

2023

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### APA Citation

Marcus, I. M., Barriquand, T. B., Thompson, R. M., Hosselkus, B. C., Hutson, C. T., Jacobs, M., McNeil, C., Newton, L., Olivarez, S. M., & Abell, J. (2023). Spatial and Temporal Variations of Microplastics within Humboldt Bay, California. *CSU Journal of Sustainability and Climate Change*, 3(1). DOI: <https://doi.org/10.55671/2771-5582.1023>

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# Spatial and Temporal Variations of Microplastics within Humboldt Bay, California

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# Spatial and Temporal Variations of Microplastics Within Humboldt Bay, CA

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This study aimed to quantify microplastic (MP) concentration and analyze the spatial and temporal variabilities of the concentrations during the tidal cycle in Humboldt Bay, California. To get an approximation of MP concentration, both water and sediment samples were taken at five different stations, twice during one tidal cycle. Sampling was conducted during two different cruises, on the 19th and 21st of September 2020. The samples were processed in the lab using a density separation procedure and filtration. MP concentrations in the different samples were determined using an average optical microscopy count. Comparison of the water column MP concentrations during ebb and flood tides shows higher concentrations during flood tide,  $49.0 \text{ particles/L} \pm 32.37$  (flood) vs  $34.4 \text{ particles/L} \pm 16.32$  (ebb), indicating that MPs are brought into Humboldt Bay from the ocean. The comparison of the MP concentrations during lower energy and higher energy conditions indicates that concentrations in the water column were elevated when there was greater tidal kinetic energy, approximated by the covariance of the measured velocity in North Bay Channel. This result was assumed to be caused by the strong tidal currents stirring up both sediments and the settled MPs into the water column. Due to lower tidal kinetic energy on the sediment sampling cruise day, we could not confirm that assumption. Water samples indicated that MPs are heterogeneously distributed in the bay, with higher concentrations found near the Entrance Channel and lower concentrations found further north in the bay. Sediment samples also indicate a heterogeneous distribution of MPs in the bay, with the lowest concentrations near the Entrance Channel, 15 particles/kg, where high tidal currents inhibit settling of particles.

## Introduction

### Plastics and Microplastics

In the past 65 years plastic pollution has risen dramatically. During that period, 6.3 billion metric tons of plastic have been produced (Dikareva & Simon, 2019). In 2016, the annual global production of plastic products was about 322 million tons (Li et al., 2018). About 60% of all plastic produced has accumulated in the environment (Dikareva & Simon, 2019). The multiple additives used to lengthen the life of plastics slow the degradation of plastic waste in the environment (Chamas et al., 2020). The duration of plastics make them an even greater concern in the marine environment. (Gall & Thompson, 2015).

Plastics are made from polymer-based materials and are processed with a range of chemical additives to make them usable, including inorganic fillers, pigments, plasticizers, and antioxidants (Lambert & Wagner, 2017). Plastic particles that are  $< 5 \text{ mm}$  are considered microplastics (MPs) (NOAA Marine Debris Program, 2015). MPs are divided into two categories, primary and secondary MPs. Primary MPs are used as resin pellets to produce larger items or used directly in cosmetic products like facial scrubs and toothpastes. Secondary MPs are formed from the disintegration of larger plastic debris (Lambert & Wagner, 2017). These secondary MPs could originate from fishing nets, industrial resin pellets, discarded plastic debris, and emissions from wastewater treatment plants (Li et al., 2018) (Lambert & Wagner, 2017).

MPs have been found throughout the ocean in the water column and sediments in varying concentrations (Dikareva & Simon, 2019).

The variation in plastic composition leads to a heterogeneous spatial distribution of MPs in the marine environment (Wagner et al., 2014). MPs are not evenly distributed horizontally or vertically in the water column, and their abundance decreases at greater distances from their source (Mendoza & Balcer, 2019). Plastic debris can also be transported by winds and direct runoff after rain events, where it eventually reaches aquatic ecosystems and accumulates (Dris et al., 2015). Environments are likely exposed to different mixtures of micro- and nano- sized particles because of the composition of the plastic material.

MPs and other marine debris can have a detrimental impact on the marine environment. Bacteria can migrate on plastics, impacting the microbiome of areas not previously affected (McCormick et al., 2014). In addition, MPs can bioaccumulate in an organism's systems and cause digestive issues, tumors, or both (Li et al., 2018). Plastics have also been found embedded in rocks on shorelines, which could impact grazers and marine invertebrates (De-la-Torre et al., 2020). In order to curb the major effects of MPs on marine ecosystems, we need to better sample and quantify the distribution of microplastics in the marine environment.

### Microplastics in Sediments

Van Cauwenberghe et al. (2015) estimated that millions of tons of plastic waste end up in the marine environment. Marine sediments are hypothesized to be major sinks of MPs. Plastics with a density greater than the average density of seawater (i.e.  $1.027\text{g/cm}^3$ ) will sink and accumulate in the sediment. Low density plastics will initially float at the sea surface or move down in the water column based on their density. Biofouling—the general accumulation of organisms on an object—causes even buoyant, lighter plastics to become denser and sink to the seafloor (Van Cauwenberghe et al., 2015).

MPs have been detected on the shorelines of all continents and the seafloor across the globe. Due to the large spatial variability of MP distribution in sediments, sediment samples must be collected from different locations in a region in order to correctly quantify the MP concentration in that region (Nuelle et al., 2014). The typical concentrations of MPs in sediments range from 1 to 100 items  $\text{kg}^{-1}$ . Wagner et al. (2014) found a maximum of 400 items  $\text{kg}^{-1}$  in coastal harbor sediments.

### Transport of Microplastics

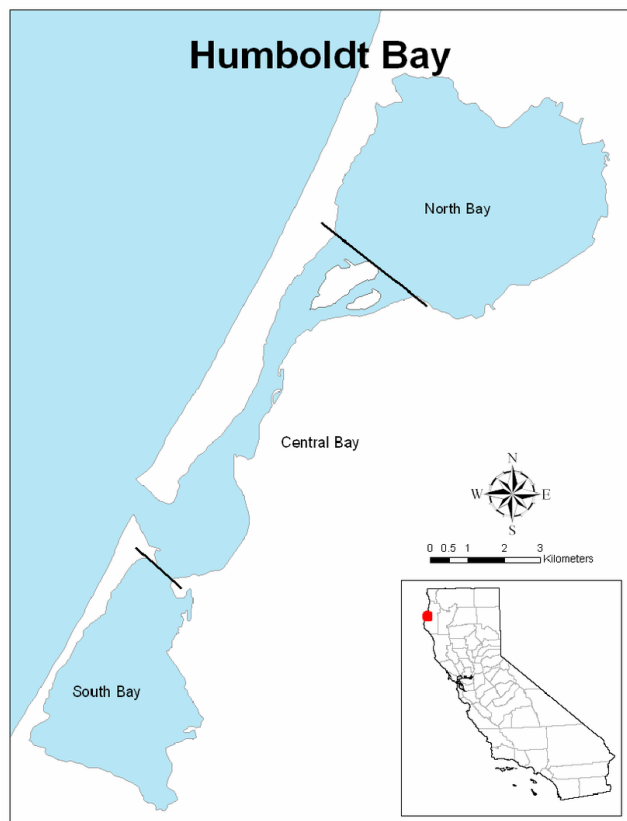
Estuarine river runoff is the primary source of MPs into the marine environment. The most abundant MP deposition into marine environments occurs during storm events directly at river and tributary mouths. After storm events in California, a six-fold increase in MP concentration is seen in surface waters of the ocean, and plastics are deposited farther from their original source (Lattin et al., 2004). The hydrodynamics of estuaries and bays affect MPs in a similar way to how sediments move in these environments when large volumes of water flow in or out due to tides or storm events (Zhang, 2017). In estuaries, MPs circulate and are distributed to the ocean through tidal mixing and currents. Once MPs have reached the open ocean, depending on their densities, they will either sink or float. The denser plastics will sink near their source, while the floating MPs will be transported by surface currents (Zhang, 2017). The floating MPs might experience biofouling during this transport, causing them to sink or become neutrally buoyant. During the sinking process, MPs will be circulated by deeper ocean currents. When the MPs become denser than the water column, they sink and settle in the sediment (Zhang, 2017). These particles can settle much farther from their source, depending on the duration of their suspension in the water column. Some MPs will never sink; they may remain floating or suspended in the water column. Ingestion by zooplankton, benthic organisms, and large marine animals is an additional source of sink for MPs in the ocean (Zhang, 2017). MP concentration in marine animals is directly correlated to the concentration of MPs in the seawater (Wright et al. 2013).

The goal of this study was to analyze the distribution of microplastics in Humboldt Bay, CA. Humboldt Bay, the second-largest estuary in California, is separated into three main sections: North Bay (NB), South Bay (SB) and Entrance Bay (EB) (figure 1).

There is a high degree of erosion and sediment transportation within EB, North Bay Channel, and Southport Channel. While the sediments deposited in the channels are predominantly sand, NB and SB are almost entirely composed of silty tidal mudflats (Costa, 1982). These tidal mudflats are extremely nutrient-rich and support an enormous variety of life, including major eelgrass habitats. Eelgrass beds increase deposition of sediment as the large leaves disrupt the flow of water, capturing sediments—and potentially plastic—within the water column (Schlosser and Eicher, 2012). The abundance of erosion causes sediment deposition at the mouth of the Entrance Channel, reducing the flux of water into and

**Figure 1.**

A map of Humboldt Bay, Eureka, CA. Three main sections of the bay are shown: North Bay, Central Bay, and South Bay (Pinnix et al., 2005). Inset depicts the state of California, with the red dot indicating the location of Humboldt Bay. Humboldt Bay's watershed spans an area of 557.6 km<sup>2</sup> (Barnhart et al., 1992). The bay is approximately 19 km long and 0.8 km–6.9 km wide, with a total surface area of 64.8 km<sup>2</sup> at high tide and 20.7 km<sup>2</sup> during low tide (Evenson, 1959). The average depth of the bay is 3.4 m, and the maximum depth is 12 m. Salt marshes make up 4% of NB and 1% of SB (Barnhart et al., 1992).



out of the bay. To maintain water flow, Humboldt Bay is dredged two miles south into SB and four miles up into EB, almost annually (Humboldt Bay Harbor District). Sediment distribution in Humboldt Bay suggests that dredging has increased the average grain size of sediment found within the bay (Stevens, 2002). Areas of Eureka and Samoa Channels become more sand dominated after dredging occurs (Stevens, 2002).

### Freshwater Inputs

Plastic is a major constituent of riverine pollution (Lambert & Wagner, 2017). In their studies of MP transportation in freshwater systems, Luo et al. (2019) found that rivers provide MPs from land-based sources to estuaries and the ocean, and the researchers deemed the concentration

of MP debris to be more detrimental to freshwater bodies than estuarine. Dikareva & Simon (2019) found that the total MP concentration in small streams varied between 17–303 items per cubic meter in the water column and 9–80 items per one kg of dry sediments, whereas Li et al (2018) found the average values of MPs in freshwater systems ranged from an undetectable concentration to almost a million pieces per cubic meter. The most abundant types of plastic found in the water column were fragments and fibers, making up 34% of all particles on average (Dikareva & Simon, 2019).

The freshwater sources of sediments are mainly the small creeks and rivers that empty into Humboldt Bay. These streams run through highly populated areas with around 65,000 residents, likely picking up contaminated runoff from streets, homes, and businesses. The major tributaries contributing to the bay are: Salmon Creek, which enters into SB; Elk River, entering into EB; and Freshwater Creek and Jacoby Creek, entering into NB (Barnhart et al., 1992). 62,532 metric tons/yr of sediments are supplied to the bay from its tributaries, the majority of which enter the bay after large winter storms, leading to heightened turbidity levels between 30–200 NTU (Houle, 2015). The biggest contributor is the Elk River, which transports sediments consisting mostly of silts intermixed with sands and clays eroded from the Wildcat Group of the Miocene-Pliocene age at a rate of 1200 Mg km<sup>-2</sup> y<sup>-1</sup> (Macdonald et al., 2016). This erosion rate is similar to those of other North Coast watersheds (Andrews and Antweiler, 2012).

### Tides and Currents of Humboldt Bay

Humboldt Bay is a well-mixed marine estuary that is tidally driven by mixed semidiurnal tides, with a mean tide height ranging between 1.5 m and 2.1 m at the channel mouth (Anderson, 2015., Crawford and Claasen, 2004). Currents entering the bay from the northwest have the greatest impact on tidal fluxes, while currents coming in from the southwest have the highest contribution of wave energy (Crawford and Claasen, 2004). Overall current flow is generally in the northeast direction, resulting in the majority of water coming in from the channel mouth to be forced into NB (Gutierrez et al., 2005). Approximately 50% of the tidal prism travels into NB, with 30% of the tidal prism flowing into SB (Costa and Glatzel, 2002).

Maximum current velocities tested within the navigation channel can reach higher than 4.1 m/s, and wave heights can be as high as 7 m. In the Entrance Channel (EC), the average velocity of the currents during an ebbing tide is 1.9

m/s with an average of 2.1 m/s during a flooding tide (Costa and Glatzel, 2002). The velocities slow down as the depth decreases through the bay. The wave energy entering through EB tends to be the strongest, especially during flood tides, where it is deflected off of the south jetty towards NB Channel (Gutierrez et al., 2005). In addition to the high wave energies coming into the bay, the placement of the jetties and the positioning of the Humboldt Bay Bar both establish a huge means of sediment transport, erosion, and mixing within EC and into NB Channel (Costa and Glatzel, 2002; Gutierrez et al., 2005). The overall circulation in the bay varies daily and seasonally (Costa and Glatzel, 2002).

### Human Impacts

Humboldt Bay is home to many industries, including local marine cargo, commercial fishing, mariculture, marine research, and recreational boating. There are two small commercial and recreational boat harbors in NB, located at Woodley Island Marina and Eureka Public Marina. The area surrounding the bay contains several sites of industrial operations: lumber mills, bulk oil storage, wrecking yards, and railroad yards which can contaminate local water sources with heavy metals, petroleum, and pentachlorophenol (PCPs). The Arcata Marsh and Wildlife Sanctuary, neighboring the top of NB, acts as a natural wastewater treatment plant for the city of Arcata and discharges the treated water into Humboldt Bay (Wastewater Treatment). Comparatively, Li et al. (2018) found that industrial wastewater treatment plants can release around 8 billion pieces of plastics per day.

Before 2013, there were sixty million single-use plastic bags used in Humboldt County and seven million or more were annually used within Arcata city limits (Ordinance No. 1434 of the City of Arcata Sec. 5476, 2013). The amount of local plastic deposited in Humboldt Bay and the ocean is still unknown. Another study found that an average of 415 pounds (188 kg) of plastics, including bottles, caps, and food packaging, washed up on North Jetty Beach annually (Plastic Pollution at Four Coastal Cali. Hotspots, 2020). Whether that plastic came from Humboldt Bay or was brought into the area from the ocean has not been determined.

This study aimed to quantify the MPs in Humboldt Bay and analyze concentration variability during a tidal cycle. To get an approximation of MP concentration, sampling was conducted of the water column and the sediments and compared during fluctuations in the tidal cycle. We also aimed to determine the directionality of plastic contribution between Humboldt Bay and the ocean, with a hypothesis that Humboldt Bay would be a net contributor of MPs to the Pacific Ocean. This prediction

was founded on the presupposition that ebb and flood tides would result in sediment and MPs becoming suspended in Humboldt Bay, thereby increasing the concentration of MPs in the water column exiting the bay (G.P. Allen et al., 1980). We expected that sediment and MPs would resettle during slack tides, increasing the MP concentration in the sediment and decreasing MP concentration in the water column. We anticipated that surface sediment MP concentrations would be greater in the extremities of the bay, where there is less tidal influence, compared to the sediment MP concentration in the tidally driven mouth of the bay.

## Methods

### Sampling Sites

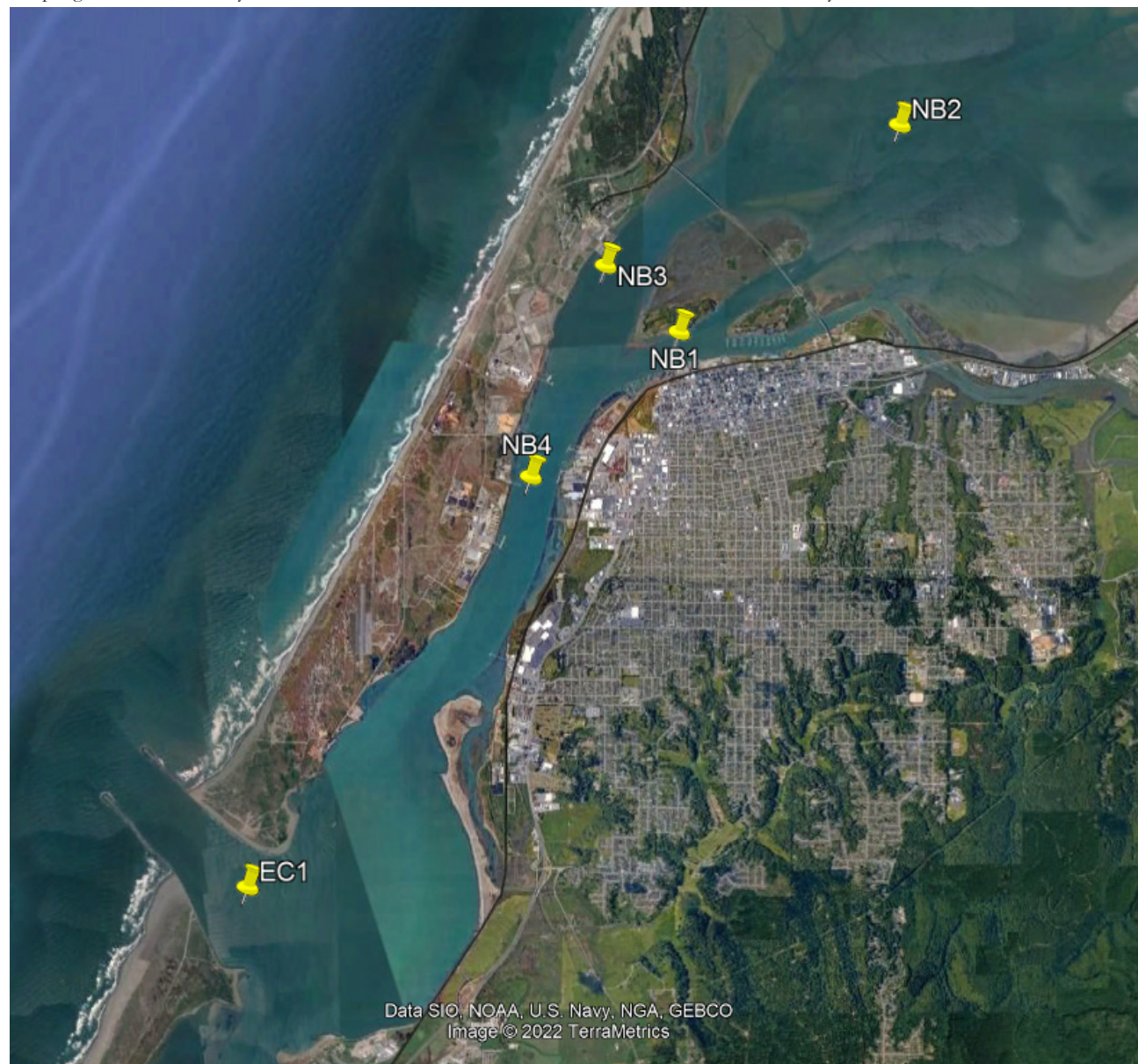
Samples were collected during two separate cruises aboard the R/V Coral Sea on September 19, 2020 and the Cal Poly Humboldt pontoon boat on September 21, 2020. Due to Covid-19 social distancing regulations aboard the vessels, two cruises were necessary. Humboldt Bay does not experience identical tides daily, so to minimize differences in sampling conditions, two days with similar tidal cycles were chosen for the separate cruises. Due to the time restraints on cruises at the time of sampling and the time necessary to complete the sampling, only flood/ebb tides were sampled for water data and only flood/slack tides were sampled for sediment data rather than sampling at all points of the tidal cycle. Samples were taken at five stations throughout Humboldt Bay (figure 2). These sites were chosen to sample distinct portions of Humboldt Bay that may experience pollution from their surroundings. For example, NB4 was near the convergence of the three main channels of Humboldt Bay, thus experiencing a variety of current direction and velocity. Ideally, stations in South Bay would have also been sampled, but due to the project's constraints around sampling protocol, this was not possible.

### Avoiding Contamination

Contamination has been a prominent issue for past studies on MPs in Humboldt Bay (Carlson et al., 2018). To minimize possible contamination from our own clothing, attire guidelines were put into place and followed by all researchers. During all sample collecting and processing, researchers wore bright orange cotton jumpsuits, and any clothing made from polyester or other synthetic textiles was prohibited. The sampling and storage equipment was cleaned thoroughly prior to and following use, and equipment was

**Figure 2.**

Station map of cruise stops on the R/V Coral Sea and Cal Poly Humboldt pontoon boat in Humboldt Bay. The yellow pins indicate the five sampling sites: EC1, the only site in the Entrance Channel; NB1-NB4, various sites within North Bay.



stored in sealed bins to prevent ambient MP contamination during storage and transport of samples. Blanks were taken across the analytical procedures and on all equipment. The processing of blanks was essential in minimizing the effect of our inevitable contamination to our samples and procedural equipment.

#### **DDI Water Blank Procedure**

The DDI water collected from the Telonicher Marine Lab (TML) was used as an absolute blank and as a density separation procedural blank. 0.5L of DDI was filtered directly onto 20 $\mu$ m glass fiber filters. Filters were dried and

weighed, and MP particle numbers were quantified using microscopy. This blank was used to quantify background MP concentration in DDI water. A separate set of 0.5L DDI aliquots was then run through the same density separation procedure as the actual water samples to quantify any possible MP contamination due to the procedure.

#### **Freshwater Reserves aboard R/V Coral Sea**

Freshwater from the hold of the R/V Coral Sea was collected in 2 L glass jars topped with aluminum foil and sealed with aluminum lids. These samples were taken back to the laboratory for blank analysis as described above for DDI.

The 0.5L aliquots were filtered directly onto combusted, 20 $\mu$ m glass fiber filters and were dried and weighed, and MP particle numbers were quantified using microscopy. This blank was used to quantify the background plastics that were introduced to the ship's freshwater from its collection pumps and storage facilities.

### Hydrocast Sample Blanks

Freshwater from the R/V Coral Sea was poured directly into semi-open Niskin bottles while the rosette was on the stern of the vessel. This mimicked the environmental conditions of sampling as accurately as possible. This sample was then run through the hydrocast sample collection procedure, followed by the density separation procedure. Filters were dried and weighed, and MP particle numbers were then quantified using microscopy. This blank quantified MPs introduced by exposure of the sample to the PVC Niskin bottles and by the handling and manipulation of the sample during collection.

### Sediment Blank Procedure

Blank samples were acquired by collecting sand from Trinidad beach due to the similar grain size of the sediment. The blanks were stored in a 2L glass jar topped with aluminum foil and sealed with an aluminum lid. These samples were placed in a muffle furnace and baked at 600°F for 1 hour to vaporize any potential MPs. Samples were then sieved in a 5.25 phi screen for 10 minutes. This sample was then mixed with 0.5 L of DDI water with 30% H<sub>2</sub>O<sub>2</sub> and ran through the

density separation procedure. This blank was used to quantify the introduced MPs from the sieving and density separation procedures.

### Niskin Bottle Water Sample Collection

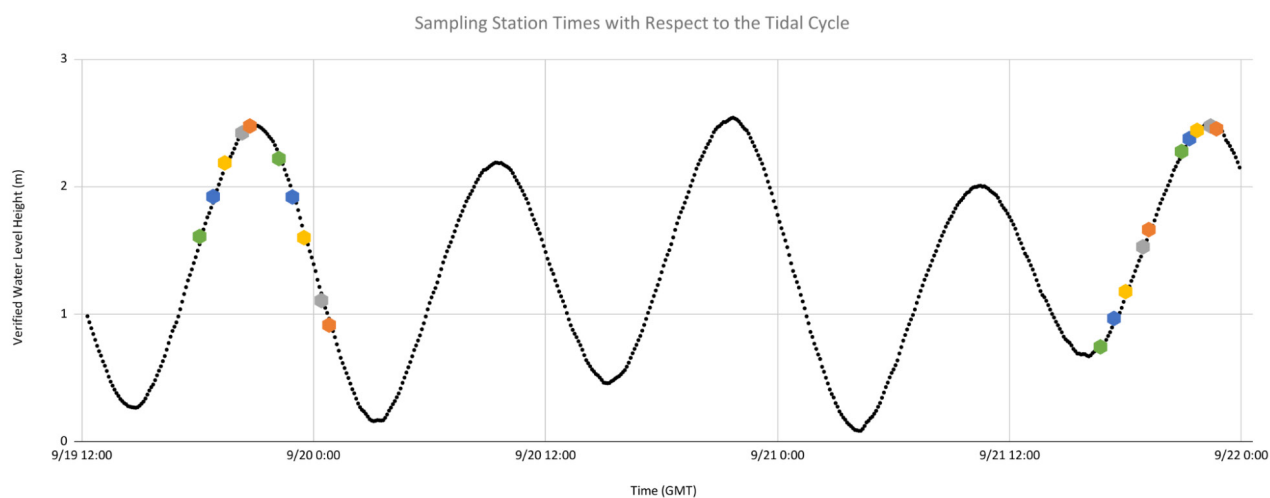
Water samples were collected on September 19, 2020, aboard the R/V Coral Sea at 5 locations throughout Humboldt Bay, each of which was sampled twice during a single tidal cycle (figure 3). Water samples were taken using a rosette armed with three Niskin bottles to collect samples at specific depths in the water column. The rosette was deployed in conjunction with a SeaBird Electronics 19 plus SeaCat CTD which measured conductivity and temperature, as well as a transmissometer which measured turbidity. The Niskin bottles were set to fire at 3 different depths: 1 m from the surface, 1 m from the bottom, and mid-depth relative to each station. Samples were taken directly from the stopcock stream exiting the Niskin with no additional plastic tubing that is commonly used to direct the stream. The samples were stored in 2L glass jars topped with aluminum foil and sealed with aluminum screw down lids. For transporting to the laboratory for processing, the samples were placed in storage bins. A 30% hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) solution was added to remove any biological organisms that could interfere with later analysis.

### Sediment Sample Collection

Prior to the cruise for sediment collection, all equipment

**Figure 3.**

Verified tidal cycle (m) at North Spit, Eureka, CA for September 19-21, 2020. The water sampling cruise aboard the R/V Coral Sea occurred on September 19th. The sediment sampling cruise aboard Cal Poly Humboldt's pontoon boat occurred on September 21st. Each color of data point represents each station: green=EC1, blue=NB4, yellow=NB3, gray=NB2, orange=NB1. Each station was sampled twice during each cruise. The station times were adjusted to plot where they were in the tidal cycle when they were plotted. The black data points represent the verified water levels taken at North Spit, Eureka, CA over the three-day study period.





pieces were cleaned with alconox and rinsed with DDI water. Sediment samples were collected on September 21, 2020, with a shipek grab aboard Cal Poly Humboldt's pontoon boat at 5 collection sites, each of which was sampled twice during a single tidal cycle (figure 3). The samples were stored in 2L mason jars topped with aluminum foil and sealed with aluminum lids. Samples were placed in storage bins for transportation to the laboratory. Sediments were collected once during flood tide and once during a high slack tide to compare the concentration of MPs during different points in the tidal cycle.

### Water Sample Laboratory Analysis

To separate the MPs from the water samples, we used a density separation process adapted from Wenfeng Wang, et al. (2018). After adding 100 mL of 30%  $H_2O_2$  to all water samples to remove organisms, the samples were left to settle for 48 hours. The full sample jar, minus the lid, was then weighed prior to pouring the sample into a separatory funnel. The empty sample jar was then weighed, and the mass of the jar subtracted from the total mass to determine the sample volume. The sample volume was then used to calculate the NaCl mass needed to saturate the sample to a density of  $1.3 \text{ g/m}^3$  (the ratio of salt to water  $\sim 360 \text{ g/L}$ ). The density of the saturated NaCl solution ( $1.202 \text{ g/cm}^3$ ) allowed less dense MPs, such as polyethylene ( $0.917\text{--}0.965 \text{ g/cm}^3$ ), polypropylene ( $0.85\text{--}0.94 \text{ g/cm}^3$ ), and polystyrene ( $1.04\text{--}1.1 \text{ g/cm}^3$ ) to be suspended in the supernatant after settling. The salt was then added to the funnel with the sample water, shaken vigorously, and then left to settle for 48 hours. The resulting supernatant was then re-mixed with additional saturated NaCl two more times. The final supernatant was then pumped through a  $20 \mu\text{m}$  glass fiber filter. Filters were then dried and weighed, and MP particle numbers were quantified using microscopy. MP concentrations were determined by dividing the volumetric quantity of MP particles by the total aliquot volumes.

### Sediment Sample Laboratory Analysis

The baking pans used for processing the sediment samples were cleaned with alconox and rinsed with DDI water. The wet sediment samples were then placed into the clean pans and dried at  $105^\circ\text{C}$  for 48 hours. The dry weight of the sediments was then taken prior to Ro-Tapping between  $-2$  and  $5.25$  phi sieves. The sediments were then dried again for 10 minutes, and a post-Ro-Tap dry weight was taken. For each sample, 500g of sediment was placed into a 1L jar with 180g of NaCl, 500mL of DDI water, and 50 mL of 30%  $H_2O_2$ . Each jar was vigorously shaken and allowed to settle for a minimum of

48 hours. The resulting supernatant was then decanted into a separate 1L jar capped with aluminum and set aside for later filtration.

### Filtration Process

Each sample was poured from their respective jar into a clean separatory funnel to begin the filtration process. The water and compacted salt were released from the spigot at the base of the separatory funnel until the volume in the funnel reached 200 mL. The excess was discarded. The remaining 200mL was then poured into the filtration setup to be filtered onto  $20\mu\text{m}$  glass fiber filters by vacuum pump. Post filtration, the filters were placed in aluminum boats and allowed to dry in a sealed, unheated oven.

### Filter Counts Procedure

To quantify the MPs on each filter, we began by pressing and sealing each filter between plastic graph paper. The filters were labeled with their respective filter numbers. Due to the limited number of people allowed in the lab following Covid-19 social distancing precautions, images of the filters were taken with a microscopy camera, to be counted offsite. Each image consisted of one  $5 \text{ mm} \times 5 \text{ mm}$  square of the filter. The images were then uploaded to a shared drive, with a total of 62 image files. At least three separate people counted

#### Figure 4.

Image of the upper left corner of Filter 15 taken with microscopy, showing a yellow fragment of hard plastic that is slightly larger than 1 mm.



particles on each filter, in an attempt to eliminate biases. MPs were identified in the images by first eliminating all particles exhibiting any cell-like organic structure. Identified MPs were then classified by size (<0.5 mm, 0.5-1.0 mm, 1.0-3.0 mm, 3.0-5.0 mm) and color (green, white/clear, red/orange, yellow, gray/black/blue) (example in figure 4). The counts were then averaged by filter and underwent a variety of statistics before producing the following plots in the results section.

## Results

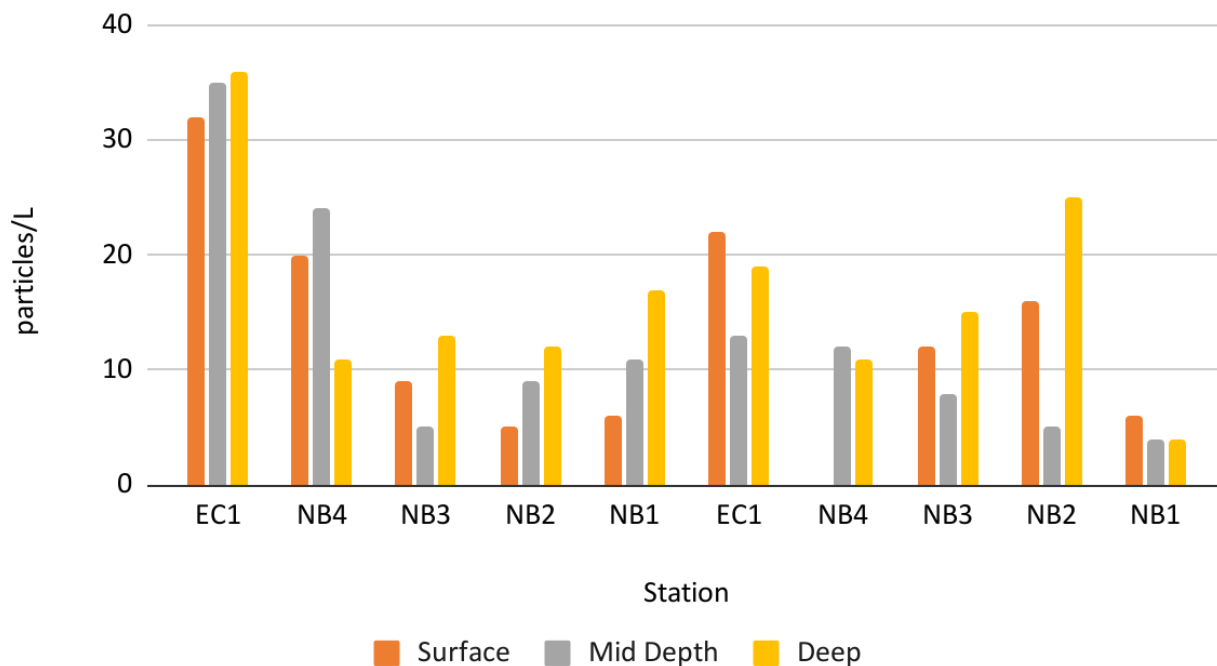
As displayed in figure 5, stations NB3, NB2, and NB1 when sampled on the flood all had fewer MP particles/L at the surface than at depth. This might have occurred by the particles being denser than the surrounding water, causing them to sink to the bottom. Overall, the number of MP particles/L is mostly consistent between the water samples taken on the flood and the ebb. It could be argued that this consistency is due to the energetic conditions experienced during both of the samplings. EC1 experienced a noticeable difference in concentration on the flood versus the ebb, which could mean the ocean is a provider of MPs to the bay. NB2, however, experienced higher concentrations on the ebb tide, possibly due to the water

becoming quite shallow. NB4 exhibiting zero MP concentration at the surface on the ebb could have occurred by the particles sinking with the release of water from the bay, or they could have been transported elsewhere.

MP concentrations in the sediment were predicted to be at a maximum during the lowest current speed, during slack tide, as the relatively lower kinetic energy without the tides would potentially allow the microplastics to settle in the sediment. Our results show that the highest microplastic concentrations occur at stations NB4 during flood and NB3 during slack (figure 7). Overall, the sediment samples do not represent the expected changes in concentration with the variation in the tide. Aside from NB3, all stations saw higher concentrations of MPs during the second leg of the pontoon boat cruise. Unexpectedly, plastics had mostly higher concentrations during flood tide rather than settling during slack tides. This could be due to the tide bringing in MPs from the ocean and depositing them into the bay, or it could be due to the heterogeneous nature of MP distribution. The inconsistent values seen at each site are possibly due to natural variations in the concentration of MPs, the shipek grab deployment locations not being precise on site, and the tidal range being smaller than ideal for sampling.

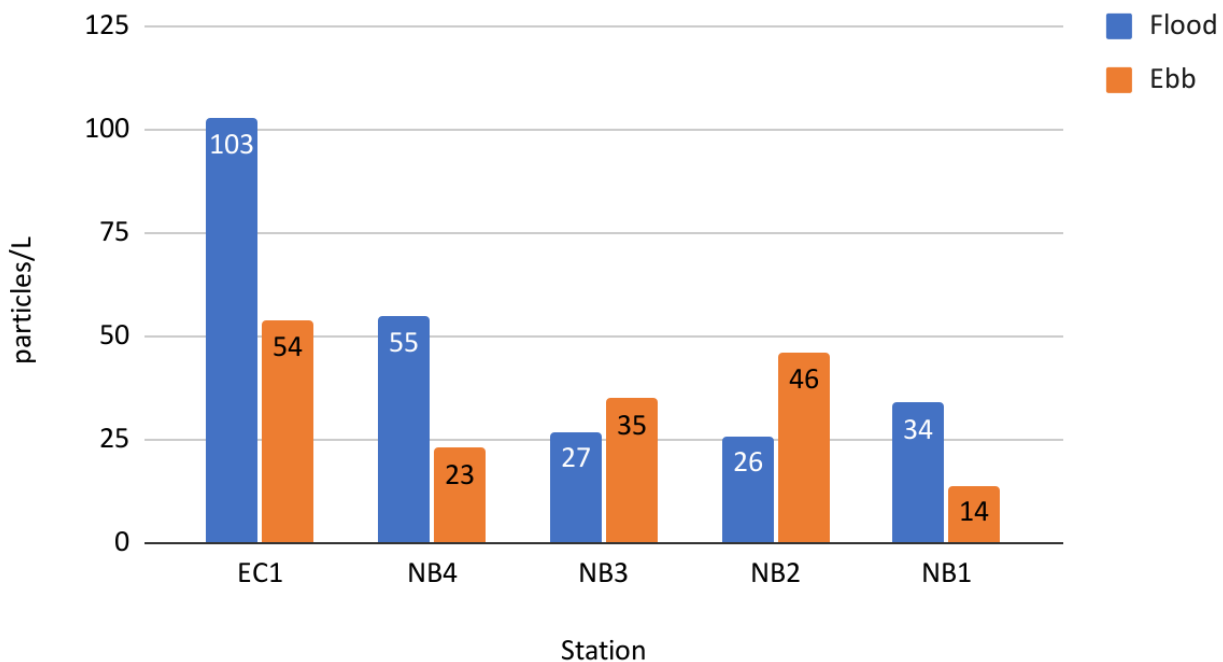
**Figure 5.**

Water Column Microplastic Concentrations by Depth. The concentration of microplastics in the surface of the water column were noticeably the greatest at EC1 and NB2. These stations corresponded to the tidal change from flood to ebb and from ebb to flood, respectively. This data was collected by the Niskin bottle sampling of different depths in the water column procedures aboard the R/V Coral Sea on September 19, 2020. Total surface average concentration: 12.8 particles/L  $\pm$  9.70. Total mid depth average concentration: 12.6 particles/L  $\pm$  9.77. Total deep average concentration: 16.3 particles/L  $\pm$  8.88.



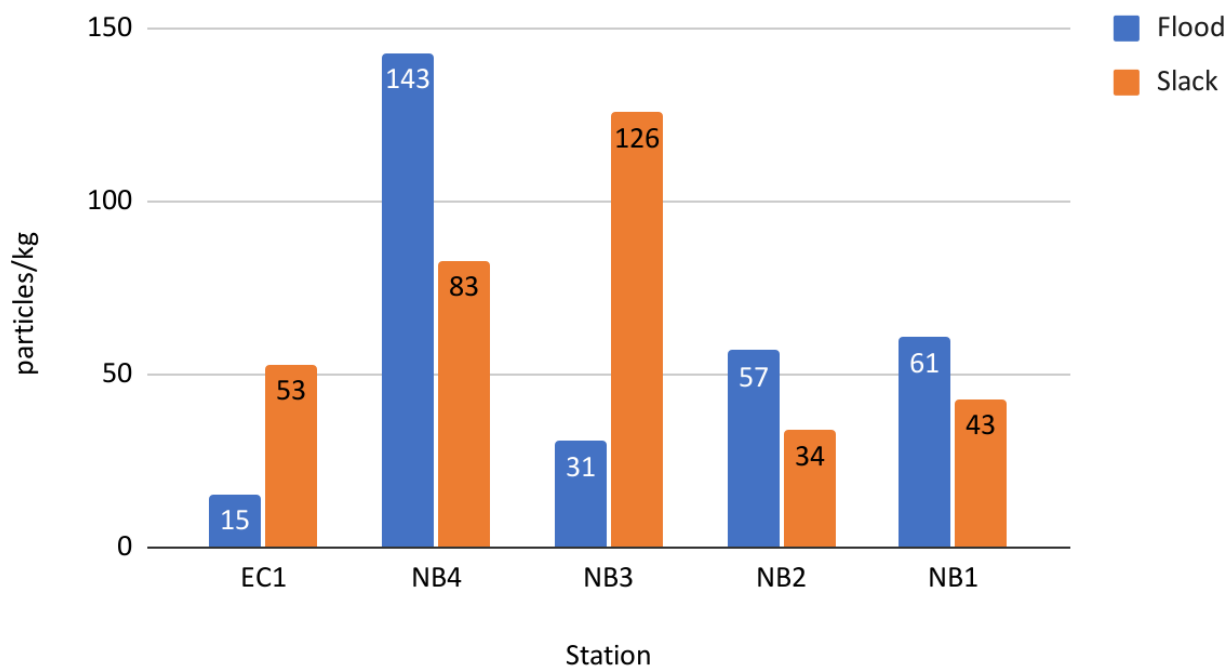
**Figure 6.**

Total MP concentration present in the water column per station on the flood versus the ebb of the tide. This data represents the sum of the MP concentrations represented in figure 5 from samples collected on September 19, 2020. Overall average concentration of 41.7 particles/L  $\pm$  25.36. The average of flood concentrations was 49.0 particles/L  $\pm$  32.37. The average of ebb concentrations was 34.4 particles/L  $\pm$  16.32.



**Figure 7.**

Sediment MP concentrations in particles/kg, from the pontoon boat cruise in Humboldt Bay on September 21, 2020. The blue bars represent sampling taken during flood tide, and the orange bars represent sampling during slack tide. MP concentrations were found to be highest in the NB4 flood sample and the NB3 slack sample. The lowest concentration was seen at EC1 during the flood tide. A standard deviation of  $\sigma = \pm 41.44$  particles/kg was calculated based on this data and an average of 64.6 particles/kg. Average on the flood was found to be 61.4 particles/kg  $\pm$  49.38. On the slack, the average concentration for all stations was 67.8 particles/kg  $\pm$  37.40.



**Figure 8.**

Study station map of Humboldt Bay with transposed approximate 5-minute cotidal lines (seen in red) based on the time of the high tide in the NOAA tidal prediction model at various stations around Humboldt Bay (“Tide Predictions - NOAA Tides and Currents”).



We hypothesized that sediment MP concentrations in the extremities of the bay would have a larger MP concentration. EC1 saw the lowest MP concentration of any station, which lines up with our hypothesis. NB2, however, was the farthest station from the mouth of the bay, yet it saw the second

lowest total concentration of the 5 stations, possibly due to the site being in the center of NB and farther from terrigenous sources. MP concentrations were found to be highest overall at NB4, which is located at a point of convergence of the three main channels in the bay.

The concentrations of MPs found in sediments were lower than the maximum of 400 particles/kg mentioned by Wagner et al. (2014), with our two highest concentrations at 143 particles/kg (NB4 flood), and 126 particles/kg (NB3 slack). The overall average sediment MP concentration was 64.6 particles/kg with a standard deviation of  $\sigma = \pm 41.44$  particles/kg, making Humboldt Bay sediment relatively plastic-free; however, the evidence of MPs might allude to higher concentrations existing in places that were not sampled during this study.

The kinetic energy associated with the tidal cycle was expected to affect MP concentrations in the sediments and in the water column. To quantify the kinetic energy, we calculated the velocity covariance, which is directly proportional to the kinetic energy. To determine the velocity covariance ( $v^2$ ), current velocity (cm/s) data was obtained from the NOAA Physical Oceanographic Real-Time System (PORTS) station at Chevron Pier in Humboldt Bay for the respective sampling times at each station (“CO-OPS Current Station Data”). To account for the noticeable delay in the dispersion of Humboldt Bay’s tide, cotidal lines of approximately every five minutes were determined from the NOAA Tide Predictions historic data from the eight stations north of the Entrance Channel in Humboldt Bay (figure 8).

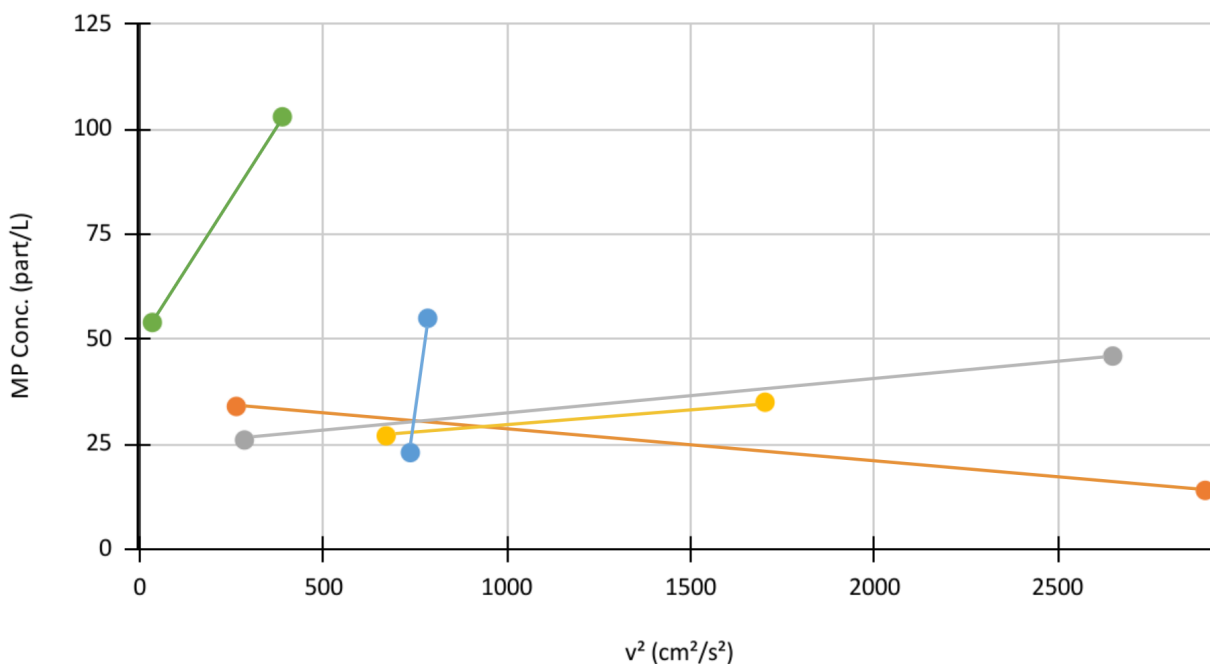
The respective velocities were then squared to find the velocity covariance and plotted against the MP concentrations

of both the water column and the sediment samples, as seen in figures 9 and 10. The expectation was that when the covariance was high, the microplastics on the seafloor would be resuspended in the water column. Thus, when the covariance was higher at a given station, the microplastic concentration in the sediments would be lower and the microplastic concentration in the water column would be higher. The covariance values on the water sampling cruise were nearly an order of magnitude greater than the covariance values from the sediment sampling cruise. This difference was expected due to the difference in the tidal range between the two sampling days. The water sampling cruise occurred during an ideal tidal range to test our hypothesis, with a higher high and a lower low tide leading to greater tidal velocities. The sediment sampling cruise occurred on a day with less ideal conditions: a smaller tidal range and smaller tidal velocities.

The velocity covariance and the MP concentration in the water column for each station follows the expected pattern of higher covariance, higher MP concentration at all stations, except NB1 (figure 9). Site NB1 was located near both the Eureka Public Marina and the Woodley Island Marina, exposing it to high amounts of boat traffic. The increased human activity at this station could have affected both the concentration of MPs in the area and the mixing of the water column between sample collections.

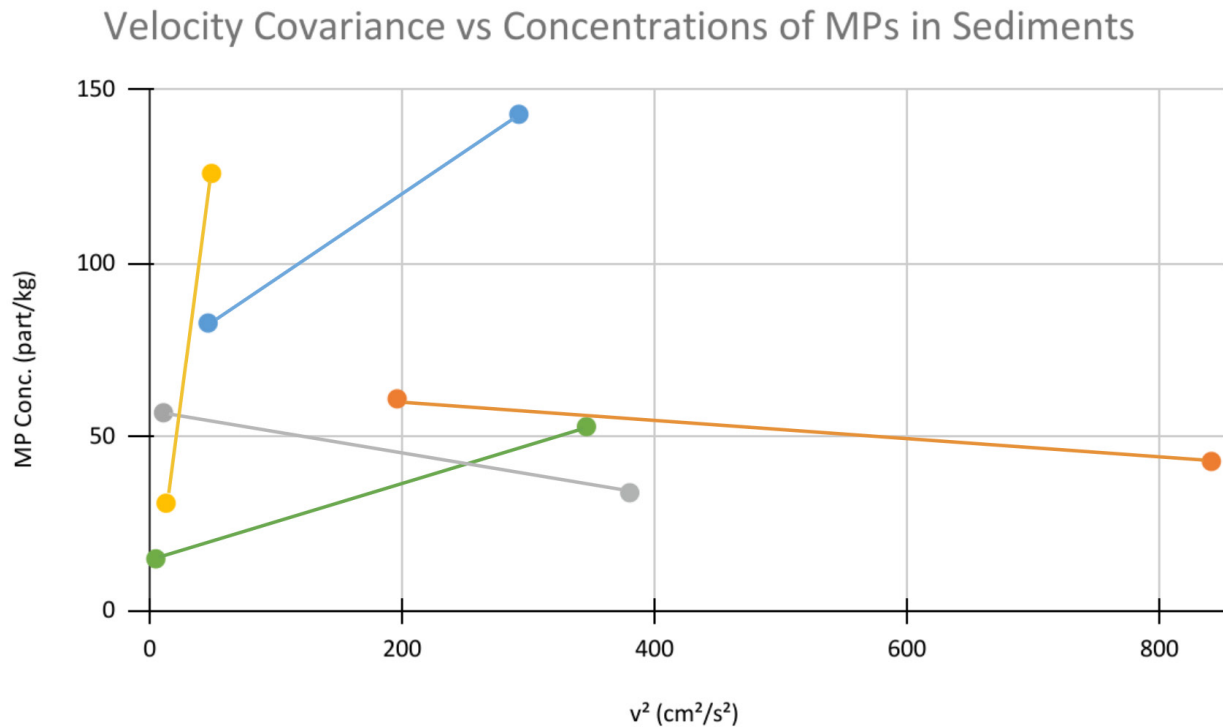
**Figure 9.**

Velocity Covariance vs Concentration of MPs in the Water Column. Each color of data point represents each station: green=EC1, blue=NB4, yellow=NB3, gray=NB2, orange=NB1. Each station was sampled twice during the tidal cycle, as seen in figure 3. Velocity data obtained from the NOAA PORTS data for Chevron Pier in Humboldt Bay (“CO-OPS Current Station Data”).



**Figure 10.**

Velocity Covariance vs Concentration of MPs in Sediment. Each color of data point represents a station: green=EC1, blue=NB4, yellow=NB3, gray=NB2, orange=NB1. Each station was sampled twice during the tidal cycle, as seen in figure 3. Velocity data obtained from the NOAA PORTS data for Chevron Pier in Humboldt Bay (“CO-OPS Current Station Data”).



### Discussion

Similar to the results found by Carlson et al. (2018), the sites with higher boat traffic had higher MP concentrations in the water column and sediments. However, EC1 concentrations contradict this pattern in the sediment data, likely because of the higher kinetic energy in the channel mouth that would not allow the MPs to settle fully into the sediment. It is also possible that MPs were present deeper down in the sediment at EC1, as well as at other sample sites, therefore they were not collected during the deployment of the shipek grab.

Future experiments would benefit by sampling on a singular cruise to fully utilize a greater tidal range, as bodies of water similar to Humboldt Bay can experience a broad range of conditions within a small time period. Unfortunately, due to Covid-19 boat time restrictions, obtaining more cyclical data was not possible with this study. In the future, to obtain a complete data set, samples should be collected on multiple days with various tidal ranges and conditions.

A possible cause for the three stations closest to the Entrance Channel experiencing a higher MP concentration on the floodtide is the MPs being supplied by the Pacific

Ocean. The Pacific Ocean currently has two Great Pacific garbage patches, one of which is between California and Hawaii. This patch could act as the source of ambient plastics that can be transported along the western coast of N. America and throughout the Pacific Ocean. Local sources that deposit directly into the ocean, including Mad River and Eel River, could also contribute to the incoming MPs. Future studies could use a spectrometer for the identification of the MPs that could help pinpoint the source of pollution. In addition, this method would help minimize human error in identifying plastics and allow for more repetition within the study. These data could be used to help identify and reduce plastic pollution in Humboldt Bay and nearby bodies of water. As this was a preliminary study that could not yield as much data as desired, more sampling is necessary to comprehend how and where MPs move in Humboldt Bay.

The prevalence, distribution, and environmental impacts of MPs are not entirely understood, which is why baseline studies, like this one, are so important. The results from this study will hopefully improve the scientific community's understanding of the dynamics of MP distribution, helping to pave the way for further research into the cumulative and projected impacts of MP pollution in estuaries and elsewhere.

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