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A baseline for microplastic occurrence in three New England estuaries

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Abstract

Although microplastics (MP) have been documented in estuarine habitats, limited published data exist for New Hampshire and northern Massachusetts hampering meaningful, regional comparison with other geographies. Here we synthesize previously unpublished data from several independent baseline studies spanning three estuarine systems including Great Bay Estuary (GBE), Hampton-Seabrook Estuary (HSE), and Great Marsh Estuary (GME) to compare geographic data for MP to other published regional studies. Data include water column in GBE ($n = 179$ from 7 sites), surface waters and salt marsh sediment cores from HSE ($n = 72$ water samples from 12 sites and $n = 77$ sediment cores from 8 sites), and surface waters from GME ($n = 42$ water samples at 17 sites). Samples were analyzed for MP characteristics initially via either automated confocal microscopy or light microscopy, allowing initial estimation of the number and size distribution of putative MP. Particles from representative samples were analyzed using laser direct infrared spectrometry (LDIR) to determine elemental analysis. MP were found in > 98% of samples collected including surface waters, water column, and marine sediments. Counts ranged from 1 to 144,000 MP particles m^{-3} and mean MP differed significantly among regions, sites within regions, and across years. In the GBE water column, MP tended to peak during June-August in 4 of the 5 years studied. Most MP were roughly circular and ~50 μm in diameter. LDIR confirmed that many types of plastics are in these estuarine waters and also revealed that despite the digestion processes, biogenic materials often remained, predominantly chitin, rubber, wood, and coal. These data allow us to address the realistic levels of risk that estuarine MP pose in NH and



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northern MA estuaries and can be used to populate existing hydrodynamic models that will predict the tributary sources, movements, and fate of MP within these aquatic habitats.

Keywords: Microplastic, estuary, rural runoff, New England

INTRODUCTION

As early as the 1970s, marine scientists noted the occurrence of microplastic particles (MP, particles < 5mm) in nearly every aquatic system studied^[1-7] and because of their persistence, MP is an emerging water pollutant of concern^[8]. Recent studies have confirmed the roles that coastal systems play in attenuating microplastics^[9], including deposition in marine sediments^[10] and salt marsh peat^[11-13]. Subsequently plastics have been found to serve as the substrate for the growth of microbes, both prokaryote and eukaryote^[6,14-17], and as such, scientists have posited that MP are responsible for movement and redistribution of organisms both benign and harmful, within and across ecosystems^[15,16,18-23]. Having codified that this new “Plastisphere”^[17] realm constitutes a new and diverse microbial niche with biogeochemical consequences^[21,24-26], there now exists a series of questions regarding the environmental fate and consequences of plastics and their epiplastic communities^[27]. Numerous studies show that zooplankton, mollusks, fishes, crustaceans, birds, and cetaceans consume MP, and those particles are found in soft tissues, muscle, liver, and gut (10-1,000 s of particles per individual)^[8,28-37]. Recent reports quantify actual levels of MP in seafood^[38-42] with specific reference to MP location (gut, gills, tissues) within organisms. Numerous studies indicate that MP may damage aquatic organisms (including higher trophic levels) by blocking digestive tracts, and by altering feeding and reproductive patterns. To understand how MP are acquired and concentrated in living resources, data are needed for the ambient occurrence of MP in aquatic systems. A recent study of MP in Great Bay Estuary sediments^[10] found levels as high as 100 ± 50 MP·g⁻¹. Results of inputting MP sediment data into a hydrodynamic and particle transport model suggested the highest MP deposition occurs in regions with weaker hydrodynamic flows and lower bed shear stress such as eelgrass meadows and along the fringes of the Bay. Other models of MP settlement in estuaries recently have been published^[43], and together such tools will enable prediction of the tributary sources, movements, and fate of MP within these aquatic habitats. To better inform such hydrodynamic models, data are needed for the ambient quantities, types, densities, sizes, *etc.* of MP currently present within these estuaries. Because MP have not been well studied in estuarine habitats of New England, additional studies were performed in three estuarine systems in this geography, including Great Bay Estuary (GBE) in New Hampshire, Hampton-Seabrook Estuary (HSE) in New Hampshire and Great Marsh Estuary (GME) in Massachusetts to gather baseline knowledge of ambient levels of MP in the water column and intertidal sediments. Provided here is a summary of MP data from samples derived from contemporaneous independent studies conducted by our group. While it is important to note there are differences in approach, analysis, and study goals reported herein, the data constitute an important regional summary and a baseline in support of future work in this region.

METHODS

Sampled regions

The coastal GBE is surrounded by rural, agricultural, and forested areas and is fed by 7 tributaries that are typically low-discharge systems, providing limited freshwater^[44]. In contrast, HSE and GME are barrier marsh systems that were formed behind the multi-state barrier beach system, serving as the primary coastal landform that protects homes, businesses, infrastructure, and estuarine resources of the marsh complex. Although these two systems each have their individual tidesheds, both ecologically and hydrologically, HSE is a component of GME, collectively spanning 10 municipalities from Cape Ann, Massachusetts to Hampton, New Hampshire. These three estuarine systems [Figure 1] contain some of the most outstanding

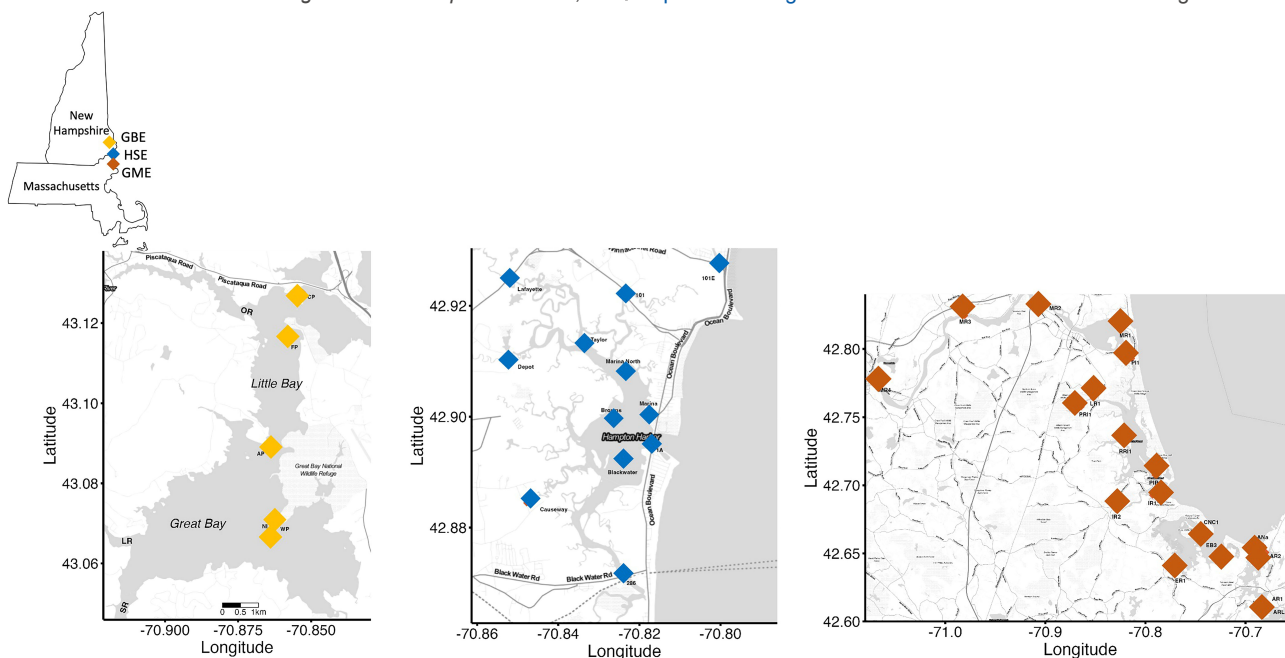


Figure 1. Map of New England (top left) and individual maps of estuarine sites where samples were analyzed for microplastics between 2018–2023. (A) GBE, New Hampshire; (B) HSE, New Hampshire; (C) GME, Massachusetts. GBE: Great Bay Estuary; GME: Great Marsh Estuary; HSE: Hampton-Seabrook Estuary.

ecological and economic resources on the Eastern Seaboard thanks to unparalleled conservation actions and coordination of multiple regional organizations and agencies.

Multiple sites and sampling approaches within three New England regions were accessed to assemble a regional summary of MP in the geography provided herein [Table 1]. Although each of these studies is reported here, it should be noted that these efforts were not necessarily coordinated efforts; thus there are differences in methodology and approach. Nevertheless, all resulting data were analyzed using a uniform approach to create this summary regional report. In GBE and HSE, both in New Hampshire, water samples of different types (bulk and manta trawl) were collected. The GBE water column samples had been collected previously (2018–2022) for a multiyear survey of oyster larva abundance. Analysis of archived samples from a previous study of sediment size characteristics provided data for MP in GBE sediments^[10]. Thus, sediment cores were collected only in HSE (at high and low marsh areas in 2021). There were no prior samples from GME, which is situated on the north shore of Massachusetts. At that site, only water samples were collected between 2021–2023 [Figure 1].

Field collections

Water sampling (GBE, HSE, and GME)

Water column samples from GBE were derived from samples archived from a prior study designed to document presence of oyster larvae. Previously preserved samples were derived from replicate horizontal tows using a 64 μm mesh net held at ~ 0.3 m below the surface, resulting in an average of 38 m^3 of water sampled per tow^[45]. In a separate study area, surface water in HSE was sampled using both manta trawl with 330 μm mesh^[46,47] and bulk sampling^[47] whereas surface water sampling in GME employed only the 330 μm mesh manta trawl. At both HSE and GME, horizontal surface-water tows were performed resulting in an average of 166 m^3 of water sampled per tow. Tows in GBE were made at slack tide and sample collections were timed to be as unaffected as possible by rainfall. Tows in HSE and GME were collected on an outgoing tide and rainfall within 48 hours was noted. The collected material was preserved and stored in the dark

Table 1. Details of samples from three New England estuaries that were investigated for microplastic content

| Sampling location | Sampling sites | Number of samples | Sampling period | Collection method | Flow rate (m ³ s ⁻¹) | Filter size cutoff |
|-------------------------|----------------|-------------------|-------------------|--------------------|---------------------------------------------|--------------------|
| GBE water (Column) | 7 | 179 | Feb-Nov 2018-2022 | Subsurface trawl | 0.5-1.0 | 5 μm - 5 mm |
| HSE water (Surface) | 12 | 72 | Jul-Sep 2021 | Bulk | N/A | 5 μm - 5 mm |
| HSE water (Surface) | 12 | 72 | Jul-Sep 2021 | Manta trawl | 0.2 ± 0.01 (0.003-0.6) | 5 μm - 5 mm |
| HSE intertidal sediment | 9 | 18 | Jul 2021 | 4 cm × 10 cm cores | N/A | 1 μm - 5 mm |
| GME water (Surface) | 17 | 42 | May-Nov 2021-2023 | Manta trawl | 0.5 ± -0.04 (0.2-1.3) | 5 μm - 5 mm |

GBE: Great Bay Estuary; GME: Great Marsh Estuary; HSE: Hampton-Seabrook Estuary.

until processed for MP. Field blanks were integrated with sampling; one blank was collected for every six environmental samples^[47-49]. Manta trawl blanks were taken by rinsing the outside of the clean net with 1 gallon of DI water and transferring liquid within the cod end into a glass jar^[47].

Bulk water sampling (HSE)

Bulk samples were collected in tandem with all manta trawl samples at HSE. At each site, 1 L glass jars triple-rinsed with filtered H₂O were used to collect water samples by submerging the jar just below the surface, allowing it to fill, and capping it underwater. Field blanks were integrated into the sampling plan, with one blank taken for every six samples collected by pouring ~1 L of DI water into a glass jar in the field^[47]. All blanks were taken at established sampling locations to best replicate actual sampling conditions^[47-49]. To assess variability among samples, replicates also were collected in the field^[50,51] immediately following the initial sample collection.

Sediment core sampling

A prior published study^[10] provides MP data for GBE. To investigate the potential for differential deposition by habitat location, HSE sediments were sampled in both high and low marsh habitats. A piston coring device (4 cm diameter and 10 cm long) was used to collect sediments proximal to surface water sampling sites in HSE [Figure 1]. The top 10 cm of the core was used for MP determinations, representing approximately 35-40 years of sediment accretion based on regional accretion rate data^[52]. Cores were held on ice for transportation to the laboratory^[46] and stored at 40 °F until processed.

Laboratory analyses

Efforts completed across the three estuaries were disparate, and various approaches were used to minimize cross contamination in the field and lab. All studies included filtering all reagents (0.2 μm), triple-rinsing glassware with filtered diH₂O, performing manipulations under a fume hood, wearing cotton garb, and covering all containers with aluminum, glass, or paper lids whenever possible. Given that this report synthesizes data from a number of discrete studies that were not uniform in design or goals, the individual methods applied in each case follow.

Water column MP (GBE)

Preserved material from water column tows was thoroughly mixed and 3-5 μm filtered (47 mm PTFE; Sigma-Aldrich, USA) in a glass funnel system. Biofilms and organic matter were removed by stoppering the funnel and adding a 1:1 mixture of KOH (30%) with NaClO (14%, Alfa Aesar) to the filter and holding at 60 °C for 24 h to digest biogenic matter^[53]. The digest was vacuum filtered followed by one rinse with

filtered diH₂O and one subsequent rinse with filtered 50% methanol. After plugging the filter again, 2 mL of Nile Red Working Solution (10 µg·mL⁻¹ in n-hexane^[54-56]) was added and allowed to incubate in the dark at 25 °C for 30 min. Following incubation, the filter was washed with 50% methanol then dried at 60 °C and kept sandwiched between two large glass slides (Home Science Tools, USA) and allowed to dry in darkness until subsequent analysis^[57].

Microplastic particle counts were estimated initially using a fluorescent staining and automated imaging approach. Confocal microscopy was performed by scanning filters (or portions thereof) using excitation: 487 nm and emission: 525 nm for Nile Red and (only for the HSE samples - see below) excitation: 409 and emission: 450 nm for DAPI using a Nikon A1R HD according to standard methods described by^[10,49,54,56,58]. Depending on the number of particles, either whole, half, or one-quarter of each filter was scanned and the images stitched together to create a composite. Automated image analysis of the full scan was accomplished using the NIS-Elements program (Nikon Instruments Inc., USA).

Following confocal analysis, filters from representative samples (samples across years, sites, and with both high and low MP counts) were handled according to the manufacturer's recommendation to remove MP and analyze them by LDIR. The prior filters were placed in a glass beaker with 2 mL of 70% ethanol and stirred for 15 min to release MP particles into solution. The solution containing MP was introduced into Fastwells™ reagent barrier (Grace Bio-labs, Bend, Oregon, USA) attached to a Kevley MirrIR™ slide (Kevley Technology, Chesterfield, Ohio, USA), covered with a petri dish, and allowed to dry completely. Prepared slides were marked with a black marker along the horizontal edge to improve sample flatness detection prior to automated infrared analysis of count, size, shape, and elemental composition using the Agilent 8700 LDIR^[59,60] and the supplied Microplastics Starter spectral library^[61] using Clarify™ software. The library contained a default set of > 50 polymer IR spectra plus spectra of biogenic particles such as coal, chitin, and rubber, and spectra of other particle types that might typically be present in an environmental sample, e.g., silica. Counts of MP in these paired samples were analyzed to determine concurrence of the two methods.

Surface water and bulk sample MP (HSE and GME)

Manta trawl samples were wet sieved through stacked 5 and 0.3 (GME) or 0.1 mm (HSE) stainless-steel mesh sieves using diH₂O. Particles remaining on the 5 mm sieve were discarded. Solids collected on the smallest sieve were transferred to a pre-weighed clean glass beaker, covered with aluminum foil, and placed in a drying oven set to 60 °C until completely dry. Due to high amounts of organic matter, samples with more than 15% settled visual organic matter were sub-sampled prior to sieving. Once dry, organic material was eliminated using 30% hydrogen peroxide (30% H₂O₂, LabChem, USA) at 60 °C^[46]. Following digestion, samples with sand or sediment were density separated using a ZnCl₂ solution (1.5 g·cm⁻³; Fisher Scientific, USA)^[62]. Following digestion and density separation, the remaining sample was vacuum filtered onto 5 µm PTFE filters. GME samples were visually assessed using light microscopy while HSE samples were stained for confocal microscopy. Bulk surface-water samples were immediately filtered onto 5 µm PTFE filters and peroxide digested (*op. cit.*). Subsampling and density separation was not performed for bulk grabs as sand and sediment levels did not require this step.

Samples from HSE were subjected to confocal imaging following the same Nile Red staining process described for GBE, after which MP in surface water and bulk samples were immediately incubated with DAPI (1 mg·mL⁻¹, Sigma-Aldrich, USA)^[63] for another 30 min in the dark to counter-stain. This procedure identified putative MP as those reflecting strongly for Nile Red (mean reflectance < 850 nm) but not reflecting strongly for DAPI (max reflectance < 170 nm). Identified particles were counted, measured, and

reflectance noted. Samples representing sites and MP count ranges were selected and processed for LDIR analysis as described for the GBE water samples. The LDIR spectra were compared to a library containing 8 different polymer spectra. Samples from GME were analyzed using light microscopy only to identify MP by type and color, and were not subject to confocal or LDIR analysis.

Sediment core processing

Sediment cores were separated into 2 cm segments, dried, homogenized^[10], and 1 μm sieved using filtered diH_2O . Filtrates were again dried, digested over two days using wet peroxide at 60 $^\circ\text{C}$ ^[46], and vacuum filtered onto a 5 μm PTFE filter, after which particles were manually identified using light microscopy. Samples containing a large amount of material following digestion were density separated using 25 mL of ZnCl_2 solution (1.5 $\text{g}\cdot\text{cm}^{-3}$; Fisher Scientific, USA)^[62]. Floating solids were collected by vacuum filtration onto a 5 μm PTFE filter. MP in this fraction also were identified by type and color using light microscopy, and were not subject to confocal or LDIR analysis.

Statistical treatments

Normal distribution of data was tested by Shapiro-Wilk normality test. Kruskal-Wallis test was performed to investigate ambient MP concentration in all sample types and the P-values were adjusted by Bonferroni correction. Data derived from confocal and LDIR analyses were compared using paired two-sample *t*-test for means. All tests were performed in R.

RESULTS

Across the 179 GBE water column samples analyzed, the range of numbers of MP detected was 0.08-4,528 MP particles m^{-3} , with a mean of 467 ± 811 (median 115) MP particles m^{-3} . Significant variation was observed across GBE sites ($\chi^2 = 15.2$, 6 df, $P = 0.002$), despite the fact that the estuary is uncharacteristically well-mixed^[44,64] [Figure 2]. In GBE, where the same sampling procedure was used every year, there was significant variation in MP counts across the 5 years ($P < 0.0001$) [Figure 2]. Within years, because of high levels of variation, there were few significant differences in temporal (monthly) or spatial MP counts in GBE [Figure 3]. The majority of particles observed in GBE were very small, $< 100 \mu\text{m}$ and fairly tightly focused around 50 μm . Elemental morphology of MP in the water column was highly variable where some samples contained only a few polymer types and others had complex mixtures of low and high density (ranging the gamut from PP to PVC), copolymers, and biogenic particles (especially cellulose, chitin, coal, and rubber) [Figure 4]. The size distributions (roughly 50 μm) and polymer variation (variable but often with more high density MP) of GBE water column MP were similar to the distributions observed in GBE sediments^[10], an observation that was expected as MP and sediments co-transport^[43]. Of recent particular interest is the occurrence of rubber particles (a.k.a. Tire Wear Particles, TWP) that have been identified in areas near urban runoff^[65]. Rubber was detected in all GBE samples (ranging from 1%-28% of particles). This proportion is similar to that observed (15%-38% of all “MP” particles) in Guanabara Bay Brazil bottom sediments^[65].

Across the 14 manta water samples from HSE, the range of MP counts was 0.45-30 MP particles m^{-3} , with a mean of 11 ± 9 (median 8) MP particles m^{-3} . For the 72 bulk water samples from HSE, the range of numbers of MP detected was 0-143,763 MP particles m^{-3} , with a mean of $27,690 \pm 29,860$ (median 10,927) MP particles m^{-3} . As observed in GBE, the majority of particles observed in HSE waters were very small, around 50 μm [Figure 5]. Elemental morphology of MP in these water samples also was highly variable generally showing representatives of at least eight polymer types. Bulk grabs in HSE contained significantly more MP per volume than those collected with a manta trawl net. Similar findings have been noted across a range of MP studies comparing discrete grabs with manta trawls^[47,66-69]. This outcome can be explained by differences in methodology; 300 mm trawl mesh captures only a subset of MP present, whereas bulk grabs of water

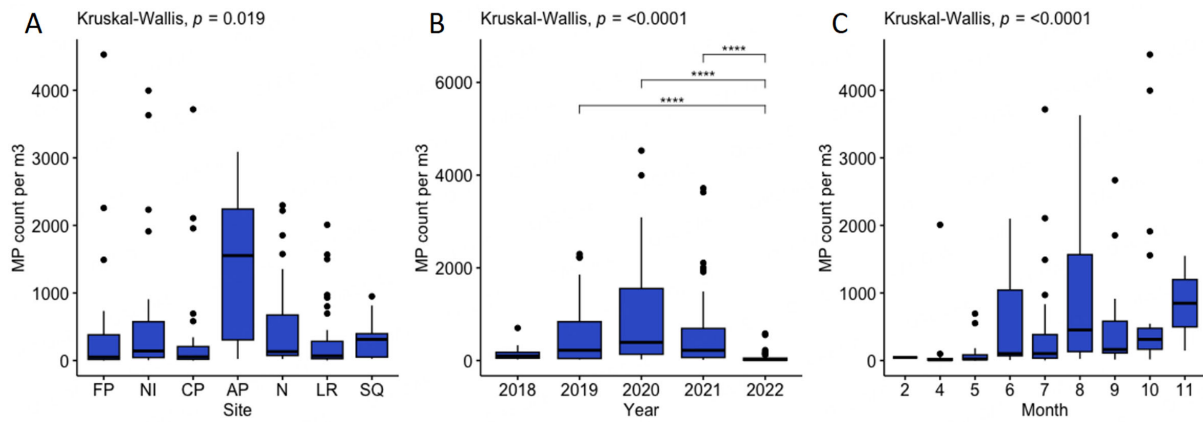


Figure 2. Overall variability in MP counts in water column samples from Great Bay Estuary shown by (A) location sampled; (B) year sampled; and (C) month. Significance level indicated by “***”. MP: Microplastics.

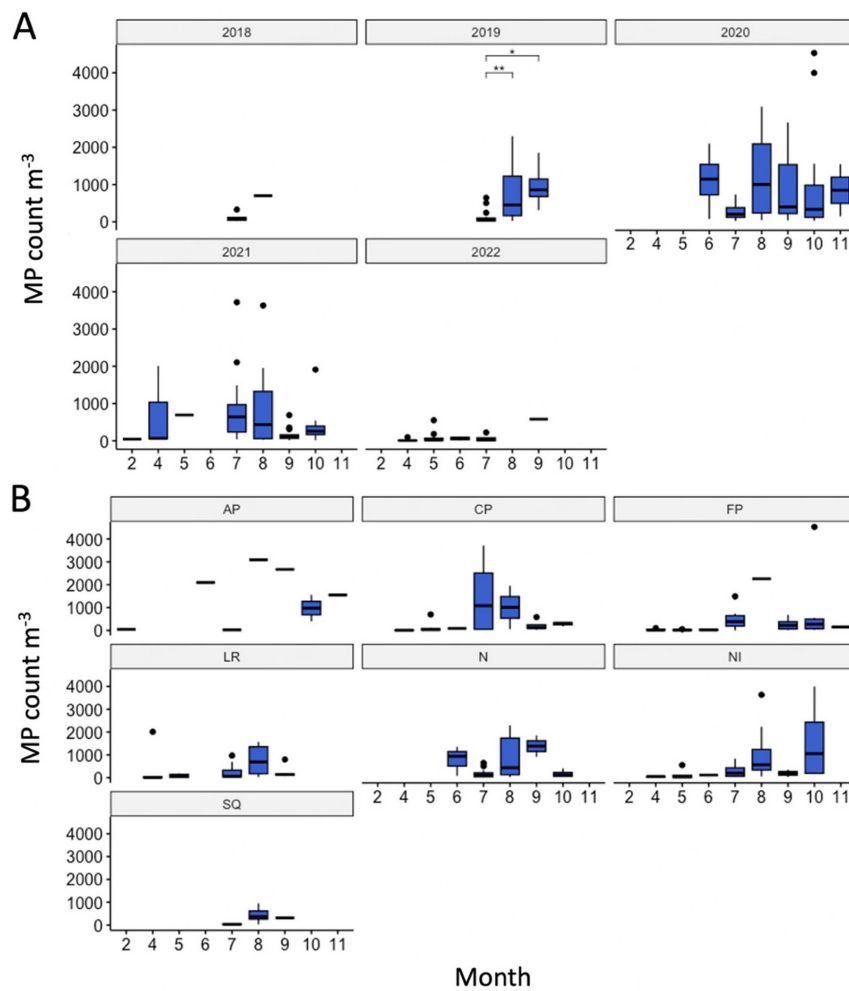


Figure 3. (A) Temporal and (B) spatial variation in MP counts from water column samples collected in GBE. Significance level indicated by “***”. GBE: Great Bay Estuary; MP: microplastics.

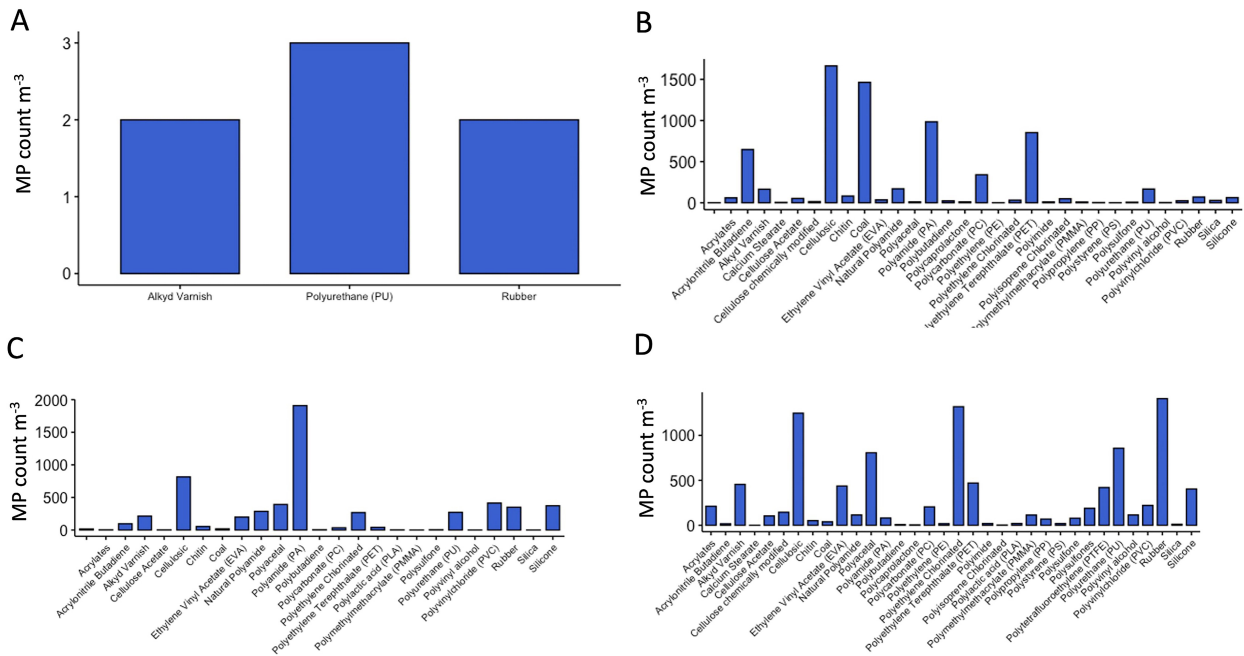


Figure 4. Spectrally determined (LDIR) elemental composition of MP detected in representative water samples from Great Bay Estuary. Panels show data for (A) Nannie Island August 2020; (B) Nannie Island April 2021; (C) Oyster River July 2020; and (D) Adams Point October 2020. LDIR: Laser direct infrared spectrometry; MP: microplastics.

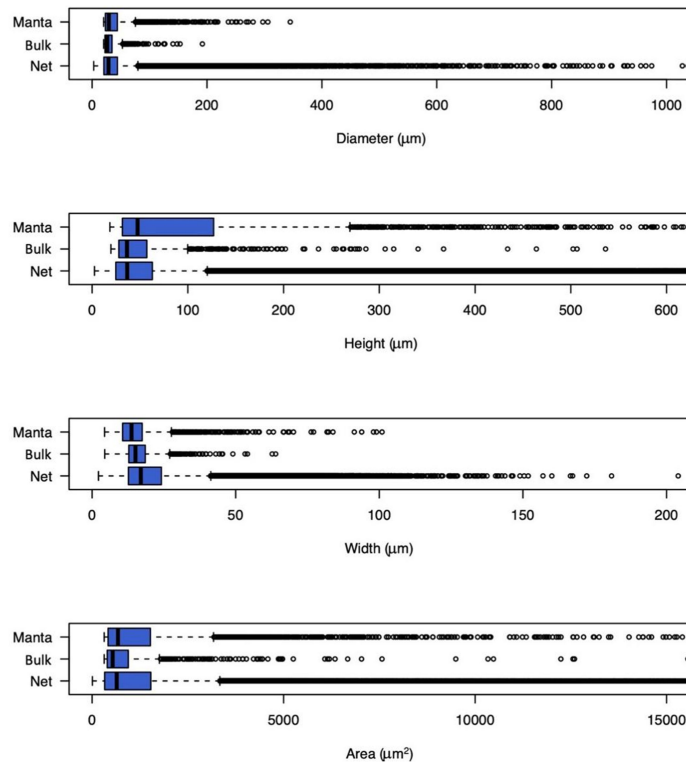


Figure 5. Physical parameters of MP collected in different estuarine water systems of New England using three types of water samplers, all estimated using a confocal microscopic analysis. Manta and bulk sample types were from Hampton-Seabrook Estuary only. Net samples were from Great Bay Estuary. MP: Microplastics.

capture the full size-range of MP^[47,70]. When comparing particle abundance derived from nets only (manta trawl in HSE versus plankton net in GBE), far fewer MP were observed in HSE than in GBE waters, very nearly 1,000× fewer particles. Those differences were most likely due to the hydrology of HSE, which has greater ocean influence and flow in/out. It also is possible that differences in particle density could account for lower MP abundance in the HSE surface sampling approach (top 0-0.2 m of the water column), vs. the slightly deeper water column GBE sampling (~0.3 m below the surface). Minor differences could be due to differences in spectral libraries used for LDIR, with the library used for the HSE samples having a smaller number of polymer types (8 vs. > 50 used for the GBE samples). The GME water samples, being analyzed only via light microscopy, yielded many fewer MP than the samples from other studies that were subjected to confocal fluorescent analysis. Across the 42 GME surface water trawl samples, the range of MP detected was 0.05-5 MP particles m⁻³, with a mean of 1 ± 0.2 (median 0.6) MP particles m⁻³. Significant variation was observed across GME sites ($P = 0.002$).

The numbers of MP vary considerably among each of these New England habitats. This is not surprising given the differences in methodologies, hydrology, and particular geographies^[71]. The MP counts also are well-within the ranges reported for many other estuaries around the globe [Table 2].

As the study of MP progresses and expands, so do the methods utilized. The fortunate application of both confocal and LDIR analysis for many of the samples accessed in this survey provided for comparison of MP counts derived from both methodologies at two sites and using three sampling methods. In every case, LDIR microscopy yielded more particles than was obtained by confocal microscopy. For samples collected with plankton nets, MP were underestimated by a factor 5× when analyzed by confocal. For bulk samples, MP were undercounted by confocal microscopy by a factor of 6×. Samples collected by manta trawl were undercounted by a factor of 10×.

Across the eight salt marsh locations tested for MP sequestration, there were significant differences in the counts of MP·kg⁻¹ ($\chi^2 = 6.8$, $df = 7$, $P = 3 \times 10^{-6}$). The majority of MP (60%-70%) found in HSE sediments were in the top 4 cm [low marsh (19,800 ± 6,900 kg⁻¹), high marsh (13,900 ± 2,800 kg⁻¹)]. More MP particles were found in HSE low marsh sediment samples (16,400 ± 3,900 kg⁻¹ dry weight) than were found in high marsh samples (10,600 ± 1,500 kg⁻¹), which floods less frequently than the low marsh on a lunar tidal cycle. The difference translated to 60%-70% more MP particles, but due to high variation among samples this difference was not significant ($P = 0.45$) [Figure 6]. The absolute numbers of MP detected in HSE marsh sediments were within the typical ranges of many other estuarine MP burdens [Table 3] and were an order of magnitude less than the numbers of MP found in GBE sediments^[10].

DISCUSSION

Use of fluorescent staining is popular for MP studies because it is inexpensive and can be accomplished with less sophisticated microscopes. This study illustrates the ease with which hundreds of samples can be analyzed by that method but by no means is an endorsement of one approach over another. A caveat is that the numbers of particles can be greatly underestimated by staining for reasons that are well-documented, namely variations in handling (time and temperature of incubation with the stain^[84]) and also the lower staining intensity of many polymers. Infrared spectral analysis of representative samples that previously were analyzed with Nile Red confocal fluorescence analysis provided a precise and accurate means to confirm size and shape analyses from the less stringent procedure and provided estimates of the proportional elemental composition of MP from waters and sediments.

Table 2. Microplastic counts in selected estuaries

| Sediment location | MP count (max particles m ⁻³) | Ref. |
|--------------------------------|-------------------------------------------|---------------------------------------|
| Hampton-Seabrook Estuary (USA) | 143,763 | This study |
| Great Bay Estuary (USA) | 4,528 | This study |
| Great Marsh Estuary (USA) | 5 | This study |
| Chesapeake Bay (USA) | 0.2 | Bikker et al. 2020 ^[72] |
| Delaware Bay (USA) | 1 | Cohen et al. 2019 ^[73] |
| Estuary (India) | 752 | Suresh et al. 2020 ^[74] |
| Estuary (China) | 4,137 | Zhao et al. 2014 ^[75] |
| Estuary (Australia) | 172 | Hitchcock et al. 2019 ^[76] |
| Estuary (Mauritius) | 412,000 | Ragoobur et al. 2023 ^[77] |

MP: Microplastics.

Table 3. Selected studies of microplastic counts in some coastal sediments

| Sediment location | MP count (max particles kg ⁻¹) | Ref. |
|--------------------------------|--------------------------------------------|---------------------------------------|
| Hampton-Seabrook Estuary (USA) | 20,000 | This study |
| Great Bay Estuary (USA) | 675,000 | Cheng et al. 2021 ^[10] |
| Narragansett Bay (USA) | 13,000 | Fulfer and Walsh 2023 ^[78] |
| Tampa Bay (USA) | 290 | McEachern et al. 2019 ^[66] |
| Coastal (Belgium) | 213 | Claessens et al. 2011 ^[79] |
| Estuary (China) | 340 | Peng et al. 2017 ^[1] |
| Salt marsh (Portugal) | 23 | Cozzolino et al. 2020 ^[80] |
| Mangrove (Singapore) | 63 | Nor et al. 2014 ^[81] |
| Lagoon (Italy) | 2,175 | Vianello et al. 2013 ^[82] |
| Estuary (Mauritius) | 135 | Ragoobur et al. 2023 ^[77] |
| Beach (Germany) | 62,100 | Liebezeit et al 2012 ^[83] |

MP: Microplastics.

Many of the data recounted here were made possible because samples for other (non-MP) studies were archived long ago and still were available. Because the studies took place over multiple years and by investigators in different laboratories, there was variation in the methodologies used to assess MP. Many water samples were collected using a plankton net whereas some other samples were collected using a manta trawl, also constructed of gradient mesh material. It is important to note that net sampling in estuarine waters is nearly always affected to some degree by the ambient levels of organic material, which can vary rapidly and greatly. Mesh can clog with phytoplankton chains, resuspended sediments, and gelatinous creatures such as ctenophores, cnidaria, and organic debris (i.e., plant material and wrack, especially in estuarine waters). When nets become clogged, particles smaller than the actual mesh size are entrained and maintained in samples. For this reason, grab samples also were utilized in the recent HSE sampling. Furthermore, the increasing accessibility of high-throughput infrared analysis allowed improvement over time in the analysis of plastic type. This allowed the finding that although confocal analysis of Nile Red stained particles is a reasonable method to begin a survey, the LDIR is consistently a more valuable method because it identifies particles that are not effectively stained and the LDIR provides a definitive morphology. In the future, as sampling continues to evolve with apparatus such as the MuMi^[85], more precise data can be obtained. Despite the limitations of net mesh sampling, this set of studies provides a remarkably consistent baseline for the presence and types of MP in these New England estuaries.

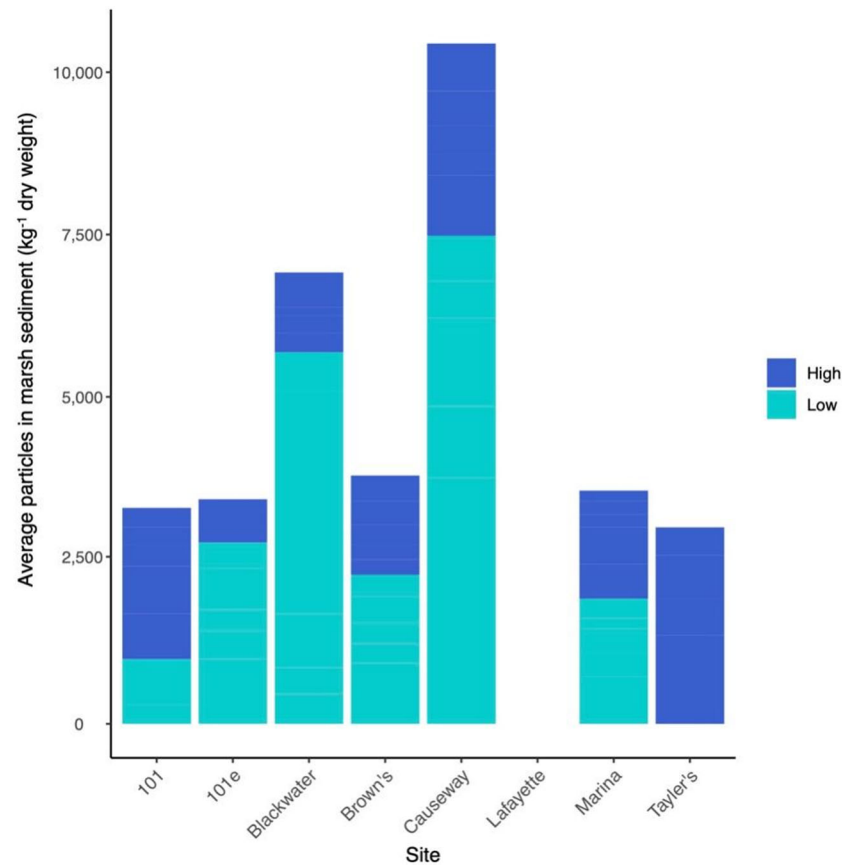


Figure 6. MP found at eight locations in coastal marsh sediments of Hampton-Seabrook Estuary, each subsampled at high and low marsh elevations. MP: Microplastics.

There are a variety of environmental and anthropogenic factors that may influence how MP enter, circulate, and alternatively are sequestered or exit an estuarine system. Variation in watershed size, number of tributaries, water depth, geomorphology, hydrology and tidal energy, and presence intertidal and subtidal coastal habitats can influence MP presence, as can human population densities, amounts and proximity of impervious surface, and the abundance of associated infrastructure. Precipitation events have been tied to increased surface-water MP abundance^[86-88], with higher numbers in nearshore waters compared to offshore waters likely due to land-based runoff^[87]. Tides have also been found to impact the distribution of MP, with greater amounts of plastic litter found on the outgoing tidal phase^[89] and larger MP particles correlated to the spring and flood tidal cycle^[90]. Coastal flooding may have a similar impact as precipitation on transporting land-based MP into estuarine environments^[86-88]. With increasing storm events and associated surge, flooding events may resuspend and force stranded near-shore marine MP deeper into the estuary^[91].

GBE is surrounded by rural, agricultural, and forested areas and is fed by seven tributaries that typically are low-discharge providing limited freshwater^[44]. GBE is unique from other estuaries in that it is well-mixed due to strong currents that introduce oceanic water into the Bay, but flushing rates and residence times are slower/longer than HSE and GME. Although the extent to which the tributaries are the primary source of MPs in GBE is unclear, water resource recovery facilities from more than 50 rural communities in New Hampshire and Maine discharge into GBE. Thus, the MP and other point and non-point source pollutants may be higher in GBE. Despite being well-mixed overall, most of the stations in GBE that were sampled are

a considerable distance from the estuary's outlet at the mouth of the Piscataqua River. Various authors suggest the flushing time for Great Bay is between 2 and 30 days^[64,92,93], depending on the tide and river discharge states involved. The most recent estimate of residence time for Great Bay is between 5 and 20 days^[92] which is a more appropriate metric for particles moving through a system. Note that residence time does not account for whether such particles re-enter the system^[94].

To our knowledge, quantitative estimates of flushing time have not been published for the HSE, nor were any calculated estimates found in grey literature. However, recognizing that 88% of the water in the estuary is exchanged on each tide and 6% of the ebb tide plume returns to the estuary on the next tidal cycle based on estimates from Public Systems of New Hampshire^[95], the flushing time is estimated to be less than one day^[96], which is considerably faster than GBE. Although there are no published residence time estimates for HSE and GME, both estuaries have a more direct connection to coastal waters and have higher flushing rates despite the considerable watershed areas and associated population densities. GBE is the smallest of the three estuarine systems studied here at an estimated area of 200 ha with relatively low population density, yet it had orders of magnitude greater MP per unit volume than the significantly larger HSE (5,300 ha) or GME (~10,000 ha) systems.

The uniquely different geomorphology of GBE as a post-glacial drowned river valley landform may play an important role. GBE lacks the extent of broad fringing salt marsh that buffer the waterways in HSE and GME. Unlike the shallow barrier-beach systems of HSE and GME that are flanked by hundreds of hectares of salt marsh^[97,98], terrestrial runoff may be washing directly into the waters of GBE with little to no natural filtration through high and low marsh habitats along much of its extent. The salt marshes in GBE are limited to relatively small, narrow and disparate patches that do not extend far seaward from upland shores and many are struggling to keep pace with sea level rise^[99]. As a result, GBE is expected to have far less capacity to eliminate MP through sequestration in vegetated wetlands given significantly less acreage of fringing saltmarsh and riparian buffer zones than the barrier beach marsh complexes of HSE and GME. In fact, the high levels of MP in GBE waters have little destiny other than to settle. This corroborates the higher sediment levels of MP in GBE^[10] than in GME (current data).

The HSE sediment MP were dramatically lower than found for GBE sediments^[10]. Although higher MP concentrations have been correlated with population density^[86,100], their presence also can be influenced by point sources such as water resource recovery facilities^[101,102]. These facilities are located on two of the seven tributaries to GBE, just upstream of several sampling areas. It is possible that these potential MP point sources, coupled with slower (and lower) tidal flushing rates and diminished ability to sequester MP in fringing salt marsh sediments, could be collectively contributing to the greater abundance of MP in GBE waters as compared to HSE and GME. Similarly, the greater MP in GBE subtidal sediment (100 ± 50 particles g^{-1})^[10] as compared to HSE intertidal marsh peat cores (2.3 ± 0.3 particles g^{-1}) may be attributed to lower flush rates and longer residence times allowing denser MP to settle. Together, the detection and comparisons of MP in these New England estuarine waters and sediments illustrate that baseline MP will vary based on anthropogenic impacts (e.g., water treatment methods), hydrology (e.g., estuary type and circulation), and habitat type (e.g., subtidal mudflats vs. intertidal salt marshes).

CONCLUSIONS

This work illustrates that the occurrence and fate of MP in the New England estuarine habitats studied vary widely and are generally comparable to the broad ranges of MP identified by other published studies of estuarine waters and sediments. These water and intertidal sediment data complement the only other published study on MP in this geographic area, a study of MP in bottom sediments of GBE^[10]. The data are

essential to populate new hydrological models that are expected to help trace the sources of MP and other pollutants in GBE. The results provided here are of immediate importance and value to resource users, harvesters, aquaculturists, and managers to inform and evaluate contaminant risks, to choose low MP sites for their activities, and provide information on the time of year with the highest or lowest MP concentrations.

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Authors' contributions

Conceptualization: Brown B, Moore G

Methodology: Brown B, Mogensen H, Sims-Harper T, Gibson J, Jarrett G, Wardinski C

Data analysis: Lee BY, Brown B, Mogensen H, Gibson J

Investigation: Sims-Harper T, Brown B, Jarrett G, Wardinski C

Original manuscript preparation: Brown B, Moore G, Gibson J

Authors have read and agreed to the submitted version of the manuscript.

Availability of data and materials

Not applicable.

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Conflicts of interest

All authors declared that there are no conflicts of interest.

Ethical approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

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