Microfiber abundance associated with coral tissue varies geographically on the Belize Mesoamerican Barrier Reef System

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ABSTRACT

Ocean plastic pollution is a global problem that causes ecosystem degradation. Crucial knowledge gaps exist concerning patterns in microfiber abundance across regions and ecosystems, as well as the role of these pollutants within the environment. Here, we quantified the abundance of microfibers in coral samples collected from the Belize Mesoamerican Barrier Reef System (MBRS) using a polarized light microscope and identified a subsample of these to the polymer level using an Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy microscope. Microfibers were found in all coral samples with rayon being identified as the most common microfiber, comprising 85% of quantified pollutants. We found a greater average abundance of microfibers in coral samples from the Sapodilla Cayes (296 ± SE 89) than in samples from the Drowned Cayes (75 ± SE 14), indicating spatial variation in microfiber abundance within coral tissue along the MBRS. These results demonstrate that corals on the Belize MBRS interact with microfibers and that microfiber abundance on reefs varies spatially due to point sources of pollution and local oceanography. As rayon from clothing typically enters the ocean through wastewater effluent, alterations to waste water infrastructure may prove useful in decreasing rayon pollution in coastal waters.

1. Introduction

Approximately eight million metric tons of plastic are expected to enter the oceans each year, adding to an estimated maximum of 245,000 million metric tons of plastic already present in the oceans (Jambeck et al., 2015). Plastic debris has accumulated in the oceans over many years from a variety of sources, including commercial fishing debris and municipal waste (Moore, 2008). Up to 95% of marine litter in any given area may be composed of plastic, with an average of 60–80% of marine litter worldwide being plastic (Moore, 2008). Such plastic debris can be degraded through hydrolysis, biodegradation, thermo-oxidative degradation, and photodegradation (Andrady, 2011). Lower oxygen concentrations and temperatures in the ocean, compared to those in terrestrial ecosystems, further perpetuate the slow degradation of plastic eventually resulting in the formation of microplastics and microfibers (Andrady, 2011), defined as particles <5 mm. Microplastics and microfibers can enter the ocean through the breakdown of macroplastics and fibers that are already present in the ocean (Andrady, 2011), or directly via rivers (Lebreton et al., 2017), wind (Browne et al., 2010), and sewage-runoff containing clothing fibers (Browne et al., 2011).

Microplastics and microfibers, are omnipresent in the oceans accumulating in even the most isolated of locations, such as the surface waters and sediment of Antarctica (Cincinelli et al., 2017; Waller et al., 2017) and ice cores from the Arctic (Obbard et al., 2014). Microfibers, including plastics and synthetic fabrics, such as rayon, are extremely abundant in the oceans with upwards of 13 million tonnes of microfibers entering the oceans each year (Mishra et al., 2019). One of the main sources of microfiber pollution is clothing and domestic laundry. Clothing is a major source of rayon, a man-made semi-synthetic contaminant that is widespread throughout the marine environment (Mishra et al., 2019), which composed 55% of analyzed particles from the coastal waters off of Plymouth, United Kingdom (Steer et al., 2017), and 63% of particles collected in the Atlantic off the southwestern coast of Europe and the western coast of Africa (Kanhai et al., 2017). Rayon and many other microfibers enter the oceans mostly through wastewater effluent and has been identified as the most common contaminant found

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in sediment and water samples from the deep ocean to shallow coastal ecosystems (Woodall et al., 2014; Yu et al., 2018). Additionally, rayon is commonly found to be ingested by a variety of marine organisms from fish to bivalves (Lusher et al., 2013; Mishra et al., 2019). Up to 20,000 pieces of plastic per km^2 of surface water were found off the coast of Honduras in the Cayman Basin area, with concentrations exceeding 200,000 pieces per km^2 in the North Atlantic Subtropical Gyre (Law et al., 2010). Upon reaching the ocean, debris can travel for thousands of miles carried by ocean currents and wind, affecting marine life by spreading diseases and pollutants and by harming their digestive systems (Kim et al., 2015; Law et al., 2010; Sheavly and Register, 2007).

Of great concern is the capacity for plastics and fibers to facilitate the dispersal of diseases and pollutants which can harm marine life. Plastics contain a variety of endocrine disrupting additives, such as bisphenol A, polybrominated diphenyl ethers, phthalates, alkylphenols, and lead (Avio et al., 2017; Diana et al., 2020; Grün and Blumberg, 2007). These additives are capable of desorbing from the plastic based on factors including their water solubility, the pore size of the plastic, pH levels, and dissolved organic carbon fractions (Teuten et al., 2009). Microbes, including potentially pathogenic strains, are able to inhabit plastics as well (Harrison et al., 2014; McCormick et al., 2014; Zettler et al., 2013). The presence of microbes and organic contaminants on plastic is of particular concern due to plastics presenting a relatively new mode of spreading microbes, diseases, and contaminants throughout the ocean as they travel via ocean currents. The ecotoxicological effect that contaminated plastics can have on marine animals is also of interest, especially considering that microplastics and their affiliated toxicants can easily be consumed by creatures of any size, ranging from whales to zooplankton (Wright et al., 2013).

Microplastic and microfiber pollution can be consumed by oceanic organisms, leading to a myriad of health and fitness concerns. Around 10% of marine fish sampled in the Gulf of Mexico had ingested microplastics (Phillips and Bonner, 2015). Large organisms such as snakes, penguins, seals, turtles, and manatees consume microfibers, occasionally leading to respiratory blockages (Mishra et al., 2019). Marine filter feeders such as mussels and oysters also consume microplastics and fibers (Khan and Prezant, 2018; Li et al., 2018; Mishra et al., 2019) in the wild, while other marine organisms, such as lugworms and sea cucumbers, were shown to consume microplastics in the laboratory (Besseling et al., 2013; Graham and Thompson, 2009). Pelagic red crabs and giant larvaceans from Monterey Bay also ingested microplastics (Choy et al., 2019). Ingestion of microplastics is linked to reduced filtering activity, lysosomal membrane destabilization, and inflammatory responses in the blue muscle *Mytilus edulis* (Von Moos et al., 2012; Wegner et al., 2012). Additionally, without a mechanism to break down microplastics, accumulation of microplastics in digestive tracts can occur with the potential to be incorporated within the tissues of filter feeders and translocated to the circulatory system (Browne et al., 2008; Wright et al., 2013). Corals can consume plastic in the laboratory (Allen et al., 2017; Hall et al., 2015; Rotjan et al., 2019), and as heterotrophs, can also ingest plastics in their natural habitat (Rotjan et al., 2019). Since corals are already under significant stress, plastic consumption poses yet another risk to these vital organisms.

Numerous natural and anthropogenic factors, including climate change, tropical cyclones, pollution, and disease, contribute to the decline of coral cover and health on reefs worldwide (De'ath et al., 2012; Hoegh-Guldberg et al., 2007; Hughes et al., 2017b; Spalding and Brown, 2015). In the Caribbean Basin, coral cover has declined from 50% to about 10% between 1977 and 2001 (Gardner et al., 2003). Such declines in coral cover may be compounded by stress associated with plastic exposure and consumption, impacting other marine life as coral reefs are a valuable habitat for fish and invertebrates (Cole et al., 2008). Corals obtain nutrients through both heterotrophic and photoautotrophic methods. As heterotrophs, corals can ingest plankton, particles, and dissolved nutrients from the water column (Houlbrèque and Ferrier-Pagès, 2009), while also meeting up to 100% of their metabolic energy needs through the translocation of photosynthetic products from their endosymbionts (Grottoli et al., 2014). In the laboratory, corals can consume microplastics at a rate similar to their feeding rate of plankton (Hall et al., 2015) leading to clogged polyps (Allen et al., 2017), and possible starvation in corals that rely on heterotrophic carbon. In the coral *Pocillopora damicornis*, microplastic exposure resulted in a suppressed immune system, a down regulation of epidermal growth factor-extracellular signal-regulated kinase signal pathway genes, and an activation of the stress response system through the up regulation of genes (Tang et al., 2018).

Many studies have focused on the consumption of microplastics by corals in the laboratory (Allen et al., 2017; Hall et al., 2015; Hankins et al., 2018; Reichert et al., 2018), with a limited number of studies focusing on the interactions between corals and microplastics in their natural habitat. Woodall et al. (2014) qualitatively analyzed octocorals from the Indian Ocean with each sample containing microplastics and rayon in their surface mucus layer (Woodall et al., 2014). A study by Lamb et al. (2018) found that macroplastic accumulation (both hydrophobic and hydrophilic plastics) on corals had the potential to increase disease likelihood by twenty-fold (Lamb et al., 2018). Rotjan et al. (2019) found that temperate *Astrangia poculata* corals collected in Rhode Island contained over 100 microplastic particles per polyp and that corals fed plastic particles preferred these to brine shrimp eggs, confirming that microplastic ingestion may have ecological and physiological consequences for corals. Microplastics have the ability to further magnify the effects of numerous other anthropogenic factors, including overfishing, commercialized recreational activities, and chemical pollution, that are already stressing fragile coral communities (Hughes et al., 2017a; Sebens, 1994).

Here we quantify the abundance of microplastic and microfiber exposure by two species of coral on the Belize Mesoamerican Barrier Reef System (MBRS). We sought to investigate whether corals on the Belize MBRS contain microplastics and other microfibers, either on their surface or within their polyps, and how the abundance and type of microfiber changes geographically in relation to local scale drivers, particularly river inputs and ocean currents. Understanding the abundance, spatial distribution, and potential sources of microplastics on coral reefs will allow for improved local and regional scale management of plastic waste in these vital coastal ecosystems.

2. Methods

2.1. Study location

The Mesoamerican Barrier Reef System (MBRS) is the most expansive barrier reef system in the Western Hemisphere, extending 1200 km from the tip of the Yucatan Peninsula in Mexico through the entire coast of Belize, and down to the Islas de la Bahia (Bay Islands) off of northern Honduras. Coral samples were collected from back reef (shoreward side of the reef crest) sites on the barrier reef off the coast of Belize with permission from the Belize Fisheries Department under a Marine Science Research permit (# 0000035-15) granted to KDC and JHB.

2.2. Sample collection

In total, eighteen 1″ diameter coral core samples of *Siderastrea siderea* ($n = 12$) and *Pseudodiploria strigosa* ($n = 8$) collected in 2015 at 2 to 5 m depth along the Belize MBRS were utilized in the current study. Sampling sites consisted of Drowned Cayes Reef, Long Caye Reef, Gladden Spit Reef, and Sapodilla Cayes Reef (Table 1; Fig. 1).

Coral samples consisted of *Siderastrea siderea* (*S. siderea*; $n = 11$ for abundance counts) and *Pseudodiploria strigosa* (*P. strigosa*; $n = 4$ for abundance counts). The coral cores were collected with a pneumatic drill, transported to the boat by divers, rinsed in seawater, stored in capped PVC tubes containing ethanol, and transported to the University of North Carolina at Chapel Hill (UNC) for analysis. Once at UNC, cores

Table 1
Sampling regime.

Sample site	N	ATR-FTIR analysis (n)	Abundance counts (n)
Drowned Cayes Reef	6	6	3
Long Caye Reef	3	3	3
Gladden Spit Reef	6	6	5
Sapodilla Cayes Reef	5	5	4
Total	20	20	15

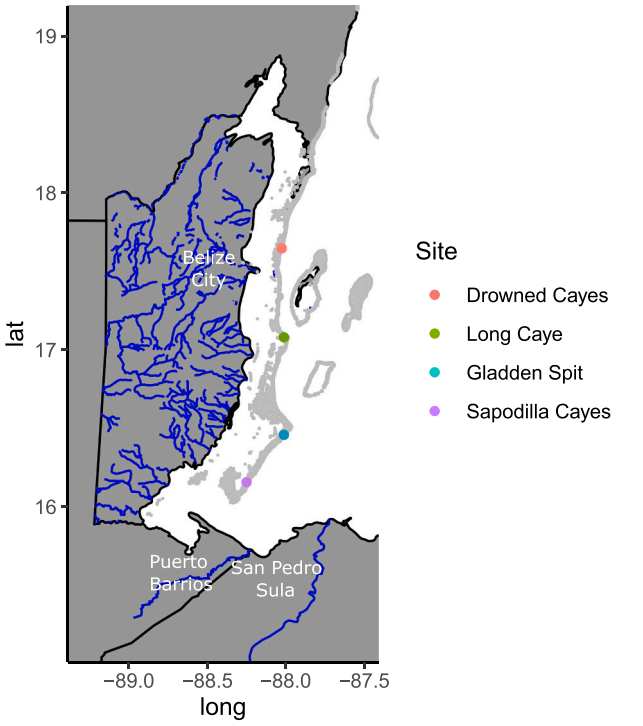

Fig. 1. Geographical location of each site from which samples were collected off the coast of Belize.

were rinsed in ethanol, allowed to air-dry and stored in a sealed polycarbonate box (all cores used in this analysis were stored in the same box). Field collection, transport, and storage were all possible sources of contamination. In this case, all core samples were treated in the same way, though contamination was still possible. Of note, no PVC was reported in any of the samples (Fig. 4). As the field and lab storage vessels were made of PVC, this suggests contamination via storage was likely minimal. Care was taken in the laboratory to minimize contamination in sample preparation and analysis (see Section 2.6).

2.3. Sample preparation

A rectangular piece of dried coral tissue and skeleton with a volume of at least 1 cm^3 was sectioned from each core using an Inland DFS-100 ReefKeeper Saw. Samples were cut in rectangles, allowing for the quantification of two-dimensional surface area using calipers. Coral samples were then placed in 10% HCL for up to 12 h to dissolve the calcium carbonate skeleton (Rotjan et al., 2019; Susic et al., 1991). When the entire coral skeleton was dissolved the sample was rinsed with Milli-Q water, filtered using a 30 μm filter, and then back washed into a sterile glass vial (Rotjan et al., 2019). Certified ACS Sodium hydroxide (NaOH) (100–300 ml) was then added to the vial to digest any organic tissue residue (Catarino et al., 2017). Vials were placed in a 60 °C drying oven for at least 12 h to allow for digestion of organic material (Catarino et al., 2017). Following this incubation, all samples were rinsed thoroughly with Milli-Q water onto a 30 μm sieve (ATM Corp) to remove NaOH residue. The cleaned sample was back washed into a glass vial. To assess fiber loss due to this filtration and back washing method, 10 replicate samples each containing 10 2 mm fibers were run through the methods resulting in a mean fiber recovery rate of 84%. Contents of the vial were pipetted onto a glass slide and placed in the drying oven set at 60 °C until the sample was completely dried onto the slide. On average, drying took about 12 h. Five laboratory method controls were created by running samples of Milli-Q water through the above procedure to determine the number of microfibers accumulated due to the sample preparation and analysis methods. The dried tissue layers of all twenty samples were analyzed using Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR-FTIR), while a sub-sample was used for microfiber abundance counts (Table 1).

2.4. Microfiber abundance counts

Microfiber counts were conducted using an OLYMPUS BX51 polarized light microscope. For each sample and control, every microfiber on the slide was counted. We defined microfibers as any fibrous object on the slide (Fig. 2). Due to the NaOH incubation, any organic matter should have been digested (Catarino et al., 2017). Thus, any objects that survived the NaOH incubation should be non-organic and are most likely not naturally found on, or within, the corals. Counts were conducted in triplicate for each sample. The mean value of microfibers counted on all control slides, 124.47 microfibers, was subtracted from the mean microfiber values of each slide. These values were then divided by the surface area of the sample to calculate the number of microfibers per cm^2. While non-fibrous contaminants were found, they were not quantified due to zoom and contrast limitations of the microscopes used in this study. As such, this study reports only on microfiber abundance and identification.

2.5. Microplastic and microfiber identification

To identify the types of contaminants in our samples we randomly selected on average 6 fibers per sample and used an Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR-FTIR) microscope (Smiths IlluminatIR coupled to an Olympus microscope) to produce infrared spectra for polymer identification. ATR-FTIR was selected for analysis as it has been used to identify polymers found in corals in previous studies (Hall et al., 2015; Rotjan et al., 2019). The

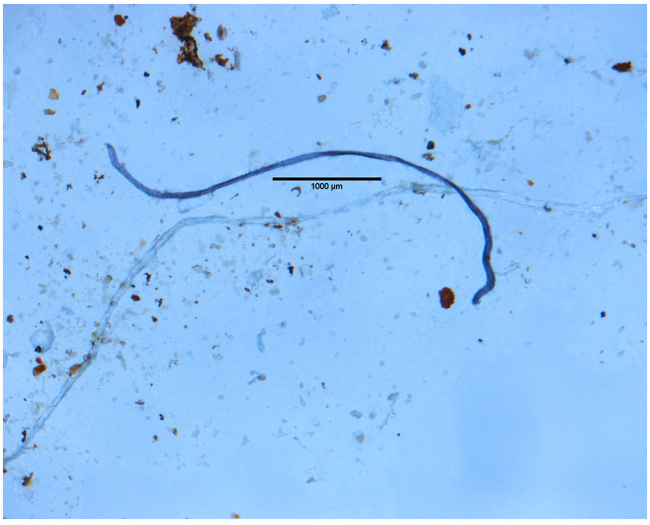

Fig. 2. Blue and translucent microfibers from *Siderastrea siderea* - both identified as rayon by ATR-FTIR analysis. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

ATR-FTIR was set to 4 cm^{-1} resolution, Objective 36×-ATR, full spectral range 650–4000. The spectra were read by an integrated software (Spectral ID) which determined possible matches by comparing the spectral peaks to the Polymer, Polymer Additives, and Plasticizers library from Thermo-Fisher Scientific. Spectral matches with a confidence greater than 60% were considered as positively identified, following cutoff values from Woodall et al. (2014). We analyzed spectral matches for objects found in 20 coral samples.

2.6. Laboratory contamination prevention

All researchers working with the samples wore natural fiber clothing (e.g., cotton) and nitrile gloves (Woodall et al., 2014). Any glassware used was washed with soap and water, placed in a 10% HCl acid bath, rinsed in Milli-Q water, dried, and baked at 400 °C for 4 h to remove all residues. The lab bench and fume hood were cleaned with ethanol to minimize the abundance of microfibers and other contaminants that may have accumulated from previous use. Tools, solutions, and samples not in use were covered or stored to reduce their exposure to the air. The 30 μm filter used was kept wrapped in aluminum foil when not in use and was rinsed thoroughly with Milli-Q water between each sample. All glass vials containing the samples were covered by watch glass covers while coral samples were being dissolved or while being heated in the oven. Before drying down the samples onto the glass microscope slides, the drying oven was cleaned and the racks were covered with aluminum foil. Opening of the oven door while samples were drying was reduced to minimize airflow into the oven. Finished slides, along with the control slides, waiting to be analyzed were stored in slide holder folders made out of pressed cardboard to reduce their exposure to the ambient environment.

2.7. Statistical analysis

Average microfibers per cm^2 ± standard error were calculated for each site. A two-way analysis of variance (ANOVA) was used to assess the effects of coral species and site on microfiber abundances (Table 2). If factors were found to be significant ($p < 0.05$) in the ANOVA, post hoc Tukey's HSD tests were used to evaluate the significance of each pairwise comparison.

3. Results

3.1. Microfiber counts

Every coral sample contained microfibers. The mean number of microfibers found in each coral sample ($n = 15$) was 165 ± 50 (SE) fibers per cm^2. Mean microfiber abundances for each individual sample (counted in triplicate) are available in Table S1.

3.2. Effects of species and collection site on microfiber counts

There were significant effects of species, collection site, and the interaction of species and collections site on the average abundances of microfibers in coral samples (Table 2; Fig. 3). *Pseudodiploria strigosa* corals had greater abundances of microfibers than did *S. siderea* corals. Average microfiber abundances (mean ± SE) were greater on the Sapodilla Cayes Reef (296 ± 89) southern Belize than on the Drowned Cayes Reef (75 ± 14) northern-central Belize while no significant differences were measured between the Long Caye and Gladden Spit corals and any other sites (Table 3; Fig. 3). Within the Sapodilla Cayes samples, *P. strigosa* corals contained significantly more microfibers than did *S. siderea* corals. In addition, *P. strigosa* corals at the Sapodilla Cayes contained greater abundances of microfibers than any of the corals at all of the other sites (Table 3; Fig. 3).

Table 2
Results of analysis of variance (ANOVA) to test the effect of species, site, and species:site on microfiber abundance.

Treatment	Df	Sum Sq	Mean Sq	F value	*p*-Value
Species	1	232,620	232,620	10.901	0.002
Site	3	207,338	69,113	3.239	0.033
Species:Site	2	346,187	173,094	8.112	0.001
Residuals	38	810,869	21,339		

3.3. ATR-FTIR identification

A total of 119 microfibers were positively identified with confidence greater than 60%. Out of the microfibers sampled, the most prominent positively identified contaminant was rayon. Rayon accounted for about 84.9% of the fibers identified ($n = 101$) (Fig. 4). Additionally, rayon was the most frequently sampled microfiber across geographical location (Fig. 4). The second most sampled contaminant was fiberglass (10.9%), and the third most sampled contaminant was Nylon (1.68%) (Fig. 4). A majority of the microfibers were a semi-transparent white color, while other fibers were either blue, pink, red, black, or purple. Controls were also contaminated with small quantities of rayon. Beyond rayon, no other fiber types were identified on control samples though it is possible that other types of fiber were present in minute proportions and were just not identified with ATR-FTIR.

4. Discussion

4.1. Corals interact with microfibers, including microplastics, on the Belize MBRS

Through the analysis of dried coral tissue samples, we show that corals do indeed interact with microplastics and microfibers on the Belize MBRS (Figs. 3 & 4). Past tank-based experiments have shown that corals can ingest microplastics, strongly suggesting their ability to ingest microplastics in their natural habitat (Allen et al., 2017; Hall et al., 2015; Rotjan et al., 2019). This interaction may occur either through ingestion or entrapment in the coral mucus layer. Every coral sample analyzed in this study contained microfibers (158 ± 30, mean ± SE), indicating that coral-fiber interactions are ubiquitous and quite abundant on the Belize MBRS. Microplastic exposure through either surface contact or ingestion increases the stress response of corals and can clog their polyps, potentially affecting their feeding rates and overall health (Allen et al., 2017; Tang et al., 2018). With the increasing abundance of single use plastic and a lack of proper waste management, the presence and effects of microplastics on marine life is of growing global concern.

4.2. Microfiber abundance may be coral species specific

Pseudodiploria strigosa corals contained a greater abundance of microfibers than *S. siderea* corals (Table 2; Fig. 3). This may be due to variance in morphology, polyp size, and/or surface area roughness causing differing numbers of particles to be trapped in the mucus layer or polyps (Veron and Stafford-Smith, 2000a,b). While both species have a mounding morphology, *P. strigosa* is a brain coral with surface grooves that contain polyps while the polyps of *S. siderea* are large and sit on a relatively smooth surface structure. These surface structure differences may alter the dynamics of the diffusive boundary layer surrounding these corals (Jimenez et al., 2011). Morphological differences have been shown to impact prey capture (Sebens et al., 1998), indicating that they may also impact interactions with pollutants such as microfibers. Such differences in microfiber abundance may lead to some coral species being more negatively affected by the presence of microfibers than others. Corals at the Sapodilla Cayes contained the greatest abundance of microplastics and *P. strigosa* corals at Sapodilla Cayes contained more microfibers than did *S. siderea* corals (Table 3; Fig. 3). These results suggest that *P. strigosa* may be more prone to interaction with

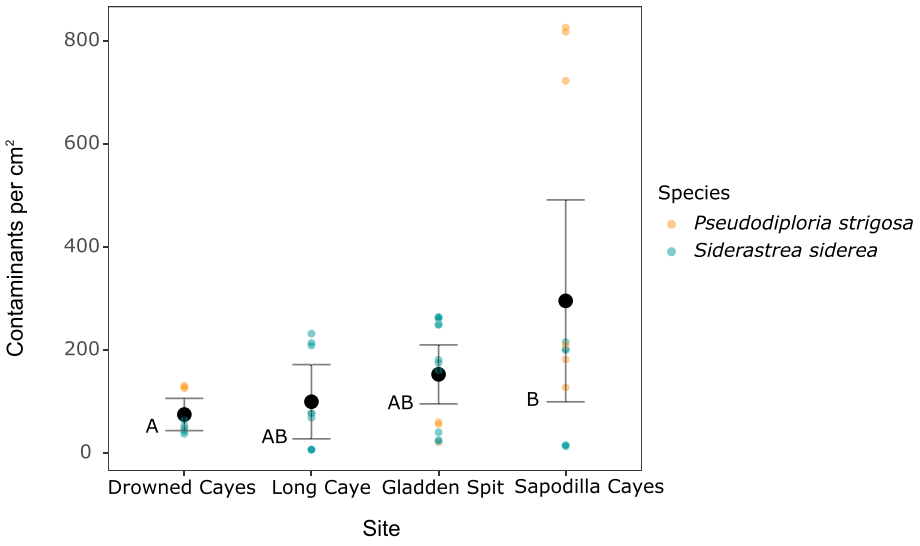

Fig. 3. Mean number (±SE) of microfibers per surface area (cm^2) for each coral sample collection site. Symbols (A, B, AB) show statistically significant differences between sites ($p < 0.05$).

Table 3
Significant results of Tukey HSD test to assess the effect of species, site, and species:site on microfiber abundance.

Treatment		Df	Lower	Upper	*p*-Value
Species	*Siderastrea siderea* - *Pseudodiploria strigosa*	−162	−262	−62.9	0.002
Site	Sapodilla Cayes - Drowned Cayes	193	20.4	366	0.023
Species: Site	*Pseudodiploria strigosa*: Sapodilla Cayes - *Pseudodiploria strigosa*: Drowned Cayes	352	21.4	684	0.030
	Pseudodiploria strigosa: Sapodilla Cayes - *Siderastrea siderea*:Drowned Cayes	433	162	703	<0.001
	Pseudodiploria strigosa: Sapodilla Cayes - *Siderastrea siderea*:Long Caye	381	134	628	<0.001
	Pseudodiploria strigosa: Sapodilla Cayes - *Pseudodiploria strigosa*: Gladden Spit	435	104	766	0.003
	Pseudodiploria strigosa: Sapodilla Cayes - *Siderastrea siderea*:Gladden Spit	301	67.1	535	0.004
	Siderastrea siderea:Sapodilla Cayes - *Pseudodiploria strigosa*:Sapodilla Cayes	−371	−641	−100	0.002

microfibers than *S. siderea*, though this result is not replicated across our other sites.

4.3. Rayon is a prominent contaminant

The most prominent type of microfiber identified from coral samples in this study was rayon (Fig. 4). Rayon is a regenerated cellulose product manufactured from naturally occurring polymers and is semi-synthetic. Despite being non-plastic, rayon was included in our results since it is a fibrous man-made object that is quite abundant within the oceans, with one study finding that over 50% of fibers seen in deep-sea sediment cores and on octocorals from South-West Indian Ocean were composed of rayon (Woodall et al., 2014). Rayon is commonly used in textiles and clothing and enters the oceans through waste water, including effluent from washing machines and textile washing (Miller et al., 2017; Mishra et al., 2019; Sheavly and Register, 2007; Woodall et al., 2014). Rayon has been identified as the most numerous contaminant in other studies focused on micro-debris in oceanic fish (Lusher et al., 2013), water, and sediments (Woodall et al., 2014). Additionally, in a study of coastal sites along the southern United States a large portion of microfibers were rayon (Yu et al., 2018). Rayon seen in this study is likely sourced from waste-water effluent from surrounding coastal areas. As such, monitoring of and improvement of wastewater treatment and effluent streams with a focus microfiber capture at the source are likely to decrease microfiber pollution on the MBRS.

A few of our samples also contained nylon, which is a common synthetic polymer used for making fabrics, including fishing line and braided ropes used in boating. Nylons are a group of plastics known as polyamides that have been found in coastal regions across the world, especially near sewage disposal sites (Browne et al., 2011; Kim et al., 2015; Wessel et al., 2016). Other plastics and contaminants identified within our samples included glass fiber (fiberglass), a polymer reinforcer used on boats and at marinas, and rubber fiber, found in elastic yarn and clothing.

Due to the sheer variety of plastics, plasticizers, and other contaminants found in our samples, marine debris is likely accumulating from a variety of anthropogenic sources along the MBRS. Despite rayon being widely reported as an ocean contaminant, little is known about the effects this man-made fiber can have on corals and other animals. Microplastics can become stuck in the polyps of corals (Allen et al., 2017), raising the question as to whether or not rayon can clog polyps as well, possibly leading to starvation. The ability of microfibers to adhere to the surface of corals and become ingested by their polyps has the potential to negatively affect photosynthesis rates of Symbiodiniaceae genera in corals. The aforementioned possibilities could allow rayon and other microfibers to negatively impact both the heterotrophic and photoautotrophic feeding of corals, thus establishing another stressor impacting coral communities. Further research must be undertaken to better understand how these contaminants are affecting corals, where the contaminants are originating from, and how to best mitigate these contaminants.

4.4. Distribution of microfibers across the Belize MBRS reveals a geographic gradient in microfiber abundance

Ocean currents are the driving force of microplastic distribution worldwide, while local scale point sources, such as coastal cities and

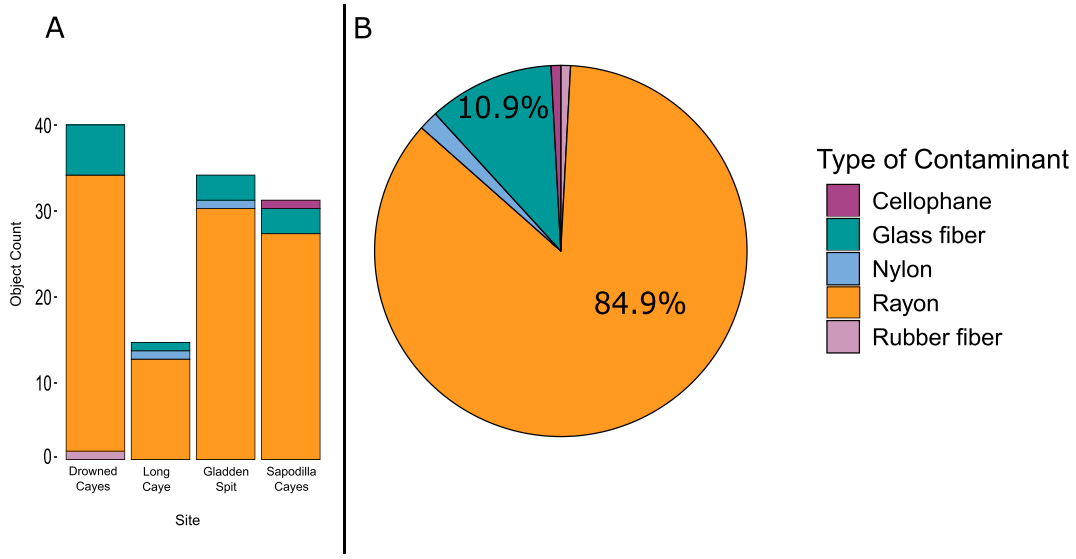

Fig. 4. ATR-FTIR spectroscopy identification of a subset of contaminants within twenty coral samples. A) Sample identification by location, and B) total abundance of five different contaminants identified in our subset of contaminants (spectral ID match of >60%).

rivers, are also substantial contributors. As such, we analyzed variation in the abundance of microfibers across geographic locations. We found that microfiber abundance was significantly ($p < 0.05$) greater in corals located on the Sapodilla Cayes (296 ± 89; mean ± SE) microfibers per cm^2 than in corals located 170 km north at the Drowned Cayes (75 ± 14) microfibers per cm^2 (Fig. 3). Additionally, we found that *P. strigosa* corals on the Sapodilla Cayes had greater abundances of microfibers than did *S. siderea* corals at the same site, however, our results are limited by a low sample size of *P. strigosa* corals (Table 3; Fig. 3). This suggests that microfiber abundance in corals may not only depend on the microfiber abundance in the surrounding seawater, but also on the specific coral species and morphology as well. It is likely that the most influential factors resulting in this geographic gradient are population density, watersheds, and the Honduras Eddy.

Belize has the lowest population density in all of Latin America (359,000 people in 2015). The country's most populous city, Belize City, is located on the coast southwest of the Drowned Cayes site used in this study. The Drowned Cayes Reef experiences a prevailing northward current, as the site resides just below where the Caribbean and Yucatan Current meet (Centurioni and Niiler, 2003; Sheng et al., 2005). Low population density, and possibly shorter water retention time due to a northward current (Centurioni and Niiler, 2003; Sheng et al., 2005), most likely account for the lower abundance of microfibers compared to the Sapodilla Cayes Reef, despite Belize's most populous city being nearby.

Each of the factors causing debris to enter the Gulf of Honduras, including improper waste management (Browne et al., 2011), watersheds, coastal towns, and ports (Nel et al., 2017), may be contributing to the significantly greater abundance of microfibers and other debris found within the coral samples from the Sapodilla Cayes compared to those on the Drowned Cayes. The Honduran Province of Cortés in northwest Honduras (~50 km southeast of the Sapodilla Cayes) contains the largest population density per km^2 in all of Honduras, roughly 26 times larger than the average population density in Belize. This province is located within the Ulúa watershed, adjacent to the Gulf of Honduras, and accounts for the greatest amount of sediment delivery by watershed to the Mesoamerican Barrier Reef System near the Sapodilla Cayes Reef, with an estimated 100 million metric tons of sediment delivered annually to the northern coast of Honduras (Burke and Sugg, 2006). In comparison, the largest annual sediment delivery from a watershed in Belize is estimated to be just 3 million metric tons, two orders of magnitude lower than that of largest sediment delivery values in Honduras (Burke and Sugg, 2006). Due to population density, sediment discharge, and river discharge in Northern Honduras, it is likely that this area serves as a larger source of debris, including microplastics and microfibers, than does Belize.

Compounding the impacts of this larger debris source on the Sapodilla Cayes relative to the Drowned Cayes is the local oceanography. The cyclonic Honduras Eddy likely causes a recirculation of debris filled water over the reef. In fact, a study by Paris and Chérubin (2008) determined that a majority of the buoyant matter found offshore on the southern portion of the MBRS originated from the northern watersheds of Honduras, including from the Ulúa and Aguán basins, with buoyant matter being retained within the Gulf of Honduras due to the Honduras Eddy (Paris and Chérubin, 2008). The Honduras Eddy is fed by a current traveling across the coast of Honduras, which has the potential to accumulate various debris from sources, such as rivers, coastal towns, and the eastern Caribbean (Carrillo et al., 2017; Cole et al., 2011; Sheng et al., 2005). As ocean currents dictate the distribution of microplastics in the ocean (Browne et al., 2010; Kim et al., 2015; Law et al., 2010), we believe that the cyclonic effects of the Honduras Eddy may lead to elevated microfiber debris levels in the Gulf of Honduras, including the Sapodilla Cayes.

Although the greater accumulation of microfibers in corals from the Sapodilla Cayes Reef may be due to the water current patterns in the Gulf of Honduras rather than population density, we cannot ignore the potential sources of pollution that the riverways, coastal towns, and ports of Honduras, Guatemala and Belize pose. Likewise, we cannot hold these three nations as solely responsible for the debris in the Gulf of Honduras considering that debris in the ocean can travel thousands of miles at the liberty of currents. In the case of the Gulf of Honduras, the Honduras Eddy and population density are most likely augmenting one another's effects on the concentration of microplastics and other debris on the southern portion of the Mesoamerican Barrier Reef System. Of importance is the potential for the Honduras Eddy to increase the likelihood of diseases, chemicals, and organic pollutants being transported to the Gulf of Honduras by microplastics due to accumulation and cycling of debris. Further research needs to be conducted to determine if increased residence of time of the debris leads to an increase in desorbed chemicals, such as plasticizers.

5. Conclusion

Corals are interacting with microplastics and microfibers in their

natural habitats, not just in the laboratory, indicating the potential for coral communities to be harmed by these contaminants. All of our coral samples contained multiple microfibers, even after subtracting out control values which accounted for ambient contaminants from the laboratory. The most frequently measured microfiber we sampled was rayon, reiterating the strong presence of rayon as a contaminant in the oceans. However, other contaminants including glass fiber and nylon were also found. We found an uneven geographical distribution of microfibers across the coast of Belize, with the Sapodilla Cayes corals containing a significantly greater abundance of microfibers than the Drowned Cayes. Geographical variance in abundance is most likely caused by the combination of population density, nearby terrestrial point-sources, and ocean currents, while microfiber uptake may be influenced by the type of coral species. Further research needs to be conducted on how the presence of microplastics and other debris is affecting the Mesoamerican Barrier Reef System and how it may disrupt coral physiology. Since many corals are able to consume objects within the water columns, our microfiber counts have the potential to serve as a proxy for the abundance of anthropogenic contaminants within the water columns at various sites. Our analysis presents a vital baseline for microfiber abundance on the MBRS that can be used for future monitoring projects, including temporal research. Ultimately, understanding the abundance, spatial distribution, and potential sources of microplastics on coral reefs will allow for improved local and regional scale management of plastic waste in these vital coastal ecosystems. As rayon is typically found in clothing fibers and enters the ocean through wastewater effluent, modifications to wastewater systems may limit future rayon pollution in coastal waters.

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CRediT authorship contribution statement

Kirsi Oldenburg: Conceptualization, Methodology, Software, Formal Analysis, Investigation, Writing – Original Draft, Visualization, Funding Acquisition; Juanita Urban-Rich: Conceptualization, Methodology, Validation, Resources, Writing- Review & Editing, Supervision; Karl Castillo: Validation, Resources, Writing – Review & Editing, Supervision; Justin Baumann: Conceptualization, Methodology, Software, Validation, Formal Analysis, Investigation, Resources, Data Curation, Writing – Review & Editing, Visualization, Supervision, Project Administration, Funding Acquisition.

Declaration of competing interest

The authors declare that no conflicts of interest exist.

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