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Fish and decapod community structure in estuarine habitats of the New Orleans Land Bridge, including a description of the life cycle of tarpon (*Megalops atlanticus*) in southeastern Louisiana

A Dissertation

Submitted to the Graduate Faculty of the University of New Orleans in partial fulfillment of the requirements for the degree of

Doctor of Philosophy in Engineering and Applied Science

by

William Stein, III M.D.

B.A. Vanderbilt University, 1970 M.D. Louisiana State University School of Medicine, 1974

May, 2013

### Dedication

This work is dedicated to Mary Betz Stein and William Stein, Jr.

To My Wife of thirty-three years who has supported me in all of my endeavors with grace, love, and pride; who has nursed my wounds and edited my papers.

And to My Father who practiced law for over 40 years while always wanting to be an engineer. He implanted in me the desire to succeed, the need to excel, and the knowledge that I could only regret what was left undone.

### Acknowledgments

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The most exciting part of my research was the time I got to spend in the marsh on my boat, "Tarpon Research". I first became acquainted with my research area over 50 years ago and the opportunity to become reacquainted has had a very special meaning to me. But all of the hours on the boat would not have been the same without a man who has become a very special friend and fellow scientist and without whose help this work would never have been completed. Galen Smith and I shared over 750 hours in the marsh in the hot sun and in the windy cold, in thunderstorms and rain, when everything broke, and when there was nothing left to go wrong.

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He always had a smile, was incredibly supportive, meticulous and exacting, and didn't make mistakes. I will miss the marsh, but I will miss those days we shared even more.

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#### Abstract

Estuarine marshes are generally considered to be productive but not necessarily diverse ecosystems. During 24 consecutive months, I collected 60,000 fishes and decapods comprising over 61 species from the New Orleans Land Bridge, an estuarine salt marsh. My research details the distribution of nekton across five contiguous but geomorphically different regions, which I defined as "Areas". This factor "Area" was significant in explaining community composition differences in 11 of the 24 months I evaluated. That is, during those 11 months community structure was different among the Areas. Specific "month" was also found to be a significant factor as community structure was found to differ among the months. No consistent abiotic factors were associated with community structure. These observations imply that a different set of factors are associated with community structure at the Area level than at the microhabitat level. Sampling of nekton in shallow estuarine salt marsh habitats was difficult. The cast net is a useful gear type for this type of sampling and can be readily standardized for each operator. Standardization of the area covered by the net allows density of collected nekton to be calculated. Little is known about the life cycle of one important estuarine dependent sport fish, tarpon (Megalops atlanticus), in southeastern Louisiana. This research details the presence of the major life stages of the species in Louisiana and suggests that it is capable of completing its life cycle in State waters. The presence of a spawning capable female and male tarpon is documented.

Keywords: Community structure, New Orleans Land Bridge, *Megalops atlanticus*, Cast net, Spawning tarpon, Tarpon life cycle

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#### Introduction

In an attempt to study the life cycle of tarpon (*Megalops atlanticus*) in southeastern Louisiana, I conducted a thorough and systematic study of the New Orleans Land Bridge (LB). Historic evidence suggested that adult tarpon had occurred in this and nearby water bodies. This region is mostly shallow salt marsh with abundant submersed aquatic vegetation and muddy substrate. Fieldwork here and elsewhere in coastal Louisiana revealed that young of the year and juvenile tarpon are not common in these large expanses of marsh. I continued this work as a comparison of the nekton communities in five geomorphologically distinct areas within the LB. Each of these areas has different amounts of habitats such as tidal sloughs, bayous, intertidal basins, shallow ponds, grass prairies, and mud flats. I attempted to determine if these habitats or the abiotic variables associated with them played a role in determining the composition of the nekton communities. While conducting this research, I also continued my studies into the life cycle of tarpon in Louisiana but I expanded my scope to encompass a larger area. To determine if all life stages of this species occurred in Louisiana, I used diverse and novel methods such as reaching out to local divers and fishers and training them to collect tarpon data as 'citizen scientists".

I collected over 60,000 fishes and decapods comprising more than 61 species during 24 consecutive months of work in the LB. Only one of these specimens was a young of the year tarpon. In Chapter 1, I discuss my nekton community work on the LB and the differences in community structure across five geomorphologically diverse areas. To complete this research in the physically unstable habitats of the LB, I developed a sampling method that relied on using a cast net to collect organisms. The cast net has not often been used for this type of sampling and little information exists on using a cast net as a gear type. In order to justify the use of the cast

net in my research, I developed a way to standardize the net in terms of area covered by different operators. The process accounts for the different capabilities of different operators and provides for the calculation of average density of sampled species. In Chapter 2, I describe the results of my work to standardize the cast net and my comparisons of it to a more commonly used gear type, the throw trap.

During my search for the various life stages of tarpon, I was informed of several young of the year tarpon being collected in a roadside ditch near Port Sulphur, Louisiana. The tarpon had been collected during another research project conducted by the Nekton Research Laboratory (NRL). Since this initial collection, I was successful in identifying not only young of the year tarpon but all life stages of the species from different areas of coastal Louisiana. With the help of numerous divers and fishers, I collected young of the year, juveniles, and adult tarpon. The only tarpon larva (leptocephalus) I could confirm was a single specimen housed in the Tulane Museum of Natural History - Royal D. Suttkus Fish Collection. In Chapter 3, I describe the life cycle of tarpon in southeast Louisiana based on these multiple collections and occurrences of this species. Before this research, little was known about the life cycle of this important game fish in Louisiana.

Finally, in Chapter 4 I detail the most significant finding of my research with tarpon in Louisiana. During the summer 2011, a member of the Louisiana Tarpon Club collected an adult during a tarpon rodeo. As with other adult tarpon I had studied, the gonads (in this case ovaries) were removed and brought to the NRL for examination. Based on close histological examination, these ovaries were determined to be from the first spawning capable female tarpon documented in the northern Gulf of Mexico. Prior to my finding, it was assumed all tarpon

spawned either off southern Florida or the Yucatan Peninsula. Here I document the collection of spawning capable tarpon, both female and male, from coastal Louisiana.

### Chapter 1

Fish and decapod community structure in estuarine habitats of the New Orleans Land Bridge Abstract:

Fish and decapod community structure in an estuarine marsh is determined, in part, by the temporal and spatial distribution of abiotic factors. I examined the distribution of fishes and decapods across an estuarine marsh in southeast Louisiana divided into five adjacent but geomorphologically different areas. Over a 24 month period, I collected 2120 community samples along with concurrent sets of abiotic variables in estuarine habitats of the New Orleans Land Bridge. In 11 of the 24 months compared, there were significant differences in community composition among the five areas. None of the measured abiotic factors were predictive of community structure and abiotic differences among areas on a monthly basis did not correspond to differences in community structure. It is likely that other pre-settlement factors (advection, distance from spawning location, etc.) determine composition of community structure in these shallow estuarine habitats.

### Introduction:

"Life and its landscape are intimately related" (Reinhardt et al., 2010). It has long been recognized that the distribution of organisms and their abundance is, to some extent, governed by the spatial and temporal variation in physical processes (Reinhardt et al., 2010). The distribution of nekton is complex at various spatial and temporal scales and densities are not distributed evenly (Minello and Rozas, 2002). A question that has intrigued me is whether or not fishes and

decapods are evenly distributed on a large scale across a geomorphologically diverse marsh; and if not, what are the factors that responsible for their distribution.

The salt water marshes of the estuaries of the Gulf of Mexico differ dramatically in size, climate, and geomorphology, all of which may influence the community structure of fishes and decapods (Rozas et al., 2012). Wetland coverage and freshwater inflow also affect the community structure (Rozas et al., 2012). Microhabitats within the marsh may differ from one section to another. Estuarine resident species may be confined only to areas of the marsh that experience stable abiotic conditions such as salinity or temperature. Many of these species may not be capable of seasonal long distance migration but may migrate to other areas of the marsh in response to local changes which will change distribution and community structure (Able and Fahay, 2010). Recruitment and settlement are not the same thing as settlement only occurs when larvae encounter favorable conditions within a certain window of time (Siegel et al., 2008). For estuarine dependent species, it is possible that recruitment can occur across an entire marsh but that settlement occurs only where local conditions are favorable (Able and Fahay, 2010). For these species, this is a complex process dependent on several additional factors which may be just as difficult to elucidate. These factors may include advection from spawning locations removed from the marsh, area currents and tides, and stochastic weather events such as storms, changes in atmospheric pressure, or prevailing wind (Boehlert and Mundy, 1988; Pineda et al., 2007). In order to address these issues, it is first necessary to know how fishes and decapods are distributed across a large marsh-scape within distinct areas rather than just within microhabitats. The New Orleans Land Bridge (LB) provides an ideal marsh for this type of study (Figure 1).



Figure 1: Map of southeast Louisiana including the Pontchartrain Basin and the city of New Orleans. The study area on the New Orleans Land Bridge is outlined in yellow.

The LB is located between Lakes Pontchartrain and Lake Borgne and is transected by two major tidal passes, Chef Menteur Pass and Rigolets Pass. The two passes connect Lake Pontchartrain and Lake Maurepas with Lake Borgne and the Gulf of Mexico (GOM). Today, two artificial structures dominate the LB. An elevated railroad right of way divides the LB into two parts, an eastern side and a western side. Some three to four hundred meters to the east of the railroad, the Gulf Intercoastal Water Way (GICWW) transects the LB from Chef Menteur Pass to Rigolets Pass. This study encompasses the LB to the east of the GICWW and all of Pearl River Island, which lies north of Rigolets Pass and east of Mud Lake and the West Pearl River.

The five morphologically distinct areas of the LB were chosen for this research project based upon their separation each from the other, easily defined boundaries between them, differences in connectivity to Lake Borgne and the Gulf Intracoastal Waterway (GICWW), and different apparent morphology. The geomorphologic development of the New Orleans Land Bridge (LB) occurred over a period of several thousand years and began at the end of the late Wisconsin Glacial Stage 18 – 20,000 years ago (Darnell, 1962; Sikora and Kjerfve, 1985; Flocks et al., 2009). At the end of the post glacial sea level rise approximately 5000 years ago, sea level reached the approximate level of the current high-stand. The embayment which became Lake Pontchartrain was partially guarded to the south by an extension of the barrier islands which formed during the post glacial eustatic sea level rise off the coast of what is now Mississippi. This barrier complex, today known as the Pine Island Barrier Islands, now forms the Pine Island Trend, an extensive sand unit which extends westward under what is now New Orleans (Saucier, 1963). During the period of sea level rise the north shore of Lake Pontchartrain was marine habitat and as sea levels rose the Gulf of Mexico (GOM) waters filled the basin. The incised Mississippi River valley was gradually in filled with deltaic sediments. The Mississippi River formed several delta lobes as it meandered across the deltaic plain. Since some 4000 years ago, the Mississippi River has formed a series of at least 5 delta lobes. The St. Bernard delta lobe, which formed some 1700 to 2600 years ago, is responsible for the formation of the LB. As sea level rose, the Pine Islands became transgressed and the LB was formed by distributaries of the St. Bernard delta lobe depositing deltaic sediments on top of the Pine Island barrier island chain. Today this area is characterized by abandoned distributaries, natural levees, and inter-levee basins. There has been little if any sedimentation since the Mississippi River adopted its present course and since its flow has been controlled by the construction of artificial levees. In recent years, dams and sills along the Pearl River have decreased sediment input from that source. The geologic history of the LB is now dominated by compaction and subsidence (Kulp et al., 2005).

Ecosystems have been changed by the intrusion of higher salinity water and accompanying changes in vegetation. Tropical and winter storms have also accelerated these changes.

Natural and anthropogenic processes have further shaped the geomorphology of the LB. Compaction and subsidence, and the absence of current sediment input, have caused the LB to experience relative sea level rise as it undergoes a natural progression from a freshwater system to a more brackish system as salinity increases and salt marshes begin to appear (Neill and Deegan, 1986). Many tropical systems over the last several decades have contributed to the loss of land and have helped change the nature of the LB marshes (Fearnley et al., 2009). Anthropogenic effects including the construction of the Gulf Intercoastal Waterway (GICWW) have also contributed to the changes in the LB by allowing greater tidal exchange with the interior of the marsh.

Habitat diversity within the LB has not been well described. Among the different types of fishes and decapods which inhabit the LB are estuarine resident and estuarine dependent species. Estuarine resident animals complete their entire life cycles within the estuary whereas estuarine dependent animals use the estuary for some part of their life cycle. Some stenohaline freshwater and salt water species are occasionally found in the estuary, as are some anadromous and catadromous species. Euryhaline transient species are occasionally encountered. For my present research, I was concerned only with the estuarine resident and estuarine dependent fishes and decapods. The purpose of my research was to determine if these fishes and decapods are in fact uniformly distributed across the five different areas of the LB or if there are differences in species composition among the areas. I then attempted to determine what abiotic factors might be driving compositional differences, when and where they existed.

### Methods:

The area included in this research is that part of the LB east of the GICWW and Mud Lake extending 27.75 km from Chef Menteur Pass to the Louisiana-Mississippi border at the northeast end of Pearl River Island (Figure 1). The study area was further divided into five "Areas" based upon natural boundaries between them (Figure 2). Pearl River Island, located east of Mud Lake and extending from the State border to Rigolets Pass was designated Area 1 (Figure 3). Pearl River Island is 9.75 km long, 1.85 km at the widest, and 0.20 km at the narrowest. It is



Figure 2: The grid lines dividing the study area into 0.65 km2 are shown in red. The five Areas are shown. Area 1: yellow. Area 2: pink, Area 3: blue, Area 4: orange, Area 5: green. Numbers shown in white are the fiducial squares originally used with the random number generator to determine Squares to be sampled on each trip.

18.5 km from Chef Menteur Pass across the Rigolets Pass to the eastern end of Pearl River Island. Area 1 overlies the eastern extent of the Pine Island Barrier Island chain. The West Pearl River enters Mud Lake to the immediate west and on the east side of the island is Lake Borgne. Through the long axis of the island running southwest to northeast is an uninterrupted elevated railroad right of way. Although several natural sloughs and bayous run through the island, several artificial canals penetrate the island to the railroad. The substrate is general soft silt and mud although exposed sand lenses are occasionally encountered. Water levels are greatly influenced by meteorological events and astronomical tides. The sloughs on the west side of the island are generally shallow and cannot be accessed during low water conditions. The east side of the island has four deeper tidal sloughs that allow access deep into the marsh. Low water conditions limit access.



Figure 3: Pearl River Island. Mud Lake and Pearl River are to the north. Lake Borgne is to the south. Yellow markers are sampled sites with sample numbers.

Area 2 comprises the marsh from Rigolets Pass southwestward to Unknown Pass (Figure 4). The eastern 2/3 portion is composed of six shallow large lakes. The western end is marsh with four main tidal sloughs. The substrate is predominantly soft mud and silt to the west and



Figure 4: Area 2 with sampled Sites and sample numbers. Unknown Pass separates this Area from Area 3. Lake Borgne is to the south, GICWW to the west and Rigolets Pass to the northeast.

hard sand to the east. Area 2 is 6.6 km long and 1.4 km at the widest. The large marsh from Unknown Pass to Bayou Platte is Area 3 (Figure 5). This marsh is comprised of inter-levee basins of shallow ponds and swales, and long tidal sloughs and bayous winding through a marsh that is four km wide and six km long. Only one deep long slough enters Area 3 from Lake Borgne, one from Unknown Pass and one from Bayou Platte. Numerous sloughs and bayous enter from the GICWW. Large areas of Area 3 are dry during low water.



Figure 5: Area 3 with sampled Sites and sample numbers. Unknown Pass separates Area 3 from Area 2. Bayou Platte separates Area 3 from Area 4.

Area 4 extends from Bayou Platte to Chef Menteur Pass (Figure 6). To the southeast, there is a natural division extending from the southwestern end of Bayou Platte on a straight line to Chef Menteur Pass. One small shallow lake provides the only connection between Areas 4 and 5 to the southeast of Area 4. This Area is shallow and has the least amount of water movement. In the summer, abundant SAV inhibits boat travel and in the winter shallow water makes large portions inaccessible. The substrate is soft mud and silt and even low velocity wind re-suspends sediments and decreases water clarity.



Figure 6: Area 4 begins at the confluence of Chef Menteur Pass and the GICWW. Only one small shallow lake connects it to Area 5 to the south east. Sampled Sites and samples numbers are shown in yellow.

Area 5 includes all of Alligator Point and is surrounded on its other three sides by Lake Borgne (Figure 7). Several deep sloughs penetrate deep into the interior and most of the lakes and swales are accessible year round. Natural levees and inter-levee basins are prominent in the northeast section. A shallow lake usually less than 0.5 m deep connects to a bayou to the southeast which is the only connection between Areas 4 and 5. Many areas of recent erosion since Hurricane Katrina are encountered oriented in an east-west direction. In the northeast, the stumps of trees and occasional logs are encountered in the shallow ponds.



Figure 7: Area 5, Alligator Point. Sampled Sites and sample labels are in yellow. This Area has multiple openings into Lake Borgne which surrounds it on three sides.

The entire study area was further subdivided into  $0.165 \text{ km}^2$  "Squares" by a series of gridlines overlain on a map of the area (Figure 2). These lines were arbitrarily drawn to run east west and north south. The Squares were the principle divisions for the sampling. Each square can be identified by counting from left to right and top down from the five fiducial markers placed on the map (Figure 2).

To test for possible community differences among these Areas, I sampled each monthly from 1 July 2010 until 30 June 2012. It was initially decided to sample each Square only once during the course of the project. Over time it became obvious that all of the Squares in an Area were not reachable by boat and that they could not all be sampled. For these reasons Areas sampled became arbitrary and some were sampled more than once. Four different squares were chosen for sampling each month in each of the 5 Areas. Initially, these squares were chosen by

use of a random number generator. A pair of random numbers was generated for each area each month. The first four pairs were used to identify the four squares on the map in each Area by counting down and across from each fiducial Square. If the Squares contained accessible marsh that had not been previously sampled, they were sampled that month. If not the next pair was used until a suitable Square was identified. In each square, five Sites were randomly chosen for sampling. At each Site, the net was thrown until two different species were collected or until the net had been thrown five times.

All sampling was done using a 5.33 m welded aluminum flat boat built for this purpose and equipped with a 70 hp two stroke outboard engine. A platform at the bow allowed a stable workspace for deploying the net. This included 3.35 m of open deck space aft of the elevated bow area which provided ample work room for sorting samples. The boat draws 13 cm with the outboard engine raised and is maneuvered in shallow water by means of a 5.8 m graphite push pole from a platform above the engine. During sampling the boat was allowed to drift in place by means of a 2 m steel rod attached to 4 meters of 1.25 cm nylon rope tied to the stern of the boat. Because of shallow water and frequently changing depths from wind, tide, and atmospheric pressure, extreme care was exercised when navigating this area. Both artificial and natural obstructions to navigation including submerged logs, shell banks, pilings, pipelines, abandoned oil field equipment, bulkheads, and storm related debris were common and required not only a thorough knowledge of the area but mandated extreme care when maneuvering.

All sampling was performed using a 1.8 m radius monofilament cast net with a 6 mm bar and 03.67 kg of lead. After the first three months, all sampling was performed by the same operator (see Chapter 2). The net was arbitrarily deployed from the bow of the boat until two different species were collected or until the net had been deployed five times. Each time the net

was deployed, care was taken not to throw it into the same area as the previous throws. Initially, the net was emptied onto the work deck of the boat and all of the animals collected and placed into a plastic one quart bag labeled with the sample number and submerged in an ice-water bath. Each of the four Squares in each Area was assigned a successive sample number and the five Site collections in each square were labeled separately and placed into a bag labeled with the appropriate number of the Square sampled. All specimens were returned to the laboratory for processing. In some cases, large numbers of single species or very large specimens that could be released alive were collected. In the first case, at least 25 randomly selected animals were collected and placed into the sample bag and the rest were counted and released. In the second case, these animals, usually large *Sciaenops ocellatus* (red drum), *Dasyatis sabina* (Atlantic stingray), or *Lepisosteus oculatus* (spotted gar), were weighed and measured on the boat and released alive.

At every Site, the following were recorded on our standard data sheet: GPS coordinates, type of location (pond, bayou, or pond edge), water depth, Secchi disc, an estimate of SAV coverage (0, 25%, 50%, 100%), the presence or absence of grass in the net, and the number and species of any animals released along with their weight, total length and standard length. The number of cast net throws at each Site was also recorded. A separate data sheet was recorded for each Square. At the first Site in each Square, Sample number, date, time sampling was begun in the Square, air temperature, wind direction and speed in mph, cloud cover, and tide stage and an estimate of the type of substrate surface were recorded. At the first, third, and fifth Sites in each Square, water temperature, salinity, dissolved oxygen, per cent oxygen saturation, and specific conductivity, were recorded. A log was kept of each trip including the date, trip number, duration of each trip, and the number of total cast net throws made at each Site.

GPS coordinates were obtained using a Lowrance model Elite 5 combination GPS, moving map display, and depth finder. Water depth was recorded to the nearest inch using a 72 inch (182.88 cm) steel rule, or in water over 6 feet by means of the Lowrance depth finder corrected for the transducer placement beneath the waterline of the boat. Air temperature and wind speed were measured with a factory calibrated Kestrel 4000. A standard Secchi disc with a rope graduated in 0.25 m was used to estimate water clarity and was read by the same person each time. All water measurements were performed with a calibrated Yellow Spring Instrument Professional Plus Model and were made as near to the middle of the water column as possible. The instrument was calibrated on a monthly basis.

The estimate of "Tide" at each Square was an estimate of whether or not water was moving into or out of the square at the time of sampling and whether or not the level of water was closer to high or low tide. Southerly winds and low pressure often produced abnormally high water levels even in the presence of a falling astronomical tide. The reverse was true in the case of a northerly wind and high barometric pressure which often made much of the marsh inaccessible during December and January. Water movement in each Square was a function of astronomical tide, wind, and inverted barometer effect.

Percent submersed aquatic vegetation (SAV) coverage was estimated as being absent, 100%, or more or less than 50%, as was estimated for the immediate area into which the net was thrown. In no case was the net purposely thrown into emergent vegetation. If the net was thrown less than two meters from the emergent vegetation at the edge of a pond, it was recorded as pond edge. A pond was recorded as the site for any large wide body of water shallower that the bayous or sloughs that entered or exited. These areas had little if any current. The term 'bayou' was used for a tidal slough or long body of water which drained a section of marsh.

Secchi disc depth was used as an indirect measurement of turbidity. In several samples, Secchi disc depth was recorded as clear to the bottom (CTB). In these cases, Secchi disc depth was estimated to be the greatest depth recorded in a Square or the greatest Secchi disc depth recorded in that Square, whichever was greatest. Unfortunately, estimates of substrate were difficult and were based upon the ease with which the steel measuring stick could be pushed into the substrate when the depth was measured. This measurement was highly subjective and on occasion was found to vary greatly over small distances.

Immediately after returning from a trip, the samples were placed in a freezer. All of the data were transferred to a computer spreadsheet and the GPS location of each sample was plotted on Google Maps Pro to ensure that it was made at the proper location. Within 48 to 72 hours, samples were thawed in the lab. All animals were identified to the species level, and 25 randomly selected of each species in each sample were weighed and SL and TL were measured and recorded. Any remaining animals were counted and weighed together by species. Individual weights were measured on a digital scale to the nearest 1/100 g and standard lengths (SL) to the nearest mm.

The five Sites in each Square sampled each month were treated as replicates and for analysis the abiotic data were averaged and the biotic data were pooled and summed. The summed biotic data were then divided for the number of cast net throws. Using the method described in Chapter 2, it was determined that the mean area covered by the operators cast net throws was  $3.9 \text{ m}^2$ . In this manner, the density of each species in each Square per 4 m<sup>2</sup> was calculated.

Data analysis was performed using Primer 6 software (Clarke and Gorley, 2006). The initial analysis was a SIMPER (Similarity Percentage) analysis to see what species were driving the community. A resemblance matrix was then created and a Multidimensional Scaling plot of the samples was made. One-Way Analysis of Similarity (ANOSIM) using Area or Month as factors were performed to see if either or both were predictors of the community. The same analysis was then performed on all of the Squares during each of the 24 months of collection. However, since the analysis was carried out for each month, the factor tested for significance was Area.

Abiotic (environmental data) included: Area, tide, wind direction, wind velocity, distance from the nearest large water body, water temperature, salinity, oxygen saturation, Secchi disc depth, water depth, and whether or not SAV was present in the net. The abiotic data were square root transformed and then normalized. A BEST analysis was performed to determine which abiotic variables expressed a pattern in multidimensional space that best matched the pattern of community data in the Squares. This analysis was first performed on all of the combined data and then on the sample data for each of the 11 of 24 months individually in which Area was a significant factor. A Euclidean matrix of abiotic factors was created for each month and pairwise comparisons of the Squares were performed for each month with Area as the factor. This was done to ascertain whether or not the Areas were significantly different within each month. Select single species average densities were calculated for each of the 24 months of data collection and for each the five Areas across all 24 months. One way ANOSIMs were run with month and Area as factors to assess their significance.

### **Results:**

Between 15 July 2010 and 29 June 2012, 424 Squares were sampled at 2120 Sites during 75 trips to the research areas. In excess of 670 hours were spent on the boat and the cast net was thrown over 4370 times resulting in the collection of 58,869 fishes and decapods comprising 58 different species of fishes in 26 families and 3 species of decapods in 2 families (Table 1). Mud crabs, *Rhithropanopeus* spp., and grass shrimp, *Palaemonetes* spp., were highly abundant and were not counted. Gulf menhaden, *Brevoortia patronus*, was the most common fish collected (38,849) and accounted for 66% of all of the animals collected (Table 1).

Table 1: List of all families and species and numbers of each collected on the New Orleans Land Bridge from July 2010 through June 2012 in 2120 collections with a 1.8 m, 6 mm bar, monofilament cast net.

Family	Species	Number Collected
Fishes		
Dasyatidae	Dasyatis sabina	6
Lepisosteidae	Atractosteus spatula	1
	Lepisosteus oculatus	18
Elopidae	Elops saurus	33
Megalopidae	Megalops atlanticus	1
Engraulidae	Anchoa hepsetus	14
	Anchoa mitchilli	4739
Clupeidae	Alosa chrysochloris	5
	Brevoortia patronus	38849
	Dorosoma cepedianum	13
	Dorosoma petenense	433
Ariidae	Ariopsis felis	14
	Bagre marinus	1
Batrachoididae	Opsanus beta	1
Mugilidae	Mugil cephalus	1456
	Mugil curema	62

Table 1: (Continued)

Family	Species	Number collected
Atherinopsidae	Membras martinica	196
	Menidia beryllina	1107
Belonidae	Strongylura marina	3
Fundulidae	Adinia xenica	41
	Fundulus grandis	767
	Fundulus jenkinsi	32
	Fundulus pulvereus	20
	Fundulus similis	10
	Lucania parva	1477
Poeciliidae	Poecilia latipinna	60
	Gambusia affinis	7
Cypinodontidae	Cyprinodon variegatus	1194
Syngnathidae	Syngnathus louisianae	11
	Syngnathus scovelli	117
Triglidae	Prionatus tribulus	4
Centrarchidae	Lepomis microlophus	1
	Lepomis miniatus	1
	Micropterus salmoides	1
Carangidae	Caranx hippos	1
	Oligoplites saurus	17
Sparidae	Archosargus probatocephali	<i>u</i> s 2
	Lagodon rhomboides	97
Sciaenidae	Bairdiella chrysoura	71
	Cynoscion arenarius	109
	Cynoscion nebulosus	113
	Leiostomus xanthurus	3462
	Micropogonias undulatus	338
	Pogonias cromis	8
	Sciaenops ocellatus	146
Gobiesocidae	Gobiesox strumosus	2
Gobiidae	Ctenogobius boleosoma	2
	Ctenogobius shufeldti	25
	Gobionellus oceanicus	14
	Gobiosoma bosc	45
	Gobiosoma robustum	2
	Microgobius gulosus	66

Table 1: (continued)				
	Family	Species	Number collected	
	Paralichthyidae	Citharichthys spilopterus	65	
		Etropus crossotus	2	
		Paralichthys lethostigma	17	
	Achiridae	Trinectes maculatus	1	
	Cynoglossidae	Symphurus plagiusa	7	
	Tetraodontidae	Sphoeroides parvus	12	
	Decapods			
	Portunidae	Callinectes sapidus	712	
	Penaeidae	Farfantepenaeus aztecus	928	
		Litopenaeus setiferus	1275	

An MDS plot of the 424 Squares did not show any obvious clustering (Figure 8). A twoway crossed ANOSIM revealed a significant difference among Area groups across all Month groups (R = 0.187, p = 0.001). The difference between Month groups across all Areas was significant (R = 0.484, p = 0.001). A preliminary analysis of the Squares with Areas and



Figure 8: Multidimensional Scaling plot of 424 Squares as a function of the 5 Land Bridge Areas. No obvious clustering is appreciated.

months combined into Seasons of three and six months each, as factors did produce statistically significant results but the R values were all less than 0.200.

A SIMPER analysis of the Squares revealed that the within Area samples had low indices of Similarity indicating a high degree of within Area variability among Squares (Table 2). *Brevoortia patronus, Anchoa mitchilli* (bay anchovy), *Lucania parva* (rainwater killifish) and *Leiostomus xanthurus* (spot) were among the most abundant species collected and made the greatest contribution to community structure in each of the Areas. However, the Areas were all different in terms of the species ranking and relative contributions across all months. Areas 1 and 2 were similar for the first 4 species in the community, but the average abundance was different between the two. Average dissimilarity indices between communities were determined in pair-wise comparison of Areas in each of the 11 months for which Area was found to be a significant factor (p < 0.05). In each of these comparisons across all 11 months, average dissimilarity was greater than 55.

A preliminary BEST analysis was performed using 10 variables (Clarke and Gorley, 2006). These included tide, wind direction, wind velocity, distance from Square to the outside border of the marsh, salinity, dissolved oxygen, Secchi disc depth, water depth, and presence of SAV in the cast net. Area and month were omitted as they were already shown to be significant. The abiotic variables shown to be most predictive of the community structure were water temperature and Secchi disc depth (r = 0.221, p = 0.001). Because water temperature was possibly a proxy for month, this variable was omitted and the analysis was run again. Dissolved oxygen and Secchi disc depth had the highest sample statistic of r = 0.154 (p = 0.01).
Table 2: Similarity indices should reflect within Area similarities between samples in Squares. The species driving the Community structure in each Area and the absolute and relative contribution is given in the table. The low similarity indices indicate that the within Area variability among Squares is high.

		Area 1			
	Average sir	nilarity: 3	0.99		
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
B. patronus	23.83	13.68	0.6	44.15	44.15
A. mitchilli	1.31	6.65	0.61	21.45	65.6
L. setiferus	0.74	3.13	0.37	10.11	75.71
L. xanthurus	1.04	2.03	0.4	6.55	82.26
M. beryllina	0.21	1.74	0.26	5.63	87.88
M. cephalus	0.4	1.49	0.39	4.8	92.68
		Area 2			
	Average sin	nilarity: 3	3.20		

	0	2			
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
B. patronus	9.66	10.55	0.59	31.77	31.77
A. mitchilli	1.42	5.86	0.61	17.64	49.41
L. setiferus	0.62	4.82	0.41	14.5	63.92
L. xanthurus	1.25	3.17	0.55	9.56	73.48
F. aztecus	0.36	2.68	0.51	8.06	81.54
M. cephalus	0.38	1.72	0.47	5.19	86.73
M. beryllina	0.25	1	0.37	3.01	89.73
C. sapidus	0.13	0.78	0.39	2.34	92.08

## Area 3

Average similarity: 28.72								
Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%				
1.43	6.01	0.63	20.95	20.95				
15.24	5.82	0.43	20.27	41.22				
1.39	4.61	0.52	16.05	57.27				
0.55	2.01	0.37	7	64.26				
0.35	1.93	0.5	6.73	70.99				
0.39	1.76	0.41	6.12	77.11				
0.45	1.73	0.34	6.04	83.15				
0.38	1.57	0.37	5.46	88.6				
0.18	1.32	0.31	4.61	93.21				
	Average sir Av.Abund 1.43 15.24 1.39 0.55 0.35 0.39 0.45 0.38 0.18	Average similarity: 22 Av.Abund Av.Sim 1.43 6.01 15.24 5.82 1.39 4.61 0.55 2.01 0.35 1.93 0.39 1.76 0.45 1.73 0.38 1.57 0.18 1.32	$\begin{array}{llllllllllllllllllllllllllllllllllll$	Average similarity: 28.72Av.AbundAv.SimSim/SDContrib%1.436.010.6320.9515.245.820.4320.271.394.610.5216.050.552.010.3770.351.930.56.730.391.760.416.120.451.730.346.040.381.570.375.460.181.320.314.61				

# Table 2: (Continued)

L. xanthurus

L. setiferus

F. aztecus

M. cephalus

C. sapidus

L. parva

Area 4						
	Average sir	nilarity: 2	8.34			
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	
B. patronus	11.11	8.34	0.5	29.43	29.43	
L. parva	1.29	6.79	0.55	23.96	53.38	
L. xanthurus	0.96	2.81	0.48	9.92	63.31	
A. mitchilli	0.69	2.75	0.42	9.71	73.01	
M. cephalus	0.34	1.91	0.39	6.76	79.77	
C. sapidus	0.25	1.42	0.47	5.02	84.79	
M. beryllina	0.39	1.23	0.24	4.35	89.14	
L. setiferus	0.13	0.82	0.28	2.89	92.03	
		Area 5				
	Average sir	nilarity: 34	4.61			
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	
B. patronus	11.19	9.04	0.49	26.1	26.1	
A. mitchilli	1.83	7.38	0.61	21.31	47.41	

3.78

3.56

3.03

2.74

1.27

1

0.55

0.37

0.33

0.36

0.52

0.26

1.87

0.31

0.56

0.2

0.36

0.19

58.33

68.63

77.39

85.29

88.96

91.84

10.92

10.3

8.76

7.91

3.67

2.88

Next, I analyzed the Squares from each of the 24 months of collections. An ANOSIM
was performed on the collections of each month to test for significance of AREA (Table 3). A
significant difference among the communities in the Areas was found in 11 of 24 months ( $p < p$
0.05). Five of six months beginning in June 2011 exhibited a significant difference in
community structure among the Areas. This pattern was repeated in only 3 pairs of the same
months out of 12 pairs.

Table 3: An Analysis of Similarity of the Squares sampled for each month was run with AREA
as the factor. Those months for which AREA was significant are shown in bold. The sample
statistic (R) and significance (p) are show for each of the 24 months. AREA was a significant
factor in the community structure in 11 of 24 months.

	Year 1		Year 2	
	<u>R</u>	<u>p</u>	<u>R</u>	<u>p</u>
Jul	0.158	0.06	0.294	0.014
August	0.152	0.076	0.273	0.012
September	0.262	0.011	0.118	0.097
October	0.34	0.008	0.272	0.012
November	0.292	0.031	0.433	0.012
December	-0.177	0.886	-0.025	0.596
January	0.557	0.001	0.186	0.132
February	0.217	0.052	0.099	0.168
March	0.007	0.421	0.232	0.016
April	0.059	0.223	0.08	0.175
May	0.001	0.424	0.084	0.185
June	0.229	0.013	0.378	0.003

The eleven months which exhibited a statistical difference in the Areas were then tested to see which abiotic factors were the best predictors of the community by means of the BEST analysis (Table 4). During the first year of data collection, September (p = 0.011), October (p =0.008), November (p = 0.031), and January (p = 0.001) all showed significant differences among the Areas. In September, wind direction, salinity, and the presence of SAV in the net were the most important factors and were unlikely to have occurred by chance (p = 0.01). Water temperature, Secchi depth, water depth, and SAV in the net were the most important factors in October, in spite of the fact that the R value was high (R = 0.455) this correlation had a 12% probability of occurring by chance alone (p = 0.12). Wind direction was again important in November and water temperature and water depth were important in October, November, and January and Secchi disc depth was important in January. All three of these months had R values

of 0.45 or greater, but may have happened by chance alone (p > 0.05).

Table 4: The abiotic factors best correlating with the Community Structure during each individual month where Area was statistically significant (p < 0.05) are shown. Spearman's rank correlation (Rho) and p values for each month are shown.

	Abiotic Factors Driving Community		
Year 1Month	Factors	<u>Rho</u>	p
September	wind direction, salinity, SAV in net	0.418	0.01
October	water temperature, Secchi disc depth, water	0.455	0.12
	depth, tide		
November	wind direction, water temperature, water depth	0.65	0.09
January	wind speed, water temperature, Secchi	0.45	0.12
	disc depth, water depth		
June	wind direction, wind speed, SAV in net	0.389	0.03
Year 2 Month			
	distance from marsh entrance, salinity, Secchi disc		
July	depth	0.459	0.01
	water depth, SAV in net		
August	tide, distance to marsh entrance, salinity, Secchi	0.491	0.01
	disc depth, water depth		
October	wind direction, wind speed, distance to marsh	0.42	0.01
	entrance, salinity, dissolved oxygen		
November	tide, dissolved oxygen, Secchi disc depth,	0.368	0.22
	water depth, SAV in net		
March	wind speed	0.331	0.16
June	water temperature, Secchi disc depth,	0.375	0.04
	SAV in net		

June Year 1 (p = 0.03), and July (p = 0.01), and August (p = 0.01) all had Rho values greater than 0.389 and all reached statistical significance (less than 5% chance of happening by chance alone). Wind direction and speed were important factors in June as well as presence or absence of SAV in the net, which was a significant factor in July as well. In July and August, distance from entrance to the marsh became significant as well as Secchi disc depth, water depth, and salinity. The pattern changed in the fall 2011. The factors were significant in October (p = 0.01) but not in November (p = 0.22). The only factor repeated in both months was dissolved oxygen content. Wind direction and speed, distance from the marsh entrance, and salinity were significant factors in October (p = 0.01). Tide, Secchi disc depth, water depth, and presence of SAV were the most important factors in November but were not significant.

March and June 2012 also showed significant differences among the Areas. The most important abiotic factor in March was wind speed but it was not significant (p = 0.16). In June, water temperature, Secchi disc depth, and presence of SAV were significant (p = 0.04).

Table 5: Pairwise tests of abiotic differences between Areas for July Year 2. Abiotic factors include tide, wind direction and speed, distance from entrance into marsh, water temperature, salinity, dissolved oxygen, Secchi disc depth, water depth, and presence of SAV in the net. All Areas are significantly different from each other except Areas 1 and 5, and 4 and 1. This pattern is repeated in each of the other 10 months for which Area is significant.

		Year 2	Pairwise Tests		
			Possible	Actual	Number>=
Areas	<u>R</u>	<u>p</u>	Permutations	Permutations <b>Permutations</b>	Observed
3,2	0.771	0.029	35	35	1
3,4	0.833	0.029	35	35	1
3,5	0.948	0.029	35	35	1
3,1	0.76	0.029	35	35	1
2,4	0.198	0.029	35	35	7
2,5	0.635	0.029	35	35	1
2,1	0.573	0.029	35	35	1
4,5	0.552	0.029	35	35	1
4,1	0.448	0.057	35	35	2
5,1	-0.021	0.571	35	35	20

Nevertheless, pair-wise comparisons for each month revealed significant abiotic

differences among the Areas in the majority of months (68/88) for which Area was found to be a significant factor in the community structure (p < 0.05; Table 5). When these comparisons were compared to dissimilarity analyses and pair-wise Area comparisons for the same months, no pattern could be discerned implying that abiotic factors as a whole were not the sole determinant of community structure (Table 6).

Table 6: Dissimilarity comparisons of Areas for July Year 2. The average abundance of species is compared per 4  $m^2$ . The dissimilarity index is an indication of how different communities of the two Areas are from each other. This pattern is repeated in each of the other 10 months for which Area is significant.

Areas 3 & 2						
Average dissimil	arity = 68.51					
-	Area 3	Area 2				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
B. patronus	2.5	5.3	29.68	1.25	43.32	43.32
M. cephalus	2.47	0.49	13.81	0.64	20.16	63.48
L. xanthurus	1.56	1.22	8.33	0.98	12.16	75.64
A. mitchilli	0.42	0.75	4.65	0.93	6.78	82.42
L. parva	0.56	0	4.44	0.81	6.49	88.91
F. aztecus	0.68	0.54	2.99	1.32	4.37	93.28
Areas 3 & 4						
Average dissimil	arity = 78.40	)				
	Area 3	Area 4				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
B. patronus	2.5	20.72	45.52	1.4	58.06	58.06
A. mitchilli	0.42	2.4	9.75	0.89	12.43	70.49
M. cephalus	2.47	0.24	9.59	0.54	12.24	82.73
L. xanthurus	1.56	0.45	4.82	0.86	6.15	88.88
Table 6:						
(Continued)						
L. parva	0.56	0.13	2.85	0.63	3.63	92.51
Areas 2 & 4						
Average dissimil	arity = 66.19	)				

	Area 2	Area 4				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Table 6:						
(Continued)						
B. patronus	5.3	20.72	46.2	1.56	69.8	69.8
A. mitchilli	0.75	2.4	7.83	0.84	11.83	81.63
L. xanthurus	1.22	0.45	4.72	0.61	7.14	88.76
M. cephalus	0.49	0.24	1.93	0.88	2.92	91.69
Areas 3 & 5						
Average dissimi	ilarity = 92.44	ł				
	Area 3	Area 5				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
A. mitchilli	0.42	21.16	51.84	2.2	56.08	56.08
B. patronus	2.5	1.28	9.47	0.66	10.25	66.33
M. cephalus	2.47	0.21	8.64	0.54	9.35	75.67
L. xanthurus	1.56	0.28	5.18	1.05	5.6	81.28
C. arenarius	0	0.63	4.46	0.56	4.83	86.11
F. aztecus	0.68	0	2.85	1.04	3.09	89.2
L. parva	0.56	0	2.58	0.7	2.79	91.99
Areas 2 & 5						
Average dissimi	larity = 86.60	)				
	Area 2	Area 5				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
A. mitchilli	0.75	21.16	50.18	2.1	57.95	57.95
B. patronus	5.3	1.28	17.28	0.95	19.96	77.91
C. arenarius	0.04	0.63	4.53	0.56	5.23	83.14
L. xanthurus	1.22	0.28	4.47	0.67	5.17	88.3
F. aztecus	0.54	0	2.35	1.09	2.71	91.02
Areas 4 & 5						
Average dissimi	larity = 82.45	5				
	Area 4	Area 5				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
A. mitchilli	2.4	21.16	36.06	1.49	43.73	43.73
B. patronus	20.72	1.28	34.96	1.23	42.41	86.14
C. arenarius	0	0.63	3.06	0.46	3.71	89.85
M. martinica	0	0.93	1.8	0.85	2.18	92.02
Areas 3 & 1						
Average dissimi	larity = 89.67	7				

Table 6: (Conti	nued)					
	Area 3	Area 1				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
B. patronus	2.5	10	27.11	0.82	30.24	30.24
Table 6:						
(Continued)						
M. cephalus	2.47	0.16	14.25	0.57	15.89	46.13
M. martinica	0	3.14	13.03	0.67	14.53	60.66
L. xanthurus	1.56	0.16	10.68	1.04	11.92	72.57
A. mitchilli	0.42	1.53	6.91	1.25	7.71	80.28
F. aztecus	0.68	0	5.82	0.9	6.49	86.77
L. parva	0.56	0	5.27	0.71	5.88	92.65
Areas 2 & 1						
Average dissim	ilarity = 82.74	1				
	Area 2	Area 1				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
B. patronus	5.3	10	42.09	1.47	50.88	50.88
M. martinica	0	3.14	13.18	0.67	15.93	66.8
L. xanthurus	1.22	0.16	9.37	0.64	11.33	78.13
A. mitchilli	0.75	1.53	6.71	1.26	8.11	86.24
F. aztecus	0.54	0	4.68	1.04	5.66	91.9
Areas 4 & 1						
Average dissim	nilarity = 80.11	l				
	Area 4	Area 1				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
B. patronus	20.72	10	50.19	1.49	62.65	62.65
A. mitchilli	2.4	1.53	10.14	0.89	12.66	75.31
M. martinica	0	3.14	9.42	0.59	11.76	87.07
L. xanthurus	0.45	0.16	2.1	0.59	2.62	89.69
F. aztecus	0.19	0	2.09	0.46	2.61	92.3
Areas 5 & 1						
Average dissim	ilarity = 86.04	1				
	Area 5	Area 1				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
A. mitchilli	21.16	1.53	48.55	1.68	56.43	56.43
B. patronus	1.28	10	16.73	0.67	19.45	75.88
M. martinica	0.93	3.14	9.51	0.73	11.05	86.93
C. arenarius	0.63	0.02	5.18	0.49	6.02	92.95

As expected, the highly significant difference between Squares when analyzed across the months was not surprising. I could only catch what fishes were available and these varied with the seasons. The difference among areas when compared across all months was significant (p = .001) but the global R value was low. Even when analyzed as Squares, the within Square variability was almost as high as the between Square variability. However, it is obvious that community structure is not the same among Areas when analyzed across all months.

Of all the species, *B. patronus* accounted for 66% of species collected. While this species was collected throughout the year in every Area, it was most commonly found in Area 1 where it accounted for over 44% of all of the animals collected and had an average density of  $23.83/4 \text{ m}^2$ . In Area 2, *B. patronus* made up 31% of the total collection with a density of  $9.66/4 \text{ m}^2$ . In every Area except Area 3, *B. patronus* was the most common animal collected. In Area 5, *A. mitchilli* had the greatest density ( $1.83/4 \text{ m}^2$ ) but only accounted for 21.3% of the animals collected and had an average density of  $0.69/4 \text{ m}^2$ . Both *B. patronus* and *A. mitchilli* were the most common fishes captured in all Areas except Area 4 where *L. parva* was second to *B. patronus*.

The greatest density of *L. xanthurus* was in Area 5 and the lowest in Area 4. *C. sapidus* (blue crab) had the greatest density in Area 3 and the lowest in Area 1. *L. setiferus* (white shrimp) had the highest concentrations in Areas 1 and 2 and the lowest in Area 4. *M. cephalus* (striped mullet) had the highest density in Area 3 and decreased in the Areas on either side.

I attempted to find out what months were accounting for the measured significant differences. In 11 of 24 months, communities among the Areas not only showed global R values greater than 0.23 but were significantly different in species composition (all comparisons: p <

0.05). These months were clustered for the most part from June through November. It is during this period that the highest numbers of larvae and small juveniles of estuarine dependent species were expected to be present in the marsh. In June of both years, *L. setiferus* was appearing in Area 1 and *F. aztecus* (brown shrimp) was still present towards the west end of the LB. By July, *F. aztecus* was appearing in all Areas, but only in the first year in Area 1. *C. arenareus* (sand seatrout) first began to appear in Area 5 in July with very few captured in Areas 1 and 2.

*M. martinica* (rough silverside) were collected in July and August in Area 1(153) and Area 5 (19). While a few were collected inside the entrance of tidal sloughs, most were caught on the shoreline of Lake Borgne. No *M. martinica* were ever collected in the marsh. Over 1100 *M. beryllina* (inland silverside) were captured in every Area although they had the highest density in Areas 3 and 4.

The community structure was shown to be different across all of the Areas when assessed across all months combined and statistically significant in 11 of 24 months. When the abiotic data were analyzed, no consistent correlation with community structure could be identified. BEST analyses on a monthly basis identified several factors which were significant, but not in all months. This analysis was run to return the most significant factors (up to 5) with the highest correlation value. The results were then tested for significance (p < 0.05) using a random iteration approach. Wind direction was identified as an important factor in four months but was only significant in three of these. Wind speed was significant in only two of the months where it was a factor. Secchi disc depth was significant in three of six months and water depth in only two of six months. The presence of SAV in the cast net was a significant factor in

four of six months. Distance from the marsh entrance was a significant factor in three of three months. Tide was never recognized as a significant factor.

On a species basis, Area was not a significant factor in the distribution of *B. patronus* (R = 0.001, p = 0.334). Month was significant (p = 0.001) but the global R was low (0.184). Densities of *B. patronus* in November and December (R= 0.764, p = 0.001) and March and April (R= 0.634, p = 0.001) were significantly different. This difference decreased steadily throughout the year and did not become significant again until February, suggesting *B. patronus* begins to appear in the marsh in March. The highest density of *B. patronus* occurred in March (80.8  $/4m^2$ ), April (62.4  $/4m^2$ ) and May (28.7  $/4m^2$ ) Year 1. The lowest occurred in November (.019  $/4m^2$ ) and December (0  $/4m^2$ ) in Year 1.

Area was even less significant for *A. mitchilli* (R = 0.004, p = 0.152). Month was significant (p = 0.001) but with a low R (0.102). Densities appeared to vary throughout the year with no pattern. It was highest in February ( $3.1 / 4m^2$ ) and November ( $1.6 / 4m^2$ ) in Year 1 and July ( $6.1/4m^2$ ), October ( $3.64 / 4m^2$ ), and January ( $2.73/4m^2$ ) in Year 2.

Estuarine dependent *L. xanthurus* showed no preference for Area (R = -0.004, p = 0.797). Month was positively correlated with density (R = 0.224, p = 0.001). Density was greatest in March (10.955 /4m<sup>2</sup>), April (4.29 /4m<sup>2</sup>) and May (2.15 /4m<sup>2</sup>) in Year 1 and March (2.25 /4m<sup>2</sup>), April (0.311 /4m<sup>2</sup>), and May (0.33 /4m<sup>2</sup>) in Year 2. Density during June (1.5 /4m<sup>2</sup>) Year 1 and June (0.168 /4m<sup>2</sup>) were noted to be different. A BEST analysis showed that water temperature and depth were significantly correlated with the number of *L. xanthurus* collected (R= 0.269, p = 0.001). This species exhibited a preference for water temperatures from 20 C to 34 C. In this case, water temperature may be a surrogate for Month. The majority of this species were collected in water between 25 and 110 cm deep.

Densities of *M. undulatus* (Atlantic croaker) peaked in February  $(0.672 / 4m^2)$  Year 1 and March  $(0.728 / 4m^2)$  in Year 2. Month was a significant factor (R = .215, p = 0.001) and supported the seasonality of this species. Area was not statistically significant (R = -0.002, p= 0.546). However, the greatest density was in Area 4 (0.255 / 4m<sup>2</sup>).

An ANOSIM of *L. parva* (global R = 0.134, p = 0.001) showed the most significant difference between Areas 1 and 4 (R= 0.359, p = 0.001). Areas 2 and 4 were significantly different (R = 0.311, p = 0.001). The average density of *L. parva* was 1.3 /4m<sup>2</sup> in Area 4 but .0159 /4m<sup>2</sup> in Area 1 and .088 /4m<sup>2</sup> in Area 2. Month was much less of a significant factor in the distribution of *L. parva* (R= 0.067, p = 0.001).

The fundulids (*Fundulus grandis* [Gulf killifish], *F. jenkinsi* [saltmarsh topminnow], *F. pulvereus* [bayou killifish], and *F. similis* [longnose killifish]) were all considered together to negate any issues that might be introduced because of misidentification. An ANOSIM run with Areas as a factor showed that Area was not a significant predictor (global R = -0.004, p = 0.834). Using months as a factor, global R = 0.051 (p = 0.001). The greatest average densities for all 4 species were in Area 5. The highest average monthly densities were in November of Year 1.

The penaeid shrimps showed a similar pattern in that for *L. setiferus*, Area was not a significant factor (R= 0.008, p = 0.045) in terms of numbers collected, but average density was greatest in Area 1 ( $0.74 / 4m^2$ ). Density appeared to decrease towards the West and but increased again in Area 5 ( $0.31 / 4m^2$ ). Month was significant for this species also (R= 0.323, p = 0.001). Density increased beginning in June, peaked in October ( $3.4 / 4m^2$  Year 1 and  $2.3 / 4m^2$  Year 2)

and then decreased through November. *F. aztecus* began appearing in April in Year 1 and February in Year 2. Greatest densities were in May  $(1.88 / 4m^2)$  in Year 1 and April  $(2.48 / 4m^2)$  in Year 2. For *F. aztecus*, the greatest densities were recorded in Area 5  $(0.561 / 4m^2)$  followed by Areas 3, 2, 1, and 4.

Month was a significant factor (R = 0.409, p = 0.001) when an ANOSIM was run on the abiotic factors as Squares. Likewise, Area was a significant factor (p = 0.001) but with a much lower R (0.092), suggesting that the differences of Squares within Areas was almost as great as the differences among Areas. The implication is that Area is not a determinant of the abiotic factors within Squares.

When Areas were tested for differences in abiotic factors, they were found to be significantly different from each other during the months in which Area was a significant factor in community structure (Table 5). There does not appear to be any correlation between the abiotic differences and the dissimilarity among Areas. Some other as yet unidentified factors must be driving community structure other than the abiotic factors examined.

### **Discussion:**

It is generally accepted that abiotic variables have an effect on the structure of estuarine communities (Jones et al., 2002; Cowan et al., 2013). Many different microhabitat types exist in estuarine marshes. These habitats may change frequently as a result of flooding events, changes in temperature, salinity, depth, turbidity, and vegetation. Many species of estuarine dependent nekton use these marshes as nursery areas (Boesch and Turner, 1984; Cowan et al., 2013). Fishes and decapods undoubtedly move about within the estuary to find the most suitable

habitats (Cowan et al., 2013). Here, I assessed the composition of fish and decapod communities across a broad stretch of estuarine marsh. The five Areas were adjacent to each other but were different in structure in terms of depth, marsh coverage, connections to Lake Borgne and tidal passes. However, they all contained similar microhabitats. My hypothesis was that community structure across the Areas would be dissimilar in that species would not be evenly distributed across the 5 Areas. This indeed turned out to be the case. It was anticipated that the differences could be accounted for by variability of abiotic factors. This did not turn out to be the case. No consistent abiotic factors were identified which could predict community structure across all months. For individual months, certain variables were predictive but only for those months.

Several abiotic variables such as salinity have been shown to have an effect on the distribution of salt marsh species (Martino and Able, 2003; Martin et al., 2009). The apparent lack of consistent predictive variables in my data set can be explained in several ways. The most obvious explanation is that community structure can be explained by some as yet untested variables. This may indeed be the case. Several other factors have been suggested to have an effect on community structure such as frequency and depth of flooding events (Rozas and Reed, 1993; Minello et al., 2012). Community structure and density of animals has been shown to vary with proximity to marsh edge and the marsh surface (Minello and Rozas, 2002). Trophic effects and the presence of food also affect distribution of fishes and decapods across an estuarine marsh (Baltz et al., 1998; Chesney et al., 2000). These factors are most active at the microhabitat level. My data suggest that a different set of factors are operating at the Area level.

Three important events happened during the time period covered by this research that may have had an impact on the results. The Deepwater Horizon oil spill in 2010 carried oil into the Mississippi Sound and Chandeleur Sound and possibly into the lower reaches of Lake

Borgne. Adult fishes can swim away from oil, but eggs, larvae, and juvenile nekton are much more susceptible (Short et al., 2003; Roth and Baltz, 2009). The effects of PAHs (poly aromatic hydrocarbons) have long term effects on nekton reproduction and populations (Rolland, 2000). Furthermore, oil and weathered oil may have long term effects on marsh grasses and thus effect microhabitats (Culbertson et al., 2008). The August 2011 West Pearl River chemical spill from the Temple-Inland paper mill in Bogalusa caused massive fish kills on the lower West Pearl River. The West Pearl empties into Mud Lake on the northwest side of Pearl River Island (Area 1). Few nektonic organisms were collected in that Area when it was sampled that month. It is unknown how long the effects of the spill persisted in the Area or how much of Area 1 was effected.

The Bonnet Carre Spillway opening in May 2011 also had an unknown effect. Higher waters and lower salinity persisted in the LB marsh for a prolonged period of time during and following this event (Georgiou et al., 2010). Data collection for this project only covered a period of two years making it difficult if not impossible to identify both short and long term effects from any of these events. One short term effect was a lowering of the salinity on the LB for several months during the period of larval influx and growth. Area was a significant factor (p < 0.05) during five of the six months from June through November after the Spillway opening. Secchi disc depth and salinity were significantly correlated with community structure each in three of the six months. The presence of SAV in the net was correlated in two of the months and the amount of SAV in the marsh may be a consequence of decreased salinity. It is unclear whether or not the sediment plume which entered Lake Pontchartrain affected Secchi disc depth on the LB.

Transport mechanisms of larval fish into the estuary may have an effect on the size of year-classes and numbers of fishes and decapods that are recruited to marsh habitats (Boehlert and Mundy, 1988; Able and Fahay, 2010). Stochastic meteorological events such as storms may also affect advection and transport and year-class sizes of different species (Able and Fahay, 2010). The topographic structure of salt marshes, adjacent shorelines, and tidal creeks affect salt marsh currents and may also play a role in transport and distribution of larvae (Torres and Styles, 2007). These effects may be magnified by changes in water depth which influence topography and bathymetry. Stochastic meteorological events are not limited to storms. Variations in the onset of seasonal warming may have effects on spawning activity and timing of larval ingress into the marsh. Additionally, extremes of temperature variation affect reproduction, egg maturation, and larval growth (Pankhurst and Munday, 2011).

Again, it is noteworthy that Area was a significant factor (p < 0.05) in the months immediately following the Spillway opening when larval nekton would have been entering the marshes of the LB. The volume of river water that was passing through Lake Pontchartrain and subsequently through the two natural passes (the Rigolets and Chef Menteur Pass) was substantial. To some extent during this period, the estuary may have temporarily become ebb tidal dominated. The Bonnet Carre Spillway is designed to carry 7,100 m<sup>3</sup>/s with all 350 bays open. During the peak of this event 330 bays were open carrying more than 94% of the design volume (USACE, 2011). The mean daily tidal prism of Lake Pontchartrain is 1.56 x 10<sup>8</sup> m<sup>3</sup> (Sikora and Kjerfve, 1985). The calculated maximum flow through the Spillway would add 6.13 x 10<sup>8</sup>m<sup>3</sup> per day, or 390% of the daily tidal prism. The mean annual discharge of all rivers into Lake Pontchartrain is in the range of 200 m<sup>3</sup>/s or less than 5% of the discharge from the Spillway (Sikora and Kjerfve, 1985). The extra volume of water exiting from Lake Pontchartrain may be enough to disrupt normal currents and tidal exchange through the natural passes and, in Lake Borgne, affect advection of larval nekton into the marsh at a time when they are most vulnerable to this effect. The effects of variation in Pearl River discharge may also influence currents and advection into the marsh. Although only a small fraction of the Pearl River discharge enters Lake Pontchartrain, variations in discharge into Lake Borgne may have local as well as distant effects. Heavy rainfall events may have a similar effect on subtidal currents.

It is possible that the sampling gear used was biased towards certain nekton (see Chapter 2). Although we saw large numbers of *S. marina* (Atlantic needlefish) and *L. oculatus*, we collected very few. It is impossible to know how much of an effect gear avoidance had on our collections. Where Secchi disc depth was significant (p < 0.05) gear avoidance may have been a factor. Clear water may make smaller nekton more susceptible to predation and result in lower density. Clear water may also cause small nekton to seek refugia in SAV or in very shallow water (Paterson and Whitfield, 2000). In any of these events, my collections would not have reflected true density.

I did not include fish size or biomass in this analysis. The inclusion of nekton size may be a factor in distribution. Larvae and smaller juveniles may exhibit a much different distribution than large juveniles. The absence of size data may have prevented site (pond or bayou) from being a significant abiotic variable in the initial BEST analyses. Larger juveniles are able to migrate within the marsh over longer distances to find ideal microhabitats than larvae or small nekton. This effect would be difficult to discern with my data without the inclusion of size data.

The presence of different species of nekton in marsh nursery areas is seasonal (Hagan and Able, 2003; Able and Fahay, 2010; Mukherjee et al., 2013). My data support this in that several of the estuarine dependent species had higher densities in some months than in others. In addition, there was a significant (p < 0.05) difference in density between Areas for some species. Variations in community structure have been attributed to fluctuations in salinity and fresh water flow (Mukherjee et al., 2013). My data suggest that this may be an oversimplification for processes on the LB. For larvae of species that enter the marshes during short periods of time, this may in part be due to issues related to transport, particularly during winter and spring storms. Species such as *A. mitchilli* and *B. patronus* which spawn in the estuary may have a different periodicity and distribution than *C. arenareus* and *S. ocellatus* which spawn offshore. Advection is much more important for these latter species. Further research is needed to more clearly understand the effects on larval transport.

Estuarine marshes are harsh environments for nekton. Stochastic meteorological and anthropogenic events are part of the "normal" for these environments. While they may affect the transport and distribution of nekton, they must be viewed as part of the whole and not as independent events. Month was found to be a significant factor in the distribution of nekton in that different species were found to peak in different months. Furthermore, species which peaked in different months tended to have higher concentrations in different areas. For shrimp, *F. aztecus* peaked earlier than *L. setiferus* and had higher densities in Areas generally to the west whereas *L. setiferus* peaked much later and had highest densities in the east and in Area 5. The estuarine dependent fish species *M. undulatus* also reached peak densities in February and March and had the highest densities in Area 4. One of the reasons for higher densities in different transport.

into the marsh. Fishes such as *A. mitchilli* and *B. patronus* tend to spawn in all areas of the estuary and at very frequent intervals. These facts probably account for their widespread presence throughout all of the Areas. The observed seasonality only reflects those periods most conducive to spawning.

In contrast to M. martinica, the resident fish species *M. beryllina* spawns in the marsh and is most common in Areas 3 and 4. These two Areas were the most removed from Lake Borgne and the least affected by winds and tides. Both of these areas had the greatest amounts of SAV. These two species occupy different niches. Where *M. martinica* was found for the most part outside of the marsh near the beaches, customary habitats for *M. beryllina* are inside the marshes. The difference in the numbers of each species collected is a consequence of very little sampling having been conducted along the beaches outside of the marshes.

The fundulids were collected most frequently when water levels were at their lowest. During these periods, when the marsh platform was shallow if not dry, they were found along the marsh edges and frequently in large numbers. It is difficult to understand why *C. sapidus* would have the highest densities in Area 3. This Area has only two deep bayous entering from Lake Borgne but has many entering from the GICWW. One explanation for this observation is that *C. sapidus* takes advantage of selective tidal stream transport for entering the marsh. This behavior may lead this species to take advantage of the two large tidal Passes rather than the smaller bayous with less tidal exchange and current depositing them into the GICWW, where the current begins to slow. From there they would have direct access into the marsh particularly into Area 3.

An interesting observation is the relationship of distance from the entrance into the marsh to community structure. This may be a result of abiotic conditions fluctuating less further into

the marsh, or it may be a result of certain species such as estuarine resident species dominating the community deeper in the marsh. Transport mechanisms resulting from tidal prism, ship assisted transport, and meteorological conditions may also affect community structure as it transitions deeper into the marsh.

The structure of nekton communities on the LB vary with month and with Area. These two factors are closely interrelated. The presence of nekton in samples reflects seasonal occurrences. However, the varying densities in the different Areas are not a result of the measured abiotic variables alone. The different Areas appear different although they are the sum of similar microhabitats. The differences in densities of different species of nekton are a function of the timing of spawning. The differences among Areas, particularly in densities of estuarine dependent species, suggest that advection and transport into the marsh are important factors. Varying currents and tides, resulting from changes in meteorological conditions, may favor advection from spawning grounds more towards one area of the marsh than another. Biotic responses to as yet undetermined stimuli may cause certain species to orient more towards one section of the marsh than towards another. In conclusion, in the LB nekton community, structure varies across a large marsh-scape and may be a consequence of effects which act at a distance from the marsh and not just a few abiotic factors whose effects are more prominent on smaller microhabitats

## Chapter 2

The Cast Net: An Overlooked Sampling Gear

## Abstract:

Sampling fishes and decapods in shallow estuarine marsh habitats is challenging because of depth variation, soft substrates that make standing difficult, and the presence of submersed aquatic vegetation. Though often overlooked, a cast net is an excellent gear type for sampling nekton species in this environment. An oft repeated complaint about the cast net is that it is difficult to deploy successfully and is not repeatable. I report here that the cast net can be deployed successfully with a minimal amount of practice and we present a method of standardization that shows most of the variation in area covered is among individual operators rather than within one individual. In terms of catch per unit effort (when compared to a 1 m<sup>2</sup> throw trap), the 1.8 m cast net collected more species, more biomass, and more pelagic animals than the throw trap. There was no statistical difference in the total number of animals collected or the total number of species. The cast net is a useful gear type for sampling nekton communities in estuarine habitats.

## **Introduction:**

Sampling of fish and decapod communities in estuarine marshes is important for assessing the health of fisheries and for monitoring anthropogenic impacts on these sensitive and productive environments (Beck et al., 2001; Solomon et al., 2006; Rotherham et al., 2007). Knowledge of the habitat requirements of all life stages of the fishes and decapods that inhabit these areas is critical in making decisions regarding preservation and restoration (Sargent and Carlson, 1987; Beck et al., 2001). The proper design of sampling regimens and the choice of gear is important to the success of any monitoring program (Rotherham et al., 2007). The choice of sampling gear is dependent on many factors including the purpose for which sampling is done, the habitat being sampled, and the species targeted (Sargent and Carlson, 1987; Rojas and Minello, 1997). Technically, sampling in estuaries can be difficult because of the variation in depth, the variety of habitats that comprise an estuary, the nature and quality of the substrate and vegetation, and the variety of species targeted (Rojas and Minello, 1997).

In June 2010, I initiated an analysis of the fish and decapod community structure of the Lake Pontchartrain/Lake Borgne Land Bridge, a highly productive estuarine marsh in the Pontchartrain Estuary of southeastern Louisiana. The sampling area comprises more than a hundred square kilometers of predominantly *Spartina* spp. (smooth cordgrass and saltmeadow cordgrass) salt marsh consisting of tidal creeks and ponds (Penland et al., 2002). Depth typically ranges from 20 cm to over 3 m. The substrate is composed primarily of poorly compacted decaying organic material and mud. It is not possible to stand or walk on the substrate or the sparse land. Tidal flow in the creeks may exceed 1 m/s especially with the combined effect of wind stress. Submersed aquatic vegetation (SAV) and algae are dense in many of the ponds making sampling difficult at times.

The cast net has been used by other investigators to collect fishes in shallow habitats and to supplement impoundment collections (Meador and Kelso, 1990; Stevens, 2006a). Common complaints about the cast net are that it is difficult to deploy and that the area covered by the net is not consistent (Leber, 1995; Emmanuel et al., 2008). Furthermore, the catch efficiency of the cast net has not been compared to other gear types such as the throw trap, another commonly

used gear type. The purpose of this paper is to present one method of standardizing the area covered by the net for different users (operators) and to compare it to the throw trap in terms of efficacy and sampling efficiency.

#### Methods:

The cast net used in this experiment was a 1.8 m 6.4 mm bar, monofilament net with 2 kg of lead per radius m. The throw trap was a 1 m square aluminum box, open at both ends and 80 cm deep. It was emptied by means of a 1 m square aluminum frame, which fit just inside the box, covered with 1 mm nylon mesh.

Standardization of the net was performed in the following manner. Three different operators each threw the net 10 times onto a flat lawn of 5 cm high St. Augustine grass. A seventy foot bucket truck was then suspended directly over each net after it was thrown. A plumb-weight suspended from the bucket was maneuvered until it was in the center of the net. A 50 cm x 50 cm white square was then placed directly under the weight and a digital photograph was taken from a height of 8 m directly over the weight to minimize any angular foreshortening.

Using Image J software, the area of the net and the area of the white square fiducial were measured in each digital image and the area of the net throw was computed from the ratio (Rasband, 1997 - 2011). The mean area covered by the net, and the variance and standard deviation were computed for each of the operators. I used repeatability, r, as a measure to quantitatively describe the variation that occurs among rather than within operators (Lessells and Boag, 1987). Mean values of the areas of each operator's throws and the variance were

obtained and compared using an ANOVA (analysis of variance) and post-hoc analysis was performed with Tukey's HSD function in "R".

Repeatability can be used to examine the consistency of the operators. Computing repeatability, r (Lessells and Boag, 1987):

$$r = S2A / (S2 + S2A)$$
 (1)

Where, S2A is between group variance and S2 is within group variance.

As part of the routine sampling program, several sites on the Land Bridge were chosen randomly for sampling prior to each trip (see Chapter 1). A shallow draft motorized aluminum flat boat was outfitted with a two meter anchor pole and three meters of rope was allowed to drift into the sampling area and pole was deployed to anchor the boat in place. For the first 20 samples, either the net or the box was arbitrarily deployed first. For the remainder, the order was randomly determined by a coin flip. After the first gear was deployed and emptied, the boat was allowed to drift on the mooring rope such that the second gear was deployed in a different site at the same location. In this manner, the same community was sampled by both the cast net and the throw trap in each area. Initially, the times required for sampling with each gear type were recorded for the first six samples with each gear type.

The net was deployed by the same operator each time. It was thrown three meters from the boat, allowed to sink, and then slowly retrieved and emptied into a large white tub. The net and deck of the boat were inspected for any fishes or decapods that may not have been dropped into the tub. All SAV was carefully removed and inspected. All fishes and decapods were placed into a zip lock bag and submerged in an ice water bath. The throw trap was deployed by two people, one on either side, over the side of the boat, into water no deeper than 78 cm. If the water level came over the side of the box, testing was terminated and the boat was moved to a new location at least 50 m distant. If the net sampling had been done first, it was repeated in the same order at the new location. The box was then swept with the net frame until three successive sweeps did not produce any new animals. All SAV was removed from the box and inspected for animals. All recovered decapods and fishes were placed in a zip lock bag and submerged in ice water.

Immediately before sampling was started at a site, a Yellow Springs Instrument (Professional model) was used to obtain water temperature, salinity, specific conductivity, oxygen saturation, and dissolved oxygen concentration. A steel rule was used to directly measure water depth to the substrate and a Secchi disc was used to obtain a measure of water clarity.

A total of 37 cast net samples and 37 throw trap samples were obtained at 37 different sites. All fishes and decapods were returned to the lab and identified to species level, weighed, and standard and total length measured and recorded. The number of each species, the total number of species, and total biomass were recorded for each sample. Grass shrimp (*Palaemonetes* spp.) and Harris mud crabs (*Rhithropanopeus harrisii*; Gould, 1841) were not included in the counts due to their large numbers in both gear types. Date, time, GPS coordinates, water depth, water temperature, salinity, dissolved oxygen, oxygen saturation, Secchi disc depth, the presence of SAV in the net or trap were recorded for each sample.

The same community was sampled with each pair of gear deployments. For this reason, the cast net and throw trap data were compared across all of the samples rather than as a pairwise

comparison of the samples. A resemblance matrix of the samples (numbers of each species) using a Bray-Curtis similarity index was created using Primer 6 software to assess the relative distances separating the samples in the same rank order as the dissimilarities (Clarke and Gorley, 2006). A multi-dimensional scaling (MDS) plot was generated to assess the separation among samples and any potential grouping of samples. This analysis was performed after excluding those samples which contained no animals. A one way analysis of similarity (ANOSIM) using cast net and throw trap as predictive variables was performed on the samples as number of animals per species per sample. A MANOVA (multivariate analysis of variance) was performed using "R" ( $\alpha = 0.05$ ), with weight, number of animals, and number of species per sample as dependent variables and gear type, cast net or throw trap, as predictor variables (Ihaka and Gentleman, 1996). The weight and number of animals in each collection were log-transformed to satisfy the assumptions of normality required for the test. Pairwise t-tests were performed if the MANOVA was found to be significant. The significance level was adjusted for each pairwise t-test ( $\alpha = 0.05/n$ ).

## **Results:**

An ANOVA of "areas" as a function of the operators demonstrated a significant difference among the operators relative to the difference within the operators (F = 32.283, p = 6.918e-08). The high value of F demonstrates the numerator is large, and the probability is less than 0.000001% that there is no difference. Since the within operator error is small compared to the between operator difference, it would appear that the operators were more consistent than different operators were alike (Vanhooydonch et al., 2005).

Repeatability, r, was significant at 0.758. A repeatability value of 0.76 means that 76% of the variation was among operators rather than within the operators. This showed that although there is a difference among operators, individual operators were consistent in their performance. There was a difference in skill level and more skillful throwers were able to cover more area, but each thrower appeared to be consistent in their throws and to cover the same area each time (Figure 1). This implied not that an operator's throws will cover the same area each time but that there will be little variation over a large number of throws. This methodology can be repeated periodically to determine the area a single operator's cast net throws cover and to correct sampling for different operators. Operators 1 and 2 were both fairly experienced and the mean area covered by their throws was 3.9 square meters (Figure 9). Operator 3 was inexperienced and exhibited a greater range, variance, and standard deviation (Figure 9).



Figure 9: Boxplot showing the results of ten cast net throws by three different operators. Operators 1 and 2 are both fairly experienced and the mean area covered by their throws is 3.9 square meters. Operator 3 is inexperienced and shows a greater range, variance, and standard deviation.

A large difference between times required for sampling with the two gear types was immediately noted and this part of the experiment was discontinued. It took less than five minutes for a single sample with the cast net, but took at least 15 minutes for a single sample with the throw trap depending upon the number of sweeps with the net that it took to clear it completely with some samples requiring more than 30 minutes to complete. Furthermore, if the depth was more than 0.78 m or if the trap sank in the soft substrate, sampling with both gear types had to be repeated.

All sampling was performed at a depth range of 0.23 m to 0.78 m and was limited by the depth of the throw trap. Water temperatures ranged from 13° C to 32.4° C. Salinity varied from a minimum of 0.71 to 10.35. Secchi disc depth varied from 0.2 m to 0.6 m. A total of 37 samples were collected with each of the two gear types and included 22 different species of fishes and decapods (Table 1). All of the cast net samples contained fishes and decapods. Five of the 37 throw trap samples did not contain any animals. The two gear types differed in the total numbers of animals collected and in the weight of the samples. The cast net collected 616 animals with a total weight of 2966 g, and a mean weight per sample of 80.18 g. The weight of the 946 animals collected in the throw trap was 187 g, with a mean weight of 5.06 g per sample. The mean number of animals collected per cast net throw was 17 (range 1-139). The mean number of species per throw was 3.08 (range 1 - 6, SD 1.31). For the throw trap, the mean number of animals collected was 26 (range 0 - 247), and the mean number of species was 2.54 (range 0 – 6, SD 1.7). Lucania parva (rainwater killifish) less than 12 mm were collected by both gear types although the smallest (10 mm) was collected by the cast net. Less than 10 mm carapace length *Callinectes sapidus* (blue crab) were collected by both gear types. The largest animals were collected in the cast net including *Mugil cephalus* (striped mullet) (SL = 220 mm;

Figure 10). The difference in weights between the two gear types indicates that there is a difference in the animals collected in the communities sampled.



Figure 10: Graph of the relative standard lengths of the fishes captured in the cast net (Blue) and the throw trap (Red). Only 25 random fish were measured in a sample if contained more. Decapods were omitted from this analysis. Cast net fishes had longer standard length. The plot of the throw trap fish was biased by the inclusion of *Syngnathus scovelli*.

Twenty-one different species were collected in the cast net and sixteen in the throw trap. *Sciaenops ocellatus* (red drum) and *Poecilia latipinna* (sailfin molly) were both collected in the throw trap and not in the cast net. The throw trap species did not include *M. cephalus*, *Brevoortia patronus* (Gulf menhaden), *Cynoscion arenareus* (sand seatrout), *Lagodon rhomboides* (pinfish), *Bairdiella chrysoura* (silver perch), or *Oligoplites saurus* (leatherjacket). Of the 946 animals collected by the throw trap, 765 were *L. parva*, with the next most common being *C. sapidus*. *B. patronus* was the most common species in the cast net collection (278) followed by *L. parva* (109) (Table 7).

Table 7: Species composition for 37 collections with each gear type including total numbers of
each species collected. The cast net collected essentially all of the animals collected in the throw
trap but surpassed the throw trap in capturing pelagic animals. The throw trap collected more
benthic animals and those associated with submersed aquatic vegetation.

Species		Cast net	Throw Trap	
Decapods				
Callinectes sapidus		42	40	
Farfantepenaeus aztecus		16	4	
Litopenaeus setiferus		14	7	
Fish				
Anchoa mitchilli		20	37	
Brevoortia patronus		278	0	
Mugil Cephalus		11	0	
Menidia beryllina		10	3	
Fundulus grandis		45	7	
Lucania parva		109	765	
Poecilia latipinna		0	1	
Cyprinodon variegatus		26	12	
Oligoplites saurus		1	0	
Lagodon rhomboides		2	0	
Bairdiella chrysoura		1	0	
Cynoscion nebulosus		4	1	
Cynoscion arenareus		2	0	
Leiostomus xanthurus		16	1	
Sciaenops ocellatus		0	8	
Ctenogobius shufeldti		3	4	
Gobiosoma bosc		3	23	
Microgobius gulosus		1	5	
Syngnathus scovelli		12	28	
	total	616	946	

There was a significant difference in species composition among the samples (ANOSIM, Global R = 0.063, p = 0.003). However, the very low global R value implies that the variation within samples is high and that the variation among samples may not be attributed to gear type.

The MDS plot indicated a clustering of the throw trap samples within the cast net samples (Figure 10). The MANOVA indicates that gear types were significantly different (p = 0.008374; Figure 12). The pairwise t-tests show that only the weight of the samples is significantly different (p = 0.0000036) and that the number of animals and species collected per sample are not statistically different (Table 8).



Figure 11: Multidimensional Scaling Plot of all of the samples collected. Cast net (n, blue triangles) and throw trap samples (t, green circles) are labeled. The throw trap samples appear as a subset of the cast net samples. Multivariate dispersion indices, n = 0.983, t = 1.009. Pairwise comparison, IMD = - 0.026.

Although the cast net collected almost all of the species collected with the throw trap, the converse was not true. The throw trap tended to collect more benthic and SAV-associated animals while the cast net collected not only those, but also more pelagic species. The one exception to this was the single throw trap deployment which accounted for the only eight *Sciaenops ocellatus* (red drum) collected in the sampling. The cast net collected more pelagic

fishes whereas the throw trap collected more benthic and SAV-associated animals (Table 7). All

of the M. cephalus, B. patronus, O. saurus, and C. arenareus were collected in the cast net.

Table 8: Pairwise t-tests for cast net and throw trap samples. The significance level was adjusted for the t-tests by dividing the original  $\alpha$  level (0.05) by the number of variables (3). Only the weight of the samples reached statistical significance. The number of species and the number animals collected was not statistically different for the different gear types.

Dependent Variable	degrees of freedom	t-value	p-value
Weight	72	5.0195	3.6E-06
Number of species	72	1.5133	0.1346
Number of animals	72	0.2824	0.7785
The significance leve	l had to be adjusted fo	r the t-tests.	This was done by taking the

The significance level had to be adjusted for the t-tests. This was done by taking the original level ( $\alpha = 0.05$ ) and dividing it by the number of variables (3).

$\alpha$ level = 0.05, adjusted for t-tests	0.016667	
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Most of the *L. parva, Syngnathus scovelli* (Gulf pipefish), and the gobies were collected in the throw trap although all of these species were represented in the cast net samples.



Figure 12: These four boxplots depict the mean and quartiles for centroid values, Log-weight, Species, and Log-Animals, for each year gear types (Cast Net and Throw Trap). Centroid values are standardized combined values which include all 3 response variables analyzed. Significant differences between gear types were found for Centroid values and Weight (MANOVA, p = 0.008374; t-test, p = 0.0000036). "\*" indicates significant difference.

## **Discussion:**

My research has countered the complaints that the cast net is difficult to deploy and standardize. I have presented one method for standardizing the area covered when the net is deployed by different operators. An oft quoted objection to the cast net is the amount of skill that is required to deploy it successfully. Just as there are different techniques for pulling a beach seine or an otter trawl, there are different techniques for throwing a cast net. Some of these have been shown to be better than others for maximizing the area covered for some

operators. The best method for deploying a cast net was not the subject of this research. Assuming that the operator's technique does not change, I have shown that it is possible to standardize the net throw for each operator, in terms of coverage area, regardless of the amount of training or experience that he or she may possess. It is reasonable to assume that each operator will be consistent in the area that is covered by a series of throws.

One method previously used to estimate the area covered by a cast net was to throw it from a john boat 14 times onto grass and measure the maximum and minimum radius for each throw. The approximate area was then calculated assuming that it approximated an ellipse. Using a 1.15 m radius net, the mean area was estimated to be  $2.8 \pm 0.2 \text{ m}^2$  (Stevens, 2006b). The calculated maximum coverage area using the reported net radius (without knowing the length of the lead line) is  $4.15 \text{ m}^2$ . The approximate coverage area was 67% of the maximum. Our method produces a more accurate estimate of the covered area by measuring the actual area rather than approximating it. Both experiments confirm that the area actually covered by the thrown net cannot be calculated from the length of the brails but must be measured directly. Furthermore, our data show that though there is variance among operators, individual operators are consistent and that as they gain experience, their coverage area does not change significantly. Experience will decrease the amount of individual variation. For this reason, it may be necessary to do an initial assessment of each operator and to perform a periodic reassessment. I have demonstrated one procedure for standardizing the area covered by the throw of a typical operator. This method will allow a semi-quantitative approach to this gear in that while an exact comparison cannot be made cast to cast, a statistically meaningful evaluation of species presence can be made across a number of samples.

I have compared sampling of fish and decapod communities with the throw trap and have characterized a particular type of habitat where the cast net is a useful sampling gear. The cast net is a good choice for sampling in shallow estuaries with soft substrate and SAV where it would be impractical to use a trawl or beach seine. It can be used in deeper water than the throw trap and in the presence of moderate currents. It is not useful for sampling in the presence of emergent vegetation or in the presence of underwater obstructions such as large or rough rocks, logs, or large debris. I found the cast net to be easy to use. In my hands, it is more versatile and faster to sample with than the throw trap. The large coverage area of the cast net allowed me to sample more than one microhabitat with each throw. This allowed me to collect more species with each throw than would otherwise have been possible. It is effective for capturing fast swimming pelagic nekton that might otherwise be capable of avoiding a throw trap.

Multiple gear types have been used to sample an area composed of different habitat types because of the limitations imposed by the use of a single gear type (O'Connell et al., 2004; Stevens, 2006a). Multiple gear types may not be either as effective or allow sampling as efficiently as the cast net depending upon the habitats sampled. Sampling may be quantitative or qualitative in nature. For some sampling regimes, the goal of the research may be to determine the numbers of animals present within a habitat (density) and their response to various treatments (Rojas and Minello, 1997). In others, the goal may be to determine the number of species within a specific area or habitat, to define the community, or to determine diversity or productivity (O'Connell et al., 2004). In each case, the choice of gear type may be different. The accuracy and precision of the gear employed is determined not only by the ability of the gear to entrap all of the animals in the area sampled (gear efficiency), but also on the ability of the operator to remove all of the animals collected from the gear (clearance efficiency; Jordan et al., 1997;

Steele et al., 2006). Once collected in the gear, all of the animals must be removed and counted (gear clearance). The suitability of various gear types to accomplish these goals has been the subject of much debate and investigation (Sargent and Carlson, 1987; Rojas and Minello, 1997; Able et al., 2005; Rotherham et al., 2007; Able and Fahay, 2010). I did not test gear clearance or clearance efficiency in this experiment. We compared gear efficiency of the cast net directly to the throw trap in a particular environment.

This comparison was conducted as part of a larger research project to study the fish and decapod community structure of the New Orleans Land Bridge. I did not intend to compare sampling gear as part of our community structure study. However, as I planned our sampling protocol, it became obvious that I would have to justify our choice of sampling gear. I chose to compare the results of my cast net collections to an equal number of throw trap collections because throw traps are considered 'standard' estuarine sampling gear. Cast net efficiency has been compared to published throw trap and seine efficiencies (Stevens, 2006b). In my research, I made a direct comparison between the two gear types. Throw traps are considered to be quantitative in that both their catch efficiency and catch clearance have been shown to be very high by numerous investigators (Jordan et al., 1997; Rojas and Minello, 1997; Minello and Rozas, 2002). Throw traps are limited by the area that they enclose and by the difficulties inherent in deployment, clearing, and transport. Larger throw traps can be employed but the typical trap covers  $1 \text{ m}^2$ . Multiple samples can be used to decrease sampling error, but these are time consuming and are still fraught with inherent errors such as trap avoidance. Determination of species density is often the goal of throw trap sampling (Rojas and Minello, 1997) whereas other studies target different measures of community composition.
The usefulness of the cast net for collecting a large size range of animals, including very mobile species such as *M. cephalus* and *S. ocellatus*, is determined by three characteristics of the net itself and by the fact that it is not limited by water depths less than the radius of the net (Emmanuel et al., 2008). Sufficient net radius size must be chosen to adequately cover the anticipated area to be sampled without being too large for the operator to handle successfully. Nor should it be too large for the area being sampled. The size of the animals collected will be determined to some extent by the mesh size of the net. I had no trouble collecting animals less than 14 mm SL with a 6.4 mm bar net, however, the smaller the net size, the smaller the animals collected, but the net will encounter more air and water resistance. In deep water, drag produced by small mesh size may cause the net to collapse prematurely thus decreasing the effective coverage area. The weight of the lead line is equally important as this will determine how fast the net sinks, how well it counteracts the drag of the mesh, and how well it will enclose animals, particularly in the presence of SAV. My experience is that the cast net cannot be used in the presence of very dense SAV or emergent vegetation, but is efficient in the presence of moderate less-dense SAV.

The cast net can be used in deeper and moving water which gives it access to additional habitat characteristics generally not available to the throw trap. The cast net can be deployed faster, covers a larger area and is less visible to fishes that may be able to avoid the bulky, slow moving, and more obvious throw trap. This assumption is supported by the observation that the cast net collects more species and more biomass than the throw trap and it successfully collects larger and more mobile fishes, such as *M. cephalus*. Stevens (2006b) also found that the cast net collected more *M. cephalus*, *Elops saurus* (ladyfish), and *Mugil curema* (white mullet) than did the throw trap while the throw trap collected more benthic animals. One reason for this may be

that the cast net can be deployed at a distance from the operator which may allow her or him to approach fishes with little disturbance.

It is noteworthy that the cast net also collected small animals as well, despite the relatively large minimum mesh size of 6.4 mm. The fishes collected in the cast net are generally of greater standard length than those collected by the throw trap (Figure 3). The cast net may be biased against certain species and habitats. My cast net collections did not contain as many of the SAV-associated species such as L. parva or as many of the benthic gobies as did the throw trap, an observation shared by Stevens (2006b). The cast net did not bring up all of the SAV it encompassed so undoubtedly additional fishes that may have been present were not collected. Fishes such as the gobies are capable of burrowing into the substrate and therefore are capable of avoiding the cast net but not the throw trap. As evidenced by the large number of gobies and L. *parva* in the collections, the throw trap appears to be biased towards benthic and smaller animals as these animals are less able to avoid the trap than larger more mobile animals. The clearing net used with the throw trap has 1 mm mesh, whereas the cast net was 6.4 mm bar, which may account for some of the differences in my study in that fewer small animals were likely to escape the throw trap than the cast net during clearing. Other aspects which I have not addressed include issues such as capture efficiency, or how well the net collects all of the animals in an area, and gear clearing efficiency, which is a measure of how many animals collected ultimately escape or are lost during the process of emptying the net.

Although my collections confirm the patchiness of species within the estuary, the absolute numbers of animals collected does not appear to be statistically different between the two gear types. Were the throw trap to be deployed four times (area ratio a nominal 4:1) undoubtedly more animals would be collected. The question is whether or not the catch data

would change significantly if the throw trap were deployed enough to make up for this difference. Practicality may limit the number of samples that can reasonably be collected with the throw trap, but this is less of a problem with the cast net. Study design may answer the question as to whether or not this bias is significant.

The use of the cast net has enabled me to obtain a large number of samples in a large geographical area across a wide range of habitats in an efficient amount of time. I was more interested in the community structure than the density of animals within a particular habitat. As is frequently the case, the larger the sample size, the more meaningful the data. I have demonstrated that the cast net was more versatile and faster to use than the throw trap or drop sampler, and was not limited by depth and boat size. In these habitats, it was less destructive of vegetation than beach seines and trawls and easier to use given the shallow depths and soft substrate. It was also less labor intensive than many other types of sampling gear. In choosing the gear type that is best suited to each application, the researcher must first consider the limitations imposed by the habitats and species to be sampled and should consider the cast net as a valid option.

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## Chapter 3

Tarpon (Megalops atlanticus) life stages in southeastern Louisiana

# Abstract:

Despite the fact that tarpon (*Megalops atlanticus*) are an important game fish in Louisiana, little is known about their about their life history in state waters. Tarpon stocks crashed in the northern Gulf of Mexico during the 1950's and 60's, though only recently has there been new interest in properly managing this species. In this paper I discuss the presence of five major life stages of tarpon in the waters of southeast Louisiana and document for the first time the presence of spawning tarpon, the presence of over- wintering adult tarpon, and the yearround presence of juvenile tarpon in Louisiana coastal habitats. These results have serious implications towards future management efforts in state waters and throughout the northern Gulf of Mexico.

## **Introduction:**

In the Gulf of Mexico, tarpon (*Megalops atlanticus*) are popular game fish sought for their spectacular leaps and tenacious fights when hooked (Babcock, 1921). In Florida, Louisiana, Central America, and on the Yucatan Peninsula, they support an important sports fishery that draws fishers from all over the world (Cruz-Ayala, 2002; Dailey et al., 2008). In the United States, tarpon are popular game fish from North Carolina to South Texas. At one time, Port Aransas, Texas, and Biloxi, Mississippi, vied for the title of "Tarpon Capital of the World" (Ault, 2008). The southeastern coast of Louisiana has been the home of numerous sports fishing

tournaments in which tarpon have been the "glamour" fish. The Grand Isle Tarpon Rodeo, the oldest fishing tournament of its kind, and several other tournaments attract anglers from all over the southern United States and beyond (Dailey et al., 2008).

Tarpon occur on both sides of the Atlantic Ocean. In the eastern Atlantic, their range extends along the coast of Africa as far northward as the Mediterranean Sea. In the western Atlantic, they range from the northern coast of Brazil to the Chesapeake Bay although they have been found as far north as Nova Scotia. Tarpon have become established on the Pacific coast of Central America in Panama, after successfully transiting the Panama Canal. They are ubiquitous in the Caribbean Sea and the Gulf of Mexico (GOM). In certain parts of northern South America, such as Columbia, and along coast of Central Mexico in Costa Rica, tarpon are fished for consumption (Cruz-Ayala, 2002; Silgado, 2002).

Female and male tarpon reach sexual maturity at 8 – 10 years although females are somewhat larger at maturity (Crabtree et al., 1995). Sexual dimorphism is demonstrated by the larger size of the female and they tend to be longer lived than males (Crabtree et al., 1995). Mature females may produce between 12 and 20 million eggs a year and live more than 50 years. It is presumed that the eggs hatch within 24 to 48 hours of fertilization although fertilized tarpon eggs have not been collected *in situ* (Jones et al., 1978). Tarpon larvae, the leptocephali, remain as icthyoplankton for as long as several months until they transform into juvenile tarpon when they enter inshore nursery areas.

Although no formal population surveys have been conducted, tarpon populations in the northern Gulf of Mexico, and in particular in Louisiana, Mississippi, and Texas, have declined precipitously since the 1960's and 1970's (Cruz-Ayala, 2002; Winemiller and Dailey, 2002;

Ault, 2008). The numbers of anglers specifically targeting tarpon likewise has decreased in the past four to five decades according to records of several tarpon rodeos (Dailey et al., 2008). The cause of this apparent decline is unknown and has been discussed by many investigators (Holt and Holt, 2002; Landry, 2002; Ault et al., 2008; Dailey et al., 2008). It has been hypothesized that the decline is a result of the loss of nursery habitat in South Florida and on the Yucatan Peninsula as a consequence of human development and the destruction of mangrove marshes. Tarpon have been demonstrated to be highly migratory (Ault et al., 2009). Tagged tarpon have traveled from Veracruz, Mexico, to Marsh Island, Louisiana, and from the east coast of Florida to the Chesapeake Bay (Ault et al., 2009). The principle of "their fish are our fish and our fish are their fish" has become the mantra for management efforts of this species, implying that tarpon migrate long distances across state and international borders and that conservation and management efforts need to involve all of the geographic stake holders (Luo et al., 2008).

In 2004, Jim Franks at the University of Southern Mississippi's Gulf Coast Research Laboratory, began collecting young-of-the-year (YOY) tarpon in tidal sloughs opening into the Mississippi Sound (Franks et al., 2009). He continued to collect the YOY in the same sloughs for six consecutive years. In addition, he collected two leptocephali on the Mississippi Coast in 2012 (Franks, personal communication). He originally hypothesized that these fish were arriving on the Mississippi Coast after being carried northward by the Loop Current as tarpon were never documented to spawn in the northern GOM (Franks et al., 2009). Smith (1980) and Crabtree (1992,1995) collected leptocephali in the southern GOM and off the southwestern coast of Florida and hypothesized that tarpon spawned off the continental shelf in these areas. They further hypothesized that YOY found on the Yucatan Peninsula and in southwest Florida were a product of this spawning activity.

It has been assumed that tarpon migrate southward from the northern GOM in the late fall and perhaps exit the GOM, except for small resident populations in south Florida (Luo et al., 2008). Tarpon are also not known to overwinter in the GOM. The purpose of my research was to examine the life cycle of tarpon in the northern GOM and in particular the southeastern coast of Louisiana. My hypothesis is that some tarpon are capable of completing their entire life cycle within Louisiana waters. At the beginning of this research effort, it quickly became obvious that it was impossible for one person in a small boat to conduct this research alone. Over two hundred volunteer anglers and divers were enlisted to gather specimens, photographs, video, and to carry out an extensive search for all tarpon life-stages. Numerous members of the fishing community including commercial fishers, bait-sellers, launch operators, recreational fishers, and scientists also participated. The use of "citizen scientists" to collect data is rapidly becoming an accepted practice and is enabling new and different types of research (Henderson, 2012). Our experience working with volunteer divers and anglers has proven invaluable to the collection of samples that would otherwise have been unavailable to us (Fogg et al., 2013).

The purpose of this research was to document the presence of five major life stages of tarpon on the southeastern Louisiana coast. Using various approaches, I here summarize my efforts to document the presence of tarpon larvae (leptocephali), YOY, juveniles, adults and overwintering adults, and spawning capable tarpon. My goal was to produce evidence that tarpon are capable of completing their entire life cycle within Louisiana waters.

## **Materials and Methods:**

Confirmation that tarpon are completing their life cycle along the Louisiana Coast required documenting the presence of five principle life stages: leptocephali (the larval stage), YOY, juveniles, adults, and spawning adults. Because free-floating tarpon eggs have never been identified, no effort was made to find these. The search for each these life stages required different approaches in addition to different types of field work. Museum collections and collection records were reviewed. Fishers and divers with experience with tarpon in Louisiana were interviewed and recruited to assist in the search. This included joining the Louisiana Council of Underwater Dive Clubs and requesting and reviewing catch records, video, and still photographs going back decades. Numerous spear fishing and sports fishing tournaments and rodeos were attended in order to examine any tarpon brought to the dock and to meet and talk to as many spear fishers and anglers as possible. Southeastern Louisiana comprises a large area of potential tarpon habitat. The search for each life stage was unique, and without the help of the volunteers, this work would not have been possible.

# Leptocephalus:

The leptocephalus is the larval life stage of the tarpon. Spawning is assumed to occur in deep waters off the continental shelf. Eggs are hypothesized to hatch within 24 to 48 hours after fertilization. Tarpon eggs are small, vitellogenic, and generally less than 700  $\mu$  in diameter. The planktonic leptocephalus increases in length after hatching and may approach 3.0 cm when fully developed prior to metamorphosis. The leptocephalus is elongated, with a small pointed head, is laterally compressed, with a forked caudal fin and approximately 54 - 57 myomeres. It

progresses through three stages of development as it ages and drifts as part of oceanic icthyoplankton. As it grows it becomes more motile, the leptocephalus acquires a heart and circulatory system, an alimentary canal, functioning gills, and a physostomous airbladder (Jones et al., 1978). As larvae approach coastal nursery areas, they undergo metamorphosis to juveniles. I was unable to conduct sampling of the off shore waters for leptocephali because of limited resources and equipment. However, other sources of possible samples were available. Trawl samples and the records of these samples from routine collections of the R.V. Cavalla conducted as part of a research program of the Nekton Research Laboratory (NRL) of the University of New Orleans were examined (O'Connell, personal communication). All of these collections were performed within the Pontchartrain estuary. Most were performed in the upper estuary although some were conducted in the area of the Chandeleur Islands. In addition, samples from cast net collections performed as part of a research program on the New Orleans Land Bridge were examined. Collections of the NRL at University of New Orleans and the Tulane Museum of Natural History - Royal D. Suttkus Fish Collection were also examined.

# YOY:

YOY tarpon have been anecdotally reported over the years by fishers and bait collectors from several areas of southeastern Louisiana. Little effort has previously been made to scientifically document the presence of this life stage. Southeastern Louisiana comprises a large area of varying habitats over thousands of square kilometers. In an attempt to locate YOY tarpon in southeastern Louisiana, I developed a systematic search using a variety of approaches.

The first approach involved a public awareness campaign and a request for samples. I distributed 250 four-color 8.5 x 11 inch flyers to sporting goods stores, boat launches, bait suppliers, boat storage facilities, fishing clubs, and individuals beginning in the fall 2009 (Figure 13). A phone number and email address were provided for reporting tarpon. I answered every





Please put me on ice and contact **Dr. Will Stein** <u>Pontchartrain Institute for Environmental Sciences</u> **NEKTON RESEARCH LABORATORY** <u>http://www.nekton.uno.edu/</u> **504 616 5262** or email <u>doctarpon@hotmail.com</u>



Figure 13: Tarpon Flyer distributed throughout southeastern Louisiana. While the response to the flyers in terms of the numbers of YOY and juvenile tarpon reported was minimal, overall response indicates they increased public awareness of the research.

response in person and I identified every submitted fish. The location of the capture, weight, length, and the identity of the person capturing the fish were also recorded.

The second approach was to identify a specific area in which to search for YOY tarpon. I chose to survey the New Orleans Land Bridge east of the Gulf Intercoastal Waterway (GIWW), from Chef Menteur Pass to the Louisiana-Mississippi State Line east of Pearl River Island (Figure 14). As part of another research project (See Chapter 2), I conducted a systematic search



Figure 14: Research area on New Orleans Land Bridge. Of 65,000 fishes and decapods collected between 22 April 2010 and 5 July 2012, only one YOY tarpon was collected (pink marker). Over 2340 sites were sampled and more than 4600 cast net throws were made over 30 months. Sampling covered 18,400 square meters of aqueous marsh.

in the following manner. The Land Bridge search area was divided into five areas based upon geomorphology. Area 1 included all of Pearl River Island. Area 2 stretched from Rigolets Pass westward to Unknown Pass. Area 3 included all of the marsh from Unknown Pass to Bayou Platte. Area 4 was bounded by Bayou Platte on the east and Chef Menteur Pass on the west. It was separated from Area 5, which included all of Alligator Point by a line extending from the angle made by the coast of Lake Borgne between the two areas extending perpendicularly to Chef Menteur Pass. Areas 4 and 5 were connected by only one short bayou. Each Area was further subdivided into 0.65 square kilometer "Squares" (Figure 14). Each month for 30 consecutive months, five sites in each of four Squares in each Area were sampled with a 1.8 meter cast net. The 6 mm bar monofilament cast net carried 4 kg of lead weights. The net throws were standardized as previously described. At each Site, the cast net was deployed until two different species were collected or until the net had been deployed five times. Abiotic data at each Site was collected including depth, Secchi depth, substrate type, salinity, specific conductivity, dissolved oxygen and percent saturation, water temperature, presence and type of submersed aquatic vegetation, the type of location (pond or bayou), tide state, air temperature, wind direction and velocity, cloud cover, and date and time of day. A total of 2340 Sites were sampled in this manner. An additional 43 Sites were sampled with a 1 meter square aluminum throw trap, 0.8 m deep. At each Site the throw trap was deployed over the side and emptied by the use of a 1 m square, 1 mm bar net on an aluminum frame that just fit within the throw trap.

The third approach was to sample sites where YOY had been previously reported or that were suggested by members of the public interviewed for this purpose. Sites in Port Sulphur, Barataria Bay, on State Highway 1 around Leeville, Biloxi Marsh, Hopedale, Grand Isle, Port Fouchon, Myrtle Grove, salt marsh south of Highway 90 east of the Rigolets Pass, and the Bonne

Carre Spillway, were all sampled multiple times during different seasons with a 1.8 m radius cast net with 6.5 mm bar or a similar 1.3 m net, whichever was appropriate for the site.

The fourth approach was to employ the assistance of members of the Louisiana Tarpon Club to look for YOY in and around Morgan City and south to the GOM. Photographic evidence was requested along with GPS coordinates for all YOY collected and several trips were made to investigate captures and to document the sites.

The fifth approach was to examine sampling records from the Louisiana Department of Wildlife and Fisheries. Records of tarpon captures as part of routine finfish and shrimp monitoring programs from southeastern Louisiana were requested and reviewed. Records of several years of sampling conducted by the Nekton Research Laboratory (NRL) in the Pontchartrain Estuary were also reviewed.

In one locality where YOY tarpon occurred, I conducted a study to determine whether or not YOY tarpon were able to over-winter in these habitats. These YOY were collected from a roadside ditch by NRL scientists conducting another research project. After the initial collection, I sampled the ditch using a cast net every two weeks from October through early December, 2010. On the occasion of the original collection, I also collected any available prey species present. On subsequent collections from the ditch, 63 YOY tarpon were retained and sacrificed for the purpose of examining their diets. Stomach contents were examined and identified to the lowest possible taxonomic level. Each time the ditch was sampled, collected fish were weighed and fork length was measured. Temperature and salinity were measured using a Yellow Springs Instrument Professional Plus. As it was not practical to continuously monitor water temperatures

in the ditch, air temperatures were obtained from a nearby NOAA weather station on Grand Isle, Louisiana, located 15 km to the southeast.

#### Juvenile Tarpon:

Juvenile tarpon were considered to be those that were year 1 or older but had not reached sexual maturity. For the purposes of this study, any tarpon collected after February and greater than 300 mm was considered to be a year 1 fish. Attempts were made to document these fish in different habitats in an effort to see if they were capable of over-wintering in Louisiana waters. The search for juvenile tarpon was conducted using the following approaches. I attempted to determine the types of habitat in which juvenile tarpon occur by visiting different areas throughout the Gulf of Mexico where juveniles had been collected previously. These efforts included collection efforts in Venezuela, Columbia, Trinidad, Costa Rica, the Yucatan Peninsula, and south Florida and the Florida Keys. Unfortunately, these efforts proved singularly unproductive; collecting juvenile tarpon, even where they were supposed to be common, proved difficult. In the few instances when juvenile tarpon were collected, the observed habitats varied considerably among collection sites.

The first approach was to sample areas where YOY tarpon were collected on previous occasions. This sampling was conducted through the winter. In addition, the search for juvenile tarpon was part of all of the sampling I conducted for all other research purposes during my thirty months of sampling. I conducted most of this sampling with a 1.8 meter cast net as previously described though occasionally in narrow shallow ditches I used the 1.3 m net. The second approach was to seek the help of sports divers and spear fishers from the Louisiana

Council of Underwater Dive Clubs (LCUDC) and anglers from the Louisiana Tarpon Club. Members of the Louisiana Tarpon Club were asked to report any landings of small tarpon that could be considered juveniles. Unfortunately, the tarpon anglers target larger adults and generally only fish from July until September. Members of the LCUDC were asked to capture small tarpon and, in particular, were asked to capture the smallest examples that they could collect. The divers were able to conduct searches for all 12 months of a year and to sample a large number of oil production platforms in the GOM extending from the South Timbalier Block leases to the Main Pass Block leases. All collected juvenile tarpon were photographed, weighed and measured (fork length) and their otoliths removed. Block number, depth, temperature, date and time, and name of diver were recorded for each fish where provided. On two occasions we were able to video the fish underwater before and after they were collected and returned to the surface.

# Adult and Spawning Capable Tarpon:

As with other age-classes of tarpon, I decided at the beginning of my study to sacrifice as few adult tarpon as possible while at the same time attempting to document spawning capable fish. Volunteers attempted to collect adult tarpon during the spawning season from March through early July and thereafter to document that spawning had ended. Our volunteer anglers were encouraged to practice catch and release. In addition to the tarpon provided by the volunteer anglers, several fishing rodeos and tournaments were attended and fourteen of the tarpon brought to the dock were dissected, sexed, and samples of the gonads were preserved in 10% buffered formalin for histologic examination. All preserved gonad samples were submitted

to Nancy Brown-Peterson at Gulf Coast Research Laboratory for histologic examination. Where possible, otoliths were obtained, labeled and stored dry. Fin clips and scales were obtained and stored in 95% ethyl alcohol. Dried scales were also obtained and stored. Date and location of collection, angler or diver's name, and any other pertinent information was recorded when it was available. It soon became obvious that anglers and divers were reluctant to reveal the locations of their "favorite fishing spots". When information concerning the collection location was felt to be dubious or was not available it was recorded as "NA".

Adult tarpon are common in the northern GOM in the summer. During the months of December through June, the LCUDC was asked to capture only two tarpon each month, if they were found, in excess of 45 kg (100 lbs.) only. This request was made for two reasons. The first was to document the presence of adult tarpon on the Louisiana coast during the winter months to document overwintering. The second was to document adult tarpon during the spawning season with the plan to capture spawning capable adults. This search was conducted during 2011 and into 2013. In addition, I asked members of the LCUDC for any videos that they had taken of tarpon during the winter. When I obtained these videos, I interviewed several members present on the dive to verify the veracity of the video.

The Official Spearfishing Records of the State of Louisiana maintained by the LCUDC were examined to determine when the largest tarpon were collected. These are public records, maintained by the LCUDC under strict requirements for certification including witnesses and certified scales. I also examined the records of the Hell Divers Spearfishing Club and several other clubs for certified collections of tarpon.

Numerous reports were received of adult tarpon along the south shore of Lake Pontchartrain during the summer and fall of 2009. During 2010 and 2011, more than twenty trips were made each year to the south shore of the Lake in search of adult tarpon from September through October. Tarpon frequently roll on the surface of the water when it is quiet and calm. The best way to find them is to sit quietly and watch for them in areas they are known to frequent. Blindly fishing for them is not productive in large bodies of water. My procedure was to travel the shoreline of the southeastern and northeastern shore of Lake Pontchartrain, in particular off Little Woods and Goose Point, both areas where local anglers often fish for them. My boat is an 18 foot, Hell's Bay Marguesas with a 115 hp outboard engine. It is ideal for this work as it is light weight and quiet. I would stop frequently, turn off the engine, and allow the boat to drift for at least fifteen to thirty minutes through the areas where the tarpon had been reported. During this time, the surface was observed for rolling tarpon with 8.5 x 40 binoculars. Similar trips were made to Chef Menteur Pass and Rigolets Pass for this purpose during routine sampling trips during 2010, 2011, and 2012. These two passes are the two main entrances into Lake Pontchartrain from Lake Borgne and tarpon moving in and out must travel through one of these two passes.

From spring 2010 through February 2013, I distributed flyers to commercial bait sellers, shrimp trawlers, and members of the public requesting reports of any sightings or captures of adult tarpon in the Lake Borgne and Lake Pontchartrain Basins. All received reports were followed up with direct interviews and attempts were made to obtain any tarpon collected.

### **Results:**

The presence of five major life stages of tarpon: leptocephali, YOY, juveniles, adults, and spawning capable adults in southeastern Louisiana was confirmed during multiple seasons in 2010 through 2012, with YOY documented in multiple locations. An experiment was conducted, *in situ*, with YOY to ascertain if they are capable of over-wintering in nursery habitat in southeastern Louisiana. Juvenile tarpon were confirmed in numerous locations including "hot water canals" and in nearshore waters of the GOM. Adult tarpon were ubiquitous during the late spring, summer, and fall. Video evidence of adult tarpon at offshore oil production platforms in the GOM, verified by members of the Louisiana Council of Underwater Dive Clubs (LCUDC), and State of Louisiana Spearfishing Records, documented the presence of tarpon in Louisiana waters in winter for the first time. Spawning tarpon have never been scientifically documented, nor is the location of tarpon spawning known. Three spawning capable tarpon were collected in Louisiana waters confirming spawning activity in Louisiana also for the first time.

### Leptocephalus:

A review of the records of samples collected by the NRL and R.V. Cavalla reveal that no tarpon leptocephali had been collected (O'Connell, personal communication). Leptocephali of lady fish (*Elops saurus*) were frequent in the samples, particularly during the late spring. These occurred as far west in the Pontchartrain estuary in the vicinity of the Bonnet Carre Spillway. Lady fish leptocephali were also reported from Bayou St. John (Smith, personal communication).

No tarpon leptocephali were found in the museum samples of the NRL. However, a single leptocephalus was found in the Tulane Museum of Natural History - Royal D. Suttkus Fish Collection samples (Forman, personal communication). This leptocephalus was collected by Wayne Forman, then a marine biologist, during routine surveillance sampling in the Freeport Canal in 1976, approximately 15 km from the GOM. Mr. Forman was interviewed and reported that this specimen was the only one that he ever collected.

No leptocephali were collected during the sampling of the New Orleans Land Bridge sites. No leptocephali were reported in the LDWF data that I received.

YOY:

I received 25 calls in response to the flyers. One was from a fisher who had collected a skipjack herring (*Alosa chrysochloris*) in Lake Borgne and believed it to be a tarpon. Another was from a man fishing in the Bonnet Carre Spillway after the Spillway opening in 2011. He had collected several American gizzard shad (*Dorosma cepedianum*) and believed them to be tarpon. Two calls were investigated and found to be lady fish (*Elops saurus*), one from Lake Pontchartrain by a fisher and one from Rigolets Pass. Only one call was received for YOY. This was from a fisherman in Myrtle Grove, Louisiana, who had caught one in the summer of 2010 in a cast net and had frozen and saved it. He had seen the flyer and obtained my contact information from a friend who was familiar with my research. I had the name "Tarpon Research" painted on the side of my boat; several fishers that I ran into asked me about my work and several reported seeing my flyers and wanted to know more about what I was doing and why.

In the spring 2011, a YOY tarpon was collected in Myrtle Grove, Louisiana (Figure 15). The tarpon was collected in the marina at Myrtle Grove and kept frozen for one year before being released to me. A second YOY was received from Bayou Black in Gibson, Louisiana. This site was a "hot water" discharge canal for a petroleum processing plant. Digital photographs and several specimens were obtained from members of the Louisiana Tarpon Club who had found tarpon of several age classes in this fresh water bayou. This site was more than 60 km from the



Figure 15: Map of YOY tarpon collections and LDWF tarpon collections. The YOY captures from 2010 – 2012 are shown in pink. The Louisiana Department of Wildlife and Fisheries captures as part of routine finfish and shrimp monitoring programs, 1990 – 2010, are shown in blue. YOY were found along the entire coast as far as 60 km from the Gulf of Mexico.

GOM. A visit to this site resulted in the hooking of several tarpon of several age classes. Only one was collected and it was not a YOY, as it was over 60 cm standard length (SL). Several

fishers were interviewed at the site and confirmed that tarpon had been present there for several years, and that YOY were frequently caught there in cast nets. As a result of this visit, several YOY were reported from the vicinity of Fourleaque Bay, collected in October 2012 by a fisherman cast netting for bait behind a dam on an "oil-company canal" (Figure 15).

In October 2010, an NRL field crew collected five YOY in a gill net while conducting sampling operations in a roadside ditch along Louisiana Highway 23 not far from the Myrtle Grove site (Figure 15). YOY were also collected at this site in 2011 and 2012. One call to a member of the Hell Divers Spearfishing Club, which was reported, did come from a man who had seen a flyer. He delivered a 25 cm YOY that had been collected in 2010 in Myrtle Grove, Louisiana, and kept in a freezer.

The Louisiana Department of Wildlife and Fisheries (LDWF) conducts extensive sampling as part of routine shrimp and finfish monitoring. Over the 20 year period from 1990 to 2010, the LDWF data contains records of 14 YOY (Table 9, Figure 15). All were collected using gillnets or otter trawls. Eight of these were collected at the same location in Bayou Rambio during five different years. Two were collected on different dates during the same year at Bayou Bell. YOY were collected by NRL field crews at Port Sulphur during three successive years and on four occasions during one year. Similarly, YOY were collected during two years in Hopedale, but not at the same location. The Aquarium of the Americas received over 100 YOY Table 9: Young of the year tarpon (YOY) collected during 2010 – 2012 research in southeast Louisiana. Young of the year tarpon collected by Louisiana Department of Wildlife and Fisheries, 1990 – 2010 during routine finfish and shrimp monitoring surveys.

Young of the Year Tarpon

Date	Location	Number	Gear
8/10	Hopedale	100-	Cast net
5/10-6/12	Bayou Black, Gibson	20-	Hook
10/5-12/8/10	Port Sulphur	68	Cast net
9/26/11	Bleux Island Plant	1	Cast net
10/15/11	Port Sulphur	3	Cast net
11/2/11	Land Bridge	1	Cast net
8/11	Grand Isle	50-	Cast net
9/11	Myrtle Grove	1	Hook
9/16/12	Hopedale	5	Hook
10/1/12	Port Sulphur	1	Electro-fishing
10/30/12	Lost Lake/Fourleague Bay	10-	Cast net

Louisiana Department of Wildlife and Fisheries YOY Tarpon captures

Date	Location	Number	Gear
11/14/1990	Barataria Bay	1	Gill Net
12/4/1990	Bayou Rambio	1	Gill Net
10/7/1991	Bayou Rambio	1	Gill Net
12/1/1995	Bayou Rambio	1	Gill Net
4/21/1998	The Pen, Barataria	1	Otter Trawl
4/6/2004	Bell Bayou	1	Otter Trawl
4/28/2004	Bell Bayou	1	Otter Trawl
10/4/2010	Bayou Rambio	1	Gill Net
10/11/2010	Lake Quitman	1	Gill Net
10/27/2010	Bayou Carlin	1	Gill Net
11/18/2010	Bayou Rambio	4	Gill Net

in 2010 which were collected by cast net in a ditch behind a marina in Hopedale, Louisiana (personal communication, John Hewitt and Glen Sanchez). In the fall of 2012, a member of the

NRL staff hooked seven YOY while fly fishing in the Biloxi Marsh (Patrick Smith, personal communication). At Bayou Black, YOY tarpon were collected yearly for four successive years for which there are records.

YOY tarpon were collected by a cast netter in the marsh north of Fourleague Bay 30 October 2012 and by the LDWF just to the north of that site (Table 9). Current reversal in the



Figure 16: The only YOY collected on the New Orleans Land Bridge out of 65,000 fishes and decapods 31 October 2011. The lower lobe of the caudal fin was lost prior to collection. At least 6 or 7 others were in the school. This tarpon may have not been able to swim fast enough to avoid the cast net.

bayous and canals of this area occurs during tropical storms and preceding winter storms. This may in part explain the presence of YOY and juvenile tarpon in these areas and farther north in Bayou Black.

The survey of the New Orleans Land Bridge resulted in the capture of more than 60,000 fishes and decapods. Of these organisms, only one was a YOY tarpon (Figure 16). On 2 November 2011 as the cast net was deployed, a school of about 7 to 9 YOY was noted just below the surface in shallow water on the side of a wide, deep bayou. Only one was collected. Further efforts to capture tarpon in that location were unsuccessful although the cast net was thrown over 25 times in the area where the school was observed. During the summer of 2010, several trips were made to the marsh south of U.S. Highway 90 on the east side of Rigolets Pass. No tarpon were observed or netted in this area despite extensive efforts and four trips into this area (Figure 15).

Following the capture of the YOY tarpon north of Port Sulphur, Louisiana, I conducted a study to determine whether YOY could overwinter in this location. Five YOY were originally collected at this location on 5 October 2010 by a Nekton Laboratory field research team sampling with a rope seine (a 5 X 2 m seine pulled with ropes attached to the brailles). The roadside ditch was on average 1 m deep and drained into a marsh 1 km to the southwest. This marsh, in turn, connected into Barataria Bay and thence into the GOM, approximately 26 km to the south. The salinity was 8 and the temperature was 27° C on the first and second visits on 19 and 26 October 2010. At the time the first YOY were collected, three rope seine hauls were made with the goal of identifying other species occurring in the ditch.

Samples were collected from the ditch on four successive trips on 26 October 2010 and on 5, 11, and 18 November 2010. On each trip to the ditch, YOY were collected with a 1.8 m cast net with either 6 or 7 mm bar. Between 14 and 17 YOY were collected on each trip and euthanized in an ice-water mixture. All tarpon were weighed and measured. Stomach contents

were also weighed and measured, with all items identified to the lowest possible taxonomic level.

While YOY were collected on each of the first four trips to the ditch, none were collected on the fifth trip on 8 December 2010. Between the fourth and fifth trips, water temperature had decreased to 10° C, the theoretical fatal temperature for YOY (Rickards, 1968 ; Howells, 1985; Jud et al., 2011). Furthermore, the weight of the contents of the tarpon stomachs decreased as water temperature fell to 17° C after the first trip. While no tarpon were collected on this fifth trip, cold shocked fishes of other species were present.

My visits to assess potential "typical" YOY and juvenile tarpon nursery areas in the Florida Keys, Columbia, Trinidad, Costa Rica, and Panama yielded no obvious consistent characteristics among sites, with the exception being that water temperatures were consistently above 20° C (although this was likely a result of the time of the year at which the visits were made). Salinities ranged from less than 1 to 34. Depth varied from less than 1 m to greater than 2 m. All of the areas where YOY were observed were in mangrove swamps and none were observed in the open ocean or tidal passes.

# Juvenile Tarpon:

In order to ascertain whether or not YOY tarpon were able to over-winter in Louisiana, I conducted a search for juvenile tarpon in multiple known habitats in other areas. My goal was to better understand the habitat requirements of juvenile tarpon and when they leave the inshore nurseries in areas where juvenile tarpon were known to occur. On the bay side of the middle Florida Keys, juvenile tarpon were found in schools of 20 or more in shallow protected waters

around small mangrove islands. They were not observed with the large schools of adult tarpon migrating through deeper waters. They are frequent in residential canals on both sides of the main islands year round. On the Yucatan Peninsula at Ascension Bay, large schools of juvenile tarpon were observed in the shallow mangrove lagoons west of Pajaros, but not on the coral flats on the ocean side of the peninsula. On the Caribbean coast of Coast Rica, juvenile tarpon were collected in the lagoons and canals of the Rio Colorado. Similar observations were made in Trinidad, Puerto Rico, Venezuela, and Columbia. Typically in these nurseries, YOY and juveniles were common in protected inshore rivers, canals, and lagoons in mangrove marshes.

I also visited the west coast of Panama in search of juvenile tarpon to learn more about possible habitats. Large numbers of juvenile tarpon were observed to be feeding on schools of mullet (*Mugil* spp.) in the Bayano River on 20 December 2009. One 11.5 kg tarpon was collected and released. The average water depth was 3-4 m, salinity was less than 1, and the temperature was 29.4° C. No YOY were observed in the river although there were local reports of their occurrence in shallow tributaries. Juvenile and adult tarpon were also reported in the river throughout the year, although adults were most common in the spring and during periods when coastal waters are cold.

Wherever juveniles were collected or observed in these areas, their habitat appeared to share several common features. The water was protected, shallow, and warm. It was generally deeper than YOY habitat, and the salinity varied from fresh to euhaline. I attempted to find analogous habitat in southeastern Louisiana, but this was difficult in that the dominant habitat is composed of black needle rush (*Juncus roemarianus*), smooth cordgrass, and saltmarsh cordgrass (*Spartina* spp.) marsh, not mangrove swamps. Furthermore, water temperatures as provided by the U.S. Geological Service in southeastern Louisiana coastal marsh, frequently

drop below 10 ° C in the winter (Jud et al., 2011; USGS, 2013). In spite of the presence of YOY in southeastern Louisiana, the seasonal cold weather meant that the life cycle of tarpon may have ended here.



Figure 17: Bayou Black juvenile tarpon collected in hot-water canal in March 2012. Though more than 60 km by water from the Gulf of Mexico, juvenile tarpon such as this specimen are routine collected on hook and line there. The year round water temperature in the canal is  $32^{\circ}$  C to  $40^{\circ}$  C from the output of a steam petroleum processing plant. The water was very clean with a salinity of 2. Blue crabs, catfish, Gulf menhaden, bay anchovies, blue gill, and black bass were collected here with a cast net.

Initially, the only place I was able to find juvenile tarpon in Louisiana was in Bayou Black near Gibson, Louisiana. Members of the Louisiana Tarpon Club reported catching YOY and juvenile tarpon there for several years (Figure 17). Photographs sent to me of tarpon collected there by members of the club strongly suggested several different age cohorts based upon obvious size differences. The majority of these fish were collected in a "hot water" canal that drained a petroleum processing plant. At least five of these fish were tagged and released, but there were no reported recaptures (Richard Hawthorne, Louisiana Tarpon Club President, personal communication). A visit to the area on 29 March, 2012, confirmed the presence of juvenile tarpon in large numbers. The water temperature was 32° C and salinity was 2.0. I hooked several juvenile tarpon with spinning gear and collected one (Figure 17). I interviewed a plant foreman of the North Terrebonne Gas Plant who informed me that the temperature in the outflow canal was closely monitored and was maintained at 32 to 40° C throughout the year. On



Figure 18: Juvenile tarpon collected 25 December 2011 in hot-water canal, Bayou Bienvenue, New Orleans. Seven more were collected the week before Christmas 2012, by the same angler at the same location. A steam power generation station outputs warm water into this canal throughout the year. The railroad right of way and the Paris Avenue Bridge are visible in the background.

Christmas Day 2011, a juvenile tarpon was reported collected in the Hot Water Canal off Bayou Bienvenue east of New Orleans. The fisher reported that he caught several but released them all alive. He collected seven more at the same location during the first week of December 2012.

Several more juveniles were collected by anglers on Christmas Day 2011, in the hot-water discharge canal at Bayou Bienvenue in eastern of New Orleans. Seven more were collected there during the first week of December 2012. These tarpon were collected in warm water refugia similar to that reported by Jim Franks (Franks, personal communication). Franks reported finding juvenile tarpon during the winter in a hot water discharge canal in Back Bay Biloxi during several successive years. These fish were caught in a specific small habitat unlike that which existed along the remainder of the coast. Water in the discharge canal was much warmer than that in surrounding areas and remained so throughout the winter (Franks, personal communication). Consistently collecting juvenile tarpon in two separate temperature refuges gives support to the hypothesis that they cannot over-winter in natural coastal habitats, but rather require artificial warm water canals. Another juvenile was collected in the marsh adjoining the south west side of Lake Borgne in September 2011. The fisher reported that this juvenile tarpon was in a school with several others of the same general size in a narrow but deep bayou within a few hundred meters of Lake Borgne. Based on this evidence, I altered my strategy to find more juveniles.

In the interest of conservation, I asked volunteers to release any tarpon collected. Volunteers were asked to photograph and weigh any juvenile tarpon collected, and note the location and date of capture in addition to any witnesses. All digital images were carefully examined to insure that the fish were tarpon. I also personally interviewed each volunteer along with any witnesses to verify date and location of capture. Any photographs that could not be verified were excluded. In all, more than 35 juveniles were identified and over 10 digital images were received. In hopes of personally observing juvenile tarpon in the GOM, I participated in a dive trip with members of the Hell Divers Spearfishing Club to oil production platforms in the

South Pass lease blocks and in West Delta. During three dives conducted 5 August 2012, no tarpon were observed.

Volunteer divers were asked to obtain video and to capture juvenile tarpon in the vicinity of oil production platforms in the GOM. During summer 2011 and through fall 2012, over thirty juvenile tarpon were collected (Table 10, Figure 19). All juvenile tarpon were weighed, measured, and the location of capture, name of the volunteer, water temperature, and



Figure1 19: Map of juvenile tarpon captures, 2010 - 2012. Most of the captures appear concentrated in a limited area because divers tended to return to the same productive areas in West Delta. Juvenile tarpon were collected across the entire coast and inland at the two "hotwater" canals in Bayou Black and Bayou Bienvenue. The juvenile collected on the west side of the Biloxi Marsh was the only one collected in Lake Borgne. None were collected in Lake Pontchartrain. The large numbers of juveniles are in close proximity to the YOY of Barataria Basin.

depth were recorded when available. Otoliths and fin clippings were obtained from all juveniles and preserved for future aging and DNA studies. The last juvenile tarpon collected as part of this Table 10: Juvenile tarpon collected in 2010 - 2012 by date and location. Gear type, fork length, weight, approximate depth and temperature given where available. Most of the juveniles were collected by divers. Many of these fish were taken in schools of more than 10 similar sized tarpon.

		Juvenile '	Tarpon C	Collected			
Date	Location	Number	Gear	Fork Length	Weight	Depth	Temp
				(cm)	(kg)	(ft)	(° F)
Max 10	Southwest Pass	101	Uook	NΙΛ	NΛ	NΙΛ	NΙΛ
5/31/2011	Jelly Bayou Black	10+	Hook	NA NA	NA NA		NA 02
6/5/2011	S Timbalier 21		Spear	NA NA	6 82	U NA	92 NA
6/6/2011	Bayou Black		Spear	NA	0.82 NA		02
7/7/2011	Lake Borgne		Hook	NA NA	8 73	5	92 NA
9/16/2011	S Timbalier 67		Spear	76.20	5.82	30	80±
9/16/2011	Shin Shoal 186		Spear	93.98	9.52	35	82-86
10/14/2011	WD 79		Spear	123.10	25.09	75-100	75-78
10/14/2011	WD 79		Spear	118 41	21.00	75-100	75-78
10/14/2011	WD 79		Spear	112.08	15 55	75-100	75-78
10/14/2011	WD 79		Spear	102.24	14.18	75-100	75-78
12/25/2011	Hot-water Canal	2	Hook	NA	NA	15	NA
3/29/2012	Bayou Black	-	Hook	NA	NA	0	92
5/15/2012	S. Timbalier 72		Spear	NA	13.68	30	NA
6/3/2012	WD 79		Spear	88.39	6.68	58	74-77
6/3/2012	WD 56		Spear	114.55	17.27	25	74-77
6/3/2012	WD56		Spear	118.11	18.55	25	74-77
6/3/2012	WD56		Spear	86.61	7.64	25	74-77
6/3/2012	SE165		Spear	71.63	3.64	50	74-77
6/3/2012	WD30		Spear	139.70	27.05	0	80
6/3/2012	WD30		Spear	96.77	9.95	0	80
7/6/2012	WD 32		Spear	80.00	5.94	45 - 50	NA
7/6/2012	WD 32		Spear	104.80	13.60	45 - 50	NA
7/21/2012	S. Timbalier 80		Hook	NA	4.77	60-80	82
8/20/2012	WD 58		Spear	89.00	9.15	25	NA
8/20/2012	S. Timbalier 54		Spear	101.00	12.39	50	NA
Table 10:							
Continued)							
8/20/2012	S. Timbalier 54		Spear	96.50	10.85	50	NA
8/20/2012	Grand Isle 19		Spear	118.00	17.05	50	NA
9/21/2012	WD 44		Spear	66.68	3.65	45	80

(Table 10: Continued)							
9/24/2012	WD 31		Spear	107.95	15.00	40	NA
9/24/2012	WD 31		Spear	100.97	11.50	40	NA
9/30/2012	WD 79		Spear	109.50	13.18	NA	NA
9/30/2012	WD 26		Spear	85.00	7.58	NA	NA
9/30/2012	WD 26		Spear	79.00	5.10	NA	NA
9/30/2012	WD58		Spear	87.00	8.00	NA	NA
9/30/2012	WD58		Spear	77.00	5.16	NA	NA
12/9/2012	WD 86		Spear	92.71	9.15	60-80	NA
12/9/2012	WD60		Spear	91.44	9.55	60-80	NA
12/9/2012	Hot-water Canal	9+	Hook	NA	NA	15	NA

study was 10 December 2012. Two juvenile tarpon were collected at different oil production platforms in the Barataria Bight on the same day. The diver in question reported observing a few additional juveniles but did not record the number. He did see one adult tarpon but was unable to get close to it. This is the first known documentation of juvenile tarpon in offshore Louisiana habitats during winter months.

# Adult and Spawning Tarpon:

Documentation of adult tarpon began in Lake Pontchartrain in early fall 2010 and continued yearly through 2012. I made approximately 10 trips during the end of August, September, and October of each year, in early morning or late evening to the "tarpon holes" on calm days, beginning the week prior to Labor Day. The "tarpon holes" are three deep dredge holes approximately 400 m from the shoreline along the south shore of Lake Pontchartrain between New Orleans Lake Front Airport and Irish Bayou. This has been the traditional time area anglers fish Lake Pontchartrain for tarpon at these sites. Reports of tarpon being collected by anglers in this area were plentiful during the years preceding Hurricane Katrina, and I hooked and lost two tarpon at this location just prior to Hurricane Katrina on 13 August 2005. For this research, I drifted through these areas for 15 to 30 minutes at each location and observed the area for "rolling" tarpon in the early mornings beginning one to two hours after dawn or beginning two hours before sunset. I have observed undisturbed tarpon rolling on the surface in quiet water at these times of the day for over thirty years. Tarpon "roll" in a manner that is easily identified once it is observed, as their dorsal and filament fins are usually easily seen and their tail and roll is quite distinctive from the occasionally confused roll of both *Atractosteus spatula* (alligator gar) and *Lepisosteus oculatus* (spotted gar), which are present in these waters. I used a quiet "Florida Flats Boat", an 18 foot total length Hell's Bay Marquesas with a poling platform above the engine which afforded an excellent view of a wide expanse of the water surface. No tarpon were observed in Lake Pontchartrain on any of these trips. During the period of this research, I was unable to document any landings of tarpon in Lake Pontchartrain.

During my research, the first documented adult tarpon in the Pontchartrain Basin was collected in Chef Menteur Pass on 25 November 2012, in the wing net of a shrimp boat pushing between the Highway 90 Bridge and the railroad bridge to the south. This tarpon was released alive but no measurements were obtained (reportedly, the crew stated that they were concerned should the capture be recorded, they would be required to put tarpon excluder devices on their nets).

Tarpon are plentiful during in the GOM during the summer and fall but are not known to occur in the northern GOM during the winter. A lengthy search and discussions with members of the Louisiana Council of Underwater Dive Clubs produced three videos showing tarpon in large numbers, congregating at oil production platforms in Main Pass Block 299 on 14 February

1999, 21 February1999 and at Main Pass Block 296 on19 March 2005. Several divers present on each of these dives were interviewed to authenticate the video. In addition, the records of the largest tarpon collected by divers were obtained from the LCUDC official lists. These data confirm that 9 of the 11 largest tarpon collected in Louisiana by spear fishers were collected during February and March (Table 11). It should be noted that these data do not include all of the tarpon collected but only the largest.

Table 11: Louisiana Council of Underwater Diving Clubs Official Records of Tarpon as of May 2006. Only the largest tarpon reported and officially weighed are listed. All but two were collected in February and March 1999 and 2004. Weights are given to tenths of a pound. (S), Free Diver; (ST), Sea Tigers; (HD), Hell Divers; (SS), Sea Scamps.

162-8 John Hagmann(SS) 2-99 160-0 Allen Walker(ST) 2-99 153-0 Steve Hartley(ST) 2-99 153-0 John Hagmann(SS) 2-99 153-0 Steve Hartley(ST) 3-04 150-8 Stan Smith(HD) 2-99 147-6 Allen Walker(ST) 2-99 146-0 John Hagmann(SS) 2-99 142-0 Louis Rossignol(HD) 2-99 142-0 Louis Rossignol(HD) 6-02 (S)43-5 David Chaix(HD) 7-05

Tarpon are known to enter estuaries, coastal rivers, and lakes (Crabtree et al., 1995). All of the adult tarpon collected by the anglers for this research were in open waters of the GOM. All adult tarpon collected by the divers occurred in water immediately adjacent to offshore oil production platforms. Unfortunately, not all of the tarpon collected by fishers were able to be examined as not all of the fishers could be convinced to cooperate with this study and some of the distances were too far from New Orleans or too little notice was given to allow time to get to

the location where the tarpon were brought ashore.

Table 12: Adult tarpon collected 2010 – 2012 by volunteer anglers and divers. Only the Chef
Menteur tarpon was released alive. Spawning capable (SC) and spent (S) tarpon are labeled.
Most of the adult tarpon were obtained from fishing tournaments in the interests of conservation

Adult Tarpon Collected							
Date	Location	Gear	Fork	girth	Weight	Spawning Status	
	South Tim		Length (cm)	(cm)	(Kg)		
7/2/2011	60	Hook	177.8	93.98	58.6	F/SC	
	South Tim	110011	17770	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	0010	1,80	
7/2/2011	60	Hook	167.64	96.52	55.5	M/SC	
7/9/11	WD 58	Hook	187.96	44.75	68.2	F/S	
7/13/11	SP 86	Spear	157.48	83.82	42.5	M/SC	
7/13/11	SP 86	Spear	149.86	78.74	33.1	Μ	
7/28/11	Rodeo	Hook	NA	NA	45.2	Μ	
7/28/11	Rodeo	Hook	NA	NA	56.6	Μ	
7/28/11	Rodeo	Hook	NA	NA	30.0	Μ	
7/13/12	SP 37	Spear	186.69	93.98	43.9	Μ	
7/28/12	Rodeo	Hook	189.23	NA	75.0	Μ	
7/28/12	Rodeo	Hook	185.42	NA	70.0	Μ	
7/28/12	Rodeo	Hook	187.96	NA	68.6	Μ	
7/28/12	Rodeo Chef	Hook	167.64	NA	53.2	М	
12/11/7/12	Menteur Pass	Trawl	NA	NA	NA	NA	

Tarpon spawning activity is believed to occur in April and extend into the summer in the GOM (Harrington, 1966; Crabtree et al., 1992; Crabtree et al., 1995; Crabtree et al., 1997). Tarpon fishing generally begins on the Louisiana coast in early June and may be hampered by bad weather in May. During 2011 and 2012, 14 adult tarpon were collected and examined

(Table 12, Figure 20). No adult tarpon were reported by volunteer anglers until June 2011, but these specimens were released and not returned to shore. The first angler-collected adult tarpon was brought in 2 July 2011 to the Port Fouchon Tarpon Rodeo (Table 3). This tarpon, a large



Figure 20: Map of winter adult tarpon and spawning capable=tarpon. The location of the spawning-capable tarpon and the winter tarpon are marked along with the sole adult tarpon collected in the Chef Menteur Pass. Prevailing currents in the Barataria Bight would carry leptocephali from the capture site of the spawning-capable tarpon into the Barataria Basin and westward along the coastline.

female, was found to have large ovaries. They were dissected and fixed in 10% buffered formalin and submitted to Nancy Brown-Peterson at Gulf Coast Research Laboratory. This tarpon was the first spawning-capable female collected in the northern GOM. A male tarpon collected on the same day in the same area was also found to be spawning-capable as was a second male collected 13 July 2011. Another female collected on 8 July 2011 and brought
ashore on 9 July was found to have enlarged ovaries which appeared to contain eggs or mature follicles (Figure 21). These ovaries were in poor condition and were not fixed properly to allow



Figure 21: This post-spawn 150 lb. female adult tarpon was collected 22.5 km south of Fouchon, Louisiana. Large tarpon are routinely collected by sport fishers off the Louisiana coast. Though not always the practice, most are now caught and released. Unfortunately, the ovaries of this specimen had deteriorated and were not fixed well enough for histology. Though the photo is labeled July 8, this specimen was not received until July 9.

for histologic examination (Figures 22, 23). Although no other spawning-capable tarpon were collected after 9 July 2011, these were the first spawning-capable tarpon documented on the Louisiana coast and provide clear evidence of spawning activity. All of the spawning tarpon were from the west of the Mississippi River on the west side of the Barataria Bight. Despite the



Figure 22: Ovary from spawning-capable adult on the left showing post ovulatory follicle and various stage vitellogenic oocytes. Testis, on the right, shows germinal epithelium and spermatozoa but no spermatogenesis.



Figure 23: Preserved partial ovary of post-spawn 68.2 kg tarpon. Preservation was insufficient for histology in part due to the delay in dissecting the specimen. What appear to be eggs or mature follicles are visible in the specimen. Fish of this size are very common on the Louisiana coast from June through October. It is not unlikely that this fish had spawned there.

evidence of tarpon on the east side of the Mississippi River during the late winter, the Main Pass Area was not sampled in the summer.

#### **Discussion:**

All life stages of tarpon have been identified and confirmed in multiple locations in Louisiana. These data support my original hypothesis that tarpon are not only capable of spawning in Louisiana but can complete their life cycle in Louisiana waters. I was not surprised when no leptocephali were collected during my sampling (Crabtree et al., 1992). The lone leptocephalus collected in 1976 reveals that they are present in Louisiana, just not in large numbers. Leptocephali have been found in offshore waters along the west Florida coast in greater numbers (Crabtree et al., 1992). The fact that more were not found in this study is more a consequence of sampling location, lack of sufficient effort, and scarcity, and reflects similar findings in other locations (Shenker, 2006; Eldred, August 1968). Furthermore, tarpon larvae undergo metamorphosis to the juvenile stage at the time of inshore migration. This transformation is necessary for pelagic larvae to make the transition to demersal habitats (Ault et al., 2008). The cue for the induction of metamorphosis is not known, so leptocephali may undergo metamorphosis prior to entering the marshes upon encountering lower salinity nearshore waters. In this case, the transition from larva to YOY would occur well before they actually enter in shore marshes. Franks collected five leptocephali on the beach in Ocean Springs during the summer 2012, but none in the tidal sloughs where he collects YOY yearly (Franks, personal communication).

Unlike leptocephali, YOY tarpon were collected regularly along the southeastern shoreline of Louisiana during my study. Appropriate juvenile tarpon habitat is extensive in coastal Louisiana and while YOY were collected at one location for three successive years, they were also collected at several other locations over the years in the Barataria Basin. Again, the paucity of collections is more likely a result of sampling effort and poor reporting than absence of fish. From the available data, I was not able to define likely nursery habitat in Louisiana as YOY were collected in isolated, mesohaline ditches, open bays (e.g., Barataria Bay), and deep tidal sloughs in the marsh. They were not collected in the open GOM, or in the deep tidal passes opening into the GOM. It was not surprising either, that they did not over-winter in the roadside ditch. When the temperature dropped below 10° C, these juveniles either died or left the ditch. It is likely that they escaped to the near shore waters of the GOM where they became the following year's juveniles. This is where large numbers of them have been verified.

Considering the extent of sampling I conducted in Land Bridge habitats (2400 individual samples), it was surprising that only one YOY was collected. The Land Bridge is a large area with different microhabitats. It is not only possible, but likely, that more YOY were present. The finding of older juvenile tarpon in the hot-water canal less than 10 km to the west suggests that their presence in this area was not limited to the one that was collected. The second juvenile collected on the east side of Lake Borgne and the YOY hooked to the south suggest that they are present in this area of the marsh also. Juveniles and YOY are fast swimmers and are wary of anything passing overhead from a fishing line to a bird or a cast net (personal observation). It is probable that the small number collected is a more a result of gear avoidance than a scarcity of tarpon.

The finding of juvenile tarpon in "hot-water" canals indicates that they are capable of locating and over-wintering in these thermal refugia. However, the finding of large numbers of juvenile tarpon in nearshore waters of the GOM, especially in the Barataria Bight, suggests that they are capable of over-wintering in these waters where temperatures may be significantly warmer and further suggests a connectivity between the YOY fish found in Barataria Basin and the juveniles in the Bight. Juvenile tarpon were collected in December 2012 in Barataria Bight, which suggests that they are over-wintering here and not migrating out of the area. The difference in sizes suggests differences in age cohorts and further suggests that the juveniles are remaining in nearshore coastal waters for several years.

Videos obtained during the winters of 1999 and 2005 showing large numbers of adult tarpon at offshore oil production platforms in the GOM demonstrates that large numbers of adult tarpon are in Louisiana waters in the winter and do not migrate out of the GOM in winter as has been suggested. These observations are supported by the LCUDC Spearfishing Records which show that the largest tarpon collected in Louisiana, with the two exceptions, were collected in February and March. The finding of adult tarpon present in Louisiana waters during the late winter and early spring implies that they are present here during the spawning season (Crabtree et al., 1992). It is not known exactly where tarpon spawn as spawning has never been scientifically documented (Ault et al., 2008; Baldwin and Snodgrass, 2008). It has been assumed from the work of Crabtree (1992, 1995) and Smith (1980) that spawning occurs off the edge of the continental shelf, many kilometers from shore. Also, nearshore spawning has been hypothesized in Puerto Rico (Zerbi et al., 2001). My identification of a spawning-capable female, the first reported in the northern GOM, and two spawning-capable adults in early

summer is therefore not surprising (Stein et al., 2012). Unsuccessful efforts by others to document spawning are probably a result of waiting until too late in the season.

In Louisiana, spawning is now believed to occur near the coast in the vicinity of the area where the spawning-capable tarpon were collected. This is consistent with the suggestion that tarpon spawn in relative proximity to juvenile habitat (Crabtree et al., 1992). Flow studies using the Finite Volume Coastal Ocean Model show that spawning in the area just to the west of the Barataria Bight would result in leptocephali being carried into the Barataria Bight and then farther westward along the shoreline (Georgiou, personal communication). This spawning activity may be the source of the YOY collected in the Barataria Basin. The occurrence of YOY during multiple years suggests that this is not a rare event. Spawning in the areas of the videos of winter tarpon (Main Pass Block 305 and Main Pass Block 299) would result in the presence of YOY in the area of Ocean Springs, Mississippi, where they have been reported over several successive years (Georgiou, personal communication). Furthermore, the same model suggests that tarpon spawning on the Louisiana coast could result in the transport of leptocephali onto the West Florida Shelf.

More juvenile tarpon were collected than YOY which was likely a result of a more concentrated effort. Nevertheless, juveniles were collected along the entire coast and inshore. The proximity of the juveniles found in the Barataria Bight to the YOY found in the Barataria Basin appears to be more than coincidental. My results suggest YOY cannot over-winter in inshore nursery areas except in mild winters. The lack of YOY in nursery areas during colder months suggests YOY are moving offshore with the onset of cold weather and maturing in the warmer nearshore waters. The discovery of spawning activity in this area combined with the FVCOM model runs further suggests that all three life stages are connected here.

Tarpon are highly migratory, and it is unknown whether or not there are one or multiple tarpon stocks in the GOM (Garcia De Leon et al., 2002; Ault et al., 2009). Tarpon from Mexico have been demonstrated to migrate to Marsh Island, Louisiana, in the summer and tarpon from Florida have been shown to migrate to this location as well (Ault et al., 2008; Ault et al., 2009). Over-wintering Louisiana tarpon may have originated from either location or represent members of a resident stock. Whatever the case, gene flow appears to be extensive within the GOM and there is one large meta-population with little divergence except perhaps in South Florida and Costa Rica (Garcia De Leon et al., 2002; Blandon et al., 2003; McMillen-Jackson et al., 2005; Seyoum et al., 2008). Genetic studies involving adult Louisiana tarpon have not included sufficient numbers to support any conclusions concerning a separate stock.

Unfortunately, at this point there can be no reasonable approximation of the number of tarpon produced on the Louisiana coast each year nor can the contribution to Gulf stocks be estimated. Little is known about such issues as density-dependent survival of juvenile tarpon or the influence of stochastic abiotic events on recruitment. In terms of life history strategy, tarpon are "extreme periodic strategists" (Winemiller and Dailey, 2002). Periodic-strategists benefit from large adult body size and delayed maturity by producing huge numbers of pelagic eggs. Survival rates of the early life stages are extremely low, although the few that ultimately reach the proper environments may grow rapidly. There is a trade-off between this high early life stage mortality and the survival of at least some of the larvae that reach an appropriate location for settlement (Winemiller and Rose, 1992). Survivorship of early life-stages in most years approaches zero and in some years it is high. Stochastic effects produce boom or bust recruitment years and this probably, in part, has the most influence on the abundance of YOY in coastal Louisiana marshes. Even in years of few YOY, nearshore schools of juvenile tarpon

appear to be comprised of several year-classes. At this point, it is unknown whether or not these schools are dominated by particular year-classes.

Strong year-classes have been documented to occur periodically with other species such as red drum (*Sciaenops ocellatus*) over broad areas as a result of factors acting over large spatial scales (Scharf, 2000). It is these unknown factors which may be responsible for the boom or bust year-classes by influencing the survival of larval fishes. There is little that fisheries managers can do to influence the survival of this life stage, however, preservation and protection of essential YOY and juvenile habitat has the potential to greatly enhance the survival of this stage and to ultimately improve tarpon stocks. One simulation of population dynamics demonstrated that a "1% increase in late juvenile survival increases age-20 abundance by 2224% (more than tenfold) over baseline (Winemiller and Rose, 1992). It may be hypothesized that the loss of Louisiana coastal marshes has decreased the survival of YOY and juvenile tarpon and decreased recruitment into Gulf stocks. My research was not able to test this hypothesis.

Based on the evidence presented in this paper, it is a reasonable conclusion that tarpon are reproducing in the northern GOM and that this spawning activity is responsible for the YOY tarpon that I have found in the coastal marshes. It is also a reasonable assumption that juvenile tarpon found in the Barataria Bight have moved from the Barataria Basin in response to falling temperatures inland. They are successfully maturing in nearshore coastal waters. The numbers that I have found here suggest that Louisiana is a significant source of tarpon that recruit to migratory coastal stocks. Any management efforts directed towards this important game fish must take these data into account.

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This work could not have been completed without the dedication and assistance of members of the Louisiana Council of Underwater Diving Clubs and, in particular members of the Hell Divers Spearfishing Club and the Sea Tigers who worked tirelessly to photograph, video, and capture tarpon during the last two years. I am also grateful to members of the Louisiana Tarpon Club and the numerous anglers who helped find the spawning-capable tarpon.

#### Chapter 4

Evidence of spawning capable tarpon (Megalops atlanticus) off the Louisiana coast

## Introduction:

Despite the fact that the tarpon (*Megalops atlanticus*) is a popular sport fish in the Gulf of Mexico (GOM) (Ault et al., 2008), little is known of its spawning behavior. Spawning *M. atlanticus* have never been documented and fertilized eggs have not been observed *in situ* (Ault et al., 2008). While it has been suggested that adult *M. atlanticus* move to deep water off the southwest coast of Florida and into the Yucatan Channel to spawn, the actual locations of spawning grounds remain unknown (Smith, 1980; Crabtree, 1995; Crabtree et al., 1997). Distribution patterns of larvae (leptocephali) have served as the basis for most of what has been inferred about the spawning areas (Smith, 1980; Crabtree et al., 1997). For example, leptocephali were collected from the southwestern GOM, the Yucatan Channel, and along the west coast of Florida, and based on their size, it was assumed that *M. atlanticus* spawned in nearby areas (Smith, 1980).

Histological examination of gonads has also been used to estimate the location of *M*. *atlanticus* spawning habitat. Females collected from the Florida Keys and Boca Grande Pass off the west coast of Florida and contained ovaries with post ovulatory follicles (POF) and advanced vitellogenic oocytes, suggesting *M. atlanticus* spawn in this region from April through July (Crabtree et al., 1997). Examination of gonads from *M. atlanticus* caught off the coast of equatorial Ceara State, Brazil suggested that spawning occurs there from October through January (de Menezes and Paiva, 1966). I report here the first evidence of spawning capable *M*. *atlanticus* off the coast of Louisiana in the northern GOM based on histological examination of gonads.

#### **Materials and Methods:**

Two large, sexually mature *M. atlanticus* (one female, one male) were collected by anglers about 30 km south of Venice, Louisiana on 2 July 2011. A second male was captured by anglers on 28 July 2011 from the same area. Fish were weighed (kg) and measured (mm fork length, FL) and gonadal tissue from each specimen was removed, weighed, and fixed whole in 10% neutral buffered formalin within 12 h of capture. Gonadal tissue was processed following standard histological techniques, embedded in paraffin, sectioned at 4  $\mu$ m and stained with hematoxylin and eosin. Reproductive phases and gamete stages were determined following Brown-Peterson et al., (2011).

#### **Results and Discussion:**

The female *M. atlanticus* weighed 56.8 kg and was 1778 mm FL. The first male weighed 55.5 kg and was 1676 mm FL; the second male weighed 56.6 kg and was 1698 mm FL. These fish were within the same size ranges as reported for spawning *M. atlanticus* from Brazil and Florida (de Menezes and Paiva, 1966; Crabtree et al., 1997). The female was classified as spawning capable based on the presence of both late vitellogenic oocytes and 24 h POF in the ovaries at the time of collection (Figure 23A). The warm water at the



Figure 24: Histological images of gonadal tissue in the spawning capable phase from M. atlanticus collected off the coast of Louisiana on 2 July 2011. A. Female. B. Male. Key: PG— primary growth oocyte; POF—24 h postovulatory follicle; Vtg2—secondary vitellogenic oocyte; Vtg3—tertiary vitellogenic oocyte; GE—germinal epithelium; SZ—spermatozoa.

time of specimen collection in combination with less than optimal fixation resulted in rapid degradation of the POF observed. However, these POFs appear similar to 24 h POF from other species collected at similar water temperatures and provide evidence that this female spawned 24 h or less prior to capture (Brown-Peterson et al., 2011). While the ovary was dominated by tertiary vitellogenic oocytes (Vtg3), there was evidence of asynchronous oocyte development because secondary vitellogenic (Vtg2) were also present (Figure 24A). The two males were also both classified as spawning capable. Testes were full of spermatozoa but there was no active spermatogenesis occurring (Figure 24B), suggesting the males were at the end of the reproductive season but still capable of releasing spermatozoa. These specimens were collected near the end of the reported spawning season for *M. atlanticus* in Florida (Crabtree et al., 1997).

The collection of both male and female *M. atlanticus* in the spawning capable phase suggests that *M. atlanticus* may be spawning off Louisiana and represents the first evidence that this species appears to be reproducing in the northern GOM. Juvenile *M. atlanticus* have been reported from Mississippi coastal locations by Franks (1970), Overstreet (1974) and Schofield et al. (2007). Various suggestions have been made to account for the presence of juvenile *M. atlanticus* on the coasts of Louisiana<sup>1</sup> and Mississippi<sup>2</sup> (Overstreet, 1974; Schofield et al., 2007; Franks et al., 2009). It is possible that these juveniles are the product of local spawning activity based on evidence provided here. Additional collections of adult *M. atlanticus* during the spring and summer from the northern GOM would help elucidate reproductive activity of this species.

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### Appendix

Friday, February 22, 2013

To Whom it may Concern:

William Stein has my permission to include, as part of his Ph.D. dissertation, the publication "Evidence of spawning capable tarpon (*Megalops atlanticus*) off the Louisiana coast" that appeared in Vol. 24 (pp 73-74) of the scientific journal *Gulf and Caribbean Research*. William was first author on this paper that was co-authored by myself, Nancy Brown-Peterson and Martin O'Connell. William conducted the majority of the research reported in this paper and wrote the paper.

Regards,

James S. Franks Senior Research Scientist Fisheries Biologist University of Southern Mississippi Center for Fisheries Research and Development Gulf Coast Research Laboratory 703 East Beach Dr. Ocean Springs, MS 39564 <u>228.872.4202</u> jim.franks@usm.edu Wednesday, February 20, 2013

To Whom it may Concern:

William Stein has my permission to use the publication "Evidence of spawning capable tarpon (*Megalops atlanticus*) off the Louisiana coast", appearing in Gulf and Caribbean Research (volume 24, pages 75-76) and co-authored by myself, James Franks and Martin O'Connell, as a portion of his dissertation. Mr. Stein did the majority of the work reported in this paper and wrote the paper; clearly this should be considered as part of his dissertation.

Sincerely,

Nancy Brown-Peterson

Nancy Brown-Peterson Research Associate Department of Coastal Sciences The University of Southern Mississippi 703 East Beach Dr. Ocean Springs, MS 39564 <u>228-872-4288</u> (office) <u>228-872-4204</u> (fax) email: nancy.brown-peterson@usm.edu

# Institutional Animal Care and Use Committee

UNIVERSITY OF NEW ORLEANS

DATE:	September 24, 2009
TO:	Dr. Martin O'Connell
FROM:	Steven G. Johnson, Ph.D. Chairman
RE:	IACUC Protocol # UNO-09-013 Entitled: Collection of larval fishes and other aquatic organisms prior to closure of MRGO – obtaining critical pre-impact data from all three Lake Pontchartrain aquatic corridors.

Your application for the use of animals in research (referenced above) has been approved beginning September 24, 2009 and expiring September 24, 2011. Please note that an annual/final report must be provided to the UNO IACUC.

The University of New Orleans has an Animal Welfare Assurance on file with the Office of Laboratory Animal Welfare (OLAW), National Institutes of Health. The assurance number is A3299-01.

M. OCH

The author was born in Natchez, Mississippi. He obtained his Bachelor's degree in chemistry from Vanderbilt University in 1970. He entered medical school at Louisiana State University School of Medicine and received his Doctorate in Medicine in 1974. He completed an internship and residency in Medicine at Charity Hospital of Louisiana in the Louisiana State University Medical School program. After practicing Internal Medicine for three years he joined the University of Texas M. D. Anderson Hospital and Tumor Institute Fellowship in Medicine and graduated in 1981. After practicing adult hematology and oncology for 27 years, he joined the University of New Orleans graduate program to pursue a Ph.D. in engineering and applied science with a concentration in earth and environmental science. He is a member of Dr. Martin O'Connell's research group.