

EXAMINING SPATIAL CONCENTRATIONS OF
MARINE MICRO-PLASTICS ON SHORELINES
IN SOUTH PUGET SOUND, WASHINGTON

by

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ABSTRACT

Examining spatial concentrations of marine micro-plastics on shorelines in South Puget Sound, Washington

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The spatial distribution and density of marine micro-plastics (5.6 mm - 0.3 mm) on 12 shorelines within three isolated finger inlets in South Puget Sound, Washington were investigated during the winter of 2013. The mean density of buried marine micro-plastics in Budd, Totten and Eld Inlets was 89, 2.1 and 2.3 items/ m² respectively. Budd Inlet contained 45 fold more micro-plastics/ m² than Eld and Totten Inlets. Within each inlet an area was located where micro-plastic concentration was significantly higher relative to other locations within the inlet. Single locations in Budd, Eld and Totten comprised 98%, 68% and 59% of the total micro-plastics collected within each respective inlet indicating that the spatial distribution of micro-plastics is not even between inlets, or among inlets, in South Puget Sound. The micro-plastics were divided into two size classes that ranged from 5.6mm-1.0mm and 1.0mm-0.3mm. A strong positive correlation between the two size classes was observed (R^2 0.96, $p=0.0001$). A strong positive correlation was also observed between the areas of high micro-plastic abundance and the population density of the inlets (R^2 0.74 $p=0.0001$). Study sites with a south aspect in South Puget Sound accumulated more micro-plastics than study sites that faced other directions, and this is believed to be driven by interaction with the predominate Southwesterly wind. The findings of this study suggest that spatial distribution between the inlets is driven by a combination of anthropogenic factors, while the spatial variability within the inlets is driven by physical factors.

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INTRODUCTION

The annual global production of plastics has increased from ~1.7 million tons in 1950 to ~280 million tons in 2011, at a rate of approximately 9% growth per year (Plastics Europe 2012). While the benefits of plastics have helped to transform society, plastic is also contaminating many parts of our environment. Discarded plastics are found all over the marine world; from heavily populated areas to the most remote beaches in Antarctica (Eriksson et al. 2013).

Micro-plastics which range from 5 mm to 0.3 mm, are either manufactured as small pieces of plastic (primary) or they are created when larger pieces of plastic degrade into smaller pieces (secondary) once disposed of in the landfill or in the natural environment (Betts 2008). Eliminating marine micro-plastics pollution is one piece of creating a healthy marine eco-system as the potential for negative environmental impacts from micro-plastics has been established in the literature. The small size of micro-plastics is believed to create a hazard to organisms through ingestion (Baird and Hooker 2000; Boerger et al. 2010; Davison and Asch 2011; Derraik 2002; Fossi et al. 2012; Graham and Thompson 2009; Kociubuk in press; Laist 1997; Murray and Cowie 2011).

The quantity, rate and locations that plastic is accumulating in different environments, such as the ocean, is not understood, thus limiting clean-up efforts. Micro-plastics were initially recognized in the marine environment in 1972 (Carpenter and Smith 1972). Since that time, with the rise in global plastic production, the quantity of plastics found in the marine environment is believed to be increasing as well (Plastics Europe 2012; Thompson et al. 2004).

Puget Sound is the second largest estuary in the United States and is located in Washington State (Gaydos 2008). Research on marine micro-plastics in Puget Sound is not existent in the published literature. This is the first study to examine marine micro-plastics in South Puget Sound. South Puget Sound is the furthest point away from the entrance of Puget Sound. This location was selected because South Puget Sound's isolated location, similar wind and current patterns in combination with differences in population between the smaller finger inlets located in the most southerly section of South Puget Sound, create conditions in the field that are very close to a controlled experiment and allow exploration of the drivers of spatial distribution of marine micro-plastics and establish baseline data to track if micro-plastics are increasing over time locally. Thus, it advances the scholarly literature by creating a baseline from South Puget Sound and identifies the primary physical and anthropogenic drivers of marine micro-plastics spatial distribution.

Aside from eliminating the source of micro-plastics, the best way to reduce marine micro-plastic pollution and their subsequent impacts to wildlife is to remove the plastic from the marine environment and dispose the plastic pollution properly. Once micro-plastics have entered the marine ecosystem, there are four locations that they are believed to be accumulating in. The sea surface, open ocean floor, near coastal sediment and shorelines (Ryan et al. 2009a). Thus the easiest location to collect and remove plastic pollution is from shorelines. Understanding the spatial distribution of marine micro-plastics along shorelines can facilitate focused clean-up efforts to increase cleanup efficiency. Identifying the physical factors that are driving the spatial distribution in a specific area creates the potential for isolating the source locations of micro-plastics.

Understanding how population and physical factors drive the spatial distribution of marine micro-plastics is the key to facilitating both effective education programs to the public about reducing inputs as well as coordinating efficient shoreline cleanup efforts. In the marine environment, the small pieces of degraded plastic are thought to be accumulating in the same areas, primarily the sea surface and shorelines that marine organisms inhabit, thus creating the potential to negatively impact the health of the entire marine ecosystem. Plastics have been demonstrated to accumulate persistent organic pollutants and trace metals (Ogata et al. 2009; Teuten et al. 2009; Teuten et al. 2007). Emerging work has demonstrated that Polybrominated diphenyl ethers (PBDEs) were transferred into the tissues of Short tailed Shearwaters from pieces of micro-plastic that they ingested (Tanaka et al. 2013)

LITERATURE REVIEW

Introduction

Plastic is one of the most prevalent materials in the world. Annual global production of plastics has increased from ~1.7 million tons in 1950 to ~280 million tons in 2011, at a rate of approximately 9% growth per year (Plastics Europe 2012). While the benefits of plastics have helped to create the current world we live in, discarded plastic is also contaminating many parts of our environment. The quantity and rate at which plastic is accumulating in different environments, such as the ocean, is not understood.

Plastics begin to degrade into smaller pieces once disposed of in the landfill or in the natural environment. In the marine environment small pieces of degraded plastic are thought to be accumulating in the same areas that marine organisms inhabit, thus creating the potential to negatively impact the health of the entire marine ecosystem (Ryan et al. 2009a). The degradation of plastic into smaller pieces creates a hazard to organisms through ingestion. Furthermore, persistent organic pollutants and trace metals have been demonstrated to accumulate in plastics. Marine organisms such as lobsters, oysters and blue mussels have been observed with marine micro-plastics in their gut contents, in the field as well as in laboratory experiments (Kociubuk in press; Murray and Cowie 2011; von Moos et al. 2012).

This literature review addresses what is currently known about the broad topic of marine micro-plastic pollution. Micro-plastics which range from 5 mm to 0.3 mm, are either manufactured as small pieces of plastic (primary) or they are created when larger pieces of plastic degrade into smaller pieces (secondary) once disposed of in the landfill or in the natural environment (Betts 2008). Initially the uses of plastics, the creation of

micro-plastics and how long those micro-plastics are believed to remain in the marine environment is discussed. Next the sorption of toxic chemicals and trace metals as well as the negative impacts that micro-plastics have on wildlife is examined. The current sampling methodology along shorelines is examined to demonstrate that the methods employed in this thesis are consistent with that which is established in the literature. The results from past shoreline micro-plastic studies that have used this common methodology and exploration of the theories that drive the spatial distribution of these results is the final topic that is examined.

Uses of Plastic

The term plastic applies to many different materials that are derived from petrochemicals produced from oil or gas (Thompson et al. 2009). Based on volume, plastics are one of the most used materials in the United States both industrially and commercially (Society of Plastics Industry 2012). 50% of annual plastic production is used in single-use disposable applications, such as packaging, agricultural coverings and disposable consumer items. Thus the majority of plastic products has very short useable lifespans and is designed to be thrown away. In the United States, 31 million tons of plastic waste was generated in 2010, which is approximately 12.4% of the total municipal solid waste for the nation (Environmental Protection Agency 2012).

Plastics in the Marine Environment

Micro-plastics were initially recognized in the marine environment during oceanographic research cruises in 1972 (Carpenter and Smith 1972). Since that time,

global plastic production has increased by approximately 9 percent per year and the quantity of plastics found in the marine environment is believed to be increasing as well (Plastics Europe 2012; Thompson et al. 2004). Multiple research studies have found varying densities of marine micro-plastics around the globe. Marine micro-plastic pollution has been found in every ocean and on isolated shorelines, such as Antarctica, midway atoll in the middle of the Pacific Ocean and on the Fernando de Noronha Archipelago in the middle of the South Atlantic Ocean, making marine micro-plastic pollution a global issue (Barnes 2002; Cooper and Corcoran 2010; Costa et al. 2010; Ivar do Sul et al. 2009; McDermid and McMullen 2004; Moore et al. 2002).

Pathways for plastics to enter the marine environment

Plastic enters the ocean through a variety of sources. Plastics can be delivered into the marine environment through disposal of ship waste, spills of feedstock pellets into water bodies and rivers, delivered through sewer systems and waste water treatment plants' outfall pipes, washed into the ocean by storm water drains and runoff, blown into the ocean from land or simply left on the shoreline by beach goers (Ryan et al. 2009a). An addition input of plastics are microscopic scrubbers which have replaced natural exfoliating materials used in many household cleaners, body washes and facial scrubs (Fendall and Sewell 2009; Gregory 1996). Once these microscopic scrubbers are used, they are washed down the drain where they pass through waste water treatment plants and are discharged into waterways.

In 2007, it was estimated that two-thirds of the Earth's population lived within 100 miles of a coastline and this number is expected to increase (*Coastal Hazards* 2007).

With the large amount of disposable products being produced globally and the large number of people living close to the coasts the potential for plastic waste to enter into the marine system from land is high. Andrady (2011) estimated that 80% of the plastic that enters the marine environment comes from land based sources. However, the quantity of plastics that enter into the marine environment has not been reliably estimated (Andrady 2011).

Micro-plastics

Much of the research examining the distribution of plastics in the ocean has focused on micro-plastics because of suspected increases in abundance and micro-plastic's similar size to food sources of many marine fauna. Micro-plastics have been recently defined as plastic that ranges from between 5 mm and 0.3 mm (Betts 2008), but historically definitions have ranged between less than 20mm and .001 mm (Hidalgo-Ruz et al. 2012). For the purpose of this literature review, the former definition will be used.

Micro-plastics can be manufactured as small pieces of plastic, such as plastic production pellets, plastic beads for bead blasting, or micro scrubbers for facial washes and dish soap (Fendall and Sewell 2009; Gregory 1996). Micro-plastics can also be created by the degradation of larger pieces of plastic discarded in the marine environment.

Trends of micro-plastics in the marine environment

The direction of long-term trends of micro-plastics accumulating in the marine environment are unclear. Different studies have found that plastics are accumulating,

staying steady or not accumulating (Goldstein et al. 2012; Gregory 2009; Law et al. 2010; Thompson et al. 2004). The changes in these trends could be influenced by changes in the way people are disposing plastics globally.

Thompson et al. (2004) re-examined zooplankton samples from neuston trawls that dated back to the 1960's from the North Sea for micro-plastics. Examination of those samples allowed the authors to see an increase in micro-plastic abundance over the last 50 years which was in concert with the increases in plastic production worldwide (Thompson et al. 2004). Similarly, in the North Pacific micro-plastics are believed to have increased by 2 orders of magnitude from 1972 to 2010 (Goldstein et al. 2012). The authors note that although they believe micro-plastic abundance is increasing, they note that sampling was not conducted from 1987 to 1999 allowing the potential that the temporal pattern found could have more variation due to shifting wind and current conditions during the study years.

In contrast, one of the longest running continuous studies (1986- 2008) on marine micro-plastics, Law et al. (2010) found micro-plastics in 68% of the 6136 tows, but found no spatial or temporal trends in the abundance of micro-plastics in the western Atlantic and Sargasso Sea, with the density of micro-plastics observed in each neuston tow remaining the same (Law et al. 2010). Finally, a study conducted over a similar period examining pre-production plastic pellets (nurdles) on shorelines in Bermuda, eastern Canada and New Zealand from the 1970's to the mid 2000's found a slight decline in the abundance of nurdles on the shorelines sampled (Gregory 2009).

These differences in trends of micro-plastics, demonstrate the variability of not only the marine system that is being studied, but also reflect the varying methodologies

being used between studies. For example, Gregory (2009) was only looking for pre-production plastic pellets (nurdles). During the time frame of his study, reduction of spills was a major priority of manufactures and this reduction could be what is reflected in his data. More benefit would have come from Gregory (2009) if all the plastic litter on shorelines was examined. Thus, the decline observed by Gregory (2009) could be simply due to the fact that he looked at nurdles, whereas the other studies examined other types of plastic. The author notes that the decrease in abundance of nurdles could be due to changes in the handling and transport. Examining all micro-plastic debris on the shorelines would have allowed the author to see if their observations were in line with increases in production of plastic products.

Another example as to how potential discrepancies in study design could affect temporal trends is Thompson et al. (2004). They reanalyzed zoo-plankton samples that were collected over 50 years, but little is known about the collection methods and if the people who initially analyzed the zooplankton samples ever discarded micro-plastics when identifying the zoo-plankton.

Aside from the potential discrepancies in methodology, the differences in these trends could be attributed to different anthropogenic influences. Just as the reduction of nurdle spills from industrial applications was seen as a reduction in marine micro-plastics by Gregory (2009) the lack of trend in the density of plastics in the North Atlantic over 22 years by Law et al. (2010) could be attributed to increased recycling and improvements in waste disposal that may cancel out the increase in the global production of plastics.

Goldstein et al. (2012) and Thompson et al. (2004) both demonstrate opposite trends from Gregory (2009) and Law et al. (2010). Both Goldstein et al. (2012) and Thompson et al. (2004) observed increases in the abundance of micro-plastics in the North Pacific and the North Atlantic respectively which they believe indicates that the global increase of plastics is creating more plastic pollution in the oceans. Thus, although a review of the academic literature suggests that there does not appear to be consistent temporal trend in micro-plastic abundance, the presence of marine micro-plastics in the water and the potential threat these small pieces pose to wildlife.

Whether the abundance of micro-plastic is constant or increasing, the processes that create micro-plastics are becoming more understood by academia. The sorption of toxic chemicals and the impacts to wildlife has been demonstrated but to be able to fully understand the temporal and spatial variability, the factors that produce micro-plastic need to be understood.

Degradation of Plastics

The processes creating micro-plastics from larger pieces of plastic are believed to be a combination of physical degradation from wave action and chemical weathering from the sun. Plastics are believed to be initially susceptible to photodegradation and physical processes (Andrady 2005; Cooper and Corcoran 2010; Webb et al. 2012). The process of photodegradation is the deconstruction by UV based processes, breaking down molecules into lower molecular weight fragments (International Union of Pure and Applied Chemistry 1996). Photodegradation causes the plastics to become brittle, then

physical processes such as wave action, cause the plastics to break into smaller pieces in the marine environment (Webb et al. 2012).

On land the complete mineralization of polymers has been observed. After break down from photodegradation, thermo-oxidative degradation is the next step that occurs quickly, when heat and oxygen accelerate the breaking of the polymer chains that created the plastic. Once photodegradation and thermo-oxidation have occurred, bacteria degradation can consume the remaining polymer chains (Shah et al. 2008; Zheng et al. 2005). However, this process has not been observed in the marine environment largely due to the long time scales it takes to breakdown plastics in the marine environment. Current estimates for mineralization, the complete reduction of a polymer to inorganic components, of plastics in the marine environment range from hundreds to thousands of years (Andrady 2005; Barnes et al. 2009). Once plastics have entered the marine environment they are believed to be a long lived hazard. However, there is debate on the time frame for full mineralization of the polymer chains back into water and carbon dioxide.

The time frame for full mineralization is hard to generalize as each additive to a plastic product changes the durability of the product. It has also been found that identical products will break into smaller pieces at many different speeds depending on the environment they are left in (A.C. Albertsson, Personal Communication, January 25th, 2013). Cooper and Corcoran (2010) believe that the degradation of plastics to micro-plastics and eventually to unseen particles via mechanical and chemical weathering will create microscopic plastic particles that will remain in Earth's marine environment indefinitely.

The time frame that plastics are re-mineralized is believed to be slower in the marine environment than on land. The cold temperatures of the oceans and the lower oxygen concentration in the deep oceans are believed to slow the degradation rates of plastics (Andrady 2011). The growth of marine organisms on plastic is called fouling. Fouling is also believed to slow the degradation rate by blocking the UV light and thus slowing photodegradation and the process of re-mineralization even further.

A 2008 study examined the degradation rates of polyethylene bags. The study found that after 40 weeks in the water less than two percent of the surface area of the polyethylene bags was lost. All samples had been fouled by organisms and the presence of these organisms is believed to slow their degradation times by blocking sunlight (O'Brine and Thompson 2010).

O'Brine and Thompson examined the breakdown of the plastic bags in sea water to micro-plastics, but they did not examine the mineralization of the materials. Roy et al. (2011) examined the complete degradability of polyethylene sheets and the recent development of biodegradable polyethylene. Polyethylene is the most common plastic in use today and the primary component in packaging and common plastic bags. They found that polyethylene bags disintegrate into pieces invisible to the naked eye, but still harmful to marine organisms. The bags did not completely remineralize over a time frame, though that time frame was not stated, that would avoid negative impacts to the marine environment (Roy et al. 2011).

Cooper and Corcoran (2010) found that polyethylene appears to be more conducive to degrading from a combination of chemical and physical processes while

polypropylene appears to degrade from physical processes first (Cooper and Corcoran 2010).

Because mass production of plastics began within the last 60 years, no long-term studies in terrestrial or marine environments have been conducted to estimate the actual time frame for which plastics will be remineralized and incorporated into biomolecules (Andrady 2011; Roy et al. 2011; Webb et al. 2012).

Dr. Ann-Christine Albertsson, a leader in plastic degradation studies and the head of the Polymer Technology Department at KTH in Stockholm, Sweden explains that there are thousands of materials with very different chemistry that can be called plastic. There are many types of plastic and each type contains different additives. These various plastics are being used and discarded in different surroundings, making their interaction with the environment very complex. For example, very simple plastics may stay for thousands of years in a landfill but disappear quickly in the next surrounding (A.C. Albertsson, Personal Communication, January 25th, 2013).

Marine Micro-plastics as a Vessel for Chemicals and Trace Metals

Common micro-plastics are less dense than water allowing them to float near the surface of the water. This characteristic of plastic is believed to be the reason that micro-plastics are initially found on the surface of the sea. Persistent Organic Pollutants, hydrophobic compounds and trace metals, have also been found accumulating in the sea-surface micro layer as well. Concentrations of toxic chemicals and trace metals have been found up to 500 times greater in the sea-surface micro-layer, than concentrations in the water underneath (Ogata et al. 2009; Teuten et al. 2009; Teuten et al. 2007). As such,

micro-plastics and Persistent Organic Pollutants are present in the same micro layer, thus causing micro-plastics to be vectors for organic pollutants, as will be discussed below.

It has been found through various studies that Persistent Organic Pollutants are attaching to plastics through sorption. Bisphenol A (BPA), chlordanes, dichlorodiphenyltrichloroethane (DDT's), dischloroethene (DDE's), hexachlorocyclohexane (HCH), nonylphenol, polychlorinated biphenyls (PCBs), Polycyclic aromatic hydrocarbons (PAHs), and Polybrominated diphenyl ethers (PBDEs) have all been found to achieve sorption into plastics in the marine environment (Bakir et al. 2012; Endo et al. 2005; Hirai et al. 2011; Ogata et al. 2009; Teuten et al. 2009; Teuten et al. 2007; Van et al. 2012). When micro-plastics were analyzed for trace metals they were found to contain Aluminum, Copper, Chromium, Cobalt, Cadmium, Iron, Manganese, Nickel, Lead and Zinc (Ashton et al. 2010; Holmes et al. 2012). These initial studies demonstrate the potential for metals as well as POP's to be transported into the food chain through the consumption by marine organisms where they would not have originally appeared.

Plastics are initially produced to be biochemically inert materials that do not affect the endocrine system because of their large molecular size which does not allow them to penetrate through cell membranes. Additives within plastic products are believed to be disruptive and create the potential for new plastic products to carry chemicals that can disrupt the endocrine system (Teuten et al. 2009). As plastic products degrade in the marine environment, their molecular weight decreases (Andrady 2011). Plastics with lower molecular weight have higher rates of sorption of chemicals than plastics with larger molecular weights (Teuten et al. 2009).

The potential for chemicals and trace metals to sorb into wildlife has been demonstrated in the laboratory. Tanaka et al. (2013) is the first study to find that chemicals from marine micro-plastics are accumulating in marine wildlife. All the Short tailed Shearwaters studied contained from 0.04 g-0.59g of micro-plastics in their stomachs. The Shearwaters tissue was examined for the chemical compounds found on the micro-plastics from their stomachs. Polybrominated diphenyl ethers (PBDEs) was found in all the Shearwaters tissues (Tanaka et al. 2013).

Although the potential for micro-plastics to be a vector for the sorption of toxic chemicals and trace metals in marine organisms is beginning to be understood, further work is needed to understand the full scope of the issue and what the biological implications are for the sorption of chemicals from micro-plastics. Micro-plastics threat to certain species through ingestion has already been well documented.

Impacts of Marine Micro-plastics on Marine Wildlife and Ecosystems

The potential for plastics to harm ecosystems is high and potential mechanisms by which they could do so are through the transport of invasive species and toxic chemicals, and being ingested by wildlife and entangled in marine debris (Baird and Hooker 2000; Bakir et al. 2012; Barnes 2002; Boerger et al. 2010; Davison and Asch 2011; Derraik 2002; Endo et al. 2005; Fossi et al. 2012; Graham and Thompson 2009; Gregory 2009; Laist 1997; Murray and Cowie 2011; Ogata et al. 2009; Teuten et al. 2009; Teuten et al. 2007; Williams et al. 2011). However, actual documentation of harm to marine species has only been demonstrated for ocean going seabirds (Colabuono et al. 2009).

Direct Impacts (Primary consumers in the food chain)

Marine organisms have the potential to consume micro-plastics. Laist (1997) was the first study to compile a list of species that interact with marine debris. Laist found over 100 animals that either ingested or were entangled in marine debris (Laist 1997).

Papers have emerged since the original 1997 census taken by Laist (Baird and Hooker 2000; Boerger et al. 2010; Davison and Asch 2011; Derraik 2002; Fossi et al. 2012; Graham and Thompson 2009; Kociubuk in press; Laist 1997; Murray and Cowie 2011; Williams et al. 2011). It has now been shown that most marine organisms are indiscriminant eaters and will consume anything that passes by that resembles a food source including micro-plastic. Large mammals such as Baleen whales down to Oysters all have the potential to ingest marine micro-plastics.

Williams et al. (2011) conducted a study off of the north end of Vancouver Island in the Eastern Pacific looking at the relationship between marine mammals and plastic marine debris. Marine mammals were present in the same areas that accumulations of plastic marine debris were found. Williams et al. (2011) found that micro-plastic abundance off the North End of Vancouver Island was similar to those found in more urban areas suggesting that the potential for ingestion by marine mammals was present even in isolated areas (Williams et al. 2011).

Many of the studies that examine consumption of micro-plastics are based on theoretical models and laboratory experiments. However, a few studies examining myctophids and lobsters used direct methods from samples collected at sea. They found micro-plastics were ingested by both species (Boerger et al. 2010; Lusher et al. 2013; Murray and Cowie 2011). The potential for micro-plastics to disrupt food webs has been

exhaustively explored but the work done in the field to truly measure is just beginning to emerge. Seabirds are the most studied consumer of marine micro-plastics.

Procellariiformes (albatross, shearwaters and petrels) are commonly found deceased with micro-plastic pieces in their stomachs. It is believed that procellariiformes are the most affected bird from marine micro-plastic pollution. This is thought to be a result of the combination of feeding habits, collecting zoo-plankton from the ocean's surface, and regurgitating the food for chicks on land. Marine micro-plastics have been observed filling the stomachs of various procellariiformes species as well as getting lodged in the ventriculus making ingestion of other food difficult (Colabuono et al. 2009)

Procellariiformes are not the only birds affected by marine micro-plastics. The most common sea bird in Puget Sound, Glaucous-winged Gulls, were examined for consumption of marine micro-plastics by a team from the Port Townsend Marine Science Center. 12% of the boluses examined contained plastics. The most common type of plastic found within the Gulls bolus was films, which were similar to disposable plastic bags and wrappers (Lindborg et al. 2012).

With many species on the lower levels of the food chain thought to be consuming plastics, the ingestions of those organisms by predators further up the food chain can be a mechanism by which higher trophic level species ingest plastics. Many micro-plastic studies hint to the idea of micro-plastics making their way up trophic levels, but do not collect samples to support their hypothesis.

Eriksson and Burton (2003) is the main study that found micro-plastic has the potential to move up trophic levels through ingestion. Scat samples from the carnivorous fur seals on Macquarie Island were collected from 1991 and 1997. In the laboratory they

found that 4% of the scat samples collected contained pieces of micro-plastic. Fur seals eat a variety of fish including the most abundant species of fish, myctophids. Eriksson and Burton believe that the plastic particles were consumed by the myctophid species *E. subaspera* which in turn was eaten by the fur seals (Eriksson and Burton 2003).

Indirect Impacts (Changes to the Marine Ecosystems)

The most commonly used and produced plastics have densities that are lighter than sea water (Morét-Ferguson et al. 2010) and have the potential to float in the ocean over long distances. This allows locations that have had no inputs of marine plastics to be invaded by foreign marine debris and the associated organisms that have attached themselves along the way. Both macro-plastics and micro-plastics have been implicated in changing the marine environment.

Pieces of plastic regardless of size are commonly colonized by many different species of barnacles, tubeworms, foraminifera, coralline algae, hydroids and bivalve mollusks. Large pieces of floating plastic such as polypropylene rope can provide cover for planktonic organisms similar to Sargassum (Gregory 2009).

Pieces of plastic have been found all over the world from remote parts of Antarctica to isolated islands in the south Pacific. Barnes (2002) explores the transport of invasive species to some of the last pristine places on Earth. Barnes found that plastic marine debris was washing up in many places but the biotic communities on the foreign marine debris were still limited by environmental conditions such as temperature in the Arctic (Barnes 2002). He concluded that with changes in climate patterns these barriers

could be weakened and invasions of biota could occur on plastic debris to some of the last pristine places on Earth in the near future.

Changes in abundance of floating substrate available in the oceans due to increased micro-plastic abundance has also allowed marine organisms that depend on substrate for breeding, to increase in population. Goldstein et al. (2012) found that the *Halobates* population in the North Pacific is increasing and they attribute that rise to the increased in marine micro-plastic (Goldstein et al. 2012). Whether micro-plastic is ingested by zooplankton, a seabird or a marine mammal it has the potential to impact the lifecycle of that organism and thus, the entire marine ecosystem. The introduction of foreign species by micro-plastics has the potential to be devastating to ecological communities. Micro-plastics are another anthropogenic threat to the marine ecosystem.

Micro-plastic Sinks and Sampling Methods Development

The main findings from the 2008 *International Research Workshop on the Occurrence, Effects, and Fate of Microplastic Marine Debris* were that, “Data that conclusively demonstrate negative impacts of micro-plastics on the marine environment are not available. This is probably the largest and most critical gap to fill. Research into collection methods, species impacts, and removal methods should focus on potential micro-plastics hotspots” (Arthur et al. 2009). Thus, interest is high in assessing where micro-plastics are accumulating, with an emphasis on shoreline locations.

There are four locations that micro-plastics are believed to be accumulating. The sea surface, open ocean floor, near coastal sediment and beaches (Ryan et al. 2009a). Because shorelines are the most accessible sink locations, beach surveys have been

conducted more often than the other locations (Browne et al. 2010; Browne et al. 2011; Claessens et al. 2011; Cooper and Corcoran 2010; Corcoran et al. 2009; Costa et al. 2010; Eriksson et al. 2013; Frias et al. 2010; Ismail et al. 2009; Ivar do Sul et al. 2009; Kusui and Noda 2003; Martins and Sobral 2011; McDermid and McMullen 2004; Rees and Pond 1995; Rosevelt 2011; Ryan et al. 2009b; Van et al. 2012; Velander and Mocogni 1999; Zurcher 2009).

The majority of shoreline sampling for micro-plastics has occurred along sandy shorelines. The highest tide line of a beach is the most common place for sampling to occur because the debris deposited is likely from the most recent tide (Martins and Sobral 2011; Velander and Mocogni 1999).

Summary of Past Buried Marine Micro-plastic Shoreline Studies

A protocol has been adopted by a few studies examining buried marine micro-plastics. One m² quadrats are placed along the highest tide line, sediment samples are taken from the top 2 cm of the beach. These samples are processed using saltwater to float out the micro-plastics which have low specific densities. The micro-plastics are identified visually or with the aid of a dissecting microscope. The use of this similar protocol has allowed comparisons of micro-plastic density along shorelines across the world (Claessens et al. 2011; Costa et al., 2010; Ivar do Sul et al. 2009; Kingfisher, 2011; Kusui & Noda, 2003).

Density and distribution in previous shoreline studies

Results from these previous studies demonstrate great spatial variability. The densities range from 2610 pieces of micro-plastic per m² along Japan's coast to 9 pieces micro-plastic per m² along the shorelines of the island Fernando de Noronha in the Equatorial Western Atlantic (Ivar do Sul et al. 2009; Kusui & Noda, 2003). These studies further illustrate that marine micro-plastic pollution placed in one location has the potential to be not only a local problem but a global one as well.

The spatial variability in types of plastics is different across the global locations that have been surveyed using the similar protocol as well. This indicates that people are using and discarding plastics in different ways in different parts of the world. The samples from north Puget Sound and Japan have higher abundance of foamed plastics. This could suggest that an activity associated with foamed plastics is performed more often near shorelines than along the central California Coast where an even split of polystyrene and other plastic pieces is found (Kingfisher 2011; Kusui and Noda 2003; Rosevelt 2011).

The most common micro-plastic found in the Puget Sound is polystyrene followed by a much lower concentration of plastic fragments and very few virgin pellets (Kingfisher 2011). On Russian and Japanese shorelines 87.1% of the plastic found was polystyrene followed by 10.6% plastic fragments and 1.8% plastic pellets (Kusui and Noda 2003).

In Belgium and Fernando de Noronha, very low numbers of polystyrene were found but high numbers of plastic fragments and pellets were observed (Claessens et al. 2011; Ivar do Sul et al. 2009). This suggests that people may be discarding larger pieces

of plastic into the marine environment, an alternative hypothesis is that both locations perform as sinks for micro-plastics coming from the Atlantic Gyres.

The factors that drive that spatial variability are a combination of physical and anthropogenic drivers working in concert to create the spatial variability across locations as well as the variability in the types of plastics that are collected.

Theories for Explaining Marine Micro-plastic Spatial Distribution

Spatial variability is a common theme in published micro-plastic studies, both documenting variability between the studies and variability within the studies themselves. Physical and anthropogenic factors are attributed as explanations for locations having different densities of micro-plastics. Physical factors used to explain the variability include the effect of wind, global currents, local currents and tidal currents.

Anthropogenic factors used to explain the variability include population density around the study area and proximity to recreational use. The presence of industrial areas is described in studies but no quantification is conducted to measure the effect on micro-plastic density. No studies have examined the affect that impervious surfaces have on micro-plastic density.

Population density

People are responsible for the inputs of marine micro-plastics. Examining the relationship between population density and micro-plastic density can provide evidence to guide clean-up efforts. Population density as an indicator of micro-plastics present was used by Browne et al. (2011). In the 18 locations sampled worldwide, a positive

correlation was found between population density around the sample location and the quantity of micro-plastics that were discovered (Browne et al. 2011). The study only examined one sample from each location and did not look at any local variability. The initial findings display that population density could be an explanatory variable in the spatial distribution of micro-plastics but local spatial variability was not taken into account.

Population density was also used as a possible explanatory variable for the spatial distribution of micro-plastics found at different shoreline locations along the Japanese and Russian coast. Kusui & Noda (2003) found that micro-plastic density was higher along the Japanese coast where the population was much higher. They found that foamed polystyrene was much higher along Japanese shorelines and was responsible for the majority of the differences in micro-plastic density between the two coasts.

In north Puget Sound, there was a very weak negative correlation between the number of plastic items collected and the distance from a population center (Kingfisher 2011). The weak correlation could be because other factors were not eliminated first when selecting site locations such as isolation and circulation. Surface water in the North Puget Sound has a very short residence time, 20 days or less, suggesting that locations where micro-plastic inputs occurred were different from where accumulation zones were located due to wind and tidal currents dispersing the micro-plastics.

Proximity to Recreational Use

Claessens et al. (2011) found that shoreline study locations within an enclosed inner harbor where 2000 moorings were located had the highest density of micro-plastics

within their study area as compared to shoreline study locations located along the open ocean. The geographic characteristics of the inner harbor being confined was also thought to be a cause of the higher density found in the inner harbor (Claessens et al. 2011). Ismail et al. (2009) also found that the quantity of plastic pellets along the shorelines in Malaysia were higher in areas that were used for recreation (Ismail et al. 2009). The increase was very slight, no statistical tests were performed and the sample size was small, but the pattern was visible.

Rosevelt et al. (2013) found that no relationship between accessibility and the density of plastic debris that was found on beaches in Monterey Bay California. The authors believe that other factors such as currents and wind could be driving the density along beaches in Monterey Bay (Rosevelt et al. 2013). In the tidal estuary Firth of Forth located in Scotland, the density of plastic debris was also not dependent on the accessibility of the beach itself (Storrier et al. 2007). Storrier et al. (2007) believe that storm conditions combined with tidal current patterns are the most likely explanation for areas of increased density on shorelines in the estuary.

Only at locations where physical geography creates a way to keep micro-plastics near their area of input can the influences of anthropogenic uses be seen. Because anthropogenic influences are responsible for the inputs of marine micro-plastics to the marine environment the simple process of tracking them becomes much harder. Once micro-plastics have entered the marine environment wind and currents disperse them creating the spatial variability that is seen worldwide.

Wind Driven

Browne et al. (2010) found that the location of the site in relationship to the prevailing wind had an effect on the quantity of micro-plastics. Locations that were downwind had higher levels of micro-plastics than locations that were up wind. Dense micro-plastic was found at higher quantities than less dense and expanded polystyrene foam at the downwind locations. The study was conducted along the Tamar Estuary in the United Kingdom, where the combination of strong winds and flat shorelines are believed to be one explanatory variable for the low abundance of foamed polystyrene which is less dense. It is believed that the wind blows the foam polystyrene out of the water, up the shorelines and off the beaches into the near shore environment. The near shore environment was not sampled, so this remains a theory to explain the low numbers of foamed polystyrene at this time.

In the Arctic and the middle of the Equatorial Atlantic a beaches aspect in relation to the prevailing wind has been demonstrated as an explanatory variable of the spatial distribution found within local study locations. On the Fernando de Noronha Archipelago, in the western side of the South Atlantic, beaches on the eastern side of the islands had significantly higher densities of micro-plastics (Ivar do Sul et al. 2009). The prevailing winds around the Fernando de Noronha Archipelago, blow from east to west. Eriksson et al. (2013) conducted daily surveys of two islands in the Antarctic. Marine debris accumulation was greater on the western side of the islands which both intercept the Antarctic circumpolar current (Eriksson et al. 2013).

Ivar do Sul et al. (2009) hypothesize that the higher density of micro-plastics on the eastern beaches could be explained by the beaches interception of the prevailing

wind. Ivar do Sul et al. (2009) also believe that the location of the Fernando de Noronha Archipelago in the path of the South Atlantic Ocean anticlockwise gyre could be an alternative cause of the higher micro-plastic concentrations on the eastern side of the archipelago (Ivar do Sul et al. 2009). The location of a shoreline in relation to the established currents has also been demonstrated as a driver for spatial variability of marine micro-plastics on shorelines worldwide.

Surface Current Based Studies

Surface currents carry large amounts of water around the world. The global surface current pattern is clockwise in the northern hemisphere and counter-clockwise in the southern hemisphere (Figure 1). These surface currents carry any floating debris placed in the water. For example, a piece of plastic washed into the ocean from an individual in San Diego can be carried by the California Current south and eventually by the North Equatorial Current to be washed on the eastern shoreline of Kauai.

Heavier accumulations of micro-plastics were found on the eastern side of Kauai Island in Hawaii that were influenced by the current from the North Pacific sub-tropical Gyre and currents traveling around the island (Cooper and Corcoran 2010; Corcoran et al. 2009).

Martins & Sobral (2011) found the location and the orientation of the beaches sampled in Portugal to be the cause of the extremely high density of micro-plastics. The north orientation of beaches places them in the path of the North Atlantic Gyre, which is known for high densities of micro-plastics and the authors believe promote higher rates of accumulation (Martins and Sobral 2011).

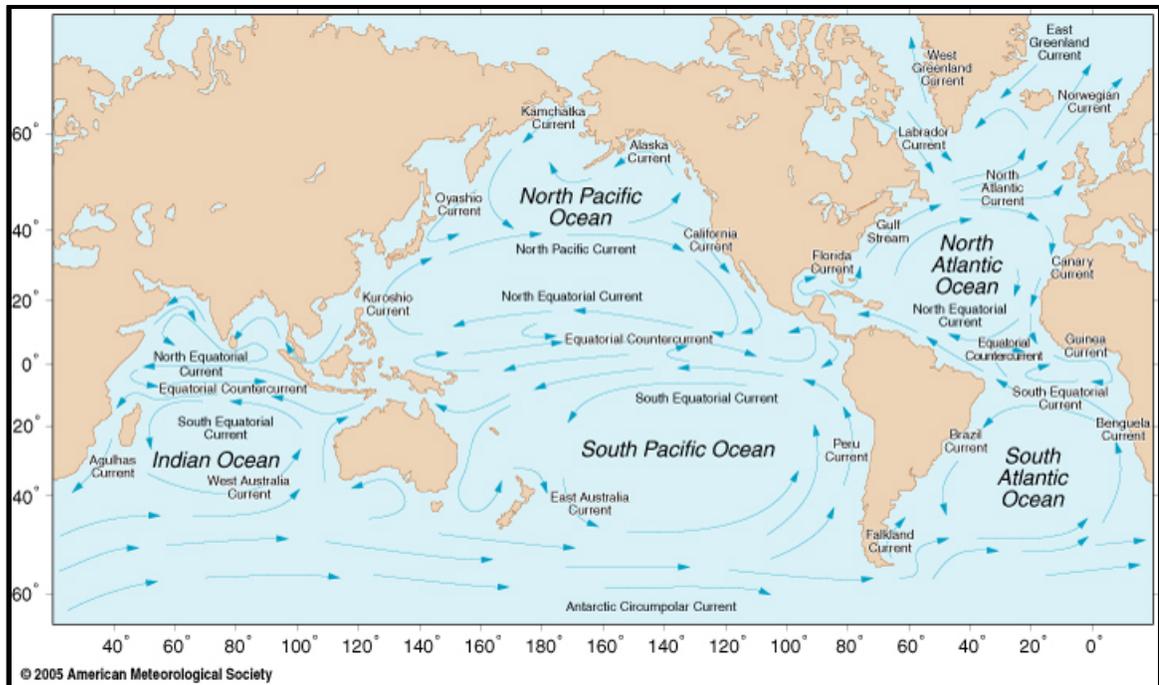


Figure 1. Global wind driven surface currents taken from NASA’s Ocean Motion and surface currents (American Meteorological Society 2005).

These studies demonstrate the importance of understanding the physical factors that drive the surface movement of the water, be that by the wind or currents. Locations that intercept the prevailing wind or currents coming from areas with high anthropogenic influence will likely have higher densities of micro-plastics on their shorelines.

Previous Studies Examining Marine Micro-plastic Spatial Distribution Summary

Examination of the previous results indicates that more than one factor is driving the variability found within each study. Utilizing a combination of factors including wind, currents, proximity to areas of high population and recreational use is believed to be the best explanation of the spatial variability in micro-plastic density within studies. Understanding the physical geography of the study location, the prevailing wind and the currents within the study area is key to understanding the local spatial variability. Once

these factors have been understood the larger issues of anthropogenic influences can be examined and sources of micro-plastics be traced back to their sources via the physical factors.

Thus, South Puget Sound is an excellent location to study the combination of anthropogenic and physical factors driving the spatial distribution of marine micro-plastics. South Puget Sound is the furthest point away from the entrance of Puget Sound. South Puget Sound's isolated location, similar wind and current patterns in combination with differences in population between the smaller finger inlets located in the most southerly section of South Puget Sound, create conditions in the field that are very close to a controlled experiment and allow exploration of the drivers of spatial distribution of marine micro-plastics and establish baseline data to track if micro-plastics are increasing over time locally. Thus, it advances the scholarly literature by creating a baseline from South Puget Sound and identifies the primary physical and anthropogenic drivers of marine micro-plastics spatial distribution.

Conclusion

Plastic production is believed to continue to increase (Plastics Europe 2012). The persistence of micro-plastics in the marine environment is not yet understood, but the estimates are in the range of tens to hundreds of years (Andrady 2005; Barnes et al. 2009). Once these plastics enter the marine environment they begin to slowly break down into smaller pieces. These small pieces pose a threat to marine organisms from ingestion, transport of toxic chemicals and the transport of invasive species.

Micro-plastics are believed to be accumulating in three locations, the open ocean floor, near coastal sediment and shorelines (Ryan et al. 2009b). The most accessible locations that micro-plastics are believed to be accumulating are shorelines. Thus shoreline surveys have been a popular way to establish baseline data for micro-plastic densities. Local and global spatial variability along shorelines has been documented.

The localized spatial variability is believed to be driven by prevailing winds and currents, while global trends in spatial variability are attributed to anthropogenic influences such as population density or levels of recreational use in a region. Understanding the local spatial variability allows a clearer picture to be drawn when establishing baseline data for marine micro-plastic densities. Obtaining clear baseline data allows the exploration of these areas again as the growth of plastic manufacturing and consumption of plastic products continues to grow globally.

METHODS

This study used an established protocol for sampling marine micro-plastics in sediment along shorelines, to investigate the spatial distribution of marine micro-plastics in South Puget Sound, Washington USA. This protocol has been implemented locally in North Puget Sound (Kingfisher, 2011) as well as internationally (Costa et al., 2010; Ivar do Sul et al., 2009; Kusui & Noda, 2003; Martins & Sobral, 2011), hence this protocol was selected for purposes of cross comparison.

This is the first study to examine marine micro-plastics in South Puget Sound. South Puget Sound is the furthest point away from the entrance of Puget Sound. This location was selected because South Puget Sound's isolated location, similar wind and current patterns in combination with differences in population between the smaller finger inlets located in the most southerly section of South Puget Sound, create conditions in the field that are very close to a controlled experiment and allow exploration of the drivers of spatial distribution of marine micro-plastics and establish baseline data to track if micro-plastics are increasing over time locally.

Overview of Puget Sound Circulation Patterns

Puget Sound is the second largest estuary in the United States with approximately 2,500 miles of shoreline. The net flow within the Puget Sound is towards the entrance at the Strait of Juan de Fuca, with this flow being driven by surface freshwater inputs from rivers, creeks and groundwater runoff pushing the surface water toward the entrance. Sea water, which is denser, enters the system at depth running in the opposite direction and draws its source from the Pacific Ocean. The flow is further influenced by wind strength

and direction (Gaydos 2008). Driven by these variables, the residence time of water in the entire Puget Sound Basin is approximately 90 days, but it ranges from 20 to 120 days in the different Puget Sound basins.

This circulation pattern is further modified by three underwater sills, left by glaciers during the last ice-age. The sills are located at the head of Admiralty Inlet to the North, the mouth of Hood Canal to the west and the Tacoma narrows to the south (Gaydos 2008).

Four distinct water bodies are created by the sills in Puget Sound: Hood Canal, North Puget Sound, the Main Basin and South Puget Sound (Figure 2).

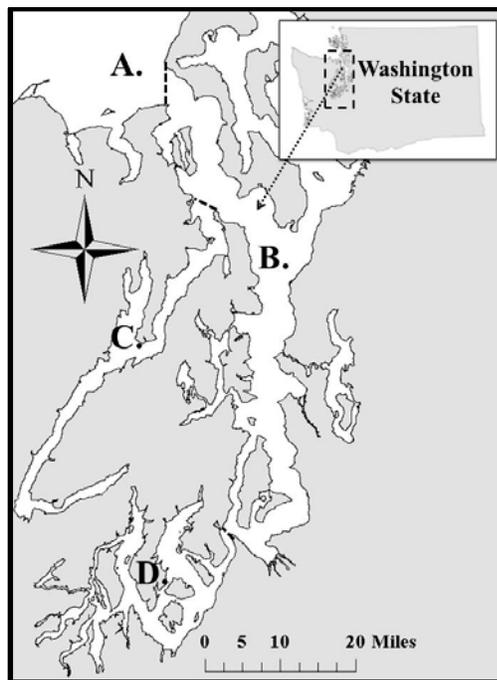


Figure 2. Four main water bodies created by underwater sills in Puget Sound, Washington State USA. **A.** North Puget Sound is the water to the north of the sill at Admiralty Inlet; **B.** The Main Basin is the largest water body in Puget Sound; **C.** Hood Canal is the water to the southwest of the sill at the mouth of Hood Canal; **D.** and South Puget Sound is the water body south of the Tacoma Narrows. Dotted lines indicate underwater sills.

North Puget Sound is the water body north of the sill at Admiralty Inlet, North Puget Sound is closest to the ocean and contains a mix of high and low population

density. Central Puget Sound also referred to as the Main Basin is the water body south of Admiralty Inlet and north of the sill at the Tacoma Narrows. The Main Basin of Puget Sound has the highest population density and two major cities are located on its east shorelines, Seattle and Tacoma. The population density for the east side of the Main Basin is 2,924 people/ km² (United States Census Bureau 2013).

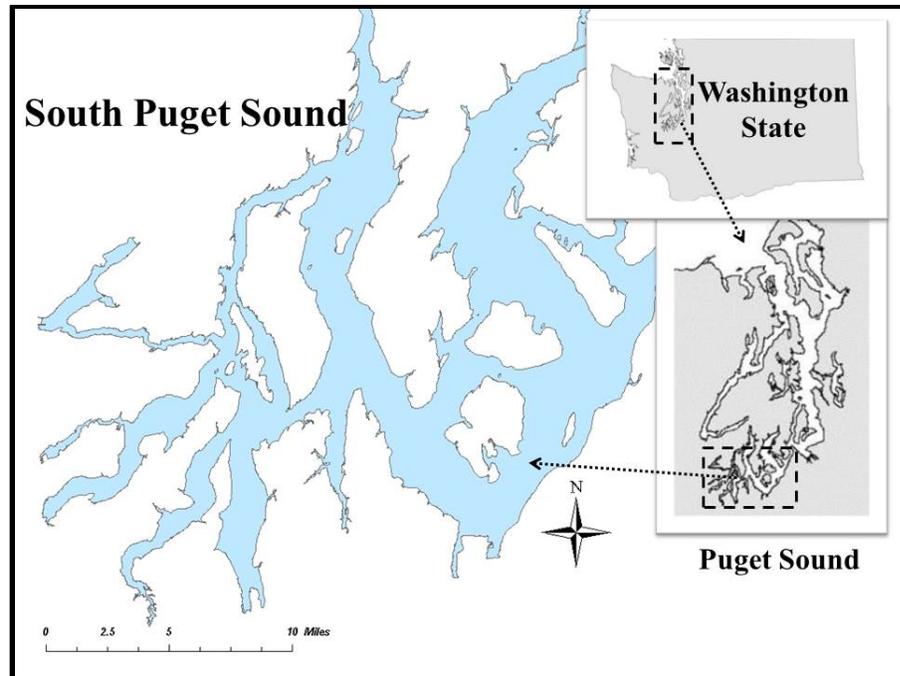


Figure 3. South Puget Sound, Washington State USA.

Finally, South Puget Sound, which is the basin examined in this study, is the water body south of the Tacoma Narrows. This basin is a complex and interconnected system of straits and fjord-like bays (Albertson et al. 2007) (Figure 3). South Puget Sound's location as the furthest basin from the ocean creates the greatest tidal range, 14.4 feet on average, in the Puget Sound (Ebbesmeyer et al. 1998). South Puget Sound is less densely populated than the Main Basin, with 1,056 people/ km² residing in the most populated area at the south end of Budd Inlet (Thurston Regional Planning Council 2012; United States Census Bureau 2013).

Description of study sites within South Puget Sound Basin

Within South Puget Sound the three most southerly inlets, Budd, Eld and Totten, were selected as study locations (Figure 4). These three inlets were selected as study locations because of their similar geographic locations, differences in population, and differences in land use. They are the furthest points from the Pacific Ocean in Puget Sound allowing the exploration of the influence that localized wind and current patterns have on the spatial distribution of micro-plastics. The differences in population density and differences in land use between the inlets allow the exploration of the influence that population density and impervious surfaces have on the spatial distribution of micro-plastics as well.

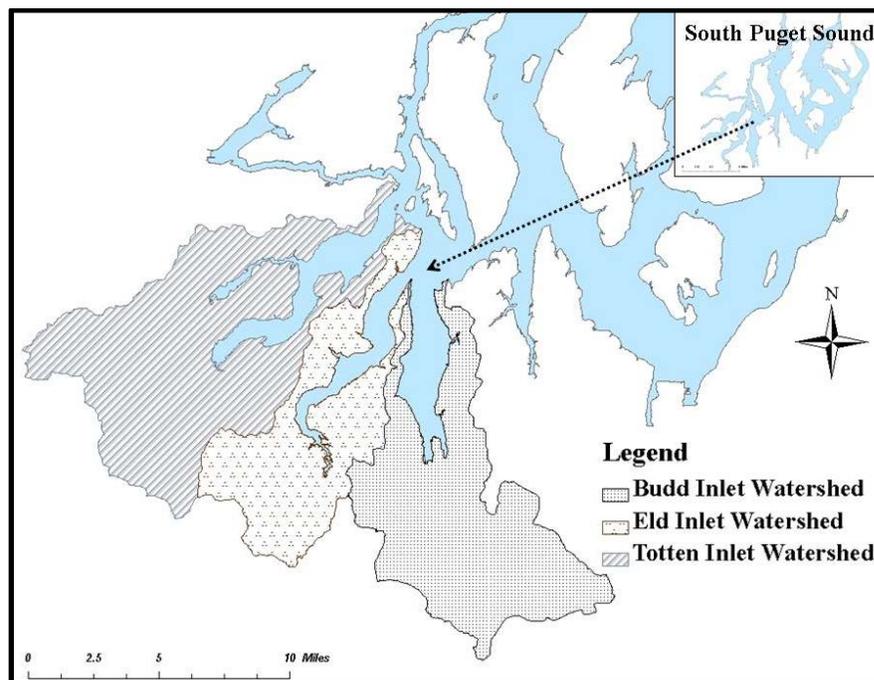


Figure 4. Budd, Eld and Totten Inlets located in South Puget Sound, Washington State USA.

Budd Inlet is the most populated watershed in South Puget Sound. As of 2010, the population density within the Budd Inlet watershed is approximately 1186 people/ km²

(U.S. Department of Commerce 2010). The land within the Budd Inlet watershed had an estimated 8% of impervious surfaces as of 2005 (Thurston County Regional Planning Council 2006). The residence time of water in South Puget Sound Basin is approximately 8-12 days (LOTT 2000). Budd Inlet has been the most studied inlet of the three finger inlets due to the metropolitan center of Olympia, being located at the south end of the inlet. The surface circulation and currents were studied as part of the recertification project for the Lacey, Olympia, Tumwater and Thurston County sewage treatment plant during 1996-1997 (Aura Nova Consultants et al. 1998) (see below).

The greatest percentage of population and impervious surfaces are located at the south end of Budd Inlet. The rest of the inlet is lightly populated and with much less impervious surfaces (Thurston Regional Planning Council 2012). All of the inlets are similar in their distribution of slightly denser populations along the shorelines that in the interior of the watersheds.

The Eld Inlet watershed has less people than the Budd Inlet watershed, with 267 people/ kilometer² and an estimated 4% of the land is covered with impervious surfaces (Thurston County Regional Planning Council 2006; U.S. Department of Commerce 2010). The residence time of water in Eld Inlet is not known.

The Totten Inlet watershed is the least populated and has the least amount of impervious surfaces. The Totten Inlet watershed has a population density of approximately 84 people/ kilometer² and an estimated 2% of the land is covered with impervious surfaces (Thurston County Regional Planning Council 2006; U.S. Department of Commerce 2010) The residence time of water in Totten Inlet is not known.

Factors affecting Circulation in South Puget Sound

The water that enters South Puget Sound, and consequently Budd, Eld and Totten inlets, comes from two sources. Dense salt water enters at depth through the Tacoma narrows at the northern tip of South Puget Sound. Annually the approximate rain fall is 50 inches in the South Puget Sound region (United States Naval Research Laboratory 2008). This less dense freshwater enters the South Puget Sound from two freshwater rivers fed by the Cascade Mountains, the Nisqually and Deschutes. Precipitation falls an average of 185 days a year, allowing freshwater to enter South Puget Sound directly, and as storm water runoff.

Though freshwater inputs contribute freshwater into the system, the circulation in Puget Sound, and consequently South Puget Sound is driven by tidal pumping. Tidal pumping is the movement of water created when the tide moves over and through changes in bathymetry (Gaydos 2008). The complex and interconnected system of straits and fjord-like bays in combination with the single entrance and exit at the the Tacoma Narrows makes the residence time of water in South Puget Sound Basin the longest in Puget Sound at approximately 120 days (LOTT 2000).

The prevailing wind in South Puget Sound is from the southwest but wind patterns are affected locally by the interactions with the main flow and local topography (United States Naval Research Laboratory 2008). This southwest wind is a product of the wind flowing around the southern end of the Olympic Mountains and up through the Chehalis River Valley (United States Naval Research Laboratory 2008).

All inlets share the same input of salt water coming from Central Puget Sound through the Tacoma Narrows. The tidal flow of salt water enters Budd Inlet on the west

side and proceeds to the south. The central gyre in the inlet rotates counter-clockwise and the water exits along the eastern side of the inlet (LOTT 2000; McGardy and Lincoln 1977) (Figure 5).

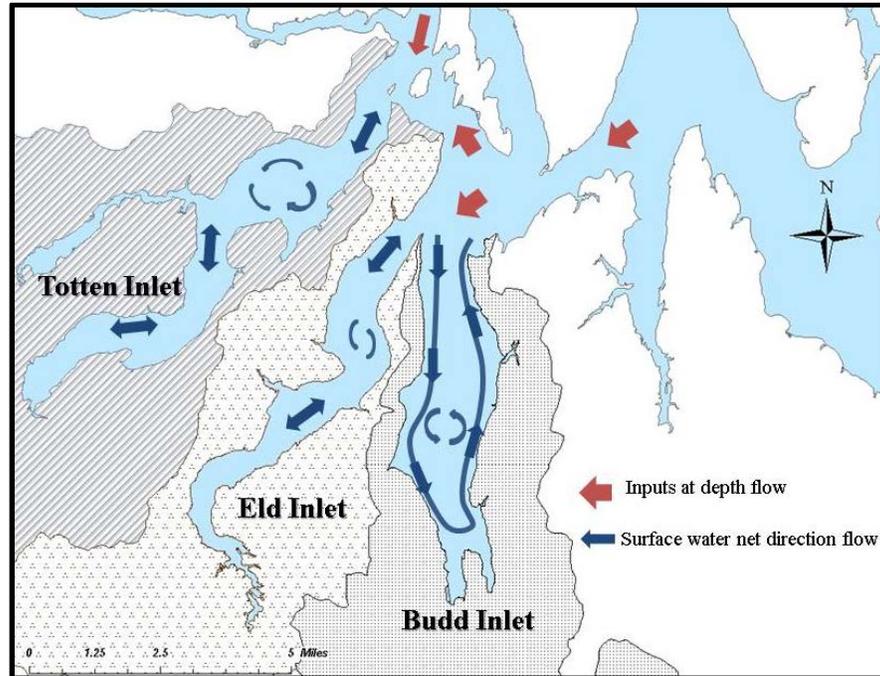


Figure 5. Tidal current circulation patterns for Budd, Eld and Totten Inlets located in South Puget Sound, Washington State USA. Budd Inlet circulation based on (LOTT 2000), Eld and Totten Inlet circulation based on (McGardy and Lincoln 1977).

The tidal currents in Eld and Totten Inlets have only been briefly studied. The main study of tidal currents in South Puget Sound was conducted using the University of Washington's Puget Sound model, using pieces of Styrofoam and a film camera (McGardy and Lincoln 1977). The general circulation patterns were identified but the detail that is available in Budd Inlet is not available for Eld or Totten Inlet.

Surface Circulation Patterns in South Puget Sound

The previous section described tidal circulation patterns at depth. However, surface circulation may differ substantially from patterns observed at depth. Much of what is known about circulation patterns in the South Puget Sound is from work conducted by Aura Nova Consultants to evaluate the effects of increasing the capacity of the sewage treatment plant located at the south end of Budd Inlet.

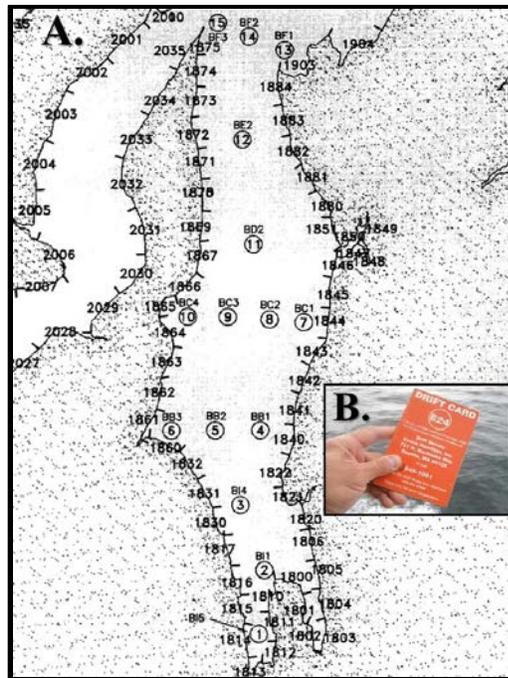


Figure 6. A. The fifteen drop locations drift cards were released in Budd inlet from October 1996 to September 1997 taken from (Ebbesmeyer et al. 1998). **B.** Figure of a drift card used by the Aura Nova Consultants in South Puget Sound taken from (Albertson et al. 2007)

Briefly, a drift card study was used to understand the surface circulation patterns in South Puget Sound. From October 1996 to September 1997, 8,950 drift cards were deployed by dropping them in 15 locations within Budd Inlet (Figure 6). These drift cards are wooden, measure approximately 3" x 5", and were coated with orange, non-toxic paint to render them readily visible to beachcombers. Each card carried a serial number preceded by 'L' for LOTT, an address, and a 1-800 telephone number enabling

beachcombers to report recoveries. The recovery reports were tabulated within 1-mi shoreline segments because most of the cards were found along short stretches of beach.

Fifty one percent of the drift cards were recovered, some were found as far as Alaska (LOTT 2000). Of the recovered cards 31% were found within Budd Inlet suggesting that not everything placed into the inlet will be washed out of the inlet (Albertson et al. 2007). Less than 2% of the drift cards released in Budd Inlet were recovered from the closest adjacent inlets, Eld and Totten (Figure 7).

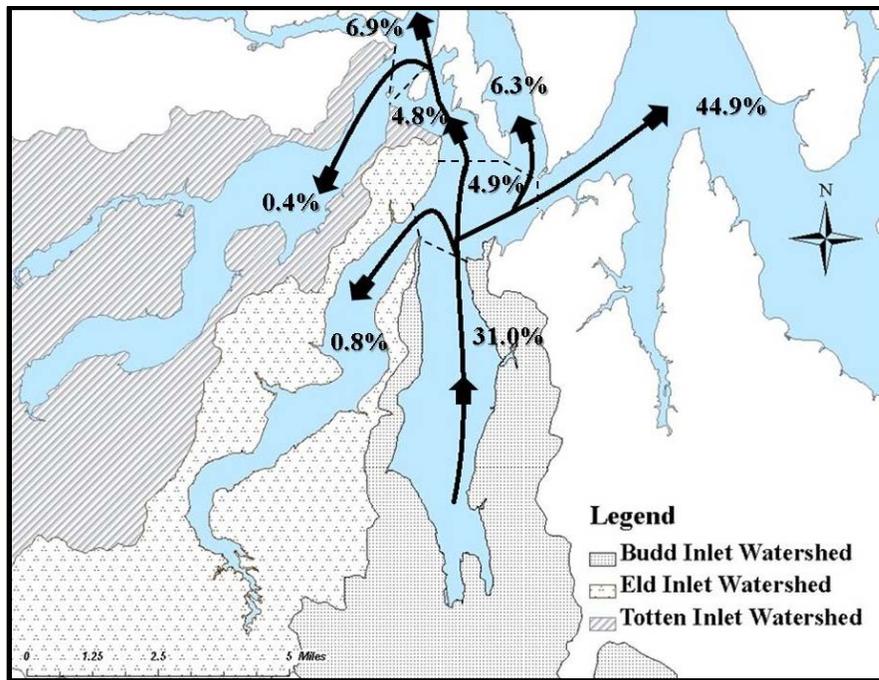


Figure 7. Drift card pathways from Budd Inlet. Dotted lines represent divisions between water bodies in South Puget Sound. Arrows represent drift card pathways and direction. Percentages shown are based on the total number of cards recovered within each water body from those released within Budd Inlet from October 1996 to September 1997. Data taken from (Ebbesmeyer et al. 1998).

Aura Nova Consultants conducted a secondary drift card study to ensure that circulation patterns were responsible for the low recovery rates of drift cards from Eld and Totten Inlets. In January, 1997 drift cards were released in Eld and Totten Inlets as well as Budd Inlet by small plane (Figure 8). Similar detection rates (51.5%) were found for the entire study area providing evidence that recovery rates were driven by the tidal currents. This also demonstrated that the net direction of floating debris deposited within the finger inlets is towards the ocean.

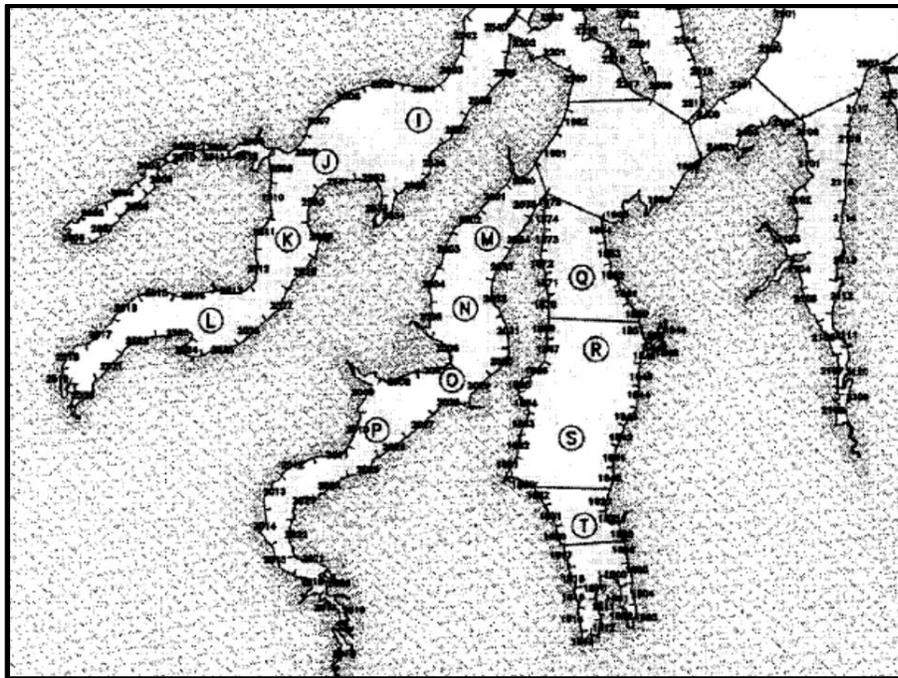


Figure 8. Drift card air drop drift card release sites. Twelve locations where drift cards were released from a small airplane on January 22, 1997: site I-L in Totten Inlet; sites M-P in Eld Inlet; and sites Q-T in Budd Inlet. Figure taken from (Ebbesmeyer et al. 1998).

Further examination of the drift card data suggests that drift cards accumulate in certain regions of each inlet. Accumulation zones were identified as regions within each inlet where a high quantity of drift cards were recovered. Non-accumulation zones, locations where little to no drift cards were recovered, were identified from the drift card data as well (Figure 9).

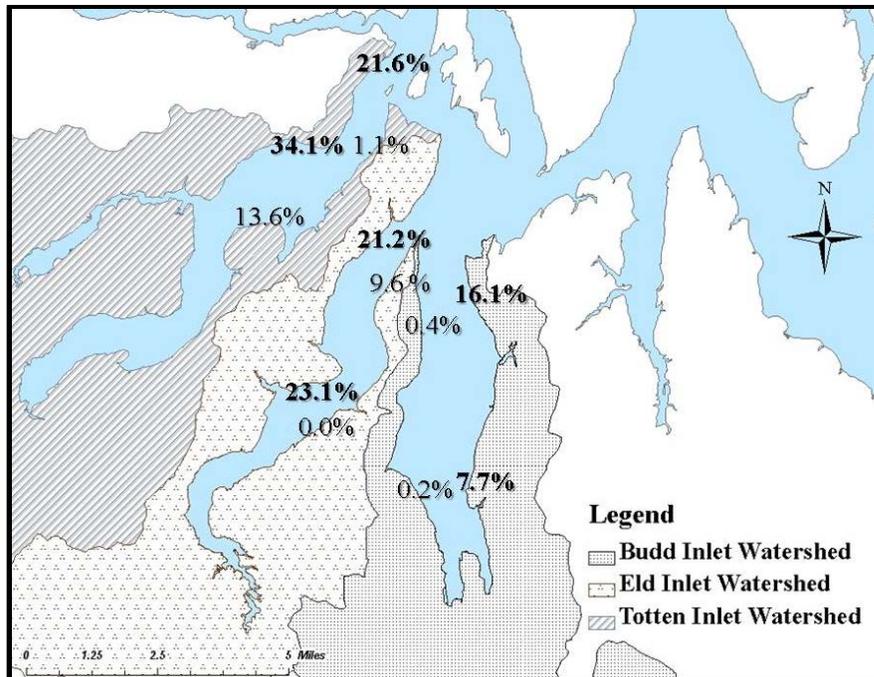


Figure 9. Accumulation and non-accumulation zones identified from the examining data collected for the recertification of Lacey, Olympia, Tumwater and Thurston County Clean Water Alliance’s waste water treatment plant, conducted by Aura Nova Consultants. Percentages represent the quantity of drift cards released in each inlet that were recovered at each location. **Bold** locations are accumulation zones. Data collected from (Aura Nova Consultants et al. 1998)

The lower percentages in Budd Inlet are due to the higher number (1,429) of drift cards recovered and released (8,950) over the period of one year (Ebbesmeyer et al. 1998). This created a more dispersed distribution of detection in relation to Eld and Totten Inlets where 200 drift cards were released once, hence a smaller proportion of drift cards (55 and 51 respectively) were collected to create higher relative percentages. Had the drift card study been conducted for a longer time period with more drift cards the results are believed by the author to more closely resemble Budd Inlet.

Site Selection

Twelve study sites were selected in the study area. Initial selection of two sites in each inlet was based on the identification of accumulation zones from the Aura Nova

Consultants data. Two more sites in each were selected on the opposite side of the inlet from the accumulation zones. The location of these sites was the same approximate distance from the mouth of the inlet. These locations are referred to as non-accumulation zones based on low detection rates of drift cards at these sites (Figure 10).

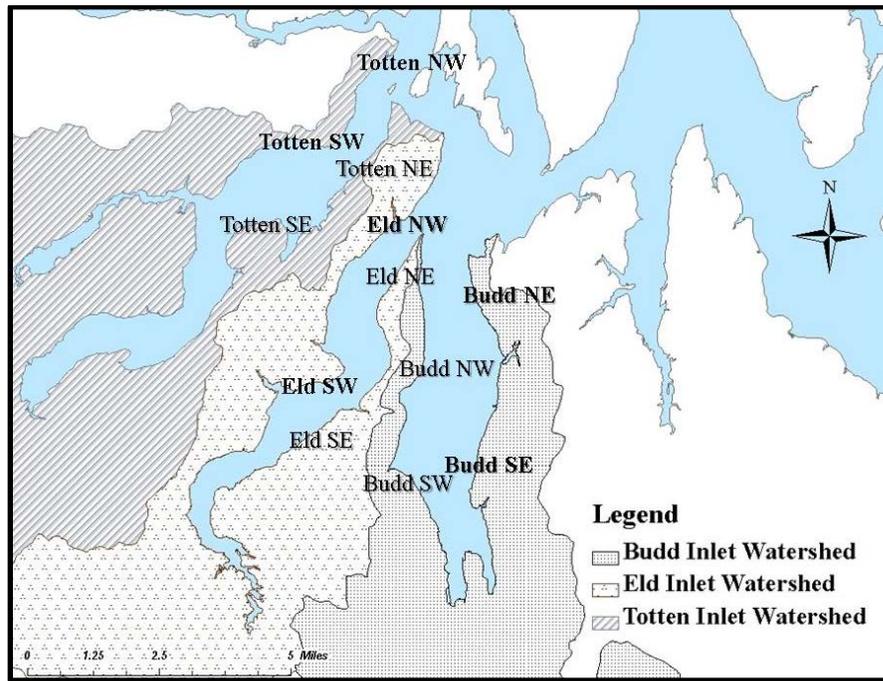


Figure 10. Twelve site locations in South Puget Sound. **Bold** locations are accumulation zones.

Field Methods

This study used an established protocol for sampling marine micro-plastics in sediment along shorelines, to investigate the spatial distribution of marine micro-plastics in South Puget Sound, Washington USA. This protocol has been implemented locally in North Puget Sound (Kingfisher 2011) as well as internationally (Costa et al. 2010; Ivar do Sul et al. 2009; Kusui and Noda 2003; Martins and Sobral 2011), hence it was selected for purposes of cross comparison.

The field survey was conducted from January 16th, 2013 to February 6th, 2013 (**Table 1**). Sampling along the highest, high tide line took place during the day after the most recent high tide had begun to recede. The survey purposely took place immediately following the highest high tide of the year, 16.9 feet which occurred on January 14th (Ecology 2013). The intention of this sampling design was to collect floating debris deposited at the same time, by the same high tide across all sites.

Table 1. Study site locations and Sampling Dates. Tide data from (NOAA Tides and Currents 2013).

<u><i>Inlet Sample Location</i></u>	<u><i>Sample Date</i></u>	<u><i>High Tide (Feet)</i></u>
Budd NE	January 16, 2013	16.51
Budd NW	January 22, 2013	13.21
Budd SE	January 16, 2013	16.51
Budd SW	January 29, 2013	15.8
Eld NE	January 23, 2013	13.18
Eld NW	January 22, 2013	13.21
Eld SE	January 23, 2013	13.18
Eld SW	January 29, 2013	15.8
Totten NE	February 1, 2013	15.94
Totten NW	February 6, 2013	14.43
Totten SE	February 6, 2013	14.43
Totten SW	February 1, 2013	15.94

At each site, ten quadrats were sampled along a one hundred meter transect parallel to the water's edge (Figure 11) and spaced 10 m apart from each other. Transects were placed along the highest high tide mark, the highest line of floating deposited material up the shoreline, as plastic is believed to accumulate in this zone (Martins and Sobral 2011).

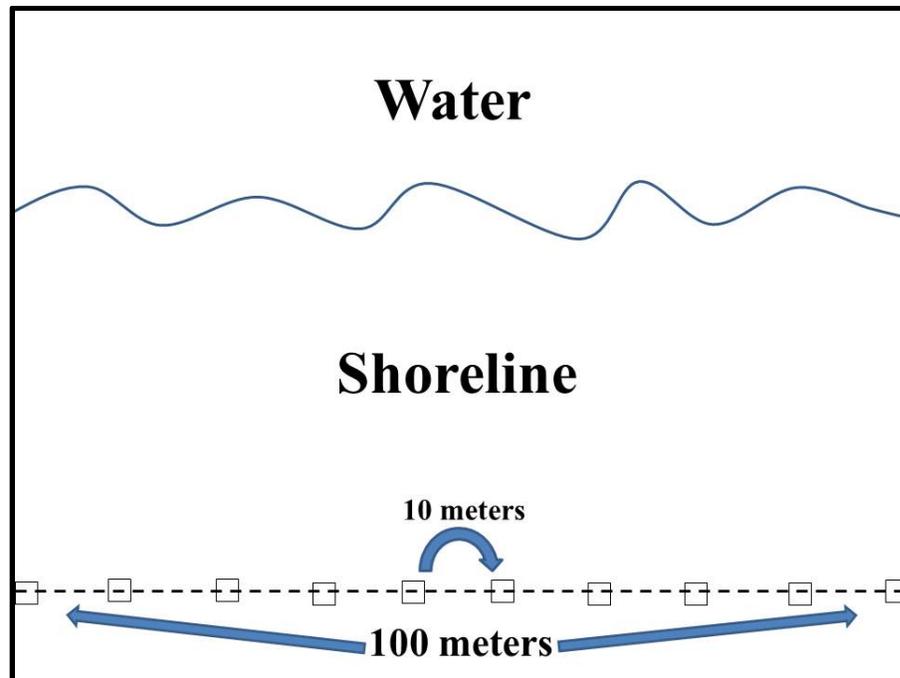


Figure 11. Transects were located along the highest high tide mark (upper wrack line) and were 100 meters in length. Each 0.5-m² quadrat was placed every 10 meters along the upper wrack line.

Within each 0.5-m² quadrat, all large rocks and woody debris were removed.

Sediment was then removed to a depth of 2cm, which was subsequently placed into a 3.8 liter bag to be processed in the laboratory.

Laboratory Methods

Micro-plastics have been recently defined as plastic that ranges from between 5 mm and 0.3 mm in diameter (Betts 2008), but historically definitions have ranged

between less than 20mm and .001 mm (Hidalgo-Ruz et al. 2012). For the purpose of this study, objects between 5.6 mm and 0.3 mm in diameter are considered micro-plastics. The 5.6mm sieve was selected due to limitations of available equipment at the time of sampling.

To isolate the micro-plastics from the sediment the beach samples were oven dried at 75°C for 24 hours and weighed using a balance to the nearest gram. The sample was then sieved, using a 5.6-mm sieve, and the portion of the sediment sample larger than 5.6-mm was discarded.

The remaining dry sediment was weighed using an analytical balance to the nearest gram. The sample was subjected to density separation using 500-750ml of 5 M of NaCl aqueous solution in a 1000ml beaker following the protocol of several previous studies including Claessens et al. (2011), Costa et al. (2010), Ivar do Sul et al. (2010), Kusui & Noda (2003) and Martins & Sobral (2011). The aqueous solution was vigorously mixed by hand using a metal spatula to float out any portion of the sample that had a low specific density.

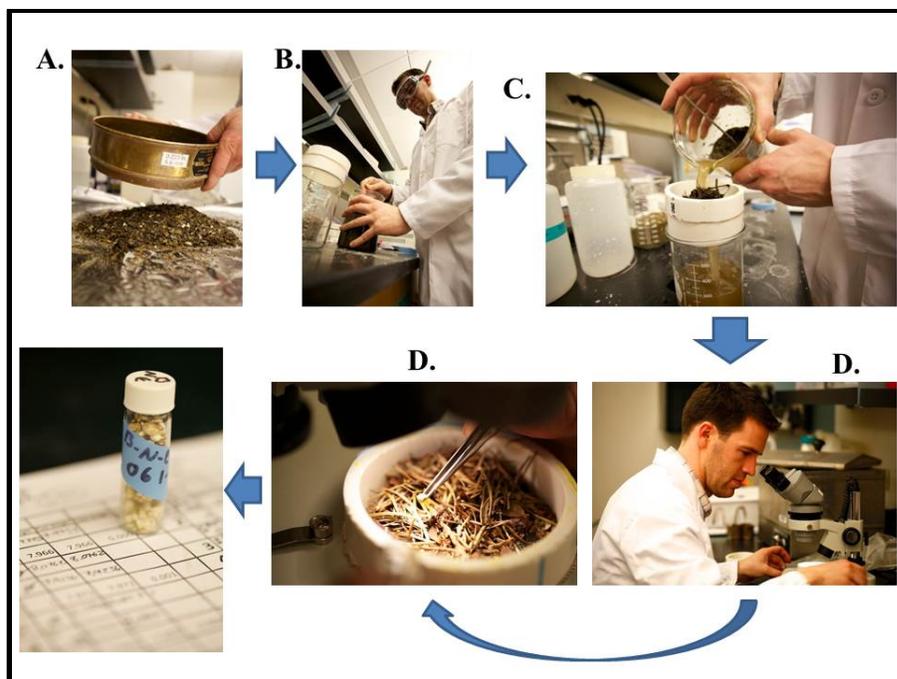


Figure 12. **A.** The dry sample was sieved using a 5.6mm sieve. **B.** The remaining sample was vigorously mixed in a 5M NaCl solution by hand using a metal spatula to separate floatable material **C.** The sample was decanted into stacked 1.0mm and 0.3mm sieves. **D.** The sample was dried at room temperature for 24 hours then identified by category using a dissecting micro-scope.

The sample was then decanted for floatable material and passed through stacked 1-mm and 0.3-mm sieves. These sieve size classes were selected to allow comparisons to be drawn with other Puget Sound studies (Baker et al. 2011). This process was repeated until no floatable material was visible at the surface, generally 3 to 4 times (Figure 12). The floatable material collected on the 1-mm and 0.3-mm sieves was air dried for 24 hours at room temperature. The floatable material collected on the sieves was visually inspected using a microscope under 40x magnification to identify the different types of the micro-plastic collected. The micro-plastic was collected using forceps and placed into a pre-weighed 4ml vial. The collected micro-plastics were weighed using an analytical balance to the nearest 0.001 mg.

For this thesis micro-plastics are identified as an object between 5.6 mm and 0.3 mm in diameter where no cellular or organic structures are visible. Types of micro-plastics were identified as follows: if fibrous the fibers should be equally thick throughout their entire length. Fibers, foamed particles and fragments are a single and homogeneous color (Hidalgo-Ruz et al. 2012). Recent studies have used five micro-plastic categories for identification which allow differences in anthropogenic uses of plastics to be compared around the globe. The five categories are; plastic fragments, foamed plastics, filaments (which are referred to as line balls in this study), films and pre-production pellets (also referred to as nurdles) (Figure 13). The first four categories of micro-plastics are created by the degradation of larger plastic items while the preproduction pellets are manufactured as micro-plastics.

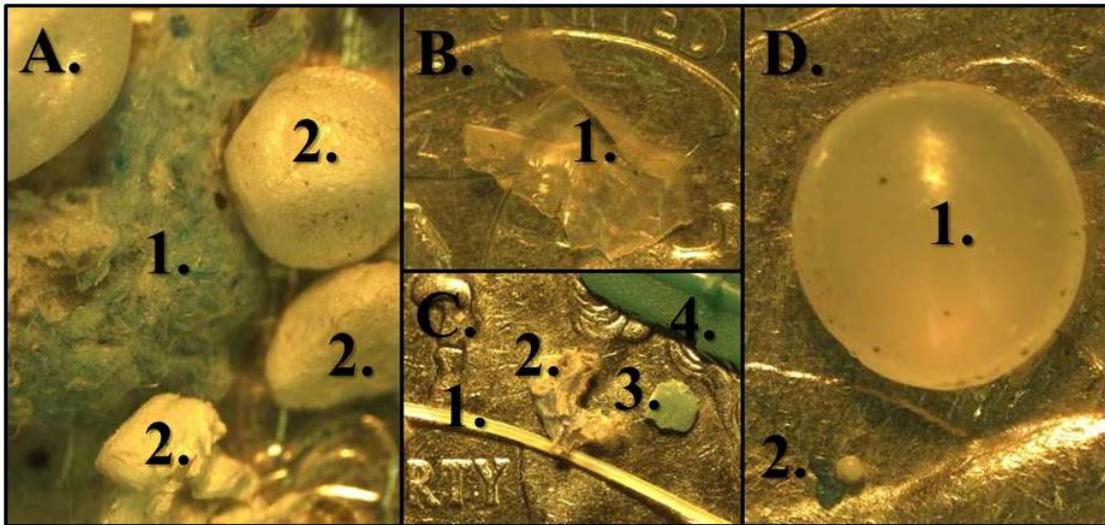


Figure 13. Micro-plastic categories. All pictures taken on quarter to give scale **A.** Five Foamed Plastics **A1.** Blue Foamed Plastic **A2.** White foamed plastics **B1.** Three Plastics Films **C.** Four Plastic Fragments **C1.** Fibrous plastic Fragment **C2.** Blue Plastic Fragment **C3.** Light Green Plastic Fragment **C4.** Green Plastic Fragment with little degradation present **D.** Four Pre-production plastic pellets (nurdles) **D1.** One Large Nurdle **D2.** Three small nurdles **E.** Line Balls (not pictured)

Data Analysis Methods

This study used GIS to determine the watershed boundaries of the three finger inlets and to determine the population density of Budd, Eld and Totten Inlets. ERSI ArcMap 10 and Microsoft Excel 2010 were used for the GIS analysis.

GIS Analysis

Watershed

Using National Watershed Boundary (WB) vector shape file data set for Washington State from the National Resource Conservation Service the watersheds of Budd, Eld and Totten Inlets were identified. The smaller watershed units within each inlet were merged to create a new ArcGIS layer for each inlet.

Population Density

2010 census block TIGER vector shape files were used for analysis, which were provided by the Washington State Office of Financial Management. The Tiger data was queried using select by location. Total population in watershed was calculated using the ArcGIS summary statistics tool. Total square kilometers of each inlet watershed was calculated using the measure tool. Population density was calculated using Microsoft Excel 2010, total population within the watershed was divided by the total square kilometers of the watershed.

Statistical Methods

Statistical analysis of the samples was conducted to establish if there was a difference in micro-plastics density between the three finger inlets. JMP Pro 10.0.2 statistical package was used. Summary statistics for micro-plastic density for the total

micro-plastics collected, large micro-plastics (5.6mm-1.0mm) and small micro-plastics (1.0mm-0.3mm) were conducted for each study site, each finger inlet and total study area. Data is presented in items/ m² and grams/ m² ± the standard error of the mean. Correlation between the large and small size classes were conducted to determine if the density of items (i.e., items or grams/ m²) was correlated between the size classes. Correlation was also performed on the relationship between population density of an inlet and the total micro-plastics items/ m² collected with the inlet.

The data was checked for normality using Shapiro-Wilk W test, for goodness of fit to the normal distribution. The data was not found to be normally distributed, so non-parametric analyses were conducted. Using the Wilcoxon test of rank sums the total density of total micro-plastics/ m² collected within the three finger inlets, the total density of micro-plastics/ m² for the suggested accumulation zone locations across the three finger inlets and the total density of micro-plastics/ m² from suggested non-accumulation zone locations in the three finger inlets were tested for statistical significance. These Wilcoxon tests of rank sums were conducted to determine if a statistical difference was present either between the inlets, between the accumulation zones or between the non-accumulation zones.

RESULTS

1,872 pieces of marine micro-plastic were collected over the 60 m² that was surveyed. The composite weighed 1.9 grams and was collected in 189,747 grams of sediment. There was great spatial variation in the density of items collected and the weight of the items collect across the study area. One location, Budd NE, contained the majority of the micro-plastic collected in the study area (Figure 14).

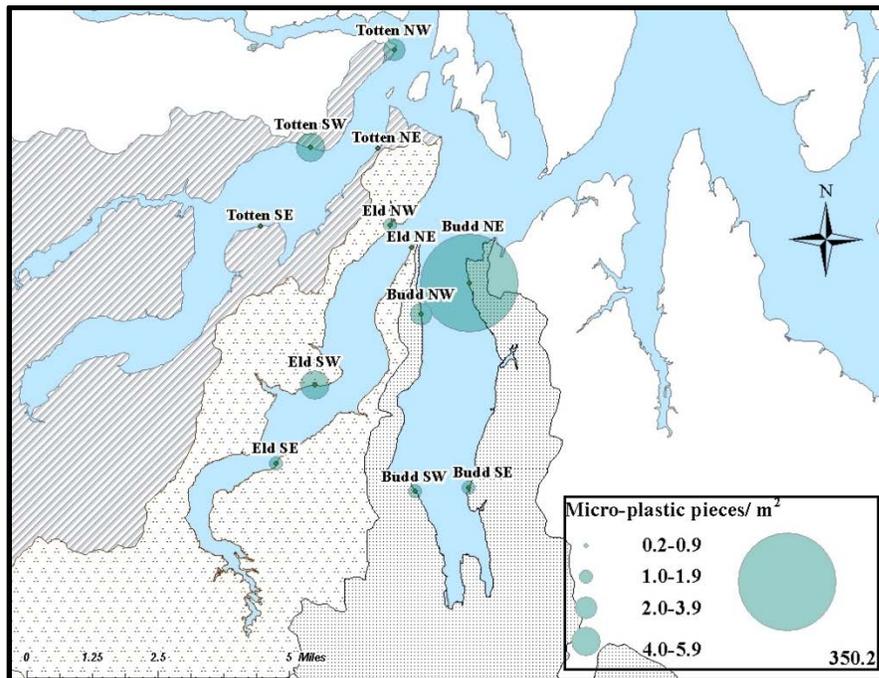


Figure 14. Map of spatial distribution of micro-plastic items/ m² collected from January 16th, 2013 to February 6th, 2013.

Overall

Overall Density of Items

An average of 31 ± 14 items/ m² (including all size classes examined during this study) was observed throughout the three finger inlets. Budd Inlet had the highest density of micro-plastics with 89 ± 40 items/ m². Eld and Totten Inlets had much lower densities with 2.1 ± 0.8 and 2.3 ± 1.1 items/ m² respectively (Table 2).

Table 2

Density of micro-plastic by various size classes (items/ m²) collected from January 16th, 2013 to February 6th, 2013.

Water Body Inlet Variable	Summary statistics		
	Mean	Std. Error of μ	Area Sampled (m ²)
All Finger Inlets			
Items (5.6 mm-0.3 mm)/ m ²	31	13.7	120
Items (5.6 mm-1.0 mm)/ m ²	25	11	
Items (1.0 mm-0.3 mm)/ m ²	6	2.4	
Budd Inlet			
Items (5.6 mm-0.3 mm)/ m ²	89	39.8	40
Items (5.6 mm-1.0 mm)/ m ²	73	32.1	
Items (1.0 mm-0.3 mm)/ m ²	16	7.8	
Eld Inlet			
Items (5.6 mm-0.3 mm)/ m ²	2.1	0.8	40
Items (5.6 mm-1.0 mm)/ m ²	1.3	0.7	
Items (1.0 mm-0.3 mm)/ m ²	0.8	0.3	
Totten Inlet			
Items (5.6 mm-0.3 mm)/ m ²	2.3	1.1	40
Items (5.6 mm-1.0 mm)/ m ²	2.2	1.1	
Items (1.0 mm-0.3 mm)/ m ²	0.1	0.1	

Overall Weights

An average of 32 ± 13 mg of micro-plastics/ m^2 was observed throughout the study area, which reflects the weight of all the size classes. Budd Inlet had the highest density by weight of micro-plastics with 91 ± 38.8 mg of micro-plastics/ m^2 . Eld and Totten Inlets had much lower densities by weight with 1.5 ± 0.7 and 2 ± 1.0 mg of micro-plastics/ m^2 respectively (Table 3).

Table 3
Means of milligrams of micro-plastic/ m^2 collected from January 16th, 2013 to February 6th, 2013.

Water Body Inlet Variable	Summary statistics		
	Mean	Std. Error of μ	Area Sampled (m^2)
All Finger Inlets			
Milligrams (5.6 mm-0.3 mm)/ m^2	31.57	13.39	120
Milligrams (5.6 mm-1.0 mm)/ m^2	31.02	13.22	
Milligrams (1.0 mm-0.3 mm)/ m^2	0.36	0.18	
Budd Inlet			
Milligrams (5.6 mm-0.3 mm)/ m^2	91.2	38.76	40
Milligrams (5.6 mm-1.0 mm)/ m^2	90.16	38.26	
Milligrams (1.0 mm-0.3 mm)/ m^2	0.94	0.54	
Eld Inlet			
Milligrams (5.6 mm-0.3 mm)/ m^2	1.48	0.7	40
Milligrams (5.6 mm-1.0 mm)/ m^2	1.35	0.7	
Milligrams (1.0 mm-0.3 mm)/ m^2	0.13	0.1	
Totten Inlet			
Milligrams (5.6 mm-0.3 mm)/ m^2	2.03	0.98	40
Milligrams (5.6 mm-1.0 mm)/ m^2	1.56	0.9	
Milligrams (1.0 mm-0.3 mm)/ m^2	0.01	0.01	

The overall standard error of the means of each inlet, for both density by number of items and density by weight, were very high because of the spatial variability within sites, between sites and between the inlets. For example, one site (Budd NE) contained 94% of all the micro-plastics found in this study. Hence the highest mean density of micro-plastic was observed at Budd NE with 350 ± 131 items/ m² and 91.2 ± 38.8 mg/ m², whereas the lowest mean density of micro-plastic for a study location (Eld NE) was 0.2 ± 0.2 items/ m² and 0 ± 0 mg/ m².

Variability of micro-plastic density (items/ m²) within Inlets

Unless otherwise indicated, “mean density of total items” refers to the density of items in both the large (5.6mm-1.0mm) and small (1.0mm-0.3mm) size classes. The strong positive relationship found between number of items in the two size classes ($R^2 = 0.96$) suggested that examination of both size classes together would be representative of the sample in order to examine the factors driving spatial variability within inlets. This is further supported by the strong positive relationship found between total number of items collected and total weight ($R^2 = 0.98$).

Budd Inlet

Budd Inlet contained 95% of the plastic collected during this study. Spatial variation within the inlet was very high as the majority of micro-plastics collected within Budd Inlet (98%) were located at the study site Budd NE. The mean density of total items collected at Budd NE was 350 ± 131 items/ m². 1% of the micro-plastics from Budd Inlet were collected at Budd NW, where the mean density was 4 ± 1 items/ m². Budd SW and Budd SE contained less than 1% of the micro-plastics collected within Budd Inlet. The

mean density of total items collected at Budd SW and Budd SE was 2 ± 1 items/ m^2 and 1 ± 1 items/ m^2 respectively (Appendix 1).

Eld Inlet

Eld Inlet contained 2.2% of the micro-plastic collected during this study. Though Eld had a lower density of micro-plastic than Budd Inlet, spatial variability was also great in Eld Inlet. The majority of micro-plastics collected within Eld Inlet (68%) were located at the study site Eld SW, where the mean density was 6 ± 3 items/ m^2 . Eld NW and Eld SE each contained 14% of the micro-plastics, with mean density of total items being 1 ± 1 items/ m^2 and 1 ± 0.5 items/ m^2 respectively. Eld NE contained 2% of the micro-plastics collected within Eld Inlet. The mean density of total items collected at Eld NE was 0.2 ± 0.2 items/ m^2 (Appendix 2).

Totten Inlet

Totten Inlet contained 2.5% of the micro-plastic collected during this study. The majority of micro-plastics collected within Totten Inlet (59%) were located at the study site Totten SW, where the mean density was 5 ± 4 items/ m^2 . Totten NW contained 24% of the micro-plastics collected within Totten Inlet with a mean density of 2 ± 1 items/ m^2 . Totten NE and Totten SE each contained 4% of the micro-plastics from Totten Inlet. The mean density of total items collected at Totten NE and Totten SE was 1 ± 1 items/ m^2 and 1 ± 0.5 items/ m^2 , respectively (Appendix 3).

Variability of micro-plastic density (items/ m^2) within Sites

Spatial variability was present within the sites (Appendix 4) both in terms of the variability of micro-plastic density, but also in the distribution of the small versus large

size classes. Sediment samples from Budd NE consistently contained both size classes, and this site was the only site where micro-plastics were detectable in all of the ten quadrats analyzed per site. The density of individual items was higher by 2 orders of magnitude in the center of the Budd NE when compared to the edges. Similarly, sites such as Budd NW, Budd SE, Budd SW, Eld SW and Eld NW contained both size classes and the spatial distribution appeared to be focused at specific areas along the shorelines. When micro-plastics were detected in Totten Inlet they were generally from the large size class. Like the observations for Budd Inlet, but at a small spatial scale, one quadrat at Totten SW contained the majority of the micro-plastics found in the inlet. Totten NW was the only location where micro-plastics appears to a more uniform distribution across an approximate 50 meter section of the beach. Finally, Eld SE was the only location where the smaller size class of micro-plastics was exclusively found. The micro-plastics found at Eld SE were in two quadrats 50 meters apart from each other along the shoreline.

Overall Distribution of types of Plastic Collected

Micro-plastics were identified by five categories; foams, fragments, films, pellets and filament balls. Of the five categories that the micro-plastics were identified by, foams comprised 87.8% of all items collected in this study. Fragments of micro-plastic comprised 11% of the total, while films 0.6%, pellets 0.3% and filament balls 0.3% were the least dominant items (Figure 15).

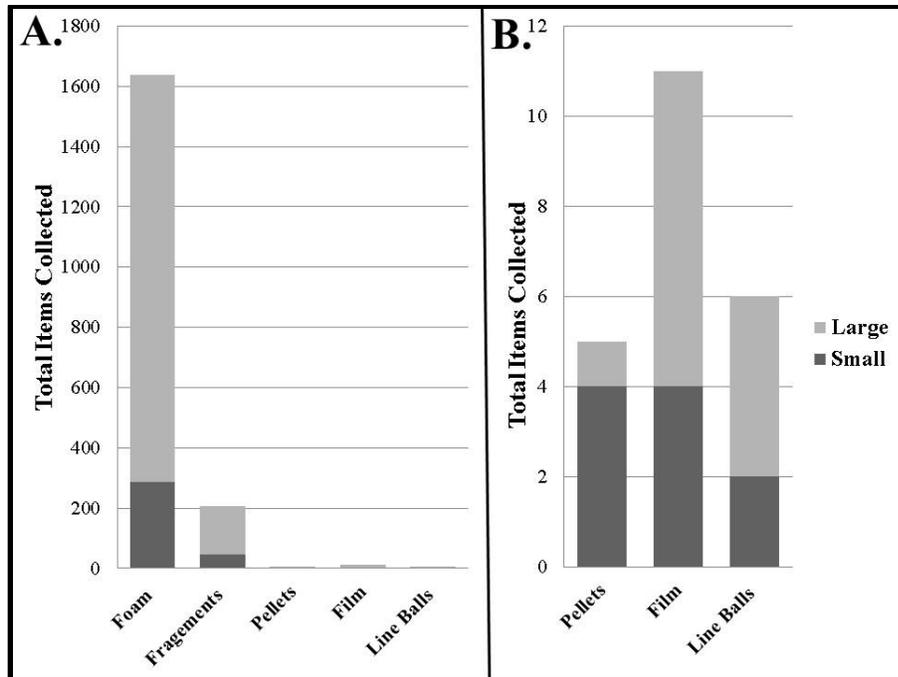


Figure 15. Distribution of categories of micro-plastic collected from January 16th, 2013 to February 6th, 2013. **A.** Distribution of all micro-plastics collected. **B.** Same data set as Figure A. but axis has been adjusted to better portray the data. Large (5.6 mm - 1.0 mm) and Small (1.0 mm - 0.3 mm).

Overall correlation between Size Classes

The size class 5.6 mm - 1.0 mm contained 81.7% of the total micro-plastic items and 99.5% of the total weight that was collected during this study, whereas the size class between 1.0 mm - 0.3 mm contained 18.3% of the total micro-plastic items and 0.5% of the total weight. There was a strong positive correlation between the size classes 5.6 mm - 1.0 mm and 1.0 mm - 0.3 mm for items/ m² (R^2 0.96, $p=0.0001$, $n=120$), indicating that the same processes were causing the accumulation of both large and the small size classes at a given site, although there are exceptions (i.e. Totten Inlet, where very small amounts of the small size class is observed and Eld SE where no micro-plastics of the large size class is observed). There was a weak positive correlation between the size classes large and small when density was explored by weight (R^2 0.05, $p=0.0146$, $n=120$) (Figure 16).

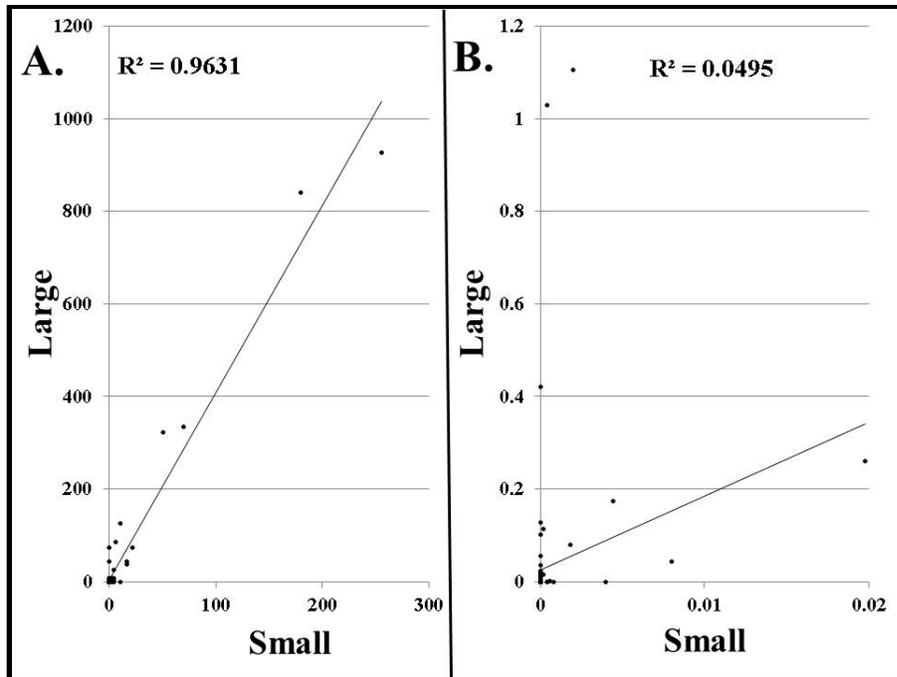


Figure 16. Correlation of the two size classes of micro-plastics collected from January 16th, 2013 to February 6th, 2013. **A.** Correlation of total items of micro-plastic/ m² between large (5.6 mm - 1.0 mm) and small (1.0 mm - 0.3 mm) size classes. **B.** Correlation of total weight of micro-plastic/ m² between large (5.6 mm - 1.0 mm) and small (1.0 mm - 0.3 mm) size classes.

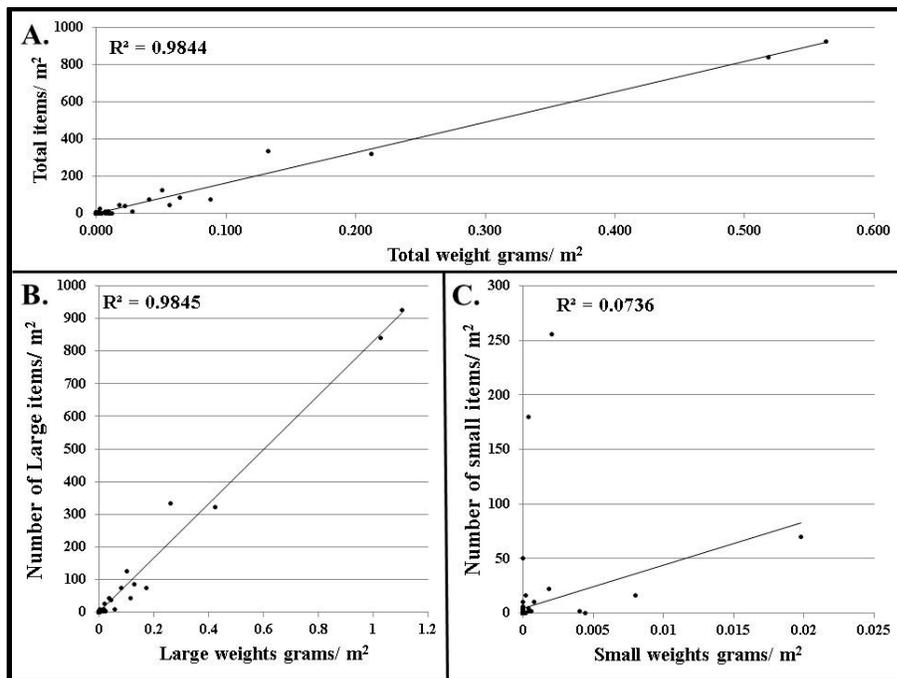


Figure 17. Correlation of relationship between items of micro-plastics/ m² and the weight of the micro-plastics in grams/ m² collected from January 16th, 2013 to February 6th, 2013. **A.** Correlation of total items of micro-plastic/ m² and total weights of micro-plastics in grams/ m². **B.** Correlation of items of micro-plastic/ m² and weights of micro-plastics in grams/ m² for the large size class. **C.** Correlation of items of micro-plastic/ m² and weights of micro-plastics in grams/ m² for the small size class.

Correlation between Weight and Items

There was a strong positive correlation between the weight of the micro-plastic collected (summing both size classes) and the quantity of items (again summing both size classes) collected for the overall study area (R^2 0.98, $p=0.0001$, $n=120$). There was also a strong positive correlation between the weight of the micro-plastic collected and the quantity of items collected for the large size class (R^2 0.98, $p=0.0001$, $n=120$), and a very weak positive correlation between the weight of the micro-plastic collected and the quantity of items collected for the small size class (R^2 0.07, $p=0.0027$, $n=120$) (Figure 17). This weak correlation was due to the lower limits of the scale used for measuring micro-plastics which made the weights of the small size class undetectable.

Micro-plastic Accumulation Zones within the Inlets

As described in the previous section, all three finger inlets studied had locations where higher accumulation of micro-plastics occurred. For the purposes of this thesis, accumulation zones are defined in this study as locations within an inlet that contain 50% or more of the total number of micro-plastic items/ m^2 collected in that inlet, which indicated that each inlet contained one accumulation zone. The study site Budd NE contained 98.1% of the micro-plastic pieces collected within Budd Inlet. Eld SW contained 68.3% of the micro-plastic pieces collected within Eld Inlet, whereas Totten SW contained 58.7% of the micro-plastic pieces collected within Totten Inlet (Figure 18).

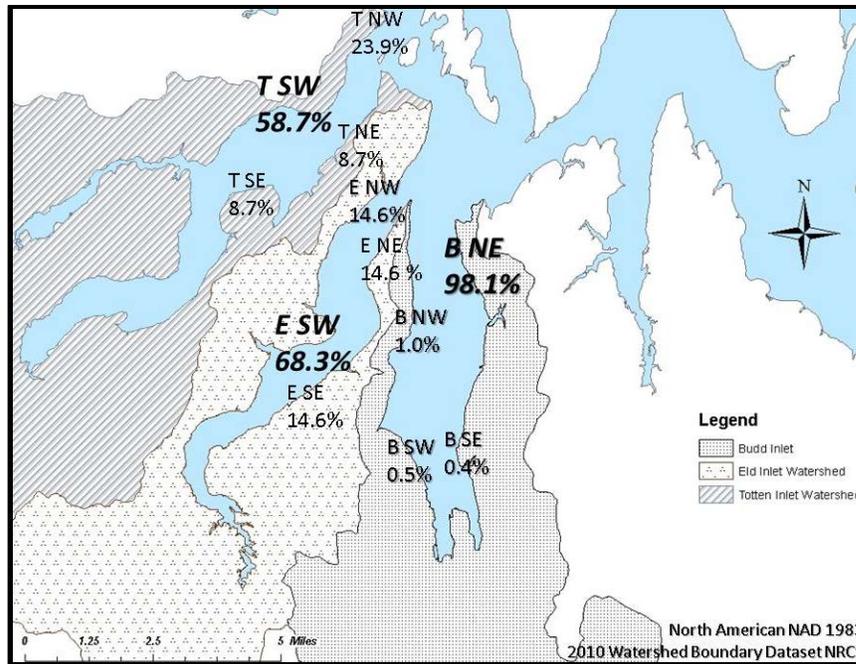


Figure 18. Accumulation zones in each finger inlet. Percentage is the quantity of micro-plastic items/ m² within each inlet **Bold** indicates an accumulation zone.

Comparison of Accumulation Zones across the Inlets

The wilcoxon test of mean ranks of the density of total micro-plastic (in units of items/ m²) in the accumulation zone sites between the inlets was significantly different ($p = 0.0001$). Tukey's honest significance difference test was performed and Budd NE was significantly different from Eld SW and Totten SW. Budd NE contained the majority of the plastics collected within the study area.

Comparison of Non-Accumulation Zones across the Inlets

Wilcoxon test was performed between areas designated as non-accumulation zones across the inlets to determine if one inlet had a higher density of total micro-plastic items/ m². The mean ranks of the non-accumulation zone sites between the inlets were significantly different ($p = 0.0484$). The mean micro-plastic density for the non-sink locations in Budd Inlet was 2.3 items of micro-plastic/ m², while the non-sink locations in

Eld and Totten inlets were 0.9 and 1.3 items of micro-plastic/ m², respectively. Wilcoxon test were further performed between Eld and Totten inlets to determine if one inlet had significantly higher micro-plastic density. The mean ranks of the non-accumulation zone sites between Eld and Totten inlets were not significantly different (p = 0.91). This suggests higher densities of micro-plastics were collected in both the sinks and non-sinks of Budd Inlet.

Watershed Demographics

Demographically, the three inlets are very different. Budd inlet had a population density of approximately 1186 people/ kilometer², while Eld inlet has a population density of 267 people/ kilometer² and Totten inlet has a population density of 84 people/ kilometer².

DISCUSSION

Micro-plastics have been demonstrated to pose a threat to wildlife through direct ingestion (Baird and Hooker 2000; Besseling et al. 2012; Boerger et al. 2010; Colabuono et al. 2009; Davison and Asch 2011; Graham and Thompson 2009; Lindborg et al. 2012) and the sorption of toxic chemicals into wildlife from those plastics that were ingested (Tanaka et al. 2013; Teuten et al. 2009). Micro and Macro-plastics are also dispersing invasive species and have the potential to disrupt ecological communities (Barnes 2002; Gregory 2009). Aside from eliminating the source of micro-plastics, the best way to combat marine micro-plastic pollution and their subsequent impacts to wildlife, is to remove the pieces of plastic from the marine environment and dispose the plastic pollution properly. The easiest location to collect and remove plastic pollution is from shorelines.

Understanding the spatial distribution of marine micro-plastics along shorelines can facilitate focused clean-up efforts to increase cleanup efficiency. For example, micro-plastics have been demonstrated to accumulate in select areas of shorelines as a result of different factors, including population density (Browne et al. 2011; Kingfisher 2011; Kusui and Noda 2003), prevailing wind conditions (Browne et al. 2010; Eriksson et al. 2013; Ivar do Sul et al. 2009) and surface currents (Cooper and Corcoran 2010; Corcoran et al. 2009; Martins and Sobral 2011). Once the spatial distribution and the physical factors that are driving that distribution in a specific area are understood, the potential for isolating the source locations from which micro-plastics originate is obtainable. Accordingly, starting at locations with high micro-plastic density and tracing the physical

factors backwards to the location of the source is a valuable tool for combating cleanup efforts.

Resources are limited for cleanup efforts as well as our current ability to trace the source locations of micro-plastic pollution in Puget Sound. Kingfisher (2011) found little relationship between proximity to population and micro-plastic density on shorelines. While Baker (2011) found a higher densities of micro-plastics in surface waters near urban locations than in remote locations, suggesting that population density is correlated to micro-plastic density.

As such, this study examined the distribution of micro-plastics between Budd, Eld and Totten inlets all located in the South Puget Sound. Budd inlet, which has the highest population density of the inlets, contained the highest density of micro-plastics by one order of magnitude. This suggests that anthropogenic factors are the main driver of spatial variability between the inlets. Within the inlets spatial variability was also very great suggesting that physical factors are the main driver of spatial variability within the individual inlets. The findings of this study also suggest that when micro-plastic pollution is present in South Puget Sound, it will accumulate in specific areas driven by physical factors such as winds and currents, (see discussion below), thus suggesting that targeted cleanup efforts should be devised in South Puget Sound.

Spatial variability between the Inlets (Anthropogenic Factors)

Budd Inlet as a whole contained 45 fold more micro-plastics/ m² than Eld and Totten Inlets (Figure 19). Further, it contained 95% of the total micro-plastics collected. The spatial distribution between the inlets appears to be driven by a combination of

anthropogenic factors, including population density, the amount of land covered with impervious surfaces, and marine utilization by recreational users.

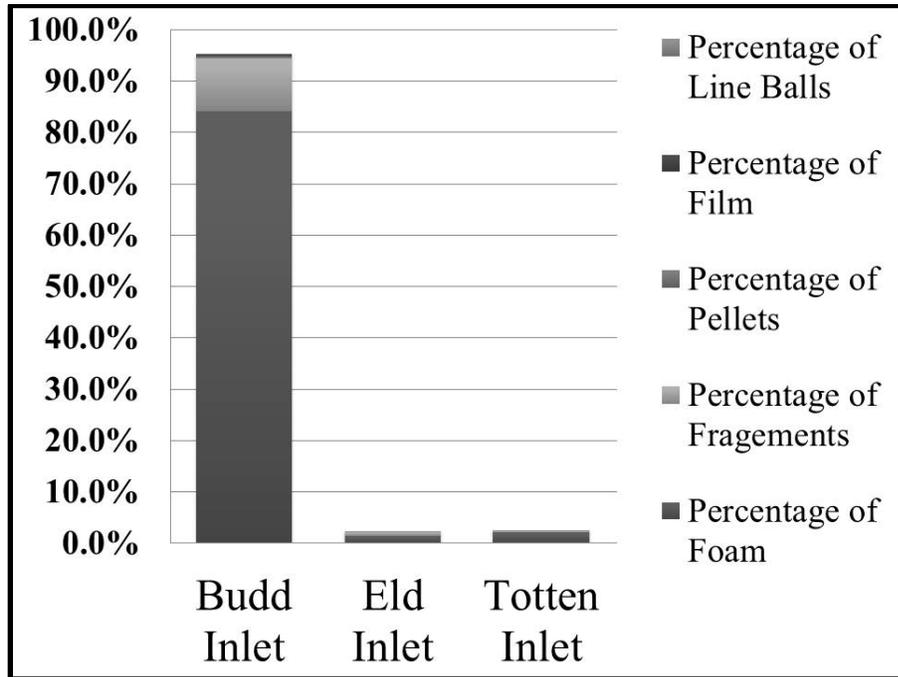


Figure 19. Distribution of total micro-plastics Items/ m² collected in the study area separated by inlet from January 16th, 2013 to February 6th, 2013.

First, the population of Budd Inlet is much higher than that of the Eld and Totten Inlets. The population density is approximately 1,186 people/ kilometer² in Budd Inlet while Eld and Totten have an approximate population density of 267 and 84 people/ kilometer². As micro-plastics are only anthropogenically produced, this suggests that population could be responsible for the high density of micro-plastics found in this inlet. The density of micro-plastics found overall within the inlet, within the accumulation zones ($p = 0.0001$) and within the non-accumulation zone locations ($p = 0.0484$) was significantly higher in Budd Inlet compared to Eld or Totten Inlet.

A strong positive correlation between population density and micro-plastics/m² was present ($R^2 = 0.98$, $p = 0.10$), although there were only three inlets examined, augmenting the p-value. Browne et al. (2010) as well as Kusui and Noda (2003) found a

similar relationship between population density and micro-plastic density. Browne et al. (2010) sampled 18 shoreline locations around the world with varying populations, a positive correlation was found between population density around the sample location and the quantity of micro-plastics that were collected. Kusui and Noda (2003) found a similar relationship between population density and micro-plastic density when they compared Japanese shorelines with high population density to less populated Russian shorelines.

The distribution of population and developed land is different between the inlets which could also be influencing the differences in micro-plastic density between the inlets. Sixty eight percent of the population in Thurston County lives in the incorporated areas, along the shorelines population density is the highest within the Olympia city limits. Olympia is located at the southernmost tip of Budd Inlet and the majority of development is densely located within that area, which is demonstrated by the differences in impervious surfaces. Impervious surfaces make up 25-40% of the land cover in the southern tip of Budd Inlet, while 2-10% of the land cover in the middle and north section of Budd Inlet contain impervious surfaces. The land use in Eld and Totten inlets closely resembles (2-10% impervious surfaces) the middle and north end of Budd Inlet (Thurston Regional Planning Council 2012).

However it is interesting to note that the highest accumulation of micro-plastics was not found in the southern Budd inlet sites. The highest accumulation was found at Budd NE, which is located at the end of Budd inlet on the eastern shoreline. The high population density in the south end of Budd Inlet, in combination with the higher density of impervious surfaces may create the opportunity for more plastics that are discarded to

enter the inlet and be transported by the prevailing southwest wind and surface currents to the Budd NE study site and beyond (see discussion below in Physical Factors driving Spatial Variability).

The inputs of micro-plastics from waste water treatment plants was originally hypothesized in this study to be a driver of micro-plastic density on shorelines in South Puget Sound. Upon further investigation the majority of the micro-plastics collected were foamed plastics and plastic fragments from the degradation of larger plastic pieces. The micro-plastics of concern from waste water treatment plants are micro-beads which are commonly used in facewash, tooth paste and dish soap (Fendall and Sewell 2009). Five of the 1,751 pieces of micro-plastics collected at Budd NE were nurdles. Four of the five nurdles collected were very small and blue in color, similar to the micro-beads found in facewash and toothpaste.

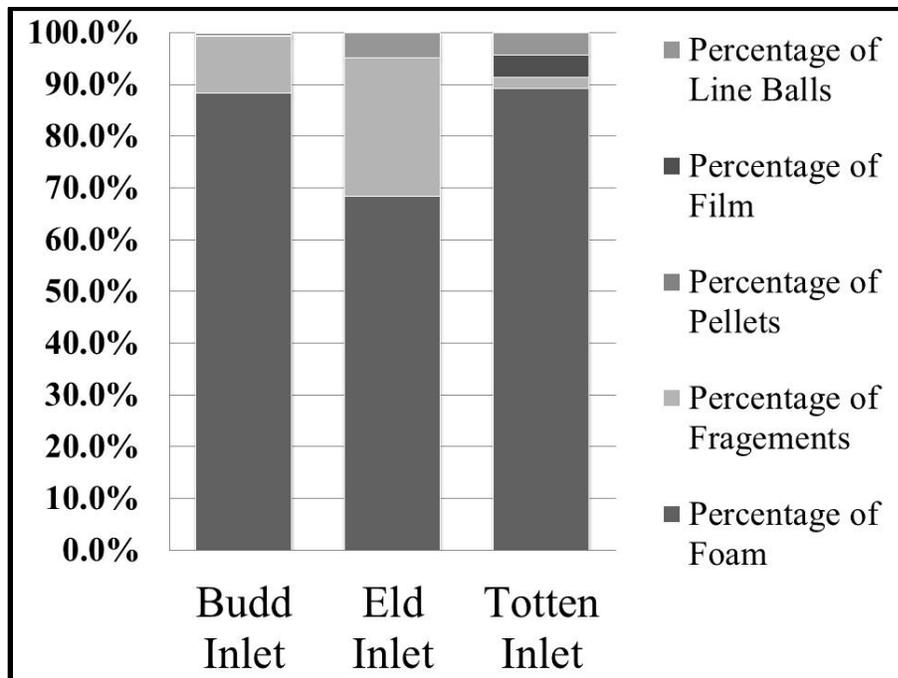


Figure 20. Distribution of micro-plastics categories collected in the study area from January 16th, 2013 to February 6th, 2013.

There are four waste water treatment plants that input water into Budd Inlet, one in Totten Inlet and none in Eld Inlet (Roberts et al. 2009). This suggests that the inputs of the waste water treatment plants have the potential to become a source for micro-plastics in the South Puget Sound, but larger plastic waste being discarded into the marine environment is currently the major issue. Foamed plastics were the dominant micro-plastic collected within all inlets (Figure 20).

Foamed plastics are one of the plastics considered disposable. Common uses for foamed plastics are to-go and carryout containers from restaurants as well as coolers for beachgoers, recreational boaters and commercial fishermen. Foam was also used in the construction of older floats and docks for marinas (Gregory 2013). Foamed plastics float higher out of the water, than other plastics due to their construction with air pockets. The compositional data suggests that these are likely sources of marine micro-plastic pollution in South Puget Sound.

Spatial Variability within Inlets (Physical Factors)

This study found that locations within South Puget Sound inlets accumulate floating debris non-uniformly. For example, the number of micro-plastics/ m² collected at the Budd Inlet North East site (Budd NE) was 158 fold that which was collected at the 11 other sites in the study, including three other sites found within Budd Inlet. Budd NE contained 181 fold more foamed plastics, 122 fold more plastic fragments and 19 fold more plastic films compared to the 11 other sites in the three finger inlets. The only plastic pellets collected during the study were collected at Budd NE, though no line balls were detected at the site. These observations beg the question as to what is causing the

preferential accumulation of micro-plastics at this site, relative to Budd Inlet as a whole and the rest of the study area. In addition to being affected by higher population in Budd inlet, the high density of micro-plastics collected at Budd NE could be attributed to a combination of two physical factors: its geographical position and aspect relative to the prevailing wind direction and the effects of localized currents.

Winds

Winds are likely to be a dominate factor in controlling the spatial distribution of micro-plastics within the inlets. The prevailing wind in South Puget Sound is from the southwest (United States Naval Research Laboratory 2008). During this study the average wind direction was from the southwest (Figure 21).

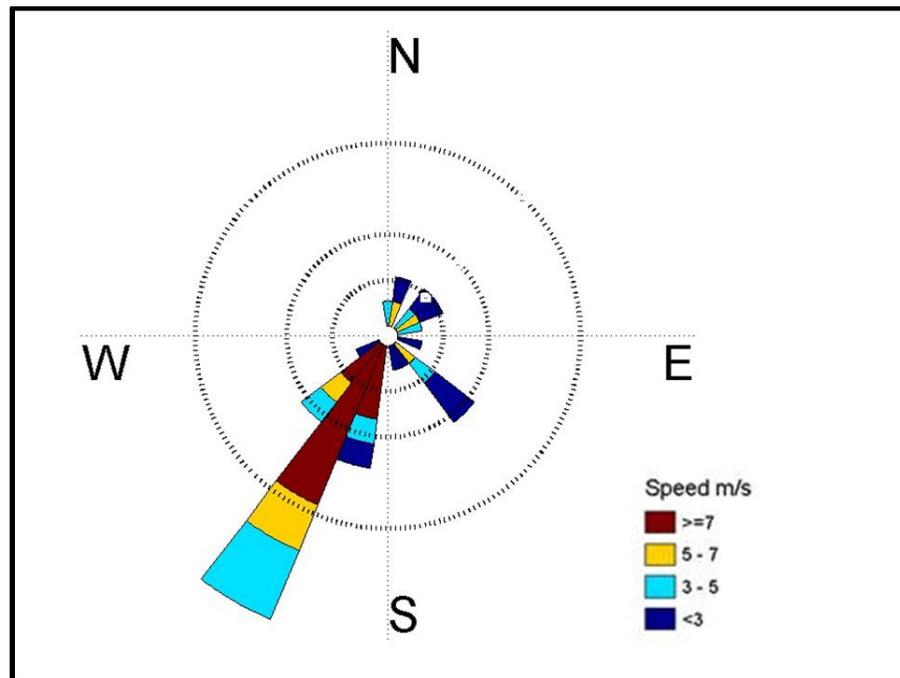


Figure 21. Wind direction in South Puget Sound, Washington State USA during the duration of this study (January 16th- February 6th, 2013). Data from (National Weather Service - NWS Seattle 2013). Reprinted with permission from S. Albertson.

The micro-plastics that float higher in or on top of the water such as foamed plastics would have the potential to be driven by wind alone. With strong southwest winds during this study, foamed plastics were found in higher densities on shorelines that have a southward aspect, such as Budd NE which faces west-southwest. This suggests that downwind shorelines from population centers could have higher densities of micro-plastics.

Totten and Eld Inlets also contained locations where higher densities of micro-plastic items/ m² were collected compared to the other sites within each inlet. Eld southwest (Eld SW) and Totten southwest (Totten SW) are both located in the southwest of their respective water bodies, which could have played a factor in the higher micro-plastic density found at each of these sites relative to the other study locations within their respective inlets. Eld SW faces south while Totten SW faces south-southwest. Eld SW and Totten SW both contained high percentages of foamed plastics, with 89% and 93% respectively. The high percentage of foamed plastics collected at the two sites could be due to the way that foamed plastics float upon the water. The strong prevailing southwesterly wind could have pushed discarded foam pieces up onto the Eld SW and Totten SW beaches where they were broken apart into smaller pieces and mixed in with the sediment by wind wave action.

Further evidence that wind may be a driving factor of the micro-plastic accumulation in the South Puget Sound can be found by examining sites Eld northwest (Eld NW) and Totten northwest (Totten NW). Both Eld NW and Totten NW face east-southeast, indicating these sites would intercept the southwesterly wind, these two sites contained 14.6% and 23.9% of the micro-plastics collected within each inlet respectively.

Foamed plastics were the only item collected at Totten NW while an even mix of plastic fragments and foamed plastics was collected from Eld NW.

Currents

In addition to winds, surface current rotation and tidal pumping are likely to be a secondary factor in controlling the spatial distribution of micro-plastics in this study. The counter-clockwise rotation of surface and sub-surface currents in Budd Inlet suggest that debris could be picked up from the higher populated south end of the inlet and deposited at the entrance to the inlet at the Budd NE site (Aura Nova Consultants et al. 1998; Roberts et al. 2009).

The currents in Eld and Totten inlets have not been studied as extensively in Budd Inlet. However, higher levels of micro-plastics were collected in levels indicated as accumulation zones in data on Totten and Eld inlets surface currents obtained from Aura Nova Consultants. The areas predicted to be accumulation zones by the recertification project for the LOTT contained much higher densities of micro-plastics than the other locations, suggesting that currents as well as wind are playing a role in accumulating micro-plastics on shorelines.

Another driver of surface water in the South Puget Sound is freshwater inputs. The net flow within any estuary is toward the entrance; this flow is primarily driven by tidal pumping in Puget Sound but the addition of fresh water on the surface does aid in this movement. The Deschutes River enters Budd Inlet at the inlets southernmost tip, and contributes to the net surface flow toward the entrance of the inlet. Though freshwater inputs have some effect on the circulation and net flow of the surface water in Puget

Sound, tidal pumping has been demonstrated to be the major driver of surface flow in South Puget Sound and Budd Inlet (LOTT 2000).

Conclusion of driving factors

A combination of local currents and prevailing wind appear to be the main drivers of the spatial distribution of micro-plastic densities within the finger inlets in South Puget Sound. The same physical processes that drove the location of accumulation zones were also responsible for the lower densities of micro-plastics at the other locations. The predominant southerly wind would have blown micro-plastics away from Budd northwest (Budd NW), Budd southeast (Budd SE), Budd southwest (Budd SW), Eld northeast (Eld NE), Eld southeast (Eld SE), Totten northeast (Totten NE) and Totten southeast (Totten SE). These same locations were also indicated as non-accumulation zones in the 1998 LOTT study, with the exception of Budd SE. This suggests that both current and winds created the conditions for lower micro-plastic densities on these shorelines.

Further Implications

The overall results for micro-plastic density of items/ m² are similar to other shoreline micro-plastic surveys. When examined per inlet the results for Budd Inlet are on the higher end of the spectrum (89 pieces of micro-plastics/m²) while the results for Totten (2.3 pieces of micro-plastics/m²) and Eld (2.1 pieces of micro-plastics/m²) are the lowest reported numbers.

Table 4

Previous studies examining micro-plastic density along shorelines. Mean density is presented.

Author	Location	Weight/ m²	Pieces/ m²
Kusui & Noda (2003)	Japan	13.6 g	2610 Pieces
	Russia	8.8 g	31.3 Pieces
Martins & Sobral (2011)	Portugal	36.4 g	185.1 Pieces
<i>Gilman (2013)</i>	<i>South Puget Sound</i>	<i>0.003 g</i>	<i>31 Pieces</i>
Costa et al. (2010)	Brazil		29 Pieces
Ivar et al. (2009)	Equatorial Western Atlantic		9 Pieces
Kingfisher (2011)	North Puget Sound	2.46 g	

The milligrams/ m² of micro-plastics found during this study was much lower than any other study that examined buried marine micro-plastics (Table 4). This can be explained by the other studies not isolating micro-plastics when creating total weights for their studies. Kusui & Noda (2003) included rubber, glass and metals in their estimates for marine debris on shorelines. Martins & Sobral (2011), included plastics larger than greater than 10mm which accounted for 89.6 percent of the weight collected during their study. The distribution of plastics that they collected during their survey was dominated by polyethylene, polyester and then polystyrene. Products made of polyethylene are commonly dense and weigh more than products made of foamed polystyrene which is created using air pockets in the foam.

The values found by Kingfisher (2011) were only reported in terms of weight so comparisons are difficult. The net flow of surface currents in Puget Sound is toward ocean so higher numbers would be expected in the North Sound than the South Sound as well. The population density on the east side of northern and central Puget Sound metro area was 2,930 people per km² in relation to the population density of South Puget Sound metro region was 1,058 people per km² (United States Census Bureau 2013). The

cumulative effect of the population all along the shorelines adds to help create the larger numbers in the North Sound.

The lower density of micro-plastic pieces/ m² found in this study are due to sampling at the head of the 120km long estuary in isolated inlets with lower relative population density. The drivers of micro-plastic density in South Puget Sound appear to be a combination of anthropogenic factors; higher total population at possible source areas and high percentage of impervious surfaces allow the micro-plastics to enter the system. Once micro-plastics have entered the system the prevailing Southwest wind in combination with the surface currents driven by tidal pumping and riverian sources push north. Shorelines that are oriented south within the inlets accumulated the majority of the micro-plastics that were collected during this study.

CONCLUSION

Micro-plastics are present on all of the shorelines surveyed across the three South Puget Sound finger inlets Budd, Eld and Totten. Two orders of magnitude more micro-plastic pieces/ m² were collect in Budd Inlet than Totten or Eld Inlets. The spatial distribution between the inlets is due to anthropogenic factors. Once the micro-plastics have entered the inlets, physical drivers control the spatial distribution. In South Puget Sound, wind appeared to be the primary driver of spatial distribution. This study creates baseline data for South Puget Sound as well as locating sites that would be ideal for further research studying temporal accumulation rates of micro-plastics for shorelines in South Puget Sound. Further work should be conducted examining other areas that micro-plastics are believed to be accumulating in. Further research into the temporal distribution of marine micro-plastic density will allow future trends to be understood. Selecting locations for future work where circulation is understood and little mixing between isolated sites allows other issues such as the impact of population density to be understood.

FURTHER WORK RECOMMENDATIONS

Further research on the spatial distribution of marine micro-plastics in South Puget Sound is recommended in three areas; the role of impervious area, the role of recreational boating and the role of bathymetry on smaller scale spatial distribution.

Total impervious area was positively correlated to micro-plastic pieces/m² in the inlets ($R^2 = 0.89$, $p = 0.21$), though the high p-value is likely due to the small sample size. The total percentage of impervious surfaces was also higher in Budd Inlet. Eight percent

of the land within the Budd Inlet watershed is covered with impervious surfaces, which can contribute to greater levels of storm water runoff. Storm water runoff can carry pieces of plastic and micro-plastic into the inlet. An estimated 4% of the land within the Eld inlet watershed is covered with impervious surfaces, while an estimated 2% of the land in the Totten Inlet watershed is impervious surfaces. The combination of high population density and high percentage of impervious area creates a situation where more plastics and micro-plastics can be washed into an inlet by storm water water runoff though this relationship has been studied very little.

In addition to factors on land, more recreational boaters are using Budd Inlet than Eld or Totten Inlets (personal observation). Budd Inlet is also the only inlet of the three studies that contains recreational marinas (Discover Boating 2013). The combination of anthropogenic factors may create a situation where more micro-plastics are accumulating within Budd Inlet due to coolers and plastic pieces falling off of boats.

Spatial variability of micro-plastic density, pieces/ m² within the sites was also present. Six of the twelve sites appeared to have patterns of areas along the shoreline that accumulated more than others. At the other six sites the spatial distribution appears sporadic. The spatial distribution at the smaller level is believed by the author to be due to changes in localized bathymetry. But further work understanding the small scale spatial distribution would contribute to the ultimate goal of tracking the sources of micro-plastics at the source.

Understanding the role that these additional factors play in the spatial distribution of marine micro-plastics will further advance knowledge in the ability to identify point sources of the pollution as well as informing organizations working on beach cleanups.

INTERDISCIPLINARY STATEMENT

More and more people live close to coastlines, creating more and more waste. Plastics persist in the marine environment for much longer than we had thought about, and accumulation is believed to be increasing. The use of cleanup efforts along beaches is one pathway to combat the growing problem of marine debris and marine micro-plastics. This study identified the presence of locations with higher density of micro-plastics than other locations, suggesting that beach cleanup efforts can have a large effect at localized areas. These findings suggest that beach cleanup efforts in South Puget Sound should be targeted at locations with a high abundance of visible micro-plastics. Targeted cleanup efforts will likely have a large impact on reducing micro-plastic pollution in South Puget Sound.

This study found that foamed plastics were the most prevalent form of plastics in our marine waters. The same high quantities of foamed plastics was found in shoreline surveys conducted in the Northern region of Puget Sound (Kingfisher 2011). A possible source of foamed plastics could be marina floats. Marinas in Puget Sound are required to convert from foam plastic docks to concrete docks in order to obtain the Clean Marina Certification from Puget Soundkeeper Alliance (Gregory 2013).

Recycling programs for foamed plastics are now in place in Thurston county as well as efforts to educate the public on alternatives to foamed plastics (Dodge 2009). In Thurston County 0.83 percent of the solid waste by weight were foamed plastics. Efforts to reduce, reuse and recycle should be continued but work should also be conducted to isolate the principle uses of foamed plastics to identify the points of entry into the marine eco-system.

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APPENDICES

Appendix 1.

Table Budd

Micro-plastic (number of items/ m²) collected from January 16th, 2013 to January 29th, 2013 in Budd Inlet. Standard error of the mean is reported as (SEM).

Inlet <i>Sample Location</i>	Summary statistics		
	Mean	Std. Error of μ	% within Inlet
Budd Inlet			
<i>Budd NE</i>			
Items (5.6mm-0.3mm)/ m ²	350	131.8	98.1%
Items (5.6mm-1.0mm)/ m ²	286	105.2	
Items (1.0mm-0.3mm)/ m ²	63	27.3	
<i>Budd NW</i>			
Items (5.6mm-0.3mm)/ m ²	4	1.4	1.0%
Items (5.6mm-1.0mm)/ m ²	3	1.2	
Items (1.0mm-0.3mm)/ m ²	1	0.5	
<i>Budd SE</i>			
Items (5.6mm-0.3mm)/ m ²	1	0.5	0.4%
Items (5.6mm-1.0mm)/ m ²	1	0.4	
Items (1.0mm-0.3mm)/ m ²	1	0.3	
<i>Budd SW</i>			
Items (5.6mm-0.3mm)/ m ²	2	0.6	0.5%
Items (5.6mm-1.0mm)/ m ²	1	0.4	
Items (1.0mm-0.3mm)/ m ²	1	0.4	

Appendix 2.

Table Eld

Micro-plastic (number of items/ m²) collected from January 22nd, 2013 to January 29th, 2013 in Eld Inlet. Standard error of the mean is reported as (SEM).

Inlet <i>Sample Location</i>	Summary statistics		
	Mean	Std. Error of μ	% within Inlet
Eld Inlet			
<i>Eld NE</i>			
Items (5.6mm-0.3mm)/ m ²	0.2	0.2	2.4%
Items (5.6mm-1.0mm)/ m ²	0.2	0.2	
Items (1.0mm-0.3mm)/ m ²	0.0	0.0	
<i>Eld NW</i>			
Items (5.6mm-0.3mm)/ m ²	1.2	0.5	14.6%
Items (5.6mm-1.0mm)/ m ²	0.6	0.4	
Items (1.0mm-0.3mm)/ m ²	0.4	0.3	
<i>Eld SE</i>			
Items (5.6mm-0.3mm)/ m ²	1.2	1.0	14.6%
Items (5.6mm-1.0mm)/ m ²	0.0	0.0	
Items (1.0mm-0.3mm)/ m ²	1.2	1.0	
<i>Eld SW</i>			
Items (5.6mm-0.3mm)/ m ²	5.8	2.9	68.3%
Items (5.6mm-1.0mm)/ m ²	4.4	2.6	
Items (1.0mm-0.3mm)/ m ²	1.2	0.4	

Appendix 3.

Table Totten

Micro-plastic (number of items/ m²) collected from February 1st, 2013 to February 6th, 2013 in Totten Inlet. Standard error of the mean is reported as (SEM).

Inlet <i>Sample Location</i> Variable	Summary statistics		
	Mean	Std. Error of μ	% within Inlet
Totten Inlet			
<i>Totten NE</i>			
Items (5.6mm-0.3mm)/ m ²	0.8	0.53	8.7%
Items (5.6mm-1.0mm)/ m ²	0.8	0.5	
Items (1.0mm-0.3mm)/ m ²	0.0	0.0	
<i>Totten NW</i>			
Items (5.6mm-0.3mm)/ m ²	2.2	0.7	23.9%
Items (5.6mm-1.0mm)/ m ²	1.8	0.6	
Items (1.0mm-0.3mm)/ m ²	0.4	0.4	
<i>Totten SE</i>			
Items (5.6mm-0.3mm)/ m ²	0.8	0.4	8.7%
Items (5.6mm-1.0mm)/ m ²	0.8	0.4	
Items (1.0mm-0.3mm)/ m ²	0.0	0.0	
<i>Totten SW</i>			
Items (5.6mm-0.3mm)/ m ²	5.4	4.3	58.7%
Items (5.6mm-1.0mm)/ m ²	5.4	4.3	
Items (1.0mm-0.3mm)/ m ²	0.0	0.0	

Appendix 4.

Variability of micro-plastic density (items/ m²) within Sites in Budd, Eld and Totten Inlets

Figure Budd

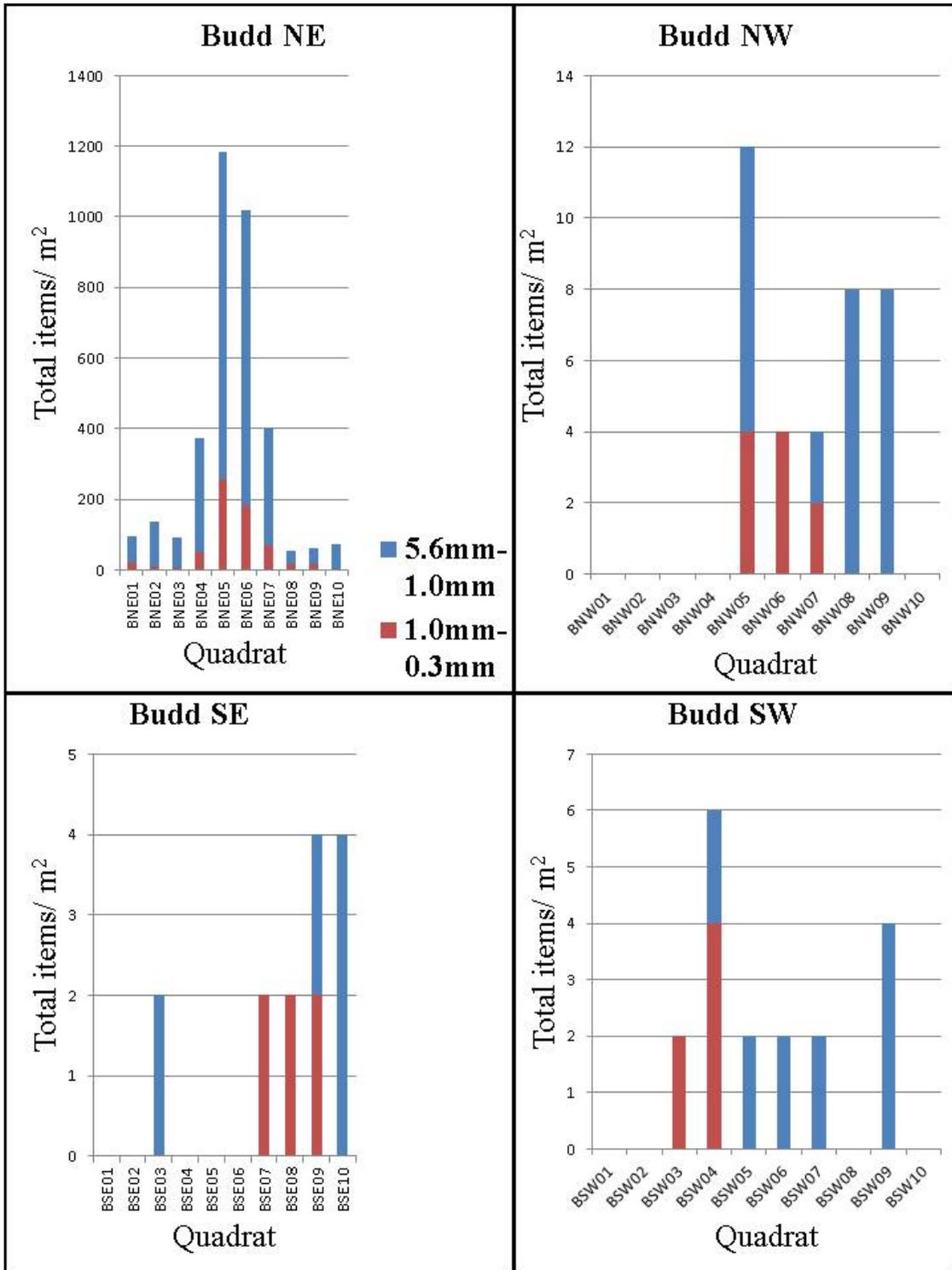


Figure Eld

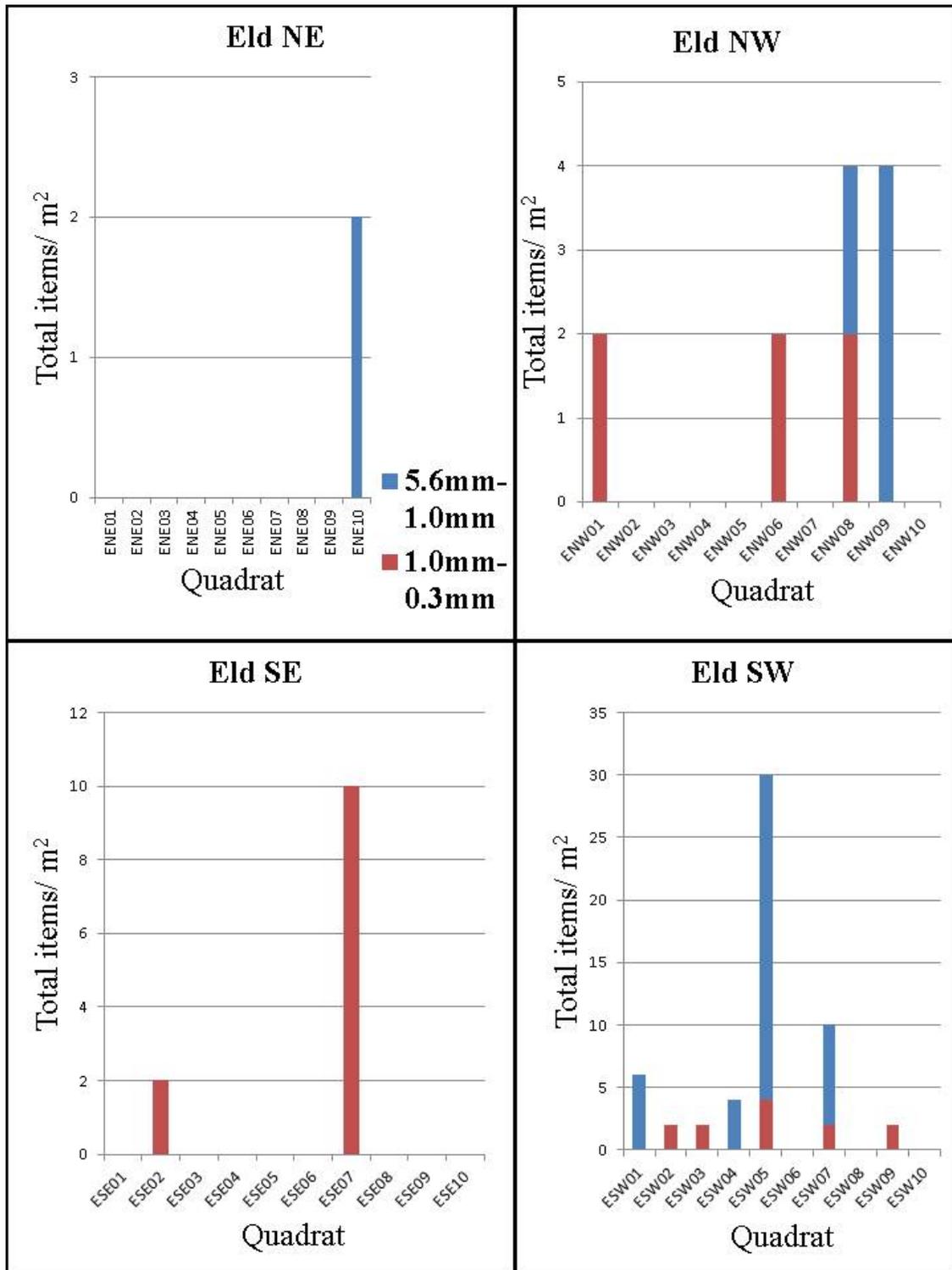


Figure Totten

