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Thesis of Katrina A. Smith

Submitted in Partial Fulfillment of the Requirements for the Degree of

Master of Science Marine Science

Nova Southeastern University Halmos College of Arts and Sciences

May 2022

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NOVA SOUTHEASTERN UNIVERSITY HALMOS COLLEGE OF ARTS AND SCIENCES

An analysis of suspended microplastics in sewage outfalls, inlets, and coastal waters of Broward county, Florida

By:

KATRINA SMITH

Submitted to the Faculty of Halmos College of Arts and Sciences in partial fulfillment of the requirements for the degree of Master of Marine Science with a specialty in:

> Marine Biology Nova Southeastern University April 2022

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Abstract

The widespread use of plastics has led to a surplus of plastic waste in landfills and the ocean. The degradation of these plastics produces microplastics, which are detrimental to the environment. Microplastics can be introduced into the ocean in several ways, including runoff and sewage outfalls; both pathways can concentrate microplastics and promote adsorption of environmental contaminants to the plastic's surface. South Florida has six sewage outfalls, and nine inlets, whose proximity to the Florida Reef Tract increases the potential exposure of microplastics to sensitive environments, such as the coral reef. A quantitative assessment of microplastics introduced into the ocean via sewage outfalls and inlets is an essential first step toward understanding potential environmental impacts. This study evaluated microplastics' concentration, composition, and spatial and temporal distribution in southeast Florida coastal waters. Plastics were quantified in water samples from eight sites. Fourier Transform Infrared (FTIR) Spectroscopy was used to identify 5.7% of the particles sampled, including the plastic polymer composition and possible source material. There were no significant differences between sampling locations, categories, or sites; however, a significant spatial influence was found. Of the particles identified, 57.63% were plastics. This study revealed that the coastal waters of Broward county have notable amounts of microplastics, and the type of polymers present suggests that sewage effluent is a major source of microplastics. The results of this study have provided new background information to guide further research and support future management strategies.

Introduction

Plastic usage has grown exponentially over the last century, and many of those plastics have been discarded into the ocean (Hidalgo-Ruz et al., 2012). Improper waste disposal has contributed to the development of floating islands of garbage composed chiefly of plastics (Martinez et al., 2009). Over time, many physical and chemical processes reduce most plastics into progressively smaller pieces, eventually becoming microplastics. Microplastics are defined as pieces less than 5 mm in size and are divided into two groups, primary and secondary, according to their original size. Primary microplastics, which were originally designed to be less than 5 mm, can enter the ocean directly through sewage effluent (Cole et al., 2011). Plastics that are reduced into smaller pieces via mechanical, chemical, and physical processes are considered secondary microplastics (Ryan et al., 2009). Investigation into the fate of both types of microplastics is critically important, as their effect on the marine environment may be significant.

Desforges et al. (2015) found that microplastics are present and widespread across the oceans, with the highest concentrations near populated areas and convergence zones. Microplastics are often removed from the water column via shoreline deposition, sedimentation, and ingestion, which has led to an underestimation of plastics in the oceans (Desforges et al., 2015). Coastal areas have been found to have some of the highest microplastic concentrations due to their proximity to known plastic sources such as municipal drainage systems and sewage effluents (Yang et al., 2021). A major contributor is the influx of plastics from sewage outfalls which contain concentrated amounts of many primary types of microplastics, such as microbeads derived from cosmetics and clothing fibers (Costa et al., 2010). Outfalls release concentrated amounts of microplastics that have been exposed to human waste, chemical contaminants, and pathogens; thus, they may pose a more significant hazard to organisms (Lares et al., 2018). Southeast Florida currently has six outfalls (Figure 1) that release sewage into the ocean off the coast of Palm Beach, Broward, and Miami-Dade Counties (Hazen and Sawyer 1994). Although sewage is filtered and treated before being released, the high flow rates can introduce large quantities of microplastics (Kelly et al., 2021). Coastal inlets are also a source of plastic debris due to maritime activities and stormwater effluent (Velez et al., 2020). Estimating the concentration and type of microplastics introduced to the ocean via outfalls and inlets is a critical first step in understanding the potential impact of outfall-associated plastics on the environment.

Figure 1. Location of the six active sewage outfalls that discharge effluent in southeast Florida coastal waters. Reproduced from Banks et al. (2008) with author permission

The distribution and abundance of microplastics throughout the marine environment are of increasing concern due to the potential for ingestion by organisms and the ability of microplastics to act as vectors for toxins (Budimir et al., 2018). The potential ecotoxicological effects of microplastics are not well understood and determining how microplastics are introduced into the environment is an essential first step in understanding the threats they pose (Costa et al., 2010). Ingestion, accidental or intentional, is the most common vector of microplastic introduction into food webs. Ingestion can cause intestinal blockage and subsequent starvation in a wide range of consumers, including polychaetes, crustaceans, and fishes (Lares et al., 2018). Hall et al. (2015) found that scleractinian corals can ingest microplastics, which can impede digestion and slow growth. Microplastics may also biomagnify, increasing the risk of ingestion by humans (Lares et al., 2018).

In addition to the physical hazards of ingestion, plastics exposed to UV radiation degrade into their component hydrocarbon compounds, which are irritants and toxins (Munier and Bendell 2018). Microplastics release higher quantities of contaminants such as hydrocarbons compared to larger plastic fragments due to their high surface area to volume ratio (Wright et al., 2013). This high ratio also increases the area for bacterial microfilm development, which can create subenvironments in which anaerobic bacteria flourish (Eckert et al., 2018). The increased density of anaerobic bacteria may then increase the transfer potential of human pathogenic and antibioticresistant plasmids (Eckert et al., 2018). Dense bacterial populations in microfilms also increase the chances of horizontal gene transfer among bacteria (Eckert et al., 2018). Microplastics can also settle into substrates creating an anoxic layer in which toxic chemicals, such as methane and hydrogen sulfide, are produced via anaerobic respiration (Munier and Bendell 2018). Over time, gravity and wave action can compact the sediment, which reduces the oxygen available to the surrounding substrate (Wright et al., 2013).

The distribution of microplastics in the water column is related to surface circulation, precipitation, and prevailing winds. The hydrodynamic wind mixing model has been used to predict microplastic levels 2.5 times higher than what has been collected in surface nets (Doyle et al., 2011, Kooi et al., 2016). Floating microplastics can travel long distances, often breaking into smaller fragments, before settling (Thiel et al., 2013). Most studies have relied on net tows to collect samples; thus, current microplastic research has largely been restricted to sampling the ocean surface. However the clogging of nets with organic material makes it challenging to obtain accurate estimates of microplastics in the water column, and current surface sampling methods may miss up to 70% of microplastics (Kooi et al., 2016). Sampling a fixed volume of water via a Surface Sampler can permit more accurate quantification of microplastics and eliminate errors due to clogging.

Reliable identification of microplastics is difficult due to variations in sampling methods and visual errors (Kappler et al., 2015). Plastic identification often relies on both physical and chemical identifiers. Microscopy can be used to visually identify plastic particles by following physical identifiers such as shape, color, and texture, but it has several limitations (Shim et al., 2017). Fourier Transform Infrared Spectroscopy (FTIR) is an analytical technique used to identify organic and inorganic materials. It is useful for identifying the composition of plastic polymers when combined with high lateral resolution. This technique uses the infrared range of the electromagnetic spectrum to induce vibrations in various chemical bonds and functional groups of the molecules as the energy is absorbed. Data is collected concurrently for each frequency of the infrared spectrum, and as the frequencies at which the vibrations occur for the chemical bonds and

functional groups are well known, the chemical composition of the material can be interpreted. The software transforms the data into a spectral graphical representation, which is then matched to a database for identification.

Significance of Work

The threat of microplastics to the environment and humans is significant (Lares et al., 2018). Between 4.8 and 12.7 million tons of plastic are introduced into the ocean each year, most of which will become microplastics (Jambeck et al., 2015). Several recent studies have found that organisms ingest microplastics at all trophic levels and can bioaccumulate at higher trophic levels (Barnes et al., 2009: Santillo et al., 2017: Rotjan et al., 2019). Microplastics aggregate biofilms on their surface; because they concentrate microbes that would generally remain dispersed, they are potential vectors for disease (Reisser et al., 2015). Recent research has found that microplastics covered in a biofilm are preferred over natural foods in the coral Astrangia poculata, leading to a diminished ability to ingest food (Rotjan et al., 2019).

Microplastics and possibly toxic chemicals they contain and adsorb can lead to compounding effects on ecosystems and human health. Because microplastics bioaccumulate and biomagnify, they are likely to pose a major threat to human health (Tsang et al., 2017). Although the full extent of the effects of microplastics on human health is still poorly understood, seafood contamination could impact the role of fish as a protein source or make coastal water hazardous to swim in, hurting both local and world economies (Barboza et al., 2018).

Monitoring the quantity and composition of microplastics introduced into the ocean via sewage outfalls and inlets is crucial to understanding their potential impact on the environment. This study assessed microplastics' spatial and temporal distribution in the southeastern Florida coastal environment. It also determined their concentration in the upper water column, particularly in waters adjacent to sewage outfalls. Microplastic data from Florida's outfalls, inlets, reefs, and surrounding areas can be used to estimate plastic contamination. Fourier Transform Infrared Spectroscopy permitted the identification of the composition of the microplastic samples, which can help determine where the plastics originated. The findings from this study could support stricter regulations on the amount of sewage discharged, which will reduce some microplastic introduction. Broader public awareness of the threat microplastics pose to the public could increase support for regulatory measures. Current legislation has reduced the output of sewage from the outfalls and is planning on ending their regular use by 2025 (FL DEP, 2004). After 2025, outfalls will only be used to reduce wastewater leaves from treatment plants during periods of high input or emergencies. It is vital to study their current impact and the impact of their use over time; this will help guide regulations for future projects.

Objectives

This study examined the possible major sources of microplastic introduction and accumulation in the coastal marine waters of Broward county, Florida. The objectives were to (1) determine if microplastics were present in coastal waters; (2) determine the concentration of each category of microplastic; (3) identify the types of plastic polymers present using FTIR; (4) determine if there is spatial variation in the concentration of microplastics, and; (5) determine if there is temporal variation in the concentration of microplastics.

Methods

Field sampling

Two main sampling locations were chosen for this study (Figure 2), which are denoted as Hillsboro (Hill) and Port Everglades (PEV). Each location included four categories: outfall, inlet, coral reef,

Figure 2. A map showing the two sampling locations in Broward county. The approxamite sampling site is shown as a dot with its category classification identified via color. Map was made using randymajors.org Map Tools ©2022.

and north. Sites were selected at each location and category, for a total of eight sites. These sites were selected due to their proximity to populated areas which are economically dependent on healthy offshore environments. The Hillsboro location sites and categories are Hillsboro Inlet (inlet), Broward Outfall (outfall), Oakland Ridge Reef (reef), and Hill Northern (north). Sites at the Port Everglades location were as follows: Port Everglades Inlet (inlet), Hollywood Outfall (outfall), Barracuda Reef (reef), and PEV Northern (north). The Northern Site for the Hillsboro location is 26.1300N, -80.08816W, and the location of the Port Everglades north site is 26.00000N, -80.09642W. Inlets and outfalls are likely sources of microplastics, so they were analyzed for them it see what concentration was coming from freshwater input. Concentrations from a nearby reef site were also examined to see if these sites were affected by microplastics. A northern site was also chosen upstream of the inlet/outfall to act as a control since they were less likely to be influenced by the sources and were therefore used as a reference. All sites are based on The National Oceanic and Atmospheric Administration's (NOAA) Florida Area Coastal Environment (FACE) program.

Sample collection

Sampling took place bi-monthly for one year, from February 2020 through December 2020, to examine microplastics' temporal and spatial composition at each site. Due to the closures related to the Covid-19 pandemic, samples were not collected at any site for the Port Everglades location for April 2020.

Water samples were taken at the surface of each site at 0-15 cm depth using a 12-foot-long Nasco swing sampler. Samples were poured into a large container which was filled until there was 4 L of water. Sub-samples were then created by distributing 1 L into one of four containers for transport and storage. Each sub-sample was stored in an individual 1 L plastic container and chilled in coolers for transport (Kooi et al., 2016). All collection equipment was washed three times with seawater from each site before samples were taken. All samples were taken during daytime hours, and the inlets were sampled two hours after high tide when water was flowing out. Per FACE protocols, when the boil (the surface disturbance above the outfall) was weak or not present, water samples were taken from the GPS coordinates of the outfall mouth. Two samples of reverse osmosis (RO) water produced at the Nova Southeastern University Oceanographic campus were also obtained at each sampling time. There were 10 samples taken for analysis at each sampling event, for a total of 56 samples for the duration of this experiment.

Sample processing

Each 1 L sub-sample was filtered separately; samples were vacuum filtered through cellulose filters with a pore size of 0.22 µm and stored in sterile polystyrene Petri dishes. Additional filter methods, such as buoyancy separation, were not used as treated sewage contains high levels of cellulose fibers as well as denser plastics from cosmetics (Lares et al., 2018). Sieve filtration was also not viable because microplastic fibers are flexible enough to fit through micron meshes and the mechanical process can break the fibers into smaller pieces, (Hidalgo-Ruz et al., 2012). Filters were air-dried at room temperature in covered Petri dishes for at least seven days. Once the filters were dry, the particles present were visually identified using the protocol established in Nor et al. (2014), counted, and sorted into five shape categories under a dissecting microscope. The categories used were microplastic fibers, fragments, foam, pellets, and other (Lares et al., 2018). Sub-samples were totaled for each site. Plastics greater than 5 mm (5,000 µm) in length were not considered microplastics and were not analyzed.

Sample identification

Simple random sampling was used to select 5-10 individual microplastic particles from each sample for identification using FTIR. Unique particles, ones that had infrequent identifiers, and those with reoccurring color and shape identifiers were also selected in addition to the random samples to account for loss and identify unique particles. There were 2,070 microplastic particles counted, of which 204 were sent to the lab for analysis. Since funding for this project was limited, available equipment capacity was around 200 samples. A total of 118 microplastic samples were successfully analyzed. There were 86 samples not identified due to samples being unable to be located on the filter paper, destruction in shipment, or instrument inability to acquire a valid signal for interpretation. Identified samples were referred to as microplastic polymers or just polymers.

Analysis was done using a Nicolet IS5 FTIR with a diamond ATR (attenuated total reflectance) crystal at Dallas College Eastfield Campus (EFC) in Mesquite, TX. The instrument uses ONMICTM software to collect, process, and interpret the information. Data were collected over a spectral range of 7800-350cm⁻¹ (Thermo Scientific Nicolet iS5 FT-IR spectrometer Brochure BR51983-E 0216M-Nicolet iS5). The instrument was calibrated according to manufacturer specifications. Background data for the instrumentation were collected to eliminate ambient peaks and were repeatedly collected between batches of similar samples (Arahman et al., 2017). Sample particles were placed into the sample port containing the diamond ATR. Each set of samples had an assigned code and a unique sample number. This information was used as the title of the FTIR file and entered into the OMNICTM software before accumulating the data. The $OMNIC^{TM}$ software was then used to identify the composition of the microplastic samples. Images of each sample were correlated to the spectrum and identified. Staff at EFC identified the microplastic samples. Identification data may include common name, chemical name, and molecular structure. Individual pieces were sorted using images and identification to determine the statistical probability that a representative image will be a specific microplastic (Table 1).

Table 1. An example of the tabulation data produced by FTIR. The first column contains a picture of the microplastic particle identified, the second column shows the spectrum given off when exposed to infrared, and the last column shows the best possible match for the spectrum received.

Image	Spectrum	ID
	O-H Carboxycil C=C-C Aromatic $C-O$ C-H phenol C-H Acid Ring O-H (Alcohol) $C-O \rightarrow$ (a.u) Absorbance Ester 4000 3600 3200 2800 800 400 2400 2000 1600 1200 Wavenumber (1/cm)	PET Polyethylene terephthalate

Data Analyses

Descriptive statistics were used to evaluate the presence of microplastics, the concentration of each shape category, and the type/abundance of polymers. The average concentration of each shape category was calculated per L of surface water. The distribution of the shapes and the type of polymers found were used to identify potential plastic sources. Conclusions were based on which plastic shapes and polymers occurred most often.

Samples from each site were treated as replicates for each sampling time point. This approach resulted in six samples for each site at the Hillsboro location and five samples for each site at the Port Everglades location. All statistical tests were conducted using RStudio. Raw data did not meet assumptions of normality or homoscedasticity. A Wilcoxon rank-sum test with continuity correction (α =0.05) was used to compare the mean microplastic concentration between the Port Everglades and Hillsboro locations. This test was used to determine if the location affected microplastic quantity.

To determine if there was spatial variation between categories (Inlet, Outfall, Reef, North), a One-Way ANOVA (α =0.05) was used. Site data from each location were combined into a single category. Log transformations were used to normalize the data. This test allowed for a comparison of the different categories to determine if the source categories (Inlet and Outfall) differed from the other categories (Reef and North). A Kruskal Wallis ANOVA on ranks test (α =0.05) was used to determine if there was spatial variation between the eight sites. This test allowed for the comparison of each category between locations.

A One-Way ANOVA (α =0.05) with a Tukey's Honest Significance (HSD) test was used to determine if there was temporal variation in the mean microplastic concentration throughout this study. Log transformations were used to normalize the data.

Results

Microplastic particles were found in every sample across all sampling sites (Figure 3). Fibers were the most prevalent particle found, and the predominant polymers were Polyacrylates, Alkyd, and Polyester.

Microplastic classification and Identification

Overall the mean concentration of microplastic particles was 11.8 ± 18 particles/L. A similar study conducted in the last six months of 2019 showed an average particle count of $8.17 \pm$ 7.29 particles/L (Wightman, 2020). Plastic from particles were initially categorized by shape using physical characteristics.

Fibers made up the majority of the microplastics with 1,378 particles (66.57%), then fragments with 485 particles (23.43%), foam with 6 particles (0.29%), pellets with 11 particles (0.53%), and other with 190 particles (9.18%). The mean concentration of fibers for all water

samples was 7.83 ± 14.79 fibers/L; fragments had a mean of 2.76 ± 5.83 fragments/L, pellet 0.06 \pm 0.18 pellets/L, foam 0.03 \pm 0.13 particles/L, and other 1.08 \pm 1.48 particles/L.

There were 22 microplastic particles found in the blank water samples and blank filters; these were not added to the total microplastic count. Six out of 10 blank filters examined had microplastic particles, and there were 13 microplastic particles found in total on the filters. Blank water samples yielded 1.83 ± 2.89 particles/ L of RO water, with blank filters having 1.3 ± 1.83 microplastic particles per filter. As microplastic concentrations in this study are considered underreported, and blanks yielded minimal contamination, filter count values were not corrected. All microplastic particles found in blank water samples and blank filters were sent to the lab for analysis. The microplastic particles from February blank 1 were identified as either a laboratory reagent or an inorganic pigment. The microplastic particles from October blank 2 could not be identified due to the signal being too weak for the FTIR machine to read. The rest of the microplastic particles identified from blank water samples and blank filters (August blank 1, October blank 1, December blank 1, blank filter sample 1, blank filter sample 5, blank filter sample 6, and blank filter sample 8) were identified as the laboratory reagent bromoform, with diphenylamine inhibitor.

A total of 2,070 microplastic particles were found in this study, of which 118 were identified to chemical composition. Most of the microplastic particles identified were plastic polymers (57.63%); the rest were grouped into generic categories. These categories are inorganic, laboratory reagent, organic, organometallic (bromopentacarbonyl bromide), salt (copper(I) thiocyanate), and synthetic organic (camphene) (Figure 4 A & B). There were 14 types of plastic polymers identified (Figure 5). The fiber shape category had the highest variety of plastic polymers. Polyacrylates were the most numerous with 18 identified samples. Polyester and alkyds were identified 11 times, cellophane was found 6 times, ethylene propylene diene 5 times, lowdensity polyethylene 3 times, polyvinylchloride 4 times, polyisobutene 3 times, polyamide 2 times, and all other plastics (polyethylene, polyethylene wax, polystyrene, rayon, and vinyl alcohol/ vinyl butyral) were found once. Cellulose, a common organic identified, and rayon have almost identical FTIR spectrums, which could lead to an underestimate of the rayon found in this study (Lusher et al., 2015). The organic category was the second largest group and consisted of substances like

Figure 4. (A)The composition distribution of the particles identified by FTIR. (B) The distribution of all identified particles categorized by shape.

tallow or hair. All particles sent for identification in the foam shape category were lost or unable to be identified via FTIR.

Many samples in the laboratory reagent group contained bromoform, which is a surface contaminant and not an accurate identification. Bromoform is used in geological tests, laboratory reagents, and as a solvent. Presence in a marine environment is most likely due to the breakdown of disinfectants used in wastewater. Bromoform was also found in the blank water samples and blank filters, and present on 11 of the microplastic particles sent for identification. Organic materials made up 19.49% of the samples identified. Several organic samples contained natural fibers such as cotton and wool, originating from laundry or boat upholstery. Organic material was

Figure 5. Polymer identification for every microplastic identified by FTIR. Polymers are grouped by shape category and stacked to show proportion of each category. Segment color corresponds to the polymer identification.

also found as a surface contaminant on nine microplastic particles. Organic contaminants on the surface can help identify microplastic sources.

Spatial analysis

There were no significant differences between locations (Wilcoxon Test, p=0.8135), (Figure 6).

Figure 6. Mean (±SD) microplastic particles found at the Hill and PEV sampling location across all months.

There were no significant differences between categories (One-Way ANOVA, p=0.8993), (Figure 7). There were no significant differences between sites (Kruskal Wallis ANOVA, p=0.8234), (Figure 8).

Figure 7. Mean (±SD) microplastic particles found across all categories and all months.

Figure 8. Mean (±SD) microplastic particles found across all sites and all months.

Temporal analysis

A highly significant difference was found in mean microplastic concentration over time (One-Way ANOVA, p= <0.01). A Tukey's HSD was used to determine what months differed from each other (Figure 9). February had a significantly higher mean microplastic concentration than all other

Figure 9. The mean (±SD) number of microplastics found for each month of sampling. Letters indicate statistically similar groups.

months, and August had significantly lower mean microplastic concentrations than June, October, and December. April was not significantly different from June, August, October, and December.

Discussion

Microplastic research is an ever-evolving field that has received much attention in recent years, with multiple studies focused on various aspects of introduction, transport, and decomposition. There are many different ways microplastics are collected and identified, making it difficult to compare research accurately, therefore standardized methods need to be developed. Uniform methodology will enable researchers to compare data and obtain an accurate picture. This

study only covered the coastal areas of Broward county; to obtain a complete picture of microplastic trends, further research is needed.

The focus of this study was to (1) determine if microplastics were present in the coastal waters of Broward county, (2) determine the concentration of each common category of microplastic, (3) identify what types of plastic polymers were present, (4) examine spatial variation in the concentration of microplastics, and (5) examine temporal variation in the concentration of microplastics. This was the first study to specifically consider outfalls and inlets as potential sources of microplastics in the surface waters of Broward county. Overall, 2,070 microplastic particles were found over the sampling period; fibers were the most prevalent category of microplastics and 14 polymers were successfully identified. The average concentration of microplastic was 11.8 ± 18 particles/L, which was higher than expected based on the limited studies available for comparison.

Microplastic Classification

Most plastics are transported to the ocean via inland, sea, and aerial sources (Yang et al., 2021). Coastal regions often have higher concentrations of microplastic due to 80% originating from a terrestrial source (GESAMP, 2016). In a review done by Yang et al. (2021), it was found that fibers were the most common type of microplastic found, followed by fragments. The results of this study correspond to these findings, as 66.57% of the microplastics found were fibers. Similar microplastic sampling in the coastal waters of the Southern Mediterranean and South Carolina estuaries also found fibers to be the most prevalent microplastic category (Gray et al., 2018, Tata et al., 2020). Fibers are considered primary microplastics, with their predominant mode of introduction being wastewater from laundry. Even though the two outfalls sampled in this study did not have significantly higher quantities of microplastics at the surface than the surrounding environment, outfalls cannot be overlooked as potential sources of primary microplastics.

Plastic polymers

Coastal regions, especially those near populated areas, have greater varieties of plastic polymers due to their proximity to sources (Yang et al., 2021). This study found 14 different plastic polymers consisting of high- and low-density plastics. The most common polymer identified was polyacrylates, a low-density plastic that is predominantly used in paints, adhesives, and textiles (Penzel et al., 2000). The sources of the polyacrylates were likely both primary and secondary sources. Other common microplastics found were alkyds and polyester; alkyds are used in varnishes, paint, and adhesives and likely entered the marine environment as secondary microplastics from abrasions to boats (Wagner et al., 2014). Polyester is mainly used in clothing and was almost certainly introduced into the ocean via an outfall. Plastics used in textiles, such as polyester, rayon, and polyamides are typically introduced directly into the ocean from sewage outfalls or can be transported from land to water via winds (Yang et al., 2021). Due to Covid-19, disposable mask use drastically increased and became an additional source for microplastic fibers. Many single-use masks are made from synthetic fibers such as polypropylene and polyester. Polypropylene was only found once during this study, higher levels will likely be seen in the future. In a marine environment, polypropylene takes longer to decompose due to salt having a high refractive index and can take hundreds of years to degrade (Ranjan and Goel 2021). During that time, polypropylene and many other plastics desorb additives, pollutants, and metals attached to their surface into the environment. Polyester was the second most common plastic polymer found, it was found as fibers, fragments, and other shape categories. As polyester is commonly found in marine ecosystems, due to its regular use in clothing and fiberglass, it most likely came from laundry or boats (Tokiwa et al., 2009).

Non-plastic Classification

There were several particles found that were not plastic polymers. These include organic herbs, oils/ fats, wool, cotton, and residual laboratory reagents. Several samples had organic surface contaminates such as *Uncaria tomentosa* (Cats Paw) and *Withania Spp.* (Ashwagandha). One sample with Ashwagandha on the surface was identified as cellophane, and this would be considered a secondary microplastic introduced via improper waste disposal. Several samples contained mortar or sand used in construction, this was likely transported into the ocean via runoff or wind. Cellulose was another common organic found and was expected because of outfall input. It is important to note that rayon and cellulose have almost identical FTIR spectrums leading to misidentification. Four instances of cellulose were found, two of which were not a standard color for cellulose and likely rayon that was misidentified by FTIR. Misidentification of microplastics

with FTIR is a common issue that often results in underestimating microplastics in a sample. Surface contamination and similar spectrums can be misleading and inaccurate. Many of the laboratory reagents found were surface contamination and not accurate identifications. This further supports the idea that this study underestimates the microplastics present in Broward county because plastic polymers could be falsely identified as laboratory reagents. A combination of identification methods such as visual microscopy, Raman spectroscopy, and thermal analysis should be used to reduce the likelihood of false identifications.

Microplastics adsorb many toxic substances and can be vectors for transporting pollutants (Raju et al., 2018). Bromoform was the chemical most frequently identified on particles in this study; it is sometimes used as a laboratory reagent and results from the degradation of certain wastewater treatment chemicals (Agency for Toxic Substances and Disease Registry, 1990). Bromoform is considered a potential human carcinogen, and animal studies have shown bromoform to affect kidney and liver function (U.S Department of Health and Human Service. Hazardous Substances Data Bank 1993). There is limited published research available; however, one study found that bromoform increased the incidence of several tumor types in animals (U.S Environmental Protection Agency 1999). Other chemical contaminants were found including copper(I) thiocyanate, an algae/bactericide; talloamphocarboxypropionate, a multipurpose polymer often used in personal care products; and camphene, a plasticizer. These chemicals' cumulative and long-term effects on the environment are not known. As microplastics degrade to progressively smaller particles, their ability to adsorb pollutants increases, as does their ability to become biologically incorporated into an organism's tissues (Snell & Hicks, 2011). Microplastics pose a persistent threat to marine ecosystems and humankind due to their ability to persist in an environment and become vectors for chemical contaminants.

Spatial Influence

No significant influence of location, category, or site on mean microplastic counts was found. Throughout this study, the amount of microplastic particles found varied and did not appear to follow spatial trends. It was expected that source categories (outfalls and inlets) would have the highest concentrations; this was only true for some sampling dates. The apparent homogeneity of microplastics in southeast Florida surface waters is likely explained by the degree of mixing which

results from strong, fast-moving currents in this area. Future research should consider taking samples at various depths in the water column to show settling rates for different polymers. Sampling offshore and in the Gulf Stream is also recommended to determine how microplastics are transported regionally.

The Hollywood and Broward outfalls were both active during the sampling period of this study. There are current expansion plans in development to increase the output of all active outfalls and increase the use of deep injection wells. The projected flow rates in 2025 have the Broward outfall producing 94 MGD and the Hollywood outfall producing 54 MGD (FL DEP, 2004). It was predicted that the outfalls would be a significant source of microplastics, thus leading to higher counts in adjacent surface waters. However, the data does support the outfalls as possible sources of microplastics due to the high number of fibers present and subsequent identification of polymers associated with clothing.

Port Everglades and Hillsboro inlets are areas of significant boat traffic and human activity, although Port Everglades is a large international shipping port and Hillsboro is a noncommercial inlet used primarily for recreation. Due to higher traffic and human activity, Port Everglades was expected to have higher microplastic counts. However, particle counts varied, with the highest counts in October and December at Port Everglades, and the highest counts in April at Hillsboro (when the Port Everglades location was not sampled). Inlets are still probable sources of microplastics as several of the identified polymers originated from antifouling paints and resins.

The northern sites in this study were selected because of their distance from what was believed to be a source of microplastics; it was assumed those sites would have the lowest concentration of microplastics. As at all other sites, however, counts for the northern sites varied. Samples taken at Hill in April, PEV in August, Hill in Oct, and Hill in Dec had the highest count for the category; likewise, samples taken at PEV in Feb, PEV in June, PEV in Oct, and PEV in Dec had the lowest counts for that category. A previous study found an accumulation of microplastics in the northern reaches of the sampling area, which suggested that the Gulf Stream and Florida current created a gyre and concentrated them there (Wightman, 2020). A study done in the Nordic Sea compared microplastic counts in the East Greenland Current to counts in the Greenland Sea Gyre and found that the gyre increased the microplastic pollution in that specific area (Jiang et al., 2020). The data from the present study does not support this; instead, the

observed variation supports the premise that microplastic pollution is subject to currents and winds, which can mix the surface of the water column in complex ways.

Temporal influence

 Microplastic counts varied over time throughout this study, with significantly higher average counts in February $(p<0.01)$ compared to all other months. Many factors, such as weather and tourism, can influence the abundance of microplastics. Tsang et al. (2020) found that microplastic concentrations peaked during the dry season in Hong Kong's coastal areas, suggesting that lower rainfall increased microplastic concentrations. In February 2020, lower than average rainfall and warmer than normal temperatures in Broward county could have led to a significantly higher concentration of microplastics (South Florida Weather, 2020). In contrast, August had significantly lower microplastic counts than June, October, and December. Hurricane Isaias passed along the east coast of Florida in early August, causing heavy rainfall and flooding. This freshwater input and the strong winds associated with a hurricane could have contributed to the lower microplastic counts in the surface waters of Broward county. To obtain a more accurate picture of microplastic input, monthly sampling covering all of South Florida, including all inlets and outfalls, should be done and could yield more detailed information regarding microplastic input.

 South Florida experienced a very warm and dry 2020. The early months of 2020 (January through mid-May) were characterized by lower than average rainfall (South Florida Water Management District, 2020). La Niña conditions emerged during August, with below-average sea surface temperatures across the central and eastern equatorial Pacific Ocean (Florida State University 2020). These conditions continued in the Northern Hemisphere for the duration of the 2020 winter season. La Niña winters tend to be warmer and drier than average winter conditions (Malone et al., 2014). These dry conditions lead to the most active hurricane season on record, this resulted in heavy rainfall and strong winds (National Weather Service, 2021). After mid-May, Broward county experienced above-normal rainfall for the remainder of the year (National Weather Service, 2021). Rainfall in 2020 was overall much higher than in 2019. Higher density microplastics, such as polyamide and polyvinyl chloride, could have been resuspended from sediments during these weather conditions.

South Florida saw a massive decrease in tourism during 2020 due to the COVID-19 pandemic. This possibly led to the high degree of temporal variability in microplastic concentration over time. The winter months are typically peak tourism season, but the pandemic shutdown caused a decrease in both domestic and international visitors in every quarter of 2020 (Visit Florida, 2021). There were 79.34 million visitors to Florida in 2020, which is 39.6% lower than the previous year (Visit Florida, 2021). While this study was not intended to assess tourismrelated pollution, the sampling timeframe allowed a unique assessment of how such a decrease affected microplastic concentrations. The counts of microplastics were expected to be lower; however, this was not the case. In a similar study conducted from July to December of 2019 in Broward county, Wightman (2019) found an average of 8.17 ± 7.29 particle/L in surface waters, which was lower than the average of 11.8 ± 18 particles /L found in this study. A comparison of corresponding months (August, October, and December) in the 2019 and 2020 studies is shown in Figure 10. No substantial differences between corresponding months are noted, except for August 2020.

Figure 10. A comparison of the average number of microplastic particles found in the months of August, October, and December from 2019 and 2020.

Conclusion

The influence of outfalls and inlets on microplastic input has not been well studied, especially in South Florida; the spatial, temporal, and polymer data collected in this study have thus provided a needed baseline for future research. By evaluating the temporal and spatial effects on microplastic distribution, this study revealed that the coastal surface waters of Broward county have a microplastic concentration that is consistent with other coastal areas. Both the particle shape category and type of polymer identified are consistent with other coastal locations. The types of polymers present also indicate that sewage effluent is a major source of microplastics, although the mixing which results from strong currents and surface winds in the study area make it challenging to determine specific point sources of microplastics.

 Microplastics are small and flexible enough to pass through water filtration systems and will be continuously introduced into the marine environment from human refuse. Accumulation in the environment, both biologically and in the sediment, will continue unless drastic measures are taken to reduce plastic production and consumption. A dynamic approach at the consumer and producer level coupled with improved support from governing bodies is the first step to limiting plastic output.

 To better understand the scope and distribution of microplastics across Broward county, analysis of long-term trends and depth profiles is needed. Future studies should implement longer sampling periods and a broader sampling scope. Additional research into the long-term effects of the hazardous chemicals found associated with microplastics would allow for a better understanding of their threat to the environment. Additionally, more stringent processing and analytical identification methods are needed for more accurate polymer identification.

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