Progress Report

A. Grant Number: NA17NOS4820086

B. Amount of Grant: Federal $80,000___ Match _$31,800___ Total___$111,800_

C. Project Title: Testing the efficacy of MPAs and the impact of invasive lionfish on mesophotic reefs in the US Virgin Islands

D. Grantee: University of the Virgin Islands' Center for Marine and Environmental Studies

E. Award Period: From: Aug. 1, 2017 to Jan. 31, 2019 (NCE to Jan. 31, 2020)

F. Period Covered by this Report: From: August 1, 2017 to January 31, 2020

G. Summary of Progress to Date:

Executive Summary

In the United States Virgin Islands (USVI), mesophotic coral ecosystems (MCE) are important deep reef habitats supporting corals and reef fish at depths greater than 30m. These reef systems are more extensive than previously thought and represent > 75% of coral reef area in the USVI. Two large marine reserves, Red Hind Bank Marine Conservation District (MCD) and Virgin Islands Coral Reef National Monument (VICRNM) include extensive MCE habitats, that are largely unstudied relative to shallow coral reefs due to diving limitations. There is an urgency to understanding MCE because long-term monitoring has found that mesophotic reefs are supporting large populations of the invasive lionfish (*Pterois volitans*), making culling-based management on shallow reefs ineffective. This research had two primary goals. The first goal was to examine the efficacy of the large MPAs in improving fishery resources by conducting a comparative assessment of biological resources including fish populations (abundance, biomass and diversity) and benthic habitats (benthic cover and coral health) inside and outside the MCD and VICRNM. The second goal was to examine changes in these resources before and after the arrival of the invasive lionfish by comparing historical data from these sites to more recently collected data several years after the arrival of lionfish.

Between February 2018 and March 2019, 121 sites were surveyed inside and outside VICRNM and MCD and compared to historical data collected in 2002-2006 and 2007-2008, respectively. Percent cover of live coral, gorgonians, and sponges declined between the first and second sampling periods inside and outside VICRNM but were relatively stable inside and outside MCD. This pattern may have largely been an artifact of when surveys were conducted since VICRNM sites were all surveyed before the 2005-2006 mass bleaching and disease event that caused large-scale mortality of shallow and mesophotic coral reefs around the USVI. The MCD surveys were conducted after this mortality event in 2007-2008. The biomass of all fishes, and piscivore and herbivore trophic groups were relatively unchanged across time periods inside and outside both

MPAs.

Lionfish densities were not significantly different inside and outside the MPA's, but were significantly higher around the MCD (inside and outside) than around the VICRNM. This may have resulted from the MCD being deeper (mean = 39.6 m) with higher rugosity (102 cm relief) than VICRNM (mean depth = 25.4 m, mean rugosity = 38 cm relief). We found a significant positive relationship between lionfish biomass and depth within the MCD but no relationship with depth and lionfish density, indicating that larger lionfish tended to be on deeper reefs. No relationship was found between density or biomass of lionfish and small-bodied prey fish, potential lionfish predators or other functional groups nor an effect on percent coral cover among our study sites. The lack of any clear patterns from these analyses suggest that at the time of our recent surveys, lionfish density and biomass has had no clear direct or indirect impacts on native fish populations or coral health on mesophotic reefs.

1. Work Accomplishments:

a. Overview of methods and results and their relationship to the general goals of the grant.

The goal of this research was to conduct a comparative assessment of biological resources including fish populations (abundance, biomass and diversity) and benthic habitats (benthic cover and coral health) inside and outside two marine reserves in the US Virgin Islands before and after the arrival of the invasive lionfish (*Pterois volitans*). The two marine reserves were the Virgin Islands Coral Reef National Monument (VINCRM) and the Red Hind Bank Marine Conservation District (MCD). Both marine reserves are composed of highly complex shallow (<25m) and mesophotic reefs (>25m). Previous assessments of biological resources were conducted in 2002 – 2004 in VINCRM (Monaco et al. 2007) and 2007 in MCD (Smith et al. 2010) and set important baselines of fish populations and benthic composition and condition. An additional early study conducted outside the MCD in 2001 and 2002 provided additional base-line fish population data (Nemeth and Quandt 2005). These assessments occurred prior to the invasive lionfish reaching the USVI in 2008. The purpose of this project was to fill important gaps in our understanding of the function of marine protected areas on reef fish populations and to understand the direct and indirect effects of the invasive lionfish on native fish populations and coral health on mesophotic coral reef ecosystems. With this study, we tested the hypotheses that, 1) MPA's will harbor a higher abundance and diversity of commercially important species than outside its boundaries, 2) sites with a greater abundance of predatory fishes, which may act as natural control of lionfish, will have lower abundance of lionfish, and 3) potential lionfish prey and condition of the coral reefs will be inversely related to lionfish abundance.

Due to the complexity of data presented in this report, the report will be organized into sections examining similarities and differences among sites and time periods. These sections include 1) general overview of results and site characteristics; comparisons of 2) benthic habitats among sites; 3) lionfish abundance, distribution and biomass among sites; 4) native reef fish biomass and diversity among sites; 5) trends in benthic habitats and fish biomass and diversity over time periods; and 6) multivariate analysis and synthesis of these data.

1) General overview of results and site characteristics

Between February 2018 and March 2019, 121 sites were surveyed inside and outside two marine protected areas in the US Virgin Islands. The marine protected areas were the Marine Conservation District (MCD), located south of St. Thomas on the edge of the Puerto Rican plateau, and the Virgin Islands Coral Reef National Monument (VICRNM), located on a mid-shelf reef south of St. John (Figs 1.1, 1.2, 1.3). These data (Table 1.1) were compared to two other studies conducted at same sites within the MCD (Smith et al. 2010) and VICRNM (Monaco et al. 2007) before the invasive lionfish arrived in the northern Virgin Islands (Table 1.2). The survey sites inside and outside the MCD, were significantly deeper (mean 39.6m, range 33-48m) than sites inside and outside VICRNM (mean 25.4m, range 20-33m) (Table 1.1).

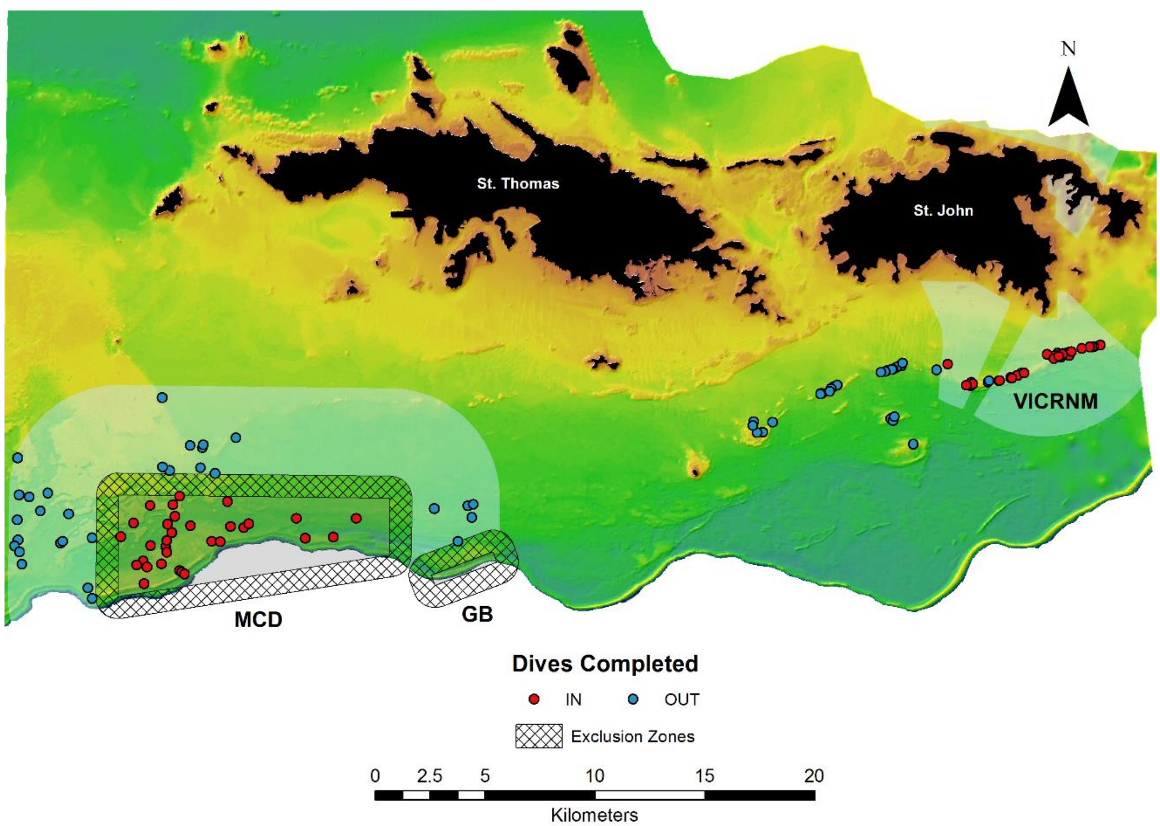


Figure 1.1. Location of 50 randomly selected points inside (red) and outside (blue) of the Red Hind Bank Marine Conservation District (MCD) and the Virgin Islands Coral Reef National Monument (VICRNM). The MCD includes a 100m buffer exclusion zone to eliminate potential edge effects. Due to potential bias from the Grammanik Bank (GB)

seasonal fishery closure a 100m buffer was also included.

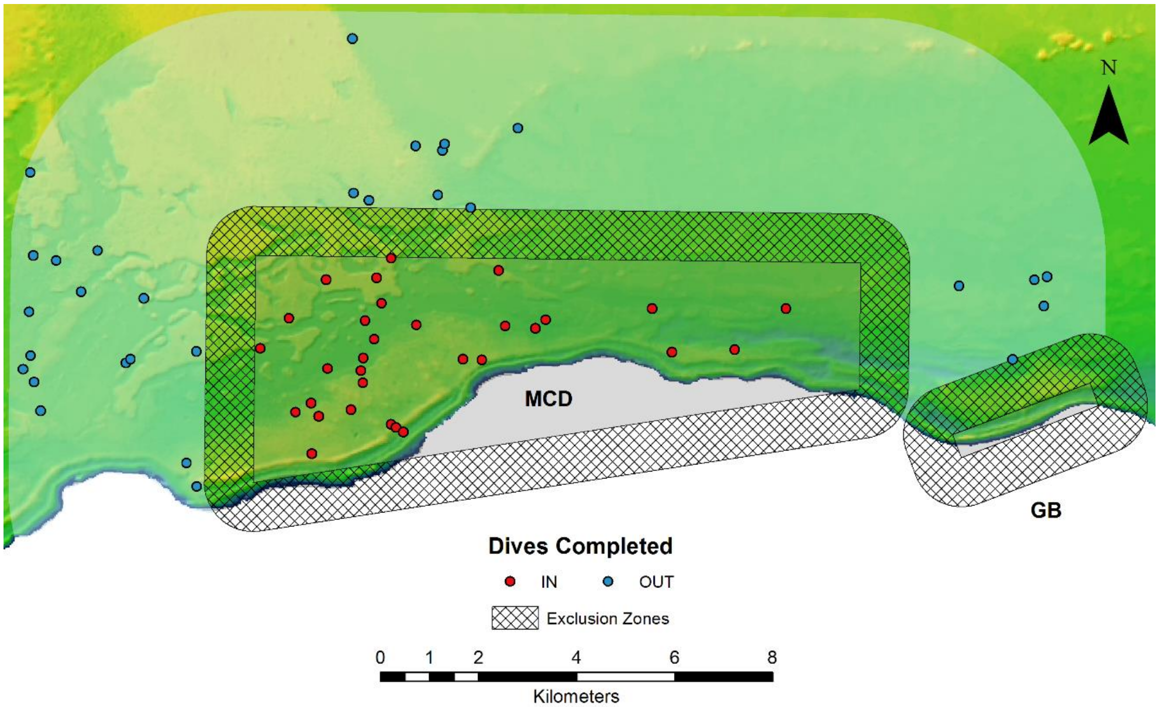


Figure 1.2. Close up of 50 randomly selected points inside (red) and outside (blue) of the Red Hind Bank Marine Conservation District. A 100m buffer exclusion zone to eliminate potential edge effects was included around MCD and Grammanik Bank seasonal fishery closure.

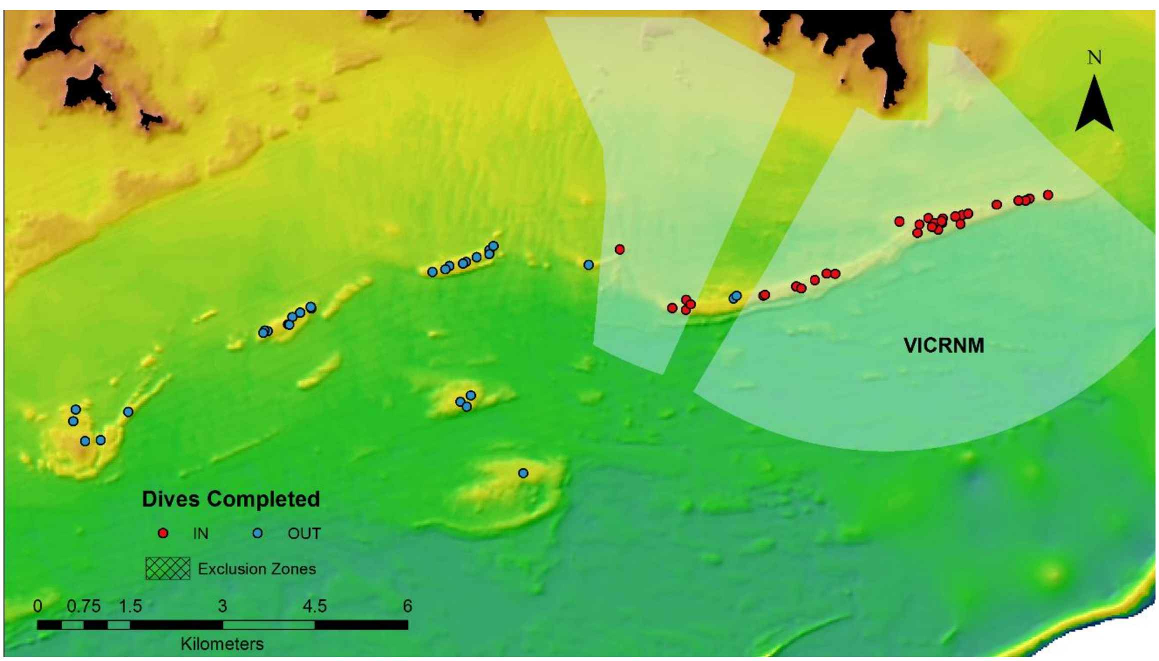


Figure 1.3. Close up of 50 randomly selected points inside (red) and outside (blue) of the

Virgin Islands Coral Reef National Monument.

Table 1.1. Summary data on physical and biological characteristics of study sites inside and outside the Marine Conservation District (MCD) and the Virgin Islands Coral Reef National Monument (VICRNM), which were sampled from June 2018 to May 2019.

2018-2019	MCD		VICRNM	
	in	out	in	out
No. of Sites Surveyed	31	29	31	30
Start & End dates	6/7/18 – 5/23/19	6/7/18 – 5/23/19	9/6/18 – 5/18/19	5/15/19 – 5/21/19
Depth Range	33-48 m	34-46 m	20-31 m	22-32 m
Total Lionfish	30	19	10	6
Lionfish Presence	58% (18 of 31 sites)	38% (11 of 29 sites)	10% (3 of 31 sites)	13% (4 of 30 sites)
Lionfish Density (#/100m^2 ± SE)	0.97 ± 0.18	0.66 ± 0.03	0.33 ± 0.23	0.21±0.11
Lionfish Size (mean ± SE)	25.5cm ± 1.2cm	21.8cm ± 1.3cm	24.0cm ± 3.4cm	28.0±1.6
Fish families (#)	23	27	30	30
Fish species (#)	77	85	88	86
Coral cover %	19.3 ± 2.7	17.9 ± 2.3	2.0 ± 0.3	6.0 ± 1.2
Macro algae cover %	41.1 ± 1.7	42.5 ± 1.9	34.7 ± 1.9	36.1 ± 1.6
Rugosity (mean ± SE; range)	102.5 ± 6.4 cm 58-213 cm	102.7 ± 8.5 cm 9-218 cm	31.4 ± 1.7 cm 13-50 cm	44.8 ± 2.4 cm 22-73 cm

Table 1.2. Summary data on physical and biological characteristics of study sites inside and outside the Marine Conservation District (MCD) and the Virgin Islands Coral Reef National Monument (VICRNM). Data from Monaco et al. (2007) for VICRNM and Smith et al. (2010) for MCD. nd=no data

2002-2007	MCD		VICRNM	
	in	out	in	out
No. of Sites Surveyed	50	nd	41	38
Start & End dates	10/04/2007 thru 01/30/2008	nd	July 2002, 2003, 2004, 2005, 2006	July 2002, 2003, 2004, 2005, 2006

Depth Range	33-48 m	nd	23-33 m	21-33 m
Total Lionfish	0	nd	0	0
Fish families (#)	35	nd	28	32
Fish species (#)	112	nd	100	109
Mean % Coral Cover (SD)	25.3 (2.1)	nd	8.00 (11.02)	25.36 (22.71)
Mean Rugosity (SD)	nd	nd	1.47 (0.63)	2.00 (0.56)

2) Benthic habitats among sites

Cover of macro algae was similar among sites (36.1% to 42.5%), but percent coral cover ranged from a low of 2.0% inside VICRNM, to a high of 19.3% inside MCD (Table 1). Percent coral cover within the MCD showed an inverse relationship with depth whereas in VICRNM coral cover increased with depth. This was largely due to the highest coral cover occurring from about 30m - 42m (Fig 2.1). Macro algae was relatively uniform among depth categories (Fig 2.1). Average vertical relief was nearly double around the MCD than around VICRNM (Fig. 2.2) which was largely due to depth differences among sites, where mean rugosity and variability increased with depth (Fig. 2.3). As expected there was a significant positive relationship between % coral cover and rugosity (Fig. 2.4).

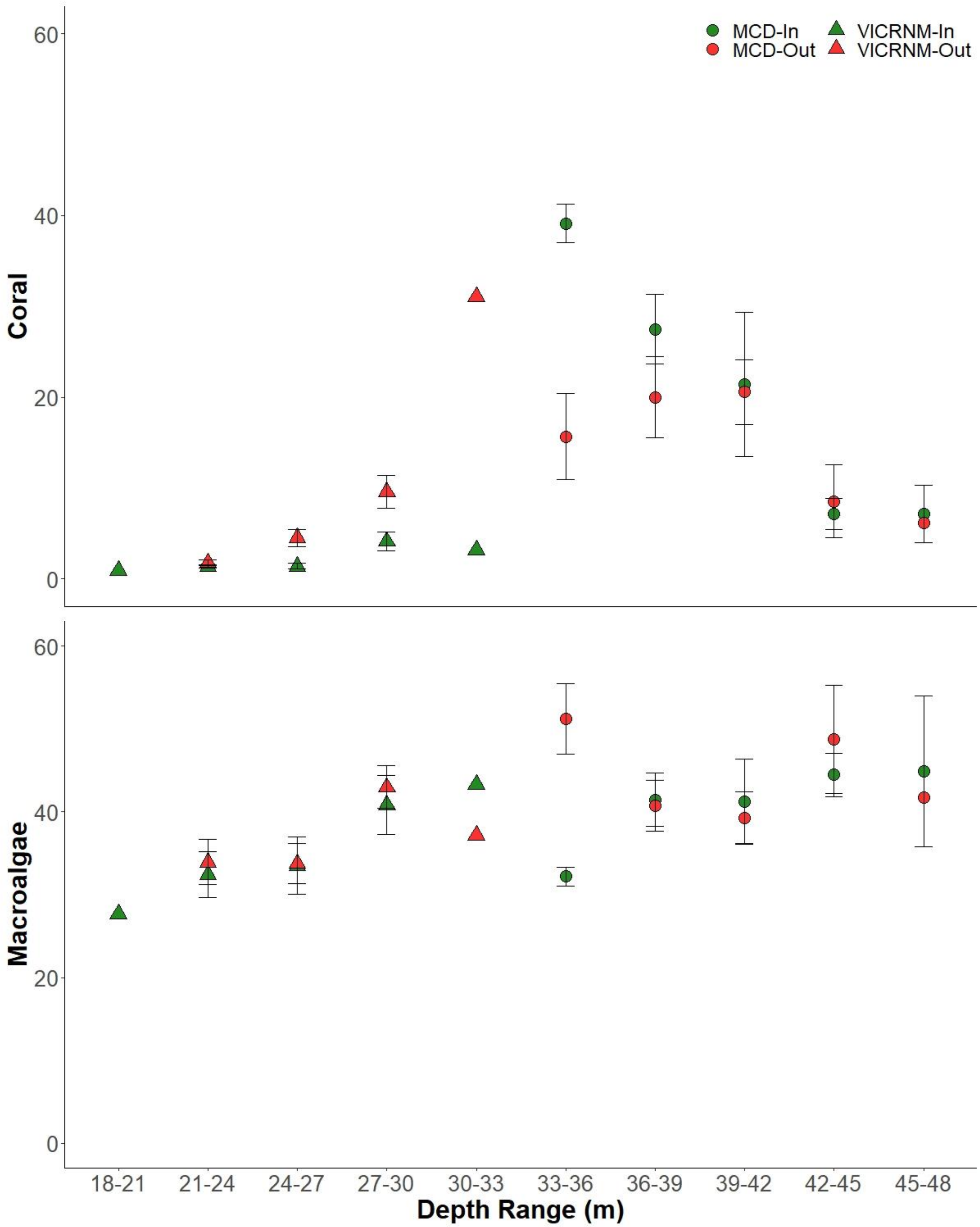


Figure 2.1 Mean (±SE) cover of live coral (upper) and macro algae (lower) inside and outside the Hind Bank Marine Conservation District (MCD) and Virgin Islands Coral Reef National Monument (VICRNM).

Depth	18-21	21-24	24-27	27-30	30-33	33-36	36-39	39-42	42-45	45-48
n	1	16	30	12	2	10	18	13	16	3

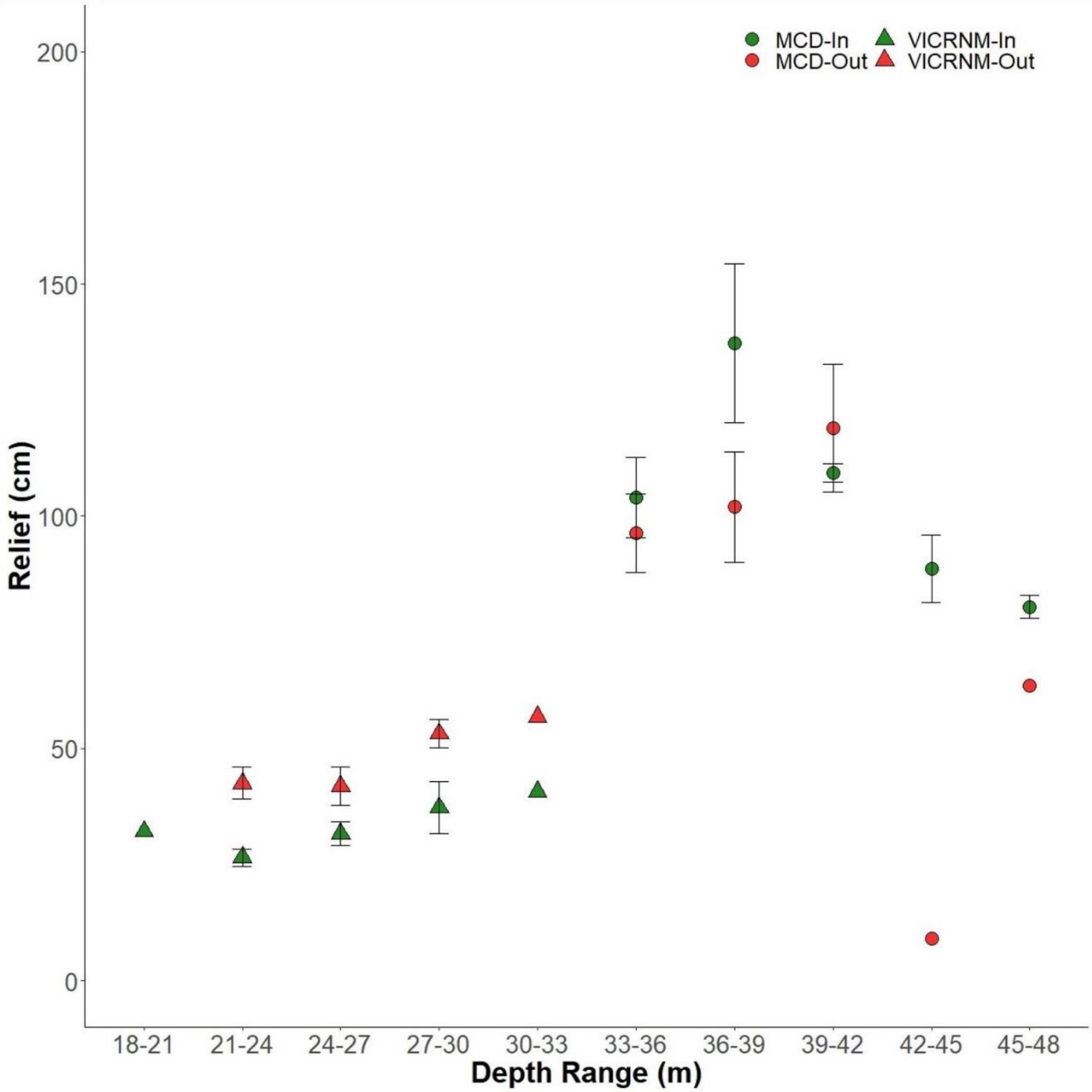


Figure 2.2 Mean (±SE) vertical relief (estimate of rugosity) of benthic structure inside and outside the Hind Bank Marine Conservation District (MCD) and Virgin Islands Coral Reef National Monument (VICRNM). Table shows number of sites sampled (n) within each depth range.

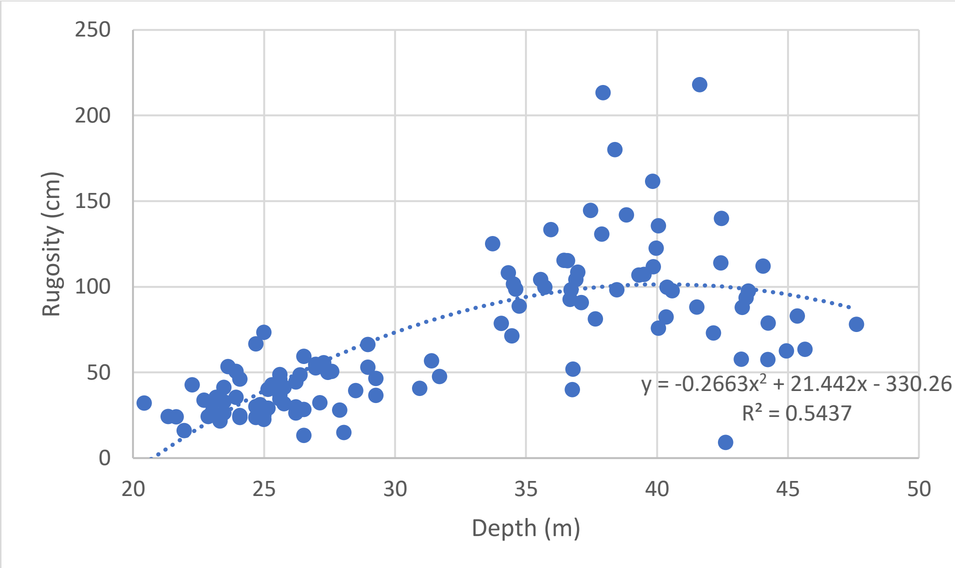


Figure 2.3. Significant relationship (F= 97.338, p<0.001) between rugosity (cm) and depth (m). Shape indicates the MPA in which the lionfish was observed, and color indicates whether the site was inside or outside the MPA.

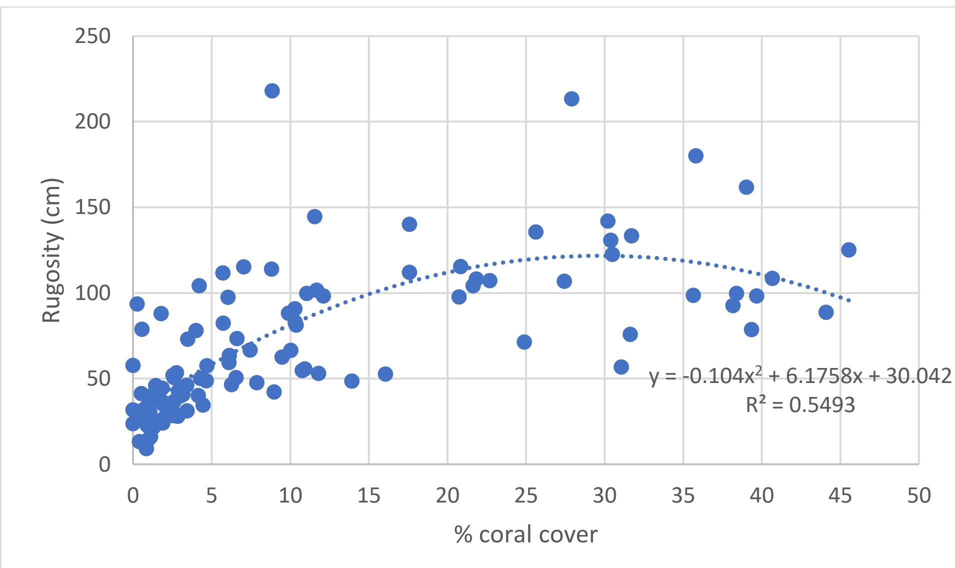


Figure 2.4. Significant relationship (F= 89.101, p<0.001) between rugosity (cm) and live coral cover (%).

3) Lionfish abundance and distribution patterns

Lionfish were most abundant in the 20 to 30cm size classes, especially around the MCD (Fig 3.1). Mean lionfish density was low, ranging from 0.21 lionfish /100 m^2 outside the VICRNM to 0.97 lionfish / 100 m^2 inside the MCD (Table 1.1). The MCD had more than twice the density and biomass of lionfish compared to VICRNM (Figs 3.2 and 3.3). Lionfish were present on 58% of sites within MCD, 38% of sites outside the MCD, 10% of sites within the VICRNM, and 13% of sites outside the VICRNM (Table 1.1). There was no relationship between depth and lionfish size (Fig. 3.4), density (Fig 3.5) or biomass with all sites combined (Fig. 3.6). We found no significant relationship between rugosity and lionfish density or biomass (Figs 3.7 and 3.8)

A two-way analysis of variance showed there was significantly higher density of lionfish around the MCD than the VICRNM ($F=8.425$, $p<0.01$), but that there was not a significant difference in density across MPA boundaries ($F=1.29$, $p=0.26$), with no significant interaction between MPA and location inside or outside the boundaries ($F=0.25$, $p=0.62$). Lionfish biomass was significantly higher in the MCD than the VICRNM ($p=0.03$), but not across boundaries of an MPA ($p=0.16$). We found a significant positive relationship between lionfish biomass and depth within the MCD ($F= 7.86$, $p = 0.013$, $R^2 = 0.33$) (Figure 3.6), but no relationship with depth and lionfish density ($R^2 = 0.01$), indicating that larger lionfish tended to be on deeper reefs.

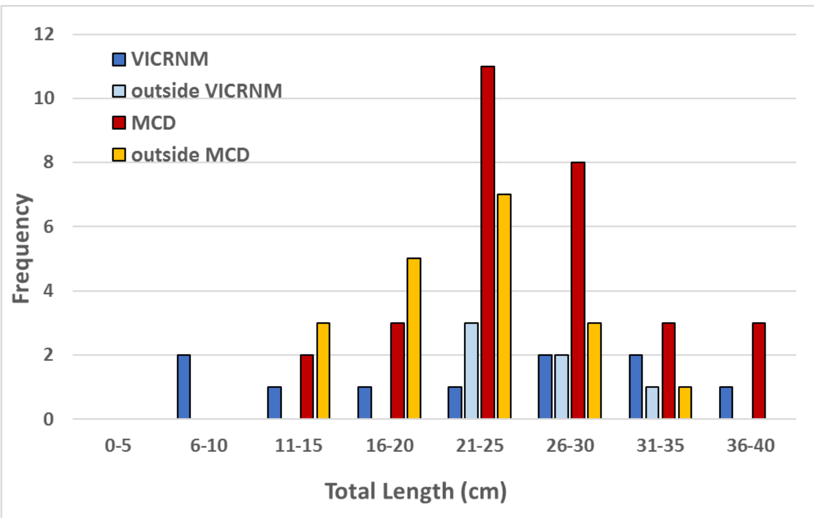


Figure 3.1. Size frequency of estimated lengths of lionfish (n=65; 5-cm bins) inside and outside the Virgin Islands Coral Reef National Monument (VICRNM) and inside and outside of the Hind Bank Marine Conservation District (MCD).

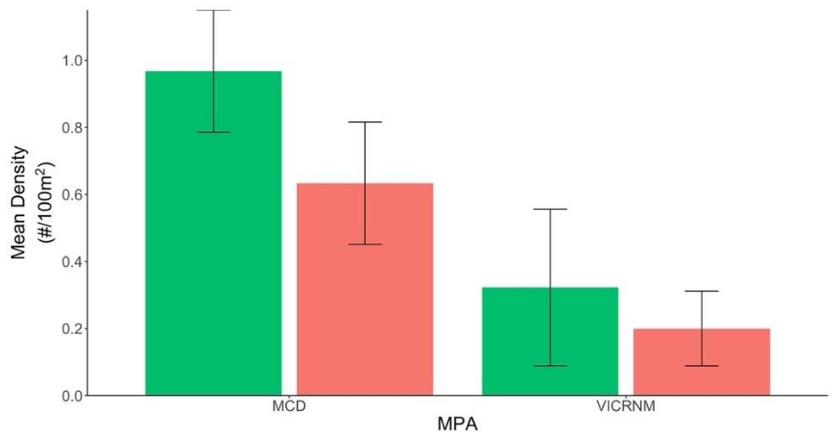


Figure 3.2. Mean (±SE) density of lionfish inside (green) and outside (red) MPA's.

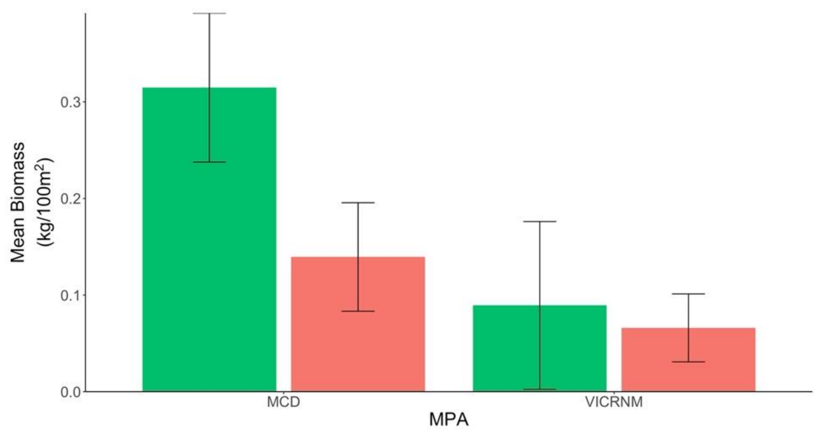


Figure 3.3. Mean biomass of lionfish inside (green) and outside (red) MPA's. Error bars represent mean ± SE.

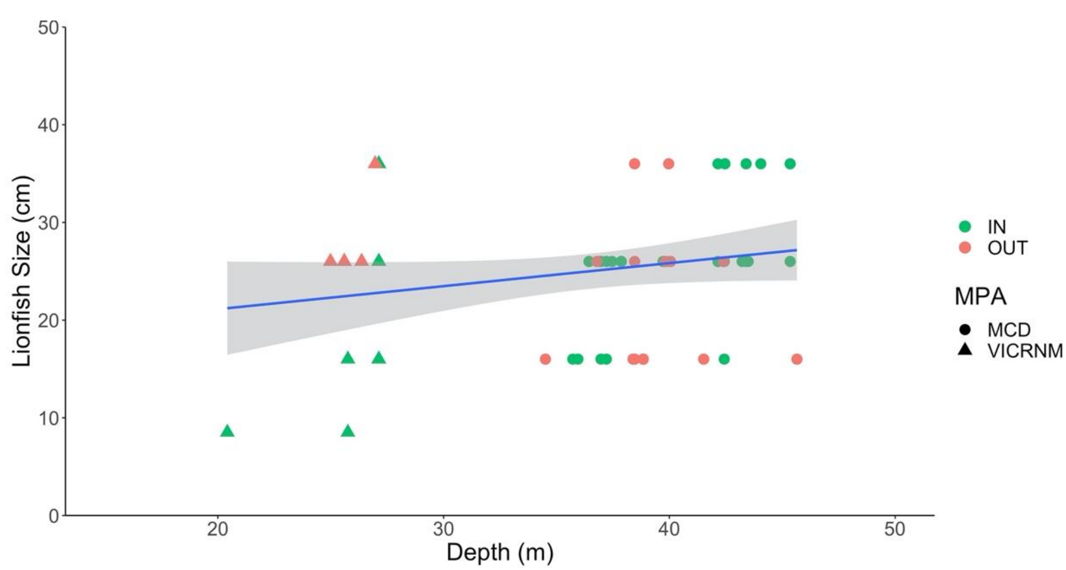


Figure 3.4. Estimated size (cm) of lionfish observed in 2018 and 2019, by depth of occurrence (m). Size was estimated in 10-cm bins, plotted as the median value of the bin (i.e. 11-20 cm plotted as 16 cm).

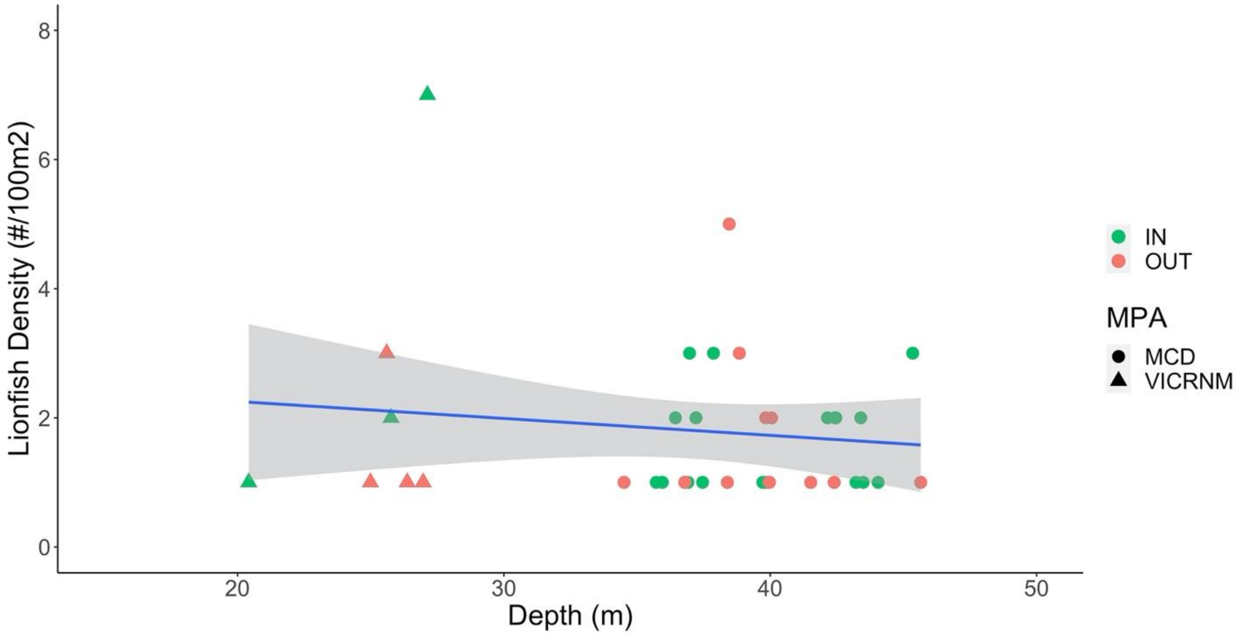
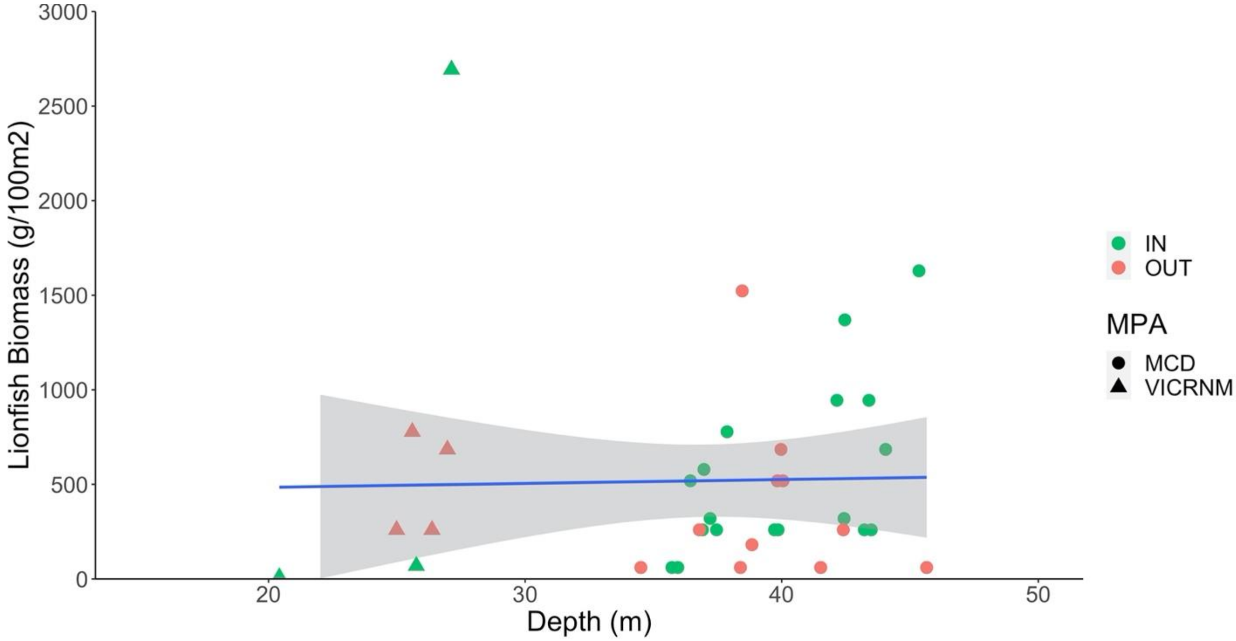


Figure 3.5. Density of lionfish (number per 100m^2) observed in 2018 and 2019, by depth of occurrence.

A.



B.

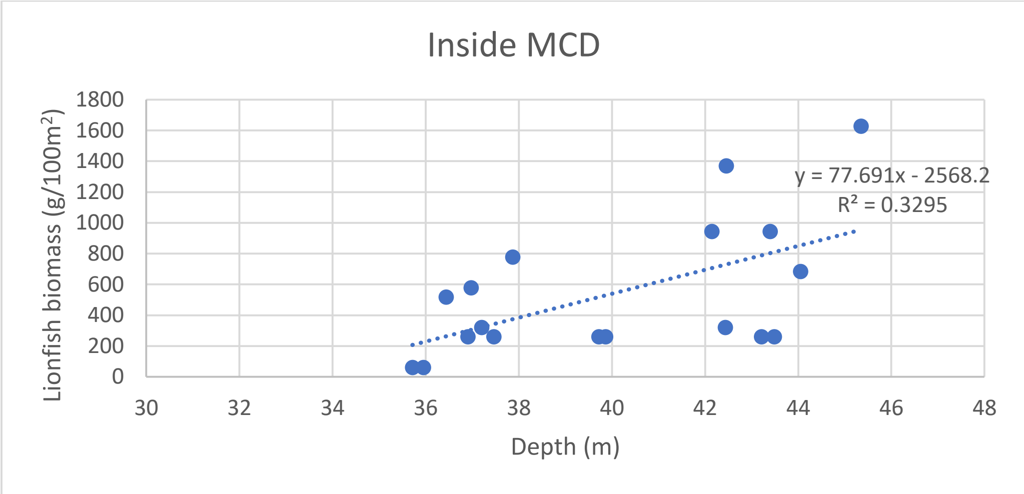


Figure 3.6. Biomass of lionfish (g per 100m^2) observed in 2018 and 2019, by depth of A) all sites and B) inside MCD only. A significant relationship was found for lionfish biomass and depth within the MCD (green dots only in A; p = 0.013, R^2 = 0.33)

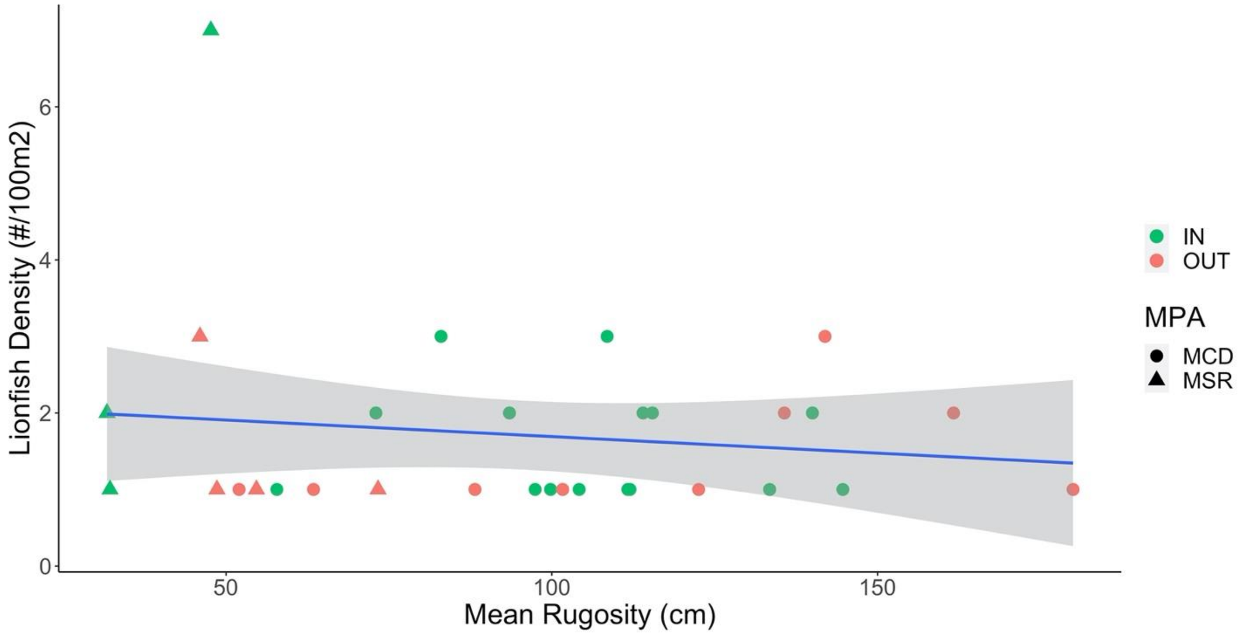


Figure 3.7. Rugosity (cm) of sites by lionfish density (#/100m2). Shape indicates the MPA in which the lionfish was observed, and color indicates whether the site was inside or outside the MPA.

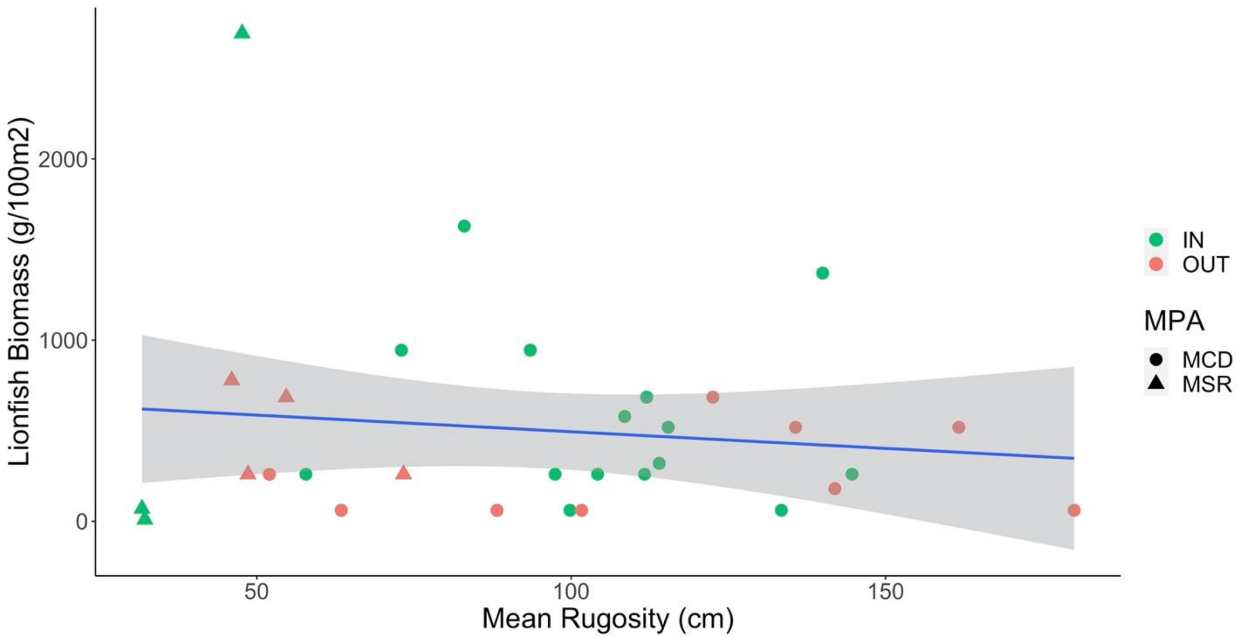


Figure 3.8. Rugosity (cm) of sites by lionfish biomass (g/100m2). Shape indicates the MPA in which the lionfish was observed, and color indicates whether the site was inside or outside the MPA.

4) Native reef fish biomass and diversity among sites

A total of 119 fish species representing 35 fish families were found on underwater visual surveys. Fish diversity was lowest inside the MCD (77 species, 23 families) and highest inside the VICRNM (88 species, 30 families) (Table 1). The patterns of fish abundance were similar inside and outside MCD but was different inside VICRNM. Higher biomass of several fish families inside VICRNM were evident, including Acanthuridae (surgeonfishes), Balistidae (triggerfish), Haemulidae (grunts), Mullidae (goatfish), and Pomacanthidae (angelfish) (Figure 4.1). Mean total fish density was highest inside the VICRNM, and lowest outside the MCD, with a two-way ANOVA showing MPA being a significant predictor ($p=0.006$, $F_1=7.87$) and inside/outside being a suggestive predictor ($p=0.06$, $F_1=3.65$) (Figure 4.2). Herbivores were present at all sites, but higher inside the VICRNM than the MCD. Piscivores were present at most sites in low densities, from 4.1 per 100 m^2 inside the MCD to 8.0 per 100 m^2 inside the VICRNM.

Native fish density was significantly different between MPAs for all native reef fish ($p<0.01$), herbivores ($p<0.0001$), and piscivores ($p<0.01$), but not significantly different for these groupings across boundaries of an MPA ($p=0.07$, $p=0.5$, $p=0.9$, respectively). These groups had higher densities at the VICRNM than the MCD (Fig. 4.2), opposite of the pattern in lionfish (Fig. 4.2). Total fish biomass was similarly high outside the MCD (10.55 ± 1.7 kg) as inside the VICRNM (10.50 ± 1.8 kg); total biomass was not significantly different between MPAs ($p=0.44$) or across boundaries ($p=0.46$) (Figure 4.3). Herbivore biomass was not significantly different between MPAs ($p=0.32$) or across boundaries (p=0.87), and while piscivore biomass was not significantly different between MPAs, but it was significantly higher outside MPAs than inside (p=0.04).

Lionfish prey, considered any fish less than 10 cm in length, had significantly higher densities in the VICRNM than the MCD (p=0.006), and significantly higher densities inside MPAs than outside boundaries (p=0.02) (Figure 4.4). Prey biomass was also significantly higher in the VICRNM (p=0.02), but not different inside/outside the MPA (p=0.22) (Figure 4.5).

Potential lionfish predators, considered any piscivore greater than 30 cm in length, were present in low densities that were not significantly different across MPAs (p=0.62) or boundaries within an MPA (p=0.97) (Figure 4.4). Biomass of potential predators was also not significantly different across MPAs (p=0.52), but was significantly higher outside MPAs than inside (p=0.03) (Figure 4.5).

Potential competitors for lionfish, defined as piscivores greater than 20 cm in length, followed similar patterns to potential predators (Figure 4.4), as many of the fish who could be predators on small lionfish also could be competitors to larger lionfish. Densities of potential competitors were not significantly different between MPAs (p=0.7) or across boundaries (p=0.8). Biomass of potential competitors was also not significantly different between MPAs (p=0.6) or across boundaries (p=0.06) (Figure 4.5).

Non-competitors for lionfish, defined as non-piscivores greater than 20 cm in length (i.e. herbivores, planktivores, detritivores, invertivores), showed no clear patterns across locations (Figure 4.4). Densities of non-competitors were not significantly different between MPAs (p=0.3) or across boundaries (p=0.5), and biomass of non-competitors was also not significantly different between MPAs (p=0.3) or across boundaries (p=0.6) (Figure 4.5). We found no significant relationship between rugosity and total fish density or biomass (Figures 4.6 and 4.7)

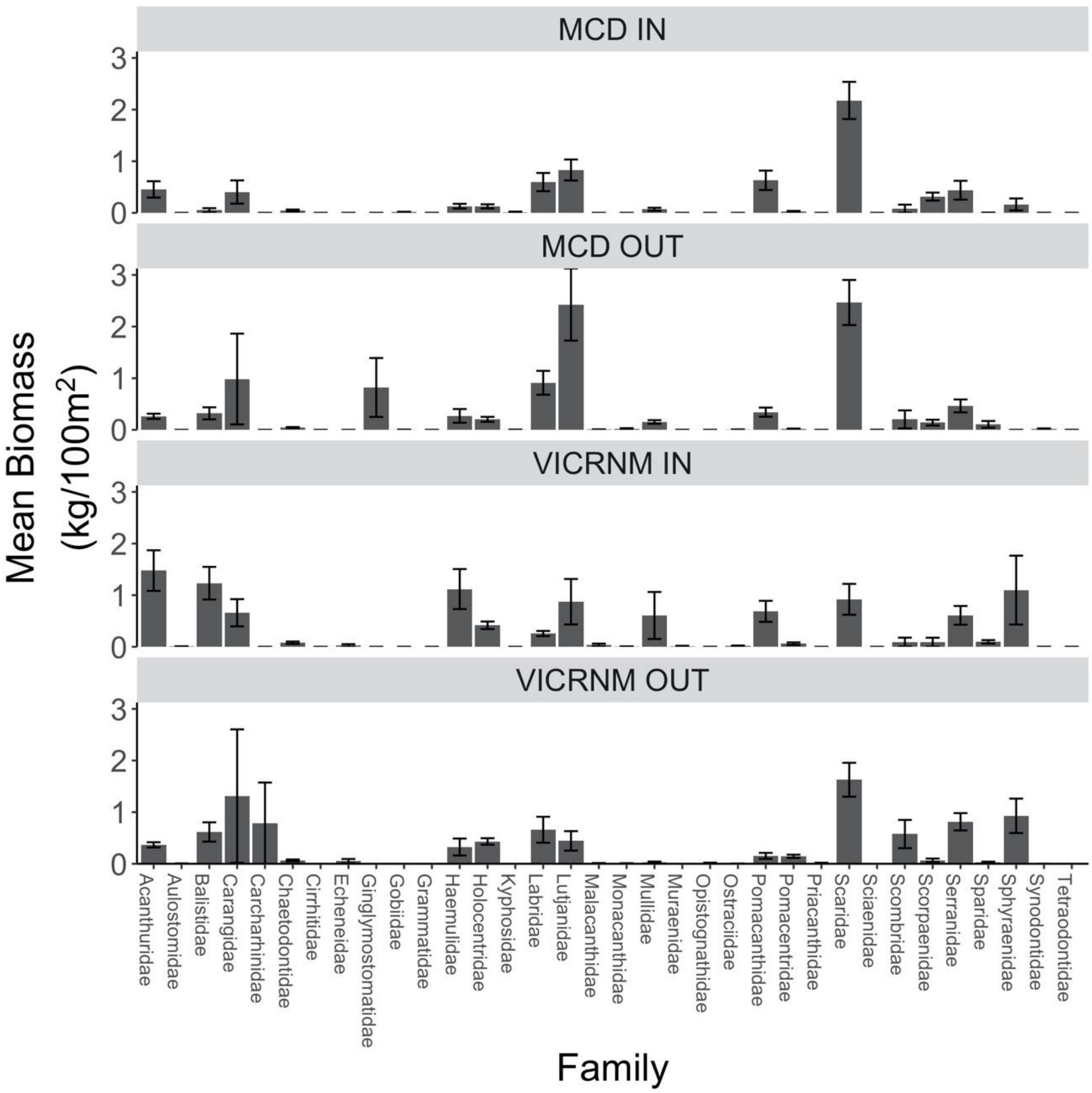


Figure 4.1. Mean fish family biomass (g/100m^2 ±SE) across sites, separated by MPA and inside/outside MPA boundaries.

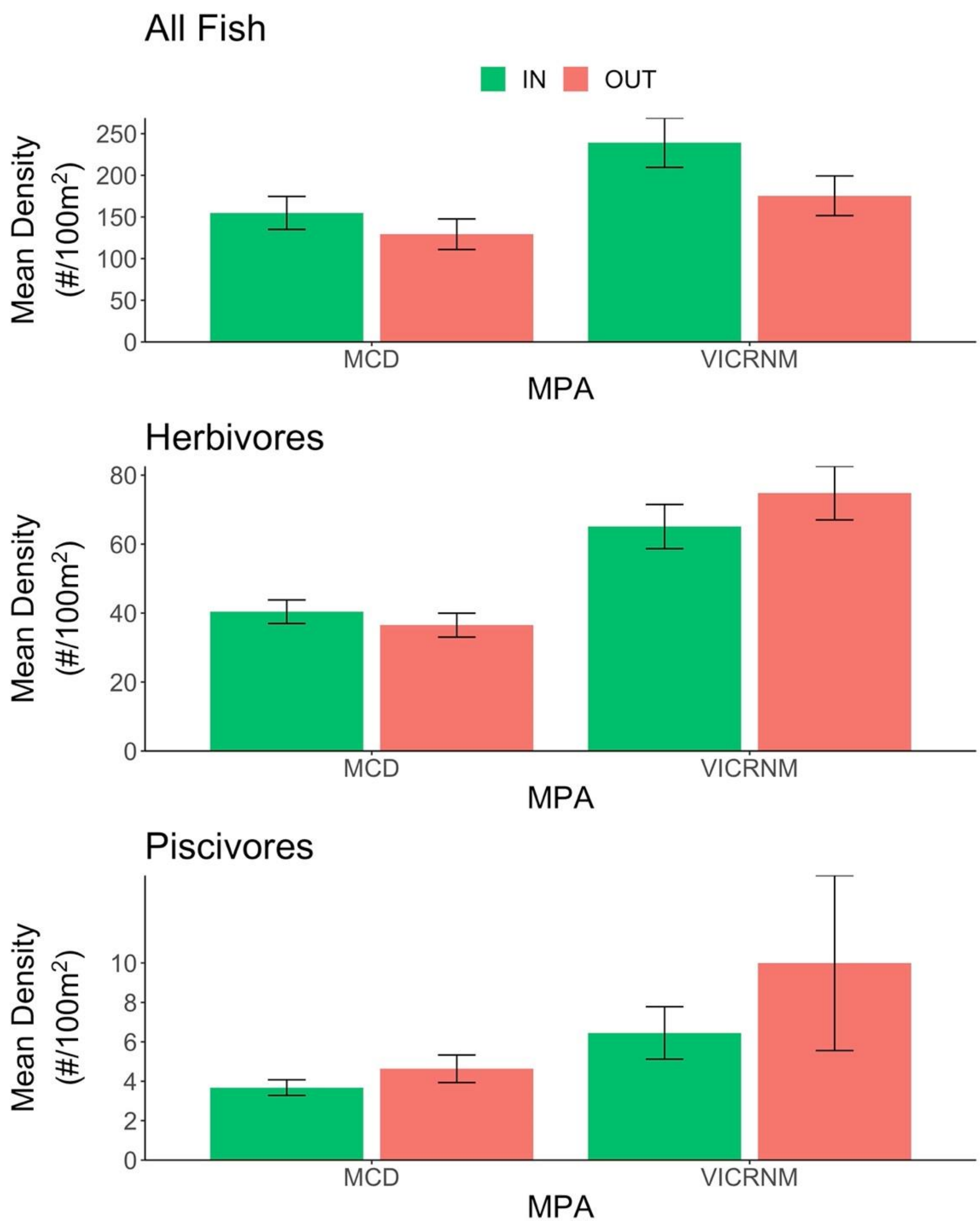


Figure 4.2. Mean fish density (#/100m^2 ±SE) across sites for all fish, herbivores, and piscivores, separated by MPA and inside/outside MPA boundaries.

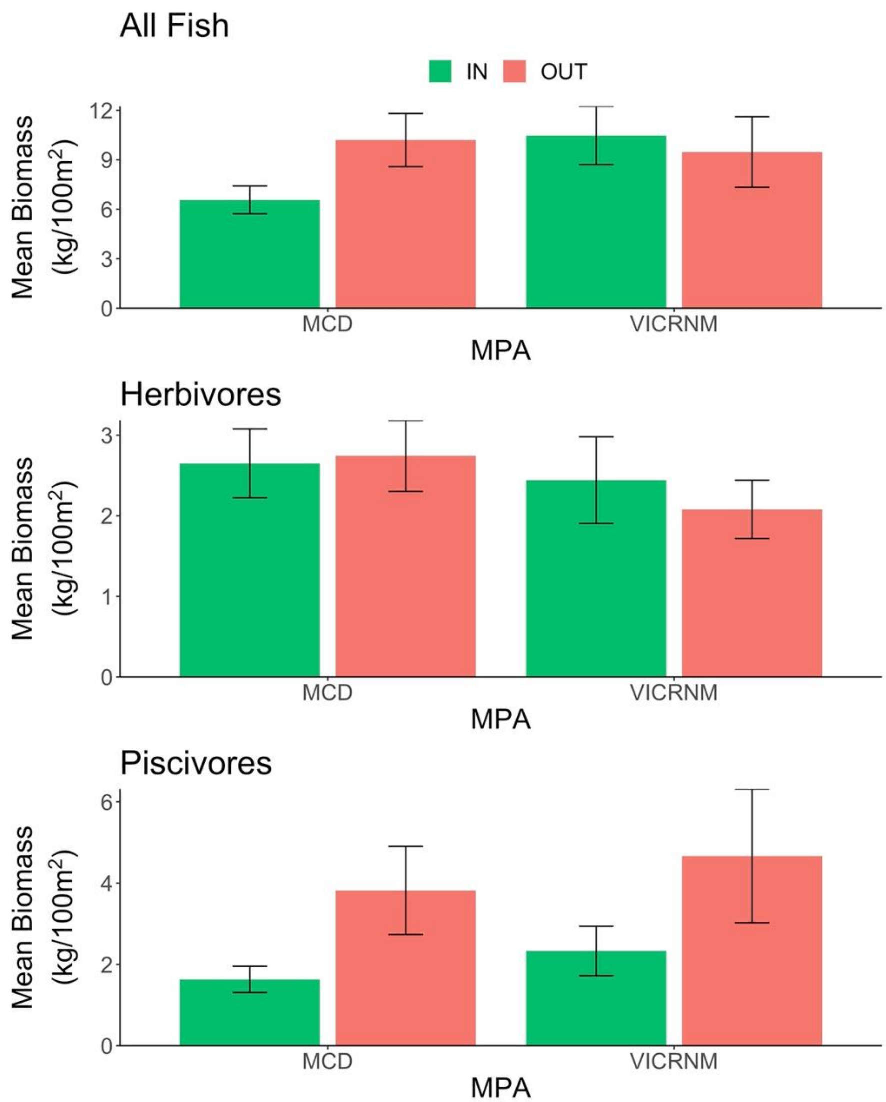


Figure 4.3. Mean fish biomass (kg/100m^2 ±SE) across sites for all fish, herbivores, and piscivores, separated by MPA and inside/outside MPA boundaries.

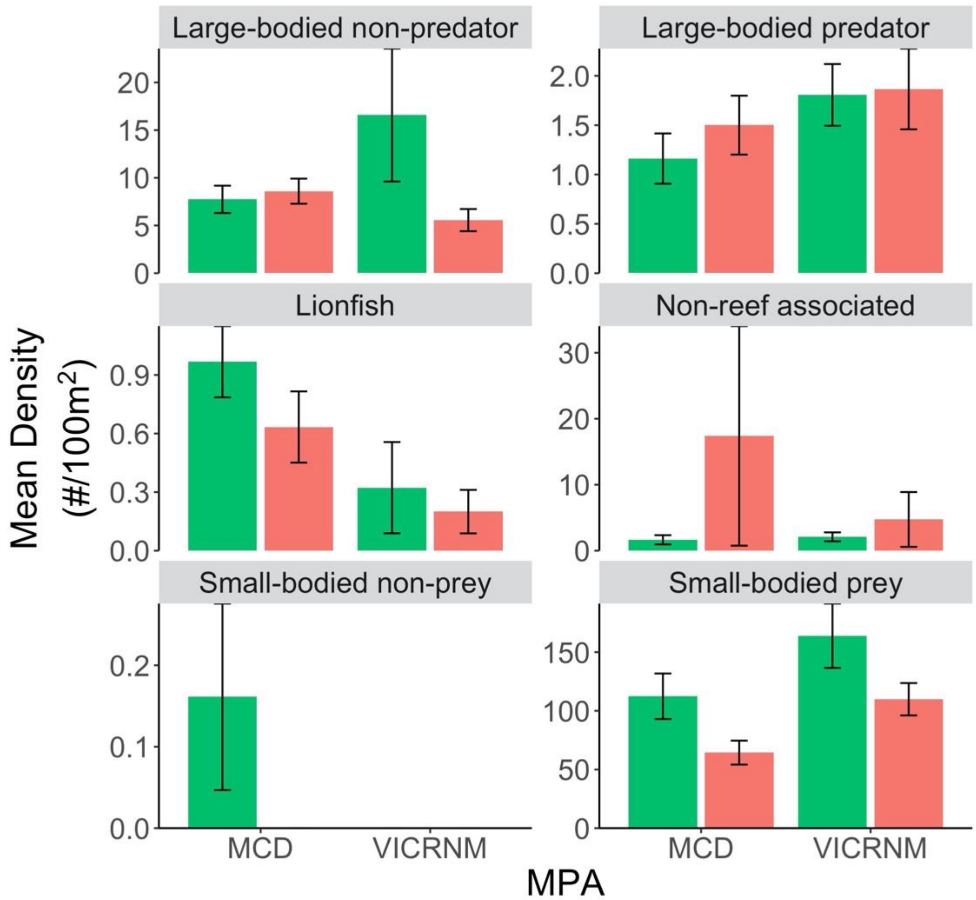


Figure 4.4. Mean fish density (#/100m^2 ±SE) across sites for all lionfish and other groups of interest, separated by MPA and inside/outside MPA boundaries. Groups were adapted from Green et al (2012).

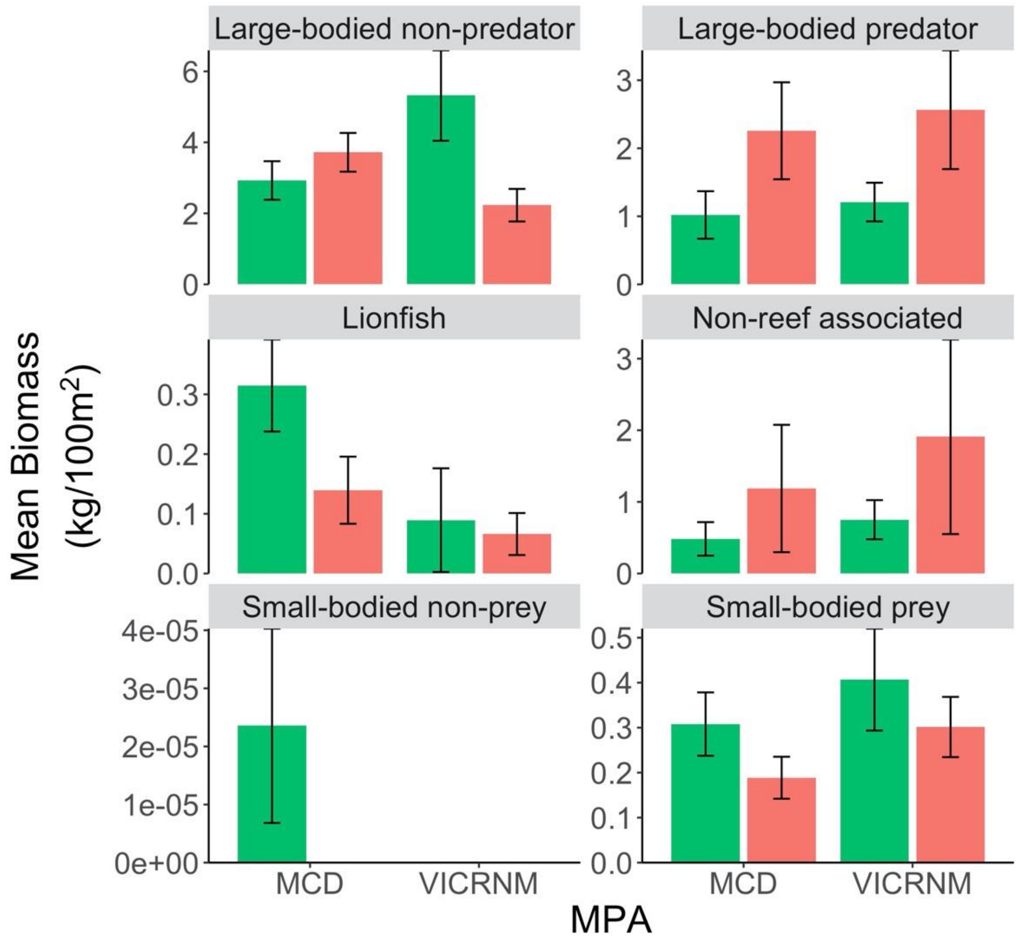


Figure 4.5. Mean fish biomass (kg/100m^2±SE) across sites for lionfish and other groups of interest, separated by MPA and inside/outside MPA boundaries. Groups were adapted from Green et al (2012).

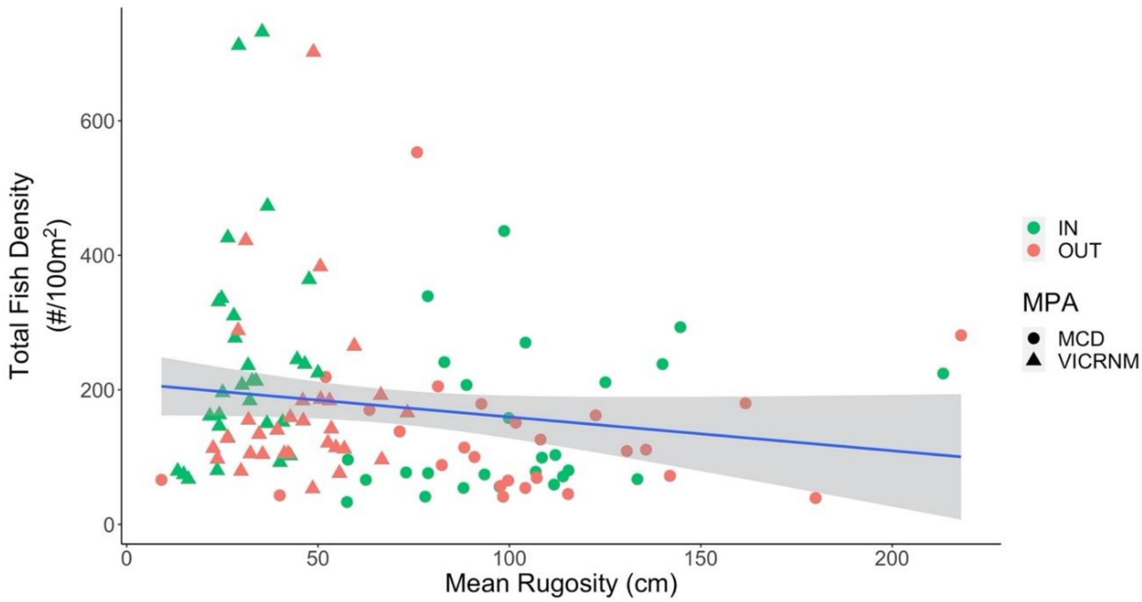


Figure 4.6. Rugosity (cm) of sites by total fish density (#/100m2). Shape indicates the MPA in which the lionfish was observed, and color indicates whether the site was inside or outside the MPA.

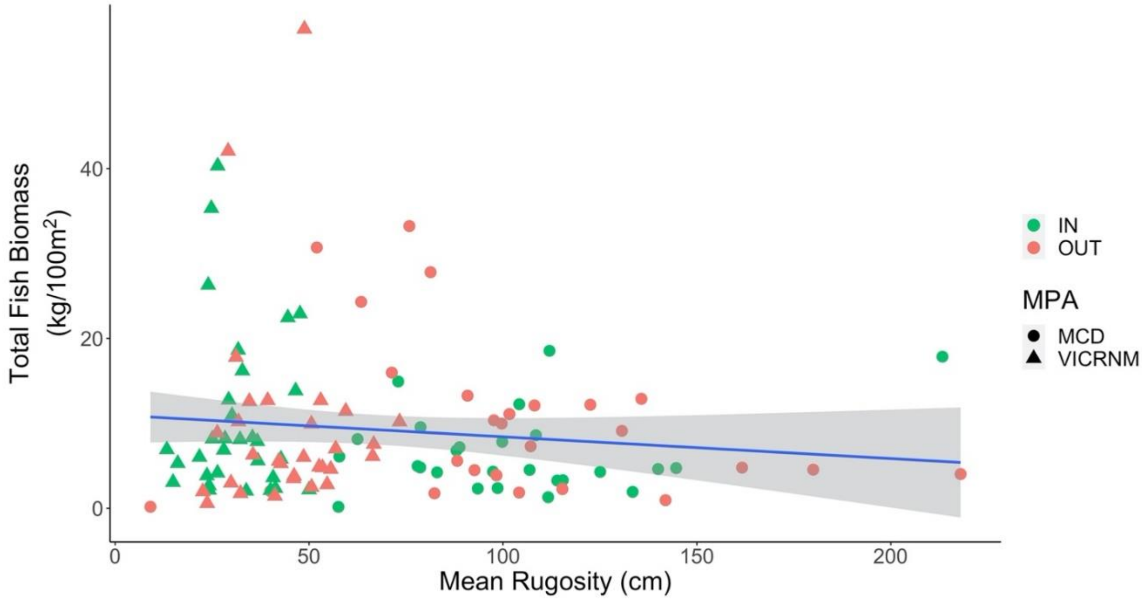


Figure 4.7. Rugosity (cm) of sites by total fish biomass (g/100m2). Shape indicates the MPA in which the lionfish was observed, and color indicates whether the site was inside or outside the MPA.

5) Changes in benthic structure and native fish abundance and diversity over time periods

Percent cover of live coral, gorgonians, and sponges declined between the first and second sampling periods inside and outside VICRNM but were relatively stable inside and outside MCD (Figure 5.1). This largely an artifact that surveys within the VICRNM were conducted before the 2005-2006 mass bleaching and disease event that caused large-scale mortality of coral reefs around the USVI. The MCD surveys were conducted after this mortality event in 2007-2008. Macro algae was stable in MCD and VICRNM but epilithic algae increased inside and outside the VICRNM (Figure 5.1). There was no apparent change in total fish density, herbivores or piscivores between first and second sampling periods in MCD. However, there was a significant decline in total fish density in the VICRNM (Figure 5.2). However piscivores within the MCD showed a trend of increasing density (Figure 5.2). Biomass across time periods were relatively unchanged in both MPAs (Figure 5.3).

Density of key lionfish functional groups (potential small bodies prey and non-prey, and large bodies competitors and predators) showed only a few significant trends from first to second sampling periods. Small bodied lionfish prey declined significantly from first to second sampling periods outside the VICRNM (Figure 5.4). Large bodied non-competitors and non-predators show an increasing trend in density from first to second time periods especially inside the VICRNM (Figure 5.4). Finally, large-bodied predators showed decline in density outside the VICRNM and inside the MCD (Figure 5.4). The biomass within these functional groups were relatively stable in MCD (Figure 5.5). However large-bodied non-predators increased and small-bodied non-prey decreased inside VICRNM (Figure 5.5). Outside the VICRNM saw increases in large bodied predators, and decreases in small-bodied non-prey and small-bodied prey (Figure 5.5).

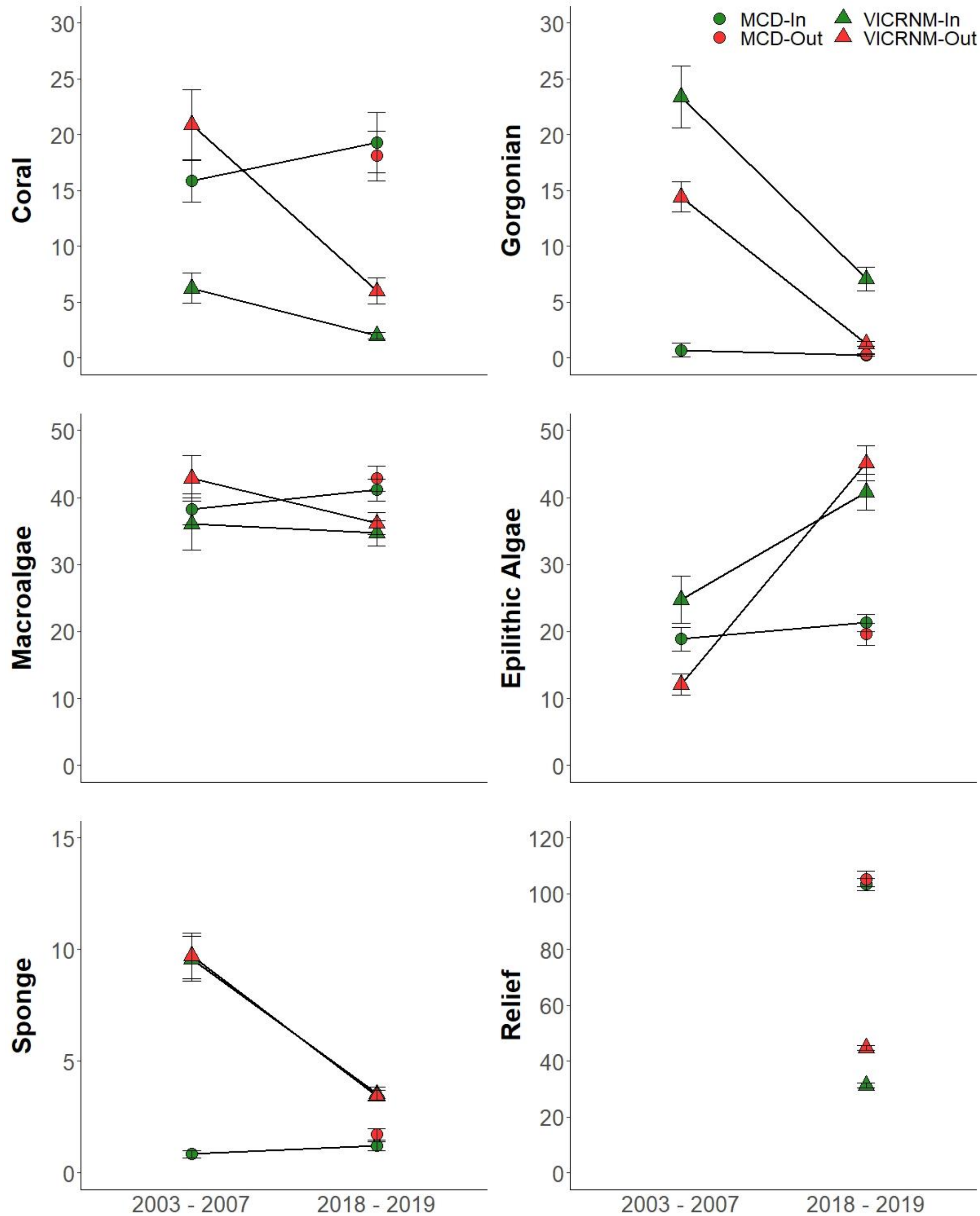


Figure 5.1 Change in mean (±SE) percent cover of coral, macroalgae, gorgonian, epilithic algae and sponge between first (2003-2007) and second survey periods (2018-2019). No data was available from 2003-2007 studies.

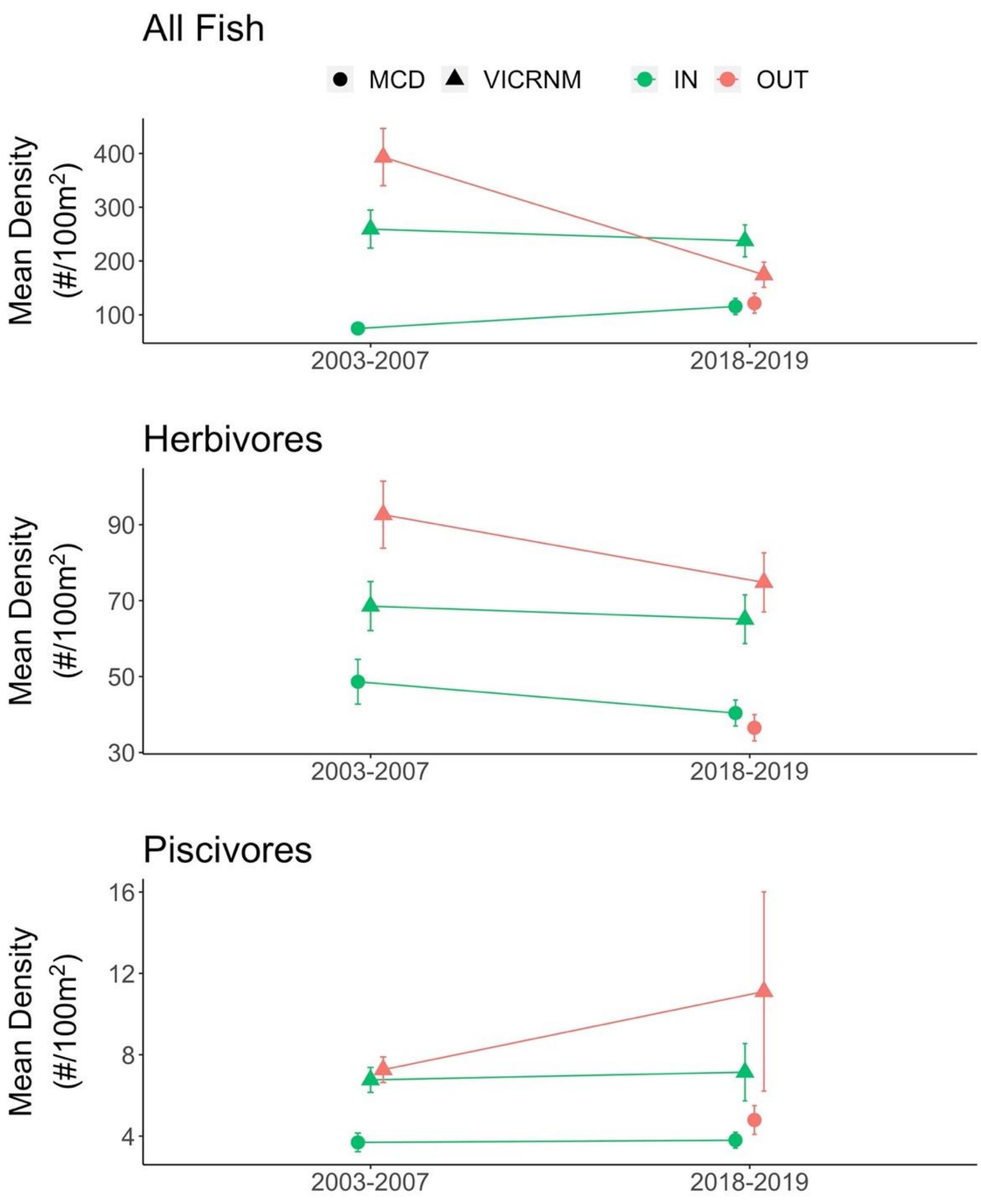


Figure 5.2. Mean density (#/100m^2±SE) of all fish (excluding gobies) summarized by time period, MPA, and inside/outside MPA boundaries. Shape represents the MPA in which the site was located, and color represents whether it was inside or outside MPA boundaries.

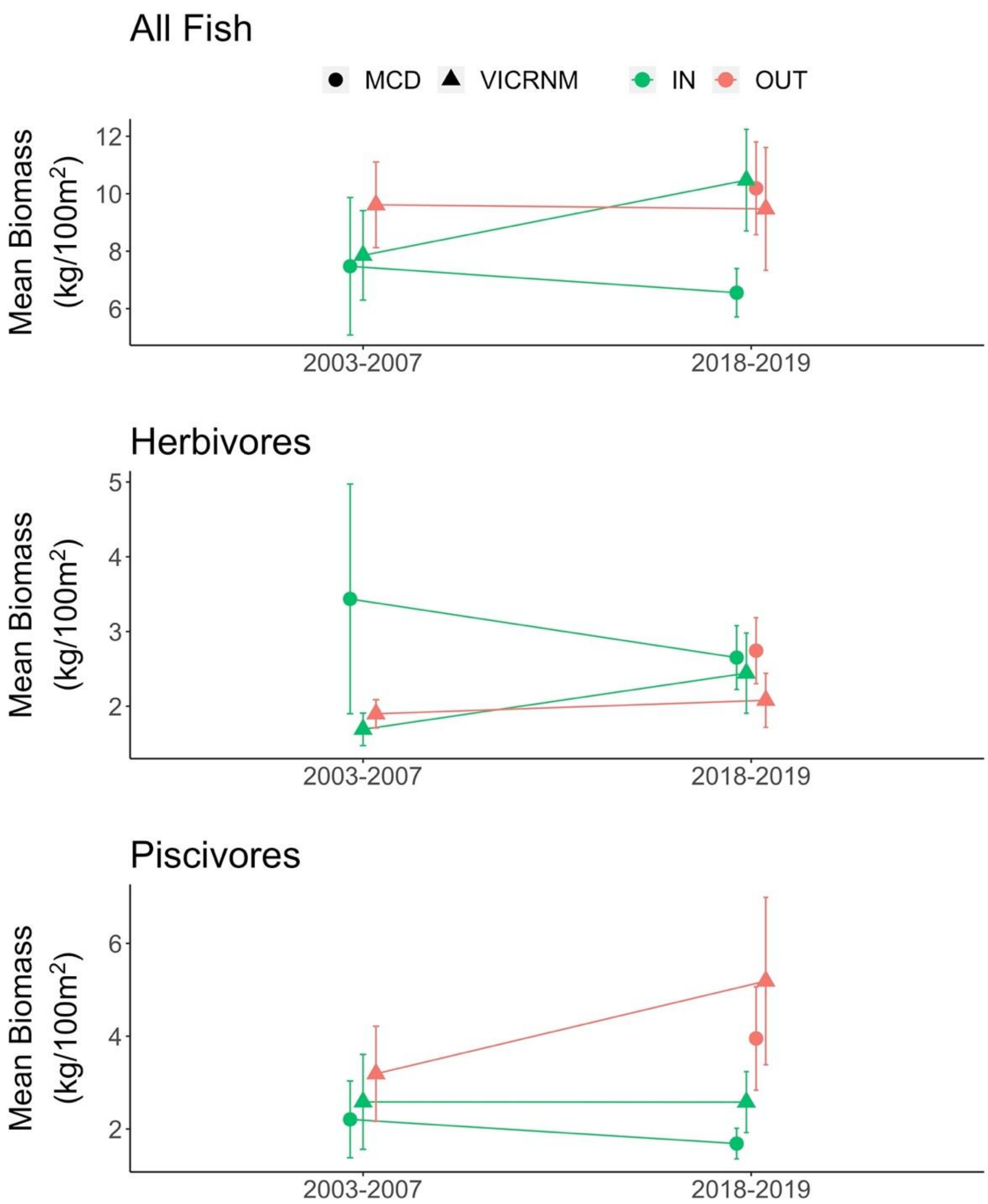


Figure 5.3. Mean biomass (kg/100m^2±SE) of all fish (excluding gobies) summarized by time period, MPA, and inside/outside MPA boundaries. Shape represents the MPA in which the site was located, and color represents whether it was inside or outside MPA boundaries.

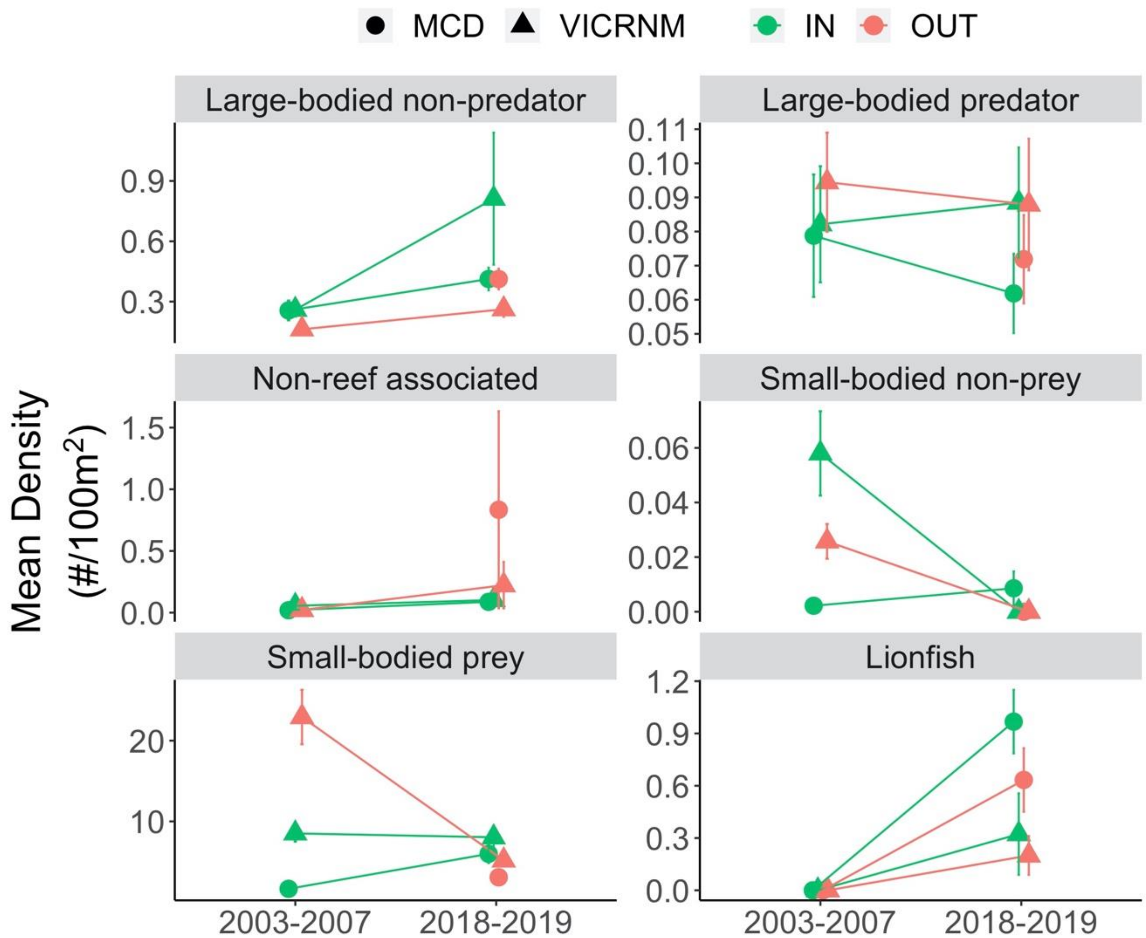


Figure 5.4. Mean fish density (#/100m^2) of groups of interest, summarized by time period, MPA, and inside/outside MPA boundaries. Error bars represent ± SE. Shape represents the MPA in which the site was located, and color represents whether it was inside or outside MPA boundaries.

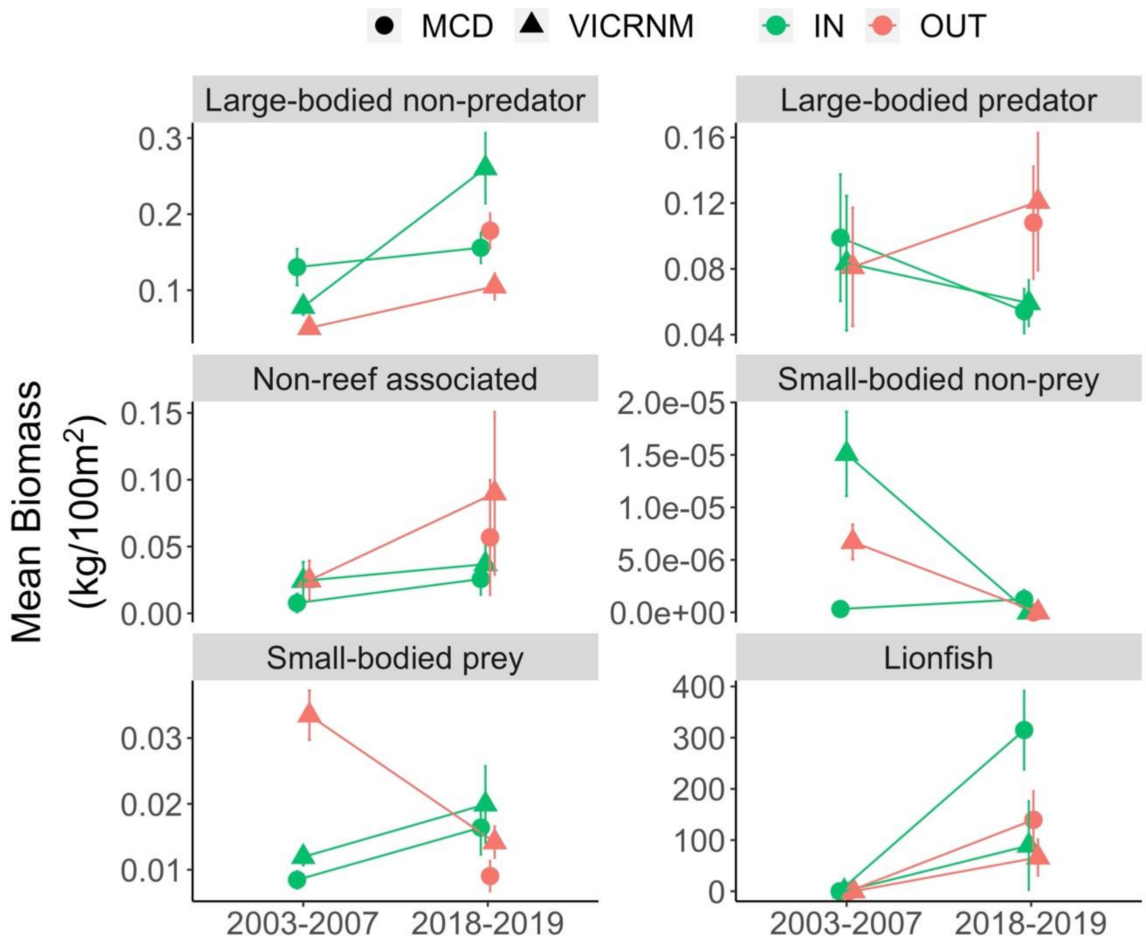


Figure 5.5. Mean fish biomass (kg/100m^2) of groups of interest, summarized by time period, MPA, and inside/outside MPA boundaries. Error bars represent ± SE. Shape represents the MPA in which the site was located, and color represents whether it was inside or outside MPA boundaries.

6) Data Synthesis

Functional groupings of fishes important to lionfish ecology (adapted from Green et al. 2012) were examined for differences among MPA's and between inside and outside of MPAs. These functional groupings included lionfish, large-bodied predators, large-bodied non-predators non-competitors, small-bodied prey, small-bodied non-prey, and non-reef-associated fishes. Non-metric Multi-Dimensional Scaling (NMDS) showed no clear differences in density or biomass of the various functional groups for inside and outside comparisons of either MPA (Figures 6.1, 6.2). Comparison of these functional groups between MCD and VICRNM was significantly different for density (df=3, p=0.002) (Figure 6.3) but not biomass (df=3, p=0.12) (Figure 6.4). The largest contributor to the differences in density was small-bodied prey, accounting for 78-84% of the differences between locations (both inside and outside MPAs). Specific regression analyses were conducted to examine the relationship between density or biomass of lionfish and small-bodied prey fish, potential lionfish predators or other functional groups and percent coral cover among our study sites. No clear patterns emerged from these analyses suggesting that at time of surveys lionfish density and biomass has had no clear impact on native fish populations or percent coral cover on mesophotic reefs.

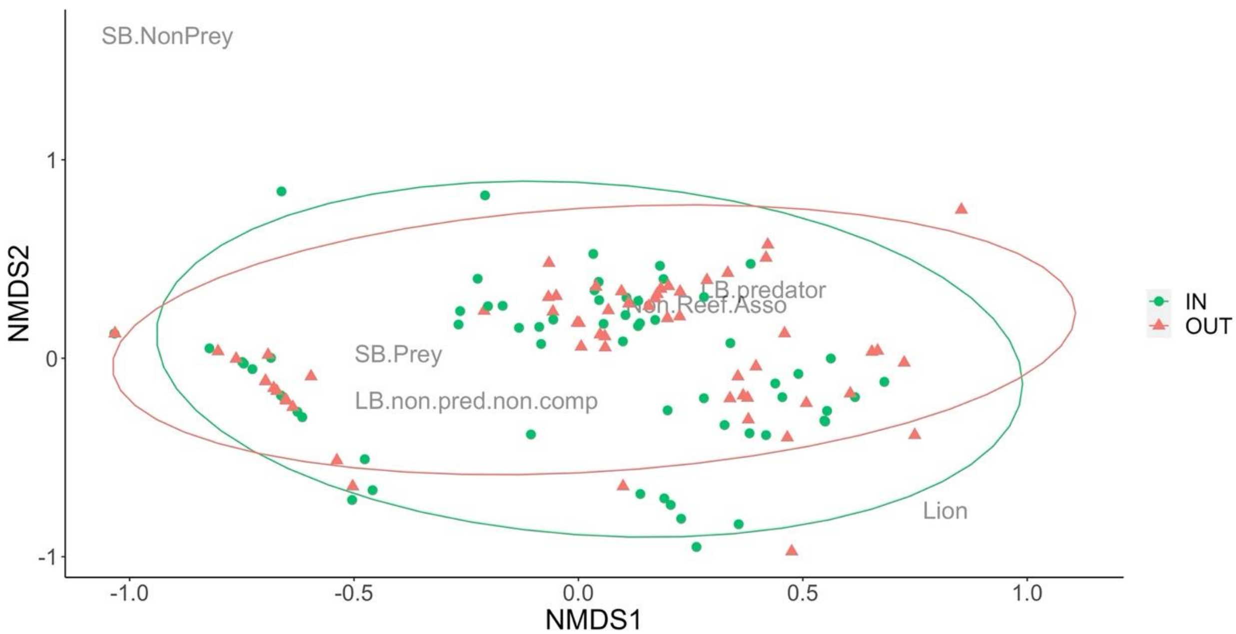


Figure 6.1. NMDS plot of **density** of various functional groups of interest inside and outside the protected area. Groups were adapted from Green et al (2012), and include lionfish (Lion), large-bodied predators (LB.predator), large-bodied non-predators non-competitors (LB.non.pred.non.comp), small-bodied prey (SB.prey), small-bodied non-prey (SB.NonPrey), and non-reef-associated (Non.Reef.Asso). Ellipses represent 95% groupings by in or out.

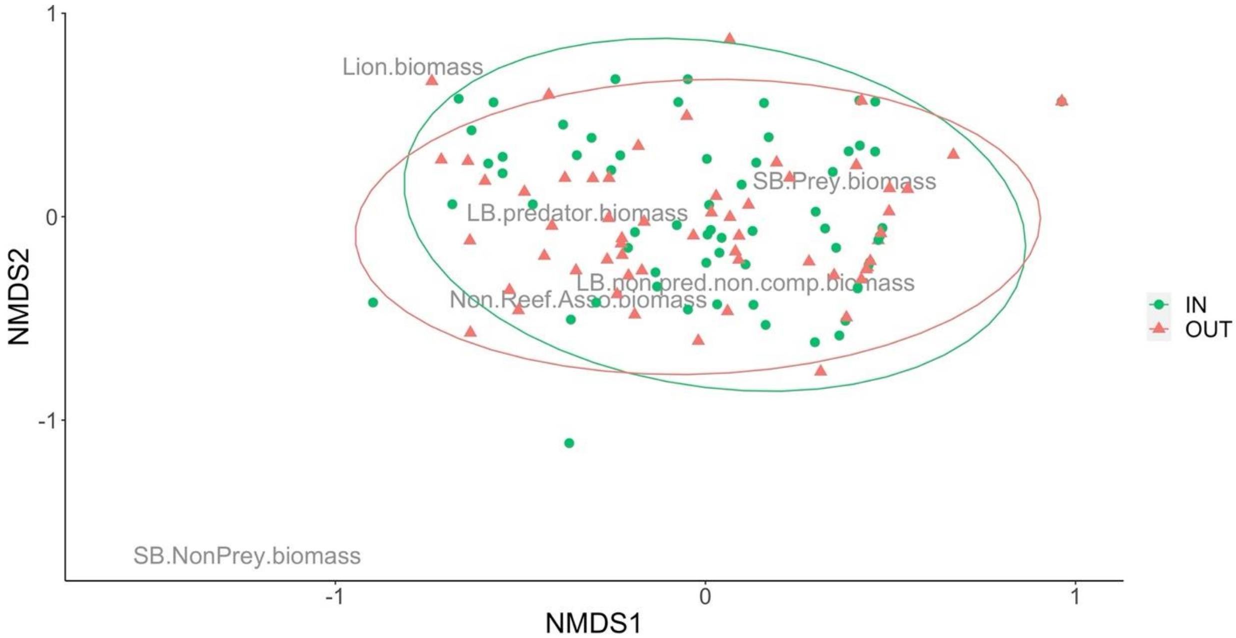


Figure 6.2. NMDS plot of **biomass** of various functional groups of interest inside and outside the protected area. Groups were adapted from Green et al (2012), and include lionfish (Lion), large-bodied predators (LB.predator), large-bodied non-predators non-competitors (LB.non.pred.non.comp), small-bodied prey (SB.prey), small-bodied non-prey (SB.NonPrey), and non-reef-associated (Non.Reef.Asso). Ellipses represent 95% groupings by in or out.

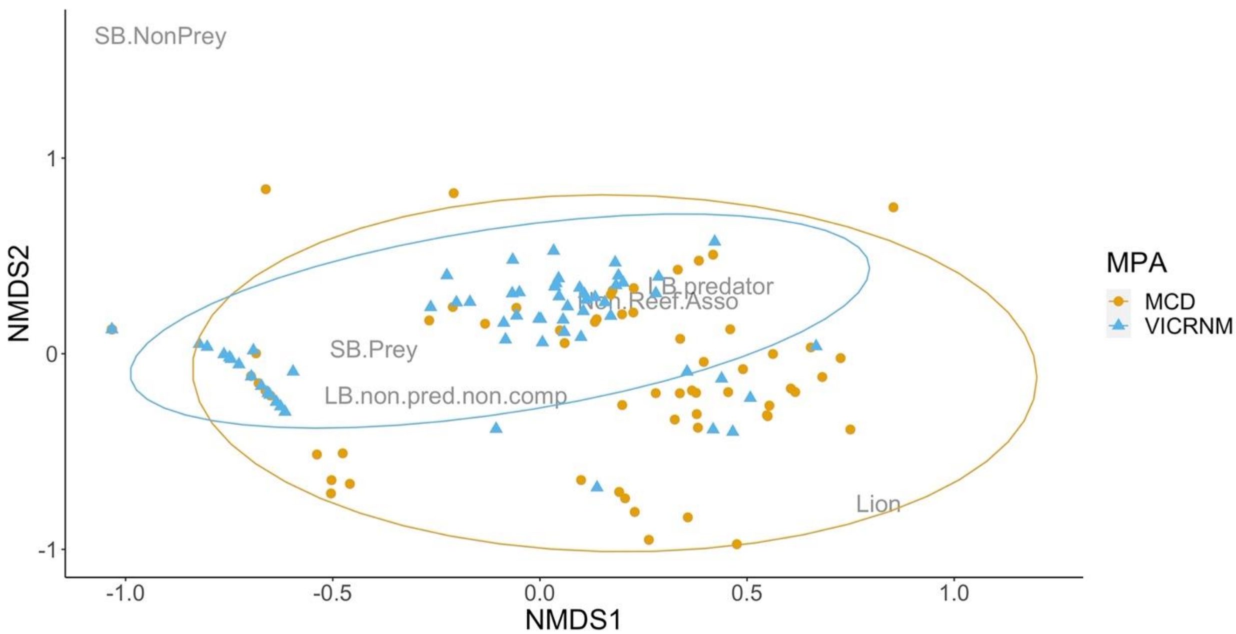


Figure 6.3. NMDS plot of **density** of various functional groups of interest by MPA. Groups were adapted from Green et al (2012), and include lionfish (Lion), large-bodied predators (LB.predator), large-bodied non-predators non-competitors (LB.non.pred.non.comp), small-bodied prey (SB.prey), small-bodied non-prey

(SB.NonPrey), and non-reef-associated (Non.Reef.Asso). Ellipses represent 95% groupings by MPA (inside and outside combined for each MPA). Locations (in terms of MPA and in/out) are different in their density of groups of interest (df=3, p=0.002). SIMPER analysis showed the largest contributor to these differences is small-bodied prey, accounting for 78-84% of the differences between locations.

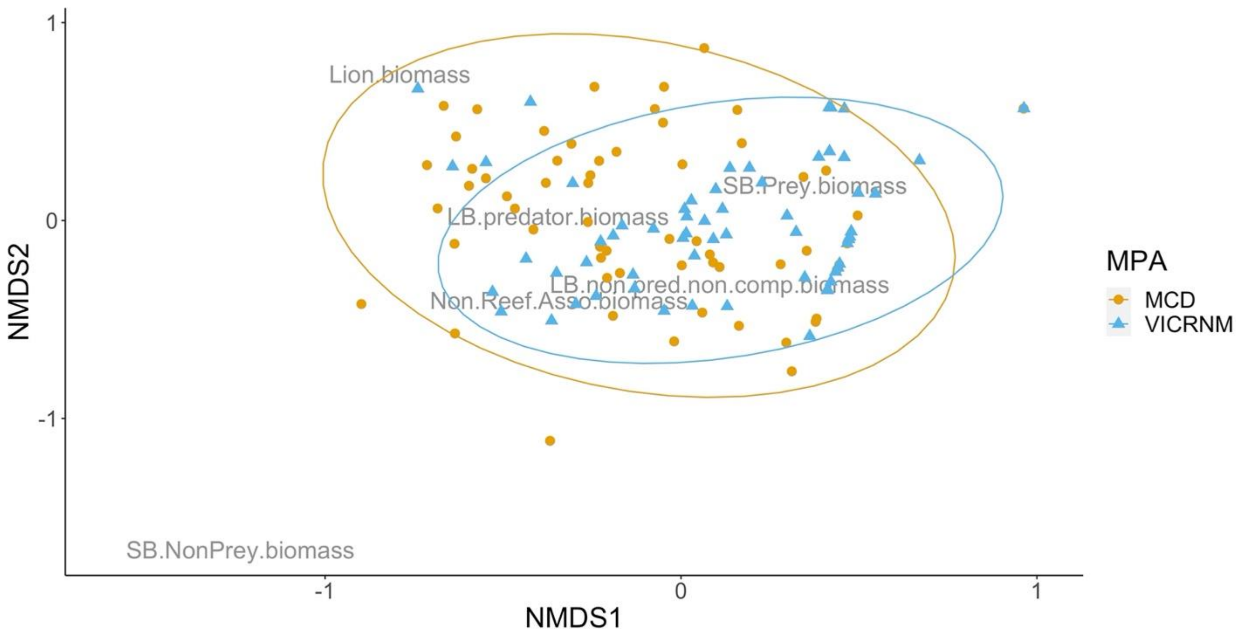


Figure 6.4. NMDS plot of **biomass** of various functional groups of interest for MCD and VICRNM (inside and outside combined for each MPA).

Literature Cited:

Monaco, M. E., A. M. Friedlander, et al. (2007). Characterising reef fish populations and habitats within and outside the US Virgin Islands Coral Reef National Monument: a lesson in marine protected area design. Fishe. Manage. Ecol. **14**: 33-40.

Nemeth, R. S. and A. Quandt (2005). Differences in fish assemblage structure following the establishment of the Marine Conservation District, St. Thomas U. S. Virgin Islands. Proceedings of the 56th Gulf and Caribbean Fisheries Institute., Tortola, BVI.

Smith, T. B., J. Blondeau, R. S. Nemeth, S. J. Pittman, J. M. Calnan, E. Kadison and J. Gass (2010). Benthic structure and cryptic mortality in a Caribbean mesophotic coral reef bank system, the Hind Bank Marine Conservation District, U.S. Virgin Islands. Coral Reefs **29**(2): 289-308.

2. Applications (address what is possible)

This section should describe specifically the outputs and management outcomes achieved. Outputs are defined as products (e.g. publications, models) or activities that lead to outcomes (changes in user knowledge or action). In cases where proposed management outcomes are not fully achieved, indicate the progress made during the reporting period. Also, indicate expected outputs and management outcomes for the next year of support.

a. Outputs

i. New fundamental or applied knowledge

Initial analysis of data showed that higher densities of lionfish and their potential prey (native fish < 10cm) were found within marine protected area boundaries, especially the MCD. This pattern could result from either lower fishing mortality of lionfish inside MPA boundaries, lionfish densities increasing due to greater abundance of prey or a combination of these two factors. Additional analysis found that there was a positive relationship between lionfish biomass and depth and rugosity, factors that may have contributed to higher lionfish biomass in MCD. we also found that since the arrival of lionfish no significant differences in potential lionfish prey or predators were affected.

ii. Scientific publications

None at this time.

iii. Patents

None

iv. New methods and technology or improved skills

None at this time.

v. New or advanced tools (e.g. models, biomarkers)

None at this time.

vi. Workshops

None at this time.

vii. Presentations

One presentation in preparation for 14th International Coral Reef Symposium, Bremen, Germany, July 2021.

viii. Outreach activities/products (e.g. website, newsletter articles)

None at this time.

b. Management outcomes I. Management application or adoption of:

i. New fundamental or applied knowledge

We found that mesophotic reefs were supporting large populations of the invasive lionfish (Pterois volitans), in particular reefs with higher relief (rugosity). This suggests that *culling-based management on shallow reefs will be ineffective if large deep-water populations of lionfish continue to persist.*

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ii. New or improved skills

Technical divers who have contributed to the project include technicians (Sarah Heidmann, Rossie Ennis, Shaun Kadison, Jason Quetel and Viktor Brandtneris) and Master's students (Alex Gutting, Allie Durdall, Rosmin Ennis, Dan Mele, Sonora Meiling, Jason Quetel and Joe Townsend),

.

iii. Information from publications, workshops, presentations, outreach products.

None at this time.

iv. New or improved methods or technology

None at this time.

v. New or advanced tools

None at this time.

c. Management outcomes - II. Societal condition improved due to management action resulting from output; examples:

i. **Improved water quality (N/A)**
ii. **Lower frequency of harmful algal blooms (N/A)**
iii. **Reduced hypoxic zone area (N/A)**
iv. **Improved sustainability of fisheries**

None at this time.

d. Partnerships established with other federal, state, or local agencies, or other research institutions (other than those already described in the original proposal).

Prepared By:

[signature]

Signature of Principal Investigator(s) — 30 April, 2020 Date

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