# 2021-2022 SE FL ECA Reef-building-coral Disease Intervention and Preparation for Restoration

# **Final Report**



Florida Department of Environmental Protection Coral Protection and Restoration Program



## 2021-2022 SE FL ECA Reef-building-coral Disease Intervention and Preparation for Restoration

Final Report

Prepared By:

Brian K. Walker, Hunter Noren, Reagan Sharkey, and Samantha Buckley,

Nova Southeastern University Halmos College of Arts and Sciences 8000 N. Ocean Drive Dania Beach, FL 33004-3078

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## List of Acronyms

FAU	Florida Atlantic University Harbor Branch Oceanographic Institute
DEP	Florida Department of Environmental Protection
ORCP	Office of Resilience and Coastal Protection
FWC	Florida Fish and Wildlife Conservation Commission
NSU	Nova Southeastern University
Coral ECA	Kristin Jacobs Coral Reef Ecosystem Conservation Area
FCR	Florida's Coral Reef
SCTLD	stony coral tissue loss disease
SE FL	Southeast Florida
NOAA	National Oceanic and Atmospheric Administration
NCRMP	National Coral Reef Monitoring Program
CIMAS	Cooperative Institute For Marine And Atmospheric Studies
GIS	Geographic Information System

#### Acknowledgements

Thank you to the Florida Department of Environmental Protection's Coral Protection and Restoration Program (DEP CPR) and NOAA CRCP for supporting these efforts. We thank the Florida Coral Disease Advisory Committee for the large number of volunteers assisting in the meeting and planning of coral disease efforts. We thank Lisa Gregg for assisting with permitting. Thanks to the DEP CPR staff including Kristi Kerrigan for contract and report-review coordination. Thanks to Miami-Dade Regulatory & Economic Resources for field assistance and boat time. Thank you to Katy Toth, Sasha Wheeler, Zach Graff, Allie Kozachuk, Amanda Zummo, Alex Wagner, Elizabeth Fromuth, Kristin Anderson, Thomas Ingalls, Alysha Brunelle, and Brooke Enright at the NSU GIS and Spatial Ecology lab.

#### Management Summary (300 words or less)

Coral diseases have caused enormous impacts to coral populations globally perpetuating the rapid need for large-scale coral reef restoration. SCTLD exemplifies that coral diseases are more devastating than ever before. However, implementing recent disease intervention techniques in the restoration toolbox reduces the necessity for costly and time-consuming post hoc restoration techniques.

Our work shows that SCTLD is still prevalent in the region, interventions are still needed, and these efforts are saving corals. This year, interventions helped maintain species diversity and ecosystem services within the Coral ECA and kept alive some of the largest and oldest animals in Florida. Monthly monitoring and treatments have reduced the loss of live tissue area and provided valuable information on the temporal and spatial variations of colonies with lesions. Corals not regularly treated with disease interventions have had drastic declines in live tissue area. Regular disease intervention is restoring colony health and saving the large priority corals, preempting post hoc restoration. For example, the current live tissue area of all previously treated large corals is 357.8 m<sup>2</sup>. This is the equivalent to the area of 2 volleyball courts of live coral tissue.

Our reconnaissance for coral restoration sites identified eleven sites with high densities of corals remaining, nineteen sites that had drastic reductions in coral density and/or richness after hurricane Irma and three years of SCTLD, and nine new large colonies of high value to the State of Florida.

The monthly monitoring data have been invaluable in linking environmental drivers to disease dynamics. Higher water temperatures, amounts of water following out of the inlets, and rainfall account for 56.4% of the temporal variability in the number of new lesions. Understanding these dynamics can help with water management strategies. Caution should be taken when choosing coral restoration sites as to avoid areas more prone to disease.

#### **Executive Summary**

SCTLD was first discovered in the Kristin Jacobs Coral ECA in 2014 and remains present in the region. As of November 19, 2021, a total of 1,218 colonies were treated by the coral disease interventions strike teams in the Coral ECA since 2018 (including the experimental sites, but not the large priority corals in section 4.2) totaling 802 meters of treatment. Between July 1 and May 31, 2022, 320 colonies were treated. The amount and species of corals treated in broad scale recon surveys indicates that there are still some rare survivors of the highly susceptible brain coral species in the area and some are still succumbing to the disease.

Priority corals are still requiring monthly visits to ensure their survival. Of the 107 monitored corals between July 1 and May 31, 2022, a total of 189 antibiotic ointment treatments were conducted on 47 colonies. Twenty-five treatments failed, equating to 87.6% effectiveness on lesions and 86.9% effectiveness on coral colonies. The number of new infections varied throughout the period. The months with the highest treatments were

August, October, and January. September 2021 was unusually low (1) from previous Septembers. Monthly visits should continue to maximize treatment success and capture new infection frequencies.

During this reporting period, NSU divers conducted a total of 86 recon and intervention dives over 27 dive days. Four newly identified recon sites: a new high density *Montastraea cavernosa* site (BS3), two rare large (over 5.8 m diameter) *Orbicella annularis* colonies, and one previously unknown large healthy *Orbicella faveolata* (~3.6 m diameter).

A desktop analysis resulted in nineteen sites with previously high coral richness and/or density that were severely impacted by hurricane Irma and disease in 2017. Out of the 233 2020 NCRMP dives and 277 strike team dives completed during this project, thirty sites were identified that contained unique or special existing corals. These sites should be recognized as candidates for future restoration activities to restore coral densities and richness to previous amounts.

This period NSU assisted the Smithsonian Marine Station at Fort Pierce with probiotics treatments at a new site, BS3. NSU also assisted Dr. Joshua Voss' lab at FAU Harbor Branch Oceanographic Institute with investigating the impact of elevated nutrient levels on the spread and progression of SCTLD in *M. cavernosa*.

The large coral new treatment data collected herein and previously were being summarized to evaluate statistical relationships to environmental predictors funded by DEP PO B9CAF9. They will also be used to evaluate the outcomes of the SCTLD Resistance Research Consortium laboratory analyses funded by DEP B8A48D and a newly awarded EPA grant (SF02D21722).

#### 1. BACKGROUND

Florida's Coral Reef is currently experiencing a multi-year disease-related mortality event, that has resulted in massive die-offs in multiple coral species. Approximately 21 species of coral, including both Endangered Species Act-listed and the primary reef-building species, have displayed tissue loss lesions which often result in whole colony mortality. First observed near Virginia Key in late 2014, the disease has since spread to the northernmost extent of Florida's Coral Reef, and southwest through the Dry Tortugas in the Lower Florida Keys. The best available information indicates that the disease outbreak is continuing to spread west and throughout the Caribbean.

In 2021-22 DEP funded award B96800 to continue Coral ECA coral disease intervention and restoration activities ongoing since 2018 (DEP PO# B2A150, B48140, B46AD7, B3C3AD, B558F2, B7B6F3) with the expectation of supporting existing collaborations and adapting to new methodologies to improve intervention success in the Coral ECA. These actions included maintaining the monitoring and continued treatment of 100 priority large corals, conducting broadscale strike team reconnaissance and disease interventions, further testing of permitted intervention techniques and materials, and the identification of unique colonies and sites (e.g. location of remaining highly susceptible species, remaining high coral density sites, restoration candidate sites). Information from these previously funded activities has strengthened local partnerships and provided data on treatment effectiveness, saved the largest large colonies from extreme tissue loss, and facilitated probiotics testing. It also provided data on tissue loss rates, survivor sites, new infection rates through time, and classifying large corals into categories based on infection rates setting up the study design for the stony coral tissue loss disease (SCTLD) Resistance Research Consortium.

This report summarizes the progress from our continued Coral ECA coral disease interventions through May 31, 2022, including the monitoring and continued treatment of the priority large corals, broadscale strike team reconnaissance and disease interventions, field activities for initial probiotics testing, and the identification of unique coral disease survivor sites.

### 2. PROJECT DESCRIPTION

One goal of this project is to perform disease interventions on the remaining reef-building coral species with active disease in the Coral ECA. These activities are essential to saving the remaining corals in SE FL affected by disease that have the potential of recovering and building new reef structure. Coral disease intervention treatments includes smothering diseased tissue, creating a disease-break to arrest disease progression, and covering the newly exposed skeleton with an antibiotic paste. The first objective is to apply these interventions to the 100 priority large corals as necessary to maintain their health and continue monthly monitoring. The second objective is to conduct broader-scale strike team disease intervention efforts in partnership with Smithsonian, FAU, DEP, Broward County, and Miami-Dade County to help save diseased colonies throughout the Coral ECA. A third objective is to further field test new permitted intervention techniques and materials (e.g.

Smithsonian probiotics) including whole colony treatments as they are conceived, developed, and permitted.

The third objective of this project was to collect information to inform and aid in planning future Coral ECA restoration efforts. Future restoration will need to identify survivors for sexual reproduction, genetic analyses, and experimentation on stress hardening and disease resistance. Therefore, identified unique coral disease survivor sites to make restoration activities more efficient. This work is partially funded by a NOAA Coral Program grant (NA19NOS4820127).

The findings of this project are being incorporated into the on-going coral disease response effort which seeks to improve understanding about the scale and severity of the Florida's Coral Reef coral disease outbreak, identify primary and secondary causes, identify management actions to remediate disease impacts, restore affected resources and, ultimately, prevent future outbreaks. As such, collaboration amongst partners and the Disease Advisory Committee (DAC) is essential to avoid duplication, share lessons learned, and ensure alignment of needs.

#### 3. METHODOLOGY

The antibiotic paste treatments were conducted under the State of Florida Special Activity License Permit SAL-21-2022-SRP which authorized the cutting of disease-breaks and the application of disease treatments containing amoxicillin and chlorine. The probiotics work was permitted under the State of Florida Special Activity License Permit SAL-22-2201-SRP.

#### 3.1. Broad-scale Coral Disease Intervention Strike Team (Task 2)

Southeast Florida coral disease intervention strike teams, consisting of personnel from NSU, Broward County, and Miami-Dade County, conducted disease intervention at various sites throughout both counties. Intervention sites were chosen based on previous information on the locations of diseased corals and high priority county sites. The NSU efforts discussed in this report targeted locations between Hillsboro Inlet and Biscayne National Park while avoiding known existing monitoring stations and experimental sites. At each location, divers towed a GPS buoy synced to a dive computer. Once a diseased coral was located, the time was taken from the dive computer. Each coral was tagged and measured, photographed, and treated. GPS coordinates were loaded into ArcGIS and the locations that corresponded to each time recorded during treatment were copied into a GIS shapefile. All treated coral locations were supplied to FWC for inclusion in the Coral Disease Intervention Dashboard:

https://novasoutheastern.maps.arcgis.com/apps/opsdashboard/index.html#/55a759f02f3c4 86eb1d29a95f80fba0a.

#### 3.2. Apply Interventions to 100 Large Corals (Task 3)

The largest known corals in the region were prioritized for monitoring and disease interventions. From September 2018 – June 2019, approximately 60 corals began being

monitored and treated on a monthly basis. This increased to 90 colonies in July 2019, but it took several periods to establish all 90. As of October 2021, there were a total of 107 corals in the priority database. All priority corals were photographed monthly and visually assessed by a diver estimating the percentage of live tissue, diseased tissue, bleached tissue, recent mortality, and old mortality. If SCTLD was found, the lesion was treated with antibiotic paste. All margins were treated with the Ocean Alchemists antibiotic ointment CoreRx B2B with amoxicillin (1:8 ratio by weight). Photographs were taken of all areas before treatment at both the 0.5 m standard distance and wider scenes. The length of each treatment was estimated using a standardized scale in the photographs. Lesion treatments were determined failures if the active disease continued progressing past the treatment failures. All prioritized colonies were tagged with a yellow tag with a unique number and instructions to photograph the coral and submit the photo to <u>www.SEAFAN.net/tags</u> (Figure 1).

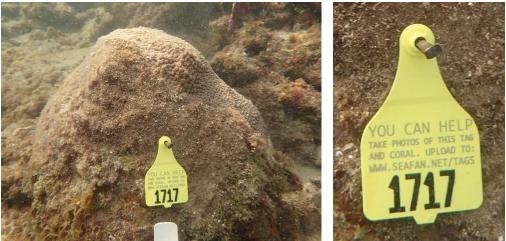


Figure 1. Example of coral tag placed on or next to each treated coral.

#### 3.3. Recon Sites (Task 4)

This report section summarizes all information gathered to date through our reconnaissance efforts useful to inform coral restoration in the Coral ECA to date. We conducted recon tasks with our DEP disease intervention strike team and NOAA NCRMP work to collect information more efficiently and cost effectively. Reconnaissance efforts were partially funded by NOAA Coral Program Award NA19NOS4820127. Disease intervention strike team operations were conducted throughout the project period in association with funded work through DEP (POs: B7B6F3 and B96800). NCRMP fish and benthic survey data were collected regionally funded by NOAA through CIMAS (NA15OAR4320064 and NA20OAR4320472).

A desktop study of previous datasets was performed that identified historic sites of high coral density and/or richness. Locations from 2017 post-Irma surveys (Walker 2018) where coral density and/or richness were drastically reduced were identified and compiled.

Concomitantly, reconnaissance throughout Broward and Miami-Dade was conducted by coral disease response strike teams to identify corals and/or sites that respond better to treatments or that have resisted infection to-date and to identify unique coral disease survivor sites. Recon was performed during strike team disease interventions, NCRMP activities, and other opportunistic times. Divers visited sites and haphazardly searched the area towing a GPS buoy to find locations of visually noticeable high coral density or richness and to identify if disease is present. If disease was found, these locations were treated and mapped. Large colonies of any species and smaller colonies of the species hit hardest by SCTLD (e.g. *M. meandrites, E. fastigiata, D. stoksii, C. natans, D. cylindrus, D. labyrinthiformis*) were mapped.

# 3.4. Field Test New Permitted Intervention Techniques and Materials (Task 5)

#### 3.4.1. Probiotics on Montastraea cavernosa

The Smithsonian Marine Station at Fort Pierce is developing several probiotics and several treatment methods to be tested on *M. cavernosa*. We previously set up two field experiment sites for testing and monitoring the corals' responses in 2019 and 2020. See Walker et al. (2021) for details on those sites.

During this project period, a third experimental site location (BS3) located on the 1<sup>st</sup> reef ledge just offshore from Commercial Pier was identified and forwarded to our partners at the Smithsonian for testing of their probiotic interventions (Figure 2).

#### 3.4.2. Nutrient enrichment on Montastraea cavernosa

The Voss lab at FAU Harbor Branch are investigating the impact of elevated nutrient levels on SCTLD spread, progression, and impact on changes in microbial communities in *M. cavernosa* found on nearshore reefs in Southeast Florida. NSU helped to identify a field experiment site for FAU, as well as contributed to boating and dive operations (Figure 3).

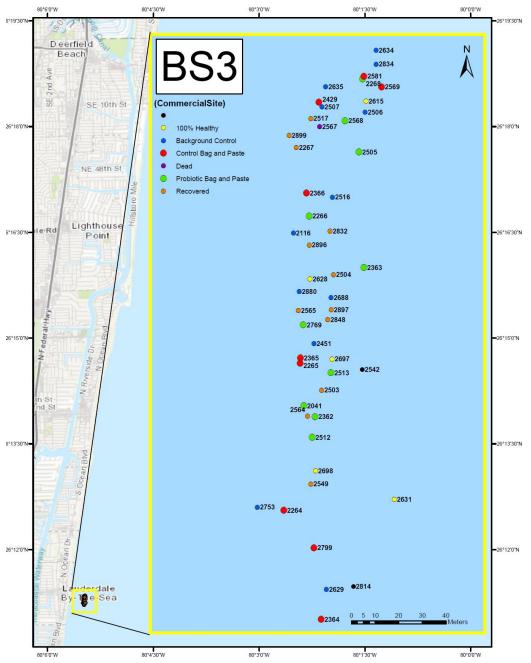
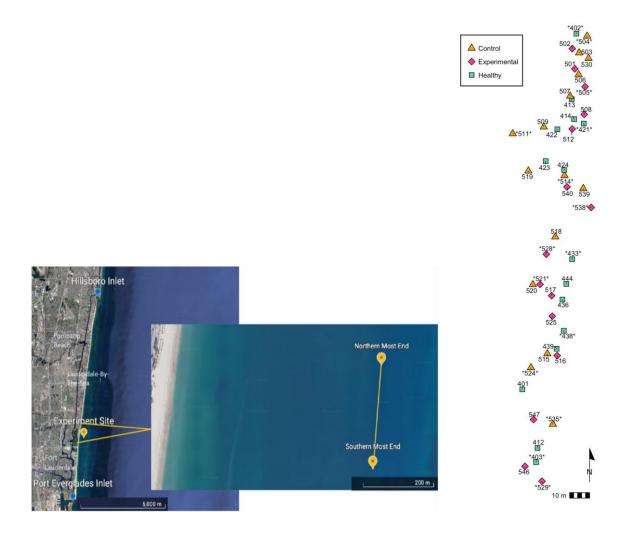


Figure 2. Map of probiotics site BS3 treatment corals established July 23, 2021.



**Figure 3.** Map of the FAU nutrient site established September 17, 2021. Numbered shapes indicate individual corals by treatment type.

#### 4. **RESULTS**

All strike team activities have been reported to FWC's coral disease intervention dashboard (<u>https://arcg.is/0L1LWX</u>). Results from 2019-2020 (PO B558F2) Task 5 to compare antibiotic paste versus chlorinated epoxy on *Montastraea cavernosa* were published in the SCTLD special of Frontiers in Marine Science Coral Reef Research. It can be found here: <u>https://www.frontiersin.org/articles/10.3389/fmars.2021.666224</u>

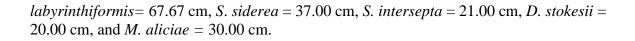
#### 4.1. Broad-scale Coral Disease Intervention Strike Team (Task 2)

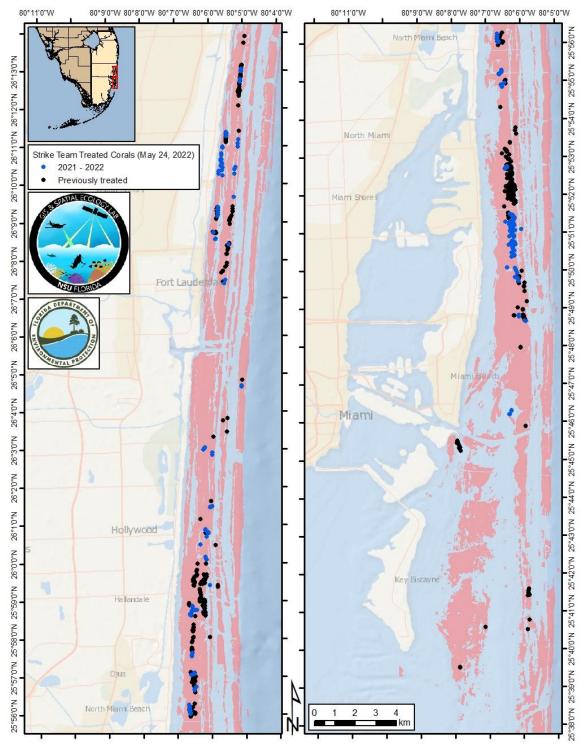
As of May 19, 2022, a total of 1,218 colonies have been treated by the coral disease interventions strike teams in the Coral ECA since 2018 (including the experimental sites, but not the large priority corals in section 4.2) (Figure 4). The total number of treatments by species were 1,012 *M. cavernosa*, 79 *O. faveolata*, 1 *O. annularis*, 32 *Colpophyllia natans*, 39 *Pseudodiploria strigosa*, 34 *Pseudodiploria clivosa*, 9 *Diploria labyrinthiformis*, 5 *Solenastrea bournoni*, 2 *Siderastrea siderea*, 3 *Stephanocoenia intersepta*, 1 *Dichocoenia stokesii* and 1 *Mycetophyllia aliciae*.

Out of the 1,218 colonies, 1,093 (89%) were treated with antibiotic ointment (897 *M. cavernosa*, 74 *O. faveolata*, 1 *O. annularis*, 30 *Colpophyllia natans*, 37 *Pseudodiploria strigosa*, 33 *Pseudodiploria clivosa*, 9 *Diploria labyrinthiformis*, 5 *Solenastrea bournoni* 2 *Siderastrea siderea*, 3 *Stephanocoenia intersepta*, 1 *Dichocoenia stokesii* and 1 *Mycetophyllia aliciae*); 109 (10%) corals were treated with chlorinated epoxy (102 *M. cavernosa*, 4 *O. faveolata*, 2 *P. strigosa*, and 1 *C. natans*); and 16 (1%) *M. cavernosa* treated with CoreRx B2B without antibiotics (that were not successful).

A total of 802.18 meters of antibiotic paste treatments, 68.59 meters chlorinated epoxy treatments, and 6.4 meters of CoreRx Base treatments were performed totaling 877.17 meters. That's 2.3 times the height of the Empire State Building. The average treatment length per coral was 74.00 cm which varied by species: *M. cavernosa* = 75.31 cm, *O. faveolata* = 94.15 cm, *O. annularis* = 149.00 cm, *P. strigosa* = 46.81 cm, *P. clivosa* = 55.81 cm, *C. natans* = 51.5 cm, *D. labyrinthiformis*= 63.38 cm, *S. bournoni* = 45.40 cm, *S. siderea* = 68.50 cm, *S. intersepta* = 21.00 cm, *D. stokesi* = 20.00 cm, and *M. aliciae* = 30.00 cm.

Between July 2021 and May 2022, a total of 320 colonies were treated with antibiotic paste by the coral disease interventions strike teams in the Coral ECA (not including the large priority corals (Section 4.2) and probiotics (Section 4.4)) (Figure 5). The total number of treatments by species were 218 *M. cavernosa*, 18 *O. faveolata*, 1 *O. annularis*, 22 *Colpophyllia natans*, 24 *Pseudodiploria strigosa*, 24 *Pseudodiploria clivosa*, 7 *Diploria labyrinthiformis*, 1 *Siderastrea siderea*, 3 *Stephanocoenia intersepta*, 1 *Dichocoenia stokesii*, and 1 *Mycetophyllia aliciae*. These totaled 226.80 meters of antibiotic ointment treatments were performed. The average treatment length per coral was 72.00 cm which varied by species: *M. cavernosa* = 73.55 cm, *O. faveolata* = 175.39 cm, *O. annularis* = 149.00 cm, *P. strigosa* = 41.625 cm, *P. clivosa* = 46.79 cm, *C. natans* = 40.4 cm, *D*.





**Figure 4**. The location of the 1,218 strike team treated corals as of May 19, 2022. The blue dots are those colonies treated during the current project period.

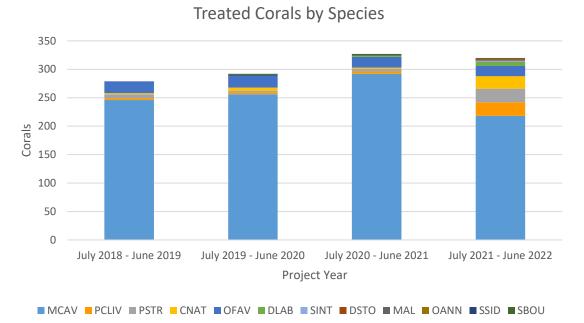
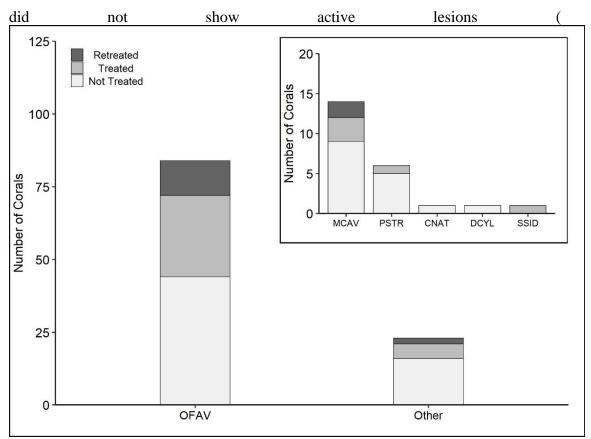


Figure 5. The number of strike team treated corals by species by project year.

#### 4.2. Apply Interventions to 100 Large Corals (Task 3)

#### a. Treatment Success

Figure 6 illustrates the locations of all 107 priority corals. Not all monitored corals required treatments this period; 47 corals (43.9%) were treated, and 60 corals (56.1%)



#### Figure 7 and

Table 1). The proportion of colonies requiring treatment were fairly similar between species, with 40 out of the 84 (47.6%) monitored *Orbicella* spp., and 5 of the 14 M. *cavernosa* (35.7%) requiring treatment.

Over the project period, a total of 189 antibiotic ointment treatments were conducted on 47 large colonies. Twenty-five treatments failed, equating to 87.6% effectiveness on lesions and an 86.9% effectiveness on coral colonies.

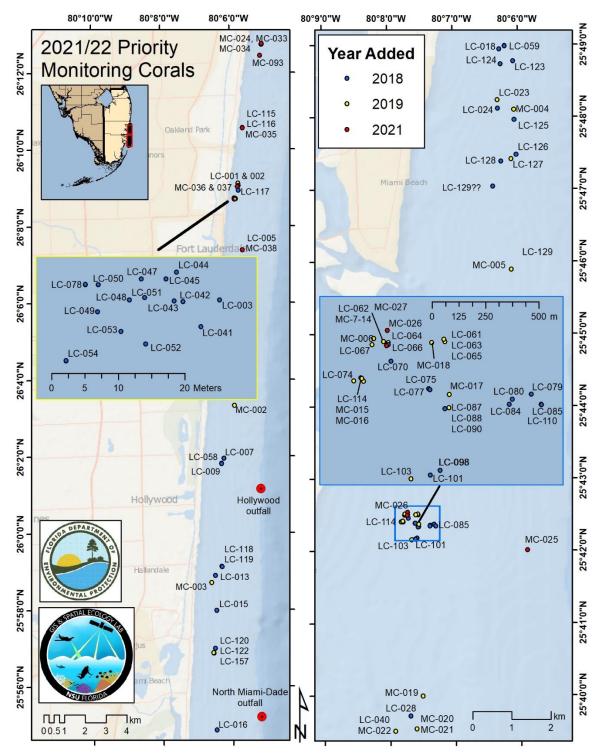
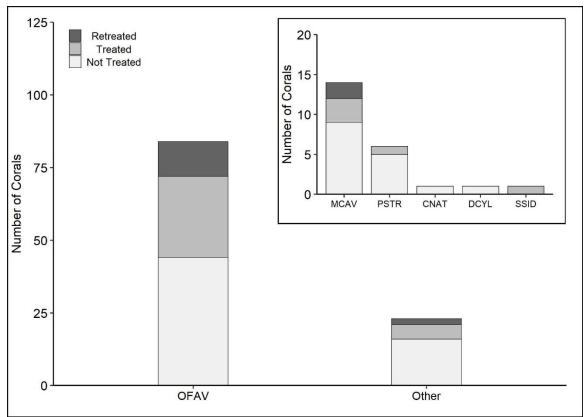


Figure 6. Map of the large priority monitoring corals colored by the year they were added to the monitoring.



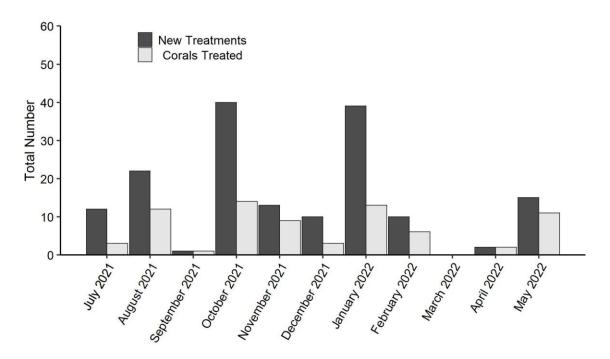
**Figure 7**. Total number of corals monitored and Not Treated (no disease), Treated (once), and Retreated for each species from June 2021 to May 2022. "Other" has been enlarged to show the number of treatments for the less abundant species.

Table 1. Total antibiotic ointment treatments by species from June 2021 – May 2022
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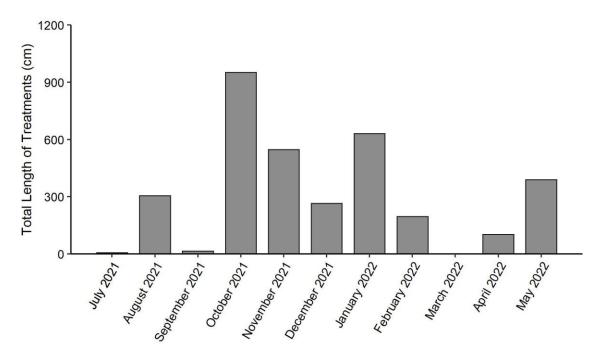
	MCAV	OFAV	PSTR	ALL SPECIES
Total Colonies	14	84	6	107
Total Treated Colonies	5	40	1	47
% Treated	35.7%	47.6%	16.7%	43.9%

#### b. Temporal Infection Patterns

At each monitoring period, all disease lesions were treated, thus the total number of new treatments indicates the amount of new disease found on the monitored corals over time after their initial visit. Figure 8 summarizes the number of new treatments required (black) and number of treated corals (grey) per monitoring period for all corals during the project period. The number of new infections and corals requiring treatment varied through time with relatively high numbers of treatments and colonies treated except for September 2021, March 2022, and April 2022. Figure 9 summarizes the amount of treatment required in each period.



**Figure 8**. The number of new treatments (black) on all corals used as a proxy for new infections and the number of treated corals (light gray) by treatment period.



**Figure 9.** The total treatment length (cm) on all corals by monitoring period. Shaded area indicates antibiotic ointment treatments.

#### 4.3. Recon Sites (Task 4)

#### 4.3.1. Desktop analysis

The desktop analysis resulted in nineteen sites with previously high coral richness and/or density that were severely impacted by hurricane Irma and disease in 2017 (

Table 2 and Figure 10) (Walker 2018). These sites should be recognized as candidates for future restoration activities to restore coral densities and richness to previous amounts. Figure 11 indicates the coral densities before and after 2017. Figure 12 indicates the coral richness before and after 2017. See Walker 2018 for site specific data.

#### 4.3.2. Reconnaissance Sites

Out of the 233 2020 NCRMP dives and 277 strike team dives completed during this project, thirty sites were identified during recon that contained unique or special existing corals Figure 13 and Table 3. Of these sites, 7 locations of abnormally high coral density were identified to consider for restoration and/or experimental activities. Table 4 contains information associated with these sites.

Two previously unknown colonies of *Dendrogyra cylindrus* (pillar coral) were found with live tissue (Figure 14 and Figure 15). The first was located South of Port Miami and had multiple live tissue isolates, it was tagged (#2025) and has been monitored monthly since February 2021 as part of our monthly large coral monitoring. On April 8, 2021, another large pillar coral colony was found with one small remnant isolate in Hollywood, FL. As the likelihood of survivorship was low, this was reported to the coral rescue team who is making plans to collect it and store in a facility to help increase their genetic diversity of captive stocks. These colonies are supposed to be added to local monitoring efforts. They might also be useful as *D. cylindrus* outplant sites to restore lost tissue.

Several sites with existing high coral density and diversity with apparently healthy, large colonies were identified. The Pompano mooring buoys have both high coral density and diversity with a number of larger, healthy-looking colonies that span a significant distance. Due to persistent disease at this site, we revisit periodically as part of our strike team intervention efforts.

The Commercial pier cluster site is an area of high *Montastrea cavernosa* density with a significant number of larger live colonies. This high-density site is located along the first reef ledge at a depth of 7 meters. Since this is a linearly distributed site, we collected North and South boundary points. This site also contains several large (3+m diameter) *Orbicella faveolata* colonies, a large *Pseudodiploria strigosa* (>1m diameter) as well as a significant cluster of *Orbicella annularis* colonies.

Another high density *Montastraea cavernosa* (BS3) site located within the Commercial Pier Site was identified and used for probiotics experiments by Smithsonian researchers.

Three new large healthy Orbicella faveolata colonies were found

Table 5). These are important because they have persisted through past bleaching and disease outbreaks. Note LC-2522 has a max planar length of 5.5 meters.

A rare, large *Orbicella annularis* (over 5.8 m diameter) was located in Broward County during our recon/intervention activities (Figure 16). This colony had multiple active disease lesions and high previous mortality. It was tagged (#5915) and treated on October 27, 2021. We plan on revisiting this colony and retreating it if necessary to ensure the survival of this important individual.

A second large *O. annularis* (over 6 m diameter) was located in the same vicinity as the colony above on the same day. This colony was ~90% alive making it worthy of monthly monitoring to maintain its health (Figure 16). Multiple individual *O. annularis* colonies were observed scattered along the reef crest between these two large colonies. Having two large *O. annularis* individuals in the same vicinity is beneficial and may provide a practical place to conduct local assisted fertilization for this species.

Divers located one previously unknown large, healthy *Orbicella faveolata* (~3.6 m diameter). Its large size and high tissue cover make it a good candidate for our large coral monitoring and/or restoration efforts. This colony was tagged (#5029), mapped, and entered into the large coral database. Additionally, divers located six large healthy *O. faveolatas* that have persisted through the SCTLD outbreak thus far. Unfortunately, for large *O. faveolatas* #5 and 6 the GPS that was being used to save colony location turned off so we plan on revisiting from the last recorded position and attempt to relocate these colonies.

A large *Pseudodiploria strigosa* nearly 100% live was reported by a public stakeholder with disease through the Southeast Florida Action Network (SEAFAN) program (Figure 17). Colonies of this size with >90% live tissue are rare in the region, thus we mobilized immediately to treat this coral. The disease was arrested immediately upon treatment and the colony has healed. We periodically visit this coral to check its condition.

Two of the high density *Montastrea cavernosa* sites that were discovered while conducting recon activities have been used for experimentation. Their high coral density made them ideal locations to test disease intervention techniques. We assisted a team from the Smithsonian Marine Station testing their new probiotic disease intervention treatments and continue to assist with monitoring at one of these sites. The colonies at the second high density site (Brooke's Site) were tagged, mapped, photographed, and used in a nutrient experiment in a collaboration with Josh Voss at FAU Harbor Branch.

We revisited the high density Mcav 1 site on March 24, 2021. While it appears that one or two colonies perished (likely due to SCTLD) the remaining colonies were healthy with no

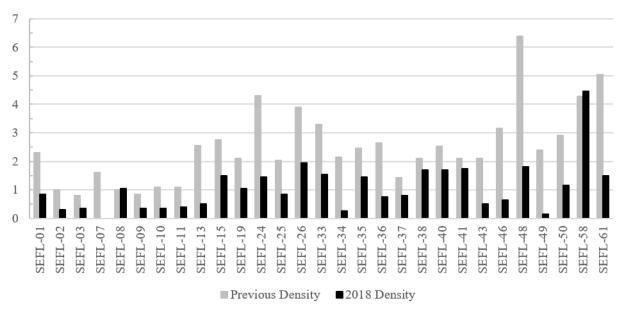
80°10'0"W 80°0'0"W 80°10'0"W W"0'0°08 139 SEFL-01 eerfield 2 rti 26°20'0"N FL-24 Ø Σ EFL-25 Coral Springs CO SEFL-23 27°0'0"N Ξ Jupiter Inlet сh 26°10'0"N SEFL-33 a a Φ SEFLESS 6 ..... Ε Pal Σ 26°50'0"N Broward County 0 orth Miami-Dade County σ Pembroke Pin 26°0'0"N z -Miraman SEFL-48 SEFL-07 a SEFL-10 ≥ 26°40'0"N 4 SEFL-46 0 υ a -SEFL-13 SEFL-48 Φ C 25°50'0"N В В SEFL-50 3.5 ε O SEFL-15 Miles \_ Kilometers a 3.5 26°30'0"N ۵. SEFL-19 iscayne 4 ÷ ٦ SEFL-61 25°40'0"N 0 S В 80°10'0"W 80°0'0"W 80°10'0"W

signs of active disease. The high coral density observed on this patch compared to the surrounding reef makes it an attractive location for *in situ* SCTLD studies.

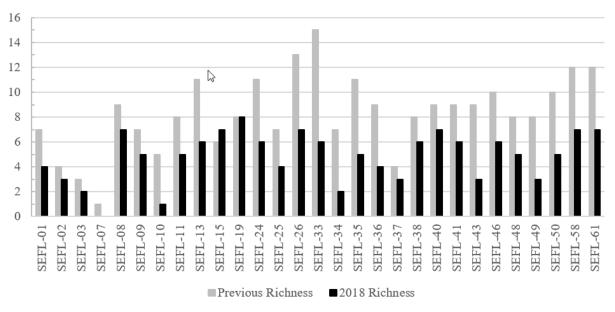
**Figure 10**. Map showing the nineteen potential restoration sites hit hard by SCTLD and hurricane Irma in 2017. From Walker 2018.

Site name County		Coordinates	Description
SEFL-01 Martin		N 27.131669,	Previous high density coral site on nearshore
SEFL-01	Wartin	W 80.134029	ridge complex
SEEL 07	Palm Beach	N 26.722063,	Previous high density coral site on nearshore
SEFL-07	Palm Beach	W 80.032167	ridge complex
SEEL 10	Palm Beach	N 26.41.0370,	Previous high coral richness site on nearshore
SEFL-10	Paim Beach	W 80.18900	ridge complex
SEEL 12	Palm Beach	N 26.610469,	Previous high coral richness and density site
SEFL-13	Palm Beach	W 80.023030	with diversity on outer reef
CEEL 15	Dalas Darah	N 26.531130,	Previous high coral density site with diversity
SEFL-15	Palm Beach	W 80.029550	on ridge
CEEL 10	D-1 D1-	N 26.479420,	Previous high coral density site with diversity
SEFL-19	Palm Beach	W 80.041899	on outer reef
GEEL 24	D 1	N 26.311350,	Previous high coral richness and density site
SEFL-24	Broward	W 80.067280	with diversity on middle reef
GEEL 25	D 1	N 26.275582,	
SEFL-25	Broward	W 80.071032	Previous high coral density site on middle reef
GEEL 26	D 1	N 26.273759,	Previous high coral richness and density site
SEFL-26	Broward	W 80.063648	with diversity on outer reef
CEEL 22	Broward	N 26.154550,	Previous high coral richness and density site
SEFL-33		W 80.088549	with diversity on inner reef
CEEL 24	Duomond	N 26.148550,	Previous high coral density site on nearshore
SEFL-34	Broward	W 80.095080	ridge complex
CEEL 25	Broward	N 26.141733,	Previous high coral richness and density site
SEFL-35	Broward	W 80.090600	with diversity on inner reef
CEEL 26	Broward	N 26.123719,	Previous high coral richness and density site
SEFL-36		W 80.094780	with diversity on inner reef
CEEL 42	Mani Dala	N 25.963020,	Previous high coral richness and density site
SEFL-43	Miami-Dade	W 80.108379	on nearshore ridge complex
SEEL 46	Miami-Dade	N 25.893110,	Previous high coral density site with diversity
SEFL-46	Miami-Dade	W 80.108019	on nearshore ridge complex
CEEL 49	Mari Dala	N 25.853100,	Previous high coral density site with diversity
SEFL-48	Miami-Dade	W 80.103699	on inner reef
SEEL 40		N 25.852940,	Previous high coral richness and density site
SEFL-49	Miami-Dade	W 80.110519	on nearshore ridge complex
CEEL 50	Mani D. 1	N 25.810650,	Previous high coral density site with diversity
SEFL-50	Miami-Dade	W 80.097700	on inner reef
CEEL (1	Mani D 1	N 25.65853,	Previous high coral density site with diversity
SEFL-61	Miami-Dade	W 80.098470	on inner reef

**Table 2**. Desktop analysis sites identified for restoration activities as having been severelyimpacted by hurricane Irma and SCTLD. From Walker 2018.



**Figure 11**. Coral density (corals/m<sup>2</sup>) by site from the previous survey (gray) versus the 2018 survey (black). From Walker 2018.



**Figure 12**. Coral richness (number of species) by site from the previous survey (gray) versus the 2018 survey (black). From Walker 2018.

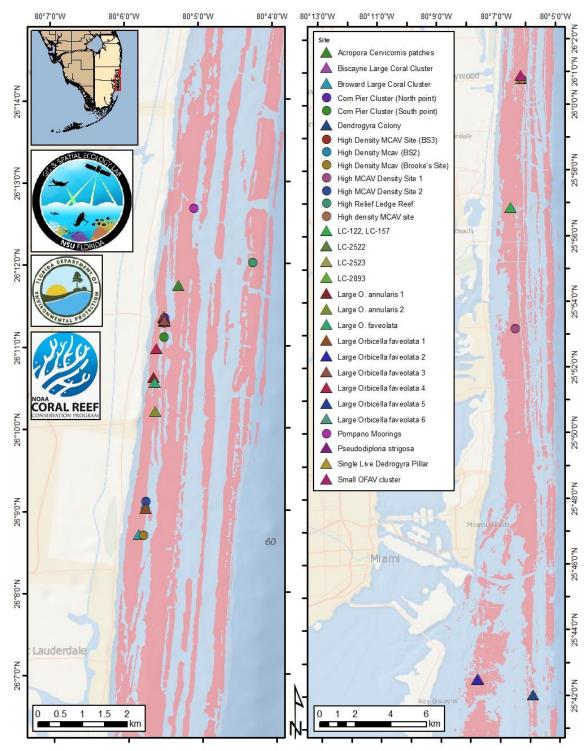


Figure 13. The location of the 30 identified recon sites.

Nr	Site name	County	Coordinates	Description
		,		Located a live Dcyl colony on a
1	Dendrogyra Colony (MC-025)	Dade	N 25.700211, W 80.098248	shallow reef in South Miami Dade
				Nice relief, high coral cover and
2	Pompano Moorings	Broward	N 26.211332, W 80.084531	diversity High rugostiy deep reef, appeared to
3	High Relief Ledge Reef	Broward	N 26.200196, W 80.071329	have coral colonies, lots of large fish
4	Broward Large Coral Cluster	Broward	N 26.145267, W 80.097367	Cluster of large OFAV
5	Biscayne Large Coral Cluster	Dade	N 25.708467, W 80.12895	Cluster of large OFAV
-				High density of MCAV on a patch
6	High MCAV Density Site 1	Dade	N 25.88624, W 80.106852	reef.
				Near BS2 High Density of large MCAV
7	High MCAV Density Site 2 (NCRMP site 3204)	Broward	N 26.151909, W 80.095806	and some CNAT, observed some SCTLD
,	Site 520+)	Dioward	W 20.131303, W 80.033800	LCM on 11/24/2020 noted many
8	LC-122, LC-157	Broward	N 25.94788, W 80.10913	diseased MCAV and Ofav
9	Commercial Pier Cluster (South point)	Broward	N 26.185208, W 80.09145	High density of MCAV
10	Commercial Pier Cluster (North point)	Broward	N 26.189059, W 80.091318	High density of MCAV
11	LC-2893	Broward	N 26.188383, W 80.091359	Large OFAV Colony
12	LC-2523	Broward	N 26.189161, W 80.091371	Large OFAV Colony
13	LC-2522	Broward	N 26.189415, W 80.091368	Large OFAV Colony
14	Pseudodiploria strigosa	Broward	N 26.189059, W 80.091318	Large PSTG Colony
15	High density Mcav (Brooke's Site)	Broward	N 26.144958, W 80.096331	High density of MCAV
				SSI Probiotic Site (High density of
16	High Density Mcav (BS2)	Broward	N 26.150397, W 80.09594	MCAV)
17	Acropora Cervicornis Multiple patches	Broward	N 26.195693, W 80.088124	Area with extensive A. cervicornis thickets
17	Actopora cervicornis multiple patches	Diowaru	11 20.195095, 11 80.088124	Old Dendrogyra colony skeleton with
18	Single Live Dedrogyra Pillar	Broward	N 26.013177, W 80.102561	single live pillar
19	High Density MCAV Site (BS3)	Broward	N 26.188045, W 80.091429	High density MCAV site along 1st reef
				Large OANN colony 91%old dead but
20	Large O.Annularis #1 (5915)	Broward	N 26.177027, W 80.09383	over 5.8m across Large OANN colony #2 over 6m
21	Large O.Annularis #2	Broward	N 26.170234, W 80.093439	across, high living tissue coverage
22	Large O. Faveolata (5029)	Broward	N 26.175988, W 80.093569	Large OFAV colony 3.6m x3.15m
	High density MCAV site (also many	Dioticita		High rugosity reef with very high
23	Ofav)	Broward	N 26.012524, W 80.10229	density MCAV (also many Ofav)
	Small OFAV cluster			
24	(3 colonies close, many nearby, near above site)	Broward	N 26.01474, W 80.102898	Unusually high density of OFAV
		Broward	1120.01171, 11 00.102030	Large <i>Orbicella faveolata</i> >90% live
25	Large Orbicella faveolata #1	Broward	N 26.150438, W 80.095793	and healthy
26	Large Orbicella faveolata #2	Dade	N 25.70833, W 80.12949	>1.5m Orbicella faveolata 60% live
			N 26 400600 ··· 00 00000	~4m Orbicella faveolata 85%live and
27	Large Orbicella faveolata #3	Broward	N 26.188699, W 80.091328	healthy >2m <i>Orbicella faveolata</i> >85%live
28	Large Orbicella faveolata #4	Broward	N 26.182942, W 80.093148	and healthy
			,	~3m Orbicella faveolata >90% live
29	Large Orbicella faveolata #5	Broward	GPS shutoff	and healthy
30	Large Orbicella faveolata #6	Broward	GPS shutoff	Orbicella faveolata >1.5m, healthy

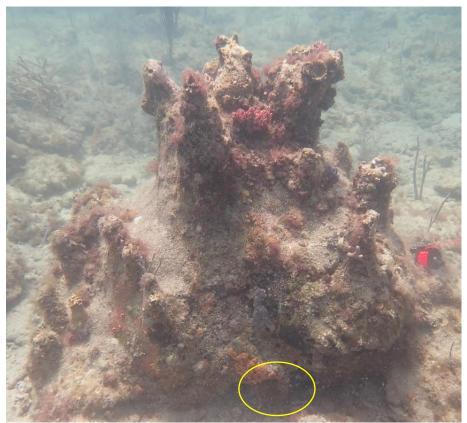
**Table 3**. Preliminary information on all of the recon sites of interest identified during this project.

Nr	Site	County	Location	Description
6	High MCAV Density Site 1	Dade	N 25.88624 W 80.106852	High density of MCAV on a patch reef
7	High MCAV Density Site 2 (NCRMP site 3204)	Broward	N 26.151909 W 80.095806	Near BS2 High Density of large MCAV and some CNAT
9	Commercial Pier Cluster (South point)	Broward	N 26.185208 W 80.09145	High density of MCAV
10	Commercial Pier Cluster (North point)	Broward	N 26.189059 W 80.091318	High density of MCAV
15	High density Mcav (Brooke's Site)	Broward	N 26.144958 W 80.096331	High density of MCAV
16	High Density Mcav (BS2)	Broward	N 26.150397 W 80.09594	SSI Probiotic Site (High density of MCAV)
19	High Density MCAV Site (BS3)	Broward	N 26.188045 W 80.091429	High density MCAV site along 1st reef
23	High density MCAV site (also many Ofav)	Broward	N 26.012524 W 80.10229	High rugosity reef with very high density MCAV (also many Ofay)

**Table 4**. Sites identified as having potential for restoration and/or experimentation. Note the Commercial pier cluster has a Northern and Southern point since it is a linear site.



Figure 14. Examples of new corals found during recon dives.



**Figure 15**. Yellow circle shows the small isolate on a large almost-dead pillar coral colony found in Hollywood, FL that was reported to the coral rescue team for collection.

			Planar			Linear	
Tag	Coral spp	%Mortality	Max Length	Max Width	Max Height	Max Length	Max Width
2893	OFAV	30	240	220	145	315	310
2523	OFAV	15	253	205	175	382	340
2522	OFAV	30	550	270	150	580	355

**Table 5.** Stats on the three new large Orbicella faveolata colonies. Measurements are in cm.



**Figure 16**. Examples of new corals found during recon dives. Top left is Large *O. annularis* #1 (5915). Top right is large *O. annularis* #2. Bottom is large *O. faveolata* (5029).



**Figure 17**. Diseased Large *Pseudodiploria strigosa* colony reported to SEAFAN by a concerned citizen on Feb 10, 2021 (top left). Strike teams treated it with antibiotic paste on Feb. 16 (top right). On May 14, the lesion was stopped and tissue was healing over the disease-break (bottom).

# 4.4. Field Test New Permitted Intervention Techniques and Materials (Task 5)

#### 4.4.1. Probiotics on Montastraea cavernosa

Results from the DEP 2020-2021 PO B7B6F3 Task 5 on the "Metabolomics of Healthy and Stony Coral Tissue Loss Disease Affected *Montastraea cavernosa* Corals" were published in the SCTLD special of Frontiers in Marine Science Coral Reef Research. It can be found here: <u>https://www.frontiersin.org/articles/10.3389/fmars.2021.714778</u>

During this period, NSU assisted the Smithsonian Marine Station at Fort Pierce with probiotics treatments at a new site, BS3 (Figure 2). Here we report on the NSU field logistics activities. Please contact the main PI for the probiotics work, Valerie Paul, for more information on those experimental outcomes.

The initial visit and site setup of BS3 occurred on July 23, 2021. This consisted of tagging diseased corals and healthy controls, taking 3D pictures of all tagged colonies, and collecting tissue/mucus samples for genetic and metabolomic testing. One week later (7/30/2021), an additional 13 corals were tagged and sampled and all corals at the site were treated with probiotics and photographed for 3D modeling. On August 31, 2021, all colonies were revisited to collect tissue/mucus samples and images for 3D modeling. One coral was added during this revisit. On November 4, 2021, all colonies were revisited to collect tissue/mucus samples and treated with probiotics for a second time. All corals were photographed on March 29th, 2022 as well as on May 5th, 2022, when the corals were once again treated for a third time.

#### 4.4.2. Nutrient enrichment on Montastraea cavernosa

This period NSU assisted FAU with investigating the impact of elevated nutrient levels on the spread and progression of SCTLD in *M. cavernosa* (Figure 3). Here we report on the NSU field logistics activities. Please contact Joshua Voss for more information on those experimental outcomes.

FAU's experimental site was set up on September 17, 2021. Forty-five corals were tagged and photographed. Of the forty-five total colonies, fifteen healthy and thirty SCTLDaffected *M. cavernosa* coral colonies were tagged and divided into three treatment groups: 1) apparently healthy, 2) SCTLD- affected controls, and 3) SCTLD-affected experimental colonies (amended with time-release nutrients). The experiment started September 22, 2021, with revisits on the 24<sup>th</sup>, 27<sup>th</sup>, 30<sup>th</sup> of September and 5<sup>th</sup> and 10<sup>th</sup> of October. The treatment was ended October 15, 2021and recovery assessments were conducted on the 3<sup>rd</sup> of November and 1<sup>st</sup> of December completing and ending the experiment.

#### 5. DISCUSSION

Coral diseases have caused enormous impacts to coral populations globally perpetuating the rapid need for large-scale coral reef restoration. SCTLD exemplifies that coral diseases are more devastating than ever before. When a virulent disease like SCTLD ravages a coral ecosystem, it significantly alters the population's demographics and causes local extinctions. However, recent disease intervention techniques are over 90% effective at stopping lesions. Implementing these in the restoration toolbox reduces the necessity for costly and time-consuming post hoc restoration techniques (e.g. microfragmentation, coral husbandry, and outplanting).

Our work shows that SCTLD is still prevalent in the region, that interventions are still needed, and that these efforts are worthwhile. This past year, interventions helped maintain species diversity and ecosystem services within the Coral ECA and kept alive some of the largest and oldest animals in Florida. Although disease interventions are very effective at stopping disease lesions, they do not provide protection against reinfection. This requires regular visits to ensure colony survival. Since disease intervention response during such an event is virtually impossible on a landscape scale, priorities must be considered. Intervention activities are usually allocated between prioritizing saving certain colonies by regular monitoring and treating large areas without the intent to monitor. The latter

involves a team of divers, known as a striketeam, covering large areas of reef, treating coral lesions that they encounter, tagging treated colonies, and recording their locations. The purpose of the striketeam is to maximize the area covered to treat as many disease lesions as possible without the intent of returning to monitor treatment success. By treating more of the remaining corals less susceptible to SCTLD, more live coral tissue and genetic diversity are saved. This allows post hoc restoration to be focused on the species more susceptible to SCTLD that are now functionally extinct.

Prioritizing the largest, oldest colonies of reef-building species is imperative to preserve the high fecundity, ecological function, and genetic diversity of the most resilient corals. Large corals have a higher reproductive potential thereby increasing the natural ability to replenish the reef as well as harness the potential for assisted reproduction through gamete collection. Their size also preserves ecological functions such as existing reef structures and habitats. As a proxy for age, coral size is an indicator of their resistance to previous perturbations which may indicate higher fitness.

Monthly monitoring and treatments have reduced the loss of live tissue area and provided valuable information on the temporal and spatial variations of colonies with lesions. Corals not regularly treated with disease interventions have had drastic declines in live tissue area. Regular disease intervention is restoring colony health and saving the large priority corals from SCTLD and is an efficient method to save live tissue area preempting slower, more costly post hoc restoration efforts.

By stopping disease lesions, *in situ* disease interventions can save a higher magnitude of live tissue in comparison to the amount that can be generated from years of microfragmentation. Our intervention activities have saved species and size classes that would take decades to achieve through restoration at a fraction of the cost that equivalent restoration would require to achieve similar coral cover. Considering the Coral ECA large coral monitoring alone, the current live tissue area of all previously treated large corals is 3,578,091 cm<sup>2</sup> (357.8 m<sup>2</sup> or 3,851 ft<sup>2</sup>). This is the equivalent to the area of 2 volleyball courts of live coral tissue. If 36 outplant plugs covered one sq ft with 100% survival and no disease, that equates to 138,636 coral restoration fragment plugs. Granted, we can't assume all tissue would've been lost without treatment or that the treatments are 100% effective or what will be lost if we stop monitoring those corals today, but it gives a rough estimate of the restoration effort required to get a similar amount of tissue. There is also the considerable advantage that the saved corals are already reproductive, and the largest ones have the most reproductive potential. Disease interventions have the added benefit of reducing disease loads as well. But the use of antibiotics poses an unknown risk in the environment.

Our reconnaissance for coral restoration sites identified eleven sites with high densities of corals remaining six years into the ongoing SCTLD pandemic. These locations are good targets for periodic strike team activities to recon for lesion outbreaks and perform interventions when necessary. These locations can also be useful in collecting hardened colonies that may be more resistant to the disease.

We also identified nineteen sites reported in Walker 2018 that had drastic reductions in coral density and/or richness after hurricane Irma and three years of SCTLD. These sites would be ideal locations for post hoc restoration activities to outplant nursery-raised colonies back to previous community levels.

We identified nine new large *Orbicella* colonies of high value to the State of Florida. These colonies can be used as genetic stock for gamete collections for assisted reproduction. They can also be monitored and treat for disease to assist in understanding disease dynamics and coral resistance to disease.

Spatiotemporal disease patterns occur on portions of Florida's Coral Reef, where SCTLD is endemic. Frequent monitoring and successful disease interventions have kept diseased corals alive, revealing variable lesion outbreak patterns where some colonies frequently or occasionally exhibit new lesions, while others appear resistant to SCTLD and have never been observed with lesions. The monthly monitoring data have been invaluable in linking environmental drivers to disease dynamics (Walker et al., 2021). Higher water temperatures, amounts of water following out of the inlets, and rainfall account for 56.4% of the temporal variability in the number of new lesions. Understanding these dynamics can help identify the disease and optimum times to treat. Caution should be taken when choosing coral restoration sites as to avoid areas more prone to disease.

#### 6. RECOMMENDATIONS

*Continue monthly monitoring and treatment of large priority corals* – Monitoring these colonies has saved many from extinction. Monthly monitoring and treatment have facilitated the classification of corals based on differing infection rates which has informed the design of the SCTLD Resistance Research Consortium.

*Continue broad-scale strike team efforts* – Conducting strike team efforts to reduce the active SCTLD prevalence and save the genetic diversity remaining on the reef.

*Continue use of antibiotic ointment CoreRx B2B and amoxicillin* (1:8 weight ratio) – Perform margin treatment and appropriate disease-break interventions using antibiotic paste on SCTLD lesions.

Continue recon for potential new monitoring colonies and restoration sites.

Continue public outreach for SEAFAN and other citizen reporting mechanisms to educate the public on reporting diseased corals.

Consult previous data collections to understand pre-SCTLD coral densities and richness.

Avoid areas prone to increased coral disease when choosing restoration sites.

#### 7. CITATIONS

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