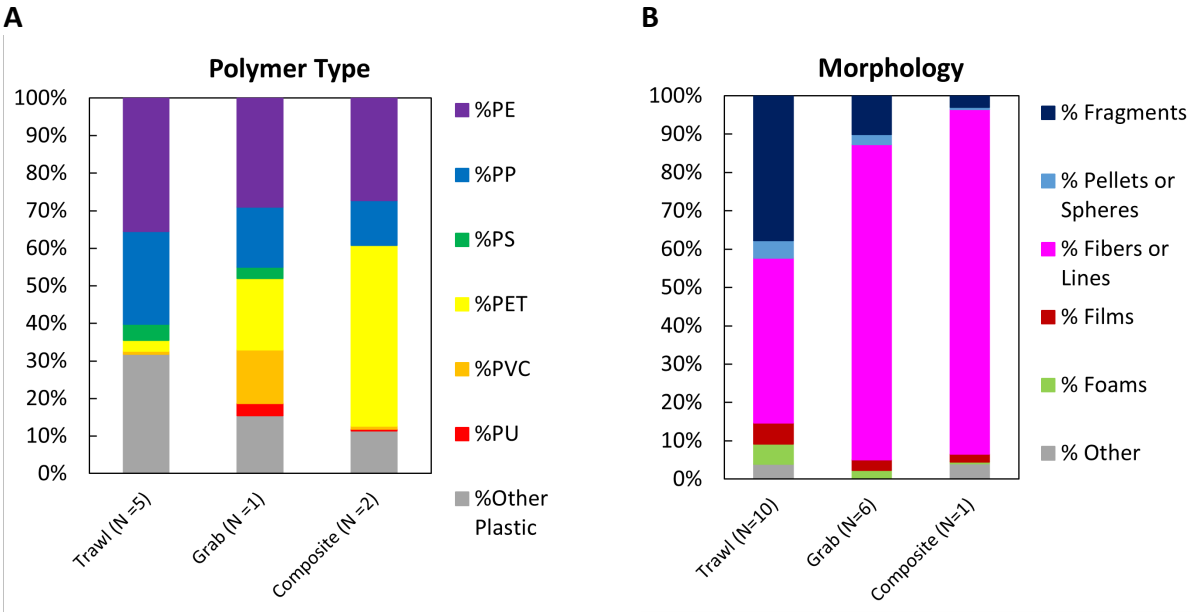
An important difference between net-based sampling methods such as manta trawls and grab sampling or pumping is that mantra trawls only collect particles larger than the mesh opening size, which is typically 330 µm. Therefore, manta trawls do not capture smaller microplastics and may undercount overall concentrations of microplastic particles in the water. This is illustrated in Figure 3-1, which compares microplastics concentrations in studies that used manta trawls to those using grab and composite sampling. This suggests that reported concentrations may not be directly comparable, depending on the sampling method used. Section 3.5.1 discusses some alignment methods that have been proposed to address this issue.



Note: Concentrations have been scaled using a log_{10}([particles/m^3] + 1) transformation. Points represent a summary of the results for each method used in a single journal article. Some papers may have used multiple methods.

Figure 3-1. Microplastic Concentrations by Method - Water

Figure 3-2 compares the distribution of particle shapes and polymers reported by studies using trawl and grab samples. Studies using grab samples reported a much higher percentage of fibers (close to 80%) compared to studies that used trawling (less than 50% fiber). In terms of polymers, trawl studies reported a significantly lower proportion of polyester. Polyester is denser than water (approximately 1.3 g/cm^3) and tends to sink, which explains why it may be collected less frequently in trawl samples (densities for many common plastic polymers are shown in Table 2-1). Trawl studies reported higher fractions of polypropylene and polyethylene, as well as other plastics including PTFE, polyamide, acrylic, polycyclohexylenedimethylene terephthalate, ethylene vinyl acetate (EVA), polydimethylsiloxane, chlorinated polyethylene (CPE), nylon, styrene-butadiene rubber (SBR), polyvinyl alcohol, and Rayon, and unknown or copolymer materials. The studies did not consistently report the same types of polymers under the "other" category.



A) Material distributions and B) shape distributions of water samples by method and size fraction. Fractions were weighted by the number of samples taken in an article. Note that some studies subsampled to determine shape and polymer type and some studies did not measure polymer type or morphology.

Figure 3-2. Shape and Polymer Fractions by Method – Water

3.1.2 Sediments

Aquatic sediments as well as terrestrial soils/sand (herein called sediments) can be sampled using a simple trowel or more specialized devices, depending on the sampling location and the study objectives (Brander et al., 2020). A summary of common sampling techniques is provided below:

- **Grab samples** require the collection of a volume of sand or sediment. This can be accomplished using a simple hand trowel for nearshore or shallow sediments, or by using more complex sampling devices to collect deep benthic sediments (e.g., Ponar grab sampler, Peterson grab sampler). Samples may be collected in bulk or volume-reduced by sieving to limit the sample to the size class of interest. Note that with grab samples, the reported concentration may vary

depending on sediment bulk density and the depth sampled; therefore, both of these co-variates should be reported.

- **Core samples** are typically used to measure vertical particle distributions in sediment. They collect a volume of sediment over a desired depth range with minimal disruption to the sediment profile. The collected sediment can then be subsampled and processed as multiple grab samples to determine particle distribution over the sampled depth range.
- **Sediment trap samples** entrain sediment particles in the air or water medium where it is placed, collecting sediment over a period of time. Unlike grab and core samples, sediment traps are often used to measure transport rather than concentration at a given time. Samples collected by this method typically report particle concentrations at intervals over time or as an average over the total sampling time.

Tables 3-3 and 3-4 summarize sediment sampling methods used by Great Lakes studies included in this review. The studies reviewed here collected aquatic sediment samples (12 of 14) as well as terrestrial sediment samples (5 of 14) from beaches. Most studies (12 of 14) collected grab samples, but a few collected core samples (4 of 14) and samples using sediment traps (2 of 14). Note that these tables include studies that analyzed both benthic and terrestrial sediments.

Table 3-3. Sediment Collection Methods (N=14*)

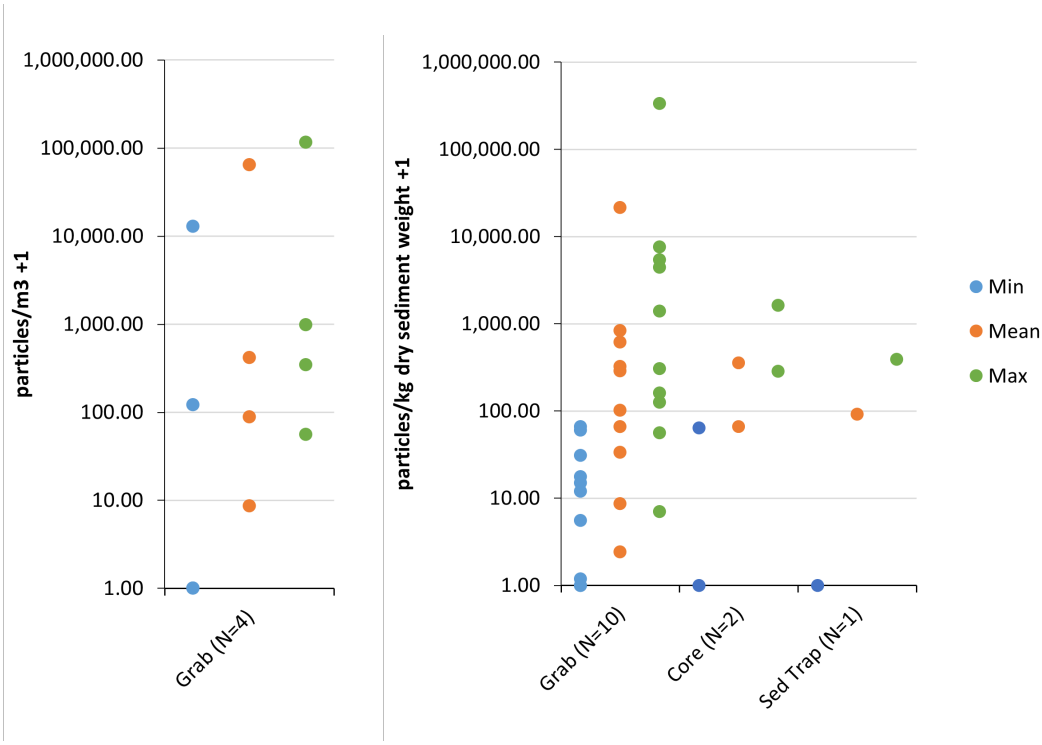
Collection Method	Number of Articles	Journal Articles
Grab	13	Ballent et al., 2016; Belontz et al., 2022; Casteñeda et al., 2014; Corcoran et al., 2015; Corcoran et al., 2020a; Crew et al., 2020; Davidson et al., 2022; Dean et al., 2018; Hou et al., 2021; Lenaker et al., 2019; Lenaker et al., 2021; Minor et al., 2020; Schessl et al., 2019
Core	4	Ballent et al., 2016; Corcoran et al., 2015; Davidson et al., 2022; Lenaker et al., 2021
Sediment Trap	2	Ballent et al., 2016; Dean et al., 2018

*Note some studies may have employed multiple methods.

Table 3-4. Sediment Collection Parameters of Studies Reviewed

Collection Method	Sample Depths	Sample Volume	Type of Equipment
Grab	0 – 3 cm	1 – 12 L	Petite Ponar Grab, Shipek Grab, Ekman Grab, Trowel
Core	0 – 30 cm	< 1 – 5 L	Glue Gravity Corer, Box Corer, Split Spoon Corer
Sediment Trap	NR	NR	Sediment Trap

Figure 3-3 compares reported microplastics concentrations in aquatic and terrestrial sediments by sampling method. This figure only includes studies that reported concentrations in terms of particles per kg of sediment weight which was 13 of the 17 total studies that analyzed sediment samples. The remaining studies (Casteñeda et al., 2014; Corcoran et al., 2015; Hou et al., 2021; and Schessl et al., 2019) reported units of particles per volume and do not include information to convert to particles per kg of sediment. Concentrations for core and sediment trap methods were similar. Grab samples had a much larger range of concentrations compared to the other methods.



*Note: Concentrations have been scaled using a log_{10}([particles/m^3] + 1) and a log_{10}([particles/kg dry sed wt] + 1) transformation. Points represent a summary of the results for each method used in a single journal article. Some papers may have used multiple methods. *Some studies (Ballent et al., 2016 and Corcoran et al., 2015) reported single concentrations for a combination of core and grab sampling methods. These studies mostly used grab sampling techniques that were supplemented with core samples, so these studies were categorized in this figure as grab samples.*

Figure 3-3. Microplastic Concentrations by Method – Sediment

Figure 3-4 compares reported microplastics concentrations in sediment by aquatic or terrestrial locations. Studies that analyzed benthic or aquatic sediment reported higher concentrations than studies that analyzed terrestrial sediments. Generally, benthic sediments are considered a sink where microplastic particles accumulate; microplastic abundance is generally higher in these sediments than in other matrices (Darabi et al. 2021). While microplastic transport can occur from sediments to other matrices, the majority of particles are transported to water bodies and then settle into benthic sediments.

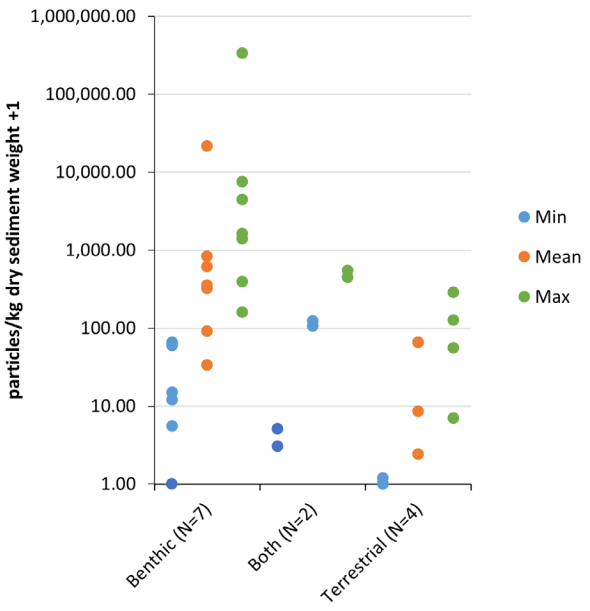
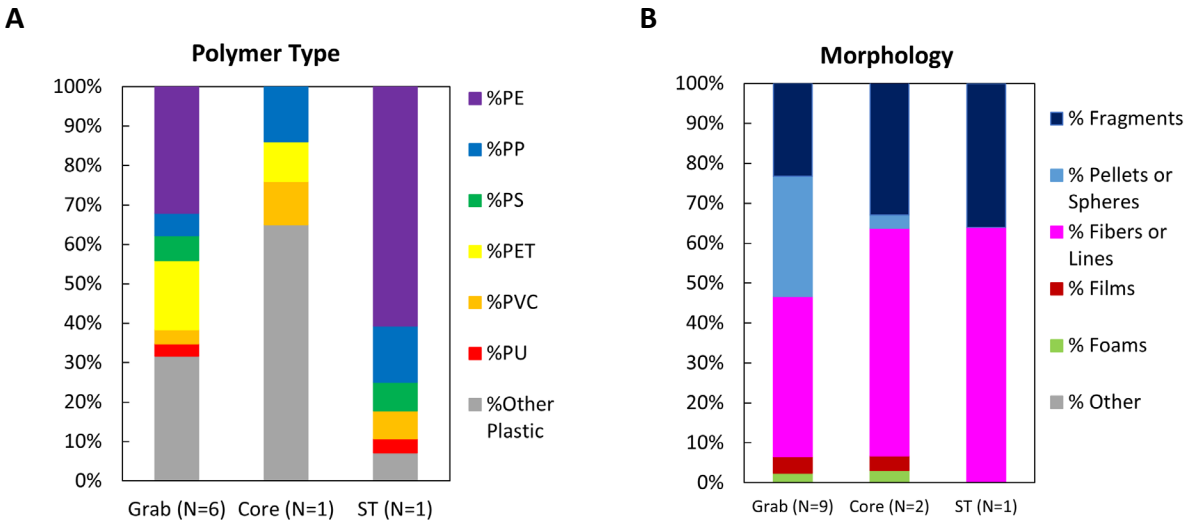


Figure 3-4. Microplastic Concentrations by Location – Sediment

Figure 3-5 compares the distribution of particle shapes and polymers reported by studies using different sediment sampling methods. In terms of polymers, studies that collected grab samples reported higher proportions of polyethylene and other polymers, while studies using core samples and sediment traps reported predominantly other plastics and polyethylene, respectively. Studies using grab samples reported a significant percentage of fibers, pellets or spheres, and fragments, while the studies that used core samples and sediment traps primarily reported fibers and fragments.



A) Material and B) shape distributions of sediment samples by method. Fractions were weighted by the number of samples taken in an article. Note that some studies subsampled to determine shape and polymer type and some studies did not measure polymer type or morphology. ST = sediment trap.

Figure 3-5. Shape and Material Fractions by Method – Sediment

3.1.3 Shoreline Debris

Shoreline debris on beaches are technically sediment samples, but instead of analyzing the amount of microplastics in a volume or mass of soil, they are instead typically taken to analyze the distribution of microplastics across a large terrestrial surface. These methods are typically limited to large visual debris that can easily be identified and collected. However, these methods can also be combined with subsampling using grab samples and statistical methods to count smaller particles, including microplastics. Concentrations of microplastic debris on a shoreline can be determined with the following methods:

- **Transects/quadrats** are methods of defining a sample area for particle collection. Transects are lines marked along a site, and surface debris within a certain distance of the transect line is surveyed and collected at regular intervals along it. Quadrats are square or rectangular areas, within which all surface debris is surveyed. Most studies visually inspected the surface and collected visible debris within approximately 2 - 5 cm (1 - 2 inches) of the surface.
- **Volunteer cleanup records** can also be used to estimate microplastic concentrations at a site. The surface area of a site can be determined through geographic methods and combined with counts of items collected to determine concentrations.

Table 3-5 summarizes the terrestrial sediment sampling methods used by studies that were included in this review. All of the studies reviewed focused on sandy beaches. Microplastic contamination on other shoreline types, including rocky or marshy shorelines, was not assessed and may be a knowledge gap.

Table 3-5. Shoreline Debris Collection Methods (N=11)

Collection Method	Number of Articles	Journal Articles
Transect/ Quadrat	9	Arturo and Corcoran, 2022; Corcoran et al., 2015; Corcoran et al., 2020a; Costello and Ebert, 2020; Davidson et al., 2022; Hoellein et al., 2014; Lazcano et al., 2020; Minor et al., 2020; Vincent and Hoellein, 2017; Vincent and Hoellein, 2021
Volunteer Cleanup Records	2	Hoellein et al., 2015; Vincent et al., 2017

Minimum concentrations for this matrix ranged from 0 – 10.5 particles/m^2, mean concentrations ranged from 0 – 544 particles/m^2, and max concentrations ranged from 0 – 728 particles/m^2. Analysis of the differences in shoreline debris data based on method, size, shape, and polymer type were not included due to the lack of studies that provided this information. Data that existed and extracted for shoreline debris studies can be found in the Rochman Lab Dataverse.

3.1.4 Biota

Table 3-6 summarizes the biota sampling methods used by Great Lakes studies that were included in this review. The majority of studies collected live samples of the species listed in Table 3-7; however, Hou et al. (2021) also compared their sample results to archived fish samples dating back to the early 20th century to study microplastic pollution trends in the last century. Holland et al. (2016) collected bird samples from hunter kills throughout Canada, and Wagner et al. (2019) used a subsample of fish specimens from a separate study conducted by the USEPA. For the purposes of this study, these papers were grouped into the Hunting/Fishing category. The remainder of the studies hand collected specimens such as roadkill, mussels, or algae.

Table 3-6. Biota Collection Methods (N=9)

Collection Method	Number of Articles	Journal Articles
Hunting/Fishing	5	Holland et al., 2016; Hou et al., 2021; McNeish et al., 2018; Munno et al., 2022; Wagner et al., 2019
Hand Collected	4	Brookson et al., 2019; Hoellein et al., 2021; Peller et al., 2021; Schessl et al., 2019

The biota sampled included birds, fish, algae, molluscs, and amphibians. Table 3-7 summarizes the species sampled and the collection methods used. Most species were collected using conventional hunting, fishing, and net techniques. Algae (*Cladophora* spp.) samples were collected from a variety of locations, including free-floating algae along the shoreline, beach wash-up, algae attached to breakwall rocks, and algae filaments from quadrats placed at various depths along the lake bottom.

Table 3-7. Biota Sampled and Collection Methods

Species	Collection Methods
Anuran	Grab sampling (roadkill)
Double crested cormorants	Grab sampling (taken from colony)
Long-tailed duck	Hunting
White-winged scoter	Hunting
Dreissenid mussels	Grab sampling (off rocks)
Banded killifish	Wading seine net
Bass sp.	Wading seine net
Brown bullhead	Electrofishing
Cisco	Gill nets
Common shiner	Electrofishing
Emerald shiner	Wading seine net, electrofishing
Fathead minnow	Wading seine net, electrofishing
Gizzard shad	Wading seine net
Lake trout	Electrofishing, gill net, hook and line
Lake whitefish	Gill nets
Largemouth bass	Wading seine nets
Longnose sucker	Gill nets
Quillback	Wading seine net
Rainbow trout	Electrofishing, gill net, hook and line
Round goby	Fishing rod, electrofishing, wading seine net
Round whitefish	Gillnets
Sand shiner	Wading seine nets
Smallmouth bass	Electrofishing, gill net, hook and line

Species	Collection Methods
Spotfin shiner	Wading seine net
Spottail shiner	Wading seine net, electrofishing
White sucker	Wading seine net, electrofishing, gill net
Yellow perch	Gillnets, electrofishing
Cladophora	Wading and diving (grab)

Concentrations by type of organism are shown in Section 4.4 of this report.

3.2 Sample Preparation Methods

All articles provided some description of sample preparation techniques which generally followed a similar pattern. After samples were collected in the field, they were typically processed to remove moisture, sediment, and organic matter before further analysis. Generally, sample preparation followed the process shown below.

- **Storage and preservation** maintain the integrity of the sample during transportation from the field to the laboratory.
- **Debris removal and size fractionation** involve removing moisture, fine particles that may stick to larger microplastic particles, and larger non-plastic debris, typically by sieving. This step may also involve separating the sample into different size classes. Sieving can also be done in the field prior to transport to the laboratory.
- **Density separation** involves separating plastic particles from similar-sized sediment and other inorganic particles.
- **Digestion** involves the removal of organic matter from the sample.

These methods are discussed in greater detail below.

3.2.1 Storage and Preservation

Samples are typically stored in capped glass or other non-plastic containers between collection and laboratory analysis. Samples may be stored in water or other media (e.g., alcohol to reduce algal growth); additionally, they can be stored at room temperature or may be refrigerated or frozen. Tables 3-8 and 3-9 describe the sample storage temperature and media used by the studies reviewed.

Table 3-8. Sample Storage Temperature (N=23)

Storage Temperature	Total Number of Articles	Surface Water	Sediment	Surface Debris	Biota
Frozen or refrigerated	12	4	5	None	6
Ambient temperature	11	7	3	1	1

Table 3-9. Sample Storage Media (N=14)

Preservation Medium	Total Number of Articles	Surface Water	Sediment	Surface Debris	Biota
Ethanol	4	3	1	None	1
Lake/river water	5	2	3	None	3
Isopropyl alcohol	3	3	None	None	None
Filtered water	2	2	None	None	None

Note that some preservation media may degrade plastic particles over time. Karr (2020) analyzed the effects of storing microplastic fibers in ethanol, which is commonly used to preserve biological samples or reduce algal growth and found evidence of mass loss across multiple polymer types after one week of storage, with greater than 10% mass loss in cellulose acetate, polyester, and phthalate polymers.

3.2.2 Debris Removal & Size Fractionation

The first step in sample processing is the removal of large debris, silt, and soil aggregates which are typically removed from the sample by "wet sieving." This involves rinsing the sample with distilled water over a sieve stack with mesh sizes corresponding to the smallest and largest particle sizes to be analyzed. Wet sieving with detergent can further facilitate size fractionation by reducing any clumping. Samples are sometimes also dried in an oven to remove moisture and can then be dry sieved to separate samples into discrete size fractions. Size fractionation can help with sorting via microscopy and also provides useful information about the size of particles. Figure 3-6 below shows minimum particle sizes reported for each study, which typically corresponds to the smallest sieve in the wet sieve stack or the mesh size used during collection.

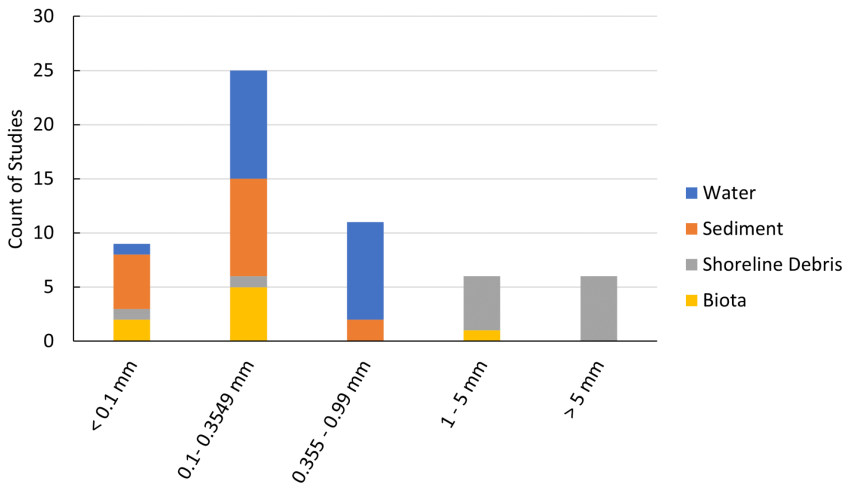


Figure 3-6. Minimum Particle Size Reported, by Matrix

Some, but not all, studies also reported particle counts by size fraction. Table 3-10 below shows the size fractions into which samples were separated by the studies that included this step. For future studies, if size fractioning occurs, it may be beneficial if the particle count within a given size range is also reported

as a percentage of all particles in the sample. Further, particle size classes should be standardized and potentially linked to meaningful ecological thresholds (e.g., see Bermúdez and Swarzenski, 2021).

Table 3-10. Sample Size Fractions Reported (N=12)

Size Distributions Reported	Number of Articles	Journal Articles
0.125 - 0.3549 mm 0.355 - 0.999 mm 1.00 - 4.749 mm > 4.75 mm	7	Baldwin et al., 2016; Cable et al., 2017; Cox et al., 2021; Eriksen et al., 2013; Lenaker et al., 2019; Lenaker et al., 2021; Mason et al., 2020
< 1 mm > 1 mm	1	Costello and Ebert, 2020
< 1.5 mm 1.5 - 3.3 mm > 3.3 mm	1	McNeish et al., 2018
< 2 mm > 2 mm	2	Ballent et al., 2016; Casteñeda et al., 2014
5 - 25 mm 25 - 100 mm	1	Arturo and Corcoran, 2022

Finally, some studies oven dried samples to remove moisture, but used varying times and temperatures (see Figure 3-7). A few studies also added vacuum filtration as an additional step to remove moisture or other preservation media. Munno et al. (2018) studied the effect of various treatment steps and noted that exposure to temperatures above 70°C led to sample loss in some cases. While higher temperatures shorten drying times which may take multiple hours with heavily saturated samples, care should be taken during sample processing to not expose microplastic particles to temperatures approaching the melting points of the polymers most likely to be present.

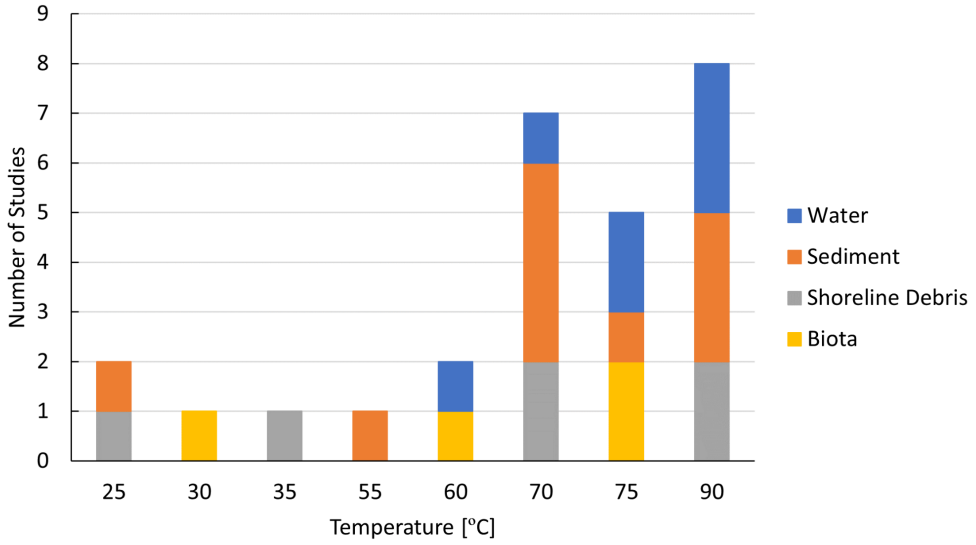


Figure 3-7. Oven Drying Temperatures Reported by Matrix

3.2.3 Separation

After moisture and debris removal and size separation, microplastic particles are separated from sediment and other non-organic particles, typically using density separation. This process involves immersing the sample in a high-density liquid, which allows the buoyant plastic particles to float while sediment and other non-organic particles sink. Various liquids with a range of densities have been used for this step by the studies reviewed, as listed in Table 3-11 below. A sodium chloride (NaCl) solution was the most common medium, followed by ultrasonic baths and sodium polytungstenate (SPT).

Table 3-11. Separation Methods (N=25)

Separation Method	Total Number of Articles	Surface Water	Sediment	Surface Debris	Biota
Sodium chloride (NaCl)	7	6	1	1	None
Ultrasonic bath, Pulsed Ultrasonic Extraction (PUE)	6	3	None	2	1
Sodium polytungstenate (SPT)	6	1	5	None	None
Zinc chloride ($ZnCl_2$)	3	None	3	None	None
Lithium metatungstenate (LMT)	1	None	None	1	None
Canola oil	1	None	1	None	None
Calcium chloride ($CaCl_2$)	1	1	None	None	None

Note: Numbers may not add up to the totals, since some studies sampled multiple matrices.

Prata et al. (2019) conducted a review of collection and analytical methods for microplastics and found that studies reported using a variety of liquids for density separation of microplastics, depending on the desired microplastics to be separated (see Table 3-12). In addition to the ability to recover plastics of varying densities, different liquids appear to provide varying degrees of consistency. Sodium iodide (NaI), for example, had better reported consistency in plastics separation than NaCl. Other considerations include cost, the toxicity of the liquid and whether it can be reused over multiple cycles.

Table 3-12. Solutions Used for Density Separation of Microplastics

Polymer	Density (g cm^{-3})	Water	NaCl	NaI	$ZnBr_2$
		1 g cm^{-3}	1.2 g cm^{-3}	1.6 g cm^{-3}	1.7 g cm^{-3}
PP	0.9–0.91	+	+	+	+
PE	0.92–0.97	+	+	+	+
PA	1.02–1.05	–	+	+	+
PS	1.04–1.1	–	+	+	+
Acrylic	1.09–1.20	–	+	+	+
PMA	1.17–1.20	–	+	+	+
PU	1.2	–	+	+	+
PVC	1.16–1.58	–	±	+	+
PVA	1.19–1.31	–	±	+	+
Alkyd	1.24–2.10	–	–	+	+
Polyester	1.24–2.3	–	–	+	+
PET	1.37–1.45	–	–	+	+
POM	1.41–1.61	–	–	±	+

Source: Prata et al., 2019

Note: + indicates separation is possible. – indicates separation is not possible. ± indicates separation may be possible. NaCl - sodium chloride; NaI - sodium iodide; $ZnBr_2$ - zinc bromide.

3.2.4 Digestion

The next step of sample preparation is to separate microplastics from natural organic particles, which can be difficult to differentiate from plastic using optical methods. Additionally, biofilms on plastic particles can interfere with chemical detection methods. Digestion can also be used to dissolve surrounding tissue or organs when analyzing microplastic contamination in biota.

Organic matter is typically removed using a digestion reaction that selectively dissolves natural organic matter. Table 3-13 lists the digestion methods used by the Great Lakes studies that were reviewed as part of this effort. Wet peroxide oxidation (WPO) was the most common technique (19 of 27 studies), followed by a smaller number of studies that used acidic or alkaline media, enzymes, and other chemical reagents.

Table 3-13. Digestion Methods (N=27)

Digestion Method	Total Number of Articles	Surface Water	Sediment	Surface Debris	Biota
WPO	19	11	6	2	5
HCl	4	1	3	None	None
Enzyme	1	1	None	None	None
Fenton's Reagent	1	1	None	None	None
KOH	1	None	None	None	1
Liquefaction (not specified)	1	None	None	None	1

Note: Numbers may not add up to the totals, since some studies sampled multiple matrices. WPO - wet peroxide oxidation; HCl - hydrogen chloride; KOH - potassium chloride.

Reviews of plastic sampling and processing methods from published literature conducted by Prata et al. (2019) and Brander et al. (2020) have documented that acid and alkaline digestion are effective at removing organic matter but may degrade certain polymers or cause color loss of microplastics, which can affect study results. Hydrogen peroxide, which is the reagent used in WPO, may be less harmful to plastics when used at lower concentrations and at lower reaction temperatures, although longer digestion times may be required to achieve sufficient removal of organic matter. However, Munno et al. (2018) found that WPO can often generate temperatures high enough to cause sample loss; therefore, care must be taken when using this method. A recent assessment found that sequential digestions using a combination of oxidative-alkaline solutions (Fenton's reagent and potassium hydroxide [KOH]) were effective in eliminating most organic matter, while also reducing impacts on microplastic particles as compared to WPO especially in the absence of adequate organic matter (Akhbarizadeh et al., 2023).

3.3 Analytical Methods

Analytical methods involve the enumeration and characterization of microplastic particles after sample processing is completed. The methods used by the studies reviewed can be broadly grouped into the following categories:

- **Enumeration** involves estimating particle counts per sample volume or mass.
- **Chemical identification** is the process to determine whether the particle is indeed a plastic material and to identify the polymer type.

These methods are discussed in greater detail below, along with a listing of the studies that used specific methods. Note that this discussion does not separate methods by the matrix (i.e., water, sediment, or biota) because by this point in the process, suspected microplastic particles have been extracted from the sample matrix and are analyzed in a similar manner regardless of the original sample type.

3.3.1 Enumeration

The studies reviewed used the following methods for particle enumeration after extraction from the sample matrix:

- **Visual identification** involves inspection of suspected plastic particles without magnification and is typically used for larger plastic particles.
- **Optical microscopy** is a common method to identify and enumerate sub-millimeter particles. It typically involves subsampling areas of a filter and manual counting of particles through a microscope lens.
- **Dye staining microscopy** is a method that stains plastic particles with a fluorescent dye. Nile Red, one of the most commonly used dyes, is selective to plastics, and stained particles can easily be identified under a fluorescence microscope.
- **Scanning Electron Microscopy (SEM)** is an imaging technique that provides high-resolution images of the sample, allowing for the identification of very small particles, the polymer type, and morphological characteristics.

Table 3-14 lists the enumeration methods used by the studies reviewed. Optical microscopy was the most common method used in 28 of 39 studies, followed by a small number of studies that used visual detection. Nile red florescence was used by one study.

Table 3-14. Enumeration Methods (N=36)

Enumeration Method	Number of Articles	Journal Articles
Optical microscopy	28	Baldwin et al., 2016; Ballent et al., 2016; Belontz et al., 2022; Brookson et al., 2019; Cable et al., 2017; Casteñeda et al., 2014; Corcoran et al., 2020b; Cox et al., 2021; Davidson et al., 2022; Dean et al., 2018; Eriksen et al., 2013; Fox et al., 2022; Grbic et al., 2020; Hendrickson et al., 2018; Hoellein et al., 2021; Hou et al., 2021; Lenaker et al., 2019; Lenaker et al., 2021; Mason et al., 2020; McCormick et al., 2014; McNeish et al., 2018; Minor et al., 2020; Munno et al., 2022; Peller et al., 2021; Schessl et al., 2019; Vincent and Hoellein, 2021; Wagner et al., 2019
Visual (naked eye)	7	Arturo and Corcoran, 2022; Corcoran et al., 2015; Corcoran et al., 2020a; Costello and Ebert, 2020; Hoellein et al., 2014; Lazcano et al., 2020; Zbyszewski et al., 2014
Dye staining (Nile red)	1	Crew et al., 2020

Cowger et al. (2020a) conducted a review of 127 peer-reviewed publications to assess the most common visual techniques used for identification of microplastics and found that optical microscopy was by far the most common technique, similar to the Great Lakes studies evaluated for this project. In their review of microplastics studies, Prata et al. (2019) found that almost all studies use visual inspection as the first step in microplastic identification and enumeration, and to identify particle characteristics such as shape and color. Some studies used other methods to verify particles were plastic alongside visual microscopy methods. These methods include scanning electron microscopy with energy dispersive X-ray spectroscopy (SEM/EDS), attenuated total reflectance (ATR)/Fourier transform infrared spectroscopy (FTIR), differential scanning calorimetry, and melt tests (Cable et al., 2017; Casteñeda et al., 2014; Eriksen et al., 2013; Fox et al., 2022; Mason et al., 2016; Mason et al., 2020; Wagner et al., 2019). Primpke at al. (2020) provides a review of the most commonly used visual methods. Table 3-15 discusses the applicability of these visual techniques.

Table 3-15. Applicability of Visual Methods

Method	Detection Limit	Notes
Visual (naked eye)	1 mm	Easy, inexpensive, does not require specialized equipment, high subjectivity
Optical microscopy	100 µm	Easy, relatively inexpensive and requires minimal equipment, subjective
Dye staining (Nile red)	3 µm	Specialized light sources and chemicals add to cost; organic matter must be completely digested to minimize false positives

Source: Primpke et al., 2020

3.3.2 Chemical Identification

The following methods were used to confirm that a particle was plastic and to determine polymer type.

- **Fourier Transform Infrared Spectroscopy (FTIR)** uses the absorption and transmission of infrared light to identify the chemical bonds present in a sample, allowing for the identification of the polymer type of microplastics.
- **Raman Spectroscopy** uses laser light to excite a sample and analyze the scattered light to identify polymer type.

- **SEM with Energy Dispersive X-ray Spectroscopy (EDS)** is a high-resolution imaging technique combined with EDS, a system that analyses X-rays dispersed during electron scanning, providing information on the composition and chemical makeup of the plastic.
- **Pyrolysis, Gas Chromatography, and Mass Spectrometry (Pyr-GC/MS)** is a destructive technique that uses high heat to break down the plastic into its component chemicals, which are then analyzed using gas chromatography (GC) and mass spectrometry (MS) to determine its physical properties and composition.
- **Melt test** is a destructive technique that involves heating particles to the melting point of suspected plastic polymers to confirm whether they are plastic or not.
- **Differential Scanning Calorimetry** involves heating a plastic sample to measure the change in temperature with heat input, especially during phase changes. The resulting curve can be compared to a library of known curves to identify the material.
- **X-Ray Fluorescence (XRF)** uses x-rays to bombard the material and make it fluoresce. The fluorescence is then analyzed to determine the chemical composition of the material.

Table 3-16 lists the chemical identification methods used by the studies reviewed. FTIR was the most common method used, followed by Raman and SEM/EDS. A smaller number of studies used other methods including pyr-GC/MS, melt test, differential scanning calorimetry, and XRF.

Table 3-16. Chemical Identification (N=25*)

Identification Method	Number of Articles	Journal Articles
FTIR	21	Arturo and Corcoran, 2022; Belontz et al., 2022; Brookson et al., 2019; Corcoran et al., 2015; Corcoran et al., 2020a; Corcoran et al., 2020b; Davidson et al., 2022; Dean et al., 2018; Fox et al., 2022; Grbic et al., 2020; Hendrickson et al., 2018; Hoellein et al., 2021; Lenaker et al., 2019; Lenaker et al., 2021; Mason et al., 2016; Mason et al., 2020; McNeish et al., 2018; Munno et al., 2022; Vincent and Hoellein, 2021; Wagner et al., 2019; Zbyszewski et al., 2014
Raman	8	Brookson et al., 2019; Ballent et al., 2016; Corcoran et al., 2015; Dean et al., 2018; Grbic et al., 2020; Hou et al., 2021; Munno et al., 2022; Peller et al., 2021
SEM/EDS	2	Minor et al., 2020; Zbyszewski et al., 2014
Pyr-GC/MS	3	Hendrickson et al., 2018; Lenaker et al., 2019; Minor et al., 2020
XRF	1	Ballent et al., 2016

*Note some studies used multiple methods. See text for acronym definitions.

In their summary of peer-reviewed studies, Cowger et al. (2020a) found that FTIR was by far the most common technique for chemical identification, followed by Raman spectroscopy, pyr-GC/MS, and EDS. Primpke et al. (2020) provide a review of the most commonly used chemical characterization methods. Table 3-17 discusses the applicability of these techniques.

Table 3-17. Applicability of Polymer Characterization Methods

Method	Minimum Particle or Sample Size	Notes
FTIR	10 µm	µFTIR techniques, combining microscopy with FTIR, can detect particles down to 10 µm. Requires specialized equipment and supplies, provides a cost-efficient way to gain information about particle numbers, polymer types, and sizes simultaneously.

Method	Minimum Particle or Sample Size	Notes
Raman	1 µm	Higher resolution, more accurate, and less prone to interference than FTIR, but requires more expensive equipment and more time for analysis. Laser light can damage particles at higher intensities, making them ineligible for further analysis.
Pyr-GC/MS	~1 µg	Single particles can be reliably identified; can be used to quantify sample sizes down to a few µg. Can be more time-consuming than FTIR or Raman if analyzing individual particles. Particles must be free of contamination. Destructive technique.
EDS	~1 nm	Requires an SEM. Light elements such as H, C, N, and O are more difficult to determine accurately than metals; further, most polymers are composed primarily of C and O, making identification challenging. Nanometer-sized particles may be difficult to identify due to elemental ratios being affected by surface oxidation or adsorbed moisture and metals.

Source: Primpke et al., 2020. See text for acronym definitions.

For samples with a large number of particles (>100), subsampling may be necessary to efficiently process the sample. De Frond et al. (2023) recommend using a random protocol for subsampling that does not rely on visual selection of particles. Additionally, the minimum subsample size varies depending on study goals; a smaller subsample size may be needed if the goal is simply to identify particles as plastic or non-plastic, compared to a study that aims to characterize polymer types.

3.4 QA/QC

Microplastic particles are ubiquitous in the environment, and many common items are made of plastic including clothing, collection nets, and other field and laboratory equipment. Therefore, the studies reviewed used a range of methods to prevent or measure microplastic contamination of samples during collection and analysis. Additionally, some studies implemented methods to assess the recovery rates of their laboratory procedures. Table 3-18 summarizes the QA/QC methods that were discussed in the studies reviewed.

Table 3-18. QA/QC Methods (N=24*)

QA/QC Method	Number of Articles	Journal Articles
Rinsing equipment with filtered or reverse osmosis water	19	Baldwin et al., 2016; Ballent et al., 2016; Belontz et al., 2022; Brookson et al., 2019; Cable et al., 2017; Corcoran et al., 2020b; Crew et al., 2020; Davidson et al., 2022; Dean et al., 2018; Fox et al., 2022; Grbic et al., 2020; Hendrickson et al., 2018; Hoellein et al., 2021; Holland et al., 2016; Mason et al., 2016; McNeish et al., 2018; Minor et al., 2020; Peller et al., 2021; Wagner et al., 2019
Use of cotton lab coats	12	Baldwin et al., 2016; Ballent et al., 2016; Belontz et al., 2022; Cable et al., 2017; Corcoran et al., 2020b; Crew et al., 2020; Davidson et al., 2022; Dean et al., 2018; Grbic et al., 2020; Mason et al., 2016; Minor et al., 2020; Vincent and Hoellein, 2021
Field blanks	11	Baldwin et al., 2016; Cable et al., 2017; Corcoran et al., 2020b; Crew et al., 2020; Davidson et al., 2022; Fox et al., 2022; Hendrickson et al., 2018; Hoellein et al., 2021; Mason et al., 2020; Minor et al., 2020; Wagner et al., 2019
Ventilation or air filtration	5	Belontz et al., 2022; Cable et al., 2017; Crew et al., 2020; Grbic et al., 2020; Mason et al., 2016
Cleaning surfaces or equipment with acid or solvents	4	Ballent et al., 2016; Eriksen et al., 2013; Holland et al., 2016; Wagner et al., 2019
Using metal or glass equipment	2	Ballent et al., 2016; Corcoran et al., 2020b

QA/QC Method	Number of Articles	Journal Articles
Method or lab blanks	19	Ballent et al., 2016; Belontz et al., 2022; Brookson et al., 2019; Corcoran et al., 2020b; Cox et al., 2021; Crew et al., 2020; Dean et al., 2018; Fox et al., 2022; Hoellein et al., 2021; Hendrickson et al., 2018; Hou et al., 2021; Lenaker et al., 2019; Lenaker et al., 2021; McCormick et al., 2014; McNeish et al., 2018; Minor et al., 2020; Peller et al., 2021; Vincent and Hoellein, 2021
Matrix spikes	3	Crew et al., 2020; Lenaker et al., 2021; Minor et al., 2020

*Note some studies used multiple methods.

Brander et al. (2020) and Miller et al. (2021) reviewed and recommended QA/QC methods for microplastics sampling, analysis, and reporting, for different matrices and methods. Their recommendations generally include the steps highlighted in the table above. However, not all the 39 Great Lakes studies reviewed reported following all the recommended QA/QC steps. For example, 2 studies confirmed using only metal or glass equipment, only 3 reported using matrix spikes, and 11 and 19 studies, respectively, reported using field and method blanks.

Cui et al. (2022) discuss the need to standardize the microplastics used to prepare matrix spikes. They recommend that microplastic standards contain at least three types of polymers with three different densities (< 1.0 g/cm^3, ~1.0 g/cm^3, and >1.0g/cm^3), three shapes (fibers, fragments, and pellets), and a similar size range as the sample. Other ongoing research (Akhbarizadeh et al., 2023) also suggests that microplastics used for spike recovery samples should possess similar characteristics as the sample being analyzed, including the presence of organic matter to mimic effects of digestion. Akhbarizadeh et al. (2023) also emphasize the need for control of sample contamination and the use of field and laboratory blanks.

3.5 Standardization and Harmonization of Methods

One of the challenges in studying microplastics in the environment is the diversity of particle sizes, shapes, and characteristics (e.g., density), and the extent to which different sampling and analytical methods can capture a representative sample of particles (Twiss, 2016; Rochman et al., 2019). For example, manta trawls and other net-based methods allow coverage of a larger geographical area but cannot capture particles smaller than the mesh size, while grab samples can capture a wider range of sizes but are more limited in geographic coverage. As a result, it is often not possible to directly compare the results from studies that collect microplastics particles using different sampling methods and to present results across different size ranges.

3.5.1 Aligning Concentration Data Across Size Ranges

In an effort to improve the comparability of monitoring studies that capture particles of different size ranges, Koelmans et al. (2020), Kooi and Koelmans (2019), and Kooi et al. (2021) propose a method to rescale particle concentrations from the reported size range to the desired (i.e., target) size range, using probabilistic modeling of microplastic particle characteristics (size, shape, and density) across various environmental compartments (freshwater surface water, marine surface water, freshwater sediments, marine sediments, freshwater biota, and marine biota). Their method involves the calculation of a correction factor based on the reported and desired size ranges (based on a typical particle distribution for environmental microplastics), and a slope factor calibrated for the compartment of interest, as follows:

$$EC_{env} = EC_{meas} \times CF_{meas}$$

and

$$CF_{meas} = \frac{L_{UL,D}^{1-\alpha} - L_{LL,D}^{1-\alpha}}{L_{UL,M}^{1-\alpha} - L_{LL,M}^{1-\alpha}}$$

Source: Kooi et al., 2021

Where:

- EC_{env} is the environmentally realistic concentration across the desired size range
- EC_{meas} is the measured concentration across the reported size range
- CF_{meas} is the measurement correction factor
- $L_{UL,D}$ is the upper limit of the desired size range
- $L_{LL,D}$ is the lower limit of the desired size range
- $L_{UL,M}$ is the upper limit of the measured size range
- $L_{LL,M}$ is the lower limit of the measured size range
- α is a slope factor, specific to the compartment of interest

Kooi et al. (2021) report different values for the slope factor for different characteristics (i.e., length, width, volume, and surface area). If the particle size classification is based on the longest dimension, i.e., length, the corresponding value of α should be used. Models typically have a lower limit where the fit is valid which ranges from 56 – 354 µm.

For example, if a surface water concentration (in particles per unit volume) measured for a range from 330 to 2,000 µm needs to be rescaled to the default definition range from 1 to 5,000 µm, then the values for $L_{UL,D}$, $L_{LL,D}$, $L_{UL,M}$, and $L_{LL,M}$ would be 5,000; 1; 2,000; and 330, respectively. For freshwater surface water, Kooi et al. (2021) report a value of α of 2.68 for particle length. The corresponding conversion factor value equals approximately 17,893.

4. Monitoring Results

As discussed earlier, 43 studies were reviewed that performed sampling within the Great Lakes and their connecting waters and tributaries. The field is still relatively new, and the number of studies published continues to increase. Of the studies reviewed, the majority were based on sampling events conducted in the mid-to-late 2010s. Delays between study sampling and publication ranged from 1 to 7 years. Figure 4-1 depicts the cumulative number of the reviewed studies published since 2008 and the number of sampling events per year.

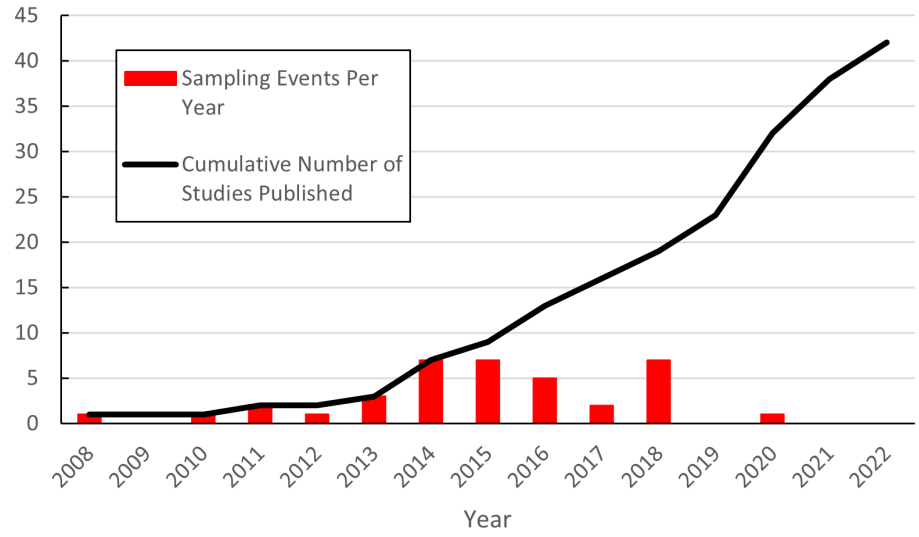


Figure 4-1. Trends in Great Lakes Microplastics Monitoring Studies

At least one study was performed for microplastics in each matrix in each Great Lake. The most studies were performed in Lake Ontario followed by Lake Michigan, Lake Erie, Lake Superior, and Lake Huron. More studies were conducted in Lake St. Clair than in any other connecting water or tributary. Surface water remains the most sampled matrix followed by sediment, surface debris, and biota. A summary of the spatial extent of microplastics sampling by lake is shown in Figure 4-2 below.

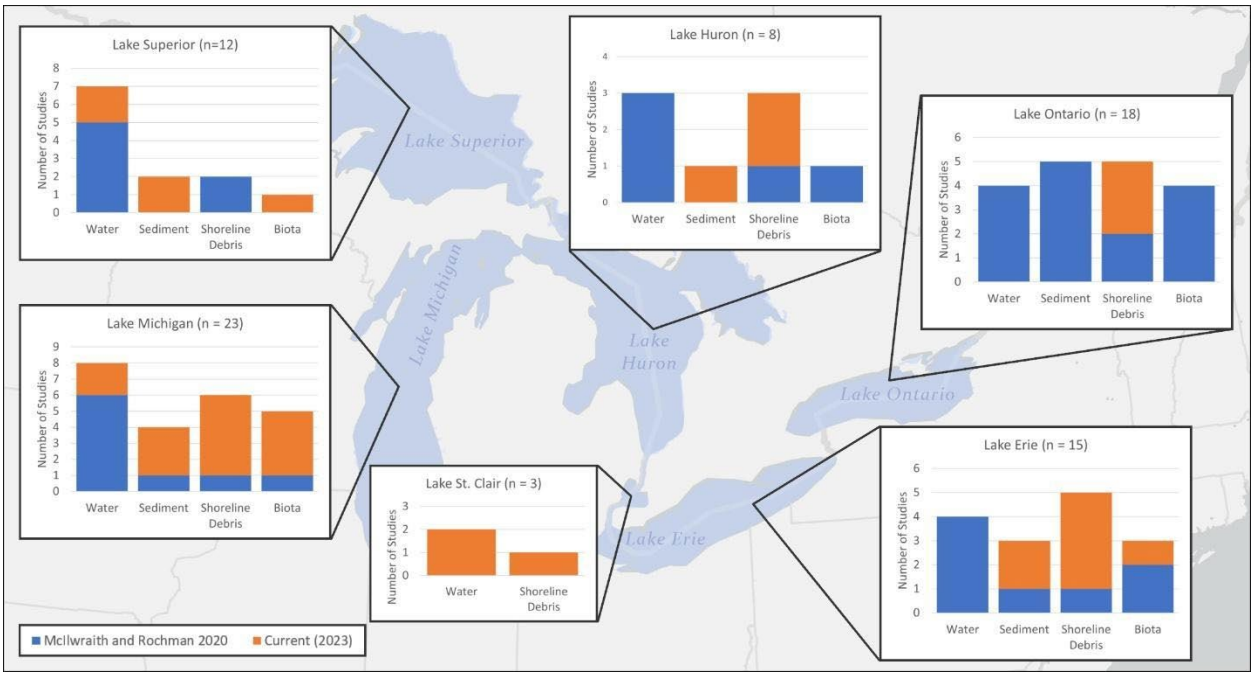


Figure 4-2. Great Lakes Microplastics Monitoring Studies by Lake (Including Lake St. Clair)

Of the studies reviewed, seven studies sampled macroplastics in shoreline debris (i.e., beaches), which were defined as sample sets that reported minimum sizes greater than 5 mm. Macroplastics were not sampled in any other matrix. Figure 4-3 summarizes the numbers of studies that studied macroplastic contamination, by lake. Note that some studies looked at multiple lakes.

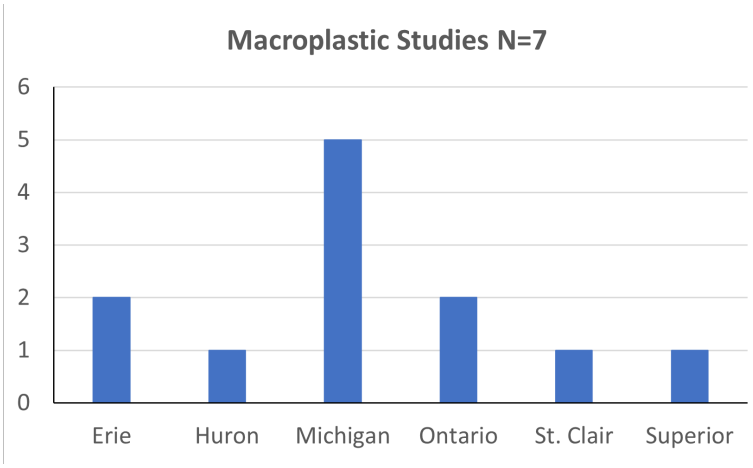


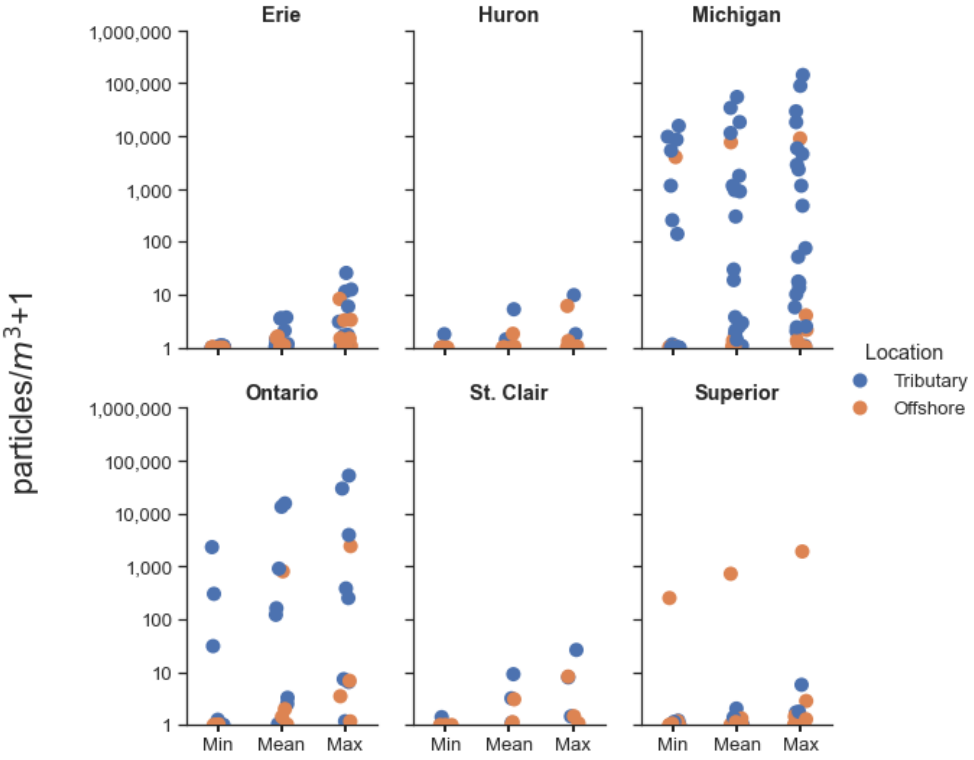
Figure 4-3. Macroplastics Monitoring Studies by Lake (Including Lake St. Clair)

4.1 Surface Water

Seventeen studies (see Table 3-1) collected and analyzed surface water samples for the presence of microplastics. Microplastics concentrations in surface water can vary significantly by method (i.e., grab sampling vs. manta trawls); as discussed in Section 3.1.1, concentrations reported by studies that

collected grab samples were generally much higher than those that collected samples from trawling, likely due to the capture of more small-diameter particles by grab sampling methods. Figure 4-4 shows results from sampling of surface water for each lake. Studies that used composite sampling were grouped into the grab sampling category for these charts.

Based on the studies reviewed, surface water samples from Lake Michigan appear to have the highest mean concentrations of microplastics, for grab samples as well as trawling. Lakes Erie and Ontario appear to have the next highest concentrations, followed by Lakes Huron and Superior. Only two studies sampled Lake St. Clair, where microplastic concentrations appear to be comparable to Lakes Ontario and Erie. Additionally, studies that sampled tributaries generally report significantly higher microplastic concentrations than studies that sampled the lakes.



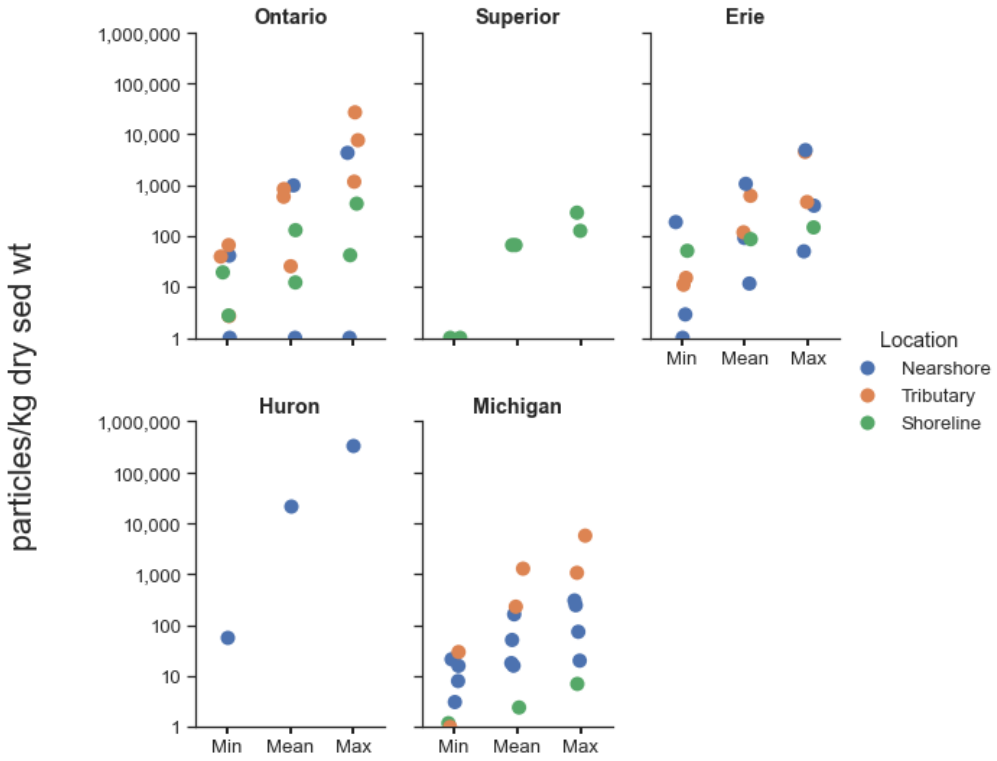
Concentrations of microplastics in water samples by lake. Points represent results from the studies reviewed for each individual sampling event and may summarize multiple samples across a single lake or other waterbody. Concentrations were scaled using a log_{10}([particles/m^3] +1) transformation.

Figure 4-4. Microplastics Concentrations in Surface Water, by Lake

4.2 Sediment

Fourteen studies (see Table 3-3) collected sediment samples and analyzed them for microplastics. Sediment concentrations showed significant variability, even within studies. Figure 4-5 summarizes sediment concentrations by lake. Because of the range of reported concentrations across studies, the data are presented using a logarithmic scale.

Based on the studies reviewed, sediment samples from Lake Michigan appear to have the highest mean concentrations of microplastics. Lakes Ontario, Erie, and Huron have the next highest reported concentrations, followed by Lake Superior. Note that only one study examined sediment concentrations in Lake Huron, and no studies sampled sediments from Lake St. Clair. Microplastic concentrations in nearshore sediment were sometimes higher than sediment from tributaries or beaches. This may suggest that lake sediments are acting as sinks for microplastic particles.



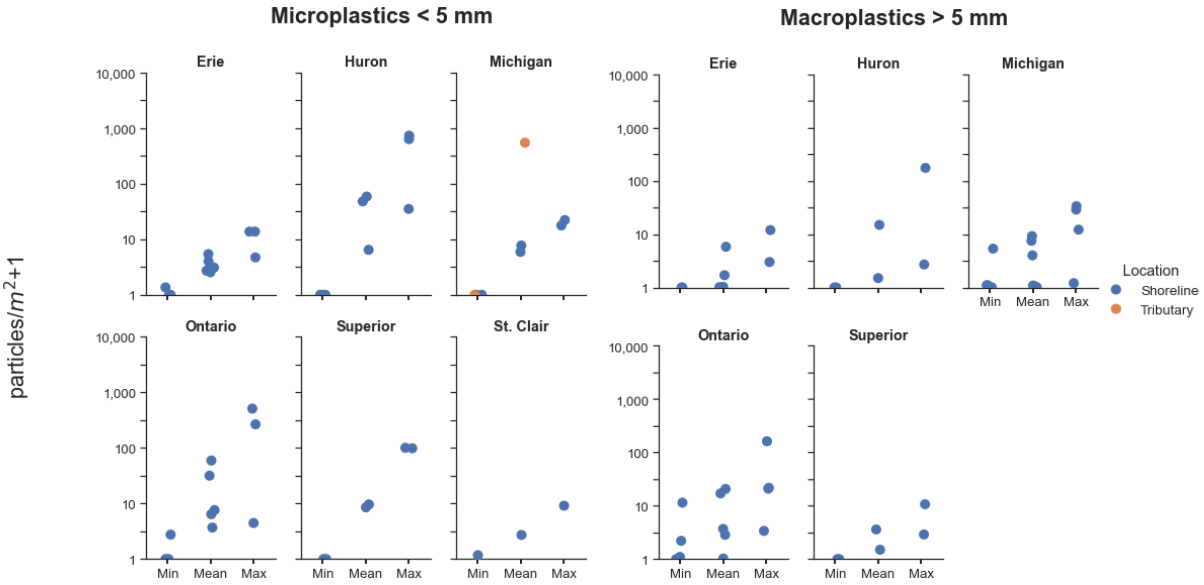
Concentrations of microplastics in sediment samples by lake. Points represent results from the studies reviewed for each individual lake sampled and may summarize multiple samples. Concentrations were scaled using a log_{10}([particles/kg dry sed wt.] +1) transformation. One study that did not report data as number of particles/kg dry sediment weight has been omitted. Note that shoreline samples in this figure are terrestrial sediment samples reported as particles/kg dry sed wt. Shoreline debris samples are discussed in the next section and are studies that reported concentrations as particles/m^2.

Figure 4-5. Microplastics Concentrations in Sediment, by Lake

4.3 Shoreline Debris

Eleven studies (see Table 3-5) looked at plastic debris on beach surfaces. The concentrations reported by these studies are shown in Figure 4-6; note that concentrations of microplastic and macroplastic surface debris are shown separately, with different scales along the vertical axes. In general, macroplastic concentrations were significantly higher than microplastic concentrations since methods used in these studies were generally not suitable for collecting small particles. Minimum size reported for microplastics in this matrix was mostly > 1 mm, with only three studies reporting minimum sizes < 1 mm.

Based on mean concentrations reported by the studies reviewed, beaches in Lakes Ontario and Huron appear to have the highest microplastics concentrations followed by Lakes Michigan, Superior, and Erie. Macroplastics concentrations appear to follow a similar trend. These findings may need to be interpreted with caution because the data were collected at different times of the year and beaches are likely to have significant variation in the amount of litter present, based on the season, the extent of visitor traffic, and cleaning operations. Vincent and Hoellein (2017), for example, observed lower levels of pollution in the summer as a result of municipal beach cleanings throughout the summer, followed by an increase in plastic litter as cleaning was reduced or halted in the autumn and spring seasons.



Concentrations of microplastics and macroplastics in shoreline beach samples by lake. Points represent results from the studies reviewed for each individual lake sampled and may summarize multiple samples across a single lake. Concentrations were scaled using a $log_{10}([particles/m^2] +1)$ transformation.

Figure 4-6. Surface Debris Concentrations on Beaches, by Lake

4.4 Biota

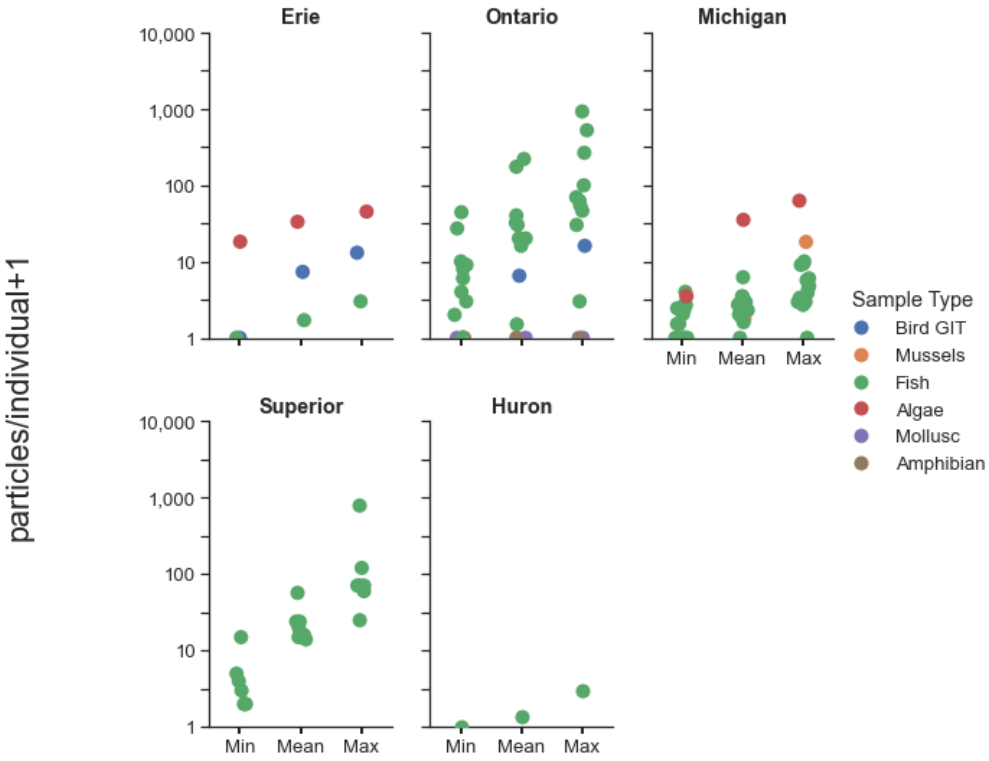
Nine studies (see Table 3-6) sampled and analyzed microplastic concentrations in biota, across 28 different species. Most of these studies were conducted in Lakes Michigan and Ontario, and no studies sampled biota in Lake St. Clair. The species of biota sampled are shown in Table 4-1 below, by lake. Fish were the most commonly sampled biota (n=4), followed by birds and molluscs (n=2, each) and algae and anurans (n=1, each). Fish were also the only class of biota sampled across all five Great Lakes. Algae were only sampled in Lakes Erie and Michigan and had similar reported mean concentrations of microplastics in both lakes. Algae samples included floating algae, filaments from the lake bottom, and growth attached to breakwall rocks.

Table 4-1. Biota Sampled by Lake

Biota Sampled	Erie	Huron	Michigan	Ontario	Superior
Algae (n=1)					
Cladophora	x		x		
Amphibian (n=1)					
Anuran				x	
Bird (n=2)					
Double crested cormorants	x			x	
Long-tailed duck				x	
White-winged scoter				x	
Fish (n=4)					
Banded killifish			x		
Bass sp.			x		
Brown bullhead				x	
Cisco					x
Common shiner				x	
Emerald shiner			x	x	
Fathead minnow			x	x	
Gizzard shad			x		
Lake trout		x			x
Lake whitefish					x
Largemouth bass			x		
Longnose sucker					x
Quillback			x		
Rainbow trout				x	
Round goby			x	x	
Round whitefish					x
Sand shiner			x		
Smallmouth bass	x				
Spotfin shiner			x		
Spottail shiner			x	x	
White sucker			x	x	x
Yellow perch				x	x
Mollusc (n=2)					
Dreissenid mussels			x	x	

Lake Ontario had the highest reported microplastic particles per individual in fish, followed closely by Lake Michigan and then by Lake Superior. Microplastic particle counts also varied by species. McNeish et al. (2018), for example, surveyed 11 fish species and reported that the most contaminated fish was the round goby (*Neogobius melanostomus*), containing an average of 19 particles/individual.

Reported concentrations of microplastics in biota are shown in Figure 4-7 below. Because of the large range of reported concentrations across studies, the data are presented using a logarithmic scale. Note that all concentrations are reported as number of particles per individual, except for algae which are reported as number of particles per g of dry weight.



Microplastics in biota samples by lake. Points represent results from the studies reviewed for each individual lake sampled and may summarize multiple samples across a single lake. Concentrations were scaled using a log_{10}([particles/individual]+1) transformation. Studies that did not report data in particles/individual (except algae which were particles/g dry weight) were omitted.

Figure 4-7. Microplastics in Biota, by Lake

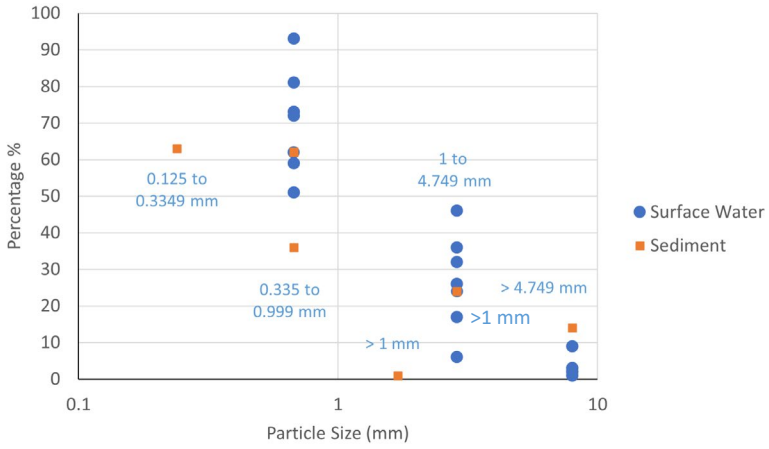
It is not clear why reported microplastic concentrations in fish do not follow trends in microplastic concentrations in lake water, where Lake Michigan had the highest concentrations followed by Lake Ontario and then Lake Erie. However, there are some confounding factors that can explain why microplastic abundance in fish and lake water are not correlated. For example, fish size may be a factor, as larger fish may accumulate a greater quantity of microplastic particles over time (see, for example, Gad and Midway, 2022). Sampling locations may also be a factor, since microplastic concentrations may be higher near urban areas and therefore sampling for water (and potentially fish as well) in these hot spots may result in higher reported concentrations than elsewhere in the lakes.

Other, recent studies have attempted to understand microplastic concentrations and sources in the Great Lakes biota and in the environment. For example, Weir et al. (2024) studied the effect of WWTP discharge on microplastic concentrations in the ambient environment and caged and resident biota in the Grand River, ON. Their results show that microplastics are elevated in a few taxa and environmental

samples downstream of WWTPs, but microplastic presence was similar across most sites in both biotic and abiotic compartments and suggest widespread contamination of the river. Kazmierczak et al. (2023) studied microplastic abundance in six fish species at sites upstream, at the effluent release, and downstream a large WWTP outflow in Chicago, IL, USA. With all species combined, microplastic concentrations were not significantly different across study sites, but microplastic abundance relative to WWTP proximity varied by species. Last, gut contents were more strongly affected by site than species.

4.5 Size, Shape, and Polymer Distributions

Twelve studies reported particles by size class (see Table 3-10); however, they did not use consistent size classes to analyze microplastic concentrations. Of the 12 studies, 8 studies (7 surface water and 2 sediment) used size classes that were either fully or partially overlapping. Size distributions for those eight studies are shown in Figure 4-8 below. In general, all studies that reported particle distributions by size class reported higher percentages for smaller sizes, suggesting that smaller particles make up a larger fraction of the overall particle distribution across matrices.



Blue dots represent surface water studies; orange dots represent two sediment studies that used different size classes. Size classes are shown on the chart in blue text.

Figure 4-8. Particle Proportions by Size Class, as Percentage of Total

Figure 4-9 summarizes the percentage distributions of microplastic particles by shape for each matrix. Note that not all studies reported shapes; the numbers at the bottom of each bar indicate how many studies enumerated shapes for each matrix. Fibers are the predominant shape, accounting for nearly 90% of the particles identified in biota, 60% of the particles in shoreline beach debris, 56% of the particles in surface water, and 46% of the particles in sediment. Fragments are the second most common shape, followed by pellets (on beaches) and spheres (in sediment). Foams and films are other shapes that were reported. Figures 3-2 and 3-4 show shape distributions broken out by sampling method, for water and sediment matrices respectively.

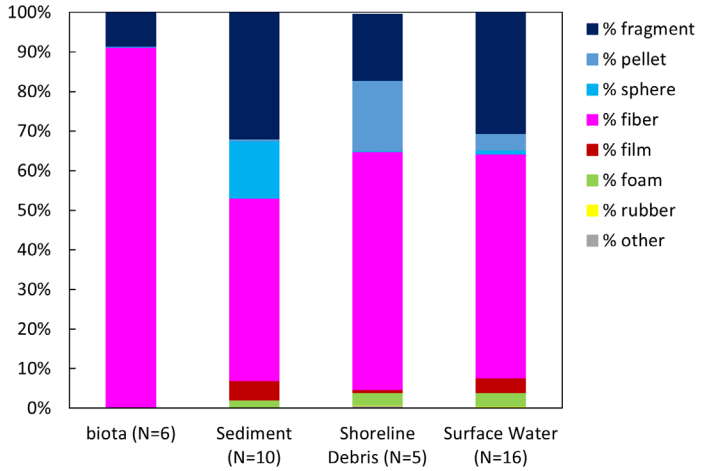


Figure 4-9. Shape Percentage Distributions, by Matrix

Figure 4-10 summarizes the percentage distributions of microplastic polymers for each matrix. Note that not all studies reported polymers; the numbers at the bottom of each bar indicate how many studies identified polymer types, for each matrix. Polyethylene was the most common polymer type in sediment, surface water, and surface debris, while "other plastic" was the most common category in biota (including acrylic, rayon, nylon, polyester, styrene-acrylonitrile, and unknown). Other commonly reported polymer types include polyester, polypropylene, and polystyrene, as well as smaller proportions of PVC and polyurethane. Figures 3-2 and 3-4 show polymer distributions broken out by sampling method, for water and sediment matrices respectively.

Lenaker et al. (2021) reported that polyester, HDPE, and semi synthetic cellulose were most common among particles in Lake Erie sediment. Polypropylene and PVC were the most common in Lake Michigan sediment. EPS and polypropylene were more abundant in surface waters, and polyester was more abundant in deeper waters and in sediment.

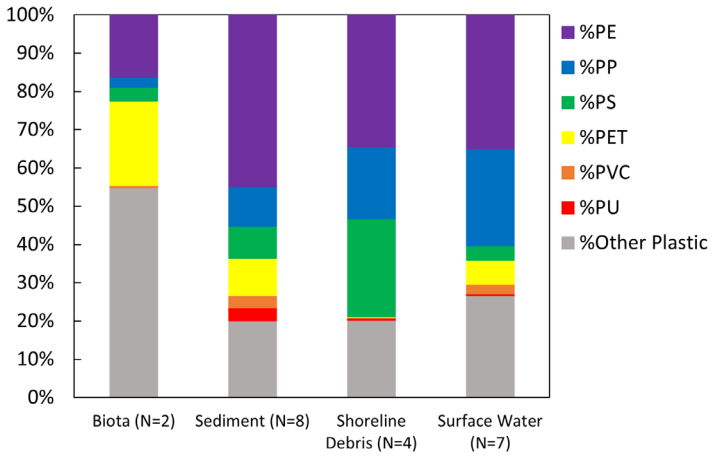


Figure 4-10. Polymer Percentage Distributions, by Matrix

4.6 Sources, Pathways, and Fate of Microplastics

4.6.1 Sources and Pathways

Marine traffic, road-wear, beach litter, urban areas, agriculture, and industry are all sources of microplastics in the Great Lakes (Earn et al., 2021). Typically, microfibers may originate from clothing and are likely to be transported via wastewater discharge or ambient air; other sources of fibers may include direct wear and tear on fishing nets, ropes, and other marine equipment and debris. Rubbery particles are typically associated with road tire wear and are likely transported via stormwater runoff; similarly, plastic pellets from industry can be easily recognized based on their shape and size. For other types of microplastic particles, however, it may be challenging to ascertain origin and pathways because plastics are so ubiquitous in the human environment (Helm, 2017).

Wang et al. (2022) conducted a systematic review of literature to identify and quantify potential pathways for microplastics to enter freshwater ecosystems. They focused on wastewater, stormwater, agricultural discharge, and industrial discharge as potential pathways. Wastewater was found to be the most well-studied pathway, reflecting in part the long-standing understanding of wastewater as a major pathway for microplastics to enter aquatic ecosystems. Stormwater was also relatively well-studied, especially as concern over tire wear increases. Note that in the Great Lakes, combined sewers are commonly present and may therefore serve as a single pathway for microplastics in stormwater and wastewater. Stormwater ponds can serve as retention sites, and as conduits for further discharge of microplastics to receiving waters. Stormwater ponds in industrial and commercial areas may have higher concentrations of microplastics compared to stormwater ponds receiving residential or highway runoff.

Industrial and agricultural discharge are less well-documented in the literature. Industrial facilities are known to be potential sources for plastic discharge, especially of pre-production pellets from plastics manufacturing facilities and fibers from textile mills. Plastics are also used in agricultural applications, e.g., as fertilizer pellets, and can degrade into microplastics over time. Additionally, WWTP biosolids that are applied to agricultural sites can contain significant amounts of microplastics, and these microplastics can enter waterbodies via stormwater discharge over time. Stormwater and wastewater were generally found to have higher concentrations of microplastics than industrial and agricultural runoff. However, flow rates are needed to characterize total loadings of microplastics to water bodies through these pathways (Wang et al., 2022).

Fahrenfeld et al. (2019) reviewed evidence and methods for source apportionment of microplastics in freshwater environments including the use of microplastic characteristics, mass balance techniques, and surface characteristics. They noted that some studies have attempted to use microplastic characteristics, including morphology and polymer type, to attribute them to a specific source. In addition, surface contaminants including metals, organic compounds and biofilms may provide some evidence of microplastic source, but complications arise as environmental contaminants can also associate with freshwater microplastics in situ. Challenges associated with source tracking based on particle characteristics include limitations to accurate polymer identification, poor understanding of weathering processes, and limited understanding of microplastic transport mechanisms within freshwater systems.

4.6.2 Fate

Once microplastics enter the aquatic environment in the Great Lakes basin, they may end up on beaches, deposited to lake sediments, suspended in the water column, consumed by organisms, or

exported out of the system downstream and ultimately in the ocean. Their transport is greatly influenced by their properties which can be altered through biofilm formation (e.g., bacteria, algae, and fungi) on their surfaces, ingestion by organisms, and weathering and degradation by various mechanisms (Helm, 2020).

The fate of microplastics within aquatic and other organisms remains a subject of active research. Recent modeling efforts have attempted to predict the effects of wind and water currents on the accumulation and transport of microplastics in the Great Lakes (Helm, 2020). These studies have predicted localized areas of microplastic accumulation in the lakes that show some agreement with observed concentrations. However, additional work remains to be done to improve our understanding of microplastic accumulation in the Great Lakes. A model of input/outputs and sinks in Lake Geneva in Switzerland, for example, suggested that sediments contain much of the plastic stock in the lake (580 tonnes) compared to surface waters (0.1 tonnes) (Boucher et al., 2019). Of the 55 tonnes estimated to enter the lake annually, 40 tonnes were calculated to enter the sediment, 10 tonnes removed via cleanups, and 5 tonnes exits the lake through the outflow.

5. Exposure to and Effects of Microplastics in Freshwater Organisms

Current research shows that microplastics can have negative effects on aquatic organisms. However, the level at which effects are observed in laboratory studies vary based on particle characteristics, test organisms, and environmental media (Campanale et al. 2020). Moreover, the types of effects studied are diverse, making it difficult to understand the main mechanisms of effects. At present, researchers think there are two pathways for toxicity that are best understood. These are food dilution, which refers to when microplastics replace nutritional diet that may lead to changes in development or reproduction, and tissue translocation, or the uptake of microplastic particles into other tissues that may lead to effects such as inflammation and oxidative stress (Koelmans et al., 2023). Additionally, microplastics may potentially leach monomers, plasticizers, or other additives when ingested by organisms. They can also expose them to other contaminants that may have sorbed to the surface of the microplastics in transit, or serve as a substrate for potentially harmful microbial communities; however, these mechanisms are still under investigation. Regardless, the characteristics of the microplastics as well as the mechanisms of effects should be considered when assessing the toxicity of microplastics.

5.1 Literature Review

Our literature review on the ecotoxicology of microplastics (described in Section 1.1) identified 62 journal articles relevant to the Great Lakes. We reviewed these articles for information such as the particles tested, the exposure concentrations, test organisms, and biological endpoints studied, and whether effects were observed. The full list of papers is included in Appendix A.3. Table 5-1 summarizes information from the literature review by taxon and plastic property, with higher numbers indicative of more studies having focused on a specific property of plastic and its effects on certain taxa.

Table 5-1. Freshwater study counts by organism group and microplastic characteristic.

	Polymer Type						Particle Shape				Particle Size			
	Polyethylene	Polypropylene	Polystyrene	Polyamide	Polyvinyl Chloride	Mixture	Fiber	Nurdle (Sphere)	Fragment	Mixture	1 nm - 100 nm	100 nm - 1 um	1 um - 100 um	100 um - 1 mm
Zooplankton	10		8				3				2		9	
Nematode	2	1	4	1	1	1		1	1	2			3	
Amphipod	1	1	1			1	1	1		1				
Phytoplankton	2		5								1			
Macrophyte	1		2					1			1		1	
Macroinvertebrate	3		3			1		3		1		1	5	2
Mollusc	1		4			1		1	1	1	1		2	
Fish (fry, small)	2	2	3	1	1			2	1		1		3	
Fish (adult, large)	2		6								1	1	1	2

The color of the cells indicates the relative frequency of each particle category for each taxon, with yellow least common and dark green most common.

Overall, studies investigated the effects of microplastics using a variety of polymer types, particle sizes, and particle shapes, with polystyrene being the most studied polymer type. In the studies reviewed, zooplankton was studied the most and amphipods and macrophytes were studied the least. Spheres

were the most studied particle shape, and the size range of 1 – 100 µm was the most studied particle size.

Native and non-native organisms related to the Great Lakes for which toxicity studies were found are listed in Table 5-2. There is a need for more toxicity studies to be done on test organisms that naturally occur in the Great Lakes, based on the literature review. Future studies informing risk should also test multiple different concentrations of microplastics to inform risk thresholds for toxicity. There should be ideally 5 to 6 concentrations with at least one control and include higher concentrations and diverse biomarkers within the design to inform effect mechanisms.

Table 5-2. Great Lakes Organisms with Available Microplastics Toxicity Data

Organism Group	Species
Fish	*Onchorhynchus mykiss* (rainbow trout)
	Ictalurus punctatus (channel catfish)
	Perca flavescens (yellow perch)
	Carassius auratus (goldfish)
	Danio rerio (zebrafish)
	Neogobius melanostomus (round goby)
	Gasterosteus aculeatus (three-spined stickleback)
	Oryzias melastigma (medaka)
	Oryzias latipes (medaka)
Plankton	*Strombidium sulcatum* (zooplankton)
	Parvocalanus crassirostris (zooplankton)
	Tigriopus fulvus (zooplankton)
	Tigriopus japonicus (zooplankton)
	Scenedesmus armatus (phytoplankton, colonial green algae)
	Brachionus plicatilis (rotifer)
	Brachionus calyciflorus (rotifer)
	Brachionus fernandoi (rotifer)
Invertebrates	*Aphylla williamsoni* (two-striped forceptail dragonfly)
	Daphnia pulex (water flea)
	Moina macrocopa (water flea)
	Gammarus pulex (gammarus shrimp)
	Chironomus riparius (midge fly)
	Sphaerium corneum (pea clam)
	Corbicula fluminea (Asian clam)
	Ceriodaphnia dubia (water flea)
	Crepidula onyx (onyx slippersnail)
	Dreissena polymorpha (zebra mussel)
	Dreissena bugensis (quagga mussel)
	Hyalella azteca (sideswimmer)
	Daphnia magna (water flea)
	Cerastoderma edule (cockle)
	Chironomus tepperi (midge)
	Hydra attenuate (freshwater polyp)
	Lumbriculus variegatus (aquatic earthworm)

Plants	*Spirodela polyrhiza* (duckweed)
	Lemna minor (duckweed)
	Potamogeton crispus (curly-leaf pondweed)
	Myriophyllum verticillatum (whorl-leaf watermilfoil)
	Hydrilla verticillata (waterthyme)
Bacteria	*Anabaena* sp. (cyanobacteria)
	Spirulina sp. (cyanobacteria)
	Microcystis aeruginosa (cyanobacteria)

5.2 Toxicity of Microplastics Explorer (ToMEx) Database

The ToMEx Database is an online repository of microplastic toxicity data developed as part of a working group led by the Southern California Coastal Water Research Program (SCCWRP). The initial version of ToMEx included data extracted from 162 microplastics aquatic organism ecotoxicity studies (Thornton Hampton et al., 2022b). While ToMEx includes studies focused on the potential human health effects of microplastics, this effort focused on ecological toxicity. In 2023, SCCWRP announced the development of ToMEx 2.0, which sought to update the database with new studies that had been recently conducted. This project worked in parallel with the ToMEx 2.0 update and resulted in the addition of over 150 ecotoxicology studies to the database.

The ToMEx 2.0 Aquatic Organism Database includes over 5,000 data points, all of which are effects tested across a range of biological endpoints. It is important to note that the database includes studies looking at both marine and freshwater organisms and includes species that may not be biologically relevant to the Great Lakes (e.g., a tropical coral reef fish). A summary of the data (automatically generated from ToMEx 2.0) organized by organism, polymer, type, particle shape, and particle size is shown in Figure 5-1. Fish, molluscs, crustaceans, and algae are the most commonly represented taxa in the database. The polymer types most commonly assessed are polyethylene and polystyrene particles. Particle shapes used in these studies are dominated by spheres and fragments. Particle sizes used are mostly 1 µm to 1 mm.

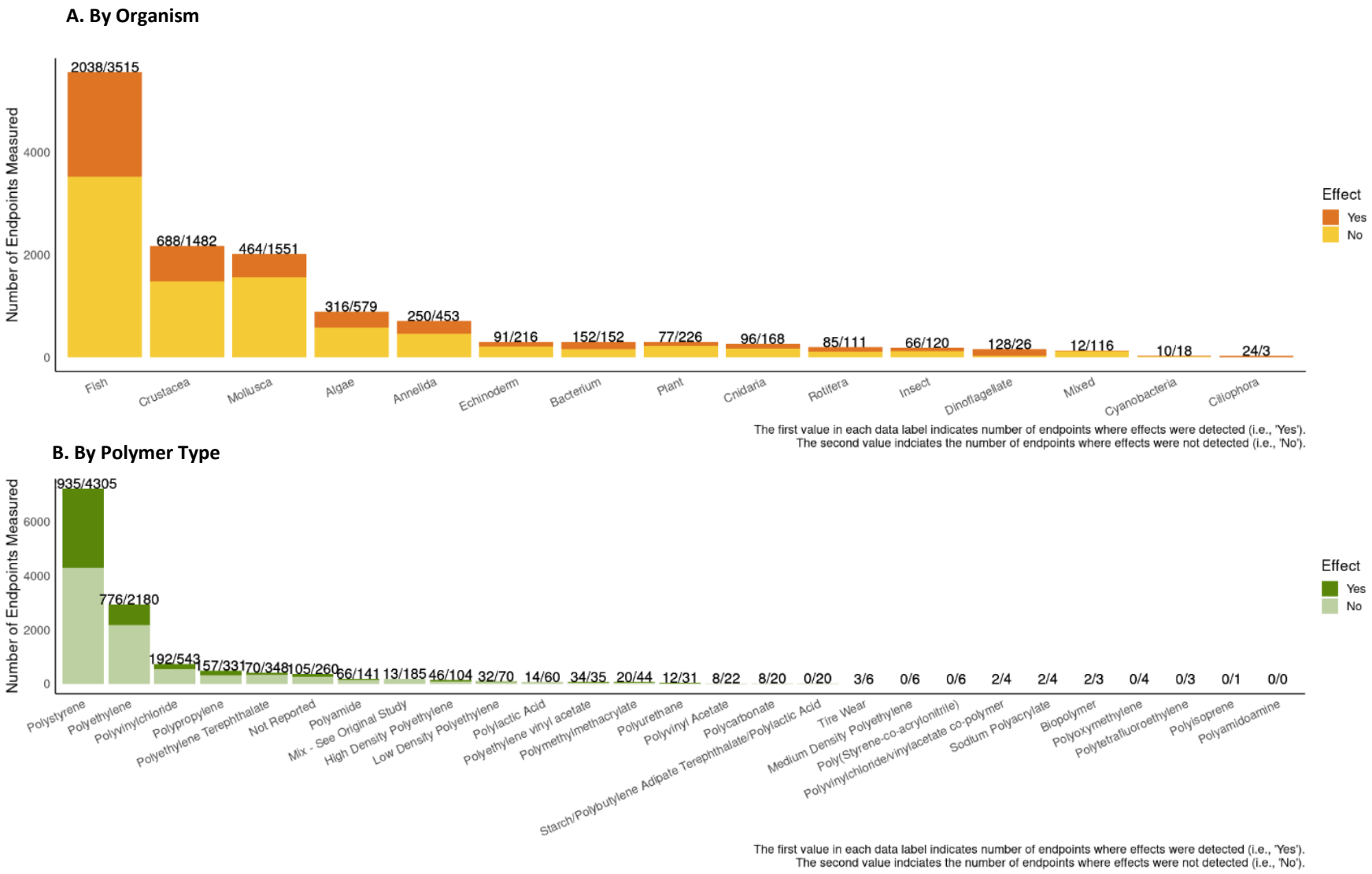
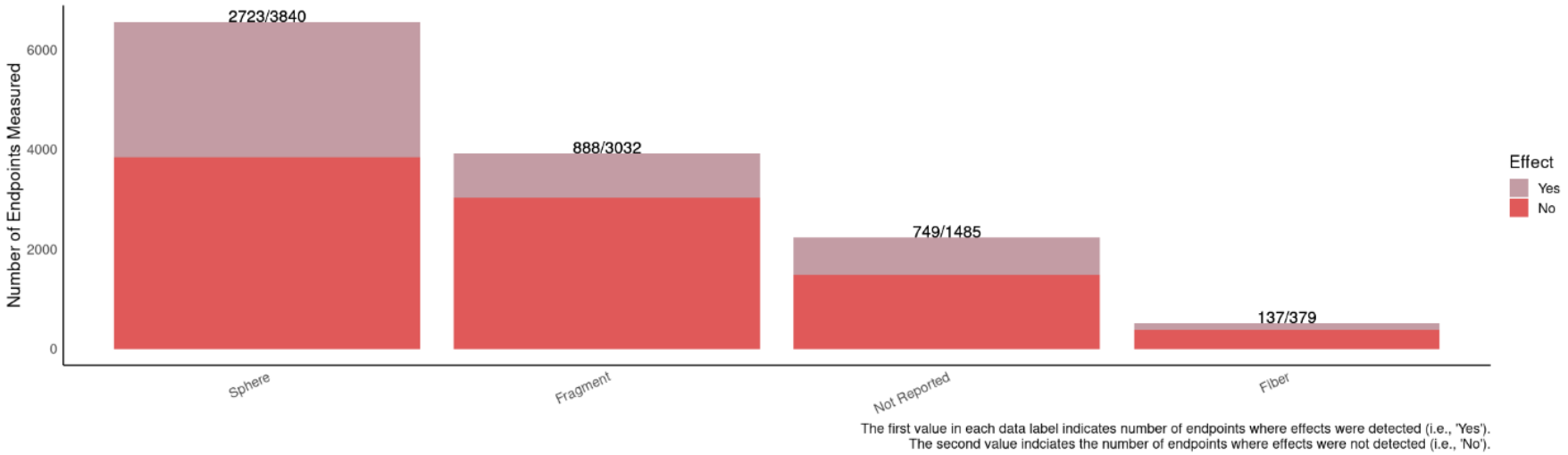


Figure 5-1. Summary of Data in ToMEx 2.0 Aquatic Organism Database Generated from Overview Tool in ToMEx 2.0 R Shiny App

C. By Particle Shape



D. By Particle Size

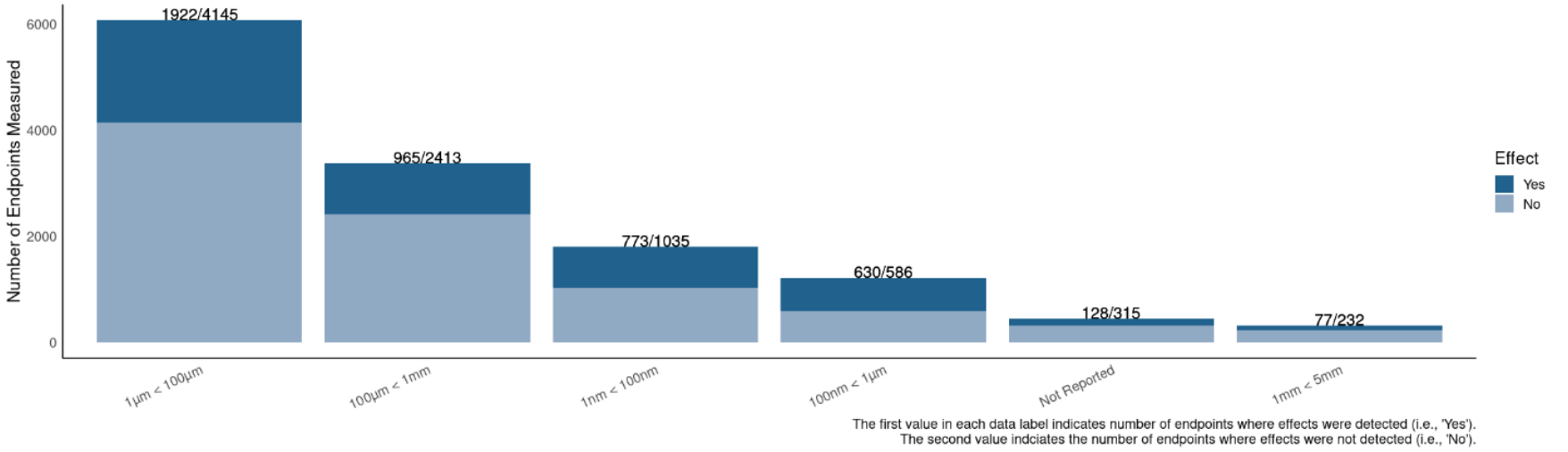


Figure 5-1. Summary of Data in ToMEx 2.0 Aquatic Organism Database Generated from Overview Tool in ToMEx 2.0 R Shiny App (Continued)

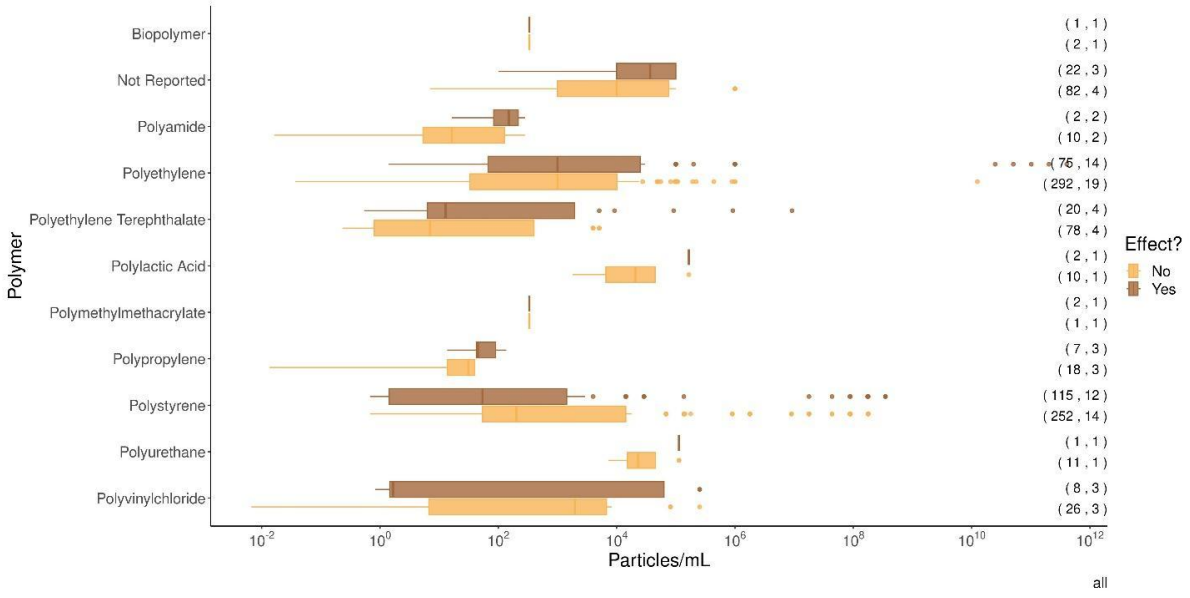
5.3 Factors that Influence the Toxicity of Microplastic Particles

5.3.1 Effects of Polymer Type

Some studies have found that the differences in polymer composition of plastic particles play a role in its toxicity. In addition to the monomers and oligomers that make up the polymer materials themselves, sorbed chemicals, or chemical additives such as plasticizers and solvents have been recognized as potential vectors of toxic chemicals to organisms (Na et al., 2021; Song et al., 2021; ITRC, 2023).

Results from the ToMEx 2.0 database filtered for only freshwater organisms are shown in Figure 5-2 and echo literature review results in that polyethylene and polystyrene were the most studied polymer types. Brown bars reflect the distribution of measurements in which an effect on the organism was measured and orange bars show the distribution where no effect was observed. It is important to note that studies typically were measuring and targeting specific biological endpoints. Just because no effect was measured, doesn't mean that the organism was not impacted in some other way. Additionally, this plot only shows the distribution of all effects demonstrated (i.e., statistically significant from the control) from microplastic exposure and does not show the severity of the individual biological endpoints.

Effects measured for most polymer types ranged from concentrations of 1 – 100,000 particles/mL (or 10^{7}-10^{11} particles/m^{3}) though this range varied based on different polymer types. The majority of effects for polyvinylchloride and polystyrene were observed at concentrations over 1 particles/mL while effects for polyethylene were mostly observed in concentrations over 100 particles/mL. However, there is lack of sufficient evidence to suggest that different polymer types are more toxic than others. There is a need for more studies that explore the effects of different polymers on biological endpoints in the same environment.



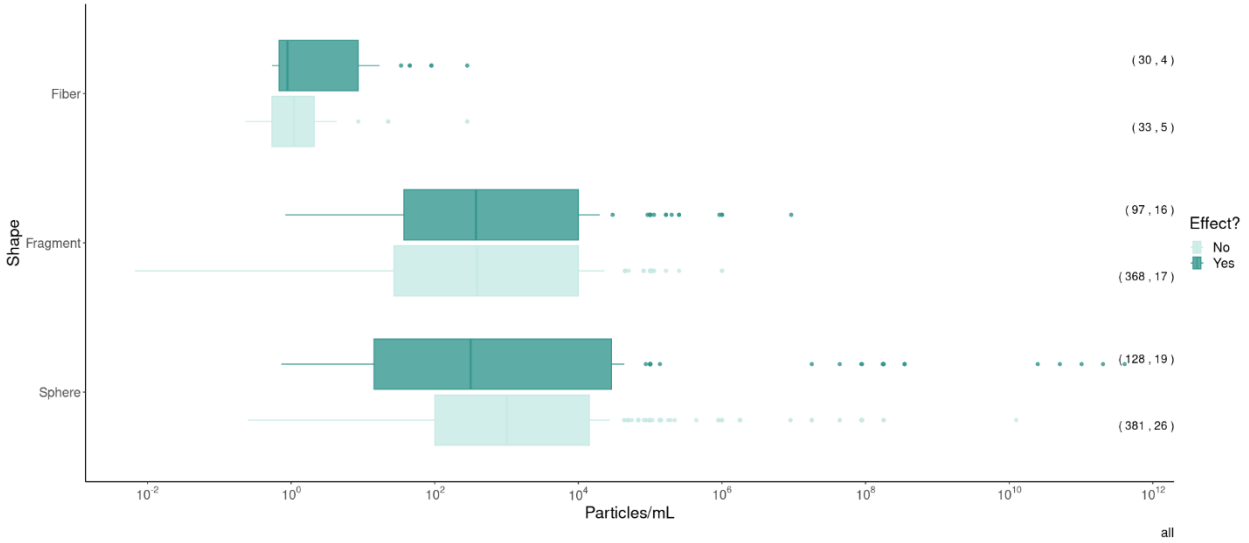
The numbers in the parentheses represent the number of measurements and the number of studies, respectively. Boxes depict the median and interquartile range of measurement data and lines depict the mins and maxes of the data. Dots represent outliers, or points outside 1.5x the interquartile range.

Figure 5-2. Distribution of Effects by Polymer Type

5.3.2 Effects of Particle Shape

Results from previous laboratory effect studies and meta-analyses suggest that complex morphologies such as fibers or fragments may be more harmful than spheres to aquatic organisms (Thornton Hampton et al., 2022a). This may be due to the ability of smoother and more rounded particles to pass through the gut more easily (Lambert et al., 2017). Sharp, rough, or fibrous particles can also cause mechanical injuries to the gut epithelium (Kutralam-Muniasamy et al., 2020, Stienbarger et al., 2021). Dose-response relationships are less clear with microplastics than with other toxic chemicals because their impacts are due to more physical and behavioral interactions with organisms and tissues than strictly chemical interactions.

Results from the ToMEx 2.0 database, relevant to morphology, filtered for only freshwater organisms are shown in Figure 5-3 below. The majority of studies used spheres and fragments over fibers. There is no discernable difference in the concentration ranges where effects were observed between spheres and fragments. Effects from studies that tested fibers were generally lower in concentration at around 1 particle/mL compared to 10 – 10,000 particles/mL for fragments and spheres.



The numbers in the parentheses represent the number of measurements and the number of studies, respectively. Boxes depict the median and interquartile range of measurement data and lines depict the mins and maxes of the data. Dots represent outliers, or points outside 1.5x the interquartile range.

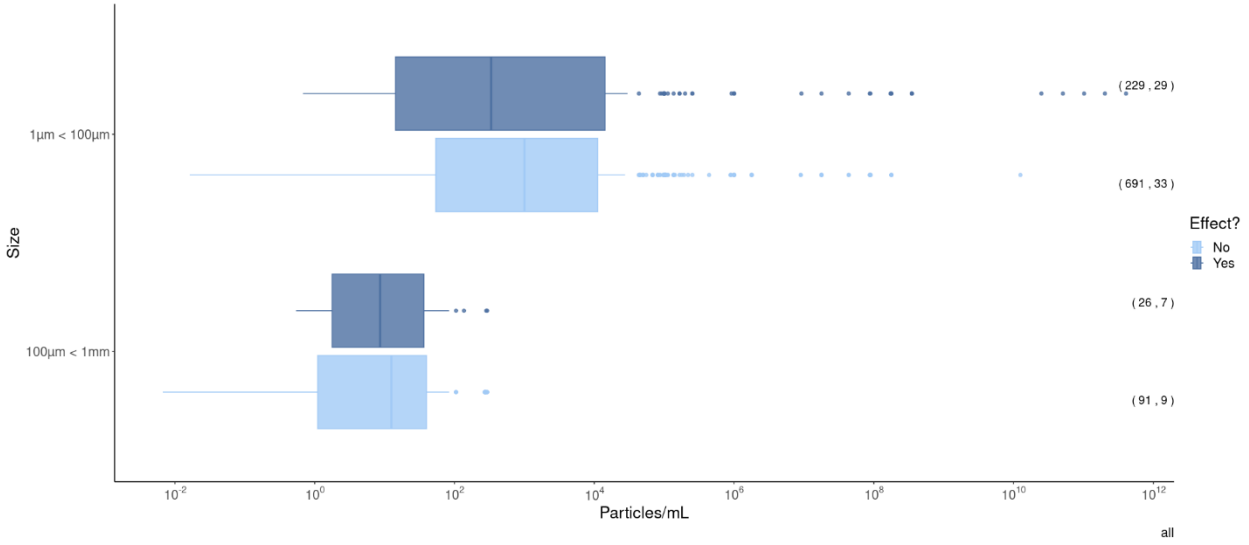
Figure 5-3. Distribution of Effects by Particle Shape

5.3.3 Effects of Particle Size

The size of plastic particles also plays a role in toxicity and smaller particles are of particular concern (Thornton Hampton et al., 2022a). Tissue translocation is primarily dependent on particle size, with smaller microplastics having the potential to translocate within an organism from one tissue to another (e.g., muscle; Mehinto et al., 2022). Smaller particles are also more likely to cause oxidative stress and inflammation once translocated (Thornton Hampton et al., 2022a). Larger ingestible particles have a greater volume (relative to an organism's size), can take up more space in the gut, and may contribute

to food dilution (Thornton Hampton et al., 2022a).

Results from the ToMEx 2.0 database, relevant to size, filtered for only freshwater organisms are shown in Figure 5-4 below. Larger particles (in the size range of 100 µm to 1 mm) appear to produce effects at lower concentrations relative to smaller particles (in the size range of 1 µm to 100 µm). It is important to note that this figure only shows the concentration distribution of all measured effects and does not show the magnitude or severity of effects. The range of concentrations over which effects were observed overlaps significantly with the range over which no effects were observed.



The numbers in the parentheses represent the number of measurements and the number of studies, respectively. Boxes depict the median and interquartile range of measurement data and lines depict the mins and maxes of the data. Dots represent outliers, or points outside 1.5x the interquartile range.

Figure 5-4. Distribution of Effects by Particle Size

5.3.4 Effects of Microplastics Blends and Mixtures

The current literature search did not identify any toxicity studies on mixtures of different types, sizes, and shapes of microplastics with respect to freshwater organisms. Most studies analyzed the impacts of a single size, shape, or type of particle. This does not reflect actual environmental conditions which can include a large variety of different types of particles from a variety of sources. It is unknown how these mixtures of different kinds of particles would impact effects on organisms. These types of studies may be useful in the future, as they would better reflect environmental conditions.

5.4 Other Considerations

The majority of toxicology studies included in the ToMEx 2.0 database, and the literature review, investigated the effects of direct exposure of microplastics to organisms and the influence of the physical and chemical properties of the microplastics on these effects. However, microplastics may also have indirect effects, and may act as vectors for more harmful chemicals and may bioaccumulate via trophic transfer through the food web, as discussed below.

5.4.1 Microplastics as Vectors

Microplastics may potentially act as vectors for transport of more harmful toxic pollutants, including unreacted monomers, additives, or other chemicals that may be released into the environment during the plastic degradation process (Wright and Kelly, 2017). The physical properties of plastic particles such as hydrophobicity and large surface area to volume ratios may also provide a suitable environment for the development of biofilms that may carry harmful pathogens or provide a chemically favorable surface for sorption of harmful pollutants in the environment (Cole et al., 2011; Wang & Wang, 2018). Organic chemicals and heavy metals have been found to readily adsorb to microplastic surfaces (Amelia et al., 2021). Particle properties such as shape, polymer type, presence of additives, and the degree of weathering can influence the particle surface area and its potential as a contaminant vector (Lambert et al., 2017). However, this literature review did not find strong evidence regarding the role of microplastics as a vector for other pollutants. Future research should focus on separating the toxicity of plastic additives and other constituents from the toxicity of sorbed environmental contaminants.

5.4.2 Trophic Transfer and Bioaccumulation

Microplastics are persistent pollutants that may build up in the organism's tissue over time through a process called bioaccumulation. Organisms may then be exposed to pollutants through consumption of other organisms that have been exposed to those pollutants. This phenomenon is known as trophic transfer, and some studies have shown this to be a potentially significant mechanism for exposure to microplastics, with resulting ecotoxicological effects observed as well (Athey et al., 2020, Steinbarger et al., 2021). As a result, there are concerns about bioaccumulation of microplastics in organisms through the food web, or the increase in microplastic concentrations in organisms at higher trophic levels. While some studies have investigated the mechanisms of bioaccumulation and trophic transfer for a variety of chemicals, very few studies have been conducted that have analyzed the kinetics of bioaccumulation of microplastic particles (MacKay and Fraser, 2000). Physical properties of microplastics such as size and shape have been shown to affect the retention of plastic particles in organisms. Entanglement of fibers and translocation of smaller particles into tissue are examples of these bioaccumulation mechanisms (Ašmonaitė and Carney Almroth, 2019).

5.5 Advancements in Microplastics Risk Assessment Frameworks

General risk assessment frameworks, with an eye toward risk management (see: https://www.epa.gov/risk/risk-management), have been challenging to develop for microplastics due to their complexity (different polymer types, shapes, sizes, etc.) and the limited ability to translate simplified lab results (e.g., single particle size, higher than environmental concentrations, single species) to broader environmental occurrences and impacts. Moreover, environmental monitoring captures varying particle sizes, shapes, and polymer types depending on the sampling and analytical methods used. As such, there is a mismatch in exposure and toxicity data. Rescaling this data to a common size range (1 – 5,000 µm) would allow sampling results from different studies to be compared directly. Moreover, lab-based environmental toxicity test data can be realigned to real-world microplastics based on characteristics such as volume (for food dilution) or surface area (for tissue translocation). This challenge has been overcome in work led by Dr. Albert Koelmans' research group (e.g., Koelmans et al., 2020; Kooi et al., 2021), and in work led by a risk assessment working group at SCCWRP (Mehinto et al., 2022; Thornton-Hampton et al., 2022).

As part of the SCCWRP working group, a multi-tiered risk management framework was developed (Mehinto et al., 2022). With the State of California in mind, the group produced a framework with five

management tiers and four risk thresholds. This framework used species sensitivity distributions and rescaling and alignment of results from diverse studies, using the rescaling and alignment approach developed by Koelmans et al. (2020) and Kooi et al. (2021) to develop probabilistic thresholds for ambient water. This management framework contains five management tiers, ranging from low to high regulatory concern, and four risk threshold values relevant to the values at which 5% (HC5) and 10% (HC10) of the species in a community would be impacted. Each management tier contains a recommended management action, which ranges from "no action required" under the "No Concern" tier to "implement pollution control measures" under the "Highest Concern" tier. These risk assessment frameworks use Species Sensitivity Distributions (SSDs) to derive risk threshold values for effects triggered by two of the most well-understood mechanisms of microplastic toxicity to date: ingestion resulting in food dilution (related to particle volume) and tissue translocation resulting in inflammation (related to particle surface area). In addition, the framework requires that exposure and effects data are both screened against QA/QC criteria and rescaled and aligned before being used for assessment.

5.5.1 Microplastic Characteristics

One key consideration in assessing microplastic risk is determining if there are key polymer types, sizes, shapes, colors, or mixtures that are more toxic than others or more likely to be consumed or retained by organisms than others. For example, differential retention could be due to their abundance or attractiveness as false prey items or their ability to be retained in guts or translocate into tissues more effectively. At present, there is not enough evidence to warrant conducting separate risk assessments based on different microplastic types. If additional information becomes available indicating that the risk associated with a particular type of particle merits expansion of the initial risk assessment, action can be taken to update frameworks.

5.5.2 Exposure Considerations

Another set of important considerations is whether there are any considerations for prioritization based on how exposure may vary among, for example, trophic level, life stage, season, the presence of other stressors, or habitat type (nearshore, offshore, surface, deepwater). Laboratory exposure experiments are performed under more consistent conditions than exist in natural settings to isolate variables, so the results of these experiments may not translate directly to natural field conditions. Multiple stressors exist in natural settings, including combinations of microplastics with different characteristics, which can have synergistic or offsetting effects on exposure. For example, lower metabolic rates in winter for organisms may result in decreased feeding and less consumption of potential food items in general, including microplastics. Alternatively, seasonal fish movement from lakes to tributaries may put them in contact with higher concentrations of microplastics at urbanized river mouths as they move from lake habitats to upstream spawning sites. Monitoring of microplastics should take exposure considerations into account such that high exposure locations or periods are not excluded, but that sampling of such hotspots or hot moments does not bias overall values for assessing risk in the lakes more broadly.

5.5.3 Differential Vulnerability

A final risk consideration is whether there are any particularly vulnerable communities, taxa, trophic levels, feeding guilds, life stages, or habitat types where monitoring efforts should be focused. Failure to consider this set of biological variables could result in false negatives when risk is assessed. That is, overall risk for microplastics could be determined to be lower than it really is due to inadequate consideration of exposure and effects on more vulnerable groups of organisms.

6. Policy and Monitoring Programs

Several existing policy frameworks, tools, and monitoring programs could be effectively leveraged or adapted in the Great Lakes region for inclusion of microplastics for assessment and management purposes including binational, federal, and state/provincial programs. Several of these are described briefly here.

6.1 Listing Microplastics as a Chemical of Mutual Concern under Annex 3 and/or as a Toxic Chemicals sub-indicator under Annex 10 of the GLWQA

Microplastics would likely fall under Annex 3 – Contaminants of Mutual Concern (CMC). A CMC nomination package for microplastics has been submitted and will undergo assessment by American and Canadian governments using the Binational Screening Criteria (see binational criteria; https://binational.net/category/chemicals-of-mutual-concern/a3-docs/). If designated as a CMC, then the GLWQA commits the Governments of Canada and the United States to prepare binational strategies for microplastics research, monitoring, surveillance, and risk management via pollution prevention and control. Even if microplastics are not included as a CMC, microplastics could become a Toxic Chemicals sub-indicator under Annex 10. This involves measuring and reporting microplastics concentrations in various media including air, water, sediment, fish, and herring gull eggs, as part of the State of the Great Lakes (SOGL) reports. The report is published every three years by the USEPA and ECCC (ECCC and US EPA, 2022). Chemical additives associated with microplastics may also need to be considered under either the Annex 3 process or the SOGL indicator reporting process. The current CMC list does include some chemicals that are used as chemical additives in plastics, such as flame retardants.

6.2 Tools for Monitoring and Risk Assessment

Tools that identify microplastics concentrations and their characteristics of concern will be needed, along with a standardized way to measure them. Three different SCCWRP working groups have developed frameworks and/or protocols for the laboratory analyses of microplastics, field sampling of microplastics, and their risk assessment and management. These tools can be leveraged in the Great Lakes Region to begin monitoring and assessing risk.

6.3 Existing Monitoring Programs

The binational Lakewide Action and Management Plan (LAMP) committees address management priorities and associated research needs for each of the five lakes on an ongoing basis under GLWQA Annex 2 (see: https://www.epa.gov/greatlakes/lakewide-action-and-management-plans-great-lakes). While they do not implement research or monitoring directly themselves, the LAMP committees and some of their member agencies would be appropriate to engage as part of a coordinated basin-wide microplastic monitoring effort. There are several existing monitoring programs in the Great Lakes that could potentially be expanded to include microplastics sampling and analysis, some of which are described below.

US Environmental Protection Agency Great Lakes National Program Office (GLNPO)
https://www.epa.gov/great-lakes-monitoring

The USEPA GLNPO's Great Lakes Monitoring Program samples water on all five Great Lakes twice a year, with additional sampling and measurement by on-board scientists of benthic organisms, chlorophyll values, and a range of other environmental parameters in the lakes. USEPA monitoring programs include the Great Lakes Fish Monitoring and Surveillance Program, the Integrated Atmospheric Deposition

Network, and the Great Lakes Sediment Surveillance Program. One lake is targeted each year for additional sampling as part of the Cooperative Science and Monitoring Initiative, which may also be an option for integrating microplastic sampling into Great Lakes monitoring on a 5-year rotating basis.

Environment and Climate Change Canada (ECCC)
https://open.canada.ca/data/en/dataset/4497ebe5-f45e-4b13-9e98-e9edd016fc66
Like USEPA, ECCC measures the natural changes and conditions of water quality to determine changes over time, at various locations, of water contaminants and/or threats and support development of science-based guidelines for water, fish, and sediment. ECCC works to identify emerging issues and threats and report and assess science results through performance indicators in an Open Science environment to support an ecosystem approach to environmental and resource management in the Great Lakes. There could be opportunities to incorporate MP sampling into this program.

Ontario Ministry of Environment, Conservation and Parks (OMECP)
https://www.ontario.ca/page/great-lakes-and-watersheds
To monitor progress on Ontario's Great Lakes Strategy, the province is developing performance measures for improvement of wetlands, beaches and coastal areas, along with protection of habitats and species. It is possible that MP sampling could be incorporated into this work. Ontario is also investing funding into new projects to improve the health of the Great Lakes, so there is potential for microplastic monitoring funding in those opportunities, too. Great Lakes and tributary monitoring by OMECP are conducted in coordination with ECCC.

U.S. Geological Survey Great Lakes Tributary Monitoring Program
The U.S. Geological Survey's Great Lakes Tributary Monitoring Program did a microplastics survey in 2016, which could be implemented again in future years to monitor any changes in microplastics in the Great Lakes over time (Baldwin et al., 2016).

Michigan Department of Environment, Great Lakes, and Energy (EGLE) Fish Contaminant Monitoring Program
https://www.michigan.gov/egle/about/organization/water-resources/assessment-michigan-waters/great-lakes-monitoring
EGLE's Fish Contaminant Monitoring Program collects and analyzes fish tissue from four of the Great Lakes for bioaccumulative contaminants of concern, which could provide opportunities to include microplastic sampling as part of that program. Water samples are currently collected each year from an upstream and downstream location in connecting channels. Samples are generally analyzed for nutrients, conventional parameters (temperature, conductivity, suspended solids, pH, dissolved oxygen), total mercury, and trace metals (cadmium, chromium, copper, lead, nickel, zinc). Microplastics could be incorporated into that water sampling program as well.

Other state fish or water quality monitoring programs that could be candidates for microplastic monitoring include:

Minnesota: https://www.ag.state.mn.us/Office/Cases/3M/docs/PTX/PTX3280.pdf
Wisconsin: https://dnr.wisconsin.gov/topic/SurfaceWater/Monitoring.html
Ohio:https://epa.ohio.gov/divisions-and-offices/surface-water/reports-data/statewide-biological-and-water-quality-monitoring-and-assessment
New York: https://www.dec.ny.gov/animals/62194.html

7. Recommendations

This section summarizes recommendations from the literature on microplastics monitoring and risk assessment. It is not meant to be an exhaustive discussion of best practices, but rather serves as a starting point for developing recommendations as part of a monitoring framework for the Great Lakes.

7.1 Monitoring and Reporting of Microplastics

Microplastics should be monitored in various matrices within an ecosystem. These should include ambient water, sediments, rivers, and biota. When monitoring, care should be taken in both sample collection and laboratory analysis. Standardized operating procedures should be developed for application across the Laurentian Great Lakes. Harmonized sample collection protocols should be tailored toward monitoring objectives. For example, managers will need to consider where, when and how often to sample. Strict QA/QC protocols will need to be followed in both the field and lab to achieve robust results. Below are several examples of QA/QC recommendations and best practices from recent studies, several of which are published in special issues in *Applied Spectroscopy* and *Chemosphere* and were products of SCCWRP working groups.

Brander et al. (2020) provide recommendations on QA/QC and best practices for microplastics sampling and analysis in a variety of matrices, including the following:

- Eliminating the use of plastic sampling and laboratory equipment, wherever possible, and using glass or metal in its place. In situations where plastic cannot be avoided (e.g., fishing nets), appropriate procedural blanks are required to quantify and correct for any contribution from the equipment.
- Collecting field and laboratory blanks and following the same protocol to process them as the sample.
- Avoiding contamination in the lab by cleaning surfaces and equipment, avoiding the use of clothing with synthetic fibers, and using laminar flow cabinets, air filters, and appropriate ventilation.
- Considering sampling techniques that reflect study goals (e.g., depth-integrated sampling using pumps vs. surface trawling, collecting an appropriate sample volume).

Miller et al. (2021) provide additional recommendations on sampling, including designing a sampling approach to satisfy study objectives, QA/QC, and reporting, including sampling water, sediment, and biota. One of their recommendations is to report particle counts and to provide sufficient information to allow results to be converted between commonly used units (e.g., from counts per dry mass to counts per wet mass, and vice versa).

Primpke et al. (2020) provide recommendations on harmonizing visual and spectroscopic analysis of microplastics. They provide an assessment of the pros and cons of various methods and discuss when each method is appropriate for use. They discuss costs associated with various methods.

Cui et al. (2022) discuss the need to standardize the microplastics used to prepare matrix spikes. They recommend that microplastic standards contain at least three types of polymer with three different densities (< 1.0 g/cm^3, ~1.0 g/cm^3, and >1.0g/cm^3), three shapes (fibers, fragments, and pellets), and a similar size range as the sample. Akhbarizadeh et al. (2023) suggest that microplastics used for spike recovery samples should possess similar characteristics as the sample being analyzed, including the presence of organic matter to mimic effects of digestion.

Cowger at al. (2020b) have developed reporting guidelines that include data related to study materials and equipment, QA/QC steps, field sampling, sample preparation, microplastics identification (visual and chemical), microplastic categorization, microplastic quantification, and toxicology considerations. These reporting guidelines are intended to improve the reproducibility and comparability of results and are available for use (along with supporting materials) at https://osf.io/jdmex/.

Kooi et al. (2021) provide a framework (discussed in Section 3.5) to harmonize reported concentrations across different size classes. This is needed to harmonize data across studies that sample and report microplastics concentrations across different size classes.

7.2 Studying Toxicity and Assessing Risk

There are several studies in the literature where the effects of microplastics have been tested on an organism. These studies, particularly those with sound quality control and analysis, can be synthesized to inform risk assessment. To do this, existing risk assessment frameworks (e.g., Koelmans et al., 2022) and management frameworks (e.g., Mehinto et al. 2022) can be adapted to the Great Lakes Region. To improve the risk assessments, more studies should be conducted that use multiple exposure concentrations within one test. Such a design will better inform thresholds for toxicity. Future work should also increase the number of studies on organisms that are biologically relevant to the Great Lakes ecosystem. Toxicity tests should investigate which microplastic characteristics (e.g., polymer type, shape, size) influence the sensitivity of test organisms to microplastic exposure (Thornton Hampton et al., 2022a).

7.3 Informing Policy

Microplastics could be managed under Annex 3 as a CMC and/or Annex 10 as a Toxic Chemicals sub-indicator under the GLWQA. These designations would motivate monitoring programs for microplastics, and necessitate a risk assessment framework aligned with existing management frameworks (e.g., SOGL). To inform these actions, we recommend the adoption of standardized laboratory protocols, monitoring protocols and guidelines, as well as a risk assessment framework for the Great Lakes basin. To help facilitate larger-scale positive outcomes, we recommend aligning and/or harmonizing with existing bodies that have similar goals (e.g., SCCWRP).

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Appendix A – Studies Reviewed

A.1 Studies Used to Extract Microplastics Monitoring Data

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