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Bacterial Communities in Multiple Tissues Across the Body Surface of Three Coastal Shark Species

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Bacteria are known to have explicit roles within the microbiomes of host tissues, therefore examining these communities may prove useful in assessing host health and responses to environmental change. The present study contributes to the emerging, yet understudied, field of microbiome research in elasmobranchs. We provide a screening of the culturable bacteria communities found on multiple tissue sites on the body surface of blacktip (*Carcharhinus limbatus*), bull (*Carcharhinus leucas*), and tiger (*Galeocerdo cuvier*) sharks near Miami, Florida. Tissue sites include mouth, gills, skin, and any visible wounds. The study adds to our understanding of the diversity of bacteria present on sharks in comparison to their natural environment. We also compare bacterial groups found within wounds in shark skin to healthy tissue sites on the same individual. Results indicate that wounds on an individual may allow for opportunistic bacteria to invade or overgrow where they would not normally be found, which may have potential health consequences for sharks that become wounded due to fishing practices. Identified bacteria belonged to the *Actinobacteria*, *Firmicutes*, and *Proteobacteria* phyla, known to be prominent bacterial groups associated with marine organisms. Results indicate shark species-specific differences in bacterial communities, including the presence of bacteria belonging to *Planococcaceae* exclusively on the skin of tiger sharks. To our knowledge, this is the first report of this family in any elasmobranch. While most tissue sites displayed commensal bacteria identified in similar studies, known pathogens belonging to *Vibrionaceae* and *Staphylococcaceae* were identified in the wounds of blacktip and bull sharks. Some bacteria may be normal residents, but the loss of protective

dermal denticles due to a wound may allow colonization by pathogens. Continued research is needed to explore microbial communities associated with sharks and their influence on host health.

Key words: Bacteria, Microbiome, Shark, Elasmobranch, Skin, Wound.

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BACKGROUND

One emerging measure of health status in animals is the characterization of their microbiomes (Hollister et al. 2014). The microbiome has been most recently defined as a characteristic microbial community that occupies a well-defined habitat (Berg et al. 2020). Members of this community are typically prokaryotes (bacteria and archaea), eukaryotes (fungi, protists, algae), and viruses (Pogoreutz et al. 2019).

Communities of microorganisms are critical to the health of the host through involvement in host functions, such as nutrient supplementation, successful development, and disease susceptibility (Doane et al. 2017). Host-microbiome dynamics are described in two categories: first, as symbiosis, in which the organisms are involved in the normal physiological functions and metabolic interactions, and secondly as dysbiosis, in which the relationship or interactions are heavily altered, possibly related to a major stress or infection event (Apprill 2017). Bacteria are known to have explicit benefits to their host, therefore, examining bacterial communities across a microbiome may prove to be a useful tool in assessing host health (Apprill 2017). However, to measure these parameters, there must first be an effective means to characterize this microbiome.

While symbiotic relationships between marine animal hosts and microorganisms have been studied for decades, technological advancements have opened the door to our understanding of how these microorganisms are involved in host health (Apprill 2017). Previous research on corals, sponges, and teleosts has paved the way for methodology and demonstrated the ability to use the microbiome for health assessment in marine species (Ingram 1980; Thompson et al. 2015; McDevitt-Irwin et al. 2017). Bacterial abundance is now known to vary at the milliliter scale, and this variability rises in response to increases in the concentration of particulate organic matter in seawater (Long and Azam 2001). Therefore, research investigating bacterial communities within the microbiome of aquatic species has

begun to shed light on some of these host-microbiome dynamics. In coral species, different coral-associated bacteria are hypothesized to play varying roles in coral health, suggesting that coral reef microbial communities may serve as indicators of environmental stress and individual health (McDevitt-Irwin et al. 2017). In fact, many coral-associated bacteria defend their host by exuding antimicrobial compounds to prevent invasions from known *Vibrio* pathogens (Rypien et al. 2010). In aquaculture settings, studying fish microbiomes is important for assessing stock health and applying necessary treatments to disease (Olafsen 2001). It is well documented that bacteria present on the skin of teleosts include symbiotic microorganisms with antimicrobial properties (Ingram 1980).

In sharks, however, the role that microbiomes play in host health is poorly understood, and research on the topic is scarce. Mounting evidence from other marine groups indicates that a shift in the normal bacterial communities can leave their host vulnerable to disease or infection and that these shifts can be related to environmental conditions such as water quality, prey availability, and temperature change (Rosenberg et al. 2007; Ghanbari et al. 2015; Merrifield and Rodiles 2015). While outbreaks of bacterial disease are relatively uncommon in wild populations of elasmobranchs, they have been previously observed in captive settings. The most common *Vibrio* spp. isolated from captive sharks is *Vibrio carchariae*, which has been repeatedly implicated as the cause of meningitis in sand tiger (*Carcharias taurus*), lemon (*Negaprion brevirostris*) and sandbar (*Carcharhinus plumbeus*) sharks, as well as the spiny dogfish (*Squalus acanthias*) (Terrell 2014). Sharks are also commonly observed in the wild with wounds caused by encounters with other sharks or humans, yet rarely do these present as infected (Doane et al. 2017). A shark's skin may provide a habitat for host-associated bacteria that confer additional protections to their host against infection from outside pathogens, and these skin-associated microbes must be able to survive in the unique environment of shark skin. The composition and abundance of a host's microbiome varies through both space and time in response to ecological interactions between the host and environment (Van Opstal and Bordenstein 2015) and microbiomes have the potential to influence health, physiology, behavior and ecology of marine species. It is presumed that symbiotic microbial associations in various shark tissues may contribute to protective mechanisms against pathogens and disease in these animals, which might alter current understandings of how sharks adapt to anthropogenic and natural changes in their environment.

The purpose of this study was to provide a snapshot of culturable bacterial communities associated with the microbiome on the body surfaces of three coastal shark species sampled in the wild: the blacktip (*Carcharhinus limbatus*), bull (*Carcharhinus leucas*) and tiger shark (*Galeocerdo cuvier*). While this study only scratches the surface of what potential microbial communities may exist in different tissue sites of the three study species, the authors hope it will contribute to the minimal

literature on the topic and serve as a starting point for future studies to investigate the role microbiomes play in shark health.

MATERIALS AND METHODS

Sample collection

In total, 18 sharks were opportunistically and non-lethally sampled for this study: 5 blacktip sharks, 6 tiger sharks, and 7 bull sharks (Table S1). Sampling began October 2018 and ended in July 2019 off Miami, Florida (Fig. 1). Field sampling occurred with The University of Miami Shark Research and Conservation Program (SRC) during weekly sampling trips (as described in Tinari and Hammerschlag 2021). Sharks were captured using a circle-hook drumline system to minimize stress on the animal (described in Gallagher et al. 2014). To assess the microbiome, cell swabs were taken from the area between the lip and teeth (hereafter referred to as "mouth"), the inner side of the gill flaps (hereafter referred to as "gills"), the area of skin just below the dorsal fin laterally (hereafter referred to as "skin"), and lastly any area of visible injury (hereafter described as "wound"). Cell swabs were collected using 18 cm Falcon cell scrapers and gently rubbed against each tissue site for collection. The scraper was then placed into a 50 ml Falcon conical tube. Additionally, a 10 ml water sample was collected at the location where each shark was sampled to identify microbes present in the surrounding environment. All samples were kept on ice until transfer to the lab within 8 hours of collection.

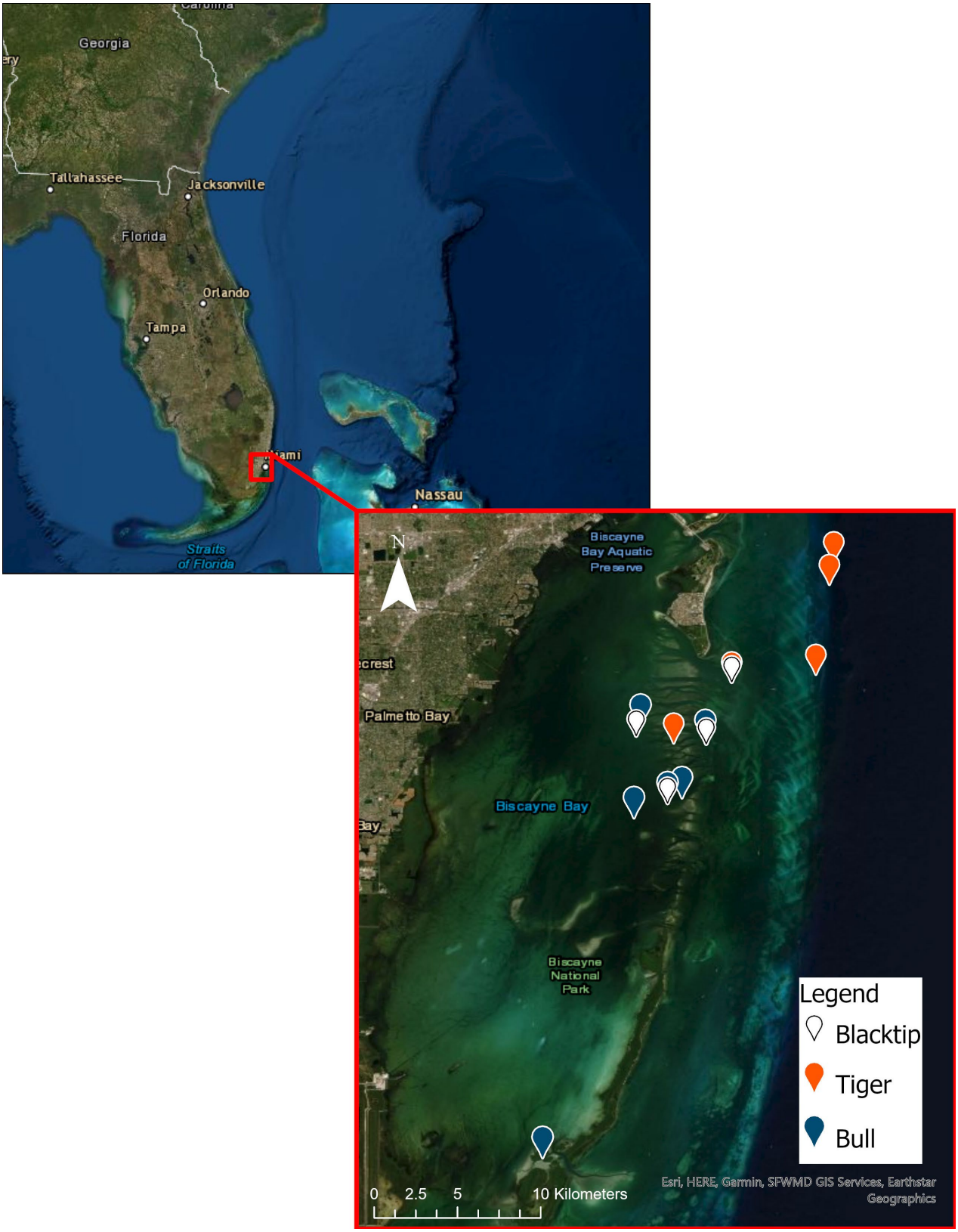

Fig. 1. Location of all sampled sharks within the study. Bull (blue), tiger (orange), and blacktip (white) sharks were all encountered off the coast of Miami, Florida, United States. Map created with ArcGIS Pro.

Sample Culture

Cell scrape samples were used to inoculate marine agar plates to culture bacteria (HiMedia Zobell Marine Agar, catalog number 95021-752). Two plates per sample were prepared and placed in an incubator at 28°C for 48 hours to allow colony growth. Each visually distinct colony (by color and texture) was further sub-cultured at 28°C for an additional 48 hours. Culture time and temperature reflect previous strategies for bacterial cultures (Lagier et al. 2015).

An inoculation loop was used to transfer bacterial colonies from the agar plate onto sterile water droplets placed on microscope slides. One microscope slide was prepared for each bacterial colony isolated and subsequently stained to identify gram-negative or gram-positive bacteria (Smith and Hussey 2005). The same process was repeated for the water samples using the inoculation loop to directly transfer one droplet of water to the agar plate.

For each shark species, samples from 2 individuals for which bacterial colonies were observed across all tissue sites underwent DNA extraction to determine which bacterial groups were present in this subset of 6 sharks. DNA extractions were performed on each bacterial colony sample using the protocol for the Qiagen QIAamp DNA Mini Kit (catalog number 51304). The subsequent DNA sample was tested for purity and concentration using a Thermo Scientific NanoDrop Spectrophotometer. Samples were then stored at -20°C until PCR amplification was performed. Forward primer 338f (5'-ACT CCT ACG GGA GGC AGC AG-3') and reverse primer 806r (5'-GGA CTA CHV GGG TWT CTA AT-3') were used to amplify 500bp of the V3-V4 region of the universal bacterial 16S rRNA gene. PCR protocol for the Advantage 2 PCR Kit by Clontech (category number 639206) was followed using the following cycle parameters: 94°C for 1 minute, followed by 30 cycles of 94°C for 1 minute, 30 seconds at 47°C and 30 seconds at 72°C.

The PCR product was run on 1% agarose gel containing SYBR-Safe DNA gel stain (Invitrogen, catalog number S33111) along with a wide range DNA ladder in the first well for reference. The gel was run at 130v for 40 minutes and was then visualized for bands under UV transillumination in a Gel Doc system. DNA samples that yielded bands of the anticipated product size were cleaned using the Qiagen QIAquick Gel Extraction Kit (category number 28704) before being sent to Eurofins Genomics USA for Sanger dideoxy sequencing. Sequencing results were run through the NCBI BLAST database for microbes. Search parameters for alignments were limited to an E value of $\leq$ 0.0 and $\geq$ 95%

identified to the genus level. These sequence data were archived in the GenBank database with accession numbers (Table S2).

Statistical Analyses

To assess bacterial diversity across the three shark species, Shannon's Index for Biodiversity (H) was run using the genera of bacteria identified through DNA sequencing (Table S3).

RESULTS

All sequenced samples identified to the genus level for the subset of sharks belonged to *Actinobacteria*, *Firmicutes*, and *Proteobacteria* phyla which are known to be associated with marine organisms (Tables 1, 2, 3).

Table 1. Distribution of bacteria genera for the phyla Actinobacteria for blacktip (Bl), bull (Bu) and tiger (Ti) sharks. "X" represents presence of bacteria

	Actinobacteria														
Genus	Mouth			Gills			Skin			Wound			Water		
	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti
Agrococcus				X											
Clavibacter				X											
Cryobacterium				X											
Curtobacterium				X											
Herbiconiux				X											
Leifsonia				X											
Mycetocola				X											
Zimmermannella				X											
Arthrobacter			X				X								
Kocuria			X				X								
Micrococcus			X				X								
Nesterenkonia			X				X								
Rothia			X				X								

Table 2. Distribution of bacteria genera for the phyla Firmicutes for blacktip (Bl), bull (Bu) and tiger (Ti) sharks. "X" represents presence of bacteria

	Firmicutes														
Genus	Mouth			Gills			Skin			Wound			Water		
	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti
Macrococcus										X					
Salinococcus						X									
Bacillus	X		X	X			X								
Exiguobacterium	X								X						
Fictibacillus				X											
Lysinibacillus									X						
Paenisporoarcina									X						
Planococcus									X						

Table 3. Distribution of bacteria genera for the phyla Proteobacteria for blacktip (Bl), bull (Bu) and tiger (Ti) sharks. "X" represents presence of bacteria

	Proteobacteria														
Genus	Mouth			Gills			Skin			Wound			Water		
	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti	Bl	Bu	Ti
Thalassospira				X										X	
Enterovibrio													X		
Vibrio										X	X		X		X
Photobacterium														X	
Luteimonas		X			X										
Stenotrophomonas		X			X			X							
Pseudoxanthomonas		X						X							
Xanthomonas		X			X			X							
Psychrobacter	X	X	X		X	X	X	X							
Pseudomonas														X	
Pseudoalteromonas			X			X			X	X				X	
Shewanella															X

Cultured samples of blacktip sharks belonged to 8 families (Fig. 2). Wounds on blacktip sharks were comprised of 3 families not present on the other tissue sites of the animal (*Staphylococcaceae*, *Vibrionaceae*, and *Pseudoalteromonadaceae*) (Figure 2). Bacterial DNA isolated from bull sharks

belonged to 3 families: *Vibrionaceae*, *Xanthomonadaceae* and *Moraxellaceae* (Fig. 2). The mouth, gills, and skin were nearly identical in bacterial composition
and distribution. However, the wounds only contained bacteria belonging to *Vibrionaceae*. Bacterial DNA isolated from tiger sharks belonged to 6 families: *Moraxellaceae*, *Pseudoalteromonadaceae*, *Bacillaceae*, *Pseudoalteromonadaceae*, *Micrococcaceae*, and *Staphylococcaceae*. The samples cultured from the mouth were the most diverse, showing four different families of bacteria (Fig. 2). The samples cultured from the gills shared the families *Moraxellaceae* and *Pseudoalteromonadaceae*, but *Staphylococcaceae* was unique to this tissue site. The samples cultured from the skin of tiger sharks harbored bacteria from only *Bacillaceae* and *Planococcaceae*. Bacillaceae was also present in the mouth, but the *Planococcaceae* family was unique to the skin.

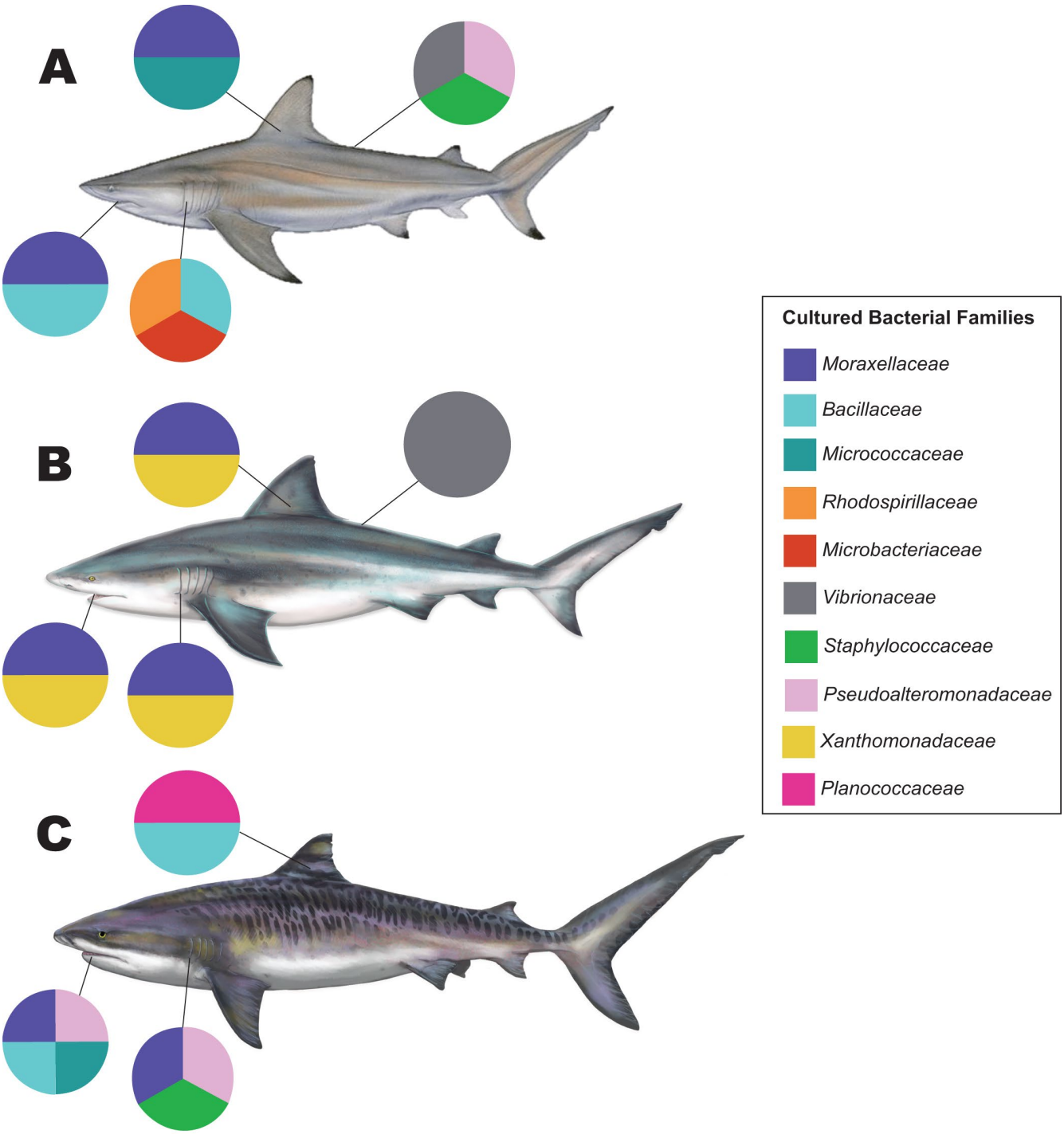

Fig. 2. Depiction of bacteria families found across tissue sites of each species: blacktip sharks (A), bull sharks (B), and tiger sharks (C). Pie charts are not proportional and only indicate presence of bacteria identified through DNA sequencing. (Blacktip shark animation from Wikimedia commons, bull and tiger animations by Kelly Quinn).

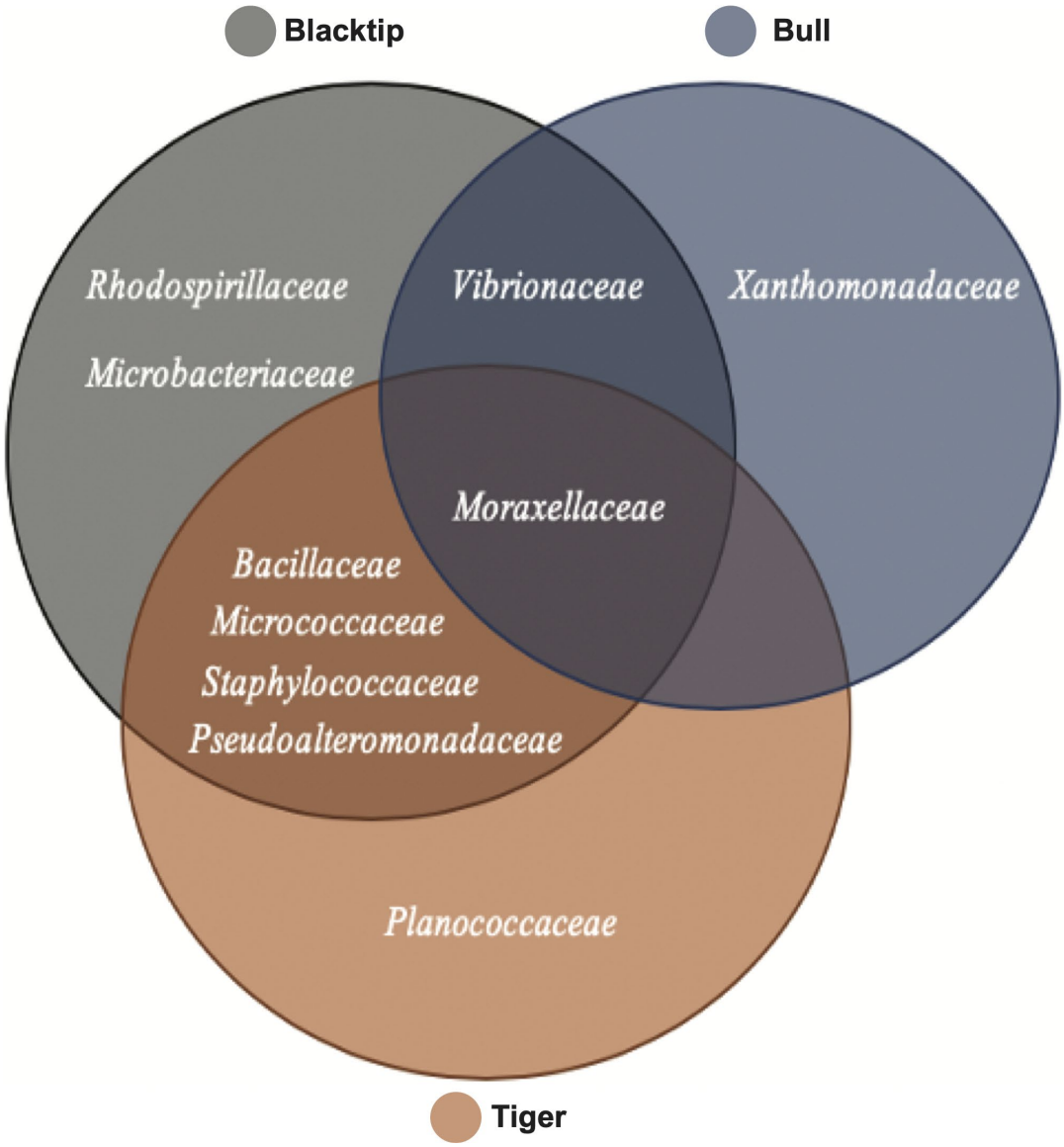

Fig. 3. Venn diagram showing the families of bacteria shared between sharks. Blacktip sharks are represented in gray, bull sharks are represented in blue, and tiger sharks are represented in orange.

Collectively, bacterial DNA isolated from water samples belonged to 5 families: *Shewanellaceae*, *Vibrionaceae*, *Pseudomonadaceae*, *Pseudoalteromonadaceae* and *Rhodospirillaceae*. *Shewanellaceae* and *Pseudomonadaceae* were not identified in samples from tissue sites on any shark species. The most frequently identified in the water samples, *Vibrionaceae*, was also identified in the wounds of both blacktip and bull sharks.

The present study provided a screening of culturable bacteria associated with multiple tissue sites from wild populations of blacktip, bull, and tiger sharks sampled off Miami, Florida. The shark species with the highest bacterial diversity was the blacktip shark (H = 2.95), followed by the tiger

shark (H = 2.47) and bull shark (H = 1.59). The surrounding water samples were less diverse (H = 1.83) than blacktip and tiger sharks but more diverse than the bull sharks. Blacktip and tiger sharks in this study shared the most bacterial families, even though they were found in different areas of the study site (Fig. 3). The similarities between the two may suggest these sharks encounter similar environmental conditions, but future research would be needed to explore this and compare their microbiomes.

DISCUSSION

Blacktip Sharks

Of the study species, blacktip sharks had the highest diversity of cultured bacteria. Results identified commensal bacteria within the healthy tissue sites that are known to produce compounds with antimicrobial, antifungal, and antibacterial activity. *Psychrobacter* was identified in the mouth and skin, and this genus has been previously identified on the skin of whales, bony fish, and blacktip reef sharks (*Carcharhinus melanopterus*) (Pogoreutz et al. 2019). In the skin of bony fishes, isolates of *Psychrobacter* have been shown to inhibit the growth of aquatic fungal pathogens (Lowrey et al. 2015). The presence of *Psychrobacter* in the intestinal tract of scalloped hammerhead sharks (*Sphyrna lewini*) has also been used as an indicator for pollution, as changes in the abundance of this bacteria correspond to environmental changes (Azevedo et al. 2013; Juste-Poinapen et al. 2015). Blacktip sharks are a highly migratory species that are exposed to a wide range of environmental conditions throughout the east coast and Florida (Keeney et al. 2005). Like other marine species, this commensal bacteria may be a good indicator for environmental changes that blacktip sharks experience. A diverse bacterial community may be important for this highly migratory species as they experience a wide range of habitat preferences.

While the healthy tissue sites of blacktip sharks cultured only commensal bacteria, the wounds possessed both commensal and pathogenic bacteria. *Pseudoalteromonas* was identified in the wounds of blacktip sharks and bacteria belonging to this group are known to play a variety of beneficial roles to their host, including producing antimicrobial compounds, aiding in prevention of biofouling, and inhibiting the biofilm of human pathogens (Holmström et al. 2002; Papa et al. 2015; Offret et al. 2016). *Pseudoalteromonas* has previously been identified as a core member of the microbiome of blacktip reef sharks and thresher sharks (Doane et al. 2017; Pogoreutz et al. 2019). In this study, *Pseudoalteromonas*

was only identified in the wounds of blacktip sharks rather than healthy tissue sites and raises the possibility that its presence in the wounds may serve some protective role in potentially competing with pathogenic bacteria present at these sites. *Pseudoalteromonas* species have been shown to actually kill *Vibrio* bacteria by digesting cell walls and the subsequent inactivation of pathogens (Richards et al. 2017). In aquaculture settings, the use of *Pseudoalteromonas* probiotics have reduced *Vibrio* growth and lowered the risk of pathogenic infections (Morya et al. 2014; Wang et al. 2018). Further supporting our theory is the fact that *Pseudoalteromonas* was not cultured from the water samples surrounding blacktip sharks, suggesting that this bacteria could be the host's natural response to outside pathogens.

The remaining bacteria identified in the wounds belong to known pathogenic genera *Vibrio* and *Macrococcus*. Although bacterial disease is considered relatively uncommon in elasmobranchs, *Vibrio* species have historically been recognized as the most significant group of marine pathogens (Bertone et al. 1996). This group of Gram-negative bacteria can cause high mortality rates in marine fishes that manifests as a hemorrhagic septicemia with extensive hemorrhaging and skin lesions (Thune et al. 1993). *Vibrio* was cultured from the wounds of blacktip sharks and did not present in any of the other tissue sites. Also identified within the wounds was *Macrococcus,* which has previously been associated with fish tumors by Vijayakumar Ramalingam et al. (2015). Because the sharks in this study did not appear to be negatively affected by these wounds, their core microbiome may be providing protection against these opportunistic pathogens, as demonstrated by the presence of commensal bacteria in the wound.

Bull Sharks

Like the blacktip sharks, *Psychrobacter* was also identified on the healthy tissue sites (mouth, gills, and skin) of bull sharks. This suggests that like blacktip sharks, *Psychrobacter* may be a normal resident of the bull shark microbiome. Another commensal bacteria identified in the mouth and skin of bull sharks, *Pseudoxanthomonas*, has the ability to degrade organic pollutants and mercury (Mahbub et al. 2016). Bull sharks are a cosmopolitan species that are often found near-shore and in estuarine and riverine waters (Bass et al. 1973; Compagno, 1984). They are one of the few elasmobranch species capable of moving into freshwater for extended periods of time and are a common inhabitant of Florida's coastal waters (Ortega et al. 2009; Hammerschlag et al. 2012). This wider range of habitat preferences may leave them susceptible to urbanized areas that have more pollutants in the water, and this commensal bacteria may play a role in degrading those pollutants. The sole genus identified in the wounds of the bull sharks was *Vibrio*, which was also identified in the wounds of blacktip sharks.

Unlike the wounds of blacktip sharks, no commensal bacteria were identified in the wounds of bull sharks. However, our culture technique targeted marine bacteria specifically, so it is possible that bacteria associated with euryhaline environments may have been missed and the reason that the bull shark microbiome was significantly less diverse than the other two study species.

Tiger Sharks

Planococcus was identified on the skin of tiger sharks and was not identified in the other two study species. This commensal bacteria includes species capable of producing antibiotic compounds that have also been isolated from marine sponges (Austin and Billaud 1990; Kaur et al. 2012). To the authors' knowledge, this is the first report of any member of the *Planococcaceae* family in an elasmobranch. Tiger sharks possess a highly productive mucoid layer in comparison to the other two study species (authors, direct observation), which potentially provides a habitat for diverse bacteria. *Planococcus* may be an important commensal bacteria that aids in the protection against outside pathogens unique to this species, due to this higher production of mucus. Another commensal bacteria, *Pseudoalteromonas*, was identified on all tissue sites of the tiger sharks and was previously only identified in the wounds of blacktip sharks of this study. Future studies should explore the bacterial communities found within tissue sites of tiger sharks to better understand the role these commensal bacteria play in their microbiome.

Water

Bacteria identified in water samples that were also present on shark tissues were members of the *Vibrionaceae* and *Pseudoalteromonadaceae* families. *Vibrionaceae* was previously identified in the wounds of blacktip and bull sharks. It is possible that the normal microbiome or structure of the epithelia of sharks would normally prevent the intrusion of this pathogen, so the loss of integrity at these compromised areas allowed *Vibrio* from the surrounding water to transfer to the sharks. *Pseudoalteromonadaceae* was also identified in the mouth and gills of tiger sharks, but within blacktip sharks this bacteria only presented itself within the wounds. This finding suggests that members of *Vibrionaceae* may not be normal inhabitants of the microbiome of blacktip and bull sharks, but opportunistic invaders from the surrounding water that have entered through a compromised area. Of the subset of sequenced DNA cultured from sharks in this study, pathogenic bacteria were only identified from the wound and water samples. While pathogenic bacteria were identified in the water

surrounding the tiger sharks, this species did not culture pathogenic bacteria on any of their tissue sites (Table 4).

Table 4. Presence (1) and absence (0) of pathogenic bacteria identified from cultured bacteria through DNA sequencing

Shark ID	Species	Mouth	Gills	Skin	Wound	Water
N392645	Blacktip	0	0	0	1	0
N392649	Blacktip	0	0	0	1	1
N389751	Bull	0	0	0	1	1
N389792	Bull	0	0	0	N/A	1
N393575	Tiger	0	0	0	N/A	1
N393619	Tiger	0	0	0	N/A	1

CONCLUSIONS

Sharks have evolved mechanisms to aid in protection against a wide variety of pathogens, including the production of mucus supplemented with antimicrobial properties (Magnadóttir 2006). In addition to this mucus layer, sharks also possess thick dermal denticles to protect them from injury; coupled together, these mechanisms may provide sharks with a strong protection against outside pathogens or infection. Our findings suggest that the loss of integrity of the normal epithelial structure from a wound likely allows pathogenic bacteria in the surrounding water, such as *Vibrio* and members of *Staphylococcaceae*, to make an opportunistic home in these wounds. In addition, the presence of certain commensal bacteria in healthy tissue sites, such as *Psychrobacter*, may be used as indicators of pollutants or other anthropogenic stressors in the surrounding environment as in previous studies (Azevedo et al. 2013; Juste-Poinapen et al. 2015). The habitat preferences and migratory patterns of blacktip, bull, and tiger sharks likely contribute to their core microbiomes. Changes in bacterial communities may be a response to changes in the immediate environment or a response to a wound, and future studies can use these deviations to assess shark health on both an individual and population level.

The microbiome of sharks, and more specifically the comparison of bacterial communities within a wound and healthy tissue sites on the same individual, is severely understudied. This study provides only a snapshot of culturable bacteria found within the microbiome of blacktip, bull, and tiger

sharks in the study area at the time of encounter. We recognize that our study is limited in its conclusions due to our culture methods and small sample size, but our study is building a foundation for future studies to explore the microbiomes of sharks and how they may influence host health. Because each individual was only encountered once and within a specified study location, we cannot definitively conclude what bacteria are residential versus invasive across time or location. Microbiomes change across environments, as most notably demonstrated in the Atlantic salmon (*Salmo salar*) microbiome during migrations from river to ocean habitats (Lokesh and Kiron 2016). Additionally, time and temperature of culturing methods controlled what bacteria cultured. While time and temperature were optimized to include as many marine bacteria species as possible, it is likely that other bacteria were present but did not grow well under our specified culture conditions. For example, freshwater bacteria that may have been within the microbiome of bull sharks could have been missed. Future studies should expand sample size and location and sampling the same individuals multiple times could provide a complete picture of the microbiome.

We hope research in this area will continue to provide insight into the role bacteria play in shark health, and our study may serve as a starting point for future work. Establishing the core microbiome for shark species will be important for future work to differentiate between commensal bacteria and opportunistic pathogens, and to understand how the microbiome may shift in response to environmental changes or injury through habitat use or anthropogenic stressors.

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Competing interests: CB LM and NH declare no competing interests.

Availability of data and materials: All bacteria DNA sequences in this study are available in the NCBI GenBank under the accession numbers MW172981-MW173019 (also attached as supporting material).

Consent for publication: All authors have reviewed the final manuscript and give consent for publication and declare that the manuscript has not been sent to other journals for consideration at this time.

Ethics approval consent to participate: Not applicable.

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Supplementary materials

Table S1. Information for Study Sharks. (download)

Table S2. DNA Sequence Accession Numbers. (download)

Table S3. Statistics for Shannon's Index Calculations. (download)