

Ecological and molecular characterization of a coral black band disease outbreak in the Red Sea during a bleaching event

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ABSTRACT

Black Band Disease (BBD) is a widely distributed and destructive coral disease that has been studied on a global scale, but baseline data on coral diseases is missing from many areas of the Arabian Seas. Here we report on the broad distribution and prevalence of BBD in the Red Sea in addition to documenting a bleaching-associated outbreak of BBD with subsequent microbial community characterization of BBD microbial mats at this reef site in the southern central Red Sea. Coral colonies with BBD were found at roughly a third of our 22 survey sites with an overall prevalence of 0.04%. Nine coral genera were infected including *Astreopora*, *Coelastrea*, *Dipsastraea*, *Gardineroseris*, *Goniopora*, *Montipora*, *Pavona*, *Platygyra*, and *Psammocora*. For a southern central Red Sea outbreak site, overall prevalence was 40 times higher than baseline (1.7%). Differential susceptibility to BBD was apparent among coral genera with *Dipsastraea* (prevalence 6.1%), having more diseased colonies than was expected based on its abundance within transects. Analysis of the microbial community associated with the BBD mat showed that it is dominated by a consortium of cyanobacteria and heterotrophic bacteria. We detected the three main indicators for BBD (filamentous cyanobacteria, sulfate-reducing bacteria (SRB), and sulfide-oxidizing bacteria (SOB)), with high similarity to BBD-associated microbes found worldwide. More specifically, the microbial consortium of BBD-diseased coral colonies in the Red Sea consisted of *Oscillatoria* sp. (cyanobacteria), *Desulfovibrio* sp. (SRB), and *Arcobacter* sp. (SOB). Given the similarity of associated bacteria worldwide, our data suggest that BBD represents a global coral disease with predictable etiology. Furthermore, we provide a baseline assessment of BBD disease prevalence in the Red Sea, a still understudied region.

Subjects Biogeography, Ecology, Marine Biology, Microbiology

Keywords Coral reef, Coral disease, Red Sea, Coral bleaching, Microbiology, Metabarcoding

INTRODUCTION

The rise of coral disease outbreaks contributes to the decline of coral reefs globally (Cróquer & Weil, 2009; Harvell et al., 2009; Hoegh-Guldberg, 2012; McLeod et al., 2010; Randall &

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Van Woesik, 2015) and coral disease appears to be the most destructive factor on many reefs. For instances, the Caribbean has been named a “disease hot spot” due to the fast emergence, high prevalence, and virulence of coral diseases in this region (*Rosenberg & Loya, 2013*). Coral disease outbreaks in the last decades in the Caribbean have resulted in significant losses in coral cover, diversity, and habitat (*Aronson & Precht, 2001; Bruckner, 2002; Hughes, 1994; Precht et al., 2016; Weil, 2002*). Following the mass-bleaching event in 2005 in the US Virgin islands, coral disease outbreaks reduced coral cover by more than 50% (*Cróquer & Weil, 2009; Miller et al., 2009*).

Coral diseases were first reported in the Caribbean in the 1970s, including black band disease (BBD), which is considered the most studied coral disease due to its widespread occurrence on reefs around the world (*Bourne, Muirhead & Sato, 2011; Richardson, 2004*). Black band disease has been reported from reefs throughout the Caribbean, the Indo-Pacific regions, the Red Sea, and the Great Barrier Reef (*Al-Moghrabi, 2001; Dinsdale, 2002; Green & Bruckner, 2000; Kaczmarek, 2006; Lewis et al., 2017; Montano et al., 2012; Page & Willis, 2006; Sutherland, Porter & Torres, 2004; Weil et al., 2012*). BBD is the first described coral disease (*Antonius, 1973*), affecting scleractinian and gorgonian corals (*Green & Bruckner, 2000; Sutherland, Porter & Torres, 2004; Weil, 2004*). BBD prevalence generally is considered low (*Dinsdale, 2002; Edmunds, 1991; Weil, 2002*); however, this disease is a serious threat to coral reef ecosystems worldwide due to its persistence, leading to coral mortality in the long-term (*Bruckner & Bruckner, 1997; Green & Bruckner, 2000; Kaczmarek, 2006; Kuta & Richardson, 1996; Page & Willis, 2006; Sutherland, Porter & Torres, 2004; Zvuloni et al., 2009*). Susceptibility to BBD differs between coral taxa and may result in long-term changes to coral community structure (*Bruckner & Bruckner, 1997; Page & Willis, 2006*). The abundance of BBD is affected by several environmental factors, including seawater temperature, water depth, solar irradiance, host population diversity, and anthropogenic nutrients (*Al-Moghrabi, 2001; Kaczmarek, 2006; Kuta & Richardson, 2002; Montano et al., 2013*). Interestingly, seasonal temperatures influence BBD prevalence, with increased virulence during warmer summer months (*Richardson & Kuta, 2003; Rützler & Santavy, 1983; Willis, Page & Dinsdale, 2004*), as for example in the Maldives where sea surface temperatures above 28 °C promoted BBD infections (*Montano et al., 2013*).

BBD manifests as a dark band that migrates across the coral colony at a rate of >1 cm/day (*Richardson, 1998*) leaving behind bare skeleton. The base of the BBD mat is anoxic and high in sulfide levels, causing damage and necrosis to coral tissue (*Ainsworth et al., 2007; Carlton & Richardson, 1995; Richardson et al., 1997*). The BBD mat is composed of a polymicrobial consortium, dominated by filamentous cyanobacteria, sulfate-reducing bacteria (SRB), including members of *Desulfovibrio* spp., sulfide-oxidizing bacteria (SOB) (*Beggiatoa* spp.), and other heterotrophic bacteria (*Cooney et al., 2002; Miller & Richardson, 2011; Sato, Willis & Bourne, 2010*). As a result of diel light changes, the microbial members of the BBD mat undergo vertical migrations, which causes the harmful microenvironment on top of the coral tissue (*Carlton & Richardson, 1995; Miller & Richardson, 2011; Richardson, 1996*). Oxygen depletion and high sulfide concentrations are produced by SRB, which is lethal to the coral tissues and considered the most important factor in BBD pathogenicity (*Glas et al., 2012; Richardson, 1996; Richardson et al., 1997; Richardson et al., 2009*). Although

the functional composition of the BBD mat is conserved, the diversity of the microbial consortium in BBD differs according to geographic location and coral species (Cooney *et al.*, 2002; Frias-Lopez *et al.*, 2004; Sekar *et al.*, 2006).

The occurrence of BBD in the Red Sea was first recorded by Antonius (1988) where the severity of BBD was measured from rare to moderate and mostly correlated with elevated temperatures and seawater pollution. However, baseline data on BBD prevalence in the Red Sea is still lacking. To fill this gap, we conducted surveys to determine the distribution and prevalence of BBD across central Red Sea reefs spanning 4 degrees of latitude. We also detected a bleaching-associated outbreak of BBD on a coral reef in the southern central Red Sea and characterized the microbial community of BBD microbial mats from *Coelastrea* sp., *Dipsastraea* sp., *Goniastrea* sp., and *Platygra* sp. using high-throughput sequencing. We compared the microbial consortium to that reported from other regions of the world in order to identify biogeographic patterns in the main BBD consortium members.

MATERIAL AND METHODS

Black band disease surveys

Coral community structure and BBD prevalence was recorded at 22 sites spanning approx. 535 km along the coast of Saudi Arabia in the Red Sea (Fig. 1, Table 1). At least six reefs per region (Yanbu, Thuwal, Al-Lith) were surveyed with three additional reefs in Thuwal and one reef in Jeddah (80 km from Thuwal) that were surveyed as time permitted. The reefs sampled/assessed in this study do not fall under any legislative protection or special designation as a marine/environmental protected area. Under the auspices of KAUST (King Abdullah University of Science and Technology), the Saudi Coastguard Authority issued sailing permits to the sites that include coral collection. At each site, divers counted coral colonies by genera along two replicate belt transects (25 m × 1 m). At the same time, point-intercept method was used to characterize the substrate at 25 cm intervals. All corals with BBD lesions were identified along wider 25 × 6 m transects and photographed. Depending on depth and time limits, the length of transects were adjusted as necessary. Survey sites ranged in depth between 3 and 7.6 m and all surveys were conducted from 19 October to 3 November 2015. The diver surveys were used to determine average percent coral cover, coral community composition, and colony densities. Underwater time constraints prevented counting all colonies within the larger 25 × 6 m belts surveyed for disease. Therefore, BBD prevalence was estimated by calculating the average colony density (by genus) within the 25 × 1 m transect and then extrapolating the colony counts to the wider 25 × 6 m disease survey area and using this as the denominator of prevalence calculations, i.e., (number of colonies with BBD lesions/total number of estimated colonies) * 100 (Aeby *et al.*, 2015a). At the outbreak site, diseased coral colonies were so numerous that only 49 m² of the transect could be surveyed. The frequency of disease occurrence (FOC) was calculated by dividing the number of sites having corals with BBD lesions by the total number of sites surveyed. At the localized BBD outbreak site a chi-square goodness-of-fit test was used to examine differential distribution of the number of BBD versus healthy colonies among the coral genera affected by the disease. The chi-square test

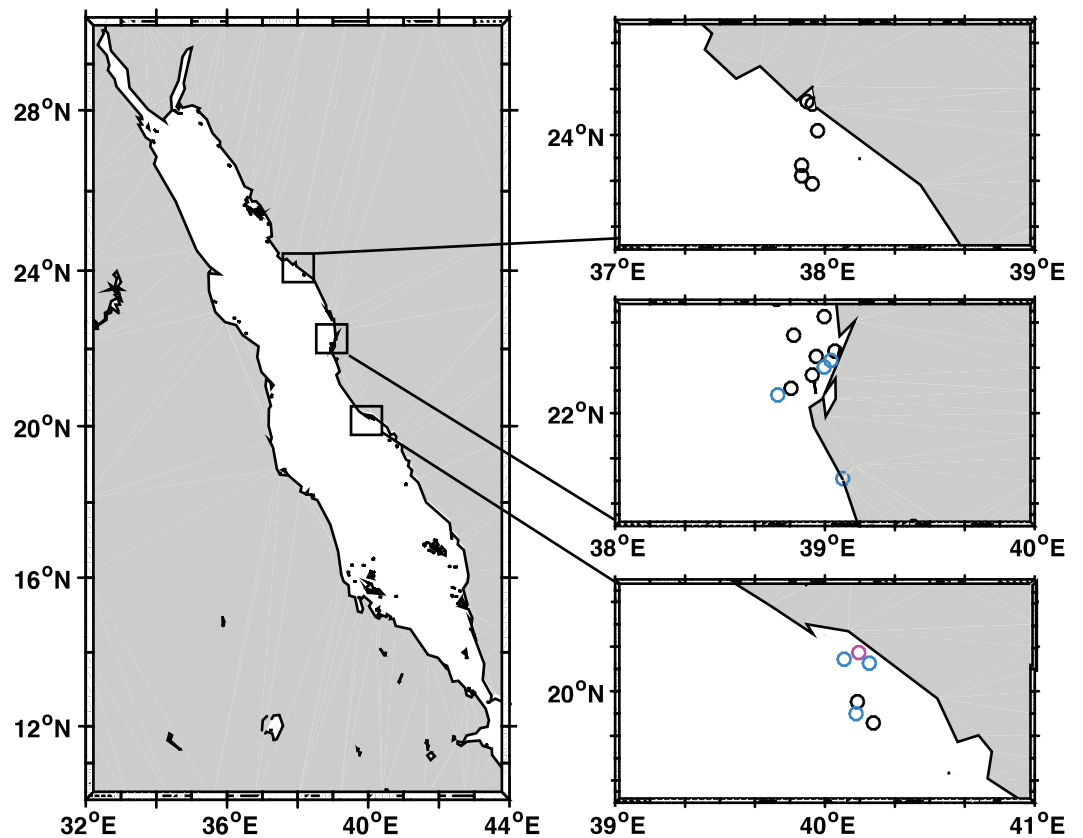


Figure 1 Black band disease survey locations of 22 reef sites along the central Red Sea coast of Saudi Arabia. Survey points marked for Yanbu region (north), Thuwal (central), and Al-Lith (south). Sites without black band diseased coral colonies marked in black, sites with one or two diseased colonies in blue, and the site where a localized outbreak of BBD was observed is marked in pink.

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compares the observed vs. expected number of infected colonies based on the abundance of each coral genus in the field. Statistical analysis was performed using JMP statistical software (v. 10.0.2, SAS Institute Inc., Buckinghamshire, UK).

Sample collection of black band disease microbial mats and 16S rRNA gene sequencing

Microbial mats were collected from BBD infected coral genera (one colony of *Coelastrea* sp., two colonies of *Dipsastraea* sp., three colonies of *Goniastrea* sp., and one colony of *Platygra* sp.) at the site of the observed BBD outbreak (Al-Lith fringing reef 1) in November 2015. Microbial mats were siphoned off the coral surface with Pasteur pipettes and transferred into ziplock bags under water. Sample replication was limited by obtainable coral species on this reef site due to environmental conditions.

Samples were homogenized using bead-beating via TissuLyser II (Qiagen, Hilden, Germany) twice for 30 sec at 30 Hz, 20 μ l of the homogenate were boiled in sterile Milli-Q water at 99 °C for 5 min and subsequently 1 μ l was directly used as PCR template. To amplify the variable region 4 of the 16S rRNA gene, the following primers were

Table 1 Survey of black band disease (BBD)-affected coral colonies at 22 reef sites in the central Red Sea. Coral genus counts denote number of BBD-affected colonies.

Region	Reef site	GPS (latitude, longitude)	Depth (m)	Area colony count survey (m ²)	Area BBD survey (m ²)	<i>Montipora</i>	<i>Dipsastraea</i>	<i>Psammocora</i>	<i>Gardineroseris</i>	<i>Astreopora</i>	<i>Pavona</i>	<i>Platygyra</i>	<i>Coelastrea^a</i>	<i>Goniopora^a</i>	Total no. of BBD	Total no. surveyed	BBD prevalence (%)
Yanbu	Marker 32	23.8664, 37.8913	4	25	75.6										0	726	0
	Marker 35	23.8207, 37.9350	4.6	25	78										0	930	0
	Abu Galaba	23.7891, 37.9393	3	21	61.8										0	848	0
	Fringing reef 1	24.1362, 37.9396	5.2	25	150										0	1,308	0
	Marker 10	24.0189, 37.9666	4.6	25	126										0	1,749	0
	Fringing reef 2	24.1452, 37.9149	4.6	25	150										0	1,830	0
Thuwal	Abu Madafi	22.0766, 38.7751	4	25	300		1								1	2,184	0.05
	Al Fahal	22.1119, 38.8411	4.6	23.5	300										0	6,000	0
	Al-Mashpah	22.0772, 38.7744	6.7	25	300										0	4,200	0
	Inner Fsar	22.2358, 39.0304	4.6	25	300					1					1	6,852	0.01
	Shaab	22.2012, 38.9992	4.6	25	300				1						1	5,778	0.02
	Shi'b Nazar	22.3409, 38.8521	4.9	23.5	300										0	3,294	0
	Tahlah	22.2750, 39.0497	5.2	25	300										0	3,780	0
	Qita al Kirsh	22.4257, 38.9957	4.6	25	300										0	5,748	0
	Um Alkthal	22.1653, 38.9391	7.6	25	300										0	5,208	0
	Jeddah	La Plage	21.7092, 39.0832	4.6	25	300							1		1	474	0.21
Al-Lith	Abu Lath	19.9554, 40.1543	5.5	20	240										0	5,556	0
	South Reef	19.8985, 40.1514	3.7	20	240				1						1	3,720	0.03
	Al-Lith 3	19.8608, 40.2282	5.5	20	240										0	4,320	0
	Qita Al Kirsh	20.1407, 40.0931	3	20	240					1					1	6,588	0.02
	Fringing reef 1	20.1732, 40.1613	4.5	20	49	1	15	2		1	1	1	1	1	23	1,281	1.72
	Whaleshark reef	20.1230, 40.2118	1.8	25	150				1						2	1,716	0.12

Notes.^aColonies of *Coelastrea* and *Goniopora* were only found outside the survey area.

used: 515F [5'TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGTGCCAGCMGCCGCGGTAA'3] and 806RB [5' GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGGGACTACNVGGGTWTCTAAT'3] ([Apprill et al., 2015](#); [Caporaso et al., 2012](#); [Kozich et al., 2013](#)). Primer sequences contained sequencing adaptor overhangs (underlined above; Illumina, San Diego, CA, USA). Triplicate PCRs were performed for all samples with 0.2 μ M of each primer in a total reaction volume of 25 μ L using the Qiagen Multiplex PCR Kit. The following cycling conditions were used: 95 °C for 15 min, followed by 27 cycles of 94 °C for 45 s, 50 °C for 60 s, 72 °C for 90 s, and a final extension step of 72 °C for 10 min. Amplification was checked visually via 1% agarose gel electrophoresis. Triplicate samples were pooled and cleaned with ExoProStar 1-Step (GE Healthcare, Little Chalfont, UK). An indexing PCR was performed on the cleaned samples to add Nextera XT indexing and sequencing adaptors (Illumina, San Diego, CA, USA) following the manufacturer's protocol and followed by sample normalization and library pooling. 16S rRNA gene amplicon libraries were sequenced on the Illumina MiSeq platform using 2*300 bp overlapping paired-end reads with a 10% phiX control at the KAUST Bioscience Core Laboratory. Sequence data determined in this study are available under NCBI Bioproject ID: [PRJNA436216](#).

Sequence data processing and bacterial community analysis

Processing of raw sequence data was conducted in mothur (version 1.36.1; [Schloss et al., 2009](#)). Using the 'make.contigs' command, sequence reads were joined into contigs. Contigs longer than 310 bp and ambiguously called bases were excluded from the analysis. Subsequently, sequences that occurred only once across the entire dataset (singletons) were removed. The number of distinct sequences were identified and counted, and the total number of sequences per sample was determined using the 'count.seqs' command.

The remaining sequences were aligned against SILVA database (release 119; [Pruesse et al., 2007](#)). Sequences were pre-clustered allowing for up to a 2 nt difference between the sequences ([Huse et al., 2010](#)). Chimeras were removed using UCHIME as implemented in mothur ([Edgar et al., 2011](#)). Next, sequences were classified with Greengenes database (release gg_13_8_99; bootstrap = 60; [McDonald et al., 2012](#)), followed by the removal of chloroplast, mitochondria, Archaea, and eukaryote sequences. Further, we found three abundant bacterial families (Dermabacteraceae, Dietziaceae, Brevibacteriaceae) that were present in all disease samples and at high abundance in our negative control. The negative control was a sample containing water as a template for the PCR reaction. As these bacterial families are also known as kit/reagent/lab contaminants, they were excluded from the dataset ([Salter et al., 2014](#)). Some additional bacterial taxa that were found in high numbers in the negative control with low abundance in coral samples were excluded (Comamonadaceae, Halomonadaceae, Staphylococcaceae). For further analyses, sequences were subsampled to 7,328 sequences per sample, which is the lowest number of sequences in a sample, and then clustered into OTUs (Operational Taxonomic Units) at a 97% similarity cutoff. Reference sequences for each OTU were determined by the most abundant sequence ([Data S1](#)). Alpha diversity indices (i.e., Chao1 [Chao, 1984](#), Simpson evenness, and Inverse Simpson Index [Simpson, 1949](#)) were calculated as implemented in mothur.

For detecting similarity of the three main microbial consortium members in BBD microbial mats to previously reported taxa from other studies, the representative sequences of the most abundant OTUs were BLASTed against the NCBI database (<https://blast.ncbi.nlm.nih.gov>) using a 98% similarity cutoff. Subsequently, our sequences were compared to matches of highly similar bacterial taxa by obtaining the respective coral species, their location, and colony health status. Furthermore, low abundant OTUs not previously reported from BBD, but with related properties to SRB, SOB, or Cyanobacteria were also BLASTed against the NCBI database.

16S rRNA sequences of SOB and SRB were aligned and neighbor-joining trees were constructed based on Jukes-Cantor model with MAFFT (Kato, Rozewicki & Yamada, 2017; Kuraku et al., 2013). All positions containing gaps and missing data were excluded and phylogenetic trees were visualized using Archaeopteryx.js.

RESULTS

Distribution and prevalence of black band disease on Red Sea reefs

We identified 30 coral genera within transects across 22 reef sites (Fig. 1) with an average density of 16 coral colonies / m² (SE ± 1.2) and an average coral cover of 43.8% (SE ± 4.3). Colonies with BBD were found at 8 of 22 sites (Table 1, Fig. 1). Over all study sites, nine coral genera were infected and include *Astreopora*, *Coelastrea*, *Dipsastraea*, *Gardineroseris*, *Goniopora*, *Montipora*, *Pavona*, *Platygyra*, *Psammocora*. Approximately 74,090 colonies were examined for disease and overall BBD prevalence over all sites was low (0.04%) because most sites had no signs of BBD. At the sites where BBD occurred, seven of the eight sites had one to two colonies infected within survey areas (up to 300 m²) (avg. prevalence = 0.064%) and one site had a localized BBD outbreak (Al-Lith fringing reef 1) where 21 infected colonies were found within 49 m² of the transect (prevalence = 1.7%) and an additional two colonies outside the transect (Table 1). At this site, 18 coral genera were found within transects, but only nine coral genera exhibited signs of disease suggesting differential BBD susceptibility among coral genera ($\chi^2 = 45.67$ $df = 6$, $P < 0.001$). *Dipsastraea* appeared to be the most susceptible (prevalence = 6.1%) with more diseased colonies than expected based on its abundance within transects (Table 2). *Dipsastraea* represented 18.6% of the coral colonies within transects but 68.2% (15 of 22) of the BBD colonies.

Bacterial community composition of black band disease microbial mats

Besides the ecological survey of BBD prevalence, we investigated the microbial consortium of the BBD mat of corals from the outbreak site in the southern central Red Sea that was also subject to a bleaching event (Al-Lith fringing reef 1). We assessed whether the same bacterial taxa are associated with BBD in the Red Sea in comparison to other sites globally. Seven coral BBD microbial mat samples from the outbreak site included one colony of *Coelastrea* sp., two colonies of *Dipsastraea* sp., three colonies of *Goniastrea* sp., and one colony of *Platygyra* sp., which together yielded 555,093 raw 16S rRNA gene sequences with a mean length of 298 bp (Table 3). After quality filtering and exclusion of chimeras and

Table 2 Survey of black band disease-affected coral genera at an outbreak site in the southern central Red Sea (Al-Lith fringing reef 1, Saudi Arabia).

Coral species	No. of coral colonies/survey area (20 m ²)	% of coral community	No. of BBD cases/survey area (49 m ²)	Prevalence %
<i>Astreopora</i>	23	4.24	1	1.8
<i>Coelastrea</i> ^a	0	–	1	–
<i>Dipsastraea</i>	101	18.63	15	6.1
<i>Goniopora</i> ^a	0	–	1	–
<i>Montipora</i>	11	2.03	1	3.7
<i>Pavona</i>	5	0.92	1	8.2
<i>Platygyra</i>	28	5.17	1	1.5
<i>Psammocora</i>	28	5.17	2	2.9
Other coral genera	346	63.80	0	0
Total^b	542	100	23	1.7

Notes.

^aColonies of *Coelastrea* and *Goniopora* were only found outside the survey area and were not counted towards totals.

Table 3 Summary of sequencing information and alpha diversity measures of bacterial communities associated with black band disease microbial mats from coral colonies at an outbreak site in the southern central Red Sea (Al-Lith fringing reef 1, Saudi Arabia).

Sample	No. of sequences	No. of OTUs ^a	Chao1 ^a	Inv. Simpson ^a	Simpson evenness ^a
<i>Coelastrea</i>	17,869	149	203	14.10	0.095
<i>Dipsastraea 1</i>	7,328	146	240	22.33	0.153
<i>Dipsastraea 2</i>	15,919	122	160	7.71	0.063
<i>Goniastrea 1</i>	16,201	120	152	8.18	0.068
<i>Goniastrea 2</i>	15,743	113	161	9.49	0.084
<i>Goniastrea 3</i>	14,466	136	159	11.57	0.085
<i>Platygyra</i>	20,037	98	125	8.50	0.087

Notes.

^aAfter subsampling to 7,328 sequences. Total number of OTUs: 315.

contaminant sequences, we retained 107,613 sequences for analysis of the BBD microbial mat microbiome. To assess bacterial community composition, sequences were classified to family level considering bacterial families that comprised >1% of the total sequence reads (Fig. 2). The presence of cyanobacteria, SRB, and SOB was confirmed but at varying abundance. For instance, Cyanobacteria such as Phormidiaceae ranged in proportion between 0 and 3.9%, SRB such as Desulfovibrionaceae between 0.9 and 33.8%, and SOB such as Campylobacteraceae between 15 to 45%. After subsampling to 7,328 sequences per sample, we found 351 distinct OTUs across the entire dataset (Data S1). Species richness (Chao1) and bacterial diversity (Inverse Simpson) were relatively similar between samples, ranging from 98 to 149 OTUs per sample (Table 3).

Black band disease representative bacterial consortia

We compared the sequences from representative bacterial BBD consortium members found in four coral genera in the southern central Red Sea to sequences obtained from other

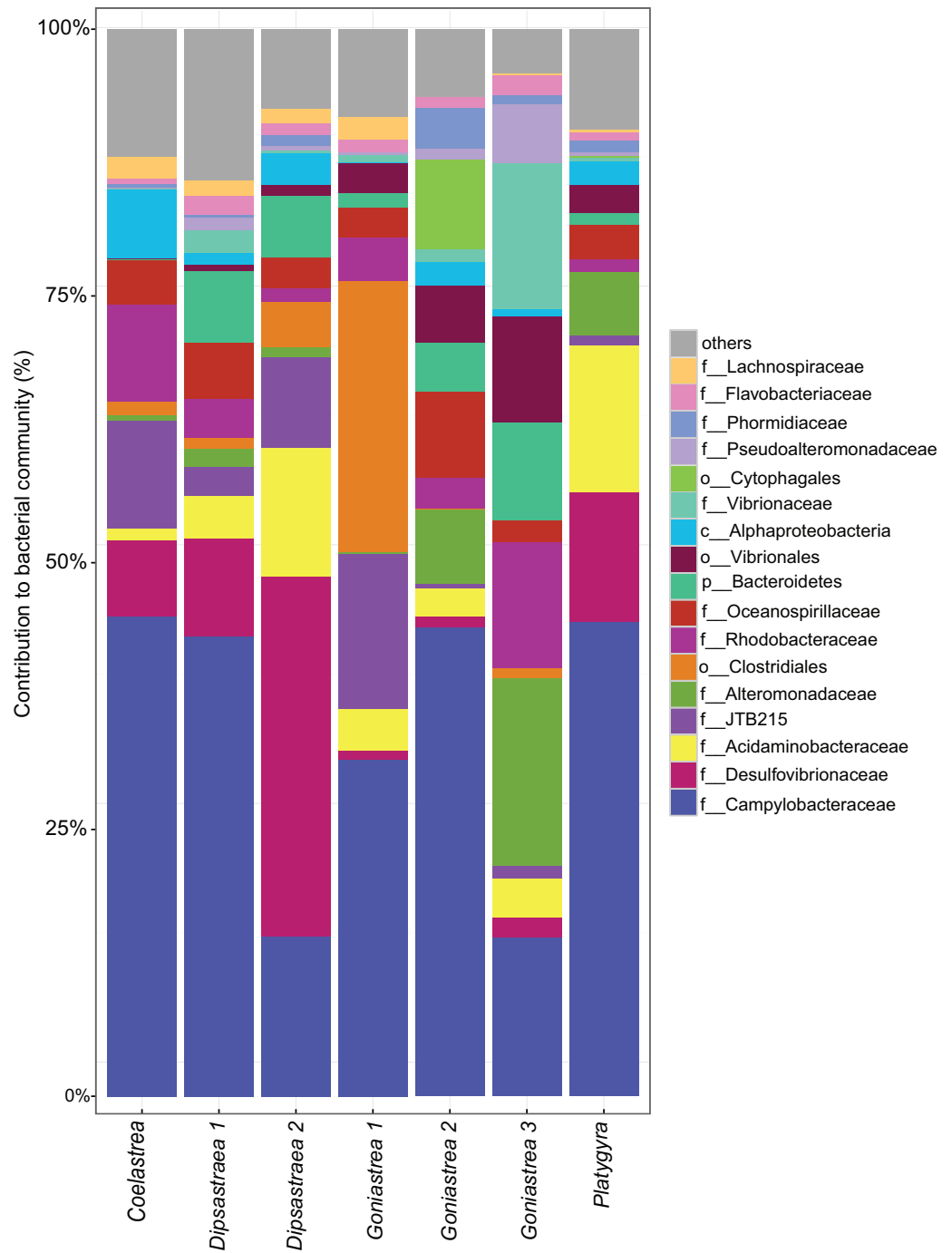


Figure 2 Bacterial community composition of black band disease microbial mats from four coral genera. (one colony of *Coelastrea*, two colonies of *Dipsastraea*, three colonies of *Goniastrea*, and one colony of *Platygyra*) from an outbreak site in the southern central Red Sea (Al-Lith fringing reef 1, Saudi Arabia). Taxonomy stacked column plot on the phylogenetic level of family or to lowest resolved taxonomic level (f, family; o, order; p, phylum). Each color represents one of the 17 most abundant families. Remaining taxa are grouped under category 'others'.

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locations and coral taxa that were affected by BBD on a global scale. Coral disease microbial mat-associated OTUs that represent the three main bacterial consortium members in BBD were successfully identified in our samples:

Sulfide oxidizing bacteria (SOB)

Beggiatoa sp., a common BBD-SOB member was absent in our samples, despite microscopic white filaments in the disease lesions which suggested its presence. Another SOB-consortium member *Arcobacter* sp. was present in all samples, which has been associated previously with BBD and with white plague disease (WPD) (Sunagawa et al., 2009). The SOB-classified OTUs were the most abundant taxa in the dataset. Several OTUs were found to be associated with all coral genera (i.e., *Coelastrea*, *Dipsastraea*, *Goniastrea*, and *Platygra*) with proportions of up to 22.6% in all coral samples (OTU0001, 2, 4, 11, 16: all *Arcobacter* sp., OTU0010: *Sulfurospirillum* sp.). These SOB-associated OTUs were found to be similar to those found in different places around the world (e.g., Philippines (Garren et al., 2009) and in the Caribbean including the Netherlands Antilles (Klaus, Janse & Fouke, 2011), US Virgin Islands (Cooney et al., 2002), and Puerto Rico (Sunagawa et al., 2009) (Table 4), where they were associated with varying coral species (Fig. 3A).

Sulfate-reducing bacteria (SRB)

Two abundant OTUs were found to be associated with BBD samples. These OTUs were annotated to *Desulfovibrio* sp. (OTU0005, OTU0006) with abundance ranges of 0.01–30.7% in all coral samples. Similar SRB-OTUs were found in the Caribbean (Sekar, Kaczmarek & Richardson, 2008; Sunagawa et al., 2009) and Japan in different coral species (e.g., *Montipora* sp., *Orbicella faveolata*, and *Siderastrea sidereal*, Table 4). OTU0005 (*Desulfovibrio dechloracetivorans*) clustered together with SRB previously found in corals diseased with WPD (Sunagawa et al., 2009) and BBD (Sekar et al., 2006), while OTU0006 (*Desulfovibrio marinisediminis*) clustered away (Fig. 3B), indicating that this is not a typical BBD consortium member.

Cyanobacteria

One cyanobacterium (OTU0023, *Oscillatoria* sp.) was found at proportions of up to 4% in our coral samples. Cyanobacteria of the same genus (99% sequence similarity) have previously been found in BBD infected *Pavona* sp. in the GBR (Buerger et al., 2016) and from other regions, e.g., the Caribbean (Casamatta et al., 2012), Hawaii (Aeby et al., 2015b), and Palau (Sussman, Bourne & Willis, 2006) (Table 4).

Others

Although not belonging to the three main BBD bacterial consortium members, Firmicutes have previously been reported in coral BBD (Barneah et al., 2007; Cooney et al., 2002; Klaus, Janse & Fouke, 2011). Members were also found in our dataset at proportions of up to 24.8% (OTU0003, OTU0009, OTU0013, OTU0018). Furthermore, the Firmicutes-associated OTUs in our data were similar to those found in *Porites* white patch syndrome (PWPS) (Séré et al., 2013) and WPD (Roder et al., 2014; Sunagawa et al., 2009) (Table 4).

We also retrieved sequences of *Vibrio* sp. (OTU0015, OTU0029) from our dataset, at proportions of up to 12.8%. These OTU sequences also had a high similarity (98–99%)

Table 4 Summary of bacterial taxa (OTUs) associated with black band disease (BBD) in corals from the southern central Red Sea and comparison with similar taxa from around the world, based on BLAST results (accession number, identity) of the BBD consortium of sulfide-oxidizing bacteria (SOB), sulfate-reducing bacteria (SRB), cyanobacteria, Firmicutes, and *Vibrio* sp.

OTU	Count	Taxonomy	Identity	GenBank Acc No.	Reference	Health state	Host & location
SOB							
Otu0001	913	<i>Arcobacter</i> sp.	99%	EF089456	<i>Barneah et al. (2007)</i>	BBD	<i>Favites</i> and <i>Dipsastraea</i> , Red Sea
				KC527436	<i>Roder et al. (2014)</i>	WPD	<i>Pavona duerdeni</i> and <i>Porites lutea</i> , West Pacific
				HM768631	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
Otu0002	625	<i>Arcobacter</i> sp.	99%	GU319311	<i>Meron et al. (2010)</i>	Healthy	<i>Acropora eurystoma</i> , Red Sea
				FJ203140	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean
				AB235414	<i>Yasumoto-Hirose et al. (2006)</i>	–	Non-coral species
Otu0004	336	<i>Arcobacter</i> sp.	99%	KT973145	<i>Couradeau et al. (2017)</i>	–	Non-coral species
				JF344171	<i>Acosta-González, Rosselló-Móra & Marqués (2012)</i>	–	Non-coral species
				FJ949362	<i>Suárez-Suárez et al. (2011)</i>	–	Non-coral species
Otu0010	246	<i>Sulfurospirillum</i> sp.	98%	LC026456	K Yamaki, F Mori, R Ueda, R Kondo, U Umezawa, H Nakata & M Wada (2015, unpublished data)	–	Non-coral species
				AF473976	<i>Cooney et al. (2002)</i>	BBD	Faviidae, Caribbean
				GU472074	L Arotsker, D Rasoulouniriana, N Siboni, E Ben-Dov, E Kramarsky-Winter, Y Loya & A Kushmaro (2010, unpublished data)	BBD	–
Otu0011	208	<i>Arcobacter</i> sp.	99%	HM768558	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, and Gorgoniidae, Caribbean
				GQ413587	<i>Garren et al. (2009)</i>	–	<i>Porites cylindrica</i> , West Pacific
Otu0016	117	<i>Arcobacter</i> sp.	98%	LC133150	S Iehata, Y Mizutani & R Tanaka (2016, unpublished data)	–	Non-coral species
				HE804002	C Chiellini, R Iannelli, F Verni & G Petroni (2012, unpublished data)	–	Non-coral species
				KF185679	J Vojvoda, D Lamy, E Sintes, JA Garcia, V Turk & GJ Herndl (2013, unpublished data)	–	Non-coral species
SRB							
Otu0005	322	<i>Desulfovibrio dechloracetivorans</i>	98%	AB470955	K Yoshinaga, BE Casareto & Y Suzuki (2008, unpublished data)	Healthy	<i>Montipora</i> sp., West Pacific
				FJ202627	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean
				EF123510	<i>Sekar, Kaczmarek & Richardson (2008)</i>	BBD	<i>Siderastrea siderea</i> , Caribbean

(continued on next page)

Table 4 (continued)

OTU	Count	Taxonomy	Identity	GenBank Acc No.	Reference	Health state	Host & location
Otu0006	294	<i>Desulfovibrio marinisediminis</i>	99%	MF039931	R Keren, A Lavy, I Polishchuk, B Pokroy & M Ilan (2017, unpublished data)	–	Non-coral species
				KY771114	H Zouch, F Karray, A Fabrice, S Chifflet, A Hirschler, H Kharrat, W Ben Hania, B Ollivier, S Sayadi & M Quemeneur (2017, unpublished data)	–	Non-coral species
				KT373805	K Alasvand Zarasvand & VR Rai (2015, unpublished data)	–	Non-coral species
Cyanobacteria							
Otu0023	81	<i>Oscillatoria</i> sp.	99%	KU579394	<i>Buerger et al. (2016)</i>	BBD	<i>Pavona</i> , Great Barrier Reef
				HM768593	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
				GU472422	L Arotsker, D Rasoulouniriana, N Siboni, E Ben-Dov, E Kramarsky-Winter, Y Loya & A Kushmaro (2010, unpublished data)	BBD	–
Firmicutes							
Otu0003	344	family JTB215	99%	DQ647593	R Guppy & JC Bythell (2006, unpublished data)	–	–
				KC527313	<i>Roder et al. (2014)</i>	WPD	<i>Pavona duerdeni</i> and <i>Porites lutea</i> , Caribbean
				HM768569	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
Otu0013	199	<i>Fusibacter</i> sp.	99%	GQ413281	<i>Garren et al. (2009)</i>	–	<i>Porites cylindrica</i> , West Pacific
				FJ202930	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean
Otu0018	112	<i>Fusibacter</i> sp.	99%	KF179748	<i>Séré et al. (2013)</i>	PWPS	<i>Porites lutea</i> , Western Indian Ocean
				GU472060	L Arotsker, D Rasoulouniriana, N Siboni, E Ben-Dov, E Kramarsky-Winter, Y Loya & A Kushmaro (2010, unpublished data)	BBD	–
				EU780347	SE Godwin, J Borneman, E Bent & L Pereg-Gerk (2008, unpublished data)	SWS	<i>Turbinaria mesenterina</i> ,
				EF089469	<i>Barneah et al. (2007)</i>	BBD	<i>Favites, Dipsastraea</i> , Red Sea
Otu0022	82	family Lachnospiraceae	99%	HM768582	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
			98%	AF473930	<i>Cooney et al. (2002)</i>	BBD	Faviidae, Caribbean
				DQ647585	R Guppy & JC Bythell (2006, unpublished data)	–	–
Otu0027	62	<i>Fusibacter</i> sp.	99%	JX391361	YYK Chan, AL Li, S Gopalakrishnan, RSS Wu, SB Pointing & JMY Chiu (2012, unpublished data)	–	Non-coral species
				HM768587	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
				FJ202981	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean
Otu0031	48	<i>WH1-8</i> sp.	99%	KF179804	<i>Séré et al. (2013)</i>	PWPS	<i>Porites lutea</i> , Western Indian Ocean
				KC527300	<i>Roder et al. (2014)</i>	WPD	<i>Pavona duerdeni</i> and <i>Porites lutea</i> , West Pacific
				FJ203165	<i>Sunagawa et al. (2009)</i>	WPD	Faviidae, Caribbean

(continued on next page)

Table 4 (continued)

OTU	Count	Taxonomy	Identity	GenBank Acc No.	Reference	Health state	Host & location
Otu0039	30	<i>Defluviitalea saccharophila</i>	99%	DQ647556	R Guppy & JC Bythell (2006, unpublished data)	–	–
				FJ202907	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean
				AF473925	<i>Cooney et al. (2002)</i>	BBD	Faviidae, Caribbean
<i>Vibrio</i> sp.							
Otu0015	170	<i>Vibrio</i> sp.	99%	KT974549	<i>Couradeau et al. (2017)</i>	–	Non-coral species
			98%	GU471972	L Arotsker, D Rasoulouniriana, N Siboni, E Ben-Dov, E Kramarsky-Winter, Y Loya & A Kushmaro (2010, unpublished data)	BBD	–
			98%	MF461384	<i>Keller-Costa et al. (2017)</i>	–	<i>Eunicella labiata</i>
Otu0029	56	order Vibrionales	99%	JQ727003	<i>Witt, Wild & Uthicke (2012)</i>	–	Non-coral species, GBR
				HM768601	<i>Klaus, Janse & Fouke (2011)</i>	BBD	Faviidae, Meandrinidae, Gorgoniidae, Caribbean
				FJ202558	<i>Sunagawa et al. (2009)</i>	WPD	<i>Orbicella faveolata</i> , Caribbean

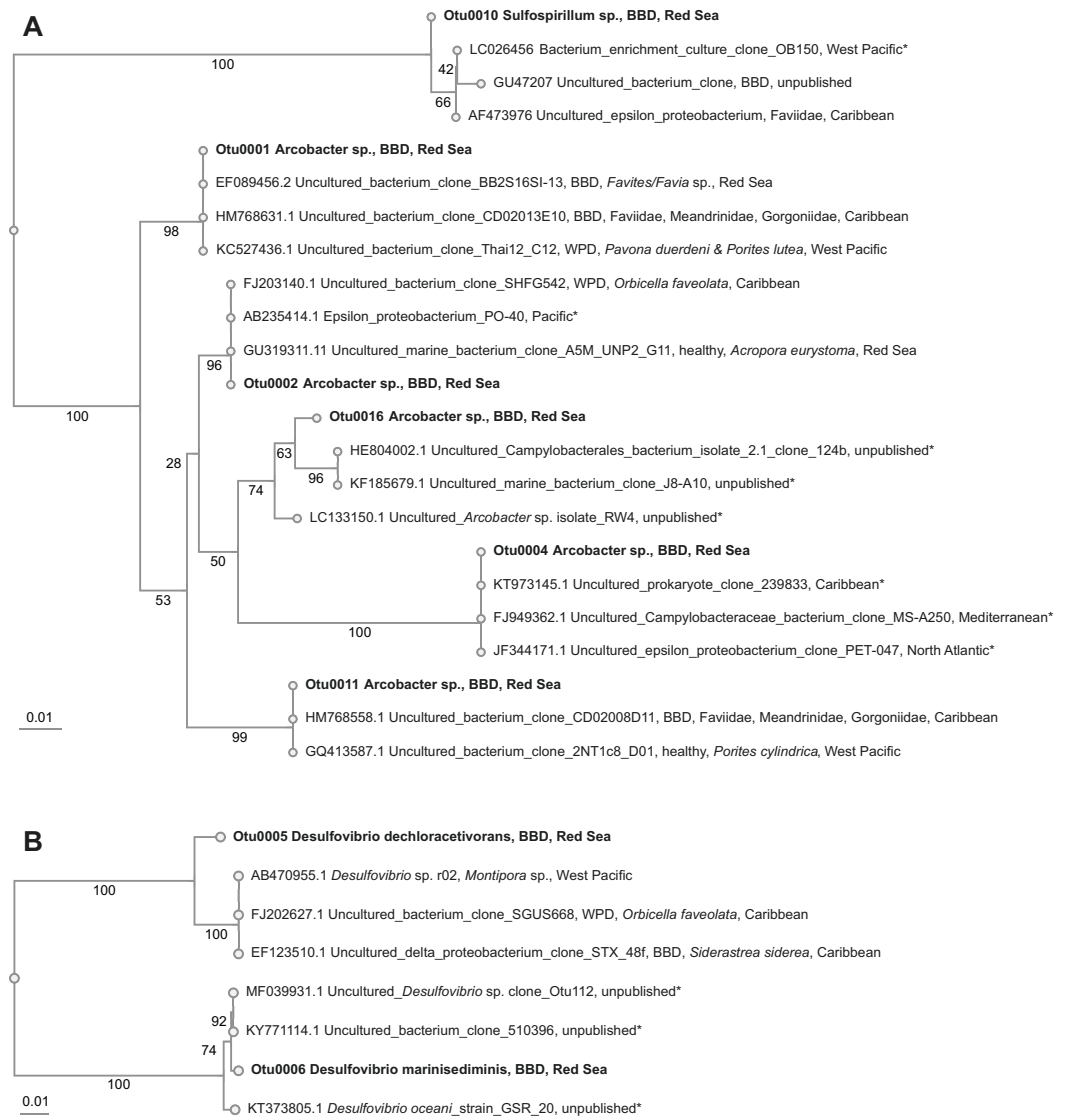


Figure 3 Overview and phylogenetic relationship of coral black band disease bacterial consortium members from the southern central Red Sea (Al-Lith, Saudi Arabia) and other regions. (A) Sulfide-oxidizing bacteria (SOB); (B) Sulfate-reducing bacteria (SRB). Phylogenetic trees were calculated using the neighbor-joining method, bootstrap values are indicated at the branches. The phylogenetic trees show NCBI accession numbers and sample name, health state of the coral species, host name, and region. Sequences from this study are in bold. The ‘*’ indicates that the bacterial species were not found in coral species.

Full-size DOI: 10.7717/peerj.5169/fig-3

to sequences from BBD and WPD (Klaus, Janse & Fouke, 2011; Sunagawa et al., 2009) (Table 4).

DISCUSSION

In this study, we report on the distribution and prevalence of coral black band disease in the Red Sea. Our surveys ranged from 19.9 to 24.1 degrees of latitude and confirm the

presence of BBD across the central Red Sea. Molecular characterization of the bacterial community identified the three main bacterial members of the disease consortium across coral species at a BBD outbreak site in the southern central Red Sea.

Black band disease distribution and prevalence in the Red Sea in comparison to other global sites

BBD is a global disease found in numerous regions, but its prevalence on coral reefs is generally low compared to other diseases such as white syndrome (WS) (*Dinsdale, 2002; Edmunds, 1991; Page & Willis, 2006; Willis, Page & Dinsdale, 2004*). The low prevalence recorded in this study is similar to levels reported elsewhere across the globe (*Sutherland, Porter & Torres, 2004*) with localized outbreaks of BBD also reported in the GBR (*Sato, Bourne & Willis, 2009*), Hawaii (*Aeby et al., 2015b*), Jamaica (*Bruckner & Bruckner, 1997*), Venezuela (*Rodríguez & Cróquer, 2008*), and the Red Sea (*Al-Moghrabi, 2001*). In the Red Sea, BBD was first discovered in the 1980s (*Antonius, 1981*) and our study confirms that BBD is a chronic threat to coral reefs in the Red Sea with localized outbreaks continuing to occur.

BBD is not a selective disease; multiple species and various levels of severity can affect colonies within and between coral species and across reefs (*Bruckner, Bruckner & Williams, 1997; Dinsdale, 2002; Green & Bruckner, 2000; Peters, 1993*). This was also observed in our study, where multiple species were infected, but with differences in prevalence among coral taxa. At the outbreak site, we found BBD prevalence to be highest in the genus *Dipsastraea*, which suggests that this genus may be an important host for BBD in the Red Sea. Our observations match previous reports and shows that this pattern is consistent through time (*Antonius, 1985*). Interestingly, although differential susceptibility to BBD among coral taxa has been found globally, the most vulnerable taxa differ by region. For example, in the Caribbean *Montastraea/Orbicella* are commonly infected (*Bruckner & Bruckner, 1997; Porter et al., 2001*), *Montipora* in Hawaii (*Aeby et al., 2015b*), and *Acropora* on the GBR (*Page & Willis, 2006*). It would be fruitful to examine the underlying defense mechanisms in the different coral taxa that lead to these differences in BBD occurrence.

BBD, climate change, and coral bleaching

The occurrence of BBD has been linked to elevated seawater temperatures (*Boyett, Bourne & Willis, 2007; Kuta & Richardson, 2002; Muller & Van Woesik, 2011*). The occurrence of a BBD outbreak during a bleaching event in the present study reflects previous reports from the Caribbean, where the positive correlation between bleaching events and BBD incidence was proposed first (*Brandt & McManus, 2009; Cróquer & Weil, 2009*). For instance, in the Florida Reef Tract, the prevalence of BBD increased from 0 to 6.7% following bleaching events in 2014 and 2015 (*Lewis et al., 2017*). Also, *Cróquer & Weil (2009)* found a significant linear correlation between coral bleaching and the prevalence of two other virulent diseases (yellow band disease and white plague) affecting *Montastraea/Orbicella* species. This further supports a strong relationship between bleaching events and the emergence of some coral diseases on a global scale. Understanding how climate change-related thermal anomalies and coral bleaching drive the emergence and virulence of coral diseases is essential for future research.

It has further been suggested that other anthropogenic activities, such as coastal pollution or ocean acidification, contribute to the increase of coral disease incidents ([Jackson et al., 2001](#); [Muller et al., 2017](#); [Rosenberg & Ben-Haim, 2002](#)). The surveyed outbreak area was adjacent to the outflow of a large aquaculture facility, which might have further aggravated the effects of the bleaching event due to increased nutrient availability ([Roder et al., 2015](#); [Ziegler et al., 2016](#)). In comparison, other reefs in the Al-Lith area that were farther away from the coast displayed similar levels of bleaching, but BBD prevalence stayed at baseline levels in these locations. This suggests that bleaching alone was not the only factor that could have contributed to the BBD outbreak. The synergistic effects of high temperatures and nutrient pollution find further support in the Caribbean where BBD prevalence increased in reef sites with direct sewage input compared to control sites ([Sekar, Kaczmarzky & Richardson, 2008](#)) and in the Bahamas where BBD migration was faster in nutrient-enriched areas ([Voss & Richardson, 2006](#)). Further work is needed to directly examine the relationship between bleaching, nutrient stress, and BBD susceptibility.

Bacterial community composition of BBD microbial mats from the southern central Red Sea reflects global microbial patterns with local characteristics

Our results verify the presence of the three main consortium members in BBD microbial mats (Cyanobacteria, SOB, SRB) of corals from the southern central Red Sea. We identified *Oscillatoria* sp. as a BBD-associated cyanobacterium, which is similar to the BBD-associated cyanobacteria in other regions of the world ([Aeby et al., 2015b](#); [Arotsker et al., 2015](#); [Buerger et al., 2016](#); [Casamatta et al., 2012](#); [Cooney et al., 2002](#); [Frias-Lopez et al., 2003](#); [Gantar, Sekar & Richardson, 2009](#); [Glas et al., 2010](#); [Meyer et al., 2016](#); [Miller & Richardson, 2011](#); [Rasoulouniriana et al., 2009](#); [Sato, Willis & Bourne, 2010](#); [Sussman, Bourne & Willis, 2006](#)). However, we retrieved only a low number of cyanobacterial sequences, although cyanobacterial filaments were visually abundant in the sampled microbial mats, which could possibly be related to primer amplification bias. In addition, members of the SOB and SRB functional groups (*Arcobacter* sp. and *Desulfovibrio* sp., respectively) from BBD microbial mats in the southern central Red Sea were similar to those found in other BBD-affected corals worldwide ([Barneah et al., 2007](#); [Cooney et al., 2002](#); [Klaus, Janse & Fouke, 2011](#); [Sekar, Kaczmarzky & Richardson, 2008](#)). This confirms that BBD-associated bacteria are not restricted to a specific coral species or region ([Barneah et al., 2007](#); [Cooney et al., 2002](#); [Dinsdale, 2002](#); [Frias-Lopez et al., 2003](#)). Interestingly, we did observe white filaments within lesions that were morphologically similar to *Beggiatoa*, a sulfide-oxidizing bacterium associated with BBD in other regions ([Cooney et al., 2002](#); [Miller & Richardson, 2011](#); [Sato, Willis & Bourne, 2010](#)). However, we found no sequences aligning with *Beggiatoa* in our study. This suggests that either the white filaments were not *Beggiatoa* or that the methods used were not adequate to extract and identify *Beggiatoa*. [Aeby et al. \(2015b\)](#) sequenced *Beggiatoa* from BBD lesions in Hawaii by first culturing the white filaments from lesions and then using universal bacterial primers 8F and 1513R for sequencing. However, they found that no DNA sequences were available for *Beggiatoa* found in BBD from other

regions even though numerous studies using molecular techniques have been published. Further work is needed to clarify these discrepancies.

Besides the three main bacterial consortium members that dominate BBD microbial mats, we detected other bacterial families as part of the BBD consortium. Members of the Firmicutes were abundant in BBD microbial mats, which is consistent with other studies (Arotsker *et al.*, 2016; Arotsker *et al.*, 2009; Barneah *et al.*, 2007; Cooney *et al.*, 2002; Frias-Lopez *et al.*, 2002; Miller & Richardson, 2011; Richardson, 2004; Sekar, Kaczmarsky & Richardson, 2008). In addition, we detected the presence of *Vibrio* species. The pathogenicity of this genus has been documented previously in corals and other marine organisms (Ben-Haim, Zicherman-Keren & Rosenberg, 2003; Harvell *et al.*, 1999; Kushmaro *et al.*, 1996), and more broadly *Vibrios* have been characterized as opportunistic taxa (Cervino *et al.*, 2004; Rosenberg & Falkovitz, 2004; Thompson *et al.*, 2004; Ziegler *et al.*, 2016). To date it is unknown whether this group plays a role in the etiology of BBD (Arotsker *et al.*, 2009; Barneah *et al.*, 2007) (Meyer *et al.*, 2016), or whether the high number of *Vibrios* could be related to seasonal increases in the coral microbiome and coral bleaching (reviewed in Rosenberg & Koren, 2006; Tout *et al.*, 2015).

CONCLUSIONS

Our study represents the first comprehensive assessment of Black Band Disease in the central Red Sea. Elucidation of the bacteria associated with BBD microbial mats of corals at a southern reef site confirms that BBD represents a disease with predictable etiology where the three main bacterial players are globally distributed with regional differences. Notably, our reef survey data, in line with data from other regions, identify BBD as a widespread disease, but as one with low prevalence in comparison to other coral diseases. Additional surveys including other coral diseases as well as pathogen infection experiments with Red Sea corals could further increase our understanding of coral stress tolerance in this understudied coral reef region. Importantly, the prevalence of BBD might increase with ongoing ocean warming and thermal anomalies, as supported by the here-documented disease outbreak coinciding with a thermal anomaly and widespread coral bleaching. The collection of long-term monitoring disease data in the Arabian Seas is important in order to establish baselines, which can then assist in more accurate prediction of disease prevalence and potential impact of climate change on coral communities in this region.

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ADDITIONAL INFORMATION AND DECLARATIONS

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Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Ghaida Hadaidi performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, approved the final draft.
- Maren Ziegler conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the paper, approved the final draft.
- Amanda Shore-Maggio performed the experiments, analyzed the data, approved the final draft.
- Thor Jensen performed the experiments, approved the final draft.
- Greta Aeby and Christian R. Woolstra conceived and designed the experiments, performed the experiments, analyzed the data, contributed reagents/materials/analysis tools, prepared figures and/or tables, authored or reviewed drafts of the paper, approved the final draft.

DNA Deposition

The following information was supplied regarding the deposition of DNA sequences:

Sequence data determined in this study is available at NCBI under BioProject ID [PRJNA436216](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA436216). Abundant coral bacterial microbiome OTU reference sequences are available under GenBank Accession numbers [MH341637–MH341689](https://www.ncbi.nlm.nih.gov/nuclseq/MH341637-MH341689).

Data Availability

The following information was supplied regarding data availability:

The OTU abundance table is provided as a [Data S1](#).

Supplemental Information

Supplemental information for this article can be found online at <http://dx.doi.org/10.7717/peerj.5169#supplemental-information>.

REFERENCES

- Acosta-González A, Rosselló-Móra R, Marqués S. 2012.** Characterization of the anaerobic microbial community in oil-polluted subtidal sediments: aromatic biodegradation potential after the Prestige oil spill. *Environmental Microbiology* 15:77–92 DOI 10.1111/j.1462-2920.2012.02782.x.
- Aeby GS, Tribollet A, Lasne G, Work TM. 2015a.** Assessing threats from coral and crustose coralline algae disease on the reefs of New Caledonia. *Marine and Freshwater Research* 34:393–406 DOI 10.1071/MF14151.
- Aeby GS, Work TM, Runyon CM, Shore-Maggio A, Ushijima B, Videau P, Beurmann S, Callahan SM. 2015b.** First record of black band disease in the Hawaiian archipelago: response, outbreak status, virulence, and a method of treatment. *PLOS ONE* 10:e0120853 DOI 10.1371/journal.pone.0120853.
- Ainsworth T, Kramasky-Winter E, Loya Y, Hoegh-Guldberg O, Fine M. 2007.** Coral disease diagnostics: what's between a plague and a band? *Applied and Environmental Microbiology* 73:981–992 DOI 10.1128/AEM.02172-06.
- Al-Moghrabi S. 2001.** Unusual black band disease (BBD) outbreak in the northern tip of the Gulf of Aqaba (Jordan). *Coral Reefs* 19:330–331 DOI 10.1007/s003380000127.
- Antonius A. 1973.** New observations on coral destruction in reefs. In: *Tenth meeting of the association of Island Marine Laboratories of the Caribbean*. Mayaguez: University of Puerto Rico.
- Antonius A. 1981.** Coral reef pathology: a review. In: Gomez ED, Birkeland CE, Bud-demeier RW, Johannes RE, Marsh Jr JA, Tsuda RT, eds. *Proceedings of the 4th international coral reef symposium, Vol. 2. Marine Science Center, University of the Philippines, Manila, Philippines*. Waco: International Society for Reef Studies.
- Antonius A. 1985.** Coral diseases in the Indo-Pacific: a first record. *Marine Ecology* 6:197–218 DOI 10.1111/j.1439-0485.1985.tb00322.x.
- Antonius A. 1988.** Distribution and dynamics of coral diseases in the Eastern Red Sea. In: Choat JH, Barnes D, Borowitzka MA, Coll JC, Davies PJ, Flood P, Hatcher BG, Hopley D, Hutchings PA, Kinsey D, Orme GR, Pichon M, Sale PF, Sammarco P, Wallace CC, Wilkinson C, Wolanski E, Bellwood O, eds. *Proceedings of the 6th international coral reef symposium: vol. 2: contributed papers. Townsville, Australia*. Waco: International Society for Reef Studies, 293–298.
- Apprill A, McNally S, Parsons R, Weber L. 2015.** Minor revision to V4 region SSU rRNA 806R gene primer greatly increases detection of SAR11 bacterioplankton. *Aquatic Microbial Ecology* 75:129–137 DOI 10.3354/ame01753.
- Aronson RB, Precht WF. 2001.** White-band disease and the changing face of Caribbean coral reefs. In: Porter JW, ed. *The ecology and etiology of newly emerging marine diseases*. Dordrecht: Springer, 25–38.
- Arotsker L, Kramarsky-Winter E, Ben-Dov E, Kushmaro A. 2016.** Microbial transcriptome profiling of black band disease in a Faviid coral during a seasonal disease peak. *Diseases of Aquatic Organisms* 118:77–89 DOI 10.3354/dao02952.

- Arotsker L, Kramarsky-Winter E, Ben-Dov E, Siboni N, Kushmaro A. 2015.** Changes in the bacterial community associated with black band disease in a Red Sea coral, *Favia* sp. in relation to disease phases. *Diseases of Aquatic Organisms* **116**:47–58 DOI [10.3354/dao02911](https://doi.org/10.3354/dao02911).
- Arotsker L, Siboni N, Ben-Dov E, Kramarsky-Winter E, Loya Y, Kushmaro A. 2009.** *Vibrio* sp. as a potentially important member of the Black Band Disease (BBD) consortium in *Favia* sp. corals. *FEMS Microbiology Ecology* **70**:515–524 DOI [10.1111/j.1574-6941.2009.00770.x](https://doi.org/10.1111/j.1574-6941.2009.00770.x).
- Barneah O, Ben-Dov E, Kramarsky-Winter E, Kushmaro A. 2007.** Characterization of black band disease in Red Sea stony corals. *Environmental Microbiology* **9**:1995–2006 DOI [10.1111/j.1462-2920.2007.01315.x](https://doi.org/10.1111/j.1462-2920.2007.01315.x).
- Ben-Haim Y, Zicherman-Keren M, Rosenberg E. 2003.** Temperature-regulated bleaching and lysis of the coral pocillopora damicornis by the novel pathogen *vibrio coralliilyticus*. *Applied and Environmental Microbiology* **69**:4236–4242 DOI [10.1128/aem.69.7.4236-4242.2003](https://doi.org/10.1128/aem.69.7.4236-4242.2003).
- Bourne DG, Muirhead A, Sato Y. 2011.** Changes in sulfate-reducing bacterial populations during the onset of black band disease. *ISME Journal* **5**:559–564 DOI [10.1038/ismej.2010.143](https://doi.org/10.1038/ismej.2010.143).
- Boyett HV, Bourne DG, Willis BL. 2007.** Elevated temperature and light enhance progression and spread of black band disease on staghorn corals of the Great Barrier Reef. *Marine Biology* **151**:1711–1720 DOI [10.1007/s00227-006-0603-y](https://doi.org/10.1007/s00227-006-0603-y).
- Brandt ME, McManus JW. 2009.** Disease incidence is related to bleaching extent in reef-building corals. *Ecology* **90**:2859–2867 DOI [10.1890/08-0445.1](https://doi.org/10.1890/08-0445.1).
- Bruckner A, Bruckner R. 1997.** The persistence of black band disease in Jamaica: impact on community structure. In: Lessios HA, Macintyre IG, eds. *Proceedings of the 8th international coral reef symposium vol. 1. Smithsonian Tropical Research Institute, Panama*. Waco: International Society for Reef Studies, 601–606.
- Bruckner AW, Bruckner RJ, Williams JEH. 1997.** Spread of a black-band disease epizootic through the coral reef system in St. Ann’s Bay, Jamaica. *Bulletin of Marine Science* **61**:919–928.
- Bruckner. 2002.** Priorities for effective management of coral diseases. US Department of Commerce, National Oceanic and Atmospheric Administration, National Marine Fisheries Service Available at <https://www.fisheries.noaa.gov/resource/document/priorities-effective-management-coral-diseases>.
- Buerger P, Alvarez-Roa C, Weynberg KD, Baekelandt S, Van Oppen MJ. 2016.** Genetic, morphological and growth characterisation of a new *Roseofilum* strain (Oscillatoriales, Cyanobacteria) associated with coral black band disease. *PeerJ* **4**:e2110 DOI [10.7717/peerj.2110](https://doi.org/10.7717/peerj.2110).
- Caporaso JG, Lauber CL, Walters WA, Berg-Lyons D, Huntley J, Fierer N, Owens SM, Betley J, Fraser L, Bauer M, Gormley N, Gilbert JA, Smith G, Knight R. 2012.** Ultra-high-throughput microbial community analysis on the Illumina HiSeq and MiSeq platforms. *The ISME journal* **6**:1621–1624 DOI [10.1038/ismej.2012.8](https://doi.org/10.1038/ismej.2012.8).

- Carlton RG, Richardson LL. 1995.** Oxygen and sulfide dynamics in a horizontally migrating cyanobacterial mat: black band disease of corals. *FEMS Microbiology Ecology* **18**:155–162 DOI [10.1016/0168-6496\(95\)00052-C](https://doi.org/10.1016/0168-6496(95)00052-C).
- Casamatta D, Stanić D, Gantar M, Richardson LL. 2012.** Characterization of *Roseofilum reptotaenium* (Oscillatoriales, Cyanobacteria) gen. sp. nov. isolated from Caribbean black band disease. *Phycologia* **51**:489–499 DOI [10.2216/11-10.1](https://doi.org/10.2216/11-10.1).
- Cervino JM, Hayes RL, Polson SW, Polson SC, Goreau TJ, Martinez RJ, Smith GW. 2004.** Relationship of *Vibrio* species infection and elevated temperatures to yellow blotch/band disease in Caribbean corals. *Applied and Environmental Microbiology* **70**:6855–6864 DOI [10.1128/AEM.70.11.6855-6864.2004](https://doi.org/10.1128/AEM.70.11.6855-6864.2004).
- Chao A. 1984.** Nonparametric estimation of the number of classes in a population. *Scandinavian Journal of Statistics* **11**:265–270.
- Cooney RP, Pantos O, Tissier MDAL, Barer MR, O'Donnell AG, Bythell JC. 2002.** Characterization of the bacterial consortium associated with black band disease in coral using molecular microbiological techniques. *Environmental Microbiology* **4**:401–413 DOI [10.1046/j.1462-2920.2002.00308.x](https://doi.org/10.1046/j.1462-2920.2002.00308.x).
- Couradeau E, Roush D, Guida BS, Garcia-Pichel F. 2017.** Diversity and mineral substrate preference in endolithic microbial communities from marine intertidal outcrops (Isla de Mona, Puerto Rico). *Biogeosciences* **14**:311.
- Cróquer A, Weil E. 2009.** Changes in Caribbean coral disease prevalence after the 2005 bleaching event. *Diseases of Aquatic Organisms* **87**:33–43 DOI [10.3354/dao02164](https://doi.org/10.3354/dao02164).
- Dinsdale E. 2002.** Abundance of black-band disease on corals from one location on the Great Barrier Reef: a comparison with abundance in the Caribbean region. In: Moosa MK, Soemodihardjo S, Soegiarto A, Romimohtarto K, Nontji A, Soekarno, Suharsono, eds. *Proceedings of the ninth international coral reef symposium, 23–27 October 2000*. Waco: International Society for Reef Studies, 1239–1243.
- Edgar RC, Haas BJ, Clemente JC, Quince C, Knight R. 2011.** UCHIME improves sensitivity and speed of chimera detection. *Bioinformatics* **27**:2194–2200 DOI [10.1093/bioinformatics/btr381](https://doi.org/10.1093/bioinformatics/btr381).
- Edmunds PJ. 1991.** Extent and effect of black band disease on a Caribbean reef. *Coral Reefs* **10**:161–165 DOI [10.1007/bf00572175](https://doi.org/10.1007/bf00572175).
- Frias-Lopez J, Bonheyo GT, Jin Q, Fouke BW. 2003.** Cyanobacteria associated with coral black band disease in Caribbean and Indo-Pacific reefs. *Applied and Environmental Microbiology* **69**:2409–2413 DOI [10.1128/AEM.69.4.2409-2413.2003](https://doi.org/10.1128/AEM.69.4.2409-2413.2003).
- Frias-Lopez J, Klaus JS, Bonheyo GT, Fouke BW. 2004.** Bacterial community associated with black band disease in corals. *Applied and Environmental Microbiology* **70**:5955–5962 DOI [10.1128/AEM.70.10.5955-5962.2004](https://doi.org/10.1128/AEM.70.10.5955-5962.2004).
- Frias-Lopez J, Zerkle AL, Bonheyo GT, Fouke BW. 2002.** Partitioning of bacterial communities between seawater and healthy, black band diseased, and dead coral surfaces. *Applied and Environmental Microbiology* **68**:2214–2228 DOI [10.1128/AEM.68.5.2214-2228.2002](https://doi.org/10.1128/AEM.68.5.2214-2228.2002).

- Gantar M, Sekar R, Richardson LL. 2009.** Cyanotoxins from black band disease of corals and from other coral reef environments. *Microbial Ecology* **58**:856–864 DOI [10.1007/s00248-009-9540-x](https://doi.org/10.1007/s00248-009-9540-x).
- Garren M, Raymundo L, Guest J, Harvell CD, Azam F. 2009.** Resilience of coral-associated bacterial communities exposed to fish farm effluent. *PLOS ONE* **4**:e7319 DOI [10.1371/journal.pone.0007319](https://doi.org/10.1371/journal.pone.0007319).
- Glas MS, Motti CA, Negri AP, Sato Y, Froscio S, Humpage AR, Krock B, Cembella A, Bourne DG. 2010.** Cyanotoxins are not implicated in the etiology of coral black band disease outbreaks on Pelorus Island, Great Barrier Reef. *FEMS Microbiology Ecology* **73**:43–54 DOI [10.1111/j.1574-6941.2010.00874.x](https://doi.org/10.1111/j.1574-6941.2010.00874.x).
- Glas MS, Sato Y, Ulstrup KE, Bourne DG. 2012.** Biogeochemical conditions determine virulence of black band disease in corals. *The ISME journal* **6**:1526–1534 DOI [10.1038/ismej.2012.2](https://doi.org/10.1038/ismej.2012.2).
- Green EP, Bruckner AW. 2000.** The significance of coral disease epizootiology for coral reef conservation. *Biological Conservation* **96**:347–361 DOI [10.1016/S0006-3207\(00\)00073-2](https://doi.org/10.1016/S0006-3207(00)00073-2).
- Harvell CD, Kim K, Burkholder JM, Colwell RR, Epstein PR, Grimes DJ, Hofmann EE, Lipp EK, Osterhaus ADME, Overstreet RM, Porter JW, Smith GW, Vasta GR. 1999.** Emerging marine diseases—climate links and anthropogenic factors. *Science* **285**:1505–1510 DOI [10.1126/science.285.5433.1505](https://doi.org/10.1126/science.285.5433.1505).
- Harvell D, Altizer S, Cattadori IM, Harrington L, Weil E. 2009.** Climate change and wildlife diseases: when does the host matter the most? *Ecology* **90**:912–920 DOI [10.1890/08-0616.1](https://doi.org/10.1890/08-0616.1).
- Hoegh-Guldberg O. 2012.** Coral reefs, climate change, and mass extinction. In: Hannah L, ed. *Saving a million species: extinction risk from climate change*. Washington, D.C.: Island Press/Center for Resource Economics, 261–283.
- Hughes TP. 1994.** Catastrophes, phase shifts, and large-scale degradation of a Caribbean coral reef. *Science* **265**:1547–1551 DOI [10.1126/science.265.5178.1547](https://doi.org/10.1126/science.265.5178.1547).
- Huse SM, Welch DM, Morrison HG, Sogin ML. 2010.** Ironing out the wrinkles in the rare biosphere through improved OTU clustering. *Environmental Microbiology* **12**:1889–1898 DOI [10.1111/j.1462-2920.2010.02193.x](https://doi.org/10.1111/j.1462-2920.2010.02193.x).
- Jackson JBC, Kirby MX, Berger WH, Bjorndal KA, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes JA, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR. 2001.** Historical overfishing and the recent collapse of coastal ecosystems. *Science* **293**:629–637 DOI [10.1126/science.1059199](https://doi.org/10.1126/science.1059199).
- Kaczmarek L. 2006.** Coral disease dynamics in the central Philippines. *Diseases of Aquatic Organisms* **69**:9–21 DOI [10.3354/dao069009](https://doi.org/10.3354/dao069009).
- Katoh K, Rozewicki J, Yamada KD. 2017.** MAFFT online service: multiple sequence alignment, interactive sequence choice and visualization. *Briefings in Bioinformatics* Epub ahead of print Sep 6 2017 DOI [10.1093/bib/bbx108](https://doi.org/10.1093/bib/bbx108).

- Keller-Costa T, Eriksson D, Gonçalves JMS, Gomes NCM, Lago-Lestón A, Costa R. 2017.** The gorgonian coral *Eunicella labiata* hosts a distinct prokaryotic consortium amenable to cultivation. *FEMS Microbiology Ecology* **93**:fix143–fix143 DOI [10.1093/femsec/fix143](https://doi.org/10.1093/femsec/fix143).
- Klaus JS, Janse I, Fouke BW. 2011.** Coral black band disease microbial communities and genotypic variability of the dominant cyanobacteria (CD1C11). *Bulletin of Marine Science* **87**:795–821 DOI [10.5343/bms.2010.1050](https://doi.org/10.5343/bms.2010.1050).
- Kozich JJ, Westcott SL, Baxter NT, Highlander SK, Schloss PD. 2013.** Development of a dual-index sequencing strategy and curation pipeline for analyzing amplicon sequence data on the MiSeq Illumina sequencing platform. *Applied and Environmental Microbiology* **79**:5112–5120 DOI [10.1128/AEM.01043-13](https://doi.org/10.1128/AEM.01043-13).
- Kuraku S, Zmasek CM, Nishimura O, Katoh K. 2013.** aLeaves facilitates on-demand exploration of metazoan gene family trees on MAFFT sequence alignment server with enhanced interactivity. *Nucleic Acids Research* **41**:W22–W28 DOI [10.1093/nar/gkt389](https://doi.org/10.1093/nar/gkt389).
- Kushmaro A, Loya Y, Fine M, Rosenberg E. 1996.** Bacterial infection and coral bleaching. *Nature* **380**:396–396 DOI [10.1038/380396a0](https://doi.org/10.1038/380396a0).
- Kuta KG, Richardson LL. 1996.** Abundance and distribution of black band disease on coral reefs in the northern Florida keys. *Coral Reefs* **15**:219–223 DOI [10.1007/bf01787455](https://doi.org/10.1007/bf01787455).
- Kuta K, Richardson L. 2002.** Ecological aspects of black band disease of corals: relationships between disease incidence and environmental factors. *Coral Reefs* **21**:393–398 DOI [10.1007/s00338-002-0261-6](https://doi.org/10.1007/s00338-002-0261-6).
- Lewis CL, Neely KL, Richardson LL, Rodriguez-Lanetty M. 2017.** Temporal dynamics of black band disease affecting pillar coral (*Dendrogyra cylindrus*) following two consecutive hyperthermal events on the Florida Reef Tract. *Coral Reefs* **36**:427–431 DOI [10.1007/s00338-017-1545-1](https://doi.org/10.1007/s00338-017-1545-1).
- Meron D, Atias E, Iasur Kruh L, Elifantz H, Minz D, Fine M, Banin E. 2010.** The impact of reduced pH on the microbial community of the coral *Acropora eurystoma*. *The ISME Journal* **5**:51.
- McDonald D, Price MN, Goodrich J, Nawrocki EP, DeSantis TZ, Probst A, Andersen GL, Knight R, Hugenholtz P. 2012.** An improved Greengenes taxonomy with explicit ranks for ecological and evolutionary analyses of bacteria and archaea. *The ISME Journal* **6**:610–618 DOI [10.1038/ismej.2011.139](https://doi.org/10.1038/ismej.2011.139).
- McLeod E, Moffitt R, Timmermann A, Salm R, Menviel L, Palmer MJ, Selig ER, Casey KS, Bruno JF. 2010.** Warming seas in the coral triangle: coral reef vulnerability and management implications. *Coastal Management* **38**:518–539 DOI [10.1080/08920753.2010.509466](https://doi.org/10.1080/08920753.2010.509466).
- Meyer JL, Gunasekera SP, Scott RM, Paul VJ, Teplitski M. 2016.** Microbiome shifts and the inhibition of quorum sensing by black band disease cyanobacteria. *The ISME Journal* **10**:1204–1216 DOI [10.1038/ismej.2015.184](https://doi.org/10.1038/ismej.2015.184).

- Miller AW, Richardson LL. 2011.** A meta-analysis of 16S rRNA gene clone libraries from the polymicrobial black band disease of corals. *FEMS Microbiology Ecology* 75:231–241 DOI [10.1111/j.1574-6941.2010.00991.x](https://doi.org/10.1111/j.1574-6941.2010.00991.x).
- Miller J, Muller E, Rogers C, Waara R, Atkinson A, Whelan KRT, Patterson M, Witcher B. 2009.** Coral disease following massive bleaching in 2005 causes 60% decline in coral cover on reefs in the US Virgin Islands. *Coral Reefs* 28:925–937 DOI [10.1007/s00338-009-0531-7](https://doi.org/10.1007/s00338-009-0531-7).
- Montano S, Strona G, Seveso D, Galli P. 2012.** First report of coral diseases in the Republic of Maldives. *Diseases of Aquatic Organisms* 101:159–165 DOI [10.3354/dao02515](https://doi.org/10.3354/dao02515).
- Montano S, Strona G, Seveso D, Galli P. 2013.** Prevalence, host range, and spatial distribution of black band disease in the Maldivian Archipelago. *Diseases of Aquatic Organisms* 105:65–74 DOI [10.3354/dao02608](https://doi.org/10.3354/dao02608).
- Muller EM, Leporacci NM, Macartney KJ, Shea AG, Crane RE, Hall ER, Ritchie KB. 2017.** Low pH reduces the virulence of black band disease on *Orbicella faveolata*. *PLOS ONE* 12:e0178869 DOI [10.1371/journal.pone.0178869](https://doi.org/10.1371/journal.pone.0178869).
- Muller EM, Van Woesik R. 2011.** Black-band disease dynamics: prevalence, incidence, and acclimatization to light. *Journal of Experimental Marine Biology and Ecology* 397:52–57 DOI [10.1016/j.jembe.2010.11.002](https://doi.org/10.1016/j.jembe.2010.11.002).
- Page C, Willis B. 2006.** Distribution, host range and large-scale spatial variability in black band disease prevalence on the Great Barrier Reef, Australia. *Diseases of Aquatic Organisms* 69:41–51 DOI [10.3354/dao069041](https://doi.org/10.3354/dao069041).
- Peters EC. 1993.** Diseases of other invertebrate phyla: porifera, cnidaria, ctenophora, annelida, echinodermata. In: Couch JA, Fourine JW, eds. *Pathology of marine and estuarine organisms*. Boca Raton: CRC Press. Waco: International Society for Reef Studies, 393–449.
- Porter JW, Dustan P, Jaap WC, Patterson KL, Kosmynin V, Meier OW, Patterson ME, Parsons M. 2001.** Patterns of spread of coral disease in the Florida keys. *Hydrobiologia* 460:1–24 DOI [10.1023/a:1013177617800](https://doi.org/10.1023/a:1013177617800).
- Precht WF, Gintert BE, Robbart ML, Fura R, Van Woesik R. 2016.** Unprecedented disease-related coral mortality in Southeastern Florida. *Scientific Reports* 6:31374 DOI [10.1038/srep31374](https://doi.org/10.1038/srep31374).
- Pruesse E, Quast C, Knittel K, Fuchs BM, Ludwig W, Peplies J, Glöckner FO. 2007.** SILVA: a comprehensive online resource for quality checked and aligned ribosomal RNA sequence data compatible with ARB. *Nucleic Acids Research* 35:7188–7196 DOI [10.1093/nar/gkm864](https://doi.org/10.1093/nar/gkm864).
- Randall CJ, Van Woesik R. 2015.** Contemporary white-band disease in Caribbean corals driven by climate change. *Nature Climate Change* 5:375–379 DOI [10.1038/nclimate2530](https://doi.org/10.1038/nclimate2530).
- Rasoulouniriana D, Siboni N, Ben-Dov E, Kramarsky-Winter E, Loya Y, Kushmaro A. 2009.** *Pseudoscillatoria coralii* gen. nov. sp. nov., a cyanobacterium associated with coral black band disease (BBD). *Diseases of Aquatic Organisms* 87:91–96 DOI [10.3354/dao02089](https://doi.org/10.3354/dao02089).

- Richardson LL. 1996.** Horizontal and vertical migration patterns of *Phormidium coralyticum* and *Beggiatoa* spp. associated with black-band disease of corals. *Microbial Ecology* **32**:323–335 DOI [10.1007/bf00183066](https://doi.org/10.1007/bf00183066).
- Richardson LL. 1998.** Coral diseases: what is really known? *Trends in Ecology & Evolution* **13**:438–443 DOI [10.1016/S0169-5347\(98\)01460-8](https://doi.org/10.1016/S0169-5347(98)01460-8).
- Richardson LL. 2004.** Black band disease. In: Rosenberg E, Loya Y, eds. *Coral health and disease*. Berlin: Springer, 325–336.
- Richardson LL, Kuta KG. 2003.** Ecological physiology of the black band disease cyanobacterium *Phormidium coralyticum*. *FEMS Microbiology Ecology* **43**:287–298 DOI [10.1016/S0168-6496\(03\)00025-4](https://doi.org/10.1016/S0168-6496(03)00025-4).
- Richardson LL, Kuta K, Schnell S, Carlton RG. 1997.** Ecology of the black band disease microbial consortium. In: Lessios HA, Macintyre IG, eds. *Proceedings of the 8th international coral reef symposium vol. 1. Smithsonian Tropical Research Institute, Panama*. Waco: International Society for Reef Studies, 597–600.
- Richardson LL, Miller AW, Broderick E, Kaczmarczyk L, Gantar M, Stanić D, Sekar R. 2009.** Sulfide, microcystin, and the etiology of black band disease. *Diseases of Aquatic Organisms* **87**:79–90 DOI [10.3354/dao02083](https://doi.org/10.3354/dao02083).
- Roder C, Arif C, Bayer T, Aranda M, Daniels C, Shibl A, Chavanich S, Voolstra CR. 2014.** Bacterial profiling of white plague disease in a comparative coral species framework. *The ISME Journal* **8**:31–39 DOI [10.1038/ismej.2013.127](https://doi.org/10.1038/ismej.2013.127).
- Roder C, Bayer T, Aranda M, Kruse M, Voolstra CR. 2015.** Microbiome structure of the fungid coral *Ctenactis echinata* aligns with environmental differences. *Molecular Ecology* **24**:3501–3511 DOI [10.1111/mec.13251](https://doi.org/10.1111/mec.13251).
- Rodríguez S, Cróquer A. 2008.** Dynamics of black band disease in a *Diploria strigosa* population subjected to annual upwelling on the northeastern coast of Venezuela. *Coral Reefs* **27**:381–388 DOI [10.1007/s00338-007-0341-8](https://doi.org/10.1007/s00338-007-0341-8).
- Rosenberg E, Ben-Haim Y. 2002.** Microbial diseases of corals and global warming. *Environmental Microbiology* **4**:318–326 DOI [10.1046/j.1462-2920.2002.00302.x](https://doi.org/10.1046/j.1462-2920.2002.00302.x).
- Rosenberg E, Falkovitz L. 2004.** The *Vibrio shiloi/oculina patagonica* model system of coral bleaching. *Annual Review of Microbiology* **58**:143–159 DOI [10.1146/annurev.micro.58.030603.123610](https://doi.org/10.1146/annurev.micro.58.030603.123610).
- Rosenberg E, Koren O. 2006.** Vibrios in coral health and disease. In: *The biology of vibrios*. Washington, D.C.: ASM Press.
- Rosenberg E, Loya Y. 2013.** *Coral health and disease*. Heidelberg: Springer-Verlag, Berlin.
- Rützler K, Santavy DL. 1983.** The black band disease of atlantic reef corals. *Marine Ecology* **4**:301–319 DOI [10.1111/j.1439-0485.1983.tb00116.x](https://doi.org/10.1111/j.1439-0485.1983.tb00116.x).
- Salter SJ, Cox MJ, Turek EM, Calus ST, Cookson WO, Moffatt MF, Turner P, Parkhill J, Loman NJ, Walker AW. 2014.** Reagent and laboratory contamination can critically impact sequence-based microbiome analyses. *BMC Biology* **12**:87 DOI [10.1186/s12915-014-0087-z](https://doi.org/10.1186/s12915-014-0087-z).
- Sato Y, Bourne DG, Willis BL. 2009.** Dynamics of seasonal outbreaks of black band disease in an assemblage of *Montipora* species at Pelorus Island (Great Barrier Reef,

- Australia). *Proceedings of the Royal Society B: Biological Sciences* **276**:2795–2803
DOI [10.1098/rspb.2009.0481](https://doi.org/10.1098/rspb.2009.0481).
- Sato Y, Willis BL, Bourne DG. 2010.** Successional changes in bacterial communities during the development of black band disease on the reef coral, *Montipora hispida*. *The ISME Journal* **4**:203–214 DOI [10.1038/ismej.2009.103](https://doi.org/10.1038/ismej.2009.103).
- Schloss PD, Westcott SL, Ryabin T, Hall JR, Hartmann M, Hollister EB, Lesniewski RA, Oakley BB, Parks DH, Robinson CJ, Sahl JW, Stres B, Thallinger GG, Van Horn DJ, Weber CF. 2009.** Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Applied and Environmental Microbiology* **75**:7537–7541 DOI [10.1128/aem.01541-09](https://doi.org/10.1128/aem.01541-09).
- Sekar R, Kaczmarek LT, Richardson LL. 2008.** Microbial community composition of black band disease on the coral host *Siderastrea siderea* from three regions of the wider Caribbean. *Marine Ecology Progress Series* **362**:85–98 DOI [10.3354/meps07496](https://doi.org/10.3354/meps07496).
- Sekar R, Mills DK, Remily ER, Voss JD, Richardson LL. 2006.** Microbial communities in the surface mucopolysaccharide layer and the black band microbial mat of black band-diseased *Siderastrea siderea*. *Applied and Environmental Microbiology* **72**:5963–5973 DOI [10.1128/AEM.00843-06](https://doi.org/10.1128/AEM.00843-06).
- Séré MG, Tortosa P, Chabanet P, Turquet J, Quod J-P, Schleyer MH. 2013.** Bacterial communities associated with Porites white patch syndrome (PWPS) on three Western Indian Ocean (WIO) coral reefs. *PLOS ONE* **8**:e83746 DOI [10.1371/journal.pone.0083746](https://doi.org/10.1371/journal.pone.0083746).
- Simpson EH. 1949.** Measurement of diversity. *Nature* **163**:688 DOI [10.1038/163688a0](https://doi.org/10.1038/163688a0).
- Suárez-Suárez A, López-López A, Tovar-Sánchez A, Yarza P, Orfila A, Terrados J, Arnds J, Marqués S, Niemann H, Schmitt-Kopplin P. 2011.** Response of sulfate-reducing bacteria to an artificial oil-spill in a coastal marine sediment. *Environmental Microbiology* **13**:1488–1499.
- Sunagawa S, DeSantis TZ, Piceno YM, Brodie EL, DeSalvo MK, Voolstra CR, Weil E, Andersen GL, Medina M. 2009.** Bacterial diversity and white plague disease-associated community changes in the Caribbean coral *Montastraea faveolata*. *ISME Journal* **3**:512–521 DOI [10.1038/ismej.2008.131](https://doi.org/10.1038/ismej.2008.131).
- Sussman M, Bourne DG, Willis BL. 2006.** A single cyanobacterial ribotype is associated with both red and black bands on diseased corals from Palau. *Diseases of Aquatic Organisms* **69**:111–118 DOI [10.3354/dao069111](https://doi.org/10.3354/dao069111).
- Sutherland KP, Porter JW, Torres C. 2004.** Disease and immunity in Caribbean and Indo-Pacific zooxanthellate corals. *Marine Ecology Progress Series* **266**:273–302 DOI [10.3354/meps266273](https://doi.org/10.3354/meps266273).
- Thompson JR, Randa MA, Marcelino LA, Tomita-Mitchell A, Lim E, Polz MF. 2004.** Diversity and dynamics of a North Atlantic coastal *Vibrio* community. *Applied and Environmental Microbiology* **70**:4103–4110 DOI [10.1128/AEM.70.7.4103-4110.2004](https://doi.org/10.1128/AEM.70.7.4103-4110.2004).
- Tout J, Siboni N, Messer LF, Garren M, Stocker R, Webster NS, Ralph PJ, Seymour JR. 2015.** Increased seawater temperature increases the abundance and alters

- the structure of natural *Vibrio* populations associated with the coral *Pocillopora damicornis*. *Frontiers in Microbiology* **6**:540–552 DOI [10.3389/fmicb.2015.00432](https://doi.org/10.3389/fmicb.2015.00432).
- Voss JD, Richardson LL. 2006.** Nutrient enrichment enhances black band disease progression in corals. *Coral Reefs* **25**:569–576 DOI [10.1007/s00338-006-0131-8](https://doi.org/10.1007/s00338-006-0131-8).
- Weil E. 2002.** Coral disease epizootiology: status and research needs. Coral health and disease: developing a national research plan. Coral Health and Disease Consortium, Charleston, South Carolina, 14.
- Weil E. 2004.** Coral reef diseases in the Wider Caribbean. In: Rosenberg E, Loya Y, eds. *Coral health and disease*. Berlin: Springer, 35–68.
- Weil E, Irikawa A, Casareto B, Suzuki Y. 2012.** Extended geographic distribution of several Indo-Pacific coral reef diseases. *Diseases of Aquatic Organisms* **98**:163–170 DOI [10.3354/dao02433](https://doi.org/10.3354/dao02433).
- Willis BL, Page CA, Dinsdale EA. 2004.** Coral disease on the great barrier reef. In: Rosenberg E, Loya Y, eds. *Coral health and disease*. Berlin: Springer, 69–104.
- Witt V, Wild C, Uthicke S. 2012.** Terrestrial runoff controls the bacterial community composition of biofilms along a water quality gradient in the great barrier reef. *Applied and Environmental Microbiology* **78**:7786–7791.
- Yasumoto-Hirose M, Nishijima M, Ngirchechol MK, Kanoh K, Shizuri Y, Miki W. 2006.** Isolation of marine bacteria by in situ culture on media-supplemented polyurethane foam. *Marine Biotechnology* **8**:227–237.
- Ziegler M, Roik A, Porter A, Zubier K, Mudarris MS, Ormond R, Voolstra CR. 2016.** Coral microbial community dynamics in response to anthropogenic impacts near a major city in the central Red Sea. *Marine Pollution Bulletin* **105**:629–640 DOI [10.1016/j.marpolbul.2015.12.045](https://doi.org/10.1016/j.marpolbul.2015.12.045).
- Zvuloni A, Artzy-Randrup Y, Stone L, Kramarsky-Winter E, Barkan R, Loya Y. 2009.** Spatio-temporal transmission patterns of black-band disease in a coral community. *PLOS ONE* **4**:e4993 DOI [10.1371/journal.pone.0004993](https://doi.org/10.1371/journal.pone.0004993).