

Bacterial Communities Associated with Healthy and Diseased Corals during a Heatwave Event in the Northern Red Sea, Egypt

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ABSTRACT

Coral disease is one of the major threats to coral assemblages globally and can significantly influence the coral microbiome as a result of climatic and anthropogenic stressors. Coral disease surveys were conducted at three patch reefs in the northern Red Sea of Hurghada coast. Surveys occurred during a heatwave event in August 2021, when the average surface water temperature reached about 29 °C. This study compared the differences in bacterial communities associated with healthy and diseased coral tissues of nine reef-building coral genera collected from the surveyed reefs. The five most common coral diseases identified were black band disease (BBD), white syndrome (WS), growth anomalies (GA), pink line syndrome (PLS), and skeletal eroding band (SEB). In general, the microbial abundance in infected parts of corals was higher than that in healthy parts. Bacteria associated with the black band disease on *Platygyra lamellina* showed that the number of total bacteria found in the tissue were higher than that in the mucus of the corals. Furthermore, the cultural *Vibrio* populations (TCBS) of tissue samples were higher than that in mucus samples, and the *Vibrio* population of the infected coral tissue and mucus was greater than the healthy tissue and mucus. Bacterial 16S rRNA gene clone libraries derived from 12 cultivable isolates of coral mucus and tissue revealed a distinct partitioning of bacterial genera into healthy and diseased samples. Species identified from the healthy samples were dominated by *Paracoccus yeei*, *Staphylococcus aureus*, and *Acinetobacter* sp., while bacteria associated with BBD-affected coral samples were *Acinetobacter* sp., *Desulfovibrio* sp., *Bacillus farraginis*, *Vibrio hepatarius*, *Vibrio brasiliensis*, *Arcobacter* sp., and *Micromonospora* sp. The present study provides a baseline assessment of coral disease incidence and associated microbial communities in a northern Red Sea region, which is expected to rise as a consequence of increased frequency and severity of climatic and non-climatic stressors.

INTRODUCTION

Diseases affecting corals are a major threat to coral reefs, and outbreaks of these diseases can happen anywhere in the world. Since 2014, stony coral tissue loss disease has devastated coral reefs across the Florida Reef Tract and beyond, spreading to neighboring Caribbean locations (Alvarez-Filip *et al.*, 2019). The occurrence of coral disease outbreaks has grown over the years and has been connected to anthropogenic

consequences such as overfishing, pollution from plastics and other man-made materials, dredging activities, runoff from land, and rising water temperatures (**Lamb *et al.*, 2018**). Threats to coral reefs can vary widely depending on where in the world they are located. Corals thrive in the Red Sea because it is a body of water that is partially contained, has limited interchange with the Indian Ocean, has minimal influxes of freshwater (less than 30 millimeters per year), and has significant evaporation rates (**Aeby *et al.*, 2021**). Most coral reefs across the world experience temperatures no higher than 29 °C and salinities of about 36 parts per thousand (ppt) (**Kleypas *et al.*, 1999**). Temperatures around 32°C in the summer and approximately 18°C in the winter, with salinities of 40 ppt or more, are usual in the Red Sea (**Voolstra *et al.*, 2021**). Over three hundred and forty different varieties of reef coral can be found in the Red Sea's thriving coral reefs. Natural temperature, salinity, and nutrient gradients run from north to south in the Red sea (**Berumen *et al.*, 2019**). Sea surface temperatures (SSTs) range from an average of 26.3 ±1.1 °C in the far north to an average of 31.3 ± 1.1 °C in the south. There has been widespread bleaching of Red Sea corals, with a corresponding latitudinal gradient in coral response. While **Osman *et al.*, (2018)** noted that degree heating weeks exceeding the bleaching threshold of four occurred throughout the Red Sea during the last bleaching event in 2015, where they occurred only in the center and southern regions of the Red Sea. While the Red Sea's coral reefs are subject to temperature and salinity extremes, they also get very little sedimentation, turbidity, or nutrient enrichment from terrestrial runoff or rivers. Damage to coral reefs and an uptick in the incidence and severity of coral diseases are both caused by land runoff (**Shore-Maggio *et al.*, 2018**). The use of microorganisms as a health indicator for coral reef ecosystems has much promise. A coral reef's microorganisms play a crucial role in the reef's biogeochemical cycle, as they live in close proximity to the reef's benthos and make substantial contributions to host health and ecological homeostasis (**Glasl and Bourne, 2017; Glasl and Webster, 2018**). Changes in community composition can be an early indicator of environmental change, as microbial communities are constantly being reshaped to better exploit available resources (**Garza *et al.*, 2018**). Coral-associated microbial communities, for instance, have been shown to undergo compositional and functional modifications along gradients of anthropogenic impact (**Ziegler *et al.*, 2016**) and with changes in water quality (**Angly *et al.*, 2016**). Nonetheless, the diagnostic potential of microorganisms for coral reef monitoring remains primarily conceptual, with only a few studies expanding on their potential value (**Glasl *et al.*, 2017**). The microbial score, for instance, evaluates the effects of humans on coral reefs by comparing the metabolic rates of microorganisms and fish (**McDole *et al.*, 2012**). Despite the great benefit provided by microbes to coral reefs, some of them may pose a great risk of acute disease infection. It can be tricky to pinpoint the source of some dangerous germs, and the ocean's environmental factors that amplify the spread of disease are also poorly understood. Diseases in corals show few phenotypic differences, which may indicate distinct diseases. The bacterial make-up of various coral

species varies, as does the bacterial make-up of the water column as a whole. However, further research is needed to determine its cause and the effects of pathogenic bacteria on coral (Sassi *et al.*, 2015). This study aims to investigate three areas of coral reefs along the northern Red Sea of Hurghada coast. The research involves identifying different types of coral reefs and studying the bacterial diseases affecting these reefs. The goal is to isolate and genetically characterize these bacterial diseases and assess their impact on the various coral reef communities.

MATERIALS AND METHODS

Coral disease sampling and bacteriological study

A total of 23 samples were collected from healthy and diseased portions of coral colonies during the summer (August) of 2021 from a depth of about 1-6 m at three reef patches, namely El-Shamaly, El-Shabrouh, and El-Hellaly, of Hurghada coast, Red Sea Egypt, (Fig. 1). Pieces of both healthy and diseased coral were taken from the same reefs but from different colonies of the same genus. Fragments of coral were transported in seawater collected from the same spot and placed in sterile bottles.

At the surveyed site, microbial mats from coral genera with infection parts were collected. Using a sterile 50 ml syringe, coral mucus was extracted from various sick coral colonies. The coral samples collected included nine stony corals (*Platygyra*, *Echinopora*, *Dipsastraea*, *Goniastrea*, *Porites*, *Galaxea*, *Montipora*, *Acropora*, *Stylophora*, *Platygyra*) from the different locations. Mucus samples were collected from seemingly healthy and infected corals from the upper portion of the coral colony or polyp (Fig.2). Sterilized flasks were used to hold the mucus samples. As soon as possible, these samples were put in an ice chest and sent to the microbiology lab at the National Institute of Oceanography and Fisheries (NIOF), Egypt.



Fig. 1: A map of the study area showing the sampled reef sites for coral disease monitoring; (1) El-Shamaly reef, (2) El-Shabrouh reef, and (3) El-Hellaly reef.

Using autoclaved artificial seawater for the serial dilutions, 100 μ l of the diluted solution was then dispersed on marine agar (HiMedia Laboratories, Mumbai, India) plates its component (mineral salts, peptone and yeast extract). Colony-forming units (CFU/ ml) were used to calculate the bacterial count after one week of incubation at room temperature

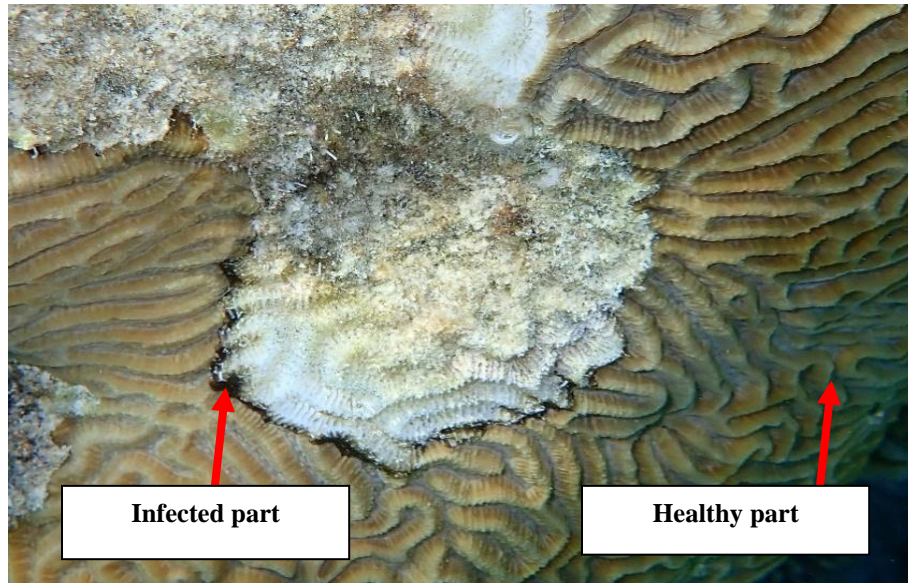


Fig. 2: Surface sampling of *Platygyra* coral infected with black band disease from both healthy and diseased areas.

Bacteria associated with the black band disease (BBD) on *Platygyra lamellina*

The Al-Shamaly reef station provided the location for the collection of samples taken from corals that had been impacted by the BBD. The three *Platygyra* sp. corals that were used for the sample were numbered and marked so that subsequent samples could be taken from them. In order to follow up the disease after a month another samples were taken in September, 2022 at a time when the disease was in its most active state and the black band was morphologically apparent (**Fig. 3**). Syringes capable of holding 50 milliliters were used to collect mucus or tissue samples which were then brought straight to the laboratory. Thiosulfate citrate bile sucrose agar (TCBS) (Biolife, Monza, MI, USA) media plates its component (yeast extract, peptone, sodium thiosulfate, sodium citrate, ox bile, sucrose, sodium chloride, iron (III) citrate, bromothymol blue, thymol blue, agar) (**Zampieri et al., 2021**) (Merck) supplemented with a pH indicator for *Vibrio* selection, and Marine Broth and Marine Agar 2216 (HiMedia Laboratories, Mumbai, India) plates (at the indicated final concentration of 10%) were employed to isolate microorganisms (DSMZ GmbH). After a week of incubation at 30 °C, all of the plates were read. The majority of high-dilution-appearing colonies were restreamed and then used for DNA isolation.



Fig. 3: Black Band Disease (BBD) on *Platygyra* sp.

DNA extraction and Sequence Analysis

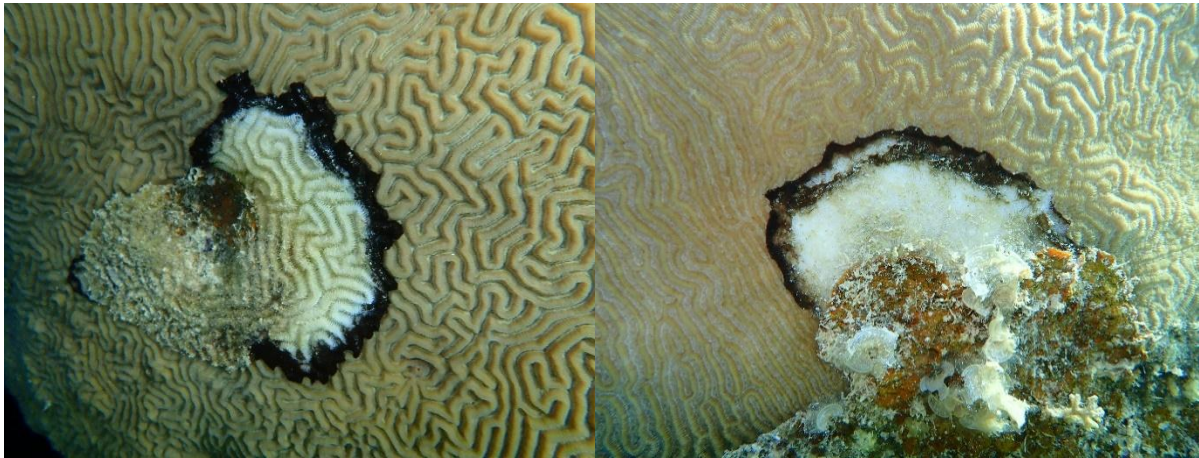
The Nucleospin Kit (NucleoSpin Co., Germany) was used to extract bacterial genomic DNA from BBD ambient samples. Drying chilled samples at room temperature was followed by adding them to the lysis buffer and crushing them with a brand-new, single-use plastic rod. After that, samples were put in lysing matrix tubes to extract DNA. The DNA was eluted with 50 ml sterile elution buffer, verified by electrophoresis in agarose gels (1.5% wt./vol) stained with Gel Red TM (USA) and finally stored at -20°C . 16S rRNA genes were amplified from DNA extracts by PCR using universal primers 314F (CCTACGGGNGGCWGCAG) and 805R (GACTACHVGGGTATCTAATCC) (Albertsen *et al.*, 2015).

Master Mix (Promega, Madison, WI), 0.5 mM primers, and 10 ng template DNA were used in 25 ml PCR experiments. Thermocycler PCR System 2700 incubated reaction mixtures. PCR amplification settings included a 4-minute denaturing stage at 95.5°C , 30 cycles at 94°C for 30 sec, 55°C for 60 sec, and 70°C for 90 sec, and a 15-minute extension step at 72°C . Electrophoresis and spectrophotometry tested DNA-templated PCR products for quality, size, and quantity. GENEIOUS TM Pro (V.5.6.3) sequencing program modified and aligned isolates pair sequences. BLAST at the National Centre for Biotechnology Information (www.ncbi.nlm.nih.gov) was used to compare all high-quality consensus sequences (HQ.65%) to known 16Sr DNA sequences. Sequences that matched at a similarity level of 1 (97–100%) were thought to belong to the same species. Sequences that matched at a similarity level of 2 (93–96%) were thought to belong to the same genus. Sequences that matched at a similarity level of 3 (93%) were thought to be below the similarity of the genus level (Rossi-Tamisier *et al.*, 2015). The rRNA sequences of putative bacteria that were closely linked to each other were aligned using Geneious alignment and then rearranged by hand. The Neighbor-Joining method of GENEIOUS TM Pro (V.5.6.3) was used to make an evolutionary tree.

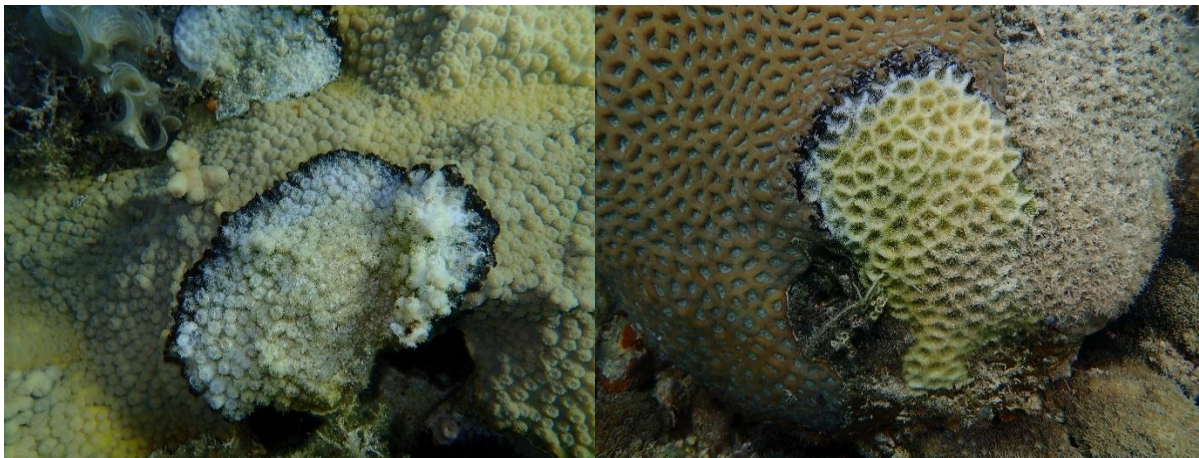
RESULTS

Sample characterization

From the most dominant coral taxa in the Red Sea in Egypt, samples of both healthy and ill corals were obtained. All of the sampling locations have coral reefs with the same topology. These locations are home to a wide variety of coral species that can take on many different lifeforms, including branching, massive, submissive, and encrusting varieties. The afflicted corals were damaged by a variety of illnesses that are frequently observed in the Red Sea. Over all study sites, nine coral genera were infected and include *Platygyra*, *Echinopora*, *Dipsastraea*, *Goniastrea*, *Porites*, *Galaxea*, *Montipora*, *Acropora*, *Stylophora* and *Platygyra* appeared to be the most susceptible to black band disease at all reef sites. A total of nine different coral diseases were recorded. The next common coral disease was white syndrome. The physical appearances of the recorded coral diseases are shown in **Figs. 4A & B.**

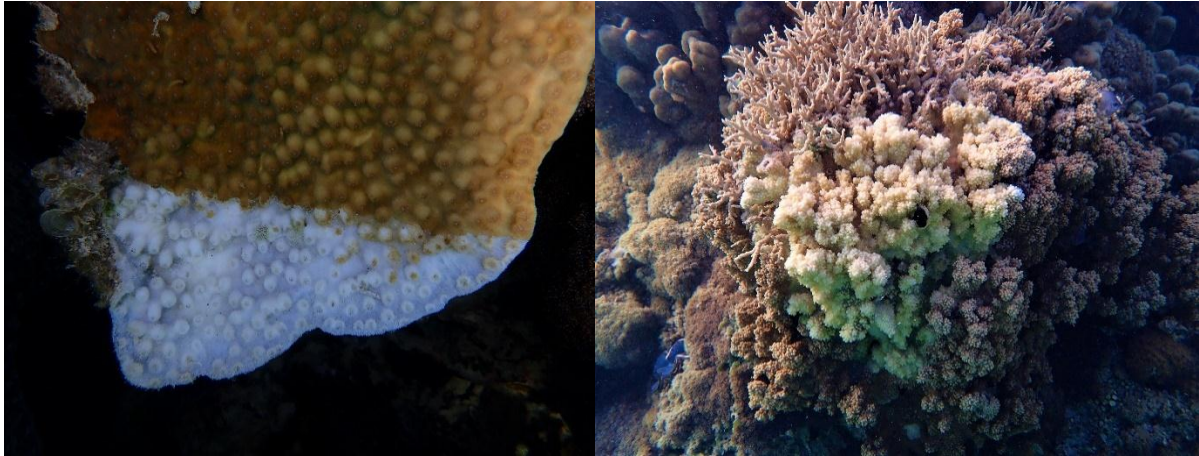


Black Band Disease (BBD) in *Platygyra* sp.



Black Band Disease (BBD) in *Echinopora* sp.

Black Band Disease (BBD) in *Favites* sp.



White Syndrome (WS) in *Echinopora* sp.

White Syndrome (WS) in *Montipora* sp.

Fig. 4A: Most susceptible coral genera and common coral diseases in the study area.



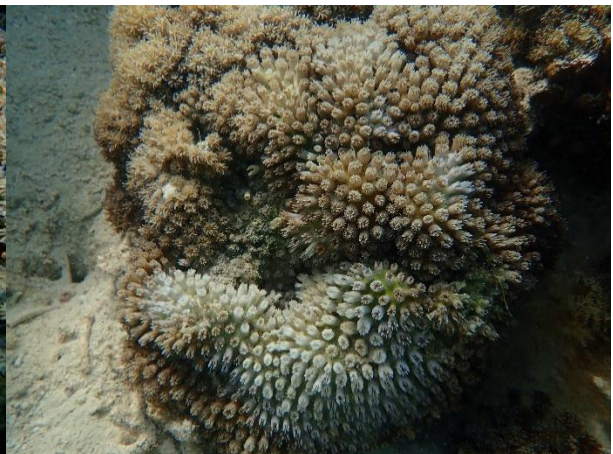
White Syndrome (WS) in *Acropora* sp.



White Syndrome (WS) in *Porites* sp.



White Syndrome (WS) in *Dipsastraea* sp.



White Syndrome (WS) in *Galaxea* sp.



Pink-line Syndrome (PLS) in *Porites* sp.

Skeletal Eroding Band (SEB) in *Echinopora* sp.

Fig. 4B: Most susceptible coral genera and common coral diseases in the study area

Microbial abundance

The abundance of coral-associated bacterial communities in each diseased coral species sampled was quantified using the total bacterial count. Across the studied sites, the microbial abundance in infected parts of corals were higher than that in healthy parts. The results of the bacterial count in healthy and diseased areas are detailed in Table 1.

The bacterial count in healthy parts varied from that in infected areas. In the first site (El-Shamaly reef), the bacterial count for the healthy part associated with the disease of Black band disease was 3×10^4 , 2×10^4 , 3×10^4 and 4×10^4 for *Platygyra*, *Echinopora*, *Dipsastraea*, and *Galaxea* species, respectively. In the healthy parts of White syndrome disease, counts of 1×10^2 , 2×10^2 , 3×10^2 , 2×10^2 and 7×10^3 were recorded for *Echinopora*, *Stylophora*, *Acropora*, *Montipora*, and *Galaxea* species, respectively. The healthy area of Growth anomaly disease recorded counts of 8×10^2 and 3×10^2 for *Platygyra* and *Acropora* species, respectively. *Echinopora* recorded a count of 3×10^2 in the skeletal eroding band disease healthy area. In the second site (El-Shabroun reef), counts of 2×10^4 and 1×10^3 were recorded for *Platygyra* and *Echinopora* species, respectively in the healthy parts of corals affected by Black band disease. Moreover, counts of 2×10^2 and 14×10^3 were recorded for *Acropora* and *Porites* species respectively in the healthy area of corals affected by White syndrome disease, and 5×10^2 for *Platygyra* species in the healthy parts of Growth anomaly disease area. In the third site (El-Hellaly reef), counts of 2.6×10^2 , 2.5×10^2 , and 1.9×10^2 bacterial cells were recorded for *Platygyra*, *Echinopora*, and *Dipsastraea* species, respectively, in the area affected by Black band disease, and 1×10^7 for *Platygyra* species in the healthy parts of Growth anomaly disease area. Regarding the bacterial count in infected coral areas, it was evident that the bacterial count for black band disease in the first site was 12×10^8 , 6×10^9 , 16×10^4 , and 11×10^2 for *Platygyra*, *Echinopora*, *Dipsastraea*, and *Galaxea*, respectively. For corals infected by white syndrome disease, the bacterial counts were 3×10^4 , 6×10^5 , 7×10^5 , 5×10^4 , and 22×10^5 in

Echinopora, *Stylophora*, *Acropora*, *Montipora*, and *Galaxea*, respectively. In the case of growth anomaly disease corals, the bacterial count was 16×10^4 and 6×10^4 for *Platygyra* and *Acropora* species, respectively. The skeletal eroding band disease reefs recorded 8×10^4 bacterial cells for *Echinopora* species. In the second site, there were recorded counts of 10×10^5 and 7×10^4 bacterial cells for *Platygyra* and *Echinopora* species, respectively, in the infected corals by black band disease. White syndrome disease corals recorded 4×10^6 and 4×10^8 bacterial cells for *Acropora* and *Porites* species. The bacterial count for *Platygyra* species in growth anomaly disease corals was 7×10^7 . Lastly, at the third site, the bacterial counts were 1×10^2 for *Platygyra*, *Echinopora*, and *Dipsastraea* in black band disease corals and also 3×10^9 for *Platygyra* in growth anomaly disease corals.

Table 1: The total bacterial count attached to the different types of coral reefs in the study area, where the samples were collected in the healthy and infected areas of each type.

Stony corals	Total Bacterial Count (CFU/ ml)							
	BBD		WS		GA		SEB	
	Healthy	Infected	Healthy	Infected	Healthy	Infected	Healthy	Infected
El-Shamaly reef								
<i>Platygyra</i>	3×10^4	12×10^8	-	-	8×10^2	16×10^4	-	-
<i>Echinopora</i>	2×10^2	6×10^9	1×10^2	3×10^4	-	-	3×10^2	8×10^4
<i>Dipsastraea</i>	3×10^5	16×10^4	-	-	-	-	-	-
<i>Stylophora</i>	-	-	2×10^2	6×10^5	-	-	-	-
<i>Acropora</i>	-	-	3×10^2	7×10^5	3×10^2	6×10^4	-	-
<i>Montipora</i>	-	-	2×10^2	5×10^4	-	-	-	-
<i>Galaxea</i>	4×10^5	11×10^2	7×10^3	22×10^5	-	-	-	-
El-Shabroun reef								
<i>Platygyra</i>	2×10^4	10×10^5	-	-	5×10^2	7×10^7	-	-
<i>Echinopora</i>	1×10^3	7×10^4	-	-	-	-	-	-
<i>Acropora</i>	-	-	2×10^2	4×10^6	-	-	-	-
<i>Porites</i>	-	-	14×10^3	4×10^8	-	-	-	-
El-Hellaly reef								
<i>Platygyra</i>	2.6×10^2	1×10^2	-	-	1×10^7	3×10^9	-	-
<i>Echinopora</i>	2.5×10^2	1×10^2	-	-	-	-	-	-
<i>Dipsastraea</i>	1.9×10^2	1×10^2	-	-	-	-	-	-

BBD : Black band disease

WS: White syndrome disease

GA: Growth anomaly disease

SEB: skeletal eroding band disease

Determine of bacteria associated with BBD-diseased coral colonies of *Platygyra*

Table 2 compares the number of the total bacterial count in coral samples with the count of specific bacterial population grown on TCBS medium. The data showed that the number of total bacteria found in the tissue were higher than that in the mucus of the corals. Also the healthy coral mucus contained about two times of total bacteria more

than in the infected coral mucus, while the opposite was observed in the tissue samples where the infected tissue have higher total count than healthy tissue. The cultural *Vibrio* populations (TCBS) of tissue samples were higher than that in mucus samples, also *Vibrio* population of the infected coral tissue were greater than the healthy tissue. The cultural *Vibrio* populations (TCBS) of tissue samples ($4.6 \pm 2.5 \times 10^3$) were higher than that in mucus samples ($2.5 \pm 1.3 \times 10^3$), Furthermore *Vibrio* population of the infected coral tissue and mucus ($4.6 \pm 2.5 \times 10^3$ and $2.5 \pm 1.3 \times 10^3$, respectively) were greater than the healthy tissue an mucus ($2.6 \pm 2.1 \times 10^3$ and $1.8 \pm 3.3 \times 10^3$, respectively). In every instance, the overall number of bacteria associated with the tissue was found to be significantly higher than the number of bacteria found in the mucus.

Table 2: The bacterial counts in infected and healthy parts from *Platygyra*

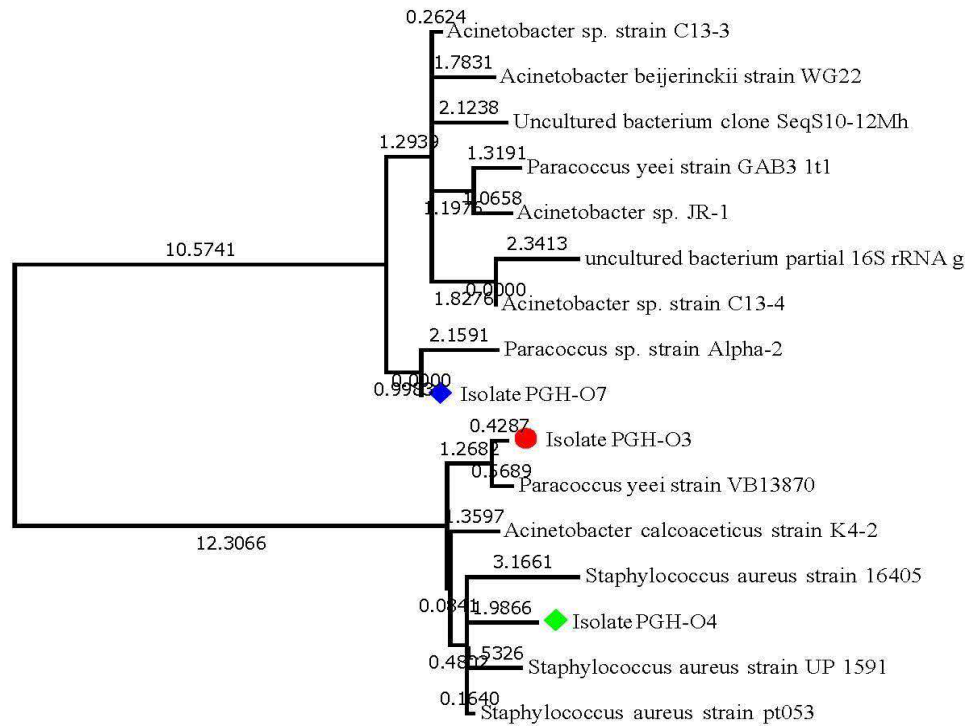
Sample	Type	Viable counts (CFU/ ml)	
		Marine Agar	TCBS Agar
Infected part	Mucus	$1.7 \pm 1.5 \times 10^4$	$2.5 \pm 1.3 \times 10^3$
	Tissue	$5.9 \pm 2.9 \times 10^5$	$4.6 \pm 2.5 \times 10^3$
Healthy part	Mucus	$0.4 \pm 1.6 \times 10^5$	$1.8 \pm 3.3 \times 10^3$
	Tissue	$1.6 \pm 4.2 \times 10^5$	$2.6 \pm 2.1 \times 10^3$

Identification of bacterial communities associated with BBD-diseased coral colonies of *Platygyra*

Twelve morphologically unique isolates were identified after the initial screening of 421 isolates taken from coral samples. Three of these isolates were taken from healthy areas of the coral, and the remaining nine were taken from infected parts. A phylogenetic analysis was performed on each of the 12 clones after they were all sequenced. Table 3 and Figs. **5A and 5B** demonstrate the wide variety of bacterial phylotypes that can be found associated with *Platygyra lamellina* Black Band Disease (BBD). Bacterial species identified from healthy coral samples include *Paracoccus yeei*, *Staphylococcus aureus* and *Acinetobacter* sp. while bacterial associated with BBD-diseased coral samples were *Acinetobacter* sp., *Desulfovibrio* sp., *Bacillus farraginis*, *Vibrio hepatarius*, *Vibrio brasiliensis*, *Arcobacter* sp. and *Micromonospora* sp.

Table 3: List of strains isolated from samples of *P. lamellina* Black Band Disease (BBD) of healthy and diseased samples

Samples	Isolate code	No. of base pairs sequenced	Most closely related hit in GenBank by BLAST analysis	Sequence Similarity (%)	GenBank Accession no. Most closely related
Healthy area	PGH-O3	420	<i>Paracoccus yeei</i>	99	MT254796
	PGH-O4	1201	<i>Staphylococcus aureus</i>	99-100	AP023034
	PGH-O7	683	<i>Acinetobacter</i> sp.	99-100	MT255191
Infected area	PGI-O1	591	<i>Desulfovibrio</i> sp.	100	AY907535
	PGI-O6	544	<i>Bacillus farraginis</i>	92	AB734966
	PGI-O7	516	<i>Vibrio hepatarius</i>	100	OP198433
	PGI-O8	939	<i>Vibrio brasiliensis</i>	99-100	MT368025
	PGI-O12	660	<i>Arcobacter</i> sp.	99-100	LC133150
	PGI-O15	774	<i>Micromonospora</i> sp.	100	EF466033
	PGI-O16	701	<i>Nocardiopsis</i> sp.	99-100	MH779065
	PGI-O19	998	<i>Staphylococcus</i> sp.	99.5	AY189747
	PGI-O23	978	<i>Aquimarina</i> sp.	96	CP03196

**Fig. 5A:** Phylogenetic tree based on 16S ribosomal DNA sequence data comparing strains from a healthy area to the most closely related bacteria in GenBank.

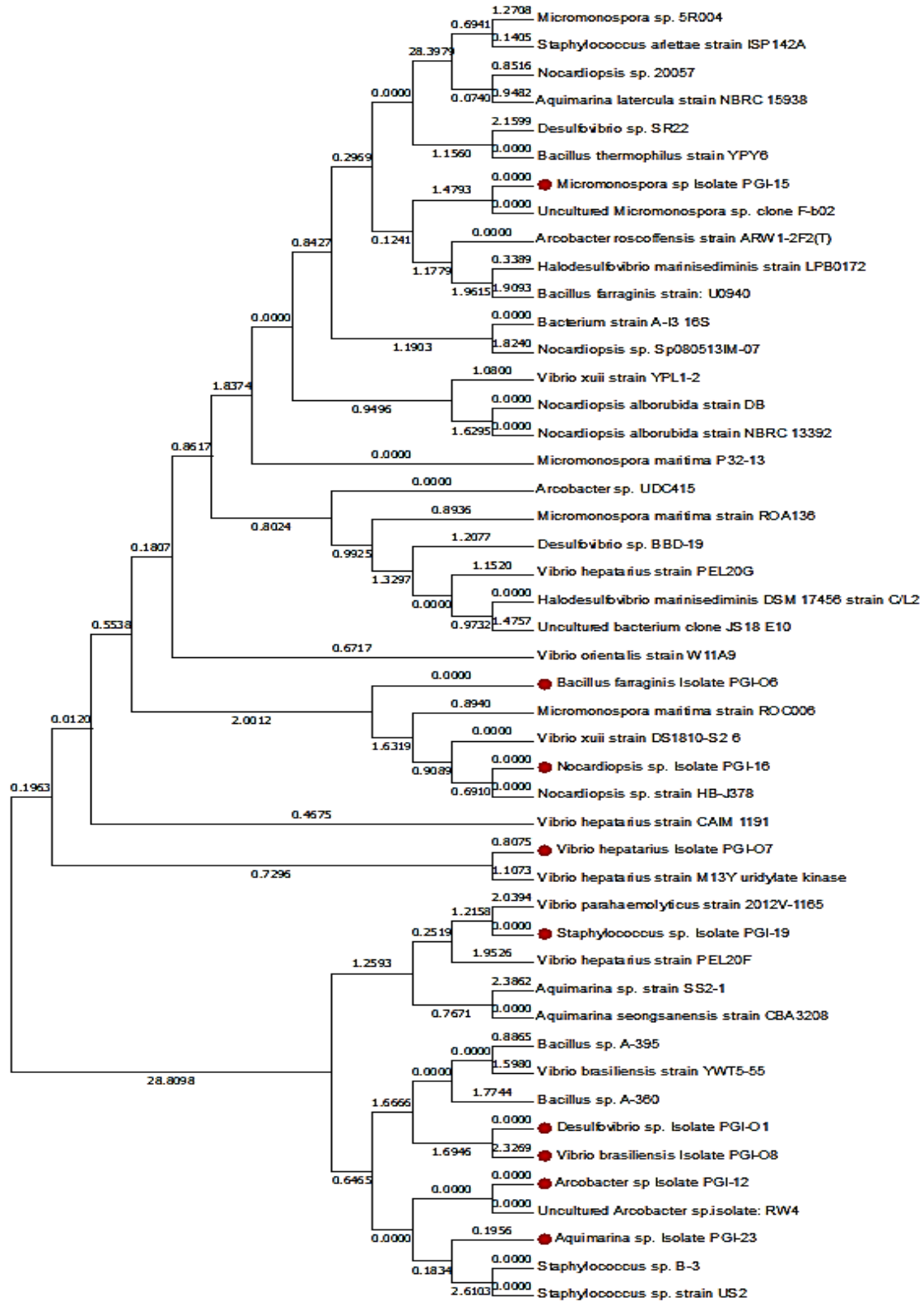


Fig. 5B: Phylogenetic tree based on 16S ribosomal DNA sequence data comparing strains from an affected area to the most closely related bacteria in GenBank.

DISCUSSION

Coral reefs are intricate ecosystems that play a crucial role in maintaining the well-being of the oceans. But in recent years, bacterial diseases have posed a serious danger to Red Sea coral reefs. The bacteria responsible for these infections are causing the coral reefs to become diseased, leading to a decline in their health and a reduction in their ability to provide essential ecological services (**Guzman *et al.*, 2020**). The rise in seawater temperature, excessive solar irradiation, increased seawater depth, outbreaks of red tide, and the presence of anthropogenic waste all contribute to the emergence of diseases in coral reefs (**Moriarty *et al.*, 2020**).

The results of our field surveys have provided some baseline information on the prevalence of coral diseases that are having an impact on reefs in Egypt's northern region of the Red Sea. The results of this study make it abundantly clear that the prevalence of diseases differed significantly between reefs. It's possible for both biotic and abiotic variables to play a role in disease development. Pathogenic microorganisms, such as viruses, bacteria, and fungi, are the root of biotic disorders. Environmental stressors, such as changes in ambient conditions or exposure to contaminants, can be either naturally occurring or produced by humans, both of which contribute to the emergence of abiotic disorders (**Vos *et al.*, 2015; Farag *et al.*, 2016**).

Black band disease (BBD) is appeared in *Platygyra* sp., *Echinopora* sp., *Dipsastraea* sp. and *Goniastrea* sp., BBD is firstly appeared in different reefs by 1970 according to Peters, (2015) as black mat a few millimeters to centimeters wide (**Peters, 2015a**). Illustrations of various BBD species illustrate how this condition manifests itself. A group of microorganisms, including filamentous cyanobacteria, sulfate-reducing bacteria, sulfide-oxidizing bacteria, fungi, and protozoans, is responsible for causing bleaching. Deep in the band adjacent to the tissue, bacteria produce anoxia, hydrogen sulfide, and microcystins, killing the coral tissue and allowing the microorganisms to feed off the organic compounds released by the dead coral cells. Most corals had lesions and fragments of microbial mats were floating around in the water, making for an ideal environment for the spread of this illness. The vulnerability of corals to infection with the microbial mats may be further exacerbated by other stresses such as nutrients, light levels, increased water temperatures, and bleaching. (**Barneah *et al.*, 2007; Richardson *et al.*, 2007; Stanić *et al.*, 2011**). This is clearly achieved by this study which in first site, the BBD appeared in *Platygyra* sp., *Echinopora* sp., *Dipsastraea* sp., *Goniastrea* sp., second site appeared in *Platygyra* sp., *Goniastrea* sp., and BBD in aquatic region of Hurghada reef station appeared in *Platygyra* sp. and *Goniastrea* sp.

Growth anomalies (GA) disease is a case of cellular proliferative disorders discovered via reefs worldwide with polyps and skeletal developments. Current study recorded GA in This disease appeared only in *Platygyra* sp. over three sites. GA is caused by biotic factors as parasitic crustaceans, bacteria (**Domart-Coulon *et al.*, 2006**), fungi (*Phanerochaete carnosae*) (**Osman *et al.*, 2011**) and parasitic algae (**Work *et al.*, 2008**) with correlation by human population size. The lesions of GA are transmitting by directly contact in two small colonies of *Platygyra* sp. and one arising without contact by affected corals (**Kaczmarzky and Richardson, 2007**).

White syndrome disease (WS) is a white line appeared in skeleton firstly as consequence of microorganism's assemblage and pigmented tissue leading to white band of necrosis advances over tissue. The WS disease signs are predator damaging and acute tissue loss caused by human activities (**Peters, 2015b**). The appearing of WS in studied sites are noted for *Platygyra* sp., *Stylophora* sp., *Acropora* sp., *Montipora* sp., *Echinopora* sp. and *Galaxea* sp. The WS disease appear due to enumeration of gram-negative bacteria in calicoblastic region of reefs as *Pseudomonas* spp. and *Vibrio* spp. which contaminate *Acropora* sp. and others (**Gil-Agudelo *et al.*, 2006 ; Polson, 2007**). **Miller *et al.*, (2014)** mentioned in their studies on WS disease that, there are bacteria called rickettsia-like microorganism that were isolated from mucocytes of coral reefs, this infection is chronic which reefs cannot resist it leading to a rapid loss of live reef. This shows the extent of disease dispersion during the study, which in first site are contaminate *Stylophora* sp., *Acropora* sp., *Montipora* sp., *Echinopora* sp. & *Galaxea* sp., and in second site the contamination represented in *Platygyra* sp.

Skeletal eroding band (SEB) disease is demonstrated only in site one in *Platygyra* sp. and *Echinopora* sp. Low SEB prevalence in the other sites may be a reflection of decrease contamination factors. Also, current status of other sites can protected reefs from serious bleaching. **Willis and Van Andel, (2004)** mentioned in his study that, skeletal eroding was the most prevalent disease in the tested sectors during the year 2002-2003 by 5.4% of all corals. Likewise, **Winkler *et al.*, (2004)** mentioned that, 38% of the tested corals in the Red Sea were infected with the same disease. These studies were not persistent with current study which the disease was the least prevalent disease.

Considering the bacterial species that were isolated from the study area. *Vibrio hepatarius* and *Vibrio brasiliensis* are gram negative species of bacteria that have been identified as contaminants in coral reefs in the Red Sea. They isolated form oysters, seawater and marine organisms with disease causing in fish, bivalves and shellfish. These bacteria are known to cause disease in various marine organisms, including corals which causing tissue necrosis, bleaching, and overall decline in coral health. The contamination of coral reefs by *V. hepatarius* and *V. brasiliensis* is a significant concern, as these bacteria can have a detrimental impact on the health of the reef ecosystem (**Pootakham**

et al., 2017). Factors such as rising sea temperatures, pollution, and overfishing can contribute to the proliferation of these bacteria, making it important to address these issues in order to protect the health of coral reefs (Pootakham *et al.*, 2017).

In coral reefs, *Acinetobacter* sp. has been associated with coral diseases such as white syndrome and brown band disease which they documented in current study. These diseases cause tissue necrosis and mortality in coral colonies, leading to significant declines in coral cover and biodiversity (Lins *et al.*, 2010). *Desulfovibrio* sp. and *Bacillus farraginis* are genus of sulfate-reducing and gram-positive bacteria that are commonly found in marine sediments and can cause infections to marine fauna. In coral reefs, they have been linked to the development of black band disease, coral bleaching, white pox disease, white patches of exposed skeleton on coral colonies and a band of black microbial mat that migrates across the coral surface, leading to tissue necrosis and mortality (Hadaidi *et al.*, 2018). The presence of *Acinetobacter* sp., *Desulfovibrio* sp., and *Bacillus farraginis* in coral reefs in the Red Sea can have a significant impact on the health of these ecosystems. These bacteria can cause diseases that lead to tissue necrosis, mortality, and declines in coral cover and biodiversity. The factors that contribute to the proliferation of these bacteria in coral reef environments are not fully understood, but may include human activities such as pollution and overfishing, as well as natural factors such as changes in water temperature and acidity. To address the threat of bacterial diseases to coral reefs, preventative measures such as reducing pollution and regulating fishing practices are important. Additionally, the development of treatments and management strategies for coral diseases is crucial for protecting the health and resilience of these important ecosystems in the Red Sea and beyond (John *et al.*, 2022).

CONCLUSION

This research on the bacterial communities associated with healthy and diseased corals during a heatwave event in the Northern Red Sea, Egypt, provides valuable insights into the intricate relationship between corals and their microbial partners. The study contributes to our understanding of the impact of heat stress on coral health and highlights the importance of preserving diverse and stable bacterial communities for coral reef resilience. Ultimately, this knowledge can guide conservation efforts and policy decisions aimed at safeguarding coral reefs in the face of ongoing climate change challenges

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