THE EFFECTS OF HARD-BOTTOM HABITAT DEGRADATION ON THE ECOLOGY AND BIOLOGY OF THE FLORIDA STONE CRAB *MENIPPE MERCENARIA* IN THE FLORIDA KEYS

By

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To my parents
ACKNOWLEDGMENTS

I would like to thank my parents, without whom I would not be the person I am today. Growing up in the outdoors brought about a passion for nature and wildlife, which has lead me to pursue marine science as a career. I would like to thank my wife for supporting me and helping me with my studies over the past two years. I would also like to thank my advisor, Dr. Donald Behringer, for supporting, guiding and helping me throughout my graduate studies at the University of Florida. I also thank my committee members, Dr. William Lindberg and Dr. William Patterson, for their guidance and knowledge on the subject matter as well as helping me further my studies with new ideas. The endless encouragement and help from these individuals throughout these past few years has made all the difference.
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THE EFFECTS OF HARD-BOTTOM HABITAT DEGRADATION
ON THE ECOLOGY AND BIOLOGY OF THE FLORIDA STONE CRAB
MENIPPE MERCENARIA IN THE FLORIDA KEYS

By
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The stone crab, *Menippe mercenaria*, supports one of the most economically important fisheries in the southeastern United States. Hard-bottom habitat in the Florida Keys is an important habitat for *M. mercenaria* and is characterized by large sponges, octocorals, and macroalgae on a porous limestone bottom. Cyanobacteria blooms, consisting primarily of *Synechococcus* sp., have periodically occurred in Florida Bay for at least the past several decades, resulting in mass sponge mortalities, most notably the loss of loggerhead sponges. Juvenile and young adult *M. mercenaria* are predominantly found residing in loggerhead sponges or solution holes. I examined the potential impacts of sponge loss and overall habitat degradation by comparing stone crab nutritional condition, trophic position, and site fidelity between degraded and non-degraded regions of the Bay. I also investigated whether *M. mercenaria* potentially use chemical cues from sponges to navigate in hard-bottom habitat.

*Menippe mercenaria* from degraded and non-degraded habitats were of similar nutritional condition regardless of index used. Using stable isotope analysis, I found *M. mercenaria* from degraded versus non-degraded habitats to be feeding at the same trophic level, but the carbon sources supporting crabs in these regions differed significantly. This difference could be
attributed to a shift in prey choice or a shift in the isotopic signatures of primary producers within the system. *Menippe mercenaria* could also be foraging more opportunistically within degraded regions of Florida Bay. Stone crabs were attracted to chemical cues from loggerhead sponges over seawater alone, indicating that they may indeed use cues exuded by sponges to navigate hard-bottom and perhaps reduce the time to locate shelter.

The impact of habitat degradation caused by cyanobacterial blooms on this important benthic species appears to be minimal. *Menippe mercenaria* maintained a similar nutritional condition despite a shift in trophic niche, indicating acclimation to their altered habitat through a shift in diet. Site and den fidelity remained similar regardless of the loss of loggerhead sponges; probably due to the abundance of solution holes available as shelters throughout hard-bottom. Overall, results indicate a high resilience to this particular natural disturbance by *M. mercenaria.*
Anthropogenic impacts to ecosystems are increasing globally as the human population of our planet continues to rise. As we continue to burn fossil fuels, CO$_2$ levels have risen exponentially leading to an increase in atmospheric and ocean temperatures. These changes, among others, are expected to magnify the frequency and intensity of algal blooms into the future (Aquino-Cruz et al. 2018, Paerl and Huisman 2008, Moore et al. 2008). Harmful algal blooms, otherwise known as “HABs”, are of particular concern, not only affecting human health (Heisler et al. 2008), but also damaging important ecosystems (Tiling and Proffitt 2017) and the services they provide. The phytoplankton that make up HABs, such as cyanobacteria, dinoflagellates, and diatoms (Glibert et al. 2005), are naturally occurring in freshwater and marine environments, but manifest into blooms when stimulated and fueled by a nutrient supply, which occurs naturally (Anderson et al. 2002) or by anthropogenic means (Fogg 1969, Paerl 2011, Reynolds 1987). Excess nutrient loading, or eutrophication, is the enrichment of a body of water and results from agricultural, wastewater, and industrial runoff (Anderson et al. 2002, Vitousek et al. 1997). The two major nutrients that typically fuel algal blooms, nitrogen and phosphorus, are normally the limiting macronutrients within freshwater or marine systems (Howarth 2008, Paerl et al. 2011). Harmful algal blooms can be grouped into two categories, toxic and non-toxic. Toxic algal blooms can cause serious threats to humans and wildlife through inhalation or ingestion of contaminated water or shellfish (Carmichael 2001, Ibelings et al. 2014). At high volumes, non-toxic blooms can also be detrimental to aquatic systems, depleting oxygen levels and blocking sunlight to the benthos which can devastate benthic habitat, causing further harm to aquatic organisms (Anderson et al. 2002).
The adverse effects and implications of HAB’s in aquatic environments have become increasingly recognized as the frequency and intensity of blooms have increased. Many economically and ecologically important species, as well as the communities they help support, have been affected by these blooms. For example, in 1998, the dinoflagellate, *Ostreopsis* cf. *ovata*, caused a major die off of the sea urchin, *Echinometra lucunter*, in Rio de Janeiro (Ferreira 2006). This dinoflagellate also was shown to affect fertilization and early development of another sea urchin, *Lytechinus variegatus*, along the coast of Brazil (Neves et al. 2018). Another toxic dinoflagellate, *Alexandrium excavatum*, was shown to induce heavy mortality in finfish larvae, including the commercially important American mackerel in the Gulf of St. Lawrence, Canada (Robineau et al. 1991). Harmful algal blooms can also affect benthic habitat directly, causing indirect effects on species that depend on it. In the mid 1980’s, a brown tide of *Aureococcus anophagefferens* occurred in several bays along the mid-Atlantic. This led to high mortalities of eelgrass, *Zostera marina*, increasing light absorption within the water column, which in turn caused recruitment failure of the commercially valuable bay scallop, *Argopecten irradians* (Bricelj et al. 1987, Cosper et al. 1987, Dennison et al. 1989). In Narragansett Bay, Rhode Island, the same bloom forming species led to reproductive failure and high mortalities in the blue mussel, *Mytilus edulis* (Tracey 1988). Impacts such as these can be seen around the globe affecting both habitat and organisms, which can further harm major fisheries and tourism. For example, the high frequency of HABs experienced in Florida has continued to have major implications on the environment and local economy (Larkin and Adams 2007).

With nearly 21 million people living in Florida, anthropogenic factors can play a large role in the high frequency of HAB’s throughout the state. High agricultural, industrial, and sewage runoff into Florida’s many water bodies facilitated by heavy rainfall contribute high
amounts of nutrients into systems leading to an increase in the frequency of algal blooms (Hu et al. 2004, Lapointe and Bedford 2010, Phlips and Badylak 1999). The dinoflagellate, *Karenia brevis*, also known as red tide, occurs on the Gulf coast of Florida almost yearly. This species is associated with neurotoxic shellfish poisoning (NSP) which can be obtained through ingestion of brevetoxins in contaminated shellfish (Backer 2009, Steidinger 1993). *Karenia brevis* also produces aerosolized brevetoxins that can be inhaled by humans and wildlife causing respiratory issues (Kirkpatrick et al. 2004). High mortalities have been reported in fish, manatees and double-crested cormorants along the coasts of the Gulf of Mexico due to these highly toxic blooms (Bossart et al. 1998, Kreuder et al. 1998, Trainer and Baden 1999). *Karenia brevis* has also been shown to lower food consumption and decrease survivorship among Florida stone crabs, *Menippe mercenaria*, on the west coast of Florida (Gravinese et al. 2018).

One group of HAB-causing phytoplankton, the cyanobacteria, are widely responsible for effecting marine and estuarine communities (Paerl and Huisman 2005, Paerl et al. 2011). In 2006, the cyanobacterium, *Lyngbya majuscule*, bloomed in the Indian River Lagoon. Shading from the bloom decreased the density of *Halodule wrightii*, a common seagrass, as well as the density of a common bivalve, *Macoma constricta* (Tiling and Proffitt 2017). Cyanobacteria have been found to produce the neurotoxin beta-N-methylamino-L-alanine (BMAA) which can biomagnify in aquatic food webs and cause neurodegenerative diseases such as Parkinson’s and Alzheimer’s (Brand et al. 2010). Harmful BMAA concentrations were found in species consumed by humans, such as pink shrimp and blue crabs from Biscayne Bay, Florida. Another cyanobacterium, *Lyngbya* sp., is a benthic form that often blooms on the coral reef tract in south Florida and can negatively impact corals and other invertebrates (Paul et al. 2005). Non-toxic
cyanobacteria such as *Synechococcus* sp. can also have deleterious effects on habitat, such as has occurred in Florida Bay in recent decades.

Florida Bay, which lies between the mainland of Florida and the Florida Keys (Fig. 1-1), is well known for its productivity, biodiversity, and ecological importance as a nursery habitat for many species of invertebrates and fish (Homquist et al. 1989, Thayer and Chester 1989, Zieman et al. 1989). Seagrass beds, primarily made up of *Thalassia testudinum*, cover approximately 60-70% of the Bay (Zieman et al. 1989) providing habitat and foraging grounds for many species. Another 30-40% of the Bay consists of hard-bottom habitat (Behringer and Butler 2006, Bertelson et al. 2009), an integral marine feature of the Florida Keys. Hard-bottom habitat is characterized by sponges, solitary hard corals, octocorals, and patches of macroalgae on a porous limestone bottom covered in a thin layer of sediment (Butler et al. 1995, Behringer and Butler 2006). A diversity of high-density sponges is synonymous with this bottom, which helps create its complex three dimensional structure. Loggerhead sponges, *Speciospongia vesparium*, are the largest and most abundant of the sponge species (Peterson et al. 2006) and are used as shelter by many benthic invertebrates, such as the Caribbean spiny lobster, *Panulirus argus* (Butler et al. 1995) and Florida stone crab, *Menippe mercenaria* (Behringer and Hart 2017). Many other smaller benthic organisms, such as snapping shrimp, live within the internal channels of this sponge as well (Pearse 1950).

Over the past several decades, Florida Bay has been affected by a series of cyanobacteria blooms, dominated mainly by the picoplankton sized cyanobacterium, *Synechococcus* sp., which has drastically changed the ecology of the Bay north of the middle Keys (Butler et al. 1995, Phlips and Badylak 1996, Wall 2012). Over the past century, the natural flow of freshwater from the Everglades into Florida Bay has been altered, leading to changes in the community structure.
of both ecosystems. Nutrient loading from Taylor Slough combined with sediment resuspension in the north central region of the Bay are responsible for elevated levels of cyanobacteria blooms throughout the year (Phlips and Badylak 1996) and wind events often push these blooms southwards towards the Florida Keys. The network of shallow mud banks throughout the north central region of the Bay also contribute to the longevity of blooms with very low water exchange with the open sea (Phlips et al. 1999). These blooms have coincided with massive sponge mortalities in the northeast region of the Bay that have killed over 40% of loggerhead sponges and over 70% of other sponge species (Butler et al. 1995, Butler et al. 2015). The loss of loggerhead sponges in impacted regions has led to a dramatic decrease in available shelters and overall habitat for many benthic species. In particular, these blooms have had negative effects on the distribution and abundance of the Caribbean spiny lobster (Butler et al. 1995). Another ecologically and economically important species that may be impacted by these blooms is the Florida stone crab, *M. mercenaria*, but data are currently lacking to discern the effects of sponge loss and habitat degradation on stone crabs in this system.

The Florida stone crab, *M. mercenaria*, and the Gulf stone crab, *Menippe adina*, occur throughout the southeastern United States and Caribbean. *Menippe mercenaria* ranges from Florida to North Carolina, also occurring throughout the Caribbean, Yucatan Peninsula and Belize (Ong and Costlow 1970, Say 1818). *Menippe adina* occurs westward from northwestern Florida along the Gulf of Mexico coast into Mexico (Williams and Felder 1986). The stone crab is the largest xanthid crab within its range (Lindberg and Marshall 1984) and is characterized by a pair of large chelipeds which account for over 50% of the overall weight of the crab (Fig. 1-2). Stone crabs are almost entirely carnivorous and use their large crusher claw to break open and feed on bivalves and gastropods (Menzel and Nichy 1958, Powell and Gunter 1968, Lindberg
and Marshall 1984, Brown and Haight 1992). Unlike crustaceans such as the Caribbean spiny lobster, which aggregates in shelters with conspecifics (Childress and Herrnkind 1997), stone crabs are solitary and share shelters only during mating periods (Wilber 1989). *Menippe mercenaria* also dominate shelter access and outcompete other crustaceans such as *P. argus* (Behringer and Hart 2017). Juvenile and young adult *M. mercenaria* inhabit the shallow hard-bottom habitat of Florida Bay taking advantage of the multitude of solution holes and large excavated loggerhead sponges as shelter. Larger individuals will emigrate to deeper sea grass beds where they can excavate their own burrows as they outgrow the confines of hard-bottom shelters.

Stone crabs support one of the most valuable commercial fisheries in the southeastern United States and the Caribbean. The Florida fishery is the largest stone crab fishery in the region, supporting an estimated $25 M annual commercial fishery. Monroe and Collier counties lead the state in landings, producing more than half of the overall harvest (FWC 2017). Both species of *Menippe* are managed as one fishery. The stone crab fishery is unique in that only the claws are taken and crabs are released to regenerate new claws. This practice would seem to make this fishery more sustainable than others, but studies have shown that the removal of claws can cause high mortality, alter diet, and alter prey size selection (Duermitt et al. 2015). The fishery is managed differently in each state with some states allowing removal of both claws, while others allow removal of only one. Florida allows the harvesting of both claws if they are both of legal size. On average, male crabs will enter the fishery during age three and females during age four (Gerhart and Bert 2008). If cyanobacteria blooms are affecting population size or nutritional condition of *M. mercenaria* in Florida Bay, the largest regional contributor to this valuable fishery could suffer.
I hypothesized that habitat degradation caused by persistent cyanobacteria blooms could affect the nutritional condition of *M. mercenaria* by limiting prey availability or prey choice in impacted regions of the Bay. A change in prey could also alter stone crab trophic position causing further alterations throughout the food web. Shifts in prey availability or choice could also affect site or shelter fidelity, ultimately causing crabs to emigrate from degraded regions to more suitable habitat. Emigration or decreased nutritional condition could both have deleterious effects on the stone crab fishery. Cyanobacteria blooms could also indirectly affect *M. mercenaria* by killing sponges. If stone crabs use chemical cues exuded by hard-bottom organisms, such as sponges, as a way of finding suitable habitat, navigating, or foraging, then the degradation of hard-bottom could also have an ecological impact on behavior. I used three indices to determine nutritional condition and employed stable isotope analysis of muscle tissue samples to determine the trophic niche of *M. mercenaria* in degraded versus non-degraded regions. A mark-recapture study was used to compare site and shelter fidelity between degraded and non-degraded regions.
Figure 1-1. Map of Florida Bay (USA) lying between the mainland of Florida and the Florida Keys. Photo courtesy of U.S. Geological Survey.
Figure 1-2. A large male *Menippe mercenaria* captured in Florida Bay in 2017. Photo courtesy of author.
CHAPTER 2
THE EFFECTS OF HARD-BOTTOM HABITAT DEGRADATION ON NUTRITIONAL CONDITION AND TROPHIC ECOLOGY OF MENIPPE MERCENARIA

Introduction

Habitat loss has been shown to have a major impact on species richness and biodiversity in marine and terrestrial ecosystems (Ehrlich 1988, Dulvy et al. 2003). Not only can disturbances to ecosystems affect organisms directly, but they can also cause changes in the community structure. Anthropogenic stressors, such as nutrient input into coastal waters, have been shown to alter ecosystem dynamics and condition such as productivity to higher trophic levels, low dissolved oxygen content, and habitat loss in nearshore communities (Nixon et al. 1986, Short and Burdick 1996, Breitburg et al. 1997). These alterations, among others, can result in drastic changes to trophic structure as well as the condition of individual species.

When characterizing the condition of an organism, it is also important to take into account prey that may be affected due to disturbances. For example, nutrient loading in Waquoit Bay, Massachusetts has led to a decline in eelgrass habitat and an increase in macroalgae biomass. This transition from eelgrass to macroalgae dominated communities has had further implications on trophic structure and a decline in the abundance of bay scallop, Argopecten irradians (Valiela et al. 1992). Effects such as these can have further implications on upper trophic level organisms that rely on bay scallops as a food source, causing a bottom-up effect. In another example, decreased nutritional condition and subsequent decline in Steller sea lion, Eumetopias jubatus, populations in the Gulf of Alaska and off the Aleutian Islands was attributed to a decline in the quality of prey available (Trites and Donnelly 2003). The condition of a species may also be affected by habitat alteration or degradation. Nearshore habitat alterations caused by coastal industrialization and chemical contaminants were shown to impact the growth and condition of juvenile sole, Solea solea, in the English Channel. Sole grew slower
and had lower condition indices in areas with higher amounts of runoff and chemical contaminants (Amara et al. 2007).

The loss of habitat due to cyanobacteria blooms in Florida Bay could be affecting *M. mercenaria* directly or indirectly. *Menippe mercenaria* predominantly feed on hard-shelled bivalves throughout their range (Menzel and Nichy 1958, Powell and Gunter 1968, Lindberg and Marshall 1984, Brown and Haight 1992) with other potential prey in Florida Bay consisting of gastropods and echinoderms (Pharo pers. obs.). Large areas of seagrass habitat in the Bay, often adjacent to hard-bottom, can have > 75 taxa of gastropods and > 25 taxa of bivalves (Nizinski 2007) making these areas ideal foraging grounds. Species such as turban snails, hard clams, and oysters can be plentiful in non-degraded hard-bottom regions of the Bay, but appear to be far less common where blooms have drastically changed benthic trophic communities and bottom structure (Pharo pers. obs.). Any potential change in prey abundance or diversity could negatively affect the nutritional condition of *M. mercenaria* in degraded regions of Florida Bay. A shift in prey or primary producers could also alter the trophic niche and nutritional condition of *M. mercenaria*. Inter- and intra-species variability in lipid, protein, and fatty acid concentrations have been shown to occur in several species of mollusks (Soriguer et al. 1997), and such differences can then affect nutritional condition of predators such as *M. mercenaria*. I compared nutritional condition and trophic position of *M. mercenaria* between degraded and non-degraded regions of Florida Bay using several condition indices and muscle stable isotope analysis.
Methods

Objective 1: To Determine the Effects of Hard-Bottom Habitat Degradation on the Nutritional condition of M. mercenaria.

During the summer of 2017, crabs (n=123) were sampled from degraded and non-degraded regions of Florida Bay by hand. Degraded regions of the Bay were chosen based on previous literature and knowledge of non-degraded hard-bottom communities in the past. To determine the effects of hard-bottom habitat degradation on the nutritional condition of M. mercenaria, three indices were measured.

Immediately after removal from the water, 0.1 mL of hemolymph was extracted from the most proximal leg sinus of the crab using a 27-g 1-mL tuberculin syringe and placed on an industrial fluid refractometer to determine its refractive index (RI) to 0.5 units. The RI has been used to assess nutritional condition in lobsters and other decapods since it is a reliable proxy for serum protein (Musgrove 2001, Behringer and Butler 2006, Gutzler and Butler 2017); however, serum protein levels of crabs are known to change with molt stage (Smith and Dall 1982) and there is currently no method for assessing the molt stage of M. mercenaria.

The dry weight index (DWI) was also measured and compared between M. mercenaria in degraded and non-degraded regions to indicate any changes in nutritional condition. This index has been shown to be the preferred indicator for nutritional condition in crustaceans as it is not affected by molt stage (Gutzler and Butler 2017). Crabs were iced for 10 min, euthanized and dissected to obtain the hepatopancreas (Fig. 2-1). The hepatopancreas is an important organ in decapods which stores lipids, carbohydrates and protein, provides digestive enzymes (Gibson and Barker 1979), and is directly associated with nutritional condition (Rosemark et al. 1980). Starvation studies in the western rock lobster, Panulirus longipes, have shown a stark decrease in the mass of the hepatopancreas when starved (Dall 1974). The hepatopancreas of each crab was
weighed wet and then placed in a freeze drier for 72 h to obtain the dry weight. After dissection of the hepatopancreas, the remaining carcass was placed in a drying oven for 24 h at 60° C to determine its dry weight for use in an additional index. Hepatopancreas dry weight and crab wet weight were used to determine the DWI calculated by Equation 2-1:

\[
\text{Dry Weight Index} = \frac{\text{Hepatopancreas Dry Weight}}{\text{Animal Wet Weight}} \times 100
\]  

(2-1)

The weight to carapace width (CW) ratio is another index that has been widely used in nutritional condition studies of other decapods (Catacutan 2002, Oliver and MacDiarmid 2002). CW was measured to 0.1 mm using Vernier calipers and dry weights were measured to 0.001 g. The dry weight was the preferred choice to be used in this index as the wet weight of decapods fluctuates with molt stage and water intake.

All statistical analyses were conducted using R software. Data used in the three indices were found to be non-normal using a Shapiro-Wilks test. Transformations on the data were attempted to achieve normality; however, data were not normal, therefore, indices were analyzed using a non-parametric Kruskal-Wallis test.

**Objective 2: To Determine the Effects of Hard-Bottom Habitat Degradation on the Trophic Ecology of M. mercenaria**

Stable isotope analysis is a widely used method in assessing trophic relationships within communities (Peterson and Fry 1987, Wada et al. 1991, Post 2002). Stable isotopes of elements, such as carbon and nitrogen, are used to determine the niche of different organisms and these isotopes fractionate (relative change in abundance of the isotopes) when incorporated into another organism. Isotopes are commonly referred to in terms of delta (δ) values which are the measure of heavy and light isotopes. Delta values can be calculated by Equation 2-2:

\[
\delta X = [(R_{\text{sample}} / R_{\text{standard}})-1] \times 10^3
\]  

(2-2)
where $X$ is $^{13}$C or $^{15}$N and $R$ is the ratio of $^{13}$C/$^{12}$C or $^{15}$N/$^{14}$N (Peterson and Fry 1987). Carbon is typically used to indicate sources of primary production (Fry and Sherr 1984). $\delta^{13}$C values will often be similar to the diet source of an individual, fractionating very little depending on the protein content of prey (Fry and Sherr 1984, Peterson and Fry 1987). $\delta^{15}$N values indicate an organism’s trophic level within the community and fractionate by approximately 3 ‰ per trophic level (Minagawa and Wada 1984, Peterson and Fry 1987). The higher up in the food web an organism is, the higher its $\delta^{15}$N value will be.

A subset of *M. mercenaria* (n=60) from the nutritional condition study (Obj. 1) were used to compare the trophic position of crabs between degraded and non-degraded regions. Muscle tissue samples were collected from the claws of crabs from degraded (n=30) and non-degraded regions (n=30). Muscle tissue was used because of its slower turnover rate compared with other tissue types, thus its stable isotope values can give an idea of long term (weeks to months) trophic ecology (MacNeil et al. 2006, Madigan et al. 2012). Crabs were selected across similar ranges of weight and CW to decrease variability between samples based on crab size. Tissue samples were dried in a drying oven at 60°C for 24 h and then ground to a powder in a mill (Wig-L-Bug; Crescent Dental). Samples were sent to the University of Florida Department of Geological Sciences Stable Isotope Mass Spectrometry lab for stable isotope analysis. $\delta^{13}$C and $\delta^{15}$N values of individual crabs were compared between degraded and non-degraded habitat to test for differences. $\delta^{13}$C and $\delta^{15}$N values were also compared between sexes and size classes to determine if either factor affected the carbon source or trophic position, respectively, of *M. mercenaria*. All statistical analyses were conducted in R using a non-parametric Kruskal-Wallis test.
Samples of turtle grass, *Thalassia testudinum*, red macroalgae, *Laurencia* sp., brown branching sponge, *Ircinia* sp., and turban snails, *Lithopoma tectum*, were also collected from hard-bottom habitat in degraded and non-degraded regions. Multiple composites were made consisting of a range of 2-5 individuals each from these species sampled in degraded and non-degraded habitat. δ¹³C and δ¹⁵N values were also obtained for these composites and a dual isotope plot was constructed with *M. mercenaria* included to visualize the trophic niche of *M. mercenaria* in relation to the community.

**Results**

**Objective 1: To Determine the Effects of Hard-Bottom Habitat Degradation on the Nutritional Condition of *M. mercenaria***

Crab size ranged from 34.8 to 101.7 mm CW and dry weight from 6.1 to 178.1 g. The sex ratio of male to female was 1:1.46. Weight to CW ratios (χ²=0.577, df=1; p=0.448) (Fig. 2-2), hemolymph refractive index (χ²=0.118, df=1; p=0.731) (Fig. 2-3), and DWI (χ²=0.641, df=1; p=0.423) (Fig. 2-4) of *M. mercenaria* were not significantly different between degraded and non-degraded regions. However, the DWI differed significantly between sexes (χ²=4.327, df=1; p=0.038) (Fig. 2-5), with the DWI of females being significantly less compared to males. This is most likely due to females utilizing energy and lipids from the hepatopancreas for reproduction. To account for this difference, the DWI of each sex was compared separately between degraded and non-degraded regions (Fig. 2-6). Females showed no significant difference in DWI in degraded versus non-degraded habitats (χ²=0.136, df=1; p=0.713), although hepatopancreas weight varied based on reproductive stage. No significant difference was found in DWI for males between degraded and non-degraded habitats, indicating that nutritional condition was similar (χ²=0.002, df=1; p=0.969).
Objective 2: To Determine the Effects of Hard-Bottom Habitat Degradation on the Trophic Ecology of *M. mercenaria*

The CW of crabs selected for stable isotope analysis ranged from 34.8 to 97.8 mm and the dry weight ranged from 6.1 to 147.1 g. The sex ratio of males to females was 1:1.5. The weight to CW ratio of *M. mercenaria* used (n=30 in degraded and n=30 in non-degraded) did not differ significantly between degraded and non-degraded regions ($\chi^2=1.259$, df=1; p=0.262) (Fig. 2-7). Stable isotope analysis revealed a significant difference in the trophic niche of *M. mercenaria* between degraded and non-degraded regions (Fig. 2-8). The mean (± SD) $\delta^{13}$C values of *M. mercenaria* in non-degraded and degraded regions were -12.78 ± 0.86 and -9.76 ± 1.30, respectively. The $\delta^{13}$C value of *M. mercenaria* in non-degraded regions was significantly more negative than that of crabs in degraded regions ($\chi^2=40.415$, df=1; p<0.001) (Fig. 2-9A). The mean (± SD) $\delta^{15}$N values of *M. mercenaria* in non-degraded and degraded regions were 6.78 ± 0.34 and 6.72 ± 0.53, respectively. $\delta^{15}$N values showed no significant difference between degraded and non-degraded regions, but showed more variability in degraded regions ($\chi^2=0.813$, df=1; p=0.367) (Fig. 2-9B). Comparisons were also made to distinguish whether trophic ecology was related to sex (Fig. 2-10). Neither $\delta^{13}$C ($\chi^2=0.018$, df=1; p=0.892) (Fig. 2-11A) nor $\delta^{15}$N ($\chi^2=0.001$, df=1; p=0.976) (Fig. 2-11B) values differed significantly between female and male *M. mercenaria*.

Crabs were divided into two size classes, large and small, relative to the median carapace width (72.6 mm) of the 60 samples taken. Comparisons were made between size classes to determine whether size was correlated with $\delta^{15}$N or $\delta^{13}$C values (Fig. 2-12). Crabs within the small size class ranged from 34.8 to 72.6 mm CW with a mean (± SD) of 59.4 ± 9.94 mm. The larger size class ranged from 72.6 to 97.8 mm CW with a mean (± SD) of 81.3 ± 6.16 mm. $\delta^{13}$C values did not differ significantly between small and large *M. mercenaria* ($\chi^2=0.483$, df=1;
p=0.487) (Fig. 2-13A). $\delta^{15}$N values also did not differ significantly between size classes ($\chi^2=3.526; df=1; p=0.06$) (Fig. 2-13B); however, the nitrogen values were more variable among smaller crabs.

**Discussion**

Habitat degradation in Florida Bay appeared to have little impact on the nutritional condition of *M. mercenaria*, regardless of the metric used. However, stable isotope results suggest a difference in the trophic niche of *M. mercenaria* between degraded and non-degraded regions with an apparent shift in $\delta^{13}$C values. The $\delta^{15}$N values of *M. mercenaria* also appear to be much more variable in degraded regions of the Bay.

Previous studies have reported a decline in species abundance and diversity after habitat loss or degradation (Butler et al. 1995, Deegan et al. 2002, Hiddink et al. 2006), but few compared species-specific nutritional condition between disturbed versus undisturbed environments. Nutrient loading and chemical contaminants within sediments along industrialized shorelines were shown to decrease growth rates and lower condition of juvenile sole, *Solea solea*, in the English channel compared with shorelines with minimal anthropogenic impacts (Amara et al. 2007). Ecosystem changes due to anthropogenic stressors have also been shown to affect trophic structure and cause declines in abundance of important lower trophic level species (Valiela et al. 1992). It appears the reoccurring cyanobacteria blooms in Florida Bay may also be responsible for altering the trophic structure within degraded regions, but not necessarily affecting the nutritional condition of *M. mercenaria*.

The similar nutritional condition of *M. mercenaria* between degraded and non-degraded regions of the Bay suggests a resilience to this particular disturbance possibly due to a similarity in prey availability between regions. Common prey items such as gastropods and bivalves (Lindberg and Marshall 1984, Brown and Haight 1992) may be unaffected by cyanobacteria.
blooms and thus just as abundant in degraded regions as in non-degraded regions. Previous studies have shown *M. mercenaria* outcompete *P. argus* for shelter (Behringer and Hart 2017) and degradation has been shown to affect lobster population densities (Butler et al. 1995), so there is a possibility that prey availability would increase for *M. mercenaria* in the absence of lobsters in degraded regions of the Bay.

If *M. mercenaria* do have access to similar prey within degraded and non-degraded regions, the shift in $\delta^{13}$C values could indicate a potential change at a lower trophic level within the community. The primary producer supporting the primary consumers or detritivores on which *M. mercenaria* prey may have changed due to the chronic effect of reoccurring blooms, and these changes are reflected bottom up through higher trophic levels. When multiple species were plotted in a dual isotope plot (Fig. 2-14), it appeared that *M. mercenaria* in non-degraded regions shared similar $\delta^{13}$C values to those of the macroalgae, *Laurencia* sp., suggesting this species may be the primary producer indirectly supporting crabs through prey in non-degraded regions. *Laurencia* sp. has been shown to be the main source of primary production supporting hard-bottom communities (Behringer and Butler 2006) and occur as large mats in non-degraded regions; however, these algal mats are far less apparent in degraded hard-bottom (Pharo pers. obs.). *M. mercenaria* in degraded regions exhibited $\delta^{13}$C values intermediate to *Laurencia* sp. and *Thalassia testudinum*, suggesting both primary producers may be supporting crabs indirectly in degraded regions. It could be that *M. mercenaria* in degraded regions may be foraging more heavily in adjacent seagrass beds when compared with crabs in non-degraded hard-bottom. Mere geographical variability in $\delta^{13}$C values of sediment or primary producers could also contribute to the observed differences.
Alternatively, *M. mercenaria* could be exhibiting a dietary shift if certain prey items are negatively impacted by cyanobacteria blooms. Previous research has shown that molluscan species, such as bay scallops and blue mussels, are negatively affected by the bloom forming algae, *Aureococcus anophagefferens*, along the mid-Atlantic (Bricelj and Lonsdale 1997, Tracey 1988, Shumway 1990). In Florida, clearance rates of hard clams, oysters, and scallops have been shown to be affected by the toxic bloom forming algae, *Karenia brevis* (Leverone et al. 2007). If certain molluscan species are similarly affected by cyanobacterial blooms in Florida Bay, a shift in diet to a more robust molluscan species could be occurring in bloom impacted regions.

*Menippe mercenaria* could also be exhibiting a shift in diet to more robust species such as sea urchins or sea cucumbers, both of which crabs were observed to prey upon (Pharo pers. obs.). The higher $\delta^{13}C$ values seen in *M. mercenaria* in degraded regions of the Bay could be due to a change in diet caused by the effects of the cyanobacteria blooms on species normally preyed upon in regions not impacted by blooms. Furthermore, the variability observed in $\delta^{15}N$ values of *M. mercenaria* from degraded regions may be due to a more opportunistic or variable diet. Variability in $\delta^{15}N$ values of the same species have been attributed to diet variability among individuals (Deniro and Epstein 1981). Due to possible effects of blooms on prey species, *M. mercenaria* may be feeding on a variety of different organisms at different trophic levels throughout degraded regions.

Despite extensive habitat degradation, *M. mercenaria* have been able to acclimate to this disturbance and still maintain a similar condition. The most likely scenario being a more opportunistic feeding strategy as seen by the results of stable isotope analysis, with *M. mercenaria* in degraded regions utilizing adjacent seagrass habitat and feeding on a wider variety of organisms. Further research should be focused on the impact of cyanobacterial blooms on
lower level trophic organisms in hard-bottom habitat, mainly prey species of *M. mercenaria*. Examining the effects of cyanobacteria blooms on bivalves, gastropods, and echinoderms that *M. mercenaria* would normally prey upon would give insight into other species that may be impacted by the blooms. Collection of lower trophic level organisms and stable isotope analysis could also be used to help further explain the alterations in trophic structure between degraded and non-degraded regions. Furthermore, directly observing foraging habits of *M. mercenaria* between degraded and non-degraded regions would yield valuable information on the effects of cyanobacteria blooms on feeding behavior. Although algal blooms have been known to cause decreased condition and mortality in many species (Paul et al. 2005, Leverone et al. 2007), the ecological impacts of this particular cyanobacteria bloom do not seem to directly affect the nutritional condition of *M. mercenaria*. 
Figure 2-1. Underside of a stone crab carapace with hepatopancreas being removed for analysis. Photo courtesy of author.
Figure 2-2. Box plots of the weight to carapace width ratios of *M. mercenaria* between degraded and non-degraded regions. The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-3. Box plots of the hemolymph refractive index of *M. mercenaria* between degraded and non-degraded regions. The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-4. Box plots of the dry weight index of *M. mercenaria* between degraded and non-degraded regions. The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-5. Box plots of the dry weight index of *M. mercenaria* compared between males and females. The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-6. Box plots of the dry weight index of female and male *M. mercenaria* between degraded and non-degraded regions. The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-7. Weight versus carapace width for *M. mercenaria* samples (n=60) for which muscle tissue was analyzed for $\delta^{13}$C and $\delta^{15}$N values at degraded and non-degraded sites.
Figure 2-8. Dual isotope plot comparing δ¹³C and δ¹⁵N values of *M. mercenaria* between degraded and non-degraded regions. Each point represents an individual crab. The ellipses around both degraded and non-degraded points represent the 95% Confidence Intervals of the data.
Figure 2-9. Box plots of the $\delta^{13}$C and $\delta^{15}$N values of *M. mercenaria* between degraded and non-degraded regions. $\delta^{13}$C values are depicted on the left (A) and $\delta^{15}$N values on the right (B). The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-10. Dual isotope plot depicting δ¹³C and δ¹⁵N values between male and female *M. mercenaria*. Each point represents an individual crab. The ellipses around both degraded and non-degraded points represent the 95% Confidence Intervals of the data.
Figure 2-11. Box plots of the $\delta^{13}$C and $\delta^{15}$N values of *M. mercenaria* between females and males. $\delta^{13}$C values are depicted on the left (A) and $\delta^{15}$N values on the right (B). The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-12. Dual isotope plot depicting $\delta^{13}$C and $\delta^{15}$N values of *M. mercenaria* between size classes. Each point represents an individual crab. The ellipses around both degraded and non-degraded points represent the 95% Confidence Intervals of the data.
Figure 2-13. Box plots of the $\delta^{13}$C and $\delta^{15}$N values of *M. mercenaria* between size classes. $\delta^{13}$C values are depicted on the left (A) and $\delta^{15}$N values on the right (B). The dark shaded horizontal line indicates the median of the data set. The box represents the middle 50% of scores for the data, while the ‘whiskers’ extending from the box represents the maximum and minimum values of the data set. Individual points represent outliers.
Figure 2-14. Dual isotope plot comparing $\delta^{13}$C and $\delta^{15}$N values for multiple species between degraded and non-degraded habitat. Ellipses represent 95% confidence intervals for species. Each point for Laurencia sp., Lithopoma sp., and Thalassia testudinum represent composite samples made of 5 individual samples each. Points shown for Menippe mercenaria represent one individual crab each.
CHAPTER 3
THE EFFECTS OF HARD-BOTTOM HABITAT DEGRADATION ON SITE AND DEN FIDELITY OF MENIPPE MERCENARIA IN FLORIDA BAY

Introduction

Movement patterns and fidelity to a particular habitat or shelter can have a significant effect on the range and survival of a species. For example, these characteristics can affect the ability of species to colonize new areas (Brousseau et al. 2002), survive harsh winter temperatures (Wilber 1986), or escape hypoxic areas (Lenihan and Peterson 1998). Movement can also be facilitated by habitat fragmentation (Micheli and Peterson 2001, Hovel and Lipcius 2001), food availability (Bearzi et al. 2006), and ability to find a mate (Diesel 1986). Factors such as these can determine the extent to which a population can expand or contract. However, if conditions are favorable in a particular habitat and resource availability is adequate, organisms may show strong fidelity to areas. Adaptability also plays a large role in the structuring of populations. The ability of an organism to utilize or adapt to multiple habitat types or conditions can determine the dispersal of a population. Different marine organisms exhibit a variety of different patterns of fidelity and movement.

Several species of fish have been shown to exhibit strong site fidelity to specific intertidal rock pools and were also shown to return to original pools after being relocated 5-30 m away (White and Brown 2013). The mangrove swimming crab, *Thalamita crenata*, uses visual cues within its home range to relocate its shelter after foraging, showing high site fidelity (Cannicci et al. 1995). Loggerhead turtles, *Caretta caretta*, do not necessarily show high fidelity to one area, but are known for their homing abilities allowing them to locate the same foraging grounds and nesting beaches repeatedly (Bowen 2004). *P. argus* has also been shown to have a homing ability (Hernkind and McLean 1971) and is able to forage and relocate its shelter afterwards or use several shelters constantly moving back and forth among them (Bertelsen and Hornbeck...
2009). In the northwest region of Florida, *M. mercenaria* was found to travel relatively short distances between dens, exhibiting short term fidelity to particular inshore oyster reefs (Wilber 1986). *Menippe mercenaria* was also shown to exhibit low fidelity to single dens in a manipulated shelter study. However, crabs were resighted within surrounding areas on subsequent surveys (Lindberg et al. 1990). Prey depletion within the area was hypothesized as the reason for the dispersal of crabs. Alternatively, the Asian shore crab, *Hemigrapsus sanguineus*, shows very low site fidelity and high adult mobility making it a very successful invader (Brousseau et al. 2002). Many of these examples of dispersal and homing abilities can be attributed to visual cues, however, several species in the marine environment rely on olfaction to locate suitable habitat.

In aquatic environments, olfaction plays a large role in the movement and behavioral patterns of many species. Chemoreception is used for hunting, predator avoidance (Turner et al. 2003, Berger and Butler 2002, Behringer and Hart 2017), finding mates (Díaz and Thiel 2004, Atema 1986, Hassler and Brockmann 2001), and disease avoidance (Behringer et al. 2018), among other important roles. Crustaceans, such as the Caribbean spiny lobster use chemical cues for many functions including avoidance of diseased conspecifics or to seek out healthy conspecifics to shelter with (Anderson and Behringer 2013). Chemoreception is also used by many species to cue settlement from their pelagic larval habitat (Forward et al. 2001, Raimondi 1988). For *M. mercenaria*, chemical cues from the brown algae *Sargassum fluitans* have been shown to induce metamorphosis in juvenile *M. mercenaria* on the west coast of Florida (Krimsky and Epifanio 2008). Considering the importance of chemoreception to many crustaceans, including *M. mercenaria*, it would seem plausible that *M. mercenaria* would use
chemical cues exuded by benthic substrate as a means to navigate, find shelter, or find suitable hard-bottom in which to reside.

The loss, degradation, or fragmentation of ecosystems could alter normal behavioral patterns, leading to changes in movement or the reliability of chemosensory cues. Destruction of important ecosystems has been shown to alter fish and crustacean communities and diversity throughout many ecosystems (King 1998, Lenihan and Peterson 1998). Dramatic declines in the abundance of seagrass-canopy-dwelling crustaceans were reported after massive seagrass die-offs in Florida Bay due to algal blooms, however, benthic crustaceans increased in abundance (Matheson et al. 1999). One of the many side effects of algal blooms are hypoxic or anoxic areas (Paerl et al. 2001), which can kill or cause species to leave affected areas seeking more suitable habitat. Off the coast of North Carolina, oyster reef habitat degradation in conjunction with hypoxic or anoxic zones was shown to cause the blue crab, Callinectes sapidus, to completely abandon their burrows in search of better habitat (Lenihan and Peterson 1998).

Within the hard-bottom habitat of Florida Bay, a wide variety of sponges cover the benthos. A few species, such as Ircenia campana, are known as ‘stinker’ sponges and give off a pungent odor when taken out of the water. These sponges saturate the water column with chemicals that are most likely used by many marine organisms. Furthermore, juvenile and young adult M. mercenaria are found residing in excavated loggerhead sponges (Fig. 3-1A) or solution holes within the porous limestone sea floor (Fig. 3-1B) of the Bay. The loss of shelter could also cause M. mercenaria to leave degraded areas in search of more suitable habitat. Presumably, organisms will only inhabit areas with high prey availability. Prey diversity and abundance seem to be relatively high with several species of bivalves and gastropods residing in the sediment or on sponges and other structure (Pharo pers. obs.). However, prey density and diversity appear to
be much lower in impacted regions of the Bay, possibly due to the lack of habitat or due to the
direct effects of the cyanobacteria blooms. This potential difference in prey availability or
abundance could cause M. mercenaria to emigrate from degraded regions to locate more
desirable habitat with higher prey availability. The loss of these sponges, potential prey and
overall habitat due to persisting cyanobacterial blooms could be affecting fidelity and
chemosensory abilities of M. mercenaria. Here I investigated the effects of hard-bottom habitat
degradation in Florida Bay on fidelity and chemoreception of M. mercenaria.

Methods

Objective 1: To Determine the Effects of Hard-Bottom Habitat Degradation on the Site and
Den Fidelity of M. mercenaria.

During summer 2017, eight sites were selected in areas of Florida Bay impacted (Fig 3-2A) and non-impacted (Fig 3-2B) by cyanobacteria blooms in recent years (e.g., 2007, 2013). Three sites were identified in non-degraded areas and five in degraded areas. Areas were chosen based on the presence and abundance of crabs to facilitate the study. Degraded sites were chosen based on previous literature and the extent of the recent blooms. Sites were 30 m x 30 m plots demarcated with weighted polypropylene line and cinderblocks. After plots were laid out, dens within sites were located. Dens consisted primarily of crevices in or under loggerhead sponges and solution holes in the porous limestone bottom. Only dens with M. mercenaria present on the first survey day were used in the study.

Sites were surveyed by a team of three divers swimming back and forth between plot borders within 1 m of each other to detect any shelters that were within the plots. Crabs were collected by hand and lured from dens with the use of a sausage placed at the shelter entrance as bait. Once captured, crabs were measured at the widest part of the carapace (CW), sexed, and tagged while still under water. Tags consisted of American lobster claw bands color coded with
electrical tape to distinguish individual crabs from one another (Fig. 3-3A). Visible injuries and shell disease (Rosen 1970, Iversen and Beardsley 1976) were also noted. After tagging, crabs were placed back into their respective dens. Each occupied den was marked with a weight and flagging tape with the corresponding color combination of the claw band written on the flagging tape (Fig. 3-3B) to distinguish which crab belonged to which den. Sites and dens were then surveyed again on days two, three, four and eight. On subsequent surveys, crabs were noted as being present in their original den, present in another den within the plot, or absent from the plot. New crabs that had moved into plots throughout the course of the eight days were also noted, but not tagged.

A generalized linear mixed model (GLMM) in R program was used to analyze the difference in fidelity to individual dens and sites between degraded and non-degraded sites. In the model, ‘site’ was treated as the random effect to account for within site variance. Fidelity between sexes was also analyzed given that male stone crabs are much more mobile than females (Wilber 1989, Lindberg et al. 1990). A post hoc analysis of variance (ANOVA) was used to determine any significant difference in fidelity between degraded and non-degraded sites.

A tag retention study was conducted in which 22 crabs were collected by hand from hard-bottom habitat. Crabs were tagged as above and held in aquaria in the laboratory for 8 d. Crabs were also fed various gastropods and bivalves throughout the course of the study. Tag retention was recorded at the end of the 8 d period.

**Objective 2: Response of M. mercenaria to Chemosensory Cues from the Loggerhead Sponge, Spheciospongia vesparium.**

Loggerhead sponges (*Spheciospongia vesparium*) are the largest sponge species in the hard-bottom habitat of Florida Bay (Peterson et al. 2006) and are typically indicative of healthy, non-degraded regions. *Menippe mercenaria* often utilize this species as shelter, excavating into
the sponge itself or occupying crevices below it. All sponges are filter feeders and pump water, potentially releasing chemical compounds into the water column that could attract stone crabs.

Studies were conducted at the Keys Marine Laboratory in Layton, Florida. In the fall of 2016, loggerhead sponges ~30 cm in diameter were cut ~5 cm from the base of the sponge and attached to 30 cm x 30 cm concrete pavers using zip-ties. Pavers with cut sponges were placed in non-degraded hard-bottom areas to heal and attach to the paver over the next seven months (Fig. 3-4A). For the chemosensory experiment, two Y-mazes (Rebach 1996, Ratchford and Eggleston 1998, Diaz and Thiel 2004) were set up with two 100-L head tanks each (Fig. 3-4B). Y-mazes (79 L) were constructed with 1.3 cm plywood and measured 94 cm long x 62 cm wide x 20 cm tall with a central divide 72 cm long x 18 cm high (Fig. 3-4B). One head tank contained only seawater to act as a control while the other contained the sponge treatment. The seawater used in the trials was twice filtered to remove any large particulates. Shelters were constructed of 3 stacked bricks (20 cm x 10 cm x 5 cm) and placed at the end of both chambers to attract crabs seeking shelter.

*Menippe mercenaria* (n=29) were collected from hard-bottom habitat by hand and placed in holding tanks prior to use in the experiment. Experiments were conducted at night to simulate when crabs would be foraging and returning to shelter. For each trial, a sponge was placed in one head tank for 30 min to allow chemical compounds being pumped from the sponge to diffuse into the water. Head tank nozzles were then opened, allowing sponge treated and untreated water to flow into either side of the choice chamber and into the center of the shelters at a rate of 5 ml/s. A dye test was used on each Y-maze to confirm that water and cues remained on their respective sides and moved at a similar rate toward the drain at the base of the maze. Prior to experimental trials, each crab was placed at the base of the maze behind a divider and allowed to
acclimate for 5 min. The divider was then lifted and the trial began. Crabs typically made a choice of side after 30 min, after which final choice was recorded. A proportion test was used in R to analyze the results.

**Results**

**Objective 1: To Determine the Effects of Hard-Bottom Habitat Degradation on the Site and Den Fidelity of *M. mercenaria***.

The tag retention study resulted in 86% retention over the course of 8 d, so it was possible during the field study that some crabs lost their tags. However, based on retention rates this was minimal and lost tags were often found outside the burrow of the crab that lost it. A total of 106 crabs were tagged in the field, 40 in degraded sites and 66 in non-degraded sites. The CW ranged from 30.7 to 93.1 mm in degraded and 50.4 to 94.7 mm in non-degraded sites. The sex ratio of males to females was 1:3.1 within degraded and 1:1.2 in non-degraded sites. The GLMM results indicated no significant difference in den fidelity between degraded and non-degraded sites ($\chi^2=0.829$, df=1; $p=0.396$) (Fig. 3-5), nor was there a significant difference in site fidelity among sites ($\chi^2=0.844$, df=1; $p=0.996$) (Fig. 3-6). When male and female crabs were analyzed separately, no significant difference was found in female den fidelity ($\chi^2=0.814$, df=1; $p=0.367$) (Fig. 3-7A), nor was there a significant difference in site fidelity ($\chi^2=0.232$, df=1; $p=0.630$) (Fig. 3-7B) between degraded and non-degraded sites. Similarly, no significant difference was found in males for den fidelity ($\chi^2=0.321$, df=1; $p=0.571$) (Fig. 3-8A) or site fidelity ($\chi^2=0.036$, df=1; $p=0.849$) (Fig. 3-8B) between degraded and non-degraded sites.

**Objective 2: Response of *M. mercenaria* to Chemosensory Cues from the Loggerhead Sponge, *Spheciospongia vesparium***.

A total of 28 Y-maze trials were performed. The CW of *M. mercenaria* ranged from 50.7 to 102.4 mm. Crabs chose the sponge treatment over seawater alone 67.9% of the time; however,
the difference between treatments was of borderline significance ($\chi^2=3.571$, df=1; p=0.058) (Fig. 3-9).

**Discussion**

Den and site fidelity of *M. mercenaria* were found to be similar between degraded and non-degraded sites, and there was no difference in fidelity between sexes either. Despite the large loss in loggerhead sponges in Florida Bay, which are used as shelter by many organisms (Pearse 1950, Butler et al. 1995, Behringer and Hart 2017), *M. mercenaria* were able to utilize the abundance of solution holes within degraded sites. Chemosensory trials also indicated an ability of *M. mercenaria* to detect chemical cues emitted by loggerhead sponges and the subsequent attraction to these cues.

Habitat loss and degradation have been linked to population declines and decreased species diversity in ecosystems globally (Brooks et al. 2002, Gray 1997, Valiela et al. 1992). For example, results of many studies have shown stark declines in fish abundance and diversity on coral reefs after bleaching events and subsequent loss of habitat for reef dwelling species (Bonin 2011, Pratchett et al. 2008, Pratchett et al. 2011). In the hard-bottom habitat of Florida Bay, habitat degradation and the loss of sponges was shown to greatly reduce populations of *P. argus* due to a decline in available shelters in degraded regions (Butler et al. 1995). However, results reported here indicate habitat degradation in the Bay did not affect den or site fidelity of *M. mercenaria* or its ability to find suitable shelter, most likely due to the availability of solution holes and their ability to excavate burrows in seagrass.

*Menippe mercenaria* did not emigrate from degraded regions at a significantly faster rate than those in non-degraded regions indicative of an acclimation to an altered habitat. However, the results suggest that *M. mercenaria* spend more time in degraded regions than non-degraded regions, albeit this result was not statistically significant. Presumably, the abundance of
resources, such as prey and shelter, within non-degraded habitat gives *M. mercenaria* the ability to move around and forage in a larger area, resulting in limited fidelity to a particular site or shelter. The lack of these resources in degraded regions could be forcing *M. mercenaria* to forage more opportunistically, possibly taking advantage of adjacent seagrass habitat as foraging grounds. *Menippe mercenaria* may also be feeding on a wider variety of organisms in degraded regions, as shown by the variability in δ¹⁵N values of crabs in these regions, allowing them to acclimate and remain in these regions. After massive seagrass die-offs in Florida Bay, also associated with cyanobacteria blooms, benthic crustaceans were shown to increase in abundance (Matheson et al. 1999), possibly moving in to capitalize on available space. A similar situation could be occurring for *M. mercenaria* in affected regions of the Bay. Bottom trawling and subsequent habitat disturbance and destruction has been shown to illicit the immigration of the hermit crab, *P. bernhardus*, into recently disturbed areas to feed on injured or dead organisms caused by trawling (Ramsay et al. 1996). This behavioral shift in foraging could also be occurring in degraded regions, causing *M. mercenaria* to forage in adjacent areas or feed on organisms injured by blooms. It could also be that the loss of sponges and other substrate has left prey items much more vulnerable and available to *M. mercenaria*. Common prey items, such as the gastropod, *Lithopoma tectum*, are found residing on many different sponges. The decline in sponge biomass may have caused this species to forage on open bottom leaving them more vulnerable to predation. Whether it is the relative similarity in prey availability or the shift in diet and adaptability to an altered ecosystem, fidelity of *M. mercenaria* in degraded sites remained similar to crabs in non-degraded sites. *Menippe mercenaria* may also be using chemical cues in hard-bottom habitat as a means of relocating suitable habitat or shelter after foraging.
Chemoreception can play a large role in aquatic habitats and affect behavioral patterns of many organisms including *M. mercenaria* (Behringer and Hart 2017, Krimsky and Epifanio 2008). The results of the Y-maze experiments, although of borderline significance, strongly suggest that *M. mercenaria* may be attracted to chemical cues exuded by loggerhead sponges, potentially as an indication of suitable habitat nearby. Substrate other than sponges, such as corals and the red macroalgae (e.g., *Laurencia* sp.), could also be emitting chemical cues used by *M. mercenaria* for navigation. Numerous organisms, such as the mangrove swimming crab, *Thalamita crenata*, and Caribbean spiny lobster, *Panulirus argus*, use visual cues or an internal map to locate shelters (Cannicci et al. 1995, Boles and Lohmann 2003). However, many species rely heavily on chemoreception for navigation and homing (Croll 1983). The loss of sponges and other substrate could be impacting movement patterns of *M. mercenaria* in degraded areas of Florida Bay. Furthermore, chemical cues, such as those given off by the brown alga, *Sargassum fluitans*, have been shown to elicit settlement and subsequent metamorphosis in *M. mercenaria* on the west coast of Florida (Krimsky and Epifanio 2008) so the loss of habitat in Florida Bay could be affecting settlement of larvae and population distribution.

Cyanobacterial blooms and the subsequent loss of hundreds of km² of hard-bottom habitat may affect the ecology of *M. mercenaria* in different ways. The fidelity shown by crabs in degraded regions is comparable to non-degraded regions; however, the attraction that *M. mercenaria* appears to show to the chemical cues emitted by loggerhead sponges may play an important role in their ability to find suitable habitat, such that the loss of habitat could be affecting chemoreception in *M. mercenaria*. Further research should investigate the general movement, home range and foraging behavior of *M. mercenaria* in hard-bottom habitat as well as their ability to relocate shelter after foraging via visual or chemical cues. General movement
of *M. mercenaria* could answer an important question regarding whether crabs are staying within one large patch of hard-bottom habitat or if they are moving in between habitats. Furthermore, the foraging behavior of *M. mercenaria* could give insight to where they chose to forage at night and what habitats they utilize as foraging grounds in degraded versus non-degraded regions.
Figure 3-2. Degraded and non-degraded sites of Florida Bay used in fidelity study. Degraded sites were located off Islamorada, FL in the impacted regions of blooms (A). Non-degraded sites were located off Long Key and Marathon, FL in non-impacted regions of blooms (B). Photos adapted from Google Earth.
Figure 3-3. Tags and weights used to mark individual crabs. Color coded American lobster claw band used to tag largest claw on each crab (A). Stone crab tagged in a marked loggerhead sponge den (B). Photos courtesy of author.
Figure 3-4. Sponges and Y-mazes used in chemosensory trials. Loggerhead sponge zip-tied to a paver for use in chemosensory trials (A). Y-maze experiments with head tanks containing seawater or sponge treated water (B). Photos courtesy of author.
Figure 3-5. Mean percentage of crabs recaptured within their original dens over the course of 8 d in degraded and non-degraded sites. Error bars represent the 95% confidence intervals.
Figure 3-6. Mean percentage of crabs recaptured on sites over the course of 8 d in degraded and non-degraded sites. Error bars represent the 95% confidence intervals.
Figure 3-7. Fidelity of female crabs to individual dens and sites over the course of 8 d in degraded and non-degraded sites. Mean percentage of female crabs recaptured within their original dens (A) and on sites (B). Error bars represent the 95% confidence intervals.
Figure 3-8. Fidelity of male crabs to individual dens and sites over the course of 8 d in degraded and non-degraded sites. Mean percentage of male crabs recaptured within their original dens (A) and on sites (B). Error bars represent the 95% confidence intervals.
Figure 3-9. Results of the final selections of *M. mercenaria* in chemosensory trials.
Menippe mercenaria is an important and abundant benthic predator throughout its range. In the Florida Keys, *M. mercenaria* supports higher trophic level organisms such as the red drum, *Sciaenops ocellatus*, as well as a multimillion dollar commercial fishing industry (FWC 2017). Natural disturbances to species, such as *M. mercenaria*, could alter trophic structure within hard-bottom communities, causing further trophic cascades as has been shown in other species in the Florida Keys (Butler et al. 1995). Trophic cascades have been reported in many ecosystems, marine and terrestrial, and can have deleterious effects, restructuring communities and altering overall energy flow (Heck and Valentine 2006, Shurin et al. 2002, Wilmers et al. 2012). Therefore, it is imperative we understand the effects of persisting cyanobacteria blooms on the ecology and biology of this ecologically and commercially valuable species.

The effects of cyanobacterial blooms in Florida Bay, although devastating to the benthos, do not seem to negatively impact the condition of *M. mercenaria*. Nutritional condition index comparisons between degraded and non-degraded regions of the Bay appear to be quite similar, indicating no effect or an acclimation to this particular disturbance. If common prey items such as bivalves and gastropods are also unaffected by blooms this could explain the similarity in condition of *M. mercenaria*. No comparative studies between regimes have been done on the abundance and condition of lower trophic level organisms within degraded regions of Florida Bay, leaving this an open question.

Alternatively, *M. mercenaria* could be foraging more opportunistically in degraded regions of the Bay possibly utilizing nearby seagrass beds as foraging grounds. This could be explained by the apparent shift in trophic niche of *M. mercenaria* to a more positive δ\textsuperscript{13}C value in degraded regions similar to seagrass δ\textsuperscript{13}C values. The variability in δ\textsuperscript{15}N values could also be
attributed to *M. mercenaria* feeding on a number of different trophic levels within degraded regions.

The loss of habitat also does not seem to affect the fidelity of *M. mercenaria* to a given area. Although hundreds of km² of sponge habitat has been lost due to reoccurring cyanobacteria blooms, the abundance of solution holes within hard-bottom areas may provide adequate shelter to *M. mercenaria* in these regions. However, the loss of sponges could be affecting crab’s ability to find suitable habitat by limiting chemical cues emitted by substrate, such as sponges, in degraded regions. The results suggest *M. mercenaria* is attracted to loggerhead sponges over seawater alone, indicating an attraction towards healthy non-degraded hard-bottom. This could elicit a problem, increasing time to find suitable habitat or shelter after foraging, leaving crabs vulnerable to predation for longer periods of time. However, given that *M. mercenaria* show short term fidelity within degraded regions of Florida Bay, it is possible that crabs are using visual cues or an internal ‘map’ of their home range to locate their shelter (Cannicci et al. 1995) as opposed to relying on chemical cues alone.

In this study, I aimed to discover the effects of habitat degradation due to cyanobacteria blooms on the ecology and biology of the Florida stone crab in Florida Bay and although *M. mercenaria* appear to exhibit a high resilience to this particular natural disturbance, further studies are needed to address other questions. Several studies have shown the diet and prey preference of *M. mercenaria* on the west coast of Florida (Menzel and Nichy 1958, Powell and Gunter 1968, Lindberg and Marshall 1984, Brown and Haight 1992), but there is a lack of knowledge on the ecology of stone crabs in the Florida Keys, the largest provider of stone crab claws in the industry. Future research should be focused on potential prey of *M. mercenaria* in Florida Bay and the effects of cyanobacteria blooms on these species. This information could
lead to a better understanding in the shift of trophic niche of *M. mercenaria* between degraded and non-degraded regions. Although HABs have been shown to negatively impact important marine species throughout the world (Bricelj et al. 1987, Butler et al. 1995, Gravinese et al. 2018), it would appear that the cyanobacteria blooms occurring in Florida Bay have little impact on *M. mercenaria*, based on the metrics evaluated here.
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BIOGRAPHICAL SKETCH

Devon Pharo was born in Stuart, Florida in 1991. He grew up exploring everything Florida had to offer from the beaches to the reefs in south Florida and the Florida Keys. His family would often travel throughout the Caribbean, furthering his love of tropical marine environments. He became enrolled at Santa Fe College in 2009 before transferring to the University of Florida (UF) in 2011 to study Biology. Devon pursued his diving career at UF’s dive program and became friends with a graduate student who would later introduce him to Dr. Behringer. He volunteered in the Fisheries and Aquatic Sciences Department before graduating in 2013 with a BS in biology and a minor in wildlife ecology and conservation. After graduating he was employed at UF as a technician on several wildlife-related projects. In 2016, he was hired by Dr. Behringer to work on an experiment examining the effects of *Panulirus argus* virus 1 (PAV1) on early benthic juvenile Caribbean spiny lobsters. He worked in the Florida Keys for 6 months on this project before applying to graduate school with Dr. Behringer’s lab. He was accepted and immediately began working on a project examining the effects of hard-bottom habitat restoration efforts in the Florida Keys. While working, Devon noticed the abundance of young adult Florida stone crabs residing in hard-bottom communities within Florida Bay and was intrigued by similar abundances of crabs in regions of the Bay degraded by reoccurring cyanobacteria blooms. Hypotheses generated lead to the thesis research described in this document.