Using prey fish species as bioindicators of anthropogenic stress and predictors of predator density and diversity on coral reefs in Bonaire, N.A.

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Abstract

Bioindicator species have been used to determine changes in water quality and the effect of pollution at sites of environmental concern. Increasingly degraded water quality throughout the Caribbean is leading marine park managers and scientists to use bioindicator organisms to rapidly detect differences in water chemistry by determining connections between environmental parameters and changes in reef fish communities. This study sought to determine bioindicator prey species that could provide early detection of changes as a result of anthropogenic activities in the coastal waters of Bonaire, N.A. The effects of these parameters on the density and diversity of reef fish species was compared between 4 sites of "more (MI)" and 4 sites of "less (LI)" anthropogenic impact (<200 m and >200 m from of coastal development, respectively). Fish communities were surveyed using a modified version of the AGRRA methodology during the morning and evening. Two 30x2 m transects at 12 m depth were used at each site to survey both prey and predator fish species. Water chemistry including nutrient, bacterial and sedimentation levels were also analyzed to attempt to determine the factor(s) driving the changes. This study revealed significantly greater densities and a higher diversity of prey and predatory fish species at MI sites versus LI sites during the morning and the evening. The species that was found at greatest densities for both LI and MI sites was *Stegastes partitus*, with significantly more *S. partitus* at MI sites during both the morning and evening. Thus, *S. partitus* may be a possible bioindicator of stressors on the reefs in Bonaire. The use of *S. partitus* as a bioindicator of anthropogenic stress may help increase the effectiveness of marine management protocols in Bonaire and provide a basis for determining bioindicator species for monitoring coastal water quality throughout the Caribbean. None of the water chemistry parameters studied differed between MI and LI sites, therefore, the driver(s) of the differences in prey species (e.g. *S. partitus*) may be unaccounted for in this study as a result of time lags in the coral reef ecosystem.

Introduction

In the last decade, concern over the impacts of human population growth and development on coral reef systems has grown worldwide. Coral reefs are quickly disappearing due in large part to destructive fishing, pollution, global climate change and coastal development (Norse and Crowder 2005). These anthropogenic pressures have led to a phase shift from coral to algal dominated reefs in the Caribbean (Burke and Maidens 2004). Coastal development has been cited as one of the main threats facing the Caribbean due to the increased nutrient and sediment runoff associated with it (Burke and Maidens 2006). The unprecedented rate of coastal development is increasing sedimentation in near-shore waters, smothering reef-building corals and reducing light necessary for photosynthesis (Rogers 1990). Losses of reef-building corals can have ecosystem-wide cascading effects by altering the reef's structural framework, thus reducing the abundance or changing the biodiversity of fish species (Rogers 1990). Anthropogenically induced decreases in biodiversity are plaguing coral reefs globally (Markham 1996). Nutrients, heavy metals, pesticides and hydrocarbons from runoff and development have also been shown to degrade coral reefs at local scales (Fabricius 2005). As coral reefs thrive in oligotrophic waters, excess nutrients may increase macroalgal growth resulting in a phase shift from coral dominated reefs to algal dominated reefs (Burke and Maidens 2004).

It has been suggested that the direct (e.g. chemical analyses) monitoring of pollutants in marine systems may not provide enough information about their ultimate effects on the biology of coral reef ecosystems (Naranjo et al. 1996). Therefore, indentifying biological entities, or bioindicator species, that readily respond to environmental stressors may be useful in the early detection and monitoring of pollutants and their effects on reef communities. Marine bioindicator species have previously been used to detect environmental stressors. Burger et al. (2007) identified and utilized bioindicator fish and bird species at multiple trophic levels to detect radionuclides in the waters of the Aleutian Islands, AK. They found that 80% of the fish species and 20% of the bird species studied had elevated levels of Cesium^{137} in their tissues. These data were used as a baseline for the continued monitoring of the pollutant's spread and effects on other local marine organisms. In New Caledonia, Southwest Pacific, the tropical brown alga *Lobophora variegata* showed promise as a bioindicator species for excess silver (Ag) in coastal waters. *L. variegata* rapidly concentrates Ag within its tissues. Metian and Warnau (2008) found that after only 28 d the alga retained 7000x more Ag than ambient water. Since excess Ag is often associated with sewage sludge, *L. variegata* may be useful as a bioindicator of improper wastewater treatment. In Bonaire, Netherlands Antilles Williams (2009) looked at Christmas tree worms (*Spirobranchus giganteus*) as potential bioindicators of environmental stress. These sessile, filter feeding worms were documented at significantly greater densities in living coral near commercial and residential sites (<200 m away) with elevated phosphates and finer sediment particles possibly attributed to eutrophication and/or run-off caused by increased coastal development (Williams 2009).

Sessile organisms have commonly been used to assess the utility of bioindicators to identify marine pollutants and stressors (e.g., Cooper et al. 2008; Metian and Warnau 2008; Williams 2009). The stationary nature and modes of nutrient uptake in these organisms (e.g., filter feeding) prove useful in determining localized changes in water quality. However, few marine studies have assessed whether mobile organisms are useful bioindicators of stress (except Burger et al. 2007), and, moreso, useful bioindicators without the need for direct handling or tissue sampling. Though coral reef fishes are not sessile, many species maintain fairly permanent territories on the reefs. For instance, damselfishes (*Pomacentridae*) have been documented protecting microhabitats of algal "gardens" within defined areas of the reef, and several species of parrotfish (*Scaridae*), hamlets (*Serranidae*), surgeonfish (*Acanthuridae*) and butterflyfishes (*Chaetodontidae*) maintain semi-permanent home territories (DeLoach and Humann 1999). Although mobile, such territorially-limited fish species may respond noticeably and rapidly to local or regional anthropogenically induced environmental changes and raises the question, could one or more of these species play the role of a bioindicator prey species? If prey fish are affected by these parameters could predators be affected too? Stevenson et al. (2007) found that several reefs in the Pacific had a greater biomass of predatory fishes and exhibited increased biomass in lower trophic level herbivorous fishes that are often the prey of choice for these predators. These data corroborated with a study performed in the Northern Hawaiian Islands which found that where apex predator biomass was >50% of the total fish biomass there was elevated biomass of herbivorous fishes at lower trophic levels (Friedlander and DeMartini 2002). These studies suggest that areas with higher densities of prey fishes will also have higher densities of predatory fishes. Therefore, if natural and/or anthropogenic stressors affect the composition of prey fish communities on reefs, it is possible that predatory fishes will be indirectly affected by these stressors as well.

The southern Caribbean island of Bonaire, N.A. has been experiencing deterioration of its near-shore fringing coral reefs since the 1970s (Bak et al. 2003). Growing resident and tourist populations and extensive coastal development have been suggested as potential causes of observed increases in sediment, nutrient and bacterial loads in the leeward coastal waters of the island (Rini 2008; Sinnott 2009; Williams 2009). The lack of a proper wasterwater treatment system has led to the widespread use of unlined septic tanks and cesspits on the island (van Sambeek et al. 2000). This, coupled with the island's base of porous limestone (BNMP 2006) allows nutrient and bacteria-"heavy" water to enter directly into the ocean. Building and road construction have contributed to nutrient loading and sedimentation in coastal waters (BNMP 2006). As Bonaire economically relies on its coral reefs (e.g., dive tourism, artisanal fishing) losses or changes in coral communities due to anthropogenically-induced stressors could be devastating. The determination of bioindicator reef fish species to effectively identify reef areas high in nutrients and sediments could potentially increase the effectiveness of the conservation efforts of the Bonaire National Marine Park. As changes in fish communities, particularly densities and diversity of predatory fishes can be easily identified by scientists, diving tourists and fisherman alike, the determination of bioindicator species could serve as an early biological detection system for changes in water quality.

In this study the following questions were addressed: 1) will sites more impacted by coastal human activity have higher densities and diversity of bioindicator prey species than those less impacted by coastal human activity, 2) will sites with higher densities and diversity of bioindicator prey species have higher densities and diversity of Bonaire's common predatory fishes, 3) do the water conditions (e.g., excess nutrients, enterococcal bacterial loads, finer sediment particles) positively relate to bioindicator prey fish densities and thus affect the densities and diversity of predatory fishes?

Materials & Methods

Study Sites

In order to assess the effects of stressors such as excess nutrients, sedimentation, and *Enterococcus* bacteria on the densities and diversity of potential bioindicator prey species sites defined as "More Impacted (MI)" and "Less Impacted (LI)" by the long-term water quality monitoring project (LTR) of the Council for International Educational Exchange Research Station Bonaire (CIEE-RSB) were chosen. A MI site is defined as one located <200 m from a commercial establishment such as a resort, restaurant, or store and sites defined as LI are surrounded on all sides by >200 m of rural or residential land (Rini 2008). For this study four MI sites including Donkey Beach, Eighteenth Palm, Cliff and Bari Reef and four LI sites including White Slave, Jerry's Reef on Klein Bonaire, Andrea II and Witches Hut were chosen (Fig. 1). At all LTR sites, excess nutrients (phosphates, nitrates, nitrites, ammonia) and *Enterococcus* bacterial loads are determined through water sampling as well as sedimentation rates and particle size distribution are determined using a permanent sediment trap at 12 m on a biweekly basis.

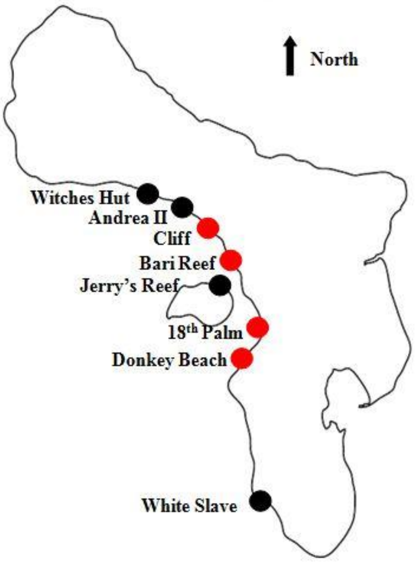


Fig. 1 Map of Bonaire, N.A. Red dots indicate "more impacted" study sites and black dots indicate "less impacted" study sites (<200 m and >200 m from sites of coastal development, respectively)

Potential Bioindicator Fish

In order to identify appropriate bioindicator fish species the process developed by Burger et al. (2006) was used as a guideline. Burger et al. (2006) recommend a literature review on the preferred prey of large predators commonly found at the study sites of interest, as well as *in situ* observations of prey fishes found at each location. Biological, methodological, and societal relevance of the species were considered in accordance with the guidelines provided. A pool of eligible bioindicator prey species was selected from the literature and relevance review and compared with the species documented *in situ*. Those fish considered in the literature to be prey of Bonaire's common predatory fish and found in elevated densities (>20 fish 100 m^{-3}) were used as bioindicator species for this study.

Monitoring

Once potential bioindicator prey fish species were identified through the literature review, their densities and diversity were recorded at MI and LI locations. In order to assess densities and diversity of predator and prey fish species at the study sites, SCUBA was used to complete 30x2x12 m (720 m^3) transects to both the north and south of CIEE-RSB's permanent sediment trap at 12 m depth.

The distance of 30 m was chosen to ensure that observations of fish would be in an area close enough to the permanent sediment traps of CIEE-RSB and any potential environmental stressors documented through CIEE-RSB's LTR would be viable as parameters for potential differences in fish communities. Following transect deployment, a modified version of the AGRRA fish survey methodology was conducted using a 1 m T-bar (Kramer et al. 2005) to survey the predetermined potential bioindicator prey and predator fish species within the afore mentioned volume of space. Predators in Bonaire were predetermined to include: groupers, snappers, moray eels, barracuda, tarpon, bar jack, Caribbean reef squid, boga and black margate. Transects were surveyed in the morning (0730 h) and the evening (1930 h) for all sites in order to determine if densities and diversity of bioindicator prey and predatory fish species varied temporally. Each site was surveyed in the morning and evening on the same day to further avoid temporal variances. During each pass along the transect one diver was assigned to record "very abundant" fishes in order to allow other fish species to be surveyed by the second diver more easily. During morning dives this species was the bicolor damselfish, *Stegastes partitus*, and for evening dives this species was the blackbar soldierfish, *Myripristis jacobus*.

Additionally, during morning dives a 10 min sit-and-observe session was performed at each location prior to transect deployment. This provided an acclimation period for the fish as well as an opportunity to survey each site for any "shy" prey or predatory fishes that tend to avoid the transect line. These observation sessions were performed by two divers positioned back-to-back directly over the CIEE-RSB permanent sediment traps at a 12 m depth so as to encompass a 360° view of the site with their combined range of vision. All fish species observed were identified and recorded.

Water Quality

In order to determine the presence or absence of potential stressors at MI and LI sites the data on excess nutrients, sedimentation and bacterial loads were acquired from CIEE-RSB's LTR data set during the time period of January-March 2010. The collection and analyses of nutrient, bacterial (*Enterococcus*), and sediment samples for the CIEE-RSB LTR data were done using the methodology described by Sinnott (2009) and Williams (2009).

Statistical Analyses

In order to assess any differences between the densities and diversity of prey and predator species at LI and MI sites for morning and evening data tow two-way analysis of variance (ANOVA) were run. A two-way ANOVA was also used to compare the densities and diversity of bioindicator prey species between LI and MI sites. Additionally, a two-way ANOVA was used to assess differences in the densities of bicolor damselfish, a potentially useful bioindicator species (see results and discussion) between LI and MI sites. Two-tailed T-tests were used to assess differences in nutrient levels and bacterial loads between LI and MI sites. A two-way ANOVA was used to determine differences in sediment particle size distributions and sedimentation rate between site types.

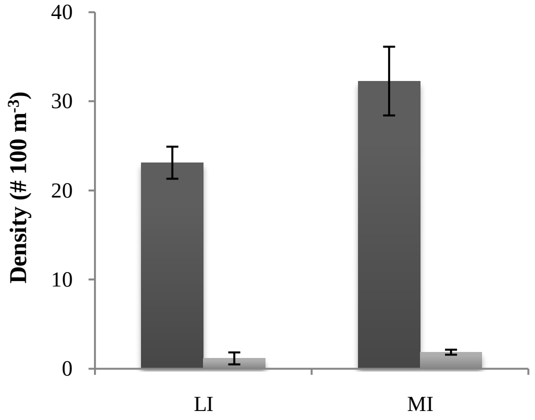
Results

Monitoring

Fish densities were significantly higher at MI sites (two-way ANOVA, site x fish type p=0.002, Fig. 2a) during the morning. The mean density (± SD) of prey fishes in the morning at MI sites was 32.256 ± 3.854 as compared to 23.099 ± 1.799 at LI sites. The mean density (± SD) of predatory fishes in the morning at MI sites was 1.840 ± 0.281 as compared to 1.146 ± 0.672 at LI sites. In the evening MI sites had higher prey and predatory fish densities (2.170 ± 0.611and 0.590 ± 0.216, respectively) than LI sites (1.632 ± 0.657 and 0.590 ± 0.216, respectively, Fig. 2b). The interaction between site and fish type was not significant (two-way ANOVA, p=0.621). A higher number of predatory fishes were present at MI sites (27.000 ± 13.342, mean ± SD), than LI sites (14.500 ± 3.697, Fig. 3) during morning sit-and-observe sessions (T-test p=0.121).

Overall, predator species were significantly more diverse than prey species during the morning, while prey were significantly more diverse than predators in the evening (two-way ANOVA, p=0.003 and p=0.004, respectively, Fig. 4a & b).

(a) Morning



(b) Evening

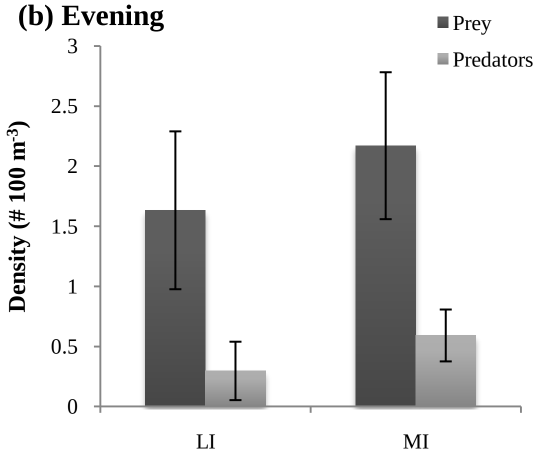


Fig. 2 **(a)** Mean density (# 100 m^{-3} ± SD) of prey and predatory fishes at less and more impacted sites (>200 m and <200 m from sites of coastal development, respectively) during the morning (two-way ANOVA, site p<0.001, fish type p<0.001, and site x fish type p=0.002) **(b)** Mean density (# 100 m^{-3} ± SD) of prey and predatory fishes at both less and more impacted sites during the evening (two-way ANOVA, site p=0.107, fish type p<0.001, site x fish type p=0.621)

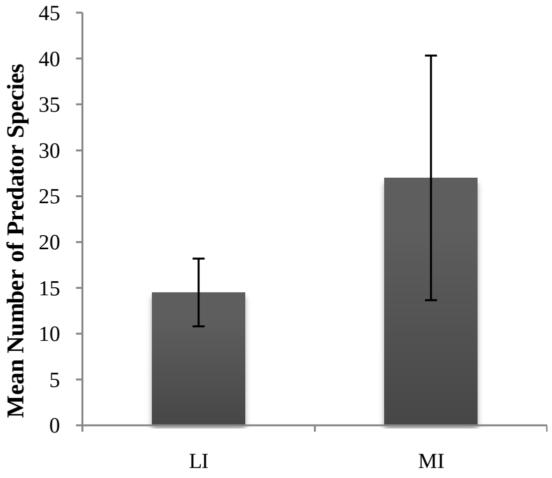
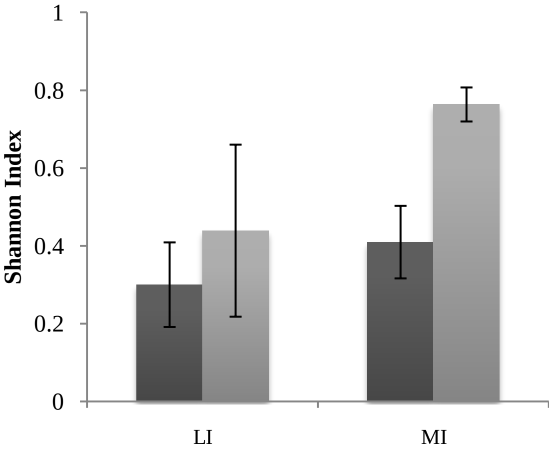


Fig. 3 Mean total number of predatory fish (± SD) recorded during the sit-and-observe session at less and more impacted sites (>200 m and <200 m from sites of coastal development, respectively) during the morning (T-test p=0.121)

In the morning there was a significantly higher overall diversity at MI sites when compared to LI sites (two-way ANOVA, site p=0.007). This, however, was not found to differ significantly at night (two-way ANOVA, site p=0.174).

(a) Morning



(b) Evening

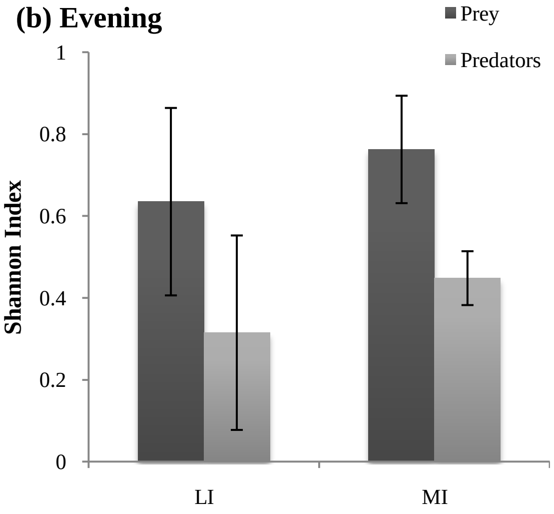


Fig. 4 **(a)** Mean diversity (Shannon Index ± SD) of prey and predatory fishes at less and more impacted sites (>200 m and <200 m from sites of coastal development, respectively) in the morning (two-way ANOVA, site p=0.007, fish type p=0.003, site x fish type p=0.133). **(b)** Mean diversity (Shannon Index ± SD) of prey and predatory fishes at less and more impacted sites in the evening (two-way ANOVA, site p=0.174, fish type p=0.004, site x fish type p=0.976)

Potential Bioindicator Fish

Upon a more detailed analysis of individual prey species, there appeared to be higher densities of *S. partitus* than any other species encountered. *S. partitus* was observed at densities of >20 fishes 100 m^{-3} for three of the four MI sites, thus meeting the previously described requirements for a bioindicator species. Mean densities (± SD) of *S. partitus* during the morning at LI sites (17.682 ± 2.749) were significantly lower than at MI sites (22.856 ± 2.643, two-way ANOVA, site x time p=0.019, Fig. 5). However, at night *S. partitus* was nearly undetectable at both LI and MI sites (0.017 ± 0.035 and 0.026 ± 0.017, respectively).

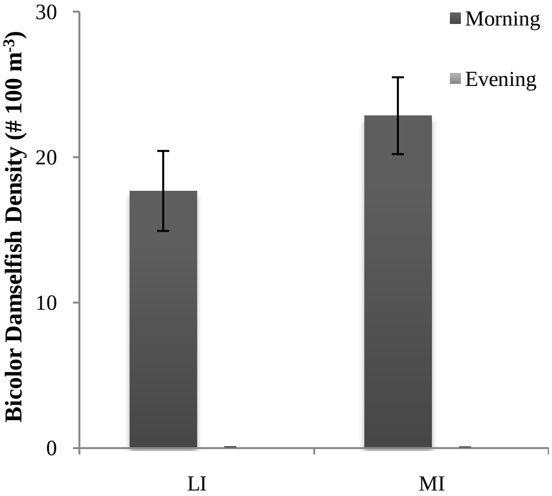


Fig. 5 Mean density of *Stegastes partitus* (± SD) at less and more impacted sites (>200 m and <200 m from of coastal development, respectively) during the morning and evening (two-way ANOVA, site p=0.019, time p<0.001, site x time p=0.019)

Water Quality

Overall, in nutrient levels, bacterial loads, sedimentation rates and sediment particle size distributions were found between LI and MI sites for several weeks prior to and during this study. In fact, nitrate and nitrite were not detected at any sites, while ammonia and phosphate concentration levels (mean ppm ± SD) were slightly elevated at MI sites (ammonia=0.075 ± 0.150; phosphate=0.092 ± 0.010) as compared to LI sites (ammonia=0.050 ± 0.058; phosphate=0.088 ± 0.025), but were not significantly different (T-test p=0.766 and p=0.766, respectively; data not shown). *Enterococcus* bacterial loads (MPN ± SD) appeared elevated at MI sites (0.583 ± 1.058) compared to LI sites (0.338 ± 0.307), but were not significantly different (T-test p=0.671; data not shown)

There were no significant differences in sedimentation rates (mean g day^{-1} ± SD) between MI (0.060 ± 0.018) and LI (0.078 ± 0.032) sites (T-test p=0.375; data not shown) and there was no significant difference among the sediment particle sizes between site type (MI: <10 µm≈65.7%, 10-50 µm≈26.8%, 51-100 µm≈5.2%, 101-250 µm≈2.1%, 251-500 µm≈0.2%, >500 µm≈0.0%; LI: 10 µm≈63.2%, 10-50 µm≈28.5%, 51-100 µm≈5.9%, 101-250 µm≈2.2%, 251-500 µm≈0.2%, >500 µm≈0.0%, two-way ANOVA, site x particle size p=0.974; data not shown).

Discussion

The goal of this study was to determine bioindicator species that could be useful to scientists and marine park managers to rapidly assess changes in water quality known to be detrimental to coral reefs. The analysis of densities and diversity of prey and predator species was completed between LI and MI sites to determine a potential bioindicator species. The higher densities and diversity of prey fishes (in particular *S. partitus*) at MI sites during the morning suggested that indentifying potential bioindicator prey fishes in Bonaire, N.A. was feasible. Additionally, higher abundances of predatory fishes observed during morning sit-and-observe sessions at MI sites lended support to the idea that higher predatory fish densities may positively relate to higher prey fish densities. Sih (1984) found that for *Notonecta*, an aquatic insect predator, concentrating hunting efforts in areas of higher prey densities increased predator feeding success because the prey was more abundant. Thus, higher densities of predatory fish on a reef may indirectly predict the stressors affecting the site.

The history of environmental stress and/or other parameters not considered in this study may be driving the difference in prey densities, between LI and MI sites. There are several possible explanations why no differences in nutrient concentrations, bacterial loads and sedimentation were seen. The months of November-January is the rainy season in Bonaire. However, the months prior to this study were unseasonably dry (A. Hollebone personal communication). This has most likely limited the amount of terrestrial runoff containing nutrients, bacteria and sediments entering the coastal waters. However, as time lags and feedback loops are common in natural ecosystems, it is probable that the current water chemistry was not driving the observations made in this study, but possibly it is the water chemistry from months to years past. Time lags in the addition of nitrogen fertilizers exemplify this concept. At the Cedar

Creek Natural History Area in Minnesota a five year study on nitrogen addition revealed an increase in perennial ragweed in year one followed by a decrease to control levels, an increase in bluegrass during year two, and finally, a delayed increase in blackberry during year five (Magnuson 1990). A time lag was additionally observed in meadows dominated by the sedge *Kobresia*, a species that prefers areas free of snow in the winter months. Webber et al. (1976) found that increasing the snow pack in a normally snow-free meadow at first appeared to vigorously increase the growth of the *Kobresia*. However, the plants allocated more energy to growth than reproduction and within 10 yr the large colony had completely disappeared. It is clear that immediate biological responses to environmental conditions are not always common to natural ecosystems.

In sharp contrast to this study, previous analyses of water quality in Bonaire have shown marked differences between LI and MI sites (Rini 2008; Sinnott 2009; Williams 2009). Willams (2009) found elevated levels of phosphates and finer grained sediments at MI sites. Sinnott (2009) found that at a depth of 12 m there were significantly higher concentrations of ammonia (NH_4^+) at MI sites and significantly smaller sediment particle size (characteristic of construction sites) at MI sites at a depth of 12 m. Rini (2008) found significantly elevated concentrations of *Enterococcus* bacteria at MI versus LI sites. Based on the potential for time lags in coral reef ecosystems, it is probable that these previously documented differences in water chemistry between LI and MI sites may be at least partially responsible for the differences in fish densities and diversity observed in this study.

Additional parameters not explored could be solely or in combination responsible for the density and diversity results of this study. Differences in potential substrate type between sites may explain the significantly higher overall densities and diversity at MI sites. Risk (2003) found a positive correlation between fish species diversity and a higher degree of topographic complexity in The U.S. Virgin Islands. Preliminary work in Bonaire comparing reef habitat complexity at six different sites (including three used in this study) suggested no correlation between habitat type and fish species richness (Yanson 2009), but this work did not assess differences in the evenness of the species or the diversity of fish communities. Additional study is needed to investigate these parameters.

Another parameter that could be examined in future studies is the presence of organics in the water column. Preliminary studies on the levels of organics in the water are being performed in Bonaire as part of the Light and Motion Sensor Program (LMSP). There are permanent sensors at several reefs in Bonaire located at 5, 12 and 20 m depths on the LMSP mooring line that monitor temperature and light intensities, as well as detect organic matter in the water column (Jones et al. 2008). Organics can increase surface productivity and fuel changes in benthic environments of marine systems (Suess 1980). These organics, as a direct or indirect food source, could be driving the differences seen in fish densities and diversity, particularly prey fish species, between LI and MI sites.

The determination of a bioindicator fish species for Bonaire specific anthropogenic stressors was an integral part of this study. The only species that met the defined guidelines of a bioindicator species and appeared at densities >20 fishes 100 m^{-3} was *S. partitus*. This species was documented at significantly higher densities at MI sites during the morning than LI sites. Sinnott (2009) and Williams (2009) found higher levels of nutrients (ammonia and phosphates), at MI versus LI sites. These compounds may be responsible for increases in macroalgal growth on the coral reefs of the Caribbean (Burke and Maidens 2004). Sinnott (2009) found significantly higher macroalgal cover at MI sites versus LI sites at 12 m in Bonaire, which corresponds with the higher densities of *S. partitus* at MI sites. Alga are “gardened” and fed upon by *S. partitus* (DeLoach and Humann 1999). Thus, it is possible that higher percent of macroalgal cover may be driving the greater densities of *S. partitus* observed at MI sites. This is another potential area of study for future projects on bioindicator species in Bonaire. This determination of *S. partitus* as a possible bioindicator fish may increase the effectiveness of the Bonaire National Marine Park’s efforts to protect reef communities from anthropogenic stressors. Reef sites with elevated densities (>20 fishes 100 m^{-3}) of *S. partitus* may indicate where regulations on coastal development are needed.

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