PHYSIS

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CIEE Research Station

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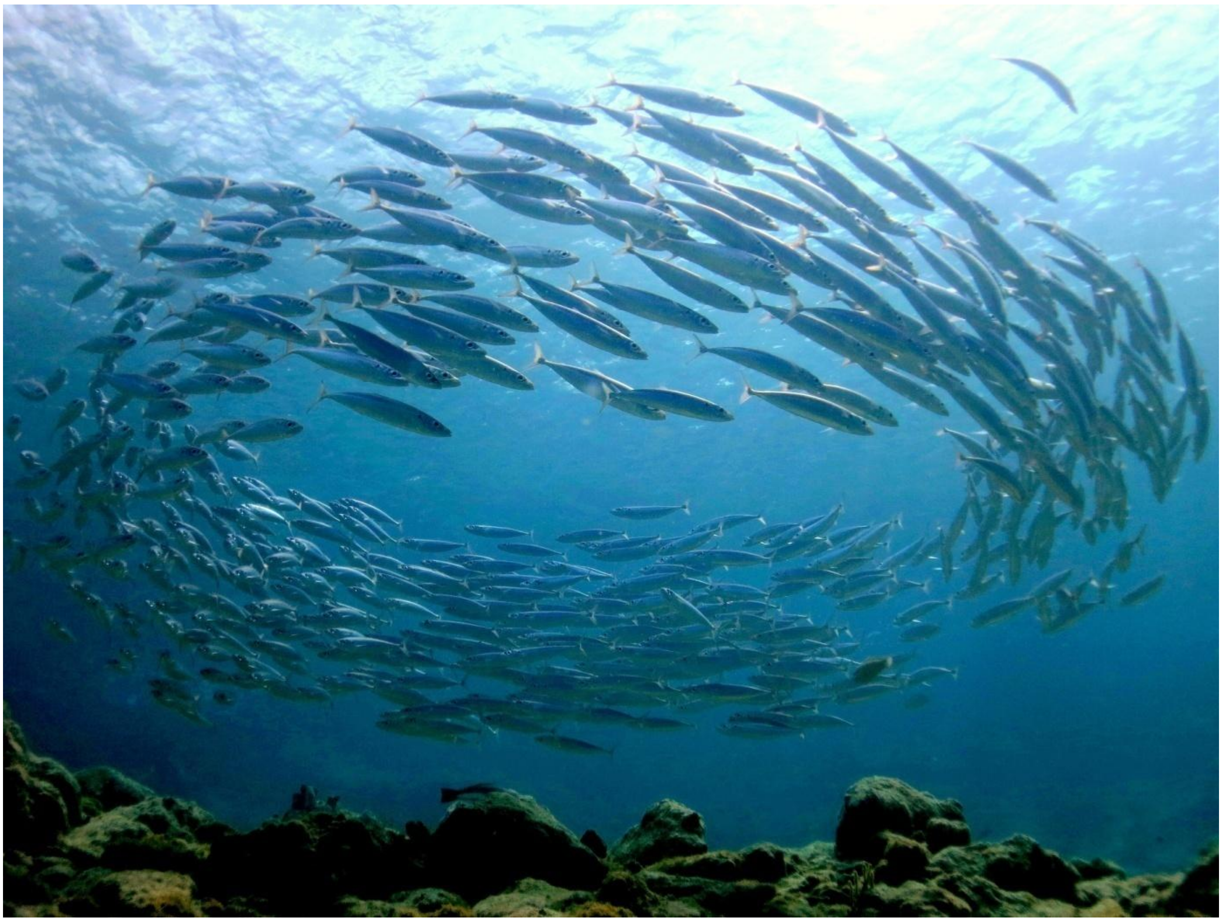
Physis

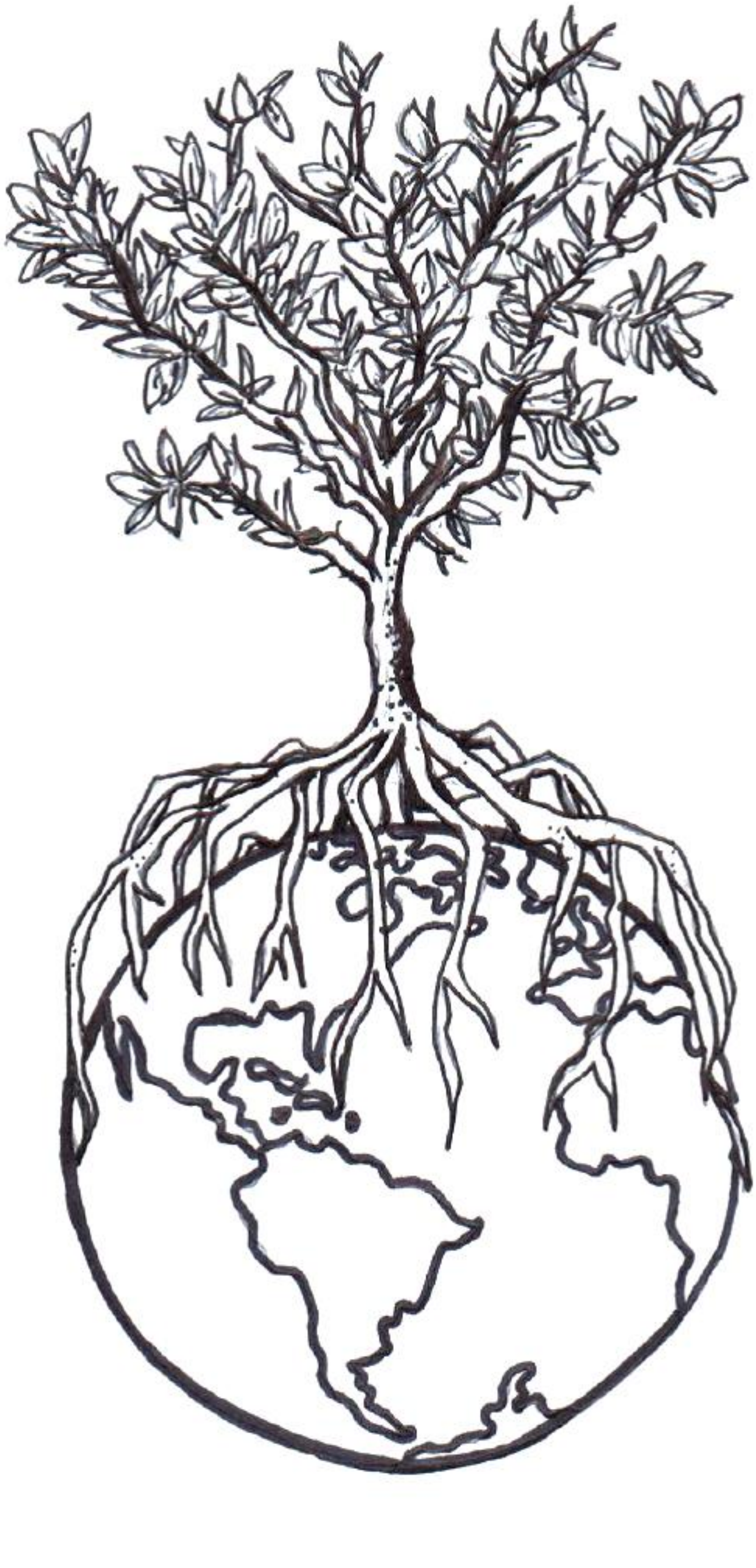


Journal of Marine Science

CIEE Research Station

Tropical Marine and Conservation Program

Volume 13 Spring 2013



PHYSIS φύσις

In the *Odyssey*, Homer uses the term *Physis* just once, to simply describe nature. Nature is a breathtaking phenomenon of the physical world that includes plants, animals, landscapes, and the systems present. The Greeks used this term not only to describe nature, but also to portray the stability of the natural world. Nature is a product of the Earth, as opposed to a creation of man, yet we are deeply and intrinsically connected to it. We have destabilized nature through our dependence on it, but nature has responded through the process of *Physis*—self-healing that has occurred regardless of our impact. Through our strong will to advance in society, we have reached a point where nature cannot heal fast enough to counter the destruction of our actions.

Our Earth is dramatically blue. Water shapes our planet and provides life, something that is unlike anywhere else in this universe. In the words of Herman Melville, "…we ourselves see in all rivers and oceans [the] images of the ungraspable phantom of life; and this is the key to it all." The ocean has challenged our worldview—the great depths holding unknown secrets, which inspired humanity to push the limits of our understanding of nature. The border between land and sea has historically been viewed as a hard and fast line between worlds: our terrestrial homeland and the vast, mysterious ocean. Earth is so vast, that it is believed to be resilient. However our actions on land are having increasing impacts on the marine ecosystem. The ocean covers more than 70% of our Earth, and yet at an alarming amount we are diminishing its resources. It is our duty and privilege to take the challenge to begin protecting it.

"People protect what they love."
-Jacques Yves Cousteau

Physis evokes an image of something lost to us in our advances: natural beauty unencumbered by climate change, human degradation, light pollution, and metropolitan noise. The state and restoration of our blue planet must become a priority in our lives. The balance that *Physis* illustrates will come not by manipulation of biological or ecological functions, but by stepping back, reducing our harmful impacts and letting ecosystems find their long lost equilibrium.

In the fifteen weeks that we have spent on Bonaire, we have been able to see the effect that we as a society have had on our reefs. Through assessments and data analysis, we have been able to quantify the damage we have created in our environment, and we have seen the consequences of our actions. This reflexivity—the ability to see the effect of our actions in the delicate balance that is necessary to sustain the ocean environment—has shed light on the need for us to speak out, to educate, and be educated on our role. We can no longer expect the oceans to sustain us and remain unchanging. However, we can re-create a positive relationship with the oceans not through trying to cover past mistakes, but through removing our pressures and presence to allow nature to heal itself. This semester, we were able to study and witness extraordinary events from monitoring sea turtles to the intricate relationships between corals and algae to the self-healing capabilities of sea-pearls. Through our semester, we were able to bear witness to the incredible power of *Physis.*

Gabrielle Lout
Amber Packard
Madeline Roth

FOREWORD

The Council on International Educational Exchange (CIEE) is an American non-profit organization with over 150 study abroad programs in 40+ countries around the world. Since 1947, CIEE has been guided by its mission:

“***To help people gain understanding, acquire knowledge, and develop skills for living in a globally interdependent and culturally diverse world*.**”

The Tropical Marine Ecology and Conservation program in Bonaire is a one-of-a-kind program that is designed for upper level undergraduates majoring in Biology. The goal of the program is to provide an integrated program of excellent quality in Tropical Marine Ecology and Conservation. The field-based science program is designed to prepare students for graduate programs in Marine Science or for jobs in Marine Ecology, Natural Resource Management and Conservation. Student participants enroll in six courses: Coral Reef Ecology, Marine Ecology Field Research Methods, Advanced Scuba, Tropical Marine Conservation Biology, Independent Research in Marine Ecology/Biology and Cultural & Environmental History of Bonaire. In addition to a full program of study, this program provides dive training that results in certification with the American Academy of Underwater Sciences; a leader in the scientific dive industry.

The student research reported herein was conducted within the Bonaire National Marine Park with permission from the park and the Department of Environment and Nature, Bonaire, Dutch Caribbean. Projects this semester were conducted on the leeward side of Bonaire where most of the population of Bonaire is concentrated. Students presented their findings in a public forum on the 17th of April, 2013 at the research station.

The proceedings of this journal are the result of each student’s research project, which is the focus of the course that was co-taught this semester by Rita B.J. Peachey, PhD; Catherine Jadot, PhD; and Enrique Arboleda, PhD. In addition to faculty advisors, each student had an intern that was directly involved in logistics, weekly meetings and editing student papers. The interns this semester were Katie Correia, Fadilah Ali and Brian Strehlow. Ryan Patrylak was the Dive Safety Officer and provided scientific dive training and oversight of the research diving.

Thank you to the students and staff that participated in the program this semester! My hope is that we succeeded in our program goals and CIEE’s mission.

Dr. Rita Peachey

FACULTY

Dr. Rita Peachy is the Resident Director in Bonaire. She received her B.S. in Biology and M.S. in Zoology from the University of South Florida and her Ph.D. in Marine Sciences from the University of South Alabama. Dr. Peachey's research focuses on ultraviolet radiation and its effects on marine invertebrate larvae and is particularly interested in issues of global change and conservation biology. She teaches Independent Research and Cultural and Environmental History of Bonaire. Dr. Peachey is president of the Association of Marine Laboratories of the Caribbean.

Dr. Catherine Jadot is the Tropical Marine Conservation Biology Faculty. She holds a PhD in Eco-Ethology, a MSc. in Oceanography and a MSc. in Zoology. Before joining CIEE Bonaire she worked for various universities and agencies in Belgium, France, the Azores, Dubai, Trinidad, the Cayman Islands, the Bahamas and Turks and Caicos. Her research interest are marine resources management, near-shore habitat enhancement and restoration. Catherine has authored and co-authored numerous scientific papers and technical reports on issues related mainly to eco-ethology, ecological restoration, fisheries management and coral reef ecosystems.

Dr. Enrique Arboleda is the Coral Reef Ecology Faculty for CIEE and co-teaches Independent Research and Marine Ecology Field Methods. He is a Marine Biologist from the Jorge Tadeo Lozano University (Colombia), holds a specialization on Biodiversity and Evolutionary Biology from the University of Valencia (Spain) and obtained his PhD at the Stazione Zoologica di Napoli (Italy) working on photoreception of sea urchins. He worked as a Post-Doctoral fellow at the Max F. Perutz Laboratories (Austria) investigating chronobiology on marine invertebrates before moving to Bonaire. Dr. Arboleda's research interests include adaptation, plasticity upon disturbance, competition, reproductive strategies and how ecological, molecular and physiological responses, like those associated to an abrupt climate change, can drive evolution by natural selection.

FACULTY AND STAFF

Ryan Patrylak is the Diving Safety Officer and Lab Technician at CIEE Bonaire. He holds a B.S. degree in Coastal Studies from the University of Connecticut. After graduation he moved to the Orkney Islands, Scotland where he was a research intern for Divers Alert Network. After the internship he worked as a Fisheries Biologist/ Observer with the Connecticut Department of Environmental Protection and the National Marine Fisheries Service for two years before moving to Bermuda. In Bermuda he held the position of Diving Safety Officer at the Bermuda Institute of Ocean Sciences for almost two years before coming to CIEE Bonaire. He is an active PADI Instructor, DAN Instructor and AAUS Science Diver.

Amy Wilde is the Administrative Assistant for CIEE. She holds a B.S. degree in Business Administration, as well as, a Masters of Science in Management Administrative Sciences in Organizational Behavior, from the University of Texas at Dallas. She has worked in call center management for the insurance industry and accounting for long term care while living in Texas. Amy currently provides accounting and administrative support for staff and students at CIEE and she is the student resident hall manager.

Catalina Sanchez is the CIEE Education and Research assistant. She holds a B.Sc. on Ecology from the FUP in Colombia and is currently working on her thesis towards a M.Sc. on Environmental Sciences with the University of Buenos Aires (Argentina). After working on mangrove conservation for her B.Sc. thesis, she has assisted with the management of diverse projects and acted as Coordinator of the Unit for Information Services in a biotech company in Colombia. She is a newly certified diver enthusiastic about bringing the Bonairean community, especially youngsters, closer to CIEE's activities

INTERNS

Fadilah Ali is the Conservation Biology Intern at CIEE Bonaire. She recently completed her undergraduate studies at the University of Southampton where she pursued a Masters in Environmental Science (M.Env.Sci) with a specialisation in Biodiversity and Conservation. Currently she is conducting research for her Ph.D. in Ocean and Earth Sciences focusing on the Lionfish Invasion in the Caribbean. Fadilah volunteered at CIEE in the summer of 2010 and then came back in 2011 to conduct more research on lionfish and to assist with the program as an intern.

Katie Correia is the Coral Reef Ecology Intern at CIEE Bonaire. She holds a B.S. degree in Marine Science/Coastal Geology from Coastal Carolina University and is currently working towards achieving her M.S. in Coastal Zone Management/Marine Biology from Nova Southeastern University. She is an active PADI Divemaster which she has utilized during her previous employment experiences in the Florida Keys as a Field Technician for Mote Marine Laboratory and as a Biological Scientist for Florida Fish and Wildlife Conservation Commission.

Brian Strehlow is an Intern at CIEE. He has a B.S. in Biology with a double major in Latin American and Iberian Studies from the University of Richmond (UR). After graduation, he continued his undergraduate research full-time as a laboratory assistant at UR. His research focuses on the symbiotic relationship between Zooxanthellae and marine sponges. His fieldwork was conducted primarily at Mote Marine Laboratory in Summerland Key, Florida. He has also participated in reef surveys and conservation efforts in the bay islands of Honduras through Operation Wallacea.

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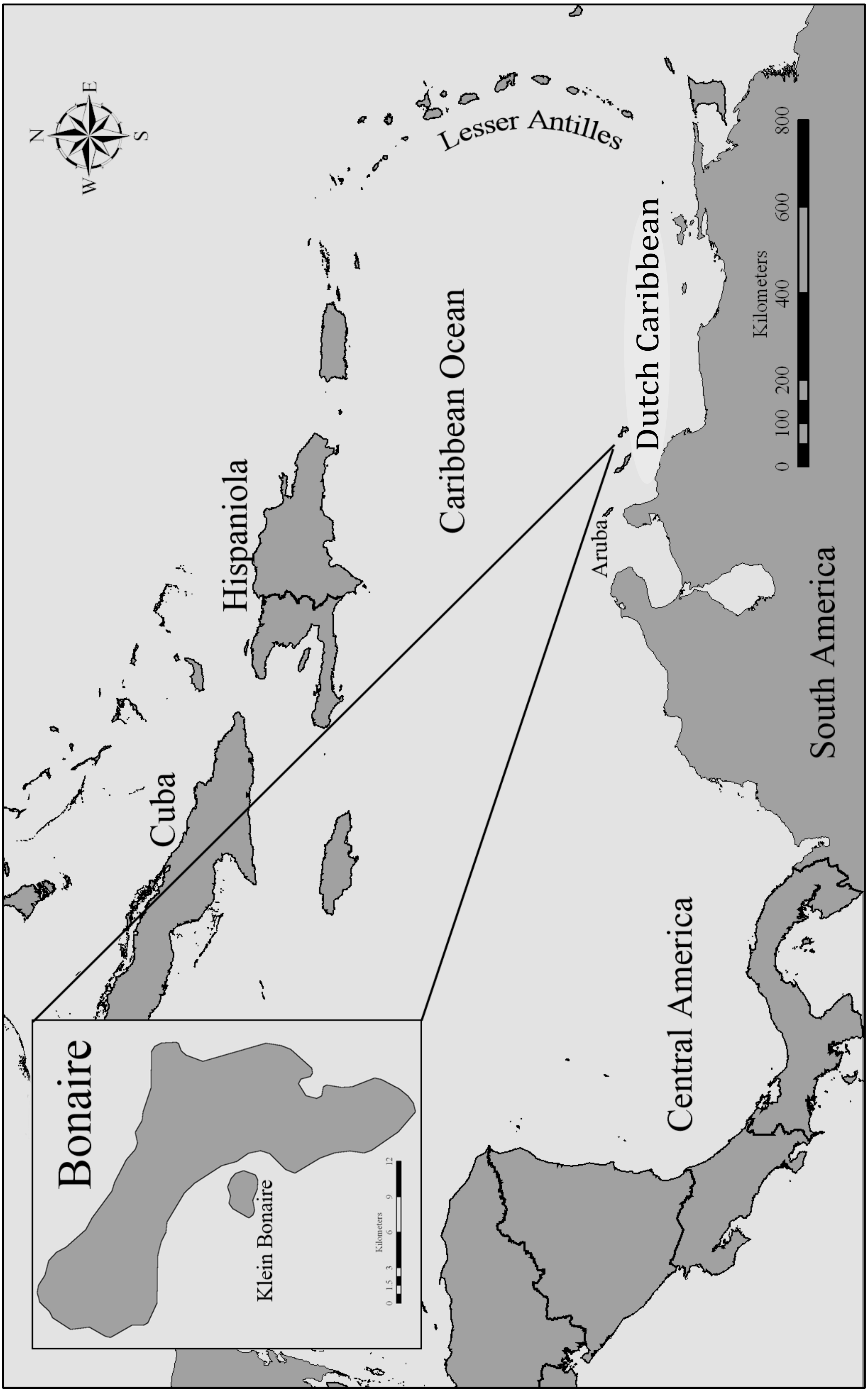
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REPORT

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Abundance and size distribution of the bearded fireworm *Hermodice caruncula*ta on sand flats and coral reefs in Bonaire

Abstract *Hermodice caruncula*ta the bearded fireworm is abundant in Bonaire's coral reefs. The corallivorous fireworm is a voracious eater, and a generalist predator. *H. caruncula*ta's foraging behaviors play a role in coral reef community structure and building. This study looked at the abundance of the bearded fireworm in two environments, coral reefs and sand flats, during dusk and night hours. Within these two substrates size, abundance, and fluorescent color morphologies of the fireworms were studied. Sizes were separated into four length categories: <3 cm, 3-6 cm, 7-9 cm, >9 cm. Due to the active nature of fireworms and sampling at night, BlueStar flashlights and yellow barrier filters were used to locate the fireworms in the dark. Field surveys were conducted using 10 m transects and a t-bar to estimate abundance in both environments. Wire box traps were also placed along the coral reef and sand flats to estimate abundance of fireworms in the area. *H. caruncula*ta were found to be less abundant on coral reefs at dusk than at night. Furthermore, fireworms in the size class >9 cm were only found on coral reefs, indicating an ontogenetic shift in habitat and size. An ontogenetic shift was also found in the fluorescent color morphologies. Green fluorescence was most abundant in the 0-6 cm size range, and completely absent in the >9 cm size class. The green body with orange bands was an intermediate fluorescent pattern found predominantly in 3-9 cm size range. The orange body with orange bands coloration was found in the largest size class, possibly being the terminal fluorescent phase.

Keywords Bearded fireworm • *Hermodice caruncula*ta • Coral reefs • Sand flats • Fluorescence

Introduction

Bearded fireworms (*Hermodice caruncula*ta*)* are annelids in the class polychaeta and can be commonly found on reefs and littoral areas across the Caribbean and Western Atlantic Ocean (Lewis 2009; Pérez and Gomes 2012). These worms range in size from a few millimeters to 35 cm in length. Most *H. caruncula*ta found on coral reefs are in the range of 5-10 cm in length (Trauth 2007). The bearded fireworm gets its common name from the sharp white setae (hair-like bristles) along both sides of its body. The setae are fine and can easily break off and cause a sharp burning sensation (Kicklighter and Hay 2006). The setae found on the fireworm are thought to deter predators from consuming them. This defense allows the fireworm to forage more freely than worms that are more palatable.

*H. caruncula*ta tends to be more active at night, foraging on top of the reef in the open for more valuable prey (Witman 1988; Kicklighter and Hay 2006). *H caruncula*ta moves from feeding on open horizontal or vertical surfaces to less exposed surfaces, such as in crevices, or under rocks (Witman 1988). *H. caruncula*ta is a corallivorous worm, meaning it feeds on coral tissue. It is a voracious eater, which also feeds on a variety of coral reef organisms many of which belong to the phylum Cnidaria (Pérez and Gomes

2012). It has been shown that fireworms are also detritivores, meaning they will eat dead and decaying organic matter (Jumars 1993, Lewis 2009).

A recent study by Wolf and Nugues (2012) showed an ontogenetic shift in the diet of *H. carunculata*. Fireworms under 5 cm were observed feeding under cover more often, while fireworms above 5 cm feed more on top of coral and in the open. Fireworms under 5cm are more likely to stop and feed on spat (coral recruits) than fireworms above 5 cm, becoming a cause for early-post settlement mortality (Witman 1988; Wolf and Nugues 2012). As well the average feeding duration increased with size. Coral recruitment is vital for the regeneration of coral reefs after a large disturbance. Since the bearded fireworm has such a varied feeding behavior they may play an important role in influencing coral reef community structures (Wolf and Nugues 2012).

Sussman et al. (2003) indicated that *H. carunculata* is a vector for coral disease. In the winter months the bearded fireworm becomes a virus reservoir, while in the spring and summer months the fireworm becomes a vector for disease. In the summer month's fireworms transfer coral disease while foraging on coral that is already stressed. Corals can become stressed from a number of different factors such as bleaching, disease, or predation. In a study done by Wolf et al. (2012) suggested that *H. carunculata* preferred eating decaying or diseased coral, rather than healthy coral. These predation patterns of feeding on stressed corals have been linked to increased stress on bleached and diseased corals.

Witman (1988) found fireworms to feed exposed on horizontal or vertical surfaces at night more often than during the day. Fireworms seen feeding during the day quickly returned to a crevice or under a rock once finished feeding. *H. carunculata* is most active at night, yet little is known about their night time activity (Trauth 2007). The bearded fireworm fluoresces when exposed to blue light. Utilizing this technique at night makes even the smallest individuals easily visible during darkness. The fireworm's body fluoresces under UV light, while the bristles do not exhibit this trait. The bearded fireworm has several fluorescent variations, showing green, orange, yellow (Mazel 2007), and blue colors.

Habitat selection varies throughout different life stages of many marine invertebrates. Factors such as shelter, protection against predators, and food availability can determine habitat choices and distribution of a species. Roughly 0.09% of the ocean is coral reefs; yet, this small amount supports approximately 25% of marine animals (Lewis 2009). Thus the health and abundance of coral reefs is important. Looking at the abundance of *H. carunculata* in both coral reef and sand habitat will help determine the influence they could have on community structure and development.

This paper will address the following questions about *H. carunculata*: Is the bearded fireworm more abundant in a sand environment or a coral reef environment? What is the size distribution and abundance of the fireworm in these two environments? How does the abundance of *H. carunculata* vary between dusk and night on the two substrates?

H_1: *Hermodice carunculata* will be more abundant in a sand flats than in a coral reef environment.

H_2: *Hermodice carunculata* between <6 cm will be more abundant on sand substrate, while *H. carunculata* >6 cm will be more abundant on coral reefs.

H_3: *Hermodice carunculata* will be more abundant at night than at dusk on both coral reef and sand flats.

Since *H. carunculata* has an ontogenetic shift in its diet as it grows larger (Wolf et al. 2012), it is important to understand if they also have an ontogenetic shift in their habitat, as they grow larger. This study will help to enhance the understanding of fireworm distribution and size variation between the two environments as well as their impact on coral reef and sand flat communities.

Materials and methods

Study site

All experimental work was conducted at Yellow Submarine dive site (12°09'36.47"N, 68°16'55.16"W), located along the western shore of Bonaire, Dutch Caribbean. Diving was done two days a week, during five weeks, over two different ecosystems: coral reefs and sand flats. Each day was dedicated to one of the two ecosystems. The two transect locations were chosen at random, by swimming three minutes to the north of the dive site entrance to a location with less dive traffic.

Transects

Two 10 m transects were conducted on two different substrates: sand flats and coral reef. For each transect, two passes were made using a 1 m t-bar for a reference point, with 0.5 m on each side of the transect tape. The first pass was made approximately 20 minutes after sunset and the second pass 30 minutes after the first. The 30 minute period between each pass allowed for the survey to be completed, and for dusk to become night.

BlueStar UV flashlights (www.nightsea .com) and yellow barrier filters were used to observe the fluorescence of *H. carunculata*. The blue filter converts UV light into blue light that stimulates the fluorescence found in the fireworm. By using the yellow barrier goggle the fluorescence reflected from the fireworm can be observed.

During each transect the surveyor recorded, size to the nearest centimeter, fluorescent color variation, and benthic habitat the fireworm was found on. *H. carunculata* was categorized into four size classes: <3 cm, 3-6 cm, 7-9 cm, >9 cm. An initial pre-sample was done to establish size categories of the fireworms. Transects were conducted in the same manner as the study, and size was recorded for each individual fireworm to the nearest centimeter. Size classes were determined based on the occurrence of each color variation in association with the size.

To measure the size distribution on both sand flats and coral reefs size classes <3 cm and 3-6 cm were combined into <6 cm, and 7-9 cm and >9 cm was combined into >6 cm. The size classes were combined to see if there was an ontogenetic shift from smaller fireworms living in the sand flats, to larger fireworms living in coral reefs.

Wire boxes

Using a method similar to Wolf et al. (2012), wire boxes measuring 20x20 cm with 1 cm mesh size, were used to estimate the abundance of *H. caruncul**ata*** in both sand and reef ecosystems. Two wire boxes were deployed on the sand and two on the reef. Of the two boxes, one was filled with synthetic algae (synthetic bath sponge) as well as live algae collected from the surrounding benthos. The second box was identical to the first, with the addition of lionfish meat. The wire boxes with meat were sampled and replaced with fresh lionfish meat during each sample period. The wire boxes without meat were left for two weeks between each sampling. The wire boxes with meat were sampled more often to replenish the meat inside. The contents were brought to the laboratory for analysis before being released back to the organisms’ place of collection. To avoid re-sampling of the same fireworms, every collection and placement of new boxes was moved 15 m along the substrate in a horizontal manner identical to the transects. The abundance of worms, their size, and color variation were recorded for every wire box collected for sampling. BlueStar flashlights and yellow barrier filters were used to identify the fluorescent color morphologies.

Laboratory studies

Fireworms collected in the wire boxes were transferred to the laboratory for documentation of fluorescent color morphologies. A dissecting scope and a Canon powershot S100 were used to capture images of the fluorescent fireworms. Fluorescent color was photographed in a dark room, with two BlueStar flashlights being used as the light source. Yellow barrier filters were attached to the camera lens.

To aid in taking clear pictures of the fireworms fluorescence, 1% ethanol was used to temporarily relax and slow the movement of the fireworms. Fluorescent morphologies were documented for the four different size classes to observe if patterns were consistent within the various size classifications (Fig. 1).

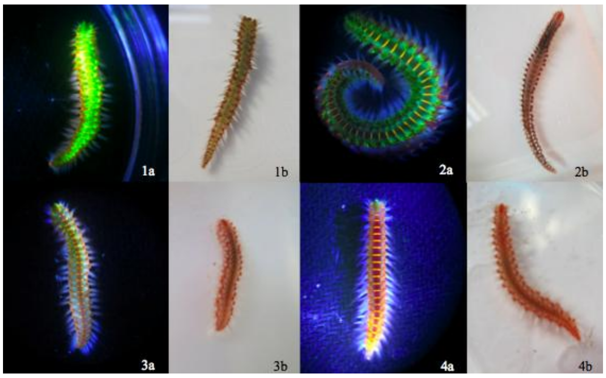

Fig. 1 Fluorescent color variations of *Hermodice carunculata*. An all green fluorescent fireworm in the <3 cm size class displays the green fluorescent color category (1a). The same fireworm is shown without fluorescence, displaying a slight green pigment along the body (1b). A fireworm in the 6-9 cm size class represents the green body with orange bands (GOB) fluorescent color category (2a). To the right the same fireworm non-fluorescent (2b). A fireworm in the 3-6 cm size category also represents the GOB category (3a). Same fireworm is shown non-fluorescent (3b). A fireworm in the 3-6 cm size class represents the orange body with orange bands (OOB) fluorescent color category (4a). Shown to the right is the same fireworm under non-fluorescence, showing a red color variation along the body (4b)

Data analysis

Data collected from the field was analyzed using a two-tailed t-test. The t-test was used to analyze the differences between: coral reef abundance at dusk and coral reef abundance at night, sand flat abundance at dusk and sand flat abundance at night, as well as total coral reef abundance and total sand flat abundance. A Mann-Whitney U-test was used to test for a difference between cages with meat and without meat on sand flats and coral reefs.

Results

Field surveys

The data collected on sand flats and coral reefs, as well as dusk versus night, were analyzed using a two-way t-test. There was no significant difference between the total abundance of *H. carunculata* on coral reefs (1.97 per m^2, SD 2.39) versus sand flats (2.17 per m^{-2}, SD 1.19) (Table 1). There was a significant difference in total abundance of fireworms on coral reefs at dusk (0.60 per m^2, SD 0.54) and total abundance on coral reefs at night (3.35 per m^2, SD 2.77) (p=0.026). There was no significant difference between the abundance of fireworms on sand flats at dusk (1.80 per m^2, SD 1.13) and sand flats at night (2.55 per m^2, SD 1.19) (Table 1).

Table 1 Two-way t-test of the abundance of *Hermodice carunculata* on coral reef and sand flat substrates. Time of dusk was 20 minutes after sunset, while night was 50 minutes after sunset. Total abundance of each substrate was taken from both night and dusk. The four size classes (<3 cm, 3-6 cm, 7-9 cm, >9 cm) were grouped into two larger size classes (<6 cm, >6 cm)

	P-value	df
Coral reef total vs. Sand flat total	0.793	15
Coral reef dusk vs. Coral reef night	0.026	15
Sand flat dusk vs. Sand flat night	0.217	15
Coral reef dusk vs. Sand flat dusk	0.021	15
Coral reef night vs. Sand flat night	0.471	15
Coral reef <6 cm vs. Sand flat <6 cm	0.448	15
Coral reef >6 cm vs. Sand flat >6 cm	0.044	15

There was a significant difference between abundance of fireworms on coral reef at dusk compared to sand flats at dusk (p=0.021). Coral reefs at night and sand flats at night had no significant difference between abundance (Table 1).

The bearded fireworm was not found to be more abundant in size class <6 cm on sand flats (1.01 per m^2, SD 0.68) than coral reefs (0.79 per m^2, SD 1.16). There was a significant difference between fireworms of the >6 cm size class found on coral reefs (0.2 per m^2, SD

0.35) versus sand flats (0.07 per m^2, SD 0.17) ($p=0.044$).

Cages

The wire cages deployed on sand flats had a large number of fireworms in both cages with meat and cages without meat. The bulk of fireworms found in the cages with meat were in the 3-6 cm size class (0.74 per m^2, SD 0.49) (Fig. 2). Cages in sand flats without meat still had a large number of fireworms (0.73 per m^2, SD 0.59), but were slightly lower than cages with meat (0.43 per m^2, SD 0.48) (Fig. 3). There was no significant difference between the number of fireworms in sand flat cages with meat or without meat ($p=0.142$, $n_1=6$, $n_2=2$).

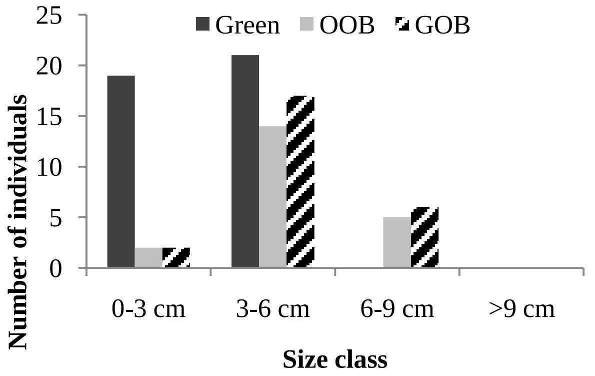

Fig 2 Number of *Hermodice carunculata* found in cages on sandy flats with lionfish meat added. Fluorescent color variations: green, orange body with orange bands (OOB), green body with orange bands (GOB)

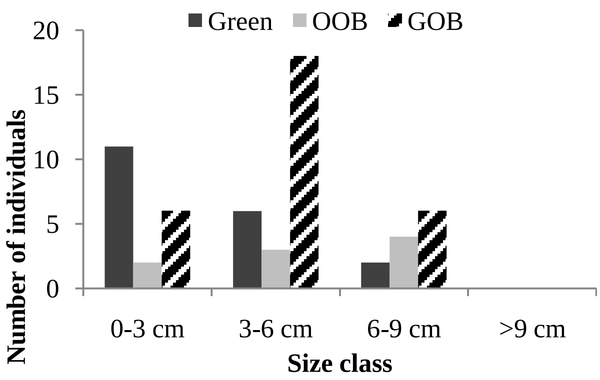

Fig. 3 Number of *Hermodice carunculata* found in cages on sandy flats. Fluorescent color variations: green, orange body with orange bands (OOB), green body with orange bands (GOB)

Fluorescence

Fluorescent color variation was recorded on all fireworms seen in the field along with size. Fluorescent color variations were separated into three categories: green, orange body with orange bands (OOB), and green body with orange bands (GOB) (Fig. 1). A fourth color variation seen outside the transects, was characterized by a blue body with blue bands (Fig. 4).

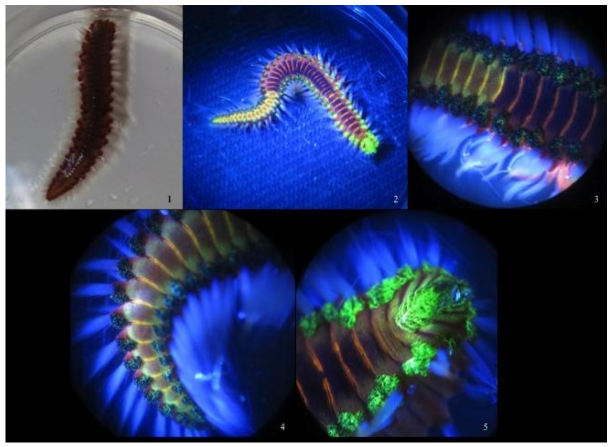

Fig. 4 Fluorescent fireworm under UV light. Panel 1 show the non-fluorescent image of the same fireworm. Panel 2 shows and image of the fireworm showing the blue body with orange bands (BOB) fluorescent variation. Panel 3 shows a close-up of the shift between the orange body with orange bands (OOB) to BOB pattern. Panel 4 shows an up-close image of the tail displaying the OOB pattern. Panel 5 shows an image of the BOB pattern on the head

This color variation was seen twice during spawning events at full moon nights (Fig. 5). Fireworms were observed spawning an hour after sunset on the night of the full moon and three days after.

Only two color patterns were seen in the >9 cm size class, OOB and GOB (0.038 per m^2, SD 0.09; 0.14 per m^2, SD 0.31 respectively). Of the two fluorescent color variations seen in size class >9 cm, it was predominantly OOB (0.14 per m^2, SD 0.31) (Fig. 6).

The different color variations were broken up into percent abundance in each size class (Fig. 6). The green fluorescent variation had the largest percentage at 74.27% in size class <3 cm (1.54 per m^2, SD 1.16). The size class

3-6 cm GOB and OOB had an even number of individuals at 39.4% (0.61 per m^2, SD 0.58 for both).

Fig. 5 Fireworms observed spawning an hour after sunset on the full moon and three days after. Panel A shows a fireworm stretching into the air possibly sensing for a potential mate. Panel B shows fireworms congregating on top of *Montastraea annularis*. Panels C and D show the fireworms spawning

As the fireworms grow larger, size class 7-9 cm, there is a shift in abundance to OOB at 46.9% (0.19 per m^2, SD 0.27). Only two color patterns were seen in the >9 cm size class, OOB and GOB (0.038 per m^2, SD 0.09; 0.14 per m^2, SD 0.31 respectively). Of the two fluorescent color variations seen in size class >9 cm, it was predominantly OOB (0.14 per m^2, SD 0.31) (Fig. 6).

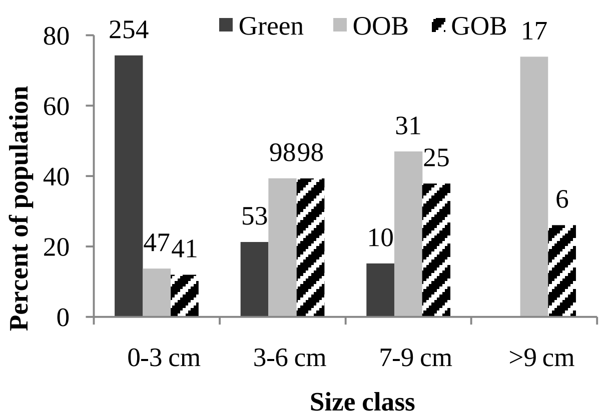

Fig. 6 Fluorescent color variation in *H. carunculata* within the four size categories. Numbers on top of each column represent the number of individual fireworms observed in given size and color category. Fluorescent color variations: green, orange body with orange bands (OOB), green body with orange bands (GOB)

Images taken with a dissecting scope show the details of *H. carunculata*. The fireworm has two eyes positioned at the front of the caruncle on each side. Protruding from the front of the caruncle two antennae are seen with a third protruding from the top of the caruncle. *H. carunculata* has a segmented body, which can be seen from both the dorsal and ventral sides. The mouth of the fireworm is located along the ventral side. Gills are found on each body segment of the fireworm at the base of the setae (Fig. 7).

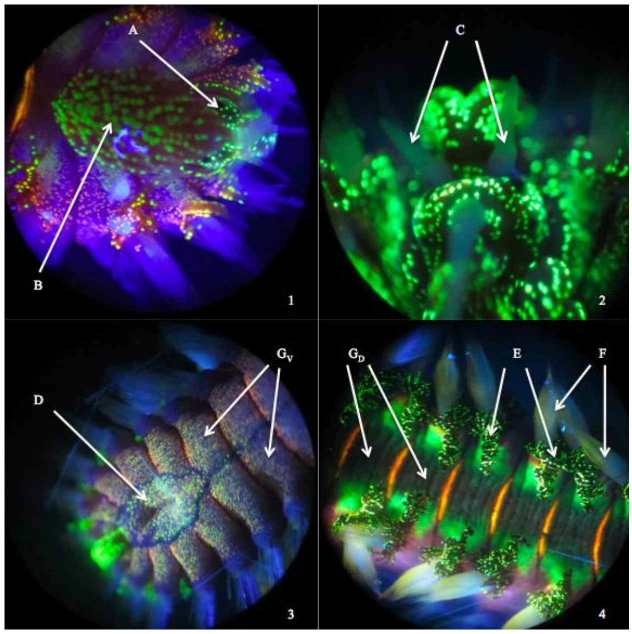

Fig. 7 Fluorescent fireworm under UV light. Images were taken with a dissecting microscope at a magnification of 4 (3,4) and a magnification of 6 (1,2). Panel 1 shows the two eyes, (A) two located on each side of the caruncle (B) (1). Panel 2 is a close-up on the two antennae (C). Panel 3 shows the ventral side of the fireworm showing its mouth (D). Panel 3 is showing the setae protruding from each segment (F). The fireworm has gills at the base of the setae on each segment (E). Segments of the fireworm have clear lines on both the dorsal and ventral sides (G_V, G_D)

Discussion

With the use of fluorescence this study looked at the size distribution, abundance and color variations of fireworms on two substrates, sand flats and coral reef, between dusk and night.

The first hypothesis was rejected: there was no significant difference between abundance in sand flats and coral reef environments. Originally it was thought that fireworms would be more abundant on sand flats as there was a higher level of recruitment.

Due to the complex habitat structure of coral it would seem larger fireworms would prefer these areas as there is more room for growth and expansion, as well as a larger food source.

The second hypothesis was partially supported: there was a significant difference between the two larger size classes (>6 cm) on coral reefs and sand flats, but there was no significant difference between the two smaller size classes (<6 cm) on coral reefs and sand flats. Larger fireworms were found more often on the coral reef environment than the sand flats. This could be due to a shift in diet, as the fireworms get larger. Fauchald and Jumars (1979) showed that fireworms found in the sand had different stomach contents than fireworms found on coral reefs and suggested that smaller fireworms found on sand flats are carrion-feeders while larger fireworms on coral reefs fed on prey.

The third hypothesis was partially supported: there was a significant difference between abundance of fireworms at dusk versus night on coral reefs, but there was no significant difference of fireworm abundance between dusk and night on sand flats. It is possible that the difference between day and night are more significant than dusk and night because fireworms are fluorescent. Fluorescence does not appear during the night, as there are no UV wavelengths to be reflected by the fireworm. A study conducted by Witman (1988) showed that fireworms on the coral reef remained mainly hidden during the day. When fireworms were seen eating exposed during the day, they returned to a hiding place soon after feeding. As well the fireworms seen eating during the day were generally ranged from 5-10 cm in length. While at night they did not return to hiding places, and once finished feeding, they continued onto the next food source.

Two fireworms were found in the cages placed on the coral reef environment, which suggests that the fireworms found a better environment to reside on the coral reef. In the sand flats the numbers rose drastically. The majority of fireworms found in the sand flat cages were in the first two size classes (<3 cm, 3-6 cm). Wolf et al. (2012) showed that predation on fireworms was highly dependent on size. Fireworms above 4cm were not preyed upon, while smaller fireworms were preyed upon by numerous species such as the yellowhead wrasse (*Halichoeres garnoti*) and the bluehead wrasse (*Thalassoma bifasciatum*). The abundance of fireworms in the cages on sand flats could be an indication for the need of refuge. Smaller fireworms were preyed upon more often than larger fireworms, this could be due to the fluorescent color variation found on smaller fireworms or possibly less developed defenses in the smaller individuals.

Three fluorescent color variations were observed during the transect: green, green body with orange bands (GOB), and orange body with orange bands (OOB). As *H. carunculata* changes its feeding behaviors with size, it also changes fluorescent color patterns. The three color patterns made a subtle shift from predominantly green color variation on the smaller size class (<3 cm), to GOB in middle size classes (3-6 cm, 7-9 cm), to predominantly orange body with orange bands in the large size class (>9 cm). The green color variation was absent from the >9 cm size class, indicating that an ontogenetic shift in color pattern is present. The blue coloration, although rare and seen only during spawning aggregations, raising additional questions. Fireworms were seen congregating on top of *Montastraea annunlaris*. This fourth color variation could be the terminal or sexually mature stage in fireworms, possibly indicating the difference between males and females, however more information is needed to verify this hypothesis. Other species such as the parrotfish and some wrasses have a terminal or sexually mature stage with distinct color morphology. This could be true in fireworms as well. More research is needed to see if there is a correlation between this fourth color morphology and spawning. Reasons for this change are unknown, but many marine species change color morphologies with size, either due to predation, dominance, or sexual maturity. It could also be an important tool in better understanding the ecology of other coral reef organisms that are also active at night.

The distribution of larger fireworms on coral reefs could be based on food availability or habitat preference, as the results from this study shows fireworms of the size class >9 cm were absent from sand flats. Fauchald and Jumars (1979) showed a shift in diet as fireworms became larger. This supports the general trend of larger fireworms being absent from sand flats and present on coral reefs as their diet shifts from carrion-feeders to predators. It is common among many size oriented marine organisms for a behavioral shift to occur alongside a shift in diet. As well, predation risks are generally related to body size, thus many species will change habitat use as predation pressure changes.

H. carunculata is a known predator to hermatypic corals. Wolf et al. (2012) found that fireworms below 2 cm influenced a high mortality rate in coral recruits, while fireworms above 6 cm chose other sources of food and had little effect on coral recruits. The study also found that predation on adult corals was absent. This indicates that as healthy corals mature they either become less desirable to fireworms, or they improve their defenses (Pérez and Gomes 2012). The lack of a significant difference between the abundance of *H. carunculata* on coral reefs and sand flats in the size classes 0-6 cm could be due to their diverse feeding behavior. Smaller fireworms may be able to utilize the resources in the sand flats as well as coral reefs while in order to support the growth and nutritional requirements of a larger fireworm a habitat change to coral reefs is important. A study done by Fauchald and Jumars (1979) shows that semi-digested coral matter in the water will attract other fireworms in the surrounding area. Corals that have disease or are stressed are generally victim to predation. The abundance of larger fireworms on coral reefs could be directly linked to the abundance of coral, and the source of decaying coral available. As fireworms are also detritivores they do not solely rely on coral as their main food source, but it is a primary source of nourishment (Wolf and Nugues 2012).

As coral reef health declines world wide, the loss of coral reef community structure are impacting the coral reef inhabitants. This study indicates that *H. carunculata* has an effect on coral recruitment success, thus playing a large role in community structure. More research is needed to help understand the role of fireworms on coral reefs to better predict the impacts they will have when corals begin to decline. It is suggested that with the varied diet of the fireworm they will adapt well to change, but it would be interesting to see if this is true and if so, how they may affect the survival of hermatypic corals.

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REPORT

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Tubastraea coccinea: Distribution and substratum preference of an exotic coral in Bonaire, Dutch Caribbean

Abstract The study of introduced species has gained popularity in recent years. A species introduced to a new area can have negative effects on the native ecosystems, as well as positive interactions with local fauna. The success of an exotic species depends on many factors. Those that are most successful at expanding possess mechanisms of reproduction, settlement, and distribution that aid in competing for space and resources. *Tubastraea coccinea*, also known as orange cup coral, is native to the Indo-Pacific and was introduced in Bonaire in the 1940s. Little is known about the effects *T. coccinea* has on the local marine community. It has a very opportunistic nature and has become a dominant scleractinian coral in the subtidal zones occupying shallow, heavy surged waters. *T. coccinea* is an azooxanthellate coral, which explains its ability to occupy habitats not desirable by other corals requiring sufficient nutrients and sunlight for photosynthesis. The purpose of this investigation was to determine the distribution and abundance of *T. coccinea* along various sites in Bonaire and observe its habitat preferences. Six sites in Kralendijk, Bonaire were surveyed by snorkelers, who counted various sized colonies and substratum occupancy. *T. coccinea* was found at all six surveyed sites, being most abundant at sites with very shallow shores and heavy surge. It preferred man-made pilings underneath docks as its habitat. This confirms that *T. coccinea* is established in Bonaire. By observing the distribution and preferences, the successful nature of *T. coccinea* throughout the Caribbean can be better understood.

Keywords *Tubastraea coccinea* • exotic species • abundance

Introduction

Introductions of exotic species have long been recognized as a possible threat to natural marine communities (Silva et al. 2011; Sampaio et al. 2012). To monitor and understand an exotic species' presence in the area; distribution, reproduction and settlement patterns are crucial to study (Paula and Creed 2005). These factors can be used to understand the species range expansion and possible future impacts to the native communities. The azooxanthellate coral *Tubastraea coccinea* has been present in Bonaire and surrounding islands dating back to 1943 (Vermeij 2005). It is considered an exotic species to the area, but little is known about its distribution, reproduction, and settlement. It has become one of the dominant species in the shallow subtidal zones along the coast. Due to its opportunistic nature it has established itself in habitats not normally favorable for most corals.

T. coccinea are non-reef building corals that extend their polyps and feed mainly at night. It is a heterotrophic species that does not have a symbiotic relationship with zooxanthellae, like most corals do. Due to the lack of zooxanthellae it is not limited to the areas it can settle. It does not require light needed for photosynthesis and can live in heavy sediment areas. It has long been found in caves, wrecks, and hidden under rocks (Vermeij 2005). It has been observed to compete with benthic invertebrates for space and can harm surrounding fauna with chemical release (Ferreira et al. 2003).

T. coccinea planulae larvae also display mechanisms that can aid in their survival and settlement possibilities. *T. coccinea* larvae tend to swim for a few minutes before falling to the bottom. The time spent swimming before settling range up to 3 days but larvae can remain competent for up to 100 days (Glynn et al. 2008). Recruits commonly settle in tight clusters near established adult colonies and the recruits present a rapid growth rate. One day after settling, they increase their diameter by 1.40mm (Glynn et al. 2008). The combination of relatively effective larvae settling and quick growth aids in the success of reproduction and expansion of *T. coccinea* on certain substrates.

T. coccinea has expanded its range to all continents with the exception of Antarctica, yet little is known about its reproductive nature (Paz-Garcia et al. 2007). It is known to be a brooding species, releasing planulae larvae. It spawns numerous times over a year based on the lunar cycle (Glynn et al. 2008). *T. coccinea* has relatively high fecundity rates in comparison to other reef corals, which could explain its widespread distribution and success (Glynn et al. 2008). Along with its high reproduction rate, it has the ability to asexually bud and create a "runner." This runner is a thin tissue lacking polyps that extends from the original colony until it finds available and favorable substrate (Vermeij 2005). A new polyp then forms at the end of this runner. This mechanism adds to *T. coccinea's* rapid expansion and settlement among a range of substrates (Vermeij 2006).

T. coccinea possess many mechanisms that have increased their success in the Caribbean. Although it has become a dominant species in the intertidal zone in Bonaire, little is known of its behavior and impacts. The following hypotheses have been formulated:

H_1: *Tubastraea coccinea* colonies will be most abundant in areas of heavy surge in very shallow waters.

H_2: *Tubastraea coccinea* will be most abundant on rocky substratum.

The aims of this study were to measure the relative abundance of *T. coccinea* at various sites in Bonaire and gain knowledge on its patterns of settlement, such as substratum preference. The data collected will help the scientific community better understand *T. coccinea*'s rapid expansion and strategies that have made it a successful introduced species.

Materials and methods

Survey sites

Six sites along Bonaire's west coast were surveyed. The sites were chosen based on their close proximity to the capital city of Kralendijk, as well as a known presence of *T. coccinea* (Fig. 1). The northern three sites were shallower and had consistent heavy surge, while the southern three sites were in calmer waters with less tidal movement.

Fig. 1 Map of the coast of Kralendijk, Bonaire. Sample sites are marked by points from South to North: Plaza Resort, Dive Inn, Main Town Pier, South Pier, Something Special and Eden Beach (modified from www.maps.google.com)

Abundance survey

At each of the sites, snorkelers surveyed the littoral/subtidal zones to identify the relative abundance by counting *T. coccinea* colonies and characterizing the size of the colonies. In a 30m by 10m area, the colonies were counted and categorized as young (1-5 polyps), intermediate (6-10 polyps), and well-established colonies (>10 polyps). The total numbers of colonies counted were used to calculate the relative abundance. The abundance was calculated using an index: Abundant (>50 polyps), frequent (50-25), rare (25-5), and absent (<5). The total number of polyps in the area was calculated per square meter to obtain the density. The substratum type for each colony was recorded as rock, rubble, manmade pilings, or dead coral.

One of the sampling sites (Something Special) presented a particular situation: The site has a dock with pilings covered by *T. coccinea*, creating a vertical plane not considered on the other sites when the 30m by 10m horizontal area was surveyed. To evade this situation, the pilings' surface area was calculated (11.5 m^2) and added to the 30m by 10m area before calculating density.

Slope preference

To determine the slope preference of *T. coccinea* at the three sites, angles of the well-established colonies (i.e. colonies with more than 10 polyps) were measured. The angles of the established colonies were measured by placing a protractor at the base to measure the outward hemisphere of the coral colony. The average preference of each of the colonies was then calculated.

Fluorescence

Coral colonies were observed under fluorescent light in order to establish fluorescent capabilities and to localize small coral recruits not visible with white light.

Data analysis

Data was analyzed for trends or correlations. The counted colonies were used to estimate the relative abundance at each site. The percent cover and density (m^2) was calculated for the 30m by 10m areas surveyed. The slope preferences at all the six sites were averaged to estimate the overall slope preference of *T. coccinea* in Bonaire.

Results

General abundance

T. coccinea was found at all six sites along the Kralendijk's coast ranging from very abundant to rare at sites. It was found in least abundance at Main Pier (15 colonies) and Dive Inn (3 colonies) sites (Fig. 2). *T. coccinea* was most abundant at Something Special with 393 colonies (Fig. 3). This site had the most densely packed colonies with a more than one colony/m^2 and 48.9% cover (Fig. 4). This site also possessed the most well established colonies (261colonies). *T. coccinea* was also very abundant at Plaza Resort with 183 colonies present. This site had the most intermediate colonies (91 colonies) of all six sites. South Pier was very abundant possessing 136 colonies, 96 of these being established colonies. Plaza Resort and Something Special possessed the most young colonies in comparison to the other sites (36 and 72 colonies respectively). Dive Inn and Main Pier did not contain any young colonies. South Pier and Eden Beach contained more established colonies (96 and 35 colonies) than young (18 and 9 colonies) or intermediate colonies (22 and 21 colonies). Overall, *T. coccinea* was most abundant at the southern most three sites in Bonaire (Fig. 1).

Substratum angle

T. coccinea was measured to occupy all possible angles on various substrates in Bonaire. More colonies were found occupying vertical substrate structures, like pilings (62.3%), than the other horizontal structures, such as, rock and dead coral (Fig. 5).

Substrate preference

The colonies surveyed occupied all the recorded substrates. *T. coccinea* was most abundant of man-made substratum, or dock pilings with 495 colonies present (62.3%). It was also very abundant on large dead coral heads and rock (33.5% or 166 colonies).

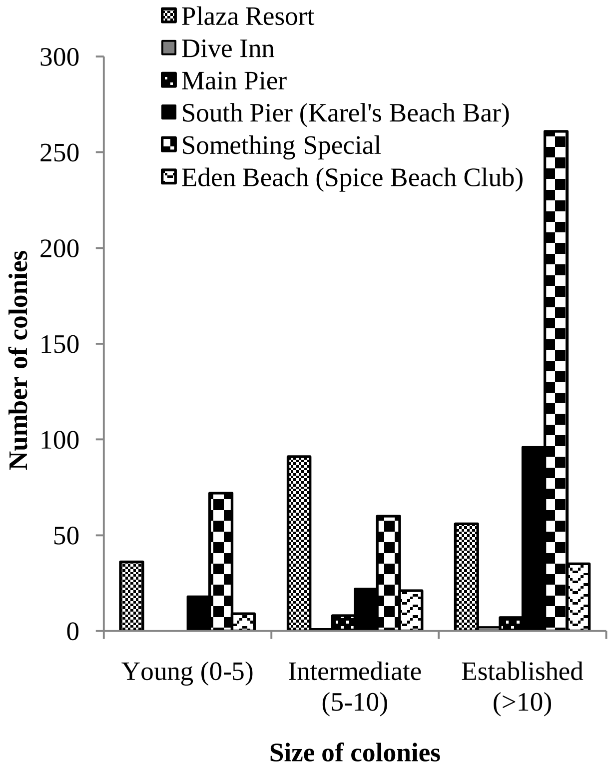

Fig. 2 Number and size of *T. coccinea* colonies at six sites in Kralendijk, Bonaire. Young colonies contained 0-5 polyps, intermediate colonies contained 5-10 polyps, and established colonies contained >10 polyps

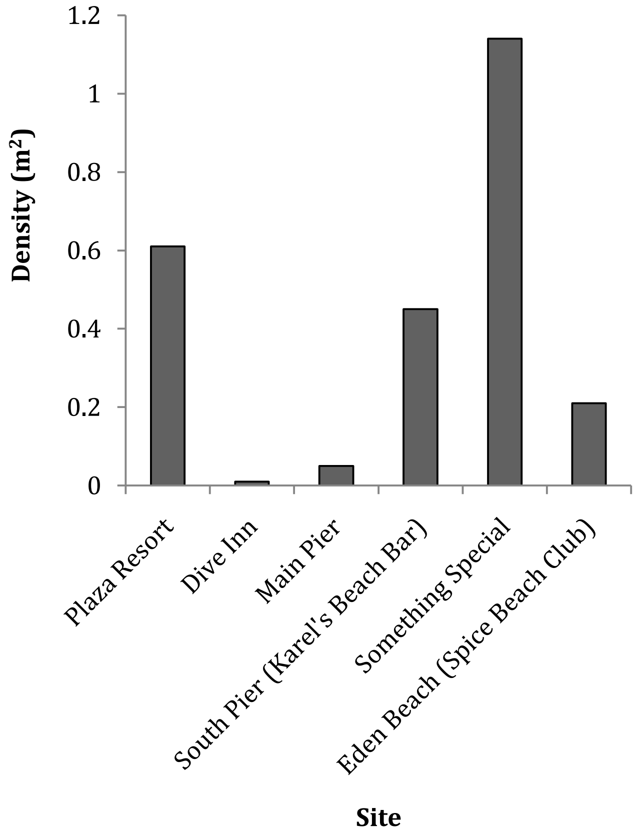

Fig. 4 Density of *T. coccínea per* m^2 at six sites in Kralendijk, Bonaire

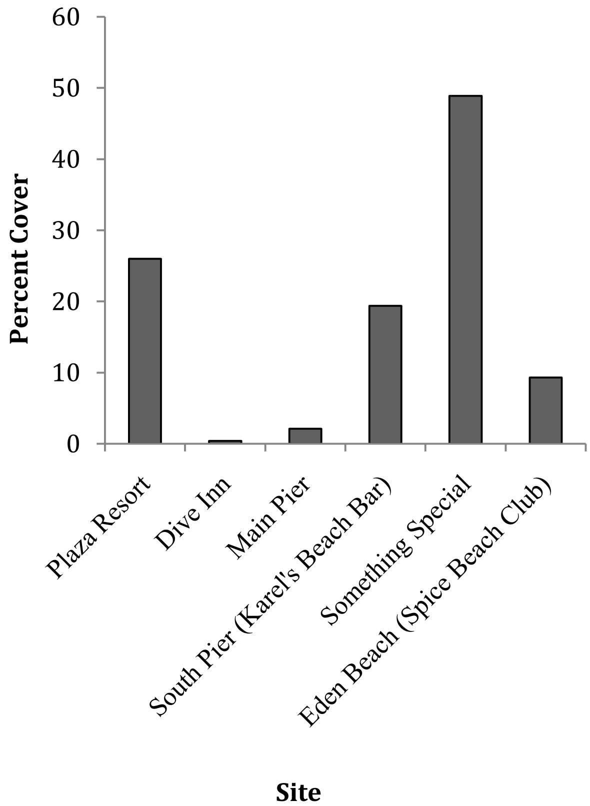

Fig. 3 Percent cover of *T. coccinea* measured in a 30m by 10m survey area at six sites in Kralendijk, Bonaire

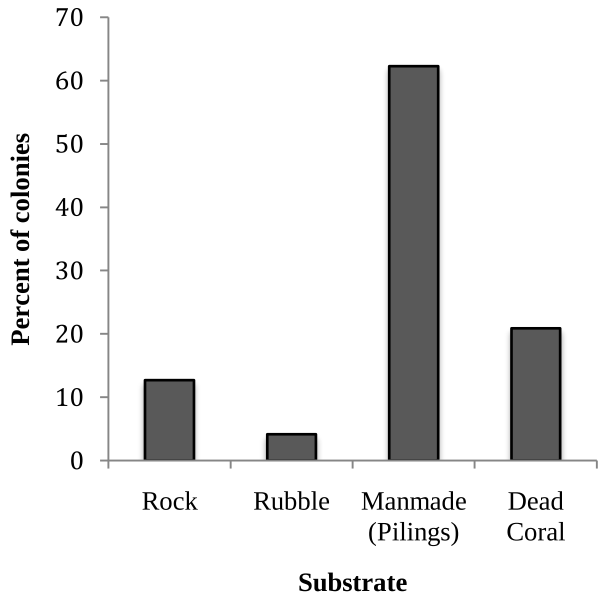

Fig. 5 Total number of *T. coccinea* colonies present on various types of substrate at six sites in Kralendijk, Bonaire

Very little was found on rubble (4.2%), due to it lack of firm structure (Fig. 5).

Something Special was a site dominated by pilings, which contributed to the increased percentage of *T. coccinea* favoring man-made substratum. This site drastically skewed the preference of *T. coccinea* due to it being the only site possessing manmade pilings. If the data from Something Special is removed, *T. coccinea* would prefer dead coral with 55.3% of surveyed colonies present on this substrate, as opposed to only 20.8% total colonies being present on dead coral when manmade pilings are included.

Fluorescence

The use of fluorescence under UV light to detect coral recruits is widely used in coral reef ecology (Piniak et al. 2005). However, the colonies and recruits of *T. coccinea* were observed to not fluoresce under UV lighting. At night while feeding, the stalks or feeding tentacles did not have any color change under UV light instead of white light. Therefore, very small coral recruits could not be observed or measured.

Discussion

Based on the data collected from the six survey sites, the first hypothesis was accepted, while the second hypothesis was rejected. *T. coccinea* was found in highest abundance at sites with shallow, heavy surge areas as hypothesized. South Pier, Something Special, and Eden Beach were sites with shallow structures inhabited by *T. coccinea* that experience consistent strong surge. This environment is unfavorable for other corals, which could explain *T. coccinea's* high density. At these sites, other fauna was not abundant. Due to the constant water movement, these sites contained large amounts of sediment, which could be seen on the sea bottom in the survey areas. Main Pier, Dive Inn, and Plaza Resort survey areas possessed calmer waters, away from the intertidal zone. These sites also possessed more fish, invertebrates, and had some presence of corals, unlike the sites where *T. coccinea* was heavily abundant.

The distribution was not only dependent on the water circulation at the various sites, but the substratum present at each site. The sites were dominated by rocky substratum, with the exception of Plaza Resort and Something Special. The data rejects the hypothesis regarding *T. coccinea*'s preference on rocky substratum. *T. coccinea* naturally inhabits caves and overhangs, which explained the above hypothesis, but instead it was found in greatest abundance on manmade substrate and dead coral. Something Special was dominated by manmade pilings, which explains the high presence of *T. coccinea* on this substrate. Plaza Resort survey area was dominated by dead coral, which in fact is known to represent an environment very desirable for *T. coccinea* by providing crevices and shaded areas for this coral to flourish (Vermeij 2005).

Something Special was a unique site due to the fact that it was completely made up of pilings. *T. coccinea* was most abundant at this site out of the six survey areas and favored manmade substratum more than any other substratum. The pilings were also shaded underneath a dock. *T. coccinea* was completely absent on areas of the pilings in direct sunlight (Fig. 6, image 1), which illustrates *T. coccinea's* natural occurrence in caves and non-photic areas (Vermeij 2005). Various studies in Brazil have shown *T. coccinea*'s presence on oil platforms (Paula and Creed 2005). By settling these structures, *T. coccinea* finds a niche not suitable for other corals or invertebrates.

While surveying *T. coccinea*, relationships with other fauna were observed. Although little is known about *T. coccinea*'s interactions with surrounding species, it is known to compete for space with sponges and invertebrates (Fig. 6, image 2). A known competitor observed in Brazil is the lumpy sponge, *Desmapsamma anchorata* (Meurer et al. 2010), which is a species also present in the Caribbean.

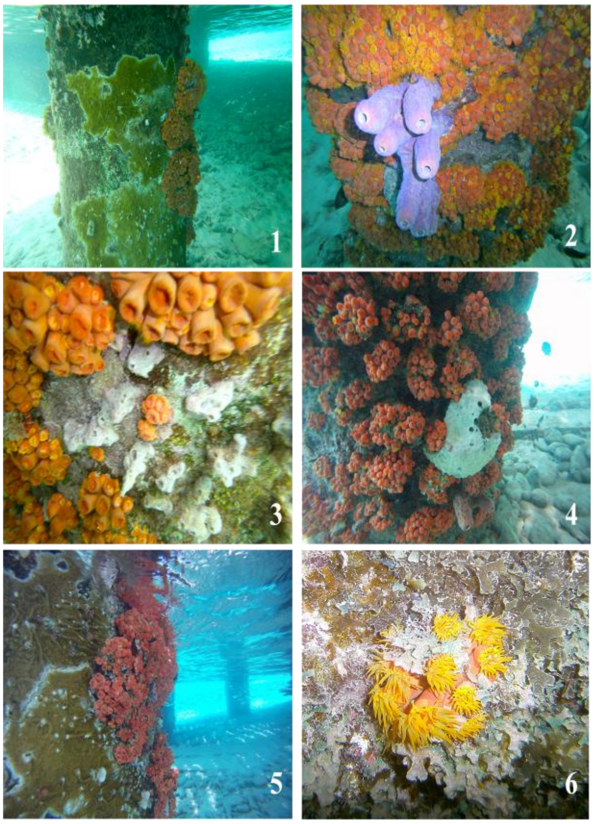

Fig. 6 Images of *T. coccinea* and observed surrounding fauna at survey sites, Something Special (images 1-5) and South Pier (Karel's Beach Bar, image 6)

This is a fast growing sponge, which frequently inhabits similar areas as *T. coccinea*. Dive Inn had a presence of this sponge, which could explain the low abundance of *T. coccinea* at this site. It was also found at Something Special on the pilings out competing *T. coccinea*. The sponge was present and growing outwards, as *T. coccinea* lacked any polyps in very close proximity to the sponge (Fig. 6, image 3). At Something Special and South Pier, *Ircina stroblina*, a ball sponge, was seen in close proximity to many *T. coccinea* colonies (Fig. 6, image 4). Competition for space was evident as the ball sponge was seen overgrowing *T. coccinea* taking any available space around the coral colonies. Along with sponges and many types of macroalgae, fire coral was observed growing near *T. coccinea* (Fig. 6, image 5). Specifically, *Millepora complanata* was the only other coral abundant at the sites. This species is also known to establish itself in less desirable areas for reef corals, or taking over recent dead coral.

While surveying *T. coccinea* at South Pier during the first week of March, the waters were very murky and nutrient filled. This was recorded and after one week, the site was observed for any changes: *T. coccinea* was not observed to have any physical changes, but there was a vast increase in the growth of macroalgae (Fig.6, image 6). This could have been explained by sewage run off or heavy rains, but since there was no rain events in the previous weeks, sewage deposits may be the reason for heavy nutrients at this site. This is a regular occurrence in Bonaire, due to the lack of sewage regulations (Lukasiewicz 2012). This could be used to explain *T. coccinea*'s resistant nature and ability to withstand introduction to various environments.

T. coccinea's success in Bonaire's intertidal is evident, yet its settlement patterns are still unknown (Paz-Garcia et al. 2007). Due to the time constraints of this study, the spawning and planulae larvae could not be observed. Observing the larvae in lab conditions could have provided significant information about the mechanisms of reproduction and settlement (Glynn et al. 2008). *T. coccinea* is known to asexually bud and use "runners" to rapidly expand (Glynn et al. 2008). Runners were not observed at any of the sites. It would be valuable further research to observe the larvae and their preference of substratum when settling (Piniak et al. 2005). As mentioned previously, recruits also were not visible under ultraviolet light. The lack of fluorescence of *T. coccinea* is a very interesting characteristic that also would need to be studied more thoroughly. The mechanism for coral fluorescence is not completely understood, but the lack of fluorescence in *T. coccinea* is not common for corals. The composition of its tentacles and polyps should be further studied for this characteristic.

Even though *T. coccinea* is found in western and eastern Atlantic, Indian Ocean, western and central Pacific and eastern Pacific Oceans, it is still very understudied (Paula and Creed 2005). Its distribution in Bonaire supports previous studies regarding its distribution patterns in shallow, intertidal waters. While its distribution here is out of its native range, it is similar to other areas it has been introduced to. *T. coccinea* has been observed to have changed its behavior in response to its introduction and has filled available niches. In its native range it inhabits caves and deeper waters as stated before, while in the Caribbean it favors shallow, intertidal waters. Exotic species are very successful when they possess the mechanisms to adapt to several environments and make due with the resources they have. *T. coccinea* is a great example of this behavior and can be a species for further comparison and understanding of exotic introductions. From this study it can be deduced that *T. coccinea* is abundant on numerous substrates in Bonaire. Sinces it introduction, it has been most abundant in shallow, heavy surge locations most often present of manmade substratum. These results confirm and expand on the existing data supporting *T. coccinea*'s expansion in the Caribbean. Understanding new exotic species is critically important to protect the present ecosystem and give indication of health on Bonaire's coral reefs.

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Physis (Spring 2013) 13: 17-22

REPORT

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Sediment rates and composition surrounding a disturbed outlet in Bonaire, Dutch Caribbean

Abstract The small island of Bonaire has seen a dramatic increase in visitation due to diving tourism. The economy of Bonaire depends on this influx of visitors and therefore has had an increasing demand for the development of new housing, resorts, and commercial areas. This study examines how the increased human development affects sediment deposition and nutrient load in an area surrounding a combined marina and salina outlet in Bonaire. Sediment load, sediment compositions, and interstitial water quality (water found in the sediment) were analyzed to determine if sediment or nutrient load had an effect on the reef community in the area. The results of this study show no significant trend suggesting reef degradation north of the marina due to point source pollution from the salina outlet. The results, however, only apply to the limited 26 day sampling period. There is a possible seasonal link that could not be explored here without further year-round data collection. These results show that while sediment, nitrogen and phosphate are present in the marine environment north of the marina outlet, it cannot be specifically linked to the marina using data collected in this study. Interstitial water analysis showed no detectable refined oil. The methodology implemented here could be modified to collect additional data in future research.

Keywords Sedimentation • Interstitial water • Coral reefs

Introduction

In the Caribbean, diving tourism has been a main driver behind the increased visitation to the area. Bonaire, a small island in the southern Caribbean, has seen a dramatic increase in visitation since the 1970's (Hawkins et al. 1998). This is predominantly due to its healthy coral reef habitat and dive friendly community. To accommodate this influx of visitors there is increasing demand for the development of new housing, resorts, commercial areas and industrial zones. Housing developments and resorts have changed the landscape considerably, disturbing the limited topsoil and changing the hydrology of many ecologically sensitive areas.

This new construction raises concerns about potential impacts to the coral reefs surrounding the island. The relationship between the terrestrial and marine environments has been overlooked in the past due to the limited interactions with the underwater world. It wasn't until the advent of SCUBA diving that we were able to have intimate interactions with the underwater world. Unfortunately, it has now become clear that modern development on land is having serious consequences on marine environments. Anthropogenic disturbances in coastal environments, such as the recent construction seen in Bonaire, can have direct and indirect impacts on marine ecosystems (Costanza et al. 1997). Sedimentation, pollution, and physical disruption are all threats to coral reefs that can be directly related to anthropogenic activities such as land development, agricultural practices, wastewater dumping, and destructive fishing practices (Bannister et al. 2012, Yoshikawa, 2004).

This study examines how one documented effect of human development, increased sediment load, is affecting the outlet of a salina with an associated marina in Bonaire. Sediment influxes are a serious concern for coral reefs worldwide, and have not been extensively studied for Bonaire's reefs (Fabricius KE 2005; Carballo et al. 2006; Bannister et al. 2011; Tseng et al 2011; Bégin et al . 2013). Specifically this study will look at fine-grained terrigenous sediment (sediment originating from land). This type of sediment has been often been found to be detrimental to coral (Tseng et al. 2011), sponges (Bannister et al. 2011; Carballo et al. 2006) and other benthic organisms (Perry et al. 2011). Terrigenous sediment particles tend to be very small, less than 63 μm (Carballo JL 2006), and interfere with the physiological functions of filter feeders (eg. clog pores). For instance, Bannister et al. (2011) found that fine-grained sediments increased respiration rates in a common reef sponge and therefore high sedimentation can infer "sub-optimal" conditions for sponges. In addition, fine-grained sediments can cause physiological stress and death in coral colonies (Fabricus et al. 2003). Fabricus et al. (2003) concluded that clay sediment rates of 2-12 mg cm^{-2} day^{-1} reduced the ability of *Acropora millepora* to settle and survive. There are regulations in place for developers and construction companies aimed to help retain Bonaire's limited, dry top-soil (STINAPA construction handbook 2010) but their effectiveness remains undocumented.

The need for study on sedimentation rates in Bonaire's coastal waters is increased by the fact that Bonaire is home to a unique type of saline lake ecosystem called a salina. These hypersaline, estuary-like areas are home to many species of seabirds (e.g. flamingos, snowy egret, common tern, etc.) as well as brine shrimp and various salt tolerant algae (Simal, 2009). These fragile ecosystems are catchment basins for rainwater and runoff, which can be particularly problematic when the surrounding landscape is physically modified or polluted. A marina has been established at this outlet making the interactions between the salina and surrounding waters more complex. It is the aim of this study to document the changes in sediment and water quality in relation to the distance away from the outlet of this combined salina and marina outlet.

The coral reefs surrounding Bonaire are within a protected marine park, however, they are not immune to anthropogenic pressures. The coral reefs on Bonaire have suffered from declining health and a decrease in coral cover (Hawkins et al. 1998). The area directly in front of the marina outlet as well as the area directly north of this have been particularly degraded with an area completely lacking coral. Many possible influencing factors have been proposed for the decline in the health of coral worldwide but this study aims to examine what effects sedimentation may be having on Bonaire's coral reefs. Sedimentation on coral reefs from terrestrial erosion and runoff have been proposed as a possible driver of coral reef community change (Bannister et al. 2012; Fabricius 2005; Tseng 2011). This study aimed to examine if the rate of sedimentation and percent grain-size composition was a major driver behind the decline in reef health near this highly modified outlet.

Interstitial water condition is a concern in addition to the sedimentation rates and particle size distribution. Interstitial water is water that resides in the spaces between sand and sediment particles on the ocean bottom. Nutrients and pollutants often settle onto the sea floor with sediments (Szmant & Forrester, 1995). After a polluting event, sediment can also hold pollutants long after the initial polluting event has ceased and pollutants can still be detected in the interstitial water (Salomons 1985). Therefore, interstitial water can help determine if past polluting events may have had a lasting effect in the area.

The information collected here may be useful for park managers in the future to help understand how construction plans might alter the marine environment. It may also help determine if this is an important area of study and long term monitoring or if the effects of sediment in this area could be negligible. Therefore, this paper investigates the following hypothesis:

- H_1:Total sediment rates will be highest at the salina outlet and show a decreasing gradient further away from the outlet due to higher amounts of sediment washing out of the marina than surrounding areas.
- H_2:Terrigenous fraction of sediment load will be highest at the salina outlet as well and will decrease with increasing distance.
- H_3:There will be a pollution gradient leading away from the salina outlet.

Materials and methods

Study site

Bonaire is a small island in the southern Caribbean located 50 km off of the Venezuelan coast. The study area was chosen in the west coast of the island where surf is fairly calm and many anthropogenic influences can be measured. The study site was chosen to test if there was significant sediment input into surrounding waters from North Salina and the associated marina (Fig 1). Preliminary observations showed an obvious change in landscape and reef community from the marina outlet towards the northern sites. Southern sites (1 & 6) were dominated by *Acropora* rubble covered in turf algae. The landscape changed to cyanobacteria covered rubble about 50 m north of the initial sites (sites 2 & 7). It was not until 150 m away from the outlet (sites 4 & 9) that sponges were observed. Towards the end of the study area (sites 5 & 10) corals could be seen with associated invertebrates and reef fish.

North Salina itself and the landscape surrounding have been significantly modified; housing developments have been constructed as well as a road that cuts off the natural connection of the salina to the ocean. A small marina has been established at the outlet of the salina so the level of disturbance was predicted to be higher here.

Sedimentation rate

Simple tube traps were deployed at each of the sample sites (see Fig 1). There were two lines of traps set out starting at the marina outlet: five at 8 m depth and five at 16 m depth. The traps were set at 50 m intervals heading north for a 26-day sampling period starting on the 6^{th} of March 2013.

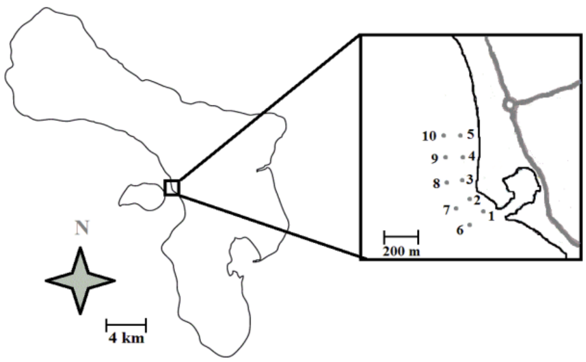

Fig. 1 Five study sites were chosen at both 8 m and 16 m north of a salina/marina outlet on the leeward side of Bonaire, Dutch Caribbean.

The traps had a diameter of 7 cm and a height of 16 cm. Each trap was outfitted with a 1 x 1 cm mesh cap to keep out fish and other potential inhabitants. After 26 days, the traps were capped underwater and removed. The samples were rinsed three times, dried in an oven set to 41°C for 24 hours, and weighed. For this dry-weight measurement grain size was not taken into consideration.

The area of the mouth of the trap was used as the trapping area (38.5 cm^2) and the sedimentation rate was determined by using the formula (Bannister et al. 2012):

$$\left(\frac{(\underline{\textit{sediment weight/sampling period}})}{\textit{trapping area}} \right) *1000$$

Sediment size composition

Sediment samples were taken at each site using a 60 mL syringe. Samples were collected below the sediment trap to avoid any disturbance of the sediment in the trap itself. Each sample was rinsed three times with fresh tap water, dried for 24 hours at 41°C in an oven and weighed.

Using a Retsch AS 200 sieve, the dry sediments were separated by size classifications which included: >2 mm, between 2 mm and 1 mm, between 1 mm and 500 µm, between 500 µm and 250 µm, between 250 µm and 125 µm, between 125 µm and 63 µm and < 63 µm. The proportion by weight was determined for each particle size and the percent composition was determined for the particle size that is < 63µm.

Water quality

Water samples were taken from the interstitial water (water inside the sediment) at each site using a 60 mL syringe with a 60 µm filter. Each sample was tested for refined oil (polynuclear aromatic hydrocarbons or PNAs), nitrate (NO_3^-), nitrite (NO_2^-), and phosphate (PO_4^{3-}) using a Turner Designs Trilogy Fluorometer and suggested protocols.

Data analysis

Correlation tests and regression were used for each group of data collected to see if a gradient existed.

Results

Sedimentation rate

No significant relationship was found between the trap distance from the outlet and the sedimentation rate for the traps at 8 m or 16 m depths (R^2= 0.043 at 8 m and R^2= 0.292 at 16 m) (Fig 2).

Sediment size composition

No significant difference was identified in the fraction of < 63 µm grain class in core samples taken from each site when compared to distance away from North Salina outlet (R^2=0.296 at 8 m and R^2=0.273 at 16 m) (Fig 3).

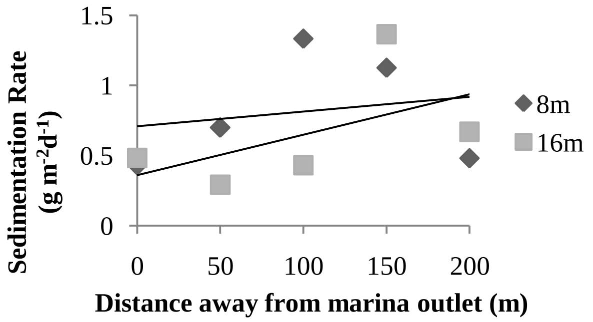

Fig. 2 Sedimentation rate during the 26 day sampling period for sediment traps set at 0, 50,100, 150 and 200 m from the North Salina outlet and separated into depth groups (8 m and 16 m)

Water quality

Refined oil, PO_4^{3-}, NO_2^- and NO_3^- concentration were tested from each water sample. No correlation was found in any of the samples (R^2<0.8) (Fig 3). Refined oil concentrations ranged from 2.53 µM to 4.08 µM. Average PO_4^{3-} concentration was 0.256 µM (SD= $4.09*10^{-5}$) NO_3^- concentration was fairly uniform with a high of 0.04820 µM and a NO_2^- concentration with a high of 0.0281 µM.

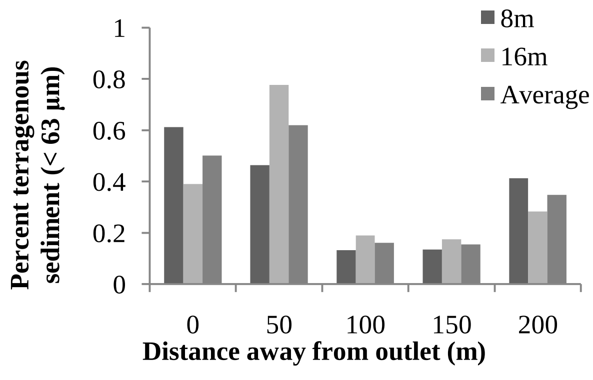

Fig. 3 Depth groups (8 m and 16 m) and averages of sediment grains (<63 µm) percentages were separated from lager core samples from each study site on March 20, 2013.

Discussion

The results showed no significant difference in either sediment (Fig 3 & Fig 4) or water quality when compared with distance. The data collected suggest that the marina and associated salina did not contribute significantly to the sediment, NO_3^-, NO_2^-, or PO_4^{3-} concentration in the adjacent ocean waters during the 26-day sampling period. Refined oil test also was not detectable and suggested protocols indicate that the low levels of florescence detected in each of the samples could have been induced by substances other than PCA's. These results are contrary to the original hypotheses that, considering the high levels of anthropogenic influence in the area, sediment and pollution would be high.

The consistence of measured parameters throughout the sampling area seems to support the conclusion that another driving force could explain the changing underwater landscape that was not explored during this study. It is possible that an event such as the creation of the marina could have had a lasting impact on the area and that these impacts were not testable using the methods presented here. The abundance of rubble in the southern part of the study area indicates that there may have been significant disturbance by a heavy storm event or hurricane (Scheffers & Scheffers 2006). These events could have caused detectable disturbances at or immediately following the event that could not be detected here.

Future research including temperature and salinity measurements may be useful in determining the causes of benthic community change in the area.

The methods employed here may not have been ideal for the study area chosen or the time of year in which this study was conducted. Resuspension of fine grained sediments is often a problem encountered when using STT's (Storlazzi et al. 2011). This likely had little effect on the results presented here as the fine grained sediments in core samples did not differ significantly between sites. The likeliness that fine grained sediment particles will either remain suspended or resuspend with wave action means that it is possible for these particles to travel further and therefore would not have been detected within this study area. In the future, the sample area should be increased from 200 m and extend south as well.

In addition, the changing currents and high boat traffic in the area may complicate the distribution patterns of sediment. In this 26-day study, no replicates were conducted. Since the ocean current conditions here are so variable in direction and strength, it would be necessary to repeat the trapping period several times throughout the year to see if there are seasonal changes in the sedimentation rate (rainy vs. dry seasons). It is not uncommon to see this area flood during periods of heavy rain. Using the sediment trapping techniques presented here during heavy rains and flooding events may help researchers collect useful data in future studies.

Due to the time and technical constraints of this study, the reef change along the study sites could not be explained. However, futher studies need to be done in order to definivly discard sedimentation as a driving force of this change.

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REPORT

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Identifying the cultural value of Bonaire's marine resources

Abstract The island of Bonaire in the lesser Antillean islands is economically dependent on the tourism industry. A majority of tourists who visit the island are scuba divers, drawn by Bonaire's coral reefs. Increased development, tourism, fishing, and pollution threaten Bonaire's marine resources. Survey questionnaires and oral interviews were conducted to determine the interactions between the inhabitants of Bonaire and marine resources, specifically the coral reefs. Four sub-cultures (fishermen, divers, researchers, and others) were identified through their use of marine resources. These subcultures were asked to identify major threats to the reefs and changes in the reefs over the past decade. Overall, 81% of Bonairean residents were able to identify changes to the reef over the past decade, and 77% of residents were able to identify at least one threat to the reef. The freelisted responses were analyzed to determine how sub-cultures interact with marine resources, and although the nature of interactions varied between sub-cultures, the threats listed were similar, identifying a shared communal knowledge of reef ecological importance. Through the advent of the Bonaire Marine Park and the increased access to scuba technology, the Bonairean culture has adapted to include the reefs as part of the cultural identity. Residents on Bonaire have a vested interest in the preservation of Bonaire's marine resources. Public education and increased access to research conducted on the island is suggested to promote community engagement with marine resource management.

Keywords Marine Resource Use • Freelist • Reef Threats

Introduction

The Caribbean (including continental Central America) is one of the world's most tourism dependent areas, as well as a premier diving destination (Hawkins et al. 1999; Green and Donnolly 2003). In many areas of the Caribbean, marine protected areas (MPAs) have been established to help conserve the natural environments surrounding islands. Many surveys and assessments of MPAs have shown that coral reefs protected as MPAs have more numerous and diverse species of fish, especially those species most commonly harvested. There are also increases in coral cover and complexity (Green and Donnelly 2003). The island of Bonaire (Dutch Caribbean), located in the Lesser Antillean island chain off the North coast of Venezuela, established the Bonaire Marine Park (an MPA) in 1979 to preserve the island's marine resources and coral reef environment (Hawkins et al. 1999; Green and Donnelly 2003; Stinapa 2010; Lacle et al. 2012).

After the formation of the marine park, ecotourism based on scuba diving grew rapidly throughout the 1980s and 1990s (Abel 2003). Today, Bonaire's economy is primarily based on tourism due to the pristine marine environment (Hawkins et al. 1999; Lacle et al. 2012). Bonaire is a popular destination for recreational diving and snorkeling (Lacle et al. 2012). The tourism provided by the natural resources on the island benefits the island's inhabitants both economically as well as providing incentives to preserve natural resources, however tourism has been shown to cause damage to coral reefs (Hawkins et al. 1999; Abel 2003; Green and Donnelly 2003).

When Hawkins et al. (1999) conducted research at various dive sites on the island of

Bonaire, they found that frequently dived sites were subjected to higher levels of disturbance indicated by changes in coral communities and a greater loss of massive coral colonies. They suggested that divers exacerbated the stress and tissue damage on corals at different dive sites, causing an increased susceptibility of the corals to infection. Although they did not believe that the divers were the direct cause of coral mortality, the stressors on reefs such as bleaching events and disease acted synergistically with the damage done by divers, which causes greater damage to the reef (Hawkins et al. 1999). A 1992 estimate of the carrying capacity of the marine park found that the 200,000 dives, conducted by 19,000 divers, was at the dive carrying capacity of the marine park (Green and Donnelly 2003). Since the 1992 estimate of the island's diver carrying capacity, the number of divers visiting Bonaire annually has increased to anywhere from 38,000 divers to 70,000 (Lacle et al. 2012; Stinapa 2010). The reefs around Bonaire have been extensively studied and have been found to be an important ecosystem which functions in the flow of energy and resources on Bonaire (Hawkins et al. 1999; Abel 2003). Divers are a cause of damage to the reef, and although Bonaire is dependent on dive tourism as a major source of income, it is necessary to continue to conserve natural resources and inform divers of their impact on the reefs.

Dive tourists represent a major source of income, however, they are not the only users of the marine park. Fishermen are also Marine Park users. The fish upon which fishermen are economically dependent are a common property resource (i.e. a resource available to everyone in the community) (Acheson 1981). Due to the uncertainty of common property resources, which could potentially be available and used by anyone, fishermen "are locked into a system in which it is only logical that they increase their exploitation without limit," (Acheson 1981). Although fishing only represents a small source of income for the island of Bonaire, fishing is part of the local culture. When Lacle et al. (2012) surveyed Bonaireans about their fish consumption habits, 94% of households reported that they had eaten locally caught fish. Furthermore, their study also found that one third of households participated in recreational fishing. In order to prevent the depletion of a cultural resource, it is necessary to conserve fish stocks for fishermen as well as for the tourism industry (Lacle et al. 2012).

Bonaire is a leader both in the conservation of natural resources and the management of MPAs, however the marine resources face pressures from development and pollution, which cannot be directly managed through the marine park (Lacle et al. 2012). When surveyed, the residents of Bonaire stated they felt cruise ships, tourists, and development all posed major threats to the marine environment. They further stated that they wanted a healthy coral reef, but they could not identify important aspects for managing the marine resources (Lacle et al. 2012). The surveyors concluded that Bonaireans were invested in protecting and conserving the island's natural resources because the resources not only play an important role as an ecosystem, but also as a means of subsistence for food and recreational activity.

Based on the results found by Lacle et al. (2012), it is clear that Bonaireans are invested in the protection and conservation of the island's resources. In order to understand how to better implement conservation strategies and programs on the island, it is necessary to identify how the different sub-cultures (researchers, professional divers, fishermen, and nature enthusiasts) interact with each other and Bonaire's natural resources. Therefore it is hypothesized that:

- H_1: Although not all Bonaireans are directly dependent upon marine resources, all participants will have an understanding and appreciation of the ecological importance of Bonaire's reefs
- H_2: There will be a significant difference in how sub-cultures interact with Bonaire's marine resources

Materials and methods

Study site

Due to the need of a convenience sample for survey participants, the majority of research was conducted within the public areas in the city of Kralendijk, on the island of Bonaire (Fig. 1). Some preliminary research involved traveling to Lac Bay to attend an environmental outreach event in order to pre-test survey instruments.

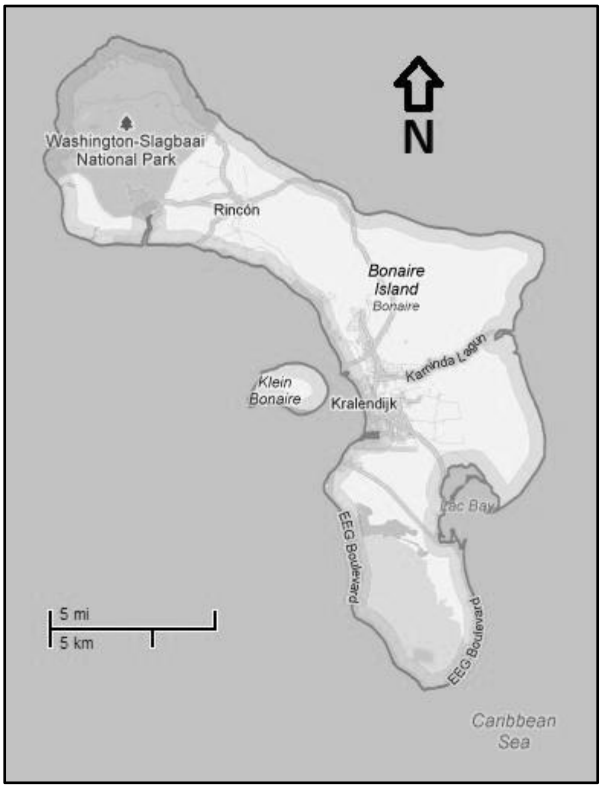

Fig. 2 Map of Bonaire, Dutch Caribbean. (Modified from: https://maps.google.com/)

Data collection

Ethnographic surveys have been shown to be successfully employed when identifying resource use and relationship of people to ecosystems and natural processes (Berkes et al. 2000; Grant and Miller 2004; Aswin and Lauer 2006). Due to the effectiveness of survey work, as seen in the research conducted by Green and Donnelly (2003) and Lacle et al. (2012), a survey was constructed with the purpose of identifying the relationships between Bonaireans and marine resources. Following the methods outlined by Schensul et al. (1999), the questions in the survey were based on the author's prior observations from the field, as well as the factors outlined in the surveys conducted by Lacle et al. (2012). The surveys were given to a convenience sample of the study population as a self-administered questionnaire, a recommended practice when time constraints are present during the research (Schensul et al. 1999). Approximately 75 questionnaires were collected from participants in Kralendijk, representing 0.5 percent of Bonaire's total population (Lacle et al. 2012).

In order to supplement the data collected from questionnaires (Schensul et al. 1999; Aswin and Lauer 2006), oral interviews were conducted with a fisherman/employee in the tourist industry, a professional diver, a scientist, and a citizen scientist/nature enthusiast. The questions in the interview were extrapolated from the questionnaires, however, additional prompts were issued to increase the interviewer's comprehension of responses. Further questions were also included to expand the data concerning the relationship between sub-cultures.

Within both the questionnaire and surveys, freelist prompts (i.e. inventory prompts) were included as open-ended questions due to the validity of identifying familiarity and boundaries of cultural domains (Garibaldi and Turner 2004; Quinlan 2005). Due to the lack of cultural familiarity of the interviewer, the freelists provided cultural insight into various subfactors of the domain experienced by each sub-culture (Quinlan 2005).

Preliminary research

Participant Observation was conducted over a period of eight weeks to familiarize the interviewer and surveyors with the Bonairean culture (Schensul et al. 1999). A pre-testing with a pilot survey, as suggested by Lacle et al. (2012) was conducted with a smaller study sample outside of Kralendijk to address issues with interviewers and the survey itself.

Data analysis

Bivariate and multivariate analyses were conducted to determine the relationships between survey variables (Schensul et al. 1999); furthermore, a salience analysis was conducted in order to rank the responses from freelist questions by frequency and importance to respondents (Quinlan 2005). All statistics were calculated with the statistical platform Microsoft Excel and SPSS. Network mapping was conducted through coding of oral interview recordings and review of participant responses, as well as further participant observation by the interviewer (Trotter 1999). The primary sources of bridges between social networks were identified qualitatively, as well as natural groupings, behaviors spread across groups, and relationships between groups (Trotter 1999).

Results

Questionnaire results

Ocean usage

Fifty-six percent of Island residents reported that they visited the ocean at least once per week (n= 62). On the other hand, only 11% reported that they visited the ocean less than once per year. Overall, 18% of Bonairean residents reported that they never snorkel (n=62), but 30% of people who were born on the island snorkeled at least once a week (n= 62). 59% of people originating in Bonaire reported that they dive several times per month, and 56% of island residents reported that they dove at least several times per month, and 32% reported that they dove at least several times per week (n=62).

Fishing

Overall, 84% of local residents reported that they have eaten locally caught fish (n=62). Furthermore, 86% of residents who had eaten locally caught fish reported to eat it several times per year (n=62). Twenty-seven percent of residents said they received their fish from a local fisherman (n=62). Twenty percent of residents said they caught the fish themselves (n=62), and 13% of residents said that they did not know where their fish came from (n=62) (Fig. 2).

Of people born on Bonaire, 77% reported never eating lionfish (n=17). There was a significant negative correlation between nationality originating within the Caribbean and eating lionfish (n=73, p=0.002). There was also a significant positive correlation between people who ate fish and people who identified as Bonairean (n=70, p<0.05).

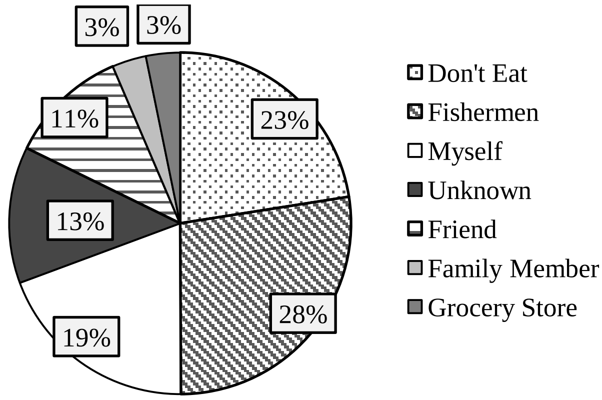

Fig. 2 Sources of locally caught fish consumed by Bonairean residents (n=51)

Threats

Out of all Bonairean residents (those residing permanently on the island), 82% acknowledged that there are threats to Bonaire's reefs (n=62). Out of these, 44% of residents identified Pollution as the primary cause of damage to the reef (n=62). 21% of residents identified Divers as the primary cause of damage (n=62). 11% identified fishermen as the primary cause of damage to the reef (n=62), and 24% identified something else or a combination of factors as the primary cause of damage to the reef (n=62) (Fig. 3).

There was a significantly positive correlation (p<0.001, n=56) between people who were born outside Bonaire and their own identification as a Bonairean. 36% of people who were originally from Bonaire agreed that there were no threats to reefs (n=17).

Furthermore, 47% of people who identified their origin in Bonaire stated that there was no change in the reefs over the past decade (n=17).

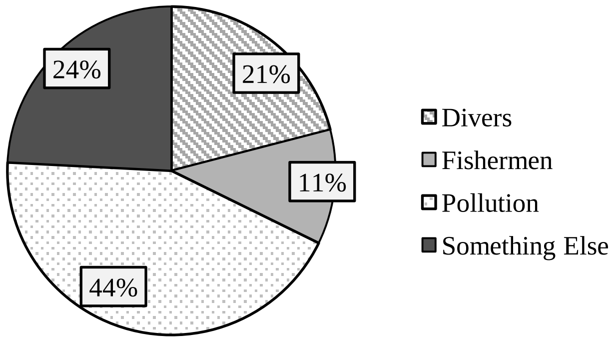

Fig. 3 Primary causes of damage to the reef identified by Bonairean residents (n=62)

Salience analysis

The surveyed population was asked to identify both threats to Bonaire's reefs and changes in the reef that had occurred over the last decade. These responses were then ranked for each respondent following the method outlined by Quinlan (2005). For instance, if the respondent identified two threats (e.g. Divers and Anchors), the first threat would be ranked 2/2 and the second ranked 1/2. The sum of these rankings was then calculated, and divided by the total number of respondents who identified threats in the surveys. The resulting value, called the salience value, is shown in Tables 1 and 2 for identified threats and changes, respectively. A salience value closer to one indicates a more frequently and higher ranked threat.

The responses from survey participants were then used to create word clouds (http://www.wordle.net/) where the size of the word corresponds to the frequency with which it appears in the freelisted responses. The most common words associated with threats to Bonaire's coral reefs were 'Pollution', 'Fishing', 'Divers' and 'Lionfish' (Fig. 4). The most common words associated with noticed changes over the past decade were 'Coral Loss', 'Hurricanes', 'Bleaching', 'Less Coral', and 'Less Fish' (Fig. 5).

Table 1 Composite salience of threats for all survey respondents

Threat	Sum of Ranked Values	Salience Value
Pollution	20.07	0.386
Divers	10.74	0.207
Invasive Species	8.59	0.165
Garbage	6.80	0.131
Tourism	6.35	0.122
Climate Change	5.55	0.107
Sewage Runoff	5.48	0.105
Overfishing	4.86	0.093
Fishing	4.40	0.085
Sunlotion	3.90	0.075

Table 2 Composite salience for top 10 reef changes over the past decade for survey respondents

Change	Sum of Ranked Values	Salience Value
Fewer Corals	15.83	0.337
More Algae	4.25	0.090
Storms	3.25	0.069
Decreased Fish Populations	3.00	0.064
Water Pollution	2.80	0.060
Bleaching	2.66	0.057
Lionfish	2.26	0.048
Less Visibility	2.20	0.047
People	2.16	0.046
Fewer Fish/Fewer Reefs	2.00	0.043

Network analysis

A network analysis was conducted by the author between sub-groups based on the top five salience values for each sub-culture as well as responses from the oral interviews. Fig. 6 is the mapped network analyses between sub-cultures including the top threat identified in the freelisted responses.

Fig. 4 Word cloud generated from freelisted threats to Bonaire's Reefs (n=168 threats identified)

Fig. 5 Word cloud generated from freelisted changes in reefs over the past decade (n=94 changes identified)

Discussion

The results of the study did support the two hypotheses set by the author. When analyzing if Bonairean residents did have an understanding of the ecological importance of the reefs, a freelist analysis of threats and changes indicated that 81% of residents (n=62) had experienced changes in the reefs over the past decade. Furthermore, 77% of residents identified that there were threats to Bonaire's reefs and listed at least one threat (n=62). The sub-cultures did interact differently with Bonaire's marine resources, however, many of the threats that the different groups identified were related (see Fig. 6 and Table 3), indicating that there is a shared ecological understanding of the ecological importance of the reefs on Bonaire.

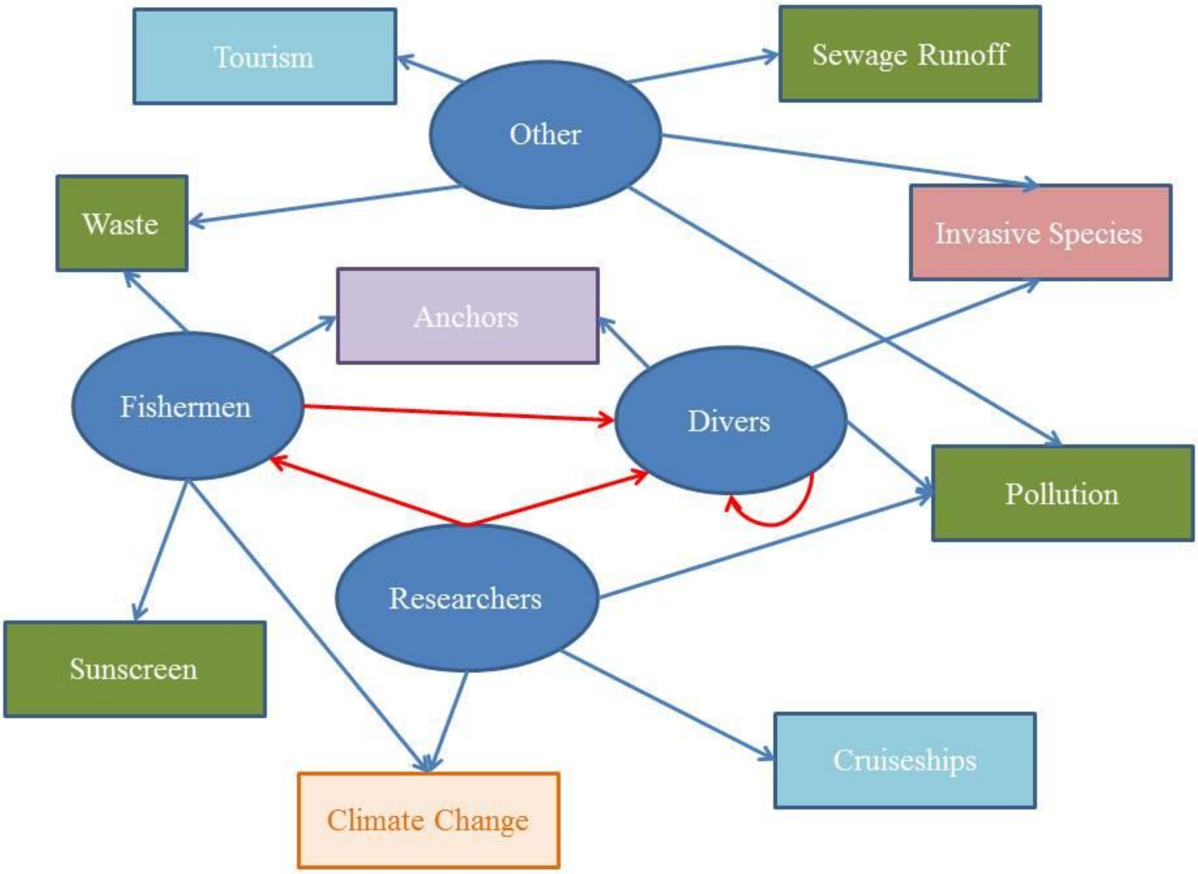

Fig. 6 Network analysis between different sub-cultures (identified in blue circles) and the top five freelisted threats for each sub-culture. One-way arrows indicate threats (named in boxes) identified by the sub-culture. Red arrows indicate the relationships between sub-cultures when one sub-culture identifies another as a threat to reef health. Threats were coded in the same color if they were linked (e.g. waste is a form of pollution, etc.)

Table 3 Top Ranked Threats from Freelisted Threats for Each Sub-Culture

Ranked Threat	Divers (n=12)	Fishermen (n=1)	Researchers (n=6)	Other (n=31)
Threat 1	Pollution	Divers	Pollution	Pollution
Threat 2	Fishing	Sunlotion	Fishing	Invasive Species
Threat 3	Divers	Anchors	Divers	Tourism
Threat 4	Invasive Species	Climate Change	Cruiseships	Sewage Runoff
Threat 5	Anchors	Waste	Climate Change	Waste

Reef cultural significance

When interviewing local island residents, several responses came up indicating that there was very little historical association with the ocean apart from subsistence fishing. One resident who has lived on the island for the past five decades stated that the people who originally lived inland, such as in the village of Rincon (see Fig. 1), did not traditionally interact with the ocean on a daily basis. Furthermore, diving is relatively new to the island, becoming widely popular with the advent of the marine park (Personal Oral Interview; Abel 2003; Green and Donnelly 2003). A conservationist who has immigrated to the island stated in an oral interview that people enjoy the ocean and are definitely aware of the reefs because they are so prevalent in the culture that Bonaire presents to tourists. Another interviewee elaborated on this idea by stating that the local people really don't have a historic interaction with the reef even though the reefs are perhaps "the most important economic asset Bonaire has" (Oral Interview). Although the reefs were not traditionally an important part of the culture on Bonaire, the culture has adapted to include them because they are the drivers of the tourist based economy. Garibaldi and Turner (2004) state that in order to protect ecological resources, it is important to strengthen the relationship between local people and said resources.

From the answers given in oral interviews, it is apparent that the historical cultural significance of the reefs was primarily based on fishing. However, due to the necessity of economic stability, the island of Bonaire has become dependent on the preservation of the reefs and people have adapted to integrate the reefs into the tourist culture. Berkes et al. (2000) state that the way people use resources as an indicator of their observational knowledge of resources and their belief system regarding those resources. In other words, integrating how the Bonairean community has traditionally interacted with the reefs can help adapt management strategies that promote resilience of marine resources. People born outside of Bonaire do identify as Bonairean, suggesting that the cultural identity has evolved to include many cultures and therefore is changing how people interact with the natural environment. Furthermore, as the Bonairean culture has evolved to include reefs as part of Bonairean cultural identity, people have become increasingly aware of the threats to the reefs.

The presence of a moral fishing economy

Out of the 22 respondents who said that they fished, 18% (4 respondents) reported that they sold the fish they caught. Out of these four respondents who do sell their fish, two respondents fished to earn money whereas the other two fished for recreation/enjoyment. Ten of the 22 respondents who have fished identified that they did not fish for food, but for recreation and /or enjoyment. Furthermore, when looking at where residents receive their fish, 42% of respondents stated that they received the fish from a friend, family member, or local fishermen (Fig. 2). The distribution of fish amongst community members in Bonaire could be indicative of the presence of a moral economy, in which resources are redistributed based on relation and kinship values (McCormack 2007). These resources are spread throughout the community based on personal relationships, leading to a more humane but less individual based market economy.

Moral economies often arise in communities that have been colonized, where communities use tradition to challenge the colonial economies based on self-capitalization (McCormack 2007). This tradition counters changes in class systems brought on by capitalist economies and helps counter wealth inequalities and loss of communal resources such as access to fish populations (McCormack 2007). Of course, fishing is a small part of the economy of Bonaire, but it is important to realize that aspects of the commercial and recreational fishing industries could be linked to the presence of a moral economy within Bonairean society. A local fisherman responded through an oral interview that it was a familial tradition to fish. Furthermore, most of the fish caught were given away because there was little need to derive an economic benefit from fishing. Instead, fishing was used as a way to carry on traditional activities. As indicated by the importance of fishing as a traditional activity, the response of the local fisherman is further evidence for the presence of a fishing moral economy. Only one of the 22 respondents who said they fished was a full time fisherman. People who ate locally caught fish identified themselves as Bonairean, indicating that fishing is a vital part of the Bonairean culture. Understanding how the Bonairean culture identifies with fishing can help to look at marine resource use and how resources are distributed throughout the community.

Sub-culture salience threats

Out of all the Bonairean residents surveyed (n=51), 92% responded that there were threats to the reefs and they had experienced changes in the reefs over the past decade. Pollution appears as one of the top five threats (based on salience value) for three out of four sub-cultures (Table 3) indicating that pollution is one of the threats that the Bonairean community is most familiar with and identifies as extremely important (Fig. 4). Pollution was identified as the number one threat overall (Table 1). The fishermen sub-culture (n=1) was the only sub-culture to not identify pollution as a threat, however, waste and sunlotion were both identified as specific threats that can be considered under the category of pollutants. Furthermore, it is hard to use the responses from only one fisherman to represent the threats perceived by all fishermen.

The threats 'Divers', 'Waste/Sewage Runoff', and 'Fishing' are also very prominent categories for the sub-cultures (Table 3). Divers were ranked as the second highest threat to the reef overall (Table 1), and their impact was mentioned by all oral interviewees as a direct cause of damage to the reef structure. The Bonaire Marine Park receives 38,000 annual visitors, twice the number of visitors that were estimated to be the carrying capacity of the park in 1992 (Green and Donnelly 2003; Stinapa 2010). The data collected by Hawkins et al. (1999), which indicated that divers were exacerbating reef stressors, supports divers being identified as a major threat to Bonaire's reefs.

One of the conclusions drawn by Lacle et al. (2012) is that there are tensions between the local population and tourism on the island because residents are economically dependent on tourism, but they felt that expanding tourism had a negative effect on Bonaire's nature. Furthermore, residents also identified that they were interested in better waste management and a restriction on development (Lacle et al. 2012). Waste, sewage runoff, development, and tourism were all listed in the top ten freelisted reef threats supporting the data collected by Lacle et al. (2012). When interviewing a community member, the lack of a waste water treatment facility was prominent in the conversation because a de-salinization plant allowed the island to support a greater number of tourists which in turn generated more waste flowing untreated through the ground water onto the reefs (Oral Interview). The Bonairean community is aware of this threat and has seen declines in the reef that are a result of increased stressors such as pollution (Fig. 5) (Hawkins et al. 1999).

Overall, fishing was identified as the ninth most prominent threat to the reefs (Table 1). A clear distinction was made between commercial fishermen and recreational fishermen (Fisherman oral interview). Commercial fishermen were identified with pelagic fishing, which is not directly linked with use of Bonairean resources but instead with common property resources (Acheson 1981). There is some netting of bait fish on the reef, but most of the damage (identified through oral interviews with each sub-culture) was thought to be done by hooks, nets, and lines getting left on the reef by recreational fishermen. This fishing gear poses the threat of entanglement, which can lead to issues such as the suffocation of sea turtles (Conservationist oral interview). It was suggested by a local fisherman that there should be regulations set on recreational fishing because commercial fishermen are seeing recreational fishermen damaging the reefs through ill practice.

Final thoughts

The results from the study emphasize the importance of Bonaire's reefs in relation to Bonairean identity and the perceived threats to the reefs. Although the reefs have not always been a key part of Bonairean culture, the island has adapted to incorporate reefs into the cultural appeal for tourists. The creation of the Marine Park emphasizes the importance that the reefs have to Bonaire, but with increasing threats from factors such as storms and climate change, it is important to look at how island residents interact with the reefs. Once this is relationship is understood, actions can be taken to limit the negative interactions between people and the reefs, whether they be direct or indirect. Residents were aware of the threats facing the reefs, and although some threats, such as sewage treatment, can only be reversed through participation of local governmental organizations, others, such as the reef damage caused by divers, can be regulated through education and outreach from all sub-cultures.

When interviewing a local member of a reef monitoring foundation, there was some discussion about how research was conducted and presented to the general public. Bonaire's reefs have been the focus of many scientific studies (see Hawkins et al. 1999; Abel 2003; Green and Donnelly 2003) but the results of these studies do not always reach the general public. When speaking with local fishermen, interviewers were denied interviews and survey participants because fishermen have been interviewed and surveyed by researchers and have seen no results from the studies. As a result, only one commercial fisherman was surveyed. When Lacle et al. (2012) conducted survey research, they found that residents were willing to pay for further marine management, but they were unable to identify necessary aspects of marine management.

This study was conducted within a small percentage of the Bonairean population, so a wider pool of respondents may lead a greater understanding of how residents interact with marine resources. Very little anthropological research has been conducted on the island resulting in little background literature on Bonairean society and values. The Bonairean culture has adapted to assign both economic and environmental value to the reefs. Therefore, it is evident that results from scientific studies should be made available to island residents because residents have a vested interest in preserving the reefs. As a result, management plans should be constructed with local input to further community engagement with conservation efforts.

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REPORT

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Changes in the healing rate of *Ventricaria ventricosa* in acidified ocean water

Abstract Given the amount of CO_2 currently being absorbed by the ocean, there is a great deal of research studying the effects of ocean acidification on a variety of species. Considering the relationship between pH and levels of calcium present in the ocean water, the healing process of *Ventricaria ventricosa* is hypothesized to be negatively affected by the decreased pH that is projected for the ocean. *V. ventricosa* is a green alga (*Chlorophyta*) and one of the largest unicellular organisms. When punctured, *V. ventricosa* forms an aggregation ring around the wound which contracts in order to heal the membrane. This research measured the healing rate of *V. ventricosa* in present day ocean water (pH=8.05) as well as acidified ocean water (pH less than 7.0). Individuals of *V. ventricosa* in present pH water conditions were able to heal the puncture wound within 120 minutes, while the individuals in acidified ocean water were not able to heal themselves within the same time frame. It is unknown whether the *V. ventricosa* would eventually heal themselves over a longer period of time or given a greater volume of ocean water; however it is apparent that the decreased levels of calcium in the acidified water had a negative effect on the healing process of *V. ventricosa*. Ocean acidification is likely to affect the basic biological functioning of a variety of marine life, which will face severe difficulties adapting to the acidified ocean water.

Keywords Sea pearls • Ocean acidification • Aggregation ring

Introduction

Ventricaria ventricosa is a unicellular green alga that is found on Caribbean reefs from depths of three feet to depths of 250 feet. Typically, *V. ventricosa* are solitary but sometimes grow in clusters (Humann and Deloach 2002; prs obs). It is one of the largest unicellular organisms, with a diameter of up to ten centimeters (Shepherd et al. 2004), but is more commonly one to five centimeters in diameter (Humann and Deloach 2002). Due to the large size of *V. ventricosa* many studies have investigated the functions and structural aspects of its membrane (Astbury et al. 1932; Preston and Astbury 1937; Tepfer and Cleland 1979). Although unicellular, *V. ventricosa* has a coenocytic structure (i.e. the cell has undergone division but not cytokinesis), and therefore there are many nuclei, chloroplasts, mitochondria and other organelles throughout the cell. This coenocytic structure as well as the structure of the cell membrane allows *V. ventricosa* to heal itself within hours of being punctured (La Claire II 1982; Shepherd et al. 2004). If the puncture wound is less than 100 micrometers in diameter, *V. ventricosa* forms a ring of migrating chloroplasts around the wound, which will then contract centripetally until it closes. Considering that *V. ventricosa* is a unicellular alga, in order to survive when punctured it is necessary to concentrate energy on closing the wound (La Claire II 1982). It is thought that calcium plays a vital role in the healing process. Sugiyama et al. (2000) performed a study using an anti-calcium-dependent protein kinase (anti-CDPK) in order to determine the molecular mechanism of healing. They found that a particular CDPK is an important receptor in the healing process;

therefore, it has been confirmed that calcium is necessary for the wound to heal. By blocking the uptake of calcium (using the anti-CDPK) the aggregation ring around the wound is unable to contract. *V. ventricosa* is also able to heal larger wounds (greater than 150 micrometers) by the formation of protoplasts (La Claire II 1982; Sugiyama et al. 2000). Based on personal observations made in the laboratory, *V. ventricosa* is able to heal itself within hours of a puncture wound, which makes it ideal to study in the laboratory.

The balance of calcium in ocean water is related to the pH. The ocean has been absorbing more CO_2 from the atmosphere due to the increase of anthropogenic activity since 1901. This absorption has caused a 30% increase in hydrogen ion concentrations and future projections predict that the decrease in pH will continue to occur over the next 50 years at a rate faster than has occurred in the past several million years. The current pH is 8.05 and is expected to decrease by 0.15-0.35 by 2100(Schmidt and Ridgwell 2013). As the pH of the oceans is slowly decreasing due to ocean acidification, the availability of calcium in the reef ecosystem is also decreasing (Ocean Acidification 2012; Feely et al. 2006). This change in the availability of inorganic matter has been shown to affect many marine species in a variety of ways, although the majority of studies have focused on scleractinian corals and the affect of decreasing calcium levels on their skeletal growth rates (Wisshak et al. 2012). With decreasing concentrations of calcium in the water, scleractinian corals are unable to grow as fast, giving macroalgae more space to colonize on the reef. Other than indirectly affecting the growth of *V. ventricosa*, it is important to examine the effects of calcium on the wound healing process in *V. ventricosa*. As stated earlier, calcium plays an important role in the healing process and as the pH of a reef decreases, calcium becomes less abundant in the water; therefore it is hypothesized that

H_1: A decrease in pH will cause an increase in the amount of time required to heal puncture wounds in *V. ventricosa* but not prevent the wound from healing

Materials and methods

Study site

V. ventricosa was collected from the reefs of Bonaire at Yellow Submarine and Something Special dive sites between 20 and 100 feet. Yellow Submarine dive site is located at 12°09'36.47"N, 68°16'55.16"W and Something Special is located at 12°09’43.69"N, 68°17’07.79"W (Fig. 1).

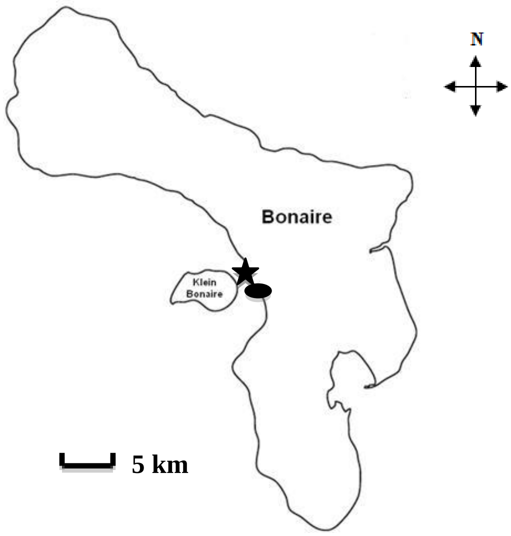

Fig. 1 Map of Bonaire, Dutch Caribbean. Star indicates the location of the dive site Something Special and the circle indicates the location of the dive site Yellow Submarine (Modified from Tyrie 2008)

Data collection

When *V. ventricosa* were found on the reef, data collection included depth, dominant coral and dominant surrounding other macroalgae as well as whether or not they were found in clusters. *V. ventricosa* was removed from the substratum by hand and placed in a labeled bag. In the laboratory, *V. ventricosa* were initially placed in separate containers with ocean water. The water was replaced every four days. The *V. ventricosa* were then placed into a small petri dish completely submerged in ocean water, and punctured with a dissection needle.

A Fisher-Scientific Micromaster microscope was used to observe the puncture wound and subsequent aggregation ring formation at a magnification of 40x. Pictures were taken every five minutes using an AmScope microscope digital camera in order to determine how quickly the aggregation ring contracted. The aggregation ring was measured in each photo using ImageJ 64. Once control measurements were taken for *V. ventricosa*, new individuals were collected and placed in a tank all together. The previously separated *V. ventricosa* were also put in a tank all together and the pH and temperature of the tank was measured using a YSI water quality field meter. The pH of the experimental tank was decreased below 7.0 by bubbling carbon dioxide (CO_2) into the water in order to determine the effects of ocean acidification. The *V. ventricosa* were given 84 hours to acclimate to the new pH and the pH was measured every day. If the pH had risen above 7.0 then more CO_2 was bubbled into the tank. After the acclimation period, the *V. ventricosa* were punctured, completely submerged in the acidified ocean water, and observed as before. Measurements and pictures were once again taken at the beginning of the experiment and then every five minutes until the wound was successfully closed.

Data analysis

Once the data was collected the ImageJ 64 was used to determine the most accurate measure of the area of the aggregation ring. The area of the aggregation ring was measured three times and the average area was used for each image. The percentage of the initial area was then determined for each image. The percentage was then plotted over time and a linear regression was performed for each alga. The slopes of each individual alga's healing process from the control experiment and the increased pH experiment were then compared.

Results

In the field, *Ventricaria ventricosa* were commonly found between depths of 20 and 100 feet. The most prevalent surrounding coral was *Undaria agaricites* and the most prevalent surrounding macroalgae was *Dictyota* spp. It was often more commonly found growing in clusters as opposed to growing solitarily.

Collected *V. ventricosa* were tested under two different water conditions. One tank contained ocean water that was not manipulated (pH=8.05) and one tank contained ocean water where CO_2 was bubbled

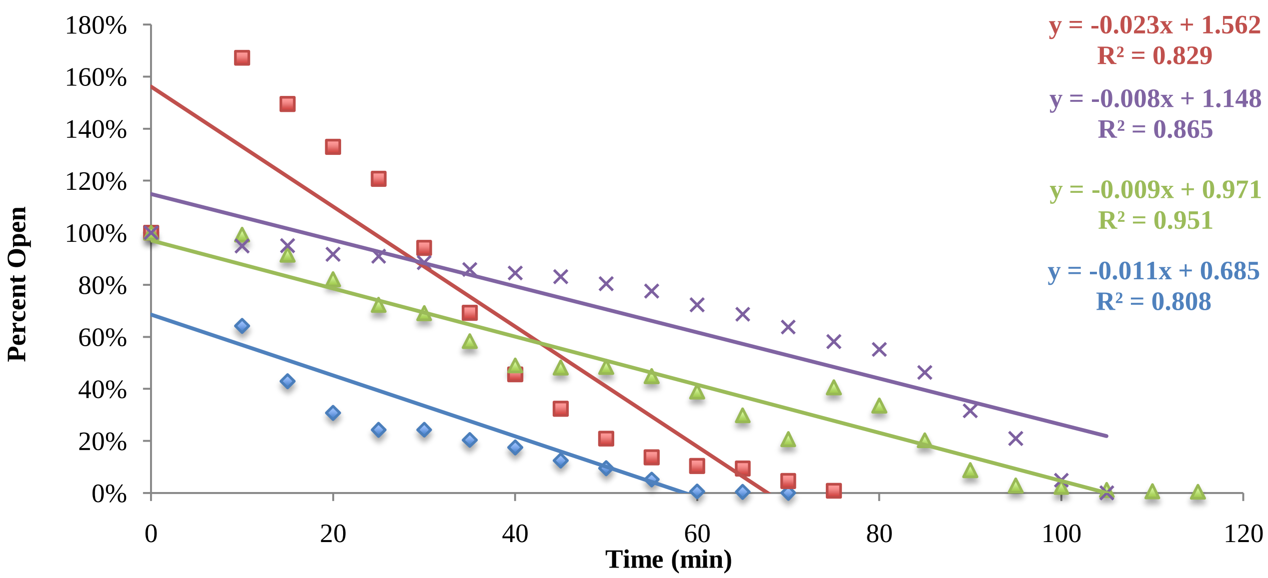

Fig. 2 Healing process of *Ventricaria ventricosa* in present day ocean water (pH=8.05). Each line represents an individual alga. All successfully healed, although trial 2 healed at a faster rate than trials 1, 3 and 4. The time frame for total closure was between 60 and 105 minutes

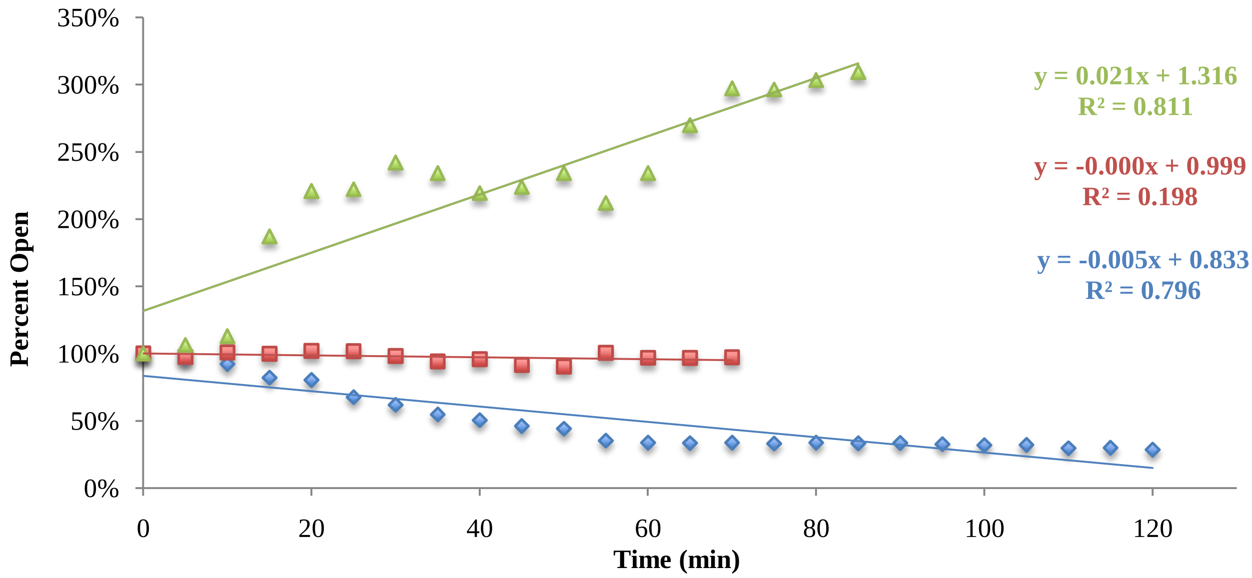

Fig. 3 Healing process of *Ventricaria ventricosa* in manipulated ocean water (pH<7.0). Each line represents an individual alga. None of the *V. ventricosa* successfully healed in the allotted time period. T rials 1 and 2 both closed the puncture very slowly, however trial 3 increased the area of the aggregation ring in the time period studied

through daily to maintain a pH less than 7.0. Fig. 2 shows the four *V. ventricosa* whose healing was measured under present day conditions.

All were successfully healed between 60 and 105 minutes. The rate of healing was similar for trials 1 (slope= -0.0588), 3 (slope= -0.0467) and 4 (slope= -0.045). Trial 2 healed at double the rate (slope= -0.1202) of the other three trials.

Fig. 3 shows the healing process of *V. ventricosa* when their environment had a decreased pH. The *V. ventricosa* were only observed for 120 minutes in order to be consistent with the previous studied data (present day condition). Each individual alga responded differently to the change in pH. Trial 1 slowly contracted the aggregation ring around the puncture wound. Trial 2 formed an aggregation ring but it never contracted, and trial 3 formed an aggregation ring that increased in size. In all trials, the aggregation ring was beginning to break apart by the end of the 120 minutes.

The range of the healing rate of the *V. ventricosa* in the present day pH was from -0.045 to -0.1202. The range of the healing rate of the *V*, *ventricosa* in the ocean acidification pH was from -0.0057 to 0.0216 (Table 1).

Table 1 Comparison of slopes for individual healing rates of *Ventricaria ventricosa*.

	Present Day pH	Ocean Acidification pH
Trial 1	-0.0588	-0.0057
Trial 2	-0.1202	-0.0007
Trial 3	-0.0467	0.0216
Trial 4	-0.0450	X

Discussion

The healing process of *Ventricaria ventricosa* occurred at a similar rate for the majority of the alga in the control group with present day pH. Trial two showed a large increase in area initially, and then healed at a faster rate than the other three trials. It would be interesting to test more alga to determine if the aggregation ring frequently gets larger initially or if it is a rare occurrence. Each alga was able to heal completely over a period of 120 minutes, although two algae healed in 80 minutes or less, which was consistent with the findings of La Claire II (1982).

The healing process of the *V. ventricosa* in the ocean acidification trials varied between individuals. The consistent finding in all trials was that the *V. ventricosa* in acidified water (pH<7.0) were unable to heal the puncture wound in the same amount of time as the *V. ventricosa* in the present day water. In all trials the *V. ventricosa* in the ocean acidification water the aggregation ring around the puncture wound appeared to be breaking apart. If this is true, then the alga is unable to heal the wound in acidified ocean water. This further supports the conclusion that calcium is an important factor in the alga's healing process. It does not support the hypothesis that the alga would still be able to heal the wound in acidified water; however in order to determine this the alga would have to be observed over a longer period of time. It was shown that the rate of healing of *V. ventricosa* decreased dramatically in the acidified water.

Based on the images and videos taken throughout the healing process, it is clear that the aggregation ring is formed by chloroplasts moving to and surrounding the wound site. As time went on, the surrounding area of the wound got lighter in color, which may correspond to the removal of organelles from the opening (La Claire II 1982). Further studies could examine the color and appearance of the surrounding membrane in a given time frame after the healing of the wound in order to determine how long it takes for the organelles to return to that area of the cell.

Overall this research proved to be challenging in a variety of ways. Initially many different sizes of *V. ventricosa* were punctured in order to determine the effect on the rate of healing. Through trial and error it was discovered that under present day conditions the larger *V. ventricosa* took much longer than two hours to heal and many did not heal at all. This may have been because there was not enough calcium (or other inorganic matter) in the water they were submerged in. In some cases the puncture wound was difficult to locate because of microalgae growing on the *V. ventricosa*. In these cases the *V. ventricosa* were placed in a separate container where they were still able to heal themselves, although the healing process was not recorded. Another challenge of this experiment was maintaining the pH of the ocean acidification tank. It was checked daily, although for future experiments it is advised that the pH be monitored more closely. Future studies could also include exposing the *V. ventricosa* to a wider range of pH (i.e. 6.5, 7, 7.5, 8, 8.5) to establish a more precise relationship between pH and the healing rate of *V. ventricosa*.

Along with many other organisms, the future projection of changes in pH of the ocean seems to have an effect on the healing process of *V. ventricosa*. Many people concerned with coral reef ecosystems may not find this problematic due to the fact that a healthy reef generally has low levels of macroalgae. However, as research continues on ocean acidification, more and more species are being found to be negatively affected. There are rare cases in which organisms are able to adapt, however this appears to be the exception rather than the rule (Foo et al. 2012). In order to maintain the current biodiversity found in coral reef ecosystems, the rate of CO_2 absorption would have to decrease dramatically.

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REPORT

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Underrepresentation of eels in AGRRA and REEF fish surveys in Bonaire

Abstract Eels play an important role in coral reef ecosystems as part of nuclear hunting groups and as top predators. They are often underrepresented in fish population surveys due to their hiding behavior. There is consensus among the scientific community that most visual survey methods are not able to give an accurate measurement of the eels in the area surveyed. This study conducted surveys using the methods of two environmental and research organizations: the Reef Environmental Education Foundation (REEF) and the Atlantic and Gulf Rapid Reef Assessment (AGRRA). The surveys conducted in this study looked specifically for eels and the results were compared with historic data collected using both methods. This comparison indicates how accurate these methods are at representing eel populations in Bonaire. Using the same protocols but looking specifically for eels this study found eels more frequently and in greater numbers than the REEF surveys from the online database. The density of eels found using the AGRRA protocol was higher but not significantly different than the density reported in a similar study from 2011. The conclusions from this study are that the REEF methodology, while excellent at reporting species diversity, in not a very accurate method when it reports eel densities. The AGRRA method is about as accurate as a visual survey method can be for eels and allows the data to be reported in a much more usable manner.

Keywords REEF • AGRRA • visual survey

Introduction

Coral reefs are very diverse ecosystems that have lots of checks and balances wrought into their structure (Polovina 1984). Corals, phytoplankton and algae are primary producers kept in balance by grazers and coralivores (Glynn et. al. 1982, Polunin and Klumpp 1992). Populations of these primary consumers are in turn checked by predators such as sharks, groupers, snappers, and eels (Polovina 1984). Eels are mostly nocturnal in the Caribbean and as such are not as visible as some other predatory fish; however they play an important role in the ecosystem (Parrish et al. 1986, Young and Winn 2003).

There have been many observations of eels hunting in cooperation with other predators (Strand 1988). In these interactions the eels are foraging the small cracks and crevices of the reef for small fish, while the following snappers or jacks scare prey into holes on the reef (Hixon and Beets 1993). If a prey fish leaves its hiding place on the reef to escape the eel, it runs straight towards the following predators (Strand 1988). This nuclear hunting behavior is another reason that eels are an important factor in the ecosystem; they have an effect on the populations of prey species beyond what they eat themselves because they aid the hunting of other predators (Parrish et al. 1986).

Marine ecologists worldwide are trying to ascertain the current status of our coral reefs using standardized survey methods. Some of these surveys are intended for use by citizen-scientists, allowing recreational divers to assist in determining the reef’s condition. The two programs that are analyzed in this study are the Atlantic and Gulf Rapid Reef Assessment

(AGRRA) and the Reef Environmental Education Foundation (REEF). These organizations have multiple surveys with different target taxa, namely coral or fish surveys.

This study attempts to determine if eels are over looked in the fish surveys when they are hiding in the benthos. AGRRA and REEF surveys may lead to an incorrect representation of the number of eels in the surveyed area. This is due to the fact that generally the surveying diver does not spend a large amount of time looking in and under the corals or rocks that make up the reef structure where many eels are hiding. The majority of the surveys are conducted in the daylight, so the number of observed eels is likely to be lower than the actual number of eels in the survey area due to their hiding behavior.

The data from the AGRRA and REEF surveys are handled in different ways; REEF data is compiled in an online database that is accessible to the public (REEF 2013), while the AGRRA data belong to the surveyor (Lang et al. 2010) and will only be publicly accessible if the surveyors publish their work. The REEF database has nearly 170,000 surveys recorded to date providing a huge amount of data that can be very useful to scientists in the Caribbean. However REEF data, reported with species densities only being reported as part of a general category (single, few, many, or abundant), may inhibit the analysis of populations of species that are often seen in the few category (2-10 individuals) (REEF 2013). This means that in a survey recording only three eels of a certain species it will be reported in the database in the same category as a survey with 10 eels. This is a huge increase in the number of eels that is not reflected in the database. This is one of the important limitations of the REEF method that makes it difficult to detect changes in populations of some species. For these reasons this study will investigate if:

H_1: REEF surveys give a disproportionally low representation of eel populations on Bonaire

H_2: AGRRA surveys give a disproportionally low representation of eel populations on Bonaire

Materials and methods

REEF protocol

The REEF methodology is referred to as the “roving diver method” and it does not structure the citizen scientist’s dive. It requires the surveyor to record all the species of fish they see on the dive and their abundance (REEF 2013). The description of the method on the website does encourage surveyors to look in and under the reef structure for fish, but it doesn’t require the surveyor to do this over the entire area that is surveyed (REEF 2013).

There are five levels of REEF citizen-scientists; the first three are novice levels while the last two are expert. REEF divers are required to positively identify fish and are given lists of species that they are required to know for each level of certification. Level three novice divers must have completed 25 surveys and be able to identify around 200 species, while level five experts can identify over 450 species of fish and must have completed at least 50 surveys. In order to be certified to a higher level, divers must pass an identification test of up to 100 fish.

All the data collected by the divers is entered into a form online that compiles and analyzes all the data. This data can be sorted by the viewer to look at survey site or fish family, and then by date of survey and surveyor level. The database reports the sighting frequency of each species as well as the density index. This index is recorded as a number from one to four with one meaning single, two meaning two to ten, three meaning 11-100, and four meaning more than 100. The average density that is shown in the database shows the density number that the species is most commonly observed in.

AGRRA protocol

The AGRRA fish method requires divers to swim a 30m belt transect using a 1m T-bar to measure a 2m wide belt, recording the number and estimated size of each AGRRA fish species they come across (Lang et al. 2010). AGRRA has compiled a list of species considered important, either commercially, ecologically or notably invasive species (Lang et al. 2010). Surveyors use 10cm increments on the T-bar to estimate size classes of fish they record in 10cm categories (Lang et al. 2010).

Study site

All surveys for both the REEF and AGRRA methodologies were conducted at the Yellow Sub (12°09'36.47"N 68°16'55.16"W) dive site near Kralendijk, Bonaire, from 25-60ft deep. (Fig. 1)

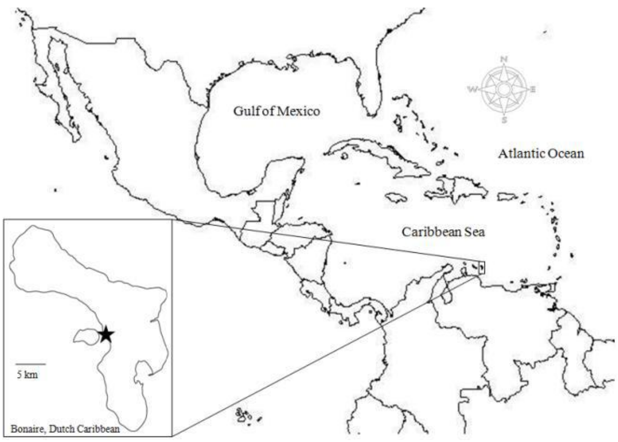

Fig. 1 Black star shows the location of Yellow Submarine dive site in Bonaire in the Netherland Antilles

Data analysis

Data collected during REEF surveys was transcribed into REEF density categories. Data were then compared to 114 surveys previously conducted from 2008-2013 at the Yellow Sub dive site. A one-sample t-test in Excel was used to analyze this comparison.

Data collected during AGRRA transects were used to calculate density (eels $100m^{-2}$). The AGRRA data from this study were compared to data from a Bonaire status report conducted in Steneck et al. (2011) using an independent sample t-test in Excel.

Results

REEF protocol

The sighting frequency of four common eel species (spotted moray; *Gymnothorax moringa*, sharptail eel; *Myrichthys breviceps*, goldetail moray *Gymnothorax miliaris*, and chain moray *Echidna catenata*) in this study was higher than recorded in the REEF database (Fig. 2). It was not possible to run statistical analysis on the results because the raw data from REEF was not attained due to time constraints; however it was possible to determine that sighting frequencies from this study were higher than sighting frequencies in the database for *G. moringa* (100% - 77.7%) and *M. breviceps* (53.3% - 42.0%), but were the same or slightly lower for *G. miliaris* (33.3% - 34.8%) and *E. catenata* (40.0% - 33.0%). Sighting frequencies at the expert level were almost always greater than those at the novice level (Fig. 2).

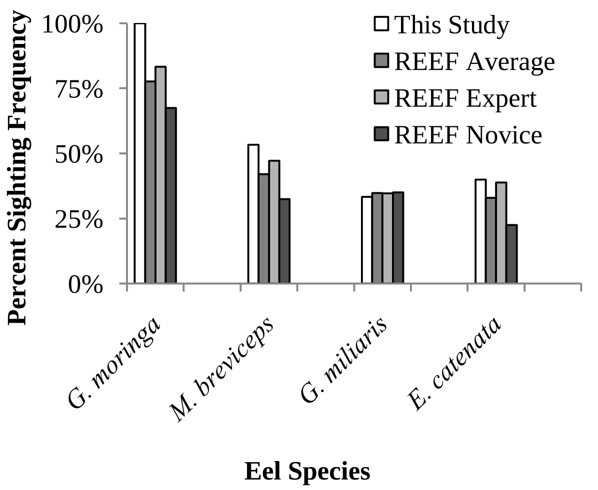

Fig. 2 Comparison between sighting frequency of four common eels (*Gymnothorax moringa*, *Myrichthys breviceps*, *Gymnothorax miliaris*, and *Echidna catenata*) found in this study and REEF database (2008 - 2013) at Yellow Submarine dive site, Bonaire. The REEF data show the Expert and Novice levels as well as the average of the two

The density index (Den) used by REEF is calculated using the following formula:

$$Den = \frac{(S*1)(F*2)(M*3)(A*4)}{\text{Number of surveys in which species was present}}$$

Where S= #surveys with 1 eel, F= #surveys with 2-10 eels, M= #surveys with 11-100 eels, and A= #surveys with +100 eels.

The density indexes in this study were significantly higher than those reported in the database for *G. moringa* (Den 1.9 - Den 1.6 respectively, df=15, p=0.0002) and *M. breviceps* (Den 1.8 - Den 1.3 respectively, df=15 p=0.0285), but were not for *E. catenata* (Den 1.2 - Den 1.2). The density index for *G. miliaris* was lower than reported in the database (Den 1.0 - Den 1.2), but it could not be analyzed statistically because no variance within the sample was recorded (Fig. 3).

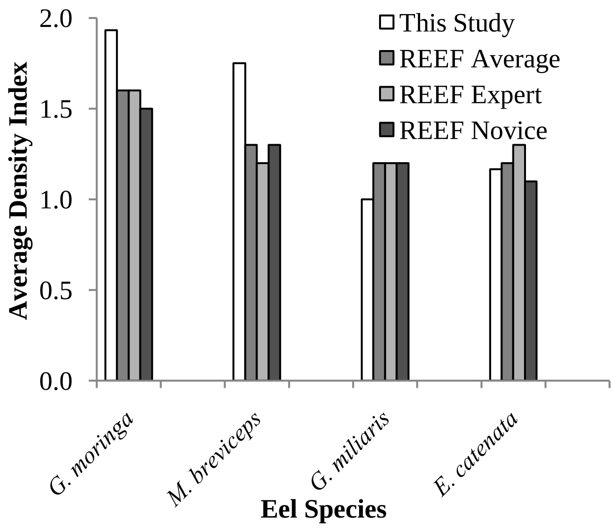

Fig. 3 Comparison between density index of four common eels (*Gymnothorax moringa, Myrichthys breviceps, Gymnothorax miliaris,* and *Echidna catenata*) found in this study and REEF database (2008 - 2013) at Yellow Submarine dive site, Bonaire. The REEF data show the Expert and Novice levels as well as the average of the two

AGRRA protocol

The density of eel species found in this study's AGRRA surveys were higher than those from Steneck et al. 2011 at two nearby locations (0.625 eels $100m^{-2}$ - 0.450 eels $100m^{-2}$ respectively) (Fig. 4), however the difference proved not to be statistically significant.

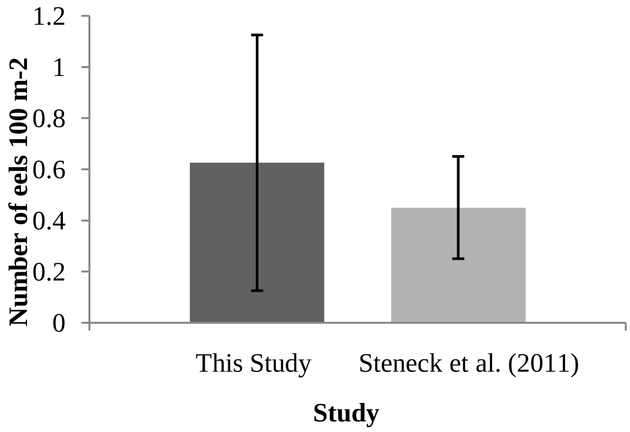

Fig. 4 The *black bar* shows eel density found at Yellow Sub dive site, Bonaire. The *grey bar* shows eel density reported in Steneck et al. 2011 at two similar sites. Error bars show standard deviation

Discussion

REEF

The hypothesis that REEF's method underestimates the eel population in Bonaire is supported by the results of this study but not, however universally. The two most common eel species (*G. moringa* and *M. breviceps*) were found both with a higher frequency and in higher concentrations; however the less common eels (*G. miliaris* and *E. catenata*) were found with similar frequency and in roughly the same densities. It has been concluded that this is most likely due to sample size.

Several other eels species reported in the REEF database that were not mentioned in this study due to their low sighting frequency (<20%) and the number of surveys that this study conducted. The only species of eel that was reported in the REEF database at a frequency greater than 20% that was not observed in this study was the brown garden eel (*Heteroconger longissimus*). It is thought that they were never found during these surveys due to their inhabitance of sand patches deeper than 30ft. The reef crest at Yellow Submarine dive site is at about 25ft and there are few sandy patches shallower than 60ft Therefore, even though the sighting frequency of *H. longissimus* is reported at ~ 35% in the REEF database, they were never recorded during this study.

Holt et al. (2013) shows that the REEF volunteer survey method is better at representing species diversity than methods which restrict the survey area; however the way REEF database reports the density index makes it almost impossible to compare it to any other density number found using a different method. The database also reports in very wide density categories, indicating a very limited ability to detect changes in a population if it stays within a given range. A 70%-80% decrease in the number of eels could be reported the same. This means that the REEF method is not very useful for tracking fine changes of density within a given population. The most applicable information from REEF is the sighting frequency which allows large scale density to be inferred on hundreds of species.

The fact that REEF uses citizen-scientists to collect data allows for the collection of huge amounts of data in short periods of time over wide ranges of habitats. This resource is very valuable, but could be made more relevant if the categories in which species are reported were made more accurate, for instance if the surveyors recorded densities up to 10 or 15 as a precise number and then the categories began. This would allow a greater ability to detect changes in the populations of any species that is normally seen in low numbers.

AGRRA

There were more eels found in the AGRRA surveys of this project than those in Steneck et al. (2011), but these differences were not found to be statistically significant. This does not support the hypothesis that the AGRRA surveys underestimate the populations of eels in Bonaire; however there have been studies done using modified visual census methods that have found eel concentrations as high as 4.48 per $100m^{-2}$ (Gilbert et al. 2005), which is much higher than what was reported in either this study or Steneck et al. (2011). Therefore the hypothesis is not directly supported or refuted by the data from this study, but there are strong indications from previous studies (Christensen and Winterbottom 1981) that the AGRRA method and most other visual survey methods underestimate eel populations.

The reason that even this study looking for eels specifically could not find as many as were found in Gilbert et al. (2005) is thought to be that all the transects surveyed in Gilbert et al. (2005) were counted twice, once in the day and again that night. They recorded size and location of each eel counted in the daytime survey to avoid counting individuals again at night, but they still found a many more eels. The discrepancy between their results and the results of this study are likely to be because there is increased eel activity after sundown, and none of the surveys in this study were conducted at night.

The only eel species that were recorded on the AGRRA transects were the spotted moray (*G. moringa*) and the goldentail moray (*G. miliaris*). Gilbert et al. (2005) found both of these species as well, but chestnut morays (*Enchelycore carychroa*) and viper morays (*Enchelynassa formosa*) were also recorded during the night portions of the surveys. Both of these eels are small (<2ft long) and rarely seen during the day. This further reinforces the main difference between these two studies - one surveyed at night while the other did not.

It would be interesting to analyze the AGRRA method by conducting an AGRRA survey, and then on the same transect conduct the modified census method used in Gilbert et al. (2005). This could provide a very accurate indication of how accurate AGRRA is at determining eel populations.

Conclusions

While both the AGRRA and REEF survey methods are very useful to researchers, they may not report eel populations very accurately. This could be moderated by changing how REEF records and reports their data, but it has been found (Christensen and Winterbottom

1981, Parrish et al. 1986) that all visual surveys underestimate populations of eels on a coral reef.

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REPORT

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Effectiveness of the burglar alarm hypothesis: a comparison between bioluminescent displays in dinoflagellates and abundance of copepods at various depths

Abstract The burglar alarm hypothesis states that dinoflagellate bioluminescence is used to avoid predation from copepods by taking an altruistic approach. This study aims to test the effectiveness of the in-situ dinoflagellate defense mechanism in the waters of the western coast of Bonaire, (Dutch Caribbean). According to the burglar alarm hypothesis, areas of higher bioluminescence should have lower abundances of copepods, however there is little evidence supporting this. To analyze the effectiveness of the burglar alarm hypothesis, horizontal tows were used to collect plankton samples at four depths: the surface, 10, 20, and 30 feet. Dinoflagellate bioluminescent display frequency was recorded immediately after collection in a dark room. Copepods and Dinoflagellates were then identified under a compound microscope and abundance was analyzed. Two-way ANOVA tests showed significant relationships between the decreased ratio of copepod to dinoflagellate abundance associated with increased bioluminescence frequency and increased bioluminescence frequency in relation to increased depth. No significant difference was found in bioluminescent displays in comparison with various weather conditions. This study shows that the burglar alarm has positive effects in maintaining dinoflagellate populations. The environment at depth allows for more dinoflagellate bioluminescent displays to occur than at the surface. Implications of light pollution may cause variations in bioluminescence at shallower depths, lessening the effectiveness of the luminescent defense mechanism of dinoflagellates. Comparisons of biolumi-nescence between the areas exposed to light pollution and areas sheltered from ambient light can be used to analyze how the primary trophic level will change with increasing human impacts.

Keywords Bioluminescence • Dinoflagellates • Burglar Alarm Hypothesis

Introduction

Over 700 different genera of organisms, both terrestrial and marine, have the ability to display a biological light, known as bioluminescence, which can be displayed in the absence of external light (Widder 2010). Bioluminescence is an internal biochemical process allowing organisms to produce a cold-light through the luciferase catalyzed chemical oxidation of luciferin (Goto 1968). The chemical processes and physical displays vary between the wide range of organisms with the adaptation, however the functionality of these luminescent presentations serve similar purposes: mate attraction, food location, communication and or predator avoidance.

Although it is more common to find bioluminescent marine organisms below the epipelagic due to the lack of sunlight, bioluminescence can easily be found at the sea surface in various planktonic marine organisms. Several species of dinoflagellates have developed a bioluminescent defense mechanism for the purpose of predator avoidance, known as the burglar alarm (Widder 2010, Burkenroad 1943). According to the burglar alarm hypothesis, the luminescent display produced by the dinoflagellate is used to startle the copepod, allowing the dinoflagellate that exhibited the light and other dinoflagellates in a close proximity to flee. One variation of this hypothesis explains that the bioluminescent display is used to attract the copepod as well as its predator. The light displays the position of the dinoflagellate to the copepod and to the copepod's predator (Abrahams et al. 1993). This multi-trophic level interaction assumes that the dinoflagellate is sacrificing itself, for the benefit of the population, a phenomenon known as altruism. A second variation of this theory states that dinoflagellates, which get eaten by copepods, are able to luminesce while inside the translucent body of the copepod, thus attracting the copepod's predator to its location (Fogg 1991).

Higher fish feeding rates on copepods occur when dinoflagellate luminescent displays are present compared to when the displays are absent (Abrahams 1993). Copepod swimming behavior during feeding is also shown to be more rapid and erratic in the presence of bioluminescent dinoflagellates as opposed to feeding on non-bioluminescent dinoflagellates (Busky et al 1983). Abrahams (1993) and Busky's (1983) studies support the burglar alarm hypothesis, indicating that luminescent displays by dinoflagellates have an effect on the behavior of copepods and their predators. These results support the theory that dinoflagellate bioluminescence is used in a multi trophic level interaction to avoid predation.

The purpose of this study was to provide a basis for understanding the ecological aspect of the bioluminescent behavior of dinoflagellates and the abundance of its natural predator, the copepod. Thus far, only one study, by Pilla (2012), relating to bioluminescent dino-flagellates in Bonaire has been conducted; dinoflagellate bioluminescent activity was shown to be significantly higher in the evenings (18:30-20:00) compared to the mornings (6:30-8:00). Copepods and dino-flagellates play an important role in the trophic structure of coral reefs and influence the abundance and behavior of organisms in higher trophic levels (Fogg 1991). By studying the ecological function of dinoflagellate bioluminescence in waters surrounding Bonaire, the relationship between dino-flagellate and copepod population dynamics can be determined more accurately. This study intends to answer the following three questions: 1) Is there a relationship between the frequency of dinoflagellate bioluminescent displays and the abundance of copepods? 2) How does depth affect the frequency and intensity of dinoflagellate bioluminescence? and 3) Does sunlight influence dinoflagellate bioluminescence therefore the effectiveness of the burglar alarm hypothesis? The following hypotheses are proposed:

H_1: Areas with high intensity/frequency bioluminescent dinoflagellates will have a lower abundance of copepods than areas with low intensity/frequency bioluminescent dinoflagellates.

According to Burkenroad's (1943) burglar alarm hypothesis, dinoflagellate bioluminescence is used as a defense mechanism against copepod predation. Assuming this relationship exists, the abundance of copepods should be decrease with an increase of more luminescing dinoflagellates.

H_2: Bioluminescent display frequency will increase with depth.

Copepods exhibit diurnal vertical migration patterns; they remain deeper in the water column during daylight and migrate towards the sea surface at night. Dinoflagellates deeper in the water column have a greater exposure to copepods than dinoflagellates that remain higher. To combat this increased predator-prey interaction, dinoflagellates that are deeper in the water column should have more frequent and brighter bioluminescent displays. In addition, there is an increase of light pollution from boats and buildings on the shoreline of our study site that may interfere the dinoflagellates' defense mechanism. If there is ample light illuminating the dinoflagellates, the bioluminescence won't be as bright due to ambient light which is also present. Ambient light not only affects the circadian rhythm of dinoflagellate bioluminescence (Fritz et al. 1990), it also makes the luminescent displays appear more dim and less frequent.

H_3: Bioluminescent displays should be more intense and frequent on sunny days than cloudy/rainy days at all depths.

Dinoflagellates use stored energy from photosynthesis to produce bioluminescence at night. With a greater exposure to sunlight, the dinoflagellates should have more energy to produce bioluminescent displays (Lambert 2006).

Materials and methods

Study site

Due to the presence of bioluminescent dinoflagellates, all field research for this study was conducted at the Yellow Submarine Dive Site (12°09'36.47"N, 68°16'55.16"W) on the west coast of Bonaire, Dutch Caribbean (Fig. 1). Plankton tows were taken along the start of the reef crest at the Yellow Sub Site.

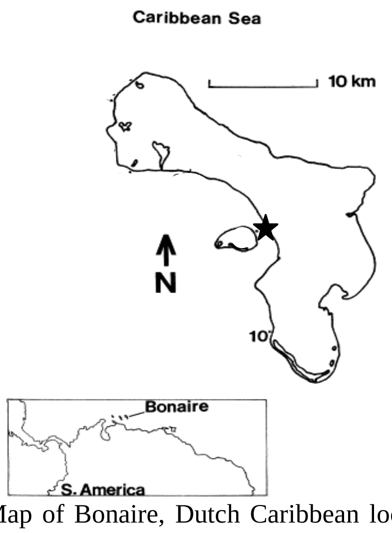

Fig. 1 Map of Bonaire, Dutch Caribbean located in the Caribbean Sea (Hall 1999). The sampling site, Yellow Submarine Dive Site (12°09'36.47"N, 68°16'55.16"W), is indicated by the ★ (star) on the map along the western coast of Bonaire

Plankton collection

Plankton samples were taken twice per week in the evening from 20:00-22:00 when bioluminescent activity is the greatest (Pilla 2012). To compare abundance and bioluminescence at various depths, four separate horizontal plankton tows were conducted at the sea surface, 10, 20, and 30 feet.

A diver pulled a plankton net (30 µm mesh net has an opening of 13 cm in diameter, equipped with a Wildco® flow meter at consistent depth until 300 m^3 of seawater had been filtered (Fig. 2). After the tow, each sample was placed in a separate 300 mL lightproof container, to prevent light pollution that could have from influencing or altering the biochemical processes of bioluminescence in dino-flagellates. Samples remained sealed in these containers from the time of collection to the time of bioluminescent analysis.

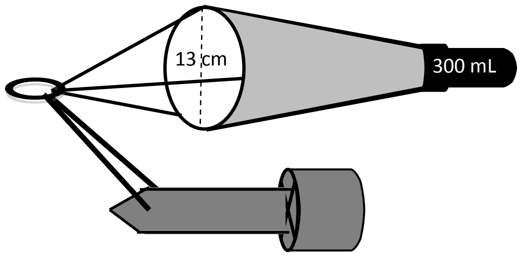

Fig. 2 Sampling device used to collect plankton contains a 30 µm plankton net with a 13 cm diameter opening connected to a 300 mL collection container. The volumetric flow meter is attached and hangs directly below the plankton net. The diver towed the sampling device throughout the water column until 300m^3 of seawater was filtered through the net

Dinoflagellate bioluminescence

Immediately after plankton collection, bioluminescence was analyzed in a dark room. To each sample, contained in a shallow clear container, a total of 6 mL of acetic acid (vinegar) was added in 10 second increments to stimulate the luminescent activity. Individual visible bioluminescent displays were counted continuously for the time period of 60 seconds.

Length of luminescence was measured on a descriptive scale of short (less than one second), medium (one to two seconds), and long (more than two seconds). Plankton samples were placed back into the same container after bioluminescence was tested.

Copepod and dinoflagellate abundance

After testing bioluminescence, the samples were rinsed with ethanol and passed through a 30 µm mesh strainer to filter out the plankton. Plankton were filtered onto a petri dish and 0.1 mL of 70% rose bengal was added to dye organisms present in the sample. Each petri dish was photographed under a 4x magnified dissecting scope to analyze dinoflagellate and copepod abundance on ImageJ©. Dino-flagellates and copepods were counted and identified to the lowest taxonomic level using this software and a 40x compound microscope.

Data analysis

Two-way ANOVA tests were run on the following data: 1) abundance of dinoflagellates and depth, 2) bioluminescent displays and ratio of copepods to dinoflagellates, 3) biolum-inescent displays and depth.

A Mann-Whitney U test was used to analyze frequency of bioluminescent displays and weather conditions (i.e. sunny v. cloudy and rainy). The statistical tests were used to evaluate the strength of those three relationships and used to determine any factors that may have an influence on the effectiveness of the dinoflagellate burglar alarm hypothesis.

Results

Plankton identification

Several geneses of bioluminescent dinoflagellates were identified in the samples including *Alexandrium*, *Ceratium*, *Ceratocorys*, *Gonylaux*, *Noctiluca*, *Ornithocerus*, *Peridinium*, and *Procentrum*. Three orders of copepods were also identified in the samples: Calanoid, Cyclopoid, and Hapacticoid. Using a 40x compound microscope, three different species of the previously mentioned dinoflagellates and one species of copepod were photographed (Fig. 3).

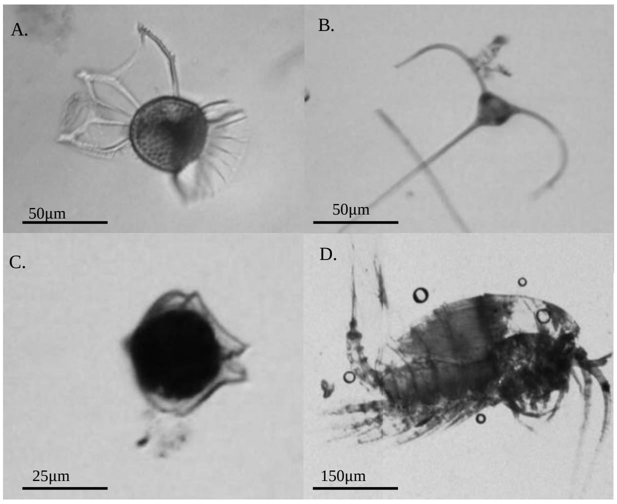

Fig. 3 Microscopic photographs of several of the planktonic organisms that were identified in the samples taken from the waters along the western coast of Bonaire, Dutch Caribbean. Reference bars show length in micrometers (µm). The following plankton were identified to the lowest taxonomic level possible using a 40x compound microscope: *Ornithocerus* spp. (A), *Ceratium* spp. (B), *Gonyaulax* (C), and Calanoida (D). Other species of dinoflagellates and copepods were analyzed in the samples, but are not shown in the figure above

Dinoflagellate and copepod abundance

There is a significant correlation between the mean abundance of dinoflagellates in regards to depth (df=31, F=4.92, p=0.007). As depth increases, so does the mean of dinoflagellates. The ratio of copepod to dinoflagellate abundance was used instead of the abundance of copepods. The ratio of copepod to dinoflagellate abundance decreased as the frequency of dinoflagellate bioluminescent displays increased (Fig. 4). The variance in the ratio of copepods to dinoflagellates had a significant difference as the number of bioluminescent displays changed (df=31, F=4.15, p=0.015).

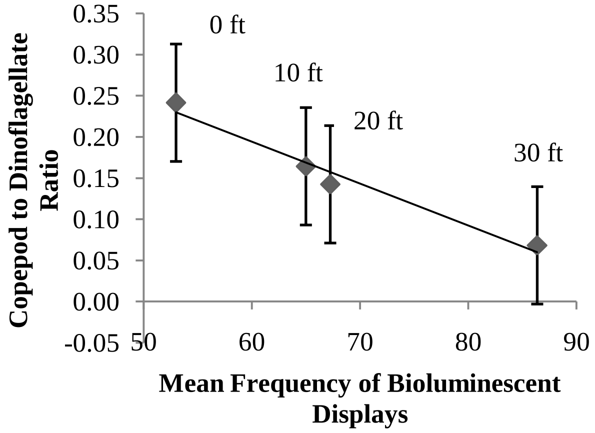

Fig. 4 Ratio of abundance of copepods to the abundance dinoflagellates as the frequency of dinoflagellate bioluminescent displays change. Data was taken from 32 different samples at four depths between 20:00-22:00 in the waters off the western coast of Bonaire, NA. The numerical label on the plot indicates depths in feet of each data point. The ratio of copepod to dinoflagellate abundance decreases as the frequency of dinoflagellate bioluminescent displays increase (df=31, F=4.15, p=0.015)

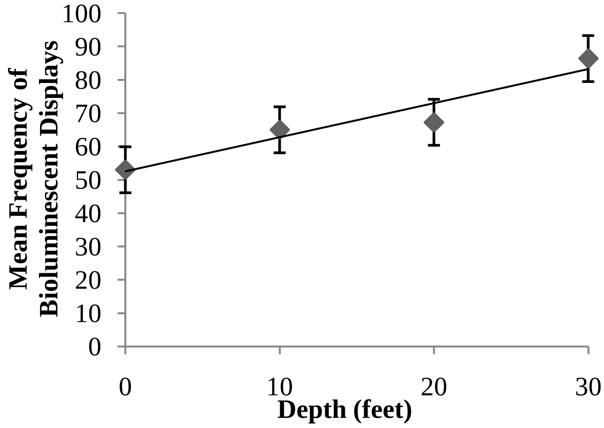

Fig. 5 Mean frequency of dinoflagellate bioluminescent displays at depths of 0, 10, 20, and 30 feet in the waters of the western coast of Bonaire, NA. Error bars indicate ± standard deviation. Data was taken from 32 different samples at four depths between 20:00-22:00. As the depth increases, so does the average number of bioluminescent displays emitted by dinoflagellates (df=31, F=3.23, p=0.037)

Bioluminescent displays

The average number of luminescent displays was calculated for each sample and compared against each other using a two-way ANOVA test. The number of dinoflagellate bioluminescent displays increased as depth increased (Fig. 5). There was a significant difference between the frequencies of luminescent displays at each sampling depth (df=31, F=3.23, p=0.037).

During bioluminescence testing, luminescent activity in samples from 30 feet depth was repeatedly brighter and longer than the samples at the three shallower depths (Table 1). The majority of individual displays at 30 feet were classified as long, while the majority of displays in the samples from shallower depths were categorized as medium or short. It was observed that the deepest sample in each test group was noted for having the highest intensity of luminescence.

Table 1 Average length of the mean bioluminescent displays in each sample depth (in feet) tested. Bioluminescent categories are based on descriptive categories of short (less than one second), medium (one to two seconds), and long (more than two seconds)

Sample Depth (ft)	Display Length
0	Short
10	Medium
20	Medium
30	Long

Weather conditions

There was no significant difference between the number of luminescent displays on sunny days and on cloudy/rainy days (n_1=24, n_2=8, U=103, p>0.05). The difference between the mean frequencies of luminescent displays on sunny and cloudy and rainy days is less than two bioluminescent displays (Fig. 6).

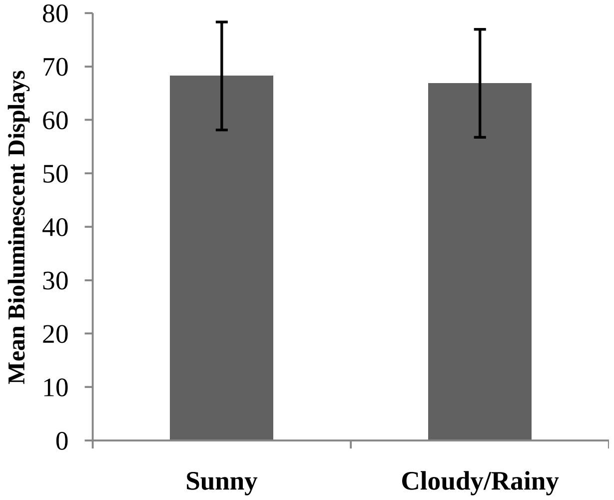

Fig. 6 A graph of the average number of dinoflagellate bioluminescent displays on sunny days versus cloudy/rainy days in the waters off the western coast of Bonaire, NA. Error bars indicate ± standard deviation. Data was taken from 32 different samples (N_1=24, N_2=8) at four depths between 20:00-22:00 (p>0.05)

Discussion

The ratio of copepod to dinoflagellate abundance decreased as the frequency of dinoflagellate bioluminescent displays increased. This trend was proven to be statistically significant, rejecting the null and supporting the first hypothesis. The bioluminescent "burglar alarm" first proposed by Burkenroad (1943) was shown to be a successful defense mechanism used to decrease copepod predation during the night and maintain dinoflagellate populations. It is still unclear whether or not dinoflagellate bioluminescence is used to startle the copepod or used to attract the copepod's predator or a combination of the two. Isolated laboratory studies indicate that this defense mechanism has a multi-trophic effect; however there is no evidence that this is the natural interaction that takes place. More studies focusing on abundances of planktivores in areas with bioluminescent dinoflagellates compared to areas with non-bioluminescent dinoflagellates would help determine the effect that the burglar alarm has on various trophic levels.

Luminescent activity increased as the depth of each sample group increased. The correlation between bioluminescent displays and depth showed a statistically significant relationship, rejecting the null and supporting the second hypothesis. Variations in luminescence at depths may be caused by the amount of ambient surface light present during the night. As depth increases more light wavelengths are absorbed, which indicates that there was unequal light exposure between sample groups.

If enough ambient is light present, it may alter the natural circadian rhythm of the dinoflagellate, thus changing their normal luminescence. Bioluminescent activity in dinoflagellates functions on a circadian rhythm: in the evening the regulatory protein is activated and in the morning the protein is inhibited (Mittag et al. 1994). Dinoflagellates exposed to 72 hours of normal day and night cycles had more luminescence than dinoflagellates in 72 hours of constant light (Mittag et al. 1994). Similarly, dinoflagellates samples collected at night which were exposed to 30 minutes of light showed lower bioluminescent activity than dinoflagellate samples that were left in darkness (Behrmann and Hardeland 1999). This indicates that light pollution at night has a detrimental effect on the defense mechanism of dinoflagellates.

During bioluminescence testing, luminescent activity in samples from 30 feet depth was repeatedly brighter and longer than samples at the three shallower depths. The majority of individual displays were longer, lasting more than two seconds, while the majority of displays in the other samples were less than one second. Brightness was not quantified, although it was observed that the deepest sample in each test group was noted for having the highest intensity of luminescence. This may be attributed to the occurrence of ambient light pollution at night being stronger on the surface, than at depth.

Decreases in bioluminescence may also be caused by a higher frequency of predation at the surface than at depth. Copepods have a single eye at the center of their head, which they use to visually locate prey (Land 1988). The presence of ambient light can pose a problem by illuminating the location of dinoflagellates to its predators, making the brightened phytoplankton more vulnerable to predation. According to the burglar alarm theory, there should be more bioluminescence at the surface to combat the increased predation, however the highest frequency of bioluminescent displays was observed in samples taken at 30 feet, furthest from the surface (Burkenroad 1943). Implications with light pollution may cause the defense mechanism to be ineffective in reducing copepod predation.

Frequency of bioluminescent displays does not differ between sunny and cloudy and rainy days. There is not enough data to reject the null hypothesis, so it cannot be concluded that there is an association between bioluminescence patterns and weather conditions. Luminescent displays are effected with the alteration of normal day and night cycles, however bioluminescent activity in cloudy and rainy weather conditions may not show a large enough difference from sunny conditions for the methods used in this study to detect. Further studies using more advanced methods of calculating luminescence, such as a luminometer, may show a difference between weather conditions (Lambert 2006). Comparisons between weather conditions may be better evaluated during seasonal weather changes, where more prominent rain exposure occurs.

Recent studies have shown that chlorophyll catabolism of luciferin creates an alternative pathway which produce a biochemical cold-light (Fresneau 1986). Chlorophyll catabolism is dependent on the photosynthetic energy available, which can alter the given amount of sunlight the dinoflagellate is exposed to. This is not the main pathway used for dinoflagellate bioluminescence; minute differences in luminescence may correlate with patterns in weather conditions.

Further studies on the implications of light pollution may be helpful in understanding the limitations of the burglar alarm hypothesis in dinoflagellates. Alterations in the ability of the dinoflagellate defense mechanism may cause changes in their ecological role, mainly focusing on the structures of phytoplankton blooms (Busky 1983). Comparisons of bioluminescence between the areas exposed to light pollution and areas sheltered from ambient light can be used to analyze how the primary trophic level will change with increasing human impacts.

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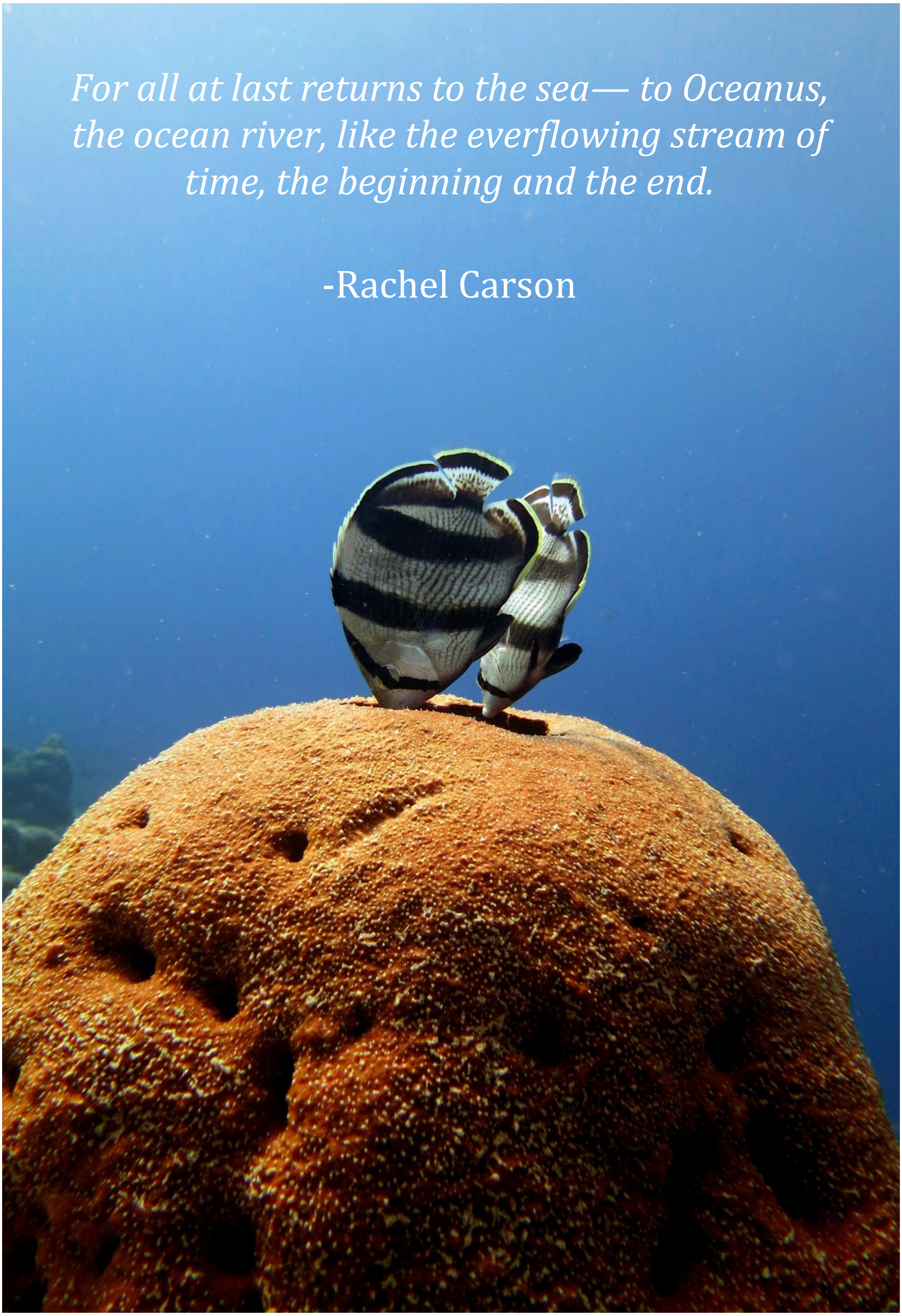
For all at last returns to the sea— to Oceanus, the ocean river, like the everflowing stream of time, the beginning and the end.
-Rachel Carson