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The University of Southern Mississippi

SEASONAL DIFFERENCES IN DIET OF TWO PREDATORY FISHES IN

RELATION TO REEF TYPE IN THE INSHORE

NORTHERN GULF OF MEXIC0

by

Brinton Thomas Barnes

A Thesis Submitted to the Graduate School of The University of Southern Mississippi in Partial Fulfillment of the Requirements for the Degree of Master of Science

Approved:

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ABSTRACT

SEASONAL DIFFERENCES IN DIET OF TWO PREDATORY FISHES IN RELATION TO REEF TYPE IN THE INSHORE NORTHERN GULF OF MEXICO

by Brinton Thomas Barnes

May 2014

Relationships of various structural features between reefs and their developing benthic and fish communities have an immense biological and ecological importance for reef restoration and rehabilitation. Therefore, objectives of this study were to establish how abundance (CPUE) and diet composition (%IRI) changes seasonally within Spotted Seatrout, *Cynoscion nebulosus*, and Sand Seatrout, *Cynoscion arenarius*, to view which trophic levels are interacting in relation to different reef type (high relief profile vs. low profile relief). A Kruskal-Wallis one-way ANOVA was performed on non-normal abundance data and determined no significant differences for reef type and season for both piscivorous species. Both *Cynoscion* spp. had relatively similar mean ranked CPUE across reef type and season suggesting their transient ubiquitous distribution may be influenced by prey availability rather than reef profile. Diet composition was analyzed by a PERMANOVA, HMD, MDS, and SIMPER analysis. Between the main terms, only season was significant for both species while the interaction was only significant for Spotted Seatrout. Various fishes and crustaceans were the main prey taxa in both species suggest that both species are opportunistic foragers where gap limitations on available prey may be the

only restriction on diet. Prey availability most likely was driven from seasonal changes within the Mississippi Sound and further studies must include prey density in relation to diet composition for each *species*.

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iv

TABLE OF CONTENTS

LIST OF TABLES

Table

LIST OF ILLUSTRATIONS

Figure

CHAPTER I

INTRODUCTION

Reefs are the most complex ecosystems in the world with more than twice the number of trophic contributors as most other systems (Dunne et al. 2004). The intricate interactions between species and the ecological processes make reef systems difficult to study (Seaman and Sprague 1991). Riding (2002) defined natural reefs as sedentary calcareous deposits created by sessile organisms, and they are presently classified as one of the most imperiled systems on the planet (Ammar 2009). These complex ecosystems have been found to be sensitive to a host of pressures ranging from water quality and seasonal changes to SCUBA diving and multiple types of pollution (Peters et al.1981; Heyward 1988; Carter 1990; Pittock 1999).

Artificial reefs may be useful as a restoration tool to improve recruitment and reproduction of reef-associated species due to the worldwide loss of natural reefs (Bohnsack and Sutherland 1985; Pickering et al. 1998; Sosa-Cordero et al. 1998; Sponaugle et al. 2012; Bryan et al. 2013). Artificial reefs are recently defined "as objects of natural or human origin deployed to influence physical, biological, or socioeconomic processes related to living marine resources" and are classified by Seaman and Jensen (2000:5) as primary (planned) or secondary (unplanned). Some materials used for planned man-made reefs include concrete, stone, FAD (floating artificial devices), tires, stabilized ash waste, PVC, wood, trees, steel grids, and netting (Baine 2001) whereas unplanned artificial reefs include shipwrecks, tanks, oil jetties, and gas platforms

(Bohnsack and Sutherland 1985; Bryan et al. 2013). Concrete, stone, and rock are the most common materials used; however, concrete is highly recommended for artificial reef construction due to it being able to be specifically shaped and durable in seawater (Baine 2001). In addition, benthic and fish community development associated with three artificial reef materials (concrete, cars, and painted steel) were compared to coral reefs and found coral reefs were most similar to concrete reefs, thus, suggesting artificial reefs should be constructed by concrete to closely mimic a natural habitat (Fitshardinge and Baily-Brock 1989). However, concrete does alter water quality by increasing alkalinity and pH due to leaching of calcium hydroxide, thus, affecting organism settlement on artificial reefs (Anderson 1996).

The dominant uses of artificial reefs in coastal management are to 1) improve recreational SCUBA diving and fishing (Bortone et al. 1994a), 2) deter trawling (Relini 2000), and 3) increase fisheries yield and production (Bohnsack and Sutherland 1985; Chua and Chou 1994; Fabi and Fiorentini 1994; Sponaugle et al. 2012). Since fishes aggregate near reefs (Grossman et al. 1997), deployment of well managed man-made reefs may compensate for lost marine habitat and result in a decrease of anthropogenic damage by diverting pressures away from natural reefs (Rilov and Benayahu 1998; Ammar 2009; Sponaugle et al. 2012). The rehabilitation process may well occur after anthropogenic stressors are reduced, therefore, conserving biodiversity and increasing natural conservation (Wilhelmsson et al. 1998; Seaman and Jensen 2000; Ammar 2009). Scientifically, artificial reefs can be useful in testing

ecological hypothesizes especially when compared to natural reefs (Ammar 2009).

Even though artificial reefs have the potential to shelter fish (Bohnsack and Sutherland 1985; Bortone et al. 1994a, 1994b), there is no substantial scientific data supporting that they significantly increase fish abundance (Grossman et al. 1997; Wilson et al. 2001). Difficulties with settling this debate rely on poor scientific design (Lindberg and Loftin 1998), complicated fish behaviors that are influenced by anthropogenic and environmental factors (Grossman et al. 1997), and high environmental variability of marine ecosystems (Osenberg et al. 2002). Additionally, observed increases in fish abundance may be from attracting fish from other nearby reefs, since most predatory fish are highly mobile (Grossman et al. 1997; Lindberg 1997; Powers et al. 2003). Lastly, studies lacking comparisons of how artificial habitats influence neighboring natural reefs also limit conclusions (Bohnsack et al. 1994; Tupper and Hunte 1998); hence, Badalamenti et al. (2002) and Perkol-Finkel and Benayahu (2004) point to a recent increase of interest in interactions between artificial and natural reefs.

 For example, Perkol-Finkel et al. (2006) determined that artificial reefs will mimic adjacent natural reefs when both are constructed of similar structural features; conversely, if structurally different, the communities will also differ. Some structural features affecting species diversity, size distributions, and densities of benthic invertebrate and fish populations include substratum composition and texture, spatial orientation, and structural complexity (Duedall

3

and Champ 1991; Perkol-Finkel and Benayahu 2005; Perkol-Finkel et al. 2006). According to Guichard et al. (2001), Holbrook et al. (2002), and Werner et al. (2011), artificial and natural reefs of complex structure (or additional inhabitable space) and substratum tend to attract more invertebrates and fishes than structures which are simple. Therefore, the relationship of various structural features between reefs and their developing benthic and fish communities have an immense biological and ecological importance for reef restoration and rehabilitation (Perkol-Finkel and Benayahu 2004, 2005).

Piscivorous fishes that have been recognized as top opportunistic carnivores (Perret et al. 1980; Hettler 1989; Rakocinski et al. 2002), and thus selected for this study, are Spotted Seatrout (*Cynoscion nebulosus* Cuvier 1830) and Sand Seatrout (*Cynoscion arenarius* Ginsburg 1930). Both species represent highly valuable sport fishes and may significantly affect the structure of estuarine communities (Lassuy et al. 1983; Ditty et al. 1991; Purtlebaugh and Rogers 2007). Moffett et al. (1979) and Lassuy et al. (1983) describe the distribution of each species to be similar throughout estuarine habitats of the Gulf of Mexico. Spotted Seatrout spend considerable amount of time within a single estuary (Helser et al. 1993) around grassy areas or salt marshes (Peterson and Turner 1994), whereas Sand Seatrout inhabit unvegetated mud substrates with mesohaline salinities near tidal creeks, small rivers, and at the mouths of large rivers (Purtlebaugh and Rogers 2007). Younger individuals of both species feed on mysids, amphipods, copepods, and polychaetes then become more piscivorous during adulthood, although penaeids get more common in the diet

throughout both species' life spans (Darnell 1958; Moffet et al. 1979; Sheridan and Trimm 1983). Feeding habits and diet composition are highly variable, in part, because prey availability changes seasonally (Tabb 1961; Coma et al. 2000), with geographic location, type of estuary (Darnell 1958; Overstreet and Heard 1982), water circulation, sediment type, surrounding vegetation (Hettler 1989) and water quality (Knapp and Purtlebaugh 2008).

In this study, abundances and predator-diet relationships were quantified for two dominant piscivorous species on two different artificial reef types: high profile rubble reefs and low profile oyster beds. The main objectives in this study were to 1) determine if there were differences in catch-per-unit-effort (CPUE, fish/hr) of these two common inshore piscivorous fishes by season and reef type, separately, and 2) determine if there was a difference in diet composition (%IRI) by reef type and season.

Objectives of Study

Objective 1: Does CPUE (fish/hr) of Spotted Seatrout and Sand Seatrout change seasonally by reef type or their interaction.

Objective 2: Determine if diet composition of Spotted Seatrout and Sand Seatrout changes seasonally by reef types or their interaction*.*

CHAPTER II METHODS Study Sites

Spotted Seatrout and Sand Seatrout were collected near the Mississippi coastline at Katrina and Square Handkerchief (both high profile reefs) and USM and Legacy reefs (both low profile reefs) (Figure 1). All four sample sites were

created by the Mississippi Department of Marine Resources (MDMR) to improve finfish habitat and are maintained sporadically throughout their history when funds are available. The largest concrete high profile reef was Square Handkerchief reef (19,526.1 m²) followed by Katrina reef (11,900.1 m²) whereas the low profile reefs (USM and Legacy) are smaller: 4,034.8 m^2 and 3,199.1 m^2 , respectively. The oldest site was USM reef (deployed in 1993) followed by

Square Handkerchief (2002) and then the newest were Legacy (deployed in 2007) and Katrina reefs (2007). Additional background information for the reef sites is included in Table 1.

Table 1

Latitude/longitude, distance from offshore, and distance from nearest major river for all four artificial reefs. (Sq. Hand. = Square Handkerchief)

Sample Collection

 Spotted Seatrout and Sand Seatrout were collected seasonally from August 2011 through August 2013. A gill net (228.6 m by 1.83 m) was deployed at each reef and allowed to soak for one hour. The gill net had 5 mesh sections each 45.7 m long with increasing mesh sizes: 5.1 cm, 6.4 cm, 7.6 cm, 8.9 cm and 10.2 cm. Salinity, dissolved oxygen (mg/L), and water temperature (°C) were recorded at the center of the net at the surface and bottom. Sampling occurred three times per season at each reef type during periods of tidal changes to obtain additional samples.

Diet Methods

Spotted Seatrout and Sand Seatrout were collected and placed on ice in the field. Each individual fish was returned to the lab and measured (TL, mm) and weighed (kg) before stomachs were removed from the body cavity. Each digestive tract was removed based on Bowen (1990) with the exception that only stomachs were placed in 0.47 L plastic jar with 10% formalin for at least a week. Once stomachs were fixed, each was blotted with a paper towel and allowed to dry in a desiccator. A summer 2011 subsample for both species (3 stomachs/ reef) was used to determine a final desiccator drying time (hrs) by taking an initial weight (nearest 0.001 g) and reweighing every 24 hrs thereafter until the stomach weights stabilized. The final desiccation times for stomachs were established to be 48 hrs, at which time weight loss (percent loss of the initial weight) was relatively uniform among weighing periods. Gut contents were then emptied into a Petri dish to be separated, categorized, and identified to the lowest taxonomic resolution using Needham and Needham (1962), Heard (1982), and Hoese and Moore (1998). Afterwards, contents were weighed to the nearest 0.001 g and stored in 70% ethanol. As performed by Lindquist et al. (1994), gut contents were dried at 40°C for 5 hrs or until a constant weight was achieved.

Statistical Analysis

Spotted Seatrout and Sand Seatrout CPUE data were skewed and did not meet the assumption of normality. Homoscedasticity of CPUE for reef type and season was reached for Spotted Seatrout (p> 0.05); however, for Sand Seatrout

reef type was homogenous ($p > 0.05$) but not for season ($p < 0.01$). Thus, a nonparametric Kruskal-Wallis one-way ANOVA (Zar 1999) was used to evaluate CPUE differences by reef type and season, separately. The mean catch-per-uniteffort data was plotted for clarity for both species.

As defined by Hyslop (1980), frequency of occurrence was calculated by counting the number of stomachs containing a prey item and converted to a percentage. Percent number was tabulated by determining the number of prey items in each category and then expressed as a percent of the total number of prey items (Crisp et al. 1978; Lindquist et al. 1994). Finally, percent dry weight (g) was determined by expressing the weight of each prey item as a percent of the total ingested prey weight (Bowen 1990). Hyslop (1980) assessed the advantages and limitations of these calculations. The measurements were used to determine an index of relative importance (IRI), where the sum of percent weight and percent number is multiplied by the percent frequency of occurrence for each prey category (Cortez 1997). To provide a more accurate description of dietary importance and allow for easier comparison among food types, IRI values were further manipulated into %IRI for each prey item. Pinkas et al. (1971) outlined the equations used in this common analysis and are

$$
IRI = %O_i(%W_i + %N_i)
$$

and

$$
\%IRI=100 \times IRI_i / \Sigma IRI_i
$$

where *i* is one of *j* different prey types, *%O* represents frequency of occurrence, and *%Wi* and *%Ni* characterize the proportion of stomach content by weight and number in which both consists of prey *i.* However, %IRI is known to underestimate the influence of lower taxonomic categories due to a multiplicative effect that %O has on the index within a single study (Hansson 1998). Additionally, Macdonald and Green (1983) claimed compound indices are redundant and offer very little new scientific information. Thus, only %IRI was used to directly compare and facilitate comparisons for both species by season and reef type as recommended by Cortez (1998).

Stomachs were binned with a max of 6 stomachs per bin (at least 3 stomachs per bin) to recalculate %IRI for each prey item and used to test for reef type and season (Fall = October-November, Winter = December-February, Spring = March –May, and Summer = June-September) effects in diet composition of Spotted Seatrout and Sand Seatrout. Ontogenetic effects were not considered in this analysis since all but one specimen were <150 mm TL, where a diet shift to mainly fish and shrimp occurs for both species (Moody 1950; Reid et al. 1956; Darnell 1958; Moffet et al 1979; Perret et al. 1980; McMichael and Peters 1989). The main and interactive effects were tested by a two-way non-parametric permutation multivariate analysis (PERMANOVA; permutations = 999; Anderson et al. 2008), where the permutation test creates a distribution of F values (Pseudo-F) and obtains a P-value (Pseudo-P) for non-normally distributed data (Anderson 2001). Only three seasons were considered for the analysis because few stomachs were available during the winter. Pair-wise *a posteriori* comparisons were made using a multivariate analogue of the t-test (pseudo-t) for each level of significantly different main effects and interaction terms. If there was significant interaction term, then pseudo-t tests were used to compare levels within each term of the interaction (Anderson et al. 2008). Based on the fourth root transformed Bray Curtis similarity matrix, Homogeneity of Multivariate Dispersion (PERMDISP; hereafter HMD) was estimated and used to describe the deviation from centroids in diet composition among reef type and season separately (Anderson et al. 2008).

Non-metric Multidimensional Scaling (MDS) was used to visually separate diet composition of Spotted Seatrout and Sand Seatrout between seasons and reef types. These were followed by Similarity Percentages (SIMPER) analysis to disaggregate the similarity matrix to identify which diet components were most responsible for any dissimilarity between reef and season (Clarke and Warwick 2001). Kruskal-Wallis one-way ANOVA was conducted with SPSS software (version 12.0, SPSS, Inc., Chicago, III) (Green and Salkind 2008) whereas MDS, PERMANOVA, and SIMPER similarity analyses were completed with PRIMER software (version 6.01.2; PRIMER-E Ltd, Plymouth, UK).

CHAPTER III

RESULTS

A total of 293 fish were captured from August 2011 through August 2013: 164 (56%) Spotted Seatrout and 129 (44%) Sand Seatrout. The size of Spotted Seatrout ranged from 220 to 535 mm TL with a mean (± STD) length of 327 mm TL ± 63.7 mm. The range for Sand Seatrout was from 148 to 365 mm TL with a mean (\pm STD) length of 286 mm TL \pm 39.4 mm. On low profile reefs, there were 94 (57%) Spotted Seatrout and 80 (62%) Sand Seatrout caught, whereas on high profile reefs 70 (43%) Spotted Seatrout and 49 (38%) Sand Seatrout were caught. Seasonally, there were no fish caught during winter despite effort. During the summer, there were 73 (44%) Spotted Seatrout and 55 (43%) Sand Seatrout, whereas in spring 83 (51%) Spotted Seatrout and 36 (28%) sand seatrout were caught. During the fall there were only 8 (5%) Spotted Seatrout and 38 (29%) Sand Seatrout. Total numbers of fish caught by reef type and season are presented in Appendix A-C for Spotted Seatrout and Appendix D-F for Sand Seatrout.

All CPUE data were considered in the analysis for both species by reef type or season. The CPUE range for Spotted Seatrout was from 0 to 23.6 fish/hr on low profile reefs with a mean $(\pm$ SEM) CPUE of 3.3 \pm 1.2 fish/hr. On high profile reefs, Spotted Seatrout CPUE had a smaller range at 0 to 18.0 fish/hr and a mean (±SEM) CPUE of 2.7 ± 1.0 fish/hr. The CPUE ranges for Sand Seatrout were more variable across reef type than Spotted Seatrout: 0 to 16.8 fish/hr on low profile reefs and 0 to 46.0 fish/hr on high profile reefs. On low profile reefs,

Sand Seatrout mean (±SEM) CPUE was 2.4 ± 1.0 fish/hr and on high profile reefs the mean (\pm SEM) CPUE was 3.0 ± 1.8 fish/hr (Figure 2). Differences in mean ranked CPUE by reef type were not different for Spotted Seatrout (Kruskal-Wallis test, Chi-square = 0.090 , df = 1 , p = 0.765) or Sand Seatrout (Kruskal-Wallis test, Chi-square = 0.012 , df = 1 , p = 0.911).

Figure 2. Catch-per-unit-effort (fish/hour) for *Cynoscion nebulosus* and *Cynoscion arenarius* by reef type. Error bars are one standard error of the mean.

Additionally, Spotted Seatrout CPUE was highest during the fall with a mean (\pm SEM) CPUE of 4.6 \pm 2.8 fish/hr but a range of 0 to 14.0 fish/hr. Spring had the second highest CPUE for Spotted Seatrout with a mean (±SEM) CPUE of 3.1 ± 1.4 fish/hr despite a wider range of 0 to 23.6 fish/hr. Summer CPUE had

the smallest mean $(\pm$ SEM) CPUE at 2.5 \pm 1.0 fish/hr and a range from 0 to 18.0 fish/hr. Catch-for-unit-effort of Sand Seatrout was slightly different by season than Spotted Seatrout. Fall had the highest CPUE with a mean (±SEM) CPUE of 10.5 ± 8.9 fish/hr and a range of 0 to 46.0 fish/hr. Finally, summer had a mean (\pm SEM) CPUE of 2.1 \pm 0.9 fish/hr with a range of 0 to 16.8 fish/hr whereas spring had a mean (\pm SEM) CPUE of 1.5 \pm 0.8 fish/hr and a range from 0 to 12.4 fish/hr (Figure 3). Differences in mean ranked CPUE by season were also not significant for Spotted Seatrout (Kruskal-Wallis test, Chi-square = 0.736 , df = 2 , $p = 0.692$) or Sand Seatrout (Kruskal-Wallis Test, Chi-square = 3.587 , df = 2 , $p = 0.166$).

Diet analysis was performed on 183 (62% of total fishes collected) stomachs: 101 (62%) Spotted Seatrout and 82 (64%) Sand Seatrout. No fishes were caught during the winter, thus it was not included in the diet analysis. There was a total of 16 prey taxa among the two predatory species including six families of teleosts and four families of crustaceans. The identifiable vertebrate prey taxa included Bay Anchovy (*Anchoa mitchilli*), anchovy (*Anchoa* spp*.*), Gulf Menhaden (*Brevoortia patronus*), Atlantic Bumper (*Chloroscombrus chrysurus*), and seatrout (*Cynoscion* spp). Gastric mills (gizzard-like muscle) and fish otoliths were combined with unidentifiable fish prey category for descriptive statistics in this study. Identifiable invertebrates were brown shrimp (*Farfantepenaeus aztecus*), grass shrimp (*Palaemonetes* spp*.*), mud crab (*Eurypanopeus depressus*), and portunid crabs (*Callinectes* spp.). A remaining major prey category, amorphic debris included detritus, stones, and unidentified material.

Regardless of reef type or season (Figure 4), the majority of the diet of Spotted Seatrout consisted of fishes, with the addition of other prey categories

Figure 3. Catch-per-unit-effort (fish/hour) for *Cynoscion nebulosus* and *Cynoscion arenarius* by season. Error bars are one standard error of the mean. No fish of either species were caught during the winter months.

in the summer. High profile reefs during the fall had a 100 %IRI but only 2 stomachs were analyzed (Figure 5) and dominated by unidentifiable fish (Appendix B). During summer and spring on both reef types, the %IRI for fish taxa was higher than 80%. Spring low profile reefs had a higher %IRI for fish taxa (99.57 %) (Figure 4) than on high profile reef (84.62 %IRI) (Figure 5), where unidentifiable fish dominated both reef types. However, all three metrics (%F,

%N, %W) mainly influence %IRI for low profile reefs, whereas for high profile reefs %W was the predominant influence (Appendix C). Summer had revealed

Figure 4. Index of relative importance (percent) for *Cynoscion nebulosus* of the major prey taxa identified in stomachs at low profile reefs across all seasons. an opposite trend, where the high profile reef %IRI for fish taxa (91.2%) (Figure 5) was higher than for low profile reefs (82.4%) (Figure 4) and both reefs displayed more diversity of fish prey items than for spring (Appendix C). High profile reefs during summer months were dominated by unidentifiable fish and *Cynoscion* spp. but driven differently (%F and %W, respectively) (Appendix A). Other fish prey taxa on high profile reef during the summer included *A. mitchilli*, unidentified Sciaenidae, and *C. chrysurus* (in decreasing %IRI). Numerically, and

by occurrence, *A. mitchilli* (55.49 %IRI) dominated the diet of Spotted Seatrout on low profile reefs during the summer, while other fish prey taxa included unidentifiable fish, *Anchoa* spp., and *B. patronus* (Appendix A).

Figure 5. Index of relative importance (percent) for *Cynoscion nebulosus* of the major prey taxa identified in stomachs at high profile reefs across all seasons.

Similar to fish taxa, crustaceans or amorphic debris were not found in any stomachs of Spotted Seatrout during the fall on either reef type (Figures 4 and 5; Appendix B). Shrimp was the only crustacean consumed during summer and spring, but diets in summer months were dominated by *F. aztecus* where low profile reefs (15.51 %IRI) (Figure 4) were influenced by %W and high profile reefs (8.52 %IRI) (Figure 5) by %O and %N (Appendix A). In contrast,

unidentifiable shrimp were dominate by %O (Appendix C) in the spring on high profile reefs (2.21 %IRI) (Figure 5), whereas on low profile reefs, stomachs contained a single *Palaemonetes* spp., where none were found on any other reef or season (Appendix C) and the %IRI of this prey was too low to be shown in Figure 4. Amorphic debris was higher for Spotted Seatrout, %N and by %O (Appendix C), in the spring on high profile reefs (Figure 5); however, low profile reefs diets contained very little amorphic debris during summer and spring (Figure 4).

The MDS of the %IRI of the prey taxa for Spotted Seatrout for reef type and season indicated the 2-D fit was suitable (stress = 0.12). The ordination plot showed considerable separation across reef type and season (Figure 6). Diet $(\%IRI)$ differed by season (PERMANOVA; Pseudo-F = 10.65, P (perm) = 0.002) and the reef type*season interaction term (Pseudo-F = 4.56 , P (perm) = 0.002); however, reef type was not significant (Pseudo-F =0.98, P (perm) = 0.464). Square root of the estimates of components of variation indicated season (27.63) accounted for more variation in diet (%IRI) than the interaction term (23.74) or reef type (-1.10). Pair-wise pseudo-t tests were used to decompose the interaction term and indicated %IRI similarity between reef types was significantly greater in the spring (Pseudo-t = 3.00 , P (perm) = 0.008) than in the summer (Pseudo-t = 1.33, P (perm) = 0.198). Mean similarity was 73.71 in spring but only a 45.90 between reef types in the summer. However, when only season was considered, %IRI was significantly greater during summer and spring on low $(Pseudo-t = 4.00, P (perm) = 0.001)$ and high profile reefs $(Pseudo-t = 1.88, P)$

(perm) = 0.043) with a 40.7 low profile mean similarity between season and 42.04 between seasons for high profile reefs. The HMD showed no significant difference for reef type (Pseudo-F= 1.80, P (perm) = 0.38) and mean deviation from centroids for low profile reefs was 28.29 (± 4.18) and for high profile reefs

Figure 6. MDS ordination plot for diet composition data of *Cynoscion nebulosus* by reef type and season. (ss = spotted seatrout, $O =$ oyster bed (low profile) reefs, R = rubble (high profile) reefs, S = summer, SP = spring, B = replicate 1, replicate 2, etc.). Fall was not considered for *C*. *nebulosus*.

was 35.63 (± 2.23). Conversely, there was a significant difference for season (Pseudo-F = 22.20, P (perm) = 0.001) with a mean deviation from centroids of 35.12 (\pm 3.83) for summer and 13.81 (\pm 2.64) for spring. Fall was not considered for this analysis of *C. nebulosus.* Two-way SIMPER comparisons indicated seven prey taxa (*A. mitchilli, F. aztecus*, amorphic debris, unidentified fish, unidentified penaeids, *Cynoscion* spp., *B. patronus*) contributed 90.87% to the diet composition of Spotted Seatrout during the summer whereas in the spring there

Table 2

Mean pair-wise fourth root transformed diet composition (%IRI) between seasons (summer, spring) of Cynoscion nebulosus from the Mississippi Sound based on SIMPER analysis. SD = standard deviation, UID = unidentified. Taxa are listed in order of their contribution to the mean dissimilarity between seasons with a cutoff when the cumulative percent contribution approaches 90%. Fall was not included in this particular analysis.

were only three prey taxa (amorphic debris, unidentified fish, unidentified penaeids) (Table 2).

Similar to Spotted Seatrout, the majority of the diet of Sand Seatrout comprised fishes but the fish taxa %IRI > 80% in the spring, and fall rather than in the summer on both reef types (Figures 7 and 8). By all three metrics, unidentifiable fish (95.85 %IRI) dominated low profile reefs (98.75 %IRI) during spring and other prey taxa of less importance included *B. patronus* and unidentifiable Sciaenidae. High profile reefs (81.70 %IRI) were dominated numerically and by occurrence of unidentifiable fish (55. 84 %IRI), whereas unidentifiable Sciaenidae (25.86 %IRI) was mainly influenced by %W (Appendix F). During the fall, the reefs had an opposite diet trend from the spring when high profile reefs (99.29 %IRI) (Figure 8) had a higher %IRI for fish taxa than low profile reefs (82.32 %IRI) (Figure 7). By %W, *B. patronus* dominated the diet of Sand Seatrout on high profile reefs and lesser importance prey taxa consisted of unidentifiable fish, *Anchoa* spp., *A. mitchilli*, and unidentified Ariidae and Gobidae. Conversely, diets during the fall from low profile reefs had less fish prey taxa richness and were dominated via %W by unidentified Sciaenidae (60.35 %IRI) with less important fish prey taxa only included unidentifiable fish (Appendix E). The summer months for both reef types had lower fish importance than any other season. For both reefs, %N and by %O, were dominated by unidentified fish; however, the high profile reefs (52.51 %IRI) showed a lower %IRI than low profile reefs (72.31 %IRI) (Figures 7 and 8). Low profile reefs had different fish prey taxa than high profile reefs, which included unidentifiable

Sciaenidae, *Cynoscion* spp., and unidentified Clupeidae and Ariidae (Appendix D).

On both reef types, crustaceans were the second most common prey taxa for Sand Seatrout during summer and fall months whereas amorphic debris was more common in the spring (Figures 7 and 8). Crustaceans were more common in the summer (Appendix D), when *F. aztecus* by %W dominated both reefs

Figure 7. Index of relative importance (percent) for *Cynoscion arenarius* of the major prey taxa identified in stomachs at low profile reefs across all seasons.

(Figures 7 and 8); however, in the fall, crustaceans were represented by a few crab species. For example, stomach contained *E. depressus* during the fall on low profile reefs (17.68 %IRI) (Figure 7), while on high profile reefs (0.071 %IRI) (Figure 8), a single stomach contained *Callinectes* spp., and both prey taxa were mainly influenced by %O and %N (Appendix E). Similar to summer months, stomachs from low profile reefs during the spring were dominated via %O by unidentifiable Penaeidae but with 0.28 %IRI which was too small to show up on Figure 7, whereas no stomachs contained crustaceans on high profile reefs (Figure 8). Alternatively, high profile reefs (18.30%IRI) (Figure 8) contained more

Figure 8. Index of relative importance (percent) for *Cynoscion arenarius* of the major prey taxa identified in stomachs at high profile reefs across all seasons. amorphic debris than low profile reef (0.98 %IRI) (Figure 7) during the spring and both were driven by %O (Appendix F). Comparable to spring, amorphic debris during the summer was higher on high profile reefs (19.44 %IRI) (Figure 8) than on low profile beds (3.98 %IRI) (Figure7) with both influenced by

Figure 9. MDS ordination plot for diet composition data of *Cynoscion arenarius* by reef type and season. (SS = sand seatrout, $O =$ oyster bed (low profile) reefs, $R =$ rubble (high profile) reefs, $S =$ summer, $SP =$ spring, $F =$ fall, $B =$ replicate 1, replicate 2, etc.)

%N and %O (Appendix D and F). Amorphic debris was not discovered in any

stomachs during the fall on either reef types (Appendix E).

The MDS of the %IRI of the prey taxa for Sand Seatrout of low and high profile reefs seasonally, indicated the 2-D fit was appropriate (stress = 0.11). Similar to Spotted Seatrout, the ordination plot showed separation between reef type and season (Figure 9). Diet composition (%IRI) of Sand Seatrout differed by season (PERMANOVA; Pseudo-F = 3.42 , P (perm) = 0.005) but not by reef type (Pseudo-F = 1.15, P (perm) = 0.318) nor the reef type*season interaction term (Pseudo-F = 0.83, P (perm) = 0.549). The square root of the estimates of components of variation showed season (24.23) accounted for more variation than reef type (5.20) and the interaction term (-7.73). Pair-wise pseudo-t test for season indicated the summer and fall comparison (Pseudo-t = 1.64, P (perm) =

0.027) contributed the most to the variation with a 29.19 mean similarity followed by the spring and fall comparison (Pseudo-t = 2.01 , P (perm) = 0.03) with a 38.86 mean similarity. The summer and spring comparison (Pseudo-t = 1.53, P (perm) = 0.117) did not significantly contribute to the variation in diet composition (%IRI) with a 53.35 mean similarity. The HDM test showed no significance for reef type (Pseudo-F = 4.62 , P (perm) = 0.067). The mean deviation of the centroids of low profile reefs was 30.37 (± 2.46) and high profile reefs was 38.59 (± 2.92) . Similar to reef type, there was no significance by season (Pseudo-F = 1.16, P (perm) = 0.472) with mean deviation from centroids of 28.03 (\pm 3.76) for summer, 32.72 (\pm 5.49) for fall, and 22.29 (± 3.97) for spring. Two-way SIMPER analysis indicated seven prey taxa (*F. aztecus*, unidentified Penaeidae shrimp, amorphic debris, unidentified Sciaenidae, *B. patronus,* unidentified fish, *Cynoscion* spp*.*) contributed 93.40% during the summer and spring combination (Table 3). In addition, during the spring and fall combination there were also seven prey taxa (amorphic debris, *B. patronus*, unidentified Sciaenidae, *Anchoa* spp., unidentified fish, *A. mitchilli*, unidentified Gobidae) that contributed 90.71% to the diet composition of Sand Seatrout (Table 4).

Table 3

Mean pair-wise fourth root transformed diet composition (%IRI) between seasons (summer, spring) of Cynoscion arenarius from the Mississippi Sound based on SIMPER analysis. SD = standard deviation, UID = unidentified. Taxa are listed in order of their contribution to the mean dissimilarity between seasons with a cutoff when the cumulative percent contribution approaches 90%.

Table 4

Mean pair-wise fourth root transformed diet composition (%IRI) between seasons (spring, fall) of Cynoscion arenarius from the Mississippi Sound based on SIMPER analysis. SD = standard deviation, UID = unidentified. Taxa are listed in order of their contribution to the mean dissimilarity between seasons with a cutoff when the cumulative percent contribution approaches 90%.

CHAPTER IV

DISCUSSION

Catch-Per-Unit-Effort

Both species showed no differences in mean ranked CPUE among low and high profile reefs in the north-central Gulf of Mexico. Only CPUE for Spotted Seatrout on low profile reefs during the spring was barely higher than all other values from either reef type, suggesting Spotted Seatrout and Sand Seatrout are generalist opportunistic foragers and reef type appears not to matter. Generally, individual CPUE data varied between season or reef type within this study; therefore, both *Cynoscion* spp. can be considered transient, which has been found in other systems (Breitberg et al. 1999; Coen et al. 1999; Harding and Mann 2001a, 2001b, 2003; Simonson and Cowan 2008; Simonsen et al. 2013). A relatively similar study of diets of transient fishes in Chesapeake Bay reported comparable findings (Harding and Mann 2001a). Their study analyzed differences between artificial oyster reef and sand bar habitats and suggested the ubiquitous distribution among all habitats for both *Cynoscion* spp. However, Harding and Manning (2001b) studying Bluefish and Striped Bass (2003) indicated a difference of abundance between artificial reefs vs. mud-bottom reference sites. Conflicting patterns among species suggest opportunistic predators probably occupy similar habitats and trophic niches (Peterson 2003). Their data suggest variations in prey availability (Overstreet and Heard 1982; Music and Pafford 1984; Burke 1995; Baltz et al. 1998; Harding and Mann, 2001a, 2001b, 2003; Knapp and Purtlebaugh 2008) may obscure habitat use patterns as observed in this study regarding use of reef types.
Reef profiles may not influence relative abundance of transient fishes as habitat in this study. For example, physical attributes of artificial reefs can directly affect total fish assemblages and indirectly influence CPUE (Bohnsack et al. 1991). Structural complexity, such as varying inhabitable space and size, can change overall fish assemblages (Harman et al. 2003; Garcia-Sais 2010) and have a strong association with larger predators (Hixon and Beet 1989). One may have expected a difference in CPUE by reef type in this study since the high profile reefs would have more and larger inhabitant space than the low profile reefs for refuge of larger prey items, thus increasing the availability of more larger nutritional prey taxa for *Cynoscion* spp. and overall affecting their relative abundance across reef type. As already mentioned, the two selected species are highly transient within an estuary as suggested by similar mean CPUE across reef type and season; therefore, a limiting factor of their abundance and indirectly diet may only be gap limitation for both *Cynoscion* spp, than reef profile. In addition to vertical profile differences, high profile reefs covered more area than the low profile reefs in this study potentially allowing for higher CPUE among larger and smaller fishes. Bryan et al. (2013) concluded that low profile artificial structures offer habitat for smaller solitary species whereas high profile reefs provide highly complex structure for more variety of species, thus altering fish abundance. However, the high profile reefs in the previously mentioned study was a vessel reef where assemblages may differ greatly compared to inshore reefs (Bryan et al. 2013); therefore, structural complexity in some estuaries may

not greatly influence fish assemblages and CPUE for the selected inshore reefs as observed in the present study.

One of the possible factors that may have influenced CPUE among reef types is age of the reef, which was not controlled for in this study. For example, there was a wide range of reef ages recorded in the present study where one of each reef type (USM and Square Handkerchief reefs, 21 and 12 years old) was older than the other two reefs (Legacy and Katrina reef, both 7 years old). This difference of age may influence productivity of the reefs and thus prey availability. Other studies comparing species diversity observed a significant difference by reef age and indirectly affecting fish abundance (Hasting et al. 1976; Hastings 1979; Sanders et al. 1985; Bortone et al.1994b). Settlement of fishes and the attraction of transient species may be rapid but obtaining an equilibrium reef community has been found to take multiple years (Fager 1971; Bohnsack and Talbot 1980). Thus, reef age may suggest future differences among reef type when all sampled reefs in the present study are at equilibrium. However, the Gulf coast states are within Hurricane Alley where tropical storms and hurricanes most likely keep succession at an early stage; thus, the selected reefs in this study may not ever reach an equilibrium community (Connell 1976; Connell and Slatyer 1977), which may create more confounding variables. In addition to structural complexity and reef age, construction material, depth, and orientation of reef sites (Bryan et al. 2013) may also influence CPUE.

Selected *Cynoscion* spp. were not observed during winter months, which may have been due to seasonal movements of both species into areas that were

30

not sampled for this study (Gunter 1938, 1945; Tabb 1966; Benefield 1971; Perrett and Caillouet 1974; Moffet et al. 1979; Shlossman and Chittenden 1981; Helser et al. 1993; Cowan and Shaw 1988). For example, Sand Seatrout migrate into the gulf during the summer months probably in response to extreme high water temperatures (Simmons 1951; Simmons and Hoese 1959); however, this trend was not observed in this study. Similar to Spotted Seatrout, some Sand Seatrout remain in the seaward portion of two Florida bays where they may occupy deeper water within the bay during winter months (Tabb 1966; Knapp and Purtlebaugh 2008) where sampling did not occur in this study (December-February). Differences in offshore migration may reflect the age of Sand Seatrout where large fish move farther offshore and smaller, immature fish move into deeper waters within bays or estuaries (Ditty et al. 1991; Knapp and Purtlebaugh 2008). Even though age was not considered during the current study, Spotted Seatrout and Sand Seatrout caught were mainly young adults, suggesting the lack of occurrence during winter may have been due to movement into bays/channels or offshore.

The mean CPUE was higher during the fall for Sand Seatrout than any other season, while Spotted Seatrout CPUE was relatively similar across seasons (besides winter). This particular mean CPUE was driven by a single high catch of Sand Seatrout during a fall trip; however, all other trips none were caught causing a higher variability of CPUE between seasons. The high profile reefs were over 3 km away from the coastline, whereas low profile reefs were markedly closer to shore (< 0.5 km). Therefore, Sand Seatrout might be moving

out of the estuary and utilizing resources on high profile reefs before going further offshore during winter months. This may explain the relatively higher CPUE and variability observed on high profile reefs for Sand Seatrout during the fall compared to CPUE for Spotted Seatrout. Additionally, high profile reefs had twice as large CPUE range than low profile reefs further suggesting Sand Seatrout may use the seaward high profile reefs during the fall. In contrast, McDonald et al. (2009) conducted seasonal trawl surveys off the coast of Texas for Sand Seatrout and found that summer and spring abundances were significantly higher than fall; however, the summer and spring trend was not observed in this study where CPUE for both *Cynoscion* spp. for fall was higher than summer and spring catches.

Reef sites were all <40 km from a major river basin. The original individual unranked CPUE data was highly variable among reef type and season; thus, differential fresh water discharge could have created sporadic fish movements of transient *Cynoscion* spp. and indirectly their abundances by altering abiotic and biotic characteristics within the estuaries for both species (Rabalais et al. 2002; Garcia et al. 2003; Chanton and Lewis 2002; Maes et al. 2004). For example, Mazeaud et al. (1977) and Moore (1979) suggested the salinity and temperature changes caused by freshwater sources can cause lethal stress in some estuary fishes. Knapp and Purtlebaugh (2008) observed reduced relative abundance of Sand Seatrout in two Florida estuaries due to an increase in river discharge altering salinity and temperature patterns. They also showed that spring catches were four times greater than other seasons and catches declined significantly in

late summer when water temperature and salinity were too high for essential metabolic rates. Wuesnschel et al. (2004) and Vetter (1982), with Spotted Seatrout and Sand Seatrout respectively, indicated temperature affected metabolic stress rather than salinity; thus, varying temperatures may have explained an observed difference of fish abundance during the cooler months within this study.

Diet Composition

The number of prey taxa in each study species was generally low (11 for Spotted Seatrout and 14 for Sand Seatrout), probably due to focusing only on adults. Younger seatrouts consume a variety of small diet items, and as they grow, their diet shifts to mainly mysids, fish, and shrimp. However, adult seatrout exhibit diets consisting of fewer items of mainly fish and crustaceans (Moody 1950; Reid et al. 1956; Darnell 1958; Moffet et al. 1979; Sheridan and Livingston 1979; Perret et al. 1980; McMichael and Peters 1989; Russell 2005; Simonsen and Cowan 2008). The diet trend of mainly fish and crustaceans was observed in this study and various %IRI values among reef type and season further indicate that both species are opportunistic feeders, thus reinforcing findings of previous studies (Moody 1950; Darnell 1958; Tabb 1961; Perret et al. 1980; Overstreet and Heard 1982; Hettler 1989; Llanso et al. 1998; Simonsen and Cowan 2008). Generally, neither fish indicated a strong selectivity or avoidance to a particular diet item further indicating their generalist, opportunistic foraging strategy. Exploitation of a variety of food resources was represented by the wide dispersions in the ordination plots for both species, which is expected by

opportunistic feeders (Llanso et al. 1998). Overall, within the Mississippi Sound, the apparent diet restriction for opportunistic foragers for either *Cynoscion* spp., besides seasonal prey availability, may be gap limitations where the size of the prey in relation to mouth gape size (or consumer size) could be the only limitation on diet preference. Akin and Winemiller (2008:144) suggested consumer and prey body size is "a key variable influencing trophic interactions and the structure aquatic food webs," particularly Spotted Seatrout and Sand Seatrout which have similar trophic position based on stable isotope analysis.

Unfortunately, high %IRI values were observed for unidentified fish prey taxa in this study for both piscivorous species, limiting the diet analysis when determining what prey taxa caused major differences between reef type and season. Unidentified fish contributed to the majority of fish prey taxa, which may have occurred from the transient behavior of both species allowing them to feed in multiple habitats before moving onto the reefs. This may have allowed for additional time for further digestion of identifiable fish taxa to become unidentified. Previous literature also suggests that fish digest faster than crustaceans (Beukers-Stewart and Jones 2004) and even more quickly during warmer months because of higher metabolic rates (Savage et al. 2004). In this study, these trends were observed from the majority of unidentifiable fish taxa occurring more frequently during the warmer months (summer and spring) than the fall. Additionally, unidentified fish taxa may have been the result of the impossibility of sampling during all periods of tidal changes on all four reefs, therefore, resulting in unidentified fish prey taxa being more common with various

degrees of digestion. However, the main identifiable fish and crustacean prey taxa included pelagic, transient species supporting non-selective feeding and suggest that both estuarine species forage within the water column. Preceding studies have reported the most common diet items for Spotted Seatrout (Overstreet and Heard 1982; Simonsen and Cowan 2008) and Sand Seatrout (Darnell 1958; Moffet et al. 1979; Sheridan 1979; Sheridan et al. 1984) were engraulids (anchovies) and Gulf Menhaden which both are common pelagic foraging, transient prey taxa and were observed in this study. Based on the SIMPER analysis, Bay Anchovy and Brown Shrimp contributed to the most differences in Spotted Seatrout diets between summer and spring where the prey taxa are common secondary consumers within the Mississippi Sound and are dependant upon the detritus food web. Prey taxa contribution was relatively different for Sand Seatrout diet during the summer and fall where amorphic debris and Gulf Menhaden contributed to the diet difference where Gulf Menhaden is also considered secondary consumers and rely on the detritus food web.

Although this study examined only adult fishes of various sizes and many diet items were considerably digested, Overstreet and Heard (1982) compared diets of Spotted Seatrout in Mississippi Sound seasonally and reported %O of prey items was higher during spring and summer than during fall and winter. This was also observed in this study by the PERMANOVA model and HMD indices where the only seasons considered in these analyses were spring and summer since no stomachs were examined during the fall and winter for Spotted

Seatrout. *Anchoa mitchilli* and *F. aztecus* contributed the most to Spotted Seatrout diet composition in this study during the summer than spring. Additionally, Overstreet and Heard (1982) reported that during winter, adult Spotted Seatrout consumed quite frequently polychaetes and crustaceans compared to penaeids. Polychaetes were not found in this study; however, polychaetes have also been reported by McMichael and Peters (1989), Llanso et al. (1998), and Russell (2005). Only McMichael and Peters (1989) were studying early life history for larval and juvenile Spotted Seatrout suggesting polychaetes are consumed throughout Spotted Seatrout life cycle and their importance may be due to seasonal prey availability (Blaber and Blaber 1980; Overstreet and Heard 1982; Llanso et al. 1998; Coma et al. 2000; Russell 2005) and specific habitats used (Franca et al. 2012).

Spotted Seatrout and Sand Seatrout consumed mainly Brown Shrimp for crustacean prey taxa but was more important in the summer than in spring (expressed by %IRI and SIMPER analysis) and was consistent with Lorio and Schafer (1966) and Overstreet and Heard (1982). The higher summer importance of Brown Shrimp in the diets follows previous literature, which suggests that Brown Shrimp tend to burrow in the sediment at lower water temperatures and prefer warmer temperatures for optimal growth after reproduction (Eldred et al. 1961; Aldrich et al. 1968; Zimmerman et al. 1984). Therefore, brown shrimp are in higher abundance and more susceptible for predation during the summer as observed within the diets of the selected generalist species. Vose and Bell (1994) reported a variety of caridean shrimp in

larval and juvenile Spotted Seatrout diet occurring around seagrass beds. However, in the present study caridean shrimp, such as *Palaemonetes* spp*.,* were scarcely observed, most likely because they are associated with seagrass and salt marsh habitat where sampling did not occur as fishes collected for this study were adults (>150 mm, TL) and feed on larger prey further suggesting the main limiting factor on diet is gap limitation. However, the single grass shrimp found in the diet of a Spotted Seatrout was from a low profile reef, which is closer to the shore where seagrass and salt marsh habitat dominate the coastline.

The overall seasonal diet shift I observed was likely due to prey availability and because there were fewer fish and shrimp prey in estuaries during the fall and winter suggesting an increase of community activity on both reef profiles (Tabb 1961; Perry and Boyes 1978; Dietz 1976; Lehnert and Allen 2002; Russell 2005). Lehnert and Allen (2002) reported fish catches to be two to five times greater during the warmer months than in the winter on an intertidal oyster bed in the North Inlet estuary, South Carolina. Additionally, they determined that juvenile and adult crab and shrimp were more abundant during the spring than juvenile and adult fishes; however, in this study there were generally more crustaceans observed during summer and spring. Specifically for Spotted Seatrout, a diet shift was observed from penaeids during the summer to crabs in the spring while Sand Seatrout foraged on shrimp throughout summer and spring suggesting that season influenced the differential diet composition for both selected generalist species.

Diet studies for adult Sand Seatrout are more limited than Spotted Seatrout; nevertheless, a comprehensive feeding study in Apalachicola Bay, Florida analyzed 79% of 1,545 individuals of various sizes and ages and found that 62% fed on fishes (%W), frequently anchovies (*A. mitchilli*) and 26% mysidaceans (Sheridan and Livingston 1979; Sheridan 1979). The authors observed an inverse relationship of mysids to fishes as the size of Sand Seatrout increased. Sheridan (1979) and Sheridan and Livingston (1979) suggested that Sand Seatrout diets might be driven by a seasonal progression of dominant fishes within an estuary. For example, planktivores are abundant in spring and fall when Gulf Menhaden and anchovies are in peak abundance in Apalachicola estuary, respectively. Further evidence was observed in Moffet et al. (1979) where %O of engraulids was 31.1% of the total fish percentage (45.6%) of the diet composition for Sand Seatrout in Galveston Bay, Texas. Overstreet and Heard (1982) reported a greater %O of crustaceans in Sand Seatrout than Spotted Seatrout and Silver Seatrout where diets consisted of penaeids 53 %O and caridean shrimp 7 %O. However, Day et al. (1973) and Dierner et al. (1974) reported crustaceans (but not penaeids) found in Sand Seatrout caught off of Louisiana and Texas waters, further suggesting that diet is mainly influenced by gap limitations on the prey availability within a particular habitat. Moffet et al. (1979) found few Portunidae (1.9 %O), generally similar to this study, which also included a single *E. depressus* and has not been observed in adults in other diet studies. Overall, I observed a similar planktivore and crustacean diet trend compared to other studies between the seasonal diet differences observed in the

PERMANOVA model and HMD indices when each season had a different prey item contributions to diets based on SIMPER analysis. In contrast, Pelaez-Rodriguez et al. (2005) discovered no seasonal difference in diet for Sand Seatrout caught offshore at Veracruz, Mexico, suggesting minimal differences in only benthic prey across nortes (windy), wet, and dry seasons.

Spotted Seatrout is a desirable recreational and commercial regulated targeted species whereas Sand Seatrout is a recreational species but not commercial targeted or regulated despite bycatch in shrimp trawls. For both transient piscivorous fishes, life history has been generally well described but there is more datum for Spotted Seatrout in the Gulf of Mexico than Sand Seatrout. However, there is a need for additional Mississippi estuarine-specific abundance and diet composition for both species while focusing on seasonal and habitat type preference (not just reef type). Therefore, to further analyze abundance and diet composition future studies should 1) widen the extent of sampling to include juvenile fishes, 2) examine and include capture-sites abiotic characteristics, 3) obtain spatial and temporal prey abundance estimates, and 4) perform analyses considering habitat and prey relationships for individual fish species.

APPENDIX A

Prey items found in stomachs of *Cynoscion nebulosus* collected during summer months (June- August) on low and high relief reefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID= unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/number of stomachs with prey)

APPENDIX B

Prey items found in stomachs of *Cynoscion nebulosus* collected during fall months (September-November) on low and high relief reefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID= unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/number of stomachs with prey)

Appendix B (continued).

APPENDIX C

Prey items found in stomachs of *Cynoscion nebulosus* collected during spring months (March - May) on low and high relief reefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID = unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/number of stomachs with prey)

Appendix C (continued).

APPENDIX D

Prey items found in stomachs of Cynoscion arenarius collected during summer months (June- August) on low and high relief reefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID = unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/ number of stomachs with prey).

Appendix D (continued).

APPENDIX E

Prey items found in stomachs of *Cynoscion arenarius* collected during fall months (September-November) on low and high reliefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID = unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/ number of stomachs with prey).

 46

APPENDIX F

Prey items found in stomachs of *Cynoscion arenarius* collected during spring months (March - May) on low and high relief reefs. %O = percentage by frequency of occurrence, %N = percentage by number, %W = percentage by weight, %IRI = percentage of index of relative importance, UID = unidentified prey, and Fish = Actinopterygii. (Numbers in parenthesis: total number of stomachs/ number of stomachs with prey).

APPENDIX G

IACUC APPROVAL FORM

The University of Southern Mississippi

Institutional Animal Care and Use Committee

118 College Drive #5147 Hattiesburg, MS 39406-0001 Tel: 601.266.6820 Fax: 601.266.5509 www.usm.edu/spa/policies/animals

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE **NOTICE OF COMMITTEE ACTION**

The proposal noted below was reviewed and approved by The University of Southern Mississippi Institutional Animal Care and Use Committee (IACUC) in accordance with regulations by the United States Department of Agriculture and the Public Health Service Office of Laboratory Animal Welfare. The project expiration date is noted below. If for some reason the project is not completed by the end of the three year approval period, your protocol must be reactivated (a new protocol must be submitted and approved) before further work involving the use of animals can be done.

Any significant changes (see attached) should be brought to the attention of the committee at the earliest possible time. If you should have any questions, please contact me.

PROTOCOL NUMBER: 10101602 PROJECT TITLE: Ecosystem-Based Management - Model-Based Toolset PROPOSED PROJECT DATES: 10/16/2010 to 10/31/2012 PROJECT TYPE: New Project PRINCIPAL INVESTIGATOR(S): Richard Fulford, Ph.D. COLLEGE/DIVISION: College of Science & Technology DEPARTMENT: COA FUNDING AGENCY/SPONSOR: Northern Gulf Initiative, NOAA/Mississippi **Department of Marine Resources** IACUC COMMITTEE ACTION: Designated Reviewer Approval PROTOCOL EXPIRATION DATE: 09/30/2013

Vhbst (Scten

Robert C. Bateman, Jr., Ph.D. **IACUC Chair**

 $10 - 27 - 10$

Date

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