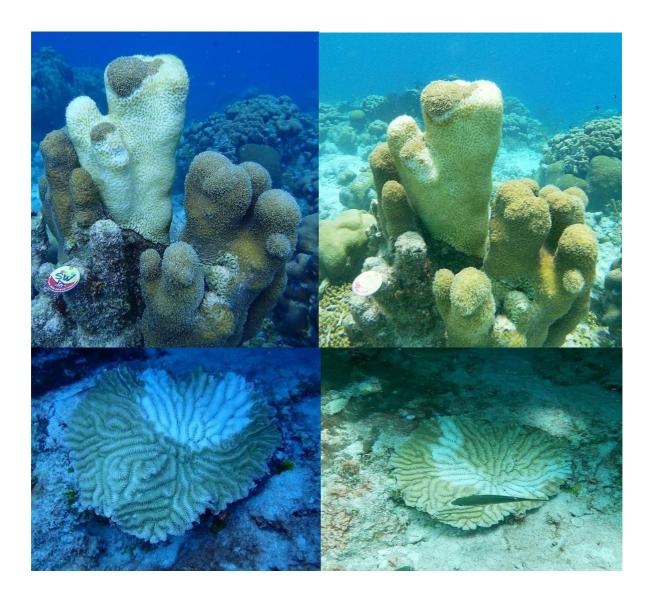


Stony Coral Tissue Loss Disease: Action Plan for the Bonaire National Marine Park



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Introduction

Since its discovery in Florida in 2014, stony coral tissue loss disease (SCTLD) has spread rapidly throughout the Caribbean region, severely impacting reef health and biodiversity. Tissue loss diseases are the most virulent and damaging of the documented coral diseases (Aeby et al., 2019), often resulting in severe mortality rates. While tissue loss diseases are not new, the widespread decline of the shallow reef building Acroporas is widely credited to a tissue loss disease (white band disease), SCTLD is distinguished by the wide number of stony coral species affected and the rapid spread both on single colonies and through the reefscape. Neither the underlying pathogen causing SCTLD, nor the transmission vector(s) have been identified, but work is ongoing. Preliminary work suggests a bacterial origin for the disease as antibiotic treatments have been highly effective at halting lesion progression (Aeby et al., 2019; Meyer et al., 2019). Studies have shown that SCTLD can be transmitted by direct contact and passively through the water column (Aeby et al., 2019; Precht et al., 2016; Sharp et al., 2020). Based on models of the spread between reefs, SCTLD progression is likely to be associated with slow-moving bottom currents and the accompanying sediment particles (Muller et al., 2020). SCTLD affects over 20 hard coral species and, at the time of writing, the disease has been confirmed in 25 countries and territories. With how rapid this disease spreads, it is only a matter of time before it is found on Bonaire's reefs.

On Friday July 22nd 2022 STINAPA biologist, Roxanne Francisca, was following up on concerning reports on a suspicious disease at the dive site 'Karpata'. During this inspection we found what we suspected to be SCTLD on a pillar coral (*Dendrogyra cyllindrus*). After consulting with on-island and regional partners, concern that this could be SCTLD grew and, as a precautionary measure, both Karpata and La Dania's Leap (one dive site south) were closed.

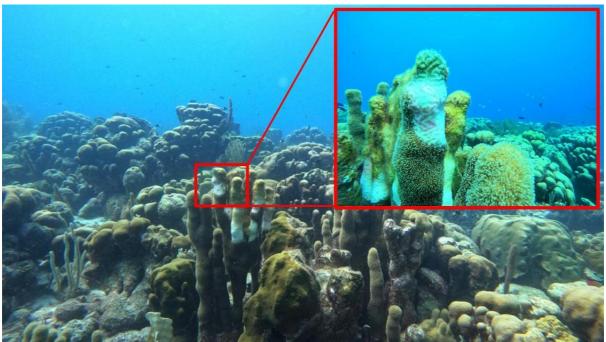


Figure 1. Pillar coral colony at Karpata on which the first concerning lesions were seen. The disease subsequently killed nearly all of the tissue on the affected branch.



To quantify the possible extent of the spread, marine park rangers inspected the majority of Bonaire's reefs looking for signs of this disease. The inspections started at Karpata to establish the spread at this dive site; in the first week inspections were also conducted at La Dania's leap and the King Willem Alexander no dive reserve (upstream and downstream from Karpata respectively). In the same report it was also mentioned that SCTLD-like symptoms were also sighted at Klein Bonaire at the dive site Knife and neighboring dive sites. These were also inspected by the marine park within the same week as Karpata; while there was non-SCTLD disease present at many of the reefs, we did not see any spread of disease that resembled what has been reported on other islands that have already been hit with SCTLD.

In the following weeks, to cover as many dive sites as possible, marine park rangers conducted two dives every morning inspecting every other dive site on Klein Bonaire and along Bonaire's west coast. It quickly became apparent that the disease found at Karpata was hitting the pillar corals singularly hard thus, during inspections, we focused particularly on this species and monitored these actively especially considering that pillar corals, and maze corals, are species that are highly susceptible to SCTLD and are often the first affected on the reef.

During this intensive pre-invasion monitoring we observed corals with similar lesions as those at Karpata at the dive sites Oil Slick, Kalli's Reef, and Small Wall. We also returned to Karpata weekly on Fridays to monitor the spread on the coral colony as well as over the reef. Following rapid expansion of the lesions, resulting in tissue loss on a large part of one branch of the colony on which the strange lesions were first detected. At this point the decision was taken to remove the diseased branch in an effort to curb the spread; following the removal of this branch, the spread of the disease stopped. During follow up weekly monitoring, some suspicious lesions were also observed on another pillar coral colony as well as on an elliptical star coral (*Dichocoenia stokesii*); from the full report, seven sites had corals in the survey area exhibiting SCTLD-like symptoms. Most of these corals, however, were not species with high susceptibility to SCTLD. In other words, they were not species that were the first to be affected during outbreaks in other regions. For a full report on what was found during these inspections, please see appendix I.

With the detection of the possible outbreak of SCTLD, and in line with recommendations from regional partners, we felt it necessary to close (quarantine) the dive site Karpata and La Dania's Leap; this in order to track the spread on this reef, but also in an effort to prevent the disease from being spread to other sites by marine park users. We also reiterated the advice to all users of the park to actively disinfect their gear as often as possible and certainly prior to their first dive, and after their last dive, on Bonaire. We feel that the protection of nature should always supersede the recreational use of nature, especially in a situation like this where it is uncertain what we are dealing with and the severity of the disease outbreak was still unknown. The dive sites remained closed until it became clear that (1) the disease had not spread to the BNMP except for some concerning observations at Kalli's Reef and Small Wall and (2) the disease was not actively spreading at Karpata. We therefore decided to reopen all dive sites on Friday September 23rd 2022.

We have established in the meantime the following:



- Weekly monitoring by BNMP rangers with a biologist present as long as this is deemed necessary;
- Marking of corals to be able to monitor the rate of spread on a coral affected with disease;
- Awareness campaign to the users that we have disease present on our reefs and may need to deal with an outbreak of a highly lethal coral disease in the form of SCTLD;
- Rinse protocols are in place and discussed with the (dive and cruise) branches;
- Protocol regarding diving in the north (where disease suspected) is being discussed with the (dive and cruise) branches first a dive in the east, south, west or at Klein Bonaire (disease not suspected) and after this a dive in the north;
- Users of the BNMP are actively reporting disease present in reefs by making use of AGRRA portal;
- Enforcement is picked-up and actively discussed with the Public Prosecutor by the possible use of a shortened charge based on art. 4 Island Decision Underwater Park Bonaire:
- Frequent monitoring of possible sightings at other dive sites by our rangers and/or biologists has been established and data has been processed (see Appendix I);
- Collaboration with NOAA and AGRRA to possibly sample corals affected by white plague and SCTLD to aid in diagnosis using tissue sampling.

After consulting with regional experts from various organizations, we were informed that it is, at this moment, impossible to diagnostically determine if we are dealing with a particularly aggressive form of white plague disease or SCTLD. We are working with NOAA and AGRRA to collect tissue samples that will help them to hopefully be able to distinguish SCTLD from

other diseases. In the meantime, SCTLD on Bonaire can only be confirmed once we have a disease that is spreading rapidly and affecting multiple species and sites.

Already during the pre-invasion monitoring, signs of bleaching were apparent and, as of October 2nd, a bleaching alert level 2 (where significant bleaching is expected to occur and some coral mortality is likely) was issued for Bonaire. In other places, it has been observed that during bleaching events the spread of the disease was slowed down, but that the rate of the disease spread increased following the event; during this year's bleaching monitoring we will be recording presence of disease as well.



Figure SEQ Figure * ARABIC 2. Partially bleached boulder star coral (Orbicella annularis) found during pre-invasion bleaching monitoring.

Once we received additional reports of potential SCTLD at reefs south of Karpata, we discussed closing dive sites in the north. In the end, we opted not to do this until we had more information about the occurrence of the disease on our reefs. How SCTLD spreads remains unclear, however there are strong indications it can spread through the water and through direct contact. We have therefore reiterated the advice given by the international SCTLD working group to all users of the BNMP in an effort to curb the spread as much as possible. We advise users to start their dives around the rest of the island and do the more northern dives in at the



end of their day/trip. This to try to limit the possibility of the disease spreading to reefs in the West, South, East, and Klein Bonaire. For boat dives the advice is similar; first visit Klein Bonaire and then the northern part of the island. And we advise divers to decontaminate gear after arriving in Bonaire and after each diving day.

We have involved the different users (by reaching out to their branches) as much as possible to be able to share information about our work.. It is important to connect with the different users of the BNMP to have their help with detecting the disease. The information shared with the branches, through letters and social media posts, about the disease and how to report it can be found in appendix II.

Per the publication of this report (November 2022) we are still monitoring our reefs but we have opened up the dive sites 'Karpata' and 'La Dania's Leap' again by placing back the moorings for the boats and opening up the entrance of these dive sites. The reasoning being that the disease observed at these sites does not present with the typically observed spread of SCTLD. It has so far only really affected one species, the pillar corals, and is not spreading at the speed observed of SCTLD. It is possible we are dealing with a particularly aggressive form of white plague that is attacking pillar corals first, but due to the lack of diagnostic tools for identifying coral disease we can't be certain one way or the other. We are very concerned about the disease situation on our reef (especially in the north and on the pillar corals) and will continue to monitor the situation. As part of an ongoing elkhorn and staghorn mapping project, in collaboration with Reef Renewal Foundation Bonaire and Wageningen University & Research, we are mapping the distribution of remaining pillar corals in the BNMP. This will allow us to be better prepared for potential targeted conservation actions down the line.

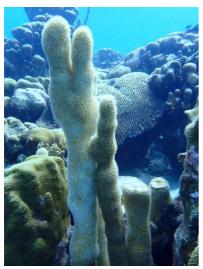






Figure 3. After the removal of the diseased branch from the pillar coral on which the disease was first identified, colony wide partial bleaching was observed; nearby pillar colonies were also showing signs of bleaching. At present, the colony seems to have recovered and the disease is no longer spreading.



BNMP Action Plan to Prevent and Manage SCTLD and other emerging coral diseases

Pre-invasion

In the pre-invasion phase, before the disease is confirmed on the reef, we are currently monitoring our reefs closely and are focusing especially on those areas in the north where the first corals with concerning symptoms were detected; from the first rounds of disease monitoring, it seems like there is more disease present overall in the BNMP than what has been reported previously in the literature. Whether this is an actual increase or an artifact of the monitoring protocols is unclear. Going forward, during routine monitoring already taking place we will try to align the protocols in a way that will provide us with more information. We have decided not to limit the use of the dive sites at the north but we are regulating the use by implementing rinse protocols. We are seeking the help of the user community of the BNMP to report to AGRRA with pictures (and locations) of disease present on corals and/or in our reefs.

It is still unclear how the disease spreads between and within reefs. As it's been hypothesized, the disease can spread through the water column as well as by direct contact, the two vectors that could potentially be introducing and spreading the disease on Bonaire that we are able to control are visitors coming from regions where the disease has been confirmed and ballast water disposal by ships. The former is being tackled through the gear rinse recommendations provided to operators and visitors as mentioned above. Ballast water disposal, while a threat for other regions, is deemed a minor concern for Bonaire.



Figure SEQ Figure * ARABIC 4. Location of Bonaire's 5 commercial ports.

Bonaire has 5 commercial ports, Kralendijk, WEB, BOPEC, Cargill, and the airport jetty at Donkey Beach, of which one, BOPEC, is not currently in use. The Marine Environment Ordinance prohibits damaging or destroying the Bonaire National Marine Park while the Harbor Ordinance allows full discretion to the Harbor Master to prevent any and all ships from docking, as a preventative measure, if there is cause to believe there might be a problem with the ship. The local government has also approved the 'Ballast Water Declaration' which states that the captain needs to sign for the following:

'Ballast water must be taken on board at least 12 miles offshore and contain clear oceanic water devoid of any obvious riverine or coastal influence.'

The Ballast Water Management Convention came into effect on September 8th 2017 and requires all vessels to have a fully operational ballast water treatment plant on board by (at the latest) September 2024. Vessels are required to exchange ballast water as far from the nearest land as possible and always in waters at least 50 nautical miles from the nearest land and in



water of at least 200 meters depth; taking into account any particularly sensitive areas of marine protected areas designated in the region. A ship shall not be required to deviate from its intended voyage, or delay the voyage, in order to comply with the rules- and regulations.

Bonaire has strict ballast rules and regulations, some of the strictest in the region, which are limiting the threat of SCTLD spreading to the Bonaire National Marine Park via the discharge of ballast water. Additionally, due to the nature of the cargo exchanges happening on Bonaire, where more often than not cargo is being unloaded, ships are often taking on and not discharging ballast water within our territorial waters. It is important to communicate this to make sure, as much as possible, that everyone is adequately informed and we can avoid unneeded discussions.

Monitoring

The dive sites Karpata, Kalli's Reef and Small Wall were monitored weekly, mostly on Friday, to determine the spread of coral disease present in these reefs. Once the spread slowed/stopped we scaled back the monitoring. Going forward we will monitor disease that rapidly spreads at certain dive sites or reefs on a weekly basis. We are doing this to determine whether or not we have SCTLD and to attempt to stay ahead of the infection when/if it appears on Bonaire's reefs.

During the bleaching monitoring of 2022, we will also focus on the levels and types of disease present on the reefs to get a better understanding of the background disease prevalence on the reef. These surveys will be conducted on the leeward side of the island at the dive sites, from north to south, Playa Funchi, the Rei Willem Alexander No-Dive Reserve (hereafter referred to as RWA Reserve), Karpata, Oil Slick Leap, Reef Scientifico, Kas di Regatta, Invisibles, and Vista Blue; the Klein Bonaire sites from east to west were Ebo's Special and Mi Dushi. These surveys should be conducted before SCTLD is present on our reefs (some monitoring has already happened after we thought we had an outbreak of STCLD). After we have an outbreak of SCTLD, we will continue monitoring these sites to see how the disease prevalence changes and document mortality caused by the disease. We will also ask stakeholders and/or partners (volunteers) to assist with the monitoring. After SCTLD is confirmed, and spreading, we will return to these sites a year later to see how many corals and/or reefs have died due to SCTLD.

Pre-invasion disease surveys will be conducted in tandem with the bleaching monitoring and at a minimum, in the event there is no bleaching, biennially. Surveys done during bleaching monitoring will be done at depths of 25m, 10m, and 5m (if there are corals present) using a line intercept method. A transect tape of 30m is laid down on the reef and every coral colony falling beneath the survey tape will be classified by species and size class. Additionally, we will collect data on disease (if present which type), live tissue under the transect, and bleaching status (if relevant). Photos, for the permanent record, will also be collected. Once compiled and analyzed the data will be publicized in a report produced by STINAPA. In the event there is no bleaching, disease prevalence will be determined from photo quadrats taken during routine reef monitoring conducted by our partners. If and when the disease is detected on Bonaire, more targeted monitoring to determine spread and mortality will be conducted.

Training



SCTLD can affect over 20 species of coral but we know that some species are more susceptible to the disease than others. We are especially monitoring the reefs for signs of the disease on the highly susceptible coral species which are pillar, maze, flower, and elliptical star corals.



For effective monitoring, it is important that our rangers are trained to assist (better) with research and monitoring that needs to happen on our reefs due to outbreak of disease. This is a point of focus for our Nature Unit (team of biologist) and needs to picked-up as soon as possible so that for future disease outbreaks we can mobilize quickly and as a unit. This requires training in the field and education for our rangers to be able to differentiate the different types of corals present in our reefs. It is also necessary to impress upon our ranger team, and the wider public, the importance of this research, as participating in research without understanding the need to do so is not giving the connection with the problem as is desired. We need to prevent this from happening at all times.

Communication & Disease Reporting

Our department of Communications will draft, whenever possible and necessary, a monthly update to be shared with our government, stakeholders, partners, dive operators and tourists. It is important to not only report in words but also in images (to show what is actually happening in our reefs). As it is difficult for us to have eyes on all the reefs on Bonaire, we need to keep asking the users of the BNMP to report disease at the AGRRA coral outbreak site. With the help of our partners, stakeholders, and visitors, we can more efficiently monitor the spread of coral disease in our reefs. We are working with the AGRRA portal to report and track signs of disease on corals and/or in our reefs. Our team of biologists are tracking the reports and will, when necessary, enlist the marine park rangers to assist with monitoring of a specific coral and/or reef in the BNMP. It is important to communicate effectively with stakeholders (particularly divers, snorkelers and cruise ship companies) about our corals and reefs. We need to focus on this by communicating directly with them (and on our social media outlets) about the spread of disease and the results of research done.



SCTLD Prevention & Interventions

During the initial phase of infection, we will attempt to quarantine and eliminate the disease. When deemed necessary, our rangers will remove (parts of) corals that are affected with a rapidly spreading coral disease. We have seen that this can help with stopping the spread of disease on the coral and the reef as was the case at Karpata. We are currently looking into the use of antibiotics and delivery agents to be able to treat corals affected with SCTLD. In other regions, the use of Coral Cure Ointment Base2B, a compound produced by Ocean Alchemists and used to improve the delivery of treatments for coral disease, with antibiotics has proven effective at halting the spread of lesions on a single colony. While this method is not a cure or a vaccine, it can be an effective treatment option; the combination of Base2B with the antibiotic amoxicillin has proven to be over 85% effective at treating SCTLD lesions. For a full breakdown of the Base2B with amoxicillin treatment application, please see Appendix III. We have an initial order for the compound and will, when deemed necessary, use it to treat rare coral species and very large and/or old colonies affected with SCTLD.

Considering the current extent of the reefs, it will not be feasible to treat every reef or every coral if and when SCTLD starts to spread on Bonaire. To be able to effectively treat high priority/high value sites will require the help of our stakeholders, partners, and volunteers. STINAPA will prioritize the treatment of colonies in the no-dive reserves by our staff; for additional reefs we hope to enlist the help of the dive operators to treat their house reefs.

A treatment plan, evaluated by our biologists and regional partners will need to be created that takes into account, for each participating operation, what frequency and intensity of treatment application is feasible. STINAPA rangers and biologists will provide instructions and training on how to apply the Base2B amoxicillin mixture. A Memorandum of Understanding (MoU) with anyone applying treatment options will need to be created in order to regulate the use of antibiotics for the treatment of SCTLD on coral colonies in the BNMP. It merits emphasizing that treatment with antibiotics, while beneficial to the colony, will not contain the spread of this disease and should be considered as an "ultimum remedium"; this is the last resort option. Prevention of the disease remains the number one priority and goal.

Next to treatment options, there are also coral rescue options such as ex-situ preservation (in aquaria) and collection of genetic materials for larval propagation. Considering the resources currently available on island and within the organization, we have opted not to pursue the option of ex-situ conservation in aquaria. As an organization we are open and willing to participate in any such programs initiated by other parties, such as the local and national governments, but do not have the capacity nor the knowledge required for maintaining corals in aquaria for long periods of time. Considering the duration of the outbreak in Florida, this could require a decades-long commitment and is a project that will require significant funding and expertise.

Another avenue being explored for reef rescue and restoration following SCTLD outbreaks is larval propagation, particularly for species with low population numbers and high susceptibility to the disease. STINAPA Biologist Caren Eckrich, as well as Reef Renewal Foundation



Bonaire (RRFB) coordinator Francesca Virdis, attended the Reef Futures workshop in September 2022 to learn more about this option. Not only do we consider RRFB a valuable partner for the management and monitoring of the state of our reefs, STINAPA supports RRFB's larval propagation activities and will assist/supplement wherever and whenever necessary. We have already started some preliminary mapping and monitoring of the pillar corals in Bonaire; identifying colony locations and spawning times will facilitate conservation and restoration efforts down the line. If, following SCTLD, it is deemed necessary, larval propagation as a way to rehabilitating reefs following disease outbreaks is something we are very open to exploring.

There is a growing body of literature on the benefits of improved water quality on reef health and biodiversity; multiple theories suggest that reef resilience to disturbances can be managed through water quality improvements. STINAPA, through various projects, is engaged in research and monitoring aimed at monitoring the water quality in the BNMP and finding targeted interventions that can help to maintain and/or improve water quality. One project we are directly involved in, with more immediate practical application, is the ban on discharge of waste water by boats using the overnight moorings in town. As of September 1^{st,} boats without a holding tank are no longer welcome to use the overnight moorings and need to dock in one of the harbors of Bonaire. Boats with a holding tank need to use the pump out facilities at Harbor Village Marina. No discharge is allowed. We are currently waiting for the Public Prosecutor to approve a shortened charge to use when art. 4 Island Decision Underwater Park Bonaire is violated. This article states the following:

'It is forbidden without a permit of our local government to direct or indirect discharge waste water, biological or chemical agents, which can harm the environment or can change the physical composition of the water, to deposit or to discharge, to leak or to do or let it get into the Bonaire National Marine Park.'

Conversations are also ongoing to examine the possibilities to extend the prohibition of use of moorings by boats without a holding tank to our private and public moorings. As we recognize that the wastewater being discharged is only part of the picture, we will also intensify the supervision, and where necessary, the enforcement, on buildings along the shoreline dumping wastewater in the Bonaire National Marine Park.

<u>Legal Preparation – Antibiotic Use</u>

The use of antibiotics in an open water system like the Bonaire National Marine Park is prohibited; to be able to implement treatment options, permission will need to be granted by the local government (and, if deemed necessary, the national Dutch government). STINAPA currently manages the BNMP, by mandate of the local public entity of Bonaire (OLB), and has the authority to take decisive action, such as the removal of infected coral colonies, when deemed necessary. However, the use of antibiotics, as an activity that does not fall under regular management actions, requires the approval of the local government. Permission needs



to be requested and granted prior to an official, and widespread, outbreak of SCTLD. In the case of an outbreak, antibiotics will be used to:

- Treat specific coral species at higher risk of extinction (either locally or regionally)
- Treat large (and thus old) coral colonies
- Treat reefs with high nature value (high biodiversity, relatively healthy reefs)

The reasoning behind this is based on the vast area of highly biodiverse coral reefs (over 27km²) surrounding Bonaire; it will be impossible to treat every affected coral colony. Decisions to prioritize those corals/reefs with the highest potential to survive and repopulate depleted reefs need to be made. This way we can effectively and efficiently treat those reefs that harbor the highest biodiversity, while also giving the reef a chance to recover following the SCTLD outbreak. This is also in line with the Treaty of Biodiversity.

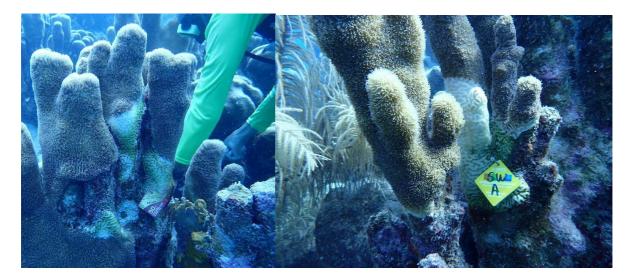
Summary

The current focus to prepare for a possible outbreak of SCTLD should be on the following aspects:

- Keep emphasizing the need to rinse dive gear (per protocol) and the need for the last dive and/or snorkel to be in the north (when visiting multiple dive sites);
- obtain permission in advance for the use of antibiotics to combat coral disease;
- order BES2B and antibiotics:
- create an action plan around the use of treatment and antibiotics that focuses on the where, how, when, who, why and so on;
- create an action plan for priority sites that will be monitored for a possible outbreak of SCTLD;
- training of the rangers to be able to assist with monitoring;
- training of the rangers to be able to assist with treatment of coral(s) (colonies) and to be able to inform dive shops about the use of treatment;
- organize an information session about the use of antibiotics on coral(s);
- as much as possible, monthly communication with stakeholders about the monitoring and research done by rangers and biologists of STINAPA on coral disease;
- proceed with the water quality program that is already being executed.

This document is to be used as a living document to be adopted and/or adapted as deemed necessary. Past outbreaks have shown how detrimental fast moving and highly lethal disease can be for the reef. At present we are unsure of the nature of the disease plaguing some of the corals in the BNMP; there are indications it could be an aggressive, fast moving, form of white plague, but it could also be an outbreak of SCTLD that is behaving a-typically. Because we don't know, and there are no diagnostic tests available to confirm one way or the other, it is necessary to work from the precautionary principle and take all measurements deemed fit to manage any concerning disease outbreaks in the Bonaire National Marine Park.





We thank you for your interest in the content of this action plan and we trust we have informed you sufficiently for now. Please do not hesitate to contact the manager of the Bonaire National Marine Park (marinepark@stinapa.org) if you have any questions and/or remarks.



Appendix I - SCTLD Pre-invasion Monitoring Results

The ever-increasing Caribbean population size, and in tandem the level of anthropogenic pressure and activity in watersheds, has been suggested to be linked with the susceptibility of corals to various disease outbreaks. Since the discovery of disease on coral reefs, and following the first regional outbreak of white band disease (WB) in the early 1980's that resulted in the mortality of over 80% of elkhorn (*Acropora palmata*) and staghorn (*Acropora cervicornis*) corals, we have witnessed expansion of both the geographic ranges as well as the possible host species. In the Caribbean, the most common diseases found on hard corals (stony corals) are: white plague (WP), white-band disease (WBD), black-band disease (BBD), yellow-blotch disease (YBD), dark-spot disease (DSD) and very rarely; red-band disease (RBD). On average, disease prevalence on the reefs has historically been low, but with some seasonal variability, variability within and between reefs, and annual variation. Many of the diseases mentioned above, with the occasional exception of WP, affect only a handful of species and move relatively slowly.

In 2014, a novel disease outbreak was discovered on reefs in Virginia Key (FL, USA). Initially the disease spread slowly through the region and preferentially affected pillar corals (*Dendrogyra cylindrus*), maze corals (*Meandrina meandrites*), and elliptical star corals (*Dichocoenia stokesii*). These species were therefore classified as high vulnerability/high susceptibility species. However, symptoms of the disease were soon found on other species as well as the disease made its way across the reef and through the region. At present the list of affected reefs counts 25 countries and territories. In other places SCTLD lesions have been observed on over 20 of the 45 hard coral species found on Bonaire; luckily species belonging to the Acropora group seem to have a higher resistance to the disease.

In other regions where the disease has invaded and spread, the change to the reefscape has been profound and severe; significant ecological shifts and changes in reef composition, and therefore function, have been documented in other areas. Many colonies, of all sizes, died and so far the disease shows no signs of stopping. The pathogen(s) and underlying mechanisms resulting in infections remain unknown, but research is ongoing. It is expected to take decades for affected reefs to be able to recover. Knowing the devastation that this disease can result in for the reef, once the first symptomatic colonies were discovered STINAPA acted proactively by closing the affected site. Karpata, as well as the site to the south (closures of the more northern sites were not necessary as Karpata is bounded in the north by the no dive reserve). We also immediately started monitoring on reefs around the island to quantify the extent and spread of the disease.

Survey Methodology

To be able to quickly cover as many sites as possible an adapted roving diver survey (RDS) wallmethod was used. During the months of July and August STINAPA marine park rangers conducted preliminary surveys at three different depths (15m, 10m, and 5m) and quantified the number of diseased colonies observed per minute swimming. Due to Bonaire's remarkable coral cover, and to keep the process workable, the total number of colonies surveyed was not accounted for. At each depth, two transects were conducted by swimming for five minutes in one direction (usually northward) and five minutes in the opposite direction (usually southward). Pictures were taken of all colonies that showed SCTLD-like tissue loss symptoms. For this survey, black band disease, red band disease, and dark spot disease were therefore excluded. Pictures were analyzed per site, identifying species and determining which disease



was found on each infected coral. The effort needed to survey each site was used to determine the prevalence of disease per site by dividing the total number of disease sightings over the effort in minutes.

In addition to the surveys, weekly monitoring of the affected colony at Karpata as well as concerning corals at Kalli's Reef and Small Wall were also conducted following observations of disease impacting the pillar corals at these sites. Opportunistic visual inspections were also conducted at a few sites along the east coast when weather and time permitted them.

Results

Fifteen sites were surveyed on the West coast of Bonaire and Klein Bonaire to determine the prevalence of coral disease. Coral diseases found in the survey areas included white plague (WP), yellow-band disease (YB), unknown diseases (UNK), and possible stony coral tissue loss disease (SCTLD). Of all the sites surveyed, for 15 sites the data was reliable enough to be included. For an overview of all the sites surveyed and the diseases found, please see table 1.

Table 1. Overview of diseases found and disease prevalence per site surveyed

Site	WP	YB	UNK	SCTLD	Total	Effort	Prevalenc
							e
Queen Maxima Marine Reserve	1	0	2	0	3	60	.05
BOPEC	1	0	1	0	2	60	.03
King Willem Alexander Reserve	1	0	1	1	3	60	.05
Karpata	5	0	1	0	6	60	.10
Bloodlet	3	3	3	0	9	30	.30
Tolo	0	0	0	2	2	60	.03
Country Garden	7	0	0	1	8	30	.27
1000 steps	1	1	0	1	3	30	.10
Jeff Davis	0	0	0	1	1	30	.03
Knife	2	1	1	0	4	60	.07
Carl's Hill	1	1	0	0	2	30	.07
Munks Haven	2	0	1	1	4	30	.13
Mi Dushi	0	0	0	0	0	30	.00
Chez Hines	2	3	1	0	6	30	.20
Corporal Meiss	0	0	0	0	0	30	.00

Of all diseased corals encountered during the surveys, 49% were affected by WP, while YB was the second most commonly occurring disease present on 21% of the diseased corals found in the survey. In 17% of the cases the disease could not be reliably identified; these were classified as unknown. At 6 sites, colonies with lesions that resembled SCTLD, accounting for 13% of all disease sightings, were found. In all instances except Tolo only one suspicious colony was found. At Tolo 2 colonies were identified. None of the lesions were found on species categorized as highly susceptible species. At two dive sites, Mi Dushi and Corporal Meiss, no diseases were found during the surveys. All these results are summarized in figure 2.



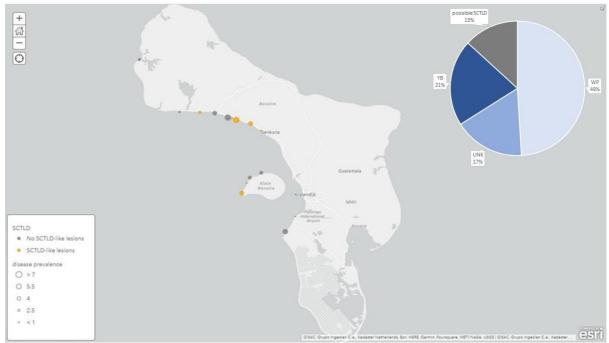


Figure 5. Disease prevalence at 15 sites on the west coast of Bonaire. The size of the markers denotes the total disease prevalence; orange-colored markers indicate sites where SCTLD-like lesions were detected during the survey. The pie chart shows the relative frequency, in percentages, of the occurrence of the different diseases surveyed.

During the weekly monitoring at Karpata, Kalli's Reef and at Small Wall we saw the disease initially spreading on the affected colonies, followed by the detection of more infected pillar colonies, and one elliptical star coral (only at Karpata). For Karpata, the decision was made to remove the actively diseased coral branch in an effort to limit the spread. Following this the colony showed signs of partial bleaching, but as per the last inspection in October, the disease no longer appears to be spreading either on the colony or across the reef. Similar observations were made for the pillar coral colonies monitored at Kalli's Reef and Small Wall.

Discussion & Conclusion

After discovering the first affected colony, local and regional experts were consulted in an effort to determine if we were dealing with a case of SCTLD. At present there is no official consensus on the identity of the disease(s) found during these surveys. As SCTLD can't be confirmed by diagnostic tests, the determination is often made based on the pattern of the spread. This leads us to believe that we either do not have SCTLD in Bonaire detected yet or the disease is behaving markedly different from how it has spread in other areas. While the initial lesions did present in similar ways as they have in other infected areas, there has been no subsequent spread across the reef scape. After analyzing the data and the species on which SCTLD-like symptoms were detected, disease was predominantly found on boulder corals belonging to the Orbicella spp., not categorized as high susceptibility species. The severity of the disease outbreak seems to increase during periods with warmer waters associated with mass bleaching events; Bonaire is currently under a mass bleaching advisory, heightening the sense of urgency. During the bleaching monitoring we will also be repeating these disease surveys. As concerning cases arise these will be evaluated on a case-by-case basis to determine the best path forward. We will also continue our cooperation with local and regional partners and experts to determine the best path forward to continue to safeguard the reef.



Appendix II – Outreach to the users

Dear partners,

Through this letter we wish to provide you with an update on the stony coral tissue loss disease (SCTLD) situation in Bonaire. On Friday July 22nd, following reports from concerned divers, we conducted a visual inspection at Karpata. During this inspection we found what we suspected to be SCTLD and took the difficult decision to close this dive site to curb the spread of the disease. For the past 3 weeks, STINAPA has been closely monitoring coral disease on the entire west coast and on Klein Bonaire. Many of you (and concerned divers) have sent in hundreds of photos. Currently we have some corals on 2 reefs that have what looks like SCTLD, but so far it does not seem to be spreading to other corals and has not been observed on other reefs. For these reasons, after consulting with local, regional, and international experts, it's at the moment unclear if what we detected at Karpata is indeed SCTLD or a particularly aggressive form of white plague, a disease that has been present on our reefs for quite a while now.

As a precaution the diseased corals of concern, those on which particularly rapid tissue loss was observed, were removed. We will keep Karpata and La Dania quarantined (no diving/snorkeling/swimming) to observe if the disease continues to spread and determine the best way forward. We will keep you informed of future developments. In the meantime, we greatly appreciate your help in tracking the health of our reefs by filling out a quick survey (https://www.agrra.org/coral-disease-outbreak/). This helps us to keep track of what's happening on our reefs and very much appreciate your contribution to the database.

STINAPA requires all users to keep up with the recommended decontamination protocols (https://www.agrra.org/wp-content/uploads/2019/03/Florida-coral-disease-decontamination-protocol.pdf). The general guidelines being: required decontamination of all gear prior to the first dive in Bonaire as well as at the end of each diving day. A dilute bleach solution is recommended with a thorough rinse in freshwater. Ideally, dive shops will rotate three tanks: an active rinse tank (with bleach), a pure freshwater rinse tank, and a non-active tank (one day in the sun will allow the bleach to become non-toxic) that will be emptied at the end of the day. Separate smaller tanks/buckets may be used with antibacterial soap for very sensitive gear. This water should be disposed of properly. In addition, we strongly recommend only diving one area (North, Middle, South or Klein) per day to limit potential spread of the pathogen.

We hope to have informed you sufficiently and please do not hesitate to contact us with any questions you might have.

Sincerely,

Judith Raming,



Manager Bonaire National Marine Park STINAPA

Additional letter to inform your guests how they can help by encouraging them to fill out the survey

Help us track the health of Bonaire's reefs!

STINAPA is asking citizen scientists and anyone who is interested to pay attention during their dives to corals that are particularly susceptible to SCTLD. Species like the pillar, maze, and flower coral are very vulnerable to SCTLD and are usually the first species that start to show symptoms. Filling out the survey is very easy and only takes two minutes, just follow this link: https://www.agrra.org/coral-disease-outbreak/

Once on the page scroll down to the green button that says "SUBMIT Basic Coral Disease & Bleaching Survey".

SUBMIT Basic Coral Disease & Bleach Survey

For Bonaire most surveys will be done on the 'forereef'; if unsure of what disease it is, please tick the box indicating 'Corals with Recent Mortality'. Whenever possible, be specific in the notes on the depth and location of the colonies of interest (for example: swim 7 minutes east of the mooring line and the colony is at a depth of 6m). If you are also submitting pictures, ideally an image of the entire colony as well as close-ups of the diseased region are submitted. For pillar corals we also appreciate observation of healthy colonies; we are trying to document the current extent of this species that has been declining rapidly in the Caribbean. Once submitted, AGRRA compiles all of the survey results and observations so that anyone can view them. This is an excellent way to help us track the health of the reef and we greatly appreciate your help contributing to the database.

Pillar coral (polyps out in the daytime)	Maze coral	Flower coral



Appendix III - Antibiotic treatment methodology

We are currently considering the use amoxicillin treatment to be able to treat rare coral species and very large and/or old colonies affected by SCTLD. In order to use this treatment option, it is important to train rangers, biologists, and volunteers in the use of this treatment. Below you will find the Amoxicillin treatment protocol currently being in other regions dealing with SCTLD outbreaks.

- 1. Protocol for Topical Antibiotic Treatment
 - 1. Create an amoxicillin mixture utilizing powdered amoxicillin and Ocean Alchemist / Core Rx Base 2b. Take appropriate precautions for working with chemicals/pharmaceuticals.
 - 2. Mix powdered amoxicillin into the Base in a 1:8 by weight ratio.
 - a. For a single small coral (eg in a nursery or a single target), 2.5 g amoxi + 20 g of Base is appropriate. For field treatments at a high-density site, a single experienced diver (~ 6 hours of bottom time) can use ~50 g amoxi + 400 g of Base.
 - b. For smaller amounts, a balance is needed to weigh out the ratio. For larger amounts, Base jars come in 400g amounts, so 2 jars can be mixed with 1 jar of 100g amoxicillin and stirred on the boat.
 - c. Mixing can be done with a metal spatula, butter knife, etc. This can easily be done on the boat for larger quantities.
 - 3. Pack the mixture into a syringe. 60cc is recommended for lengthy field days. A catheter (tapered) syringe can be helpful as it can be cut higher up if application is difficult.
 - 4. Pack a goody bag with antibiotic syringes and modeling clay. Syringes are positively buoyant; modeling clay is negatively buoyant. Be careful of how you secure and close your bag. Sticking globs of modeling clay onto the syringes is suggested.
 - 5. Select your lesion and use the syringe to apply base over the lesion margin. Follow with a finger to press down onto the immediately surrounding area (it will be ~1 cm wide, with approximately half of that anchoring onto recently dead skeleton and the other half overlaying the live tissue). It adheres better to the skeleton than to the tissue, and may ultimately require some manipulation with your fingers to apply it. Small pieces may detach during application, but can generally be caught and remolded into to the application.
 - a. Compound sticks to nitrile gloves, which are not recommended. Other glove materials have not been tried but may be effective.
 - 6. Alternative or additional intervention can be accomplished by creating and applying the compound to a firebreak about 5 cm away from the disease margin. An underwater angle grinder provides a rapid and clean trench, but this can also be accomplished with a hammer/chisel. Use the syringe to squeeze the amoxicillin mixture into the resulting trench, and then consider covering it with modeling clay. The clay helps hold the mixture in place, as it tends to wash out of the trenches easier than off of the margins.