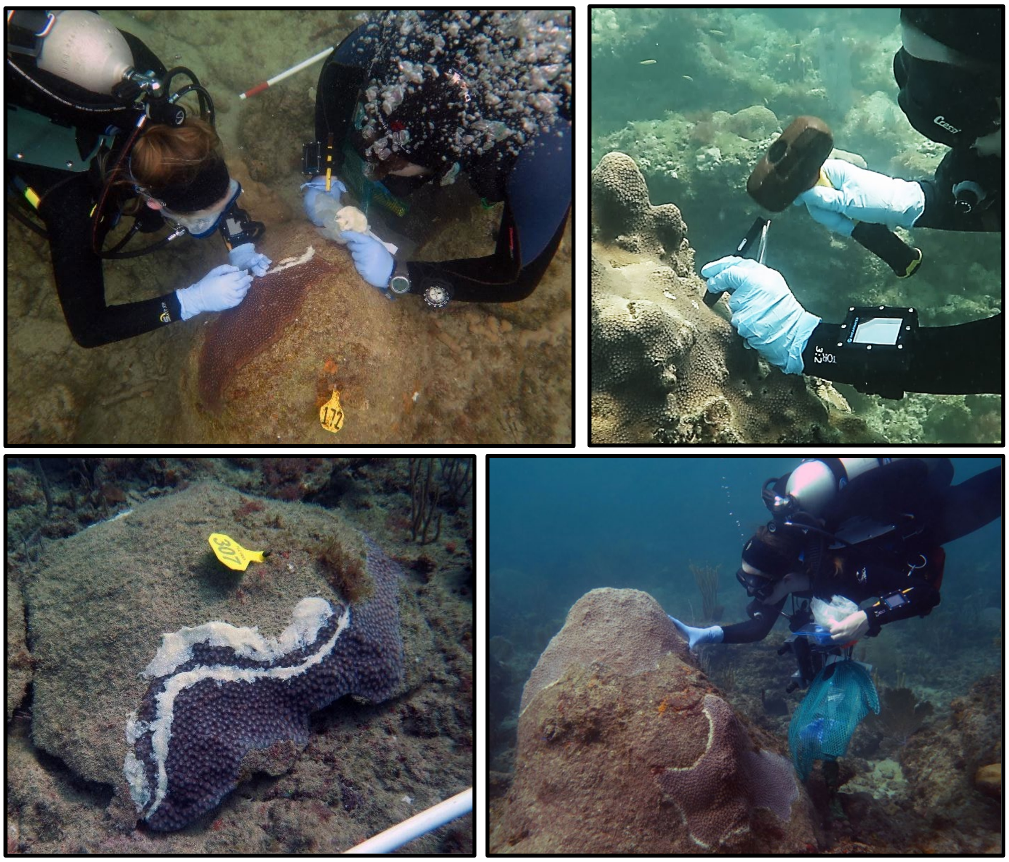
2021
Advancing Coral Intervention and Resilience in Southeast Florida

Final Report



Florida Department of Environmental Protection
Office of Resilience and Coastal Protection

Advancing Coral Intervention and Resilience in Southeast Florida

Final Report

Prepared By:

Joshua D. Voss, Erin Shilling, Gabrielle Pantoni,
Ryan Eckert, & Alexis Sturm

Florida Atlantic University
Harbor Branch Oceanographic Institute
5600 US Highway 1 North
Fort Pierce, FL 34946

June 30, 2021

Completed in fulfillment of PO B7C241

Florida Department of Environmental Protection
Office of Resilience and Coastal Protection
1277 N.E. 79th Street Causeway
Miami, FL 33138

This report should be cited as follows:
Voss J., Shilling E., Pantoni G., Eckert R. and A. Sturm. 2021. Advancing Coral Intervention and Resilience in Southeast Florida. Florida DEP. Miami, FL., pp. 1-18

This report was prepared for the Florida Department of Environmental Protection, Office of Resilience and Coastal Protection by Florida Atlantic University. Funding was provided by the Florida Department of Environmental Protection Award No. B7C241. The views, statements, findings, conclusions and recommendations expressed herein are those of the authors and do not necessarily reflect the views of the State of Florida or any of its sub-agencies.

Acknowledgements

We appreciate the collaboration with Florida Department of Environmental Protection's Office of Resilience and Coastal Protection and Coral Reef Conservation Program (DEP CRCP) who supported this research. The Florida Fish and Wildlife Conservation Commission (FWC) has served as a key collaborator on this project. Ashley Carreiro, Elle Cohen, Allison Klein, Ian Combs, Matt Roy, and Jimmy Nelson at FAU Harbor Branch were also key contributors to this project. Michael Studivan, both while at FAU Harbor Branch and now at RSMAS/CIMAS, contributed to the experimental intervention. We thank DEP CRCP staff, particularly Kristi Kerrigan, Jenna Dilworth, and Jennifer Coley for coordinating this award. This study leveraged support from the NOAA OAR Omics program to JDV through CIMAS. Samples within St. Lucie Inlet Preserve State Park were collected under permit 01221915 to Joshua Voss. Coral samples collected outside of the park in the Southeast Florida Coral Ecosystem Conservation Area were collected under Special Activity Licenses SAL-19-1702-SRP and SAL-20-2022-SRP.

Table of Contents

1. BACKGROUND

Florida's coral reefs are currently experiencing a multi-year outbreak of a coral disease described as stony coral tissue loss disease (SCTLD). While disease outbreaks are not unprecedented, this event is unique due to the presence of multiple symptoms and etiologies that have affected at least 21 species of coral across Florida's Coral Reef. The disease is highly prevalent and estimated to have resulted in the mortality of millions of corals across the newly designated Southeast Florida Coral Reef Ecosystem Conservation Area (Coral ECA), Biscayne National Park (BNP), and the Florida Keys. Hurricane Irma also recently impacted the entire FRT in September 2017, with subsequent freshwater discharge impacts particularly acute on coral reefs in Martin County. The efforts reported here focus within the Southeast Florida Coral ECA as part of a larger effort to understand the impacts of disease on coral health and to determine mitigation efforts that may prevent losses of coral reef resources. As part of the coral population genetics objectives, this study leveraged funding from the NOAA Ocean Exploration and Research program to Joshua Voss and FAU Harbor Branch under award NA14OAR4320260 through the Cooperative Institute for Ocean Exploration, Research, and Technology.

1.1. Project Goals & Objectives

This project included multiple complementary approaches to understand, reduce, and mitigate coral reef ecosystem declines in SE FL. Continued monitoring of coral disease incidence and prevalence in the northern portion of the Coral ECA was coupled with experimental tests of intervention strategies designed to 1) reduce coral tissue loss, 2) reduce the likelihood of total colony mortality, 3) reduce the probability of transmission to nearby colonies, and 4) reduce population declines in known areas of infection. This project was designed to improve understanding of the current spatial extent of the disease outbreak, prevalence, species affected, and the physiological responses of corals to disease.

Five primary tasks were established for this period of performance:

- Task 1: Project coordination and permitting
- Task 2: SCTLD surveys and survivor reconnaissance
- Task 3: Disease intervention strategies
- Task 4: Coral population genetics to inform restoration activities and management strategies
- Task 5: Reporting

The outcomes of this project contribute to ongoing and future coral disease response efforts which seek to improve understanding of the severity of the coral disease outbreak and additional impacts to Florida's Coral Reef, identify management actions to remediate disease impacts, and, ultimately, prevent or mitigate the effects of future outbreaks. The project was designed with input from state and federal agency representatives and Martin County stakeholders to improve adaptive management regarding coral susceptibility to disease and impacts from infection. Finally, this project was designed to improve the predictive capacity regarding coral susceptibility to disease and impacts from infection.

2. METHODOLOGY

This project combined repeated surveys, 3D imaging, experimental disease intervention, and coral sampling to provide data on SCTLD dynamics, intervention success, and corals' physiological responses. Table 1 below summarizes the operational activities at each of the project sites. Project sites on St. Lucie Reef were chosen from long-term monitoring sites in our lab with over 10 years of survey data. SEFL sites in Palm Beach County with the highest stony coral cover were selected from a larger number of Hurricane Irma impact survey sites used in 2017 to allow for a continuous monitoring time series in these locations. Broward County sites were chosen due to their relatively high stony coral and SCTLD abundance.

This project was significantly affected by the COVID-19 pandemic. Operational diving activities and laboratory access were limited by various state, local, and university procedures. Only university designated essential personnel and essential projects were permitted from March 2020–August 2020. As a result, we focused available staff and field capability on fate tracking, experimental intervention, and gene expression efforts during this time. During COVID restrictions, SCTLD roving diver surveys were deprioritized at Martin and Palm Beach county sites where SCTLD prevalence was low or absent as of February 2020.

2.1. SCTLD Surveys and Reconnaissance

Four locations across the northern portion of Florida's Coral Reef were selected for coral health and disease surveys: St. Lucie Reef, Jupiter, Palm Beach, and Lauderdale-by-the-Sea (Fig 1). St. Lucie Reef is located in Martin County, Jupiter and Palm Beach are both located in Palm Beach County, and Lauderdale-by-the-Sea is located in Broward County. Following Hurricane Irma in September 2017, a rapid-response damage and disease survey effort was completed throughout Southeast Florida (Walker 2018). The resulting data from these initial surveys were used to inform decisions of which sites to target for continued monitoring and fate-tracking.

Roving diver disease surveys were conducted to assess the greatest reef area possible, quantifying disease prevalence over an estimated range of 100–2000 m^2 per survey based on conditions, principally underwater visibility. SCUBA divers swam for 20 min and recorded the species and disease status of every living coral colony ≥ 10cm in diameter. Paling, partial bleaching, and bleaching were also noted within surveys. From those data, SCTLD abundance and richness were calculated. Statistical tests were run in the R statistical environment.

To understand the current condition of St. Lucie Reef, we supplemented the roving diver surveys with 3D mosaic imaging over 10 m x10 m plots in several locations across SLR. Though our initial plan was to conduct reef-wide 3D mosaics via diver propulsion vehicles, consistent visibility limitations required a change in our survey design. As a result several individual 10m x 10m reef mosaics of SLR were created, but a unified map of the entire SLR reef system is not possible given the limitations of sea state, visibility, and current imaging technologies available to us.

Table 1. Operation activities at each project site. Superscripts in the Activity and Dates column indicate when each research activity was conducted.

Site Name	Lat	Long	Region	County	Activity	Dates
SLR Central	27° 07.900'	-80° 08.042'	St. Lucie	Martin	Surveys[1], P. astreoides sampling[2]	12/10/20[1], 2/5/21[2], 5/7/21[1]
SLR South	27° 07.286'	-80° 07.650'	St. Lucie	Martin	Surveys	12/10/20, 1/19/21, 5/7/21
SLR Ledge	27° 06.712'	-80° 07.531'	St. Lucie	Martin	Surveys	12/10/20, 1/19/21, 5/7/21
SEFL04	26° 56.6225'	-80° 1.3183'	Jupiter	Palm Beach	Surveys[1], Sponge Sampling[2]	9/10/20[12], 1/14/21[1], 3/19/21[1]
SEFL05	26° 55.6467'	-80° 1.8060'	Jupiter	Palm Beach	Surveys[1], Sponge Sampling[2]	9/10/20[12], 1/14/21[1], 3/19/21[1]
SEFL06	26° 53.8641'	-80° 0.9830'	Jupiter	Palm Beach	Surveys[1], Sponge Sampling[2], P. astreoides sampling[3]	9/10/20[12], 1/14/21[13], 3/19/21[1]
SEFL08	26° 42.6260'	-80° 0.9490'	West Palm Beach	Palm Beach	Surveys[1], Sponge Sampling[2]	9/2/20[12], 1/15/21[1], 3/19/21[1], 5/12/21[1]
SEFL11	26° 40.7100'	-80° 1.0950'	West Palm Beach	Palm Beach	Surveys[1], Sponge Sampling[2]	9/2/20[12], 1/15/21[1], 3/19/21[1], 5/12/21[1]
SEFL12	26° 39.1432'	-80° 1.2409'	West Palm Beach	Palm Beach	Surveys[1], Sponge Sampling[2], P. astreoides sampling[3]	9/2/20[12], 1/15/21[13], 3/19/21[1]
SEFL16	26° 31.4131'	-80° 1.9015'	Boynton	Palm Beach	P. astreoides and Sponge Sampling	1/21/2021
SEFL18	26° 29.6155'	-80° 2.2509'	Boynton	Palm Beach	Sponge Sampling	1/21/2021
SEFL20	26° 27.2298'	-80° 2.7642'	Boynton	Palm Beach	Sponge Sampling	1/21/2021
T328	26° 10.567'	-80° 05.633'	Lauderdale-by-the-Sea	Broward	Surveys[1], Interventions[2], Sponge Sampling[3], Intervention Follow-Up[4]	9/18/21[12], 12/11/20[13], 3/2/21[14]
BC1	26° 08.855'	-80° 05.766'	Lauderdale-by-the-Sea	Broward	Surveys[1], Interventions[2], Sponge Sampling[3], Intervention Follow-Up[4], P. astreoides Sampling[5]	9/18/21[12], 12/11/20[13], 3/2/21[145]
FTL4	26° 08.197'	-80° 05.843'	Lauderdale-by-the-Sea	Broward	Surveys[1], Interventions[2], Sponge Sampling[3], Intervention Follow-Up[4]	9/18/21[12], 12/11/20[13], 3/2/21[14]

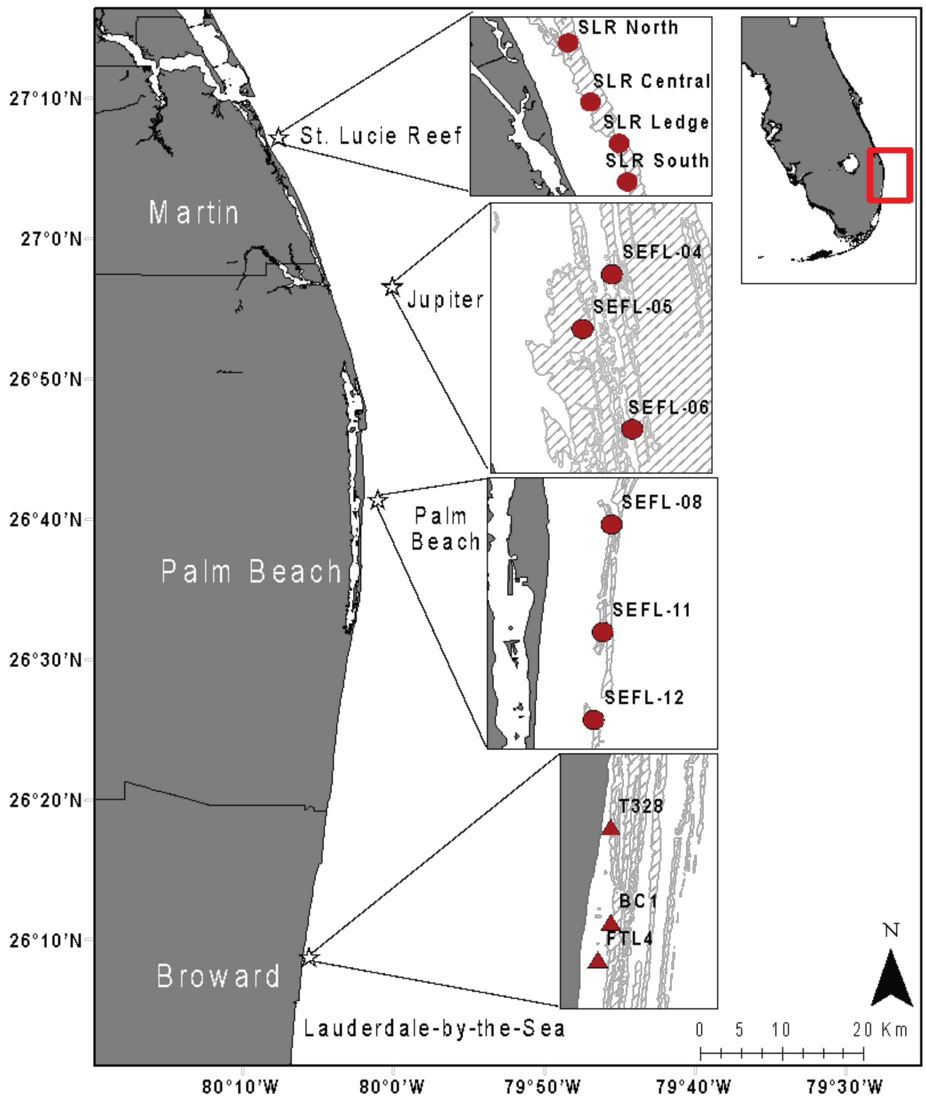
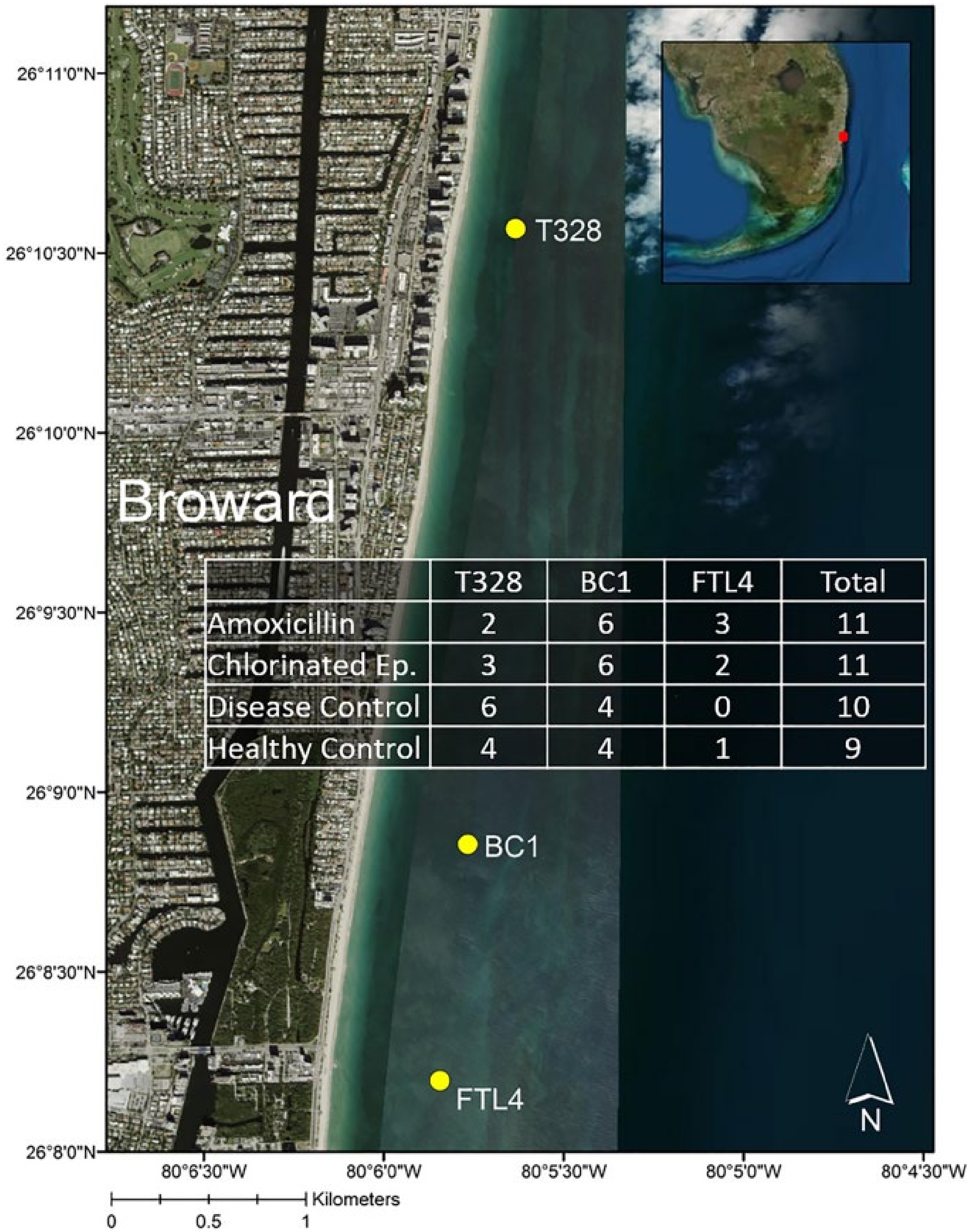


Figure 1. Map of study locations throughout the northern portion of Florida's Coral Reef. Red circles indicate roving diver survey sites and red triangles indicate sites where both roving diver surveys and the SCTLD intervention treatment experiment occurred.

2.2. Experimental Disease Intervention Strategies

We experimentally assessed the effectiveness of two intervention treatments on SCTLD-affected *Montastraea cavernosa* colonies in situ. Colonies were tagged and divided into three treatment groups: (1) chlorinated epoxy, (2) amoxicillin combined with CoreRx/Ocean Alchemists Base 2B, and (3) untreated controls (Figures 2, 3). For complete details of the experimental design, see Shilling et al. 2021 and the 2020 final report to DEP from Voss et al. The experimental colonies were monitored periodically over 11 months to assess treatment effectiveness by tracking lesion development and overall disease status. This monitoring has continued through this period of performance. Videos were recorded for 3D model generation to measure colony sizes and tissue loss over time. Stills were extracted from the videos using the software FFmpeg, and models were generated through a four-step process using the software Agisoft Metashape. All tracing and quantifying of tissue areas from coral colony models was conducted in the application software Rhinoceros 3D. Models and tissue areas were only generated for initially SCTLD-affected coral colonies in this experiment.



	T328	BC1	FTL4	Total
Amoxicillin	2	6	3	11
Chlorinated Ep.	3	6	2	11
Disease Control	6	4	0	10
Healthy Control	4	4	1	9

Figure 2. Satellite image of Broward County coast with sites utilized in this experiment denoted with yellow dots. The table lists sample sizes in each treatment group at each site, as well as totals.

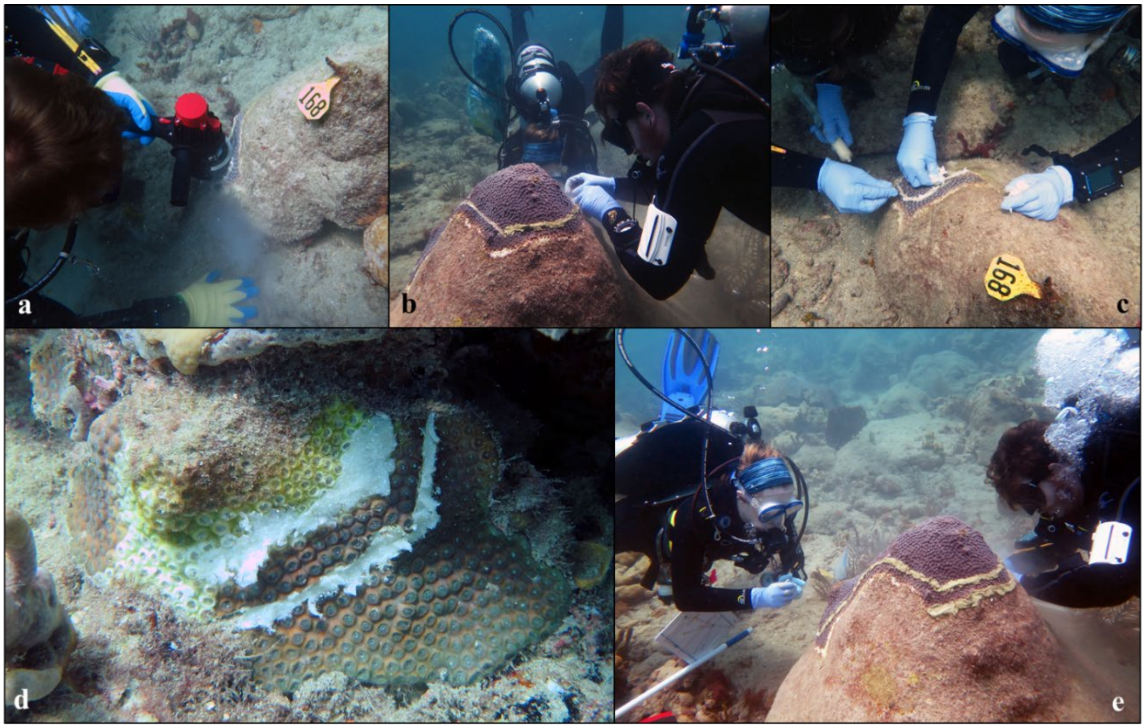


Figure 3. Process of treating SCTLD-affected coral colonies in situ. (a) Diver creating a trench around the SCTLD lesion using an angle grinder. (b) Filling a trench with chlorinated epoxy treatment. (c) Filling a trench with a Base 2B plus amoxicillin mixture. (d) A SCTLD-affected coral colony that has been treated with Base 2B plus amoxicillin mixture. (e) A SCTLD-affected coral colony partially treated with the chlorinated epoxy.

2.3. Coral and Sponge Population Genetics

During this period of performance, we focused on analysis of previous collected *Montastraea cavernosa* samples and collection/analyses of additional samples of *Porites astreoides* and *Xestospongia muta* (Table 1). For all these samples, ~5 cm^2 tissue fragments were collected and preserved in either Trizol or Zymo DNA/RNA Shield. The samples were extracted using a modified dispersion buffer/phenol–chloroform-isoamyl alcohol extraction and cleaned using the Zymo DNA Clean and Concentrator Kit. DNA extracts were digested with BcgI enzyme and 2bRAD libraries were prepared following Wang et al. (2012) including some modifications to optimize the libraries. Notably, 12 uniquely indexed 3' adaptors were incorporated, allowing 12 sample ligations to be pooled prior to amplification. Fully degenerate 5' adapters were also included, allowing PCR duplicate removal from downstream analyses. Additionally, triplicate libraries were prepared for three samples and used as a sequencing quality check and to identify natural clones. Sequencing was conducted on an Illumina NovaSeqS1 flow cell at the University of Texas at Austin's Genome Sequencing and Analysis Facility.

2.4. QA/QC

All roving diver, fate tracking, and intervention experiment data were entered into Access or Excel where QA/QC and data summaries were performed. Once entered, data were reviewed to ensure consistency with data sheets. During the summary table creation, the data were once again reviewed for consistency between teams especially for coral species and disease identifications. Intervention activities were also uploaded to the FWRI Intervention Dashboard. In some cases, site pictures were reviewed to help this QA/QC process.

3. RESULTS

3.1. SCTLD Surveys and Reconnaissance

SCTLD prevalence from June 2020 to June 2021 was relatively low at our St. Lucie Reef, Jupiter, and Palm Beach sites (all less than 0.4%, Figure 4). However, consistent levels of SCTLD were observed at our Lauderdale-by-the-Sea sites (mean 2.9%-3.7%), and these were statistically greater than any other sites. Nonetheless, the SCTLD prevalence levels observed in June 2020 to May 2021 were significantly lower than levels observed in previous years at LBTS (PERMANOVA; $p < 0.01$, Figure 5). SCTLD prevalence was significantly different between locations (PERMANOVA; Pseudo-$F = 3.1_{3,24}$, $p < 0.01$) and over time (PERMANOVA; Psuedo-$F = 3.41_{5,24}$, $p < 0.02$, Figure 4). Pairwise comparisons indicated that SCTLD prevalence in LBTS was significantly greater than in any other site in June 2020 to May 2021.

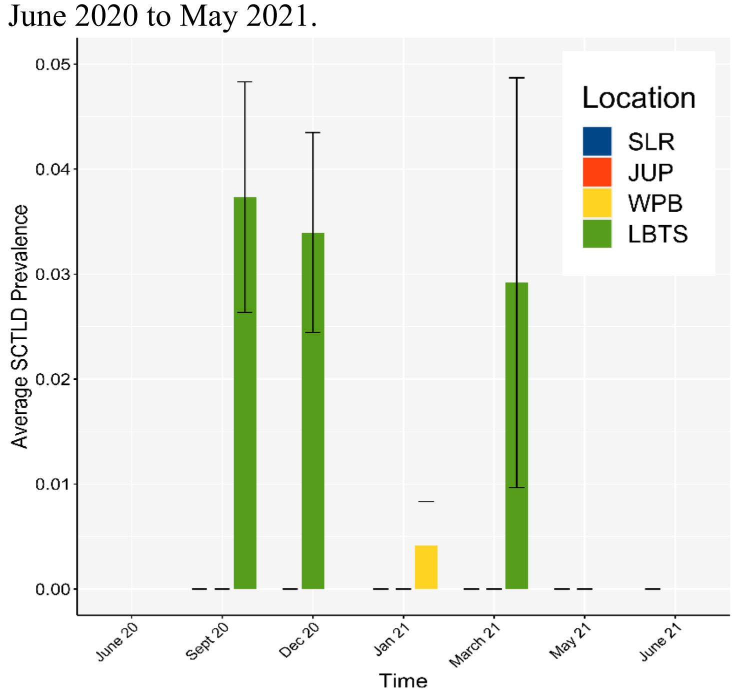


Figure 4. Mean SCTLD prevalence ± SD from roving diver surveys at St. Lucie Reef (SLR), Jupiter (JUP), West Palm Beach Breakers (WPB), and Lauderdale-by-the-Sea/Pompano (LBTS). 2020-2021. Note no surveys were completed in June 2020 due to COVID-19.

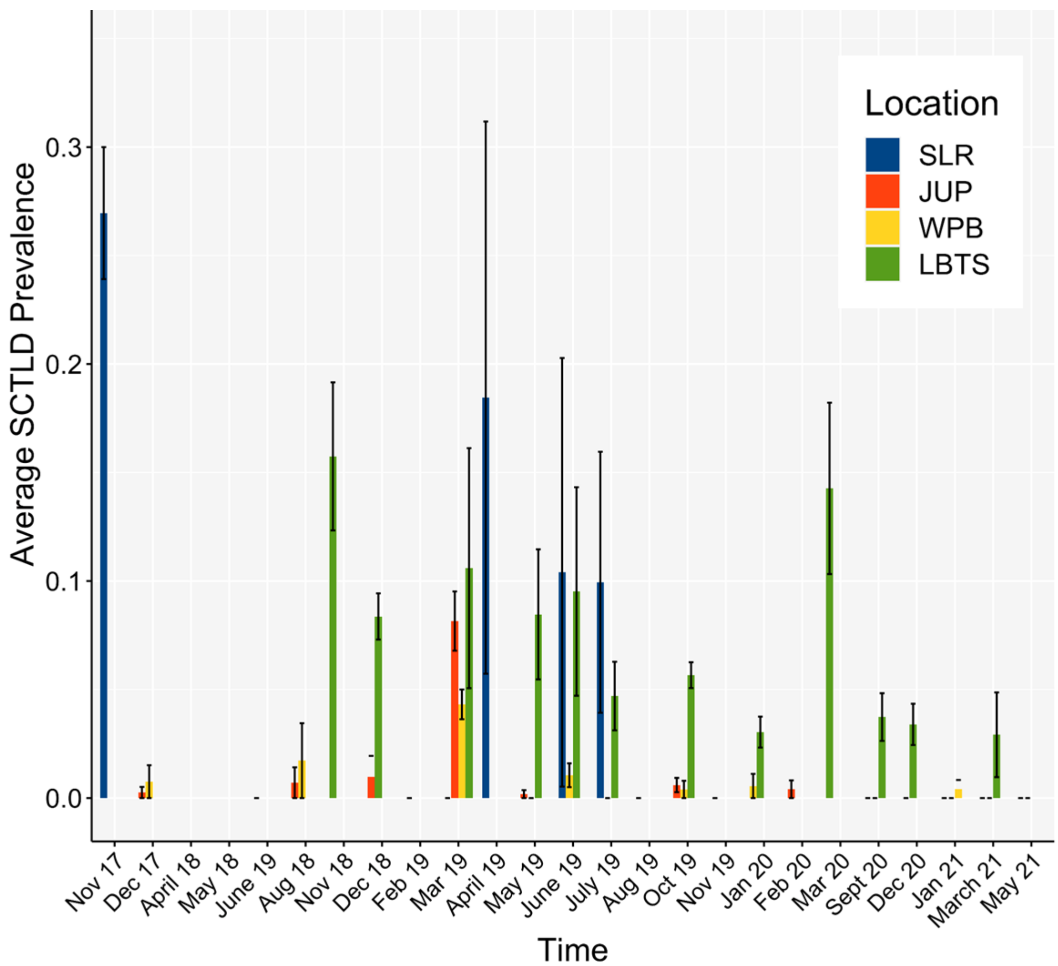


Figure 5. Mean SCTLD prevalence ± SD from roving diver surveys at St. Lucie Reef (SLR), Jupiter (JUP), West Palm Beach Breakers (WPB), and Lauderdale-by-the-Sea/Pompano (LBTS). 2017-2021

3.2. Experimental Disease Intervention Strategies

In this 2020-2021 we treated 20 additional coral colonies with amoxicillin plus Base 2b and tracked 41 colonies from previous efforts. All healthy control *M. cavernosa* colonies remained healthy throughout the course of the intervention experiment, and therefore were not included in the following analyses. Research site location had no significant effect on the development of new lesions on a colony over time, cumulative new lesion development over 46 weeks, the total lesions present on a colony at any time point, nor SCTLD status of a colony at any time point (Kruskal-Wallis tests, all $p > 0.05$). There was no significant difference between the initial numbers of lesions on experimental colonies between sites or treatment groups (Kruskal-Wallis tests, all $p > 0.05$).

Amoxicillin plus Base 2B treated corals were successful with 95% of treated lesions remaining inactive at the end of the study. The success of amoxicillin treated lesions was significantly higher than untreated lesions and chlorine treated lesions (Fisher's exact test,

$p < 0.001$; pairwise Fisher's test, all $p < 0.001$; Figure 6). There was no significant difference detected between the quiescence rates of chlorine and untreated lesions at the end of the experiment. Treatment significantly influenced the SCTLD status of a colony until the 46-week time point (Fisher's tests). From the first monitoring at three weeks to the third monitoring at 9 weeks, the amoxicillin treated colonies were more likely to be completely quiesced than the chlorine treated or untreated colonies.

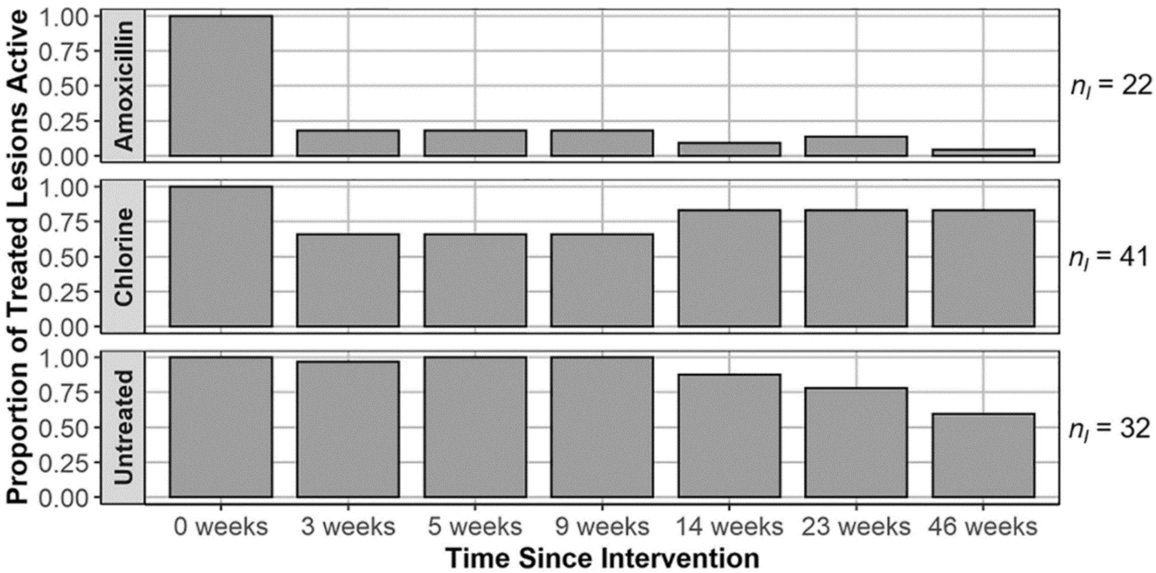


Figure 6. Status of initially treated disease lesions on colonies by treatment group at each monitoring event, shown in proportions of total, with n_l indicating total number of lesions present across all colonies in the treatment group. "Untreated" refers to untreated SCTLD-affected control colonies.

3.3. Coral Population Genetics

Genotyping profiles for *M. cavernosa* developed through analyses of single-nucleotide polymorphisms generated by 2bRAD sequencing demonstrated distinct populations throughout Florida's Coral Reef. The most likely number of populations estimated by the Evanno and Puechmaille methods from admixture models generated with NGSAdmix was K=4. While *M. cavernosa* populations in SE FL were relatively distinct from those in the Florida Keys, there was surprising similarity between populations in SE FL and those in shallow areas of the Dry Tortugas South TER (aka Riley's Hump) indicated by the darker blue profiles in the admixture compositions for each colony (Figure 7). Likewise, corals in Jupiter demonstrated moderate similarity to some of the corals sampled in the shallow Lower Keys as indicated by the presence of aqua-dominated individuals in each location. The unexpected patterns suggest that local biodiversity maintenance and future plans for restoration activities should take this heterogeneity into account.

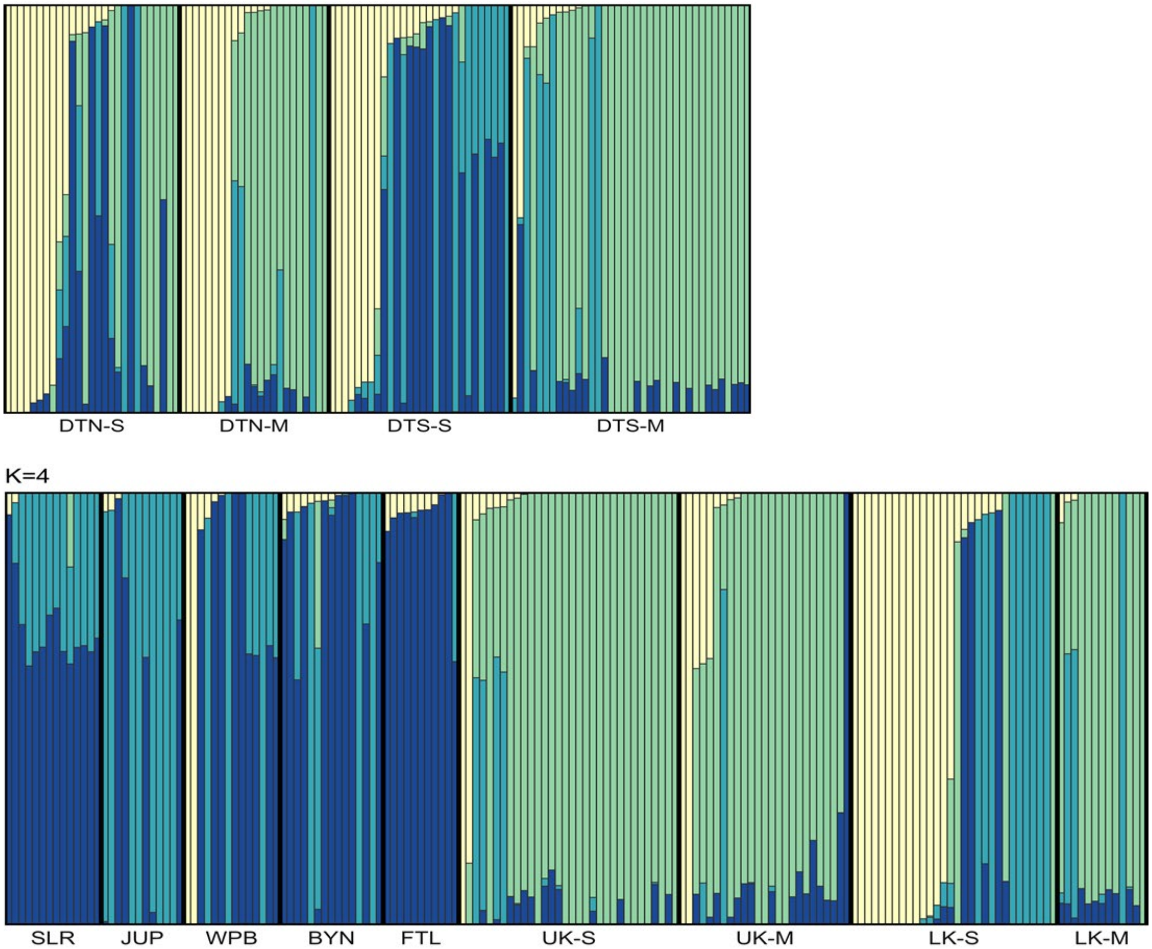


Figure 7. Population structure models for *M. cavernosa* generated via admixture analysis conducted on genotype likelihoods with the program NGSAdmix. The K=4 populations are indicated by different colors (yellow, green, aqua, dark blue). Each bar represents a single coral colony and the proportion of each color with a single bar represents the likely admixture contributions of each population to each coral. Site abbreviations are: St. Lucie Reef (SLR), Jupiter (JUP), West Palm Beach (WPB), Boynton (BYN), Ft. Lauderdale (FTL), Upper Keys Shallow and Mesophotic (UK-S, UK-M), Lower Keys Shallow and Mesophotic (LK-S, LK-M), Dry Tortugas North Shallow and Mesophotic (DTN-S, DTN-M), Dry Tortugas South Shallow and Mesophotic (DTS-S, DTS-M).

Samples for both *P. astreoides* and *X. muta* were collected across sites in SE FL from June 2020 through May 2021. For both species 2bRAD sequencing was completed and for *X. muta* subsequent data analyses have been completed. Populations of *X. muta* in SE FL appear to be comprised of two primary populations, one found in Jupiter, Palm Beach, and Boyton, and the other in Ft. Lauderdale (Figure 8). These data will be combined with samples and data collected in the Florida Keys NMS to resolve spatial patterns throughout Florida's Coral Reef.

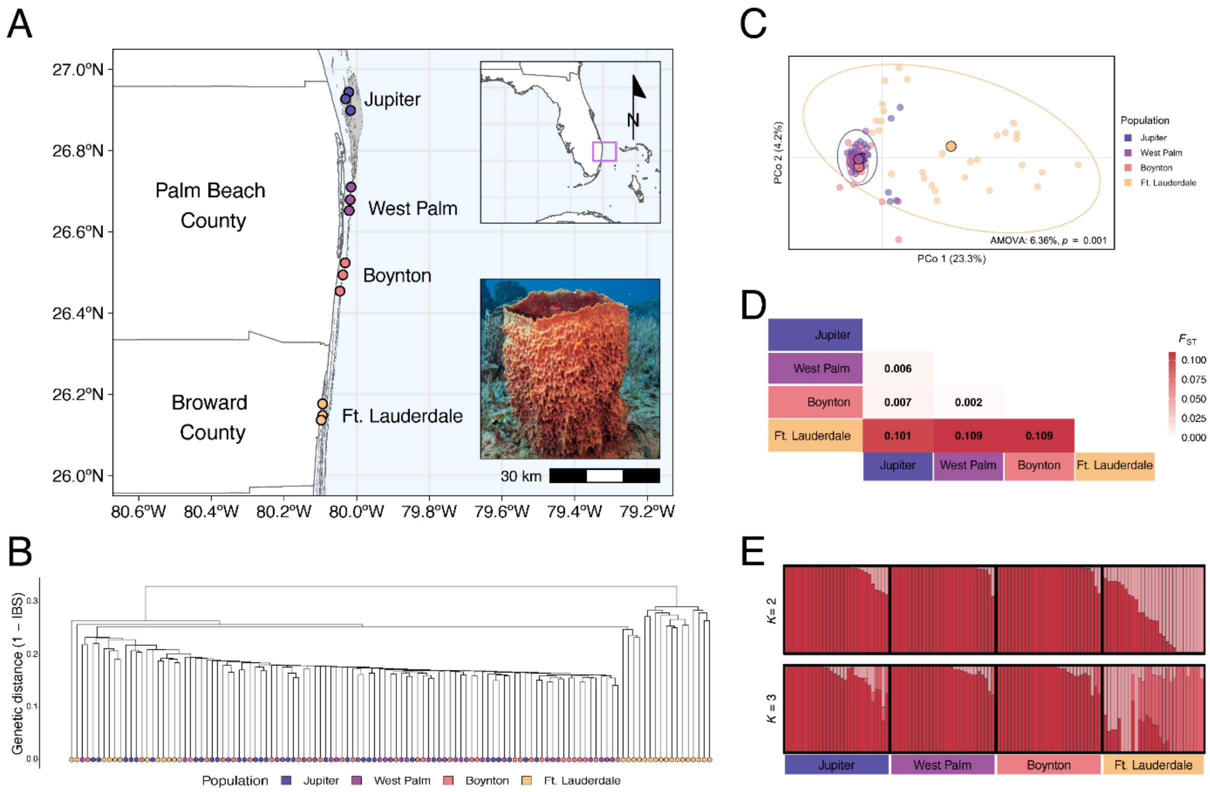


Figure 8. *Xestospongia muta* population genetic analyses. A. *X. muta* sampling sites across southeast Florida. B. Cluster dendrogram of *X. muta* samples based on pairwise Identity by State (IBS) matrix. C. Principal coordinates analysis of *X. muta* samples based on pairwise IBS matrix, prediction ellipses assume a multivariate t-distribution. D. Heatmap of *X. muta* population differentiation based on pairwise populations fixation indices (FST). E. Southeast Florida *X. muta* population genetic structure models for optimal number of genetic clusters (K = 2, K = 3).

4. PRELIMINARY CONCLUSIONS

This study demonstrated that tissue loss disease incidence and prevalence may be highly variable over space and time on coral reefs in SE FL. For example, stony coral tissue loss disease (SCTLD) was observed continually throughout the project period among corals at our Lauderdale-by-the-Sea sites, while for June 2020–May 2021 SCTLD was essentially absent in the Palm Beach and Martin County sites.

Previously, we hypothesized that St. Lucie Reef may have been buffered from tissue loss impacts by 1) relative distance from other infected coral communities, and/or 2) stress hardened coral colonies resistant to disease. However, the previous observation of high disease prevalence and 85% losses of coral colonies counter these hopeful hypotheses. The losses at St. Lucie Reef cannot be attributed to disease impacts alone. Impacts from Hurricane Irma and subsequent discharges from the St. Lucie Estuary were also critical drivers that contributed to a severe multiple stressors scenario. The temporal confounding of these events makes interpretation of the proximal causes of coral loss difficult.

Subsequent monitoring throughout this period of performance revealed that SCTLD was no longer present at St. Lucie Reef. However, this appears to be a function of lack of susceptible hosts rather than any improvement in STLD status overall. No apparent natural recovery is occurring in the *M. cavernosa* or *Pseudodiploria clivosa* populations on SLR. Therefore, this site was selected as a location for the Restoration Team Trials experiment in collaboration with FWC and DEP. In contrast, *P. astreroides* populations continue to expand.

In our *in situ* SCTLD intervention experiment, the Base 2B plus amoxicillin treatment was significantly more effective at treating individual SCTLD lesions on *M. cavernosa* colonies than the chlorinated epoxy or leaving the colonies untreated. This study supports and reinforces other reports of successful antibiotic application for the treatment of coral disease (e.g. Neely, Walker). The chlorinated epoxy in this experiment was ineffective as a treatment for SCTLD lesions on *M. cavernosa.* In instances where time and effort underwater are constrained, application of Base 2B plus amoxicillin to more SCTLD affected colonies should be prioritized over supplementing the antibiotic treatments with trenching. However, a controlled experiment comparing trenching and Base 2B plus amoxicillin treatments versus Base 2B plus amoxicillin alone is recommended to further support these observations and assess the relative risks and trade-offs associated with mechanical trenching.

The success of Base 2B plus amoxicillin treatments is encouraging in the face of a disease outbreak that is continuing to devastate Caribbean coral reefs. However, potential secondary impacts of amoxicillin treatments on SCTLD-affected corals remain uncharacterized. We recommend that future research efforts should focus on assessing the potential unintended consequences of antibiotic treatments on corals, their microbial communities (including Symbiodiniaceae), and neighboring organisms. Additionally, further efforts are needed to optimize dosing and delivery methods for antibiotic treatments on SCTLD-affected corals and scale up intervention treatments effectively.

Finally, our population genetics assessments to date indicate that *M. cavernosa* demonstrated a distinct regional population structure with some very interesting links between the shallow Dry Tortugas and Lower Keys to reefs in SE FL. These genetic differences should be strongly considered when developing coral restoration strategies for Florida. To maintain biodiversity of this species, these individual populations should all be protected and individually targeted for conservation and restoration approaches. In contrast there appears to be less diversity among *X. muta* populations, but significant regional population structure is also found in this species. Ongoing analyses of *X. muta* and *Stephanocoenia intersepta* from the Florida Keys National Marine Sanctuary, *P. astreoides* in SE FL, and comparative analyses to samples collected in Cuba, Belize, Mexico, Pulley Ridge, and the Flower Garden Banks NMS will quantify the level of genetic connectivity across the wider Gulf of Mexico region and help to determine the possibility of regional refuges or restoration actions.

5. RECOMMENDATIONS

Recommendation 1*: Prioritize disease mitigation/intervention efforts to reduce losses of key coral reef ecosystem components.* Base 2B plus amoxicillin demonstrated success against SCTLD lesions on *M. cavernosa* with a 95% success rate. We recommend expanding treatment research activities and intervention efforts to protect existing coral tissue/cover, particularly in Lauderdale-by-the-Sea, the Lower Keys, and Dry Tortugas.

Recommendation 2*: Develop strategic capabilities and teams to respond to new likely areas of SCTLD (e.g. Tortugas, Cuba) and assess risk at remote reef ecosystems (e.g. Alacranes, FGBNMS).* Rapid response could be the key to prevent SCTLD epidemics. In Florida SCTLD took hold before we had a viable, effective treatment option. But now that we do, responders properly trained and equipped to conduct amoxicillin interventions could make a very significant impact on coral ecosystems that show early phase SCTLD levels.

Recommendation 3*: Evaluate the role of water quality in SCTLD dynamics.* Several coral diseases are known to be exacerbated by nutrient pollution. Correlative data also suggests that SCTLD prevalence in SE FL may be related to septic system abundance. Controlled experiments to assess the role of nutrient pollution or other changes in water quality and needed for SCTLD.

Recommendation 4*: Determine impacts of Base 2B plus amoxicillin treatments.* Since Base 2B plus amoxicillin appears to be the only treatment currently identified as effective in the field through controlled, year-long experiments (see above), we recommend continued use of this approach. However, both ethical and regulatory issues require that we systematically characterize the potential impacts of antibiotic treatments on the host coral, its algal symbionts, its microbiome, the microbiomes and macro organisms in the surrounding area, and the relative abundance and expression of antibiotic resistance genes.

Recommendation 5: *Advance coral conservation initiatives with support from Magnuson-Stevens Act and implement actions/regulations for the Southeast Florida Coral Reef Ecosystem Conservation Area. Develop and implement a management plan.* The threat posed to Florida's coral reefs by the tissue loss disease are severe. Any additional efforts to reduce stressors or known impacts to coral reef communities should be implemented to enhance the likelihood of coral resilience and recovery, particularly with respect to water quality. Furthermore, efforts to develop more robust coral restoration programs should include research toward sexual propagation, ex situ and in situ nurseries, subsequent outplanting, and testing of outplant resilience to SCTLD.

Recommendation 6: *To support effective management for coral reef populations and communities in Florida, additional information on population connectivity and source-sink dynamics is needed.* After severe disturbance events like the SCTLD outbreak, allocated effort/ resources to particular regions should be based on predicted coral recruitment and recovery. Likewise, effective coral restoration strategies will require knowledge of genetic stocks among various coral populations to design effective restoration strategies.