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Intervention of stony coral tissue loss disease in Utila, Honduras using Base2B and amoxicillin treatments

Intervención de la enfermedad de pérdida de tejido de corales pétreos en Utila, Honduras usando tratamientos con Base2B y amoxicilina

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Abstract / Introduction. Coral diseases are on the rise due to anthropogenic stressors, and treatment is challenging due to the complexity of replicating natural reef environments. Stony coral tissue loss disease (SCTLD), detected in 2014 off the coast of Florida, has become one of the most lethal modern coral diseases, spreading throughout the Caribbean to Utila, Honduras. **Methods.** Monthly monitoring and in-situ treatments for SCTLD on 97 coral colonies over 12 months were carried out using amoxicillin and Base2B in two sites of Utila, Honduras. The paste is applied around the margins of the lesion with careful attention to application technique to maximize efficiency. **Results.** Of the 97 monitored corals 91.75% (n=89) of the colonies survived, with the two sites having survival rates of 97.96% and 85.42%. A total of 842 lesions were treated with only 45 lesions remaining active by the end of the monitoring period. Although treatment intervention halted the lesions it was directly applied to, it did not stop the appearance of new SCTLD lesions on coral colonies. **Conclusion.** It was found that the treatments themselves are highly effective. Previously infected and healed corals were observed spawning indicating that the treatment works in keeping the corals healthy enough to reproduce. This treatment is suggested as early-stage intervention to help keep as many genotypes of highly susceptible species viable as possible.
Keywords Antibiotic, Coral reefs, Honduras, SCTLD, Treatment

Resumen / Introducción. Las enfermedades en los corales están aumentando debido a factores estresantes antropogénicos y el tratamiento es un desafío debido a la complejidad de replicar los entornos naturales de los arrecifes. La enfermedad de pérdida de tejido de los corales pétreos (EPTC), detectada en 2014 frente a la costa de Florida, se ha convertido en una de las enfermedades nuevas de los corales más letales y se ha extendido por todo el Caribe hasta Utila, Honduras. **Métodos.** Se llevaron a cabo monitoreos mensuales y tratamientos in situ para EPTC en 97 colonias de coral durante 12 meses utilizando amoxicilina y Base2B en dos sitios de Utila, Honduras. La pasta se aplica alrededor de los márgenes de la lesión prestando especial atención a la técnica de aplicación para maximizar la eficacia. **Resultados.** De los 97 corales monitoreados, el 91.75% (n=89) de las colonias sobrevivieron, y los dos sitios tuvieron tasas de supervivencia del 97.96% y del 85.42%. Se trataron un total de 842 lesiones y solo 45 lesiones permanecieron activas al final del período de seguimiento. Aunque la intervención del tratamiento detuvo las lesiones a las que se aplicó directamente, no detuvo la aparición de nuevas lesiones EPTC en las colonias de coral. **Conclusión.** Se descubrió que los tratamientos en sí son muy eficaces. Se observó el desove en corales previamente infectados y curados, lo que indica que el tratamiento funciona para mantener los corales lo suficientemente sanos como para reproducirse. Este tratamiento se sugiere como intervención en una etapa temprana para ayudar a mantener viables tantos genotipos de especies altamente susceptibles como sea posible.
Palabras Clave Antibiótico, Arrecifes de coral, EPTCD, Honduras, Tratamiento

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INTRODUCTION

The health of the Mesoamerican Reef has significantly declined in recent years, as highlighted in the 2024 report by the Healthy Reefs for Healthy People Initiative (McField et al., 2024). In Honduras, coral cover has dropped from 27% in 2010 to just 21% today. This decline has been driven by a combination of factors, including disease outbreaks, coral bleaching events, and hurricanes, while overfishing and pollution continue to pose ongoing threats to the region's reefs. The reduction in coral cover has far-reaching implications for marine ecosystems, particularly for species that rely on coral reefs for shelter and sustenance. As coral reefs deteriorate, many reef-dependent species face the risk of population decline, leading to disruptions in marine biodiversity. This

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degradation also impacts coastal communities that depend on healthy reef ecosystems for their livelihoods, further emphasizing the urgency of addressing the threats to these vital marine habitats.

Like any community, coral populations live with the threat of diseases. The first reported coral disease was published in Squires (1965). Coral diseases likely existed long before that, however isolation and identification of coral disease pathogens is difficult. When using Koch's postulates for demonstrating the identity of a pathogenic microorganism's difficulties arise in replicating the natural reef environment including currents and aeration as well as contamination which causes complications in proving the re-isolation of pathogens from newly diseased experimental coral, making it difficult to establish a clear cause of the disease. Without a clear cause of the disease identifying possible treatments is challenging (Richardson, 1998). There has also been a rise in the prevalence of coral diseases, and this is most likely due to human influence. Many coral diseases are correlated to environmental stressors, stressors that are worsened by anthropogenic causes (Walton et al., 2018). These coral diseases are projected to become just as detrimental to coral reef health as coral bleaching (Maynard et al., 2015).

Stony coral tissue loss disease (SCTLD) is particularly prevalent and devastating. SCTLD was first detected in 2014 offshore of Virginia Key, Florida, and is one of the most lethal modern day coral diseases ever recorded (Precht et al., 2016). By 2023 the disease had spread throughout the Caribbean. The first detected case in the Bay Islands of Honduras was in Flowers Bay, Roatan, in September of 2020. SCTLD appeared on the north shore of Utila on June 28, 2021. SCTLD spread throughout the Bay Islands over a period of 13 months, impacting Roatan and Utila the most. It is estimated that the disease spread at a rate of 155 m/day around both islands and has severely impacted at least 28 different species (Truc et al., 2023).

SCTLD was originally described as "white plague-like disease or symptom" (Precht et al., 2016). SCTLD manifests as a lesion on the surface of hard corals, often bordered by bleaching as the coral colony responds to the presence of the disease. The cause and transmission sources of SCTLD are not presently known. Studies examining bacteria present at SCTLD lesions have identified five common families, but no common families were found across all coral species examined (Meyer et al., 2019). Another study examining the microbial diversity in water samples collected from regions of increasingly dense SCTLD presence found significant variations in the microbial communities present in water and sediment samples, with *Rhodobacterales* concentrations correlating to SCTLD presence (Rosales et al., 2020).

In 2019, several treatment plans were developed and tested in order to address the mass casualties caused by SCTLD since its initial discovery. Application of amoxicillin paste directly to lesion borders has shown to be most effective. Though unable to stop the development of new lesions, it does halt or slow the growth of existing lesions (Neely et al., 2021). Similar treatment plans have been replicated at various sites across the Florida Keys and Mesoamerican Reef with similar success.

To date, Honduras follows the Ballast Water Management Convention and the International Convention for the Prevention of Pollution from Ships (MARPOL), in an attempt to hinder the spread of SCTLD through ship ballasts (Lee Hing et al., 2022; Rosenau et al., 2021). From 2020 to 2021, the Bay Islands Conservation Association (BICA) monitored the arrival and spread of the disease throughout the Bay Islands with monthly surveying efforts (Truc et al., 2023). Numerous organizations have actively contributed to treatment efforts across the Bay Islands including Roatan Marine Park, Bay Islands Conservation Association (BICA), Whale Shark and Oceanic Research Center (WSORC) and local dive shops. Through this study, we seek to deepen our understanding of how these targeted treatment applications can enhance the recovery of vulnerable coral species across the reefs of Utila and to enhance our understanding of how coral species are affected by the growing number of environmental threats, such as disease outbreaks.

METHODS

Utila is an island located in the Caribbean Sea, 32 km northwest of the Honduran mainland city of La Ceiba. (McCranie et al., 2005). Utila, along with Roatan and Guanaja, compose the main groups of islands in the Bay Islands Department. It is the smallest of the three Bay Islands with an area of 42 km^2 (Goetz, 2006) and part of the Mesoamerican Barrier Reef System, the second-largest reef system in the world (Figure 1).

Figure 1. Study area, two sites of Utila, Bay Islands, Honduras.

This study focuses on the intervention and monitoring efforts targeting SCTLD conducted between March 2022 and December 2023. The dataset presented encompasses the initial 12 months of treatment applied to 97 individual coral colonies. Fieldwork involved 240 dives distributed across two sites on the island of Utila, Honduras: Spotted Bay (UTM coordinates: 501462.516, 1779553.904) and

Little Bight (UTM coordinates: 507515.404, 1777796.790). Both sites are popular recreational scuba diving locations and were characterized by the presence of susceptible stony coral species in the early stages of SCTLD infection at the start of the study. The study area was delineated using transect-based monitoring approaches. At Little Bight, three 45-meter transects were established, while Spotted Bay was surveyed using two 45-meter transects. These transects facilitated observation and documentation of disease progression, treatment efficacy, and coral recovery.

Treatment preparation and protocol is based on Base2B care instructions and Neely (2020). Amoxicillin treatment paste was prepared using Phytotechnology amoxicillin trihydrate and Ocean Alchemist / Core Rx Base2B Placebo Blend. Prior to use amoxicillin was kept in the freezer at -12° C and Base2B kept in the refrigerator at 1°C to prolong shelf life. The Base2B and amoxicillin were mixed together in an 8:1 ratio by weight (400g of Base2B to 50g of amoxicillin.) Mixing was done by hand in a metal bowl with a metal dough whisk for ten minutes. The mixture was put into 60mL/cc plastic syringes with a catheter tip cut to a diameter of 0.5 cm. The syringes were then placed into a fridge or cooler with ice for no more than 24 hours until the intervention dives.

On the intervention dives, SCTLD was identified in corals using criterion identified by National Oceanic and Atmospheric Administration (NOAA) in their case definition of SCTLD. this epidemiological criterion is based on which species show signs of lesions, and the rapid mortality loss observed across the site (Florida Keys National Marine Sanctuary, 2018). The paste was applied to a SCTLD lesion in an approximately 1 cm band around the lesion margin, with the paste about 25% on the recently dead skeleton and 75% on the live tissue. Paste should be applied with moderate force to ensure it reaches into the crevices of the coral and works best when squeezed out of the syringe in a long string and then pressed into the coral once separated from the syringe. This ensures the right amount of product is being used without overloading the coral with unnecessary treatment, while being wary of overhandling which causes treatment to break apart. Gloves were not used because of the treatment's adherence to gloves. For MCAV, SSID and DCYL species, small pieces of modelling clay were used to help anchor the treatment to the coral when the treatment would not stick on its own.

Monitoring occurred once per month at each site. Each colony selected for monitoring received a unique numbered tag for identification purposes. The tag was nailed into the non-living substrate near the base of the coral or into a non-living part of the coral itself, using a masonry nail. Colonies were selected based on a number of different factors including species, size, location, and treatability. A 45 m transect was placed at the chosen site and coral colonies were selected in the relative area of the transect. This was done to keep coral colonies within a reasonable working distance from one another and make them easier to find from month to month for monitoring purposes.

Large colonies were prioritized for their greater likelihood of resilience to long term environmental stressors that come from age. Bigger colonies also have a greater likelihood of a high reproductive output. Colonies that were selected but had no active lesions were tagged so that they could be monitored and treated if infected. Colonies needing immediate treatment with more than 75% remaining living tissue and fewer than 5 total lesions were prioritized. Having more remaining living tissue and fewer total lesions may signify greater overall health and a higher probability of treatment success. After one month, most corals clearly showed if the treatment had worked and the lesion showed no progression, or if the treatment had not worked and the lesion had progressed past the treatment line. If the treatment was successful, the coral was monitored for any new lesions. If the treatment was unsuccessful then the lesion was retreated. Retreatments only occurred if the coral colony had more than 50% remaining living tissue. Photographs were taken off the tagged colonies during every monthly monitoring dive both before and after treatment application.

RESULTS

A total of 97 coral colonies were treated and monitored for 12 months, colonies ranged across different stony coral species: *Pseudodiploria strigosa* (44.3%), *Diploria labyrinthiformis* (23.7%), *Colpophyllia natan*s (16.5%), *Montastraea cavernosa* (7.2%), *Orbicella faveolata* (4.1%), *Orbicella annularis* (3.1%), and *Siderastrea siderea* (1.0%). The average size of the coral colonies was 52.45 cm in width and 45.04 cm in height. Colonies were found in depths between 2 m to 15 m.

During the 12 months of monitoring, 8.25% of colonies died (n=8) and the remaining 91.75% of colonies survived (n=89). At the start of the monitoring a total of 17 healthy colonies were tagged. 82.35% of these colonies (n=14) were infected by SCTLD in the subsequent 12 months, only 17.65% (n=3) showed no signs of ever being infected by SCTLD. The Spotted Bay site had a survival rate of 97.96%, whereas the Little Bight site had a survival rate of 85.42%. SCTLD was determined to be the cause of death for all colonies, excepting one which appeared to be damaged by a storm.

In month two 29.90% (n=29) of corals needed retreatment as a result of failed initial treatments. Although fluctuations occur, the number of retreatment trends downwards over time. In the final month (12), 13.48% (n=12) of corals required retreatment due to failed amoxicillin application from the previous month.

However, treatment intervention did not stop the appearance of new lesions on the corals (Figure 2). Following the initial treatment, 28.87% (n=28) of the corals had newly developed lesions. At the end of the study 10.11% (n=9) of the corals had new lesions.

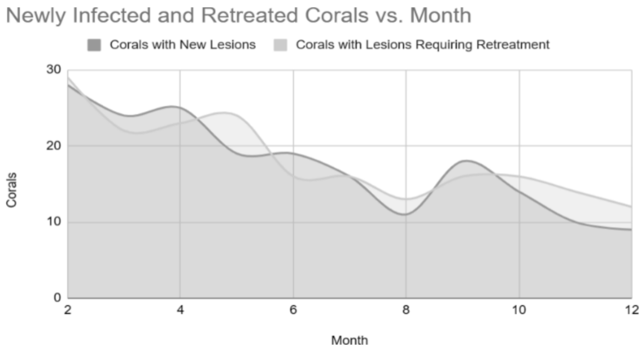

Figure 2. Retreated corals and coral with new lesions per month.

A total of 842 lesions were treated; at the end of the monitoring period only 45 lesions remained active. Treatment intervention did not stop the appearance of new SCTLD lesions on coral colonies; one month after the initial treatment 58.49% (n=62) of treatments were on newly developed lesions (Figure 3).

Appearance of new lesions did decrease over time, at the end of monitoring 42.22% (n=19) of lesions were new lesions. Lesions that didn't heal after the first treatment application were considered failed lesions, meaning that the disease continued to spread past the treatment line. One month after the initial treatment 41.51% (n=44) of applications failed. The number of failed lesions fluctuated over time, at the end of monitoring 57.78% (n=26) of the lesions were failed lesions.

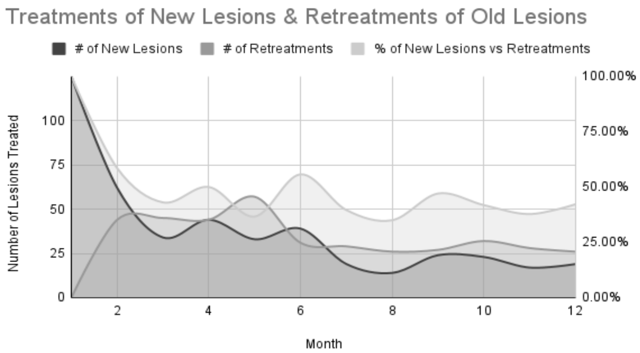

Figure 3. Treatment of new lesions or failed lesions over 12 months.

The percentage of colonies with no active disease (NAD) following initial treatment was just under half: 49.5% (n=48). Colonies' signs of active or no active disease varied over time as new lesions continued to appear, however, the percentage of corals with no active disease increased over subsequent months with an overall increase of 26.9% after 12 months. The NAD % was higher in the Spotted Bay site, with an increase of 34.4%. For the Little Bight site there was only a 17.5% increase in corals with no active disease over time (Figure 4a-4b).

Additionally, from April to July spawning monitoring for the *Diploria labyrinthiformis* species was carried out, 5 of the 10 colonies previously treated for SCTLD in the Little Bight site were monitored. Between May and June spawning was observed in 3 of the observed colonies, 11 days after the full moon and approximately 1 hour before sunset.

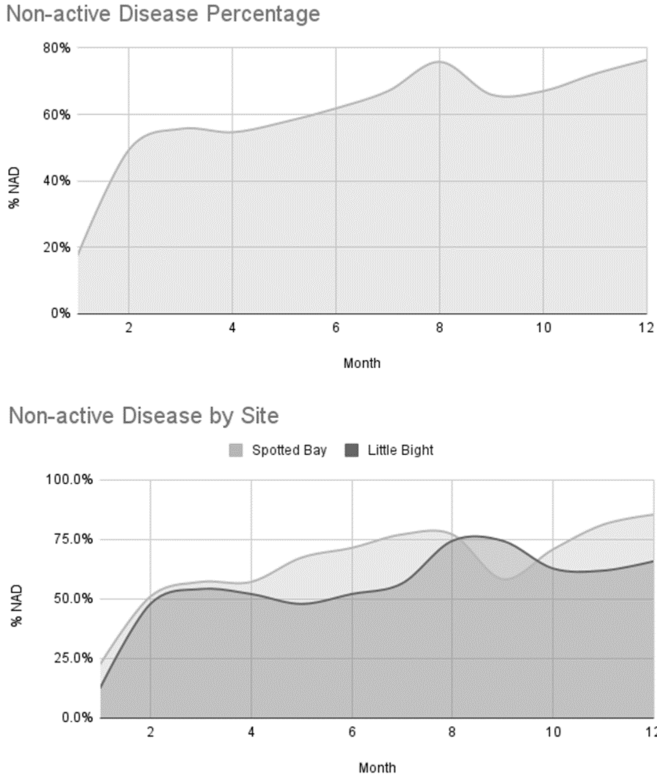

Figure 4a-4b. Non-active disease percentage over 12 months.

ANALYSIS AND DISCUSSION

Of the 97 coral colonies observed 8.25% of colonies died (n=8) and 91.75% of colonies survived (n=89). This gave an overall survival rate of 91.75% for both sites combined. This survival rate is very high and is a slightly higher success rate than the 88% of lesions prevented from expanding that the Bacalar Chico Marine Reserve (BCMR) was able to achieve with similar methods in July of 2020 (Lee Hing et al., 2022).

In a study conducted in the Florida Keys National Marine Sanctuary (FKNMS), the authors calculated a predicted effectiveness of 75-95% of amoxicillin treatments over a 3 month analysis (Neely et al., 2021). The Spotted Bay site had a survival rate of 97.96% and the Little Bight site had a lower survival rate of 85.42%. Little Bight was the initial site set up and it is likely that the difference between these two sites' survival rates could be explained by errors in treatment application.

In order to be successful, treatment application must be smooth and uniform around the entire lesion border and pressed into the coral to reach into all the grooves. It can be seen in photographic evidence that treatment applications in the first few months were not as smooth as they became in later months as experience was gained. These errors would have been minimal in the Spotted Bay site as all parties had experience with the prior treatment.

Figure 5. An example of patchy treatment application around a lesion of *Diploria labyrinthiformis* (5a). That treatment failed (5b).

At the start of the monitoring period 17 healthy colonies were tagged. These corals had no sign of disease but were tagged to be checked every month so that early detection and treatment was possible. 82.35% (n=14) of these colonies were infected by SCTLD in the subsequent 12 months as was expected and they were treated as necessary. Only 17.65% (n=3) showed no signs of ever being infected by SCTLD, this suggests that these genotypes are particularly resistant to the disease or have developed some type of defense against the disease (Meiling et al., 2021).

Of the three corals that remained healthy two were of the species *Montastraea cavernosa*. In disease progression studies *Montastraea cavernosa* has been identified as the species slowest to experience full mortality (Meiling et al., 2021). All corals have developed defense mechanisms that involve the production of mucus with distinctive properties, hosting beneficial bacteria, employing phagocytic cells, and utilizing antimicrobial chemicals to protect themselves from harmful pathogens (Mullen et al., 2004; Ritchie, 2006; Shnit-Orland & Kushmaro, 2009; Bourne et al., 2009; Mydlarz et al., 2010).

However, *Montastraea cavernosa* have significant differences in their defense capacities which could be why they are better at fighting the SCLD infection (Aeby et al., 2019). Characteristics like this are highly sought after restoration efforts and make *Montastraea cavernosa* a better fit for restoration than *Pseudodiploria strigosa* which has a higher SCTLD mortality rate than other species due to its shape causing sand to settle on it and transmit SCTLD more readily (Camacho-Vite et al., 2022). However, traits like the SCTLD disease resistance of *Montastraea cavernosa* can come with less desirable traits like a lower bleaching resistance which can still end in coral mortality (Shore-Maggio et al., 2015).

One month after the initial treatment was the first follow up and the first opportunity to see if any treatments were successful, meaning that the disease had halted at the treatment line and the remaining skeleton was covered in algae, this is also referred to as a “healed” lesion because the disease is no longer visibly present. In month two 29.9% (n=29) of corals needed retreatments because the treatment failed. This percentage continued to fluctuate month to month but in the final month only 13.48% (n=12) of corals needed retreatment. This can be attributed in part to the same idea as discussed above. Treatment applications in the first month were not as effective due to a lack of experience applying the treatment. This is true for certain species that are more difficult to apply treatment than others. The narrow grooves of the *Diploria labyrinthiformis* and the deep polyp grooves of the *Colpophyllia natans* can cause gaps in treatment application if not careful and experienced (Neely et al., 2020).

Healing a lesion on a coral unfortunately does not prevent new lesions. The exact cause of this is still uncertain, potentially meaning that the disease is systemic within a colony or that these recurring lesions are a result of reinfection from the environment. In another study, 11% of visually healthy corals were confirmed histologically to have necrosis despite their healthy appearance (Landsberg et al., 2020). It is likely that a SCTLD infection compromises a coral's immune system and makes it more susceptible to secondary infections caused by opportunistic bacteria (Shilling et al., 2021). Following the initial treatment, 28.87% (n=28) of the corals had newly developed lesions. At the end of the study 10.11% (n=9) of the corals had new lesions. This indicates that not only are the treatments helping the individual colony, but they may also be beneficial to the corals at the population-level, echoing the ideas that antibiotic applications are beneficial even to untreated corals (Forrester et al., 2022).

Suggestions for future research would be to use assisted sexual reproduction on thriving corals from the study and see if the treatments give any increased resistance to the resulting coral recruits. Also, future research could be done to continue to monitor these corals through any potential future bleaching events and see if the treatments continue to be beneficial when paired with bleaching events.

Conclusion

The antibiotic treatments are highly successful at stopping lesion progression, but they do not stop new lesions from appearing. Although highly successful, antibiotic treatments as a method for reducing the impact of SCTLD is useful on a small scale. Because reinfection is possible, and monthly monitoring is necessary for the continued health of the corals, this is a very intensive and expensive process not feasible for treating entire reefs or regions. Corals with healed lesions were observed spawning, indicating that they were in good health even after recovering from SCTLD. This treatment is suggested for use in early-stage infections to help keep highly susceptible genotypes alive and healthy on the reef while other solutions and restoration efforts are made.

Author contributions

All authors participated in the research, preparing the manuscript and approved its final version.

Conflicts of interest

None.

Ethics approval

Exempt.

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IA use

None.

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