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Patterns, dynamics and consequences of microplastic ingestion by the temperate coral, *Astrangia poculata*

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Microplastics (less than 5 mm) are a recognized threat to aquatic food webs because they are ingested at multiple trophic levels and may bioaccumulate. In urban coastal environments, high densities of microplastics may disrupt nutritional intake. However, behavioural dynamics and consequences of microparticle ingestion are still poorly understood. As filter or suspension feeders, benthic marine invertebrates are vulnerable to microplastic ingestion. We explored microplastic ingestion by the temperate coral *Astrangia poculata*. We detected an average of over 100 microplastic particles per polyp in wild-captured colonies from Rhode Island. In the laboratory, corals were fed microbeads to characterize ingestion preference and retention of microplastics and consequences on feeding behaviour. Corals were fed biofilmed microplastics to test whether plastics serve as vectors for microbes. Ingested microplastics were apparent within the mesenterial tissues of the gastrovascular cavity. Corals preferred microplastic beads and declined subsequent offerings of brine shrimp eggs of the same diameter, suggesting that microplastic ingestion can inhibit food intake. The corals co-ingested *Escherichia coli* cells with microbeads. These findings detail specific mechanisms by which microplastics threaten corals, but also hint that the coral *A. poculata*, which has a large coastal range, may serve as a useful bioindicator and monitoring tool for microplastic pollution.

1. Introduction

Plastic debris has been found on shorelines globally [1], and in remote locales including the Antarctic and Arctic, remote islands, and the deep sea (e.g. [2–12]). First noted as a potential marine problem in 1971 [13,14], plastics now reliably constitute 70% of marine litter in some areas [15]. There is no consensus on the total amount of plastic in the ocean, though models predict 4.8–12.7 million tons of plastics are added each year [16], with total accumulation by 2025 predicted to be at least 155 million tons [17]. Because plastics degradation can take 500–1000 years, nearly all plastic created on Earth still exists, often weathered down to smaller microplastics, particles less than 5 mm [18,19]. Microplastics sources include clothing, car tyres, city dust and personal care products [20], and they are transported via road runoff, wastewater, winds and waterways [19]. Because microplastics are ubiquitous [1], a critical understanding of how and when they impact marine organisms is urgently needed.

Recent studies have shown that diverse organisms ingest microplastics (e.g. [2,21,22]), and microplastics have the potential to be transferred through the

food web [23–26]. Post-ingestion, organisms may suffer from reduced nutrition, starving to death with full digestive systems [27]. Microplastic ingestion may harm organisms through chemical desorption of plasticizers, metals or organic contaminants adsorbed to the plastic [28–30]. Microplastics have been found in the digestive tracts of invertebrates and fishes from diverse habitats (e.g. [23,31]), with lethal or sublethal effects [29]. Microplastics can aggregate surface microbial biofilms, harbouring diverse communities (the 'plastisphere') distinct from surrounding seawater and particulate organic matter [32]. Microplastics enrich seawater with microbes that can hydrolyse hydrocarbon polymers [32] and are putative pathogens [32–34]. Ingestion of microplastics can thus result in increased dispersal of novel, rare seawater microbes. The ability for microplastics to vector novel microbes into consumers and subsequently transport plastisphere microbes throughout the food web is poorly understood.

Coral reefs are among the most biodiverse regions in the ocean, containing 25–33% of known marine life [35]. Macroplastics concentrations have recently been found to correlate with coral disease incidence [36]. Corals can ingest microplastics [37,38], but ingestion dynamics are probably species-specific [39]. Some corals bleach and experience tissue necrosis, whereas others produce mucus, overgrow particles or attach particles to themselves. The cold-water coral *Lophelia pertusa* experienced reduced skeletal growth rates when exposed to microplastics [40], whereas microplastic ingestion had no effect on calcification in two shallow Caribbean reef species [41]. In response to microplastic exposure, *Pocillopora damicornis* demonstrated increased stress and diminished immune function [42], but *Porites lutea* was able to upregulate mucus production and showed no negative health effects [39]. Microplastic exposure on cnidarian-algal symbioses is also varied, ranging from no change in symbiont density [42] to disruption of symbiosis [43], with bleached *Aiptasia* anemones showing decreased ability to discriminate against microfibre ingestion [44]. Coral response mechanisms to microplastics are not well understood, but some corals may use chemoreception to preferentially ingest microplastic particles [38]. It is likely that microplastics will be found in almost every animal taxon examined, creating an imperative and urgent need to understand the patterns, dynamics, mechanisms and consequences of ingestion.

A recent United Nation Environment report highlighted the critical need for monitoring, novel assays, understanding ingestion risk and consequence, investigating microplastics as a pathogenic vector and characterizing microplastic pollution patterns and ecological relevance [45]. Therefore, a system for microplastic monitoring is urgently needed. The northern star coral, *Astrangia poculata*, is a novel model for addressing urban water quality and aquatic pollution issues because it is heterotrophic, sessile and highly tolerant of diverse environmental conditions (unlike many shellfish or other sessile benthic invertebrates [46–48]). Their range includes United States (US) coastal habitats from northern Buzzards Bay (MA) to the Gulf of Mexico (FL) [49]. It also occurs along the US Gulf Coast and Bermuda and has been found to survive long-distance transport on plastic rafts [50] and turtles [51].

The objectives of this study were: (i) to determine if *A. poculata* ingests microplastics in the wild, (ii) to examine patterns of microplastics ingestion and egestion in the laboratory, and (iii) to identify consequences of microplastic ingestion on subsequent feeding or as a vector for novel microbes.

2. Material and methods

(a) Microplastics ingestion in the wild

Four colonies of *A. poculata* were collected and immediately preserved in 4% zinc-based formalin/filtered seawater (Z-fix; Anatech, Ltd). In the laboratory, fixed corals were removed from Z-fix using metal forceps, rinsed with ultraclean deionized water (DI) to remove surface debris, air-dried in a covered glass Petri dish and weighed. Polyp number was counted. Corals were then decalcified with a 0.9% HCl solution, filtered through a 20 µm sieve, rinsed and backwashed with ultraclean DI water into a clean glass beaker, pipetted onto a glass microscope slide, air-dried in a laminar flow hood [52] and imaged with polarized light (Olympus SZX12). All particles counted were 40 µm or larger and were characterized into fibres, round (ovoid) or miscellaneous shape [53]. Polymer identification was completed on the first 50 pieces encountered in each sample using a Smiths IlluminatIR II, attenuated reflectance (ATR) Fourier transform infrared spectroscopy (FTIR) with an attached Olympus scope. FTIR spectra were obtained in transmission mode and CO_2 interference was removed for clarity. Spectra were compared to standards in the ThermoFisher Scientific HR Polymer Additives and Plasticizers Library. Only particles 60 µm or larger were analysed, using the same glass slide. Control (blank) 10 ml HCl samples were processed similarly to determine procedural contamination. Control counts were subtracted from sample counts (averaging 2–8% counts removed) before samples were normalized per polyp. Controls averaged 55% cellulose (cotton), 30% nylon, 10% polyester, 5% polypropylene. An average of three polyps was dissected per colony.

(b) Collection and husbandry

Aposymbiotic *A. poculata* specimens were collected in June 2016 at Fort Wetherill Park in Jamestown, Rhode Island (RI) (41°28′40″ N, 71°21′34″ W) using SCUBA (6–10 m depth), which lies downstream of RI's largest urban centre, Providence and experiences substantial recreational and industrial shipping traffic. Underwater samples were placed into sterile whirlpak bags until processing. Aposymbiotic (white) corals were collected because they rely almost entirely on heterotrophy for nutrient acquisition [54], and the light tissue colour is ideal for imaging. In all cases, prior to experimentation, corals were laboratory-acclimated for two weeks. During acclimation, all visible epibionts (sponges, polychaetes, algae) were removed. Lighting was provided by 2 bulb high output (HO) T5 fluorescent fixtures (Hamilton Technology, Gardena, CA, Aruba Sun T5-V series) each housing a 10 000 k daylight and a 420 nm actinic bulb on a 12 L : 12 D cycle. Photosynthetically active radiation was measured (Seneye Reef Monitor), averaging 89.6 µmol m^{-2} s^{-1} ± 10.8 µmol m^{-2} s^{-1} during daylight conditions. Colonies were separated by at least 5 cm to limit intercolonial antagonism. During acclimation, colonies received targeted ad libitum feedings of copepods (Cyclopeeze, JEHM co., Inc.), with copepod length averaging 0.729 mm (s.d. = 0.297 mm). Tanks were scrubbed and siphoned to remove detritus and algae.

Colonies were fragmented to 15–30 polyps each, then allowed to recover for at least 48 h. For feeding experiments, colonies were maintained in a flow-through aquarium rack system with replicate 1 l chambers, each independently aerated, containing 500 ml of ultraviolet (UV)-sterilized, particulate-filtered seawater from Boston Harbor at 18°C in the New England Aquarium (NEAq).

(c) Microbead ingestion

Throughout, 'microbead' refers to UV-fluorescent blue polyethylene spheres (1.13 g/cc; Cospheric, LLC). Beads were pre-acclimated for 4–8 h in seawater to facilitate non-clumping; this presumably cultivated a local biofilm on beads.

Ten *A. poculata* colonies ranging between 17 and 30 polyps were simultaneously exposed to approximately 0.1 g microbeads ($X = 200.2$ µm diameter, ranging 170.5–230.8 µm) and approximately 0.1 g brine shrimp eggs (BSE) ($X = 230.6$ µm diameter, 190.7–260.3 µm) in individual 500 ml chambers for 15 min.

Particle dose was determined via microscopic quantification of particles (0.1 g of each food was equivalent to approx. 8500 particles). After feeding, corals were fixed immediately in Z-fix, decalcified and dissected for scoring. Each polyp was splayed to expose the gastrovascular cavity. Number of microbeads and BSE per polyp was scored on a dissecting stereomicroscope (Leica M165 FC). Polyp diameter was measured, and the polyp volume was calculated based on measured polyp diameter and height.

(d) Localization of ingestion

Replicate colonies ($n = 15$) in individual chambers were exposed to 0.1 g of microbeads (500 ml of seawater) with 10 ml of Cyclopeeze copepod effluent (copepod-free seawater conditioned with Cyclopeeze) to stimulate feeding behaviour for 90 min. Corals were then transferred into clean, independent 500 ml tanks for recovery periods of 0, 30, 60 and 90 min, and 24 h ($n = 3$ corals for each of the five recovery periods). Corals were immediately fixed and decalcified after recovery for dissection and scoring. Polyps were visually partitioned into three areas of the gastrovascular cavity for microbead characterization: top (mouth to the beginning of the gastrovascular cavity), middle (beginning of the gastrovascular cavity to the beginning of the septa) and bottom (beginning to the end of the septa). The number of microplastic beads in each area was scored. Microbead consumption was calculated by multiplying the average number of beads per polyp (15 polyps over three colonies, for each of the 15 colonies (45 polyps total)) by the average number of polyps per colony.

(e) Microplastics as a barrier to food intake

Twelve corals were fragmented into two pieces, for a total of 24 colonies each totalling 15–30 polyps. One fragment from each colony pair was exposed to 0.1 g BSE for 15 min; the other was exposed to 0.1 g microbeads for 15 min. A subset of three corals from each treatment was immediately fixed. Experimental corals (nine pairs fed initially microbeads or BSE) were then placed into new chambers and equally distributed to exposure of 0.1 g of either live brine shrimp, frozen Cyclopeeze or BSE for another 15 min before being fixed, decalcified and dissected. Polyp dimensions were measured.

Particle type (microplastics, BSE, live brine shrimp and frozen Cyclopeeze) in dissected polyps was scored. To verify that images could be used to accurately count particles, 34 polyps were dissected, imaged and scored for food items. Image counts were compared to manual food item counts (counted as they were being dissected). The absolute value of the difference between images and dissections was less than 1 item (mean difference was 0.9, s.e. = 0.28), suggesting the two methodologies are comparable for quantifying food items; however, we determined that photos were the only means to accurately quantify live brine shrimp and frozen copepods, as full dissections tore the prey items beyond recognition.

(f) *Escherichia coli* biofilms on microbeads

Competent *Escherichia coli* cells (50 ml) were thawed on ice from −80°C and transformed with pAM239-GFP (pMMB-derived vector encoding green fluorescent protein (GFP) and chloramphenicol resistance). Liquid cultures of GFP+ *E. coli* were grown overnight in Luria Broth (LB) on a shaking incubator at 37°C/5% CO_2. After overnight growth, cultures were diluted to $OD_{600} = 1.0$. Microbeads were added to the GFP+ *E. coli* in a sterile glass culture tube (100 µl microbeads in 1 ml culture), and soaked in liquid culture for 48 h in a stationary incubator at 37°C/5% CO_2. Cultures were then transferred to room temperature (25°C) for 7 days. Non-biofilmed microbeads were incubated in sterile LB broth and rinsed three times in phosphate buffered saline (PBS). Biofilm formation on the microbeads was confirmed via fluorescence stereoscope (Leica M165FA) and confocal (Zeiss LSM880 Airyscan) imaging (electronic supplementary material, figure S1). GFP+ *E. coli* biofilm-coated beads were visualized by stereoscope (Leica M165FA) and selected via a sterile 200 µl micropipette. To minimize LB medium and unattached cells, microbeads were rinsed twice with 10 ml of sterile PBS (pH 7.4) in a round glass dish before feeding trials.

(g) Microbead/microbe feeding trials

Corals were placed in a sterile glass dish with 300 ml of particulate-filtered Instant Ocean at room temperature and 25 µl of either rinsed GFP+ *E. coli*-biofilmed microbeads or rinsed non-biofilmed microbeads was delivered to the surface of each *A. poculata* colony. Behavioural response and microbead localization/ingestion were documented via brightfield and fluorescence microscopy.

Twelve additional corals were fed 10–25 microbeads, either with or without the chloramphenicol-resistant GFP+ *E. coli* biofilm. After 48 h, no beads were visible within any polyps. At one and two weeks post-ingestion, mortality (absence of polyp tissue in the corallite) was scored in each polyp that ingested microbeads and in neighbouring polyps. Two weeks post-ingestion, fluorescence microscopy was used to detect GFP-positive *E. coli* cells on and around the microbead-fed polyps. Following imaging and analysis, a sterile 200 µl pipet tip was used to probe each microbead-fed polyp (either biofilmed or non-biofilmed microbeads) and then streaked on LB agar/chloramphenicol plates (25 mg ml^{-1}). Plates were incubated at room temperature for 1–3 days until colony growth was visible by eye. Fluorescence microscopy was used to determine whether bacterial colonies cultured from polyps were GFP-positive (electronic supplementary material, figure S3).

(h) Statistical approach

Statistical tests were completed using R. For all datasets, a Shapiro–Wilk *W*-test was conducted to test for normality. When data were normal, *t*-tests were used, and when data were non-normal, permutation tests were used. To explore differences in microplastic shapes for field-collected corals, paired *t*-tests were conducted on colony averages. For the concurrent feeding experiment (microplastics fed with BSE), a paired permutation test was used on polyp-level data. When food items were offered sequentially, polyp-level data were averaged and colony-level data were compared using unpaired tests. Paired permutation tests were used to compare proportions of microplastics found in the top, middle and bottom of the gut for each polyp analysed during given time points. Total microplastics per polyp were compared across different time points post-feeding, using unpaired permutation tests on polyp-level data.

3. Results

(a) Ingestion of plastics in the wild

Microplastics were present in every polyp dissected from wild *A. poculata* colonies, with an average of 112 particles $polyp^{-1}$ (±5.01 s.e.) (figure 1). Of all shapes, fibres were the most abundant, averaging 73.4% of the total particles, significantly more abundant than round particles (15.6%; $T = 25.9$, $p < 0.001$) and irregularly shaped plastic particles

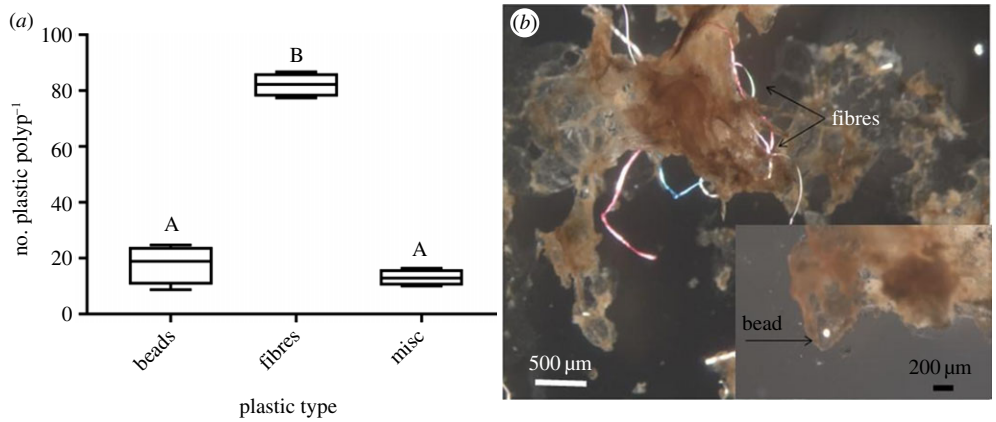

Figure 1. Microplastics in field-collected *A. poculata* corals off the New England coast (in Narragansett, RI). (*a*) Concentration of plastics per polyp, categorized by shape. (*b*) Visible fibres and beads among dissected, decalcified coral tissue. Error bars represent ± 1 s.e.m. (Online version in colour.)

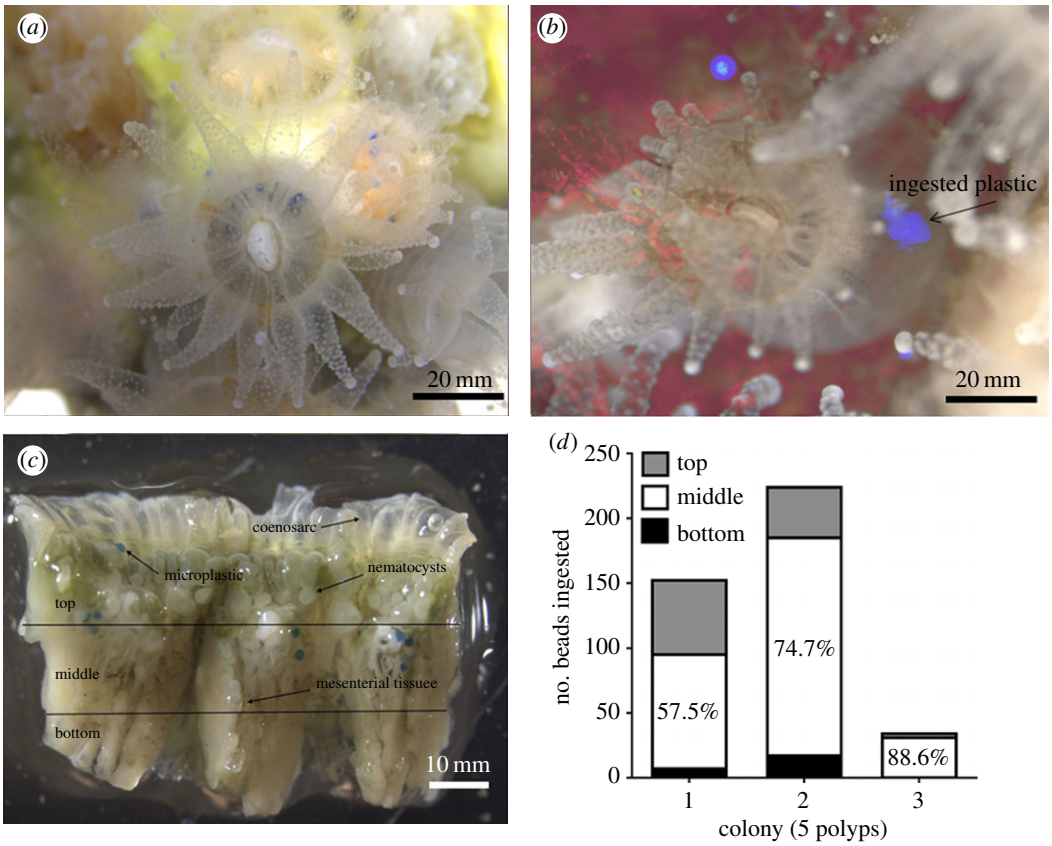

Figure 2. (*a*) *Astrangia poculata* fed blue UV-fluorescent microplastic beads (180–212 µm diameter) under brightfield and (*b*) UV filter. Arrow shows microbeads within the coral gastrovascular cavity. (*c*) Ingested beads within the polyps after 90 min (top = tentacles and mouth area; middle = mesentery; bottom = lower mesentery; tissue storage). (*d*) Microbeads consistently were localized to the mesentery in each of three sampled colonies ($n = 5$ polyps).

(11%; $T = 28.4$, $p < 0.001$). Polyamides (e.g. nylons) comprised 56% of the particles, followed by polyester (18%) and synthetic cellulose-based fibres (18%). Also present were pieces of polyvinyl chloride (3%) and fibre-reinforced plastic with epoxy resins (5%).

(b) Feeding behaviour and food preference

When fed with polyethylene microbeads ad libitum, *A. poculata* colonies ingested the beads into the gastrovascular cavity of the polyp (figure 2). Corals preferentially ingested microbeads over BSE, which are comparable in shape and size (figure 3, $T = 45$, $p < 0.004$ using paired permutation test on polyp-level data). Of 325 polyps measured, mean polyp width = 3.49 mm ± 0.04 s.e., and mean polyp volume = 30.23 mm^3 ± 1.01 s.e. Polyp size (volume in mm^3) was not correlated with the number of MP or BSE consumed ($R^2 < 0.01$ and $p = 0.86$, 0.59, respectively, using linear models in R), though there were a range of polyp sizes (electronic supplementary material, figure S2).

(c) Localization of ingestion

After 90 min, feeding was interrupted and corals were transferred into a clean chamber to examine post-ingestion microbead localization at 0, 30, 60, 90 and 1440 min (24 h) post-feeding. Immediately post-feeding, microbeads were significantly concentrated in the central mesentery (figure 2;

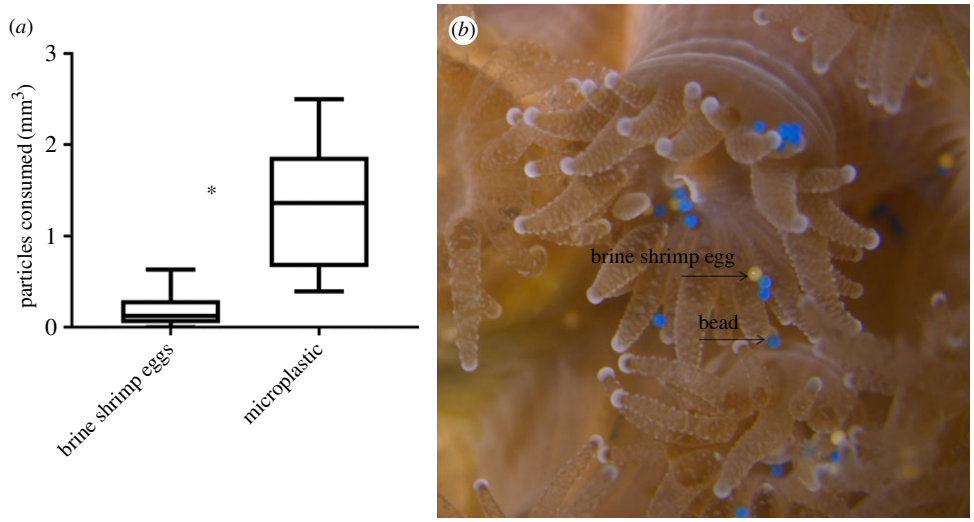

Figure 3. Comparison of brine shrimp egg (BSE) consumption versus microplastic bead (MP) consumption. (*a*) Ingestion of MPs and BSEs in a 50/50 choice assay. (*b*) Size and shape similarity of BSE and MP beads. Error bars in (*a*) represent ± 1 s.e.m. (Online version in colour.)

electronic supplementary material, table S1) and the microbead number per polyp was highest (electronic supplementary material, table S2). Between 30 and 90 min post-feeding, remaining microbeads were still concentrated in the central mesentery, but fewer beads were observed, suggesting that corals were egesting microbeads (electronic supplementary material, tables S1 and S2). Twenty four hours later (1440 min), few beads were left (electronic supplementary material, table S2), and there were no significant differences in microbead location (electronic supplementary material, table S1).

(d) Microplastics as a barrier to food intake

When corals were exposed to copepods or live brine shrimp after a 15 min exposure to either BSE or microbeads, there were no significant differences between the amounts of new prey items consumed (*t*-tests, $p = 0.3$, 0.6, respectively; figure 4*a*,*b*). Similarly, when corals were initially exposed to BSE, they continued to eat BSE when exposed again. However, no BSE was eaten by polyps after an initial 15 min exposure to microbeads (permutation test, $p = 0.05$; figure 4*c*). Similarly, corals that were concurrently exposed to both BSE and microbeads preferentially ingested microbeads (paired permutation test, $p < 0.004$; figure 5).

(e) Microplastics as microbial vectors

Confocal imaging (Zeiss LSM880 Airyscan) confirmed GFP+ *E. coli* biofilm formation on fluorescent microbeads (electronic supplementary material, figure S1). Ingestion of biofilmed microbeads was nearly immediate, happening within 15–60 s of delivery to the colony. In 100% of the feeding trials with biofilmed beads ($n = 10$ colonies), GFP+ *E. coli* cells from the surface biofilm were co-ingested with microbeads (figure 6). Biofilmed microbeads (figure 6*a*,*d*) were detectable within the polyp via microbead fluorescence (figure 6*b*) and by GFP+ *E. coli* fluorescence (figure 6*c*). Under the same settings, polyps that ingested non-biofilmed microbeads (autofluorescence negative control) displayed no detectable GFP signal (figure 6*e*,*f*). In two trials, fed polyps were recorded for 60 min following ingestion, but no microbead egestion was directly observed. No microbeads were visible within polyps after 48 h, suggesting egestion.

After two weeks, there was increased GFP signal (*E. coli*) on the surface, within and in the polyps neighbouring polyps that were fed *E. coli*-biofilmed microbeads. This was not observed in colonies fed non-biofilmed microbeads (electronic supplementary material, figure S3). There was no sign of mortality in polyps fed non-biofilmed microbeads at any time (electronic supplementary material, figure S3). Four weeks post-ingestion, mortality was observed in all polyps that ingested biofilmed microbeads and also in polyps neighbouring those that ingested biofilmed beads (electronic supplementary material, figure S3). Viable GFP+ *E. coli* was detected in all biofilmed microbead-fed *A. poculata* colonies during the two-week post-ingestion time point. No viable GFP+ *E. coli* cells were detected within polyps from colonies fed non-biofilmed microbeads (electronic supplementary material, figure S3).

4. Discussion

This study explored microplastic feeding behaviour and preference by the temperate, coastal, often-urban coral *A. poculata*, building on recent work demonstrating that *A. poculata* exhibits preferential ingestion of unfouled microplastics over fouled microplastics [38]. Our study offers several new contributions, including characterization of microplastic spheres, fibres and particles in wild-collected corals; novel experiments testing feeding behaviour and preference for biofilmed microbeads over other foods; description of ingested microbead retention and localization, and exploration of the consequences of microbead ingestion on the subsequent ingestion of nutritive prey items. Corals were fed *E.coli*-biofilmed microplastics to explore the hypothesis that microplastics vector microbes into corals. All experiments demonstrated that *A. poculata* exhibit preferential microplastic ingestion, with potentially important implications to its nutrition and microbiome.

To our knowledge, this is the first report of microplastic abundance in wild-collected corals. High concentrations of microplastics (112 particles polyp^{-1}) were found, of which the majority were fibres, consistent with other filter feeders [55]. High microplastics in *A. poculata* polyps may be caused by the proximity to a highly developed urban area, high commercial ship traffic, large polyp size and/or preferential

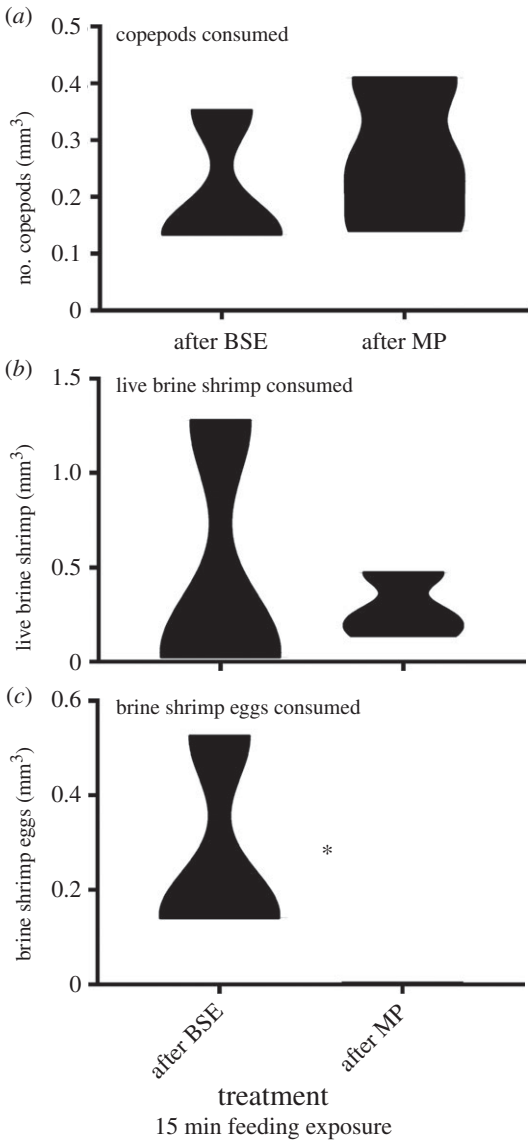

Figure 4. Violin plots demonstrating food intake following consumption of brine shrimp eggs (BSE) or microplastic beads (MP) for 15 min in *A. poculata*. Post-consumption prey items were (*a*) frozen copepods, (*b*) live brine shrimp or (*c*) brine shrimp eggs.

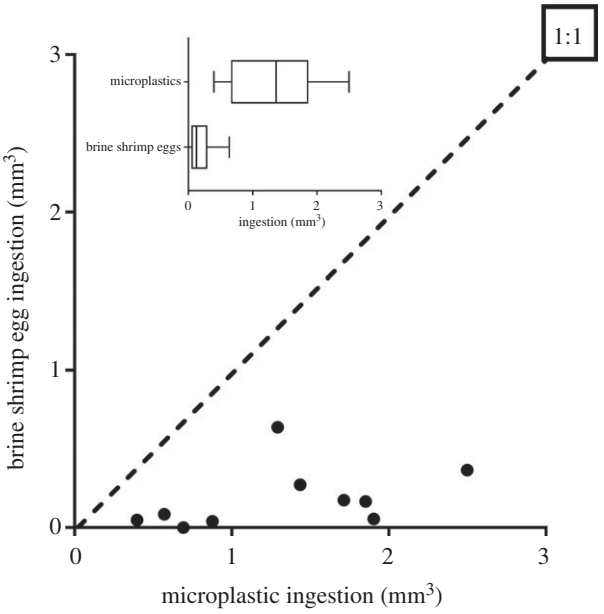

Figure 5. Comparison of brine shrimp egg (BSE) versus microplastic bead ingestion in *A. poculata* ($n = 10$ corals). Scatter plot shows paired preference, relative to a dashed line representing no preferential ingestion; any colony (dot) on the dashed line would have ingested an equivalent amount of brine shrimp and microbeads. Box plot shows the means of ingestion preference (inset).

ingestion of plastics from seawater ([28]; this study). The geographical range of *A. poculata* coincides with many coastal, urban harbours that have higher densities of floating debris than in the mid-ocean garbage patches [56]. As such, high levels of observed microplastics could be partially owing to macroplastic breakdown. Furthermore, coastally located septic and sewage systems allow microplastics to enter coastal urban waterways [57,58], and storm drain runoff allows microparticles from tyres [59] and road paints [58] to enter watersheds without wastewater filtration [56]. Further investigation in urban versus rural coastal areas is needed to determine the direct influence of various microplastic sources.

Although a wide variety of taxa have been documented to ingest microplastics, the consequences of microplastic ingestion have not yet been fully explored. When fed polyethylene microbeads ad libitum, *A. poculata* colonies centrally localized beads prior to egestion. When presented with microbeads and similar-sized BSE, *A. poculata* preferentially ingested microbeads, suggesting a high and potentially repetitive energetic cost via repeated ingestion and egestion as has previously been found in *Arenicola marina* lugworms [28].

One potential consequence of ingesting microplastics is the subsequent inhibitive feeding of nutritive prey, presenting a sort of 'double jeopardy' to corals that would potentially (i) suffer the loss of nutritive intake by eating non-nutritive microplastics, and (ii) suffer energy loss from the effort to ingest and egest microplastics. In this study, coral ingestion of microbeads inhibits subsequent ingestion of BSE, alluding to the importance of prey shape and size when the gastrovascular cavity is nearing capacity. A previous study investigated plastic spheres in conjunction with nutritive prey items (small and large diatoms in *Acartia clausi* copepods), and found similar prey selectivity based on the size of initial prey offerings [60]. Taken together, these studies underline the importance of considering the implications of microplastic shape. Because wild corals had a predominance of fibres (figure 1) over microbeads, it is possible that microplastics shape may be a main determinant of particle retention and subsequent nutritional loss. Because polyp volume did not limit microplastic ingestion (electronic supplementary material, figure S3), large polyps may be better able to cope with microplastic contamination because a larger cavity allows ingestion of nutritive prey in addition to the non-nutritive items that take up gastrovascular space.

The consumption of microplastics may also lead to the reduction of nutritive prey intake owing to a false perception of 'fullness.' For example, freshwater diving beetles that ingested microplastic-exposed zebrafish had lower subsequent ingestion rates compared to controls [61]. By contrast, some organisms, such as the copepod *Acartia clausi*, may be able to selectively avoid microplastics [60]. Corals, however, have mixed responses [39]. This suggests a need for species-specific studies on feeding behaviour and the dynamics of microplastic ingestion.

In addition to the nutritional consequences of consumption, plastic pollution has been implicated with increased prevalence

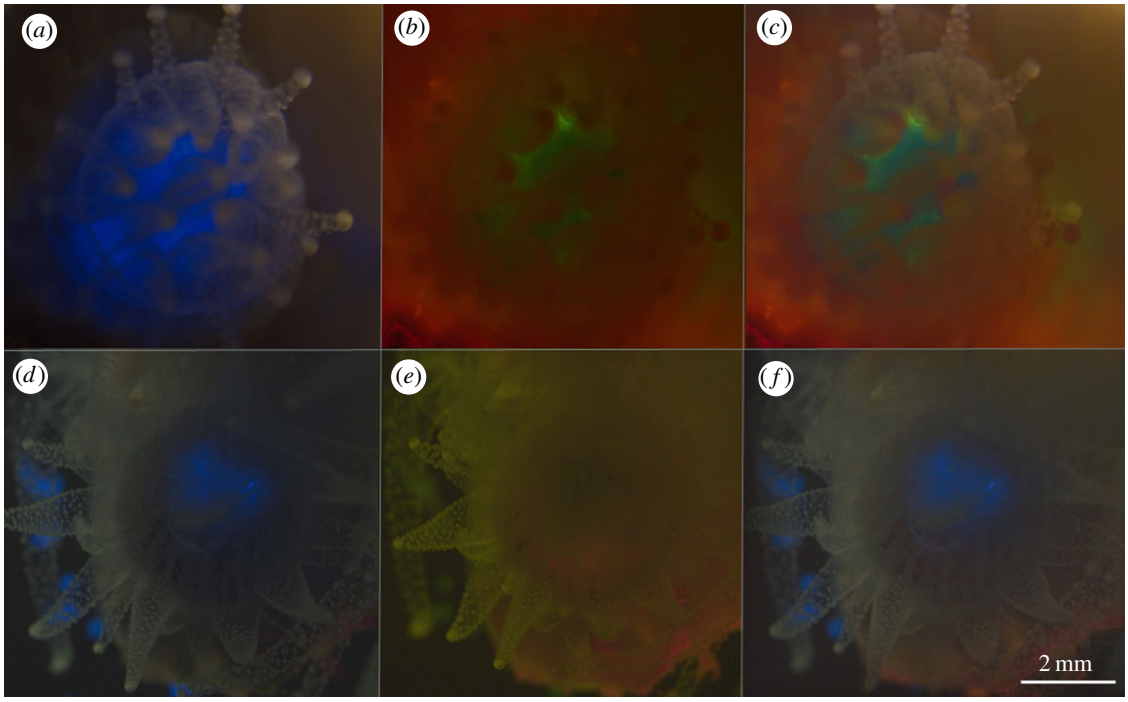

Figure 6. *Astrangia poculata* ingestion of polyethylene microbeads biofilmed with GFP+ *E. coli*. (*a*) Biofilmed microbeads detected via the UV fluorescence of the microbeads under the 4′,6-diamidino-2-phenylindole (DAPI) channel and by (*b*,*c*) GFP+ *E. coli* fluorescence. (*d*) Microbeads incubated in sterile LB broth only (negative control) imaged under the DAPI filter shows ingested beads within the polyp, and (*e*,*f*) no detectable GFP signal from the ingested non-biofilmed microbeads. (Online version in colour.)

of coral disease [36,45]. This may be a result of increased abrasion of the corals by plastic items, increasing susceptibility to microbial infection and resulting disease, or a result of pathogens vectored to the coral via microplastic [36]. Because microbial community composition of the 'plastisphere' is distinct from seawater and particulate organic matter [32] and includes putative pathogens [32–34], ingestion of novel plastics-associated microbes can potentially cause a shift in coral microbiome composition with unknown implications for host fitness and survival. In our study, microbead-delivered *E. coli* cells colonized surface mucus layers of *A. poculata*, but also, microbead-delivered microbes were retained in/on the coral. Observed polyp mortality was probably owing to microplastic-assisted delivery of *E. coli* and subsequent colonization/infection of coral surfaces and tissues. Even when *A. poculata* egested microbeads within 48 h, there was mortality in bead-fed polyps within two weeks, and within four weeks, neighbouring polyps died. However, when fed non-biofilmed microbeads, no mortality was observed. These results strongly suggest that co-ingested *E. coli* cells were responsible for increased mortality in *A. poculata* colonies (electronic supplementary material, figure S3). The *A. poculata* microbiome is relatively stable even across the symbiont state; investigating the potential for plastics to disrupt the microbiome would be an interesting future direction [62,63]. Future work identifying particular microbes that are enriched by plastics and ingested by corals via microplastics will inform models of how to mitigate impacts of microplastics on corals and coral reefs.

The consequences of microplastic ingestion evidenced by our study include retention of particles in the wild, preference for non-nutritive prey, potential limitation and inhibition of feeding on nutritive prey and potential for microplastics to vector novel or pathogenic microbes. Despite these risks and challenges, *A. poculata* continue to thrive in coastal waters, thus opening up their potential as a bioindicator tool to measure microplastic pollution in the US East and Gulf coastal waters. In our study, retention times of plastics have been measured, which will be essential for future *A. poculata*-based microplastic monitoring programmes. *Astrangia poculata*'s preference for microplastic ingestion offers the opportunity to put microplastic-free, laboratory-reared, marked colonies into the environment to monitor microplastic pollution, similar to programmes using *Mytilus edulis* blue mussels to assess chemical contaminants [64]. In addition to its resilience to microplastics pollution, *A. poculata* possesses several traits useful for a bioindicator: a large geographical range, high rates of heterotrophic feeding, a large depth range, survivability in proximity to urban wastewater input and persistence on urban substrates. As such, *A. poculata* is well positioned to serve as a potential indicator of microplastic accumulation across its wide habitat range.

Data accessibility. All data are available from the Dryad Digital Repository: https://doi.org/10.5061/dryad.3qc6328 [65].

Authors' contributions. R.R., J.U.R. and J.C. conceived the study. R.R., A.G., K.S., R.Y., E.B.L. and J.U.R. collected data used in this study. J.C. and R.R. conducted statistical analyses. R.R., K.S. and J.U.R. funded this study. All authors contributed to the writing and editing of this study.

Competing interests. We declare we have no competing interests.

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