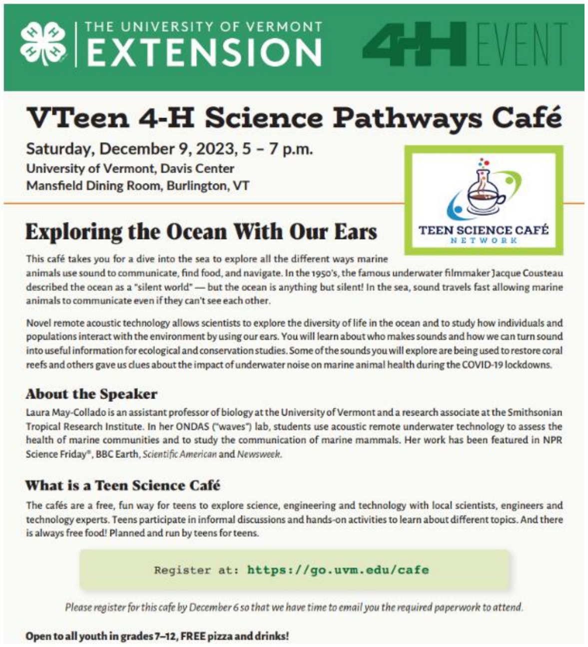
COURSE-BASED UNDERGRADUATE RESEARCH EXPERIENCE: SOUNDSCAPES AND BEHAVIOR RESEARCH
(Biol 2100 A)

Fall 2023

Course Instructor Laura J May-Collado, Ph.D.
Meeting Time: Tuesday 1:15-4:15 p.m. in 217A Marsh Life Science Bldg Website:
http://www.lauramay-collado.com/cure-lab.html


THE UNIVERSITY OF VERMONT EXTENSION 4-H EVENT

VTeen 4-H Science Pathways Café

Saturday, December 9, 2023, 5 – 7 p.m.
University of Vermont, Davis Center
Mansfield Dining Room, Burlington, VT

TEEN SCIENCE CAFÉ NETWORK

Exploring the Ocean With Our Ears

This café takes you for a dive into the sea to explore all the different ways marine animals use sound to communicate, find food, and navigate. In the 1950's, the famous underwater filmmaker Jacque Cousteau described the ocean as a "silent world" — but the ocean is anything but silent! In the sea, sound travels fast allowing marine animals to communicate even if they can't see each other.

Novel remote acoustic technology allows scientists to explore the diversity of life in the ocean and to study how individuals and populations interact with the environment by using our ears. You will learn about who makes sounds and how we can turn sound into useful information for ecological and conservation studies. Some of the sounds you will explore are being used to restore coral reefs and others gave us clues about the impact of underwater noise on marine animal health during the COVID-19 lockdowns.

About the Speaker

Laura May-Collado is an assistant professor of biology at the University of Vermont and a research associate at the Smithsonian Tropical Research Institute. In her ONDAS ("waves") lab, students use acoustic remote underwater technology to assess the health of marine communities and to study the communication of marine mammals. Her work has been featured in NPR Science Friday®, BBC Earth, *Scientific American* and *Newsweek*.

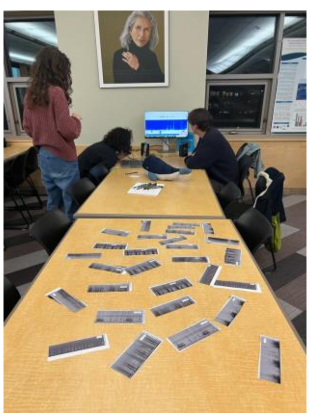
What is a Teen Science Café

The cafés are a free, fun way for teens to explore science, engineering and technology with local scientists, engineers and technology experts. Teens participate in informal discussions and hands-on activities to learn about different topics. And there is always free food! Planned and run by teens for teens.

Register at: **https://go.uvm.edu/cafe**

Please register for this cafe by December 6 so that we have time to email you the required paperwork to attend.

Open to all youth in grades 7–12, FREE pizza and drinks!



CURE Symposium: Soundscapes and Behavior

December 5 @ 1:15, MLS 224

(cookies and coffee will be available)

Time	Speaker
	Soundscapes
1:30	**Emma Wetsel**. *A characterization of the soundscape of two sites at Caño Island, Costa Rica that vary in coral reef bleaching levels*
1:45	**Katie Dimmick**. *Passive acoustic monitoring of sonorous fish at coral reef habitats off the Santa Elena Bay, Costa Rica.*
2:00	**Nichole Hardy**. *Acoustic temporal patterns and snapping shrimp level of presence in different coral reef environments, Gulf of Papagayo, Costa Rica*
2:15	**Ella Dearden**. *A characterization of the temporal changes in ambient noise levels of the Cuajiniquil Bay, Costa Rica*
	Marine Mammals
2:30	**Gabriel Falcione**. *Singing activity of humpback whales within and outside protected areas in the Gulf of Chiriquí, Panama*
2:45	**Caitlyn Archambault**. *Using unmanned aerial vehicles to estimate behavioral budgets of bottlenose dolphins in the Archipelago of Bocas del Toro, Panama*
3:00	**Ariana Casella**. *Dolphin communication before, during and after the COVID-19 Lockdown*
3:15	**Fiona Duckworth**. *Singing activity of Central American humpback whales at Cuajiniquil Bay, Costa Rica.*
3:30	**Franny Oppenheimer**. *A description of the unit repertoire Southeastern Pacific humpback whales breeding in the Gulf of Chiriquí, Panama*

A characterization of the soundscape of two sites at Caño Island, Costa Rica that vary in coral reef bleaching levels

Emma Wetsel

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Abstract: Coral reefs are critical ecosystems of global importance and are currently facing extreme and unprecedented threats from coral bleaching events due to climate change. This study investigates the impact of coral bleaching on the acoustic environment of coral reefs, focusing on two locations in the Caño Island Biological Reserve, Costa Rica: El Diablo (2017 and 2023) and La Catarata (2023). Utilizing underwater acoustic technology, we present a comprehensive analysis of variations in reef health. The findings show that there were significant differences in Acoustic Complexity Indexes observed between Catarata 2023, Diablo 2023, and Diablo 2017, suggesting varying levels of health across sites. This research underscores the potential of acoustic data as a valuable and non-invasive tool for monitoring coral reef health by unraveling the intricate dynamics of coral reef soundscapes. The study emphasizes that there is a need for subsequent research in this field to use the full potential of acoustic monitoring systems, as they could be used as valuable tools for both understanding and preserving coral reef ecosystems.

Key Words: Acoustic complexity index, acoustic events, acoustic morphotypes, Arbimon, autonomous underwater recorders

1. Introduction

Coral reefs are undergoing extreme bleaching events across the world, leading to rapid decline of coral populations and of those organisms that depend on them. Traditional visual surveys of coral reefs biodiversity are often costly and limited in time and space (Hochberg and Gierach, 2021). However, in the past two decades underwater acoustic technology and novel computational tools have allowed more rapid evaluation of coral reef biodiversity (Lin et al., 2021). In addition, the low cost associated to underwater technology facilitates to study variation of coral reef biodiversity at various temporal and spatial scales (Freeman, 2016). Recent studies have shown that healthy coral reefs systems are also acoustically noisy and diverse, and thus, noise can serve as an excellent indicator of coral reef health. For example, several studies have found that diverse soundscapes correlated with high coral cover and fish diversity (Lin et al., 2021). In addition, Piercy et al. (2014) suggested that healthier reefs produce louder and richer acoustic events than reefs experiencing bleaching (Piercy et al., 2014). Given that noise from healthy coral reefs systems is associated with recruitment of pelagic fish and invertebrate larvae understanding the extent at which of bleaching may impact the local soundscapes is key to understand coral reef community dynamics (Gordon et al., 2018).

In Costa Rica, the Biological Reserve of Caño Island, is home to a coral reef community primarily composed of crustose coralline algae and *Porites lobata* (Guzmán and Cortés, 1989). The corals around the Island have historically been impacted by large bleaching events, most notably during the El Niño event of 1982-83 (Guzmán and Cortés, 2001). However, increased numbers of sexual recruits and coral cover of around 70% in the early 2000's suggest reefs at Caño Island are recovering and may been relatively tolerant of heat stress (Guzmán and Cortés, 2001). Recent studies have found that this coral reef community sustains one of the richest fish communities in the Tropical Eastern Pacific. Salas et al., (2016) reported a total of 79 fish species from 32 families to be present at the site, with the most abundant family being Pomacentridae (Salas et al., 2016). The authors also found that Caño's fish community consist of primarily planktivorous and carnivorous species, although herbivorous grazer species are also present. Fish species richness and composition was found to be correlated with coral cover (Salas et al., 2016). Finally, the authors indicate that the total number of fish species recorded at Caño represents approximately 17% of the costal fish diversity in the Tropical Eastern Pacific, highlighting the importance of this protected area.

The purpose of this study is to examine if coral bleaching and associated fish and invertebrate community explains soundscape temporal and spatial variation. Specifically, I will study the soundscape of two locations within the reserve that vary in their degree of coral bleaching: El Diablo and La Catarata. Based on observations by collaborators this summer, Diablo and Catarata were experiencing bleaching at different scales, with Catarata showing the highest amount of coral bleached. Data from previous years at El Diablo will allow us to understand changes in the soundscape through time. I expect that sites with bleached corals will have lower acoustic complexity, events, and presence of acoustic morphotypes, driven by a decrease in the presence of acoustic species such as fish and invertebrates.

The broad significance of this proposed research is a potentially more cost-effective, less invasive, and more viable long-term method to study reef health. Acoustic data, along with other forms of remote sensing, offer a way to quickly collect vast amounts of data on environmental parameters, species composition, and behavior (Mandal and Ghosh, 2023). Acoustic data has been used historically to identify and catalog fish species based on their unique acoustic signatures, which can then be used to monitor migration and population sizes (Mandal and Ghosh, 2023). The analysis of soundscapes of coral reefs can offer a way to estimate holistic ecosystem health through the different indices mentioned above (Mandal and Ghosh, 2023) (Pieretti and Morri, 2011) (Aide et al., 2017). If a correlation between reef health and acoustic indicators is found, acoustic recordings could be used to detect poor quality reefs that should be prioritized for conservation and management efforts (Mandal and Ghosh, 2023). Furthermore, as coral reef acoustics are a potential factor of reef population dynamics, studying the acoustic indicators of reefs of varying health can advance knowledge on how reef soundscapes react to bleaching events. Implications of research include a more complete understanding of soundscape ecology within coral reef environments before, during, and after degradation occurs (Mandal and Ghosh, 2023).

2. Materials and Methods

2.1. Study Site

The study sites are located within the Caño Island Biological Reserve. Caño Island is located 0.8 km from the mainland on the Pacific Ocean side of Costa Rica (8.719N / -83.863W). El Diablo (8.70 N/-83.915W) is located on the north side of the island 2.15km offshore. La Catarata (8.710 N/-83.915W) is located on the west side of the island. Recorders were placed at both sites at a depth of 25m. The sites are primarily composed of crustose coralline algae and *Porites lobata* (Guzmán and Cortés, 1989). Many of the coral colonies present form microatolls—circular rings of living coral around a dead section of coral at the center of the colony (Guzmán and Cortés, 1989). The upper reef slope to the reef base is composed of small colonies of several coral species including coral of the *Pavona* and *Pocillopora* genus despite *Porites lobata* being predominant (Guzmán and Cortés, 1989). Reef surveys from 1998 suggest coral cover of around 70% (Guzmán and Cortés, 2001).

2.2. Recordings

Recordings of the soundscape were made using autonomous underwater recorders at a sampling rate of 48 kHz. Recordings from El Diablo in 2017 (1 site) were made using the model RUDAR-mk (sampling rate up to 96 kHz -169 dB re:1V/uPa) from Cetacean Research Technology (Cetacean Research Technology, 2023) and from El Diablo (2 sites) and La Catarata (3 sites) in 2023 were made using the model Hydromoth from Open Acoustic Devices (Open Acoustic Devices, 2023). For comparison purposes only recordings 48 hours June in 2017- and 48-hours August in 2023 will be used for this study.

2.3. Soundscape analysis

To study the soundscape, a 1-minute sample was manually taken every five minutes from each site using the program Audacity (n=6). These files will be uploaded to ARBIMON II (Sieve Analytics, 2015) for analysis using the soundscape tools to calculate the ACI and the proportion of acoustic events. Soundscapes were created for each site overall and each individual hydromoth recording. The soundscape will be measured using three metrics: the Acoustic Complexity Index (ACI) (Pieretti and Morri, 2011) and detection of acoustic events (Aide et al., 2017), The ACI is an algorithm that provides a fast and direct quantification of biological sounds based on their intensity and will be used here to determine the variability of intensities in the audio-recordings throughout the day, sites, and years. The acoustic events data will reveal what frequency bins were more impacted by bleaching.

2.4. Acoustic Morph analysis

Using the 1-minute file selection described above, files will be opened in RAVEN 1.5 (2016; Cornell Lab of Ornithology) and inspected using a spectrogram with a fast Fourier transform size of 4048 points, an overlap of 50%, and a 1024-sample Hann window to identify different types of acoustic events and annotate a potential source: fish, snapping shrimp, whale, dolphin, or boat. This data will be used to determine changes in presence of sound course by site.

3. Results

3.1 Soundscape Indexes

There was a significant difference in the ACI between Catarata 2023, Diablo 2023, and Diablo 2017 (df = 2, p = <0.0001, Fig.1). Diablo 2023 had the highest observed mean ACI and the highest ACI range of all three hydromoths. Catarata 2023 was the second highest, with a mean difference of 12.13 from Diablo 2023 (Fig.2). Both Catarata 2023 and Diablo 2023 showed peaks in ACI values between the hours of 8am to 11am, with a steady decline in the following hours. The calculated ACI values for Diablo 2017 were significantly lower than the values for 2023 and showed a peak later in the day between 11am and 3pm (Fig.1). When looking at acoustic events by frequency band, I found that in both locations Diablo and Catarata 2023, there was a higher proportion of detection of acoustic events occurred between 12am and 10am. However, there appears to be more frequency bands used in Catarata than in el Diablo (Fig.3).

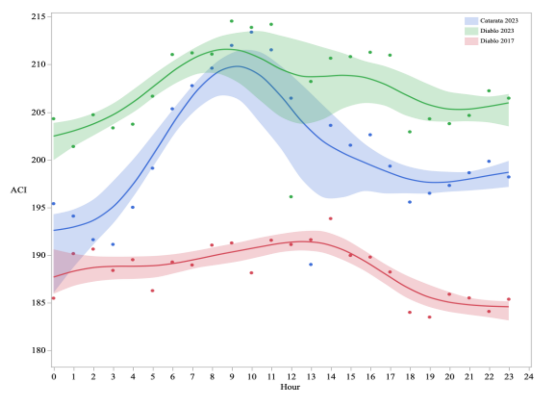


Figure 1. Variation of the Acoustic Complexity Index by time of day and location.

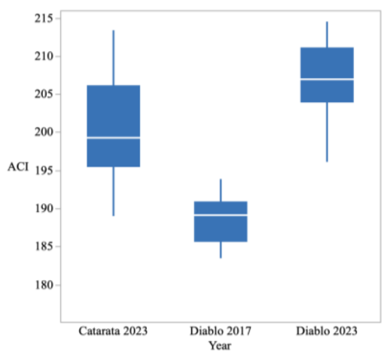
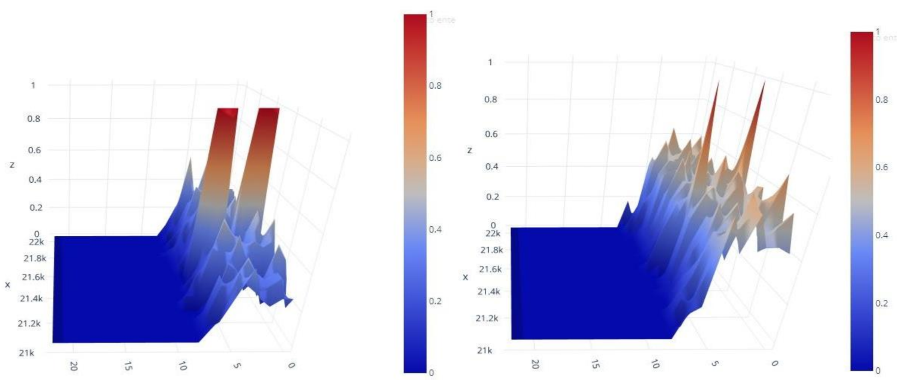


Figure 2. Mean ACI by location and year. The median ACI value is represented by the white line, the interquartile interval is represented by the blue box, and the minimum and maximum values are represented by the vertical bars



Diablo 2023 Catarata 2023

Figure 3. Proportion of detection of acoustic events by time of day (hour) and by frequency bin (kHz).

3.2 Soundscape Composition

There were significant differences in the contribution of sound sources between Catarata and Diablo 2023 between the observed peak hours of 8am and 11am (Fig.4). The proportion of acoustic files with humpback whale songs was significantly higher in Catarata than in Diablo (X^2=102.4, df=4, p<0.0001). The proportion of boats was significantly higher in Diablo than Catarata (R^2 = 0.062, df =4, p = 0.022). In contrast, the overall proportion of unknown sound sources did not vary significantly between sites (p>0.05, Fig.4).

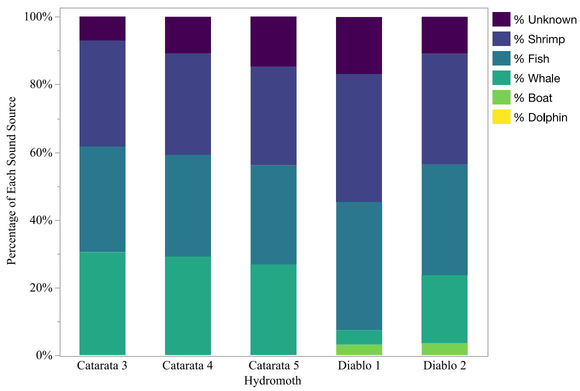


Figure 4. Percentage of Each Sound Source Across All Sites Between 8am - 11am.

When measuring the contribution of sound sources over a 24-hour period (Hydromoth 1 at Diablo 2023 and Hydromoth 2 at Catarata 2023) differences between sites were found (Fig. 5). When a contingency analysis was preformed, boats were detected significantly more at Diablo than Catarata (X^2=4.142, df =1, p = 0.0418) and the proportion of humpback whale songs was significantly higher at Catarata than Diablo (X^2=178.08, df =1, p <0.0001). There were not significant differences between the two sites for fish acoustic detections or unknown sound sources (p>0.05, Fig.5). There no variation within the site in the proportion of detections of boats, whales, fish, and snapping shrimp (p>0.05, Fig.6) in Catarata.

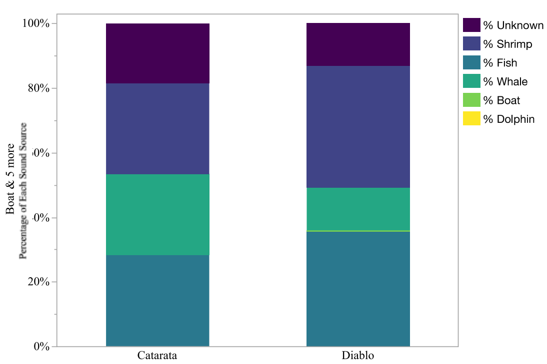


Figure 5. Percent of Each Sound Source Over 24-Hour Period at Catarata and Diablo.

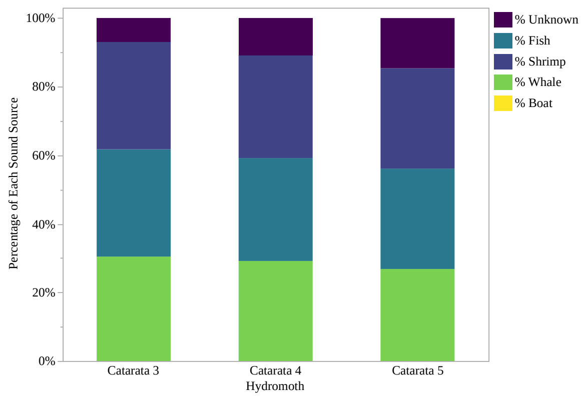


Figure 6. Percentage of Each Sound Source Across the Three Catarata Hydromoths.

4. Discussion

This study finds that Catarata and Diablo vary in two aspects of their soundscape: the variation of sound intensities measured by the ACI and the contribution of sound sources to the overall soundscape. These results suggest the two locations vary in their structure and species composition, and in the case of El Diablo that there is evidence of temporal variation as well. However, previous studies using the ACI have raised concerns that there is not as clear of a relationship between marine biodiversity and terrestrial acoustic indices (Mooney et al., 2020). Kaplan et al. (2015) suggest that ACI metrics are greatly affected by the presence of snapping shrimp, as loud, constant, and frequent sounds can bias the indices produced. Furthermore, McWilliam and Hawkins (2013) suggest that when using ACI in environments dominated by snapping shrimp—as Catarata and Diablo both were—differences between sites may be due to snapping shrimp patterns instead of overall community presence and diversity. While the significant variations between ACI values at Catarata and Diablo could reflect acoustic complexity trends within the distinctive soundscapes of the two sites, these indices do not definitively state what the source of these variations may be nor how the indices may be biased.

While I could not assess the level of bleaching in these two habitats, the differences in soundscape suggest that bleaching might be one of the factors explaining the higher ACI values in Diablo, which reported to be impacted to a lesser degree by bleaching than Catarata. Previous work suggests ACI may not be the best acoustic tool to measure coral health (Kaplan et al., 2018). A study by Bertucci et al. (2016) suggested significantly higher ACI values between two sites despite no difference in visual surveys, indicating that the calculated ACI values were not necessarily reflective of species assemblage (Bertucci et al., 2016).

As there was no statistical difference in the percentage of sound sources recorded across the three Catarata sites in 2023 aside from unknown sound sources, it cannot be concluded that varying levels of bleaching across the three sites impact the soundscape composition of a section of reef. Furthermore, previous studies suggest soundscapes can vary between recorders only a few meters apart largely due to areas of preference—or hotspots—within reef communities (Kaplan et al., 2015). Although the range in variability between locations within a habitat seem to be smaller than the range of variability between different habitats, intra-habitat acoustic variability has not been well-studied (Mooney et al., 2020). Therefore, conclusions on differences in ACI values within a reef due to coral bleaching alone may be misleading.

We also observed temporal variation in ACI in Diablo by comparing data from 2017 and 2023, suggesting changes in the marine community across this 6-year period. However, it is important to note that the comparison was done using different months and different recording equipment. The equipment in 2017 had a filter for frequencies from 0-300 Hz, which is the range in which many fish noises are produced. The 2017 data was collected June 2nd-3rd, and the 2023 data was collected August 18th-19th. Mooney et al. (2020) suggest that changes in acoustic composition can change temporally because of season, lunar periodicity, and time of day. Staaterman et al. (2014) concluded that variability in low frequencies is often driven by lunar cycles, implying that variation in lunar cycles between the 2017 and 2023 Diablo data could have impacted the resulting ACI's. Temporal changes between 2017 and 2023 at Diablo cannot be strongly concluded because of the difference in data collection dates.

Regarding soundscape composition, we found that Diablo had a higher proportion of boats and unknown sounds. This could be due to large quantities of tourists coming from the mainland towards Diablo for snorkeling. Additionally, Diablo is on the same side of the island as the station all tour boats must stop at, which could explain the increase in boat activity. There were significantly more humpback whale songs recorded at Catarata than Diablo, which is not in accordance with the ACI values. Several studies have found that in marine communities with more boat noise produce decreased soundscape complexity and variability, particularly in areas with high tourism boating traffic (Bittencourt et al., 2020). At Caño Island previous studies have highlighted the potential effects of boat presence on whale song events during popular boating hours before, during, and after the COVID-19 Pandemic lockdown (May-Collado et al., 2023). This research aligns with trends found during this study that high boat traffic may decrease humpback whale song detection.

A periodic loud humming present in the Catarata data resembled acoustic events like trawling or other boat activities. Previous studies indicate that, despite originally designed to minimize anthropogenic noise source impacts, the ACI can be impacted by intense sounds that are non-repetitive or consistent (Pieretti et al., 2011). Because it is unknown whether or not the Soundscape Composition analysis for ACI determined that this was an anthropogenic source or not, consideration should be taken for conclusions drawn on Catarata hydromoths for the entire

24 hour period. Furthermore, it should be taken into consideration that these noise sources were categorized as “unknown”, not “boats”, which could impact the statistical analysis of the sites.

Additional days of data at the two sites are needed to strengthen any conclusions drawn regarding significant differences between acoustic complexity, morphotype presence, and effects of coral bleaching across sites at Caño Island. Potential sources of error include Arbimon failing to accurately pick up and exclude anthropogenic noise sources from their ACI calculations, human error using Audacity and clipping the 1-minute files, and inconsistent analysis or categorization of morphotypes, particularly within the “unknown source” category. Future studies may consider using low-frequency recordings when assessing species composition, as Kaplan et al. (2015) suggest these lower frequency recordings between 100 to 1000 Hz were correlated with coral cover and fish density when higher frequencies produced by snapping shrimp were not. (Kaplan et al., 2015).

Conclusion

Coral reefs play a critical role in marine community health due to the large quantity of species directly dependent on reef ecosystems. Although more research is needed to draw firm conclusions on the effect of coral bleaching levels on soundscape characterization, this research reaffirms the utility of acoustic data to study reef health. Acoustic tools present a more cost-effective, time-efficient, and minimally invasive method for the rapid collection of large quantities of reef data. Additionally, soundscape analysis of coral reefs can offer a more holistic quantification of ecosystem health through varying indices. Through acoustic data collection, threatened sections of reef can be highlighted and then prioritized for conservation and management. Acoustic data, when paired with other forms of data, can help us better understand the ecological processes occurring on coral reefs, contrast these processes when reef health is threatened, and advise conservation groups and local governments on the highest priority sites to conserve. When acoustic tools are included in management decisions, a more educated and cost-effective conservation plan can be created. Through advancing technologies, we have the power to better understand what is happening beneath the ocean’s surface and how we might best protect it in the face of climate change.

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Passive acoustic monitoring of nocturnal fish off the Santa Elena Bay, Costa Rica

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University of Vermont

Abstract: Soundscapes are collections of sounds from physical, biological, and human sources. In coastal environments fish is a major contributor to the overall soundscape. This study explores the diversity of nocturnal fish sounds in the Santa Elena Bay in Costa Rica, in two locations with healthy coral reef communities. The results show that a collection of fish sounds including gulps, taps, barks, and moans. There were differences in the collection of sounds between sites, likely reflecting different fish species. This study shows the potential for passive acoustic monitoring. Future studies should associate sounds with fish species for a better use of sound in the study of coral reef health, and community composition and dynamics.

Keywords: soundscape, coral reef fish, animal communication, bioacoustics.

1. Introduction

Soundscapes are collections of sounds from physical, biological, and human sources. The contribution of each of these sources to the overall soundscape can provide important information about the health of an ecosystem. For example, several studies have found that noise associated to human activities (i.e. boat traffic) can interfere with the communication of reef fish communication, impact larvae settlement, and affect larvae development Simpson et al. 2005, 2016a,b, Slabbekoorn et al. 2010, Holles et al. 2013). Anthropomorphic noise pollution has been found to negatively affect fish health. According to a research paper published in 2013 regarding fish reaction to anthropomorphic sound, boat noise disrupts the crucial process of orientation cuing and habitat selection for fish, which increases energetic costs and predator risk (Holles et al. 2013).

There is a variety of fish species present on the Costa Rican coastline, specifically in Murciélago Islands and Santa Elena Peninsula of the Guanacaste Conservation Area. According to data presented in a 2021 research analysis paper for reef environments, there were 84 reef fish species identified (Alvarado et al., 2021). Some fish can produce sound. Normally, there are three main ways sounds are created by fish including the use of sonic muscles that are located on or near their swim bladder (drumming); striking or rubbing together skeletal components (stridulation);

and by quickly changing speed and direction while swimming (hydrodynamics). Unintentional sounds from fish are generated during swimming and feeding all the time, whereas intentional sounds from fish are generated with the purpose of attracting a mate, fighting, and other communication needs (Hossain 2). Different fish will use different mechanisms at different times at different frequencies, and although the exact species cannot usually be identified by their sound, recurring sounds in the data can be organized.

This paper aims to develop an acoustic morphotype for the reefs in Bahia Santa Elena while taking spatial and temporal variation into consideration. The goal of this work is to provide a foundation for acoustic analysis of the area's reefs as a starting point for science's understanding of the threat to a given species. Largely untouched by anthropomorphic activity, the reefs provide a good environment for studying the area's natural fish morphotypes. The Bahia Santa Elena reefs now being a Marine Management Area allow for representative samples of data. The data that will be used for this analysis include four hour long recordings from hydrophones dropped at three different reef clusters in the night time (hours not yet specified) at Bahia Santa Elena. Past research has suggested that nocturnal recordings of acoustic fish activity have been more successful (quantitatively) than the daytime activity of fish. The recordings were collected at night, as fish vocalizations are known to peak at dusk during the so called "dusk chorus" (Rossi, T., Connell, S. D., & Nagelkerken, I. (2016). Some potential fish that could be detected due to records of sound production in the tropical southeast Pacific fish include Albula vulpes (Bonefish, sound described as Click; Grunt Thump; Knock; Boom), Alectis ciliaris (African pompano), Balistes capriscus (Grey triggerfish), Balistes capriscus (Grey triggerfish), and others (FishSounds.net).

Strong biodiversity in a region is an indicator of the region's ecological health. Biodiversity is the quantity, variety and distribution across biological scales ranging through genetics and life forms of populations, species, communities and ecosystems. Biodiversity is the main factor that affects living organism's ability to positively respond and adapt to change (Hiddink 1). According to NOAA, coral reefs are one of the most biodiverse communities on our planet. Additionally, they provide over 500 million people with food, income, and protection. Coral reef community soundscapes become quieter (less detected sound) as they degrade (Lin et al. 2023).

Coral reefs are among the most highly threatened ecosystems on earth due to climate change and global warming. Declines in reef life abundance are associated with bleaching (and other diseases) driven by elevated sea surface temperatures and excess carbon dioxide in the atmosphere. Coral reefs, and the ecosystems that they house, are at risk of extinction. The threat to coral reefs worldwide are further exacerbated by local-scale anthropogenic disturbances, such as over/ illegal IUU fishing or anthrophony (man-made noise) pollution (Eakin et. al 2019). Worldwide, there is a heightened need for coral reef biodiversity data. There are several ways to measure activity and biodiversity in reefs. Acoustic soundscapes are an effective way of measuring acoustic biomass, and therefore biodiversity (Discovery of Sound in the Sea et. al

2020). The more noise, and more variety in sound, produced by reefs occupied by marine organisms, the healthier the reef is. Acoustic data collected and analyzed may add to the science and conservation community's foundational repertoire of reef fish activity, and overall ecological health/ abundance.

2. Methods and materials

2.1. Study Site

Santa Elena Bay Costa Rica is located in the province of Guanacaste, district of La Cruz. These waters became a Marine Management Area as an extension of the Santa Rosa Sector Guanacaste Conservation Area. In the waters of Costa Rica, there have been approximately 1112 different fish species detected, and nearly 7,000 total marine species (La Federación Costarricense de Pesca). There is currently no acoustic soundscape data in these specific reefs. However, according to data collected through transect samples in neighboring reefs, 84 reef fish species are said to inhabit the area (Alvarado et al., 2021). According to this same paper, fish biomass was dominated by piscivores with the highest mean, followed by macroalgae-eating herbivores, detritivores, invertebrate feeders, planktivores, and omnivores at the lowest. Grunts (Haemulidae) were abundant and dominated the fish assemblages in Murciélago Islands and the Santa Elena Peninsula (Alvarado et al., 2021).

2.2. Recordings

Recordings of the soundscape were made using autonomous underwater recorders at a sampling rate of 48 kHz using the model Audiomoth using a waterproof case from Open Acoustic Devices (www.openacousticdevices) from July 17 to 20, 2022 in three site outside Santa Elena Bay.

2.3 Acoustic Morph analysis

These 5-min files were extracted 1 hour apart and inspected in RAVEN and annotated with information about the number of significant sound detections and the corresponding established morphotype based on spectrogram appearance, perceived sound, and a number of different measurements provided in RAVEN. These measurements include begin time (s), end time (s), low frequency (Hz), high frequency (Hz), center frequency (Hz), delta frequency (Hz), filtered RMS amplitude (U), inband power (dB FS), leq (dB FS), max frequency (Hz), peak amplitude (U), SNR NIST quick (dB), and RMS amplitude (U). I will use the FishSounds database (https://fishsounds.net/index.js) to identify as many sounds as possible at the family level. Different fish species produce different sounds that can be detected by acoustic detection technology. Although the exact species cannot currently be identified off the acoustic data alone, it can be used to create a morphotype bank of different sounds (and therefore species) present in the reefs. The different sounds will be differentiated by measured duration, number of pulses, pulse rate, min/max frequency, and frequency range, as well as qualitative information about pulse rhythm.

3. Results

3.1. Morphotypes by location

Figure 1 shows that except for morpho 18, all identified acoustic morphos were present in two locations. However, the abundance of these morphos varied significantly between sites (x^2=150.8, df=1, p<0.0001) with morphos 1, 2, 6,14,17, and particularly 7 being more abundant in the location of hydromoth 3 than in the location of hydromoth 1. The types of morphotypes detected included knocks (1), gulps (2), drums(3), growls(5), gurgle-honk hybrids (6), taps(7), honk(8), unidentified calls(9), low-vibrational calls(10), high-vibrational call(11), roar(12), hums(13), croaks(14), gurgle-honk calls(18), and other undetected sounds(17).

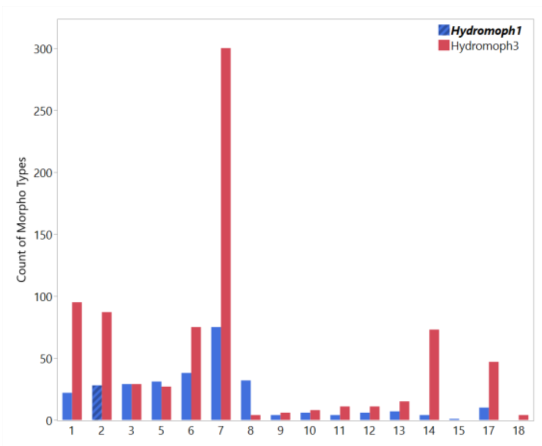

Figure 1. Morphotype (1-18) abundance (sum) measured and compared between hydromoths 1 and 2.

3.2 Acoustic characteristics of morphotypes

For comparison purposes I will focus on the three most abundant morphotypes in both locations to describe their acoustic structure in frequency and duration. We find that there is variation within morphotypes in frequency and duration between sites (Table 1, Fig.2). Morphotype 1 had a higher low frequency compared to 2 and 7, and greatly differed in high and max frequency between sites (Fig.2). This morphotype exhibited some degree of statistical significance between hydromoth locations for each acoustic variable (Table 1). Morphotype 2 had generally less outliers, as well as less statistical significance overall (Table 1, Fig.2). Morphotype 7 had a lot of variation in its high and low frequencies, and the highest recorded high frequency amongst these two morphotypes (Table1). Morphotype 7 also exhibited highly statistical significance in all acoustic variables other than delta frequency (Table 1).

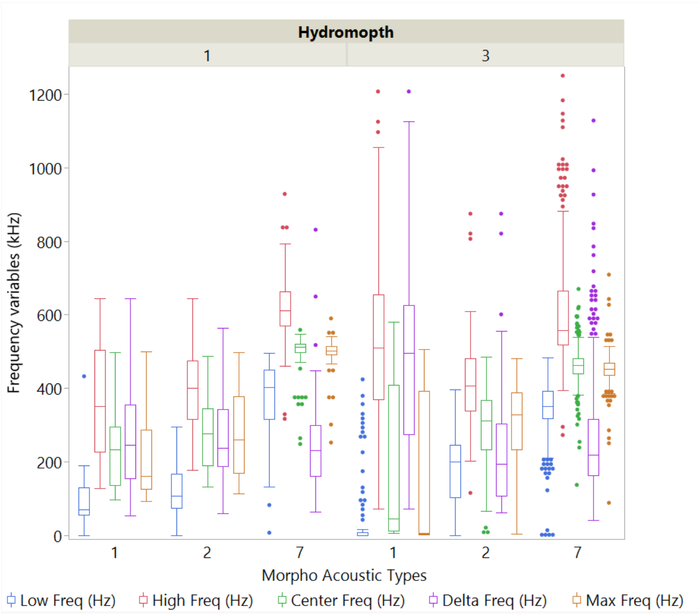

Figure 2. Acoustic structure of morphotypes 1, 2, and 7 between locations.

Table 1. Comparison of acoustic variables for morphos 1, 2 and 7 between hydromoth locations (*** highly significant, ** significant, * marginally significant at a p-value of p>0.05, all frequency variables are in kHz and duration in seconds).

Acoustic variable	**Hydromoth1**	**Hydromoth 3**	**Significance**
		Morpho 1	
Low Frequency	98.5(SD=90.8)	45.2(SD=102.6)	X^2=24.9, df=1, P<0.0001***
High Frequency	368.6(SD=155.6)	530.3(SD=239.4)	X^2=9.10, df=1, P=0.0026**
Center Frequency	243.8(SD=124.6)	184.7(SD=196.5)	X^2=4.15, df=1, P=0.041*
Delta Frequency	270.1(SD=151.7)	485.1(SD=259.4)	X^2=13.6, df=1, P=0.0002***
Delta Time	0.61(SD=0.23)	0.43(SD=0.30)	X^2=19.7, df=1, P<0.0001***
		Morpho 2	
Low Frequency	121.14(SD=67.2)	180.3(SD=104.2)	X^2=8.68, df=1, P=0.0033**
High Frequency	391.8(SD=113.8)	416.0 (SD=123.1)	P>0.05

Center Frequency	271.7(SD=96.9)	292.9 (SD=104.1)	P>0.05
Delta Frequency	270.6(SD=126.4)	235.8 (SD=161.0)	P>0.05
Delta Time	0.53(SD=0.13)	0.34(SD=0.1)	X^2=32.1, df=1, P<0.0001***
	Morpho 7		
Low Frequency	378.0(SD=107.0)	343.0(SD=77.3)	X^2=19.4, df=1, P<0.0001***
High Frequency	620.3(SD=96.6)	609.9(SD=150.9)	X^2=11.7, df=1, P=0.0006***
Center Frequency	494.5(SD=58.6)	461.9(SD=53.8)	X^2=55.6, df=1, P<0.0001***
Delta Frequency	248.3(SD=127.0)	266.9 (SD=173.2)	P>0.05
Delta Time	1.06 (SD=1.05)	0.35(SD=0.25)	X^2=101.8, df=1, P<0.0001***

4. Discussion

This study's results find that there was significantly more activity detected by the hydromoth at location #3, specifically for morphotypes 1, 2, and 7 in Santa Elena Bay Costa Rica. Morphotype 1 is sounds of general tapping, either successive or individual. These taps had large standard deviations for high, low, delta, and center frequencies, as well as delta times. This suggests either that there are different types of taps, either coming from the same fish for different purposes or from two separate fish species. There are some fish, such as Lusitanian toadfish, (*Halobatrachus didactylus)* that produce different sounds for different reasons (Sound production by the Lusitanian toadfish, *halobatrachus didactylus*, cite later). During mating season, males produce a "boat whistle", which lasts for ~0.8 seconds at about ~60 Hz frequency (Dos Santos et. al 2000, 4). This call has three segments characterized by different durations, pulse periods, amplitude, and dominant frequencies ("Discovery of Sound in the Sea" et. al 2020, 3). There can also be alterations in patterns, rate, and duration (3). Male *halobatrachus didactylus* have also been found to exhibit "short grunt" and "double-croak" sounds, which are mainly associated with nest defense against other males (4). There is not much information in scientific literature regarding the specific frequencies and variety of frequencies that fish species produce. It is difficult to study and identify fish sound emissions without disrupting their natural behavior and habitats. For this reason, the data collected and analyzed in this paper cannot be reliably attributed to specific species of fish. However, thanks to the compilation of scientific research on FishSounds.net, there is a list of fish who are known to make sounds and in some cases the type of sound is loosely detailed.

According to a reef environment analysis of Murciélago Islands and Santa Elena Peninsula in the Costa Rican Pacific, the Northern Pacific of this area (Santa Elena Bay) is dominated (biomass) by piscivores. In 2018, the most recent year assessment presented in the paper, piscivores contributed the most to similarities among localities (34.06%) across the Costa Rican Pacific (Alvarado et. al 2021). A comprehensive list of fish taxa identified across surveys of the coral reef ecosystems of the U.S and the search engine on FishSounds.net aided in creating the following list of possible morphotype sound-producers detected in the Audiomoths (Sandin et. al 2010, 12).

Within the Carangidae family, the *Alectis ciliaris* (African pompano), a piscivore, has been found to produce bark, burst, and scratch sounds. *Selar crumenophthalmus* (Bigeye scad) has been found to produce a knock sound, which could be related to morphotype 1 found in this paper's data (Looby et. Al 2023, 8). *Seriola dumerili* (Greater amberjack) has been found to produce a grunt, thump, and knock sound, which could be related to morphotype 1, 3, or 5 in this paper's data (8, 12). Morphotype 1, like morphotype 7, had large standard deviations in high, low, delta, and center frequencies, as well as delta times (8, 12). This suggests that there is also a wide range of frequencies (types) of knocks either being produced by the same family, or could be produced by two separate fish. It is possible that different species of fish (Bigeye scad and Greater amberjack) produced different knock sounds at different frequencies.

Within the Fistulariidae family, the *Fistularia commersonii* (Bluespotted cornetfish) has been found to produce unknown sound (8, 12). Within the Muraenidae family, the *Gymnothorax flavimarginatus* (Yellow-edged moray) and the *Gymnothorax meleagris* (Turkey moray) are known to make undefined sounds, along with the Scombridae family's *Katsuwonus pelamis* (Skipjack tuna) and *Thunnus albacares* (Yellowfin tuna) (8). Additionally, the *Sphyraena barracuda* (Great barracuda) and *Sphyraena helleri* (Heller's barracuda) of the Sphyraenidae family have been found to make undefined sounds (8,12). All of these fish are piscivores and have been found to be present in/ around Costa Rica's Santa Elena Bay.

This study shows the potential for passive acoustic monitoring. Future studies should associate sounds with fish species for a better use of sound in the study of coral reef health, and community composition and dynamics.

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Acoustic temporal patterns and snapping shrimp level of presence in different coral reef environments, Gulf of Papagayo, Costa Rica

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Abstract: Snapping shrimp are one of the most ubiquitous sounds in reef environments, making them a key addition to the soundscape. These shrimps are known for the high frequency, high amplitude crackling they contribute to the ocean. Because of their loud form of communication and physiology, researchers have suggested that snapping shrimp may be more resistant to noise level. However, this fails to consider anthropogenic noise which has been neglected in past publications on snapping shrimp despite its structural sound differences from the high frequency sounds these shrimp experience naturally. This study aimed to address this knowledge gap by analyzing the presence of snapping shrimp and their temporal patterns at two reef sites in the Gulf of Papgayo, Costa Rica where one (Guiri-Guiri) was expected to have higher boat presence than the other (Palmitas) due to its placement by a marina. We used acoustic event measurement to look at the amplitude of snapping shrimp snaps within a defined frequency range in order to assign snapping shrimp level of presence at each site while simultaneously tracking boat presence at each site. The results revealed no significant difference in boat presence between site meaning that no relationship between boat presence and snapping shrimp presence could be distinguished. However, we found a significantly higher level of snapping shrimp presence at Palmitas compared to Guiri-Guiri without any temporal shift, and we found differences in average snap frequency and amplitude between sites. With that being said, we could not determine if boats had an impact on snapping shrimp, but factors such as boat characteristics and environmental characteristics may have been significant or altered the results in ways we were not able to assess since we didn't collect this data. Future research should be pursued including this data in order to properly evaluate the impacts of anthropogenic noise on snapping shrimp given that previous studies have established these shrimps respond to reef habitat quality and ocean temperature making them a potentially good indicator species for monitoring.

Keywords: snapping shrimp presence, boat presence, coral reefs

1. Introduction

Soundscapes are collections of sound from physical, anthropogenic, and biological sources (Butler et al. 2017). Within marine habitats, soundscapes are particularly important and far-reaching as the environment is poor for sight-driven communication and sound moves through water 4.5 times faster than air (Hawkins et al.). The acoustic activity of a marine soundscape is described as being dependent on habitat characteristics and species composition, and the sounds making up these environments can originate from sources like rain, animal calls, and boats (Lammers et al. 2007; Kennedy et al. 2010). All of these sounds can impact species' behavior, reproduction, and survival within a given soundscape (Bohnenstiehl et al. 2016; Edmonds et al.

2016). In reefs, a major contributor to the soundscape is biological noise created by the snapping shrimp (Family *Alpheidae*) (Edmonds et al. 2016). Snapping shrimp are decapod crustaceans that measure a few centimeters in length but possess a large claw which produces one of the loudest sounds in the ocean, reaching over 200 dB (Lee et al. 2021; Bohnenstiehl et al. 2016). This sound is generated when the shrimp snaps its claw at a high speed such that a cavitation bubble forms and then collapses as water exits the pocket, radiating a distinct snap (Lee, 2021). These snaps are created rapidly by many shrimp over a sustained period of time and they vary in rate from 100 to 4,000 snaps per minute (Bohnenstiehl et al. 2016; Song et al. 2023). The frequency range of these snaps is broadband meaning they can be as low as tens of hertz or as high as over 200 kHz, but most commonly their frequency is between 2 and 5 kHz (Versluis et al. 2000; Song et al. 2023).

Snaps are utilized regularly by the shrimp to stun their prey, communicate with one another, and distinguish their territory (Lee et al. 2021). This acoustic activity is higher at low latitudes and in shallow waters where they inhabit coral and oyster reefs, mangroves, and kelp forests (Lee et al. 2021; Song et al. 2023). Several studies have documented changes in snapping shrimp temporal acoustic patterns in response to the physical and climatic characteristics of these habitats (Bohnenstiehl et al. 2016). For example, snap activity appears to correlate with temperature, tide level, and current speed (Lee et al. 2021; Song et al. 2023). Diurnal and seasonal variations in the number of snaps shrimp populations contribute to the soundscape have also been recorded; however, it's suggested these variations may be a result of more complex abiotic factors such as water temperature and light availability (Lee et al. 2021).

Because snapping shrimp communicate so loudly and lack a gas-filled chamber in the body which would allow sound pressure to affect them, they are thought to be resilient to noise (Popper et al, 2001). However, the sounds snapping shrimp are exposed to naturally through communication are very different from the low frequency, extremely high amplitude noises most anthropogenic activities emit (Edmonds et al. 2016). Among the variety of studies published on snapping shrimp, none have evaluated the effect this type of noise may have on the family (Edmonds et al. 2016). For example, the direct impact of boat noise in the marine soundscape snapping shrimp inhabit has not been thoroughly analyzed as a possible agent of acoustic temporal shift. In this study, I will analyze snapping shrimp's hourly acoustic activity on two coral reefs, with only one site being close to a boat marina such that I expect a higher presence of boats there. My aims are (1) to determine the hourly variation of snapping shrimp acoustic presence using acoustic event measurements within their frequency range (2-5 kHz) and (2) assess the potential relationship between shrimp acoustic activity and boat traffic. Independent of habitat, I expect snapping shrimp activity to be highest during the day (hour 6-18) and peak at dawn (hour 6) and dusk (hour 18) as Lillis et al. (2018) demonstrated to be their most active periods in tropical habitats. Therefore, taking into consideration the expected boat traffic in each habitat, I hypothesize that (1) snapping shrimp activity will be significantly higher in the non-marina-adjacent reef if I find it to have significantly lower boat traffic and (2) there will be a temporal shift to greater nighttime activity in the marina-adjacent reef in response if there is higher boat activity.

The broader implication of this study is to increase knowledge about the impacts of anthropogenic activity on the marine environment by focusing on one of the most predominant sounds in the ocean (Lillis et al. 2018). Given their prevalence, it's important to consider what snapping shrimp activity can say about the health of marine habitats. The Butler et al. (2017) findings showed a positive relationship between the health of a habitat and the number of snapping shrimp acoustically detected Looking toward the future, studies indicate that coral reefs will rapidly decline over the next 20 years largely because of temperature stress associated with climate change (Hoegh-Guldberg et al. 2017; Sully et al. 2019). Currently, coral reefs house some of the greatest biodiversity in the ocean and provide important ecosystem services to people, which makes monitoring them incredibly important (Harvey et al. 2018). By further understanding what factors impact the activity levels of snapping shrimp, their ubiquitous sounds could be used to monitor such environments acoustically.

2. Methods

2.1 Study Area
This study took place in the reefs of Guiri-Guiri and Palmitas within the Gulf of Papagayo off the Pacific coast of Costa Rica. The habitat in this area consists of coral reefs and mangroves, with most reefs forming around islands (Miloslavich et al. 2011). Snapping shrimp are abundant in these marine ecosystems and their 'snaps' are high enough in intensity to be detected if present in this study area (Robolledo, 2014). These sites were selected as it's expected that Guiri-Guiri has a higher presence of boats than Palmitas given the reef's placement next to a marina.

2.2. Recordings
Recordings of the soundscape were made using autonomous underwater recorders at a sampling rate of 48 kHz. Recordings at both sites were made in 2019 between December 18-31. Autonomous recorders were bottom mounted by attaching the recorder on a pole 1.5 m above the seafloor and anchoring it with a concrete block (approx. 30 kg) at 12 m depth. The sites of deployment were near patches of coral and rocky reefs. Recordings were made with a SoundTrap 300 STD (frequency range 20 Hz-150 kHz ±3 dB; self-noise of less than sea-state in the bandwidth 100 Hz-20 kHz, and sensitivity of −203 dB re V/µPa) from Ocean Instruments (http://www.oceaninstruments.co.nz/).

2.3. Snapping shrimp activity analysis
The first 5 minutes of each hour were manually taken from the available recordings between December 18-31 at both sites using sound analysis platform Audacity. These 5-minute files were then visually inspected on the same platform through a plotted spectrogram of decibels versus frequency. The frequency and decibel levels of the highest peak on each graph within the parameters of 1-5 kHz were recorded on an excel sheet. I selected this frequency range because snapping shrimp dominate the high frequency broadband within reef habitats which is generally above 1.5-2 kHz and their peak frequency for snapping shrimp was previously determined to be between 2-5 kHz (Bohnenstiehl, 2016; Song, 2023). When another sound such as boat noise or other species calls clearly entered this frequency range, that portion of the sample was omitted from analysis in order to get an uninhibited reading of the peak frequency and decibel level the snapping shrimp were reaching. Based on the peak decibel levels and audio comparison of the 5-min recordings for each day, a rating of 1-3 for snapping shrimp activity was recorded for each file, with 3 being the greatest level and 1 being the lowest level. The audio recordings were assigned these levels of presence by organizing the data from lowest to highest based on peak amplitude, and then assigning each presence rating to a third of the data. Furthermore, each 5-minute file was annotated with information about the hour, day, and site the recording took place, information about the presence (1) or absence (0) of snapping shrimp, and information

about the presence (1) or absence (0) of boats in and Excel spreadsheet. Using this data, a bar graph, line graphs and box plots were created in the program JMP Pro 15 for analysis.

3. Results

3.1. Snapping shrimp presence

The presence of snapping shrimp varied significantly between Guiri-Guiri and Palmitas (x^2=492.80, df=1, p<0.0001). As shown in figure 1, Palmitas had a higher presence of snapping shrimp while Guiri-Guiri had the lowest (Fig.1). There is an even spread of medium-level presence between sites.

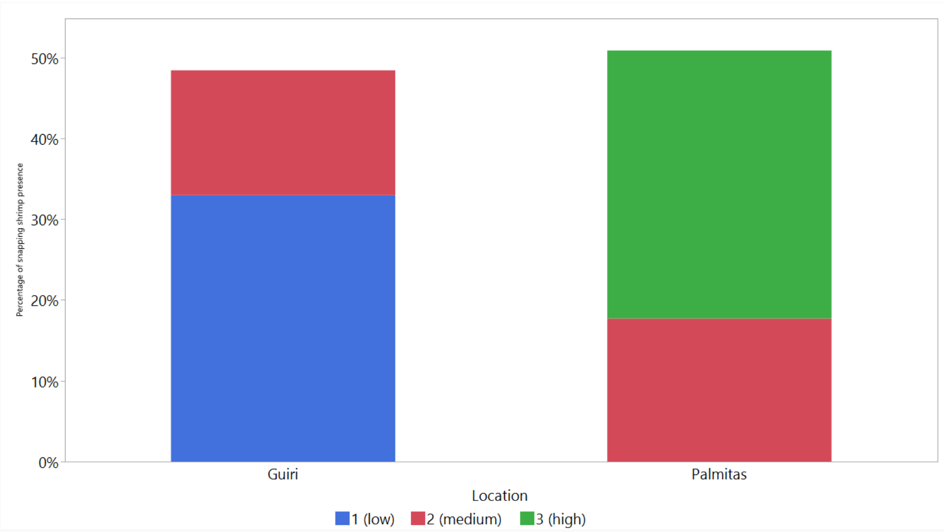
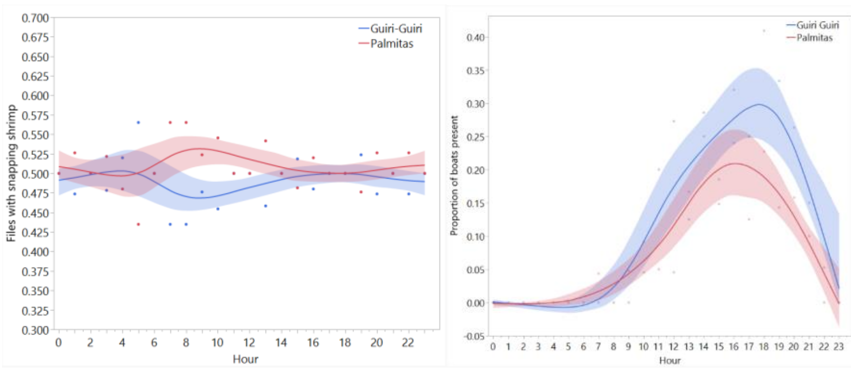


Figure 1. The proportion of snapping shrimp activity distinguished by levels 1-3 at Guiri-Guiri and Palmitas based on the peak amplitude reached within 5-minute samples from each site in the 1-5kHz frequency range (Frequency: Level 1= -50.8dB to -46.7dB, Level 2= -46.7dB to -38.2dB, Level 3= -38.2 dB to -30.6dB).

3.2. Temporal patterns of activity of snapping shrimp and boats

Overall, there were significant differences in the proportion of files with snapping shrimp presence by location (x^2=6.38, df=1, p=0.0115) (Fig.2a). Although there is a trend that Palmitas has a higher proportion of snapping shrimp between 7 a.m. and 10 a.m. this pattern is not significant (p>0.05). Similarly, Guiri-Guiri appears to have a higher proportion of boats, but this pattern is not significant (p>0.05) (Fig.2b).



a. Snapping Shrimp Presence b. Boat Presence

Figure 2. Temporal patterns in snapping shrimp presence and boat presence at Guiri-Guiri and Palmitas (a) The proportion of 5-minute sound files with snapping shrimp presence in Guiri-Guiri and Palmitas over 24 hours (b) The proportion of 5-minute sound files with boat presence in Guiri-Guiri and Palmitas over 24 hours.

3.3 Acoustic structure of snapping shrimps

There were significant differences in snapping shrimp snaps between location in peak frequency (x^2=378.46, df=1, p<0.0001) and in the amplitude of the snap between 1-5 kHz (x^2=394.19, df=1, p<0.0001) (Fig.3). Snapping shrimp snaps in Guiri-Guiri had on average a higher peak frequency but lower amplitude, while in Palmitas they had on average a higher amplitudeand lower peak frequency (Fig.3). This suggests that amplitude could be used as an indicator of snapping shrimp presence.

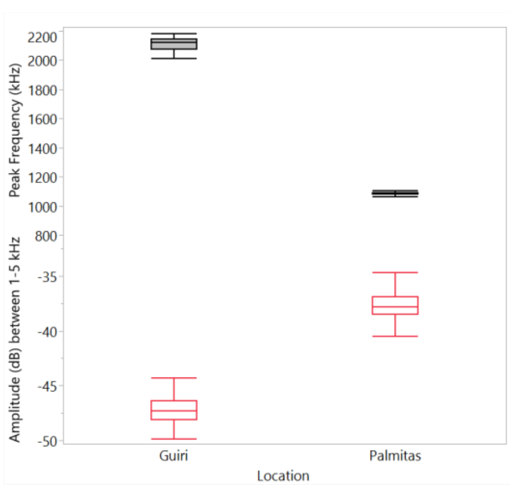


Figure 3. The average peak frequency (higher energy at given frequency) and average peak amplitude (louder amplitude has dB values closer to 0) observed at Guiri-Guiri and Palmitas.

4. Discussion

We observed significant differences in snapping shrimp presence, snap peak frequency, and snap amplitude between Guiri-Guiri and Palmitas which has important implications for further understanding the response snapping shrimp have to noise in their environment and could be integral to the future conservation of the species and their reef habitats. We did not find a significant differences in boat presence between sites or obvious temporal patterns of acoustics across populations indicating that boat presence may not be the most important factor when deriving the reason behind the snapping shrimp level of presence and their activity in a habitat.

As it stands, the field of snapping shrimp research lacks information about the impact of anthropogenic noise on their behavior (Edmonds et al. 2016). In fact, marine invertebrates in general are missing data about their response to underwater noise, which is important given their different physiology compared to the mammal and fish species which have been more broadly studied in this field (Edmonds et al. 2016). Although they lack a gas-filled chamber making them insensitive to sound pressure, Roberts et al. (2016) suggests that crustaceans like hermit crabs are sensitive to low frequency particle movement like that created by anthropogenic noise. Low frequency, high amplitude noises like those from wind turbines have been shown to delay metamorphosis among crab species, making it plausible that a crustacean like snapping shrimp could also be susceptible to the negative impacts from anthropogenic noise (Pine et al. 2012).

Although this study didn’t find a significant difference in boat presence between sites, there was higher boat presence at Guiri-Guiri. Given the significantly higher peak frequency and lower amplitude of snaps at this site, boats should not be discounted as a potential factor impacting snapping shrimp presence. Foote et al. (2004) found differences in whale call structure when they were exposed to boats making high amplitude noises of similar frequency, which suggests that the amplitude and frequency of snaps could be key to understanding the changes snapping shrimp make to adapt to anthropogenic noise level characteristics that weren’t evaluated in this study.

Abiotic factors such as temperature, tide level, and moon phase have also been shown to change the presence of snaps in an environment, but these factors were not considered in this study due to a lack of site-specific data (Lee, 2021; Song, 2023). Therefore, there may have been significant differences in environmental factors between sites which could explain the differentiation in snapping shrimp level of presence. Furthermore, since the temporal data showed no alignment between boat level of presence and snapping shrimp activity, these abiotic factors should be considered as potential drivers of the temporal data in this study. Although the temporal data described contrasts with the findings in Lillis et al. (2018) which suggests there should be peaks of snap activity at dawn and dusk, utilizing abiotic factors in future studies could account for these differences.

Integrating more information about boat noise characteristics and abiotic factors into this study could have strong implications for the future health of snapping shrimp and their use as an indicator species. Although there is not much information about the function of snapping shrimp in the ecosystem, they do have an established mutualistic relationship with the Goby fish which shows their absence could have species related impacts (Jaafar et al. 2014). Furthermore, snapping shrimp have previously responded to climate driven factors like water temperature (Bohnenstiehl et al. 2016). This, alongside findings by Butler et al. (2017) that showed increased levels of snaps per minute in healthy hardbottom habitats compared to unhealthy hardbottom habitats could mean that snapping shrimp are a metric of reef health. Therefore, their behavior and relationship to anthropogenic factors should be studied further to establish their ecological importance and their potential use as an indicator species.

5. Conclusion

Overall, the results of this study mean that although there was no significant relationship between the presence of snapping shrimp and boats, there are differences in the acoustic expression of snapping shrimp between sites that have different defining factors. This study, and further studies like this on a larger scale have important implications for the conservation of coral reefs as previous studies have shown that species fair better in the sounds associated with their natural environments (Williams et al. 2022). With snapping shrimp being such a ubiquitous sound in reefs, their presence could be integral for the future health and conservation of other species (Song, 2023).

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A characterization of the temporal changes in ambient noise levels of the Cuajiniquil Bay, Costa Rica

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Abstract: This study investigates the temporal variation of underwater ambient noise levels in the Cuajiniquil Bay, Costa Rica, using recordings from July to September 2022. The research aims to establish a baseline understanding of the bay's soundscape and biodiversity. The results show that the dominant sound sources in the bay are fish, boats, and humpback whales. Fish acoustic presence peaked at night and boat activity peaked between 6 a.m. and 6 p.m. Both were consistently present across all three months. Humpback whale songs increased from July to September and peaked in August, aligning with their expected migration into the area for breeding. Independent of month, ambient noise levels in the bay were higher at frequencies above 1000-Hz, suggesting that boats and whales are the most important contributors to the overall soundscape. This study provides the first soundscape characterization of the Cuajiniquil Bay and highlights the importance of integrating soundscape studies in ongoing biodiversity-monitoring programs and as a tool for policymakers to address the impacts of increasing industrialization on vulnerable marine communities.

Keywords*:* ecosystem diversity, coral reef health, anthropogenic noise, humpback whales, tourism

1. Introduction

Soundscapes are defined as the set of sounds in an area from geological, biological, and anthropogenic sources (Pijanowski, 2011). Features of a marine soundscape often include fish, marine mammals such as whales, wind, waves, and boat motors. Researchers use underwater autonomous acoustic recorders to study marine soundscapes at various spatial and temporal scales (White *et al.*, 2022; Marley *et al*., 2017). This type of research is important because many marine species rely on sound for numerous functions including navigation, foraging, group coordination, predation, communication, reproduction, migration, and habitat selection (Marley *et al*., 2017; Pieretti, 2017; van Opzeeland and Slabbekoorn, 2012). For humans, these behaviors are often visually obscured due to the physical restrictions presented by water; listening to the marine soundscape permits marine researchers to observe and interpret marine life more closely (Kaplan et al., 2016).

To apply soundscape data as an ecological tool, it is necessary to first understand the natural variability of noise in a given habitat (Kaplan *et al*., 2016). Fish activity, for example, routinely peaks at different times depending on the reef; McCauley & Cato (2000) found fish calling activity on the Great Barrier Reef peaks at midnight, whereas Au et al. (2012) found fish sounds on reefs in Honolulu, Hawaii, peak at twilight. Whales can also be significant contributors to the variability of noise in a habitat because they migrate seasonally for breeding (Acevedo *et al*., 2017). Once a baseline of spatial and temporal variability is established in an area, changes to the soundscape (e.g., a reduction in reef fish activity) can be monitored and correlated to causational variables (e.g., increased boat activity).

Studying marine soundscapes is also important today because ocean soundscapes are rapidly changing due to the declining biodiversity of sound-producing species as well as increasing anthropogenic noise (e.g., boat traffic, seismic surveys, echosounders) (Duarte *et al*., 2021). Propulsion of boats and shipping vessels is the leading source of human-made noise in the ocean and has contributed to a 32-fold increase in low-frequency marine noise along shipping routes in the last 50 years (Duarte *et al*., 2021; Tyack, 2008). This noise propagates mainly at low frequencies below 1000-Hz, but it is broadband and can reach frequencies as high as 10-kHz (Haver *et al.*, 2021; Tyack, 2008).

Anthropogenic noise has become recognized by marine researchers as a chronic, environmental stressor, that has the potential to harm not only individual marine animals but the ecosystem as a whole (Marley *et al*., 2017; Tyack, 2008). Cetaceans such as dolphins and whales are especially impacted because they use elaborate sound production and auditory perception mechanisms to communicate (Marley *et al*., 2017). For dolphins, close and persistent pursuit by dolphin-watching tour boats has been found to elicit increased whistle emission and whistle modulation, frequency, and duration, which are likely indicators of stress and alertness in the animals (May-Collado *et al.*, 2012; Perez-Ortega *et al.*, 2021). Coral reefs are another high-impact community to note: In coral habitats, the sounds associated with healthy reefs attract fish larvae and other groups of invertebrates to settle and remain in the habitat (Gordon *et al.*, 2019). Analysis of settlement behavior in reefs infiltrated by boat noise showed fewer fish settled than in reefs free of boat noise (Simpson *et al.*, 2016). These studies highlight the importance of including ocean noise in marine planning on various spatial and temporal scales. The response of marine mammals to human-made sound is also likely site- and species-specific, which deepens the need for habitat-specific soundscape evaluations (Pires *et al.*, 2021).

The goal of this study is to characterize the temporal variation of underwater ambient noise levels and identify the most important sources of noise using a single recorder deployed in the middle of the Cuajiniquil Bay. The Cuajiniquil Bay, located in the Gulf of Santa Elena on the North Pacific Coast of Costa Rica, is a hotspot of marine biodiversity. The bay is home to mangroves and coral reefs, two important ecosystems that function as nurseries for fish and invertebrates (Kappelle, 2016). The area is also subject to significant upwelling via wind patterns and ocean currents (Kappelle, 2016). Upwelling events carry cold, nutrient-rich, low-oxygen water to the coast, which increases phytoplankton growth and in turn supports the development of a rich ocean food web (Santora *et al.*, 2017). There is additionally a prosperous fishing community in the town of Cuajiniquil. Fish and fishing-boat activity were therefore expected to continuously contribute to the soundscape of the bay.

Seasonally, the bay is an important reproductive area to two populations of humpback whales: the Central American population from December to March (Steiger *et al*., 1991; Calambokidis *et al*., 2000; Rasmussen *et al*., 2012) and the BSG population from July to November (Acevedo-Gutierrez and Smultea, 1995; Rasmussen *et al*., 2007; Acevedo *et al*., 2017). This study examines data from July to September of 2022, therefore songs from the BSG population were expected to contribute to the soundscape, picking up especially in August as more whales arrived in the area. I hypothesized that noise levels would vary monthly reflecting changes in the presence of the dominant sound sources (Bertucci *et al.*, 2016).

Examining the temporal patterns of underwater noise levels in the Cuajiniquil Bay serves to establish a baseline of the bay's soundscape and biodiversity. This baseline can be compared to future characterizations of noise levels in the bay, revealing changes in human activity as they correlate to ecosystem health. Ocean noise research such as this has significant implications in marine conservation because it allows scientists to determine causational relationships between anthropogenic sound and marine ecosystem degradation. It accordingly guides conservationists on how to anticipate impacts of increasing ocean industrialization, and to guide policymaking and effective mitigation for vulnerable marine communities (Williams *et al.*, 2015).

2. Materials and Methods

2.1. Study Site

This study takes place along the North Pacific Coast of Costa Rica in the Cuajiniquil Bay, located in the Gulf of Santa Elena. This site is home to a rich community of marine species (Kappelle, 2016). The beaches are inhabited by organisms such as polychaetes, amphipods, and crabs, and are in many places used by sea turtles to lay their eggs. The intertidal zones are dominated by chitons and gastropods. Mangroves and coral reefs are present along the coastal waters of the bay (Kappelle, 2016, Loría-Naranjo *et al.,* 2014). Finally, the pelagic zone is home at least 19 species of cetaceans (Ramirez et al., 2023). A portion of the bay is part of the Santa Rosa National Park under management by the Área de Conservación Guanacaste since the 1970s (Maestro, 2022). The unprotected part of the bay remains subject to unregulated extraction and tourism activities (Loría-Naranjo *et al.,* 2014).

2.2. Recordings

Recordings of the soundscape were made using an autonomous and remote underwater recorder model SoundTrap 400 STD (frequency range 20 Hz-150 kHz ±3 dB; self-noise of less than sea-state in the bandwidth 100 Hz-20 kHz, and sensitivity of −203 dB re V/µPa) from Ocean Instruments (http://www.oceaninstruments.co.nz/). Recordings were made from July to September 2022, using a recording cycle of 10-minutes per hour and a sampling rate of 48 kHz.

2.3. Ambient Noise Levels

To calculate broadband ambient noise levels in the Cuajiniquil Bay, recordings taken with the SoundTrap 400 STD recorder from multiple days each month were selected: specifically, blocks of 3 days each separated by 5-day intervals. Broadband ambient noise levels were calculated as the RMSdb (average root-mean-square) by taking the first 10-minutes of every hour of the day. RMS calculations covered frequencies from 100 to 40kHz using acoustic analysis software dbWav from Marshall-Day Acoustics (https://www.marshallday.com).

2.4. Sound sources

For the same files described above, the software Audacity was used to visually and audibly analyze each 10-minute spectrogram. I manually annotated an excel file with a "1" when ambient noise of whales, boats, and any other noise sources (i.e., fish, snapping shrimp) were present at any point within the 10-minute period. A "0" was assigned when acoustic species or boats were not detected.

3. Results

3.1. Temporal composition of the soundscape

A total of 136 hours of acoustic time were analyzed throughout July (29 hours), August (57 hours), and September (50 hours) of 2022 in the Cuajiniquil Bay. Fish accounted for approximately 20% of sound in July, 20% in August, and 10% in September (Fig. 1). They were most active between the hours of 6:00 pm and 4:00 am, present in nearly all files at midnight (Fig. 2.). Southern humpback whale male songs made up only 5% of the sounds identified in July but increased to 45% in August, and 25% in September (Fig. 1). Overall, when present, their activity was relatively constant throughout the day (Fig. 2.). When accounting for days of the month, humpback whale detection was highest in the last two weeks of August, peaking around days 17 and 25, and into the first half of September, peaking around days 10 and 22 (Fig. 3).

Boat presence was constant and accounted for 35% of the sounds identified in July, 25% in August, and 30% in September (Fig. 1). Boat detections peaked between 6:00 am and 6:00 pm (Fig. 2). Dolphin detection was negligible throughout all three months (Fig. 1-2). Finally, unknown sounds were present in all three months, accounting for 40% of sounds in July, 12% of sound in August, and 25% of sound in September (Fig. 1). There were more unknown sounds at night, beginning to increase after 5:00 pm (Fig. 2).

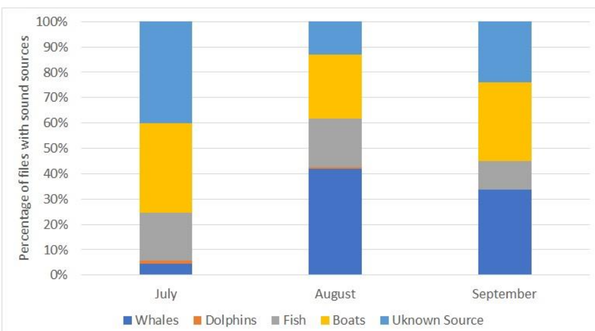

Fig. 1. Monthly composition of sound sources in the bay based on a subsample of 815 10-min files.

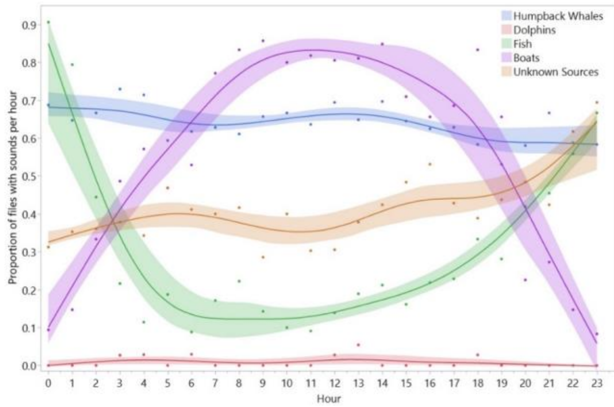

Fig. 2. Hourly distribution of sound sources in the bay.

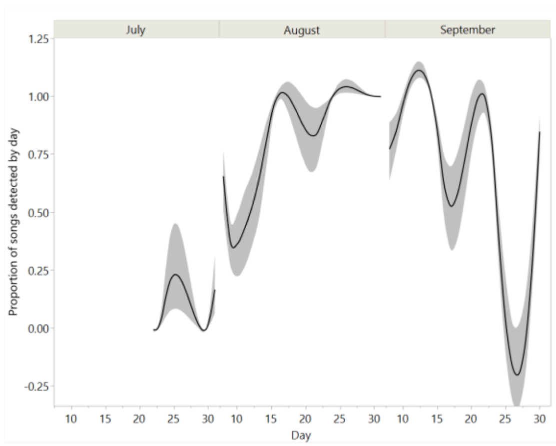

Fig. 3. Daily detection of humpback whale songs in the bay in each month.

3.2. Temporal changes in noise levels

Mean ambient noise level in the Cuajiniquil Bay varied significantly by month (F-Ratio=73.78, df=2, p<0.0001). Ambient noise levels were highest in August and lowest in July (Fig. 4). Ambient noise levels also varied significantly by day (F-Ratio=137.8, df=1, p<0.0001, Fig.5). Mean ambient noise levels peaked around days 17 and 25 in August, and days 10 and 20 in September (Fig. 5). Finally, mean ambient noise levels also varied by hour (F-Ratio=5.02, df=1, p=0.025, Fig.5).

In addition, the bay appears to be dominated by ambient noise primarily at frequency bands above 1000-Hz in all three months (F-Ratio=5443.8, df=1, p<0.0001, Fig.6). Within each frequency category, ambient noise levels at frequencies above 1000-Hz were highest in September (F-ratio=407.06, df=2, p<0.0001). In contrast, noise levels at frequencies below 1000-Hz were highest in August (F-ratio=311.6, df=2, p<0.0001) (Fig. 6).

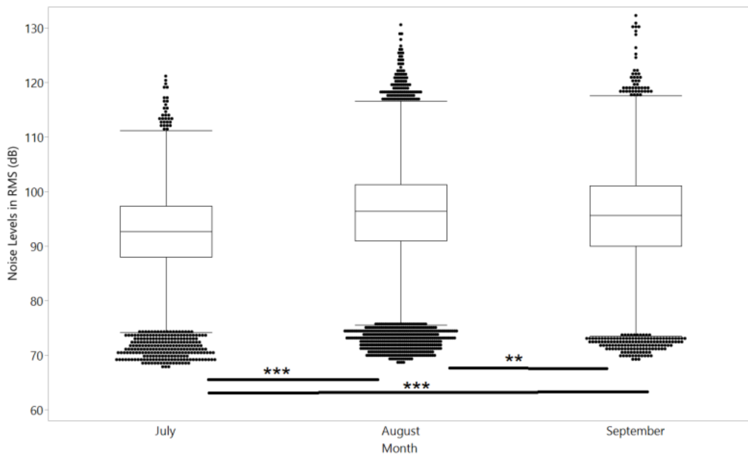

Fig. 4. Mean noise level variation by month (***= p<0.0001, **= p=0.0023).

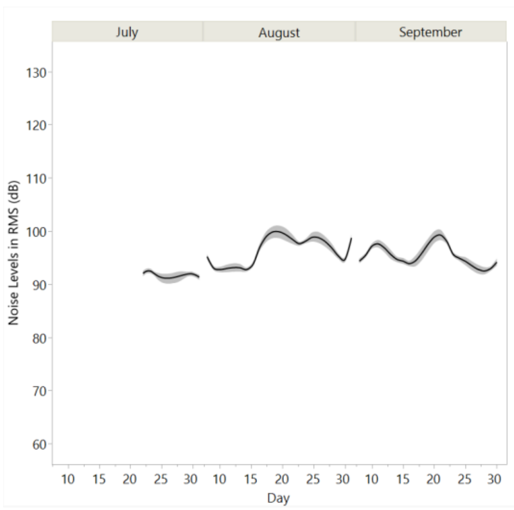

Fig. 5. Mean noise level variation by day in each month.

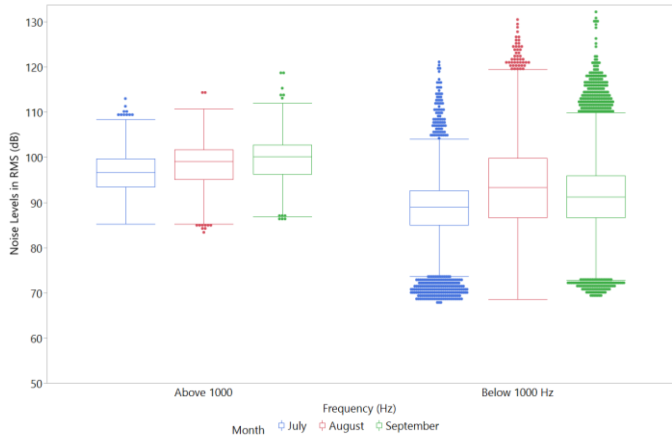

Fig. 6. Noise levels in the bay above vs. below 1000 Hz in each month.

4. Discussion

The results showed distinct temporal patterns in soundscape composition of the Cuajiniquil Bay. Fish, a consistent contributor to the soundscape, exhibited peak activity during the nighttime hours. Previous works in Australia and Hawaii have shown similar patterns on reef fish (McCauley & Cato, 2000; Au et al., 2012). In coral habitats, sounds produced by resident fish and invertebrates is a fundamental component of ecosystem functioning and thereby an indicator of ecosystem health (Gordon *et al.*, 2019; Pieretti and Danovaro, 2020; Simpson *et al.*, 2016). The consistent presence and pattern of fish activity may therefore indicate the bay is in good health.

The detection of humpback whale songs increased from July to September, with peak detection in August corresponding to the expected migration of humpback whales into the area for breeding (Acevedo-Gutierrez and Smultea, 1995; Rasmussen et al., 2007). In contrast, Chereskin et al., (2019) found that in the southern part of Costa Rica, southern humpback whale acoustic presence increased from July to October, with a peak in September (Chereskin et al., 2019). Despite this slight dissimilarity, both studies demonstrate that when humpback whales are present, they contribute significantly to the daily and hourly soundscape of the Pacific coast of Costa Rica. The results of this study further support this idea in that humpback whale song detection and mean ambient noise levels peaked on the same days (day 17 and 25 in August, and around day 10 and 20 in September). It is therefore likely that higher overall noise levels in the bay were attributed to increased humpback whale activity. Humpback whale singers are male, so song functions may include attracting females, establishing dominant or affiliative male-male relationships, or guiding aggregation during migration (Herman *et al.*, 2013; Herman, 2016).

Boat activity, a dominant anthropogenic source in the bay, demonstrated a consistent presence throughout the study period. This shows how ubiquitous anthropogenic noise is in marine environments. The location of the recorder was at the entrance of the bay, which is home to a small fishing community. While it was not the goal of this study to assess the impact of boat noise on fish activity, we show that most of the noise was at frequencies above 1000-Hz, which is the range of most small engine boats. Simpson *et al.* (2016) demonstrated that reefs infiltrated by small-boat noises showed fewer fish settled than in reefs free of boat noise. Continued research is needed to establish this possible negative relationship between fish and boats in the Cuajiniquil Bay.

Noise at frequencies above 1000-Hz was higher in all three months. However, within the two frequency categories (above vs. below 1000-Hz), some months were higher than others. This suggests that the distinct contributions of organisms and boats to the overall soundscape differed temporally (Tyack, 2008). Humpback whale songs, for example, are typically below 5000-Hz (Winn *et al.*, 1981) and boat motors are broadband in frequency but with most energy at lower frequencies (Tyack, 2008). Temporal changes in the presence of whales and boats would therefore shift the monthly mean ambient noise levels. Future research is needed to determine the contribution of each sound source to ambient noise levels in this area.

This study contributes to the establishment of a baseline soundscape of the Cuajiniquil Bay from July to September, offering a foundation for future comparisons and assessments of environmental changes in the area. The evident influence of anthropogenic noise, especially from boat activity, underscores the need to continue to monitor the habitat's soundscape. The findings emphasize the importance of integrating soundscape research into marine conservation efforts.

5. Conclusion

This investigation of the temporal variation of underwater broadband noise levels in Cuajiniquil Bay contributes valuable information to the broader field of marine soundscape research. The study highlights the complex interplay of natural and anthropogenic factors shaping the acoustic environment, providing a basis for ongoing efforts in ecosystem management. Continued research in this area in the coming years is crucial for understanding the long-term implications of changing soundscapes on marine biodiversity and informing proactive conservation measures.

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Singing activity of humpback whales within and outside protected areas in the Gulf of Chiriquí, Panama

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Abstract

Male humpback whales (*Megaptera novaeangliae*) produce complex songs throughout their breeding season. To identify temporal and spatial patterns in habitat use, presence/absence data was used from the National Park of Coiba, Panama. I compared the singing activity of humpback whales in one site inside the park and one outside the park. The results show that there is a higher song presence on the site outside the protected area. This pattern was maintained when considering the month and time of day. Because boat presence was low, I could not evaluate the effect of boats on these whales' singing activity. However, is important to note this data was collected during the Covid-19 lockdowns of 2020 when boat presence was at its lowest. The observed patterns of humpback whale singing activity cannot be explained by environmental factors either, temperature was similar among months, and the data was insufficient to evaluate the role of moon phases and tidal range. With humpback whale presence higher outside of the protected area, borders must be reevaluated for maximum protection of the breeding area.

Key Works

Humpback whale, BSG, passive acoustic monitoring, habitat use, National Park of Coiba, Panama

1. Introduction

Male humpback whales (*Megaptera novaeangliae)* produce complex songs throughout their breeding season (Smith, 2008). Combined with passive acoustic monitoring methods, researchers can use humpback whale song detection to identify temporal and spatial patterns in habitat use (Blair *et al.,* 2018; Risch *et al.,* 2012). Previous work has shown that humpback whale activities can vary with the hour of the day (Chereskin *et al*., 2019), the oceanographic properties of their environment which directly influence sound transmission (Rishch *et al*., 2012), lunar phases, day of the season, and boat activity (Sousa & Clark, 2008). These factors on humpback whale song can be used to model patterns in habitat use, allowing scientists to better inform wildlife managers how to effectively conserve this species.

In this study, I will be researching the factors determining the detection of male humpback whales' songs within and outside the National Park of Coiba, Panama. The study areas are located within the Gulf of Chiriquí, an area that receives humpback whales from the Southeastern Pacific population also known as the Breeding Stock G (BSG) (Rasmussen *et al*., 2007). These whales spend June-September in their breeding area of Central America, arriving from the Antarctic Peninsula and southern Chile (Acevedo *et al.*, 2007). BSG is estimated to be around 11,784 individuals and is the most genetically differentiated population in the Southern Hemisphere (Amaral *et al*., 2016; Félix *et al.*, 2018).

Using presence-absence data of humpback whale song detections from passive monitoring acoustic recorders at the Gulf of Chiriquí, this study seeks to understand the temporal patterns in humpback whale singing acoustic activity within and outside protected areas and in relationship to the environmental (SST, tide, moon phase) and boat traffic. Based on Sousa-Lima & Clark's (2018) research on humpback whale song activity in Brazil, I hypothesize that environmental factors such as the lunar phases could result in an increased singing during the night, which could also be due to the lower number of boats. Several studies have also demonstrated the impacts of underwater anthropogenic noise on the communication of marine mammals (Blair *et al.,* 2016; Gabriele *et al.,* 2018). In a study comparing the singing activity of humpback whales before, during after the COVID-19 pandemic lockdowns, researchers found that whale singing activity increased during the lockdown, highlighting the potential impacts of noise associated with boats (May-Collado *et al*., 2023). Singing activity has also been reported to increase at night, with twilight being characterized by intermediate singing levels (Homfeldt *et al.,* 2022). Habitat that moderates tidal amplitude has been found to be preferred by humpback whales as habitat, but no information is available on how tide influences their singing activity (Chenoweth *et al.,* 2011). The results of this study will provide baseline information about habitat use by male humpback whales during their winter months and inform future conservation and management efforts.

2. Material and Methods

2.1 Study site

Recorders were deployed in two locations Contreras Islands which are located outside the National Park Coiba and the second was located in Granito a location within the park borders. The park is located within the Gulf of Chiriquí and protects Coiba Island and 38 other islands and protects several species of threatened and migratory species. Previous work has shown the gulf is an important breeding area for BSG humpback whales. This breeding site is noteworthy because of the long migration from Antarctica and Chile that this population undertakes

(Rasmussen et al., 2007). A relatively recent update of this humpback whale population shows that more than half (54%) of all sightings included calves (Rasmussen & Palacios, 2014).

2.2 Recordings

Recordings were made using an autonomous and remote underwater recorder model SoundTrap 400 STD (frequency range 20 Hz-150 kHz ±3 dB; self-noise of less than sea-state in the bandwidth 100 Hz-20 kHz, and sensitivity of −203 dB re V/µPa) from Ocean Instruments (http://www.oceaninstruments.co.nz/). Recordings were made in Contreras (outside the park) and Granito (inside the park) from August to October 2020, using a recording cycle of 10 minutes per hour and a sampling rate of 48 kHz.

2.3 Song presence

For each 10-minute file I manually annotated in a separate Excel file with 1 the presence of whales; and for boats and with a 0 when whales or boats are not detected.

2.4 Environmental factors

The following environmental data will be collected: sea surface temperature (SST), tidal, and lunar information. SST will be obtained from this website https://seatemperatures.net/central-america-and-the-caribbean/panama/coiba-island/, and lunar and tidal data from https://tides4fishing.com/pa/oceano-pacifico/isla-cebaco.

3. Results

3.1. Humpback whale detection

A total of 2,370 hours were analyzed (Contreras h=1293 h, Granito=1077 h). After accounting for effort differences were significant differences in the proportion of files with humpback whale songs between sites (Fig.1). Contreras had a higher presence of whale songs than Granito (Two sample Z Statistic, P<0.01). When considering the month both locations had the highest presence of whale songs in August and September (P<0.01, Fig.2). This pattern is maintained when accounting for time of day (Fig.3). Overall, whale songs were detected throughout the day in both locations, but in September and October, there seems to be an increase in singing at night hours (Fig.3).

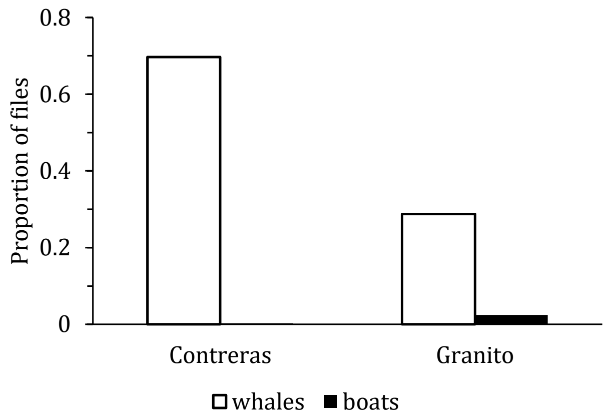

Figure 1. Proportion of acoustic files with humpback whale and boat sounds for Contreras and Granito, Panama. Data was collected from the months August-October in 2020.

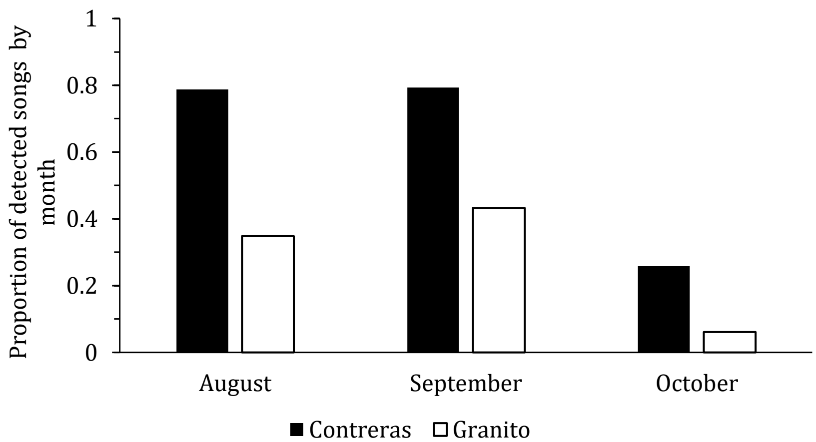

Figure 2. The proportion of detected humpback whale songs by month for sites Contreras and Granito, Panama. Data collected from 2020.

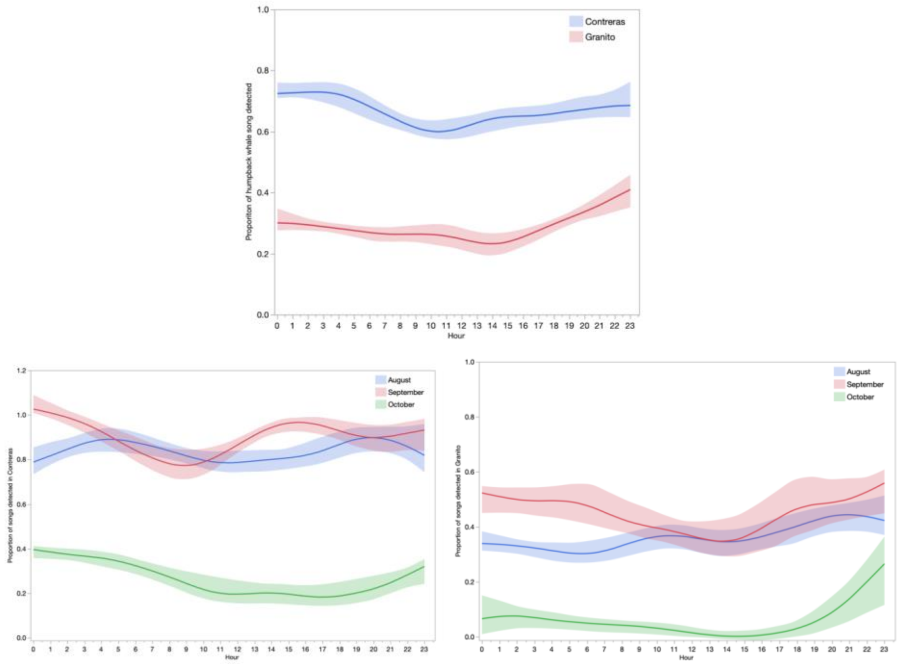

Figure 3. The proportion of humpback whale songs detected in Contreras and Granito per hour and by month.

3.2 Boat detection

Overall, the presence of boat sounds was low in both locations, but slightly higher in Granito (Contreras n=4, Granito n=26). Overall, boat presence occurred between 7 a.m. and 6 p.m. with a peak of presence at 9 a.m. (Fig 2).

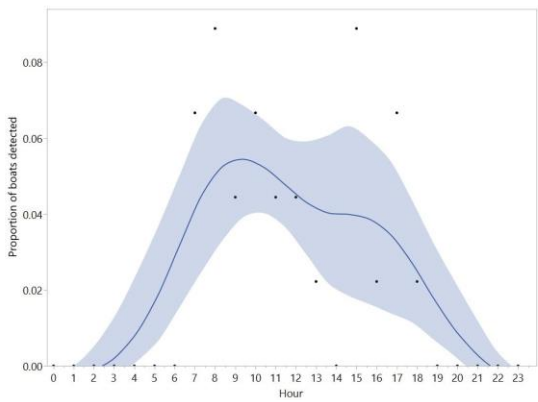

Figure 4. The proportion of boats per hour detected for Granito, Panama. Data was collected from the months August-October in 2020.

3.3. Environmental factors

There were no significant differences in sea surface temperature (SST) (Fig. 5). Furthermore, there were no significant differences in the proportion of whale song detections by moon phase. However, there is a trend to detect more whale songs from the waning gibbous to the new moon, and after that, there seems to be a slight decrease in detections. Finally, when considering the tide, there seems to be an increase in song detections at tides ranging from 1.5 to 2.5. However, tide data was highly scattered when combining song detection proportions from both sites (Fig.7). Since moon phases are split into eight categories over three months, there is potential for increased error due to the small sample size (Fig.6).

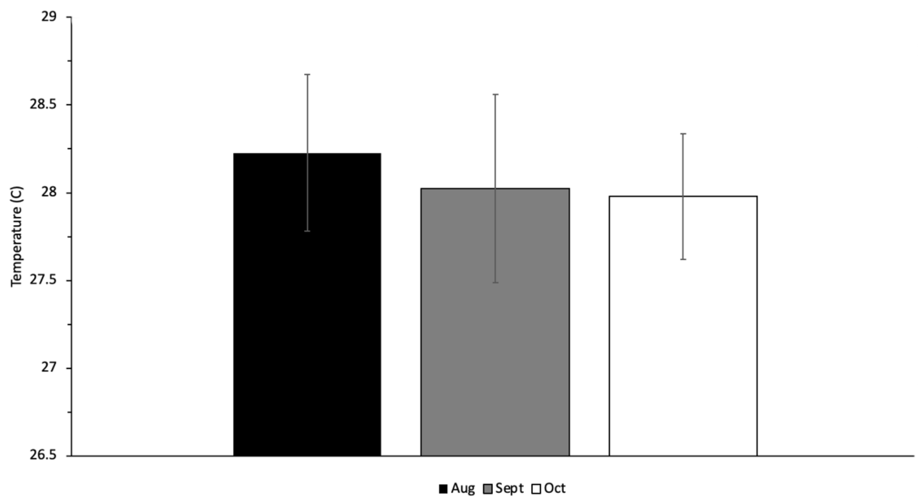

Figure 5. Mean sea surface temperature (oC) for Contreras and Granito by month in 2020. August represents the highest sea surface temperature (SST), with a steady decrease by month. Error bars were calculated from mean maximum and minimum temperature per month.

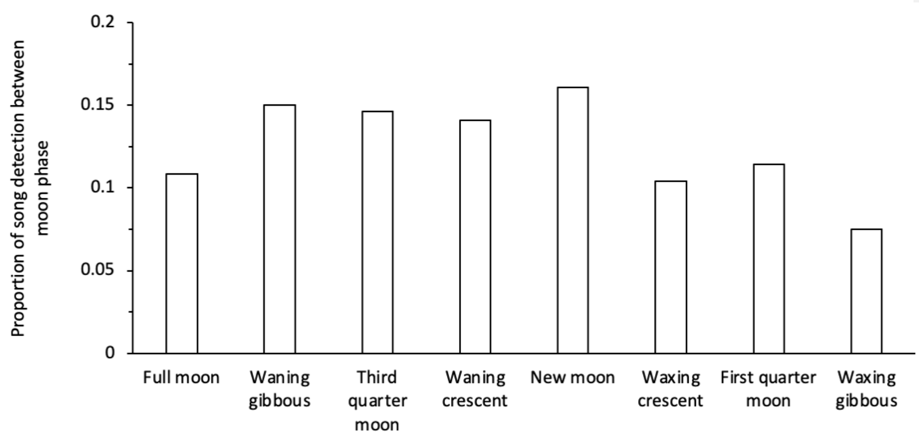

Figure 6. Proportion of humpback whale song detection relative to moon phase. Data combined for sites Contreras and Granito. Due to moon phase patterns, all phases occurred more than once for data collected except the third quarter and the new moon

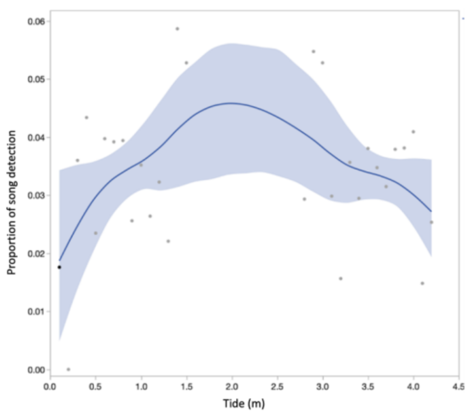

Figure 7. The proportion of humpback whale song detection compared to tide height for sites Granito and Contreras, Panama in August-October 2020.

4. **Discussion**

This study finds that the area outside the marine protected area, Contreras, had the highest proportion of humpback whale song detections independently of month. Whales sang throughout the day, with few a slight increase in singing at night hours in September and October in both sites. Given the low presence of boats and relatively similar SST, it is unlikely these patterns are explained by these factors. However, is important to highlight that this data was collected during the COVID-19 lockdowns of 2020 when boat traffic was at its lowest. May-Collado et al. (2023) found that in Cano Island National Wildlife Refuge boat traffic in 2020 decreased significantly while whale detections increased significantly throughout the day. In contrast, before and after Covid-19 lockdowns humpback whale song detections decreased when boat presence increased. The effect of boat presence on humpback whale singing activity has been shown in several other studies as well (Sousa-Lima & Clark, 2008, Blair *et al.*, 2016; Gabriele *et al.*, 2018) suggest boat traffic can impact whale singing patterns in breeding and feeding areas.

Although a higher proportion of humpback whale song activity increased from waning gibbous to new moon, the patterns observed concerning moon phases which are linked to tidal ranges merit further research. Low sample sizes of each phase due to the three-month timespan only allowed for no more than three occurrences of each phase. Other studies have found an inverse relationship between higher song detection and lower light levels (Sousa-Lima & Clark, 2008). However, a more detailed multivariate analysis would allow us to properly determine the contribution of moon phase and tide in humpback whale song-singing activity.

Although the marine-protected area in this study demonstrated a lower proportion of humpback whale singing compared to the non-marine-protected area, the influence of COVID-19 on boat activity

The high proportion of humpback whales at the site of Contreras could also provide a reason for the widening of the borders of this marine protected area. A further insight into habitat conditions between these two sites could provide more context for the difference in proportion.

Understanding how the proportion of humpback whales singing before and after COVID-19 lockdowns could help us examine the influence of marine protected areas on this important breeding ground for BSG. Since BSG is only present in this region during winter months, boat limitation during this time could aid in maintaining the singing activity during this time.

5. Conclusion

With high humpback whale activity at the study site outside of the marine protected area, extensions of the park boundary could ensure the protection of this breeding ground. Despite the limitations of COVID-19 on connections to boat activity and humpback whale singing, previous studies have found strong evidence of a negative correlation between these two variables. Limitations of large vessels could provide conservation of singing activity. The influence of environmental variables on humpback whale singing needs more research, only slight differences were found with moon phase and song activity. This study represents the baseline for male humpback whale habitat use. The continuation of this study can inform managers on how to ensure the protection of this species.

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Using unmanned aerial vehicles to estimate behavioral budgets of bottlenose dolphins in the Archipelago of Bocas del Toro, Panama

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Abstract: Attempts to study Bottlenose dolphin behavior through traditional methods of observing from boats fail to account for how anthropogenic noise sources may be impacting behavior on an individual and pod level. This study aims to compare boat observations of behavior to behavior observed from Unmanned Aerial Vehicles (UAV's). As UAV presence does not cause an immediate behavioral response within dolphin pods, the use of UAV's to observe bottlenose dolphin behavior could allow for a more complete understanding of individual and group dynamics. A better understanding of behavior, measured through behavioral activity budget estimates, could have long-term implications on management and conservation efforts within Bocas del Toro.

Key words: drones, behavior, dolphin, toothed whales, conservation.

1. Introduction

Most dolphin behavioral studies are done from boat platforms, limiting behavioral observations to only brief periods of time at the surface (Mann 1999., Ramos et al., 2023). Novel technological advances on unmanned aerial vehicles (UAVs) are enabling to have new perspectives of dolphin behavior. UAVs are inexpensive, can be flown from boat and land minimizing the observer impact while observing the animals in the field, allowing for detailed observations of their behavior (Castro et al., 2021). Furthermore, UAVs are suitable for dolphin species living in coastal and pelagic environments and allow the invasive tracking of the animals during follows (Ramos et al. 2023).

Bottlenose dolphins are social animals that live in fission-fusion societies (Mann 1999). A fission-fusion society is defined as a system in which animals may join and leave groups throughout the day making groups composition a dynamic property of this society (Evans et al., 2021, Walker et al., 2017). Dolphin group living can be influenced by differences in habitat, ecological, and behavioral conditions (Gowan et al., 2007). To understand the role of behavior in group formation, is fundamentally important to calculate activity budgets (the percentage of time that the animal spends in a certain type of behavioral state) (Huettner et al., 2021) and how investment in different behaviors change with environmental context (Walker et al., 2017).

In this study, I will calculate the behavioral activity budget of bottlenose dolphins in Archipelago of Bocas del Toro in Panama using footage from UAVs and observations from a boat-platform to determine the level at which boat-based observation might be missing important behaviors. The results of this study will help fill a gap between dolphin behavior and their welfare (Cubero-Pardo et al., 2007) and help understand how these dolphins use their space. For example, in Gulfo Dulce, Costa Rica, dolphins surface behavior correlated with prey spatial arrangement (Acevedo-Gutiérrez et al., 2000). Studies in the Bocas del Toro, also show that human activities can impact behavioral budgets. When dolphins were only in the presence of the research boat, they increase their time in foraging activity but, when in the presence of tour-boats they increase time in travel (Kassamali et al., 2019). Both studies highlight the importance of behavioral budgets in understanding habitat use and the impact of human activities.

2.1 Methods and Materials

2.1. Study site

The study tool place in the Archipelago of Bocas del Toro (BDT) in Panama. This location is home to a genetically isolated population of bottlenose dolphin population that consist of 72 to 87 dolphins, with both males and female showing high levels of philopatry (Barragan-Barrera et al., 2017, Kassamali et al., 2019). This dolphin population is also the main target for local dolphin watching activities and the effects on their acoustic behavior have been documented extensively (review by May-Collado et al., 2017)

2.2 Unmanned drone flights:
In 2023 behavioral observations were done from two platforms, a small boat and a mini-DJi drone using Ramos et al. (2023) flying protocols. Flights were made from May 13 to June 16 and lasted between 5 to 20 minutes. The drone distance from the animals which was ocean surface is 50-200m to avoid any disturbance (Giles et al., 2021). From the drone footage we obtained the following information group sizes, group composition, and behavioral dynamics (Ramos et al., 2022). Follows were done until the UVAs battery was low or the operator could not follow the dolphins due to diving too far below the surface or moving too fast for the range of the drone.

2.3. Ethogram:
To calculate behavioral activity budgets, the Behavioral Observation, Research, Interactive Software (BORIS, version 8) (Friard and Gamba 2016) was used. The drone footage open in BORIS and the activity state, code, and definition of behavior were scored in the program. The duration of each behavioral event and the time that the event occurred was extracted. The data was then used to generate ethogram tables for focal animals on each group (Kassamali et al., 2019).

2.4. Behavioral data
For each video observed in BORIS, the following data was collected, number of individuals in the group, group composition (number of adults, juveniles, calves), and their overall spatial distribution as they move through the study site. For comparison purposes between drone and boat dolphin general behavioral states for foraging, socializing, travel, milling, and rest were defined following Kassamali-Fox et al., (2019).

3. Results
A total of six days and 728.5 minutes were analyzed from May 19th, 2023, to May 25th, 2023. This is about 65.7% of the total drone data collected. Except for milling all behaviors were observed with both drone and boat platforms (Fig.1). However, a greater diversity of social behaviors was reported with the drone, including chuffing (n=6), leaping (n=1), sexual contact (n=1), tail slap (n=6) and touching (n=6). Six interactions with boats were recorded during boat surveys and only one when flying the drone.

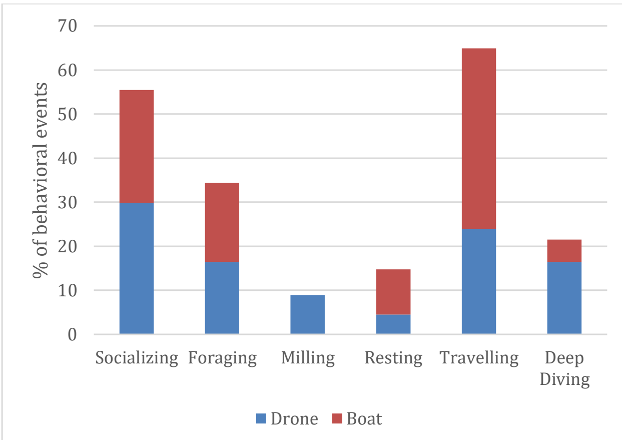

Figure 1. Percentage of dolphin behavioral of events for six behavioral contexts observed using drone and boat platforms from May 19 to 24, 2023 at Bocas del Toro, Panama.

Regarding time dedicated to each behavior, I focused on data taken only with the drone on May 20, 22, 23, and 25, and on the four most common behaviors foraging, socializing, resting and traveling. As shown in figure 2, on average full group observations of dolphins show they spent on average more time resting than socializing, traveling, and foraging. However, there was a lot of variation in how much time it was invested in resting from 10 to 3770 seconds (Table 1).

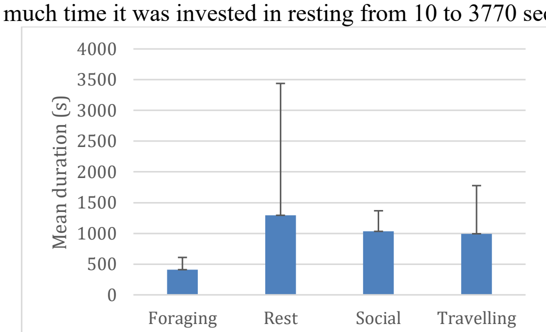

Figure 2. Time dedicated to each behavioral activity in seconds for full group observations in May 20, 22, 23, and 25 of 2023 at Bocas del Toro.

Table 1. Summary statistics of time dedicated to each behavioral activity.

Behavior	*Mean*	*SD*	*Min*	*Max*
Foraging	410.976	197.5881	203.536	596.961
Rest	1295.192	2144.037	10.944	3770.326
Social	1034.022	333.685	653.555	1276.975
Travelling	994.915	782.2911	414.314	1884.512

4. Discussion

The results of this study show that observation from drones yield more detailed behavioral data than traditional boat-base observations. Our results represent a larger number of observations can be made while using drones. Drones provide an increased repeatability of data relative to boat-based observations for an increased sample size of datasets (Torres 2018). Using drones as mode of observation provides a valuable tool in understanding the behavioral activities without being invasive such as when on a boat-based observation (Christiansen 2016). This noninvasive approach delivers a representation of how animals act while not being under duress of a boat (Dawson 2017).

Milling was a behavior noted and calculated for drones but not with boat-based observations. This adds to the hypothesis, using drones to view dolphin behavior will yield more visible behaviors than boat-based observations. Conducting regular behavioral observations can give a better indication of how external environmental changes effect dolphin welfare (Huettner 2021) and use this for conservation efforts against an increase in tour boat activity (Kassamali-Fox 2019).

Based on current research there is a lack of behavioral diversity recorded for bottlenose dolphins that need to be computed to fully analyze the welfare and behavior connection (Miller 2021). With access to drones, it opens a door to collecting greater diversity of social behaviors such as chuffing, leaping, sexual contact, tail slap, and touching. Understanding the duration of time each behavior takes can a help assess the effect of boat interactions on the welfare of the dolphins. Dolphin's time foraging significantly decreases and time spent travelling increases in the presence of a boat (May-Collado 2021). The data showed there was six boat interactions in comparison to one interaction with a drone. This correlates with figure 1 showing an increased presence of travelling for the boat. The variation in rest for figure 2 would show that dolphins spend more time resting with drone observation and would attest to dolphins not being disrupted from boat-based observations.

5. Conclusion

This study examines the utility of drones for data collection and analysis of behavioral budgets of bottlenose dolphins. To further expand the knowledge gained from behavioral budgets done using drones can connect the visual behavior being observed to the sound scape in the same location. BORIS software can run audio and video footage at the same time, allowing the usage of these two ethograms to connect the behavioral budget to the sound produced. This can show the coinciding behavior to communication for a deeper understanding of how the welfare of the bottlenose dolphin can be understood while using acoustic activity with the behavior displayed.

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Dolphin communication before, during and after the COVID-19 Lockdown

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Abstract:
Dolphins rely on sound to survive. The Covid-19 pandemic has provided a unique opportunity to study the effect of boat traffic on dolphin communication. I used data collected from remote autonomous recorders during data from pre-lockdown, lockdown, and post-lockdown periods at Dolphin Bay, Panama to this study the effect of boat traffic on dolphin acoustic presence and the acoustic structure of their communicative signals. The results indicate that boat presence increased fivefold post lockdown, and dolphin presence decreased. The pattern is maintained throughout the day. Furthermore, dolphins produce shorter and less modulated signals that were higher in most frequency variables post-lockdown. These signal characteristics are likely solutions to increased low-frequency ambient noise associated to tour-boats and may help them to compensate for signal masking. These findings highlight the impact that noise associated to tour-boats can have on dolphin habitats and their communication. Measures to regulate tour-boat presence by implement national regulations are an effective way to minimize the negative effects of noise.

Keywords: anthropogenic noise, underwater noise, Cetacea, Delphinidae, Panama, Bocas del Toro, whistle

1. Introduction

Underwater ambient noise has been increasing by 3.3 decibels since the 1950s in association with commercial shipping (Frisk, 2012). In coastal habitats, the rise of private boats, fishing, commercial shipping, and tour-boats are also reported to contribute to the increase in underwater noise around the globe (Erbe et al., 2019). The detrimental impact of increasing noise levels in the ocean has been shown on a variety of marine taxa (Erbe et al., 2019). The impact of underwater noise associated to human activities was highlighted during the Covid-19 lockdowns in 2020. Widespread mandatory lockdowns and stay-at-home orders severely limited human mobility and activities on land and in the ocean. During this period, several studies documented a decrease in ambient noise levels and changes in the communication range and signal acoustic structure of animals both in land and in the ocean (birds: Derryberry et al., dolphins: Gagnes et al., 2022, and whales: May-Collado et al., 2023).

Dolphins produce a variety of sounds to communicate, among those are a category of noises called whistles. Whistles are frequency modulated sounds that vary in duration and are used to convey information about the individual identity, behaviors, and environment. Several studies have found that dolphin whistles can vary in frequency and duration, and this variation can provide insights into their ability to communicate in various environmental conditions (May-Collado and Wartzok 2008). For example, Morisaka et al., (2005) found that dolphins produce low frequency whistles with little modulation in noisy environments, possibly as a strategy to avoid signal masking and attenuation by high frequency noise sources from small boats. This modulation is also commonly referred to as the points of infection in the whistle.

In Bocas del Toro, Panama there is a resident population of bottlenose dolphins that supports the largest dolphin watching industry in the country (May-Collado et al., 2014). Previous research has demonstrated that noise associated to tour boats impact these dolphins' communication, and results in changes in signal frequency range, modulation, and duration (May-Collado and Wartzok 2008, Perez-Ortega et al., 2021, Gagnes et al. 2022). Gagnes et al. (2022) showed that impacts of tour boats were evident given data from before and during the primary Covid-19 lockdown. During the lockdown their primary habitat at Dolphin Bay showed a significant decrease in ambient noise levels, and as a results dolphin communication range increased. In addition, the dolphins change their whistle acoustic structure (i.e., decrease in modulation), where more acoustically present, and show a more diverse repertoire of whistles suggesting a higher presence of dolphins in the bay. Looking at data regarding the frequency and acoustic quality during these varying time will give insight on indicators of stress in the population (Yang, W C, et al., 2021).

However, this study only compared acoustic activity and structure before and during the lockdowns. As many communities and tour companies recovered, this study sicks to determine the acoustic response of the dolphins to post-Covid 19 lockdowns. I hypothesize that dolphin's acoustic presence wand whistle acoustic structure will be like conditions during pre-lockdown. Specifically, I predict that dolphin acoustic presence will decrease while boat acoustic presence will increase post-lockdown. Furthermore, I predict that whistle frequency will decrease, and modulation increases to similar levels as pre-lockdown. The results of this study will show that a shift in boat traffic activity can generate changes in dolphin habitat use and in their communication.

2. Methods

The data used in this study was collected in the Bocas del Toro, Panama, on an archipelago commonly called Dolphin's Bay (Gagne et al., 2022). This bay is the home to many bottlenose dolphins, and is especially important for moms and calves, given its semi-enclosed nature (May-Collado et al., 2014). As this area is home to quite a few resident dolphins, over time it has become a commonly-used site for dolphin watching and a large attraction for tourists visiting the area. Everyday boats, at an average length of 10m, will arrive in the area between 9 in the morning to 12p.m (Perez-Ortega et al., 2021). These boats will circulate the bay until they come across a pod of dolphins, which they will then follow for approximately 36 minutes at around 50 m. I is clear that the bottlenose dolphins residing in this area have very frequent exposure to boats and humans in general.

2.2 Recordings

Audio samples were collected using underwater recorders at a sampling rate of 48 kHZ. These recorders were mounted to a pole at 1.5m above the sea floor, anchored with a concrete block to keep them stable. In order to get a good idea of the soundscape from before COVID-19, during COVID-19, and after COVID-19, three different years of audio files will be considered, 2019, 2020, and 2021. Audio files will be analyzed with the RAVEN Pro software (Center for Conservation Bioacoustics, 2014).

2.3 Acoustic Analysis

The audio files used were subject to analysis. Each 10-minute audio was screened for both dolphin noise and boat activity. If dolphin communication was noted, sound type (echolocation or whistle) was recorded. Documented whistling was analyzed using the RAVEN Pro software to observe minimum frequency (Hz) (the lowest frequency portion of the selection), maximum frequency (Hz) (the highest frequency portion of the selection), frequency range frequency (Hz) (the difference between maximum and minimum frequency), duration (s), and peak frequency (Hz) (frequency in the contour with greatest energy), according to the specifications of Gagne et al (2022).

3. Results

3.1 Presence absence of dolphins by year and Covid-19

The soundscape of Dolphin Bay varied greatly between the pre-lockdown, during lockdown and post-lockdown periods. The proportion of recordings with boats was almost four times higher and in pre-lockdown and during lockdown (Fig.1). In contrast, dolphin acoustic presence was highest during the lockdowns.

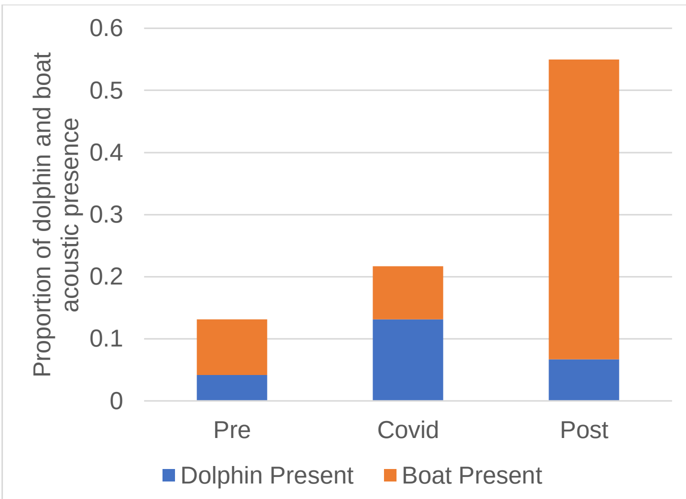
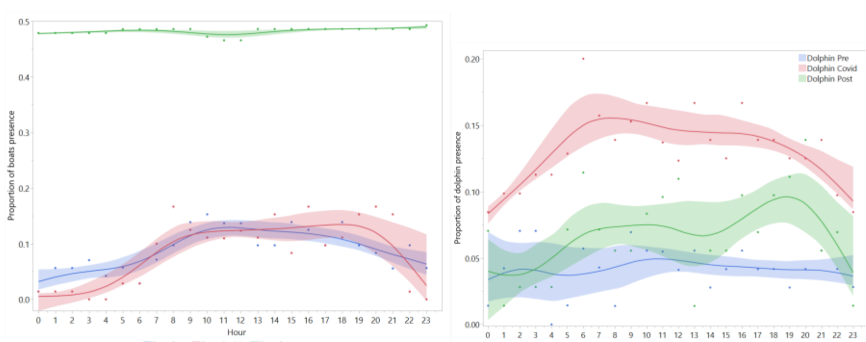


Figure 1. Proportion of dolphin and boat sounds per hour in pre-lockdown (2017-2018), during Covid-19 lockdowns (2020) and post-lockdown (2022-2023)

3.2 Presence of dolphins and boat by time of day

The patterns described above were maintained during the day. Figure 3a shows that the proportion of boat presence was higher throughout the day during post-lockdown. There were no differences in the presence of boats in pre-lockdown and during the lockdown. As shown in Figure 3b, dolphin presence was higher throughout the day during the Covid-19 lockdown. During the lockdown dolphins presence was higher at all hours but particularly between 7:00 to 11:00. In contrast pre-lockdown dolphin presence varied little, and in post-lockdown dolphin acoustic presence appeared to be higher between 16:00 and 20:00.



A. Boats B. Dolphins

Figure 3. Proportion of dolphin and boat sounds per hour in pre-lockdown (2017-2018), during Covid-19 lockdowns (2020) and post-lockdown (2022-2023). The horizontal lines represent the

mean, and the shadowed area is a smoothing spline and bootstrap confidence of fit with a lambda of 0.05.

3.3 Changes in acoustic structure

There were significant differences in the acoustic structure of whistles between pre-lockdown, lockdown, and post-lockdown. Dolphins produced whistles with significantly higher frequency in post-lockdown year than in pre-lockdown and lockdown years. During post-lockdown dolphin produced whistles with higher low frequency (X2=406.3, df=2, p<0.0001), high frequency (X2=120.8, df=2, p<0.0001), and peak frequency (X2=131.9, df=2, p<0.0001) (Fig 5). In contrast, dolphin whistle were lower delta frequency (X2=16.6, df=2, p=0.0002) and PFC Number of Inflection Points (X2=703.7, df=2, p<0.0001) and shorter in duration (X2=95.4, df=2, p<0.0001) during post lockdown than in pre-lockdown and lockdown (Fig.6).

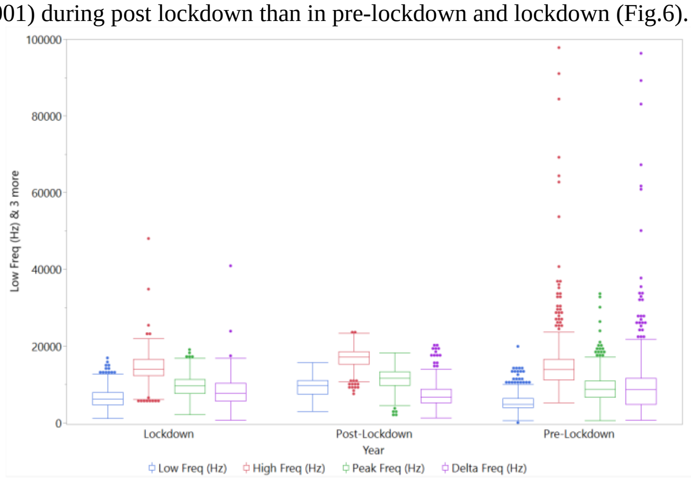
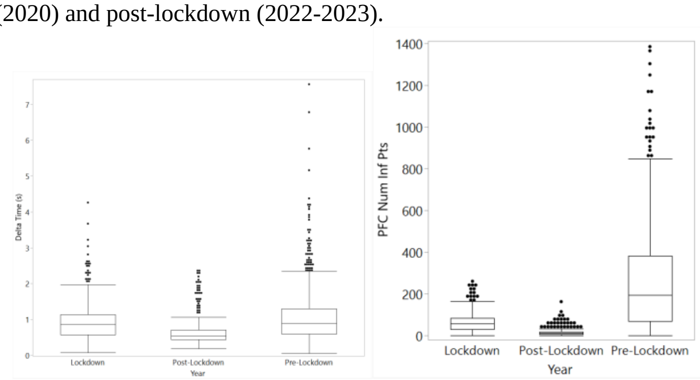


Figure 5. Dolphin frequency variation in pre-lockdown (2017-2018), during Covid-19 lockdowns (2020) and post-lockdown (2022-2023).



a. Duration b. Modulation

Figure 6. Variation in dolphin whistle duration and modulation measured as the peak frequency contour number of inflection points in pre-lockdown (2017-2018), during Covid-19 lockdowns (2020) and post-lockdown (2022-2023).

4. Discussion

Comparisons between pre-lockdown, COVID-19 lockdown, and post-lockdown periods at Dolphin Bay revealed significant changes in the acoustic presence of boats and dolphins, and in dolphin whistle acoustic structure. These changes highlight the impact that unregulated tour boat activity can have on dolphin habitat and communication.

Gagne et al., (2022), comparison of pre-lockdown and Covid-19 lockdown periods suggest noise levels decrease in Dolphin Bay during the lockdown due to a shift in boat activity from tour-boats to primarily transport. Their findings also showed that dolphins were more present as indicated by an increase in acoustic presence and a higher diversity of whistles during lockdown. These results are further supported by a study that compared areas dominated by transport boats versus dominated by tour-boat presence (Perez-Ortega et al., 2021). In both studies dolphins in the presence of transport boats dolphins produced whistles that were higher in frequency, but less modulated and lower in frequency.

Interestingly, our findings suggest that dolphins during post-lockdown experience almost five-fold increase in boat presence but remain producing whistles at higher frequency and little modulation. As in Gagne et al., (2009) we measured modulation as PFC Num Inf Points, and the authors postulated that a decrease in this variable likely suggests a decrease in stress levels as has been postulated by other authors (Esch et al., 2009). However, it is important to highlight that this variable is sensitive to background ambient noise levels, and spikes in the contour can occur when overlapping with noise.

Throughout the selection of whistles, I tried to minimize this effect by selecting only whistles with high SNR, but it is possible that the variation observed in Figure 6b in this variable is the product of noise. However, if dolphins are indeed produced less modulated whistles, a simplification of whistle contours would allow these dolphins to minimize masking from boats, as simpler whistles propagate better in noisy environments (Lesage et al., 1999; Fouda et al., 2018).

Finally, our data supports Gagne et al., (2022) results that dolphins in Dolphin Bay produced shorter whistles during the Covid-19 lockdowns. This result supports the potential impact of increased noise (due to higher boat presence) in modulation, as shorter whistles tend to be less modulated (Perez-Ortega et al., 2021).

5. Conclusion

The shifts in boat presence through the pre-lockdown, during lockdown and post-lockdown timeline, allowed research to be conducted at varying conditions. In conjunction with Gagne et al., (2022), it was found that during Covid-19 there was a greater number of vocal dolphins than there was before or after the lockdown. The detected presence of boats increased tremendously after the lockdown came to an end. The rise of ecotourism in the post covid-world is likely the culprit, as there has been recent airport expansion in the area to better accommodate for the large-scale dolphin watching taking place. The data from this study shows a range of impacts from this uptick in anthropogenic noise and could be used to inform future legislation when it comes to protecting the dolphins in Dolphin Bay, Bocas Del Toro.

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The unit repertoire of Southeastern Pacific humpback whales at their breeding area in the Gulf of Chiriqui, Panama

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Abstract: Male humpback whales (*Megaptera novaeangliae)* are known to produce long and complex songs during their breeding season. In order to understand how these songs change over time, humpback whale songs from the Southeastern Pacific population Breeding Stock G were recorded over multiple years in the Gulf of Chiriquí, Panama. A total of 70 distinct units were identified, with two units present over all years studied. The percentage of novel, stable, and recalled units varied across all years studied, with the highest rate of unit change occurring in 2013. This study provides the first unit repertoire analysis for this humpback whale population in order to understand the pace of change of humpback song, and to understand the potential effects of climate change and subsequent changes in sound propagation on humpback communication.

Key words: Bioacoustics, Whale Song, Cetaceans, Marine Biology, Population Ecology

1. Introduction

Humpback whales are highly vocal animals, with males producing complex and long song displays primarily during the breeding season. (Payne and McVay 1971, Dunlop et al. 2007). Decades of research on humpback whales show that songs are learned via "horizontal transmission", and that males within the same reproductive population conform to a conspecific song over the course of a breeding season (Herman 2017). As male humpbacks migrate, they may encounter other males from the same or other populations, providing opportunities for new song components to be introduced into their repertoire (Noad et al. 2000). Therefore, the rate at which a song changes depends highly on the level of contact between individuals (Eriksen et al. 2005).

The song structure of the humpback song was first described by Payne and McVay (1971) , as a string of hierarchical components consisting of subunits organized into units (in some cases such as Pace et al 2010, though not typically used), units organized into phrases, phrases organized into themes, and themes organized into full songs (Payne and McVay 1971, Norris et al. 2000, Cholewiak et al. 2012). These components can define populations, allowing for comparison across populations and inferences of song similarity or difference that could reflect the degree of connectivity between populations (Darling et al. 2019). In this study, units were chosen as the components for comparison because they are the smallest components to be identified and catalogued in similar studies, such as Chereskin et al. 2019, and are fundamental building blocks of the overal song structure.

There are currently 14 distinct population segments (DPSs) of humpback whales worldwide (Bettridge et al. 2015). The Southeastern Pacific population of humpback whales, also called Breeding Stock G (BSG), migrate from feeding areas in the Antarctic Peninsula and Fuegian Archipelago of Chile from June through November to the Pacific coasts of Central America (Rasmussen et al., 2007 2012). In Panama, BSG whales breed off the Gulf of Chiriquí where singers, competitive groups of males, and females with their calves are regularly observed (Acevedo et al. 2007, Rasmussen et al. 2011). Despite our growing understanding of the ecology of these whales in Panama, there is only one study describing the song of BSG humpback whales, and the study is from Costa Rica (Chereskin et al., (2019). The authors of this study found that the 2016 and 2017 BSG whales song consisted of four themes, 10 phrases, and 12 units. In a study by Magnusdottir et al. (2015) conducted off the coast of Iceland, the authors found that unit repertoire included a host of unit types that occurred frequently in songs across all years studied, while many other unit types present were not repeated. In another study, Mercado and Perazio (2022) showed a gradual morphing of units within songs, suggesting that humpback whales possess control mechanisms and vocal flexibility that can result in novel units in their repertoires. To better understand how humpback whales build their songs, it is fundamental to understand the diversity of the building blocks of their song - the unit.

It is the aim of this study to describe the unit repertoire of BSG whales at their breeding site in the Gulf of Chiriquí using a database of nine years (over a total timespan of twelve years) to estimate unit occurrence and pace of change. This will help determine the way in which humpback whales in this breeding area evolve their song. I hypothesize that, as in Iceland, some units will be more common across years than others. The proportion of new and phased-out units may be the results of contact of individuals along their migratory route, overlap with other populations at their feeding areas (where singing does occur), individuals shifting their breeding areas, and/or adjustments to adapt to environmental changes in their habitat (e.g., soundscape).

The results of this study are key to understanding the impacts of noise pollution underwater on this highly vocal animal. The effects of climate change are expected to impact their migratory behavior and their habitat (e.g., changes in sound propagation) (Meynecke et al. 2021, Derville et al. 2019, Rossi-Santos 2020). By understanding the makeup of their unit repertoire and addressing how humpback whale unit composition has shifted over time, this study will lay the groundwork for the development of models to predict the direction of these changes in the future.

2. Materials and Methods

2.1 Study Area

This study took place in the waters around the archipelago of Islas Secas in the Gulf of Chiriqui of Western Panama (depth <300m). The Gulf of Chiriquí is an important breeding area for humpback whales and has a notably high annual proportion of mother-calf pairs (Rasmussen and Palacios 2014). Findings from a 2017 photo-identification study suggest that southern humpbacks (BSG) are more likely to migrate to this area than their northern counterparts, which is consistent with a previous genetic study done in the area (Acevedo et al., 2017). This latitudinal preference may reflect behavioral or migratory patterns, or maternal fidelity and natal philopatry, both of which vary regionally (Baker et al. 1990, 1994; Medrano-González et al. 1995; Palumbi and Baker 1994; Pardini et al. 2001; Lee et al. 2007; Baker et al. 2013; Carvalho et al. 2014).

2.2 Recordings

Recordings for this study were collected manually from a boat and using various recording systems including a Cetacean Research Technology SQ26-08 plug-in powered hydrophone with an effective sensitivity of -194dB, re 1V/µPa, and a frequency response of 2 Hz to 50 KHz on a 10-meter cable. Whales were also recorded using a Zoom H4n Pro recorder at a sampling frequency of 48 kHz. Usable recordings were collected from August to early September of 2007, 2008, 2009, 2013, 2014, 2015, 2017, 2018, and 2019, and numbered 18 in all. To locate and isolate units, recording files were opened in Raven Pro 1.6 build 37 (2019; Cornell Lab of Ornithology) and a spectrogram was generated with a fast Fourier transform (FFT) size of 2,048 points, an overlap of 50%, a 512-sample Hann window All selected songs had signal-to-noise ratio (SNR) greater than 6 dB, following Chereskin et al. (2019).

2.3 Unit Classification and Analysis

From selected songs, units were identified and labeled in the spectrogram, and then classified using a modified version of the Djokic (2019) classification key. This key allows for natural variation in unit structure while still allowing for the grouping of like terms (Helweg et al., 1998; Rekdahl et al., 2018b). Units were classified based on the five major characteristics: contour type, tone type, peak frequency, duration, and harmonics presence. Contour types consist of upsweeps (contour increasing in frequency), downsweep (contour decreasing in frequency), flat (continuous frequency with the dominant frequency varying between 100Hz), arc (increasing and then decreasing in frequency), U-shape (decreasing and then increasing in frequency), frequency-variable with tail (shape “N” is repeated once or more, and the final part of the unit doesn’t resemble the rest and looks like a tail), frequency variable without tail (shape “N” is repeated once or more), frequency-variable with high tempo (indicating fast changes in frequency), and pulsed sounds (no visible contour).

In addition, contours with tonal contours were classified as purely tonal, pulsed, mixed, noisy tonal (with deterministic chaos), or raspy (sounds raspy but not pulsed, looks tonal at FFT 2048). Contours were then classified based on their peak frequency (frequency at which most of the energy is) as ~1000 Hz, < or equal to 5000 Hz, and >5000 Hz. The key was edited (from designations of ~100hz, ≤500hz, and >500hz to designations of ~1000hz, ≤5000hz, and >5000hz) to account for the abundance of units with a peak frequency class >500hz and a lack of units with a peak frequency of ≤500hz.Contour duration was classified as ~ < or equal to 1sec, >1 sec. Contour harmonics presence was classified as dense, sparse, or absence (no sidebands present).

The result of the key was a five-digit code that described each unit's contour as its name. Summary statistics were then performed to describe the number of unit types present across the years studied, and the rates of change in new unit introduction and unit phase-out. Units were classified as new (novel units not described in any of the years previous), stable (units repeated from the previous year), and recalled (units not present in the previous year but present before that). The rate of unit change was calculated discreetly for each year as the number of phased out units subtracted from the number of new units, all divided by the total number of units present.

3. Results

3.1. Unit repertoire composition

A total of 18 song files were analyzed (2007=1, 2008=3, 2009=2, 2013=1, 2014=1, 2015=3, 2017=3, 2018=3, 2019=1). From these files a total of 70 distinct unit types were identified over the span of nine separate years (Fig. 1), and only two units were present across all nine years (unit 31221 and unit 21221). The amount of discreet units present per year ranged from 6 in 2013 to 22 in 2008. This was likely due to the lack of viable song files in 2013 (only 1 was analyzed) and the abundance of viable song files in 2008 (3). On average, years with a higher number of files analyzed had a higher number of units present. Due to this discrepancy, when analyzing the unit makeup of each year (as in Figure 2), only percentage of the entire makeup was considered for each category.

Figure 2 shows the percentage of new, stable, and recalled units in the unit repertoire, and the variation in the presence of each within and between years. For example, the song of 2009 consisted of primarily stable units 59.1.% and new units made up 40.9% of the song. In contrast, in years 2014 and 2015, songs consisted primarily of new units (43.8% to 47.4%,), followed by recalled units (37% and 21%), and stable units representing only 18.8% to 31.8% .

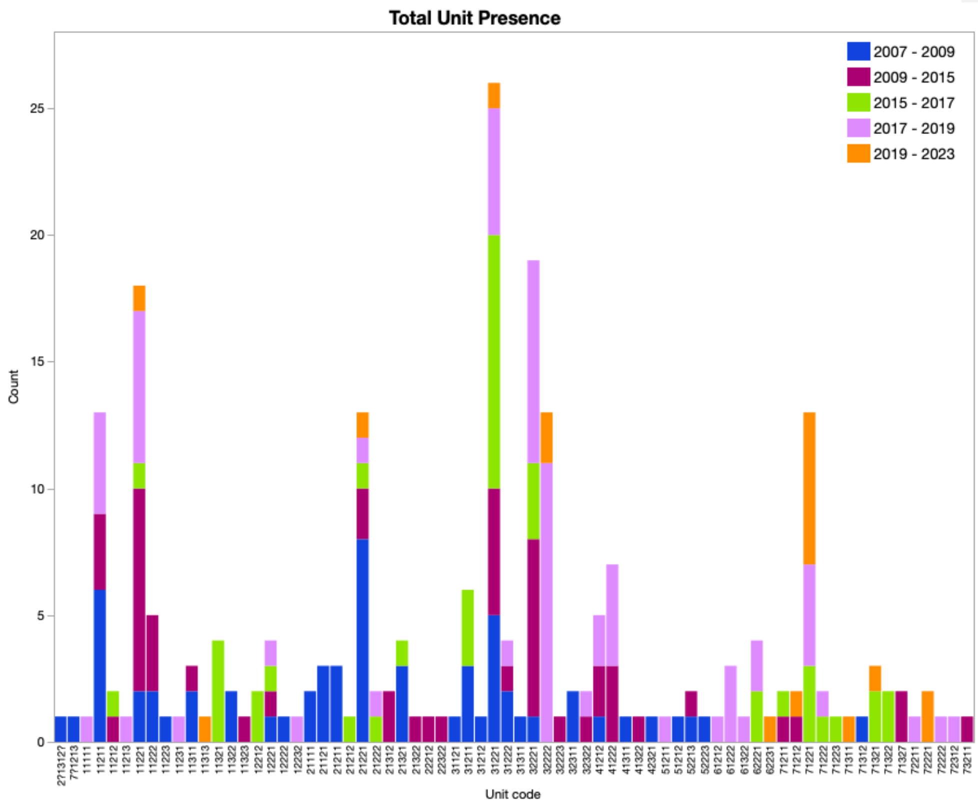

Figure 1. Total unit composition from 2007-2009, 2013-2015, 2017-2019. Each individual unit code is represented as a bar, showing the number of occurrences, and color-coded by year.

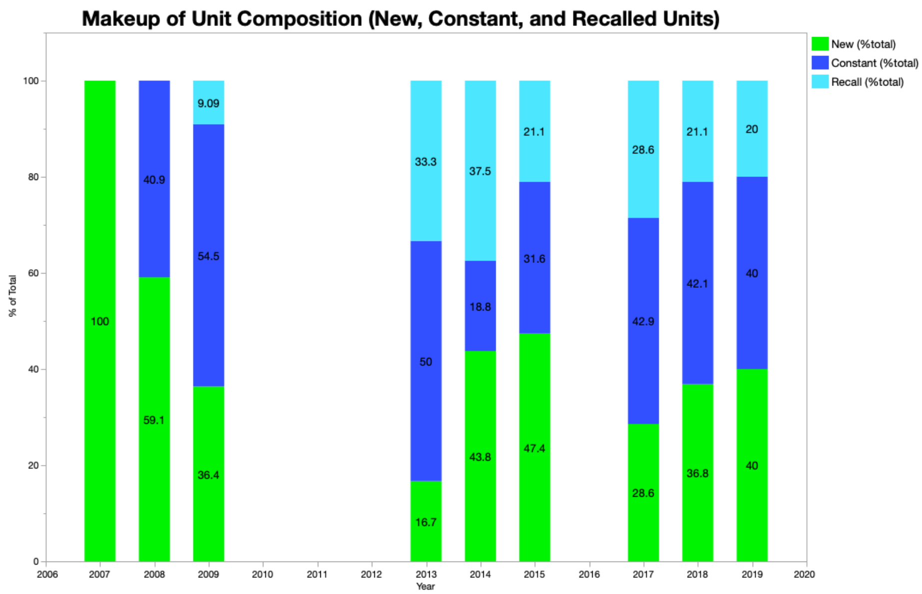

Figure 2. Percentage of new, stable, and recall units in the repertoire for each year.

3.2. Unit acoustic structure

Overall, unit contour peak frequency was relatively similar across years, while duration was more variable (Fig.3). However, in 2017 to 2019, unit contour mean peak frequency decreased while contour duration increased when compared to previous years (Fig.3). The two units present in all years both had a peak frequency equal or less than 5000 Hz. Other units, such as unit 41212, displayed dense harmonics presence compared to other units.

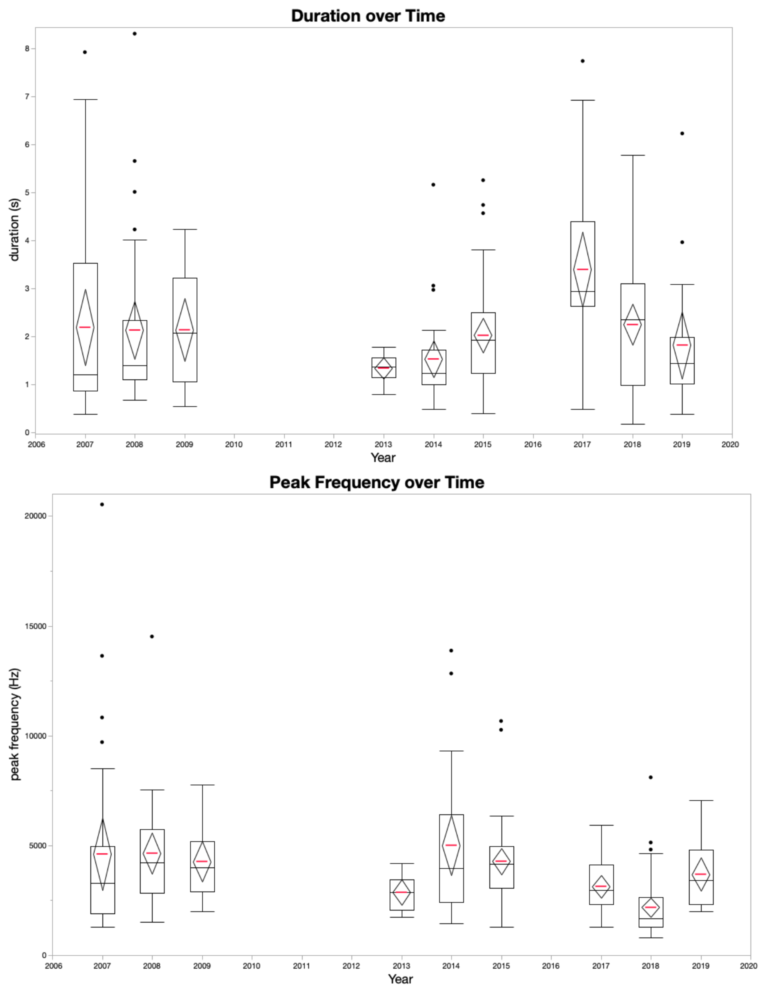

Figure 3. Peak frequency (a) and duration (b) variation across years, shown in box plots with the upper quartile, mean, and lower quartile indicated. Diamonds show 95% confidence intervals, and the red line within each diamond shows the mean for each year.

3.3. Rate of unit change

The absolute unit rate of change per year is shown in Table I. Overall, the lowest reported unit rate of change was in 2015 and highest in 2013. Negative values indicate a high rate of phase-out units, and 2019 had a rate of change of -1.1 indicating this year had a remarkably high number of phased-out units.

Year	(#N-#PO)/(#Total)		Year	(#N-#PO)/(#Total)	
2007	1		2007	1	
2008	0.27272727		2008	0.27272727	
2009	1.09090909		2009	-1.09090909	
2013	1.16666667		2013	-1.16666667	
2014	0.66666667		2014	0.66666667	
2015	0.05263158		2015	-0.05263158	
2017	0.64285714		2017	-0.64285714	
2018	0.05263158		2018	0.05263158	
2019	1.1		2019	-1.1	

a. b.

Table I. The absolute values of rate of change per year studied (a) and the actual values of rate of change per year studied (b), calculated as the number of new units minus the number of phased-out units, all over the total number of units present for that year.

4. Discussion

This study finds that Southeastern humpback whales reproducing in the Gulf of Chiriquí have a relatively rich unit repertoire, with 70 discreet unit types represented across 237 recorded units. Other studies have reported a higher number of units in a shorter period - for example, Magnusdottir et al., (2015) reported a total of 2810 recorded units in a period of five years. This discrepancy is perhaps due to the difference in methods: Magnusdottir et al. analyzed data collected over 5-month periods during the feeding season of multiple populations in one area, while this study used data collected over ~6 week periods during the breeding season on one specific population. There may also be the issue of unit classification, for which Magnusdottir et al. (2015) used different criterion (automatic unit identification in the program Ishmael - Mellinger et al., 2011) than that used in this study, as did Mednis (1991) which found 15 unit types over only one year using only contour type and frequency. Maeda et al. (2000) found only 32 unit types across 4681 recorded units over only 6 years around the Ryukyu Islands of Japan (half the length of this study, and half the number of unit types) using similarly in-depth classification methods.

The two units present in all years, 31221 and 21221, both had peak frequencies of greater than 1000 Hz and no higher than 5000 Hz. Ross (2005) calculated an average ambient noise level of 50Hz in the North Pacific increasing by 5.5 dB/decade since the 1970s, though noise generated by ocean traffic in the North Pacific has become more variable of late (Andrew et al., 2011). This is well under the peak frequencies of the most heavily-used units, indicating that selective environmental pressure factors into unit use and retention. Clark (1982) found that the calls of the bowhead whale and southern right whale remained lower in peak frequency than the ambient noise range, and Maeda et al. (2000) found that the majority of humpback units remained higher in peak frequency than the ambient noise range, both corroborating this theory. Sound propagation is known to increase with higher temperatures, and Beer's law states that sound intensity is inversely proportional to sound frequency and water density (which decreases with temperature) (Rafferty). Therefore, a lower sound frequency has a higher intensity, especially in warmer water. This, in conjunction with ambient-noise-induced constraints, may explain the prolonged use of units within a 1000-5000 Hz range.

The Chiriquí humpback whale unit repertoire also varies in composition of new, stable, and recall units vary with year, with the year 2015 experiencing the highest turnover of units. However, the overall rate of change in this population appears to be gradual, corroborated by studies of Northern populations that show a slow-evolving song, such as Zandberg et al. (2021). The results of this study contrast with a recent study where Southeastern humpback whales from Ecuador went through a song revolution in 2018. Schulze et al. (2022) reported a song spread in an eastward direction, with whales in Ecuador adopting a song from French Polynesia suggesting vocal connectivity across the entire South Pacific Ocean basis. This study's data for 2018 does not capture this change at the level of units - in fact, the rate of unit change in 2018 was among the lowest (0.52) recorded. Zandberg et al. (2021) also classifies Southern population songs as "revolutionary", experiencing a lot of change over a short period of time, contrasting the conclusions of this study. However, my results may change with an expansion into phrase and theme analysis and with future peer-review. Darling et al. (2019), has reported whale song with a varying rate of song change over three years across several studied populations of North Pacific humpback whales, and Eriksen et al. (2005) found differing rates of change across seven years studied. Generally, I found that the number of phased-out units was far greater than the number of new units each year, except in the case of 2019, where the rate of change was -1.1, indicating a much higher number of phased-out units (15). The percentage of different unit types (new, constant, recalled) also generally followed a common trend: an increase in the percentage of new units was typically accompanied by a decrease in the percentages of constant units and the percentage of recalled units.

Data was unavailable or of inferior quality for years 2010, 2011, 2012, and 2016. Comparison of unit types was limited in that a comprehensive guide is yet to be published, so all studies rely on their own criteria. Ambient noise for the exact area in which this study was conducted was also unavailable. This study grouped units by characteristics, not within subsections for individual singers, and therefore does not account for individual variation by year. Due to time constraints, consideration was limited to units only; with phrases and themes, and unit order/pattern within these components also considered, results may vary.

6. Conclusion

The unit structure of the humpback population that winters off the Chiriqui Gulf of Panama is shown in this study to be variable over time in contour shape and duration, though relatively stable in peak frequency. The rate of change in unit presence is also variable, peaking in 2015, though a common trend was followed in regards to the percentage of new, constant, and recalled units present each year. There has been a fairly steady rate of new unit introduction since 2013. From this information, we can better grasp the impact of environmental factors such as ambient noise (mostly anthropogenic), and assume a relatively high level of connectivity between populations, allowing for better and more effective conservation efforts to take place.

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Singing activity of Central American humpback whales in Cuajiniquil Bay, Costa Rica

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Abstract: During the boreal winter, the Central American population of humpback whale migrate seasonally to breeding habitats near Cuajiniquil Bay. Male humpback whales typically sing complex songs during the breeding season, and this can be used as a metric to determine presence of whales in an area. This study examines the singing activity of whales with audio recordings from December 2022-March 2023, the prime time of the breeding season. The purpose was to determine what trends exist in whale activity throughout the months and time of day, as well as to examine boat activity in the area. Overall, it was found that whale occurrence is consistently high with 96% of recordings demonstrating presence of whales, with more single singers in December, and more multiple singers in January-March. Boat presence was insignificant. This study establishes a foundational understanding of the Central American Humpback whale population's presence at Cuajiniquil Bay, offering invaluable insights for further exploration into their behavior. With the overarching goal of contributing to the conservation endeavors aimed at protecting this endangered whale population, this research paves the way for informed strategies and actions in protecting these majestic creatures and their critical habitat.

Keywords: acoustic communication, underwater noise, temporal patterns, baleen whales, soundscapes

1. Introduction

Humpback whales (*Megaptera novaeangliae*) migrate seasonally between feeding and breeding habitats (Clapham, 1996). For management purposes they are classified into 14 distinct population segments (DPSs) based on their migratory patterns. Of these 14, the Central American (CA) population continues to be classified as vulnerable (Bettridge et al., 2015). These whales migrate from feeding areas in the Northern Hemisphere, specifically California and Oregon, to breeding areas off the Pacific coast of Central America during the boreal winter (~December–April) (e.g., Steiger et al., 1991; Calambokidis et al., 2000; Rasmussen et al., 2012). This population is believed to consist of less than 1000 animals and is heavily impacted by fishing nets along its migratory route (Bettridge et al., 2015).

Humpback whales are well known for their singing behavior. The males sing long and complex songs that consist of themes, phrases, and units (Payne and McVay 1971; Cholewiak et al., 2013.). The song is thought to be a sexual display to attract females and or compete with other

Commented [LMC1]: Key words need to be different to words in the title, because the goal is to maximize its appearance in search engines, like web of science

Commented [LMC2]: Steiger, G.H., Calambokidis, J., Sears, R., Balcomb, K.C. and Cubbage, J.C. (1991), MOVEMENT OF HUMPBACK WHALES BETWEEN CALIFORNIA AND COSTA RICA. Marine Mammal Science, 7: 306-310. https://doi.org/10.1111/j.1748-7692.1991.tb00105.x

Commented [LMC3]: Calambokidis, J., Steiger, G. H., Rasmussen, K., Urban, J., Balbomb, K. C., de Guevara, P. L., Salinas, M., Jacobsen, J. K., Baker, C. S., Herman, L. M., Cerchio, S., and Darling, J. D. (2000). “Migratory destinations of humpback whales that feed off California, Oregon and WaSEPington,” Mar. Ecol. Prog. Ser. 192, 295–304.https://doi.org/10.3354/meps192295

Commented [LMC4]: Rasmussen, K., Calambokidis, J. and Steiger, G.H. (2012), Distribution and migratory destinations of humpback whales off the Pacific coast of Central America during the boreal winters of 1996–2003. Marine Mammal Science, 28: E267-E279. https://doi.or males (Herman 2017). The song is learned horizontally (e.g., Winn and Winn 1978; Mercado 2021, Rekdahl et al., 2018, Eriksen et al., 2005) and males in the same breeding population will conform into the same song for the entire season (Dunlop et al., 2007). This song conformity allows scientists to differentiate populations based on their song and evaluate potential connectivity (Cholewiak et al., 2012).

On the northeast Pacific coast of Costa Rica, the waters of the Cuajiniquil Bay are considered important for CA humpback whales (May-Collado pers.comm. 2023). However, there is little information about their singing activity in Central America. The only available study is by Chereskin et al., (2019) which took placed in the southeast Pacific coast of Costa Rica, where CA humpback whale singing activity was reported to be low. The goal of this study is to document the singing activity of CA whales in the northeastern Pacific coast of Costa Rica, specifically the Cujajiniquil Bay during the breeding season of December 2022 to March 2023. Specifically, I will be determining the variation in singing activity by month and day. Some northern hemisphere humpback whales are reportedly to increase in singing activity throughout the breeding season, often with a singing peak in March (Kugler et al., 2021). I hypothesize that CA humpback whale singing activity patterns will be like these other northern hemisphere population. The results of this study will help to evaluate the importance of this bay for CA humpback whales and inform future conservation and management efforts.

2. Materials and Methods

2.1. Study Site

This study takes place along the Northeast Pacific Coast of Costa Rica in Cuajiniquil Bay, situated in the Gulf of Santa Elena. The Gulf is part of Santa Rosa National Park and has been protected under the Área de Conservación Guanacaste since the 1970s and inscribed in 1999. The gulf is an important ecological habitat for many species that has a uniquely intact coastal-marine environment. It remains as a breeding ground for many migratory species including humpback whales (UNESCO 2023).

2.2. Recordings

Recordings were made using an autonomous and remote underwater recorder model SoundTrap 400 STD (frequency range 20 Hz-150 kHz ±3 dB; self-noise of less than sea-state in the bandwidth 100 Hz-20 kHz, and sensitivity of −203 dB re V/µPa) from Ocean Instruments (http://www.oceaninstruments.co.nz/). Recordings were made from December 2022 to March 2023, using a recording cycle of 10 minutes per hour and a sampling rate of 48 kHz.

2.3. Humpback Whale Presence

Each of the 10-min files described above were open in Audacity (3.4.2) and manually inspected for humpback whale song presence using a fast Fourier transform size of 4,096 points. Noting a 1 was used for the presence of whales and boats and the occurrence of more than one whale was

Commented [LMC5]: Herman, L. M. (2017). "The multiple functions of male song within humpback whale (Megaptera novaeangliae) mating system: Review, evaluation, and synthesis," Biol. Rev. 92, 1795–1818.https://doi.org/10.1111/brv.12309

Commented [LMC6]: Rekdahl, M. L. et al. Culturally transmitted song exchange between humpback whales (Megaptera novaeangliae) in the southeast Atlantic and southwest Indian Ocean basins. Royal Society Open Science, v. 5, n. 11, 2018b.

Commented [LMC7]: Eriksen, N., Helweg, D. A., Tougaard, J., Miller, L. A. (2005). Cultural change in the songs of humpback whales (Megaptera novaeangliae) from Tonga. Behaviour, 142(3), 305-328.

Commented [LMC8]: Dunlop, R. A., Noad, M. J., Cato, D. H., and Stokes, D. (2007). "The social vocalization repertoire of east Australian migrating humpback whales (Megaptera novaeangliae)," J. Acoust. Soc. Am. 122, 2893–2905.https://doi.org/10.1121/1.2783115

Commented [LMC9]: Find full citation here https://www.frontiersin.org/articles/10.3389/fmars.2021.735664/full recorded. A 0 was assigned when whales or boats are not detected. Whale sounds are distinct in recordings, and no other cetacean species produce similar sounds, thus the sounds from the recordings will be of humpback whales. In addition, the number of whales present in a recording was categorize as one whale when only a single song was present in the spectrogram, multiple when evidence of multiple song elements was present, and as none when no whale songs were present.

2.4 Data Analysis

Data was primarily analyzed in Microsoft excel using the PivotTable tool. Percentage of files with and without whale presence was calculated, and then compared with the percentages per each month. A summary was created of number of whales singing (multiple, one, or none) at each time of day, and was input into JMP to create graphs. This was then separated out by month.

3. Results

A total of 1,638 10-min acoustic files were analyzed (December=255, January=663, February=523, and March=197). Of this a total of 1,577 (96%) acoustic files had humpback whale song (figure 1).

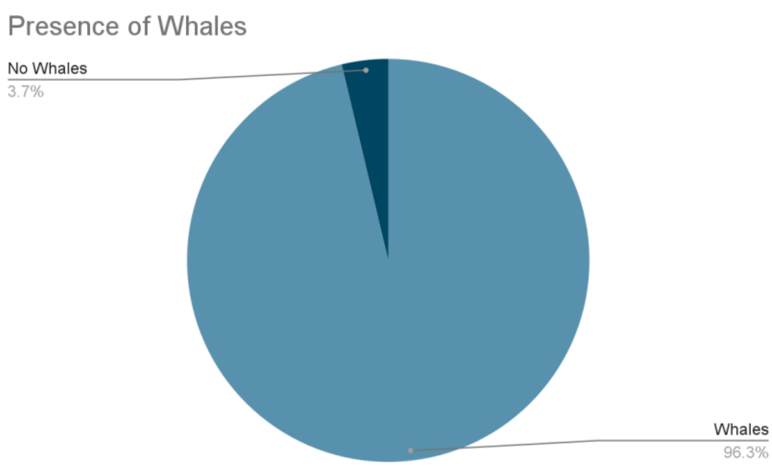

Figure 1. Comparison of files with whales present versus whales not present.

Overall, humpback whale songs were high in all four months, with slightly higher increase in February (December=96.47%, January= 93.97%, February= 99.81%, and March= 94.42%). Approximately 60% of the acoustic files contain multiple singers, 36% solo singers, and only 4% of the files did not have singers. The proportion of acoustic files by hour with multiple singers peaked at night-time hours, while the proportion of acoustic files with solo singers peaked at noon (Fig.2). When considering the differences per month of trends (figure 3) of number of singers, there were slight variation with December demonstrating more single singers, and February demonstrating more multiple singers. January's trends resembling the overall trend most closely. Boat activity was not deemed statically significant with p-values calculated of over 0.05, with only 21% of recordings demonstrating boat activity.

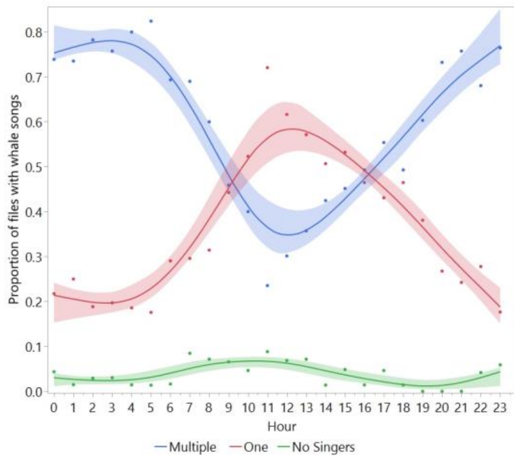

Figure 2. Overall daily trends of number of whales singing per hour from December 2022- March 2023.

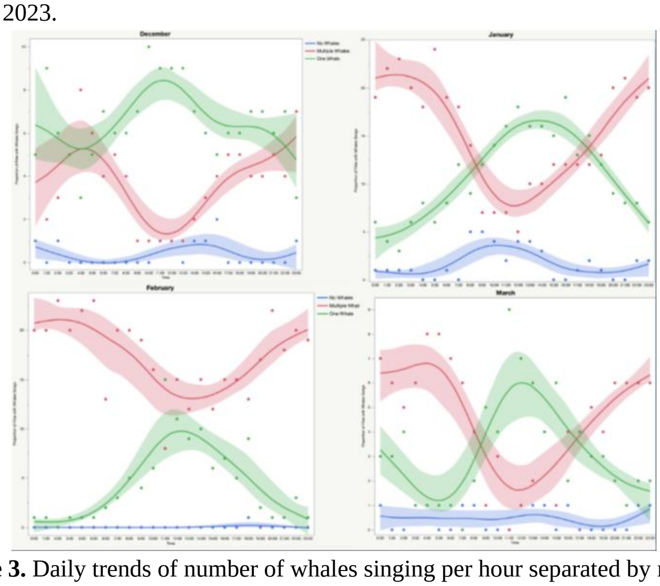

Figure 3. Daily trends of number of whales singing per hour separated by month.

4. Discussion

The results of this study show that Cuajiniquil bay is an important display area for CA humpback whale males during the breeding season of boreal winter. The whales were very active throughout the sample period with the number of whales present during this period surpassing expectations. Tit would be expected that whale activity would be less at the beginning of the breeding season in December, however it remained high every month (Steiger et al, 1991). The difference in distribution of single singers in December compared with the other months having a higher prevalence of singers might be indicative of the start to the breeding season as there are less whales singing to be picked up with. There could be a variety of factors that would change the timing of Humpback whales at the breeding ground, and it would not be the first time where whales came early to a breeding ground (Avila et. al, 2019). Overall, the data lightly supports the seasonality from previous studies. To fully understand the seasonality of this population of whales it might be beneficial to collect data from prior to the predicted period and after the predicted period in order to aim to record the arrival and departure of the CA population.

The daytime versus nighttime trends in multiple whale activity were interesting. The multiple whale trends mirrored that of what would be expected, previous work suggests that northern humpback whale populations sing primarily at night during their breeding area (Ryan et. al. 2019). In contrast, single singers are active throughout the day. This is in direct contrast with predictions of activity, however potential causes for this phenomenon might be due to competition between males, with less males singing during the day making it more common for single singers due to less competition. This would have to be studied further as there haven't been other examples of this trend. The lack of significant in the boat data is not unexpected as this time of year comes with heavier wind activity (NASA Earth Observatory 2004).

6.

5. Conclusion

The presence of CA whales in Cuajiniquil bay highlights this area's importance as an Área de Conservación, the continued protection of this area is important for this threatened population to have a chance at bouncing back. There are no current studies with recordings of this population of whales, making this study vital towards the understanding of this endangered population. Strides must be made to support the conservation of this population, and the first step is a better understanding of the behavior and migration patterns of these whales. The more that is known about this population, the better they can be protected.

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<u>C</u>ourse-based <u>U</u>ndergraduate <u>R</u>esearch <u>E</u>xperience:
PROGRAM (BIOL 2100A)
Fall 2023

Course Instructor *Laura J May-Collado, Ph.D.*
E-mail: lmaycoll@uvm.edu
CURE Website: http://www.lauramay-collado.com/cure-lab.html

Meeting Time: T 1:15-4:14

Office hours: by appointment only.

Your instructor: Before becoming an Assistant Professor, I was a Lecturer for eleven years. My teaching philosophy stems from my own undergraduate experiences at the University of Costa Rica: every course included opportunities for fieldwork, research, and development of inter- and intrapersonal 'soft skills.' As a first-generation student from a low-income family, these were my first opportunities to recognize my STEM interests. I employ this same experiential lens in the design and delivery of this Course for Undergraduate Research Experience (CURE).

Course description: CUREs are "learning experiences in which whole classes of students address a research question or problem with unknown outcomes or solutions that are of interest to external stakeholders". This approach has emerged as a promising alternative to traditional 'apprenticeship' research experience as CUREs are more accessible, engage more diverse STEM undergraduate students, and merge teaching and research. Students that participate in CUREs are reported to develop confidence in their ability to do science and express a sense of ownership of their research. A limiting aspect of many CUREs is that by design students cannot fully exercise their curiosity: research options are constrained by the faculty. This limits their discovery and creativity process and makes it more challenging to learn from failure. This 4-credit, fall semester CURE on Soundscape and Behavior (Biol-2100) CURE stands out from such designs in that each student develops an independent research project to explore their own research interests. Several studies have shown that students that are provided independent research opportunities are more motivated to learn.

Course main objective: This CURE is designed to guide students' improvement in both research skills and two intra- and interpersonal competencies that are associated with college success: <u>conscientiousness and self-efficacy</u>.

Learning goals:

1. *To offer the opportunity* to make discoveries and contributions to the scientific community, policymakers, and the public.
2. *Engage students in all aspects of research*: literature reading and discussion, asking questions that can be answered during the semester, collecting, processing, and analyzing data, learning how to interpret analytical results and how to communicate the results.

3. *Create an environment that promotes active collaboration* and contributions among students and instructor during the semester through problem solving and analysis.
4. *Learn that science is not about eureka moments!* Good science takes time, involves failure, troubleshooting, discussions, re-evaluations, and yes frustration. Good science is always challenging at different levels, from collecting the data to its analysis.
5. *Learn that there is not a single "right" way to do science!* Different questions, systems, or species will require different approaches. For example, some research questions rely on wellplanned experimental designs involving multiple controls. My research is field based which is bound to be limited by replication, sample size, and logistics. However, field-based projects are essential for our understanding of our biological world and are often the spark for more controlled experimental studies.

Course expectations:

- I expect students will be engage in a dynamic and respectful environment for scientific communication and collaboration.
- I expect students to take responsibility of their assigned projects, be independent and resourceful readers of scientific literature related to their assigned projects and demonstrate initiative in learning new programs, data collection, and analysis that can help them address their research questions.

Required texts/technology

1. **Audacity**: this is a free, open source, cross platform audio software https://www.audacityteam.org/
2. **Brightspace**: There is a Brightspace site for this class to post announcements, materials, workshop materials, and assignments.

Library

Howe Library: https://library.uvm.edu/askhowe
Dana Medical Library: https://dana.uvm.edu/help/ask
Silver Special Collections Library: https://specialcollections.uvm.edu/help/ask

4. **Required Reading** is posted in Brightspace.

Software available in lab computers

- RAVEN https://ravensoundsoftware.com/
- dBWav https://nz.marshallday.com/innovation/software/dbwav
- Luscinia https://rflachlan.github.io/Luscinia/

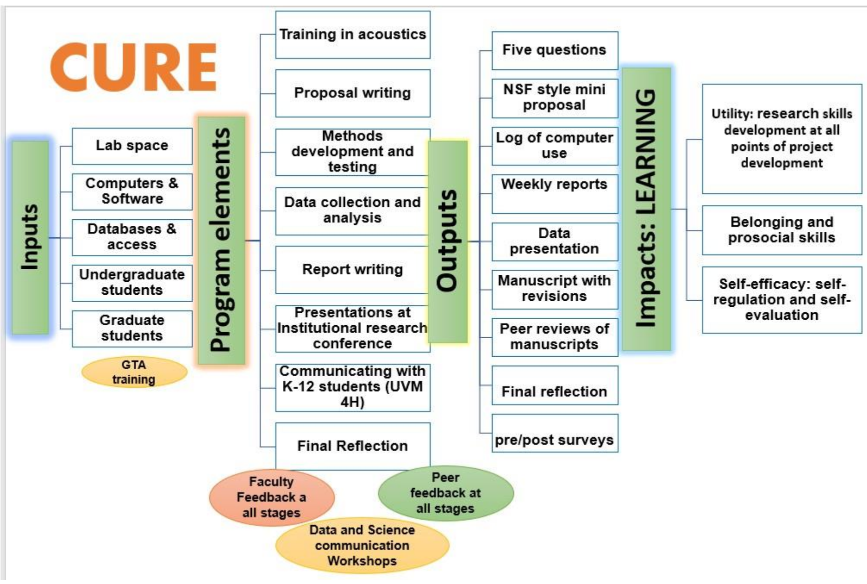
Recommended Reading Sources

Basics of Sound: https://dosits.org
Read previous CURE projects: http://www.lauramay-collado.com/cure-lab.html Acoustic Ecology: https://www.acousticecology.org/scienceprograms.html

Course Structure

Research: During this CURE course, students will develop independent research skills using original acoustic data collected by my lab and collaborators. The figure below shows the scaffolding of all phases of project development from posing questions to presenting at the UVM Student Research Conference and drafting a manuscript following the journal format of their choice. To help students develop problem-solving skills, students will participate in workshops about fieldwork methods, AI and ML, soundscapes, statistics, and phylogenetic analysis. These workshops are meant to engage students in creative thinking and knowledge of emerging computation skills, key for the current labor market. In addition to workshops, students will engage in peer evaluation of their work and meet individually with the faculty each week to review their progress. Students will also participate in workshops for scientific writing and public communication, and in a journal club to discuss scientific literature related to their research project.

Communicating your science: The research project includes work on communication of science beyond the scientific manuscript and presentation. Opportunities for learning how to communicate science outside the scientific community will take two forms. First, students will develop a 1-min video abstract about their research. This video will be posted with other student products on the CURE website. Second, students will partner with the UVM 4H program -- an extension program that provides opportunities to the public to participate in hand-on STEM activities -- to develop a team-based outreach activity for kids and families on a topic of their choice related to the research themes of the CURE in December 2023.



Belonging and prosocial skills: This course is deeply collaborative with weekly student-faculty and peer-peer meetings. Weekly in-person meetings will help students develop feelings of confidence, belonging, equity, and community that are expected to contribute to prosocial values, transcending personal goals to value the impact on others. Peer-peer interactions will be promoted through workshop group activities and weekly group meetings. During cohort meetings, students will present their progress and challenges. As a group, we will brainstorm solutions and provide feedback to each other. Awareness of the impacts on society will be discussed through the lens of "parachute science." Parachute science is the conservation model where researchers from the developed countries go to countries with limited research funding, conduct research, and leave without any investment in human capacity or infrastructure, or take control of agendas that drive undesirable conservation outcomes. This CURE involves international partners and links to international conservation efforts. As a class, we will develop an integrity protocol to ensure we fully recognize local governance, capacity, expertise, and social structures, ethically manage coauthorships, and respect our partners' conservation priorities.

Self-efficacy: Self-efficacy, the belief a student holds in their own ability to succeed, is a complex personal trait related to persistence through failure. This CURE is designed to support student growth in two components, self-assessment, and self-regulation, because they relate to acceptance of criticism and persistence. In response to the regular feedback provided from faculty, international partners, and peers, students will be revising their research efforts in each step. Student selfreflection is a component of the weekly progress meetings held individually and as a group. The faculty and graduate teaching assistants will maintain logs reflecting on their perception of students' responses to criticism and challenges. Students are also expected to manage their time and effort independently, which will be tracked through the log of computer use. At the end of the program students will have improved technical and soft skills that are transferable to various STEM fields.

What will you be working on this semester?

This semester you will become a collaborator of my ONDAS project. This is an initiative I started in 2016 to marine mammal acoustic ecology and soundscapes. My overall goal is generating a better understanding of the marine biodiversity in the region to help governments to use this information and tailor conservation strategies to preserve biodiversity.

Why use sound? Animals acquire information from their environment using their five senses (sight, smell, hearing, touch, and taste). For aquatic animals, some of the advantages of using sound are that it can be used at any time of day, travels fast in seawater and in all directions, and has low attenuation. Therefore, sound is particularly useful for nocturnal or aquatic species that cannot rely on visual cues. The integration of all sounds, animals, rain, wind, and human sounds (e.g., cars, airplanes, chainsaws), is referred to as the soundscape. Furthermore, a single species can produce different sounds depending on the context, such as trying to attract a mate, communicating with an infant or juvenile, or warning others of a predator. Fortunately, the collection of these sounds is usually species-specific, which means that the frequency range, dominant frequency, duration, and pattern of sound production are unique to each species. This maximizes communication within a species and avoids confusion among species, and it also helps scientists develop models to identify many different species.

What will you be studying? This semester we have lots of data from several collaborators and our lab. You can decide to study the acoustic repertoire of one species or to study soundscape dynamics at various temporal or spatial scales. We will be exploring this database during the first week of classes. Prior to collecting data for your project, you will be working on, I am going to ask you to sign a “Memorandum of Understanding” (MOU) on data use.

Topics to develop independent research include,

Soundscape Ecology: Biodiversity survey methods are labor-intensive and limited to a few locations and short periods of time. This is true when it comes to studying marine communities. New acoustic technology provides marine scientists the opportunity to study community dynamics using sound as a cue for biodiversity. My recent research aims to use underwater acoustic technology to study biologically important marine communities in Central America. I have accumulated over 3 million minutes of recordings from various marine communities, and one key measure of health that students can ‘easily’ measure is noise level.

Dolphin Acoustics and Behavior: Most dolphins live in complex fission-fusion societies, where animals associate with different individuals in a fluid manner. The strength of these associations appears to vary across groups and over time. In a society such as this, individuals play various roles in maintaining the integrity of the overall social structure of a population. Dolphin group structure is also maintained by learning to recognize others, and this is done by using signature whistles. For example, in bottlenose dolphins signature whistles are unique to each dolphin in the group and provide information about identify, gender, and age. These whistles are like name tags and are developed early in life using the mother (in the case of baby males) or group members' whistles (in the case of baby females) as templates. Dolphins also emit variant whistles and several other types of sounds that are species and context specific, and we know very little about the acoustic characteristics of these sounds. Question recharging function or vocal repertoire will depend on how much recording effort we have for a given species.

Humpback whale communication: Unlike dolphins, the communicative signals of baleen whales are produced at much lower frequencies and limited to specific behaviors. Also, while in dolphins both males and females emit sounds, in baleen whales only males are known to sign (there are a few exceptions though). In my lab several students are involved in studying humpback whale song activity and structure in Central America. During the first part of the year, Northern Hemisphere humpback whales from California and Oregon migrate to Central America to breed, this specific breeding population is one critically endangered. During the second half of the year the same coastal areas received Southern Hemisphere humpback whales from the Antarctica Peninsula and Chile. The song of humpback whales is quite complex, it consists of hierarchical components that are sung in the same order. Breeding populations have song components unique to them, but occasionally a new song evolves via cultural transmission. Thus, whale song structure analysis can help us study population connectivity and the role of culture in whale singing behavior. This year we have a new set of recordings from collaborators in Central America, Mexico, and Panama.

Fish Acoustic Behavior: You will be surprised at the number of fish species that emit sounds! Fish can be quite chatty! Students in this CURE course have developed projects studying the acoustic activity of toadfish. Male toadfish are territorial and emit sounds to attract females to lay eggs in their territory. Presumably, females assess the quality of the male prior releasing their eggs. We have learned that in Bocas del Toro Panama toadfish acoustic activity is primarily during dark hours, and that they respond to boat presence in various ways. Because they are important components of marine communities, toadfish presence is used to evaluate marine community's health. The toadfish is just one of several fish species we have recorded. Discovering the identity and describing the acoustic activity of other fish species in our study sites is key in understanding the dynamics of coastal marine communities. We have at least 3 million minutes of recordings, and fish are the dominant component of the soundscape at dawn and dusk, unfortunately we do not know who each sound. Nevertheless, documenting their sounds is key in understanding community dynamics.

Course Calendar (not that I reserve the right to make changes to this calendar)

Week	Where?	Chronogram

Aug 29	Lab MLS 217	• Introduction to this course Syllabus • Reading Assignment: *"How can audio recordings improve tropical biodiversity conservation?" and "Can you hear me? How do dolphins in Boca del Toro, Panama communicate in a noisy habitat?".* Links in Brightspace • Explore the projects developed by past CURE students. • Read the literature (special that produced by my lab) and explore the online resource: **write down 5 potential research questions.**
Sep5	217 MLS	• Tour of the available data • Introduction to various Acoustic software • Present your 5 questions and discuss which is the most feasible.
Sep 12	217 MLS	Proposal writing in class: • I will give you a short workshop on how to write your proposal. • Format and examples will be posted in Brightspace. • We will work out your methods. • We will set up a schedule with weekly goals and outputs. • **Submit proposal at the end of the day.**
Sep19Oct 31 Oct 3 &10 Oct 31	217 MLS	**7 weeks of data collection:** we will meet weekly to discuss in person progress, make sure to deliver weekly report. **Design 4H activity with families on December 9.** **Last day to deliver introduction and methods sections.**
Nov 7 Nov 14	217 MLS	Workshop on Data visualization and analysis: each student will work on their own data and write the result section. Workshop on How to write a scientific manuscript: we will work on the abstract and discussion, and assemblage all together.
Nov 2124		**Thanksgiving week**
Nov 28	217 MLS	Final review of the manuscript, including feedback from instructor and peers Prepare Oral presentation get feedback, practice with peers.
Dec 5	**217 MLS**	**Symposium!** **Last day to deliver CURE-Reflection Blog**
Dec 9	TBD	**4H Activity with families**

Grading

Activities	Pts (can vary)	%
5 questions	10	5
Proposal	25	15%
7 Weekly progress reports and weekly meetings with instructor (see format in Brightspace)	35 (5 pts each)	25%
5 Write up reports including final manuscript	50 (10 each)	20
4H group activity and participation	50	10
Final oral presentation (see format in Brightspace)	50	20
Final Reflection (CURE Blog) (see format in Brightspace)	30	5
Total		**100%**

Requested Formats

Proposal NSF format

The proposal must consist of the following parts (4 pages).

- **Introduction** – (1 page) ○ ***Background*** to problem with citations of papers or other sources that document the information you are presenting. This background should include the observations that lead to your question or hypothesis.
    - ***Purpose and scope*** - Statement of the purpose of your paper, this may be how you are testing your hypothesis. If you use hypothesis you need to make predictions about the hypothesis. Predictions will also go here.
    - ***Significance:*** How does your project advance knowledge on this field? How does your project benefit society?
- **Materials and Methods** – (1 page) What type of data have you found and what additional data are you going to try to find? How will the data you collect be analyzed to address your objectives, questions, or hypothesis? It is important to make it clear how the scientific method will be used to test or address either your hypothesis or the predictions you expect if the hypothesis is true.
- **Research Plan** – (1 page) Schedule of steps to be accomplished with deadline dates.
- **Literature Cited** –(1 page) Full reference to the papers cited in the introduction and materials and methods sections. Use format from Journal of the Acoustical Society of America. **See example https://asa.scitation.org/doi/10.1121/1.5139205**

MANUSCRIPT

We will be writing each section of the paper by parts as shown in the schedule above. Each section should be in the format of *Journal of the Acoustical Society of America*. Go to the journal and download the guidelines for authors.

Here is a summary of the guidelines: https://asa.scitation.org/pb-assets/files/publications/jas/JASA_AuthorChecklist1508440990393.pdf

Here is an example of a JASA published manuscript: https://asa.scitation.org/doi/10.1121/1.5139205

Other important resources

Steps to organizing your scientific manuscript. https://www.elsevier.com/connect/11-steps-to-structuring-a-science-paper-editors-will-takeseriously
How to write a scientific paper: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3474301/
Statistics for Biologists https://www.nature.com/collections/qghhqm

ORAL PRESENTATION

You will have 10 minutes, 8 minutes for your presentation, and 2 minutes for questions. **Deliver your power point presentations 1 day prior to the symposium to the instructor to be uploaded on time.**

- Please embed any videos or audio within the presentation
- Also include ALL videos & audio files in a separate folder on your thumb drive. This will enable us to correct any problems on site.

DATA USE

The data that you will be using to develop your project is my property as the PI of the projects involving the collection of this data. Some data sets are shared with collaborators that significantly contributed to data collection, and thus ownership is shared. You will sign a contract of ethical use of the data. **No sharing of data on social media or with other parties is allow without my consent. This includes photographs, acoustic files, or any other data from my databases**. We will develop a space for outreach activities and research experience communication through a blog where you can post sound files, summaries, photographs, and updates on data processing with my approval.

Writing in the Age of Artificial Intelligence: Since writing, analytical, and critical thinking skills are part of the learning outcomes of this course, all writing assignments should be prepared by the student. Developing strong competencies in this area will prepare you for a competitive workplace. Therefore, AI-generated submissions are not permitted and will be considered a violation of the cheating and plagiarism standards of the UVM Code of Academic Integrity. Violations could result in failure of the assignment or failure of the course and a notation on your transcript. - UVM Center for Student Conduct.

ACADEMIC HONESTY

Academic honesty is expected of all students. The University of Vermont has a very strict policy concerning academic honesty and plagiarism. Please see the statement on academic honesty http://www.uvm.edu/~uvmppg/ppg/student/acadintegrity.pdf.

Plagiarism constitutes a violation of Academic Honesty. Plagiarism of ANY sort will NOT be tolerated. The consequences of plagiarism or cheating range from a score of zero on the assignment, failure in the course, to filing a complaint with the University's Coordinator for Academic Honesty, which can result in expulsion from the University.

COURSE CONTENT AND DATA IS THE PROPERTY OF THE INSTRUCTOR.

Consistent with the University's policy on intellectual property rights, all teaching and curricular materials (including but not limited to classroom lectures, class notes, exams, handouts, and presentations), and **research data**, are the property of the instructor. Therefore, electronic recording and/or transmission of classes or class notes is prohibited without the express written permission of the instructor. Such permission is to be considered unique to the needs of an individual student (e.g. ADA compliance), and not a license for permanent retention or electronic dissemination to others. For more information, please see the UVM policy on Intellectual Property, sections 2.1.3 and 2.4.1

RELIGIOUS HOLIDAYS: Students should submit in writing to their instructors **by the end of the second full week of classes** their documented religious holiday schedule for the semester. Students who miss work for the purpose of religious observance will be allowed to make up this work.

STUDENT DISABILITY POLICY. In keeping with University policy, any student with a documented disability interested in utilizing accommodations should contact ACCESS, the office of Disability Services on campus. ACCESS works with students and faculty in to find reasonable and appropriate accommodations, which are communicated to faculty in an accommodation letter. Contact ACCESS: A170 Living/Learning Center; 802-656-7753; **access@uvm.edu**; or **www.uvm.edu/access.**