Marine Pollution Bulletin 135 (2018) 587–593

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

Scleractinian coral microplastic ingestion: Potential calcification effects, size limits, and retention

Cheryl Hankins[a,*], Allyn Duffy[a], Kathryn Drisco[b]

[a] United States Environmental Protection Agency, National Health & Environmental Effects, Research Laboratory, Gulf Ecology Division, 1 Sabine Island Drive, Gulf Breeze, FL 32561, USA

[b] United States Environmental Protection Agency, Region 2, 290 Broadway, New York, NY 10007, USA

ARTICLE INFO

Keywords:
Microplastic
Coral
Retention
Egestion
Calcification

ABSTRACT

The impact that microplastics (< 5 mm) have on scleractinian coral is largely unknown. This study investigated calcification effects, size limits, and retention times of microbeads and microfibers in two Caribbean species, *Montastraea cavernosa* and *Orbicella faveolata*, in a series of three experiments. No calcification effects were seen in the two-day exposure to a microbead concentration of 30 mg L^{-1}. *M. cavernosa* and *O. faveolata* actively ingested microbeads ranging in size from 425 μm–2.8 mm, however, a 212–250 μm size class did not elicit a feeding response. The majority of microbeads were expelled within 48 h of ingestion. There was no difference in ingestion or retention times of 425–500 μm microbeads versus 3–5 mm long microfibers. *M. cavernosa* and *O. faveolata* have the ability to recognize and reject indigestible material, yet, there is still a need to study effects of energetics and microplastic contamination as a result of ingestion and egestion.

1. Introduction

Ingestion of plastic pollution in the marine environment first drew attention in the 1970s with numerous reports of plastics found in seabirds (Parslow and Jefferies, 1972; Rothstein, 1973; Hays and Cormons, 1974; Baltz and Morejohn, 1976; Ohlendorf et al., 1978). Given the persistent nature of plastic, it is alarming that total global estimates of plastics produced are over 8300 million metric tons (Geyer et al., 2017), with upwards of 12.8 million metric tons entering the ocean in 2010 (Jambeck et al., 2015). The widespread occurrence of plastics in the marine environment has led to a surge of research on the potential impacts that the smaller pieces of plastics, termed microplastics (< 5 mm), may have on wildlife.

Microplastics can originate from various sources including degradation of macroplastic, industrial processes, synthetic clothing, tire fibers, and personal care products (GESAMP, 2015; Thompson, 2015; Boucher and Friot, 2017). Once in the marine environment, microplastics tend to serve as a surface for microbial community growth as well as attracting pollutants (Teuten et al., 2007; Andrady, 2011; Zettler et al., 2013). As a result, their density increases and they become more bioavailable to organisms in the environment (Ye and Andrady, 1991; Teuten et al., 2007), including benthic organisms such as scleractinian coral.

Scleractinian corals are both phototrophic and heterotrophic feeders. Corals receive nutrition via translocation of photosynthetic products produced by zooxanthellae. However, corals also rely on exogenous food sources to meet their nutritional needs which can account for 15–35% of their daily energetic demand (Houlbrèque and Ferrier-Pages, 2009). Coral are passive suspension feeders that feed on plankton passing over their tentacles. Small debris, such as microplastics, may inadvertently be captured and ingested by the coral. Hall et al. (2015) demonstrated that corals ingest microplastic and did so at rates similar to plankton uptake, which was supported in subsequent studies (Allen et al., 2017; Riechert et al., 2018). Ingestion of plastics by other invertebrates has been shown to reduce energy budgets (Wright et al., 2013; Watts et al., 2015; Sussarellu et al., 2016), which could have significant impacts on health and reproduction. Although coral calcification relies heavily on photosynthesis from zooxanthellae (Porter et al., 1989), heterotrophic feeding becomes important when light is restricted or during bleaching (Palardy et al., 2008; Grottoli et al., 2006; Anthony et al., 2009). Ingested microplastics by coral could potentially reduce the energetic demands needed for the calcification process by inhibiting digestion of exogenous food sources, especially in times of stress. The evidence of ingested microplastics by corals confirms the need for further investigations looking at physiological interactions and potential threats. The objective of this study is to evaluate potential effects and interactions of coral responses to ingested microplastics for two Caribbean, scleractinian corals, *Montastraea*

* Corresponding author.
E-mail addresses: hankins.cheryl@epa.gov (C. Hankins), drisco.kathryn@epa.gov (K. Drisco).

https://doi.org/10.1016/j.marpolbul.2018.07.067
Received 26 April 2018; Received in revised form 20 July 2018; Accepted 23 July 2018
0025-326X/ Published by Elsevier Ltd.

cavernosa and *Orbicella faveolata*. In a series of three experiments, this study (1) investigated effects ingested microbeads on calcification, (2) determined ingested size ranges (425 μm–2.8 mm) and retention times of microplastics by coral, and (3) compared ingestion and retention of microbeads versus microfibers by coral.

2. Methods

Three laboratory experiments were conducted using the Caribbean, scleractinian coral species, *Montastraea cavernosa* (large polyp coral) and *Orbicella faveolata* (small polyp coral). Coral were collected from Florida Keys National Marine Sanctuary Coral Nursery Program (Permit Numbers FKNMS-2005-057 & FKNMS-2011-036). Species were chosen to represent large and small polyp coral species. Coral specimens were maintained for at least three months in an indoor coral research facility at the U.S. Environmental Protection Agency's Gulf Ecology Division in Gulf Breeze, Florida. Corals were maintained in recirculating culture systems (~1000 L) prior to experimentation. Experiments were conducted in a separate recirculating system (~820 L). All systems were kept at a temperature of 26.0 ± 1.0 °C and salinity of 35.0 ± 0.3 ppt with metal halide lights on a 9.5:14.5 light:dark cycle. Other water quality parameters such as calcium, pH, alkalinity, magnesium, ammonia, and nitrate were all measured prior to experimentation and were within culture condition parameters (Borneman, 2001, Delbeek and Sprung, 2005, Holmes-Farley, 2004). Light intensity in culture and experimental systems ranged from 25.0–40.0 W m^{-2} depending on age of the bulbs and placement within the system. For each experiment, at least three parent colonies of each species were used to cut 4 cm^2 fragments using a Gryphon Corp. Aquasaw XL (Model C-40). Fragments acclimated in the experimental system five days prior to the start of the experiment. Coral fragments in experiment 1 were glued to acrylic pegs to sit securely on egg crate. Fragments in experiments 2 and 3 sat freely on egg crate within the chambers. The number of replicates varied between experiments based on experimental design. During all experiments 100% cotton clothing was worn. Exposure to microplastics had to be assumed as corals originated from a natural environment. Additionally, culture conditions were not strictly monitored in the years leading to the study. Microplastics used in experiments needed to be clearly identified, therefore, fluorescent microplastics were used in the smaller size classes.

2.1. Experiment 1: Effects of ingested microbeads on calcification

The first experiment determined if microplastic ingestion impacted calcification in the two coral species. Three different size classes of fluorescent, polyethylene microplastic beads consisting of mostly smooth surfaces (Cospheric®) were used in this experiment: 90–106, 425–500, and 850–1000 μm. Polyethylene is a common polymer found in marine sediment and surface waters (Teuten et al., 2007; Erni-Cassola et al., 2017). The density of each of the three size classes was 1.002 ± 0.006 g cc^{-1}. The approximate density of 26.0 °C, 35.0 ppt sea water is 1.025 g cc^{-1}, denser than the microbeads. Prior to initiating the experiment, microbeads of each size class were placed in separate 80 μm mesh containers and placed in culture for curing (i.e. growing biofilm on surface) for six weeks to decrease their buoyancy.

Ten 8 L plastic, circular chambers were used as experimental chambers and contained in a water bath to maintain temperature (26.0 ± 0.5 °C; Fig. 1). Each chamber had a 3/8″ needle valve near the bottom for chamber filling. Within each chamber, four fragments of each *M. cavernosa* and *O. faveolata* rested on egg crate (i.e. louvered ceiling panels) which sat atop an 8.5 cm tall stand. A Hydor Koralia 240 pump was used for circulation and was positioned in a hole made in the center of the egg crate, and pointed in a downward direction (Fig. 1). Curing the microbeads prior to the experiment seemed to decrease their buoyancy. Although the density of the microbeads was not determined after the curing process, the microbeads did not float at the water's surface and the circulation within the chamber was sufficient to keep microbeads in suspension. The 90–106 μm size class required the surface of the water to be agitated in order to get the beads in suspension. After agitation, all size classes generally remained in suspension as few were detected by means of ultraviolet light on chamber walls or at the water's surface.

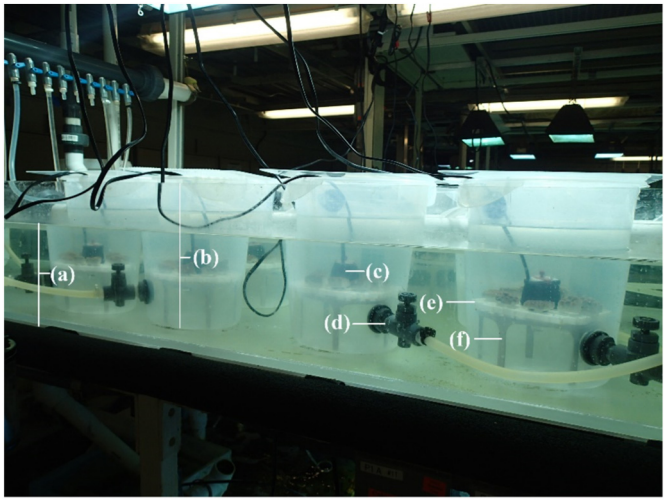

Fig. 1. Diagram of experimental system set-up including 8 L experimental chambers used in Experiment 1: (a) water bath, (b) experimental chamber with acrylic lid, (c) circulation pump, (d) needle valve, (e) egg crate, and (f) stand.

The two treatments for the experiment consisted of a control group (not exposed to microbeads) and an exposed group, with each group consisting of five replicates. Each chamber of the exposed group contained all three microbead size classes at a dose of 80 mg microbeads per size class, resulting in a final microbead concentration of 30 mg L^{-1} (10 mg L^{-1} per size class). Number of particles per liter was approximately 24 for the 850–1000 μm size class, approximately 215 particles for the 425–500 μm size class, and undetermined for the 90–106 μm size class. Before the addition of microbeads, water flow from the recirculating system was shut off to each chamber and remained off for the two-day exposure; water inside the chamber remained circulating via the pump. Each chamber was fed 5 mL from a mixture of 0.156 g Golden Pearls® (Brine Shrimp Direct, Ogden, UT) 5–50 μm coral food in 100 mL seawater to elicit a feeding response in conjunction with microplastic application. The recirculating system is equipped with a protein skimmer, phosphate reactor, as well as a 1 μm filter to eliminate all sources of nutrients and debris that may otherwise be mistaken as food. The addition of food was to simulate the presence of zooplankton on a reef to mimic natural conditions whereby microplastics may mix with plankton (Boerger et al., 2010). Salinity was not regulated during the static, two-day exposure period to eliminate the risk of losing any microplastics that might have adhered to the sensor. In preliminary tests, the maximum salinity increase within the chambers was 0.3 ppt over two days without de-ionized (DI) water adjustments.

Following the two-day exposure, coral tissue was processed to recapture microbeads. Processing occurred under UV light. All rinsing steps were performed using DI water, unless otherwise specified. Each coral fragment was removed from the chamber and placed into individual glass culture bowls. Corals were vigorously rinsed to remove microbeads that may have been stuck to the outside of the polyp or acrylic base with filtered (1 μm) seawater. The seawater used for rinsing was placed back into its respective chamber. Coral tissue was removed from its skeleton using the airbrush method first described by Johannes and Wiebe (1970). A metal funnel was placed into a 50 mL Falcon tube to collect all coral tissue. Each Falcon tube was then exposed to an ultrasonic cell disruptor (Tekmar sonic disrupter Model TM 600 with a Branson ultrasonic converter Model CV17) for 15 s at 30% amplitude to