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Evaluating Shark Community Assemblage using Underwater Video Data

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# UNIVERISTY OF CALIFORNIA SAN DIEGO

Evaluating Shark Community Assemblage using Underwater Video Data

A thesis submitted in partial satisfaction of the requirements for the degree Master of Science

in

Marine Biology

by

Madison Leigh Samilo

Committee in charge:

Dovi Kacev, Chair Sarah Aarons Tomoharu Eguchi Stuart Sandin

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University of California San Diego

# DEDICATION

This thesis is dedicated to the all the graduate students in my cohort. May you all have success and continue to make great discoveries.

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### **ABSTRACT OF THE THESIS**

Evaluating Shark Community Assemblage using Underwater Video Data

by

Madison Leigh Samilo

Master of Science in Marine Biology

University of California San Diego, 2022

Professor Dovi Kacev, Chair

Baited remote underwater video systems (BRUVs) have become a valuable method for observing large bodied marine organisms such as sharks. These systems provide several advantages over traditional methods for obtaining species distribution information, which is needed for effective management and conservation of coastal marine habitats. BRUVs are mainly utilized for assessing the distribution and abundance patterns of single species and the effects of environmental factors on these patterns (Bond et al. 2012; De Vos et al. 2015), however opportunities exist for expanded analysis from these rich collections of data. I used data from BRUV surveys within four regions of the Florida Keys to assess the co-occurrence patterns of sharks and rays, employing a method that expands on the utility of BRUVs to explore questions at the community level. I found no evidence to suggest that groups of species were gathering or avoiding each other more than expected. In species pairwise analysis, I found the pairing of specific species consistently co-occurring for common prey or habitat preference. The findings of this study will contribute additional knowledge to the work of MacNeil et al. (2020) in their effort to assess the conservation status of reef associated sharks and rays worldwide.

### **1 INTRODUCTION**

Although the decline of shark populations around the world has been researched, catch records in industrial fisheries provide much of what is known about their status and less information is available concerning their abundance, distribution, and community structure within coastal habitats (Roff et al., 2018). Similarly, studies exploring the status of chondrichthyans are limited compared to any major vertebrae lineage other than amphibians (Stuart et al. 2004; Hoffmann et al. 2010). Consequently, there exists a knowledge gap in the conservation status of reef associated sharks globally (MacNeil et al., 2020). Reef associated sharks can be defined as species that complete their life cycle on coral reefs or frequently visit them (MacNeil et al. 2020).

Given their narrow geographic distribution, coastal species are more susceptible to threats such as bycatch, habitat loss, exploitation, and climate change compared to offshore species in pelagic and deep-water ecosystems (Dulvy et al. 2014). Among coastal species, large-bodied, shallow-water species are at the greatest risk (Dulvy et al., 2014) and the loss of these predators could result in the absence of important top-down control of coastal ecosystems (Heithaus et al., 2012; Ferretti et al. 2010). The Florida Keys, as a major coral barrier reef found off the shores of North America, exemplifies the challenges associated with elasmobranch conservation (Lapointe and Matzie, 1996; *Explore: Corals and Coral Reefs*, 2010). Thus, by evaluating the cooccurrence of sharks and rays in the Florida Keys, the results of this study will contribute to the research that MacNeil et al. (2020) began in 2015 in their systematic analysis of threat for globally distributed sharks and rays using baited remote underwater video systems (BRUVs).

To address the knowledge gap and the conservation status of reef associated sharks globally, the Global FinPrint project was initiated (MacNeil et al. 2020), which united

researchers around the world to study sharks, rays, and other marine life on corals using BRUVs. Standardized BRUV surveys were conducted at more than 371 reefs in 58 nations (MacNeil et al. 2020). Although the initial publication examined the problem at the global level, finer spatial scale studies at the regional level are needed.

For larger-bodied reef fishes such as sharks and rays, BRUVs have become a valuable observational method (Meekan & Cappo 2004; Meekan et al. 2006; Malcolm et al. 2007). There are several advantages of BRUVs over traditional methods such as longline surveys and trawls (Ellis & DeMartini, 1995). Although the video resolution of BRUVs may limit their capability to conduct finer-scale, intra-species studies (Brooks et al. 2011; Kilfoil et al. 2021), BRUVs are able to record animals that would otherwise avoid active fishing surveys and divers, result in only minimal damage to the benthic environment, and avoid causing physical trauma and physiological stress to subject animals (Cappo et al. 2004, 2006; Skomal 2006, 2007; Cooke et al. 2002). Previously, BRUVs have been used to examine fish densities (Ruppert et al. 2013; Harvey et al. 2013), understand biases of sampling gears (Cappo et al 2004; Harvey et al. 2012), document fish species richness (Harvey et al. 2013; Cappo et al. 2007), and quantify elasmobranch abundances and distribution patterns (Bond et al. 2012; White et al. 2013). BRUVs are primarily used to determine single species distributions and the effect of environmental factors on these distributions whereas there is a need to assess the diversity and conservation status of overlooked chondrichthyan species (Osgood et al. 2019; Osgood et al. 2015). Data from BRUVs systems, however, can provide information that can be used to explore community dynamics and answer other broader questions concerning elasmobranch assemblage (Osgood et al. 2019; Mallet & Pelletier, 2014). Here, I present an analytical tool that determines whether or not co-occurrence can be attributed to coincidence or to specific biological or

ecological drivers in the Florida Keys. This method contributes to the expansion of the potential of data from BRUVs for multi-species inference (Mallet & Pelletier, 2014).

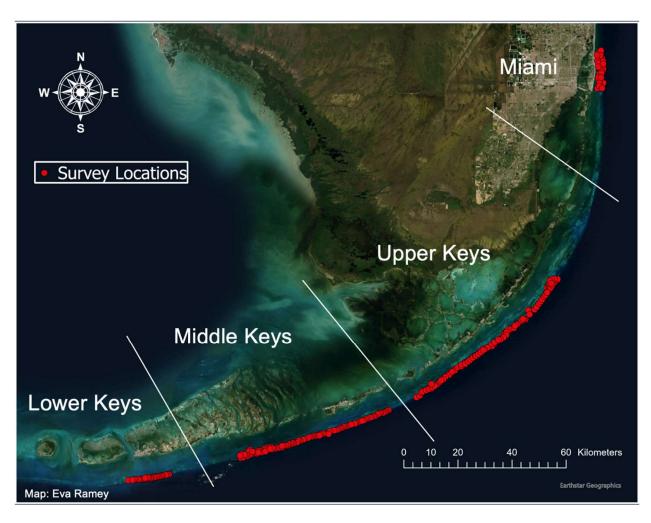
The present study assesses elasmobranch co-occurrence patterns along the Florida Keys over a three-year period using a BRUVs data set provided by the Global FinPrint program. The number of species present at a BRUV were hypothesized to be observed more often than the expected values, which were obtained by assuming species occurrence was independent. Additionally, particular combinations of species were hypothesized to be observed more often than at random.

### 2 METHODS AND MATERIALS

### 2.1 Study Site

The Florida Keys are a major barrier coral reef found off North America and represent the third largest barrier reef in the world (Lapointe and Matzie, 1996). Additionally, the Florida Keys National Marine Sanctuary protects the living organisms and habitat from fishing and construction, where protection started in 1990 (Keller and Donahue, 2006).

In order to assess the conservation status and potential of reef associated sharks and rays, in 2016, 2017, and 2019, 376 individual BRUVs were deployed in 4 regions of the Florida Keys (Figure 1). The number of BRUVs deployed annually was not equal. Annual BRUV deployments were restricted to 150-200 BRUVs per region and deployment sites were chosen based on areas where there was at least a consistent 10 square km reef tract. The recorded video from these BRUVs was standardized to one-hour intervals and then analyzed by researchers at Florida International University. The maximum number of individuals of each species that occurred on any given frame of video was recorded (MaxN).



**Figure 1:** BRUVs deployment locations along the Florida Keys (filled red circles) for 2016, 2017, and 2019. This figure was retrieved from: Ramey E., Kacev, D., Eguchi, T., Bond, M. (2022). An Assessment of Coastal Shark Distribution Patterns in the Florida Keys Archipelago Based on BRUV Survey Data (Manuscript in preparation).

### 2.2 Summary Analysis

In order to describe the co-occurrence of species at sampling locations, I analyzed the relative abundance (summed MaxN across all species) and species richness. Statistical significance was determined using a non-parametric bootstrap method. To accomplish this, the BRUVs data were sorted to find the total number of individual sightings for each BRUV. The BRUVs data were then converted into presence-absence and the total number of BRUVs having 0, 1, 2, 3, 4, 5, and 6 species co-occurring was calculated to get the observed presence-absence

values. The total number of times individuals from a species appeared, before conversion to presence-absence, was summed across all BRUVs then permuted (using sample() in r) to randomly assign all of the individual sharks and rays across a simulated 376 BRUVs. The results were then converted to presence-absence. The number of simulated BRUVs showing 0, 1, 2, 3, 4, 5, and 6 species co-occurring was evaluated. Permuting the observed individual values by unlinking individual by BRUV and taking the totals of 0, 1, 2, 3, 4, 5, 6 species co-occurring was repeated 1000 times to determine the variability in the number of co-occurrences. The permutation process generated a sampling distribution to identify the likelihood of seeing 0, 1, 2, 3, 4, 5, and 6 species co-occurring at individual BRUVs at random. We then empirically identified the 95% confidence intervals of those sampling distributions. The observed number of BRUVs given by the observed presence-absence values with each number of co-occurring species was compared to the randomly generated sampling distributions (Figure 2). A species interaction was considered significant if the observed co-occurrence value fell outside of the 95% confidence interval.

### 2.3 Species Pairwise Analysis

Pairwise species interactions were determined by calculating the observed number of cooccurrences between two species. The probability of observing a species at a BRUV site was calculated by dividing the number of BRUVs with the species present by the total number of BRUVs. To calculate the expected number of BRUVs with each two-species pairwise combination, we multiplied the expected probability of each species (assuming the two species occur independent of each other) and multiplied that product by the total number of BRUVs. The expected and observed data were recorded in a 17 by 17 matrix accounting for each species combination. Two species occurring together was considered significant if the observed value

was greater than two. The normalized value was calculated by dividing the observed number of BRUVs with each pairwise combination by the corresponding expected value for that species combination.

### 3 RESULTS

Distribution and species richness of elasmobranchs were examined at a total of 376 BRUV deployments. Overall, sharks and rays occurred in 62% of BRUV deployments. Sharks were sighted 126 times and the number of species present per BRUV deployment varied from 0-4 elasmobranch species. Of the sites that recorded a shark or a ray, 75% of them recorded only a single species. There were 13 species of elasmobranch recorded from 8 different families. When converting to presence-absence, nurse sharks accounted for 29% of the sightings and were the most abundant followed by the yellow round ray (25%) and the bonnethead shark (17%) (Table 1). Together, these species accounted for 71% of the abundance of reef-associated sharks and rays. **Table 1:** Summary statistics of occurrence of elasmobranch species in data from 376 BRUV deployments along the Florida Keys in 2016, 2017, 2019. No sightings of elasmobranchs were recorded at 143 BRUV deployments so the proportion of sightings were also calculated for BRUVs with at least one sighting.

Common name	Scientific Name	# of Individual Sightings	# of BRUVs	% out of all 376 BRUV Deployments	% out of 233 BRUVs with Sightings
Blacknose shark	Carcharhinus acronotus	3	3	1%	1%
Bonnethead shark	Sphyrna tiburo	44	40	11%	17%
Bull shark	Carcharhinus leucas	1	1	0%	0%
Caribbean Reef shark	Carcharhinus perezi	15	13	3%	6%
Eagle Rays	Aetobatus sp.	1	1	0%	0%
Freckled Guitarfish	Pseudobatos lentiginosus	1	1	0%	0%
Great Hammerhead shark	Sphyrna mokarran	3	3	1%	1%
Hammerhead shark	Sphyrna sp.	4	4	1%	2%
Lemon shark	Negaprion brevirostris	8	7	2%	3%
Nurse shark	Ginglymostoma cirratum	70	68	18%	29%
Requiem shark	Unknown sp.	1	1	0%	0%
Sharpnose shark	Rhizoprionodon sp.	12	12	3%	5%
Southern stingray	Hypanus americanus	13	13	3%	6%
Tiger shark	Galeocerdo cuvier	1	1	0%	0%
Unknown Ray	Unknown sp.	2	2	1%	1%
Whitespotted Eagle ray	Aetobatus narinari	6	5	1%	2%
Yellow Round ray	Urobatis jamaicensis	60	58	15%	25%

Some species were spotted only once during the three-year deployments which included the bull shark in 2017, the eagle ray in 2016, the freckled guitarfish in 2016, the requiem shark in 2016, and the tiger shark in 2016. Sites that had a richness of 3 or more were consistently comprised of nurse sharks or yellow round rays. The Upper and Middle Keys had the highest richness, and the Shannon-Weiner diversity index was the greatest in the Upper and Middle Keys (Ramey et al. 2020). From 2016-2019, the number and species of sharks and rays decreased as well as the catch per unit effort (Table 2). In 2019 the species recorded were the nurse shark, lemon shark, and yellow round ray. All were recorded in the Miami-Dade. In 2017 there were 9 species, in addition to 3 unidentified species. In 2016 there were 11 species in addition to 5 unidentified species of which the nurse shark was most frequently observed followed by the bonnethead shark and the yellow round ray all in the Upper to Middle Key regions.

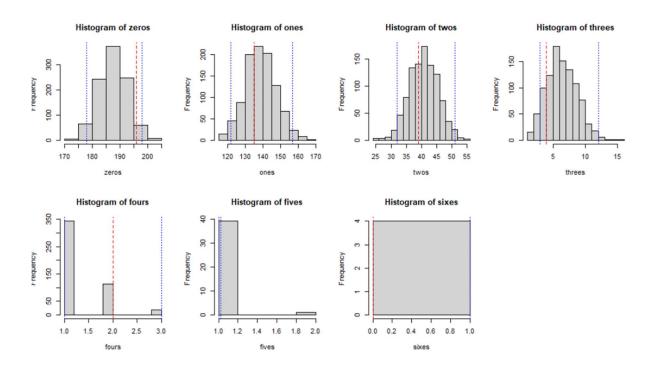
**Table 2:** A summary of BRUV deployments and the observed numbers of sharks and rays among the three deployments. The number of species is indicated by sp. CPUE is the number of observed individuals divided by the number of BRUVs.

Year	# BRUVs	# Sharks	# Rays	CPUE (indv/BRUV)
2019	38	7, 2 sp.	5, 1 sp.	0.32
2017	135	48, 8 sp.	29, 4 sp.	0.57
2016	203	107, 10 sp.	49, 6 sp.	0.77

### 3.1 Summary Analysis

When analyzing the number of species at a BRUV, no observed values fell outside of the 95% confidence intervals, indicating that the observed number of species were not statistically different from what would be expected from random. For zero observations, which occurred at 196 BRUV deployments, they also were within the 95% confidence interval. One species occurring at a BRUV was observed 135 times. The greatest frequency of co-occurrences

observed was two species occurring together 39 times. Three species occurring together was observed 4 times. Four species occurring together was observed only 2 times. Five or six species all occurring at one BRUV was never observed but did appear in the simulation results (Figure 2).



**Figure 2:** Distributions of expected number of species (0, 1, ..., 6) co-occurring at a BRUV site. Distributions were created via a non-parametric bootstrapping method with 1000 iterations. Observed values are indicated by red dashed lines and 95% confidence intervals in blue dashed lines.

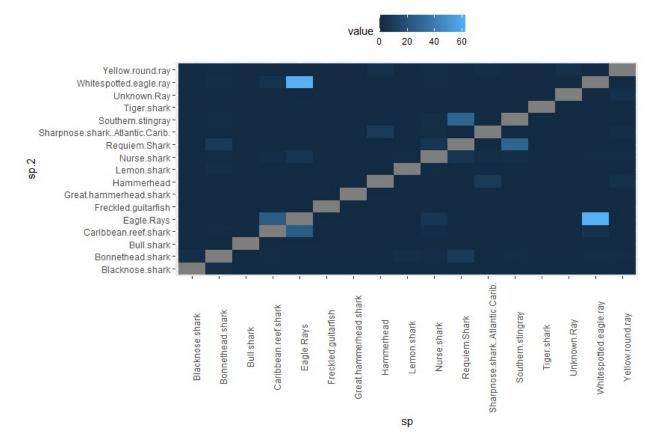
### 3.2 Assessing Species Relationships

Investigating relationships between species revealed that the number of observed pairs of species was similar to the simulated expected number for the 16 pairwise combinations that had more than 0 co-occurrences (Table 3). Excluding the co-occurrence of the nurse shark with the southern stingray, each expected value of a species combination was within 2 BRUVs of the observed value. The observed number of BRUVs that recorded the nurse shark with the southern stingray was 5, with an expected value of 2.

Shark sp. 1	Shark sp. 2	Expected	Observed
CRS	Bonnethead	1	1
Lemon	Bonnethead	1	2
Nurse	Bonnethead	8	6
Nurse	CRS	2	4
Nurse	Lemon	1	1
Sharpnose	Bonnethead	1	0
Sharpnose	Nurse	2	3
Southern	Bonnethead	1	2
Stingray			
Southern	Nurse	2	5
stingray			
Whitespotted	Nurse	1	1
Eagle ray			
YRR	Bonnethead	7	7
YRR	CRS	2	1
YRR	Lemon	1	0
YRR	Nurse	11	13
YRR	Sharpnose	1	3
YRR	Southern	2	1
	stingray		

**Table 3:** Results of the species pairwise analysis before normalization. CRS indicates Caribbean reef shark. YRR indicates yellow round ray.

When normalized, the species pair that appeared far more frequently than the expected value was the whitespotted eagle ray and eagle ray. Additionally, the eagle ray and Caribbean reef shark as well as the requiem shark and southern stingray displayed high scores (Figure 3). Species pairs that did not show as strong results as the previously mentioned pairs, but still greater co-occurrences than expected were the requiem shark with the bonnethead and nurse shark, sharpnose shark with the hammerhead shark, and the nurse shark with the eagle rays.



**Figure 3**: Normalized heat map of species interactions. Species pairs that were not seen together more than expected are darker blue whereas species that did have significantly greater sightings than expected are in lighter colors.

### 4 **DISCUSSION**

Here, I have described the patterns of co-occurrence for 13 coastal shark and ray species along the Florida Keys, highlighting the utility of BRUVs in assisting with the evaluation of biodiversity across the region. Additionally, we attempted to develop an analytical method that allows us to look at whether species co-occurrence is likely the result of coincidence, biological, or ecological drivers.

Our summary analysis revealed no evidence to suggest groups of species are gathering or avoiding each other more than expected at random. No observed value for each number of species at a BRUV site ranging from 0-6 fell outside of the 95% confidence interval. Additionally, there were many 0 observed sightings relative to the highest number of species recorded together which was four. The frequent recording of zeros could be attributed to a low abundance of sharks and rays in the Florida Keys in general.

### 4.1 Species Relationships

When investigating specific pairwise species relationships, I found the co-occurrence of several species at 50 BRUV sites. This indicates there is evidence for the pairing of specific species. However, we did not find the appearance of these combinations significantly more than the expected values. Additionally, given that elasmobranch sightings were rare, the expected values based on the randomization method tended to be low.

The most frequent of these pairings, the nurse shark and yellow round ray, was recorded at 13 BRUV sites with an expected value of 11 BRUV sites. This result was likely due to the great abundance of the nurse shark and yellow round rays off the Florida Keys year-round (Castro, 2010). A tendency of nurse sharks to be in groups suggests that the presence of one nurse shark likely indicates there are others, thus increasing the frequency of this species in video-based observations. Additionally, their diet includes small fishes, invertebrates, squid, shrimps, crabs, spiny lobsters, and sea urchins which aligns closely to the diet of the yellow round ray (Castro, 2010). In a study by Fahy (2004), the yellow round ray was the most common elasmobranch in the coral reefs and coastal habitats of southeastern Florida. Their main prey choices consist of polychaetes and, like the nurse shark, small crustaceans (Quinn, 1996). Hence the similarity in their prey selection may draw them to similar locations.

I found that the southern stingray co-occurs more frequently than expected with the nurse shark. With an observed value of five, this species pair varied the most from its expected value with a difference of three BRUV sites. In a study by Gilliam (1991), the southern stingray exhibited an asynchronous feeding behavior and spent most of its time foraging in soft sediment

algal turf and sandy shoal habitats. They were found to be generalist feeders actively feeding throughout the day with a preference for decapod crustaceans and molluscs (Gilliam, 1991; Wrigglesworth, 2019). The southern stingray shares a preference for crabs and shrimp with the nurse shark and yellow round ray, yet why the southern stingray occurs only with the nurse shark and not with the almost equally as common yellow round ray despite their similar diets is intriguing. A study conducted by Ward-Paige et al. (2011) states that the yellow round ray is found in all habitats, thus eliminating the possibility that the appearance of the southern stingray with only the nurse shark and not the yellow round ray could be attributed to habitat preference.

The results of this study demonstrate the co-occurrence of the bonnethead shark with the yellow round ray (7) or nurse shark (6). Both species pairs were observed close to their expected results and were noted frequently. The bonnethead shark, as one of the most abundant sharks in southeastern North America and another common species located off the coast of the Florida Keys, can be found in sea grass beds as well as both sandy and muddy bottoms (Castro, 2010). The diet of the bonnethead shark consists of small fishes, blue crabs, shrimp, and acorn barnacles (Castro, 2010). A study conducted by Bethea et al. (2007) examined the stomach contents of juvenile and adult bonnethead sharks in which diet included crabs, seagrass, shrimps, cephalopods, and lobsters varying with region, age, and sex. Given that all three species are common, and they share preference for similar prey, the co-occurrence of the bonnethead shark with the nurse shark or yellow round ray is likely partially driven by these factors.

The four-time co-occurrence of the Caribbean reef shark and nurse shark could be related to their habitat preferences. The observed value of this species pair was slightly higher than the expected value. Similar to the nurse shark which resides on the ocean bottom frequenting rocky areas and deeper reefs, the Caribbean reef shark is found on the ocean floor near the edges of

coral reefs resting on rocky ledges, caves, and drop offs (Castro, 2010). Appearance together is not likely attributed to similar prey preferences given that the carribbean reef shark preys on flying fishes and small scombrid fishes (Castro, 2010).

Similar prey choice could drive the presence of the sharpnose shark with the yellow round ray (3) or nurse shark (3). Both species pairs were observed slightly more than their expected values. The sharpnose shark is a predator of small bony fishes and crustaceans, a diet like that of both the nurse shark and yellow round ray (Castro, 2010). This species is also one of the most abundant sharks in eastern north America, present off the Florida Keys throughout the year (Castro, 2010). It is primarily found over continental shelves, island terraces, and deep coral reefs of 10-30 meters (Compagno 1984; Compagno et al. 2005). Additionally, in a study conducted by Carlson et al. (2008) monitoring the Atlantic sharpnose shark within the Crooked Island Sound, Florida for over three years, sightings were recorded in all habitat types like the yellow round ray. This could also explain its frequent appearance with the other two most observed species in the study.

In the normalized results, the white spotted eagle ray occurred with the eagle ray more frequently than expected the most out of all combinations. The second pair with the greatest increase from the expected to observed results was the requiem shark with the southern stingray. Both pairs include an animal where the genus or species is unidentified and thus presents difficulty in exploring reasons as to why they would be found together. Additionally, lacking this information makes it difficult to hypothesize biological and ecological reasons as to why it was only recorded once other than eagle rays sharing a preference for prey.

The species pairs that did not show as strong results as the previously mentioned pairs, but still greater co-occurrences than expected was the requiem shark with the bonnethead and

nurse shark. The identification of requiem shark is broad; therefore, it is difficult to conclude why these species were found together. However, considering the bonnethead and nurse shark share similar prey preference for crustaceans and the requiem shark was found with both, it could be suggested that prey choice contributed to their presence together (Castro, 2010).

Regarding the appearance of the sharpnose shark with the broad identification of hammerhead shark, much of what is known about hammerhead shark biology and ecology is based on studies focusing on the scalloped hammerhead (*Sphyrna lewini*) (Roemer et al. 2016). The scalloped hammerhead has been described as a generalist feeder throughout their range by Kiszka et al. (2015) and Flores-Martinez et al. (2017). They have a diet of cephalopods, crustaceans, and fish (Torres-Rojas et al. 2010). The sharpnose shark is a predator of small bony fishes and crustaceans (Castro, 2010; Bethea et al. 2006) which could possibly contribute to their co-occurrence. On the other hand, given that the great hammerhead (*Sphyrna mokarran*) has been documented feeding on stingrays (Strong et al. 1990, Chapman & Gruber 2002), it is not likely that the appearance of these two specific species together could be contributed to similar prey preference. There is evidence in the literature for great hammerheads consuming other large sharks (Mourier et al. 2013; Roemer et al. 2016).

Finally, the nurse shark with the eagle rays could be contributed to a shared preference for prey. Eagle rays are known to feed on crustaceans, while nurse sharks are also predators of this prey (Castro, 2010; Schluessel et al. 2009). Being predators of the same organisms could influence their appearance together.

Certain species were only spotted once throughout all three years of the study. These species were comprised of the bull shark (2017), tiger shark (2016), eagle ray (2016), requiem shark (2016), and freckled guitar fish (2016). The bull shark is a coastal species that enters

freshwater and lives in lagoons until sexual maturity (Castro, 2010). It is commonly found at 25-100 meters (Castro, 2010) whereas most of the BRUVs in this study were placed above 20 meters, likely contributing to why this species was not recorded often.

The single occurrence of the tiger shark could be credited to their rarity. They are considered data deficient on the IUCN (2021) red list in addition to being uncommon due to overfishing (Castro, 2010). Furthermore, the lack of their appearance with other sharks and rays could be tied to their extensive diet that includes sea turtles and other sharks (Castro, 2010).

Finally, there is little information on the diet and spatial habits of the freckled guitarfish. Considering it is listed as vulnerable on the IUCN (2021) red list and decreasing, the absence of studies on this species suggest it is not common. This likely contributed to its infrequent appearance on a BRUV.

### 4.2 Caveats

There are two potential caveats concerning the quality of the BRUVs data. Our results demonstrate a decrease in the number of species recorded each year, however, the number and location of BRUVs deployed yearly was not standardized. For instance, in 2019 BRUVs were deployed only in the Miami-Dade region and recorded the smallest number of species. The lower number of BRUVs deployed coupled with a smaller study area likely influenced the decrease in species richness that year. Our study does not look at the effects of time on species distribution. We solely make conclusions based on the presence or absence of groups or co-occurrence of certain species.

The second caveat regards the pairs of species scoring high in the normalized data. Our results include animals such as the eagle ray and requiem shark that were only spotted once throughout the study. Given that the requiem shark and eagle ray were only recorded once, we

cannot say that these pairs will occur frequently, but our results do present direction for future studies to investigate the possible relationship between the two species in each pair. These species pairs also involve animals that were unidentified to the species, which the involvement of these species could also increase the likelihood of type II error.

#### 4.3 Potential

With regards to the future, BRUVs are becoming commonly used for studying fishes in tropical systems but are not being leveraged for their full potential. They are primarily used to look at single species distributions and the effect of environmental factors on these distributions (Espinoza et al. 2014; White et al. 2013; Barord et al. 2014). However, they have the capacity for exploring community dynamics while aiding in answering much broader questions. Here, we present a tool that allows us to ask these additional questions by determining if co-occurrence can be attributed to coincidence or to specific biological or ecological drivers. Thus, our method contributes to the effort of expanding the potential of BRUVs to analyze multi-species data (Espinoza et al. 2014; Osgood et al. 2019). Previous studies utilize BRUVs to assess the distribution patterns and abundance of sharks with methods that include, but are not limited to, ANOVA (De Vos et al. (2015), generalized linear mixed models (Osgood et al. 2019), and Chi squared contingency tables (White et al. 2013). Furthermore, it is important to address that our method of analysis is not a traditional parametric test. Here, we rely on permutations compared to a 95% confidence interval. Despite being a novel technique of analysis to our knowledge, it may have more power than the traditional methods given that certain assumptions about the distribution are not required.

### 5 CONCLUSION

In this study we assessed the co-occurrence of reef associated sharks and rays within the Florida Keys using BRUVs. We did not find strong evidence supporting animals gathering or avoiding each other more often than expected based on the number of species present in our global analysis. Furthermore, we documented the frequent co-habitation by the nurse shark and yellow-round ray in our pairwise analysis likely due to their shared preference for prey. Our normalized results revealed that the whitespotted eagle ray and eagle ray as well as the requiem shark and southern stingray occurred together more often than expected. Thus, we demonstrated the importance of understanding the factors affecting the distribution of shark and ray species in the Florida Keys, which can be used for developing conservation and management plans in the future to slow the rapid decline of large teleosts and sharks worldwide and move more towards ecosystem-based fisheries management (Myers & Worm, 2003). To further the efforts of MacNeil et al. (2020), future research should include direct measurements of the effects of temperature on the distribution of sharks and rays over extended periods of time using BRUVs. Long term monitoring is essential to understanding how global processes influence susceptible marine populations, especially in the context of a changing climate. Additionally, by using BRUVs we have the ability to observe subject animals in the field with several advantages over traditional methods. Future studies could also keep the number of BRUVs and deployment locations consistent. Finally, the efficiency of the National Marine Sanctuary at protecting the precious ecosystem of the Florida Keys could be assessed (Speed et al. 2018).

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