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Effect of Fragmentation and Habitat Type on Coastal Nekton in Mississippi

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EFFECT OF FRAGMENTATION AND HABITAT TYPE

ON COASTAL NEKTON IN MISSISSIPPI

by

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ABSTRACT

EFFECT OF FRAGMENTATION AND HABITAT TYPE

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Coastal wetlands are extremely productive ecosystems that support an abundance of organisms at higher tropic levels. Coastal wetlands also act as important buffers from storms and help protect major cornerstones of coastal economies, such as tourism and fisheries. Despite the clear need for the protection of these habitats, anthropogenic use of coastal wetlands has increased in frequency and intensity resulting in the fragmentation of once continuous habitats. A central challenge to assessing the impact of marsh fragmentation is the lack of quantitative distribution and abundance data from specific habitat types. This is especially true for species that are not commercially or recreationally harvested and are, therefore, not regularly monitored by state and federal resource management agencies. This study makes use of quantitative density, habitat use, and distribution data for non-harvested marsh nekton collected in oligohaline marshes (salinity 0.5-5ppt) of coastal Mississippi. To assess how nekton assemblages varied by habitat, fragmentation level, and position in patch (core vs. edge), four sites along coastal Mississippi were sampled in the summers of 2014 and 2015. Nekton were sampled in adjacent patches of submerged aquatic vegetation and emergent vegetation using a 1 m^2 throw trap. Marsh patch fragmentation was quantified using aerial pictures taken with a GoPro camera secured to the end of a 20ft pole. Points around the patch were digitized in

TPS software and analyzed using R. The results of this study indicate that diversity and density of nekton in Mississippi marshes vary significantly based on habitat type.

ACKNOWLEDGMENTS

I would like to thank my advisor Dr. Jake Schaefer for giving me the opportunity to work and learn in his lab. I would also like to thank Dr.Jake Schaefer for helping me achieve my goal of becoming a professional fisheries biologist and for his patience during our many formative talks and with my views on nekton sampling. I would also like to thank my committee members Drs.Mark Peterson and Mike Davis for their patience and advice regarding experimental design and nekton sampling. I would also like to thank Dr. Frank Jordan for teaching me the ways of the throw trap and for cultivating my love of coastal habitats and nekton. I would also like to thank Dr.Scott Clark for his patience, time, and advice regarding graduate school and all things R related. I would also like to thank Dr. Mac Alford for all his plant identification assistance. Thanks to Dr. Kevin Kuehn for the use of his drying ovens and his wise advice regarding vegetation sampling. Thank you to Lauren Liddon for being my coastal ecology partner in crime and for tolerating countless hours of my conversation in the field. Thank you to Grover Brown and Cybil Huntzinger for all their advice and support during my time at Southern Miss. I would also like to thank the army of undergraduates including Austin King, Dave Campbell, Christopher Vanetten, and Jennifer Main; thank you for being my most enthusiastic field assistants and for making this study possible.

DEDICATION

I would like to give a special thanks to my entire family for encouraging me to follow my dreams of working with fishes and for their shining examples of perseverance and hard work. I would especially like to thank my parents for their encouragement, support, and dedication (through thick and thin) to my education in general and my dream of becoming a fish biologist.

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CHAPTER I - INTRODUCTION

Coastal wetlands are extremely productive ecosystems (Mitsch and Gosselink 1993) that support an abundance of organisms at numerous trophic levels (Stoner 1983, Beck et al. 2001, Kneib 2003). Emergent and submerged vegetation (here after SAV) provide crucial habitats and act as a nursery for small and juvenile fishes, as well as other nekton (Boesch and Turner 1984, Rozas and Odum 1988a, Cho et al. 2006). Nursery habitat can be defined as any habitat, which significantly contributes individuals to the adult population (Beck et al. 2001, Adams et al. 2006). In addition to the nursery function of coastal marshes, the abundance of habitat in general, facilitates fishes of all life-history stages to reach densities greater than those seen in non-vegetated habitats (Hosack et al. 2006). One factor partially responsible for high levels of production in coastal wetlands is the large volume of fresh water input. Freshwater input delivers fine sediment, which accretes over time in river dominated estuaries to form new wetlands. Moreover, this fresh water input carries nutrients obtained from higher up in the stream continuum, which supplement coastal production (Sklar and Browder 1998).

High levels of primary and secondary production support large commercial fisheries upon which coastal economies are often dependent (Bell 1997, Zimmerman et al. 2002). Coastal wetlands absorb significant amounts of wave energy and help protect various cornerstones of coastal economies from major storms (Klein et al. 2003, Laska et al. 2005). Given the numerous ecosystem services provided by these habitats, maintaining the physical and ecological integrity of these habitats is crucial. Despite the clear need to maintain the production and the ecosystem services offered by coastal marshes, rapidly increasing anthropogenic use and disturbances have resulted in steady

declines in these habitats (Lowe and Peterson 2014). It is estimated that 53% of the U.S population lives near the coast (Tralli et al. 2005). As anthropogenic use of, and presence in, coastal wetland increases, so too has the frequency and intensity of anthropogenic disturbances (Zedler and Kercher 2005). Coastal communities are now locked in a paradoxical relationship with the coastal wetlands on which they are economically dependent. These communities are reliant on the ecosystem service provided by these wetlands, yet their use of ecosystems services decreases the ability of the ecosystems to provide them.

Global climate change and associated sea level rise have had a profound effect on coastal wetlands. Sea level rise in coastal marshes has resulted in the conversion of historically freshwater marshes to saltwater and lead to a subsequent shift in assemblage composition (Park et al. 1991). Increased freshwater input is also predicted to increase the occurrence of hypoxic events and increase turbidity and shading of SAV. In addition, future sea level rise will result in the destabilization and loss of already threatened vegetated marsh edge (Penland and Ramsey 1990, Scavia et al. 2002).

Habitat losses in coastal wetlands can result from more direct and local changes in land use practices, such as the construction of canals for navigation and oil extraction. Such canals facilitate saltwater intrusion into historically fresh water habitats (Hitch et al. 2011). The increased salinity in these once oligohaline and fresh water habitats results in changes to both the nekton and plant community structure (Sklar and Browder 1998). In addition, the construction of these canals and similar navigation channels fragment once contiguous habitat and populations of coastal nekton (Craig et al. 1979).

As patches of coastal marsh become fragmented, patch edge (the outer boundary between vegetation and open water) increases as the interior portion of the patch decreases. Eventually, all interior habitats within the patch are dissipated and the remaining patch is comprised completely of edge (Chesney et al. 2000). The importance of "edge" in both terrestrial and aquatic habitats has been well documented. Theories that support the idea of a positive edge effect center on the basic idea that edge occurs at the intersection of multiple distinct habitat types and that this intersection results in an overall increase in habitat heterogeneity. Edges effects are often species-specific, resulting in inter and intraspecific interactions between species. For example, in frequently disturbed habitats or in generalist dominated habits, increases in edge may have no negative effect, as biota in these environments have adapted to regular perturbation. Collins and Barrett (1997) found that female meadow voles preferentially made use of patch edges during reproduction and were therefore unaffected by increased fragmentation, and, in fact, were better able to defend their territories as fragmentation increased. The resulting increases in habitat complexity often benefits more resilient edge adapted species. In contrast, specialist species with a narrow range of environmental tolerances may be negatively affected by the loss of key habitat (Rand and Tscharntke 2007). For example, Munday (2004) observed a significant decrease in populations of coral dwelling gobies and *Acropora* corals in Kimbe Bay, Papua New Guinea.

The manner in which edge is formed plays an important role in determining the nature of an edge effect. Human presence has often resulted in the rapid formation of new edge and has transformed existing habitats, increasing edge in the process (Broadbent et al. 2008). Edges associated with anthropogenic disturbance are abrupt and often

unsuitable habitat for native biota and specialist (Yates et al. 2004). Renjifo (2001) examined the role of natural and anthropogenic landscape matrixes on the bird assemblages in the Andes and found that species with narrow geographic ranges were disproportionally affected by anthropogenic fragmentation. In contrast, naturally occurring edges occur over a gradual gradient of environmental change which native biota have adapted to over evolutionary time (Dangerfield et al. 2003). The quality and type of surrounding habitat also play a role in shaping the nature of an edge effect. Edges formed adjacent to highly productive habitats (e.g. seagrass mangrove relationship) may still support relatively intact assemblages of biota. Planes et al. (2009) tracked larval *Amphiprion percula* movement between marine protected areas and found that 40% of larva settling in areas adjacent to a marine reserve came from within the reserve, suggesting that intact reserves can serve as a viable source to replenish adjacent habitats. While many habitats are composed of gradual edges, coastal marshes in the southern U.S are heterogeneous and often composed of many naturally occurring, distinct edges at the marsh water interface. This natural heterogeneity is the result of a variety of deltaic processes forming distinct patches of marsh and small rivulets over time.

Two main hypotheses have dominated coastal wetland literature in regards to the nature of fragmentation and edge. Recently the negative edge-effect hypothesis, which views edge as the result of fragmentation and habitat degradation has dominated scientific literature. Supporters of this hypothesis predict an overall decline in biodiversity as the result of habitat loss and degradation (Fahrig 1997). In addition, fragmentation may also result in the decline of species dependent on interior habitat which often decreases as edge habitat increase. Overall, declines may be both due to

habitat loss as well as increased competitive dominance of edge species (Opdam 1991). The positive edge-effect hypothesis centers on the idea that that the initial increases in edge during fragmentation may mask the negative effects of habitat loss by increasing habitat heterogeneity (Chesney et al. 2000, Yahner 1988). Despite the prominent history of this theory in landscape ecology (Leopold 1987) few examples of overall positive edge effects exist. One exception to this was observed by Miyashita et al. (2008) who found a positive relationship between edge length and the pregnancy rate of females Sika deer in Japan. Another example can be seen in Casazza and Ross (2008), who examined the assemblages of pelagic fishes near Sargassum weed lines in the Gulf Stream off North Carolina. Their study found a significant increase in the density of fishes and in the overall richness of fish assemblages in Sargassum edges relative to the adjacent pelagic environment. Given the present increase in wetland fragmentation and the predicted future loss of wetlands it is crucial that we gain an understanding of how nekton assemblages will respond. Additional information on the relative importance of "core" and "edge" habitats to the abundance and diversity of nekton is essential to furthering our understanding of marsh fragmentation.

There is also a need for studies which quantify the importance of different habitat types to coastal marsh nekton and examine how patterns of habitat use change over time. Studies such as these should aid scientist and managers in predicting how the effects of fragmentation may interact with different habitat types. While many studies have examined and compared the role of several types of coastal wetland habitats, the majority of these studies focus on the identification and comparisons of higher salinity environments, such as salt marshes and seagrass (Hanekom and Baird 1984, Orth and

Van Montfrans 1987, Ferrell and Bell 1991, Sogard and Able 1991, McIvor and Odum 1988, Rozas and Odum 1988b). Studies which compare vegetated habitats in fresh and intermediate marshes are fewer in number. The few studies that have examined these habitats have been limited to comparison of vegetated and non-vegetated bottoms.

Duffy and Baltz (1998) used drop samplers to quantify differences in fish density between various species of native and introduced SAV, in Lake Pontchartrain, Louisiana. Their results suggested that community diversity was highest in habitats characterized by *Vallisneria americana* Michx. Rozas and Minello (2006) compared nekton use of *V. americana* Michx, and non-vegetated bottom using a 1 m^2 drop sampler in oligohaline marsh, and found that Naked Goby (*Gobiosoma bosc*) and Gulf Pipefish (*Syngnathus scovelli*) were more abundant in *V. americana*. Castellanos and Rozas (2001) showed that SAV habitats supported significantly higher densities of nekton compared to non-vegetated habitats.

Hypotheses

Habitat fragmentation may have resulted in the homogenization of both the habitat and nekton assemblages present in coastal Mississippi. Thus, I predict that beta diversity (site to site change in diversity) will decrease as fragmentation increases in marsh patches (negative effect of fragmentation and increased edge). Moreover, I predict that assemblage composition will vary based on the level of patch fragmentation and position in a patch (edge vs. core).

I predict that distinct nekton assemblages will represent both emergent marsh and SAV habitats present in Mississippi's oligohaline marshes. Moreover, habitats characterized by SAV are predicted to contain a more diverse and distinct assemblage of

transient nekton (nekton species who spend only a part of their life cycle in the marshes), while emergent marsh are predicted to be represented by specialist resident nekton (nekton who spend their entire life cycle within the marsh).

Table 1

Summary of average abiotic conditions separated by watershed, month and habitat.

Watershed	**Month**	**Habitat**	**Salinity (ppt)**	**SD**	**DO (mg/L)**	**SD**	**Temperature (°C)**	**SD**	**Turbidity (NTU)**	**SD**
Pascagoula	June	SAV	1.80	0.9	5.8	1.0	28.3	1.87	8.82	0.733
Pascagoula	July	SAV	1.60	0.9	5.0	2.1	30.6	2.00	13.9	9.47
Pascagoula	August	SAV	4.45	0.1	5.6	1.8	32.3	1.38	21.6	11.6
Pascagoula	June	EME	0.945	0.6	7.1	1.5	30.3	1.38	19.9	8.86
Pascagoula	July	EME	2.75	2.3	4.7	1.4	29.6	1.23	7.74	3.86
Pascagoula	August	EME	3.00	2.2	4.7	0.9	30	1.86	11.5	3.43
Tchoutacabouffa	June	SAV	0.243	0.1	5.8	0.5	28.2	2.26	8.81	2.10

Tchoutacabouffa	July	SAV	1.75	1.1	5.9	2.2	30.8	1.58	4.65	1.13
Tchoutacabouffa	August	SAV	5.25	0	9.5	0	29.2	0	5.89	0*
Tchoutacabouffa	September	SAV	10.0	0	3.9	0.0	29	0	5.74	0*
Tchoutacabouffa	June	EME	0.055	0.02	5.6	0.5	27.7	1.05	11.3	1.48
Tchoutacabouffa	July	EME	3.54	2.74	3.8	1.1	29.5	1.32	8.56	7.58
Tchoutacabouffa	August	EME	5.37	0.01	3.5	0.4	26.3	0.11	14	0.808
Tchoutacabouffa	September	EME	9.28	0.08	9.5	0.0	31.8	0.32	30.6	24.1

Note: Rows which end in a * represent measurements taken only once during the entire study.

Table 2

List of all taxa collected and their summed density by habitat type.

Taxa	Emergent	SAV	Grand Total
Anisoptera	5	42	47
belostomatidae	4	20	24
Callinectes sapidus	45	286	331
Cyprinodon variegatus	8	20	28
Dormitator maculatus	4	24	28
Eleotris amblyopsis	0	3	3
Ephemeroptera	0	4	4
Fundulus sp.	1	0	1
Fundulus grandis	10	34	44
Fundulus jenkinsi	93	3	96
Fundulus pulvereus	9	1	10
Gambusia affinis	50	2	52
Gobiosoma bosc	0	6	6
Heterandria formosa	2	10	12
Labidesthes sicculus	0	1	1
Lepomis macrochirus	1	2	3
Lepomis microlophus	23	198	221
Lepomis miniatus	7	81	88
Lepisosteus oculatus	0	1	1

Lucania parva	90	1401	1491
Menidia beryllina	0	68	68
Microgobius gulosus	0	14	14
Micropterus punctulatus	0	3	3
Micropterus salmoides	4	62	66
Myrophis punctatus	1	34	35
Palaemonetes kadiakensis	0	5	5
Palaemonetes paludosus	12	321	333
Palaemonetes pugio	38	75	113
palaemonetes vulgaris	0	1	1
paralichthys lethostigma	0	1	1
Farfantepenaeus aztecus	0	14	14
Litopenaeus setiferus	5	12	17
Poecilia latipinna	1	0	1
Ranatra.sp	9	14	23
Shrimp larva	0	5	5
Syngnathus scovelli	0	21	21
Tadpole	0	13	13
Trinectes maculatus	0	8	8
Xanthidae	0	66	66
Zygoptera	9	170	179

Table 3

Results of a Multivariate Analysis of Variance for Distance Matrices which examined the effect of habitat (emergent marsh vs. SAV), month (June, July, August), replicate (1-6) and the interactions of all main factors.

Factors	DF	Sum of squares	Mean sum of Sqs	F.model	R^2	*P* value
Habitat	1	8.12	8.12	45.69	0.188	<0.0001
Month	3	1.77	0.59	3.32	0.041	<0.0001
Watershed	1	1.90	1.90	10.6	0.044	<0.0001
replicate	5	0.683	0.136	0.769	0.015	0.830
Habitat*Month	3	2.18	0.725	4.08	0.050	<0.0001
Habitat* Region	1	0.59	0.591	3.32	0.013	0.0039
Residuals	**156**	**27.7**	**0.178**		**0.645**	

Table 4

Results of the indicator analysis that used habitat as a grouping term. Species at the top portion of the table represent SAV species and species on the lower half of the table represent emergent marsh species.

Species	Indicator value	SAV density	Emergent marsh density	*P*
L.parva	0.936	1401	90	<0.0001
L.microlophus	0.714	198	23	<0.0001
Zygoptera	0.690	170	9	<0.0001
P.paludosus	0.658	321	12	<0.0001
M.salmoides	0.597	62	4	<0.0001
L.miniatus	0.515	81	7	0.0006
Anisoptera	0.480	42	5	0.0007
M.punctatus	0.420	34	1	0.0005
S.scovelli	0.364	21	0	0.0017
Belostomatidae	0.361	20	4	0.0325
M.beryllina	0.319	68	0	0.0103
M.gulosus	0.303	14	0	0.0095
Emergent marsh species				
F.jenkinsi	0.809	3	93	<0.0001
G.affinis	0.363	2	50	0.002
F.pulvereus	0.337	1	9	0.003

Table 5

Results of the indicator analysis using fragmentation level as the grouping factor. High fragmentation (HF), medium high fragmentation (MHF), medium fragmentation (MF).

Species	**Indicator value**	**Emergent marsh density**	***P***	**Fragmentation level**
Lepomis microlophus	0.737	23	0.004	HF
Lepomis miniatus	0.667	7	0.004	HF
Fundulus grandis	0.581	10	0.03	MHF
Cyprinodon variegatus	0.439	8	0.05	MF

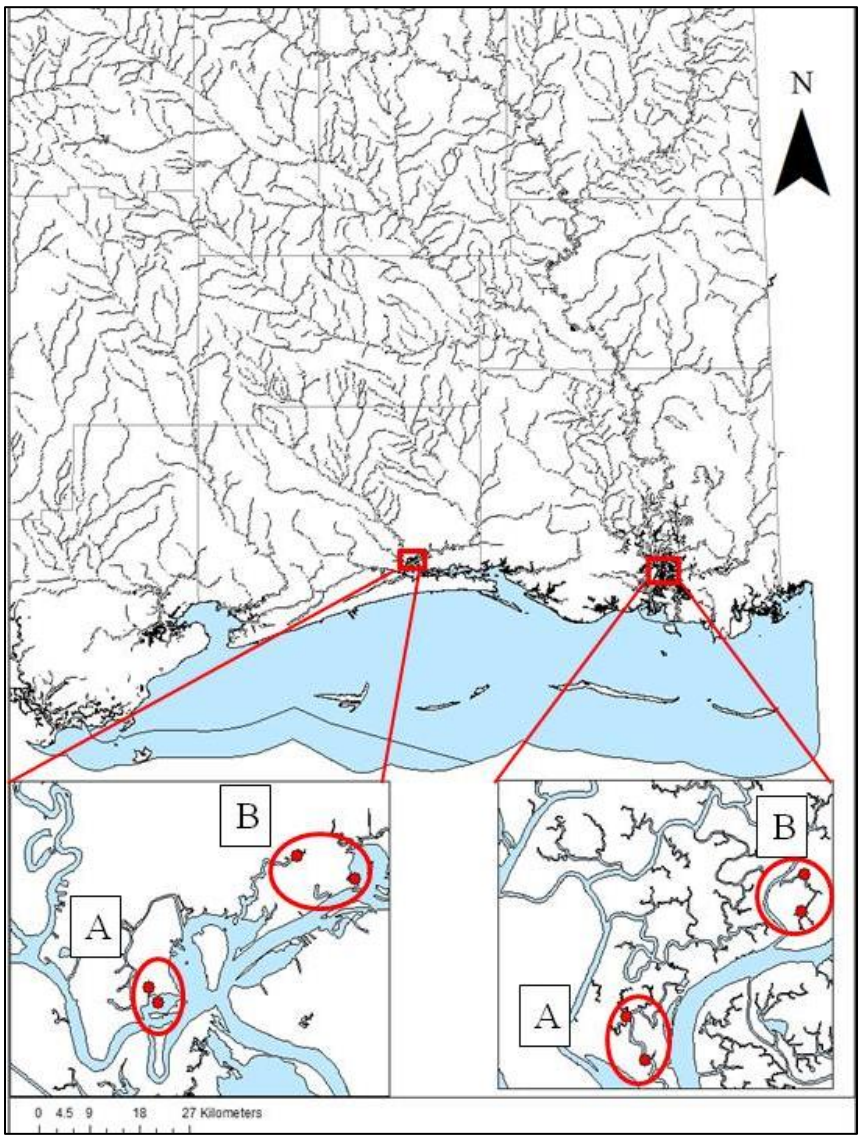
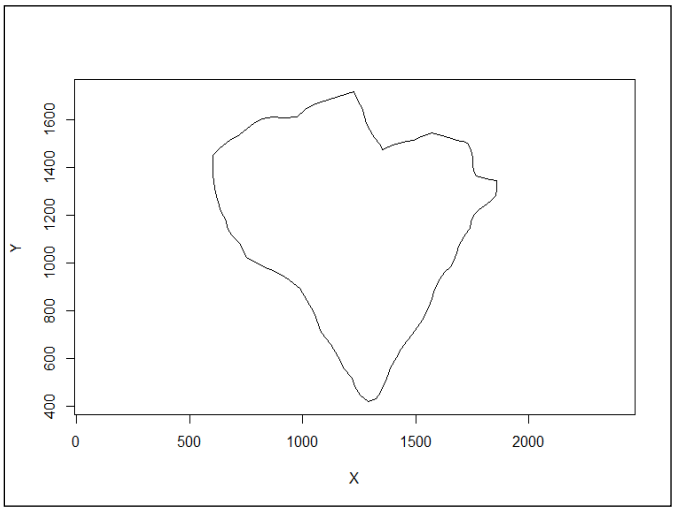


Figure 1. Study area map.

Map of the two coastal regions sampled in Mississippi oligohaline marshes. The inset map on the left shows replicate sampling areas (A and B) in the Tchoutacabouffa watershed. Each red dot represents replicate sites within each sampling area. The map on the right shows replicate sampling areas within in the Pascagoula watershed red dots again represent sampling location within each replicate area. Areas A and B in both watersheds were sample three times a year for two year.

A



B

Figure 2. **A**) Original aerial photo of patch with the 1 m^2 quadrat used for scale. **B**) Digitized points used to quantify patch area (72 m^2) and fragmentation index. The X and Y-axis represent a range of pixels per area.

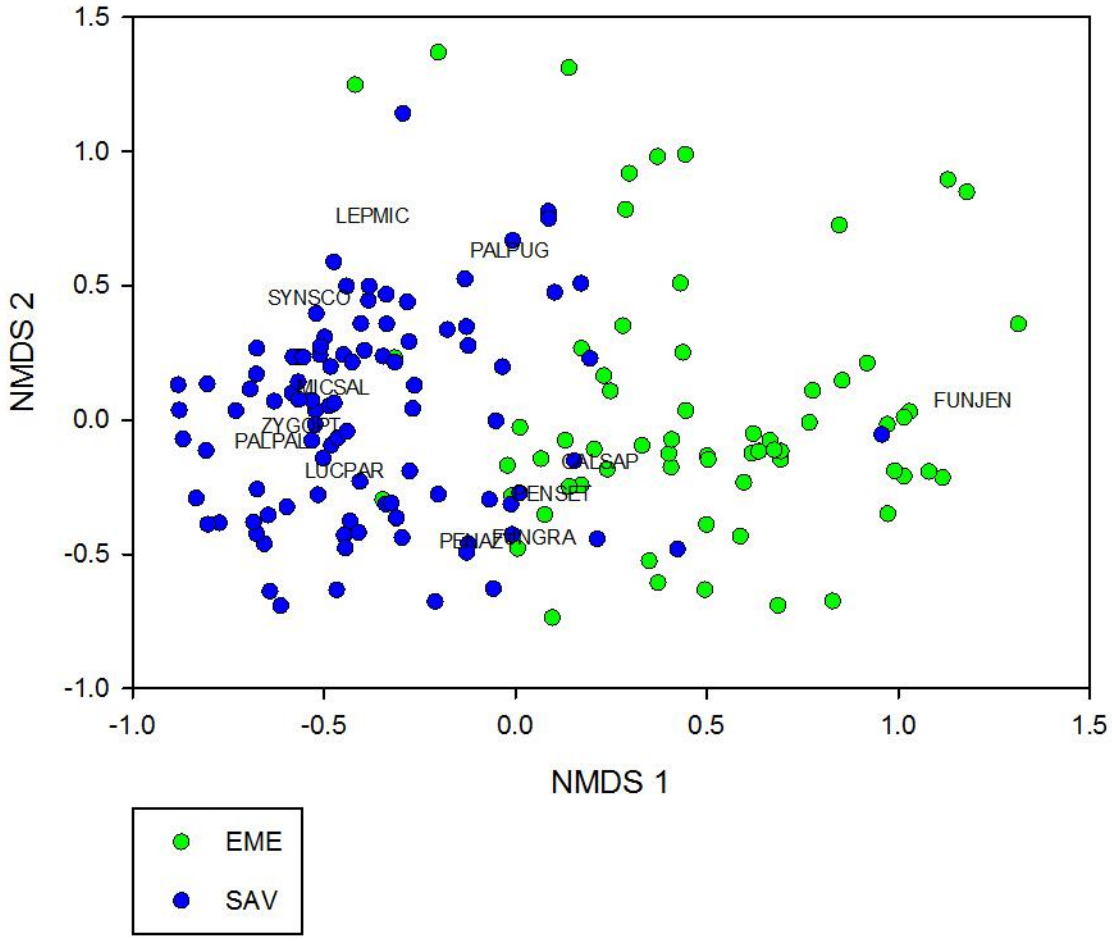


Figure 3. Results of an NMDS run on log transformed Bray-Curtis density data.

Emergent marsh sites are indicated by a green dots and SAV site are represented by blue dots. The top ten most abundant species are represented in NMDS space by species codes (the first three letters of genesis and species). K=3, stress=12.7

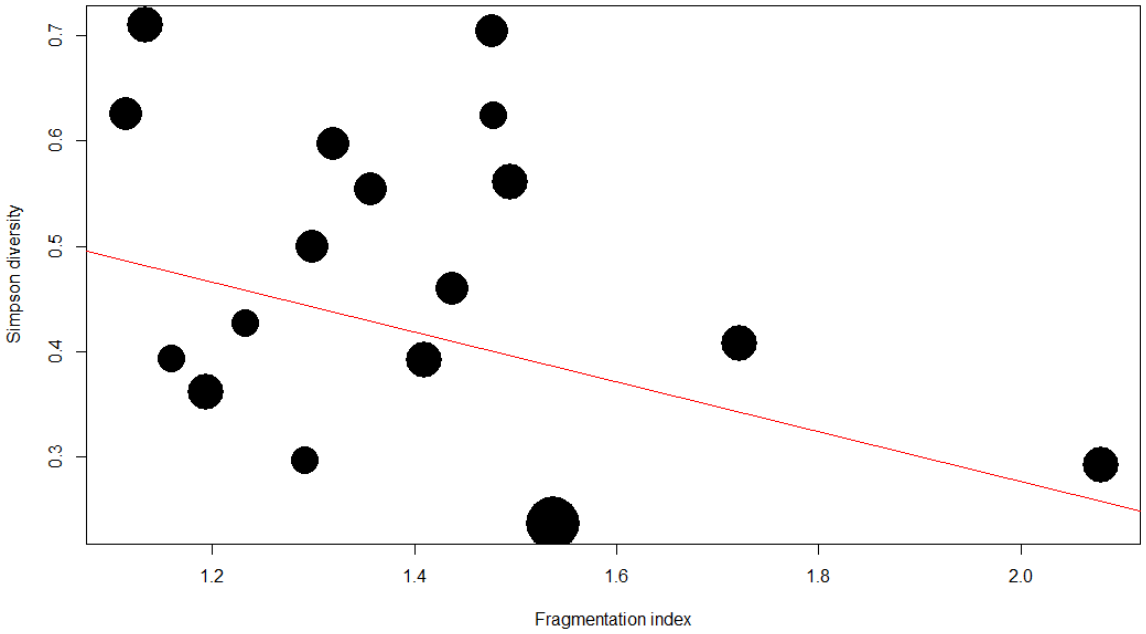


Figure 4. Regression using fragmentation index as a dependent variable and Simpson diversity as the response variable. Dot size represents emergent marsh patch area. R^2=16.4%, p= 0.041.

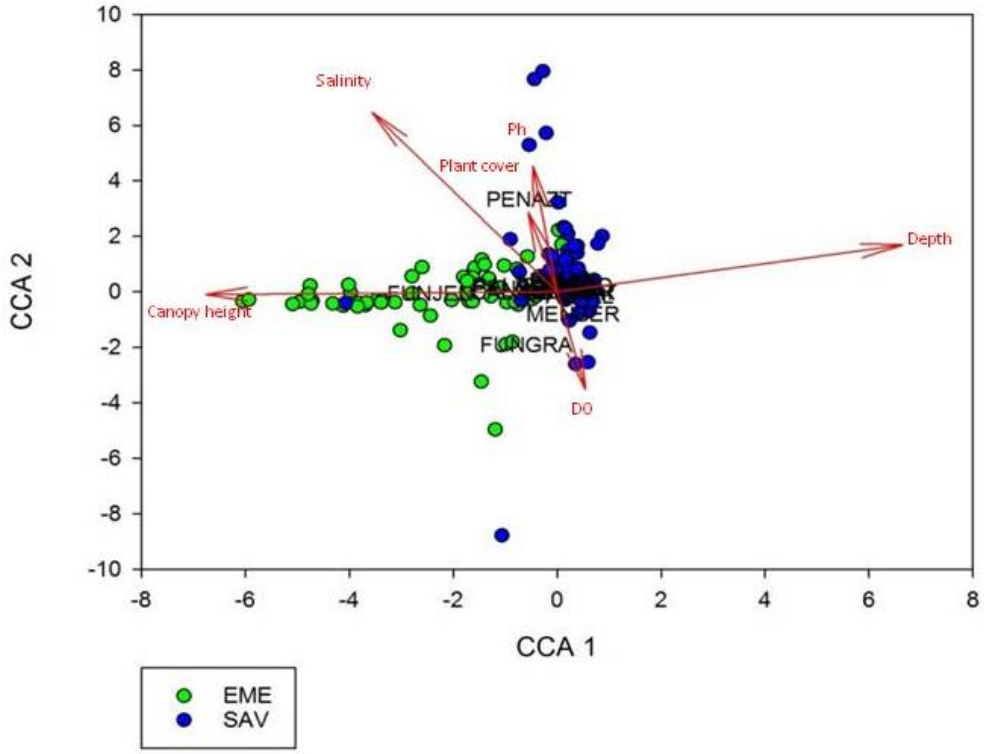


Figure 5. Ordination biplot depicting the first two axes of the CCA of the nekton density of emergent marsh (EME) and submerged aquatic vegetation (SAV) sites. Environmental variables are represented by arrows and species location by their species codes (first three letters of genus and species).

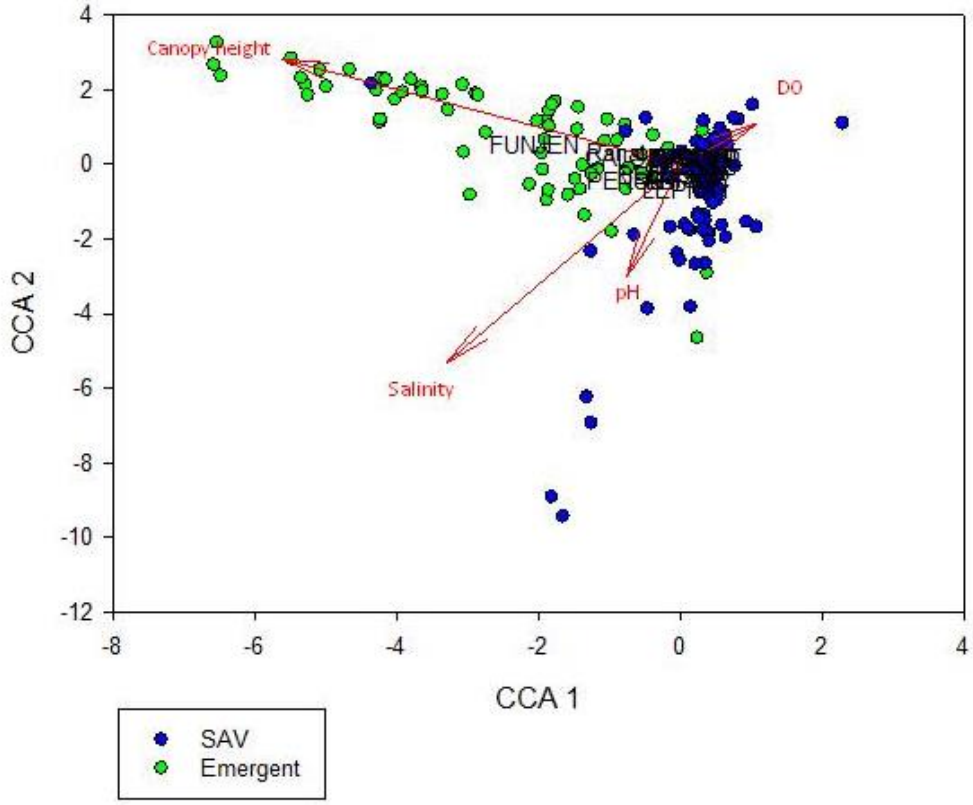


Figure 6. Ordination biplot depicting the first two axis of the partial CCA of the species assemblages of emergent marsh and SAV sites. Environmental variables are represented by arrows and species location by their species codes (first three letters of genus and species).

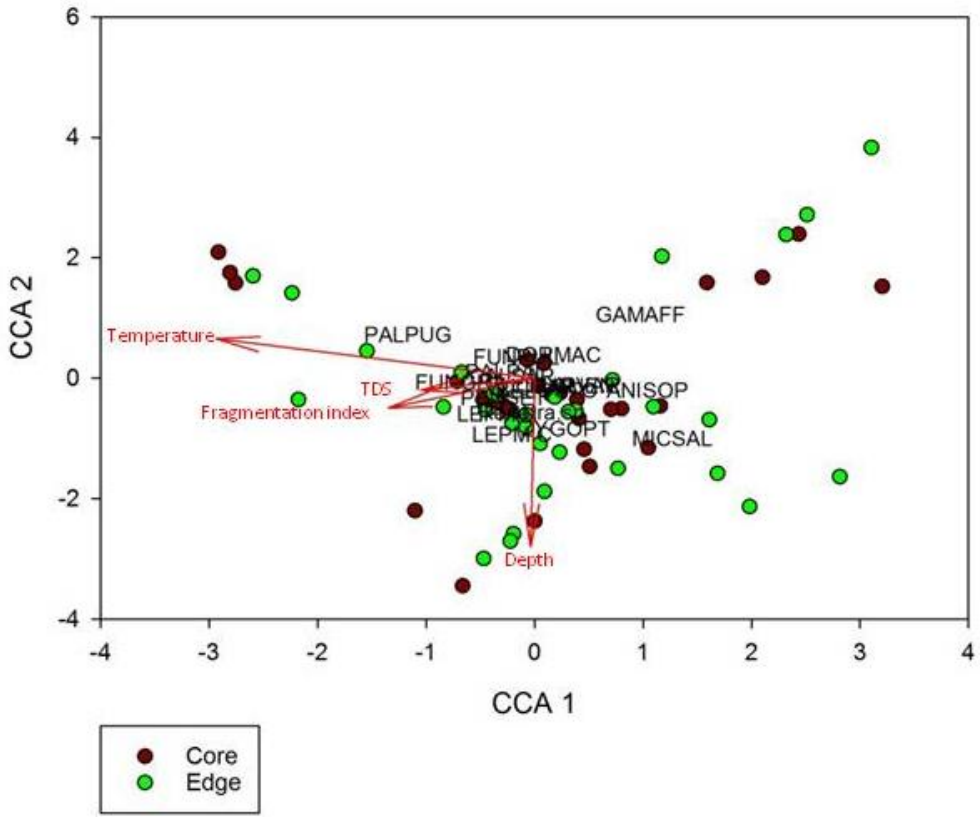


Figure 7. Ordination biplot depicting the first two axis of the CCA of the species assemblages of emergent marsh. Environmental variables are represented by arrows and species location by their species codes (first three letters of genus and species).

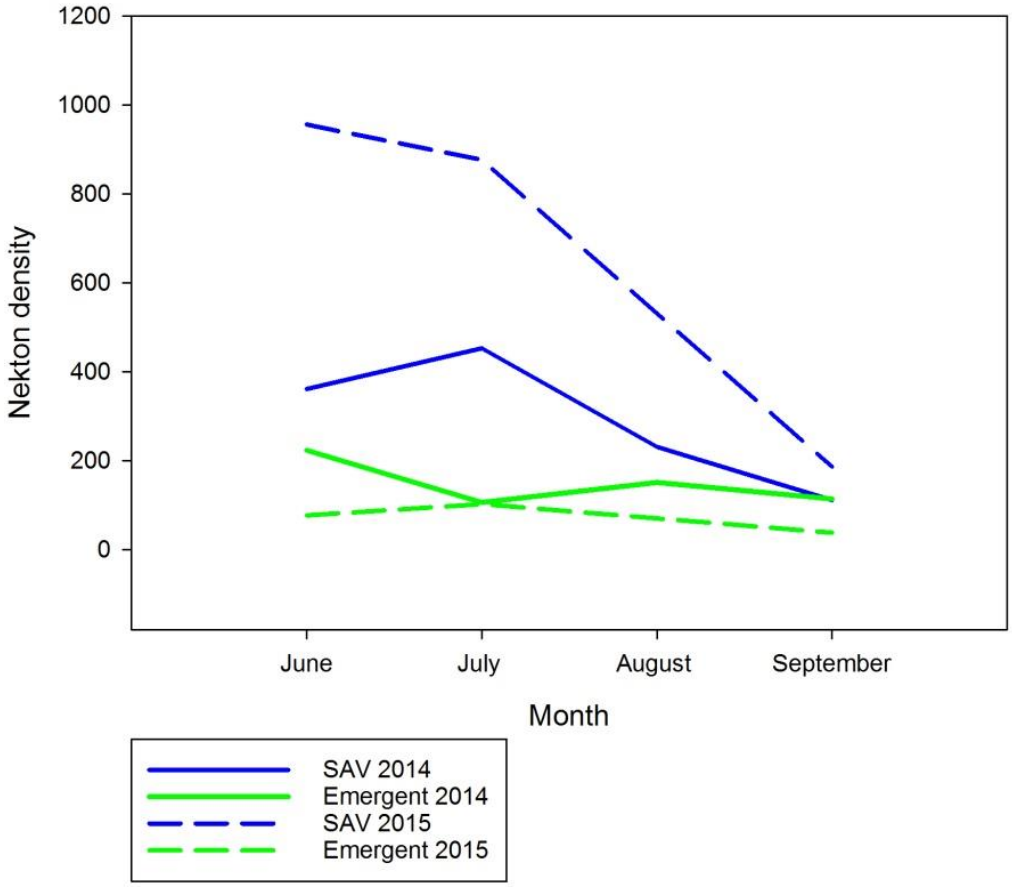


Figure 8. Graph depicts change in summed nekton abundance over summer months. Solid lines depict the summed nekton abundance in SAV and emergent marsh in the summer months of 2014. Dashed lines represent the raw nekton abundance in SAV and emergent marsh in the summer months of 2015.

CHAPTER II - METHODS AND APPROACH

Sampling Design

This study was conducted in the oligohaline marshes (salinity 0.5-5 ppt) of coastal Mississippi. A stratified sampling design was used in this study. Sites were haphazardly selected within two watersheds, Tchoutacabouffa River and Pascagoula River, based on the availability of oligohaline marshes. Based on initial site visits, two main habitat types were defined: emergent marsh (ephemeral patches dominated by *Juncus americanus* and *Sagittaria lancifolia)* and SAV (all submerged vegetation including *Vallisneria americana, Najas guadalupensis* and *Myriophyllum spicatum*). All core-edge analysis was based on data obtained from distinct patches of emergent marsh only. In contrast, the primary use for SAV data was to assess differences between nekton assemblages in SAV and emergent marsh. Two replicate sites (A and B), each of which contained the two habitat types (SAV and emergent marsh), were sampled (Figure 1) in each watershed. Sampling took place over two years during the summer months (June-August) when both plant and nekton assemblages were assumed to be at peak abundance and production (White et al. 1978, Castellanos and Rozas 2001). For the first season of sampling each site was sampled once in each month, using four throws per habitat type (eight throws per site per month). The total number of samples in year 1 was therefore: 2 localities x 2 replicate sites x 2 throws core x 2 throws edge x 2 habitat types per site x 3 months=96. In order to increase statistical power, the number of throws was increased to three (core and edge) in the second year of sampling. The sampling protocol remained otherwise unchanged, resulting in 144 samples in year 2 (2 localities x 2 replicate sites x 6 throws x 2 habitat types per site x 3 months). Several samples in both year one and two were

collected late in the season (September); these samples were determined to differ significantly from seasonal norms and were excluded from all analyses. After the exclusion of late season samples, the total samples used for analysis in this study were 184 (88 throws in year one and 96 in year two).

Physicochemical Data

Dissolved oxygen (mg/L), temperature (°C), turbidity (NTU), salinity (ppt) and pH were taken at the edge (start of vegetation) and core (at least 1m into the patch) of each habitat patch prior to sampling using YSI professional series meters. These measurements were taken at the location of the first throw for all SAV samples.

Nekton Collections

Assemblages of nekton were sampled with a $1m^2$ throw trap. Choosing a sampling method, which accurately samples the target organism and is effective in that organism's habitat, is crucial for the success of any ecological study (Rozas and Minello 1997). In many cases, towed gear, such as seines and trawls, has low catch efficiencies in marsh or other structured habitats (Wells et al. 2008). Effectiveness of towed nets can also vary based on net size, rigging method, tow speed, duration of trawl, and method of net retrieval (Rozas and Minello 1997). Large stands of both submerged and emergent marsh are difficult to sample with pulled nets because the dragging lead line is often lifted off the marsh bottom, reducing the effectiveness and the area covered by the net. Throw traps have several attributes that make them ideally suited for sampling nekton in marshes. One of these attributes is that no habitat alteration is needed prior to sampling (Rozas and Minello 1997). In addition, throw traps provide accurate estimates of density that does not vary with changes in plant stem density (Jordan et al. 1997, Rozas and Minello 1997).

Throw traps were the ideal choice for this project because they allowed for discrete sampling of both core and edge habitats. Moreover, throw traps allow nekton to be sampled in dense stands of emergent marsh.

Patches of SAV in Mississippi's oligohaline marshes are dominated by several species of aquatic vegetation (*V.americana, N.guadalupensis* and *M.spicatum, P.pectinatus*). Emergent marsh sampling focused on discrete patches of emergent marsh. These discrete patches are characterized as being separate from the main body of permanent marsh and are dominated by species such as *S.lancifolia* which senesce in winter months and returns in the spring and summer. The resulting loss of vegetated structure and increased energy expenditure by these plants make these patches particularly vulnerable to fragmentation and habitat loss relative to more permanent marsh. Edge throws were located on the edge of the emergent marsh patch (start of vegetation), while core throws were deployed in the interior of the patch (at least 1m away from edge). In the case of SAV, throws were adjacent to the edge of the marsh. Water depth (cm), plant canopy height (cm), percent cover and density of each species within the trap were measured. Plants in the trap were then uprooted, rinsed, and shaken over the trap to dislodge remaining nekton. Plants were bagged, brought back to the lab, placed in an oven at 105°C until completely dry, and weighed. A bar seine with 3.0 mm stretch mesh was passed through the trap, until three consecutive clean sweeps were obtained (Jordan et al. 1997). Collected nekton were fixed in l0% buffered formalin and later identified to the lowest taxonomic level possible, counted, weighed (g) and measured to the nearest mm standard length (SL).

Patch Data

In order to accurately measure perimeter and area of emergent marsh patches, an aerial picture was taken of each patch of emergent marsh using a GoPro camera, secured to the end of a 20ft telescoping pole. Pictures were taken prior to nekton sampling, in order to capture patch geometry before it was disturbed. A 1 m^2 quadrat was placed in the patch and used as a scale reference for the picture. Points around the patch were digitized in TPS software (http://life.bio.sunysb.edu/morph/) (Figure 2). Additionally, in emergent marsh and SAV sites, percent coverage, and density of each plant species was assessed visually using a 1 m^2 quadrat, laid across the trap at both edge and interior samples. In SAV, sample distance to the nearest marsh edge was measured using a laser range finder. Distance to the nearest patch was recorded for emergent marsh.

Fragmentation Index

To quantify the ratio of edge to core, a fragmentation index (D_L) was used. As patch perimeter (P) deviates from that of a perfect circle for a patch of area (A), the perimeter-area ratio of the patch will increase. Therefore, marsh patches with increased fragmentation should have fragmentation index values greater than 1.5. This fragmentation index is based on a shoreline index created by (Wetzel 2008), who used it to describe lake shape.

Statistical Analyses

Beta Diversity was estimated the using the average Simpson's diversity index among replicate throws. Multivariate Analysis of Variance for Distance Matrices was used to test the hypothesis that nekton assemblages differ based on habitat types (SAV and emergent marsh) and position within a patch (core and edge) (Anderson 2001). In

order to account for the disproportional effect of rare species on multivariate analyses, rare taxa with summed density of less than four individuals in the overall dataset were exclude from all analyses (Cao et al. 2001). Assemblage structure was assessed visually with Non Metric Multidimensional Scaling (NMDS) of Bray-Curtis similarity of log+1 transformed densities (K=3, metaMDS function in the Vegan package in R).

Species patterns of distribution and habitat use are often based on environmental characteristics associated with a given habitat. It is therefore crucial that ecologist make an effort to determine what abiotic characteristics play a role in determining habitat use. Based on this goal, an initial Canonical Correspondence Analysis (CCA) was used to examine the relationship between nekton assemblage composition (natural log+1 transformed abundance) and habitat data from SAV and emergent marsh (plant cover, relative % cover of each plant species, depth (cm), Temperature (ºC), salinity (ppt) and dissolved oxygen (mg/L), Ph, TDS). Variables in the global model were first exclude based on Variance inflation factor scores (VIF), highly correlated variable (>10 VIF) were eliminated (Legendre and Legendre 2012). Variables were then selected for inclusion in the final CCA model based on a forward and backward selection using the ORDISTEP function in the vegan pack in R. Ordistep uses permutated *p* values (n=1000 per step, 100 steps) and AIC values for tied *p* values as the basis for variable selection (minim *p* value of 0.004 for variable acceptance). Monte Carlo permutation tests (permutations=1000) were used to assess the significance of the overall CCA model, the first three axes, and each variable selected by ORDISTEP.

A second Canonical Correspondence Analysis (CCA) based solely on emergent marsh data was used to test the hypothesis that assemblage structure differed based on

environmental variables associated with marsh patches. In addition, a partial CCA which partialed out depth was used to separate variability resulting from depth in the data (Titeux et al. 2004).

An indicator species analysis was performed in order to identify species strongly associated with habitat types (emergent marsh and SAV) and position in patch (core and edge). A third indicator species was used to identify species characteristic of very high (1.88->2), high (1.7-1.87), medium high (1.35-1.59) medium (1.3-1.48) and low (1.11-1.29) levels of fragmentation in emergent marsh patches (Dufrêne and Legendre 1997). Indicator species analysis was also used to select species or groups of species that are characteristic of a given group of samples. The analysis produces indicator values (IV) which are the product of the relative frequency of occurrence and relative density (range 0-100%). A perfect indicator species therefore, is a species that is highly abundant and exclusively sampled in one group of samples. The significance of IV are determined by comparing observed IV to a distribution of IV obtained by randomly permuting nekton density data (permutations 10,000) (Schaefer et al. 2016). *P*-values obtained are based on the proportion of permutations that resulted in a greater than or equal indicator value than observed in the non-permuted density data (Cáceres and Legendre 2009). Linear regressions using Shannon's diversity as the response and fragmentation index as the independent variable were preformed to test the hypothesis that diversity changes with fragmentation index. Statistical analyses were performed using R software (*R* Core Team 2013). A level of 0.05 was used to determine significance.

CHAPTER III - RESULTS

Over two sampling seasons 184 throw trap samples were collected from oligohaline marshes in coastal Mississippi (88 throws in 2014 and 96 in 2015) (Figure 1). In all, 3,986 individuals making up 41 taxa (several aquatic arthropods identified down to suborder, family or genus) were collected (Table 1). SAV sites contained higher density of nekton relative to emergent marsh (2,807 total nekton in SAV and 449 emergent marshes). The *Lucania parva* was the most abundant nekton and fish species collected (1,491 total). *Palaemonetes paludosus* were the most abundant invertebrate collected and second most abundant nekton overall (333 total collected). After exclusion of rare species (occurrence < 3), 29 numerically dominant nekton taxa remained in the data set. The 29 remaining taxa collectively represented 98% of all individuals collected. Mean Shannon's diversity was 0.558 overall, 0.612 in SAV and 0.556 in emergent marsh.

The results of the NMDS suggest that there are clear differences in nekton assemblages between emergent marsh and SAV (Figure 3). The permANOVA identified significant differences between habitat types (emergent marsh and SAV), month and region in assemblage structure (habitat pseudo F =18.21 r^2=0.150 p=0.0009*;* month pseudo F = 4.17 r^2 = 0.069 p = 0.0009*;* region pseudo F = 3.93 r^2= 0.0324 p= 0.0009). There was no significant effect of replicate (Table 3). There were thirteen significant indicators of SAV habitat and three significant indicators of emergent marsh (Table 4). Only one significant indicator of Edge was identified (*P. paludosus*) and no significant indicators of core habitat were identified by the indicator species analysis. There were two significant indicators of high levels of fragmentation (*L. microlophus* and *L. miniatus*), one of medium high fragmentation (*F. grandis*) and one of medium

fragmentation (*C. variegatus*) (Table 4). The linear regression that examined the effect of fragmentation on diversity found a significant negative relationship between fragmentation index and species diversity (Figure 4).

The results of the overall CCA using nekton assemblage data collected from both emergent marsh and SAV showed distinct differences based on habitat type (Figure 5). The global CCA model (canopy height, plant cover, depth, temperature, salinity, dissolved oxygen, pH, TDS) accounted for 20.6% of the variation in nekton assemblage structure. The final CCA model contained six variables and explained 17.0% of the variation. Based on the Monte Carlo permutation tests the final CCA model explained a significant amount of variation (pseudo-$F_{6, 164} = 5.56$, $p = 0.001$). All six of the variables selected (canopy height, salinity, depth, pH, dissolved oxygen, plant cover) were found to be significant. In addition, the first three axis explained a significant amount of variation (CCA1 pseudo-$F_{1, 164} = 14.7$, $p = 0.001$; CCA2 pseudo-$F_{1, 164} = 6.40$, $p = 0.001$; CCA3 pseudo-$F_{1, 164} = 5.20$, $p = 0.001$). Based on the position of the taxa on the CCA bioplot there was significant cross over in habitat usage. Several taxa did, however, have clearly defined habitat usage trends. *Fundulus jenkinsi* was clearly associated with increased canopy height and emergent marsh habitat. *Penaeus aztecus* was associated almost exclusively with SAV habitat and increased pH and salinities. *Menidia beryllina* was also strongly associated with SAV habitat as well as with increased levels of dissolved oxygen. *Fundulus grandis* seemed to be positioned on the boundary between emergent marsh and SAV, suggesting equal usage of both habitat types.

The results of the partial CCA on both emergent marsh and SAV suggest the same distinct differences between emergent marsh and SAV. There is again significant overlap

in habitat usage among the taxa, however; *F. jenkinsi* is still strongly associated with emergent marsh habitat and increased canopy height. The global model (canopy height, plant cover, depth, Temperature, salinity, dissolved oxygen, pH, TDS) accounted for 15.9% of the variation in nekton assemblage structure and depth (the partialed out variable) accounted for 4.70% of the variation in the data. The final CCA model contained four variables and explained 12.1% of the variation. Based on the Monte Carlo permutation tests the final CCA model explained a significant amount of variation (pseudo-$F_{4, 166} = 5.72$, $p = 0.001$). In addition the first three axis explained a significant amount of variation (CCA1 pseudo-$F_{1, 166} = 11.1$, $p = 0.001$; CCA2 pseudo-$F_{1, 166} = 5.44$, $p = 0.001$; CCA3 pseudo-$F_{1, 166} = 3.70$, $p = 0.001$). All four of the variables selected (canopy height, salinity, pH, dissolved oxygen, plant cover) where found to be significant (Figure 6).

The results of the final CCA based solely on emergent marsh patches showed no clear core edge difference (Figure 7). The global model (depth, canopy height, plant cover, percent *S.latifolia*, temperature, dissolved oxygen, salinity, TDS, turbidity, pH , area, fragmentation index) contained twelve variables and explained 26.6 % of the variation. However, the factor fragmentation index was selected by ordistep as significant along with, depth, canopy height, and plant cover. In addition, fragmentation index was found to be significant by the Monte Carlo permutation tests run on factors selected by ordistep.

CHAPTER IV – DISCUSSION

The results of this study suggest that habitat type (emergent marsh and SAV) has a clear effect on the composition of nekton assemblages in Mississippi's oligohaline marshes and fragmentation has a significant negative effect on nekton diversity. SAV supported larger and more diverse assemblages of nekton taxa. These results support my original prediction that distinct assemblages would exist in Mississippi's marsh and that SAV would support a more diverse assemblage of nekton. While this pattern of nekton dependence on SAV has been clearly documented in previous studies (Rozas and Odum 1988b), it has rarely been documented in Mississippi's oligohaline marsh. Given the importance of SAV to coastal nekton, it is crucial that an increased effort be made to understand the overall distributions of these habitats, as well as the specific distributions and roles that particular SAV species play in Mississippi's coastal environment.

While emergent marsh habitats were comprised of significantly less diverse nekton assemblages, they were a crucial habitat for assemblages of resident nekton who spend their entire life cycles in and around emergent marsh habitats. In particular *F. jenkinsi* was frequently sampled in ephemeral patches of emergent marsh (predominantly *S. lancifolia*) in low salinity regions of Mississippi marsh (<5.00 ppt). The connection between emergent marsh and resident nekton has been clearly demonstrated (Kneib 1997) and several studies have focused on the reproduction, life history and distribution and abundance patterns of *F. jenkinsi* in and around Mississippi's marshes (Lang et al. 2012, Lopez et al. 2011, Peterson et al. 2003). However, this study differs from previous studies in that samples were directly obtained from both the cores and edges of shallow (<15 cm in depth) stands of ephemeral emergent marsh with throw traps. Shallow

ephemeral patches of *S.lancifolia* have been infrequently sampled in general and their value to specialist species such as *F. jenkinsi* is crucial information that may aid in conservation efforts.

Based on the results of the permANOVA there is clear temporal variation in nekton assemblage patterns. However, habitat differences persist in both space and time. The spatial and temporal variation indicated by the permANOVA may be the results of an unusually cold winter and spring in 2014, leading to a lag in plant and nekton production and subsequent interactions. In the 2015 sampling period, nekton density and diversity increased. Overall, nekton density peaked in June and July in both 2014 and 2015 in SAV and emergent marsh. However, nekton densities were higher June 2015 vs. June 2014 (Figure 8). Emergent marsh nekton density peaked in June 2014 and August 2015 and was at their lowest in July in both 2014 and 2015 (Figure 8). While it is clear the June and July are important months for nekton production, logistical issues prevented consistent sampling in August and September, decreasing the resolution with which seasonal declines in nekton density could be observed. However, several long-term studies (e.g Schaefer et al. 2016) clearly demonstrate seasonal shifts in patterns of density and diversity of coastal species.

The CCA that examined at the interactions between environmental variables and nekton assemblages by habitat type supported the idea that clear differences exist between emergent marsh and SAV based on both abiotic and biotic factors. Emergent marsh sites where characterized by increased salinity and canopy height, while SAV site had increased water depth and dissolved oxygen (Table 1). Given the frequency with

which emergent marsh patches experience desiccation, increased salinities are to be expected.

Fragmentation, along with dynamic abiotic conditions in emergent marsh patches may significantly affect the assemblage composition of these habitats. Data shows a significant negative relationship between fragmentation index and nekton diversity. This result supports my original prediction that increases in fragmentation and edge would result in decreases in nekton diversity and indicate a negative effect of fragmentation and increased marsh edge. Given these result it seems that that nekton which use shallow emergent marsh habitat are forced to contend with multiple natural and anthropogenic stressors (i.e, fragmentation and hypersaline conditions). These harsh conditions may partially explain the low overall density and diversity of nekton in emergent marsh samples. *F. jenkinsi* was sampled more often in patches of emergent marsh with lower levels of fragmentation, suggesting that intact patches of emergent marsh are crucial habitat for some species of specialist nekton . The results of the indicator species analysis found that assemblages of edge and transient nekton dominate patches of emergent marsh, which have undergone medium to high levels of fragmentation (Table 5). This result suggest that as patches of emergent marshes become increasingly fragmented, specialist species such as *F. jenkinsi* are forced out and replaced by transient edge species. Despite predictions that increased habitat complexity may result from fragmentation, data from ephemeral patches of emergent marsh in coastal Mississippi support the negative edge effect hypothesis, particularly for specialist such as *F. jenkinsi*. Reports that *F. jenkinsi* are rare and a species of concern in many states should therefore

not be surprising, given the high levels of coastal fragmentation experienced in the southern United States.

There was no clear effect of position within patch, causing me to reject my original prediction that nekton assemblages would differ based on position within patch. There are several possible hypotheses for this result. First, it is plausible given the myriad of both anthropogenic and natural stressors experienced by these habitats and their associated nekton, emergent marsh patches may be comprised almost entirely of edge quality habitat and therefore supports primarily edge tolerant species. Second, the lack of core species may be related to the fact that marsh assemblages are often comprised of species that are adapted to the dynamic edge like conditions present in Mississippi's fragmented marshes. It is also possible that given the limited time these emergent marsh patches are available (high tide) to many nekton, clear core edge delineation may not have sufficient time to form prior to patch desiccation. Resident species that are able to use the shallow marsh habitats at low tides (*F. jenkinsi*) may be the exception to this however. One possible solution to these issues is to study this system (emergent marsh patches) in a controlled environment. A mesocosum study in which water level, patch fragmentation, nekton density, and assemblage composition were controlled, may provide further insight into how nekton make use of emergent marsh.

Another possible reason for the lack of a position effect could be a lack of power (N=17 patches). Selecting the appropriate number of samples per strata is crucial in any ecological study. The results of a power analysis run on emergent marsh samples from the first season of sampling resulted in a power of 0.40. Based on these results, paired samples may have provided adequate power. However, the additions of further samples

in each stratum significantly increase power. Further sampling that is directed exclusively at emergent marsh patches and simultaneously evaluates fragmentation should provide further insight into the effect of marsh patch shape on nekton assemblage

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