Characterization of a scalloped hammerhead (Sphyrna lewini) nursery habitat in portions of the Atlantic Intracoastal Waterway

Bryanna N. Wargat

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Characterization of a scalloped hammerhead (Sphyrna lewini) nursery habitat in portions of the Atlantic Intracoastal Waterway

Bryanna N. Wargat
University of North Florida, n01392049@unf.edu

CHARACTERIZATION OF A SCALLOPED HAMMERHEAD (*SPHYRNA LEWINI*) NURSERY HABITAT IN PORTIONS OF THE ATLANTIC INTRACOASTAL WATERWAY

By

Bryanna Wargat

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CERTIFICATE OF APPROVAL

The thesis "Characterization of a scalloped hammerhead (*Sphyrna lewini*) nursery habitat in portions of the Atlantic Intracoastal Waterway" submitted by Bryanna Wargat is approved by the thesis committee.

Dr. Jim Gelsleichter Committee Chair	Date
Dr. Bryan Franks	Date
Dr. Adam Rosenblatt	Date

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Content Page

ABSTRACT

The scalloped hammerhead shark (*Sphyrna lewini*) worldwide population has been in sharp decline, and they are currently listed as a globally critically endangered species by the IUCN. This warrants a need to identify and protect critical habitats for the species, such as nurseries, which promote stable populations. A section of the Tolomato River, in northeastern Florida, has shown to host large and consistent numbers of young of year scalloped hammerhead sharks. This gave cause to determine whether this portion of the Atlantic Intracoastal Waterway (ICW) serves as a nursery habitat for the species and to understand how the sharks used the area. To declare the Tolomato River as a nursery habitat, three criteria needed to be met: the species were more commonly found in the Tolomato River as opposed to other sites, individual sharks stayed in the area for long periods of time (weeks or months), and the species used the habitat repeatedly across years. To address these criteria, a catch composition analysis, habitat preference study, mark-recapture analysis, and acoustic tracking were conducted. The results from these studies indicated that scalloped hammerhead neonates have a preference for the Tolomato River compared to other nearby estuaries. They additionally showed that individual scalloped hammerhead sharks are using the habitat for extended periods of time and the species utilizes the Tolomato River annually. These results indicate that the Tolomato River serves as a nursery habitat for the scalloped hammerhead shark. Due to the established importance of nursery habitats to the welfare of shark populations, the identification of nurseries is often required in various management plans. Thus, data from this project contributes to the management of the scalloped hammerhead shark, a species in need of protection.

INTRODUCTION

In 1996, the reauthorization of the Magnuson-Stevens Fishery Conservation and Protection Act emphasized the importance of essential fish habitat (EFH) to the maintenance of healthy fish populations; EFH's are areas that are necessary to the spawning, feeding, breeding, or growth of a marine organism (Magnuson-Stevens Fishery and Conservation Act 1996). Nursery habitats are considered to be EFH due to their importance in the life histories of various coastal shark species (Spring, 1967; Bass, 1978; Castro, 1993; Simpfendorfer & Milward, 1993; Hueter et al. 2005; Heupel et al. 2007; McCallister et al. 2013). These habitats are typically found in bays, sounds, and littoral zones (Sadowsky, 1965; Clarke, 1971; Branstetter, 1987). Pregnant, mature females may give birth in or adjacent to nurseries and their pups often stay in the area for extended periods. Some species stay in nursery habitats until they reach adulthood (Springer, 1967). In these habitats, there is often a surplus of food and protection from predators (Springer, 1967; Bass, 1978; Branstetter, 1990; Castro, 1993; McCallister et al. 2013). Consequently, nurseries can potentially improve the survival of juveniles and contribute to population stability. For these reasons, the use of nurseries has been a reproductive strategy of some chondrichthyan species for millions of years (Duncan and Holland, 2006).

Nurseries are often identified in fisheries management plans (FMPs) as they are critical to the welfare of shark populations. Various shark species have experienced population decline in recent years due to exploitation (NMFS, 2006). These animals are susceptible to population decline due to a late sexual maturity and low fecundity (Stevens et al. 2000; Dulvey et al. 2008). To promote the rebuilding of these species' populations, fishery managers protect areas of importance, such as nursery habitats (Bonfil, 1997; Kinney and Simpfendorfer, 2008). The survivorship of neonates and juveniles strongly influences the total population size of any species (Heppell et al. 1999; Cortes, 2002). Unfortunately, these ecosystems are often vulnerable to degradation, which can ultimately impact the survival of neonates; many nurseries are in close proximity to human populations as they are often found in shallow water (Rountree & Able, 1996; NMFS, 1999; Martin, 2005; Lotze et al. 2006; Jennings et al. 2008). Therefore, there is a need to identify these nursery habitats to develop effective conservation plans.

Previously, McCallister et al. (2012) reported that coastal bays and estuaries in the northeast Florida coast may provide critical nursery habitat to several economically and ecologically important shark species, including the Atlantic sharpnose shark *Rhizoprionodon terraenovae* and the blacktip shark *Carcharhinus limbatus*. More recently, anecdotal evidence has suggested that the Tolomato River may represent essential fish habitat for the scalloped hammerhead shark *Sphyrna lewini*. The Tolomato River is a river system in northeast Florida, south of the city of Jacksonville. It runs along the Atlantic coast and leads to the St. Augustine inlet. This body of water is part of the intracoastal waterway (ICW), which runs from Massachusetts to Florida. Additionally, a portion of the Tolomato River is found within the bounds of the GTM Research Reserve, which is one of 29 National Estuarine Research Reserves in the United States.

The scalloped hammerhead shark is a large, viviparous shark species that is found along continental margins and oceanic islands in the temperate and tropic zones (Compagno, 1984). Furthermore, this is one species of shark that utilizes nearshore nurseries throughout their range (Clarke, 1971; Snelson & Williams, 1981; Compagno, 1984; Branstetter, 1990; Castro, 1993; Simpfendorfer & Milward, 1993). There are six existing distinct population segments (DPS) of the scalloped hammerhead throughout the world (Miller et al. 2014). The Northwest Atlantic and Gulf of Mexico DPS of *S. lewini* is found from New Jersey to Brazil in the Western Atlantic and in the Gulf of Mexico and Caribbean Sea (Compagno, 1984); this is an area that includes the Tolomato River. Although this DPS is not considered endangered, the population has experienced exploitation in the past (Hayes et al. 2009). Due to a late sexual maturity at approximately 15 years of age and low fecundity of 10 to 40 pups every other year, the population exhibits a low growth rate (Branstetter, 1987; Duncan and Holland, 2006; Piercy et al. 2007). As a result, the species is slow to rebuild after experiencing population decline. The virgin, or unfished, population was estimated to range from 142,000 to 169,000 individuals in 1981 (Hayes et al. 2009). The population then experienced an estimated 83% decline due to fishing pressure by commercial, recreational, and IUU (illegal, unreported, and unregulated) fishing (Hayes et al. 2009; Miller et al. 2014). Since 1996, the population has begun to rebuild at a slow rate. Recent stock assessments in 2005 estimate that the Northwest Atlantic and Gulf of Mexico adult stock includes 24,850 individuals (Hayes et al. 2009). However, Hayes et al. (2009) estimates that the population still has a 95% probability of being overfished in the future. These predictions are supported by the presence of individuals from this DPS in the international shark fin trade. From a survey of *S. lewini* fins collected from the Hong Kong market, 21% originated from the western Atlantic (Chapman et al. 2009). Furthermore, it has been determined that high at-vessel fishing mortality is the largest threat to scalloped hammerhead sharks in this DPS (Miller et al. 2014). Hence, there is a need to protect the species and identify the areas that could serve as nursery habitats so that they may be incorporated into fishery management plans.

At this point in time, the Tolomato River is not designated as EFH for the scalloped hammerhead shark and is not protected as such (NOAA, 2017). Protection of this site, if identified as a nursery, would be critical in conservation efforts for a species that is considered globally critically endangered by the IUCN (Rigby et al. 2019). Therefore, there is a need to determine whether the Tolomato River serves as EFH. Although pilot work suggests that the Tolomato River may serve as a scalloped hammerhead nursery, research must be conducted to determine if the habitat meets the expectation of a nursery habitat as described in the literature. Springer (1967) first described nursery habitats as areas in which parturition occurs and where neonates and juveniles spend the first part of their lives in. Bass (1978) then defined the differences between primary and secondary nurseries. In primary nurseries, young sharks are born and present up to a year. Older juveniles are found in secondary nursery habitats. These concepts have been accepted by the scientific community, though criteria of a nursery habitat were not clear until Heupel et al. (2007) proposed a new definition. In order to be considered a nursery, a habitat should meet three criteria according to Heupel et al. (2007): the shark species in question must be more commonly found in that area in comparison to other sites, individual sharks should tend to stay in the area for long periods of time, and the habitat should be used repeatedly across years. A habitat must meet these criteria to be distinguished as a nursery under management plans.

To address these criteria, the species composition of the Tolomato River was initially analyzed to determine whether the site was annually used by the species. This was followed by a habitat preference study, mark-recapture analysis, and acoustic tracking. The preference study was completed to see if the species is found more often in the Tolomato River in comparison to nearby habitats. The mark-recapture survey was conducted to determine the extent of time individual sharks utilized the area. The acoustic tracking was completed to determine if the sharks stay in the area for extended periods of time and to assess habitat preferences. These studies are needed to properly assess the Tolomato River’s status as a nursery habitat and determine appropriate conservation measures.

METHODOLOGY

Characterization of shark fauna in the Tolomato River

Sampling

Bottom longline surveys were completed in the Tolomato River (Figure 1) from the years of 2010 to 2019 using the methods described in McCallister et al. (2013). Longlines were composed of a 250-300 meter #8 braided nylon mainline, which were anchored at both ends and marked with buoys. The line contained 50 branchlines, each composed of a 1-meter, 90-kg test monofilament leader. Each branchline consisted of a size 120 stainless steel longline snap, 4/0 swivel, and a 12/0 barbless circle hook. All hooks were baited with Atlantic Mackerel, *Scomber scombrus*. These lines were soaked for 15 minutes, as opposed to the 30-minute soak time used by McCallister et al. (2013), to minimize mortality of *S. lewini*. Set locations were haphazardly selected. This was based on varying weather conditions, tides, and maritime conditions present at the time of sampling. At each sampling site, environmental data were collected using a YSI Pro2030 (YSI, Inc., Yellow Springs, Ohio). This included bottom water temperature (°C), salinity (ppt), dissolved oxygen (mg/L), and conductivity (mS). Maximum and minimum water depth (m) were recorded for each set, and the mean depth was calculated.

For each shark caught, biological data were collected. Sharks were identified to species, measured (cm), sexed, and weighed (kg) when possible. Length measurements included precaudal length (PCL), fork length (FL), and stretched total length (STL). Life stage was classified as young of year (YOY, Age 0), juvenile, or adult. These were determined by using length at maturity as described in published literature. If the individual was categorized as Age 0, the umbilical scar status was additionally recorded when using five possible categories: 1= umbilical remains present, 2 = open or fresh scar, 3= partially open, some healing, 4= well-

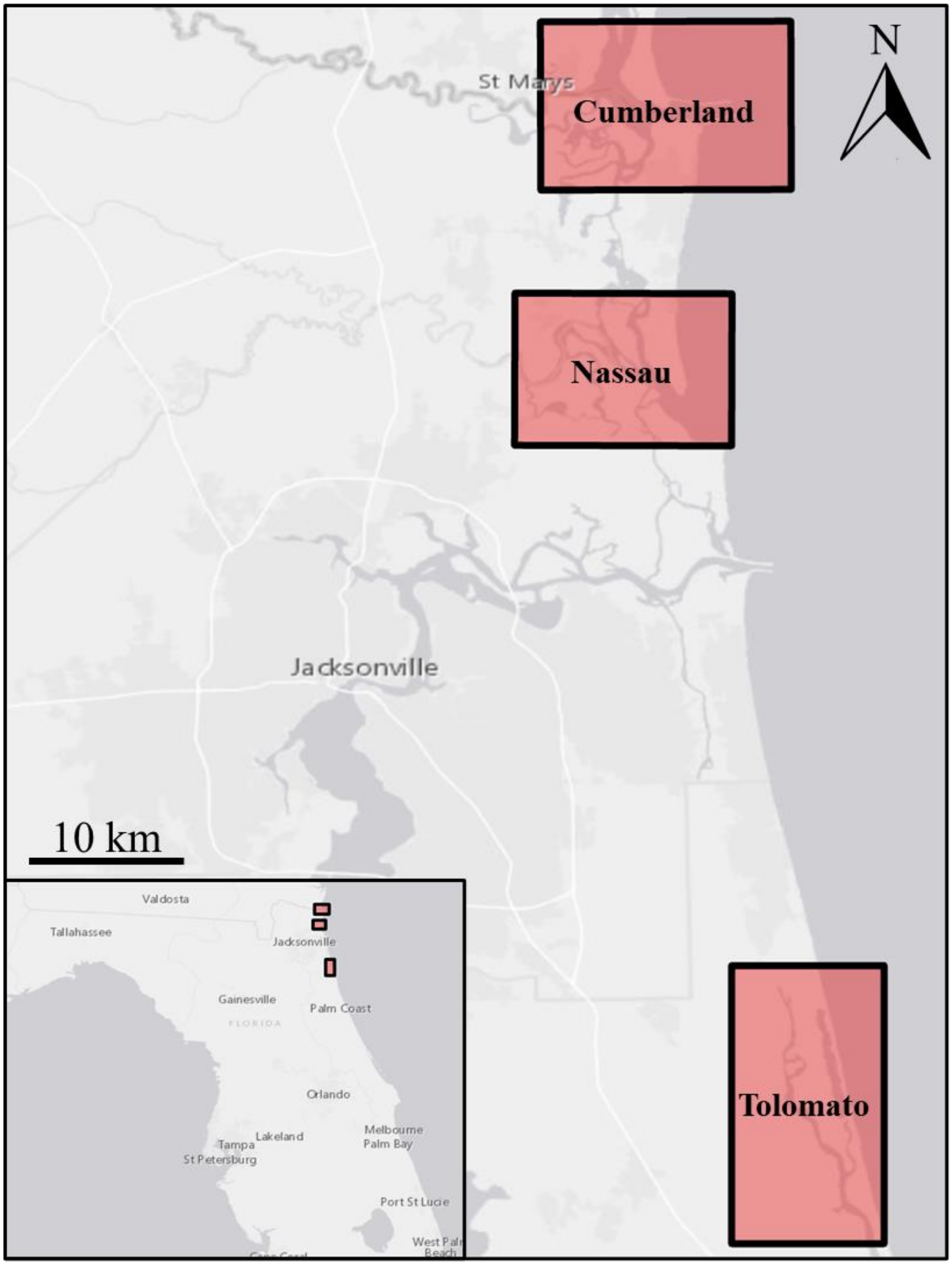

Figure 1. Map of all three study sites used to assess scalloped hammerhead shark presence in northeast Florida.

healed, scar is visible, and 5= no scar present.

Data Analysis

Based on data collected from all bottom longlines completed, catch composition was described. For YOY scalloped hammerhead sharks caught, correlations between all possible combinations of PCL, FL, and STL recorded were determined via Pearson correlation coefficient. Linear regressions were performed for FL to PCL, PCL to STL, and FL to STL. Sharks with truncated caudal fins or inaccurate measurements were excluded from these analyses. Average FL was then compared via a one-way ANOVA across the months of May through August to assess the growth of the species. Because length data was only available for two sharks during the month of September, this month was excluded from the analysis. Months were subsequently grouped into homogenous subsets based on a Tukey post-hoc test. For all morphometric analyses, results were considered significant if $p < 0.05$.

Catch rates of YOY *S. lewini* were expressed as catch per unit effort (CPUE) for each set. This was calculated as the number of YOY scalloped hammerhead sharks caught per 50 hooks. Abundance trends were examined by comparing the CPUEs across months and years via Kruskal-Wallis nonparametric analysis because data did not meet assumptions of normality and homoscedasticity. Months and years were subsequently grouped into homogenous subsets as determined by a stepwise-stepdown post-hoc test with multiple comparisons.

To evaluate habitat use and preferences, a binary logistic regression model was used to assess the effects of environmental conditions on the presence/absence of YOY *S. lewini*. Parameters included average depth, bottom temperature, salinity, dissolved oxygen, and conductivity. In this analysis, only sets for which all environmental parameters were recorded were used. Conditions were considered to significantly influence the probability of catching a scalloped hammerhead shark if $p < 0.05$. CPUE was additionally mapped geographically via ArcGIS Pro to potentially indicate areas of importance within the Tolomato River.

Comparisons of *S. lewini* abundance in the Tolomato River and other northeast Florida estuaries

Sampling

Bottom longline surveys were conducted in the Cumberland Sound/St. Marys River basin and Nassau Sound from 2009 to 2019 following the methods described in McCallister et al. (2013). Cumberland Sound can be found between Cumberland, Island, Georgia, and Amelia Island, Florida, at the mouth of St. Mary's River. Nassau Sound is south of the Cumberland Sound and is found in Florida between Amelia Island and Big Talbot Island. Nassau Sound resides as a junction of the Nassau and Amelia rivers and Sister's Creek. When including the Tolomato River study area, these three sites encompass all available inshore habitats, apart from the St. John's river, north of St. Augustine to the Florida-Georgia border (Figure 1). The surveys conducted in the Cumberland and Nassau Sounds were identical to that completed in the Tolomato River site with a single exception; that is, soak time was 30 min in duration (the shorter soak time used in the Tolomato site was established to minimize mortality of *S. lewini*).

Data Analysis

Catch data collected from all three surveys were used to test the first criterion of nursery habitat identification; that is, whether the species in question (*S. lewini*) prefers the location in question (the Tolomato River) to other available habitats (Cumberland Sound/St. Marys River or Nassau Sound). Only sets for which all environmental parameters were recorded were used in analyses. For each site, YOY scalloped hammerhead catch rates were determined and compared between the three sites using Kruskal-Wallis nonparametric analysis. Sites were grouped into homogenous subsets as determined by a post-hoc test. Catch rates were expressed as catch per unit effort. Initially, this was calculated as the number of sharks caught per 50 hook-hours in order to standardize for the difference in soak time between the sites. However, this was also completed with CPUE representing the number of sharks that were caught per 50 hooks.

A general linear model was also used to further determine whether site was an influencing factor on *S. lewini* abundance. Depth, salinity, conductivity, dissolved oxygen, and bottom temperature were used as covariates in the model to account for varying environmental conditions across the three sites. All potential interactions between covariates were included in this analysis. Factors that were not significant were removed from the model until only significant variables remained. Variables were considered to be significant if $p < 0.05$. Once all factors that significantly impacted YOY scalloped hammerhead abundance were identified, these variables were compared among the three sites. This was done to better determine whether the environmental conditions were responsible for the significant difference in scalloped hammerhead YOY abundance among the three sites.

<u>Mark-Recapture Study</u>

<u>Sampling</u>

A conventional tagging study was completed as part of the bottom longline survey conducted in the Tolomato River from 2019-2020. Weekly sampling occurred during the summer of 2019 and sporadically during 2020 because of sampling limitations posed by the COVID-19 pandemic. The longline had the same setup as the bottom longline survey of the

Tolomato River from 2010 to 2019, during which five sets were fished each day at haphazardly selected locations. For each set, biological and environmental data were collected as previously described. All scalloped hammerhead sharks caught were tagged with a numbered dart tag that was inserted into the musculature ventral to the first dorsal fin and released. Any recaptures made by UNF staff or individuals not affiliated with UNF (e.g., other biologists, anglers, etc.) were noted, and the date and location of recapture were recorded.

Data Analysis

Mark-recapture data were used to test the 2nd and 3rd criteria of nursery ground identification; that is, whether individual sharks tend to stay in the area for long periods of time, and that the species uses the area on an annual basis. Overall rate of recapture was determined. For each shark recaptured, time at large was recorded. Furthermore, the location of release and recapture sites were mapped using ArcGIS Pro, and recapture distance was determined by calculating the distance between the two points following the midline of the river.

Acoustic Tracking

Sampling

Sampling was conducted during the summer of 2020 via rod and reel fishing in which circle hooks were baited with mackerel and squid. This method of fishing was done to limit the stress put on the animals prior to tagging. Fishing locations were selected haphazardly. When a scalloped hammerhead shark was caught, biological data were collected as described above. If the shark was considered to be in a good condition, an acoustic transmitter (Innovasea V9) was attached to the first dorsal fin externally via a rototag; the tags that were deployed weighed 6.74

g and 6.94 g (Figure 2). Transmitters had an estimated battery life of 13 days and pulsed continuously at various acoustic frequencies (60, 63 kHz).

Active Tracking

In total, two scalloped hammerhead sharks were tracked, and a second track was completed for the second shark. Each shark was actively tracked from the time of capture to dusk. Only one shark was tracked at a time to avoid overlap of signals. Tracking started at the location of capture. Once the shark was released, it was manually tracked using a directional hydrophone (Innovasea VH110) and acoustic receiver (Innovasea VR100). The boat maintained a distance of at least 10 m from the sharks at all times to avoid interfering with movement patterns; though, this could not be guaranteed. Shark location was recorded every five minutes using a GPS, along with bearing and approximate distance from the boat. Depth (m) and tidal stage were additionally recorded at these intervals. Every 15 minutes, environmental data was collected via YSI Pro2030 (YSI, Inc., Yellow Springs, Ohio). This included temperature, D.O., conductivity, and salinity. Longer-term occurrences of the tagged sharks in the study area were also examined by conducting weekly surveys of transmitter detection using the omnidirectional hydrophone, as described recently in Rosende-Pereiro & Corgos (2018). If a tagged shark was detected, the VH110 directional hydrophone was used to locate the shark's position, and the shark was subsequently tracked until dusk.

Data Analysis

Initially, new positional fixes representing the position of the shark for each track were generated using estimated distance from the boat and bearing in relation to the original

Figure 2. YOY *S. lewini* with Innovasea V9 acoustic transmitter externally attached to the first dorsal fin prior to active tracking. Photograph taken by Clark Morgan.

geographic positions recorded, which represented the location of the boat. Each track was mapped via ArcGIS Pro using these positional fixes, and home range analyses were conducted. To determine the extent of each track's range, the total activity space was found using minimum convex polygon analysis via GIS. Areas outside the bounds of the river were manually removed from the total area. Activity area was also determined by using the 95% fixed-kernel utilization distribution (KUD) method with the Adehabitat package in R. 50% KUD's were also determined, which represent areas of repeated use or preference (Yeiser et al. 2008). While minimum convex polygons are used to determine the extent of an individual's range, the 50% KUD's illustrate the use of that range. Boundaries of the KUD's produced in R were mapped with GIS, and areas outside the river's boundary were clipped manually. This was accomplished by reshaping the polygon by hand, moving vertices to ensure that the area did not extend onto land. The area used by the animals was also described by determining the linearity index (LI) for each track. LI is used to determine if tracks are linear or nonlinear. While linear movements indicate directed movements, nonlinear movements indicate reuse. To calculate LI, the distance between the first and last position was divided by the total distance traveled. If this value was equal to one, the track was linear. Values near 0 were indicative of nonlinear movement paths (Rechisky and Wetherbee, 2003). If the straight-line used to determine distance between the first and last positional fixes passed over land, the distance between the two points was found by following the midline of the river.

Movement was described by rate of movement (ROM), direction of travel (upriver, downriver), and tidal stage. ROM was calculated by dividing the distance between successive positional fixes by the interval time (five minutes). Square root transformations were used so that ROM data could be analyzed parametrically. Average ROM was compared across the three tracks using an ANOVA, and homogenous subsets were indicated by a Tukey post-hoc test. Direction of travel in degrees (bearing) was calculated to describe the angle of movement between consecutive positions. Any movement northward was categorized as upriver, and any movement southward was categorized as downriver. This was based on the north-south orientation of the river. The ROM was compared between upriver and downriver travel for all tracks conducted with Student t-tests. ROM and bearing were additionally compared across all tidal stages using an ANOVA and Kruskal-Wallis test, respectively. All tracks were grouped for these analyses to account for a complete tidal cycle.

Water quality data were analyzed following relevant methods reported by Ortega et al. (2009). Average depth, temperature, salinity, D.O., and conductivity were compared among the three tracks conducted using Kruskal-Wallis tests. Then, potential preferences for each water quality variable were determined with the use of multiple linear regressions with bootstrapping. This compared the average habitat conditions (depth, temperature, salinity, D.O., and conductivity) recorded with the latitudinal position of the shark. This was chosen due to the north-south orientation of the river. Based on the R squared values generated by the models, tracks for the second animal were grouped. In all statistical tests, results were considered to be significant if $p < 0.05$.

Ultimately, these data collected from these tracks were primarily used to determine whether neonates spend extended periods of time in the proposed nursery habitat, contributing to efforts to test the 2nd criterion of nursery ground identification. This was also done to further indicate areas of importance to the species within the Tolomato River.

RESULTS

Characterization of shark fauna in the Tolomato River

A total of 444 longline sets were completed in the Tolomato River from 2010-2019 during the months of April-November. 618 sharks were caught, representing 10 species. Of those sharks caught, 248 (40.1%) were identified as scalloped hammerhead sharks. Apart from three individuals where life stage was not recorded, all *S. lewini* were categorized as YOY (Table 1). For 224 *S. lewini*, in which umbilical scar status was recorded, 0.89%, 11.16%, and 87.95% were assigned to categories 2, 3, and 4 respectively; no sharks were listed as having a 1 or 5 status. Morphometric data collected from 236 scalloped hammerhead sharks were used in length analyses. Fork lengths for these animals ranged from 29 to 45 cm. Relationships between FL and PCL (Figure 3), PCL and STL (Figure 4), and FL and STL (Figure 5) were all significantly correlated (Pearson Correlation, p= 0.000). Linear regression analyses resulted in the following: PCL = -0.2348 + 0.9085*FL (r^2= 0.964), STL = 2.963 + 1.381*PCL (r^2= 0.948), STL = 1.478 + 1.286*FL (r^2=0.959). Fork length varied significantly among the months of May through August (Figure 6; p= 0.002). September was excluded in this analysis as there were only two samples recorded for that month. Average fork length was lowest in the month of May (36.778 cm) and greatest in July (39.074 cm). The average fork length recorded for the months of June and August were not significantly different from each other, and they were grouped into the same homogenous subset as determined by the post-hoc test.

The average CPUE of scalloped hammerhead sharks was 0.553 sharks 50-hooks^{-1} (SD = 1.080) from 2010 to 2019. YOY *S. lewini* were caught consistently throughout the ten-year survey, but average CPUE varied significantly among years (Kruskal-Wallis, p = 0.000) Annual CPUE was highest in 2010 with an average CPUE of 2.729 sharks 50-hooks^{-1} and lowest in 2012

Table 1. Species composition, abundance, percent of total catch, sex, and life stage for all sharks caught in the Tolomato River from 2010 to 2019. Species are listed in order of overall abundance from most to least abundant; NS = sex unknown, NR = not recorded.

			Sex			Life stage			
Shark Species	No. caught	% of catch	Male	Female	NS	Age 0	Juvenile	Adult	NR
Scalloped Hammerhead, *Sphyrna lewini*	248	40.1	133	108	7	245	0	0	3
Atlantic Sharpnose, *Rhizoprionodon terrenovae*	152	24.6	70	75	7	144	1	5	2
Finetooth, *Carcharhinus isodon*	72	11.7	34	36	2	52	17	2	1
Blacktip, *C. limbatus*	64	10.4	25	36	3	63	1	0	0
Sandbar, *C. plumbeus*	60	9.7	34	26	0	57	3	0	0
Bonnethead, *S. tiburo*	12	1.9	4	8	0	3	9	0	0
Nurse, *Ginglymostoma cirratum*	4	0.6	2	1	1	0	3	0	1
Bull, *C. lecuas*	3	0.5	1	0	2	0	3	0	0
Lemon, *Negaprion brevirostris*	2	0.3	0	2	0	0	2	0	0
Dusky smooth-hound, *Mustelus canis*	1	0.2	1	0	0	0	1	0	0
Total	618	100.0							

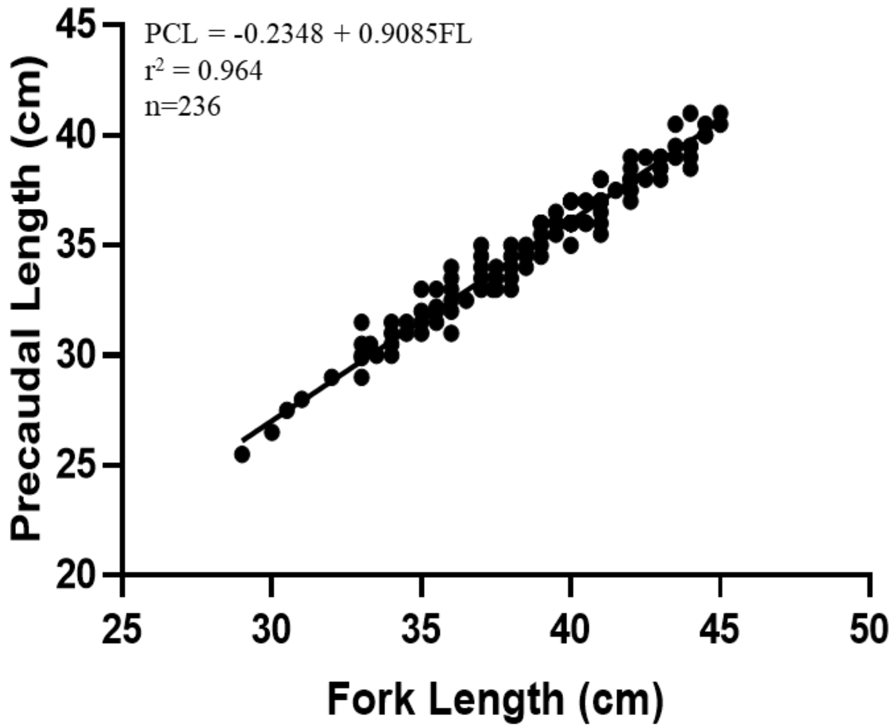

Figure 3. Relationship between precaudal lengths (PCL) and fork lengths (FL) of YOY *S. lewini* caught in the Tolomato River between 2010-2019. Results from linear regression analysis are included.

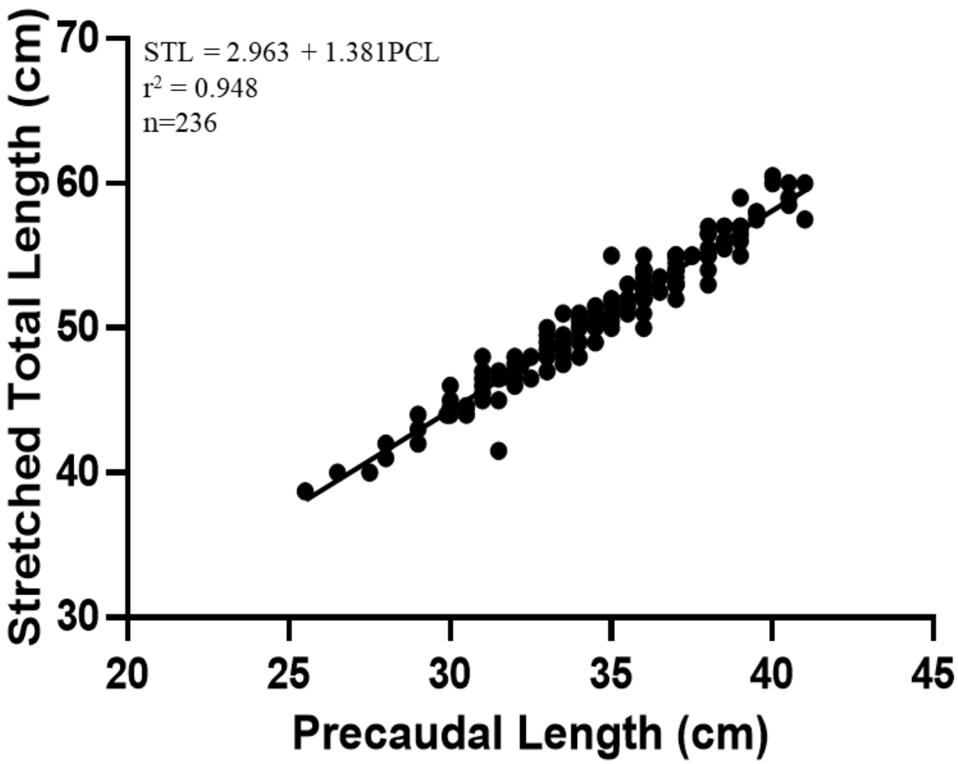

Figure 4. Relationship between stretched total lengths (STL) and precaudal lengths (PCL) of YOY *S. lewini* caught in the Tolomato River between 2010-2019. Results from linear regression analysis are included.

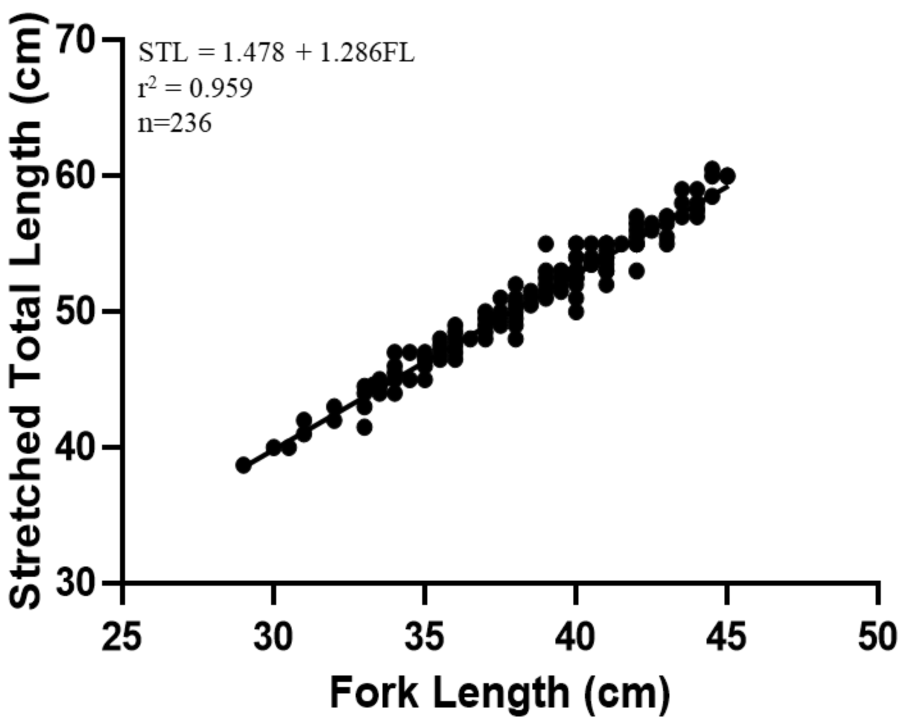

Figure 5. Relationship between stretched total lengths (STL) and fork lengths (FL) of YOY *S. lewini* caught in the Tolomato River between 2010-2019. Results from linear regression analysis are included.

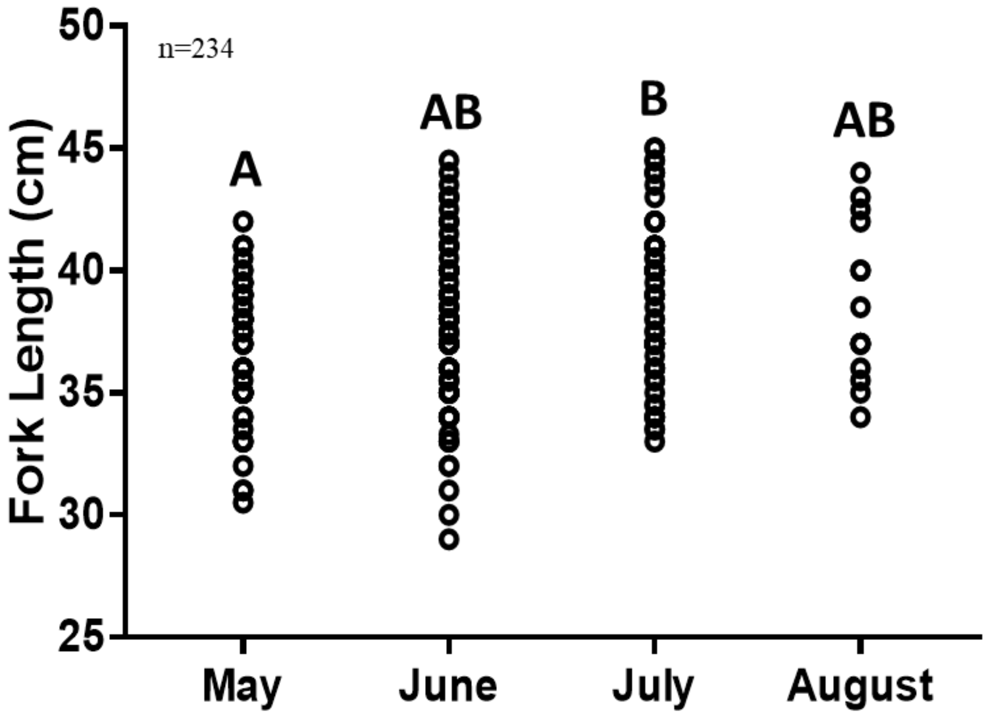

Figure 6. Fork lengths of YOY *S. lewini* caught in the Tolomato River during the years of 2010-2019 divided by month of capture. Months are grouped into homogenous subsets as determined by a Tukey post-hoc test with multiple comparisons following one-way ANOVA analysis (p=0.002). Sample size is presented.

with an average CPUE of 0.262 sharks 50-hooks^{-1} (Figure 7). Average CPUE also varied significantly among the months (Kruskal-Wallis, p = 0.000). Monthly CPUE increased from 0.737 sharks 50-hooks^{-1} in May to a maximum of 0.796 sharks 50 hooks^{-1} in June (Figure 8). Following this peak, monthly CPUE declined until no sharks were caught in October. The months of May, June, and July were grouped into one homogenous subset, while all other survey months were grouped into the other subset. No sharks were caught during the months of April, October, or November.

Sets for which all environmental parameters were recorded were used to infer environmental preferences of the species (n= 350). The binary logistic regression was performed to determine the effects of depth, temperature, salinity, D.O., and conductivity on the probability of catching a YOY scalloped hammerhead shark. The model indicated good fit (Hosmer and Lemeshow Test; p= 0.171) and correctly classified 71.7% of cases. Increasing D.O. conditions (p= 0.001) were associated with a reduction in the likelihood of catching a scalloped hammerhead shark (Table 2).

The CPUE of all sets completed over the ten-year survey were mapped via GIS and addressed visually (Figure 9). The map indicated that the majority of sets that caught scalloped hammerhead sharks occurred around Pine Island and within Pine Island Sound.

<u>Comparisons of S. lewini abundance in the Tolomato River and other northeast Florida estuaries</u>

S. lewini catch rates were compared among sets completed in the Cumberland Sound (n= 354), Nassau Sound (n= 327), and Tolomato River (n= 350). Catch rates (Sharks/ 50 hook-hours) were significantly different among the three sites (Kruskal-Wallis, p= 0.000; Figure 10). The Tolomato River experienced higher catch rates as opposed to the Cumberland and Nassau

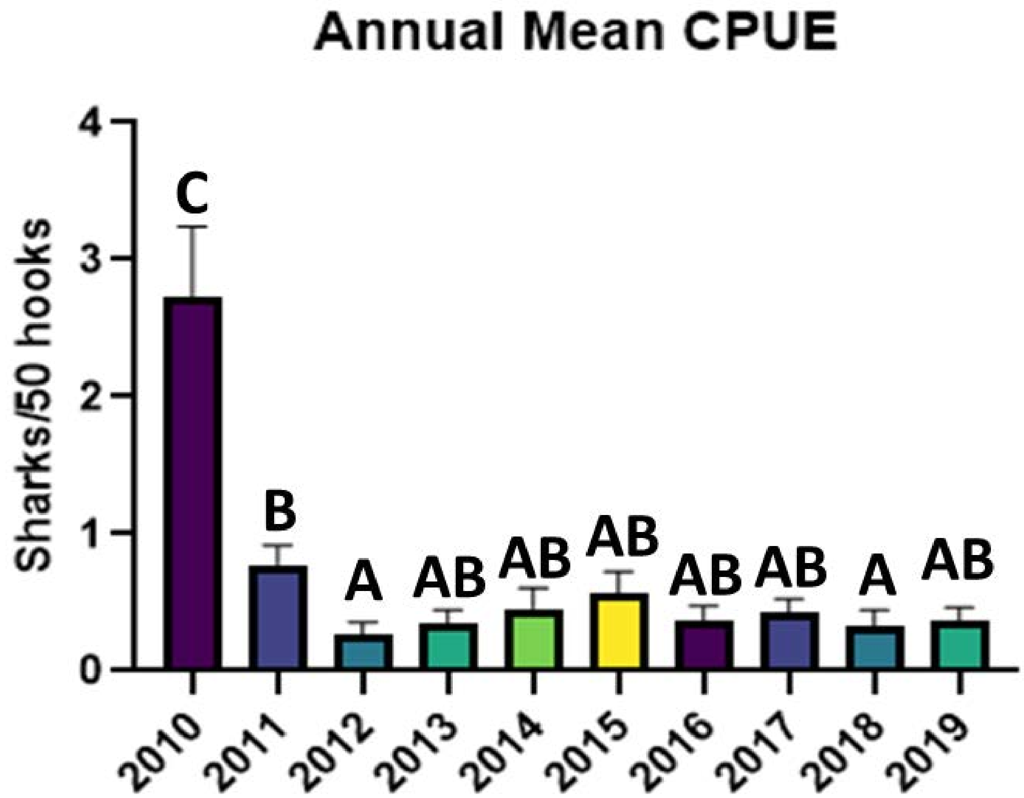

Figure 7. The average CPUE for YOY scalloped hammerhead sharks in the Tolomato River per year from 2010-2019 with error bars representing SE. Years are grouped into homogenous subsets as determined by a post-hoc test with multiple comparisons following Kruskal-Wallis nonparametric analysis ($p = 0.000$).

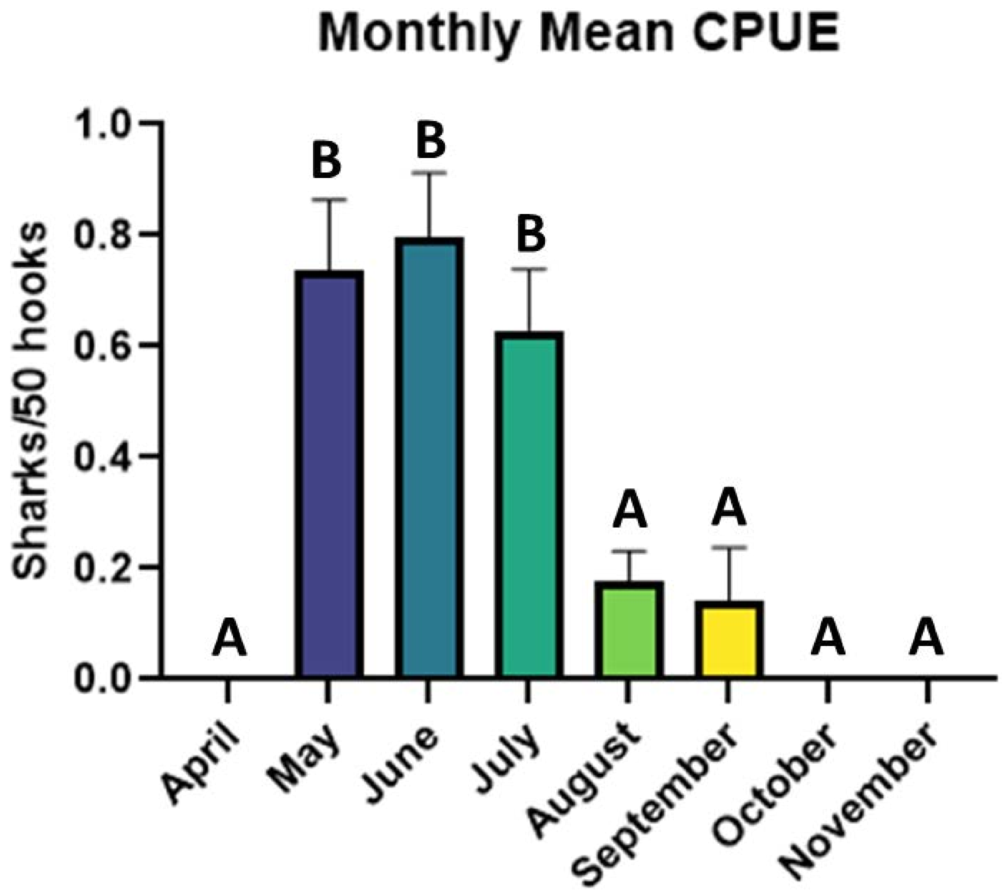

Figure 8. The average CPUE for YOY scalloped hammerhead sharks in the Tolomato River per month from 2010-2019 with error bars representing SE. Months are grouped into homogenous subsets as determined by a post-hoc test with multiple comparisons following Kruskal-Wallis nonparametric analysis (p =0.000).

Table 2. Results of binary logistic regression model analyzing the effects of environmental parameters on the presence/absence of YOY scalloped hammerhead sharks on bottom longlines conducted from the years 2010-2019 in the Tolomato River.

Environmental parameter	B	S.E.	Wald	Exp(B)	*P*
Average depth (m)	0.038	0.045	0.692	1.038	0.405
Bottom Temperature(°C)	0.060	0.071	0.709	1.062	0.400
Salinity (ppt)	0.053	0.053	1.004	1.054	0.316
Dissolved oxygen (mg/L)	-0.496	0.151	10.741	0.609	0.001
Conductivity (mS)	0.042	0.031	1.807	1.043	0.179
Constant	-4.031	2.412	2.793	0.018	0.095

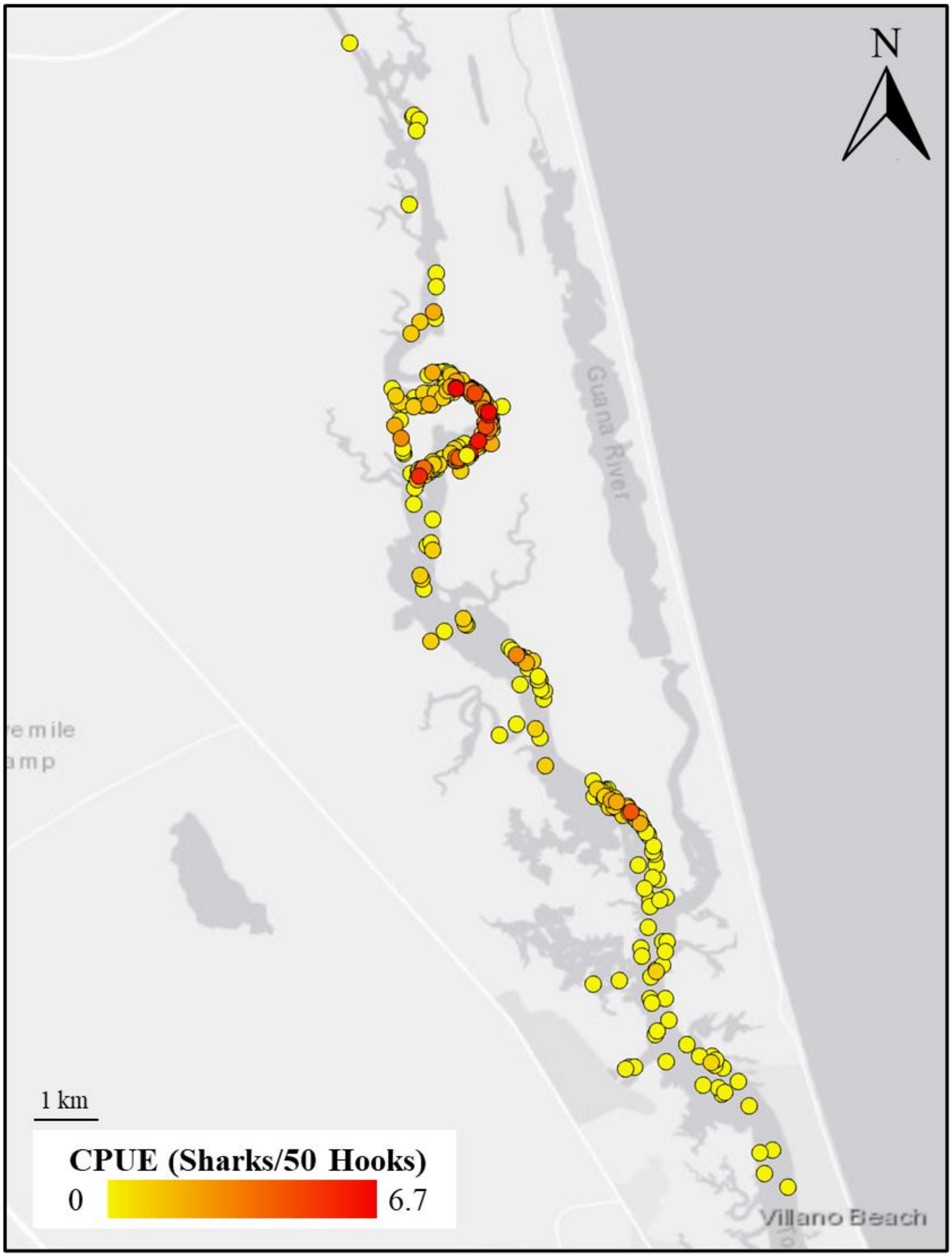

Figure 9. CPUEs associated with geographic location of bottom longline sets completed in the Tolomato River from 2010-2019. Map completed via ArcGIS Pro.

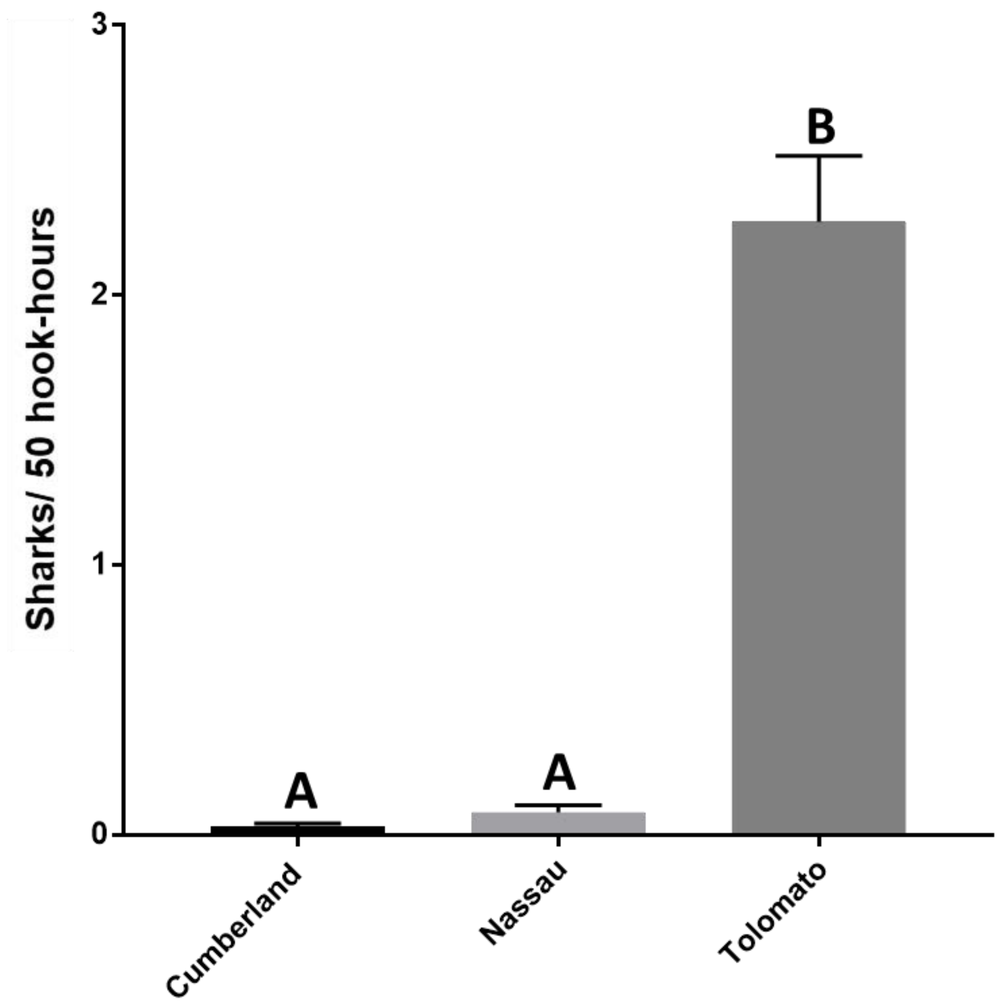

Figure 10. The average CPUE (sharks per 50 hook-hours) of sets used in GLM analysis, with error bars representing SE. Sets completed in Cumberland Sound (n=354), Nassau Sound (n=327), and the Tolomato River (n=350) are presented. Sites are grouped into homogenous subsets as determined by a post-hoc test with multiple comparisons following Kruskal-Wallis nonparametric analysis (p=0.000).

Sounds, which were grouped into the same homogenous subset. Kruskal-Wallis analysis resulted in the same outcome when the CPUE was expressed as sharks per 50 hooks ($p= 0.000$; Figure 11). As a result, this unit of CPUE was used for all subsequent analyses.

A general linear model was used to determine whether site or any other environmental factors influenced *S. lewini* abundance in the northeast region of Florida ($n= 1031$). Site, salinity, and the interaction between salinity and conductivity all had significant effects on abundance (Table 3). Average salinity and conductivity conditions are presented in Table 4. Among all the sets conducted in the northeast Florida estuaries, salinity was not significantly different between sets that caught at least one scalloped hammerhead shark and those that caught none (Figure 12). However, average salinity was significantly different among the three sites (Figure 13). Conversely, conductivity was significantly different between sets in which *S. lewini* were absent and present (Figure 14), and there was no significant difference in average conductivity between the three sites (Figure 15).

Mark-Recapture Study

During the years of 2019 and 2020, a total of 34 scalloped hammerhead sharks were caught, tagged, and released in the Tolomato River. 24 sharks were tagged in 2019, while the remaining 10 were tagged in 2020. Three males were recaptured, resulting in a total recapture rate of 8.824%. All sharks were recaptured during the same year in which they were released. Days at liberty ranged from 6 to 59 days, and distance between release and recapture locations ranged from 1.263 to 4.396 km (Table 5). Recaptures were mapped via ArcGIS Pro (Figure 16).

Acoustic Tracking

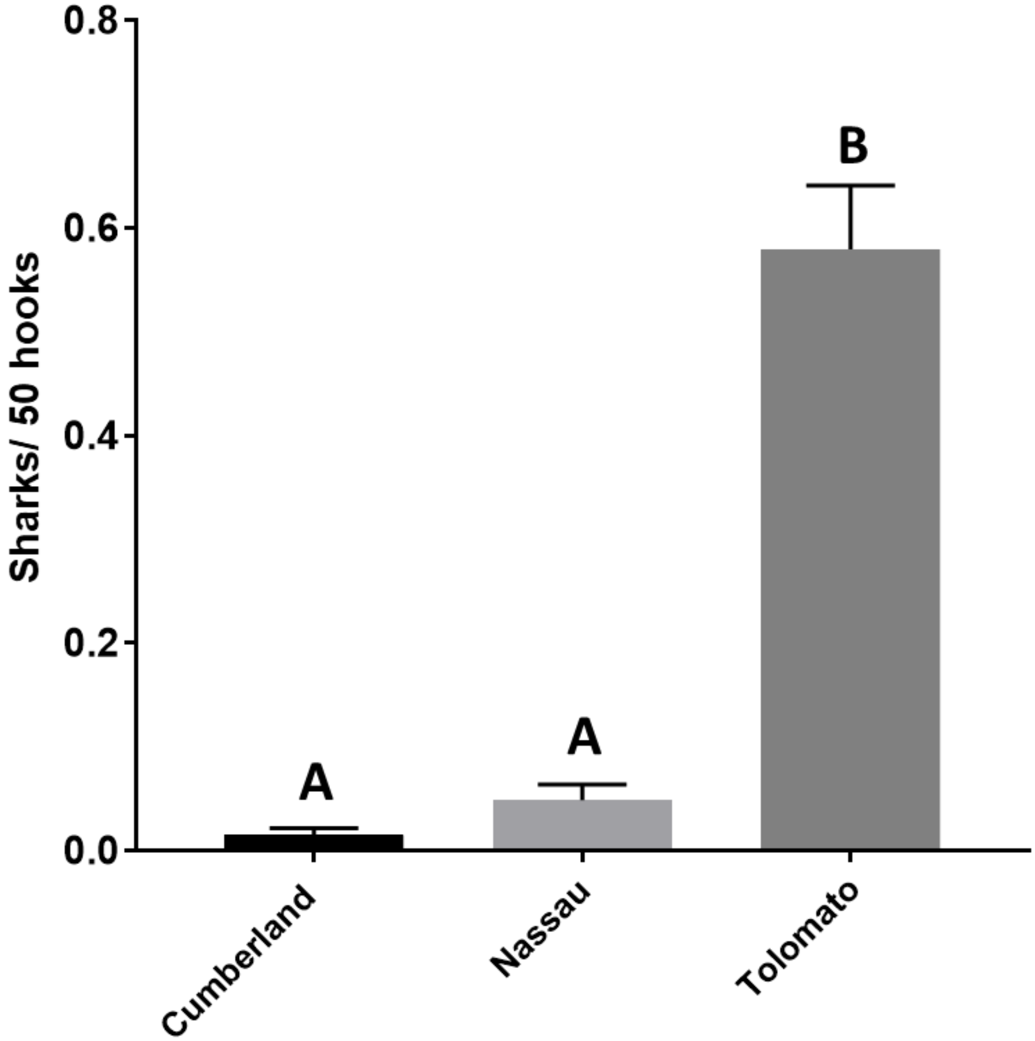

Figure 11. The average CPUE (sharks per 50 hooks) of sets used in GLM analysis, with error bars representing SE. Sets completed in Cumberland Sound (n=354), Nassau Sound (n=327), and the Tolomato River (n=350) are presented. Sites are grouped into homogenous subsets as determined by a post-hoc test with multiple comparisons following Kruskal-Wallis nonparametric analysis (p=0.000).

Table 3. Result of the general linear model used to determine the effects of site and other environmental factors on the abundance (sharks/ 50 hooks) of scalloped hammerhead YOYs in the northeast region of Florida ($F= 54.193$, $P= 0.000$, $R^2= 0.174$).

Variables	F-Value	*P*
Site	74.939	0.000
Salinity*Conductivity	24.518	0.000
Salinity	8.099	0.007

Table 4. A comparison of the average salinity and conductivity conditions recorded for sets used in GLM analysis when at least one scalloped hammerhead YOYs were caught (Present) and for sets that caught no sharks (Absent). Means were compared via Mann-Whitney U tests, and the resulting p values are displayed. Average conditions were also compared across sites via a Kruskal-Wallis analysis, and the resulting p values are displayed. Numbers in parentheses are standard errors and sample sizes.

	Present	Absent	*P*	Cumberland	Nassau	Tolomato	*P*
Salinity (ppt)	31.262 (±0.413, 128)	30.293 (±0.172, 903)	0.118	31.018 (±0.195, 354)	31.102 (±0.276, 327)	29.129 (±0.328, 350)	0.000
Conductivity (mS)	51.980 (±0.774, 128)	48.474 (±0.282, 903)	0.000	49.414 (±0.364, 354)	49.394 (±0.452, 327)	47.931 (±0.552, 350)	0.135

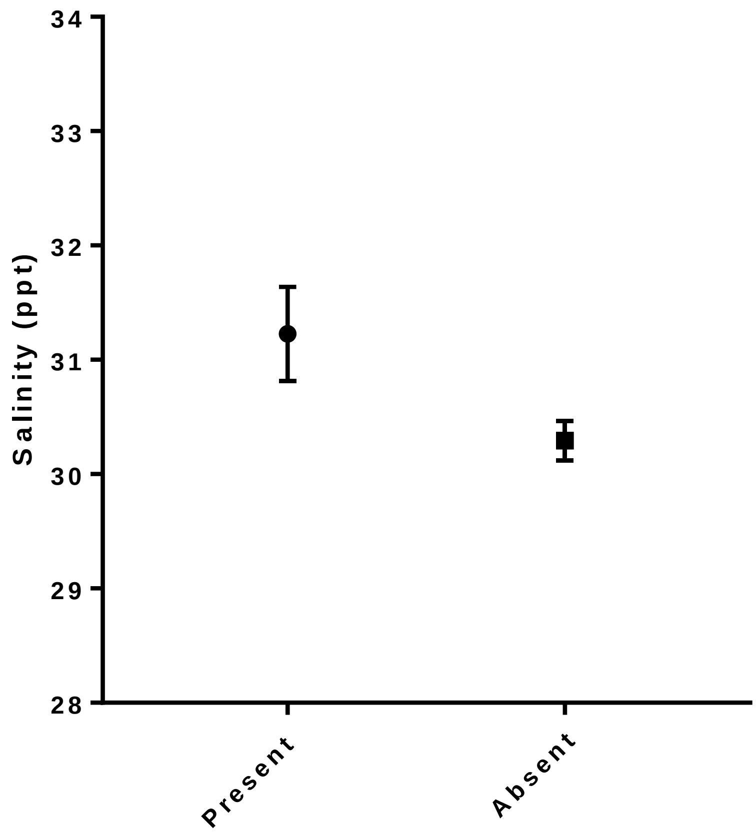

Figure 12. Comparison of the average salinity conditions recorded for sets used in GLM analysis when at least one scalloped hammerhead YOY was caught (Present) and for sets that caught no sharks (Absent) (Mann-Whitney U, $p= 0.118$). Bars represent SE.

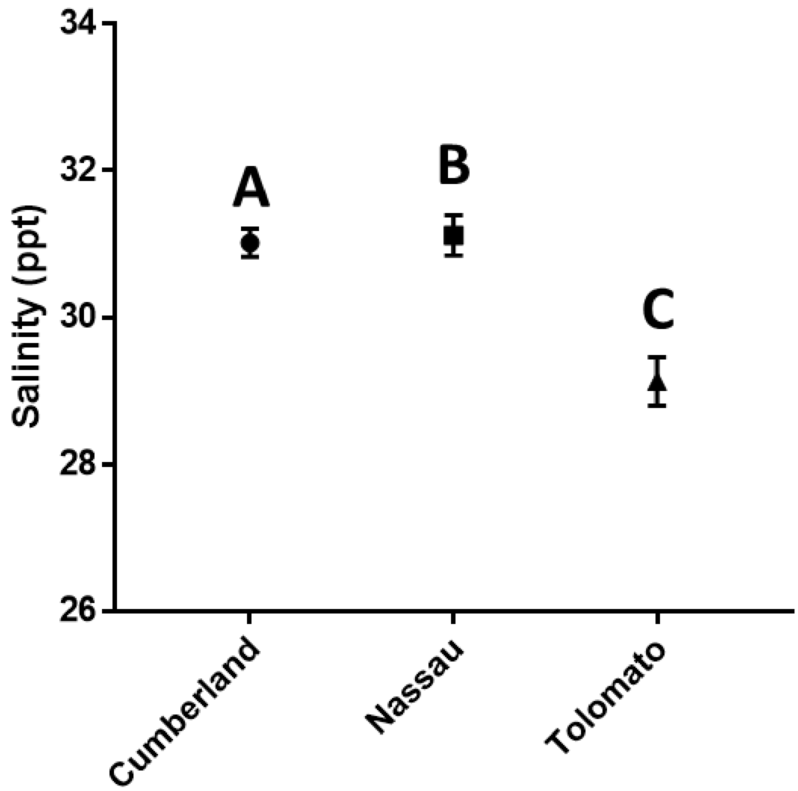

Figure 13. Comparison of the average salinity conditions recorded for sets completed in Cumberland Sound, Nassau Sound, and Tolomato River, which were used in GLM analysis. Sites are grouped into homogenous subsets as determined by a post-hoc test with multiple comparisons following Kruskal-Wallis nonparametric analysis (p=0.000). Bars represent SE.

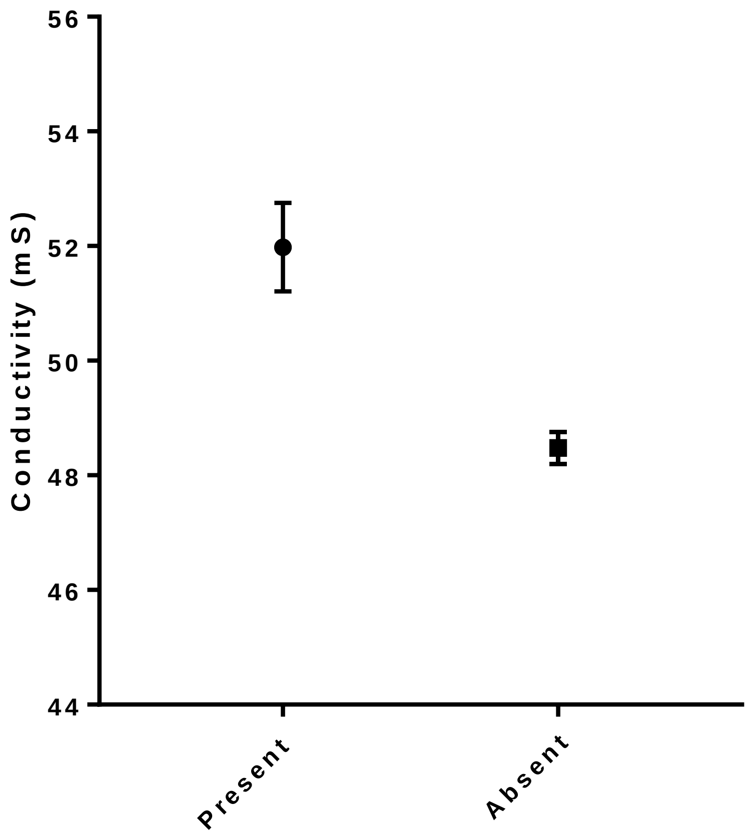

Figure 14. Comparison of the average conductivity conditions recorded for sets used in GLM analysis when at least one scalloped hammerhead YOY was caught (Present) and for sets that caught no sharks (Absent) (Mann-Whitney U, p= 0.000). Bars represent SE.

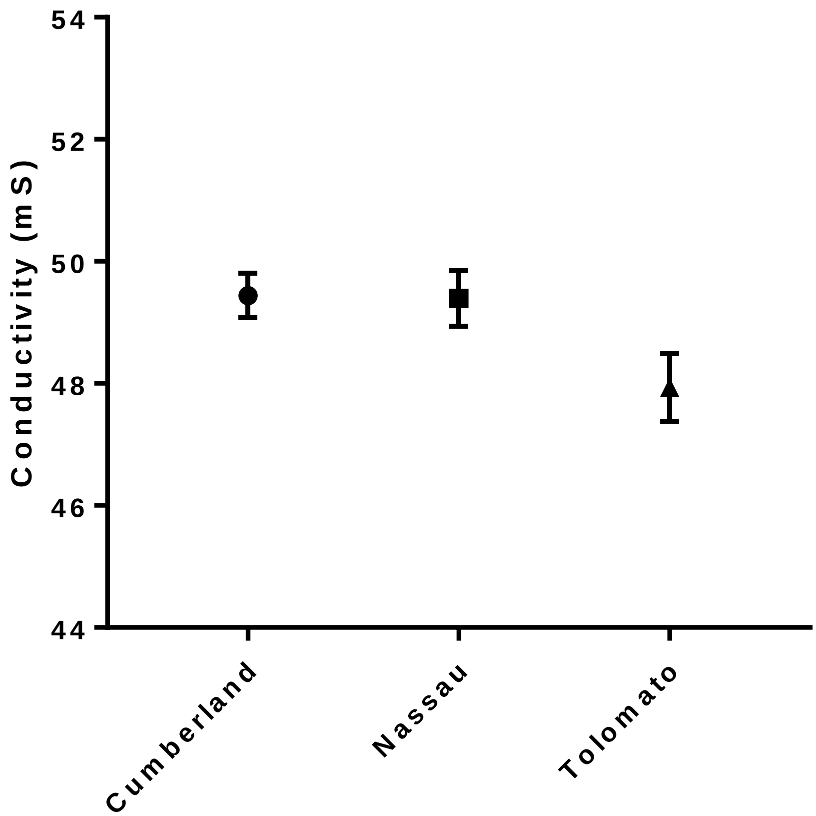

Figure 15. Comparison of the average conductivity conditions recorded for sets completed in the Cumberland Sound, Nassau Sound, and Tolomato River, which were used in GLM analysis (Kruskal-Wallis, p= 0.135). Bars represent SE.

Table 5. Scalloped hammerhead sharks recaptured between the years of 2019-2020 in the Tolomato River. Abbreviations are as follows: M = male and YOY = young of year.

Tag	Sex	Life Stage	Date tagged	Location Tagged	Location Recaptured	Days at liberty	Distance (km)
0028	M	YOY	5/14/19	30.001033, -81.335617	30.026052, -81.361433	6	4.396
0030	M	YOY	5/21/19	30.057783, -81.355200	30.051235,-81.365913	10	1.263
0044	M	YOY	8/12/20	30.050950, -81.365067	30.025833, -81.361389	59	3.183

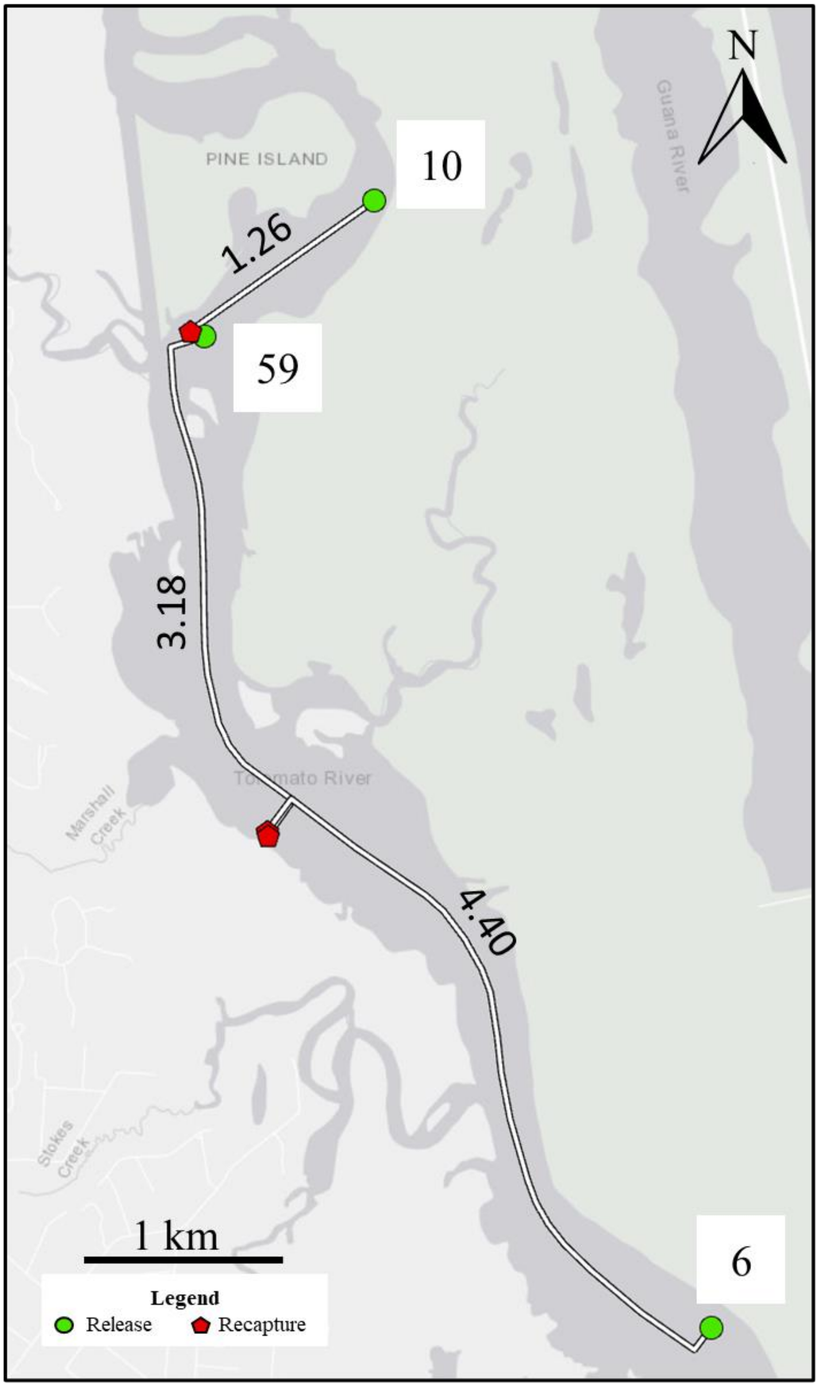

Figure 16. Mark-recapture data collected for three scalloped hammerhead YOYs between 2019-2020. Number in the white boxes near release location indicates days at liberty, and the number along the lines is the distance traveled in kilometers.

A total of 200 positional fixes were recorded for two male YOY scalloped hammerhead sharks that were actively tracked during the summer of 2020. Due to the preliminary nature of this research and the limited length of the tracks, full tracks were used in analysis. Data for the three tracks completed can be found in Table 6. The first shark (SLEW1; 360 g) was caught in the main channel of the Tolomato River and tracked for a total of six hours. During the track, the shark traveled northwards, up the river, and subsequently stayed around Pine Island (Figure 17). In total, the animal traveled 9.409 km during the extent of the track. The second animal (SLEW2; 440 g) was tracked during two different sessions for a total of 10.41 hours. During both tracks, the animal stayed in Pine Island Sound for the entire duration (Figure 17). During the first track, the animal traveled a distance of 6.632 km. The shark was tracked over 3.067 km during the second track. The positional fixes recorded for these tracks were used in activity space analysis.

SLEW1 had the largest activity spaces as determined by minimum convex polygon analysis, with a 1.747 km^2 home range (Table 6; Figure 18). 95% KUD for this animal was 4.159 km^2, while 50% KUD produced an area of 1.611 km^2 (Table 6; Figure 19). 50% KUD indicated that the animals had two core areas of use, with the larger of the two being located near Pine Island. The second largest activity space was recorded for the first track of SLEW2 (SLEW2a), with a total area of 0.401 km^2 (Table 6; Figure 18). This area overlapped slightly with the activity space generated for SLEW1. A 0.693 km^2 area was produced by the 95% KUD and a 0.213 km^2 area from the 50% KUD for this track (Table 6; Figure 20). The third track (SLEW2b) produced the smallest activity space, with 0.046 km^2 (Table 6; Figure 18). This activity space did not overlap with tracks SLEW1 or SLEW2a. The 95% KUD produced a 0.136 km^2 area, while 50% KUD produced an area of 0.039 km^2 (Table 6; Figure 21).

Table 6. Movement and activity data collected for YOY scalloped hammerheads actively tracked in the Tolomato River. Abbreviations are as follows: FL= fork length, MCP= minimum convex polygon, UD= utilization distribution, LI= linearity index.

Shark	FL (cm)	Sex	Track session	Date	Duration (h)	Distance (km)	Total Positional Fixes	Average ROM (m/min)	MCP (km^2)	95% KUD (km^2)	50% KUD (km^2)	LI
SLEW1	36.5	M	a	8/12/2020	6.00	9.409	73	26.137	1.747	4.159	1.611	0.470
SLEW2	35	M	a	8/17/2020	6.58	6.632	80	16.790	0.401	0.693	0.213	0.104
			b	8/25/2020	3.83	3.067	47	13.337	0.046	0.136	0.039	0.064

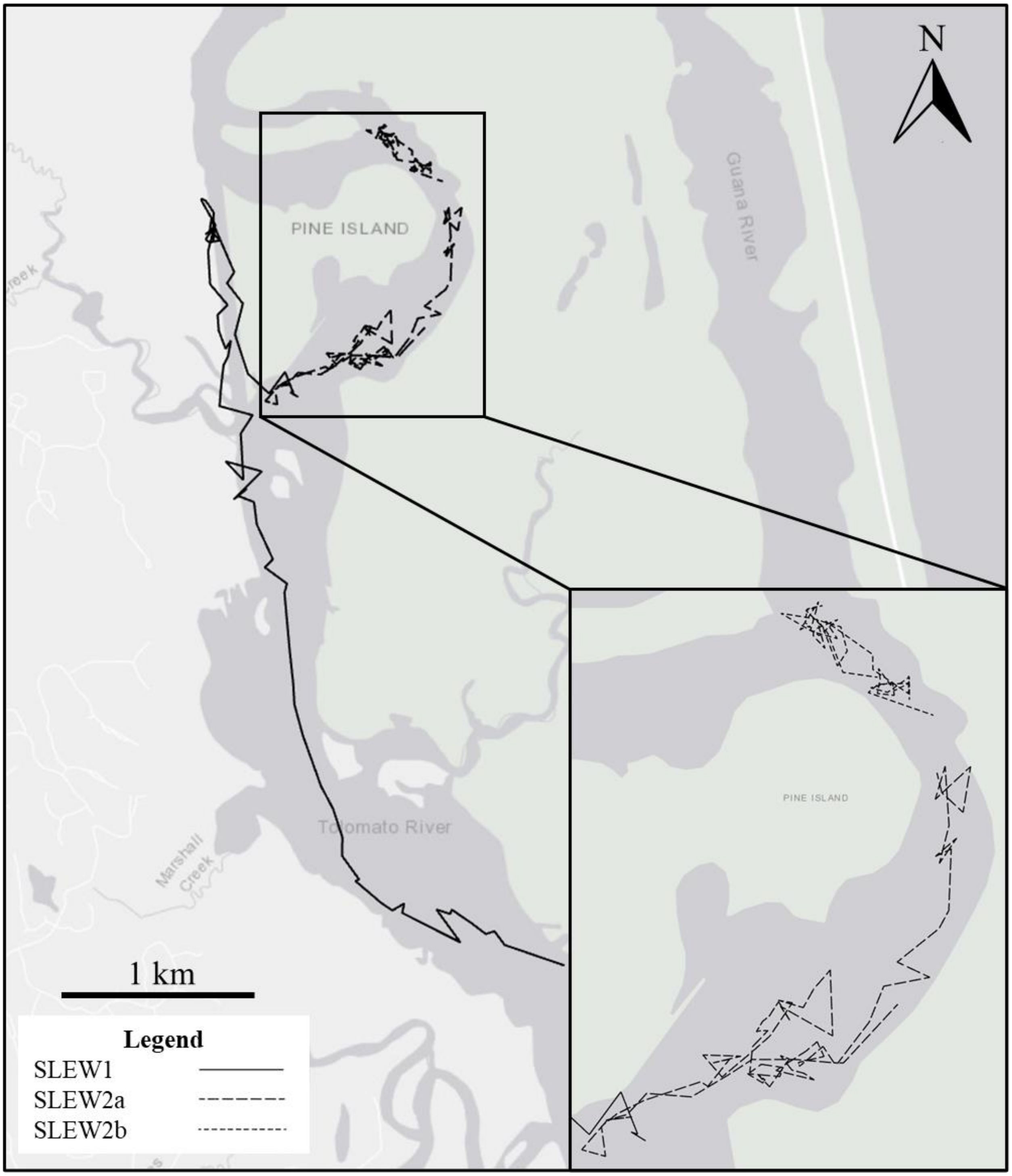

Figure 17. GIS mapping of three active tracks completed with YOY *S. lewini* in the Tolomato River during 2020.

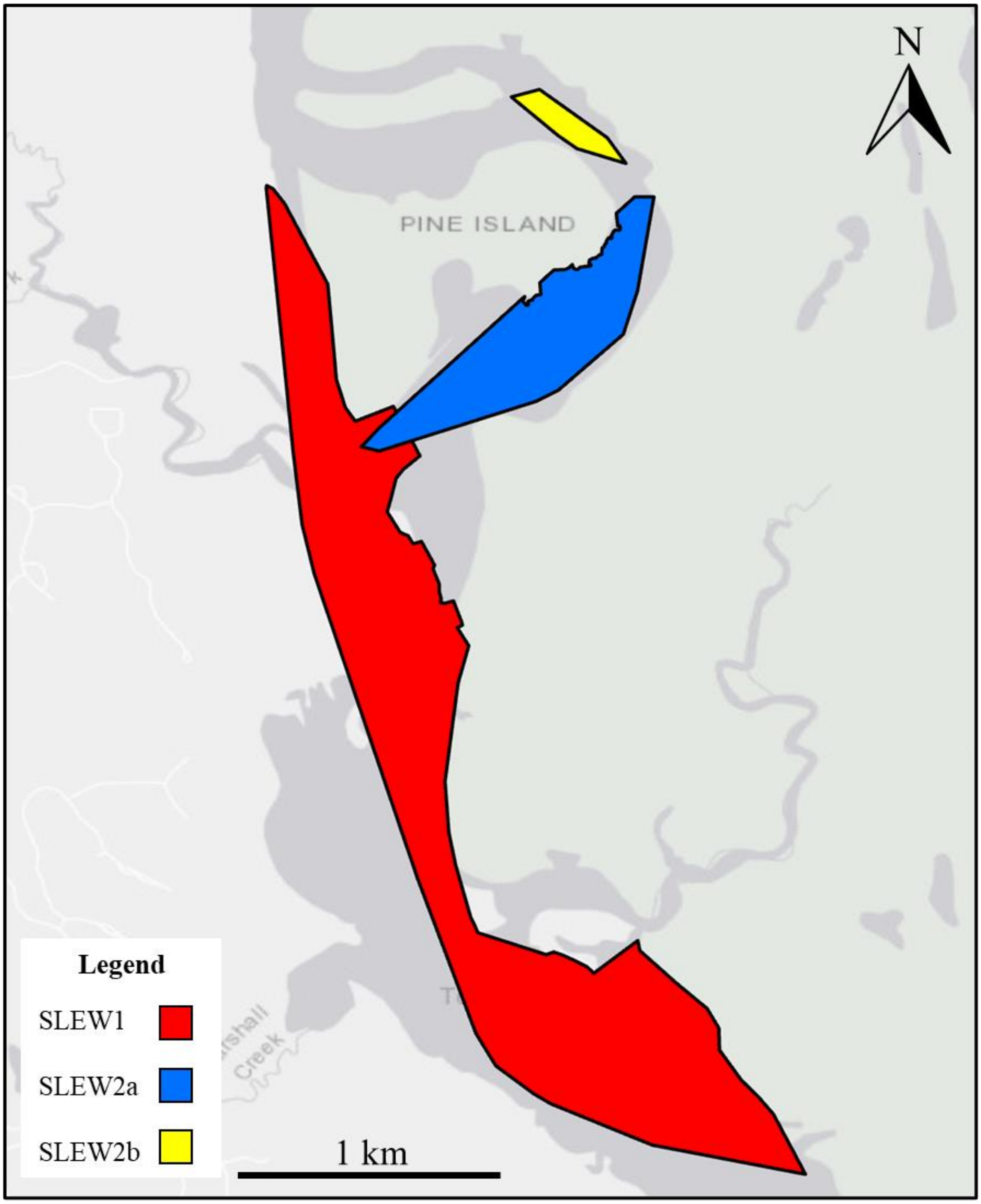

Figure 18. Minimum convex polygons (MCP) created for active tracks SLEW1 (red), SLEW2a (blue), and SLEW2b (yellow). Mapping was completed via ArcGIS Pro.

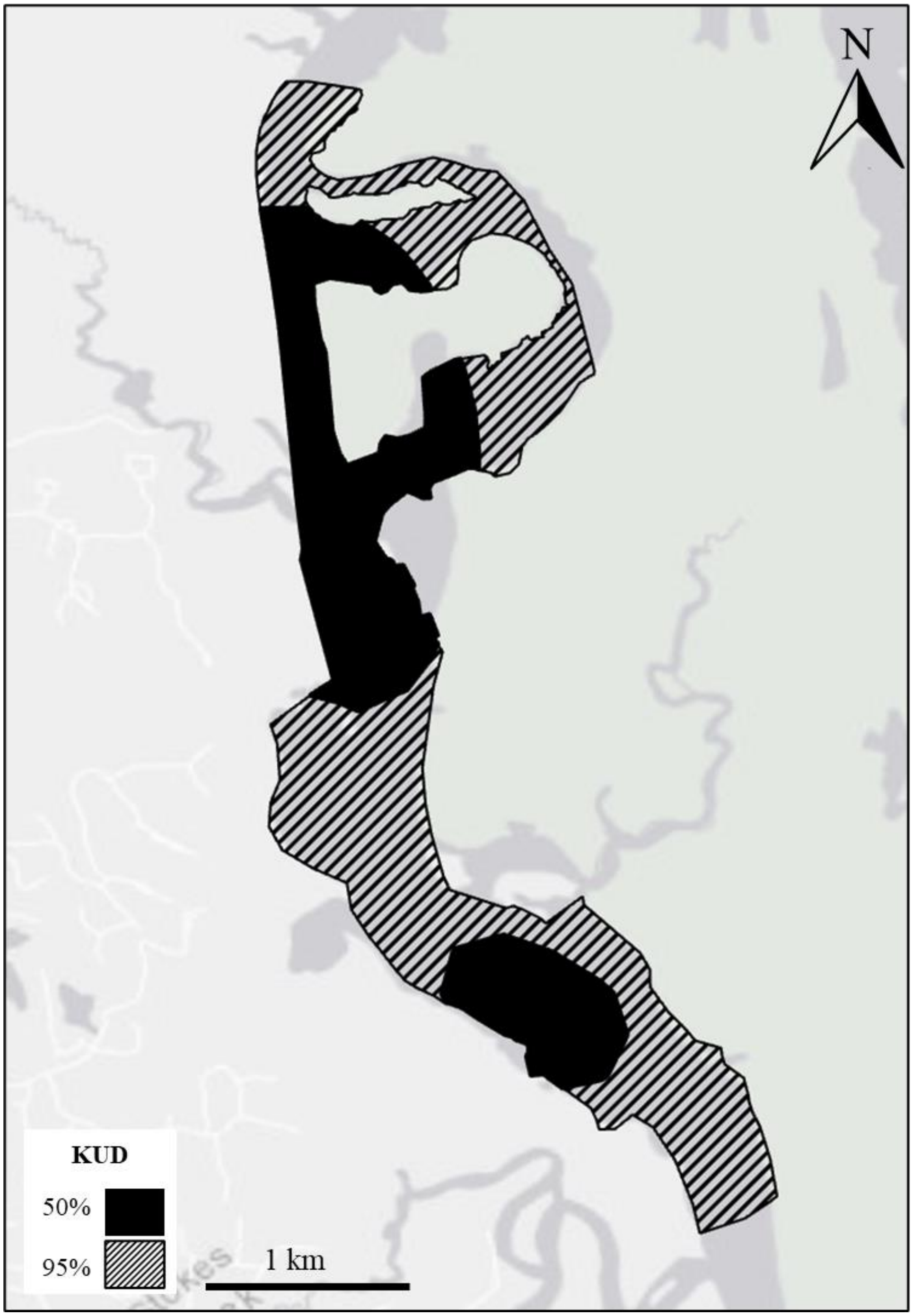

Figure 19. Kernel utilization distribution (KUD) mapped via GIS for track SLEW1. Striped areas represent 95% KUD, and black areas represent 50% KUD. Mapping was completed via ArcGIS Pro.

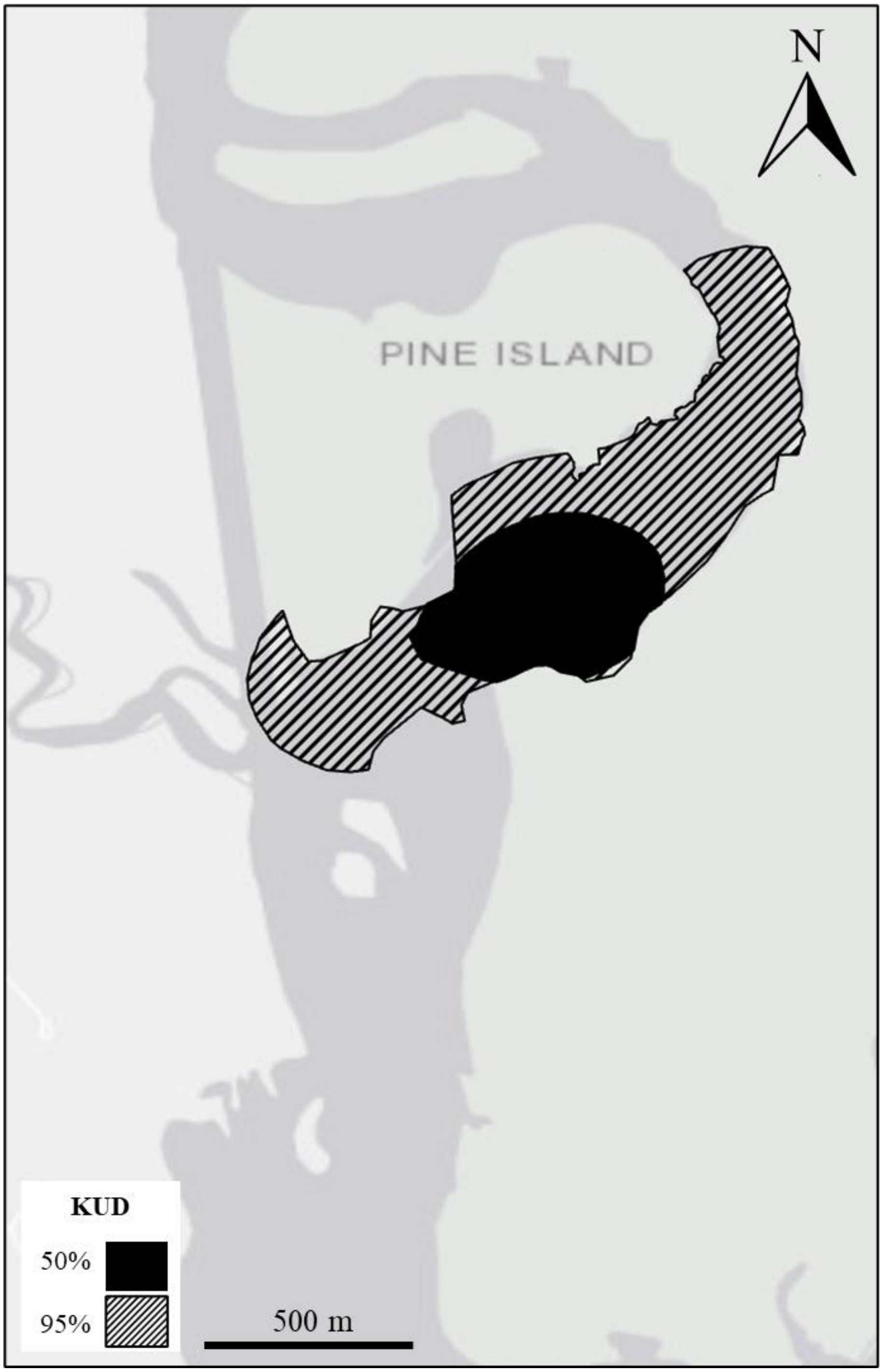

Figure 20. Kernel utilization distribution mapped via GIS for track SLEW2a. Striped areas represent 95% KUD, and black areas represent 50% KUD. Mapping was completed via ArcGIS Pro.

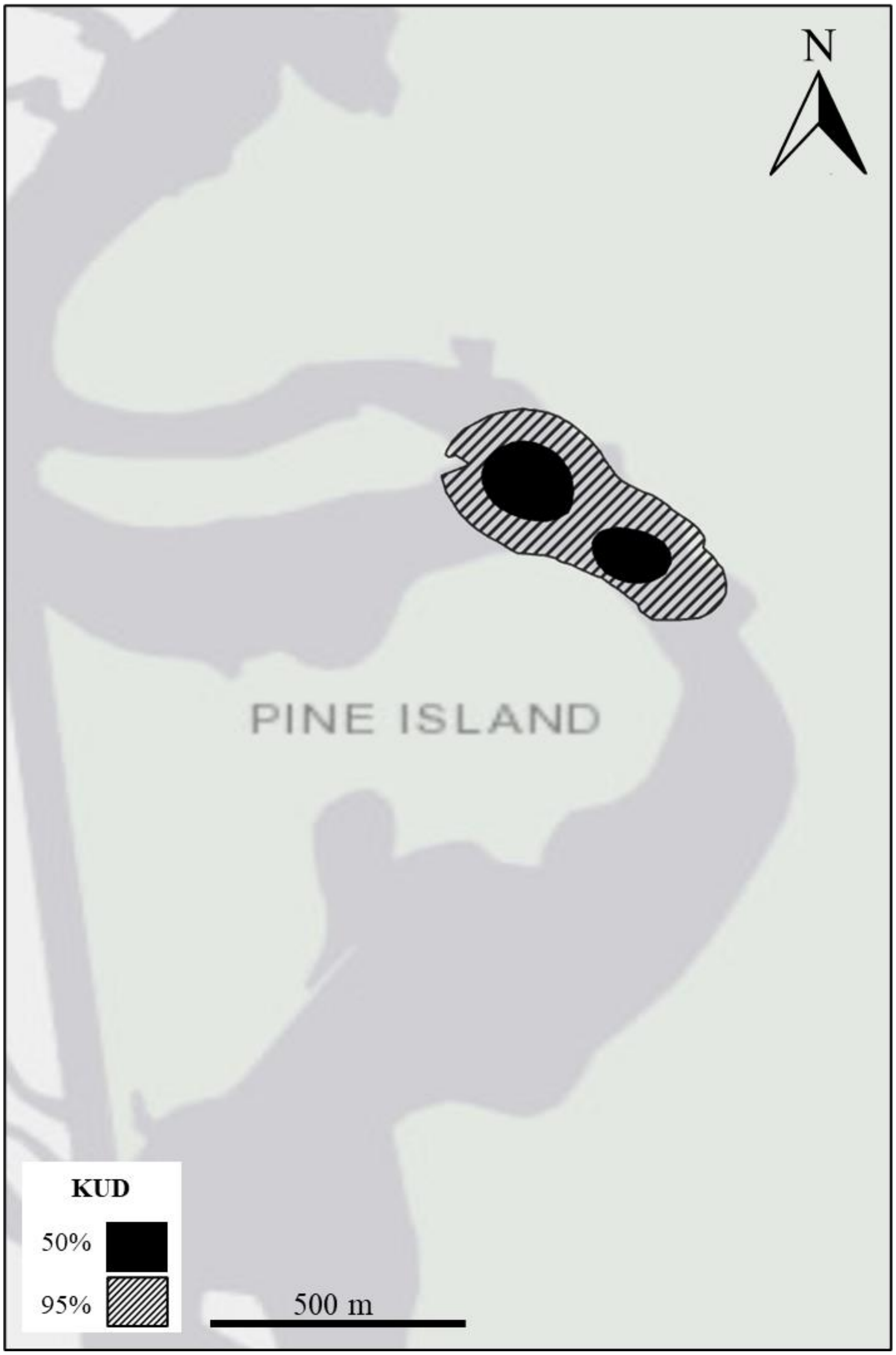

Figure 21. Kernel utilization distribution mapped via GIS for track SLEW2b. Striped areas represent 95% KUD, and black areas represent 50% KUD. Mapping was completed via ArcGIS Pro.

Movement in these areas were described by the linearity index. All linearity indexes were closer to 0 than 1 (Table 6). The SLEW1 track had a linearity index of 0.470, which was the highest of the three tracks. The linearity indexes for tracks SLEW2a and SLEW2b were 0.104 and 0.064, respectively. Movement, in terms of ROM, was subsequently analyzed.
Average ROM was significantly different (ANVOA, p= 0.000) among the tracks, with each track being put into its own homogenous subset (Figure 22). The highest average ROM was recorded for SLEW1 track at 26.137 m/min (Table 6). SLEW2b track had the lowest average ROM at 13.337 m/min (Table 6). Average ROM did not differ significantly between upriver or downriver travel for each of the tracks (Table 7). However, average ROM was significantly different across the four tidal stages when tracks were combined (ANOVA, p= 0.018; Table 8). Average ROM was highest during high tide (25.882 m/min) and lowest during the low tide (17.177 m/min). Bearing additionally was significantly different among the tidal stages despite average bearing indicating a southwards trajectory during each tidal stage (Kruskal-Wallis, p= 0.035; Table 8).

Environmental conditions recorded for the three tracks are displayed in Table 9. Average depth, temperature, salinity, D.O., and conductivity varied significantly among the three tracks (p= 0.000 for each factor; Table 10). The three tracks were put into separate homogenous groups for each environmental parameter with the exception of temperature. For this factor, SLEW2a and SLEW2b were put into the same subset. Preferences for water quality variables were indicated by multiple linear regression with bootstrapping. Temperature had a significant effect on the latitudinal position of SLEW1 during the track (p= 0.049; Table 11). For SLEW2, dissolved oxygen concentration (p= 0.047) influenced the latitudinal position of the animal during the track (Table 12).

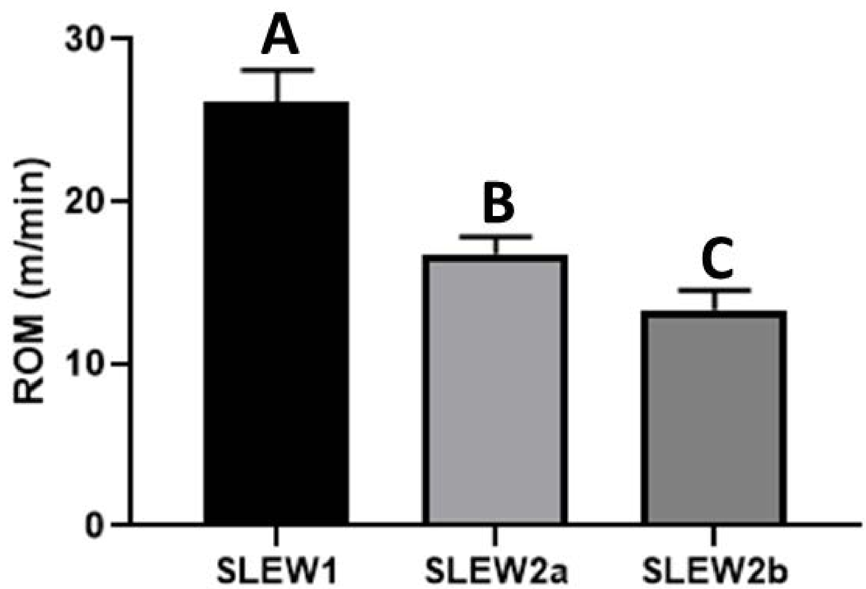

Figure 22. Comparison of average ROM (m/min) across three active tracks, with error bars representing SE (ANOVA, p= 0.000). Tracks are grouped into homogenous subsets according to a Tukey post-hoc test.

Table 7. Rate of movement (m/min) compared between upriver and downriver movement via student t-tests for tracks SLEW1, SLEW2a, and SLEW2b. P values are presented.

Track	Direction of Travel	ROM (m/min)	P
SLEW1	Upriver	27.635	0.327
	Downriver	23.356	
SLEW2a	Upriver	15.448	0.896
	Downriver	17.224	
SLEW2b	Upriver	12.695	0.893
	Downriver	12.772	

Table 8. Comparison of movement parameters recorded during the tidal stages. Rate of movement compared via a one-way ANOVA, and bearing was compared via Kruskal-Wallis analysis. P values from each test are presented.

	Tidal Stage				
	High	Outgoing	Low	Incoming	*P*
ROM (m/min)	25.882	19.348	17.177	17.754	0.018
Bearing (°)	242.156	173.959	154.015	184.653	0.035

Table 9. Mean, minimum, and maximum environmental conditions recorded during complete active tracks of SLEW1 and SLEW2.

		Depth (m)			Temperature (°C)			Salinity (ppt)			DO (mg/L)			Conductivity (mS)		
Shark	Track	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
SLEW1	a	4.86	1.30	7.10	30.38	30.10	30.70	24.90	24.10	25.40	2.94	1.70	4.70	43.34	41.86	43.98
SLEW2	a	3.07	1.20	4.90	30.73	30.40	31.90	17.95	15.60	19.80	3.65	3.00	4.40	32.50	28.46	35.15
	b	4.10	1.10	7.00	30.78	30.40	31.40	20.62	19.80	21.40	4.98	4.50	5.80	36.83	35.95	37.88

Table 10. Comparison of environmental conditions recorded during active tracks. P values indicate results of Kruskal-Wallis test. Homogenous subsets as determined by post-hoc analysis are indicated via superscripts.

Environmental Parameter	SLEW1	SLEW2a	SLEW2b	*P*
Depth (m)	4.86^{A}	3.07^{B}	4.10^{C}	0.000
Temperature (°C)	30.38^{A}	30.73^{B}	30.78^{B}	0.000
Salinity (ppt)	24.90^{A}	17.95^{B}	20.62^{C}	0.000
DO (mg/L)	2.94^{A}	3.65^{B}	4.98^{C}	0.000
Conductivity (mS)	43.34^{A}	32.50^{B}	36.83^{C}	0.000

Table 11. Multiple linear regression model indicating the influence of environmental parameters on the latitudinal location of SLEW1 during active track.

Variables	Coefficient	Standard Error	*P*
Depth (m)	0.000	0.001	0.811
Temperature (°C)	0.051	0.026	0.049
Salinity (ppt)	-0.031	0.022	0.138
DO (mg/L)	0.000	0.002	0.923
Conductivity (mS)	0.006	0.013	0.587
Constant	28.991	0.852	0.000

Table 12. Multiple linear regression model indicating the influence of environmental parameters on the latitudinal location of SLEW2 during active tracks.

Variables	Coefficient	Standard Error	*P*
Depth (m)	0.000	0.000	0.694
Temperature (°C)	-0.001	0.002	0.467
Salinity (ppt)	-0.004	0.004	0.263
DO (mg/L)	0.002	0.001	0.047
Conductivity (mS)	0.004	0.002	0.091
Constant	30.033	0.051	0.000

DISCUSSION

The goal of this study was to determine whether the Tolomato River serves as a nursery habitat for the scalloped hammerhead shark. By following the recommendations of Heupel et al. (2007), various studies were completed to verify whether the habitat exhibits characteristics typically associated with nurseries. Catch data from a ten-year survey were analyzed to assess *S. lewini* presence and annual fidelity to the river. *S. lewini* abundance was then compared across three study sites in northeast Florida to determine preference for the Tolomato River. Site fidelity of individual animals was analyzed via a mark-recapture survey, which was conducted over two years. This was concluded with the active acoustic tracks of two sharks to further understand habitat use. Results from these studies were critical in declaring the Tolomato River as a nursery.

From 2010-2019, 40.1% of the sharks caught in the Tolomato River were identified as scalloped hammerhead sharks. Of these, 98.79% were categorized as YOY's; the life history stage of the remaining *S. lewini* were not recorded. Life history stage was based upon umbilical scar status and morphometric analyses, which revealed several insights about the Tolomato River's *S. lewini* population. Based on observations of 224 individuals, 99.11% of scalloped hammerhead sharks caught had either partly healed (3) or well-healed (4) umbilical scars. Duncan and Holland (2006), utilizing comparable categories, reported that umbilical wounds reached umbilical scar status category 3 by a mean of 4 ± 2.3 days after birth. Scar status did not reach category 4 until a mean of 10 ± 3.6 days after birth. Based on these observations and the lack of mature *S. lewini* in the Tolomato River, it is likely that the YOYs caught during the survey were pupped outside of the study area and moved into the nursery habitat following parturition. The life history stage of the scalloped hammerhead sharks caught during this survey was confirmed via morphometric data.

Regarding morphometrics, relationships between PCL, FL, and STL were all highly correlated, indicating that the record of one length could theoretically be used to calculate others for YOY *S. lewini* in the future. Fork lengths recorded for 236 scalloped hammerhead sharks from the Tolomato River ranged from 29-45 cm. Plotting these fork lengths by month did not reveal any clear trends in growth, though the range in length data was consistent with expected measurements for first year animals (Piercy et al. 2007; Cuevas-Gómez et al. 2020). The range of STL of S. lewini caught in the Tolomato River (38.7-60.5 cm) fell within the lengths recorded for neonates and YOYs (35.5-93 cm STL) in a scalloped hammerhead nursery in the Gulf of Mexico (Cuevas-Gómez et al. 2020). As a result, all *S. lewini* caught at this site, for which life history stage was recorded, were considered YOY individuals. This large presence of YOY scalloped hammerhead sharks suggests that the habitat serves some importance to the species. These results further indicate that only first year sharks use the Tolomato River, which supports the notion that this site serves as a primary nursery according to the definition proposed by Bass (1978). These results were further supported by annual and monthly trends of *S. lewini* abundance.

Analysis of annual average CPUE showed that the catch of this species has been consistent across the years of the bottom longline survey from 2010 to 2019. This indicates that the species is present in this habitat on an annual basis, satisfying the third criterion of a nursery habitat. This is expected of nursery habitats as these environments are thought to be more productive relative to other habitats, thus contributing in greater quantities to the reproductive population (Beck et al. 2001). This is expected to result in natal philopatry, which is indicated by the annual use of the habitat (Beck et al. 2001). Evidence for natal philopatry in sharks has been described by Feldheim et al. (2014), in which mature female lemon sharks *Negaprion* *brevirostris* returned to their birthplace to pup. Additional evidence that the Tolomato River serves as a nursery habitat comes from trends in monthly abundance.

During a given year, the abundance of scalloped hammerhead sharks was highest in the months of May, June, and July. The months of May and June are known as the time of parturition in scalloped hammerhead sharks in the northern hemisphere (Hazin et al. 2000). Thus, if the Tolomato River was to serve as a nursery habitat, an increase in scalloped hammerhead neonates and YOYs would be expected during these months as this coincides with the time when mature individuals start to pup. This is consistent with monthly trends observed in other scalloped hammerhead nurseries. In Cape Canaveral, Florida, the majority of neonates were caught nearshore in late May to late June when assessing a *S. lewini* nursery (Adams & Paperno, 2007). This was additionally observed in a nursery habitat in the Gulf of Mexico, where neonates and YOYs were most commonly found from the months of May through August (Cuevas-Gómez et al. 2020). Likewise, the highest catch rates of this species were recorded during July in Kāne'ohe Bay, Hawaii, a well-documented scalloped hammerhead shark nursery (Duncan & Holland, 2006). Following this peak in abundance, *S. lewini* catch rates then declined in the Tolomato River in August, and catch rates remained low in September. This initial decline in catch rates may be due to high mortality rates. For example, high scalloped hammerhead shark densities in the river could result in competition for resources (Clarke, 1971; Lowe, 2002). Declines in scalloped hammerhead shark abundance were partially attributed to starvation-induced mortality, as supported by weight loss, in Kāne'ohe Bay nursery (Duncan & Holland, 2006). Therefore, it is possible that this may account for the lower catch rates at the end of the summer season. From the months of October to April, the species was absent in the river. Similar trends were found by Heupel (2007) in which juvenile blacktip sharks left summer nursery areas in the Gulf of Mexico as water temperatures declined. Winter declines in population size were also recorded for Kāne'ohe Bay, and hammerheads were expected to only reside in the nursery for 3-4 months before emigrating (Clarke, 1971). As the trends observed in the Tolomato River are comparable to those recorded in known nursery habitats, this analysis further supports that the Tolomato River serves as EFH for the species.

To better understand environmental preferences of this population, environmental data collected throughout the ten-year study were analyzed. The binary linear regression model indicated that the probability of catching a YOY scalloped hammerhead shark increased with lower D.O. conditions. Scalloped hammerhead sharks have been observed in low oxygen environments (Jorgensen et al. 2009). Seeking low D.O. regimes may allow YOYs to inhabit locations inaccessible to potential predators (Schlaff et al. 2014).

Environmental preferences may explain the higher catch rates surrounding Pine Island Sound, in the river. Mapping CPUE geographically highlighted this location as an area of importance, as the majority of scalloped hammerhead catches occurred there. Pine Island Sound is located toward the natural end of the Tolomato River; farther north, the river is replaced by a channel which was completely dredged by 1912 for the ICW (Parkman, 1983). The importance of Pine Island Sound will be further explored below.

Nurseries observed for the scalloped hammerhead shark have been generally described as relatively shallow inshore habitats, such as bays and estuaries, with temperatures upwards of 20 °C (Clark, 1971; NOAA, 2015). Several habitats in northeast Florida meet this description. Therefore, it was important to verify the Tolomato River's importance to *S. lewini*. The catch of *S. lewini* was significantly greater than that recorded in either the Nassau or Cumberland Sounds. Average CPUE (sharks 50-hooks^{-1}) recorded for the Tolomato River was 14.6 times greater than that of Nassau Sound and 38.6 times greater than the Cumberland Sound. Differences were even larger when CPUE accounted for soak time, in which Tolomato sets soak for 15 minutes rather than the standard 30-minutes soak. The general linear model additionally supported these results by indicating that site was a significant factor in determining scalloped hammerhead abundance across all the sets conducted in northeast Florida. The model additionally designated salinity and its interaction with conductivity as variables that influenced *S. lewini* abundance. In order to determine if either salinity or conductivity conditions accounted for the larger numbers of scalloped hammerhead sharks in the Tolomato River, additional analyses were completed.

Further analysis revealed that there was no significant difference in salinity when grouping all sets used in GLM analysis by the presence and absence of *S. lewini*. However, salinity did differ significantly between sites. This suggests that although salinity varied across the three sites, it did not account for the larger numbers of scalloped hammerhead sharks found in the Tolomato River. Furthermore, average salinity conditions were lower in the Tolomato River in comparison to those recorded in the Cumberland and Nassau Sounds. Therefore, it is unlikely that the salinity conditions in the Tolomato River caused the larger numbers recorded as scalloped hammerhead sharks are known to prefer salinities of 28-36 ppt, and *S. lewini* abundance has been shown to increase with salinity (Castro, 1993; Adams & Paperno, 2007; Ward-Paige et al. 2014). In contrast, conductivity did vary significantly between sets that caught *S. lewini* and ones that did not. Though, average conductivity conditions did not differ significantly among the three study sites. This shows that conductivity influenced *S. lewini* abundance regardless of location. Based on these analyses, it is clear that site was the leading cause of differences in *S. lewini* abundance and abiotic factors did not appear to influence the species preference for the Tolomato River. Thus, biotic differences could account for the higher numbers of *S. lewini* in the Tolomato River.

Nurseries have been described as having a smaller population of predators present when compared to nearby habitat (Heupel, 2007). For example, fewer large sharks were recorded in Kāne'ohe Bay nursery relative to the surrounding open-water habitat (Crow et al. 1996). The Tolomato River does have fewer potential predators of scalloped hammerhead YOYs, when compared to the Nassau and Cumberland Sounds. Juvenile and adult sharks account for 7.61% of the sharks caught in the Tolomato River from 2010-2019. In comparison, 52.99% and 68.18% of the sharks caught on bottom longline surveys in the Nassau Sound (2012-2018) and Cumberland Sound (2012-2018) were categorized as either juveniles or adults (Morgan, 2018). Thus, the smaller frequency of large sharks in the Tolomato River may account for the greater abundance of YOY *S. lewini*. Ultimately, the species was more commonly found in the Tolomato River in comparison to other, available habitat in the region. This, thereby, satisfies the first criterion of a nursery habitat.

The mark-recapture study was completed over 2019 and 2020 in order to address both the second and third criteria of nursery habitat identification. From a total of 34 sharks tagged and released in the river, three were recaptured (8.82%). This recapture rate is greater than that recorded for this species in the Kāne'ohe Bay nursery (3.7%), but less than that recorded in the Rewa Delta, Fiji (12.69%), which was also identified as critical habitat for *S. lewini* (Duncan & Holland, 2006; Marie et al. 2017). Furthermore, all sharks were recaptured in the Tolomato River during the same year in which they were released. The time at liberty recorded for these recaptured sharks (6-59 days) fell within the range recorded for scalloped hammerhead sharks in the Kāne'ohe Bay nursery (14 min-324 days; Duncan & Holland, 2006). Additionally, all recaptures occurred at the site of release, which further mirrored the study completed by Duncan and Holland (2006). It is important to note that one of the sharks in this study was released and recaptured, ten days later, in the Pine Island Sound, further indicating the area as important habitat. The other two sharks were recaptured by recreational fishermen off a pier on the river. These results support the second criterion of a nursery habitat; that individual sharks tend to stay in the area for extended periods of time. The lack of second year recaptures further supports the notion that the site is a primary nursery, in which only first-year sharks are found.

Sharks were additionally shown to stay in the Tolomato River for extended periods of time via acoustic tracking. During all three tracks conducted, the animals remained within the Tolomato River, thus further supporting the second criterion of a nursery habitat. However, due to the limited number and duration of the tracks, all additional analyses should be treated as preliminary findings. It is likely that the stress of capture and tagging influenced the animals' behavior post release. Support for these assumptions comes from a comparison of average ROM (m/min) between the first and second tracks completed for SLEW2. These tracks were completed 8 days apart, and the average ROM recorded during the second track was significantly slower than the first. Therefore, major trends among the three tracks rather than individual preferences were thought to more accurately represent *S. lewini* movement in the Tolomato River.

All three tracks had linearity indexes closer to zero than to one, suggesting the majority of the tracks exhibited reuse of space rather than linear movements. The first track (SLEW1) had the highest LI (0.470), which accurately represents the track. For the majority of the active track, the animal traveled upriver through the main channel. The track was stopped at dusk after the animal had moved back downriver and entered Pine Island Sound. In comparison, the second animal (SLEW2) spent the entirety of both tracks in Pine Island Sound. For both tracks, low LI values (0.104 and 0.064) indicated movements associated with reuse. These values were more consistent with LIs recorded for YOYs in other nursery habitats. LI values ranged from 0.01-0.37 for bull sharks in the Indian River Lagoon nursery (Curtis et al 2013).

Total activity spaces for each track reflected these trends; combined area of both SLEW2a and SLEW2b tracks was smaller than the activity space associated with track SLEW1. Average total activity space (0.731 km^2) recorded in this study was slightly smaller than that recorded in other studies. In Kāne'ohe Bay, average total activity spaces of 1.26 ± 1.12 km^2 (Holland et al. 1993) and 1.41 ± 0.41 km^2 (Lowe, 2002) have been recorded for *S. lewini* in the nursery. In a coastal nursery in Jalisco, Mexico, the average home range of YOY scalloped hammerhead sharks was 2.8 ± 1.9 km^2 (Rosende-Pereiro & Corgos, 2018). These differences are likely due to the shorter duration of the active tracks completed during this study. This was additionally seen for the 50% KUD calculated. In this study, the average area determined for the 50% KUD was 0.62 km^2. This is comparatively smaller than 50% KUD recorded for *S. lewini* in nursery studies completed by Lowe (1.31 ± 0.65 km^2 ; 2002) and Rosende-Pereiro and Corgos (1.50 ± 1.29 km^2; 2018). Despite these inconsistencies with previous research, 50% KUDs did indicate core areas of use and preferred habitat within the Tolomato River.

For each of the three tracks, 50% KUDs highlighted areas surrounding Pine Island in the Tolomato River. This supports the notion that the waters surrounding Pine Island serves as the core of the nursery habitat. Noticeably, larger numbers of scalloped hammerhead sharks have been caught in this area over the ten-year bottom longline survey. These active tracks also show that individuals spend long periods of time within this specific area. This is similar to patterns recorded for juvenile lemon sharks and blacktip sharks, which showed small areas of core use in their nursery habitats (Morrissey & Gruber, 1993; Heupel et al. 2004). Sharks of various species have been recorded to select microhabitats within nursery areas based on a variety of both biotic and abiotic factors (Heithaus, 2007). Furthermore, in previous studies, both scalloped hammerhead sharks in Kāne'ohe Bay and sandbar sharks *C. plumbeus* in the Delaware Bay made long distance trips, though they tended to reuse core areas in their nurseries (Rechisky & Wetherbee, 2003; Duncan & Holland, 2006). Similar trends were observed with the track recorded for SLEW1. Thus, Pine Island Sound likely serves as the core of the nursery habitat.

Analyzing the movement and environmental parameters recorded during the three tracks were not as informative about habitat preferences. ROM did not vary between upriver and downriver travel for any of the tracks. Though, ROM did differ significantly between the four tidal stages, with the highest average ROM being recorded during high tide and lowest during low tide. Tidal flow has shown significant effects on shark movements of other species. Juvenile bull sharks in a Florida estuary were suspected to use tides to conserve energy; it was also hypothesized that movements could be an indirect result of prey movement (Ortega et al. 2009). Bearing of travel was significantly different among the four tidal stages; however, for each stage, the average bearing indicated a southward trajectory thereby limiting the assumptions that can be made for tidal influence on direction of movement. Limitations were also found when analyzing the effects of environmental parameters on shark location.

Abiotic factors can affect digestion and osmoregulation in elasmobranchs, and movement within a habitat may be the result of physiological requirements (Schlaff et al. 2014). Therefore, the potential effects of environmental conditions on the spatial ecology of the tracked animals were assessed. All environmental parameters did vary significantly between the three tracks, and each track was put into its own homogenous subset for each factor, with a single exception of temperature. For temperature, tracks SLEW2a and SLEW2b were put into the same homogenous subset. This likely indicates the differences recorded in environmental parameters among the three tracks were due to the conditions present at the day and time of the track, rather than representing preferences of the species. For the SLEW1 track, which showed the greatest latitudinal change, temperature was shown to significantly influence the latitudinal position of the animal. However, there was only a range of 0.6 °C recorded during the track. Therefore, it is unlikely that temperature alone was the direct cause of latitudinal changes. Movement of this animal could be the response to other factors not covered in this study, such as biological ones. Though, it is important to note that the temperatures recorded during the track (30.1-30.7 °C) were slightly higher than the preferred temperatures recorded for the species (26-30 °C; Castro, 1993).

Data collected from tracks SLEW2a and SLEW2b were combined for the multiple linear regression based on analysis of the R-squared values generated, which was greater when tracks were combined, and to potentially infer habitat preferences of the individual shark. From the analysis, D.O. was identified as a factor that influenced the animal's position. During the two tracks, there was a difference of 0.010 (SLEW2a) and 0.003 (SLEW2b) decimal degrees in latitude between the northmost and southmost locations recorded during the tracks. Thus, the animal did not cover large latitudinal distances during either track as it remained within the Pine Island Sound on both occasions. Furthermore, the range of D.O. conditions recorded did not overlap between the two tracks and varied significantly. This likely indicates that the animal was not seeking certain D.O. regimes. Ultimately, a greater sample size and longer active tracks would be needed to better address the effect of environmental parameters on the localized movements of scalloped hammerhead sharks in the Tolomato River.

Results from these analyses indicate that the Tolomato River is a primary nursery habitat for the scalloped hammerhead shark, and Pine Island Sound serves as the core of the nursery.

Such findings conflict with previous work from Adams and Paperno (2007), which indicated that nearshore habitats such as beaches are more important in providing nursery habitats for scalloped hammerheads on the Atlantic coast of Florida. Though both beach and estuarine habitats were sampled during the study, their results indicated that the scalloped hammerheads were not using estuaries as nursery grounds (Adams and Paperno, 2007). The identification of the Tolomato River as a nursery habitat indicates that the species is not limited to beaches on the Atlantic coast of Florida. This information ultimately aids in the management and conservation of the species in the Northwest Atlantic and Gulf of Mexico.

Before effective management strategies can be put in place, information must be collected regarding habitat use. Boundaries of nurseries must be identified to help inform designations of EFH. Gaining this knowledge is critical to developing practical policies to protect these habitats. This is specifically important for the scalloped hammerhead shark. Adults of this species are very mobile and known to make extensive migrations (Maguire et al. 2006), therefore it is more prudent to protect nursery habitats were YOYs are predictably found. Thus, protection of their nurseries could potentially increase recruit survival, leading to larger population sizes. The results from this study not only indicate that the Tolomato River serves as a nursery habitat, but it also expands upon the knowledge of what type of habitats are used as nurseries by the scalloped hammerhead shark on Florida's eastern coast. Overall, the identification of this habitat as a nursery, and thereby EFH, aids in the conservation of a species in need of protection throughout its range.

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VITA

Bryanna Wargat

EDUCATION:

B.S. in Marine Biology, *Summa Cum Laude* with Departmental Honors

Florida Southern College

Minor: Environmental Studies

Departmental Honors Thesis: *Characterizing the histological composition of albinism in sharks*

TEACHING EXPERIENCE:

University of North Florida Graduate Teaching Assistant

Florida Southern College Teaching Assistant

ACADEMIC AWARDS

Henry and Annie Merner Pfeiffer Scholarship Recipient

Helen and Charles McConville Scholarship Recipient

UNF Coastal and Marine Biology Flagship Program Graduate Student Summer Research Award

University of North Florida Graduate School Grant

RESEARCH EXPERIENCE:

Graduate

University of North Florida Research Assistant, *UNF Shark Biology Lab*

University of North Florida Graduate Student

Advisor: Dr. Gelsleichter

Undergraduate

Florida Southern College Undergraduate Student

Advisor: Dr. Laura Habegger

REU Student, *University of North Florida*

Supervisor: Dr. Gelsleichter

MAJOR PRESENTATIONS:

2018 Oral Presentation:

Florida Southern College Annual Fiat Lux

2018 Poster Presentation:

ASLO Aquatic Science Meeting

2019 Poster Presentations:

Annual Meeting of the American Fisheries Society Southern Division

GTM NERR Conference

Annual Meeting of the American Elasmobranch Society at JMIH

2021 Oral Presentation:

2021 Spring SEERS Meeting

SCIENTIFIC MEMBERSHIP:

American Elasmobranch Society