

# **Stony Coral Tissue Loss Disease (SCTLD) Reconnaissance, Intervention, and Monitoring in Dry Tortugas National Park**

Final Report

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## Acronyms

AAUS	American Academy of Underwater Sciences
BACI	Before After Control Impact
BISC	Biscayne National Park
CRCP	Coral Reef Conservation Program
DAC	Disease Advisory Committee
DD	Decimal Degrees
DEP	Department of Environmental Protection
DMS	Degrees Minutes Seconds
DRM	Disturbance Response Monitoring
DRTO	Dry Tortugas National Park
ESA	Endangered Species Act
FAU-HBOI	Florida Atlantic University – Harbor Branch Oceanographic Institute
FDEP	Florida Department of Environmental Protection
FCR	Florida’s Coral Reef
FWC	Florida Wildlife Commission
GPS	Global Positioning System
MOCC	Motorboat Operator Certification Course
NCRMP	National Coral Reef Monitoring Program
NFWF	National Fish and Wildlife Foundation
NOAA	National Oceanic and Atmospheric Administration
NPS	National Park Service
NSU	Nova Southeastern University
ORCP	Office of Resilience and Coastal Protection
PEPC	Planning, Environment, and Public Comment process
QA/QC	Quality Assurance/Quality Control
SCHMIR	Stony Coral Health Mass Intervention Response
SCTLD	Stony Coral Tissue Loss Disease
Sf	Susceptibility factor
SFCN I&M	South Florida & Caribbean Network Inventory and Monitoring Group
SFCN	South Florida & Caribbean Network
SNP	Single Nucleotide Polymorphism
SRC	Submerged Resources Center
UM	University of Miami
UVI	University of the Virgin Islands
VI-DPNR	Virgin Islands Department of Planning and Natural Resources

## EXECUTIVE SUMMARY

Since its emergence in 2014, Stony Coral Tissue Loss Disease (SCTLD) has severely and negatively impacted coral reefs throughout Florida. This report describes the reconnaissance, intervention, and monitoring efforts conducted at Dry Tortugas National Park (DRTO) prior to the arrival of SCTLD in May 2021 and for approximately one year thereafter. Prior to the first documentation of SCTLD in the Park, coral health and demographic data were collected at 30 priority reconnaissance sites to serve as a baseline for tracking reef-wide and Park-wide changes following disease onset. After the arrival of SCTLD, project efforts quickly shifted to disease intervention and over 14,000 corals were treated in the park within about one year. Large-scale liveaboard missions were essential to intervention efforts, resulting in nearly two thirds of the total corals treated at DRTO. While long-term monitoring analyses are still forthcoming, it appears that large-scale intervention combined with local maintenance at high-priority reefs is effective at slowing disease progression, particularly at isolated and/or remote locations. Approximately one year after initial SCTLD observation, reconnaissance surveys were repeated to assess short-term changes across the 30 reconnaissance sites due to the disease. Overall trends in the data showed declines in healthy coral abundance at individual sites, across species, and within all size classes. In general, the most abundant species prior to the onset of SCTLD suffered from the largest proportional declines in healthy corals observed, with *Montastraea cavernosa* suffering from the greatest loss. Despite these declines in coral abundance, concentrated intervention efforts at high-priority sites appear to be slowing the progression of SCTLD overall. Between reconnaissance, intervention, and monitoring work, Park staff and partners completed over 500 hours of project-related work underwater, excluding the large liveaboard missions. As SCTLD continues to impact DRTO, the continuation of these activities will be essential to inform Park-level adaptive management efforts, provide information about SCTLD progression and impacts at a reef-wide scale, and guide management actions at locations that are impacted by similar outbreaks in the future.

# INTRODUCTION

## **Stony Coral Tissue Loss Disease Background**

Florida's Coral Reef (FCR), the third largest barrier reef in the world, is currently experiencing a multi-year disease-related mortality event that was first observed near the port of Miami in 2014 (Precht et al., 2016). This disease termed Stony Coral Tissue Loss Disease (SCTLD) affects approximately 22 species of coral, including ESA-listed and primary reef building species, and is characterized by steep rates of infection and mortality at impacted sites throughout FCR and the Caribbean (Precht et al., 2016; FKNMS, 2018; Walton et al., 2018; Alvarez-Filip et al., 2019; Sharp et al., 2020; Dahlgren et al., 2021; Brandt et al., 2021; Kolodziej et al., 2021). SCTLD is characterized by a clear demarcation of active disease margin between live tissue and exposed skeleton, forming single or multiple fast-spreading lesions across infected colonies. Mortality rates of infected corals are almost 100% without intervention (Neely et al., 2020, Precht et al., 2016). Impacted sites experience swift, significant declines in live coral tissue cover, with some coral species exhibiting symptoms more quickly and/or more severely than others (Walton et al., 2018; Gintert et al., 2019, Sharp et al., 2020; Heres et al., 2021; Brandt et al., 2021; Costa et al., 2021; Neely et al., 2021a; Spadafore et al., 2021). Along FCR, coral species highly susceptible to SCTLD that are affected early in the progression of disease outbreak include: *Colpophyllia natans*, *Dendrogyra cylindrus*, *Dichocoenia stokesii*, *Diploria labyrinthiformis*, *Eusmilia fastigiata*, *Meandrina meandrites*, *Pseudodiploria strigosa*, and *Pseudodiploria clivosa*; coral species that are intermediately susceptible and are affected partway to later in the progression of disease outbreak include: *Orbicella annularis*, *Orbicella faveolata*, *Orbicella franksi*, *Montastraea cavernosa*, *Solenastrea bournoni*, *Stephanocenia intersepta*, and *Siderastrea siderea* (NOAA, 2018). As SCTLD has progressed throughout Florida's Coral Reef and the Caribbean, populations of susceptible coral species have declined, leaving regions with severe losses of structural complexity, altered reefscales, and reduced biodiversity and ecosystem function (Walton et al., 2018; Gilliam et al., 2019; Aeby et al., 2019, Muller et al., 2020; Sharp et al., 2020; Heres et al., 2021; Forrester et al., 2022).

While many agencies and research groups seek to determine the as-yet unidentified causative pathogen(s) of SCTLD, the disease is evidenced to contain a bacterial component due to its response to administration of antibiotics (Meyer et al. 2019; Neely et al., 2020; Rosales et al., 2020; Clark et al., 2021; Neely et al. 2021b). Transmission has been demonstrated via direct contact as well as through the water column in neutrally buoyant particles (Aeby et al., 2019, Dobblaere et al., 2020; Eaton et al., 2021; Williams et al., 2021). There is also evidence that certain genera of algal symbionts may confer some resistance to the disease (Dennison et al., 2021; Rubin et al., 2021). In addition to topical antibiotic treatment, probiotic treatment alternatives are also in development (Paul et al. 2021). However, without additional knowledge of the causative pathogen(s) and the factors contributing to infection and/or resilience in coral colonies, the most effective and widely utilized intervention approach throughout Florida's Coral Reef has been the application of topical antibiotic to infected corals.

Disease progression modeling indicated that SCTLD would arrive at Dry Tortugas National Park (DRTO), a remote Park approximately 70 miles west of Key West, by March 2021 (Dobbelaere et al., 2020). In advance, the Park drafted and implemented the SCTLD Response Plan based on the most effective-to-date intervention approaches, historical site data, and valuable partner input.

### **Project Purpose & Ongoing Efforts**

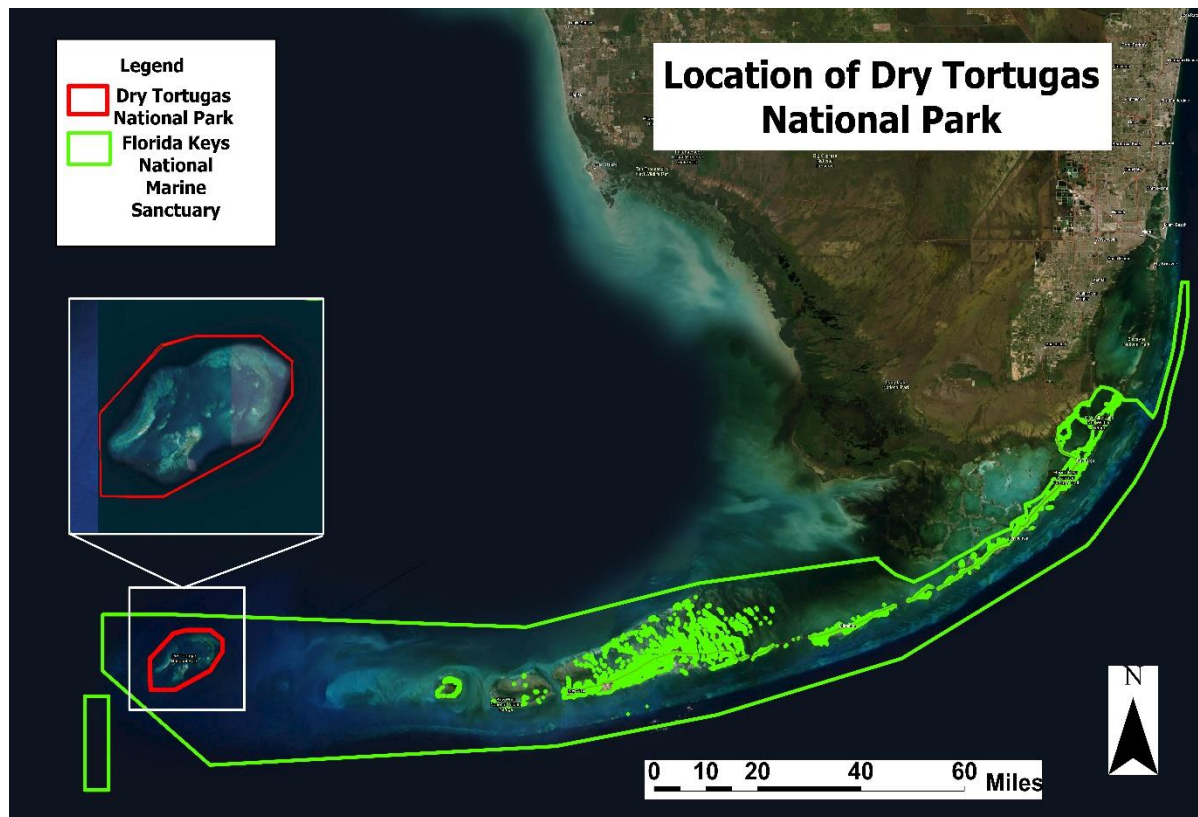
The purpose of this project is to conduct routine reconnaissance at highly susceptible and probable disease outbreak sites within the Park, respond to the SCTLD outbreak using disease intervention and treatment, and monitor impacts to marine resources and quantify the efficacy of response actions. While previous studies have concentrated on intervention treatment efficacy at the lesion and colony-level, there are few studies which assess efficacy of treatment at large, reef-level scales (Forrester et al., 2022, Neely et al., 2020). This project uniquely monitors the progression of SCTLD on a reef-wide scale across multiple time points, stages of disease, and locations. Furthermore, this project addresses a data gap within the literature by incorporating a Before After Control Impact (BACI) experimental study design to assess the efficacy of intervention across multiple treatment and control sites.

The objectives of this project are to locate and respond to SCTLD quickly, conduct intervention and treatment at high-priority reef features, and monitor the effectiveness of the intervention utilizing several complimentary monitoring techniques. Priority sites were selected for and characterized by multiple factors including increased live coral cover, a weighted coral susceptibility factor, increased coral biodiversity, increased coral abundance (especially large, reproductively viable corals), existing monitoring datasets, proximity to infrastructure and operational support, and visitor use and experience. This project supports the National Park Service (NPS) mission to protect and preserve resources at DRTO.

## METHODS

### Study Site: DRTO Description

The Dry Tortugas are the western-most extent of the FCR, located 113 km west of Key West, FL and 175 km northwest of Havana, Cuba. DRTO encompasses 296 km<sup>2</sup> (approximately 100 mi<sup>2</sup>) of mostly submerged lands and seven small islands. The Park is located at the beginning of the Florida Straits where the Loop Current, from the Gulf of Mexico, and the Caribbean Current, from the Yucatan, converge to form the Gulf Stream. DRTO is surrounded by the Florida Keys National Marine Sanctuary (FKNMS; Figure 1) and the Park's northwestern boundary abuts the Tortugas North Ecological Reserve, which includes the Tortugas western Bank. Riley's Hump, which is part of the of the Tortugas South Ecological Reserve, is located approximately 11 km to the southwest of the Park (Figure 1).

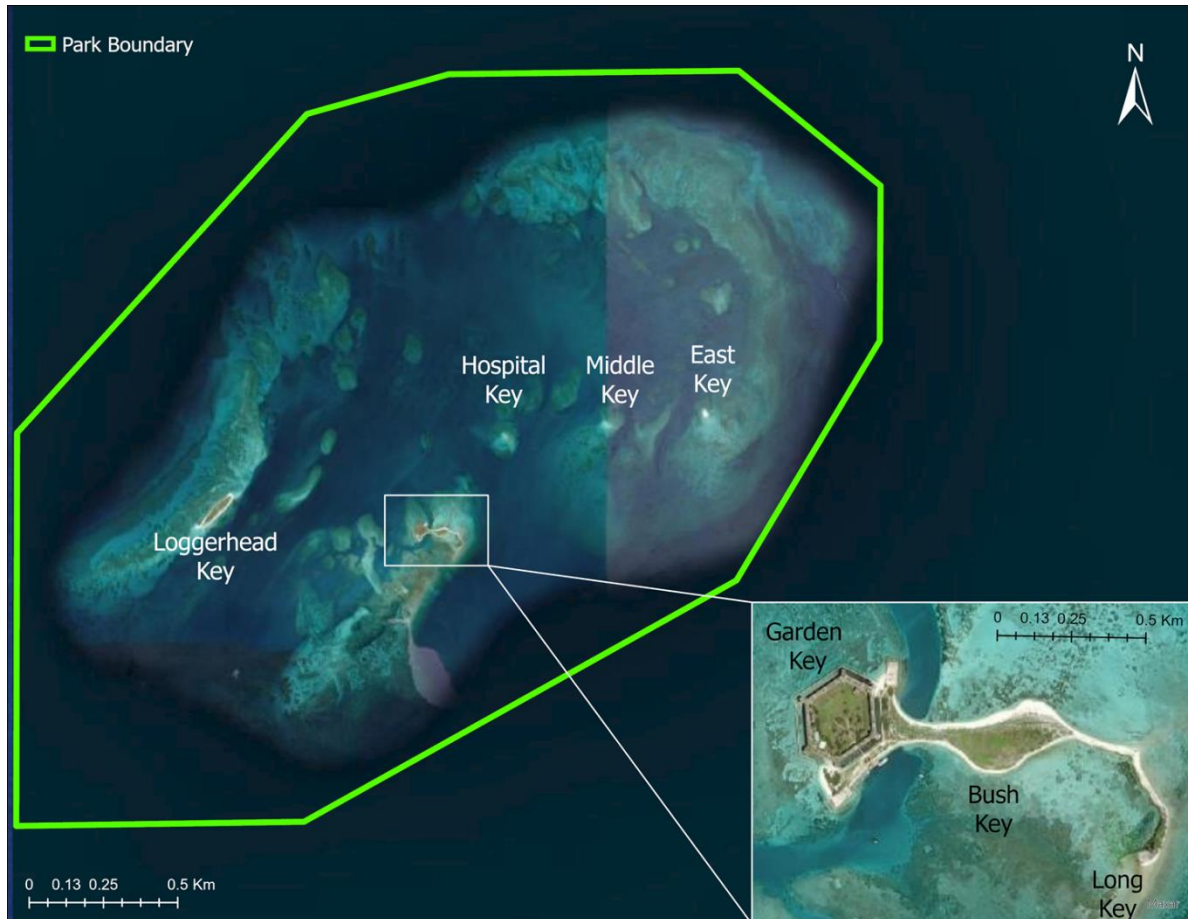


*Figure 1. Map depicting the location of Dry Tortugas National Park (red) within the Florida Keys National Marine Sanctuary (green).*

DRTO and the adjacent Tortugas Bank are characterized by highly dynamic physical oceanography and increased productivity. The dynamic currents circulate around three prominent limestone banks within the Park, which are comprised of Holocene corals and sand atop an underlying Pleistocene reef (Shinn et al. 1977). These banks create a partial atoll with an inner lagoon region separated by deeper groves and a mosaic of patch reefs (Figure 2).



Collectively, these prominent geological features are known for their abundance of commercially important reef fish and, because of their location and hydrodynamics, are believed to be important sources of recruitment for coral reef fishes and coral downstream (Domeier, 2004; Meurice, 2019).



*Figure 2. Map of DORTO boundary (green) and terrestrial lands within the Park.*

The temperatures in DORTO vary little from the balance of the Florida Keys. The highest temperatures occur in July and August (32 °C) and rarely drop below 19 °C during the winter. The Florida Keys are the driest area in Florida and due to its isolated position away from the mainland, DORTO is the driest region in the Keys. Precipitation averages 124 cm per year with most of the rain falling between May and October. Tropical storms and hurricanes can deliver excessive amounts of rain during hurricane season (June to November; peak September).

Water temperatures in DORTO are indicative of south Florida with winter temperatures in the low 20s (°C) to summer temperatures in the upper 20s (°C). Warm summertime temperatures can be punctuated by cold water upwellings that are likely caused by local eddies; however, these are currently not well understood. While considered ephemeral, these events can have impacts on coral resources (Ruzicka, R. et al. 2021). Average summer and winter sea surface temperatures have been increasing in recent years, resulting in coral bleaching events (Figure 3).

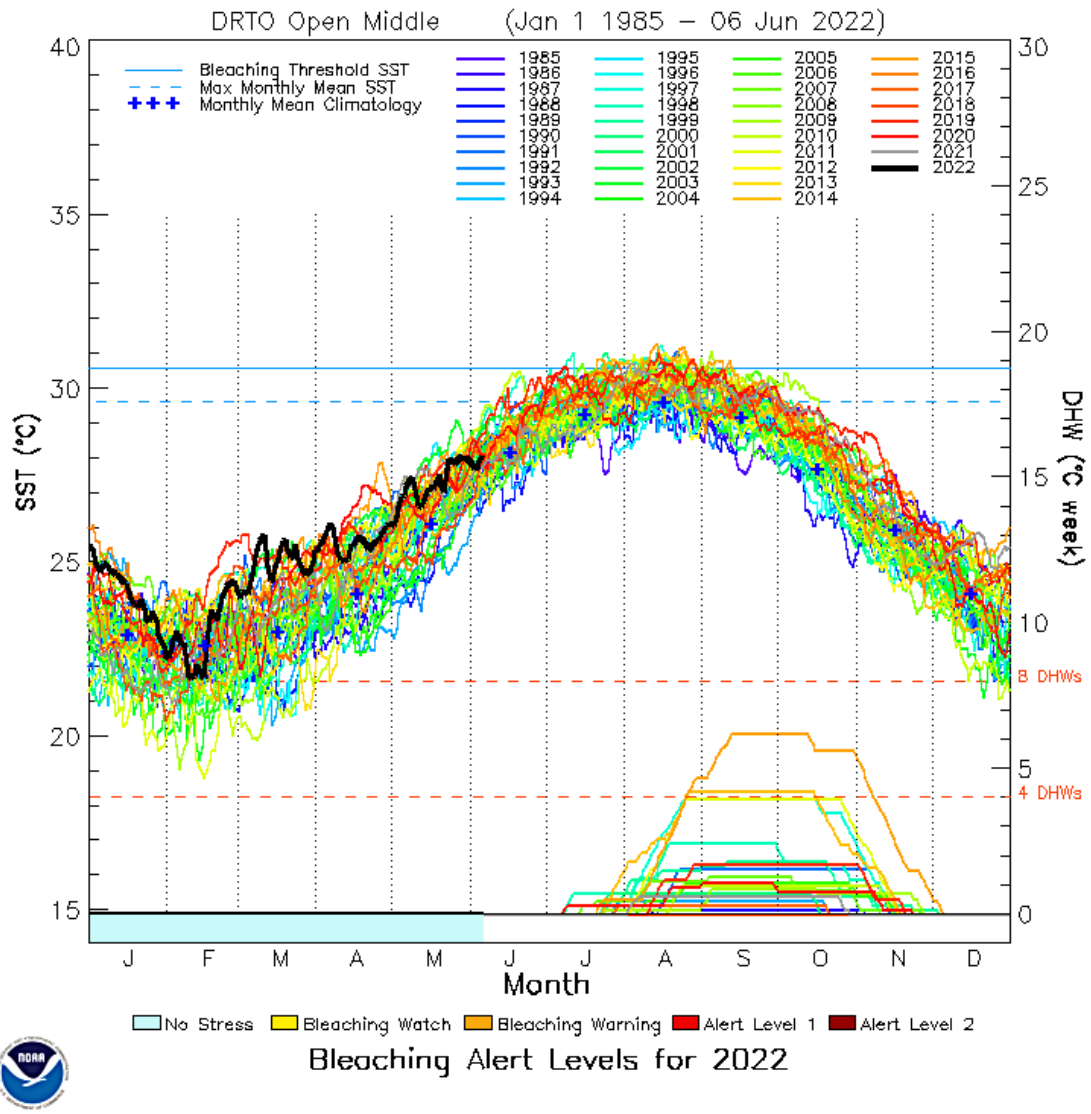


Figure 3. Sea surface temperatures at DRT0 from January 1, 1985 to June 6, 2022 (Source: NOAA, 2022)

The Park is known for its civil era fort, Fort Jefferson, which is located on Garden Key and serves as a home base for field operations. While the Park includes seven small keys, approximately 98% of the Park's square mile area is open water and patch reefs, with several extensive reef features marked by high coral cover, large colony sizes, and presence of ESA-listed coral species.

The remote location of the Park places its reefs in an advantageous position along FCR because it is removed from direct impacts facing northern reefs, such as poor water quality, coastline development, and regular dredging activities. Furthermore, as the westernmost location along FCR, DRT0 was the last location to contract SCTL.

## Reconnaissance

### *Site Selection*

To efficiently locate areas of potential SCTLD outbreak, a susceptibility factor was initially used to identify locations in the Park most likely to exhibit early symptoms of the disease. The “susceptibility factor” ( $S_f$ ) was developed by doubling the density of primary-susceptible colonies (col./10 m<sup>2</sup>) and adding the density of secondary-susceptible colonies (col./10 m<sup>2</sup>) at each site. Abundance Index values ranged from 1 to 154, with numerous sites identified as potential SCTLD monitoring locations ( $S_f > 63$ ). Using the susceptibility factor, along with historical data denoting the presence of ESA-listed species or rare genotypes, increased coral biodiversity, presence of large colonies, presence of long-term monitoring stations, high visitor use, logistical considerations (presence of moorings, ease of access, proximity to Garden Key), biological factors, and partner recommendations, the number of potential reconnaissance sites was narrowed to 30 sites (Table 1; Figure 4). Of these 30 sites, a subset of nine sites were proposed as select sentinel monitoring sites (Table 3) based on  $S_f$ , location within the Park, and importance to Park management. While reconnaissance surveys were repeatedly performed at each of the 30 reconnaissance sites, additional sites were surveyed to ensure greater distribution of reconnaissance throughout the Park.

*Table 1. DRTO SCTLD reconnaissance and monitoring (<sup>†</sup>) sites listed in decimal degrees (DD) with abundance values (\*) and susceptibility factors ( $S_f$ ).*

Site ID	Site Name	$S_f$ (col./m <sup>2</sup> )	Latitude (DD)	Longitude (DD)	Depth (ft)	Depth (m)	Coordinate Source
1	Bird Key Reef <sup>†</sup>	52	24.6117°	-82.8702°	46	14.0	FWC
2	DRTO Open East	126	24.6248°	-82.8350°	39	12.0	Response Plan V.1
3	DRTO Open Middle	127	24.6626°	-82.8128°	18	5.4	Response Plan V.1
4	DRTO Open East*	75	24.6590°	-82.7770°	48	14.6	Response Plan V.1
5	Pulaski	49	24.6918°	-82.7749°	66	20.1	Whaler GPS
6	SV01	83	24.7244°	-82.8147°	42	12.8	Response Plan V.1
7	SE of I63 RNA	87	24.7063°	-82.8805°	41	12.6	Response Plan V.1
8	Hole in the doughnut RNA	113	24.6830°	-82.9260°	59	17.9	Response Plan V.1
9	LH-05 LH Forest RNA*	50	24.6638°	-82.9270°	44	13.4	Response Plan V.1
10	SW of Loggerhead	128	24.6062°	-82.9488°	42	12.9	Response Plan V.1
11	DRTO Open O84	104	24.5736°	-82.9261°	35	10.6	Response Plan V.1

12	Near Hospital RNA	119	24.6435°	-82.8560°	56	17.2	Response Plan V.1
13	Magic Castles* †	58	24.6136°	-82.8709°	18	5.6	FWC
14	North Coal Dock	N/A	24.6297°	-82.8714°	12	3.0	Google Earth
15	South Coal Dock	N/A	24.6261°	-82.8731°	12	3.0	Google Earth
16	Little Africa	N/A	24.6358°	-82.9205°	12	3.0	Response Plan V. 1
17	The Maze †	N/A	24.6090°	-82.9495°	46	14.0	FWC
18	Texas Rock †	N/A	24.6805°	-82.8852°	49	15.0	FWC
19	Prolifera	N/A	24.6207°	-82.8697°	7	2.0	FWC
20	Patch Davis Rock †	N/A	24.6870°	-82.9071°	33	10.0	FWC
21	Mayer's Peak †	N/A	24.6080°	-82.9440°	26	8.0	FWC
22	Bird Key SFCN	N/A	24.6115°	-82.8702°	50	15.2	SFCN
23	Santa's Village 2	N/A	24.7232°	-82.8203°	53	16.2	SFCN
24	Santa's Village 3	N/A	24.7224°	-82.8283°	53	16.2	SFCN
25	Loggerhead Forest 2/LH-04 †	N/A	24.6624°	-82.9274°	54	16.5	SFCN
26	Loggerhead Forest 3/LH-01 †	N/A	24.6681°	-82.9259°	53	16.2	SFCN
27	Loggerhead Forest 4	N/A	24.6631°	-82.9310°	60	18.3	SFCN
28	Loggerhead Forest 5/LH-03 †	N/A	24.6598°	-82.9341°	53	16.2	SFCN
29	Windjammer	N/A	24.6244°	-82.9427°	22	6.8	NPS
30	Moat Wall	N/A	24.6275°	-82.8740°	12	3	Google Earth

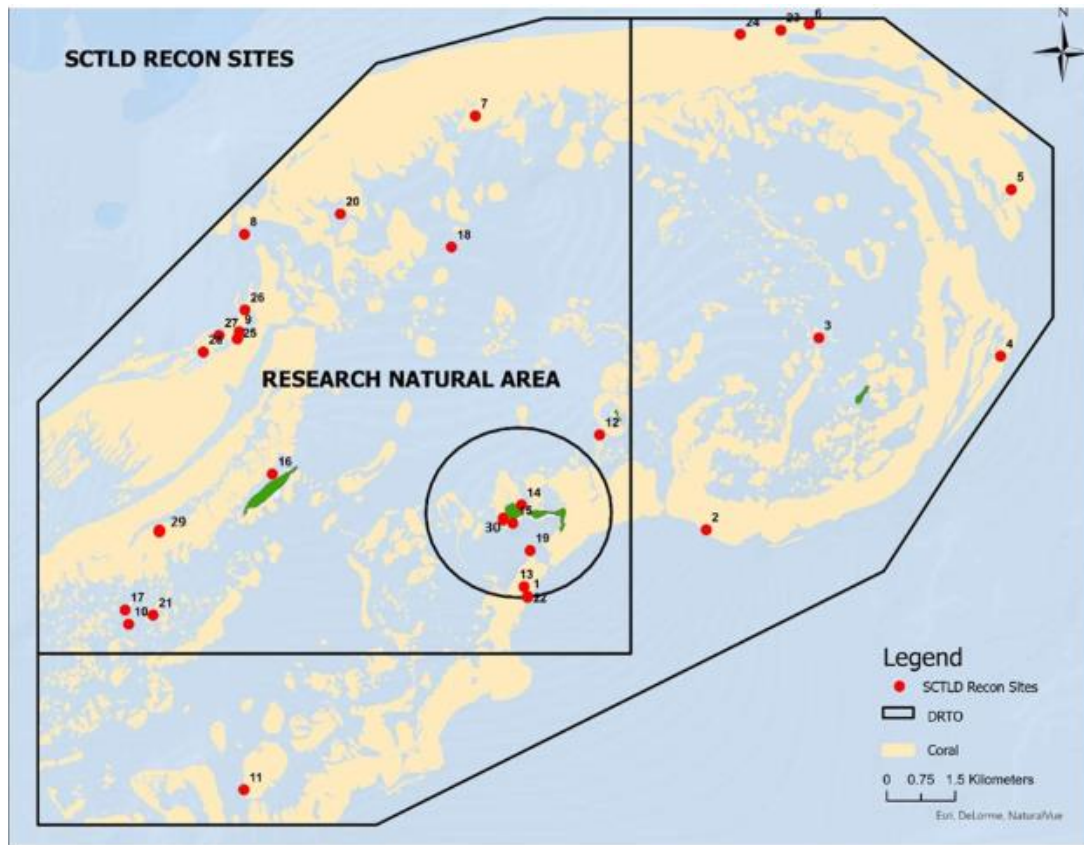


Figure 4. Map of SCTLD reconnaissance and monitoring sites in relation to Park boundary. Site numbers correspond to Table 1.

### Protocol

Reconnaissance surveys occurred in two phases. The first phase between January and December 2021 identified and tracked appearance and progression of the disease outbreak while establishing baseline datasets. Surveys took place at each site approximately once per month, although survey frequency varied at some locations. Once SCTLD was identified at a site, focus shifted to intervention at that location while reconnaissance surveys continued at other sites. The second phase of reconnaissance surveys between March and June 2022 surveyed all 30 sites approximately one year later to document changes in coral community composition that occurred since the initial outbreak.

Reconnaissance surveys were performed as roving diver surveys for a minimum of 15 minutes. Divers recorded the number of colonies of susceptible species according to size class (> 10 cm, 10-30 cm, > 30 cm) and health status (healthy, SCTLD, paling/bleaching, other disease). Because survey times varied, in some cases extending well beyond the minimum 15 minutes, all data were adjusted for time prior to analysis (DRTO SCTLD Response Plan, 2021).

## Intervention

### *High-Priority Site Selection Criteria*

While intervention was performed at any diseased location and not limited to reconnaissance sites, three locations were deemed high-priority intervention sites: Magic Castles, Bird Key Reef, and Loggerhead Forest. In addition, intervention focus was allocated to BACI impact sites: The Maze and Davis Rock.

Magic Castles is home to nine large *D. cylindrus* (ESA-listed) colonies located within approximately 0.8 km<sup>2</sup> of reef (Figure 5) at a depth of approximately 6 m to 9 m. The health and protection of these rare charismatic corals has historically been a priority for Park management and researchers, being annually monitored by FWC and regularly visited by NSU. Targeted intervention of this site occurs approximately once every two to three weeks (maximum period between treatments: 12 weeks). Since colonies are within swimmable distance of each other (approximately 1-19 meters between colonies), an underwater map with bearings and distances between numbered colonies is used to navigate the site. Although treatment applied at Magic Castles is primarily focused on *D. cylindrus* colonies, the surrounding reef is also treated to reduce transmission and/or pathogen load on adjacent reef.



Figure 5. DRT0 NPS diver surveys a *Dendrogyra cylindrus* colony at Magic Castles.



Bird Key Reef is a large spur-and-groove reef feature with high abundance of susceptible coral species at a depth between approximately 8 m to 18 m. The coral community composition includes *M. cavernosa*, *O. franksi*, *O. faveolata*, *C. natans*, and *P. strigosa*, as well as relative abundances of other coral species. The feature was roughly divided into a northern component approximately 0.9 km<sup>2</sup> (Figure 6A) and a southern component approximately 1.0 km<sup>2</sup> (Figure 6B). The site is monitored annually by SFCN and FWC at permanent transect locations and therefore has an established baseline of demographic data. Its proximity to Garden Key provides for relative ease of access. From September 1 – 9, 2021 a SCHMIR mission was performed by a collaborative team (n = 10) from NSU and FAU-HBOI at a depth ranging from 9 m to 18 m, followed by periodic maintenance intervention targeted to occur every month. This intervention tactic employed a map overlaid with a gridded cell system in conjunction with systematic swim patterns and GPS tracking via attachment to a dive flag (Figures 6 and 7). This method guided treatment of the entire reefscape.

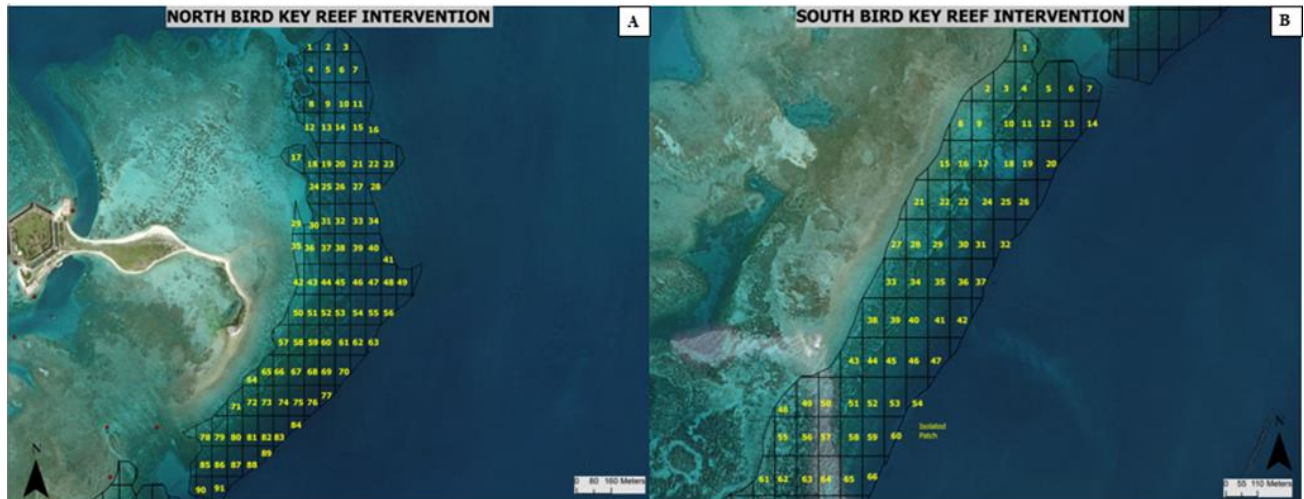


Figure 6. Regions of high coral cover at North (A) and South (B) sections of Bird Key Reef with gridded overlays for systematic treatment during SCHMIR mission and maintenance intervention dives.

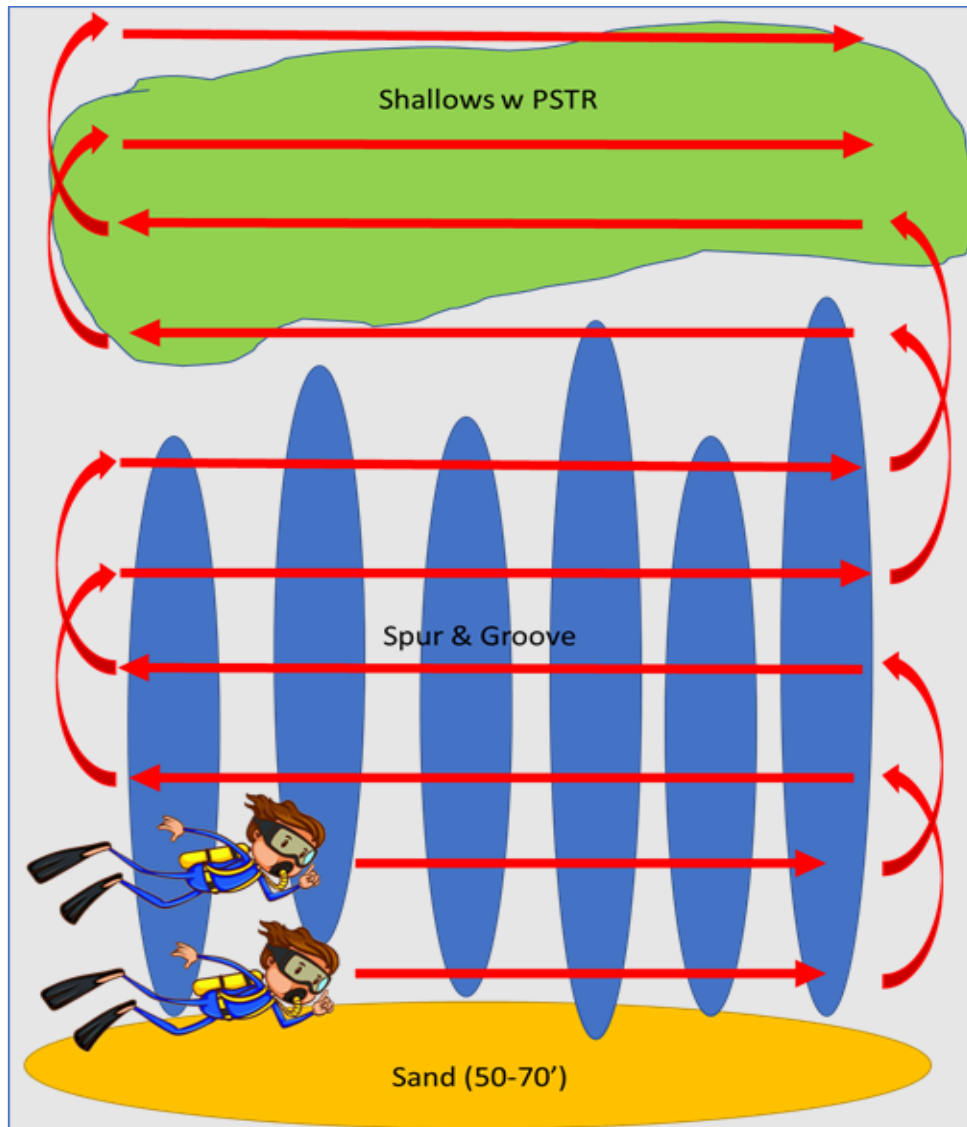


Figure 7. Systematic swim pattern within gridded cell systems for coordinated intervention tactics. Divers follow deepest section of reef and move toward shallower sections to cover large areas and ensure complete treatment. (Source: Karen Neely, NSU, 2021)



Loggerhead Forest is an expansive reef feature north of Loggerhead Key known for high coral cover and large colonies at a depth of 17 m to 21 m. The dominant coral species are *O. franksi*, *O. faveolata*, and *C. natans*. The reef feature delineated for survey and intervention is approximately 6.1 km<sup>2</sup> (red gridded polygon; Figure 8) with approximately 3.3 km<sup>2</sup> characterized by medium to high relief hardbottom (green polygon; Figure 8). Like Bird Key Reef, it is monitored annually by SFCN and FWC at permanent transects and therefore has established baseline data. While this site experiences seasonal white plague outbreaks, it was among the last of the reconnaissance sites to contract SCTLD. From June 12-14, 2022, a second SCHMIR mission was performed by a collaborative team (n = 9) from NSU, FAU-HBOI, and UVI. The effort targeted the northern reef edge at a depth ranging from 16 m to 21 m with high coral cover and high SCTLD prevalence (Figure 8). The trip was cut short due to equipment malfunction but will be rescheduled and supported by maintenance intervention efforts.

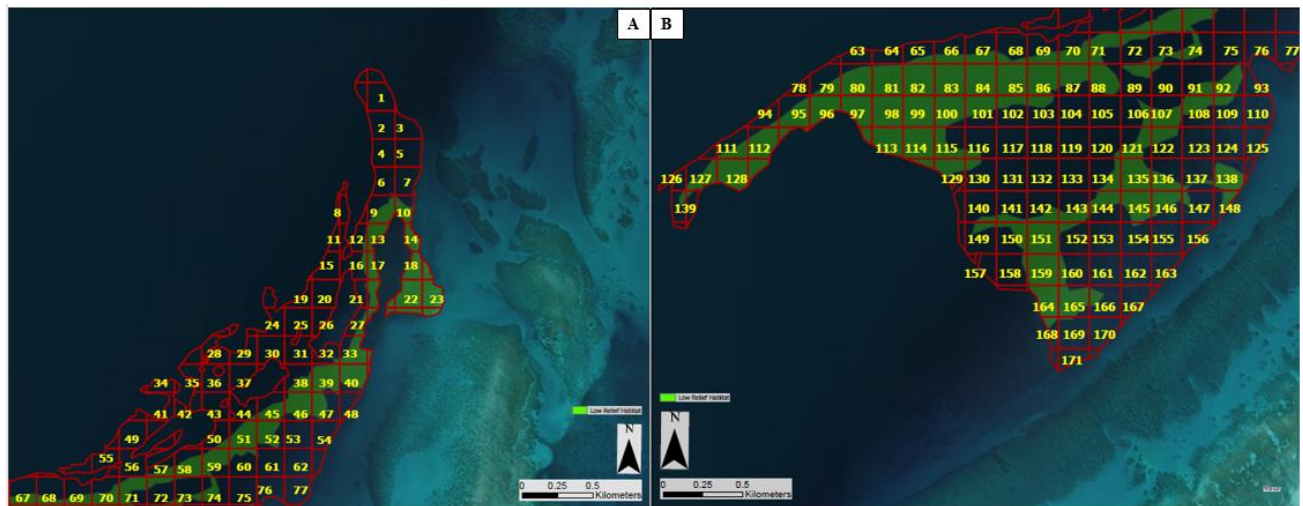


Figure 6. Regions of high coral cover at North (A) and South (B) sections of Loggerhead Forest with gridded overlays for systematic treatment during SCHMIR mission and maintenance intervention dives.

### Protocol

Intervention entails the topical application of antibiotic treatment to the disease margin of infected corals by divers (Figure 9). Treatment consists of powdered amoxicillin and Base2b mixed in a 1:8 ratio, which has been developed and supported as an effective intervention strategy to aid stony corals with SCTLD, significantly reducing the likelihood of mortality at the colony level (Neely et al., 2020). Treatment is administered by divers wielding treatment-packed caulking tubes, which make for efficient treatment transport and application methods (Figure 9).



*Figure 7. DRTN NPS diver applies amoxicillin and Base2b treatment to disease margin on a Pseudodiploria strigosa colony.*

Intervention dives at priority sites aimed to achieve as near-to-complete treatment as possible at the site level, employing underwater navigation (Magic Castles) or use of gridded map systems (Bird Key Reef, Loggerhead Forest). At all intervention sites, GPS tracks were gathered along treatment routes. Using currents during drift dives made for opportunistic treatment efficiency, allowing divers to cover large swaths of reef in relatively short periods of time. Data collected during intervention dives included number of colonies of susceptible species treated according to size class.

## Coral Rescue

Prior to the arrival of SCTLD at DRTO, two coral rescue missions were conducted in collaboration with partners from FWC and a multitude of AZA-accredited facilities. In July 2019 and May 2020, a total of over 600 corals from 18 different species were rescued from DRTO and brought to these facilities for study, propagation, and to serve as a genetic bank for species highly susceptible to SCTLD. These corals and their fate are not included within the scope of this project. However, after finding the first SCTLD-infected colony within Park boundaries in May 2021, a routine reconnaissance survey led to the discovery of a novel *D. cylindrus*-populated site on the east side of the Park (Figure 10). The new site was named Dendro City (Figure 11) due to relatively high abundance of at least 23 *D. cylindrus* colonies within the area. The colonies were located at a depth of 6 m to 11 m and were smaller than the colonies that had been previously sampled at Magic Castles, with an estimated average size of 60 cm x 60 cm x 60cm.

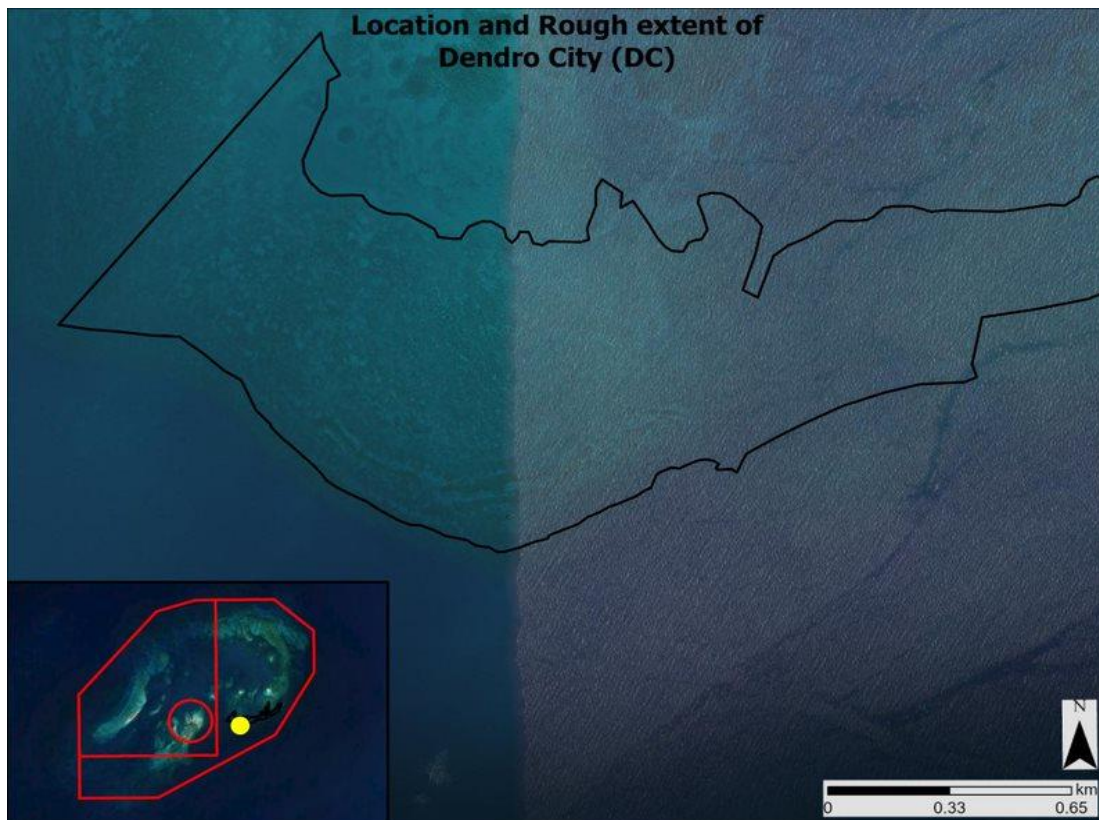


Figure 8. Map of Dendro City in relation to Park boundaries, showing location of the Coral Rescue mission to collect *Dendrogyra cylindrus* fragments.

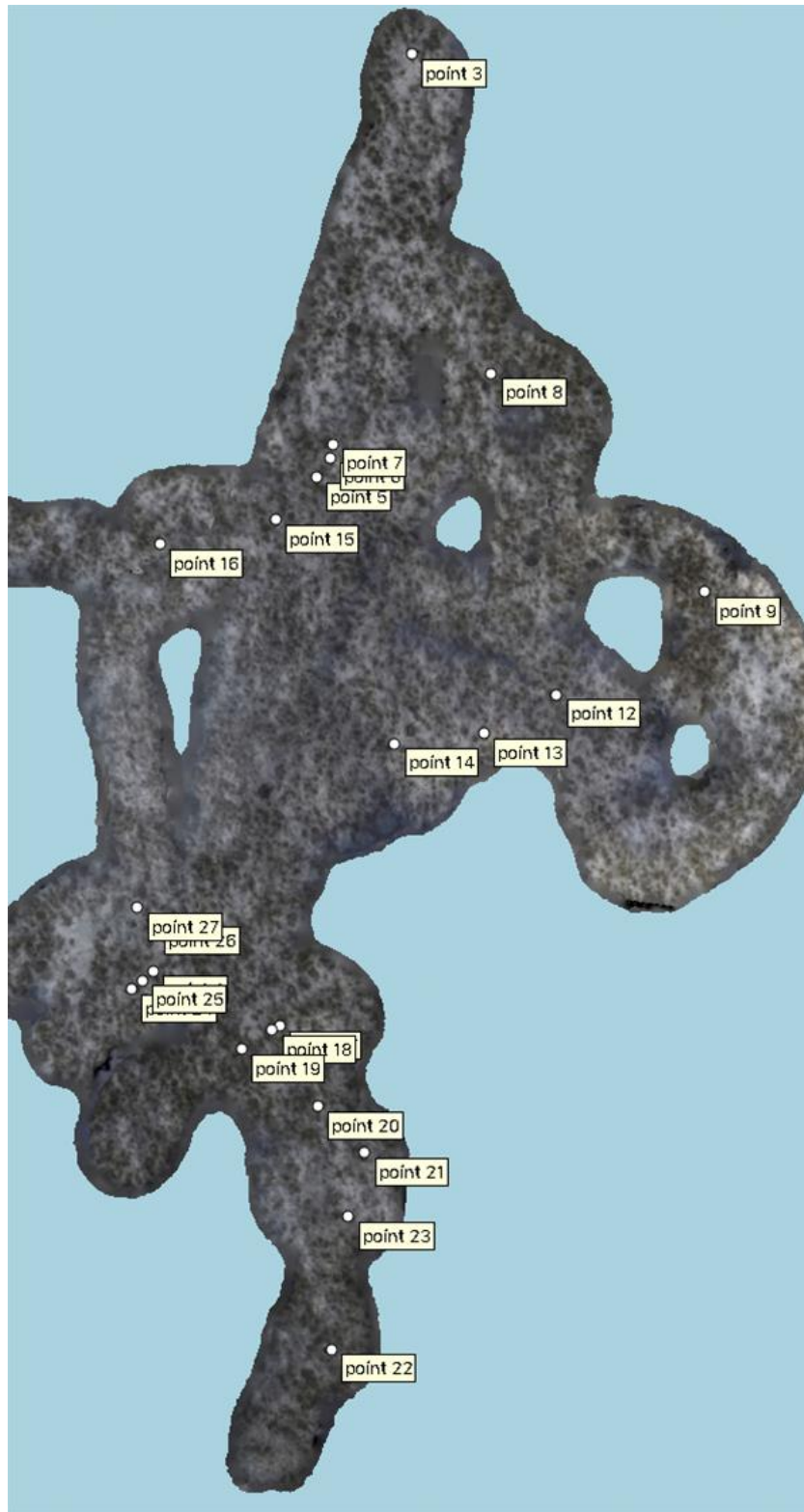


Figure 9. Orthomosaic of Dendro City showing locations of the 23 *Dendrogyra cylindrus* colonies.



While fragments from the *D. cylindrus* colonies at Magic Castles had been previously collected, research indicated that the colonies were all from a single genotype. Significant losses to *D. cylindrus* populations across FCR (Neely et al., 2021a) warranted a third coral rescue mission to potentially identify and rescue new genotypes. This action was further justified by novelty of the discovery and the distance between Dendro City and Magic Castles, potentially indicating that these newly found colonies might have unique genotypes. A rescue mission was conducted in August to collect fragments from Dendro City (n = 3) and Lone Castle (n = 1; also previously unsampled). Colonies with little to no disease were selected for collection to increase likelihood of a successful rescue. Unfortunately, despite targeted intervention efforts, Dendro City succumbed to mortality from SCTLD shortly after rescue efforts were completed.

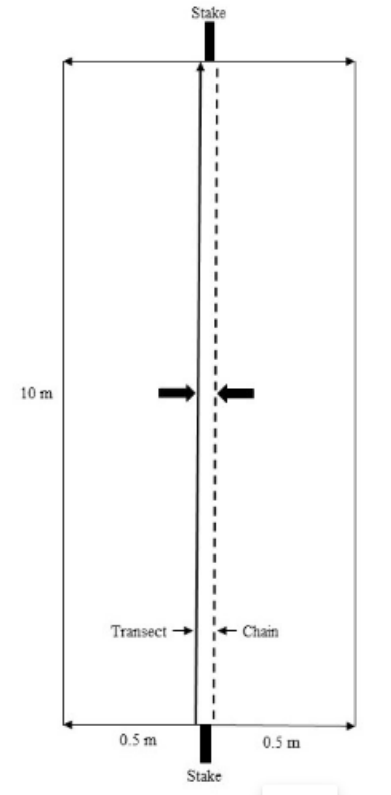
Fragments were transported by NPS in holding tanks to Key West, then transported by FWC to the Phillip and Patricia Frost Museum of Science in Miami, FL. Fragments were given a prophylactic ampicillin treatment and isolated for six weeks before being successfully transferred into general holding in September. FWC collected samples from the fragments for genotyping in December 2021 and results are pending. This information will assist with identification of approximately 192 single nucleotide polymorphism markers (SNPs) to lend greater precision in genotyping species.

## **Monitoring**

### *Coral Demographic Surveys*

To investigate potential changes in coral demographics since the disease outbreak, a subset of nine of the 30 reconnaissance sites were selected based on presence of permanent transects and historical baseline data established by FWC and SFCN, and for their wide distribution across the Park (Table 1, Figure 4). These sites were selected in consultation with SFCN I&M and FWC as locations for routine demographic surveys and repeatable time-series photos. Demographic monitoring surveys took place between April and May 2021 on FWC and SFCN's permanent transects. Surveys are scheduled to be repeated in November 2022 and May 2023.

Survey methods have been adapted from FWC's DRM surveys and CREMP procedure. Belt transects are set up between permanent stakes and a 1x10-meter area is surveyed for coral demographic data (Figure 12). For all corals  $\geq 4$  centimeters, species, size (maximum diameter and maximum perpendicular height), percent of old mortality, number of tissue isolates, and coral health condition (type, distribution, percent affected, and identification) are recorded.



*Figure 10. Belt transect for demographic surveys runs between permanent stakes with a chain underneath to demarcate center. Transect can be divided using a brass clip (arrows). (Source: DRTTO SCTLD Response Plan, 2021)*

### *SRC NPS Photogrammetry*

DRTO NPS partnered with SRC to accomplish large-scale photogrammetry surveys at the nine monitoring sites another sites of interest (Dendro City, Windjammer). Photogrammetry surveys were conducted with the multi-camera system SeaArray and surface buoy system, allowing generation of 3D underwater visualization where individual images are linked to global positioning data (Figures 14 and 15). The SeaArray can cover approximately 10,000 m<sup>2</sup> of a reef feature within a single 180-minute dive. This new technology serves as an advanced method of creating high-resolution orthomosaics at large scales, providing an innovative approach to natural resource monitoring by allowing for geographic positioning of important reef features and measuring large-scale ecological change over time. Surveys were conducted in two phases: between April and August 2021 prior to SCTLD arrival and in April 2022 approximately one year after infection. Each phase included over 24 hours of underwater dive time to conduct large-scale surveys (approximately 100 m x 100 m) at each site.



Figure 11. NPS SRC diver images reef at Loggerhead Forest using the SeaArray.

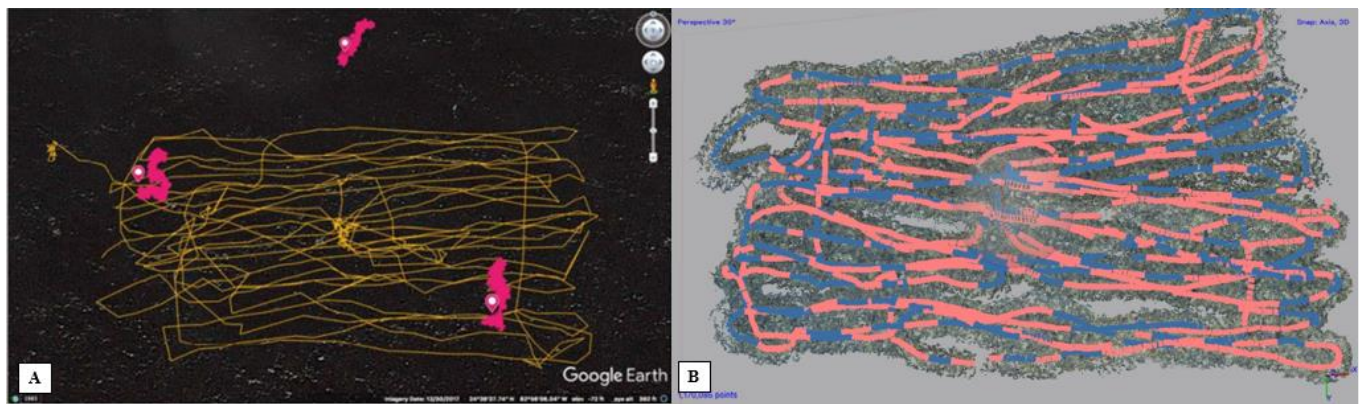
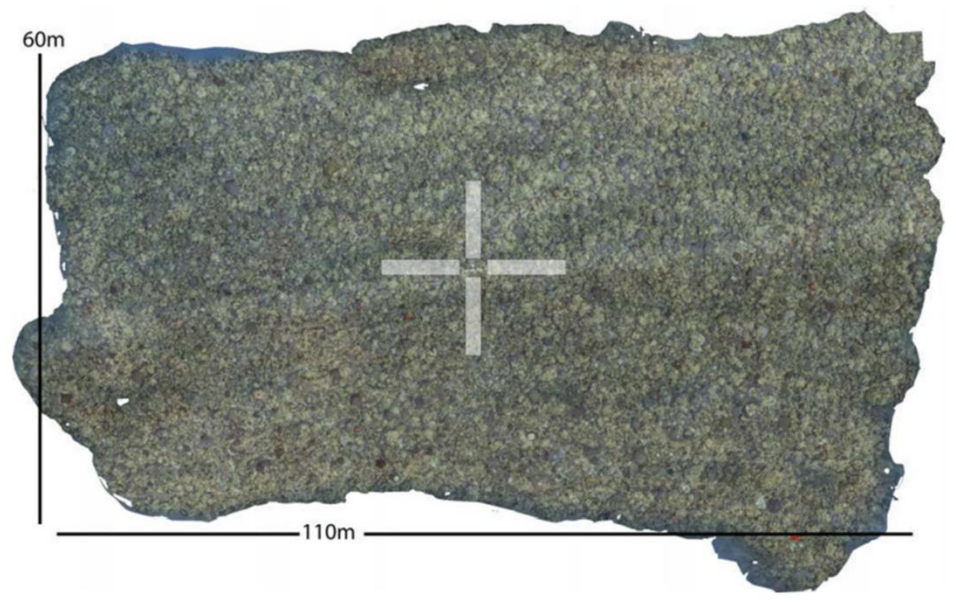


Figure 12. Output from SeaArray. (A) Initial path of SeaArray (yellow) where corals are imaged in relation to surface buoy system (pink) interacting with GPS satellites. (B) Overlay of image collection along underwater imaging path (blue and red), prior to mosaic referencing. (Source: NPS SRC, 2021)



*Figure 13. Example of final output from SeaArray, where images and individual georeferencing data have been stitched into an orthomosaic. (Source: NPS SRC, 2021)*

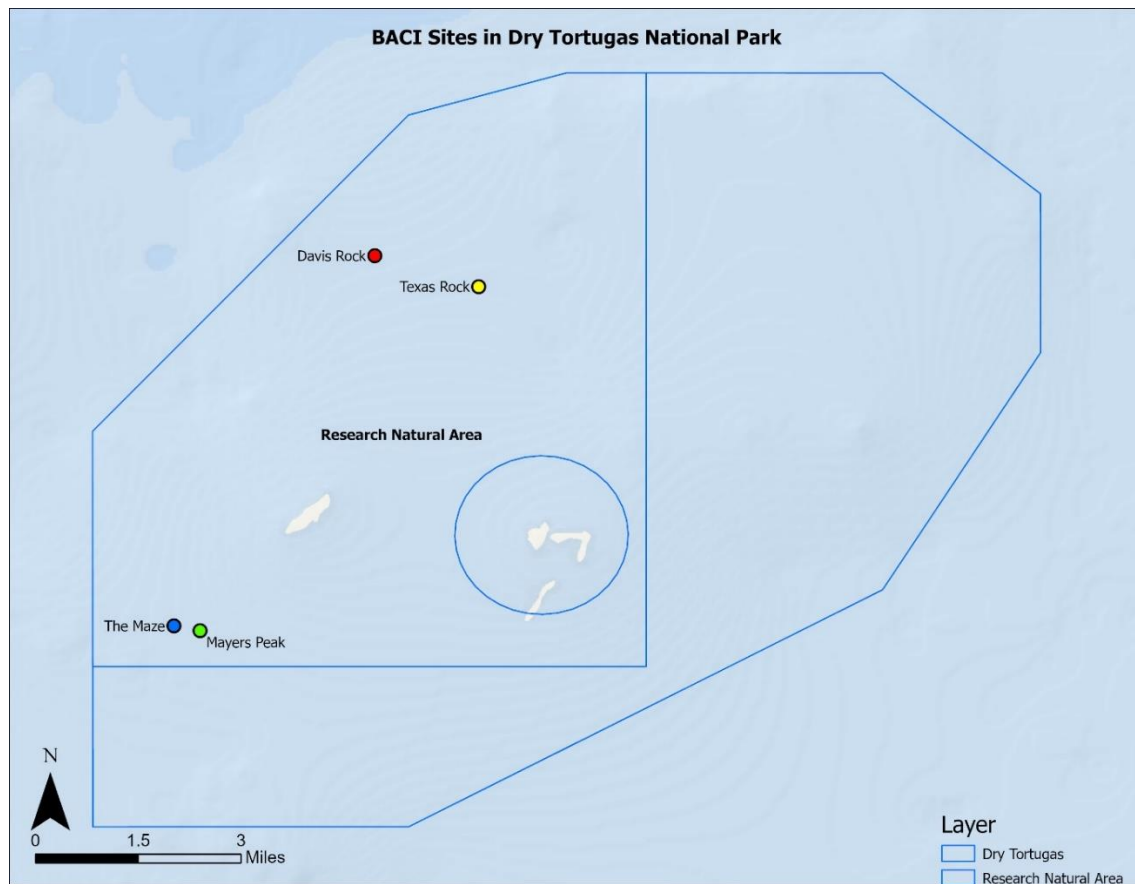
#### *DRTO NPS Photogrammetry*

The NPS staff at DRTO are currently developing their own photogrammetry program to perform frequent in-house photogrammetry surveys at monitoring sites. This program will provide insight into changes in coral communities independently of, but complementary to, SRC’s large-scale SeaArray surveys. DRTO photogrammetry surveys will be systematically conducted but not limited to the permanent transect locations at the nine monitoring sites where demographic surveys are conducted. In addition to maximizing efficiency through implementation of more rapid survey methods, the addition of photogrammetry surveys to monitoring protocols may provide a valuable comparison with demographic survey methods.

#### *BACI Study*

A subset of four of the nine monitoring sites was selected as patch or “pinnacle” reefs to be isolated and used in a Before After Control Impact (BACI) study: The Maze, Texas Rock, Davis Rock, and Mayers Peak (Table 1; Figure 16). Sites were chosen as comparable pairs with replication, where two sites serve as impact sites (intervention is conducted; The Maze and Davis Rock) and two sites serve as controls (intervention is not conducted; Texas Rock and Mayers Peak).





*Figure 14. Location of BACI impact and control sites within DRTO.*

Sites were chosen based on their close comparability in factors such as depth, coral demographics, and presence of historical data to minimize potential confounding variables between impact and control site comparisons.

The BACI study will assess the efficacy and impacts of SCTLD treatment across large, reef-level scales, providing valuable insight for current and future disease intervention management approaches. Diver-collected demographic data from permanent transects, SRC NPS photogrammetry, and DRTO NPS photogrammetry will be used to make comparisons before versus after SCTLD and between control (untreated) versus impact (treated) sites.

## Statistical Analyses

### *Project Effort*

The total number of hours underwater, dive days, and dives were calculated for reconnaissance (as of June 14, 2022), intervention (as of May 21, 2022), and monitoring (from 2021) activities. Hours underwater were calculated as man hours (hours totaled per person). Dive days were summed for any days that project-related diving or snorkeling was conducted. Dives were quantified per person and include project-related snorkeling.

### *Reconnaissance*

To summarize baseline data prior to SCTLD, coral colony abundance was calculated as the average number of corals observed per minute prior to the first observation of SCTLD in the Park on May 29, 2021. The mean number corals observed per minute  $\pm$  standard error was calculated by site (for 30 reconnaissance sites; Table 1) and by species and size class (averaged across 30 reconnaissance sites). Data were not homogenous and could not successfully be transformed, so non-parametric Kruskal-Wallis rank sum tests were used to determine the effects of site, species, and size on the number of healthy corals observed per minute. Post hoc tests were conducted with Bonferroni correction and P values were multiplied by the number of comparisons in the respective analyses.

To compare coral colony abundance before and after SCTLD, the mean number of corals observed per minute was calculated prior to the first SCTLD observation at each of the 30 reconnaissance sites (before SCTLD) and from the date of initial disease observation at each site to June 14, 2022 (after SCTLD). The mean number of corals observed per minute  $\pm$  standard error was calculated by site, species, and size class. One outlier from the pre-SCTLD data was removed from Site 6 due to an excessive amount of *M. cavernosa* colonies observed per minute by one diver. Due to the recent acquisition of reconnaissance data after SCTLD and a lack of replication, no further statistical analyses were performed on these data.

### *Intervention*

The total number of coral colonies treated was calculated for five priority sites: Bird Key Reef, Magic Castles, Loggerhead Forest, The Maze, and Davis Rock. For Bird Key Reef, totals were summed from Sites 1, 22, and other miscellaneous sites treated at Bird Key. For Loggerhead Forest, totals were summed from Sites 9 and 25-28 (Table 1). The top species observed and treated at each of these five priority sites was identified and the respective percentage of the total was calculated. The total number of colonies treated was also calculated by species and size class (summed across 30 reconnaissance sites; Table 1). All intervention totals include any retreatments of the same coral colony.

## RESULTS

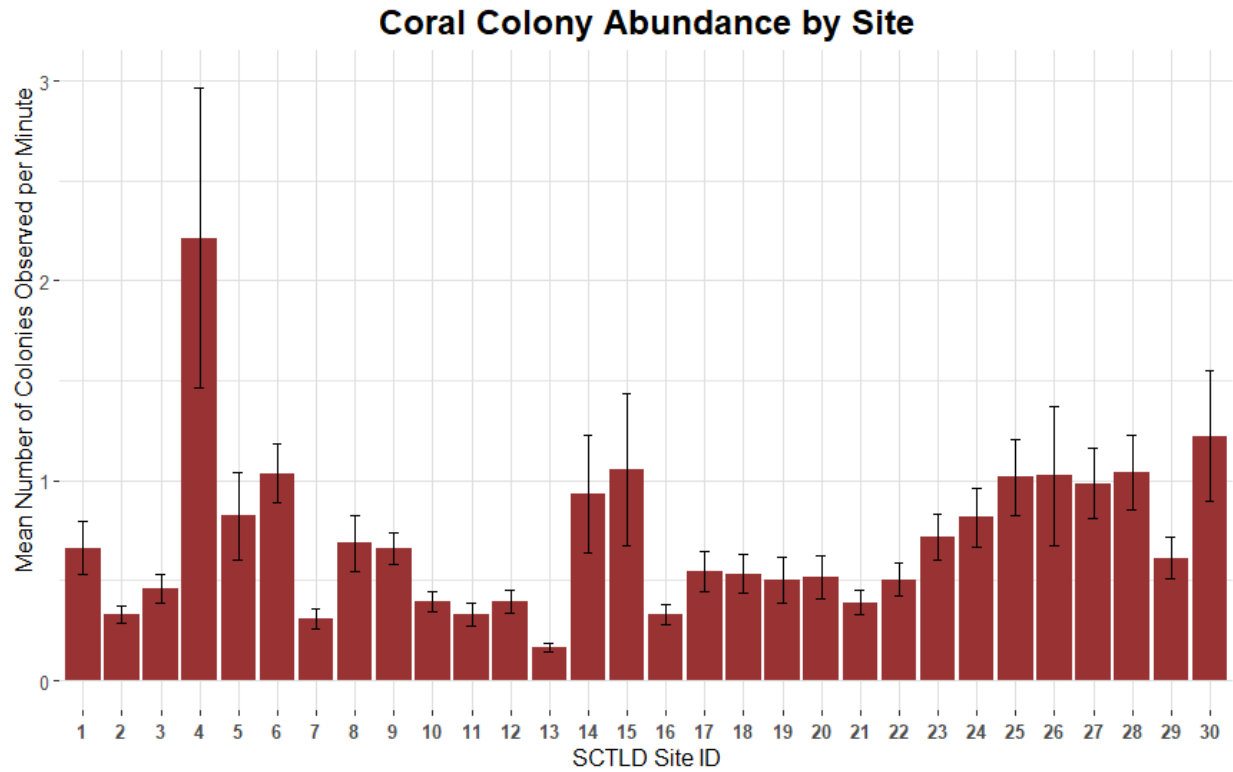
### **Project Effort**

Park staff and partners have completed a grand total of 505 hours underwater for this project. 300 of these hours were dedicated to intervention of SCTLD. Hours underwater were calculated as person hours (hours totaled per person) and are most indicative of effort as they reflect time spent working directly toward project goals. In addition to hours underwater, a grand total of 149 dive days and 848 dives per person have been achieved. The total effort for this project was summed for reconnaissance, intervention, and monitoring work.

### *SCHMIR Missions*

The two SCHMIR missions were excluded from the effort analysis above. However, we provide brief descriptive summaries from those missions to acknowledge the immense work and coordination between the respective projects and to further highlight the synergy between the efforts. The first SCHMIR mission, from September 1 – 9, 2021 was implemented by a team of 10 researchers. The team conducted a total of 265 dives equating to over 299 underwater person hours of work at Bird Key Reef. A total of 6,038 corals were treated, more than doubling the total number of treated corals throughout the whole of FCR since intervention began in late 2018. During the second SCHMIR mission, from June 12 – 14, 2022, a team of nine researchers treated a total of 2,817 corals within approximately 0.05 km<sup>2</sup> at Loggerhead Forest. A portion of the team (n = 4) returned to the Park from June 17 – 18, 2022, and treated additional corals along the Moat Wall (n = 376) and Coal Docks (n = 225).

## Reconnaissance



*Figure 15. Mean number of susceptible coral colonies observed per minute among 30 reconnaissance sites prior to first SCTLD observation at DRT0. Site numbers correspond to Table 1 and Figure 4. Susceptible species listed in Figure 18, below.*

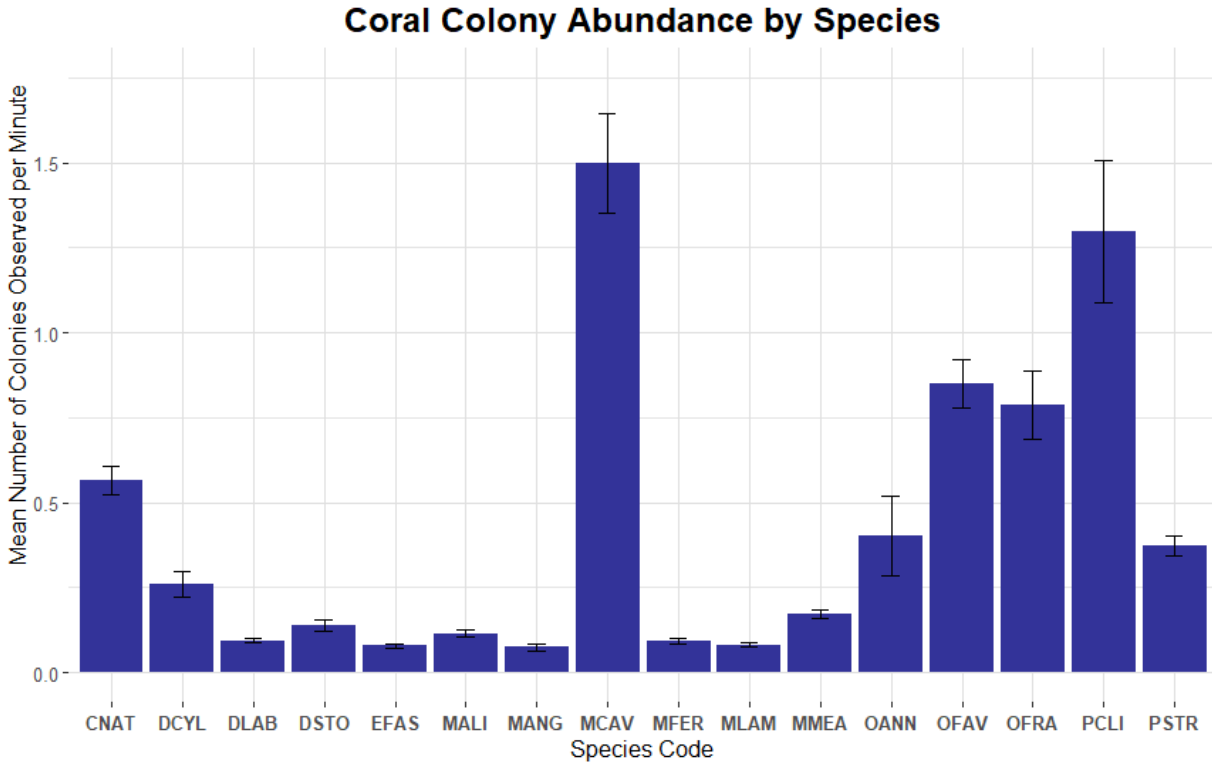


Figure 16. Mean number of susceptible coral colonies by species observed per minute among 30 reconnaissance sites prior to first SCTL observation at DRT. Species codes include *Colpophyllia natans* (CNAT), *Dendrogyra cylindrus* (DCYL), *Diploria labyrinthiformis* (DLAB), *Dichocoenia stokesii* (DSTO), *Eusmilia fastigiata* (EFAS), *Mycetophyllia aliciae* (MALI), *Mussa angulosa* (MANG), *Montastraea cavernosa* (MCAV), *Mycetophyllia ferox* (MFER), *Mycetophyllia lamarckiana* (MLAM), *Meandrina meandrites* (MMEA), *Orbicella annularis* (OANN), *Orbicella faveolata* (OFAV), *Orbicella franksi* (OFRA), *Pseudodiploria clivosa* (PCLI), *Pseudodiploria strigosa* (PSTR).

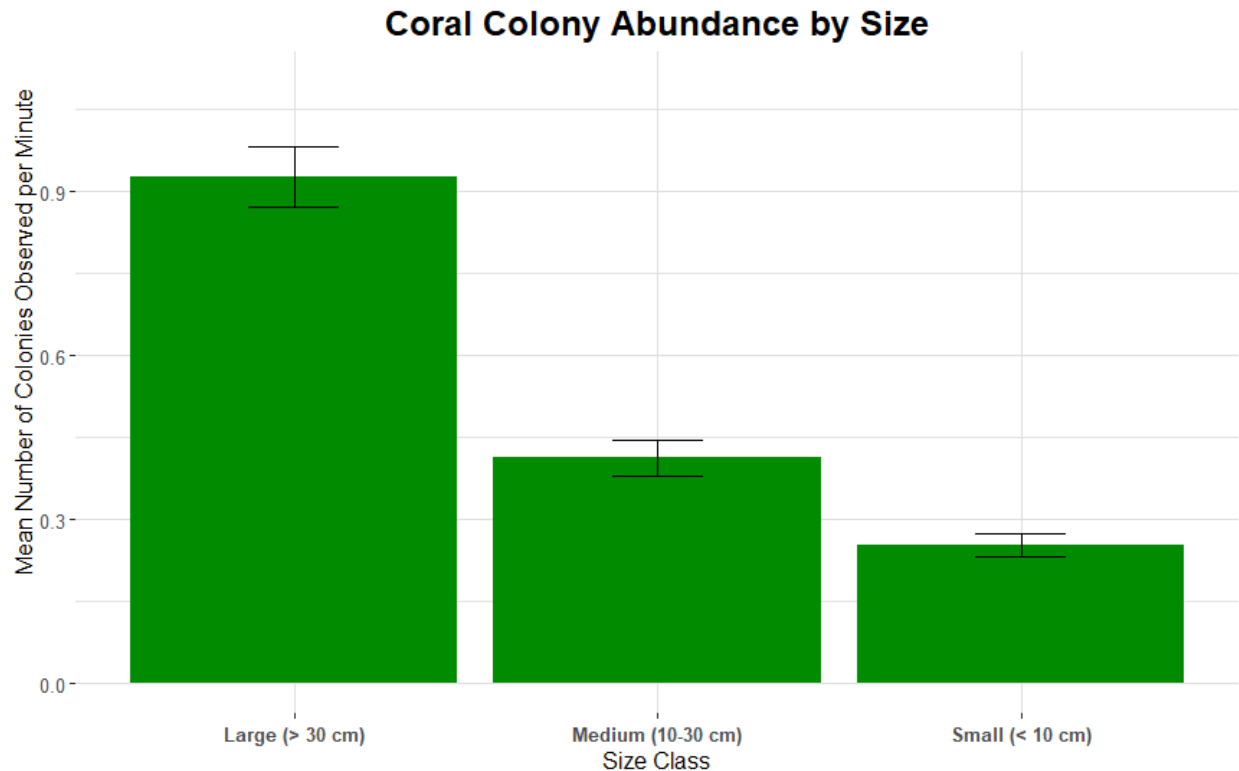


Figure 17. Mean number of susceptible coral colonies by size class observed among 30 reconnaissance sites prior to first SCTLD observation at DRT0. Susceptible species listed in Figure 18.

#### Baseline Data Before SCTLD

Baseline reconnaissance data collected prior to the onset of SCTLD showed a significant difference in coral colony abundance by site ( $p < 0.001$ ,  $\alpha = 0.05$ ,  $df = 29$ ,  $H = 118.7$ ; Figure 17). A post hoc test with Bonferroni correction revealed a significant difference between Site 13 and all sites except Sites 7, 11, 15, and 30. Results also showed a significant difference between Site 6 and both Sites 11 and 12. Differences between Magic Castles and the other 25 sites are likely due to the nature of surveys conducted at this site, which are focused primarily on locating the nine *D. cylindrus* colonies. The site requires longer survey times to collect data on fewer coral colonies, thus reducing the mean number of colonies observed per minute. Baseline data similarly showed a significant difference in coral colony abundance by species across all 30 sites ( $p < 0.001$ ,  $\alpha = 0.05$ ,  $df = 15$ ,  $H = 680.9$ ; Figure 18). A post hoc test with Bonferroni correction revealed many significant results, highlighting differences in overall species composition across the Park. The most abundant species were *M. cavernosa*, *P. clivosa*, *O. faveolata*, *O. franksi* and *C. natans*, which were observed an average of at least once every 2 minutes. Data also showed a significant difference in coral colony abundance by size ( $p < 0.001$ ,  $\alpha = 0.05$ ,  $df = 2$ ,  $H = 122.8$ ; Figure 19). A post hoc test with Bonferroni correction indicated all size classes were significantly different from one another. The most frequently observed size class was large colonies, followed by medium and small colonies (Figure 19).

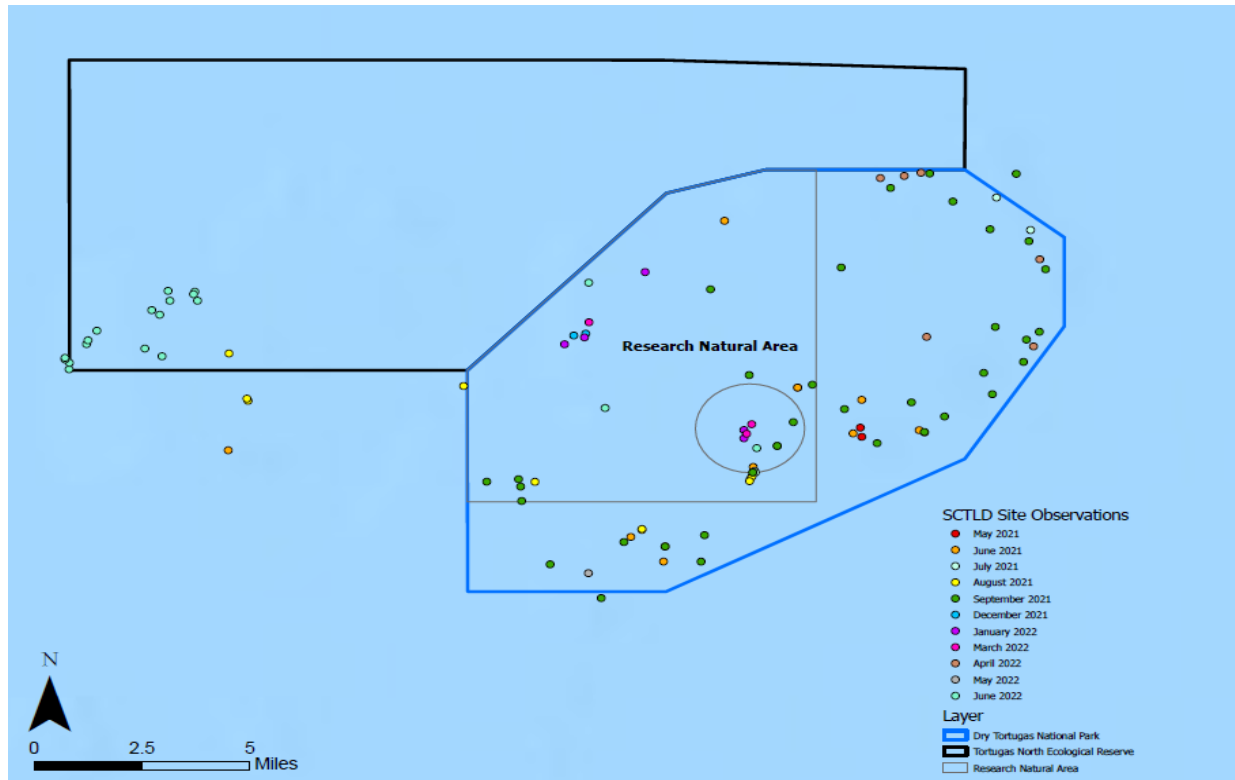


Figure 18. SCTLD progression throughout DRTN from first appearance in May 2021 to present.

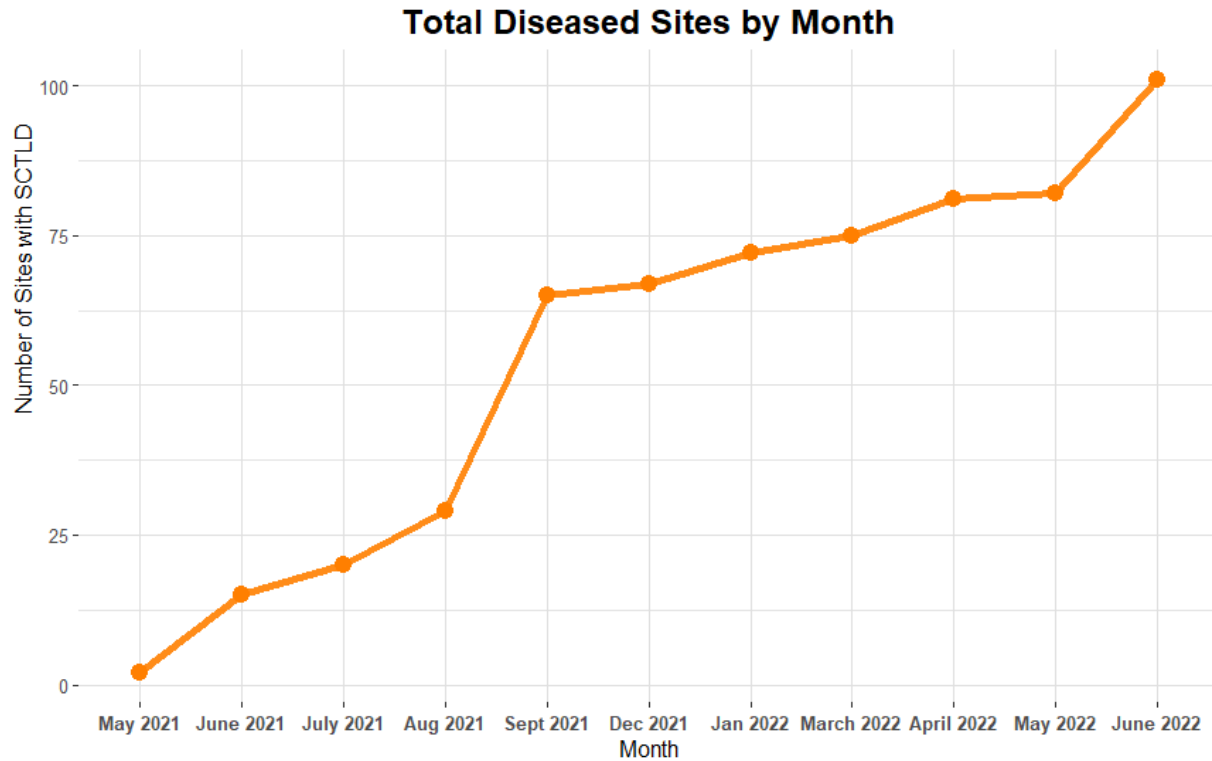


Figure 19. Number of sites within the Park observed with SCTLD since first observation in May 2021. Site numbers are based on data from NPS DRTTO reconnaissance surveys, NRCRMP surveys, and DRM research cruises.

### *SCTLD Progression & Distribution*

SCTLD was first observed in the Park at Site 2 on May 29, 2021, on a *Meandrina jacksoni* colony. As predicted by models, the disease first appeared on the east side of the Park (Table 1, Figure 4). Disease intervention began immediately, and extensive reconnaissance data was promptly collected in the surrounding area (Sites 2, 12, and Dendro City). Within one month of initial observation, SCTLD had been observed at 15 sites (Figures 20 and 21). The disease has since spread steadily across the Park and surrounding area. From August to September 2021, a sharp increase in diseased sites occurred and SCTLD was documented at 36 new sites in one month (Figures 20 and 21). By December 2021, SCTLD was documented at all nine monitoring sites and was relatively prevalent in all regions of the Park. As of June 2022, SCTLD has been documented at 101 sites, including all 30 reconnaissance sites, and the disease is estimated to be present and/or have impacted all the Park's coral reefs (Figures 20 and 21).



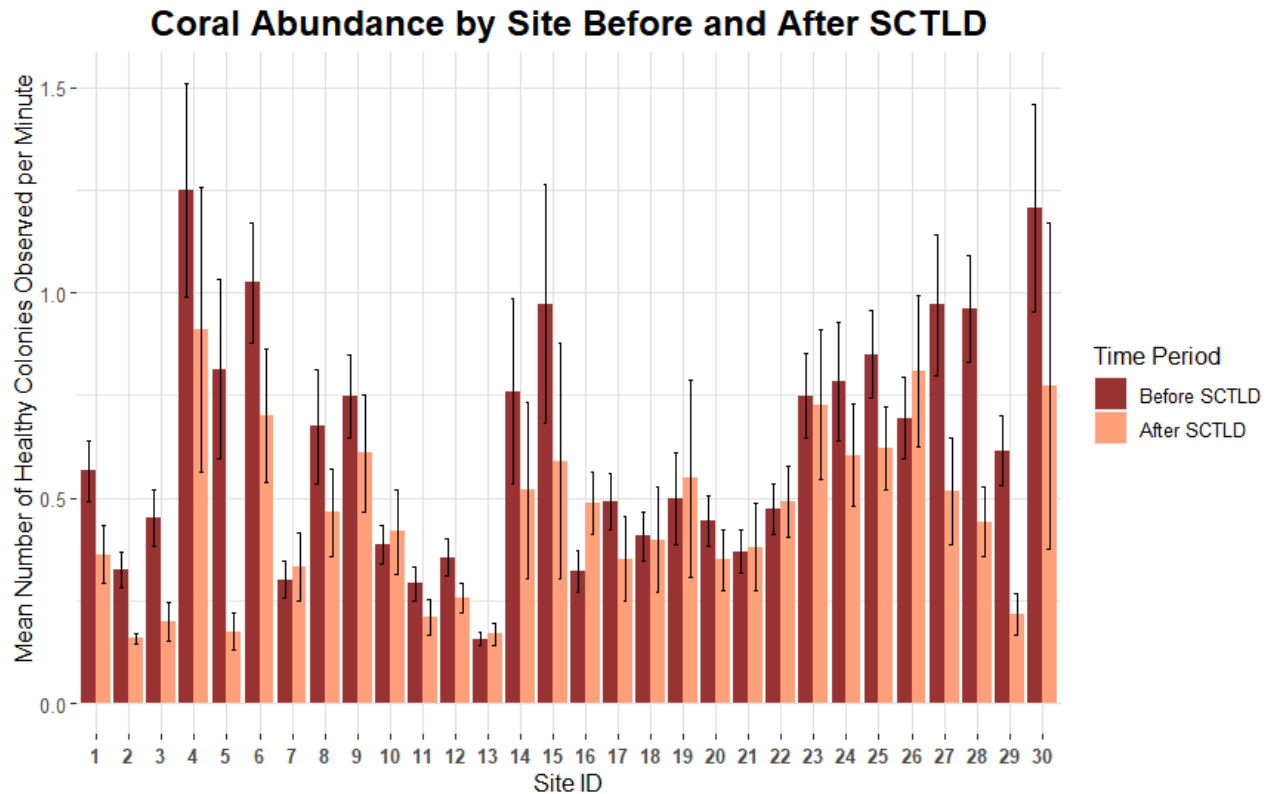


Figure 20. Comparison of mean number of healthy susceptible coral colonies by site observed per minute before (dark red) versus after (peach) SCTLD outbreak. Site numbers correspond to Table 1 and Figure 18. Susceptible coral species listed in Figure 23, below.

### Coral Abundance Before & After SCTLD

Healthy coral colony abundance generally decreased following the onset of SCTLD (Figures 22, 23, and 24). However, these results are based solely on observation of trends in the data rather than statistical tests and should be interpreted as such. Furthermore, due to a lack of replication at the site level and large standard error, the differences described across sites likely do not reflect strong trends.

22 of the 30 monitoring sites showed decreases in the number of healthy corals observed per minute following the first observation of SCTLD (Figure 22). Most of these decreases were marginal, indicating that there is likely no detectable change in coral colony abundance at these sites to date. However, five sites showed at least a 50% reduction in coral colonies observed per minute: Sites 2, 3, 5, 28, and 29 (Figure 22). In contrast to the five sites showing reduced coral colony abundance, the remaining eight sites had mostly marginal increases in coral abundance; however, Site 16 (Little Africa) showed a greater increase after SCTLD than any other site. This site is dominated by large *O. annularis* with colony boundaries that are difficult to distinguish, so the data likely reflect differences in colony distinction among surveyors.

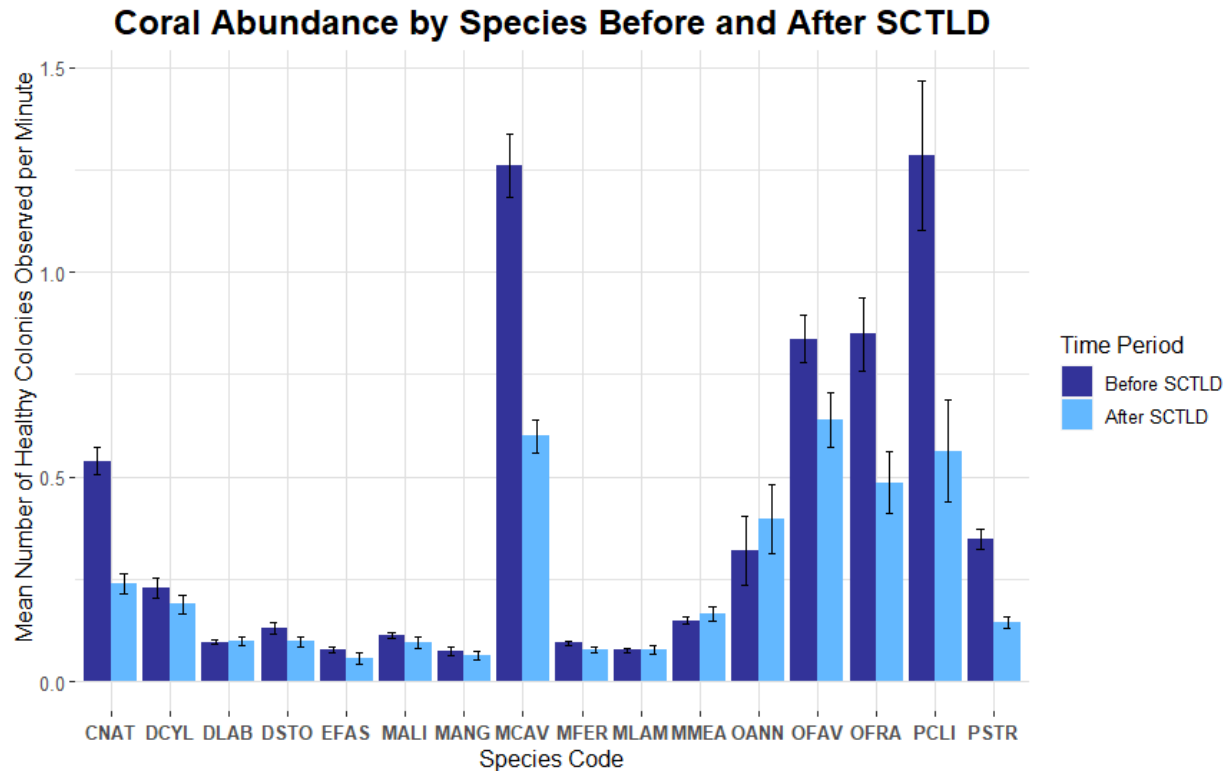


Figure 21. Comparison of mean number of healthy susceptible coral colonies by species observed per minute before (dark blue) versus after (light blue) SCTLD outbreak. Species codes include *Colpophyllia natans* (CNAT), *Dendrogyra cylindrus* (DCYL), *Diploria labyrinthiformis* (DLAB), *Dichocoenia stokesii* (DSTO), *Eusmilia fastigiata* (EFAS), *Mycetophyllia aliciae* (MALI), *Mussa angulosa* (MANG), *Montastraea cavernosa* (MCAV), *Mycetophyllia ferox* (MFER), *Mycetophyllia lamarckiana* (MLAM), *Meandrina meandrites* (MMEA), *Orbicella annularis* (OANN), *Orbicella faveolata* (OFAV), *Orbicella franksi* (OFRA), *Pseudodiploria clivosa* (PCLI), *Pseudodiploria strigosa* (PSTR).

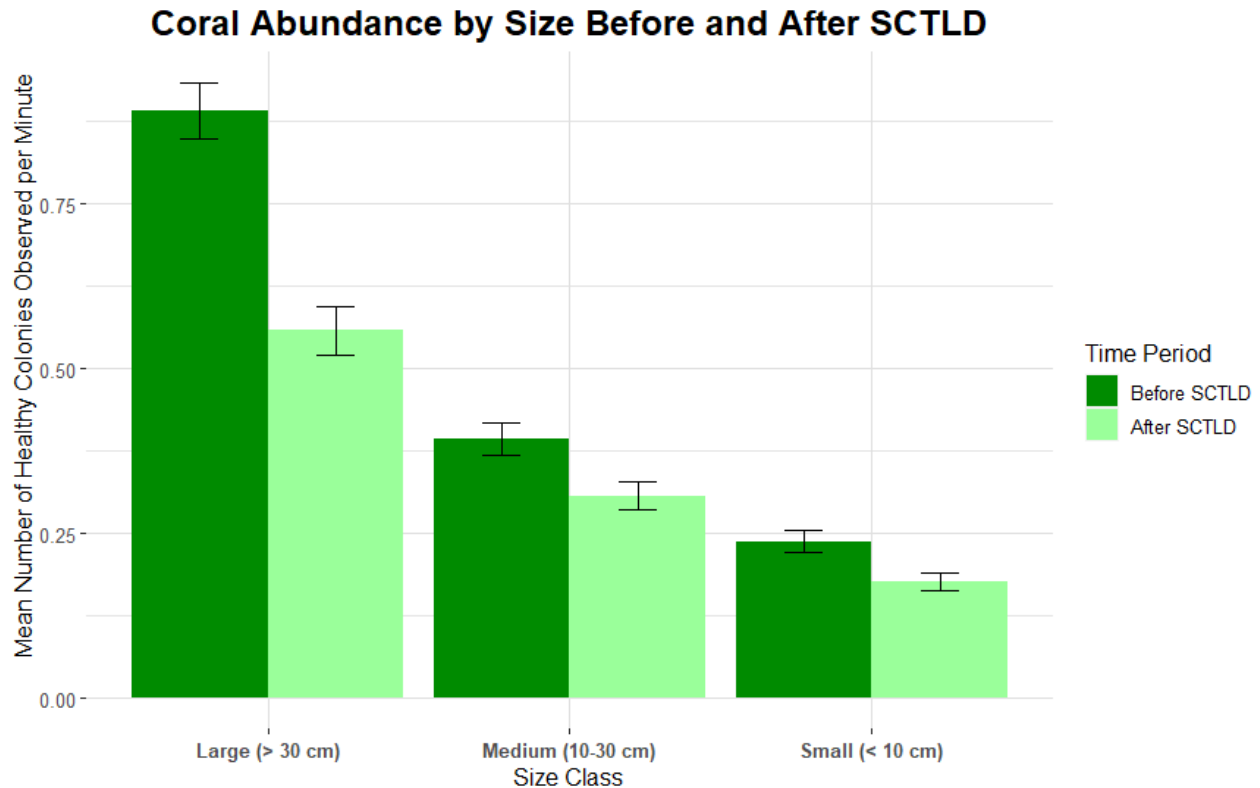


Figure 22. Comparison of mean number of healthy susceptible coral colonies by size observed per minute before (dark green) versus after (light green) SCTLD outbreak. Susceptible coral species listed in Figure 23, above.

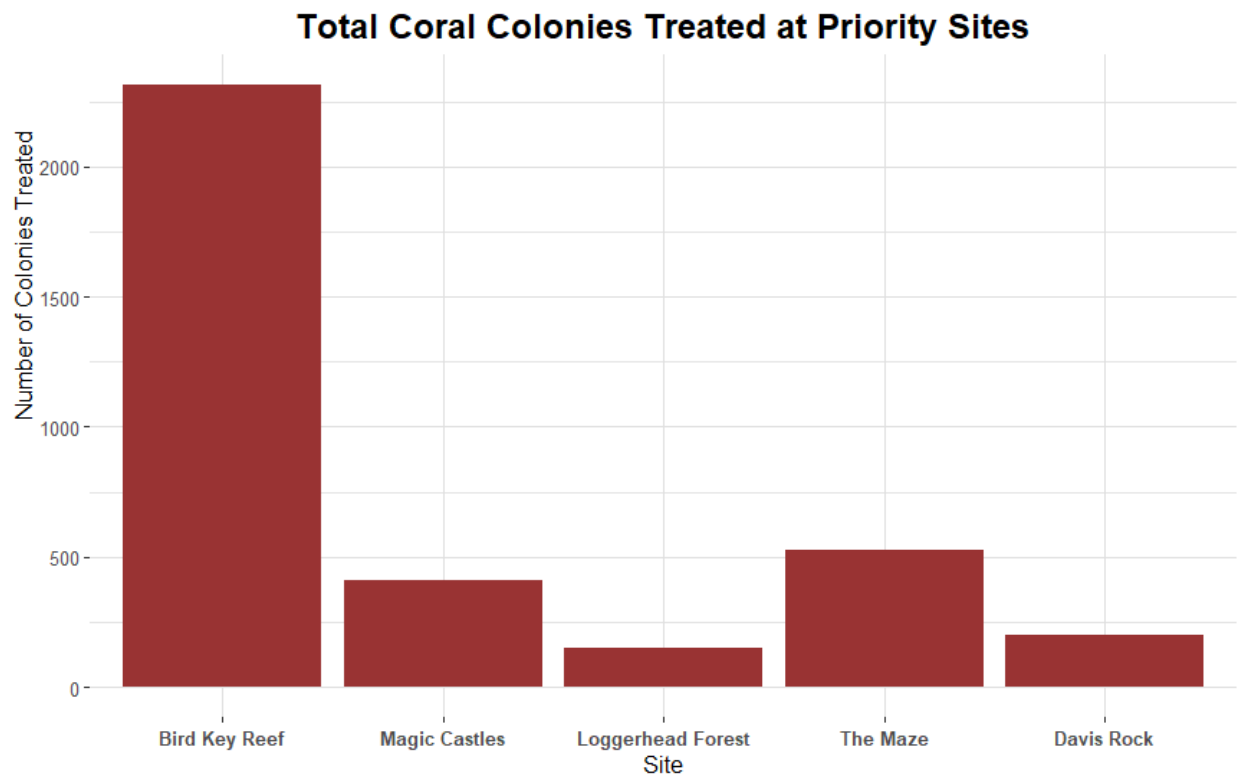
Across all sites, most species showed a negligible change in healthy colony abundance before and after SCTLD. The magnitude of change closely corresponds to the respective species abundance prior to SCLTD, with species that were less frequently observed before the onset of SCTLD showing less change than those that were most frequently observed (Figures 18 and 23). *C. natans*, *M. cavernosa*, *O. faveolata*, *O. franksi*, *P. clivosa*, and *P. strigosa* showed a decrease in healthy colonies observed per minute after the onset of SCTLD (Figure 23). All these species, except the Orbicellids, showed at least a 50% reduction in average healthy colonies observed. *O. annularis* is the only species that showed a somewhat notable increase in healthy colonies observed after SCTLD; however, this is also likely a reflection of the data collection challenges at Little Africa because this site is the only site with significant *O. annularis* presence.

Healthy colony abundance decreased for each coral size class after the arrival of SCTLD (Figure 24). Large corals were the most frequently observed size class prior to SCTLD and showed the most drastic decrease in colony abundance after SCTLD.

## Intervention

### *Intervention Totals by Site*

As of June 2022, a grand total of 14,662 coral colonies have been treated for SCTLD in the Park: 5,206 from internal operations and 9,456 from the SCHMIR missions. Intervention totals include retreatments of colonies, such as the *D. cylindrus* colonies at Magic Castles which have collectively been treated a total of 177 times. Internally, a large intervention effort was allocated to high-priority sites (Figure 25). A total of 200 person hours underwater were spent surveying and treating SCTLD at these sites: 107 hours at Bird Key Reef, 41 hours at Magic Castles, 24 hours at Loggerhead Forest, 19 hours at The Maze, and 9 hours at Davis Rock. This is nearly 40% of the total hours underwater completed internally for the entire project. Most colonies were treated at Bird Key Reef (2,315), followed by The Maze (526) and Magic Castles (412; Figure 25). It is estimated that 5,228 and 2,112 linear meters of treatment have been performed internally by NPS at Bird Key Reef and Loggerhead Forest, respectively.



*Figure 23. Total number of SCTLD-infected corals treated at priority sites: Bird Key Reef, Magic Castles, Loggerhead Forest, The Maze, and Davis Rock.*

Aside from priority sites, 1,603 colonies were treated at 10 other sites throughout the Park, including the Moat Wall, Coal Docks, Dendro City, and various other sites. 593 of these colonies were treated at the first three sites where SCTLD was observed: Site 2, Dendro City, and Site 12. This reflects the initial effort to contain the disease and to preserve the *D. cylindrus* colonies that were discovered at Dendro City.

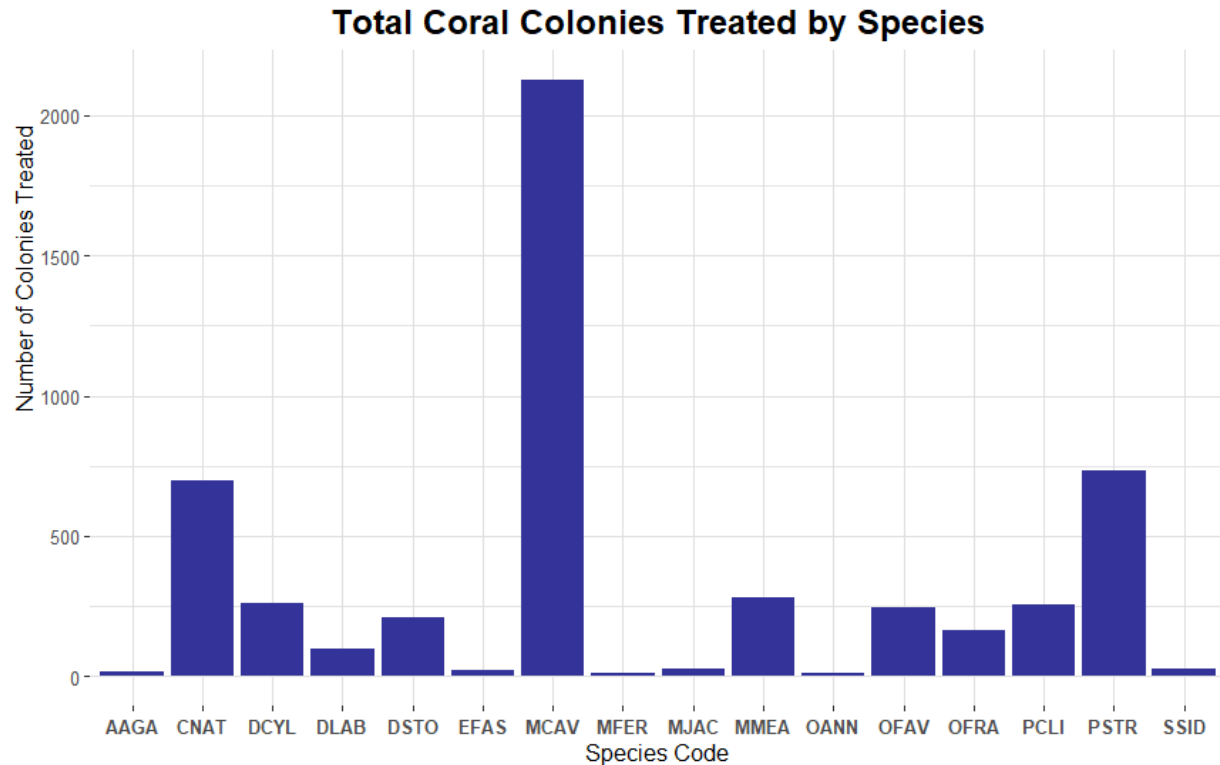


Figure 24. Total number of SCTLD-infected corals treated by species. Species codes include *Agaricia agaricites* (AAGA), *Colpophyllia natans* (CNAT), *Dendrogyra cylindrus* (DCYL), *Diploria labyrinthiformis* (DLAB), *Dichocoenia stokesii* (DSTO), *Eusmilia fastigiata* (EFAS), *Montastraea cavernosa* (MCAV), *Mycetophyllia ferox* (MFER), *Meandrina jacksoni*, *Meandrina meandrites* (MMEA), *Orbicella annularis* (OANN), *Orbicella faveolata* (OFAV), *Orbicella franksi* (OFRA), *Pseudodiploria clivosa* (PCLI), *Pseudodiploria strigosa* (PSTR), *Siderastrea siderea* (SSID).

Table 2. Percent of species most observed (light shading) and treated (dark shading) at priority sites: Bird Key Reef, Magic Castles, Loggerhead Forest, The Maze, and Davis Rock.

Site	Most Observed Species	Percent Observed	Most Treated Species	Percent Treated
Bird Key Reef	MCAV	43%	MCAV	56%
Magic Castles	PSTR	49%	DCYL	44%
Loggerhead Forest	OFAV	29%	MCAV	52%
Maze	MCAV	53%	MCAV	47%
Davis Rock	MCAV	53%	MCAV	49%

### *Intervention Totals by Species*

*M. cavernosa* was the species that was most treated for SCTLD, with 2,127 colonies treated across all sites (Figure 26). This was nearly more than three times the number of colonies treated for the next most treated species, *P. strigosa* (733 colonies) and *C. natans* (697 colonies). All other species had less than 300 colonies treated (Figure 26). *M. cavernosa* was the most observed species at three of the five priority sites and the most treated species at four of the five priority sites (Table 2). *P. strigosa* was the most observed species at Magic Castles, but *D. cylindrus* was the species most treated due to the prioritization of the nine pillar corals at that site. *O. faveolata* was the most observed species at Loggerhead Forest, but *M. cavernosa* was the species most treated (Table 2).

### **Total Coral Colonies Treated by Size**

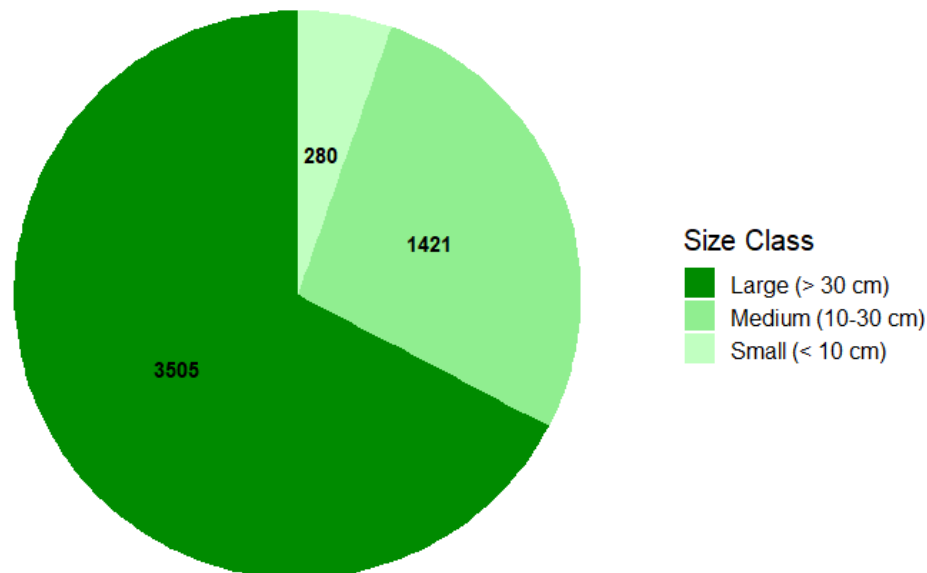


Figure 25. Total number of SCTLD-infected corals treated by size class: small (< 10 cm, pale green), medium (10-30 cm, medium green), large (> 30 cm, dark green).

### *Intervention Totals by Size*

Large corals were the size class that was most treated for SCTLD (3,505 colonies), followed by medium (1,421 colonies) and small (280 colonies; Figure 27). This corresponds to the hierarchy of size classes observed per minute prior to the SCTLD outbreak (Figure 24). More than two thirds of the colonies treated were larger than 30 cm, and only 5% of the colonies treated were smaller than 10 cm (Figure 27).

## Monitoring

### *Coral Demographic Surveys*

Coral demographic surveys were completed at all nine monitoring sites in Spring 2021 before SCTLD was observed in the Park (Figure 28). This provided valuable baseline data that will be used in the future to analyze large-scale, reef-wide changes over time following the onset of SCTLD. These data have been entered into the NPS Monitoring database and shared with FWC partners as part of the ongoing CREMP monitoring and analyses. Monitoring surveys will be repeated at all nine monitoring sites in November 2022, May 2023, and annually each May henceforth (Figure 28). These data will then be used to analyze changes in coral cover, community composition, and coral health (disease, bleaching, mortality, and interactions with other benthos) across several time points following the initial establishment of SCTLD.

Furthermore, data from the four pinnacle reefs (The Maze, Texas Rock, Davis Rock, and Mayers Peak) will be isolated and used in a separate BACI study design to assess the impact of SCTLD antibiotic treatments among similar biological communities.

### *Time Series Data*

Large coral colonies of interest, including primary susceptible SCTLD species, were fate-tracked using time series photos. Notably, the nine *D. cylindrus* colonies at Magic Castles were closely monitored to determine the efficacy of antibiotic treatments. Progression of individual lesions was tracked using photos (Figure 29) and treatment strategy and frequency was adjusted accordingly. Shortly after disease onset, treatments every 10-14 days were most effective. After significant lesion recovery, monthly treatments were sufficient. Unfortunately, from January-June 2022, the site was only visited every three months due to changes in staffing, equipment repairs, poor weather conditions, and competing priorities. During this time the condition of the colonies severely declined (Figure 30). Estimated mortality has reached over 75% on at least one colony.

Another notable fate-tracked colony, a large *M. jacksoni* at The Maze, was closely monitored until its demise. This colony was quickly and completely treated after initial observation of SCTLD, and retreated monthly thereafter. Despite careful monitoring and treatment, living tissue decreased relatively quickly until the colony was completely deceased in December 2021 (Figure 31).



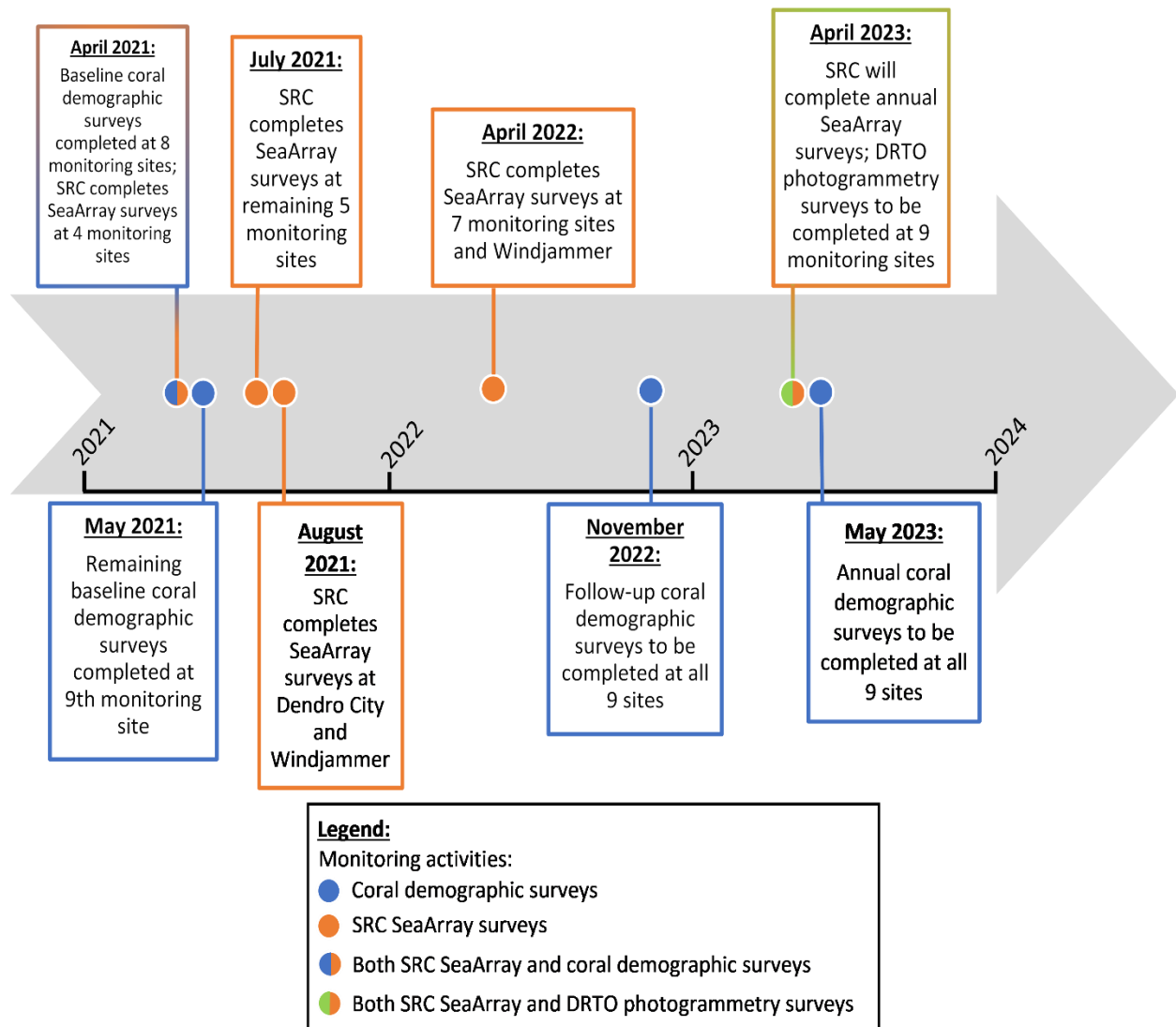


Figure 26. Timeline of monitoring activities on a monthly time scale. For exact dates that surveys were completed at each site, please refer to the SCTL Site Tracking spreadsheet.

### *SRC NPS Photogrammetry*

SRC similarly acquired baseline data from large-scale photogrammetry surveys at all nine monitoring sites (Figures 13-15). Although the photogrammetry surveys were completed after SCTLD was observed in the Park (Figure 28), data were collected prior to the arrival of SCTLD at these nine sites. After imaging the monitoring sites, SRC completed photogrammetry surveys at Dendro City and Windjammer (Figure 28). The Dendro City data were invaluable for mapping and locating 23 *D. cylindrus* colonies throughout the site for intervention (Figure 11). Without these data, many of these colonies would have gone undiscovered. Furthermore, because the photogrammetry data were collected after SCTLD was observed at Dendro City, these photomosaics will provide a reef-wide snapshot of a site during the early stages of disease onset.

To date, 251,701 images of the Park's coral reefs have been collected by SRC. Nearly one year after the initial photogrammetry surveys, SRC repeated the surveys at seven monitoring sites and Windjammer (Figure 28). Three sites were not visited due to logistical constrictions: two of the three Loggerhead Forest sites (omitted due to replication of the same reef) and Dendro City (omitted in favor of Windjammer). Windjammer was resurveyed because of its importance to Park stakeholders and stage of SCTLD progression. The site was experiencing a severe SCTLD outbreak in April, with an estimated loss of 80-90% coral cover. Therefore, the photomosaic data collected will provide a snapshot of a site during peak disease outbreak with reference to baseline data before SCTLD. In addition to the Windjammer data, photomosaics from the seven monitoring sites will be analyzed to show the progression of the disease nearly one year later. Large-scale photogrammetry surveys will be repeated annually each April to assess the progression of long-term changes on DRTO reefs due to SCTLD.

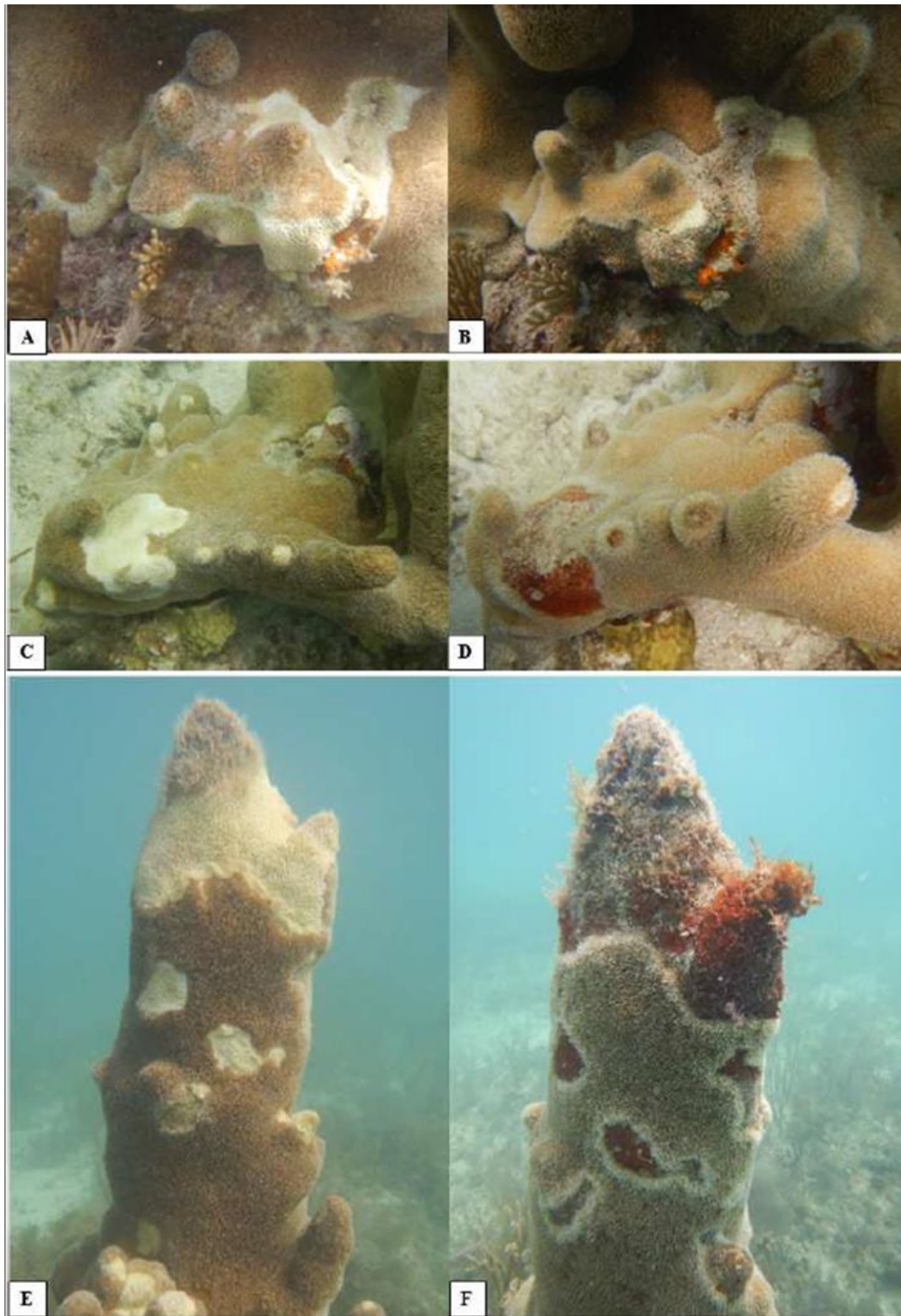


Figure 27. SCTLD lesion recovery following treatment on *Dendrogyra cylindrus* colonies at Magic Castles in September 2021 (left) and April 2022 (right), showing comparisons on colonies 2 (A & B), 6 (C & D), and 4 (E & F). (Source: FWC, 2022)

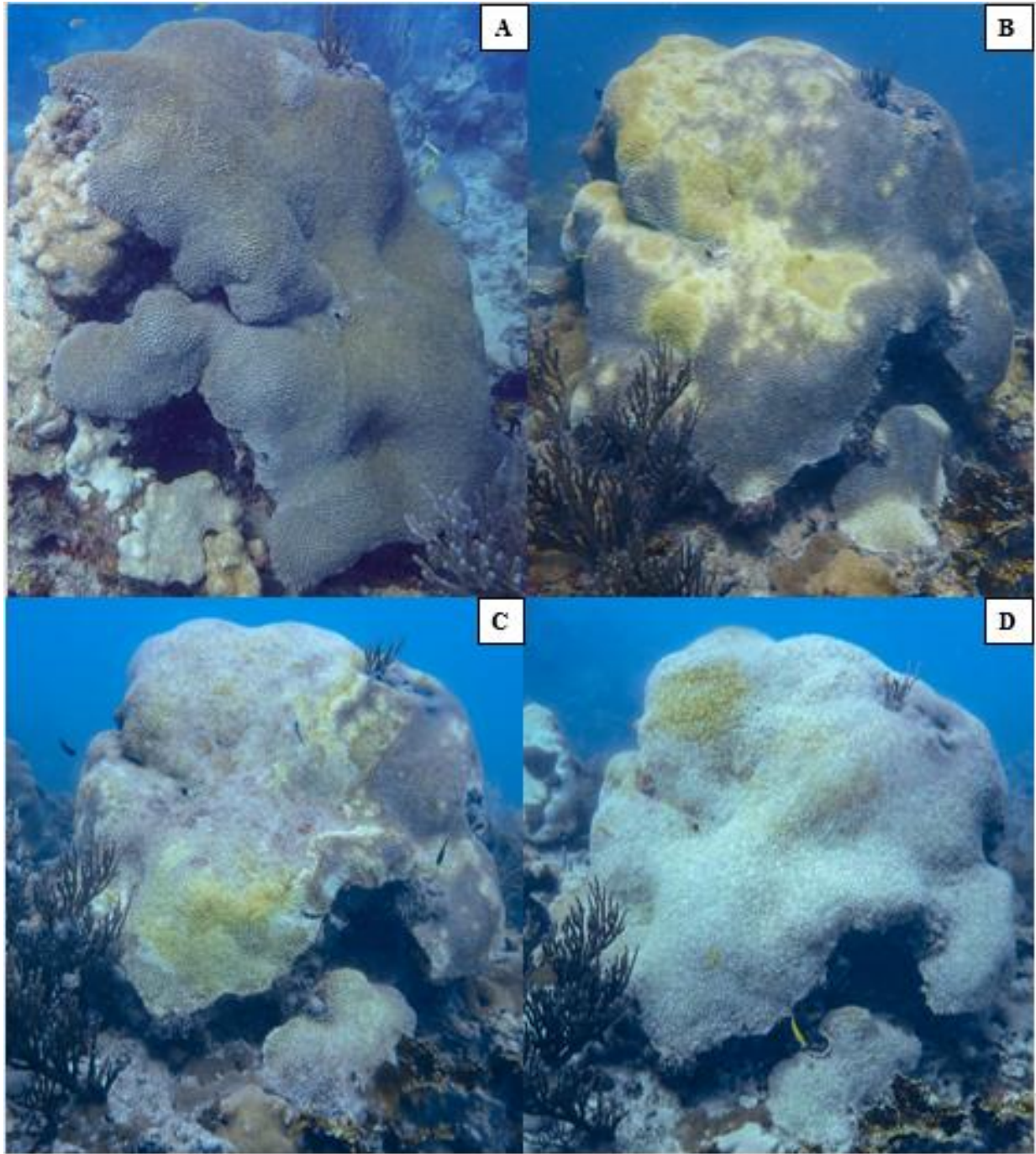


Figure 28. Severe recent SCTLD progression at Magic Castles, where (A) NPS DRTTO applies treatment to colonies 7 and 8, (B) colonies are thoroughly treated along all disease margins.

#### *DRTTO NPS Photogrammetry*

Smaller-scale photogrammetry data will be collected at the nine monitoring sites to supplement the data collected by SRC. Images of the permanent transects at eight monitoring sites (all except Magic Castles) will be collected and used to create 2D orthomosaics that will be analyzed and paired with the coral demographic data collected by divers. Orthomosaics will be generated in-house and analyzed for the same metrics as the demographic surveys, plus rugosity and percent cover of non-coral taxa such as *Diadema antillarum*, sponges, and algae. The protocol will be analogous to Scripps Institution of Oceanography photogrammetry methodology (Sandin et al., n.d.). At Magic Castles, the nine individual *D. cylindrus* colonies will be imaged to generate 3D models of the coral colonies for more precise fate-tracking. Data from the coral demographic surveys, large-scale photogrammetry, and small-scale photogrammetry will be used together to create a large picture of reef-wide changes over time following the onset of SCTLD.





*Figure 29. SCTLD progression on large infected Meandrina jacksoni colony over the course of three months with frequent treatment. (A) As of March 21, 2021, colony is healthy, (B) colony infected by September 9, (C) lesions progress significantly by October 7, (D) entire colony succumbs to mortality by December 19.*

## DISCUSSION

### **Project Effort**

This was an immense project effort, with over 500 hours underwater dedicated internally to SCTLD reconnaissance, intervention, and monitoring. Effort was largely allocated toward vigilant reconnaissance surveys to detect the first appearance of SCTLD in the Park, and disease intervention thereafter. Despite initial efforts to contain SCTLD at the first site where it was observed, the disease inevitably and rapidly spread, becoming prevalent in all regions of the Park in less than one year. This shifted intervention focus to five key priority reefs: Bird Key Reef, Magic Castles, Loggerhead Forest, and BACI impact sites at The Maze and Davis Rock. In addition to the high-priority reef sites, intervention was also conducted at secondary reef sites such as the Windjammer, Moat Wall, and Coal Docks.

As of June 2022, over 14,000 corals were treated at DRTO. The SCHMIR missions led by NSU were an extremely effective effort as almost two-thirds of the total corals treated in the Park resulted from these missions. This large and collaborative effort benefitted from extra personnel (more than double the personnel dedicated to internal Park operations) and a liveaboard vessel that allowed divers to maximize the number of dives completed. 6,038 corals were treated at Bird Key Reef during this mission, and 2,315 corals were treated internally at this site, meaning over 55% of the corals treated at the Park were at Bird Key. Bird Key Reef and Loggerhead Forest are the two largest reefscapes and the former site is at a later stage of SCTLD progression, hence the massive intervention effort to date. As the disease progresses at Loggerhead Forest, these numbers are expected to increase to similar levels as Bird Key Reef (currently about 20% of corals treated are at Loggerhead Forest). Similar results are expected after the second SCHMIR mission at Loggerhead is completed and internal operations follow the effort with concentrated intervention to increase the success rate of coral colonies treated by NSU. The large-scale intervention efforts at high-priority reefscapes followed by continuous, localized maintenance of the reef feature appear to be the most efficient and effective approach to reducing the impacts of SCTLD invasion and epidemic phases.

### **Comparisons Before & After SCTLD**

As stated, data showing total healthy corals observed per minute before and after SCTLD should be interpreted cautiously as these data are not based on statistical analyses (Figures 22-24). In particular, site-level data had minimal replication and large standard error (Figure 22), so this should be considered when interpreting results. Despite this, overall trends in the data largely show declines in healthy coral abundance after the onset of SCTLD at individual sites, across species, and within all size classes. It may be challenging to detect significant changes in coral colony abundance in general without rigorous sampling, and particularly difficult for rare and uncommon coral species. Furthermore, as this study was conducted during the invasion and into the epidemic phases of the disease outbreak, additional time may be required to begin to adequately assess and document changes to the coral community composition.

### *Comparisons by Site*

Concentrated intervention efforts at priority sites appear to be making a positive impact overall, slowing the progression of SCTLD as compared to other sites in the Park. Site 13 (Magic Castles) and Site 22 (Bird Key) showed a slight positive increase in coral colonies observed per minute following SCTLD (Figure 22). Negligible change like this, positive or negative, can likely be interpreted as no significant change in healthy colony abundance before and after SCTLD. Sites 1 (Bird Key), 17 (The Maze), and 20 (Davis Rock) showed a decrease in coral abundance; however, the magnitude of change was far less than some of the other sites in the Park (notably those with a > 50% reduction; Figure 22). Loggerhead Forest (Sites 9 and 25-28) is beginning to show overall negative trends in healthy coral abundance (Figure 22), which is reflective of stages of SCTLD progression at these sites. Site 28 is located on the far southwest site of the reefscape and has experienced the greatest reduction in the number of healthy corals observed, whereas Site 26 is located at the far northeast side and showed a small increase (no significant change) in healthy corals (Figures 4 and 22). SCTLD was not observed at Site 26 until late January 2022, and was observed at all other Loggerhead sites roughly two months prior. Therefore, these trends likely indicate that the disease is progressing through the reefscape from southwest to northeast. This may be caused by localized water movements and eddies created by the Loop Current as it passes through Dry Tortugas Bank. The second SCHMIR mission at this site was well-timed and was on track to slow the trends of decreasing healthy coral abundance that are becoming more apparent at Loggerhead Forest. The mission will be rescheduled for the earliest next opportunity and local staff will prioritize this feature in the interim.

Five sites showed a significant decrease in healthy coral abundance (> 50%) following SCTLD: Sites 2 (DRTO Open East), 3 (DRTO Open Middle), 5 (Pulaski Shoal), 28 (Loggerhead Forest), and 29 (Windjammer; Figure 22). Sites 2, 3, and 5 are located on the east side of the Park where SCTLD was initially observed (Figure 4), so the declines in coral abundance are likely due to these sites being in later stages of disease progression. As the disease progresses at sites farther to the west, similar patterns will likely become apparent. Site 29 experienced peak SCTLD progression during April 2022 and suffered a large loss in coral colonies. Because this site is a popular dive site for visitors, it is possible that the reef was impacted more severely by SCTLD due to weakened coral health from greater human influence. Moreover, because internal operations were focused on priority sites, infrequent intervention was completed at this site. This demonstrates that regular intervention effort is imperative to prevent significant loss of coral and underscores the need to support a robust, comprehensive response.

Data for remaining sites had large standard error and/or showed negligible change in the number of healthy corals observed before and after the arrival of SCLTD. Most sites with negligible change have likely not yet reached a stage of SCTLD progression sufficient to drive detectable change in the data. Interestingly, untreated control sites for the BACI study (Sites 18 and 21) showed less change in coral abundance before and after SCTLD than impact sites that were treated (Sites 17 and 20; Figure 22). This could be because impact sites were not treated from January-June 2022, underscoring the importance of regular and frequent intervention. The



differences in coral abundance changes among BACI sites are still somewhat understated, however, and may change as SCTLTD progresses at these sites. Overall, no increases in healthy coral abundance were observed following SCTLTD onset, aside from negligible increases (meaning no significant change) and the larger increase at Site 16 (attributed to differences in distinguishing colony boundaries on *O. annularis*).

### *Comparisons by Species*

In general, the most abundant species prior to the onset of SCTLTD suffered from the largest proportional declines in healthy corals observed (Figures 18 and 23). *M. cavernosa* was very abundant prior to the onset of SCTLTD (Figure 18) and was the most treated species by far (Figure 26). This species also suffered from the biggest loss in coral abundance following SCTLTD (Figure 23). Other species that experienced a significant loss in coral abundance (> 50%) include *P. clivosa*, *C. natans*, and *P. strigosa* (Figure 23). *M. cavernosa* is typically impacted by SCTLTD at the same time as Orbicellids; however, to date the Orbicellids at the Park have not been impacted as severely (Figure 23). This may indicate localized impacts that differ from larger regional trends. At Loggerhead Forest, *O. faveolata* was the most frequently observed species, but *M. cavernosa* was the most treated (Table 2), further suggesting that the Orbicellids may be at a later stage of disease progression than *M. cavernosa*. The severe impact of SCTLTD on *M. cavernosa* appears to be related to its high abundance throughout the Park. *P. clivosa*, another highly abundant species, suffered from the second biggest loss in coral abundance following SCTLTD (Figure 23). However, this species was not treated proportionally (Figure 26). This is because the majority of *P. clivosa* colonies are found at the Moat Wall and Coal Docks, which were treated less frequently than priority sites. *M. cavernosa* and *P. clivosa* suffered a very similar magnitude of loss in coral abundance despite differences in treatment. This is likely a reflection of recent onset of SCTLTD at the Moat Wall (January 2022) and Coal Docks (March 2022), rather than inefficacy of antibiotic treatments on *M. cavernosa* colonies.

Many of the highly susceptible species that are often first impacted by SCTLTD are not yet showing signs of significant change in healthy coral abundance (NOAA, 2018; Figure 23). The exceptions are *C. natans*, *P. strigosa*, and *P. clivosa*, which are more abundant, suggesting that changes in colony abundance after SCTLTD may be density dependent. Perhaps less abundant highly susceptible species, such as *M. meandrites*, *D. cylindrus*, and *D. stokesii*, are now being noticed more easily after being overlooked due to the previously high abundance of other species such as *M. cavernosa*. However, even with frequent treatment these species have been difficult to preserve. The *M. jacksoni* colony at The Maze (Figure 31) perished despite its large size and frequent and complete treatments. Furthermore, the *D. cylindrus* colonies at Dendro City rapidly succumbed to SCTLTD despite early and frequent treatment efforts shortly after disease onset. These colonies were much smaller than the pillar corals at Magic Castles and were farther apart in distance. These factors likely led to their demise because SCTLTD progressed throughout the individual colonies more quickly due to their size, and because the corals were more logistically challenging to locate and treat. Fortunately, prior to their demise, fragments from three *D. cylindrus* colonies were successfully saved and their genetics thus preserved, underscoring the importance of coral rescue missions.

As stated, colony abundance did not change at Magic Castles, indicating that treatment is at least slowing the progression of SCTLD at this site. The *D. cylindrus* colonies at Magic Castles responded well to treatment after modifying retreatment frequency (every 10-14 days for severe infections and monthly for lesion maintenance; Figure 29). Timelines for retreatment of these colonies were informed by time series monitoring photos, emphasizing the importance of thorough lesion progression and treatment efficacy monitoring on species that are highly susceptible to SCTLD. Unfortunately, any lapse in routine retreatment can result in severe decline in the condition of these colonies during the invasion and epidemic phases (Figure 30). Mortality estimates have reached over 75% on at least one colony following a three-month retreatment interval, emphasizing the need for prompt and routine intervention to improve the outlook for recovery.

### *Comparisons by Size*

Large coral colonies > 30 cm in diameter were most frequently observed before and after SCTLD (Figure 19 and Figure 24) and most treated for disease (Figure 27). Although medium and small colonies are likely more abundant than reported as they may be overlooked in favor of more obvious large colonies, large colonies are very prevalent at DRTO. Large colonies were also the size class that experienced the most drastic relative reduction in healthy coral abundance following the disease (Figure 24). This is likely due to the high abundance of large colonies throughout the Park but could also reflect challenges with treating large corals for SCTLD as it is more difficult to be thorough when applying treatment to larger disease margins. In addition, these observations could reflect changes in composition, with medium and small colonies becoming more apparent as large colonies experience mortality.

### **Monitoring**

Photogrammetry is being utilized increasingly often by underwater researchers as an incredibly useful tool to track changes over time. The orthomosaics generated by SRC and internally by DRTO staff will be important in the wake of SCTLD because they preserve a snapshot of reefs during specific time points and stages of disease progression. These data can be used in the future to answer a wide variety of research questions about SCTLD progression and impact over time. In particular, the data from the SeaArray will provide an incredibly unique opportunity to answer large-scale, reef-wide questions because the data encompasses such a large geographic area. As the Park eases into the use of this technology, it will be important to develop infrastructure to support and manage these large archives of data. DRTO staff are creating protocols to detail the collection, generation, processing, storage, and access of these data, with the intention of utilizing this technology for multiple natural resources projects within the Park.

## **Future Work & Management Recommendations**

This project has provided a summary of an intentional and immediate response to a large-scale SCTLD outbreak, with associated outcomes and important lessons learned. Unimpacted locations in the Caribbean, as well as Pacific locations with similar mass disease outbreaks, may use this information as a guide to prepare for disease outbreak prior to onset, immediately upon observation, and as the disease progresses. Importantly, SCTLD inevitably spread despite immediate treatment upon first observation in the Park and concentrated efforts to contain the disease. As the disease is waterborne, containment efforts quickly became impossible to manage. This is an important lesson as management can prepare for disease outbreak with the expectation that SCTLD will inevitably spread. With valuable input from DAC and other stakeholders, Park staff were able to identify priority sites ahead of SCTLD arrival and shift focus to these sites when appropriate. This was essential to preserve important resources when the disease inevitably spread widely enough that it could no longer be exhaustively treated.

Another valuable lesson from this project is the importance of large-scale intervention efforts, such as the SCHMIR missions. These liveaboard missions were critical to slowing disease progression across reefs encompassing a large geographical area. Having additional personnel and consecutive days dedicated to intervention on a single reef allowed for a more systematic approach to intervention, which is more effective than haphazard treatment. Importantly, these large-scale intervention missions should be promptly followed with local intervention maintenance at the site, particularly during peak SCTLD outbreak. Managers should identify high-priority reefscapes, conduct initial large-scale intervention at high-priority sites within 1-2 months of disease onset, follow up with targeted retreatments, and then conduct additional large-scale intervention at the same sites when secondarily susceptible species begin to show symptoms, approximately 3-4 months later. Targeted retreatments should be maintained throughout the invasion and epidemic phases of the outbreak, with particular emphasis on preserving highly susceptible populations and conducting intervention maintenance at high-priority reef sites that have received large-scale intervention, as capacity and support levels dictate.

One thing to consider is the effort versus benefit of long-term intervention aimed at preserving highly susceptible species such as *Meandrina* spp. and *D. cylindrus*. It became quickly apparent that these species require very frequent intervention to avoid total colony mortality. This becomes increasingly difficult when balancing other priorities and is further complicated by seasonal challenges such as poor weather conditions. As an example, Magic Castles will require frequent intervention (every 10-14 days) to manage SCTLD following the recent lapse in intervention and prevent full mortality of the pillar corals. Fortunately, coral rescue efforts are largely successful in preserving species genotypes for future restoration efforts. As SCTLD continues to ravage reefs across the Caribbean, it is critical to collect and maintain many genotypes ex-situ with the goal of identifying unique genotypes that are resistant and/or resilient to disease. This is especially important for highly susceptible SCTLD species as their abundance continues to decline across the Caribbean.

DRTO is still in the epidemic stage of SCTLD, with most of the effort currently allocated to intervention. Considering that SCTLD was detected early, and intervention will be continually completed, post-epidemic strategies at the Park may differ from other locations that were ravaged by SCTLD relatively quickly and completely. Locations in the northern Florida reef tract, such as BISC, have limited coral coverage following SCTLD and therefore have lower disease transmissibility. As a result, coral restoration strategies are somewhat straightforward and studies have shown that some restoration initiatives – outplanting coral recruits – can be effective once the disease enters the endemic phase (approximately 8 years after outbreak; Williamson et al., 2022). However, DRTO may have higher disease transmissibility for many years or indefinitely, which will require the timing and strategies of restoration activities to be carefully considered. Restoration activities will need to be opportunely timed to maximize outplant survival and minimize restoration effort required due to SCTLD mortality. Further, Park staff will need to seek expertise regarding outplant maintenance in the wake of disease. This further underscores the importance of identifying and rearing disease-resistant and/or resilient coral genotypes, which is why DRTO is part of a multi-park effort that aims to collect and identify genotypes that are resilient to SCTLD for reef restoration purposes.

As SCTLD continues to progress throughout the Park, the continuation of reconnaissance, intervention, and monitoring work is crucial. In the short-term, these data will provide invaluable insights into the progression of SCTLD at DRTO that will allow for adaptive management of the Park's coral reefs. In the long-term, these data will provide important information about the large, reef-wide progression and impacts of SCTLD across many sites and time points. Moreover, these data will inform management actions at locations that are impacted by SCTLD and similar disease outbreaks in the future. As other locations become impacted by SCTLD, it will be important for similar, reef-wide studies to be carried out for comparison to these results.

## CONCLUSION

When confronted with the possibility of widespread disease outbreaks, coral resource managers should consider the following mitigations and procedures:

1. Assess coral resources and identify at-risk populations with consideration to coral population genetics.
2. Conduct coral rescues of highly susceptible species in advance of the disease outbreak.
3. Establish a study design that allows managers to assess the efficacy of disease intervention efforts and employ adaptive management principles.
4. Identify high-priority reefscales and/or threatened coral populations for primary disease response.
5. Identify and obtain sufficient resources (including personnel) to carry out disease intervention efforts at the targeted scale and frequency.
6. Implement large-scale disease intervention at high-priority reefscales and follow with concentrated retreatments.
7. Sustain disease intervention activities throughout the invasion and epidemic phases of the outbreak, particularly for threatened coral populations.
8. Carefully consider the timing of coral restoration actions post-outbreak and ensure efficacy of proposed restoration activities in the wake of disease.

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## APPENDIX

The following are provided as supplementary materials to this report:

- Dive Logs: NPS is required to document all scientific and occupational diving. Scanned logs are provided for reference.
- Datasheets: NPS provides scans of reconnaissance and intervention datasheets for reference.
- Spreadsheets/Databases:
  - Reconnaissance Data: NPS enters reconnaissance data into the NPS Recon Database and the FWC Recon Spreadsheet.
  - Intervention Data: NPS enters intervention data into the NPS Intervention Database. NPS also makes monthly intervention data submissions to the FWC Coral Disease Intervention Dashboard.
  - Monitoring Data: NPS submits diver-collected demographic survey data into an established FWC database and can access raw data by request, quarterly. NPS SRC and NPS DRTO photogrammetry data is in processing.
- Representative and Time Series Photos: NPS has included photos and documentation of reconnaissance, intervention, monitoring and training activities.
- DRTO SCTL D Response Plan, 2021



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