2012

Effects of mimic artificial oyster reefs on the ecology of juvenile fishes in marsh ponds: a before-after-control-impact analysis

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EFFECTS OF MIMIC ARTIFICIAL OYSTER REEFS ON THE ECOLOGY OF JUVENILE FISHES IN MARSH PONDS: A BEFORE-AFTER-CONTROL-IMPACT ANALYSIS

A Thesis

Submitted to the Graduate Faculty of the
Louisiana State University and
Agricultural and Mechanical College
In partial fulfillment of the
Requirements for the degree of
Master of Science

in

The Department of Oceanography and Coastal Sciences

by
Steven B. Garner
B.S. University of West Florida, 2007
May 2012

DEDICATION

To my Dad and Grandfather,
in honor of your memory I aspire to my best,
succeed or fail,
all while in good jest.

ACKNOWLEDGEMENTS

I would like to thank my advisors Drs. Richard F. Shaw and James H. Cowan, Jr. for their insight, advice, support, funding and patience during my long stay here at LSU. I sincerely appreciate the opportunities (and second chances) they've given me.

I would like to thank Drs. Kevin Boswell and Joris van der ham as well as Kari Klotzbach for their countless hours of patience, advice, field work, and camaraderie during this project. Somehow they always found a way to make field work fun no matter how exhausted or miserable we were.

I would like to thank Kirsten Simonsen, Matthew Campbell, Kim DeMutsert, and Captain Jeff Gailliet for their help shoveling that seemingly endless pile of gravel.

I would like to thank my lab mates Alvaro Armas and Matthew Kupchick for their efforts in the field on my initial project. I would like to thank Elise Roche, Christine Ford, Jackie McCool, Caroline Tulloh, Nicole Morris, and Kyra Woods for their hard work in processing that seemingly endless pile of samples in the laboratory.

I would like to thank Dr. Ed Chesney and Craig Guthreaux for taking care of the fish for our tank experiments and Dr. James P. Geaghan for his statistical advice throughout the various stages of the project.

I would like to thank all of those listed above as well as Thomas Bianchetti, Jonathan West, Brittany Schwartkopt, Josh Saari, Courtney Saari, Michelle Zapp, Ashley Melancon, Kristy Lewis, Hillary Glenn, Grace Harwell, Todd Langland, Marshall Kormanec, and Andy Fischer for their support and friendship. I was closer to some more than others but you all made my time here at LSU a truly memorable and awesome experience. I could not have survived without all of you.

And finally, I would like to thank my family for their unconditional love and support throughout these 5 years. Making you proud has always been my greatest motivation. This project was funded by a grant from the Louisiana Department of Wildlife and Fisheries.

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ABSTRACT

I sought to assess the enhancement potential of mimic artificial oyster reefs (MAORs) on trophic dynamics of juvenile estuarine fishes in marsh ponds. Tropic dynamics were investigated by determining the impacts of MAOR addition on meiofauna and macrofauna and then comparing these results to the gut contents and condition (energy density) of four abundant estuarine fishes: Atlantic croaker (*Micropogonias undulatus*), bay whiff (*Citharichthys spilopterus*), sand seatrout (*Cynoscion arenarius*), and pinfish (*Lagodon rhomboides*). Samples were collected every other month for two years (March 2009 – 11) employing a Before-After-Control-Impact (BACI) design. Halfway through the experiment (March 2010), two mud sites in two marsh ponds were converted to MAORs and samples were collected for the remaining period of study. Meiofuanal communities were numerically dominated by nematodes and harpacticoid copepods but showed order of magnitude declines in response to MAOR addition. Shannon-Weaver diversity indices (H´) increased significantly at MAOR sites from six to 13 taxa with SIMPER analyses indicating that nematodes, copepods, tanaids, gastropods, and ostracods contributed to ≥ 95% of the cumulative dissimilarity between periods and habitats. Macrofauna communities were numerically dominated by grass shrimp (*Palaemonetes pugio*), blue crabs (*Callinectes sapidus*), and white shrimp (*Litopenaeus setiferus*), all of which decreased in density in response to MAOR addition. Shannon-Weaver diversity indices for macrofauna decreased at MAOR sites declining from 21 to eight species. Of the eight species present at MAOR sites only naked gobies (*Gobiosoma bosc*), pinfish (*Lagodon rhomboides*), gulf toadfish (*Opsanus beta*), and sheepshead (*Archosargus probatocephalus*) showed increased mean densities, lengths or weights at

MAOR sites. Based upon percent IRI, fish diets were dominated by insect larvae, calanoid copepods, amphipods, mysids, and polychaetes, but the relative proportions of each prey item differed among species. Statistical analyses of gut contents from each of the four fishes showed no significant affects associated with MAOR addition, but energy density analyses showed a significant effect of MAOR addition for pinfish. Energy densities were similar or higher at MAOR sites after addition and when compared between habitats. These data suggest little community level enhancement attributable to MAORs in marsh ponds. However, some specially adapted, reef-associated fishes may be able to effectively utilize MAOR-associated resources to enhance feeding or condition.

CHAPTER 1: GENERAL INTRODUCTION

Coastal Louisiana supports some of the most productive fisheries in the United States, with fishery yields from 2005 to 2010 totaling almost 2.7 million MT http://www.st.nmfs.noaa.gov/st1/commercial/landings/annual_landings.html. Louisiana landings contribute over 72% of the commercial catch in the U.S. Gulf of Mexico and 41% of the total monetary value http://www.st.nmfs.noaa.gov/st1/commercial/landings/annual_landings.html. The high secondary productivity of Louisiana's coastal waters is attributed to the spatial distribution and availability of nutrient rich, intertidal marshes and adjacent shallow open waters (Boesch and Turner, 1984; McIvor and Odum, 1988; Baltz et al., 1993; Cowan et al., 2008). Many of Louisiana's economically valuable fishery species are estuarine dependent and utilize the expansive marsh-estuarine complex as nurseries during postlarval, juvenile, and subadult stages (Boesch and Turner, 1984; Cowan et al., 2008).

Concurrent with the high secondary productivity in coastal Louisiana are extreme rates of wetland loss. From 1985 – 2010 land-loss in coastal Louisiana was estimated at approximately 42.92 km^2 per year, with an overall net loss of 4,877 km^2 from 1932 to 2010 (Couvillion et al., 2010). High land-loss rates have been attributed to a suite of factors including insufficient sediment delivery, subsidence, salt-water intrusion, shoreline erosion, herbivory, eustatic sea-level rise (Steyer et al., 2008), major storm events (Dingler and Reiss, 1990) and anthropogenic stressors such as levees and pipeline canals (Sasser et al., 1986; Snedden et al., 2007; Blum and Roberts, 2009). Unfortunately, rates of land-loss are expected to remain high with as much as an additional 10,000 –

13,500 km^2 of marsh-land lost by the year 2100 (Blum and Roberts, 2009).

Despite high, sustained land-loss rates, significant negative impacts to coastal fisheries have not yet been observed. Even during periods of peak land-loss, fishery yields remained high and yields of some fishery species actually increased (Cowan et al., 2008). This conundrum well illustrates our lack of understanding of the relationship between Louisiana fisheries and productivity thresholds (Cowan et al., 2008). The inability to link yields with ecological parameters (e.g., land loss) highlights the need to better understand connectivity between estuarine fishes and habitat. One experimental method to better understand this relationship is to deploy relatively complex artificial habitats where none previously existed and then determine the resultant effects and the processes that cause them.

1.1 Artificial Habitats and Habitat Enhancement

Artificial habitats have been deployed in a wide variety of locations and designs (see reviews by Grove et al., 1989; Grove et al., 1991; Pickering and Whitmarsh, 1997; Relini et al., 2007), often for the purposes of increasing catches or catch efficiency, reducing effort within a local fishery (Whitmarsh et al., 2008), or even to enhance ecosystem productivity (Relini et al., 2007). Artificial habitats have been shown to augment ecological and biological processes by reducing the intensity of negative stressors through structural resilience (Gardner et al., 1996; Hernkind et al., 1997), the dynamics of colonization (Sale and Dybdahl, 1975; Rodney and Paynter, 2006), and subsequent utilization by consumers (DeMartini et al., 1994; Johnson et al., 1994; Fabi et al., 2006). Primary and secondary consumers often utilize epiphytic, epifaunal, or fouling communities rather than direct consumption of living or decomposing host substrate

(Moncreiff and Sullivan, 2001; Fabi et al., 2006). This implies that some artificial habitats can be productive if they provide adequate availability of areal substratum for colonization and food web support. The enhancement of trophodynamics at multiple levels (e.g., Reed et al., 2006; Lingo and Szedlmayer, 2006; Perkol-Finkel, 2007) can increase diversity (Fabi et al., 2004) as well as local (DeMartini et al., 1994; Johnson et al., 1994; Relini, et al., 2007) or even ecosystem-level productivity.

The fisheries management community recognizes the potential value in enhancing existing natural, and/or degraded habitats, as well as creating new habitats through deployment of built structures. Artificial habitats have been deployed for a wide variety of management purposes in coastal estuaries along the Gulf of Mexico. For example, the harvested shell from oyster leases is usually redistributed or replaced by limestone cobble to augment not only recruitment of oyster spat but reduce crowding to optimize morphological desirability of harvested oysters (Haywood, 1999). In 2006, approximately $47 million was distributed across all states along the Gulf of Mexico through the fin- and shellfish management plan for repair and restoration of inshore artificial reefs, particularly those that mimic oyster reefs (VanderKooy and Freitas, 2006).

1.2 Oyster Reefs as Habitat

The importance of oyster reefs to estuarine ecosystems was well exemplified by the loss of approximately 98% of the original Chesapeake Bay oyster population, which resulted in significant reductions in water quality and trophic cascades (Rothschild et al., 1994; Coen et al., 1999). Because oyster reefs produce large surface areas of hard substrate, they enhance recruitment of sessile invertebrates and provide critical settlement

habitat for oyster spat (Coen and Grizzle, 2007). Settlement may be enhanced by the vertical relief and heterogeneous habitat complexity which can reduce horizontal water velocities down-current of the leading reef edge that can enhance vertical movement, create micro-turbulent flows that may deliver larvae directly to reef substrate (Eckman, 1987; Abdelrhman, 2003), and can also increase persistence after settlement (Bologna and Heck, 2000; Koehl, 2007). Enhancement of primary production may result from accumulating drifting algae (Davis et al., 2009) or when epiphytic producers occupy substrate surfaces (Reed et al., 2006; Pondella et al., 2006).

Compared with other complex habitat types, oyster reefs can produce similar and even increased densities of fishes and invertebrates (particularly structure-associated species), often exhibiting significantly higher densities when compared with non-vegetated, mud bottoms (Zimmerman et al., 1989). Complex microspaces provide food and structurally rigid refugia that may greatly reduce macrofaunal predation pressure (Hall and Bell, 1988). This results in enhanced biodiversity, especially among invertebrates, where many species of annelid worms, amphipods, isopods, crabs, shrimps, copepods, and other bivalves are often found in high densities that might not persist in adjacent non-reef habitat (Wells, 1961; Zimmerman, 1989; Peterson et al., 2003; Stunz et al., 2010).

1.3 Habitat Enhancement

Fisheries managers are particularly interested in investigating the role of artificial oyster reefs, not only in the potential to support increased productivity in estuarine environments, but in the potential for enhancement of nursery habitat for estuarine

dependent fishes (Steimle and Meier, 1997). Although resident oyster reef fishes are most reliant upon reef-associated resources, Coen et al., (1999) highlighted the potential for reef use by facultative, transient estuarine species often having more generalized requirements of complex habitats (Minello et al., 2003), many of which are economically important. Of the 15 most abundant fish species found by Baltz et al. (1993) in Louisiana estuaries, 67% were estuarine dependent transients (i.e., they are not exclusive to one habitat type within the estuary at all life stages but are dependent upon and utilize multiple habitats within estuaries during at least one life stage). In turn, a great proportion of the habitats occupied by estuarine transients are complex, structured habitats such as oyster reefs, and are used for food and refuge during pre-adult stages (Minello et al., 2003). Managed species such as Atlantic croaker (*Micropogonias undulatus*), bluefish (*Pomatomus saltatrix*), red drum (*Sciaenops ocellatus*), Spanish mackerel (*Scomberomorus maculates*), spot (*Leiostomus xanthurus*), spotted seatrout (*Cynoscion nebulosus*), striped bass (*Morone saxatilis*), and flounders (*Paralichthys spp*) have all been collected on or found to directly consume oyster reef-associated resources (Coen and Grizzle, 2007). Juvenile and sub-adult macrofauna are consistently documented in high abundances (Peterson et al., 2003; Simonsen, 2008; Stunz et al., 2010) at oyster reefs and their presence is usually attributed to high densities of forage prey.

Studies often highlight increased abundances, densities, or biomass as evidence for enhancement directly attributable to oyster reefs, especially when compared to unvegetated (Minello et al., 2003; Stunz et al., 2010) or natural mud bottoms (Simonsen, 2008). However, direct linkages between reef associated species and oyster reef resource utilization are necessary to assess their importance as fish habitat (Beck et al., 2001).

Abundant fish presence on a reef may imply utilization but does not preclude the potential for attraction without increasing production, especially at the ecosystem level (Lindberg, 1997; Lindberg et al., 2006).

1.4 Trophic Linkages and Resource Utilization

One method to directly quantify linkages between oyster reef resources and associated fishes is to determine the proportion of reef resources directly consumed by predators (DeMartini et al., 1994; Peterson et al., 2003; Simonsen, 2008). Studies of gut contents from reef associated fishes show relatively large proportions of prey directly consumed from oyster reefs (Peterson et al., 2003; Simonsen, 2008). Peterson et al., (2003) quantified reef utilization through diet analysis and extrapolated productivity throughout the potential lifetime of a restored oyster reef. Assuming protection from harvest, environmental damage, and consistent productivity rates, Peterson et al. (2003) estimated that $10m^2$ of restored oyster reef could yield as much as 2.6 kg yr^{-1} of fish and crustaceans for the functional lifetime of the reef (i.e., up to 30 years). When considering the existing areal distribution of oyster reef habitats as well as potential area for reef deployment (i.e., over natural mud or sand bottoms) throughout the Gulf of Mexico, the potential productivity becomes quite high (Peterson et al., 2003).

Deployment of mimic artificial oyster reefs provides an opportunity to determine relative habitat value while gaining valuable insight into how fishes utilize available resources. Fishes select resources that maximize trade-offs between energetic return and survival probability (Manly et al., 2002). The processes controlling resource utilization during early life are complex. As fish mature they experience drastic changes in body size, morphology, physiology, and nutritional requirements, all of which may influence

diet composition (Wuenschel, et al., 2006) and, ultimately, how resources are utilized. Because enhancement of available food resources can have direct impacts on vital rates, knowledge of habitat-specific diet composition is essential to determining the role of artificial habitat in estuarine food webs. Positive impacts upon fish diet may result from general increases in prey abundance, increases in preferred prey, increased diversity of prey items, or increases in capture efficiency. Documentation of habitat-specific resources and habitat-specific utilization are critical to management of exploited populations, can provide greater resolution than density comparisons, and can be used to calculate production values.

1.5 Thesis Goals and Objectives

Many studies cite evidence for enhancement based on increased abundances, densities or diversity of organisms at artificial reef sites, but fail to gather explicit data on growth or survival rates for species of interest. True enhancement requires the organism of study to exhibit increased vital rates such as recruitment, growth, or survival as enhancement based on abundances, densities, or diversity is equivocal at best (Lindberg, 1997). The objective of this study was to evaluate the potential for limestone cobble, deployed as mimic artificial oyster reefs (MAORs), to enhance the feeding ecology of the juvenile fish community in marsh ponds. As part of a larger study, information on vital rates, such as growth and survival was gathered but not reported on as part of this thesis. The chapters in this thesis are devoted to assessing MAOR utilization and habitat quality to provide evidence to explain potential differences, or a lack there of, observed in abundance, density, diversity, and growth or survival rates of marsh pond fishes. I sought to examine the impact of MAOR addition not only on juvenile fishes but on multiple

dimensions of the marsh pond community as the ecological impact of reef addition is often quite variable. The deployment of artificial structures aimed at enhancing young life stages is relatively uncommon as most studies utilize artificial structures for increasing catches in developing or mature fisheries. Therefore, data collection from multiple ecological viewpoints should prove useful in developing a well-rounded and informative assessment of the ecological impact of MAOR addition. To accomplish the goal of assessing utilization I first sought to determine the impact of MAOR addition on the abundant prey base potentially available to juvenile fishes in marsh ponds (Chapter 3). This would allow identification of prey taxa that were vulnerable to habitat change through increasing (MAOR addition) or decreasing (replacement of natural habitat) favorable habitat. I then sought to determine which food resources were most important to juvenile fishes in marsh pond food webs and determined if the impacts observed in the potential prey base were translated into higher trophic levels (Chapter 4). Potential impacts to marsh pond communities are certainly not limited to feeding ecology and thus I also sought to examine fish condition. Data on fish condition served two purposes: 1) by comparing fish condition between habitat types I was able to assess the relative quality of MAOR habitat versus other natural habitats and 2) in the absence of diet-related differences, condition differences could be indicative of other impacts, such as niche partitioning or predation refuge, attributable to MAOR addition. An additional aspect of this thesis was devoted to evaluating the efficacy of a relatively novel technique for assessing juvenile fish condition. Bioelectric impedance analysis provides indirect, nondestructive estimates of compositional condition that are rapidly collected, repeatable, and independent of size. This technique has recently been applied to various fishes for

condition assessments and its potential for use in studies of juvenile fishes is experimentally examined and discussed in Chapter 2.

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[1]*CHAPTER 2: USE OF BIOELECTRIC IMPEDANCE ANALYSIS TO ASSESS TOTAL-BODY CONDITION AND PREDICT ENERGY DENSITY IN JUVENILE ATLANTIC CROAKER (*MICROPOGONIAS UNDULATUS*)

2.1 Introduction

Condition indices are frequently used to compare the fitness of individuals, cohorts, or populations of fish at various life-stages. Size-based condition indices are relatively simple and require only minimal information such as length and weight (Iqbal and Suzuki, 2009; Sundstrom et al., 2009; Wanner and Klumb, 2009). When the underlying assumptions of size-based techniques are satisfied, particularly the assumption of isometric growth (see Bolger and Connolly, 1989; Cone, 1989), one can draw valid inferences about relative condition with respect to predicted values. Conclusions drawn only from regressions of length versus weight, however, may draw an incomplete picture of condition because size-based techniques do not incorporate compositional information (Setzler-Hamilton and Cowan, 1993). Condition measures such as energy density do incorporate generalized compositional information but are time-consuming, expensive, and require individuals to be sacrificed.

Bioelectric impedance analysis (BIA) is a relatively novel technique for the rapid and repeatable assessment of total body fish condition (Cox and Hartman, 2005). BIA relies on the electrical properties of biological materials to provide direct measurements of extra- and intracellular water content. Variations in the proportional composition of conductive and dielectric cellular components are directly related to changes in fish

[1]*Permission to use Chapter 2 in this thesis is shown in Appendix 4.

condition (for review of the physical properties of BIA see Kushner, 1992). BIA has been widely applied to mammals (Farley and Robbins, 1994) and human subjects (Lukaski 1987; Baumgartner et al., 1989; Kyle et al., 2004; Barbosa-Silva et al., 2005), especially in disease studies (Baarends et al., 1997; Horlick et al., 2002), and is now receiving considerable attention in applications for fish (Duncan et al., 2007; Pothoven et al., 2008; Willis and Hobday, 2008). BIA generates compositional data without sacrificing individuals, thus enabling repeated measures of the same individuals over time. BIA has been applied as an investigative technique in both laboratory and field studies of fish (Cox and Hartman, 2005; Willis and Hobday, 2008; Cox and Heintz, 2009), in aquaculture (Duncan et al., 2007), and for fish conservation purposes (Willis and Hobday, 2009).

In this study, we applied BIA techniques to juvenile Atlantic croaker, *Micropogonias undulatus*, in a controlled tank environment. The objectives of this study were: 1) to assess the efficacy of BIA in applications to juvenile fish where metabolic turnover is presumably rapid, 2) to compare the compositional resolution of BIA to traditional size-based and compositional condition techniques and 3) to predict compositional condition using BIA-derived condition estimates.

2.2 Methods

2.2.1 Experimental Setup

Wild-caught Atlantic croaker were collected near the Louisiana Universities Marine Consortium (LUMCON) facility in Cocodrie, LA in August of 2008 and held in a 600-gallon recirculation tank at LUMCON. A total of 130 fish were each injected with a

unique, non-conductive, glass-encased PIT tag for individual identification and monitoring through time. Each PIT tag was inserted into the body cavity just anterio-laterally to the anus using an injection needle. In February 2009, tagged fish (n =130) were transported to the Louisiana State University Aquaculture Center in Baton Rouge, LA and held in two, 600-gallon recirculation tanks each with a 30 L min^{-1} flow rate (approximately 1 cycle every 90 minutes). Both tanks were connected to a single recirculation system and water quality was maintained via a one cubic meter floating-bead bio-filter with daily monitoring of ammonia and nitrate levels. Salinity levels were maintained between 10 and 12 ppt. Temperature was held constant using two (four total) Finnex titanium 800-watt in-tank water heaters (± 1.1 °C) and all fish were fed to satiation once daily prior to experimentation. After a four-week acclimation period at the aquaculture facility, fish were redistributed evenly between the two tanks with each tank representing a treatment (n = 65 fish per treatment). Fish in the fed treatment were fed *ad libitum* throughout the experiment by allowing fish to feed on a predetermined amount of pellets; all uneaten pellets remaining after approximately 1 hr were removed. Fish in the starved treatment were initially fed a ration of 1% body weight per day but were reduced to a zero food ration beginning on day 15 and continuing until the end of experimentation to maximize the physiological contrast between the two treatments.

2.2.2 Size-Based Condition

Standard length (SL; mm) and weight (Wt; grams) were recorded for all fish in each treatment, every five days for 45 days (February 24 – April 10, 2009). Daily growth was determined by linear length/weight regressions.

Relative condition was calculated using the formula:

$$K_n = (W/ L^b) \times 10^n \quad (1)$$

where K_n is relative condition, W is the weight (g) of each fish, L is the standard length (mm) of each fish, n is an arbitrarily determined scaling factor, and b is the slope of the regression relationship for growth rate determined from a sample population rather than assuming a slope of $b = 3$, which is indicative of isometric growth (Bolger and Connolly, 1989; Cone, 1989). The exponent b is calculated using a log-log regression of length and weight in a power model and testing for a significant difference for the input value of b, initially set at $b = 3.0$. If a significant difference is detected, subsequent values of b are input until there is no significant difference between the input value and the observed value of b in the model.

2.2.3 Compositional Condition

Every five days, five fish were randomly selected from each treatment to be sacrificed for energy density analysis. Muscle tissue (MED; fillet only) was separated from carcass material (CED; skin, skeleton, scales, major body organs, and minimal residual muscle tissue) prior to drying. Muscle tissue and carcass material samples were each homogenized and individually dried for 48 hours at 60 °C. Energy density ($J \cdot g^{-1}$) analysis was performed separately on dried muscle tissue and carcass material samples using a Parr 6200 oxygen bomb calorimeter. Stomachs and intestines were discarded as they potentially contained unassimilated material. Total body energy density (TBED; $J \cdot g^{-1}$; dry weight) was calculated using the formula:

TBED = [(MED x dry weight (g)) + (CED x dry weight (g))] / total dry weight (g) (2)

where MED (muscle energy density) and CED (carcass energy density) are multiplied by their respective dry weights in grams, summed, and then divided by the total dry weight of the fish having discarded the stomach and intestines. MED, CED, and TBED values were reported as joules per gram and represent dry weight energy densities.

2.2.4 BIA-Based Condition

A Quantum-X bioelectric compositional analyzer (RJL Systems) was used to determine all resistance (R; ohms) and reactance (X_c; ohms) values in series. Electrode pairs were constructed using 28-gauge needle electrodes (Grass Technologies) pierced through rubber stoppers and wrapped with heat-shrink tubing around low gauge wire to provide memory and stability without human interaction during data collection. Both separation within an electrode pair and needle penetration depth were held constant at 5 mm. Electrode functionality was tested for consistency and sensitivity using a 500 ohm resistor provided by the manufacturer prior to initiation of data collection, periodically throughout the experiment, and prior to the use of any replacement electrodes.

During the tank experiment, both R and X_{cs} readings were taken concomitantly with lengths and weights and allmeasurements were taken on a non-conductive wooden board. Fish were not blot-dried to reduce slime-coat removal and decrease susceptibility to infection and parasites. For R and X_{cs} measurements, the anterior electrode pair was placed in line with the posterior-most point of the operculum at the midpoint between the first dorsal fin and the lateral line. The posterior electrode pair was placed in line with the

base of the posterior-most second dorsal fin-ray at the midpoint between the dorsal and the lateral line prior to the caudal peduncle (Cox and Hartman, 2005). The potential for adverse effects from electrode penetration was visually monitored throughout the experiment.

An additional experiment was conducted to test for possible effects of PIT tags (22 mm) and blot-drying on BIA measures. Several common estuarine fishes (n=35) were collected via otter trawl, identified to species and measured for standard length. BIA measures were taken on each fish after it was subjected to each of three treatments: 1) no blotting and no PIT tag, 2) blotting but no PIT tag, and 3) no blotting but inserted with PIT tag. PIT tags were inserted into the body cavity using an injection needle and R and X_c were measured in series.BIA readings were taken on a non-conductive board and the treatment order was alternated for each successive fish. For subsequent treatment measurements on each fish electrode needles were inserted into puncture marks from previous treatment measurements (Cox et al., 2010).

The BIA-derived condition measures, phase angle (*PA*) (Lukaski, 1987; Kyle et al., 2004; Fish and Geddes, 2008) and the composition index (*CI*) (Willis and Hobday. 2009) were calculated from R and X_{cs} values using the following formulas:

$$PA(°) = (\arctan (X_{cs}/R))\ 180^{\circ}/\pi \quad (3)$$

$$CI = L^2 X_{cs}^{-1} \quad (4)$$

Reactance in series (X_{cs}) was used as the impedance component for *CI* and fish standard length (mm) was used as a proxy for the separation distance (L) between electrode pairs.

2.2.5 Statistical Analyses

Linear autoregressions were used to examine relationships (R^2) among size-based, energetic, and BIA-based condition measures and their components, but were not used to evaluate the significance of treatment means over time. Separate significance tests were conducted for each dependent variable (SL, Wt, K_n, MED, CED, TBED, R, X_{cs}, PA, CI; Table 2.1) using repeated measures analysis of variance (ANOVA) (Littell et al., 2006) in SAS v9.1 (SAS Institute, 2002). Mean values for each dependent variable were compared between treatments over time by including the main effects: day, tank, and the interaction term (day*tank) in each model run. The main effect term "Day" was specified as the repeated term with the class variable "Fish (Tank)" specified as the subject term upon which repeated measures were conducted. For the dependent variables MED, CED, and TBED the class variable "Tank" was specified as the repeated measures term because fish cannot be repeatedly sampled for energy density as it is destructive. An autoregressive covariance structure was specified in all repeated measures tests. Table 2.1 lists the *p*-value for the interaction term only (Tank*Day) from the type III test of fixed effects for each dependent variable from repeated measures ANOVA. Mean values and standard errors were reported as least squares means (LSMeans) and all statistical tests were evaluated at a significance level of $\alpha = 0.05$.

2.2.6 Size- and BIA-Based Condition Measures to Predict Energy Densities

Linear regression analyses were performed to quantitatively assess the efficacy of the non-destructive condition measures PA and K_n to predict energy densities from both

starved and fed treatments. Energy density values for MED, CED, and TBED were individually regressed against values for *PA* and K_n. Values for *PA* and K_n were used both

Table 2.1. Output statistics for all condition measures and their components using repeated measures ANOVA (p-values are from the interaction term Day*Tank using type III test of fixed effects).

Metric	N	F-value	*p*-value
Standard Length (mm)	333	11.25	<0.001
Weight (g)	333	33.43	<0.001
Relative Condition (b = 3.5)	333	20.21	<0.001
Muscle Energy Density ($J\ g^{-1}$)	63	2.00	>0.05
Carcass Energy Density ($J\ g^{-1}$)	64	3.78	0.003
Total Body Energy Content ($J\ g^{-1}$)	63	3.65	0.004
Resistance (ohms)	333	1.65	>0.05
Reactance (ohms)	332	1.89	>0.05
Phase Angle (^{o})	333	2.98	0.005
Composition Index ($L^2\ X_{cs}^{-1}$)	333	2.95	0.005

individually and in combination to predict energy densities. For this analysis, MED, CED, and TBED values were calculated in terms of wet-weight energy density as this slightly improved model fit for the predictive equations. To further investigate the relationship between BIA and energy content, linear regressions were generated using *PA* and *CI* versus total body energy content (TBEC; Joules) for each treatment group. TBEC was calculated by multiplying the total body weight (g) by TBED ($J{\cdot}g^{-1}$).

2.3 Results

2.3.1 Size-Based Condition

Fish from the fed treatment increased in both mean weight (0.41 g day^{-1} ± 0.12 SE) and length (0.30 mm day^{-1} ± 0.09 SE), whereas fish from the starved treatment decreased in weight (-0.19 g day^{-1} ± 0.15 SE) and displayed only minimal length increases throughout the study (0.04 mm day^{-1} ± 0.10 SE; Table 2.2). Repeated-measures ANOVA indicated significant differences in growth rates between treatments over time (Table 2.1). A significant difference ($p< 0.05$) in relative condition (K_n) was observed between treatments by the third sampling event (10 days) with K_n remaining relatively constant in the fed treatment and rapidly declining in the starved treatment (Figure 2.1). Linear regression of the length/weight data indicated the slopes for both treatments were not significantly different from 3.5. Therefore, the value of 3.5 was assigned to the growth exponent '*b*' in the relative condition equation for both treatments instead of the assumed value of 3.0.

2.3.2 Compositional Condition

Mean MED values increased in the fed treatment (19.2 joules day^{-1}) and decreased in the starved treatment (-23.0 joules day^{-1}; Figure 2.2). Mean CED values increased in the fed treatment (22.2 joules day-1; Figure 2.2) and decreased in the starved treatment (-86.6 joules day^{-1}). In concurrence with its components, mean TBED values also increased in the fed treatment (22.6 joules day^{-1}) and decreased in the starved treatment (-71.5 joules day-1; Figure 2.2). Despite diverging trend lines the dependent variable MED showed no significant difference between treatment groups over time ($p>$

Table 2.2. Daily LSmeans estimates for all condition measures and their components; Tank A – fed treatment, Tank B – starved treatment.

Tank A								
Day	**0**	**5**	**10**	**15**	**20**	**25**	**35**	**45**
Fish Remaining	65	60	55	50	44	39	28	12
Standard Length (mm)	134.0	133.2	129.4	142.2	134.4	136.8	142.8	146.0
Weight (g)	51.04	54.88	48.74	64.8	55.1	57.84	65.76	69.97
Relative Condition (b = 3.5)	5.67	6.37	6.18	5.98	6.16	6.07	5.81	5.87
Muscle Energy Density (J g^{-1})	25061.11	25234.29	25467.88	25920.47	24545.10	25230.48	25445.79	26398.74
Carcass Energy Density (J g^{-1})	19892.07	20634.11	21491.49	21654.33	20240.48	21304.22	20725.53	21769.23
Total Body Energy Content (J g^{-1})	21798.35	22351.60	23149.61	23406.05	21927.47	22970.83	22554.27	23581.32
Resistance (ohms)	541.58	658.56	591.90	571.10	585.25	445.70	582.99	515.38
Reactance (ohms)	175.37	233.47	204.17	192.62	194.16	168.88	174.96	134.21
Phase Angle (°)	18.30	19.82	19.25	18.90	18.59	21.11	16.93	14.74
Composition Index ($L^2 X_{cs}^{-1}$)	100.35	79.73	83.54	103.08	103.00	104.19	148.00	170.27
Tank B								
Day	**0**	**5**	**10**	**15**	**20**	**25**	**35**	**45**
Fish Remaining	65	60	55	50	45	39	29	23
Standard Length (mm)	130.4	129.4	130.6	138.2	138.8	135.0	130.0	132.8
Weight (g)	48.88	49.34	50.16	62.04	54.5	50.6	41.07	44.35
Relative Condition (b = 3.5)	6.05	6.28	6.09	6.25	5.39	5.51	5.11	5.07
Muscle Energy Density (J g^{-1})	25806.74	25665.99	25338.76	25349.22	25078.35	25167.51	25123.04	24598.57
Carcass Energy Density (J g^{-1})	20050.61	22315.91	--	21363.04	20273.99	20323.11	17424.44	17910.37
Total Body Energy Content (J g^{-1})	22907.78	23606.84	--	23026.90	22912.59	22169.76	21336.02	19948.48
Resistance (ohms)	2746.25	3135.45	2808.05	2826.29	2811.82	2333.67	2999.93	2566.01
Reactance (ohms)	185.69	229.76	210.78	205.59	197.29	167.42	195.47	138.08
Phase Angle (°)	17.66	18.83	19.43	18.87	18.26	19.11	16.97	14.28
Composition Index ($L^2 X_{cs}^{-1}$)	91.24	71.65	80.81	85.29	118.35	104.61	94.49	146.06

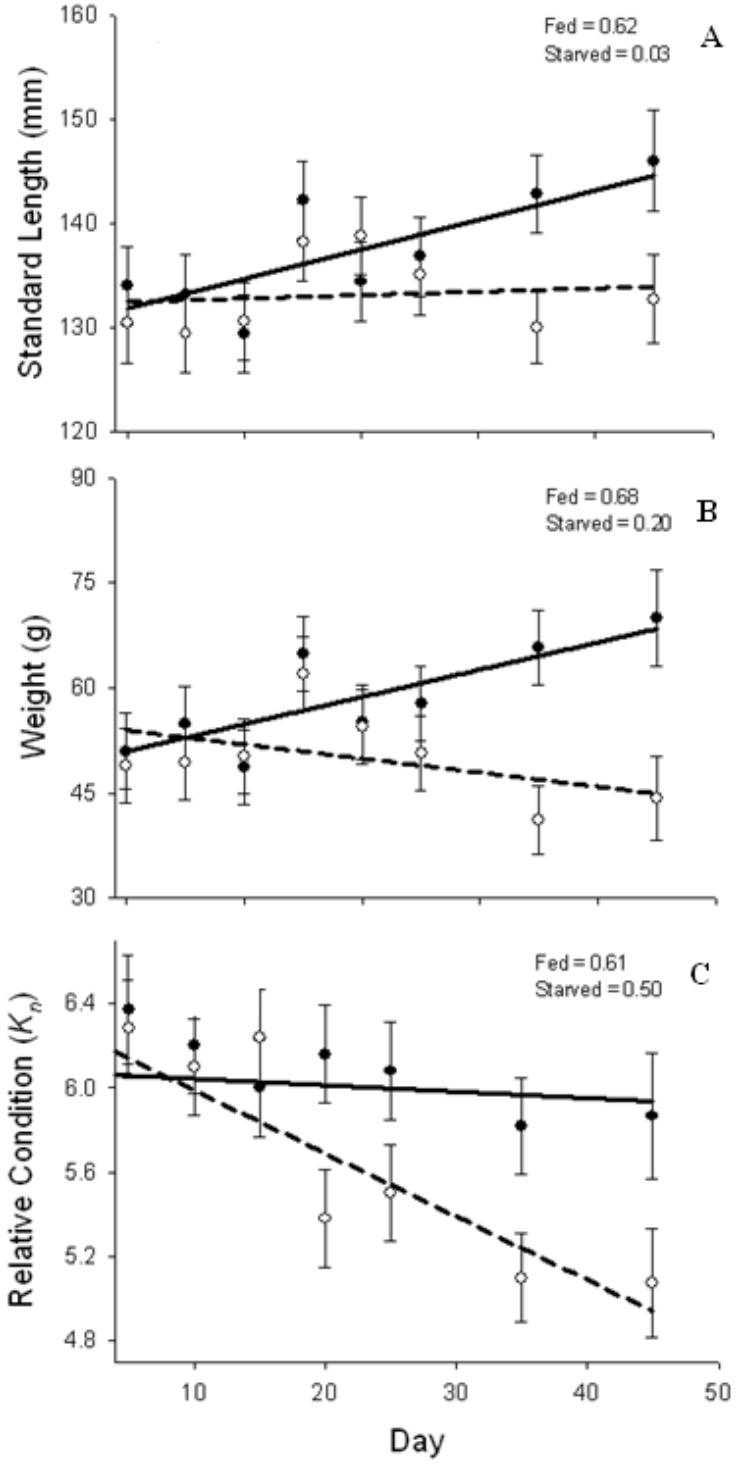

Figure 2.1. Size-based condition estimates and their components for each treatment (fed – solid line and filled circles; starved – dashed line and open circles) using linear autoregression lines overlaid onto mean values of standard length (mm: A), weight (g; B) and relative condition (K_n; C) versus time (days) with standard error bars. R-square values for each treatment are listed in the top right corner of each plot.

0.05) where as CED and TBED values were significantly different after 45 days ($p< 0.01$ and $p = 0.014$ respectively).

2.3.3 BIA-Based Condition

No significant affect of PIT tags was found on R and X_{cs}values (p> 0.05; paired t-test; SAS v9.2) between fish with and without PIT tags in the field experiment, but both R and X_{cs} values were significantly higher in blot-dried fish (p<.0001). Mean R and X_{cs} values in the blot-dried treatment were 30.4 and 17.6 ohms higher than fish that were not blot-dried. Although the effect of blot-drying was significant this source of error was relatively low (5-10% for R and X_{cs}, respectively and5% for *PA*). Both R and X_{cs} values were non-significant between treatment groups over time ($p> 0.05$; Table 2.1) in the tank experiment. Slopes for each treatment were non-parallel and decreased over time in both treatments for both R and X_{cs} (Figure 2.3). Despite the non-significance of these BIA components, both *CI* and *PA* indicated significant differences between treatments (Table 2.1). Slopes from *CI* indicated the fed treatment increased at a faster rate than the starved treatment (Figure 2.3) while *PA* values for both the fed and starved treatment decreased (Figure 2.3). Both *PA* and *CI* indicated significant divergence in feeding treatments after 25 days (Figure 2.3). Adverse effects from electrode needle penetration were minor with slight bruising occurring in only a few fish and no fish developed visible infection at penetration sites.

2.3.4 Size- and BIA-Based Condition Measures to Predict Energy Densities

Linear relationships between BIA components and energy densities were relatively low compared to previous experiments using BIA to predict proximate

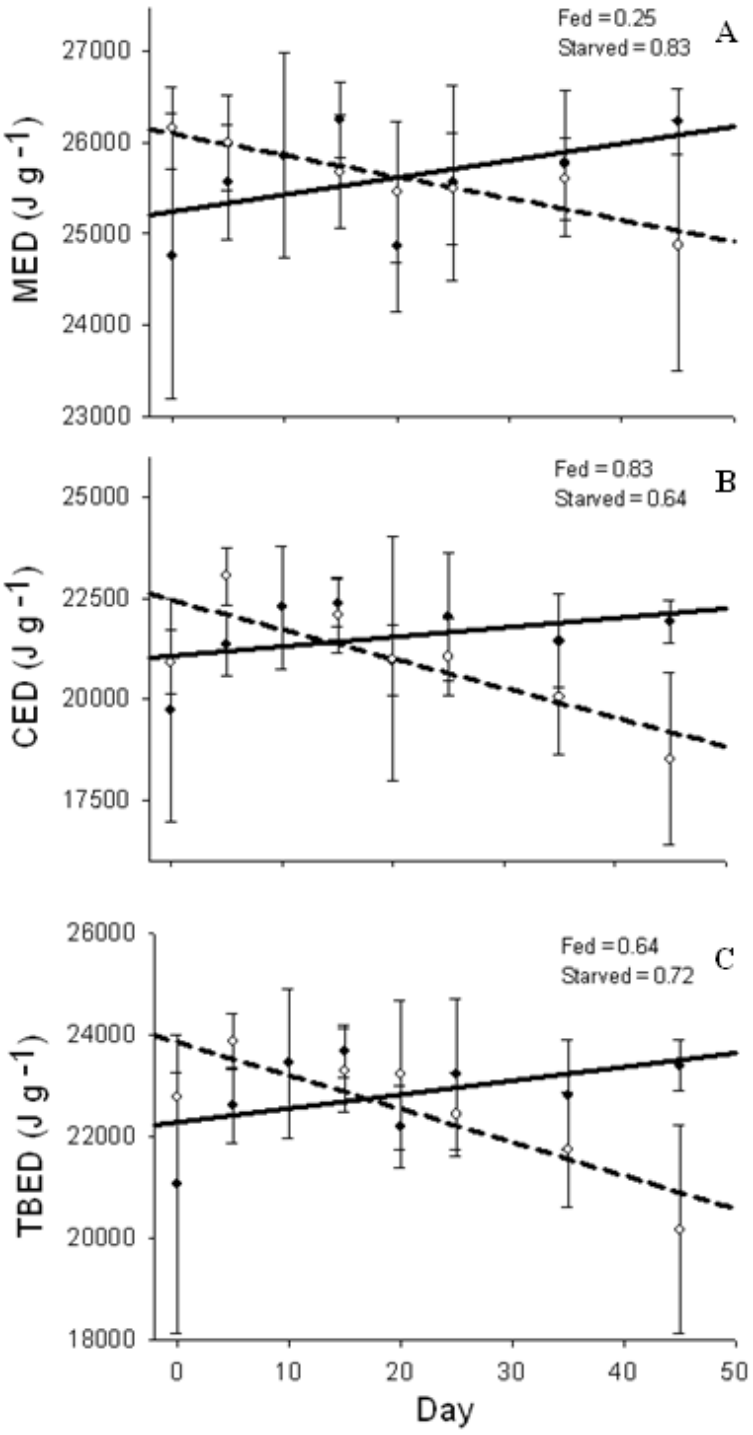

Figure 2.2. Energy-based condition estimates for each treatment (fed – solid line and filled circles; starved – dashed line and open circles) using linear autoregression lines overlaid onto mean values of muscle energy density (MED; $J \cdot g^{-1}$; dry weight; A), carcass energy density (CED; $J \cdot g^{-1}$; dry weight; B) and total body energy density (TBED; $J \cdot g^{-1}$; dry weight; C) versus time (days) with standard error bars. R-square values for each treatment are listed in the top right corner of each plot.

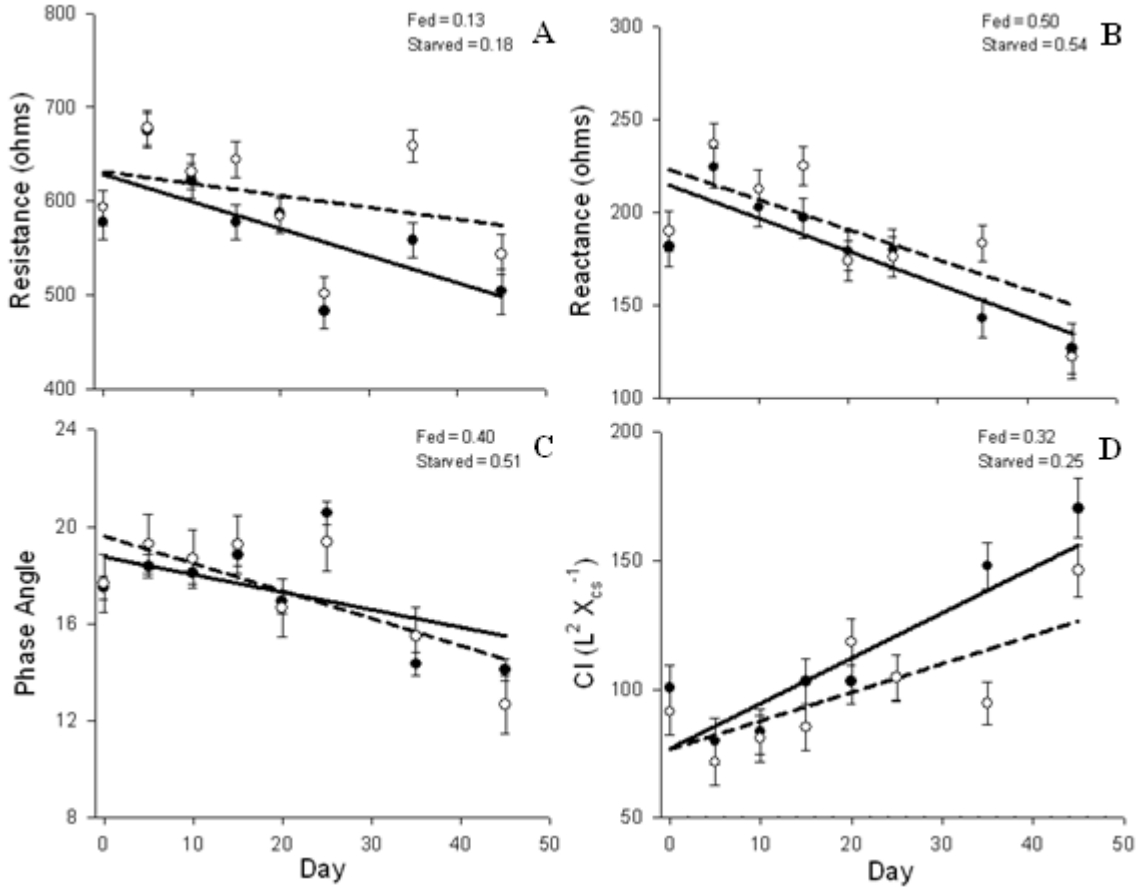

Figure 2.3. BIA-based condition estimates for each treatment (fed – filled circles; starved – open circles) using linear autoregression lines overlaid onto mean values of resistance (*R*; ohms; A), reactance (X_{cs}; ohms; B), phase angle (*PA*; C), and composition index (*CI*; $L^2 X_{cs}^{-1}$; D) versus time (days) with standard error bars. R-square values for each treatment are listed in the top right corner of each plot.

components (e.g., Cox and Hartman, 2005). Correlation measures between energy components and their size- or BIA-based estimators were much lower for fish from the fed treatment than for fish from the starved treatment (Table 2.3). In the fed treatment, relationships between estimators and energy densities were very low (r^2 = 0.0 - 0.15) for all three energetic components (MED, CED, TBED). In the starved treatment,

Table 2.3. Linear regression estimates (standard errors) for the estimators: relative condition (K_n; b = 3.5) and phase angle (PA-degrees) to predict energy components: muscle (MED; $J \cdot g^{-1}$; wet weight), carcass (CED; $J \cdot g^{-1}$ wet weight), and total body energy densities (TBED; $J \cdot g^{-1}$ wet weight); Tank A - fed treatment, Tank B - starved treatment.

Tank A

Component	Estimator	N	Intercept (SE)	K Estimate (SE)	PA Estimate (SE)	p-value	Adj. r^2
MED	K	33	2961.86 (1817.68)	574.94 (300.99)	--	>0.05	0.08
MED	PA	34	4919.04 (948.26)	--	86.32 (53.51)	>0.05	0.05
MED	K, PA	33	3001.55 (1835.25)	444.17 (361.93)	42.68 (64.23)	>0.05	0.06
CED	K	33	5116.05 (1705.93)	221.11 (282.48)	--	>0.05	0.00
CED	PA	34	4375.93 (800.64)	--	118.20 (45.18)	0.01	0.15
CED	K, PA	33	5244.02 (1579.60)	- 200.52 (311.51)	137.60 (55.28)	0.04	0.13
TBED	K	33	4451.40 (1612.12)	330.64 (266.95)	--	>0.05	0.02
TBED	PA	34	4560.34 (776.21)	--	107.54 (43.80)	0.02	0.13
TBED	K, PA	33	4550.96 (1541.61)	2.62 (304.02)	107.05 (53.95)	>0.05	0.10

Tank B

Component	Estimator	N	Intercept (SE)	K Estimate (SE)	PA Estimate (SE)	p-value	Adj. r^2
MED	K	29	2758.10 (1221.89)	618.39 (211.13)	--	0.0068	0.21
MED	PA	29	3281.10 (687.01)	--	170.95 (38.23)	0.0001	0.40
MED	K, PA	29	3071.66 (1087.47)	67.02 (266.52)	161.01 (55.47)	0.0007	0.38
CED	K	30	- 1018.44 (1100.90)	1250.64 (192.35)	--	<.0001	0.59
CED	PA	30	685.09 (683.54)	--	308.79 (38.51)	<.0001	0.69
CED	K, PA	30	- 504.38 (936.42)	482.05 (269.90)	220.22 (61.93)	<.0001	0.71
TBED	K	29	- 24.25 (1343.86)	1084.25 (232.20)	--	<.0001	0.43
TBED	PA	29	1554.16 (733.88)	--	262.55 (40.84)	<.0001	0.59
TBED	K, PA	29	378.38 (1123.44)	376.26 (275.34)	206.75 (57.30)	<.0001	0.60

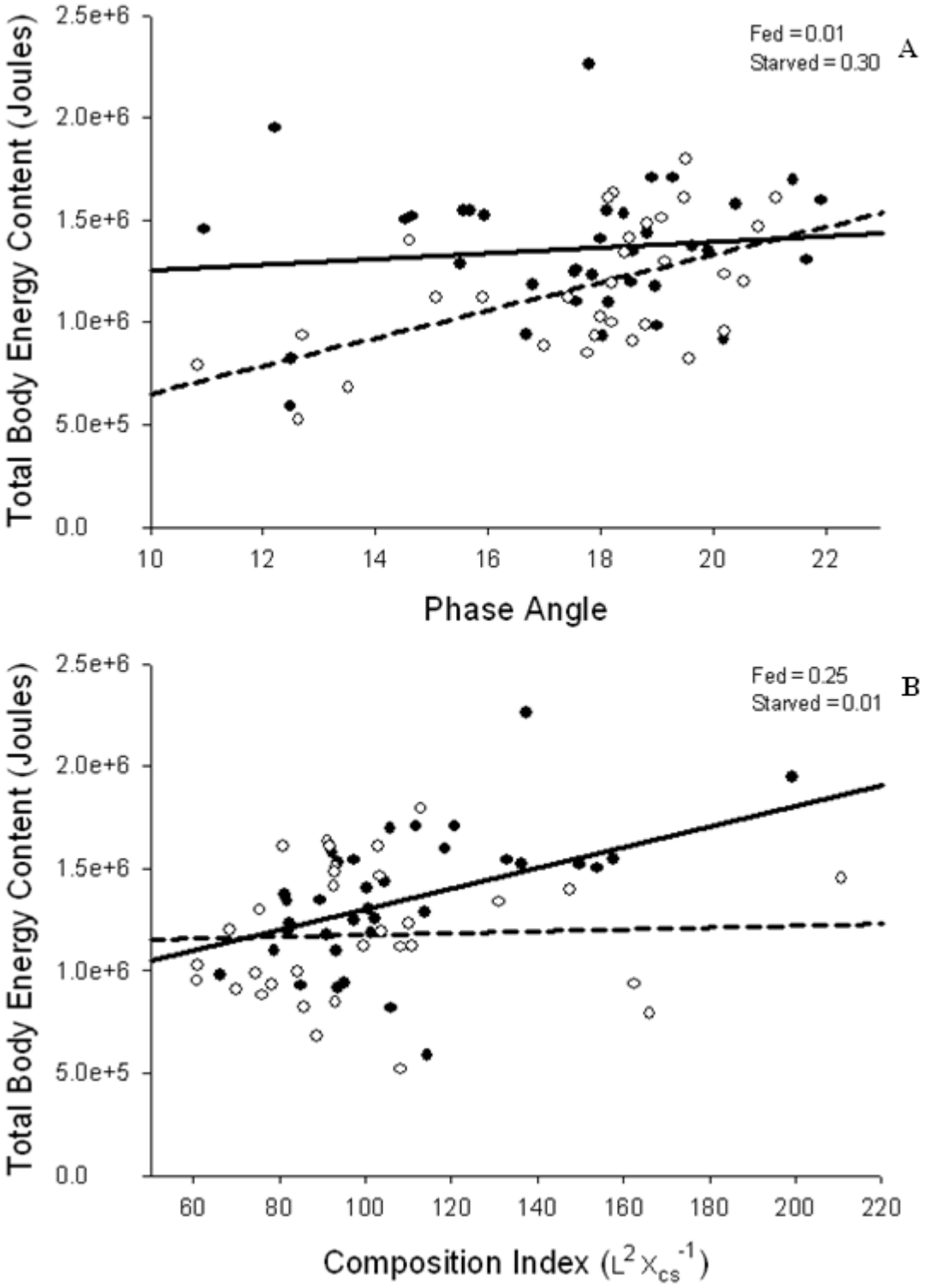

Figure 2.4. Linear regressions of phase angle (*PA*; A) and composition index (*CI;* $L^2 X_{cs}^{-1}$; B) versus total body energy content (J; total body wet weight) for each treatment (fed - solid line, filled circles) and starved treatment (dashed line, open circles). R-square values for each treatment are listed in the top right corner of each plot.

relationships were much higher, especially for CED, with r^2 values ranging between 0.21 and 0.71. Using both *PA* and $K_n$in combination to predict energy densities provided only minor increases to r-square values. Regressions of total body energy content versus either PA or CI also showed very low r^2 values (Figure 2.4).

2.4 Discussion

Throughout the study, fish from the fed treatment displayed positive growth rates while fish from the starved treatment displayed negligible (length) or negative (weight) growth. Daily growth (mm day^{-1}) of fish from the fed treatment was relatively low but within the reported range of growth rates from the literature (Miller and Able, 2002; Miller et al., 2003; Ross, 2003). Both size-based and energetic condition measures indicated significant changes in fish condition between fed and starved treatments. Declines in relative condition (K_n) of starved fish became significant within 10 days where as significant differences in compositional condition occurred after several weeks. More rapid changes in lengths and weights compared with energetic densities suggest fish were likely sacrificing growth to minimize decreases in condition.

Although BIA indicated significant differences between treatments, its utility as a reliable tool to interpret compositional condition was inconclusive in this experiment. We observed weak trends in phase angle and composition index and observed no significant trends among their components (i.e., *R* and X_{cs}) with respect to treatments. Our initial expectation was that BIA techniques (*PA* and *CI*) would resemble trends observed in K_n and energy density. Despite showing a significant difference between treatments, trend lines for *PA* indicated declining condition in both fed and starved treatments suggesting

all fish were physiologically similar throughout the experiment; a conclusion contradictory to interpretations from both size- and composition-based condition measures. The *CI* also indicated a significant difference between treatments but indicated increasing condition in the starved treatment, contrary to trend lines observed in K_n and TBED. We attribute the increasing condition values for CI in the starved treatment to the vulnerability of the composition index to size-dependent effects. In this case, fish from both treatments displayed similar X_{cs} values over time, yet the two groups diverged because of significantly different growth rates. Previous research (Cox and Hartman, 2005; Cox and Heintz, 2009) provides strong evidence for the efficacy of BIA (phase angle) in fish (particularly in salmonids), however, few studies have applied BIA to juvenile fish (Duncan et al. 2007; Hanson et al., 2010). Duncan et al. (2007) attributed low correlations among BIA estimates to low total-body lipid content and the high physiological energetic demands associated with rapidly growing juvenile cobia, *Rachycentron canadum* (see also Dabrowski, 1986). As lipids (i.e. fat) are the primary form of stored energy as well as the primary dielectric component influencing impedance metrics, a weak relationship between nutritional condition and BIA in this experiment suggests lipid contents may have been too similar or too low in the two treatments to generate detectable differences. Gallagher et al., (1991) found that Atlantic croaker stored approximately 3% of their caloric intake prior to the warmer months of summer; in contrast they stored between 7 and 10% of their nutrient intake as lipid in June and August, respectively. Adult fish of other species have been found to store more than 20% of total dry mass as lipid with peak storage usually occurring prior to reproduction (Anthony et al., 2000). As this study occurred during the months of February through

April, the initial lipid component in these juvenile Atlantic croaker may have been too low to allow sufficient compositional contrast over time necessary for detection using BIA. Although Hanson et al. (2010) found moderately strong relationships between BIA and proximate components using juvenile salmonids, condition estimates derived from lipid contents (i.e., BIA and microwave energy meters) showed almost no relationship to proximate components.

In addition to low total-body fat content, energy density analyses suggest the specific distribution of fat throughout the body may have contributed to confounded BIA values. Lipid material is more energetically dense than other catabolic energy sources (39.3 $kJ \cdot g^{-1}$ of lipid; 17.6 $kJ \cdot g^{-1}$ of carbohydrate; 18.0 $kJ \cdot g^{-1}$ of protein; Schmidt-Nielsen, 1997). In this experiment energy density declined more rapidly in the carcass material than was observed in muscle tissue. Given that the majority of carcass material was comprised of the skeleton and scales, both of which are extremely poor sources of energy and are typically broken down only after periods of extreme starvation, the relatively rapid decline in CED suggests the majority of fat mobilization was associated with the liver or viscera. These two organs often contain the primary tissues associated with lipid storage in fish (Black and Love, 1986; Black and Skinner, 1986; Rios et al., 2006). Our electrode array oriented along the anterior-posterior axis may not have detected the lipid storage (Cox et. al., 2010) and subsequent catabolism and therefore may not be appropriate when major lipid storage is not associated with muscle tissue.

We do not attribute the weak BIA relationships observed in this experiment to confounding affects from PIT tags or salinity (i.e., not blot drying). The field experiment indicated no significant affect of PIT tags on BIA readings. Although significant, the

affect of salinity was relatively weak and treatment-wide, only affecting the absolute values of BIA without compromising the integrity of the tank experiment. In addition, salinity in the field experiment was higher (~25 ppt) and probably had a greater affect on BIA values compared with the lab experiment (10-12 ppt).

This research provides insight into the potential physiological limitations and efficacy of BIA in estimating total-body condition of juvenile fish. This study is in congruence with Duncan et al., (2007), and Hanson et al., (2010) and highlights the dependency of BIA upon physiological contrasts of dielectric components necessary for accurate condition estimates. It is also important to consider changes in the relative abundance and compartmental dynamics of dielectrics within the body at various life stages. Previous research suggests strong relationships (r^2 = 0.97-99) between BIA and proximate body compartments (Cox and Hartman, 2005; Cox and Heintz, 2009). However, fish species used in previous experiments may have been better suited for BIA techniques due to naturally inherent compositional qualities (e.g., higher fat content) or suitable life-stages. Therefore, developing a complete understanding of the relationships between bioelectric impedance and ontogeny is critical as BIA has much potential value for a wide variety of biological and ecological applications, especially when using limited or endangered species.

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CHAPTER 3: EFFECTS OF MIMIC ARTIFICIAL OYSTER REEFS ON MEIOFAUNA AND MACROFAUNA IN MARSH PONDS: A BEFORE-AFTER-CONTROL-IMPACT ANALYSIS

3.1 Introduction

3.1.1 Artificial Habitats and Habitat Enhancement

Artificial habitats have been deployed in a wide variety of locations and designs (see reviews by Grove et al., 1989; Grove et al., 1991; Pickering and Whitmarsh, 1997; Relini et al., 2007), often with intent to increase catches, reduce effort within a local fishery (Whitmarsh et al., 2008), or enhance ecosystem productivity (Relini et al., 2007). Artificial habitats have been shown to augment ecological and biological processes, and can reduce the intensity of negative stressors due to their resilience and functionality (Gardner et al., 1996; Hernkind et al., 1997), the dynamics of colonization (Sale and Dybdahl, 1975; Rodney and Paynter, 2006), and subsequent utilization by consumers (DeMartini et al., 1994; Johnson et al., 1994; Fabi et al., 2006). Primary and secondary consumers often utilize epiphytic, epifaunal, or fouling communities rather than direct consumption of living or decomposing host substrate (Moncreiff and Sullivan, 2001; Fabi et al., 2006). This implies that some artificial habitats can be productive if given they provide adequate availability of areal substratum for colonization and food web support. The enhancement of trophodynamics at multiple levels (e.g., Reed et al., 2006; Lingo and Szedlmayer, 2006; Perkol-Finkel, 2007) can increase diversity (Fabi et al., 2004) as well as local (DeMartini et al., 1994; Johnson et al., 1994; Relini, et al., 2007) or even ecosystem level productivity.

3.1.2 Oyster Reefs as Habitat

The importance of oyster reefs to estuarine ecosystems was well exemplified by the loss of approximately 98% of the original Chesapeake Bay oyster population, which resulted in significant reductions in water quality and trophic cascades (Rothschild et al., 1994; Coen et al., 1999). Because oyster reefs produce large surface areas of hard substrate, they enhance recruitment of sessile invertebrates and provide critical settlement habitat for oyster spat (Coen and Grizzle, 2007). Settlement may be enhance by the vertical relief and heterogeneous habitat complexity which can reduce horizontal mean water velocities down-current of the leading reef edge that can enhance vertical movement, creates micro-turbulent flows that may deliver larvae directly to reef substrate (Eckman, 1987; Abdelrhman, 2003), and can increase persistence after settlement (Bologna and Heck, 2000; Koehl, 2007). Enhancement of primary production may result from accumulating drifting algae (Davis et al., 2009) or when epiphytic producers occupy substrate surfaces (Reed et al., 2006; Pondella et al., 2006).

Compared with other complex habitat types, oyster reefs can produce similar and even increased densities of fishes and invertebrates (particularly structure-associated species), often exhibiting significantly higher densities when compared with non-vegetated, mud bottoms (Zimmerman et al., 1989). Complex microspaces provide food and structurally rigid refugia that may greatly reduce macrofaunal predation pressure (Hall and Bell, 1988). This results in enhanced biodiversity, especially among invertebrates, where as many as 300 species of annelid worms, amphipods, isopods, crabs, shrimps, copepods, and other bivalves are often found in high densities that might

not persist in adjacent non-reef habitat (Wells, 1961; Zimmerman, 1989; Peterson et al., 2003; Stunz et al., 2010).

One method to directly quantify linkages between oyster reef resources and associated fishes is to determine the proportion of reef resources directly consumed by predators (DeMartini et al., 1994; Peterson et al., 2003; Simonsen, 2008). Deployment of artificial oyster reefs provides an opportunity to gain valuable insight into how fishes utilize available resources and compare the value of artificial oyster reefs relative to other natural habitats. Fishes select resources that maximize trade-offs between energetic return and survival probability (Manly et al., 2002). The processes controlling resource utilization during early life are complex. Fishes experience drastic changes in body size, morphology, physiology, and nutritional requirements, all of which may influence diet composition (Wuenschel et al., 2006) and, ultimately, how resources are utilized. Because enhancement of available food resources can have direct impacts on vital rates knowledge of habitat-specific diet composition is essential to determining the role of artificial habitat in estuarine food webs. Positive impacts upon fish diet may result from general increases in prey density, increases in preferred prey, increased diversity of prey items, or increases in capture efficiency. Documentation of habitat-specific resources and habitat-specific utilization are critical to management of exploited populations, can provide greater resolution than density comparisons, and can be used to calculate production values.

3.1.3 Research Goals

The goal of my research was to determine and evaluate the effects of MAORs (Mimic Artificial Oyster Reefs) on the potential prey community of juvenile estuarine fishes. I conducted a BACI (Before-After-Control-Impact) field experiment where meiofauna (infauna and demersal epifauna) and macrofauna (demsersal epifauna) were evaluated before and after the addition of MAORs to homogeneous mud-bottom habitat in marsh ponds. The following null hypotheses were evaluated:

H_0 1) the addition of MAORs had no effect on meiofauna community composition and density,

H_0 2) the addition of MAORs had no effect on macrofauna community composition and density, and

H_0 3) the addition of MAORs had no increase in diversity, density, or size of potential prey resources.

More specifically, I propose an alternate hypothesis that the proportional density and diversity of epibenthic and structure-associated meiofauna will increase, while infaunal and mud-associated meiofauna will decline in response to the addition of MAORs. Impacts to macrofauna are more difficult to predict but will most likely be seen in: 1) opportunistic species capable of utilizing multiple habitat types, especially structurally complex habitats, or 2) species that depend upon structure for refuge and foraging that could not exist in unstructured habitat. Potential impacts will likely result directly from changes in the physical characteristics of the habitat itself (i.e., change from mud to limestone cobble) and indirectly through secondary interactions associated with reef utilization.

3.2 Methods

3.2.1 Study Area

The study area is located near Empire, Louisiana, approximately 27 miles northwest of the Head of Passes in the Mississippi River's Balize delta, and consists of four, intertidal marsh ponds adjacent to Adams Bay (Figure 3.1). The four experimental ponds are referred to as Ovary pond (OP), Perfect pond (PP), Triangle pond (TP), and Big pond (BP; Figure 3.1). Marsh ponds were oligohaline (5-25 ppt), characterized by shallow depths (~1 m relative to mean high water), mud bottoms, and were surrounded by emergent vegetation consisting mostly of smooth cordgrass (*Spartina alterniflora*). The experimental ponds were relatively large, ranging in surface area from 6,600 m^2 – 16,800 m^2, and have only a single connection to nearby open waters.

3.2.2 Field Methods

- Sampling Design and Artificial Habitat Deployment

Four experimental ponds were sampled once every other month in a randomly selected order for the duration of two years, from March 2009 – March 2011. No sampling occurred in March 2010 as cobble material was added at the conclusion of the first sampling year. The "before" period consisted of sampling events from March 2009 – January 2010, and the "after" period consisted of sampling events from May 2010 – March 2011. A sampling event consisted of one pond being sampled each day over a successive four-day period. Sampling occurred at the same point in the monthly tidal

cycle and at the same point in each daily tidal cycle. The initial monitoring period, along with modification and the subsequent experimental period, allowed for direct

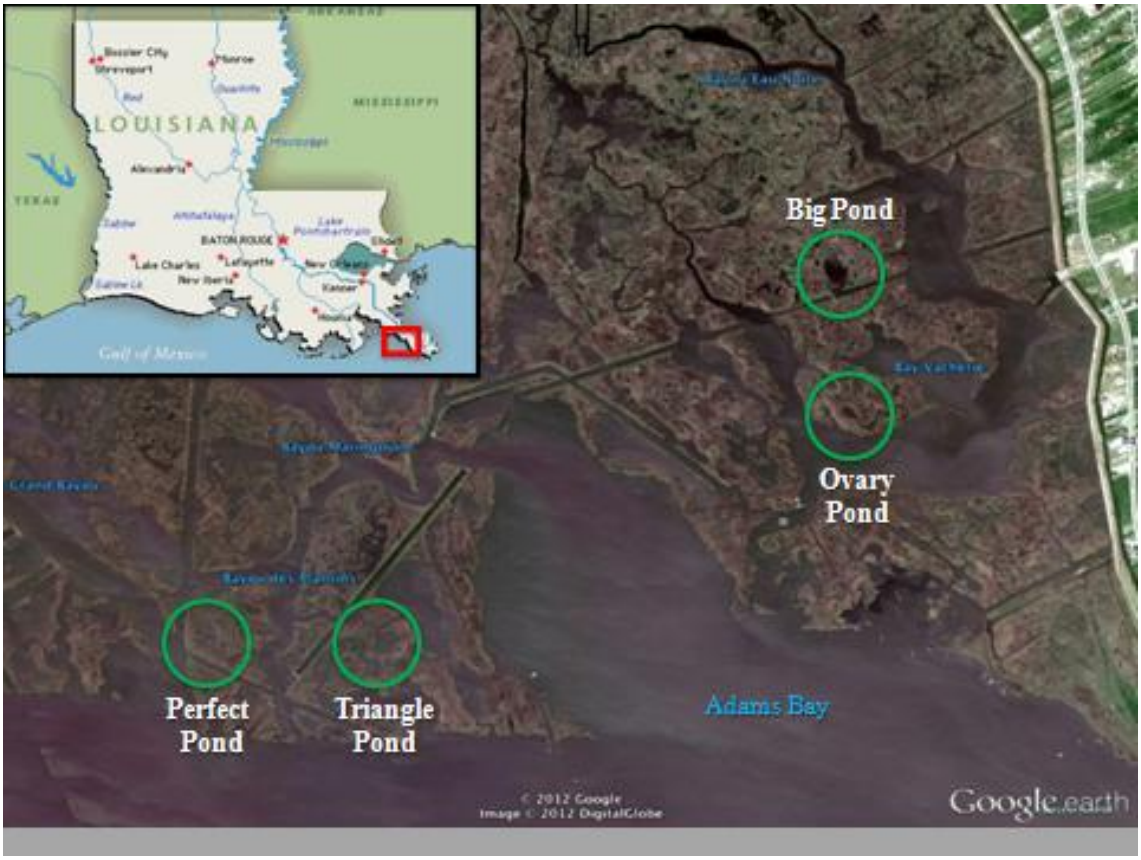

Figure 3.1. Map of the four experimental marsh ponds within marshes adjacent to Adams Bay, Empire, Louisiana.

comparisons of factors in time (before or after) and space (control or impact) as is required for BACI experimental designs. Within each of the four ponds, five fixed sites were selected for sampling for the duration of the study (Figure 3.2). During the "before" period each pond contained four mud-bottom sites and one non-vegetated, marsh edge site (Figure 3.2; sites within each pond are referred to in accordance with their orientation to the compass rose). In March 2010, (the end of the 'before' sampling period) north and

east sites in both PP and OP received #57 (3/4-1") cobble designed to mimic natural oyster reef substrate. Of the four sites that received limestone cobble treatments, all

Figure 3.2. Site map of Perfect pond containing five fixed sampling sites: 1) north – MAOR (during 'after' period only); 2) east – MAOR (during 'after' period only); 3) south – mud bottom; 4) west – mud bottom, 5) edge – unvegetated marsh edge. Perfect pond size was approximately 16,800 m^2.

consisted of mud bottom during the "before" period. Thus, in PP and OP, one of the five sites within each pond remained non-vegetated marsh edge, two of the four sites that were previously mud became MAOR, and two of the four sites remained mud bottom. No mud sites in BP or TP (control ponds) and none of the edge sites in any pond received limestone cobble treatments. The sediment surface area covered by limestone cobble at

each site after habitat deployment was approximately equal to 3% of the total surface area of that pond. Prior to MAOR deployment, mesh netting was placed across the sediment surface according to each site's dimensionality to prevent reefs from sinking into the mud bottom. Mesh netting was held in place using Polyvinyl Chloride (PVC) stakes that were removed immediately after reef deployment. Limestone cobble was evenly distributed (as possible) so that all MAORs were approximately 5 cm in height above the sediment surface. This resulted in MAOR dimensions of approximately 15 x 15 x 0.05 m and 22 x 22 x 0.05 m in OP and PP, respectively. Marking stakes were left at the four corners of each cobble plot to allow accurate sampling of MAOR habitats.

3.2.3 Environmental Variables

Temperature, water depth, salinity, and pH were measured using a YSI 6920 V2 multi-parameter hydrosonde. Readings were taken every five minutes for the duration of each daily sampling event. The hydrosonde was only deployed during the act of sampling and not continuously deployed for the entire duration of each four-day event. Sampling occurred at approximately the same point in the tidal cycle (6-8 hours before high tide) on each successive day during each four-day sampling trip. Sampling on each day began when water depths were deemed sufficient for equal habitat availability to fishes to reduce the effect of hydrologic drivers and better reflect potential habitat preferences.

3.2.4 Data Collection

- Sample Collection and Processing

Several months prior to the beginning of the first sampling trip, lift trays (plastic trays measuring 74 x 66 x 15 cm; 4,884 cm^{-2}) were lined with 1 mm^2 plastic mesh, filled with ambient sediment using a PONAR grab and placed at each site. Lift trays were gently pressed into the sediment so that the sediment surface in each lift tray was approximately level with the surrounding sediment surface. To sample each site, lift trays were carefully raised to just below the water's surface so that residual water within each lift tray would drain vertically through the sediment rather than horizontally across the sediment surface and minimize advective loss of organisms. Macrofauna were removed by hand from the entire lift tray surface. Meiofauna were then collected by horizontally scooping the top two centimeters of sediment from a randomly chosen quadrant (37 x 33 x 2 cm; 1221 cm^{-2}) of each lift tray. To determine the impact of MAOR addition, #57 limestone cobble was added to lift trays at north and east sites in PP and OP in March 2010, after completion of the "before" sampling period (March 2009 – January 2010). Lift trays at MAOR sites also contained a thin layer of mud beneath the cobble to prevent loss of organisms through the mesh lining. Meiofauna and macrofauna samples were stored in Ziploc bags, placed on ice, and frozen at the field station prior to transport to the laboratory at Louisiana State University. For mud and edge sites, samples were defrosted and excess water was removed by slowly pouring through a 250 μm sieve and then rinsed back into the sample with minimal water. Samples were then homogenized by stirring and sub-sampled by removing 10 grams of sediment (wet weight) three times from each sample bag. Samples were then stained with Rose Bengal biological stain to aid in identification. After staining, mud samples were rinsed over the 250 μm sieve to remove excess stain. Organisms were then identified to lowest taxonomic level and enumerated.

For MAOR samples, cobble was rinsed over the 250 μm sieve and removed. The remaining material (organisms and residual sediment) was processed in full without sub-sampling. Meiofauna from MAOR samples were also stained using Rose Bengal. Wet weights and standard lengths were measured for macrofauna samples, and all count data were standardized to $1m^2$.

For the purposes of this thesis, the term meiofauna is used to refer to both invertebrates between 0.5 and 0.1 mm that are typically associated with the benthos (Levinton, 1982), such as nematodes and copepods, but also refers to organisms that are commonly classified as mesozooplankton (invertebrates between 20 and 0.2 mm that are typically associated with the water column; e.g., amphipods and mysids). Mesozooplankton were included as "meiofauna" because the majority of sizes collected were within the size range of meiofauna and were also commonly associated with the benthos. A single term was also used to reduce confusion when describing results. The term macrofauna (macrobenthos) refers to animals whose shortest dimension is greater than or equal to 0.5 mm (Levinton, 1982) and can be seen with the naked eye.

3.2.5 Data and Statistical Analyses

- Environmental Variables

Environmental variables were used to compare controlling conditions during the experiment that might have affected the observed results. Environmental variables were analyzed separately as response variables using a mixed-model ANOVA in SAS. Each mixed-model test included one of the four environmental variables sampled as the response variable and included three factors as explanatory categorical variables: factor

A: Period (fixed with a = 2 levels; before or after), factor B: Month (random with b = 4 levels; May, July, September, or November; nested within factor B), factor C: Pond (random with c = 4 levels; Big, Ovary, Perfect, or Triangle) and their interaction terms. When main effects terms were significant (i.e., Period*Pond or Month*Pond), ponds were then compared using pairwise tests of Tukey-adjusted LSmeans. As environmental variables were primarily collected for the purpose of testing the assumption that hydrographic conditions in ponds were similar during any given sampling event, the sub-level factors Location and Site were included in the data as "replicates" but were not included in the statistical model.

- Meiofauna and Macrofauna

Meiofauna and macrofauna community structure were analyzed using PRIMER 6 with the PERMANOVA add-on package, which is specifically designed for analyses of community composition and density in ecological studies (Clarke and Warwick, 2001; Anderson et al., 2008, DeMutsert, 2010). All meiofauna count data were standardized to 1 m^2 log (n+1) transformed, and used to create Bray-Curtis resemblance matrices. Five factors were included in the PERMANOVA analyses: factor A: Period (fixed with a = 2 levels; before or after), factor B: Month (random with b = 4 levels; May, July, September, or November; nested within factor B), factor C: Pond (random with c = 4 levels; Big, Ovary, Perfect, or Triangle), factor D: Location (fixed with d = 3 levels; edge, control, or impact; nested within factor C), and factor E: Site (random with e = 5 levels; north, south, east, west, or edge; nested within factor D). The terms Habitat (3 levels: mud, edge, or MAOR) and Interaction (6 levels: before-control, before-edge, before-impact, after-control, after-edge, and after-impact) were included for SIMPER comparisons of

dissimilarity. Although the experimental design contained elements of a traditional BACI analysis, I wanted to include as much spatial and temporal variation into the analyses as possible. Therefore, when testing the overall effects of MAOR addition on meiofauna and macrofauna, the statistical design was essentially analyzed as a split-plot design with a time component (but is referred to as a BACI design). The simple BACI design factors representing the time and space variance components were included (i.e., Period and Location; as in traditional simple BACI designs) but additional levels (listed above) were added to the statistical model in PERMANOVA. The factor Month was also included in the temporal portion of the model (either as a repeated statement or random statement depending on the test) to better structure the temporal variation. As each pond contained the sub-level factors Location and Site (within each location) each pond represented a plot, and each sub-level represented a sub-plot. The "split" was determined by the addition of MAORs and observed in both the Period and Location factors (i.e., the factor Period was split into "before" and "after" and the factor Location was split into "control," "impact," or "edge"). PERMANOVA, which is a semi-parametric equivalent of a MANOVA, was used to test the full model but only the interaction term Period*Location (significance indicates effect of MAOR addition) was of major statistical interest as is typically evaluated in BACI experimental designs. PERMANOVA was run using 9999 permutations and tests were evaluated at a significance level of $p = 0.01$. Significance tests on each combination of the factors Period and Location were performed using ANOSIM (two-way crossed with replicates; $p = 0.1\%$), which is a non-parametric equivalent of an ANOVA. The SIMPER procedure was used to determine which taxonomic groups contributed most to dissimilarities between habitat types and between

impact sites before and after MAOR addition. Diversity indices for the major taxa of meiofauna and species of macrofauna were analyzed separately in a general linear mixed-model ANOVA. The Shannon-Weaver diversity index score for each lift tray sample was used as the response variable and the main effects terms Period, Month, Location and the interaction terms Period*Location and Period*Month*Location were included in the model as explanatory variables. The term Month was included in the repeated statement and the term site was nested in pond and included in the random statement. The univariate procedure in SAS was used to check for normality. Backward, stepwise elimination procedures in multiple regression were used to test for significant relationships between environmental variables and meiofauna and macrofauna.

Appendix 1 lists the statistical technique used for each set of analyses conducted in this experiment including response and explanatory variables, general model with effect terms for each test, and analyses techniques used for any additional treatment comparisons.

3.3. Results

3.3.1 Environmental Variables

Water temperature (°C), depth (m), salinity (ppt), pH, and dissolved oxygen (DO; $mg \cdot L^{-1}$) data were collected every other month from September 2009 – January 2010 (before artificial reef deployment) and May 2010 – March 2011 (after artificial reef deployment). Mean monthly values for each environmental variable are listed in Table 3.1. No data were collected from March 2010 because limestone cobble for artificial reefs was being deployed during much of this month. Data were only collected from Big and

Ovary ponds in March 2011 due to an equipment malfunction. In addition, only the months of September and November were included for pond comparisons between periods as these were the only two months in which hydrographic data were collected during both "before" and "after" periods (i.e., no data were available for May, July, or March in the before period, and no data were available for January in the after period).

Mean water temperature, depth, salinity, pH, and DO were all significantly different between periods ($p<0.001$), months ($p<0.0001$), and ponds ($p<0.0001$; mixed-model ANOVA). Water temperature followed seasonal trends with minimum values in winter (January) and maxima in summer (July and September; Figure 3.3). Salinity was lowest in summer (July) and peaked in the fall (November; Figure 3.3). Water depth and pH were relatively variable throughout the study period and followed no apparent seasonal trends (Figure 3.3). However, changes in pH values may have been associated with DO, especially in winter months when water levels were low and filamentous algae were abundant. The addition of limestone cobble to Ovary and Perfect ponds did not cause any consistent change in pH values during the "after" period as trends in pH between these two ponds were not similar over time (Figure 3.3).Trends in pH appeared more closely related to geographic location as ponds in close proximity to one another (Ovary and Big ponds and Perfect and Triangle ponds; Figure 3.1) displayed more similar trends over time than ponds farther apart (Figure 3.3). Mean water temperature ranged from 14.9 °C (Jan. 2010) to 30.0 °C (July 2010); mean water depth ranged from 0.55 m (March 2011) to 0.97 m (Sept. 2009); mean salinity ranged from 4.6 ppt (July 2010) to 21.6 (Nov. 2009); mean pH ranged from 7.2 (Nov. 2009) to 7.9 (Jan. 2010); and mean DO ranged from 27.9 $mg \cdot L^{-1}$ (Sept. 2009) to 128.4 $mg \cdot L^{-1}$ (July 2010; Table 3.1).

3.3.2 Meiofauna

A total of 133 samples containing meiofauna were collected during ten sampling trips: March, May, July, September, November 2009 and January 2010 (before MAOR deployment) and May, July, September and November 2010 (after MAOR deployment). Samples collected during March 2009 and January 2010 were excluded from the analyses because they were taken during the "before" period only; there was no sample from the "after" period for comparison.

Infaunal meiofauna were by far the most numerically abundant group throughout the study. Nematodes were the most abundant taxon accounting for 92% of the total meiofauna in all samples and 94%, 97% and 84% in mud, edge, and MAOR habitats, respectively (Table 3.2). During the "after" period, nematode densities increased at control and edge sites but decreased by two orders of magnitude at MAOR sites (Table 3.3). Nematode densities peaked in September and November and were lowest in July in both years (Figure 3.4).

Harpacticoids were the second most numerous taxon comprising 4% of the total meiofauna in all samples and 5%, 2% and 8% of the total meiofauna density at mud, edge, and MAOR habitats, respectively (Table 3.2). Harpacticoid densities were similar between periods at control and edge sites but decreased by an order of magnitude at MAOR sites in the "after" period. Harpacticoid densities showed a single seasonal peak in September 2009 but peaked separately at edge sites in July and control sites in November during the "after" period (Figure 3.4).

Table 3.1. Monthly mean values for water temperature (°C), depth (m), salinity (ppt), pH, and dissolved oxygen ($mg \cdot L^{-1}$) for Big, Ovary, Perfect, and Triangle ponds before and after MAOR addition.

Environmental Variable	Period	Month	Big	Ovary	Perfect	Triangle
Water Temp (°C)	Before	September	30.45	29.29	30.96	29.38
		November	19.40	21.67	16.80	16.23
		January	14.84	21.67	15.94	16.07
	After	May	27.44	26.48	25.49	26.24
		July	31.17	30.11	30.25	32.28
		September	31.24	28.91	28.53	29.77
		November	19.97	22.10	22.56	23.13
		March	20.91	24.17	.	.
Depth (m)	Before	September	1.15	0.84	0.89	0.95
		November	0.91	0.86	0.85	0.81
		January	0.81	0.52	0.75	0.67
	After	May	0.67	0.73	0.73	0.79
		July	0.95	0.69	0.76	0.69
		September	0.76	0.70	0.89	0.76
		November	0.87	0.72	0.90	0.86
		March	0.55	.	.	.
Salinity (ppt)	Before	September	11.89	11.28	11.57	11.79
		November	23.45	21.68	20.79	20.22
		January	15.13	14.67	14.50	14.50
	After	May	8.82	7.44	6.96	7.15
		July	4.54	4.53	4.75	5.08
		September	9.90	10.14	10.62	10.43
		November	18.87	19.88	20.07	19.87
		March	18.50	17.75	.	.
pH	Before	September	7.23	7.35	7.90	7.57
		November	7.03	7.10	7.45	7.50
		January	7.72	8.00	8.12	8.08
	After	May	7.60	7.60	7.73	7.61
		July	7.81	7.56	8.24	8.39
		September	7.16	7.47	7.98	7.54
		November	7.63	7.66	7.73	7.70
		March	7.38	7.66	.	.
Dissolved Oxygen ($mg \cdot L^{-1}$)	Before	September	27.89	35.47	89.03	51.25
		November	74.58	71.52	95.62	89.52
		January	91.19	111.34	96.17	97.12
	After	May	81.11	73.07	75.86	81.44
		July	85.98	64.15	97.42	128.43
		September	43.46	46.29	78.91	57.78
		November	85.20	75.43	84.61	82.91
		March	75.73	111.28	.	.

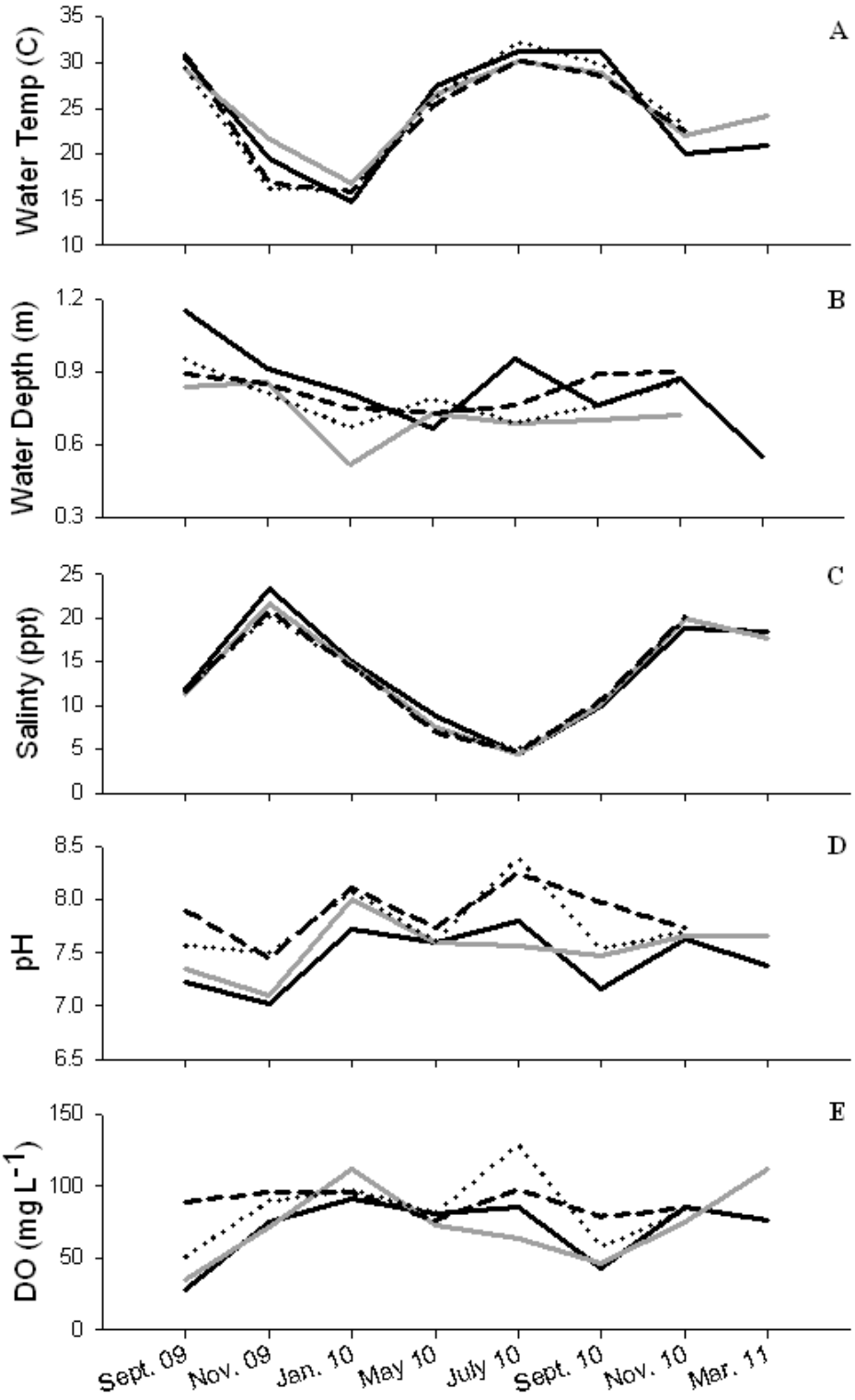

Figure 3.3. Water temp (°C; A), depth (m; B), salinity (ppt; C), pH (D) and dissolved oxygen ($mg \cdot L^{-1}$; E) profiles for each pond in each month; Big (black line), Ovary (gray line), Perfect (dotted line), and Triangle (dashed line).

The number of taxa found at MAOR sites showed marked increases from six taxa in the "before" period to 13 taxa groups in the "after" period. Nematodes, harpacticoids, calanoids, cyclopoids, amphipods, and isopods were present in both periods at MAOR sites. Tanaids, gastropods, polychaetes, mussels, clams, insect larvae, and ostracods were present at MAOR sites in the "after" period only with tanaids being the most abundant and frequent immigrant taxon.

Shannon-Weaver diversity indices (Figure 3.5) were lowest at edge sites (H′=0.67) but relatively similar between habitat types during the "before" period. During

Table 3.2. Mean density of individuals m^{-2} (± S.E) of meiofauna collected at each habitat type.

Taxonomic Group	Density (individuals m^{-2})		
	Mud	Edge	MAOR
Nematodes	88796.9 (197879.9)	96960.9 (121123.9)	4975.4 (5249.1)
Calanoids	313.4 (751.1)	94.2 (159.3)	15.6 (23.1)
Harpacticoids	4667.7 (7025.2)	2461.9 (3084.7)	496.1 (713.1)
Cyclopoids	47.1 (182.0)	9.4 (47.1)	3.0 (9.9)
Amphipods	168.5 (319.6)	164.8 (286.3)	21.4 (40.6)
Isopods	29.7 (109.9)	61.2 (242.9)	2.3 (3.7)
Gastropods	5.0 (38.1)	4.7 (23.5)	92.2 (238.9)
Polychaetes	61.9 (426.7)	131.8 (586.8)	14.6 (24.7)
Tanaids	47.1 (171.1)	315.4 (1297.5)	248.2 (252.5)
Mussels	0.0 (0.0)	0.0 (0.0)	11.4 (30.7)
Clams	0.0 (0.0)	0.0 (0.0)	1.7 (3.9)
Insect Larvae	1.2 (12.1)	0.0 (0.0)	1.9 (6.2)
Ostracods	0.0 (0.0)	0.0 (0.0)	34.7 (82.1)
Total	94138.5 (199916.4)	100204.2 (121490.2)	5918.4 (5642.6)

Table 3.3. Monthly mean densities of individuals m^{-2} (± S.E) of meiofauna collected at MAOR sites before (B) and after (A) deployment.

Taxonomic Group	Density (individuals m^{-2})							
	MAOR (Before)				MAOR (After)			
	May	July	September	November	May	July	September	November
Nematodes	15573.3 (6490.0)	6197.9 (709.4)	34510.4 (25593.2)	30156.1 (15607.1)	5317.0 (3678.2)	3290.1 (475.4)	2831.4 (2926.8)	7706.0 (8629.7)
Calanoids	0.0 (0.0)	0.0 (0.0)	205.9 (278.1)	29.4 (58.8)	5.0 (8.7)	19.7 (1.3)	37.1 (31.8)	0.0 (0.0)
Harpacticoids	5178.0 (1838.3)	4001.2 (1529.9)	7502.3 (3088.8)	941.5 (658.7)	85.8 (135.6)	208.4 (183.2)	131.9 (116.0)	1311.7 (842.4)
Cyclopoids	39.2 (67.9)	39.2 (67.9)	29.4 (58.8)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	9.9 (17.3)
Amphipods	235.4 (311.4)	117.7 (203.8)	117.7 (166.4)	117.7 (135.9)	50.1 (62.3)	0.0 (0.0)	0.0 (0.0)	31.9 (45.9)
Isopods	0.0 (0.0)	0.0 (0.0)	58.8 (117.7)	0.0 (0.0)	1.9 (3.3)	0.0 (0.0)	5.6 (4.8)	0.5 (0.9)
Gastropods	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	25.8 (51.6)	273.7 (402.0)
Polychaetes	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	47.4 (19.2)
Tanaids	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	546.5 (147.1)	378.4 (277.5)	1.4 (2.8)	206.1 (176.4)
Mussels	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	6.1 (12.2)	31.0 (53.4)
Clams	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	5.6 (5.5)
Insect Larvae	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	6.1 (11.0)
Ostracods	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	112.7 (123.5)
Total	21025.9 (8363.2)	10356.1 (963.3)	42424.5 (28290.2)	31244.7 (15319.4)	6006.2 (3728.5)	3896.6 (382.4)	3039.4 (2959.9)	9742.6 (8717.6)

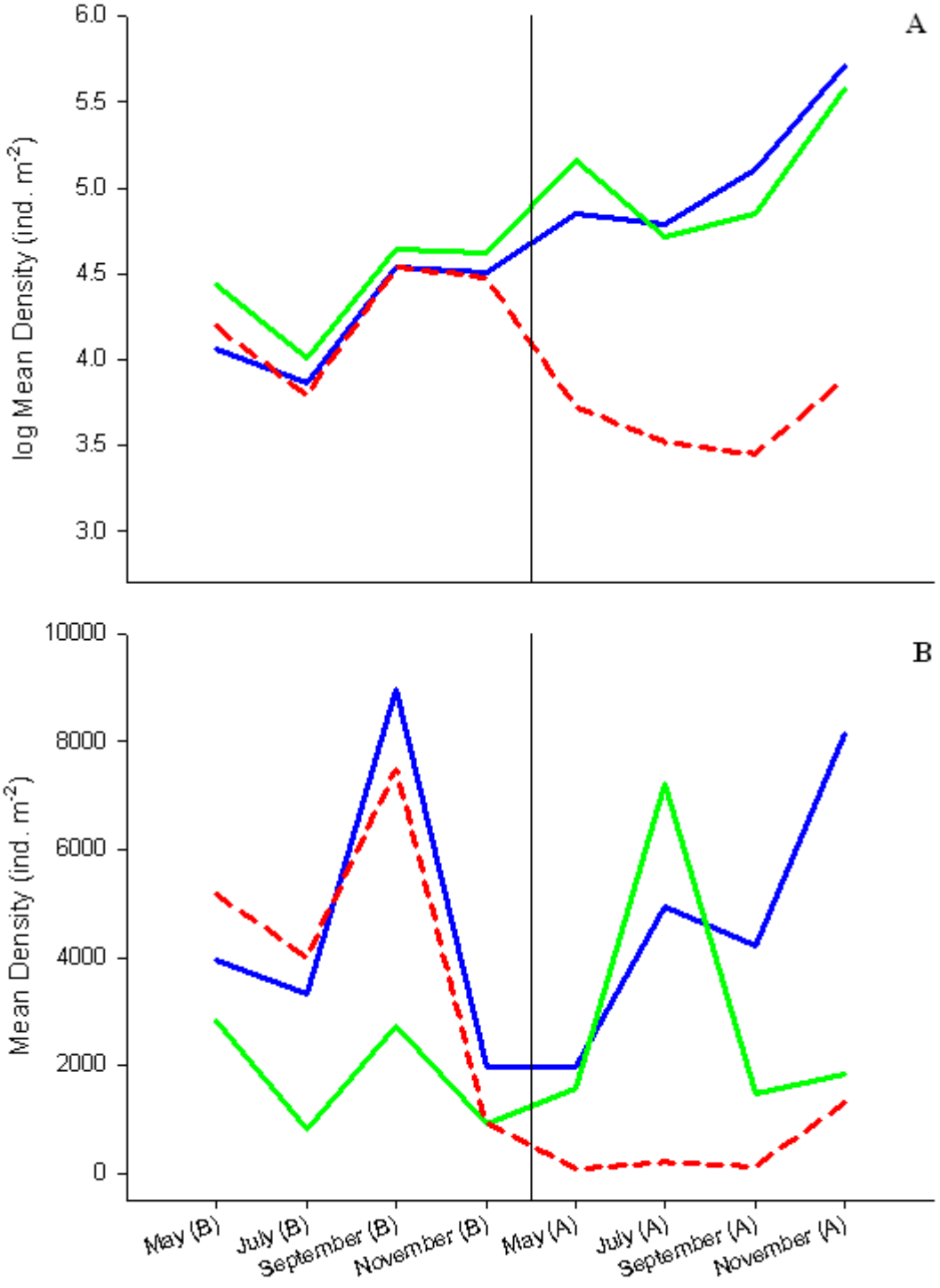

Figure 3.4. Monthly log(n+1) transformed, mean densities of nematodes (A) and harpacticoid copepods (B) at control (blue), edge (green) and MAOR (red) sites. Vertical black line indicates addition of cobble during March 2010.

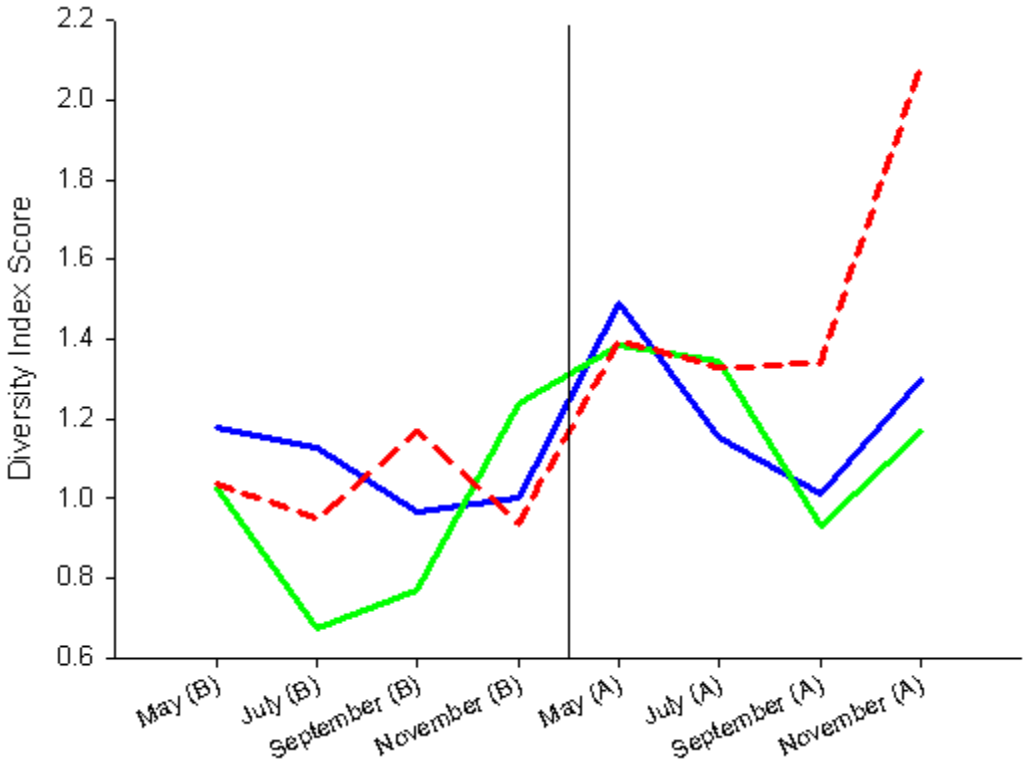

Figure 3.5. Mean Shannon-Weaver diversity index scores of log(n+1) transformed meiofauna densities (individuals m^{-2}) by month, before (B) and after (A) MAOR deployment, from control (blue), edge (green), and MAOR (red) locations. Solid vertical line indicates addition of cobble during March 2010.

the "after" period diversity scores at MAOR sites (H′=2.09) were considerably higher than control and edge sites (H′≈1.3), especially in November. Tests of Shannon-Weaver diversity indices indicated the interaction terms Period*Location and Period*Month*Location were significant (p=0.02 and 0.002, respectively). Tukey-adjusted pairwise comparisons (using LSmeans) of diversity values between locations (control, impact, and edge) indicated that diversity values for MAOR sites in the "after" period were significantly different from MAOR sites in the "before" period ($p<0.0001$) and significantly different from control and edge sites in both periods ($p<0.05$).

Table 3.4. PERMANOVA output of main effects terms and their interactions for log(n+1) transformed meiofauna densities from lift tray samples ($\alpha = 0.01$).

Factor	df	SS	MS	Pseudo-F	p-value
Period	1	4803.8	4803.8	3.96	0.0040
Pond	3	2296.7	765.6	1.92	0.0238
Month(Period)	6	6723.2	1120.5	3.23	0.0004
Location(Pond)	6	5483.2	913.9	2.30	0.0002
Period*Pond	3	1033.6	344.5	1.21	0.2671
Site(Location(Pond))	10	2341.2	234.1	0.83	0.6934
Period*Location(Pond)	6	4543.0	757.2	2.20	0.0009
Month(Period)*Pond	18	6372.4	354.0	1.26	0.1608
Period*Site(Location(Pond))	10	1860.7	186.1	0.66	0.8622
Month(Period)*Location(Pond)	29	8722.6	300.8	1.07	0.3855
Month(Period)xSite(Location(Pond))	37	10482.0	283.3	2.10	0.2113
Total	132	68800.0			

Table 3.5. SIMPER output of the taxonomic groups that explain > 95 % of the dissimilarity in meiofaunal densities (individuals m^{-2}; square root transformed) at MAOR sites before and after deployment.

Taxonomic Group	Density (individuals m^{-2})		% contribution to dissimilarity	% cumulative contribution
	Impact (Before)	Impact (After)		
Nematode	141.47	62.88	47.58	47.58
Harpacticoid	61.36	17.08	27.86	75.44
Tanaid	0.00	12.58	7.33	82.77
Amphipod	8.16	2.76	4.85	87.61
Calanoid	3.60	2.70	2.92	90.53
Gastropod	0.00	4.66	2.61	93.15
Cyclopoid	2.33	0.61	1.72	94.87
Ostracod	0.00	2.89	1.48	96.36
Average dissimilarity = 52.64				

Statistical comparisons of the interactions between the main effects terms Period, Month, Pond, Location, and Site indicated significant differences between locations in the "before" and "after" periods (i.e., significant Period*Location term; Table 3.4). Pairwise comparisons of the levels of the term Interaction indicated the "after-impact" level was significantly different than the levels "before-control," "before-edge," "before-impact," "after-control," and "after-edge" (p= 0.1%; one-way ANOSIM).

SIMPER analysis of MAOR locations before and after MAOR deployment indicated nematodes, harpacticoids, tanaids, calanoids, gastropods, cyclopoids, and ostracods comprised more than 95% of the cumulative dissimilarity between periods (Table 3.5). Tanaid, gastropod, and ostracod densities increased while nematode, harpacticoid, calanoid, and cyclopoid densities decreased at MAOR sites in the "after" period. Comparisons of SIMPER output between the three habitat types (mud, edge, and MAOR) showed similar results to those of the MAOR sites alone. Nematodes, harpacticoids, tanaids, calanoids, amphipods, and gastropods still contributed the most to the 95% cumulative dissimilarity between MAOR and non-MAOR habitats but polychaete densities contributed to dissimilarity between edge and MAOR habitats while ostracods did not contribute to dissimilarity between mud and MAOR habitats (Table 3.6). Only nematodes, harpacticoids, amphipods, calanoids, and tanaids contributed to the 95% cumulative dissimilarity between the two natural habitats (i.e., edge and mud). Cumulative dissimilarity percentages were extended to the 95^{th} percentile due to the numerical dominance of nematodes and harpacticoids in all samples.

Of the thirteen taxonomic groups identified in lift tray samples, five groups contributed to cumulative percent dissimilarity in all comparisons: nematodes,

harpacticoids, amphipods, calanoids, and tanaids. Of these five taxonomic groups, significant portions of the variation in densities were related to environmental factors, macrofauna densities, or other meiofauna (backward stepwise elimination in regression). Approximately half the variation in nematode densities was significantly related to densities of harpacticoid and cyclopoid copepods ($p<0.001$; adjusted-$R^2 = 0.43$). Approximately half the variation in harpacticoid densities was significantly related to densities of nematodes, cyclopoids, tanaids, and water temperature ($p<0.0001$; adjusted-$R^2 = 0.43$). Variation in amphipod densities was significantly related to densities of brown shrimp (*Farfantepenaeus aztecus*), blue crabs (*Callinectes sapidus*), gobies, xanthid crabs and water temperature ($p<0.0001$; adjusted-$R^2 = 0.26$). Calanoid densities were significantly related to densities of harpacticoids, gobies and water depth ($p<0.001$; adjusted-$R^2 = 0.37$). Finally, tanaid densities were significantly related to densities of calanoid copepods, gray snapper (*Lutjanus griseus*), grass shrimp (*Palaemonetes pugio.*), xanthid crabs, and water depth ($p<0.0001$; adjusted-$R^2 = 0.39$).

3.3.3 Macrofauna

A total of 124 macrofauna samples were collected during the months of May, July, September, and November in both the "before" and "after" period. A total of 26 separate taxa were identified, most to the species level (Table 3.7). Grass shrimp (69.4%), blue crabs (8.0%), estuarine mud crabs (*Rithropanopeus harrisii* - 6.8%), naked gobies (*Gobiosoma bosc* - 4.8%), white shrimp (*Litopanaeus setiferus* - 3.6%), and brown shrimp (3.6%) were the six most abundant species, comprising 96.2% of the total organisms collected by number. Blue crabs (38.0%), Xanthid crabs (estuarine mud crabs

Table 3.6. SIMPER output of the taxonomic groups that explain >95% of the dissimilarity in meiofaunal densities (individuals m^{-2}; square root transformed) at each habitat type (mud, edge, and MAOR).

Taxonomic Group	Density (individuals m^{-2})		% contribution to dissimilarity	% cumulative contribution
	Mud	MAOR		
Nematode	223.83	62.88	56.97	56.97
Harpacticoid	60.29	17.08	20.53	77.50
Tanaid	2.29	12.58	5.90	83.40
Calanoid	9.86	2.70	4.16	87.56
Amphipod	7.89	2.76	4.13	91.69
Gastropod	0.31	4.66	2.15	93.84
Cyclopoid	2.38	0.61	1.56	95.41
Average dissimilarity = 57.58				
	Edge	MAOR		
Nematode	269.79	62.88	69.33	69.33
Harpacticoid	43.71	17.08	12.35	81.69
Tanaid	5.67	12.58	5.71	87.40
Amphipod	7.24	2.76	3.32	90.72
Calanoid	5.57	2.70	2.34	93.06
Gastropod	0.43	4.66	1.86	94.92
Polychaete	3.47	2.09	1.44	96.36
Average dissimilarity = 60.99				
	Mud	Edge		
Nematode	223.83	269.79	68.49	68.49
Harpacticoid	60.29	43.71	14.26	82.76
Amphipod	7.89	7.24	5.09	87.85
Calanoid	9.86	5.57	4.63	92.48
Tanaid	2.29	5.67	2.81	95.29
Average dissimilarity = 37.27				

and *Eurypanopeus depressus* - 12.5%), pinfish (*Lagodon rhomboides* - 10.9%), brown shrimp (7.6%), sheepshead (*Archosargus probatocephalus* - 7.1%), gulf toadfish (*Opsanus beta* - 6.9%), white shrimp (5.1%), and gray snapper (*Lutjanus griseus*- 2.9%) comprised 91% of the total weight of species collected.

The number of species observed at MAOR sites decreased from 21 to eight species between the periods "before" and "after" (Table 3.8). Grass shrimp, blue crabs, white shrimp, estuarine mud crabs, naked gobies, pinfish, gulf toadfish, and sheepshead were collected from MAOR sites after MAOR deployment. Of the eight species present at MAOR sites during the "after" period only naked gobies, gulf toadfish and sheepshead increased in mean density. Mean weights of blue crabs, estuarine mud crabs, naked gobies, gulf toadfish, pinfish, and sheepshead increased at MAOR sites during the "after" period.

Densities of the three most abundant species at MAOR sites during the "before" period (i.e., grass shrimp, blue crabs, and white shrimp) all decreased in density at MAOR sites after MAORs were deployed. In between-habitat comparisons, total density was highest at mud sites, but total length and weight were highest at MAOR sites. Densities of naked gobies, sheepshead, and gulf toadfish were higher at MAORs than other habitats. Lengths and weights of blue crabs, naked gobies, pinfish, sheepshead, and gulf toadfish were also higher at MAORs than other habitats (Table 3.9).

Shannon-Weaver diversity indices (Figure 3.6) were lowest at edge sites in all months but November during the "before" period. Control and MAOR locations were relatively consistent during the "before" period with diversity values decreasing as the season progressed (i.e., May through November). Diversity scores during the "after"

period trended towards a common value in September but diverged again in November with control locations (mud) showing higher mean values than edge and MAOR sites during all months. Diversity trends at edge and MAOR sites were relatively similar over time during the "after" period with minimum values in May (H´= 0.5 and 0 respectively) and peaking in September (H´= 0.99 and 1.07 respectively). Control locations were similar over time during the "after" period with the lowest values in September (H´= 1.12) and peaking in May (H´= 1.33). Significance tests of Shannon-Weaver diversity indices indicated significant interaction terms: Period*Location (p=0.03) and Period*Month*Location (p=0.04; MIXED). Tukey-adjusted pairwise comparisons (using LSmeans) of diversity indices between locations indicated diversity values for MAOR locations in the "after" period were not significantly different from MAOR locations in the "before" period ($p>0.05$) and were only significantly different from control locations in the "before" period ($p=0.03$).

Statistical comparisons of macrofauna densities indicated no significant differences between locations or sites in the "before" and "after" periods (non-significant Period*Location and Period*Site terms; Table 3.10). Macrofauna did exhibit significant seasonal trends in density as both the Month*Pond and Month*Site interaction terms were significant ($p<0.01$). Pairwise comparisons of densities for the term Interaction indicated "after-impact" sites were significantly different from both "before-impact" and "before-control" sites (p = 0.01%; one-way ANOSIM). However, ANOSIM also indicated that control locations were significantly different between periods (p = 0.01%). Similar to densities, statistical comparisons of macrofauna lengths and weights also showed significant seasonal trends (i.e., significant interaction term

Table 3.7. List of 26 macrofauna species collected in combined lift tray samples, ranked by numerical abundance (counts), from marsh ponds in Adams Bay, Louisiana.

Species	Common Name	Abundance		Occurrence	
		Number	Portion	Frequency	Portion
Palaemonetes pugio	Grass shrimp	2588	0.694	83	0.182
Callinectes sapidus	Blue crab	300	0.080	99	0.217
Rhithropanopeus harrisii	Estuarine mud crab	254	0.068	80	0.175
Gobiosoma bosc	Naked goby	180	0.048	43	0.094
Litopenaeus setiferus	White shrimp	136	0.036	28	0.061
Farfantepenaeus aztecus	Brown shrimp	135	0.036	46	0.101
Gobiesox strumosus	Skilletfish	26	0.007	11	0.024
Anchoa mitchilli	Bay anchovy	15	0.004	8	0.018
Gobionellus boleosoma	Darter goby	15	0.004	11	0.024
Eurypanopeus depressus	Flatback mud crab	15	0.004	5	0.011
Bairdiella chrysoura	Silver perch	13	0.003	2	0.004
Lagodon rhomboides	Pinfish	12	0.003	11	0.024
Chaetodipterus faber	Atlantic spadefish	11	0.003	5	0.011
Panopeus obesus	Saltmarsh mud crab	7	0.002	1	0.002
Lutjanus griseus	Mangrove snapper	5	0.001	5	0.011
Archosargus probatocephalus	Sheepshead	4	0.001	4	0.009
Eleotris pisonis	Spinycheek sleeper	3	0.001	3	0.007
Alpheidae sp.	Pistol shrimp	2	0.001	1	0.002
Opsanus beta	Gulf toadfish	2	0.001	2	0.004
Gobionellus oceanicus	Highfin goby	1	<0.001	1	0.002
Hypleurochilus geminatus	Crested blenny	1	<0.001	1	0.002
Myrophis punctatus	Speckled worm eel	1	<0.001	1	0.002
Panopeus simpsoni	Oystershell mud crab	1	<0.001	1	0.002
Erotelis smaragdus	Emerald sleeper	1	<0.001	1	0.002
Gobiosoma robustum	Code goby	1	<0.001	1	0.002
Brevoortia patronus	Gulf Menhaden	1	<0.001	1	0.002
Mugil cephalus	Striped mullet	1	<0.001	1	0.002
Total		3731			

Table 3.8. Mean density of individuals per m^2 (± S.E.), lengths (mm ± S.E.) and weights (g ± S.E.) of 26 macrofauna species collected at MAOR sites before (B) and after (A) deployment.

Species	Density (m^{-2})		Length (mm)		Weight (g)	
	B	A	B	A	B	A
Crustaceans (9 species)						
Grass shrimp, *P. pugio*	69.8 (66.9)	20.6 (53.2)	11.2 (3.4)	3.8 (6.2)	0.1 (0.1)	0.1 (0.1)
Blue crab, *C. sapidus*	4.4 (3.6)	1.9 (2.4)	26.8 (19.3)	28.9 (39.4)	2.1 (3.0)	4.6 (9.0)
White shrimp, *L. setiferus*	3.3 (5.1)	0.3 (0.9)	8.6 (13.9)	4.2 (13.4)	0.5 (1.1)	0.3 (1.0)
Brown shrimp, *F. aztecus*	1.1 (1.5)	0.0 (0.0)	9.3 (13.5)	0.0 (0.0)	0.6 (1.2)	0.0 (0.0)
Estuarine mud crab, *R. harrisii*	0.8 (1.2)	0.8 (1.0)	5.9 (7.6)	8.5 (11.3)	0.4 (0.6)	0.7 (1.5)
Oystershell mud crab, *P. simpsoni*	0.1 (0.5)	0.0 (0.0)	3.5 (14.0)	0.0 (0.0)	1.7 (6.9)	0.0 (0.0)
Total Crustaceans	79.5 (71.3)	23.6 (52.8)	65.3 (31.5)	45.4 (34.6)	5.4 (7.5)	5.7 (8.6)
Fishes (18 species)						
Darter goby, *G. boleosoma*	0.8 (1.2)	0.0 (0.0)	6.9 (12.5)	0.0 (0.0)	0.1 (0.1)	0.0 (0.0)
Atlantic spadefish, *C. faber*	0.3 (0.6)	0.0 (0.0)	4.4 (12.2)	0.0 (0.0)	0.2 (0.4)	0.0 (0.0)
Naked goby, *G. bosc*	0.1 (0.5)	6.5 (6.5)	0.0 (0.0)	24.2 (17.2)	0.0 (0.0)	0.3 (0.3)
Pinfish, *L. rhomboides*	0.1 (0.5)	0.1 (0.5)	0.0 (0.0)	14.7 (46.4)	0.0 (0.0)	3.8 (12.1)
Skilletfish, *G. strumosus*	0.1 (0.5)	0.0 (0.0)	2.3 (9.1)	0.0 (0.0)	0.1 (0.1)	0.0 (0.0)
Speckled worm eel, *M. punctatus*	0.1 (0.5)	0.0 (0.0)	21.3 (61.0)	0.0 (0.0)	0.2 (0.5)	0.0 (0.0)
Spinycheek sleeper, *E. pisonis*	0.1 (0.5)	0.0 (0.0)	4.8 (19.3)	0.0 (0.0)	0.3 (1.2)	0.0 (0.0)
Silver perch, *B. chrysoura*	1.4 (4.6)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
Gulf toadfish, *O. beta*	0.0 (0.0)	0.1 (0.5)	0.0 (0.0)	14.4 (45.4)	0.0 (0.0)	2.7 (8.5)
Sheepshead, *A. probatocephalus*	0.0 (0.0)	0.3 (0.6)	0.0 (0.0)	27.5 (59.4)	0.0 (0.0)	9.4 (23.7)
Total Fishes	3.0 (5.7)	7.0 (6.8)	39.7 (61.9)	80.8 (82.2)	0.7 (1.5)	16.2 (24.9)
Total	82.7 (71.8)	30.6 (56.1)	105.0 (74.9)	126.1 (87.9	6.3 (7.5)	21.9 (26.4)

Month*Location; p=0.0001). Pairwise comparisons of lengths and weights for the term Interaction indicated MAOR sites in the "after" period were significantly different from control and edge sites in the "before" period ($p < 0.1\%$). During the "after" period, macrofauna lengths at MAOR sites were not significantly different from control or edge sites ($p>0.1\%$) but macrofauna weights were significantly different from control sites ($p=0.01\%$). SIMPER analysis indicated an average dissimilarity value of 86.5% at impact sites between "before" and "after" periods with grass shrimp, naked gobies, and blue crabs comprising 80% of the cumulative dissimilarity in macrofauna densities (Table 3.11). In addition to those three species sheepshead, speckled worm eel, white shrimp, estuarine mud crab, gulf toadfish and pinfish contributed to 80% of the cumulative dissimilarity in lengths and weights at MAOR sites between periods (Table 3.11). Comparisons of SIMPER output between habitat types (i.e., mud, edge, and MAOR) showed similar results with blue crabs, naked gobies, sheepshead, pinfish, brown shrimp, estuarine mud crabs, white shrimp, gulf toadfish and macrofauna contributing to 80% of cumulative dissimilarity in macrofauna densities, lengths, and weights (Tables 3.12-3.14).

Of the 26 species of macrofauna identified in lift tray samples, six groups consistently contributed to the cumulative dissimilarity in all comparisons: grass shrimp, naked gobies, blue crabs, xanthid crabs, white shrimp, and brown shrimp. For white shrimp, brown shrimp, and xanthid crabs a significant portion of the variation in density could be explained by densities of other macrofauna but the proportion of the variation explained (adjusted-r^2) was usually low (backward stepwise elimination in

regression). Environmental variables were not significantly related to any of these six species of macrofauna. Both white and brown shrimp densities showed significant relationships to grass shrimp and the combined group "all shrimps" ($p<0.0001$; adjusted-$r^2 = 0.19$ and 0.25 respectively). Densities of xanthid crabs were significantly related to densities of amphipods and gastropods ($p<.0001$; adjusted-$r^2 = 0.19$). Densities of grass shrimp, naked gobies, and blue crabs were not significantly related to any macrofauna or environmental explanatory variables.

Table 3.9. Mean density of individuals per m^2 (± S.E.), lengths (mm ± S.E.) and weights (g ± S.E.) of all 26 macrofauna species collected at each habitat type.

Species	Density (m^{-2})			Length (mm)			Weight (g)		
	Mud	Edge	MAOR	Mud	Edge	MAOR	Mud	Edge	MAOR
Crustaceans (9 species)									
Grass shrimp, *P. pugio*	39.7 (54.7)	7.2 (15.0)	20.6 (53.2)	9.3 (6.5)	6.9 (6.7)	3.8 (6.2)	0.1 (0.1)	0.1 (0.1)	0.0 (0.1)
Blue crab, *C. sapidus*	4.0 (3.0)	3.1 (3.6)	1.9 (2.4)	28.0 (22.2)	18.6 (16.9)	28.9 (39.4)	4.2 (14.0)	1.4 (3.6)	4.6 (9.0)
Estuarine mud crab, *R. harrisii*	3.3 (5.1)	3.1 (3.5)	0.8 (1.0)	10.8 (8.8)	8.2 (7.8)	8.5 (11.3)	0.7 (1.1)	0.4 (0.5)	0.7 (1.5)
Brown shrimp, *F. aztecus*	2.2 (4.3)	0.3 (0.8)	0.0 (0.0)	12.9 (15.5)	5.6 (10.8)	0.0 (0.0)	0.9 (1.6)	0.3 (0.7)	0.0 (0.0)
White shrimp, *L. setiferus*	2.0 (5.5)	1.1 (3.6)	0.3 (0.9)	6.9 (14.0)	4.2 (10.1)	4.2 (13.4)	0.6 (2.4)	0.2 (0.5)	0.3 (1.0)
Flatback mud crab, *E. depressus*	0.2 (1.4)	0.1 (0.3)	0.0 (0.0)	1.3 (7.5)	1.1 (4.2)	0.0 (0.0)	0.5 (4.4)	0.0 (0.2)	0.0 (0.0)
Saltmarsh mud crab, *P. obesus*	0.1 (1.1)	0.0 (0.0)	0.0 (0.0)	0.2 (1.6)	0.0 (0.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)	0.0 (0.0)
Pistol shrimp, *Alpheidae* sp.	0.0 (0.3)	0.0 (0.0)	0.0 (0.0)	0.1 (1.4)	0.0 (0.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)	0.0 (0.0)
Oystershell mud crab, *P. simpsoni*	0.0 (0.2)	0.0 (0.0)	0.0 (0.0)	0.6 (5.6)	0.0 (0.0)	0.0 (0.0)	0.3 (2.8)	0.0 (0.0)	0.0 (0.0)
Total Crustaceans	51.7 (56.9)	14.9 (16.0)	23.6 (52.8)	70.0 (30.1)	44.6 (25.6)	45.4 (34.6)	7.3 (14.7)	2.4 (3.7)	5.6 (8.6)
Fishes (18 species)									
Naked goby, *G. bosc*	2.1 (4.0)	0.8 (1.8)	6.5 (6.5)	10.1 (17.7)	6.1 (12.5)	24.2 (17.2)	0.2 (0.4)	0.1 (0.1)	0.3 (0.3)
Skilletfish, *G. strumosus*	0.4 (1.6)	0.1 (0.3)	0.0 (0.0)	3.4 (12.5)	0.0 (0.0)	0.0 (0.0)	0.1 (0.6)	0.0 (0.0)	0.0 (0.0)
Silver perch, *B. chrysoura*	0.4 (2.1)	0.0 (0.0)	0.0 (0.0)	0.3 (3.4)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
Darter goby, *G. boleosoma*	0.3 (0.7)	0.0 (0.0)	0.0 (0.0)	3.1 (9.2)	0.0 (0.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)	0.0 (0.0)
Bay anchovy, *A. mitchilli*	0.2 (0.9)	0.1 (0.3)	0.0 (0.0)	2.2 (8.8)	1.1 (6.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)	0.0 (0.0)
Atlantic spadefish, *C. faber*	0.2 (0.9)	0.0 (0.0)	0.0 (0.0)	1.3 (7.8)	0.0 (0.0)	0.0 (0.0)	0.1 (0.6)	0.0 (0.0)	0.0 (0.0)
Pinfish, *L. rhomboides*	0.2 (0.5)	0.1 (0.3)	0.1 (0.5)	5.9 (24.3)	3.8 (20.3)	14.7 (46.4)	0.9 (5.4)	0.6 (3.0)	3.8 (12.1)
Gray snapper, *L. griseus*	0.1 (0.3)	0.1 (0.3)	0.0 (0.0)	3.2 (16.1)	3.1 (16.3)	0.0 (0.0)	0.3 (1.6)	0.3 (1.5)	0.0 (0.0)
Sheepshead, *A. probatocephalus*	0.0 (0.2)	0.0 (0.0)	0.3 (0.6)	0.3 (2.6)	0.0 (0.0)	27.5 (59.4)	0.0 (0.0)	0.0 (0.0)	9.4 (23.7)
Spinycheek sleeper, *E. pisonis*	0.0 (0.2)	0.1 (0.3)	0.0 (0.0)	1.3 (9.3)	1.9 (10.3)	0.0 (0.0)	0.1 (0.5)	0.1 (0.3)	0.0 (0.0)
Crested blenny, *H. geminatus*	0.0 (0.2)	0.0 (0.0)	0.0 (0.0)	1.8 (12.6)	0.0 (0.0)	0.0 (0.0)	0.2 (1.3)	0.0 (0.0)	0.0 (0.0)
Emerald sleeper, *E. smaragdus*	0.0 (0.2)	0.0 (0.0)	0.0 (0.0)	0.6 (5.6)	0.0 (0.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)	0.0 (0.0)
Gulf toadfish, *O. beta*	0.0 (0.2)	0.0 (0.0)	0.1 (0.5)	1.8 (18.1)	0.0 (0.0)	14.4 (45.4)	0.6 (6.5)	0.0 (0.0)	2.7 (8.5)
Speckled worm eel, *M. punctatus*	0.0 (0.2)	0.0 (0.0)	0.0 (0.0)	3.4 (25.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.2)	0.0 (0.0)	0.0 (0.0)
Highfin goby, *G. oceanicus*	0.0 (0.0)	0.1 (0.3)	0.0 (0.0)	0.0 (0.0)	1.3 (7.1)	0.0 (0.0)	0.0 (0.0)	0.0 (0.1)	0.0 (0.0)
Gulf menhaden, *B. patronus*	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.7 (7.3)	0.0 (0.0)	0.0 (0.0)	0.0 (0.4)	0.0 (0.0)	0.0 (0.0)
Striped mullet, *M. cephalus*	0.0 (0.0)	0.1 (0.3)	0.0 (0.0)	0.0 (0.0)	5.5 (28.9)	0.0 (0.0)	0.0 (0.0)	1.1 (5.4)	0.0 (0.0)
Total Fishes	3.9 (5.7)	1.2 (1.8)	7.0 (6.8)	39.4 (53.3)	22.9 (40.0)	80.7 (82.2)	2.5 (8.8)	2.0 (6.2)	16.2 (24.9)
Total	55.6 (59.3)	16.1 (16.5)	30.6 (56.1)	109.5 (65.3)	67.5 (54.1)	126.1 (87.9)	9.8 (20.4)	4.4 (7.2)	21.8 (26.4)

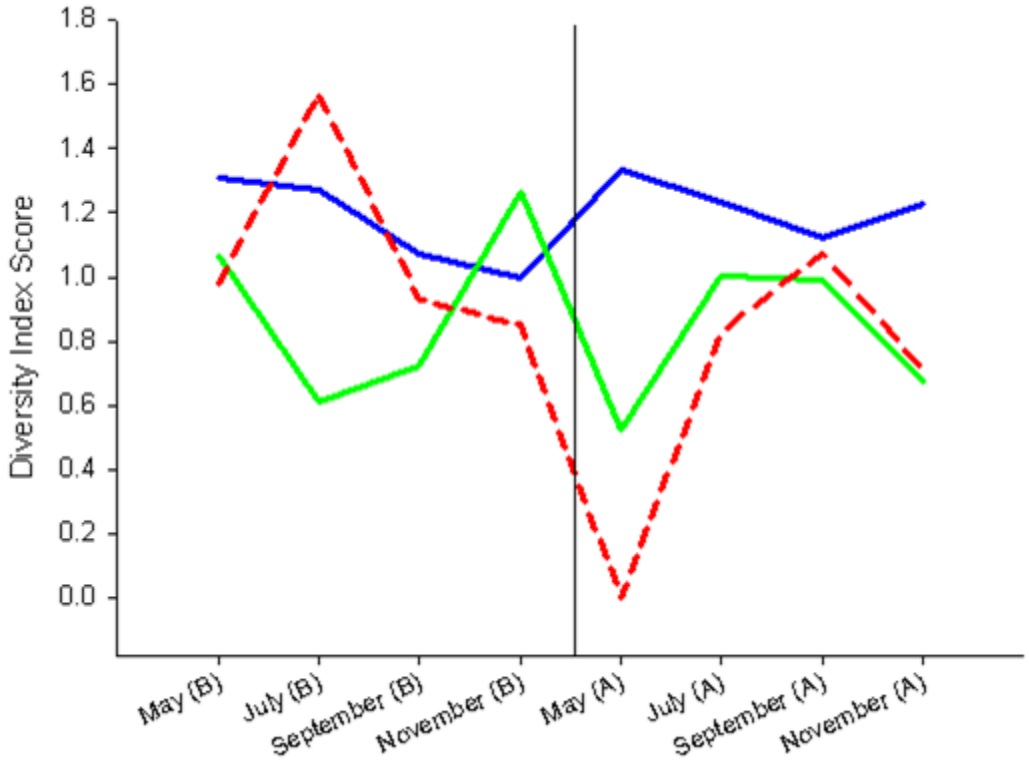

Figure 3.6. Mean Shannon-Weaver diversity index scores of log(n+1) transformed macrofauna densities (individuals m^{-2}) by month, before (B) and after (A) MAOR deployment, from control (blue), edge (green), and MAOR (red) locations. Solid vertical line indicates addition of cobble during March 2010.

Table 3.10. PERMANOVA output of main effects tests and their interactions for log(n+1) transformed macrofauna densities from all lift tray samples ($\alpha = 0.01$).

Factor	df	SS	MS	Pseudo-F	p-value
Period	1	17943.0	17943.0	3.527	0.0882
Pond	3	19035.0	6344.9	2.501	0.0007
Month(Period)	6	29463.0	4910.5	2.264	0.0010
Location(Pond)	6	16810.0	2801.6	1.306	0.1467
Period*Pond	3	4853.7	1617.9	0.765	0.6586
Site(Location(Pond))	10	11060.0	1106.0	0.962	0.5453
Period*Location(Pond)	6	12397.0	2066.1	0.931	0.6347
Month(Period)*Pond	18	39606.0	2200.3	1.914	0.0006
Period*Site(Location(Pond)	10	15357.0	1535.7	1.336	0.1206
Month(Period)*(Location(Pond))	26	49934.0	1920.5	1.670	0.0023
Total	124	268850.0			

Table 3.11. SIMPER output of the species that explain > 80 % of the dissimilarity in macrofaunal densities (individuals m^{-2}), lengths (mm) and weights (g) at MAOR sites before and after MAOR deployment.

Species	Impact (Before)	Impact (After)	% contribution to dissimilarity	% cumulative contribution
	Density (individuals m^{-2})			
Grass shrimp, *P. pugio*	69.81	20.61	66.81	66.81
Naked goby, *G. bosc*	0.14	6.46	9.97	76.78
Blue crab, *C. sapidus*	4.40	1.92	6.09	82.88
Average dissimilarity = 83.59				
	Length (mm)			
Blue crab, *C. sapidus*	26.80	28.87	20.46	20.46
Naked goby, *G. bosc*	0.00	24.18	13.91	34.37
Sheepshead, *A. probatocephalus*	0.00	27.51	10.20	44.57
Speckled worm eel, *M. punctatus*	21.26	0.00	7.02	51.59
White shrimp, *L. setiferus*	8.61	4.23	6.95	58.55
Grass shrimp, *P. pugio*	11.17	3.83	6.81	65.36
Estuarine mud crab, *R. harrisii*	5.85	8.47	6.14	71.49
Gulf Toadfish, *O. beta*	0.00	14.36	5.91	77.40
Pinfish, *L. rhomboides*	0.00	14.66	5.70	83.10
Average dissimilarity = 83.62				
	Weight (g)			
Blue crab, *C. sapidus*	2.08	4.56	30.00	30.00
Sheepshead, *A. probatocephalus*	0.00	9.42	16.08	46.08
Pinfish, *L. rhomboides*	0.00	3.84	9.25	55.33
Gulf Toadfish, *O. beta*	0.00	2.69	8.96	64.28
White shrimp, *L. setiferus*	0.52	0.32	7.76	72.04
Estuarine mud crab, *R. harrisii*	0.38	0.67	7.09	79.13
Naked goby, *G. bosc*	0.00	0.28	4.28	83.41
Average dissimilarity = 92.29				

Table 3.12. SIMPER output of the species that explain > 90 % of the dissimilarity in macrofaunal densities (individuals m^{-2}) between habitat types (mud, edge, and MAOR).

Species	Density (individuals m^{-2})		% Contribution to dissimilarity	% Cumulative Contribution
	Mud	MAOR		
Grass shrimp, *P. pugio*	39.75	20.61	50.07	50.07
Naked goby, *G. bosc*	2.06	6.46	14.21	64.27
Blue crab, *C. sapidus*	3.99	1.92	9.20	73.48
Estuarine mud crab, *R. harrisi*	3.35	0.82	9.19	82.67
Brown shrimp, *F. aztecus*	2.22	0.00	6.59	89.26
White shrimp, *L. setiferus*	1.98	0.27	3.90	93.16
Average dissimilarity = 80.56				
	Edge	MAOR		
Grass shrimp, *P. pugio*	7.21	20.61	34.93	34.93
Naked goby, *G. bosc*	0.76	6.46	21.09	56.02
Blue crab, *C. sapidus*	3.14	1.92	15.82	71.84
Estuarine mud crab, *R. harrisi*	3.08	0.82	15.05	86.89
White shrimp, *L. setiferus*	1.10	0.27	5.07	91.97
Brown shrimp, *F. aztecus*	0.35	0.00	2.02	93.99
Average dissimilarity = 81.58				
	Mud	Edge		
Grass shrimp, *P. pugio*	39.75	7.21	49.66	49.66
Estuarine mud crab, *R. harrisi*	3.35	3.08	12.88	62.54
Blue crab, *C. sapidus*	3.99	3.14	11.14	73.68
Brown shrimp, *F. aztecus*	2.22	0.35	7.26	80.94
White shrimp, *L. setiferus*	1.98	1.10	5.82	86.76
Naked goby, *G. bosc*	2.06	0.76	5.62	92.38
Average dissimilarity = 78.09				

Table 3.13. SIMPER output of the species that explain > 80 % of the dissimilarity in macrofaunal lengths (mm) between habitat types (mud, edge, and MAOR).

Species	Length (mm)		% Contribution to dissimilarity	% Cumulative Contribution
	Mud	MAOR		
Blue crab, *C. sapidus*	27.99	28.87	21.12	21.12
Naked goby, *G. bosc*	10.08	24.18	14.11	35.24
Sheepshead, *A. probatocephalus*	0.26	27.51	10.73	45.96
Pinfish, *L. rhomboides*	5.88	14.66	7.96	53.92
Brown shrimp, *F. aztecus*	12.92	0.00	7.78	61.70
Estuarine mud crab, *R. harrisii*	10.76	8.47	7.33	69.03
White shrimp, *L. setiferus*	6.91	4.23	6.63	75.67
Gulf Toadfish, *O. beta*	1.81	14.36	6.61	82.27
Average dissimilarity = 78.28				
	Edge	MAOR		
Blue crab, *C. sapidus*	18.60	28.87	23.03	23.03
Naked goby, *G. bosc*	6.09	24.18	16.84	39.87
Sheepshead, *A. probatocephalus*	0.00	27.51	11.63	51.50
Estuarine mud crab, *R. harrisii*	8.23	8.47	8.49	59.98
Pinfish, *L. rhomboides*	3.83	14.66	7.91	67.89
White shrimp, *L. setiferus*	4.21	4.23	6.88	74.77
Gulf Toadfish, *O. beta*	0.00	14.36	6.80	81.57
Average dissimilarity = 81.45				
	Mud	Edge		
Blue crab, *C. sapidus*	27.99	18.60	20.16	20.16
Brown shrimp, *F. aztecus*	12.92	5.56	12.30	32.45
Naked goby, *G. bosc*	10.08	6.09	10.63	43.08
Estuarine mud crab, *R. harrisii*	10.76	8.23	9.38	52.46
White shrimp, *L. setiferus*	6.91	4.21	9.13	61.59
Grass shrimp, *P. pugio*	9.33	6.91	7.34	68.93
Pinfish, *L. rhomboides*	5.88	3.83	4.90	73.82
Gray snapper, *L. griseus*	3.24	3.08	3.70	77.53
Darter goby, *G. boleosoma*	3.14	0.00	2.75	80.27
Average dissimilarity = 69.83				

Table 3.14. SIMPER output of the species that explain > 80 % of the dissimilarity in macrofaunal weights (g) between habitat types (mud, edge, and MAOR).

Species	Weight (g)		% Contribution to dissimilarity	% Cumulative Contribution
	Mud	MAOR		
Blue crab, *C. sapidus*	4.18	4.56	27.73	27.73
Sheepshead, *A. probatocephalus*	0.00	9.42	15.73	43.47
Pinfish, *L. rhomboides*	0.90	3.84	11.05	54.52
Gulf toadfish, *O. beta*	0.65	2.69	9.04	63.56
Estuarine mud crab, *R. harrisii*	0.69	0.67	8.43	71.99
Brown shrimp, *F. aztecus*	0.94	0.00	7.37	79.36
White shrimp, *L. setiferus*	0.59	0.32	7.35	86.71
Average dissimilarity = 91.55				
	Edge	MAOR		
Blue crab, *C. sapidus*	1.45	4.56	26.55	26.55
Sheepshead, *A. probatocephalus*	0.00	9.42	16.71	43.26
Pinfish, *L. rhomboides*	0.56	3.84	11.39	54.65
Estuarine mud crab, *R. harrisii*	0.38	0.67	9.86	64.51
Gulf toadfish, *O. beta*	0.00	2.69	9.46	73.97
White shrimp, *L. setiferus*	0.16	0.32	8.56	82.54
Average dissimilarity = 92.02				
	Mud	Edge		
Blue crab, *C. sapidus*	4.18	1.45	30.53	30.53
Brown shrimp, *F. aztecus*	0.94	0.26	15.01	45.53
Estuarine mud crab, *R. harrisii*	0.69	0.38	13.56	59.09
White shrimp, *L. setiferus*	0.59	0.16	8.76	67.86
Pinfish, *L. rhomboides*	0.90	0.56	6.01	73.87
Gray snapper, *L. griseus*	0.31	0.29	4.69	78.56
Naked goby, *G. bosc*	0.16	0.05	3.49	82.05
Average dissimilarity = 83.03				

3.4 Discussion

3.4.1 Sampling Design and Statistical Inference

- BACI Design

Previous literature has identified potential limitations of BACI experimental designs primarily attributable to type I errors, sampling designs that are incapable of accounting for ecological variance, and difficulties with interpretation of results (Hewitt et al., 2001; Stewart-Oaten et al., 1986; Stewart-Oaten and Bence, 2011). Of particular concern in executing viable statistical inferences using BACI designs are potential violations of the assumptions: 1) interval or ratio scale response variables; 2) equal variance across time and space variable combinations; 3) independence of samples and associated error structures before and after within time and space combinations; and 4) approximate normal distributions for response variables in space (Hewitt, 2001; Schwarz, 2011). In addition, as both locations (control, edge, or impact) and sites (north, south, east, west, and edge) were contained within each pond, some degree of pseudoreplication does exist within this experiment (Hurlburt, 1984).

I believe that the statistical design used for these analyses satisfies the concerns and objections associated with simple BACI designs for the following reasons: 1) a traditional "simple" temporal BACI design was not used but rather samples were collected during multiple months before and after perturbation; 2) a traditional "simple" spatial BACI design was not used but rather samples were collected from impact and control locations at spatial levels both larger and smaller than the level of impact (i.e., impact locations were sub-units of ponds and sites were sub-units of impact locations)

with adequate replication; 3) the type I error rate was reduced by collecting multiple explanatory ecological variables (i.e., species) and including them into a single analysis (i.e., PERMANOVA); 4) many of the assumption violations typically associated with parametric analysis, such as ANOVA, when analyzing BACI data are not necessary in semi-nonparametric and nonparametric tests such as PERMANOVA and ANOSIM; and 5) comparisons between factors and their levels in PERMANOVA are made using dissimilarity matrices which utilize differences between temporal and spatial units simultaneously, as was recommended to control for autocorrelation by Stewart-Oaten et al., (1986).

- Environmental Parameters

As differences in the observed data can result from ecological impacts other than the impacts controlled in the experiment (Stewart-Oaten, 1986; Stewart-Oaten and Bence, 2001), environmental parameters were collected simultaneously with experimental data as a means to measure conditions that may have influenced observed results. When analyzed, significant differences between one or more ponds were observed for all four environmental variables suggesting experimental units were not under statistically similar conditions across space. However, I disregard these differences and attribute significant differences to type I error resulting from extremely high sample size, as the observed environmental conditions in marsh ponds were very similar. Ponds were paired in different geographical locations (approximately 1 km between pairs) but were in relatively close proximity within a pair (approximately 0.25 km between ponds within a pair). Thus, concerns regarding pseudoreplication (Hurlburt, 1984) should be satisfied through a relatively large distance between pairs, and concerns regarding the expectation

of similar environmental conditions across experimental units (Hurlburt, 1984; Stewart-Oaten et al., 1986; Stewart-Oaten and Bence, 2001) should be satisfied by the relatively small distance between pairs of ponds and between ponds within a pair.

When interpreting the results from the community composition and diversity data it is important to keep in mind the criteria for evaluating enhancement success in this particular study. Enhancement success of the potential prey community was evaluated based upon: 1) the magnitude of change in community composition (density estimates) of potential prey items at MAORs compared to control sites and natural habitats, 2) magnitude of change in diversity of potential prey items at MAORs compared to control sites and natural habitats, and 3) the relative magnitude of positive versus negative effects from either or both of the first two criteria. In addition to these three criteria, a comprehensive evaluation of enhancement must also include an investigation of fish utilization at MAORs as even significant increases in potential prey do not ensure enhanced food web interactions at higher trophic levels. Chapter 4 in this thesis examines utilization of MAORs by abundant fishes, how utilization may impact fish condition, and examines comparative habitat value.

3.4.2 Meiofauna

Community composition and density at control sites (i.e. mud) were similar to previous studies of meiofauna in protected, soft-sediment habitats. Previous works cite meiofauna densities averaging approximately 10^6 individuals m^{-2} with nematodes and harpacticoid copepods as the overwhelmingly predominant taxa (McIntyre, 1969; Chandler and Fleeger, 1983; Fleeger et al., 1984; Fleeger 1985; Coull, 1999). Phillips and

Fleeger (1985) reported meiofauna densities ranging from 5.3×10^5 to 3.1×10^6 individuals m^{-2} in a shallow marsh pond in coastal Louisiana. In this study, meiofauna densities were consistently lower than literature values but peaked at approximately 5.0×10^5 individuals m^{-2}. Seasonal changes and high variability in absolute values within and between years is common due to the gregarious nature of many meiofaunal taxa, relatively fast turnover rates, and heterogeneity of taxonomic dispersion due to physical and environmental factors (Eckman, 1983; Fleeger et al., 1984; Coull, 1999). Although marsh ponds were relatively protected from most hydrodynamic disturbers (e.g., wind and waves), sampling usually coincided with peak tidal ranges resulting in complete drainage of some ponds at low tide during winter months. Fleeger et al., (1984) reported peaks and minima in meiofauna densities during low and ebb tides, respectively; finding that even relatively low tidal flow velocities were capable of significantly redistributing meiofauna. In this study, meiofauna sampling usually occurred during slack high tide and the higher flow velocities prior to high tide likely distributed some meiofauna throughout the water column. Top-down control is dismissed by many studies as a significant regulating or negative factor on meiofauna populations (Coull, 1999) but intense predation certainly occurs in shallow marsh ponds. Shallow marsh ponds in coastal Louisiana provide nursery habitat to small nekton as evidenced by high densities of fishes and invertebrates. Although predation may not be the main driver of meiofauna populations in these marsh ponds, very high densities of both demersal invertivores and pelagic filter feeding fishes may have also contributed to below-average meiofaunal densities.

Significant changes in community composition of meiofauna were associated with the addition of MAORs. Meiofauna normally associated with soft, mud bottom habitats decreased densities in samples collected at MAOR sites while densities of previously infrequent epibenthic meiofauna increased. The two predominant taxa, nematodes and harpacticoid copepods, showed order of magnitude decreases at MAOR sites despite higher overall densities of these two taxa during the "after" period. Tanaids, gastropods, polychaetes, bivalves, and ostracods showed increased densities at MAOR sites, especially in samples at the longest time interval from MAOR deployment (November 2010 – eight months after deployment). In contrast to density reductions, taxonomic diversity increased at MAOR sites from six to 13 taxa. However, densities of MAOR-specific taxa did not compensate numerically for the observed decreases in nematodes and harpacticoid copepods.

Decreases in predominant meiofauna taxa concomitant with increases in infrequent taxa in response to MAOR addition are not unexpected. Changes in habitat type and complexity are likely to provide better habitat for some meiofauna while decreasing habitat quality for others. In general, meiofauna show increases in density in response to increases in habitat complexity (Coull, 1999) but the diversity of study-specific characteristics makes this generalization dubious. The wide variety of artificial substrates as well as differences in grain size of simple bottom habitats make direct, qualitative comparisons between studies difficult. Community composition and density of organisms in the initial simple habitat (i.e., mud or sand) is heavily influenced by grain size (Coull, 1999). Therefore, the same artificial substrate added to a sandy bottom may impact taxonomic composition and density differently than if added to a finer-particle

mud bottom (Phillips and Fleeger, 1985; Coull, 1999). In response to gravel additions to littoral sandflats, Simenstad et al., (1991) found increased densities of harpacticoid copepods, no change in amphipod densities, and decreased densities of cumaceans. Hicks (1989) found significant reductions in harpacticoid densities when artificial seagrass was added to simple sand bottoms. In comparisons of meiofauna assemblages in a Louisiana estuary, Atilla et al., (2003) found stark contrasts between sandy-sediments dominated by infauna (i.e., nematodes) and pier pilings dominated by epiphytic copepods. Between-study comparisons are also complicated by differences in specificity of meiofauna as studies often focus on taxa of interest rather than all abundant groups (e.g., excluding nematodes).

Although there are numerous factors potentially impacting meiofauna communities these results were most likely attributable to the physical change from fluid, two-dimensional, small particle sediment to structurally rigid, three-dimensional cobble. Changes to vertical and structural complexity have marked impacts on hydrodynamics including water velocity and direction (Eckman, 1983). Grain size, and subsequently interstitial space, is determined in part by flow rates that can impact mobile meiofauna differently, depending on their ecology and morphology. Artificial reefs typically have larger particle size and less interstitial space than sand or mud which are likely to negatively impact smaller infauna that dominate sand/mud habitats. Nematodes, usually the predominant meiofauna taxon by both numbers and biomass, often display lower densities at artificial reef sites (Danovaro et al., 2002). In this experiment overall nematode densities increased in the second year but were orders of magnitude lower at MAOR sites during the “after” period. Atilla and Fleeger (2000) demonstrated how

complex habitats with relatively low surface area to volume ratios (e.g., the cobble stones used in this experiment) support lower meiofauna densities than substrates with higher surface area to volume ratios. The complex habitat formed by the cobble stone matrix is probably less suitable to infaunal species than the interstitial matrix formed by fine particle sediments. Numerous, micro-scale interstitial spaces found in muddy or sandy sediments would be replaced with infrequent, meso-and macro-scale interstitial spaces within cobble plots. Additionally, the rigidity and structural integrity of limestone cobble would disrupt normal burrowing behavior exhibited by infaunal species. Increased densities of tanaids, gastropods, polychaetes, bivalves, and ostracods within cobble plots suggest MAORs provide better habitat for larger epiphytic and epibenthic meiofauna but the specific mechanisms driving community differences are unclear.

Potential influences upon meiofauna community composition in response to MAOR construction include: 1) changes to hydrodynamic parameters resulting from increased vertical and structural complexity, 2) changes in chemical composition of sediments and micro-scale water quality, 3) changes in food distribution and composition, and 4) changes in foodweb dynamics through predator/prey interactions (Danovaro et al., 2002). Despite significant relationships between abundant meiofauna taxa and various explanatory variables, adjusted-r^2 values were low (26-43%). Low correlation values suggest that the environmental parameters and potential predators sampled in this study were not the main drivers of meiofauna densities, and support the contention that meiofauna are not regulated by top-down or bottom-up control. Additional data, for example about grain size or flow velocities, may have improved this study.

3.4.3 Macrofauna

Community composition and density of macrofauna in this study were lower than reported literature values but were within the expected ranges for total density, individual species densities, and species diversity. Rozas et al., (2005) identified 33 species of macrofauna from drop samples of vegetated marsh and pond areas in Breton Sound, Louisiana finding similar total numbers (4596 individuals in 100 samples, compared to 3731 individuals in 124 samples in this study). Baltz et al., (1993) reported a total of 57 fish species ranging in density from 1 to 889 individuals m^{-2}, in for the edge community in Louisiana salt-marshes. Zimmerman and Minello (1984) identified 29 separate species in a study of macrofauna in vegetated versus non-vegetated habitats in a Texas salt-marsh, with grass shrimp having the highest individual species densities (70 m^{-2}). In this study a total of 27 macrofauna species were identified, and densities ranged from 1 to 205 individuals m^{-2} with an average density of 46 individuals m^{-2}.

Macrofauna densities were likely reduced by a combination of gear selectivity, proximity of ponds to sources of emigration, and predation. Stepwise regression analysis indicated environmental factors and taxa/species interactions significantly affected observed densities of macrofauna but the proportion of variation explained in stepwise regression analysis was very low (≤ 0.25). Because lift trays were completely filled with sediment and had relatively low areal coverage, escapement of certain species may have been high. Low-mobility and cryptic forms of macrofauna such as xanthid crabs and gobies probably exhibited lower escapement rates than pelagic, free swimming forms such as menhaden and anchovies. Zimmerman and Minello, 1984, Baltz et al., 1993 and Rozas et al., 2005 all used large, cylindrical drop samplers to trap the entire water column

and reduce escapement. The seclusion of marsh ponds from major channels and bays may also have reduced absolute values by lowering immigration rates to marsh ponds. Previous studies included samples from sites in direct connection with large channels, which may have maintained higher immigration rates and densities. The marsh ponds I studied had only a single entrance/exit and were surrounded by dense vegetation primarily comprised of *Spartina alterniflora*. These marsh ponds also exhibited relatively high densities of predatory fishes (evidenced from other gear types in this study but not discussed), which may have effectively reduced absolute densities of macrofauna, especially invertebrates. Although they were not significantly related to macrofauna densities, environmental variables such as salinity, water temp, and depth have been found to contribute significantly to the variability in macrofaunal communities in salt-marshes in previous studies and likely regulated absolute densities in this study as well (Zimmerman and Minello, 1985; Baltz et al., 1993; Rozas et al., 2005).

General trends in community composition and density of macrofauna associated with MAORs are most likely attributable to factors associated with the temporal scale at which this experiment was conducted. Visual monitoring of lift tray samples indicated little to no colonization of cobble stones throughout the majority of the experiment. While fouling organisms (e.g., barnacles, oysters, tube building invertebrates, and epiphytic algae) may colonize new substrates in relatively short time periods (i.e., months; Brown and Swearingen, 1998) very few stones showed any colonization, and abundances were very low when colonization was present. Colonization at MAOR sites was not expected to compare with mature natural and artificial oyster reefs after only one year. The intense macro- and microstructural complexity of sessile and subsequent

successional colonizing invertebrates at natural and artificial oyster reefs is achieved only after multiple seasons and reproductive cycles. Successional communities in shallow estuarine habitats in coastal Louisiana are typically dominated by only a few species with highest recruitment rates occurring in late winter and early spring (i.e., February; Brown and Swearingen, 1998). Although MAORs were deployed in March, the seclusion of marsh ponds and time of deployment may have reduced exposure rates of colonizing larvae. Epiphytic colonization could provide short-term microscale complexity but turbidity levels greatly reduce algal growth for most of the year.

Where meiofauna showed diversification and moderate regime shifts in response to MAOR addition, macrofauna were reduced in both number and diversity. Of the 27 species identified, only four species display positive biological impacts at MAOR sites during the "after" period. In contrast to decreases in density and diversity, total length and weight of macrofauna was greater at MAOR sites during the "after" period. Individual species lengths and weights were higher at MAOR sites during the "after" period for blue crabs, estuarine mud crabs, naked gobies, pinfish, gulf toadfish and sheepshead; three of which have strong structural affinities. Naked gobies utilize the cracks and crevices found in complex habitats such as oyster reefs to decrease predation at all life stages (Breitburg, 1991). Gulf toadfish are cryptic carnivores that utilize complex habitats to ambush a wide variety of prey items. Sheepshead are omnivores that feed heavily on shelled invertebrates, such as barnacles and oysters, when abundant. Pinfish are also omnivorous and can occupy a wide variety of habitat types but typically display higher densities in structurally complex habitats (Zimmerman and Minello, 1984; Baltz et al., 1993).

The significant decrease in density of meiofauna suggests an overall negative impact on feeding potential of juvenile marsh pond fishes but does not preclude the potential for enhancement. Positive impacts to structure-associated macrofauna at MAOR sites suggest that only a minority group of fishes with specific biological and ecological characteristics were able to effectively utilize MAOR-associated resources. Although spatially and temporally limited, the addition of MAORs may have provided additional resources for feeding or refuge as well as relief from competitive interactions. Significantly larger fishes and invertebrates at MAORs suggests ontogenetic shifts in behavior or morphological feeding plasticity may allow some macrofauna to successfully exploit local shifts in prey resources (Cutwa and Turingan, 2000). The presence of larger individuals at MAOR sites suggests some degree of stage-specific utilization and possible enhancement, but cannot be confirmed without explicit data on resource utilization or concomitant data on growth rates or other life parameters.

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CHAPTER 4: THE EFFECTS OF MIMIC ARTIFICIAL OYSTER REEFS ON SELECT ESTUARINE FISHES IN MARSH PONDS: A BEFORE-AFTER-CONTROL-IMPACT ANALYSIS

4.1 Introduction

4.1.1 Artificial Habitats and Habitat Enhancement

Artificial habitats have been deployed in a wide variety of locations and designs (see reviews by Grove et al., 1989; Grove et al., 1991; Pickering and Whitmarsh, 1997; Relini et al., 2007), often with intent to increase catches, reduce effort within a local fishery (Whitmarsh et al., 2008), or enhance ecosystem productivity (Relini et al., 2007). Artificial habitats have been shown to augment ecological and biological processes by reducing the intensity of negative stressors through structural resilience (Gardner et al., 1996; Hernkind et al., 1997), the dynamics of colonization (Sale and Dybdahl, 1975; Rodney and Paynter, 2006), and subsequent utilization by consumers (DeMartini et al., 1994; Johnson et al., 1994; Fabi et al., 2006). Primary and secondary consumers often utilize epiphytic, epifaunal, or fouling communities rather than direct consumption of living or decomposing host substrate (Moncreiff and Sullivan, 2001; Fabi et al., 2006). This implies that some artificial habitats can be productive if they provide adequate availability of areal substratum for colonization and food web support. The enhancement of trophodynamics at multiple levels (e.g., Reed et al., 2006; Lingo and Szedlmayer, 2006; Perkol-Finkel, 2007) can increase diversity (Fabi et al., 2004) as well as local (DeMartini et al., 1994; Johnson et al., 1994; Relini, et al., 2007) or even ecosystem level productivity.

The fisheries management community recognizes the potential value in enhancing existing natural, and/or degraded habitats, as well as creating new habitats through deployment of built structures. Artificial habitats have been deployed for a wide variety of management purposes in coastal estuaries along the Gulf of Mexico. For example, the harvested shell from oyster leases is usually redistributed or replaced by limestone cobble to augment not only recruitment of oyster spat but reduce crowding to optimize morphological desirability of harvested oysters as well. In 2006, approximately $47 million was distributed across all states along the Gulf of Mexico through the fin- and shellfish management plan for repair and restoration of inshore artificial reefs, particularly those that mimic oyster reefs (VanderKooy and Freitas, 2006).

4.1.2 Oyster Reefs as Habitat

The importance of oyster reefs to estuarine ecosystems was illustrated by the loss of approximately 98% of the original Chesapeake Bay oyster population, which resulted in significant reductions in water quality and trophic cascades (Rothschild et al., 1994; Coen et al., 1999). Because oyster reefs produce large surface areas of hard substrate, they enhance recruitment of sessile invertebrates and provide critical settlement habitat for oyster spat (Coen and Grizzle, 2007). Settlement may be enhanced by the vertical relief and heterogeneous habitat complexity which can reduce horizontal water velocities down-current of the leading reef edge that can enhance vertical movement and create micro-turbulent flows that may deliver larvae directly to reef substrate (Eckman, 1987; Abdelrhman, 2003). Reefs also increase persistence after settlement (Bologna and Heck, 2000; Koehl, 2007). Enhancement of primary production may result from accumulating

drifting algae (Davis et al., 2009) or when epiphytic producers occupy substrate surfaces (Reed et al., 2006; Pondella et al., 2006).

Compared with other complex habitat types, oyster reefs can produce similar and even increased densities of fishes and invertebrates (particularly structure-associated species), often exhibiting significantly higher densities when compared with non-vegetated, mud bottoms (Zimmerman et al., 1989). Complex microspaces provide food and structurally rigid refugia that may greatly reduce macrofaunal predation pressure (Hall and Bell, 1988). This results in enhanced biodiversity, especially among invertebrates, where as many as 300 species of annelid worms, amphipods, isopods, crabs, shrimps, copepods, and other bivalves are often found in high densities that might not persist in adjacent non-reef habitat (Wells, 1961; Zimmerman, 1989; Peterson et al., 2003; Stunz et al., 2010).

4.1.3 Habitat Enhancement

Fisheries managers are particularly interested in investigating the role of artificial oyster reefs, not only in the potential to support increased productivity in estuarine environments, but in the potential for enhancement of nursery habitat for estuarine dependent fishes (Steimle and Meier, 1997). Although resident oyster reef fishes are most reliant upon reef-associated resources, Coen et al. (1999) highlighted the potential use by facultative, transient estuarine species often having more generalized requirements of complex habitats (Minello et al., 2003), many of which are economically important. Of the 15 most abundant fish species found by Baltz et al. (1993) in Louisiana estuaries, 67% were estuarine dependent transients (i.e., they are not exclusive to one habitat type

within the estuary at all life stages but are dependent upon and utilize multiple habitats within estuaries during at least one life stage). A great proportion of these transients depend on complex habitats such as oyster reefs for food and refuge during pre-adult stages (Minello et al., 2003).

Studies often highlight increased abundances, densities, or biomass as evidence for productivity increases directly attributable to oyster reefs, especially when compared to unvegetated (Minello et al., 2003; Stunz et al., 2010) or natural mud bottoms (Simonsen, 2008). However, direct linkages between reef associated species and oyster reef resource utilization are necessary to assess their importance as fish habitat (Beck et al., 2001). Abundant fish presence on a reef may imply utilization but does not preclude the potential for attraction without increasing production, especially at the ecosystem level (Lindberg, 1997; Lindberg et al., 2006).

4.1.4 Trophic Linkages and Resource Utilization

One method to directly quantify linkages between oyster reef resources and associated fishes is to determine the proportion of reef resources directly consumed by predators (DeMartini et al., 1994; Peterson et al., 2003). Studies of gut contents from reef associated fishes show relatively large proportions of prey directly consumed from oyster reefs (Peterson et al., 2003; Simonsen, 2008). Peterson et al., (2003) quantified reef utilization through diet analysis and extrapolated productivity throughout the potential lifetime of a restored oyster reef. Assuming protection from harvest, environmental damage, and consistent productivity rates, Peterson et al. (2003) estimated that $10m^2$ of restored oyster reef could yield as much as 2.6 kg yr^{-1} of fish and crustaceans for the

functional lifetime of the reef (i.e., up to 30 years). When considering the existing areal distribution of oyster reef habitats as well as potential area for reef deployment (i.e., over natural mud or sand bottoms) in estuaries along the Gulf of Mexico, the potential productivity becomes quite respectable (Peterson et al., 2003).

Deployment of mimic artificial oyster reefs provides an opportunity to determine relative habitat value while gaining valuable insight into how fishes utilize available resources. Fishes select resources that maximize trade-offs between energetic return and survival probability (Manly et al., 2002). The processes controlling resource utilization during early life are complex. As fish mature they experience drastic changes in body size, morphology, physiology, and nutritional requirements, all of which influence diet composition (Wuenschel, et al., 2006) and, ultimately, how resources are utilized. Because enhancement of available food resources can have direct impacts on vital rates, knowledge of habitat-specific diet composition is essential to determining the role of artificial habitat in estuarine food webs. Positive impacts upon fish diet may result from general increases in prey abundance, increases in preferred prey, or increased diversity of prey items and sizes. Documentation of habitat-specific resources and habitat-specific utilization are important to management of exploited populations by providing greater resolution than density comparisons alone, and can be used to calculate biomass production values (Peterson et al., 2003; Powers et al., 2003).

4.1.5 Nutritional Condition of Estuarine Fishes

Energy density is a robust indicator of total body condition as it is sensitive to changes in proximate components such as lipids and proteins (Anthony et al., 2000).

Therefore, condition comparisons can provide a measure of relative habitat value and can be used to assess the magnitude and ecological significance of diet shifts when observed in gut content studies (Lloret and Planes, 2003; Nemerson and Able, 2005). Changes in total body condition may result from increased feeding efficiency (i.e., greater energetic return per energy expenditure) or changes in the nutritional value of resources consumed. For example, meiobenthic nematodes, which can far outnumber other meiobenthic taxa in sediments, can comprise as much as 90% of the energy content available to fishes (Scholz, et al., 1991), but may not be efficiently consumed by many species, especially relatively large predators. Additionally, the per-gram caloric content of benthic amphipods is only half that of pelagic amphipods, copepods, and decapod larvae (Wissing, et al., 1973). Therefore, habitat-specific, differential size distributions and nutritive prey values may provide disproportionate energetic return to feeding fishes.

4.1.6 Research Goals

The goal of my research was to determine whether MAORs (Mimic Artificial Oyster Reefs) enhance juvenile fish nursery function within marsh ponds. My objectives in this chapter were to:

1) determine how dietary utilization by fishes differs between artificial reef sites and other natural habitats in terms of prey type, quantity consumed, and dietary importance;
2) determine if the artificial reef-associated effects on the potential prey community observed in the previous chapter are observed at higher trophic levels and how this affects food-web interactions;

3) a) assess comparative habitat value of MAORs versus other natural habitats using composition-based fish condition as a proxy for relative habitat value; and

3) b) in the absence of dietary impacts use condition differences as an alternative indicator of impacts attributable to MAORs.

4.2 Methods

4.2.1 Study Area

The study area was located near Empire, Louisiana approximately 27 miles northwest of the Head of Passes in the Mississippi River's Balize delta, and consists of four intertidal marsh ponds adjacent to Vacherie and Adams Bays (Figure 4.1). The four experimental ponds are referred to as Ovary pond (OP), Perfect pond (PP), Triangle pond (TP), and Big pond (BP; Figure 4.1). These marsh ponds are oligohaline and characterized by shallow depths (~1 m relative to mean high water), unvegetated mud bottoms, and are surrounded by emergent vegetation consisting mostly of smooth cordgrass (*Spartina alterniflora*). Ponds are relatively large, ranging in surface area from 6,600 m^2 – 16,800 m^2, and have only a single opening to nearby open waters.

4.2.2 Field Methods

- Sampling Design and Artificial Habitat Deployment

Four experimental ponds were sampled once every other month in a randomly selected order for the duration of two years, from March 2009 – March 2011. No sampling occurred in March 2010 as cobble material was added at the conclusion of the first sampling year. The "before" period consisted of sampling events that occurred from

Figure 4.1. Map of the four experimental marsh ponds within marshes adjacent to Adams and Vacherie Bays near Empire, Louisiana.

March 2009 - January 2010, and the "after" period consisted of sampling events from May 2010 – March 2011. A sampling event consisted of one pond being sampled each day over a successive four-day period. Sampling occurred at the same point in the monthly tidal cycle and at the same point in each daily tidal cycle. The initial monitoring period, along with modification and the subsequent experimental period, allowed for direct comparisons of factors in time (before or after) and space (control or impact) as is required for BACI experimental designs. Within each of the four ponds, five fixed sites were selected for sampling for the duration of the study (Figure 4.2). During the "before" period each pond contained four mud bottom sites and one non-vegetated, marsh edge

site (Figure 4.2; sites within each pond are referred to based upon orientation to the compass rose). In March 2010, (the end of the "before" sampling period) north and east sites in both PP and OP received #57 limestone cobble designed to mimic natural oyster reef substrate. Of the four sites that received limestone cobble treatments, all consisted of mud bottoms during the "before" period. Thus, in PP and OP, one of the five sites within each pond remained non-vegetated marsh edge, two of the four sites that were previously mud became MAOR, and two of the four sites remained mud bottom. No mud sites in BP or TP (control ponds) and none of the edge sites in any pond received limestone cobble treatments. Because ponds were not equal in size, MAOR dimensions were scaled to one-tenth the size of the pond receiving the artificial habitat. The sediment surface area covered by limestone cobble at each site after habitat deployment was approximately equal to 1% of the total surface area of that pond. Prior to MAOR deployment, mesh netting was placed across the sediment surface to prevent reefs from sinking into the mud bottom. Mesh netting was held in place using Polyvinyl Chloride (PVC) stakes that were removed immediately after reef deployment. Limestone cobble was evenly distributed (as possible) so that all MAORs were approximately 5 cm in height above the sediment surface. This resulted in MAOR dimensions of approximately 15 x 15 x 0.05 m and 22 x 22 x 0.05 m in OP and PP, respectively. Marking stakes were left at the four corners of each cobble plot to allow accurate sampling of MAOR habitats.

4.2.3 Environmental variables

Temperature, water depth, salinity, and pH were measured using a YSI 6920 V2 multi-parameter hydrosonde, which was attached to a PVC pole and placed in the center

Figure 4.2. Site map of Perfect pond containing five fixed sampling sites: 1) north – MAOR (during 'after' period only); 2) east – MAOR (during 'after' period only); 3) south – mud bottom; 4) west – mud bottom, 5) edge – unvegetated marsh edge. Perfect pond size was approximately 16,800 m^2.

of each pond when sampled. Readings were taken every five minutes for the duration of each daily sampling event. The hydrosonde was only deployed while sampling. Although seasonal variation in mean water depth could not be controlled, all sampling occurred at approximately the same point in the tidal cycle (6-8 hours before high tide) on each successive day during each four-day sampling trip. Sampling on each day began when

water depths were deemed sufficient for equal habitat availability to fishes to reduce the effect of hydrologic drivers, to better reflect potential habitat preferences.

4.2.4 Data Collection

Atlantic croaker, bay whiff (*Citharichthys spilopterus*), sand seatrout (*Cynoscion arenarius*), and pinfish (*Lagodon rhomboides*) were selected for gut content analyses based upon feeding ecology and their relatively high abundance in marsh ponds. Fishes were collected by deploying a 15.24 m bag seine (bag size 1.44 m^2) with 0.63 cm mesh, twice at each site within each pond (minimum of 10 tows per pond, 40 tows per sampling trip). For marsh-edge sites, one end of the seine net was placed at the marsh-edge and held stationary. The other net-end was then fully extended perpendicular to the marsh edge and towed along the shoreline while the one end of the net remained fixed at its initial location. Once the towed end of the net reached the marsh edge, both net ends were towed together along the marsh-edge to the midpoint of the net forming a circle. The net was then pulled onshore and all nekton removed. Care was taken to perform the second seine tow over an area that was not sampled by the first seine tow at each site during a single trip while still remaining within the dimensions of that fixed site; to avoid depletion affects. All fishes collected for gut content analysis were preserved in 95% ethanol, labeled, and stored for report to the laboratory. For relatively large fishes, stomachs were removed from the body cavity, labeled with the fish's relevant information (species name, morphometrics, sampling site, and date) and preserved individually in ethanol-filled jars. Gut contents were identified to the lowest practicable taxon, enumerated, and measured for length and width using an ocular micrometer.

Micrometer measurements were converted to millimeters and the volume (mm^3) of each prey item was then calculated using the formula for the volume of a cylinder.

Electivity indices were calculated using density estimates of infaunal and epibenthic meiofauna collected in lift trays (Chapter 3) as well as emergent meiofauna and mesozooplankton from plankton tows. Emergent meiofauna and mesozooplankton were collected using a square plankton net of dimensions (1.0 m width, 0.5 m height, 3 m length; 1 mm square mesh diameter). Plankton nets were deployed in duplicate when water depth was approximately 0.5 m so that the entire water column was sampled. Nets were deployed by loosely draping a loop over a PVC pole above the water surface. A small boat used to deploy the net then drifted to the end of two 7.62 m ropes attached to each side of the net frame. Additional PVC poles were then used to maintain boat position during net retrieval. Upon, retrieval the net was rinsed to move captured organisms into the cod end. Samples were immediately preserved in 95% ethanol.

4.2.5 Data Processing and Statistical Analyses

- Environmental Variables

Environmental variables were analyzed as separate response variables using a mixed-model ANOVA in SAS. Each mixed-model test included one of the four environmental variables sampled as the response variable and included three factors as explanatory variables: factor A: Period (fixed with a = 2 levels; before or after), factor B: Month (random with b = 4 levels; May, July, September, or November; nested within factor B), factor C: Pond (random with c = 4 levels; Big, Ovary, Perfect, or Triangle) and their interaction terms. When main effects terms were significant (i.e., Period*Pond or

Month*Pond), ponds were then compared using pairwise tests of Tukey-adjusted LSmeans. As environmental variables were primarily collected for the purpose of testing the assumption that hydrographic conditions in ponds were similar during any given sampling event, the sub-level factors Location and Site were included in the data as "replicates" but were not included in the statistical model.

- Diet Composition and Electivity

Percent number (*%N*), percent volume (*%V*), and percent frequency of occurrence (*%FO*) were calculated for each prey type for each of the four fishes. Percent number was calculated by dividing the cumulative total of all prey in each prey category by the cumulative total of all prey in all prey categories in all stomachs of a single fish species. Percent volume was calculated by dividing the cumulative volume of all prey in each prey category in all stomachs by the cumulative volume of prey in all prey categories. Percent frequency of occurrence was calculated using the formula:

$$\%FO = \left(\frac{Number\ of\ stomachs\ containing\ one\ prey\ category}{Number\ of\ stomachs\ containing\ prey}\right) \text{x } 100$$

The variables (*%N*, *%V*, and *%FO*) were then used to calculate both an index of relative importance (IRI) as well as a percent IRI for all prey items (McCawley and Cowan, 2007). The IRI was calculated using the formula:

$$IRI = (\%N + \%V)\ x\ \%FO$$

The percent IRI was then calculated using the formula:

$$\%IRI = \left(\frac{IRI\ for\ each\ prey\ category}{Sum\ of\ all\ IRI\ values}\right) x\ 100$$

As some prey types are relatively small but consumed in large quantities while others are relatively large but consumed infrequently, the *IRI* and *%IRI* are considered more robust than numbers or volumetric data alone as they incorporate numbers, volume, and frequency of prey items into a single metric.

To investigate the contribution of each prey type to overall diet quality the index of caloric importance was calculated using the formula derived by McCawley and Cowan (2007):

$$ICI = (\%W + C)\ x\ \%FO$$

Where W is the relative weight of each prey category, C is the dry weight energy density of each prey category (Joules g^{-1}), and FO is the frequency of occurrence. Relative weights for each prey category were calculated according to the methods used in Stobberupp et al. (2010). Dry weight energy density estimates were obtained from the literature (Wissing et al., 1979; McCawley et al., 2003; Hansen et al., 1997). Because relative weight formulas could not be found for all prey categories, categories with a percent IRI value less than 0.1 percent were eliminated from the ICI and percent ICI calculations.

The percent ICI was then calculated using:

$$\%ICI = \left(\frac{ICI\ for\ each\ prey\ category}{Sum\ of\ all\ ICI\ values}\right) x100$$

Electivity indices were calculated, using Ivlev's electivity index, to determine if fishes exhibited preference for any prey item or if prey were simply consumed opportunistically (Ivlev, 1961; Lechowicz, 1982):

$$E_i = (r_i - p_i) / (r_i + p_i)$$

where p_i is the relative proportion of each prey item in the environment and r_i is the relative proportion of each prey item in the stomachs of each fish species. Prey items that are consumed in greater proportion than found in the environment (i.e., positive electivity values) are considered preferred; prey items consumed in lesser proportion than found in the environment (i.e., negative electivity values) are considered avoided; and prey items consumed in proportions relatively similar to their abundance in the environment (i.e., electivity value of zero) are considered to be consumed at random (Lechowicz, 1982).

To determine the proportional density of prey items in the environment, lift tray samples were standardized to 1 m^2 using the methods presented in Chapter 2. Meiofauna densities in plankton tow samples were estimated by multiplying area of the net opening (0.5 m^2) by distance towed (7.62 m), then standardized to 1 m^{-3}. Density estimates (individuals·m^{-3}) were then converted to areal densities (individuals·m^{-2}) by multiplying by the mean water depth (m) in each pond during each sampling event. Densities of meiofauna from plankton tows and lift trays were then summed for each prey item and log(n+1) transformed (due to disproportionately high nematode density). Percent density was then calculated for each prey item in the environment using the log-transformed density estimates. Numbers consumed of each prey type were used (as opposed to using volume consumed or the IRI) to calculate the percent prey consumed in fish stomachs for use in calculating the electivity indices (Ivlev, 1961).

Stomach content data were analyzed using PRIMER 6 with PERMANOVA, which is specifically designed for analyses of multivariate data in ecological studies

(Clarke and Warwick, 2001; Clarke and Gorley, 2006; Anderson et al., 2008, DeMutsert, 2010). In PRIMER, individual stomachs are treated as replicates and are used to create Bray-Curtis resemblance matrices. The matrices are then analyzed for statistical significance using PERMANOVA (a semi-parametric equivalent of MANOVA). Five factors were included in the PERMANOVA analyses: factor A: Period (fixed with a = 2 levels: before or after), factor B: Month nested within Period (random with b = 4 levels: May, July, September, or November), factor C: Pond (random with c = 4 levels: Big, Ovary, Perfect, or Triangle), factor D: Location nested within Pond (fixed with d = 3 levels: edge, control, or impact), and factor E: Site nested within Location (random with e = 5 levels: north, south, east, west, or edge). The terms Habitat (3 levels: mud, edge, or MAOR) and Interaction (6 levels: before-control, before-edge, before-impact, after-control, after-edge, and after-impact) were included for SIMPER comparisons of dissimilarity. Although the experimental design contained elements of a traditional BACI analysis, I wanted to include as much spatial and temporal variation into the analyses as possible. Therefore, when testing the overall effects of MAOR addition on meiofauna and macrofauna, the statistical design was essentially analyzed as a split-plot design with time components. The simple BACI design factors representing the time and space variance components were included (i.e., Period and Location), along with the additional levels listed. The factor Month also was included in the temporal portion of the model to better structure the temporal variation. As each pond contained the sub-level factors Location and Site (within each location) each represented a plot, and each sub-level represented a sub-plot. The "split" was determined by the addition of MAORs and observed in both the Period and Location factors (i.e., Period was split into "before" and "after" and Location

was split into “control,” “impact,” or “edge”). PERMANOVA was used to test the full model but only the interaction term Period*Location (significance indicates effect of habitat addition) was of major statistical interest as is typically evaluated in BACI experimental designs. PERMANOVA was run using 9999 permutations and tests were evaluated at a significance level of $p = 0.01$. Significance tests on each combination of factors Period and Location were performed using ANOSIM (two-way crossed with replicates; $p = 0.1\%$), which is a non-parametric equivalent of ANOVA. The SIMPER procedure was used to determine which prey groups contributed most to dissimilarities between MAOR sites before and after limestone cobble addition, and between habitat types. Differences in mean lengths of fishes were analyzed using Tukey-adjusted least square means (LSmeans).

- Energy Density

Energy densities (Joules·gram^{-1}; dry weight) were compared among the four fish species using total body fish condition as a proxy for habitat quality. Energy density values were measured directly using a Parr 6200 oxygen bomb calorimeter. As bomb calorimetry analysis requires a minimum sample dry weight of 0.6-1.2 g, each sample usually consisted of multiple fish that had been dried for 48 hrs at 60 °C, then homogenized using mortar and pestle. Each sample was tested for energy density in triplicate when sample weights were sufficient. Samples consisted of whole fish previously analyzed for gut content analysis and whose intestines had already been removed and discarded.

Differences in fish energy density were analyzed separately for each species to test the effects of MAOR addition and to compare habitat types. Because there was only one response variable (energy density) a simpler BACI statistical design was used due to limitations in the degrees of freedom. The main effects Period, Location, and the interaction term were included in a general linear mixed-model in SAS (mixed-model ANOVA). The main-effect term Month was also included in the model and listed in the repeated statement to structure the natural variation of energy densities over time. Habitat-specific energy densities were compared using ANCOVA (mixed-model ANOVA) with the main effects Period, Habitat and the interaction term and analyzed for significance using the type III sums of squares. The factor Month was included in the random statement as not all habitats (i.e., MAORs) were sampled repeatedly over time making the repeated statement invalid. Mean values for energy density were determined using LSmeans.

Appendix 1 lists the statistical technique used for each set of analyses conducted in this experiment including response and explanatory variables, general model with effect terms for each test, and analyses techniques used for any additional treatment comparisons.

4.3 Results

4.3.1 Environmental Variables

Water temperature (°C), depth (m), salinity (ppt), pH and dissolved oxygen (DO; $mg \cdot L^{-1}$) data were collected every other month from September 2009 – January 2010 (before artificial reef deployment) and May 2010 – March 2011 (after artificial reef

Table 4.1. Monthly mean values for water temperature (°C), depth (m), salinity (ppt), pH and dissolved oxygen ($mg \cdot L^{-1}$) for Big, Ovary, Perfect, and Triangle ponds before and after MAOR addition.

Environmental Variable	Period	Month	Big	Ovary	Perfect	Triangle
Water Temp (°C)	Before	September	30.45	29.29	30.96	29.38
		November	19.40	21.67	16.80	16.23
		January	14.84	21.67	15.94	16.07
	After	May	27.44	26.48	25.49	26.24
		July	31.17	30.11	30.25	32.28
		September	31.24	28.91	28.53	29.77
		November	19.97	22.10	22.56	23.13
		March	20.91	24.17	.	.
Depth (m)	Before	September	1.15	0.84	0.89	0.95
		November	0.91	0.86	0.85	0.81
		January	0.81	0.52	0.75	0.67
	After	May	0.67	0.73	0.73	0.79
		July	0.95	0.69	0.76	0.69
		September	0.76	0.70	0.89	0.76
		November	0.87	0.72	0.90	0.86
		March	0.55	.	.	.
Salinity (ppt)	Before	September	11.89	11.28	11.57	11.79
		November	23.45	21.68	20.79	20.22
		January	15.13	14.67	14.50	14.50
	After	May	8.82	7.44	6.96	7.15
		July	4.54	4.53	4.75	5.08
		September	9.90	10.14	10.62	10.43
		November	18.87	19.88	20.07	19.87
		March	18.50	17.75	.	.
pH	Before	September	7.23	7.35	7.90	7.57
		November	7.03	7.10	7.45	7.50
		January	7.72	8.00	8.12	8.08
	After	May	7.60	7.60	7.73	7.61
		July	7.81	7.56	8.24	8.39
		September	7.16	7.47	7.98	7.54
		November	7.63	7.66	7.73	7.70
		March	7.38	7.66	.	.
Dissolved Oxygen ($mg \cdot L^{-1}$)	Before	September	27.89	35.47	89.03	51.25
		November	74.58	71.52	95.62	89.52
		January	91.19	111.34	96.17	97.12
	After	May	81.11	73.07	75.86	81.44
		July	85.98	64.15	97.42	128.43
		September	43.46	46.29	78.91	57.78
		November	85.20	75.43	84.61	82.91
		March	75.73	111.28	.	.

deployment). Mean monthly values for each environmental variable are listed in Table 4.1. No data were collected from March 2010 because limestone cobble for artificial reefs was being deployed during much of this month. Data were only collected from Big and Ovary ponds in March 2011 due to an equipment malfunction. In addition, only the months of September and November were included for pond comparisons between periods as these were the only two months in which hydrographic data were collected during both "before" and "after" periods (i.e., no data were available for May, July, or March in the before period, and no data were available for January in the after period).

Mean water temperature, depth, salinity, pH, and DO were all significantly different between periods ($p<0.001$), months ($p<0.0001$), and ponds ($p<0.0001$; mixed-model ANOVA). Water temperature followed seasonal trends with minimum values in winter (January) and maxima in summer (July and September; Figure 4.3). Salinity was lowest in summer (July) and peaked in the fall (November; Figure 4.3). Water depth and pH were relatively variable throughout the study period and followed no apparent seasonal trends (Figure 4.3). However, pH may have been associated with high DO values, especially during winter months when water depth was low and filamentous algae were abundant. The addition of limestone cobble to Ovary and Perfect ponds did not cause any consistent change in pH values during the "after" period as trends in pH between these two ponds were not similar over time (Figure 4.3).Trends in pH appeared more closely related to geographic location as ponds in close proximity to one another (Ovary and Big ponds and Perfect and Triangle ponds; Figure 4.1) displayed more similar trends over time than ponds farther apart (Figure 4.3). Mean water temperature ranged from 14.9 ºC (Jan. 2010) to 30.0 ºC (July '10); mean water depth ranged from 0.55 m

(March 2011) to 0.97 m (Sept. 2009); mean salinity ranged from 4.6 ppt (July 2010) to 21.6 (Nov. 2009); mean pH ranged from 7.2 (Nov. 2009) to 7.9 (Jan. 2010); and mean

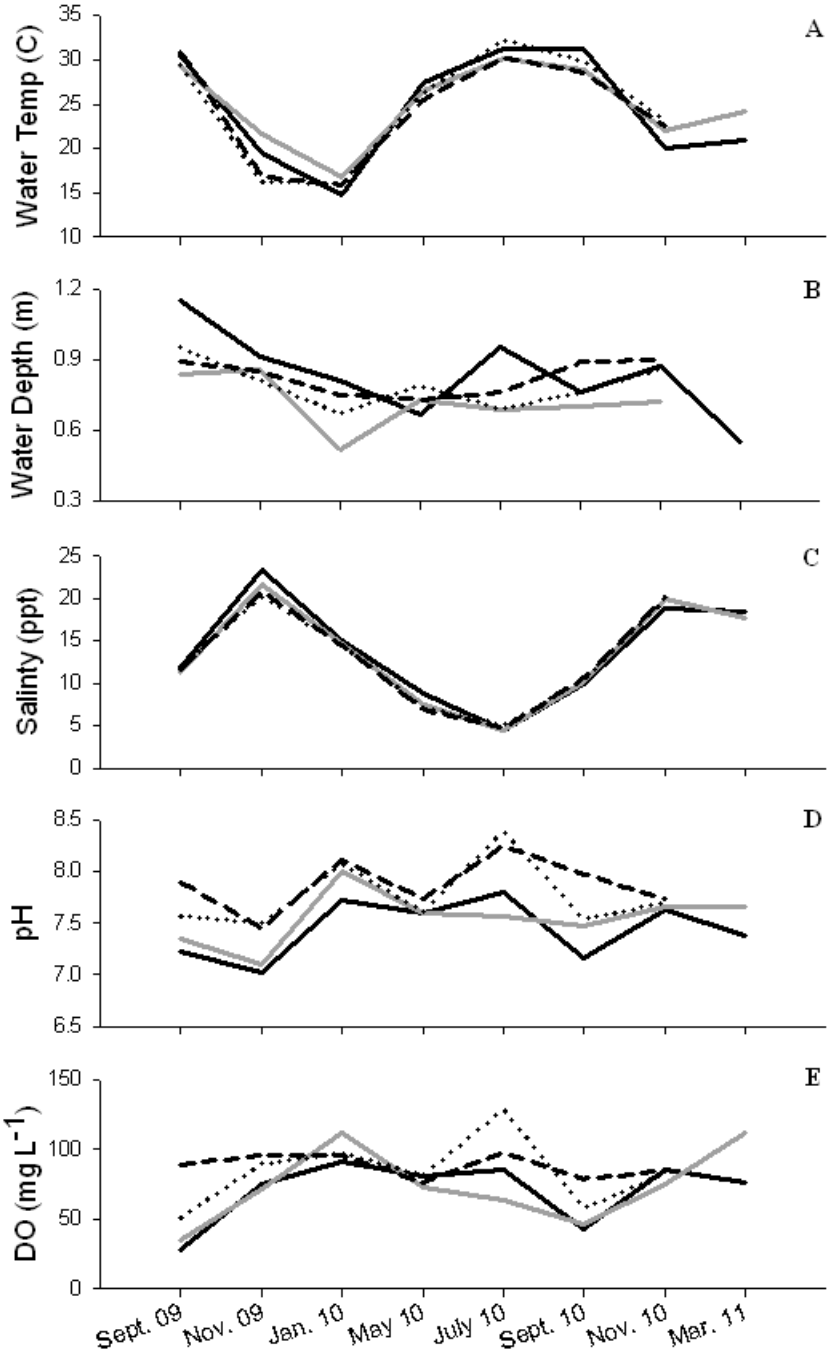

Figure 4.3. Water temp (°C; A), depth (m; B), salinity (ppt; C), pH (D) and dissolved oxygen ($mg \cdot L^{-1}$; E) profiles for each pond in each month; Big (black line), Ovary (gray line), Perfect (dotted line), and Triangle (dashed line).

DO ranged from 27.9 $mg \cdot L^{-1}$ (Sept. 2009) to 128.4 $mg \cdot L^{-1}$ (July 2010; Table 4.1).

4.3.2 Diet Composition

A total of 749 Atlantic croaker, 429 bay whiff, 370 sand seatrout, and 226 pinfish were collected, with the highest number of each species being collected at mud sites, except in pinfish, which were collected most from edge sites (Tables 4.2-4.3). Atlantic croaker were collected during the months of March, May and July; bay whiff, sand seatrout and pinfish were collected during the months of May, July, and September. All four fish species were not collected in sufficient numbers for statistical analysis in all months; only one pinfish was collected during July 2009, only one Atlantic croaker was collected during September 2009, and zero sand seatrout were collected during March 2011. Species accumulation curves indicated that sufficient numbers of stomachs were collected to achieve an asymptotic value on an S-curve (Ferry and Cailliet, 1996). Species accumulation curves indicated Atlantic croaker diets became asymptotic on 35 unique prey items, bay whiff diets on 23 unique prey items, sand seatrout diets on 26 unique prey items, and pinfish diets on 25 unique prey items (Figure 4.4). Forty-two (5.6%) Atlantic croaker, 34 (7.9%) bay whiff, 82 (22.2%) sand seatrout, and 31 (13.7%) pinfish stomachs contained no prey items.

Despite the diversity of abundant prey types, diets were generally dominated by only a few taxa, with a single prey type comprising as much as 80% of the total diet. Mysids, calanoids, and amphipods were important diet items by *%N* and fish prey and shrimps were important by *%V* for all four species (Figure 4.5). Opportunistic fishes (i.e., Atlantic croaker and pinfish) consumed a greater variety of prey items including both

pelagic and benthic forms. Diets of these fishes were evenly distributed with 5-6 different prey types having *%IRI* values between 9 and 33%. Insect larvae, copepods, amphipods

Table 4.2. Mean length (mm; ±SE), weight (g; ±SE), and the number of stomachs collected (including empty stomachs) from each habitat type for all four fish species.

Species	Habitat	N	Length (mm)	Weight (g)
Atlantic Croaker	Mud	519	51.8 (15.6)	2.68 (3.71)
	Edge	150	52.8 (14.3)	2.96 (2.42)
	MAOR	80	48.6 (13.5)	2.31 (1.85)
	Total	749		
Bay Whiff	Mud	305	48.6 (16.2)	1.61 (1.51)
	Edge	72	51.5 (12.9)	1.68 (1.10)
	MAOR	52	43.6 (15.9)	1.16 (1.25)
	Total	429		
Sand Seatrout	Mud	241	44.7 (17.2)	1.49 (1.44)
	Edge	95	43.2 (10.2)	1.41 (1.05)
	MAOR	34	36.5 (8.0)	0.76 (0.50)
	Total	370		
Pinfish	Mud	69	61.0 (15.0)	6.61 (3.87)
	Edge	111	50.8 (19.1)	4.84 (6.16)
	MAOR	46	45.0 (14.0)	2.92 (3.05)
	Total	226		

and polychaetes comprised the majority of Atlantic croaker diets. Pinfish diets were less evenly distributed with plant material (includes both living plant material and algae) comprising almost a third of the total diet (*%IRI*). More specialized predators (i.e., bay whiff and sand seatrout) consumed fewer prey types and diets were primarily dominated by pelagic prey. Bay whiff and sand seatrout diets were dominated by only 2-3 prey types

Table 4.3. Mean standard length (mm; ±SE), weight (g; ±SE) and the number of stomachs collected (including empty stomachs) during each month before and after MAOR addition.

		Before				After			
		March 09	May 09	July 09	Sept. 09	May 10	July 10	Sept 10	March 11
Atlantic Croaker	N	168	112	22	1	141	88	7	210
	SL (mm)	47.6 (10.9)	54.4 (9.9)	88.4 (22.5)	119.0	53.2 (6.5)	70.0 (6.9)	85.4 (8.2)	39.4 (7.6)
	Wt (g)	2.11 (1.4)	2.74 (1.4)	10.64 (17.1)	.	2.51 (0.9)	5.94 (2.0)	10.12 (3.8)	1.09 (0.7)
Bay Whiff	N	31	111	76	32	62	64	22	31
	SL (mm)	37.7 (10.9)	41.3 (10.4)	60.0 (7.7)	55.7 (18.4)	41.0 (8.4)	63.0 (11.6)	62.0 (11.6)	24.3 (3.4)
	Wt (g)	0.77 (0.8)	0.82 (0.6)	2.28 (0.9)	2.50 (1.9)	0.75 (0.5)	2.98 (1.5)	2.76 (1.8)	0.17 (0.1)
Sand Seatrout	N	6	80	108	47	35	57	37	0
	SL (mm)	35.0 (6.8)	37.2 (8.5)	45.7 (12.0)	57.1 (26.2)	42.8 (11.4)	37.2 (10.7)	45.6 (12.6)	0.0
	Wt (g)	0.79 (0.4)	0.83 (0.8)	1.69 (1.3)	2.05 (1.9)	1.41 (1.1)	0.98 (0.9)	1.80 (1.4)	0.00
Pinfish	N	10	11	1	6	57	70	32	39
	SL (mm)	32.2 (3.3)	53.3 (8.4)	73.0	98.5 (8.6)	44.4 (8.1)	62.9 (6.3)	71.5 (6.7)	28.6 (4.3)
	Wt (g)	0.80 (0.3)	4.18 (1.7)	11.23	30.58 (9.4)	2.31 (1.2)	6.97 (2.3)	9.25 (3.1)	0.52 (0.2)

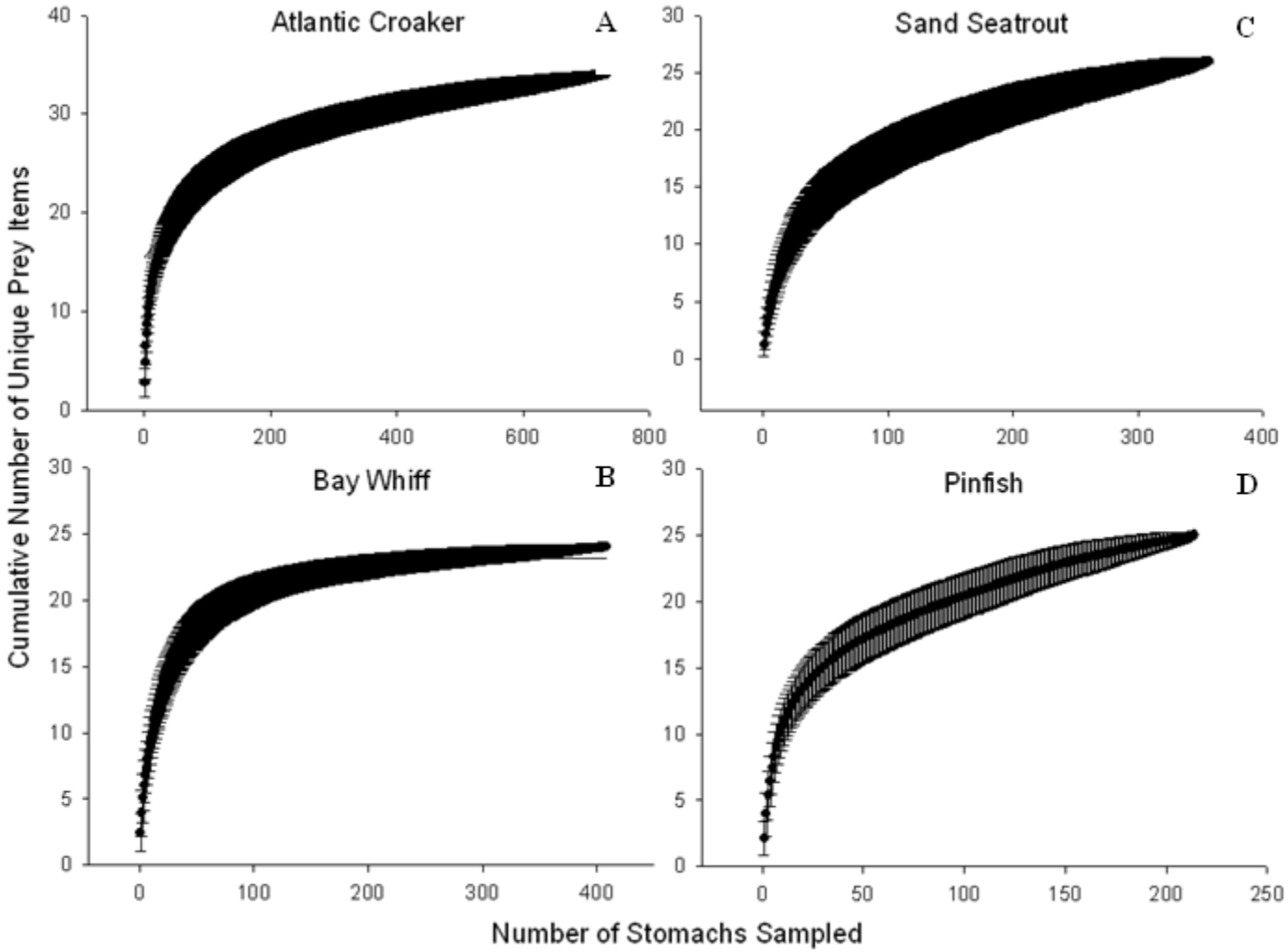

Figure 4.4. Prey accumulation curves for Atlantic croaker (A), bay whiff (B), sand seatrout (C), and pinfish (D; PRIMER; S-curve with standard error bars: n=999 permutations).

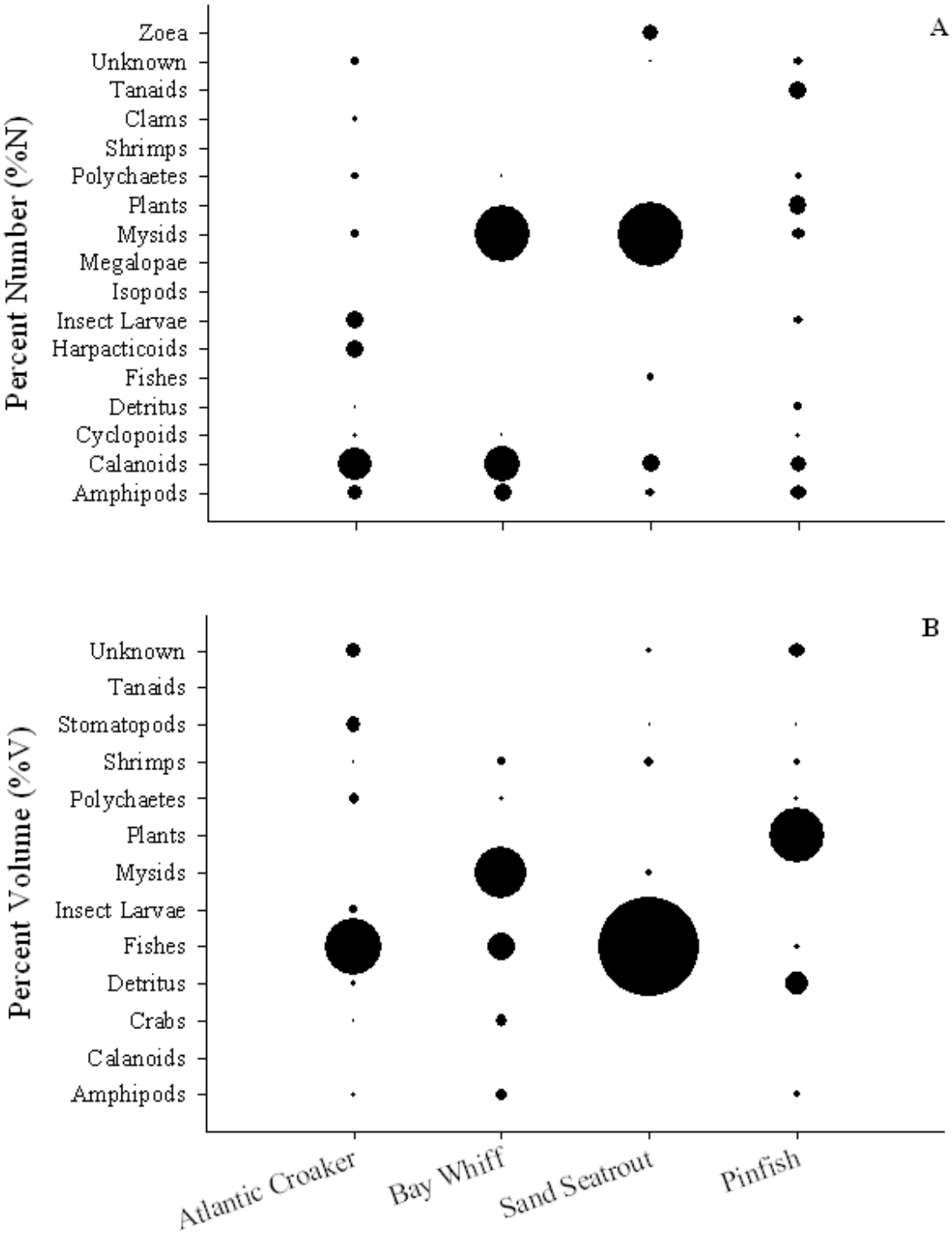

Figure 4.5. The contribution of each prey item by *%N* (A) and *%V* (B) to the overall diet of Atlantic croaker, bay whiff, sand seatrout, and pinfish.

with mysids comprising more than 70% of the total diet in both species *(%IRI*). Calanoid copepods comprised the second most important prey type in bay whiff diets while fish prey comprised the second most important prey type in sand seatrout diets (*%IRI*). Density estimates used to estimate electivity indices for each prey type are listed in appendices 2 and 3. Appendix 2 lists density and relative proportions of meiofauna in the environment (marsh ponds) collected using plankton tows. Appendix 3 lists the relative proportions of prey items in stomach contents from each habitat type. A detailed discussion of observed results for meiofauna collections is presented in Chapter 3.

- Atlantic Croaker

Of the three habitat types, the highest mean lengths, weights, and numbers of Atlantic croaker were collected from mud sites. Atlantic croaker from MAOR sites had the smallest mean length and weight (Tables 4.2 and 4.3), but lengths and weights were not significantly different between habitats ($p>0.05$; ANOVA). Overall, diets were dominated in number (*%N*) by calanoid copepods at 51.4% and in volume (*%V*) by fish prey at 44% (Table 4.4). Insect larvae, calanoid copepods, amphipods, polychaetes, and harpacticoid copepods comprised 84.2% of the total diet (*%IRI*). Neither of the interaction terms (i.e., Period*Location or Period*Site) that would indicate an affect of MAOR addition were significant ($p> 0.01$; PERMANOVA). However, significant monthly shifts were observed for the interaction terms Month*Pond and Month*Site ($p = 0.0001$). No prey type was consumed in all months but insect larvae and harpacticoid copepods were consumed in most months (Table 4.5).

Habitat-specific diets also indicated opportunistic feeding. Habitat-specific *%N* was similar to the overall diet, with the same six prey types dominating stomach contents. However, habitat-specific consumption of fish prey (*%V*) was much reduced at edge habitat compared with the other two habitat types (Figure 4.6). At edge habitat, polychaetes represented the greatest portion of the diet but no single prey type dominated the diet by *%V* (Figure 4.6). Diets at mud, edge, and MAOR sites were similar for *%IRI* values (Table 4.6). Despite similar mean densities, SIMPER analysis indicated insect larvae to be the largest contributor to cumulative dissimilarity between mud and MAOR habitats, and the second largest contributor to cumulative dissimilarity between edge and MOAR habitats (Table 4.7). Of the nine prey types that contributed to >80% cumulative dissimilarity between habitat types, only fish prey showed increased consumption by *%V* in Atlantic croaker stomachs at MAOR sites. All other prey types decreased in consumption by *%V* at MAOR sites compared to mud and edge sites (Figure 4.6). Comparisons amongs diets realized "before" and "after" at MAOR sites followed similar trends, with all prey types decreasing during the "after" period, except for fish prey and detritus (Table 4.8). Estimates of habitat-specific prey quality (*%ICI*) indicated amphipods, calanoid copepods, harpacticoid copepods, and insect larvae were consistently the most energetically valuable prey types in Atlantic croaker diets across habitats (Table 4.9).

Electivity indices indicated Atlantic croaker fully selected for (E=1.0) plant material and stomatopods, strongly selected for (>0.3) insect larvae and zoea, strongly avoided (E< -0.3) gastropods and egg masses, and fully avoided (E=-1.0) anthomedusae (Table 4.10). Oher prey types were consumed in proportion to their density in the

environment (0.3>E>-0.3). Habitat-specific electivity indices indicated branchiurans, cyclopoids, insect larvae, mysids, and zoea were strongly selected for by Atlantic croaker at MAOR sites.

Table 4.4. The relative importance of prey categories as percent number (*%N*), percent volume (*%V*), percent occurrence (*%FO*), and percent index of relative importance (*%IRI*) for all four fish species. The number of stomachs sampled that contained prey items is listed in parentheses after each fish common name.

Predator	%N	%V	%FO	%IRI
Atlantic Croaker (n=707) *Micropogonias undulatus*				
Insect Larvae	8.1	6.7	40.8	19.7
Calanoids	51.4	0.6	33.7	15.7
Amphipods	5.8	3.5	31.8	14.1
Unknown	3.4	11.0	27.8	13.5
Polychaetes	2.9	8.1	28.3	11.7
Harpacticoids	7.9	0.1	39.4	9.5
Mysids	4.2	1.2	13.5	5.2
Detritus	1.3	4.4	14.9	2.9
Clams	3.5	1.3	7.1	2.3
Fishes	0.5	44.2	6.5	2.0
Cyclopoids	2.6	0.0	13.2	0.7
Tanaids	0.7	0.3	6.1	0.6
Shrimps	0.2	2.0	2.4	0.6
Zoea	5.8	0.2	2.5	0.5
Stomatopods	0.0	11.6	0.6	0.4
Plants	0.2	0.2	1.7	0.2
Mussels	0.1	0.0	1.8	0.2
Sediment	0.2	0.5	1.7	0.1
Crabs	0.1	3.0	1.0	0.1
Isopods	0.2	0.5	2.1	0.1
Branchiurans	0.0	0.2	0.7	0.0
Nematodes	0.8	0.0	2.2	0.0
Insects	0.0	0.1	0.7	0.0
Eggs	0.0	0.2	0.1	0.0
Nemerteans	0.0	0.0	0.1	0.0
Ostracods	0.0	0.0	0.4	0.0
Gastropods	0.0	0.0	0.1	0.0

Table 4.4. cont.

Bay Whiff (n=395) *Spilopterus citharichthys*	%N	%V	%FO	%IRI
Mysids	36.7	48.3	79.0	73.8
Calanoids	40.0	3.7	43.3	13.7
Amphipods	11.4	11.5	32.2	8.0
Polychaetes	1.2	3.4	17.5	1.9
Crabs	0.7	8.5	13.2	0.8
Insect Larvae	1.0	1.5	11.9	0.6
Cyclopoids	5.9	0.2	10.6	0.4
Shrimps	0.5	4.9	7.5	0.4
Fishes	0.3	16.4	6.3	0.2
Unknown	0.3	0.7	5.8	0.1
Detritus	0.2	0.4	2.8	0.1
Harpacticoids	0.5	0.0	7.2	0.0
Zoea	1.1	0.1	3.8	0.0
Tanaids	0.1	0.1	2.8	0.0
Isopods	0.1	0.5	1.5	0.0
Clams	0.0	0.0	0.3	0.0

White Trout (n=288) *Cynoscion arenarius*	%N	%V	%FO	%IRI
Mysids	60.2	10.0	58.5	79.5
Fishes	3.0	75.0	24.2	10.6
Zoea	19.0	0.3	13.8	3.8
Calanoids	10.5	0.1	11.4	2.1
Unknown	0.9	4.2	8.0	1.3
Amphipods	3.6	0.6	13.5	1.2
Detritus	0.5	0.2	5.2	0.5
Shrimps	0.6	7.1	5.9	0.5
Crabs	0.8	0.7	6.1	0.4
Tanaids	0.4	0.1	3.8	0.2
Polychaetes	0.2	0.1	0.7	0.0
Stomatopods	0.0	1.6	0.3	0.0
Insect Larvae	0.1	0.0	1.0	0.0
Harpacticoids	0.1	0.0	0.7	0.0
Cyclopoids	0.1	0.0	0.7	0.0
Clams	0.0	0.0	0.3	0.0

Table 4.4. cont.

Pinfish (n=195) *Lagodon rhomboides*	%N	%V	%FO	%IRI
Plants	7.1	43.4	31.8	32.4
Unknown	3.7	11.6	28.2	17.9
Tanaids	7.5	1.5	31.8	12.8
Amphipods	6.9	6.9	24.6	10.4
Detritus	3.0	17.8	20.5	9.4
Polychaetes	2.2	3.7	16.4	4.7
Mysids	7.1	1.1	15.9	4.4
Insect Larvae	3.7	0.6	24.6	4.2
Calanoids	49.6	0.2	13.8	2.6
Cyclopoids	6.7	0.0	7.7	0.4
Isopods	0.9	0.5	5.1	0.3
Fishes	0.2	4.2	2.6	0.2
Harpacticoids	0.5	0.0	4.1	0.1
Branchiurans	0.2	0.7	2.1	0.0
Sediment	0.2	0.1	1.0	0.0
Shrimps	0.1	4.7	1.5	0.0
Clams	0.3	0.1	1.0	0.0
Stomatopods	0.0	2.8	0.5	0.0
Insects	0.0	0.0	0.5	0.0
Ostracods	0.0	0.0	0.5	0.0
Crabs	0.0	0.0	0.5	0.0

- Bay Whiff

Bay whiffs having the highest mean lengths and weights were collected from edge sites but the highest numbers of bay whiffs were collected from mud sites (Tables 4.2 and 4.3). Mean lengths and weights of bay whiffs were significantly smaller at MAOR sites ($p<0.05$) but were not significantly different between mud and edge habitats ($p>0.05$; Tukey adjusted LSmeans). Neither of the habitat-effect interaction terms (i.e., Period*Location or Period*Site) were significant nor were there any significant interaction terms that would indicate monthly diet shifts (i.e., Month*Locaiton or Month*Site; $p> 0.01$; PERMANOVA). Overall, diets were limited in diversity with mysids, calanoids, and amphipods comprising over 95% of the diet by *%IRI* (Table 4.4).

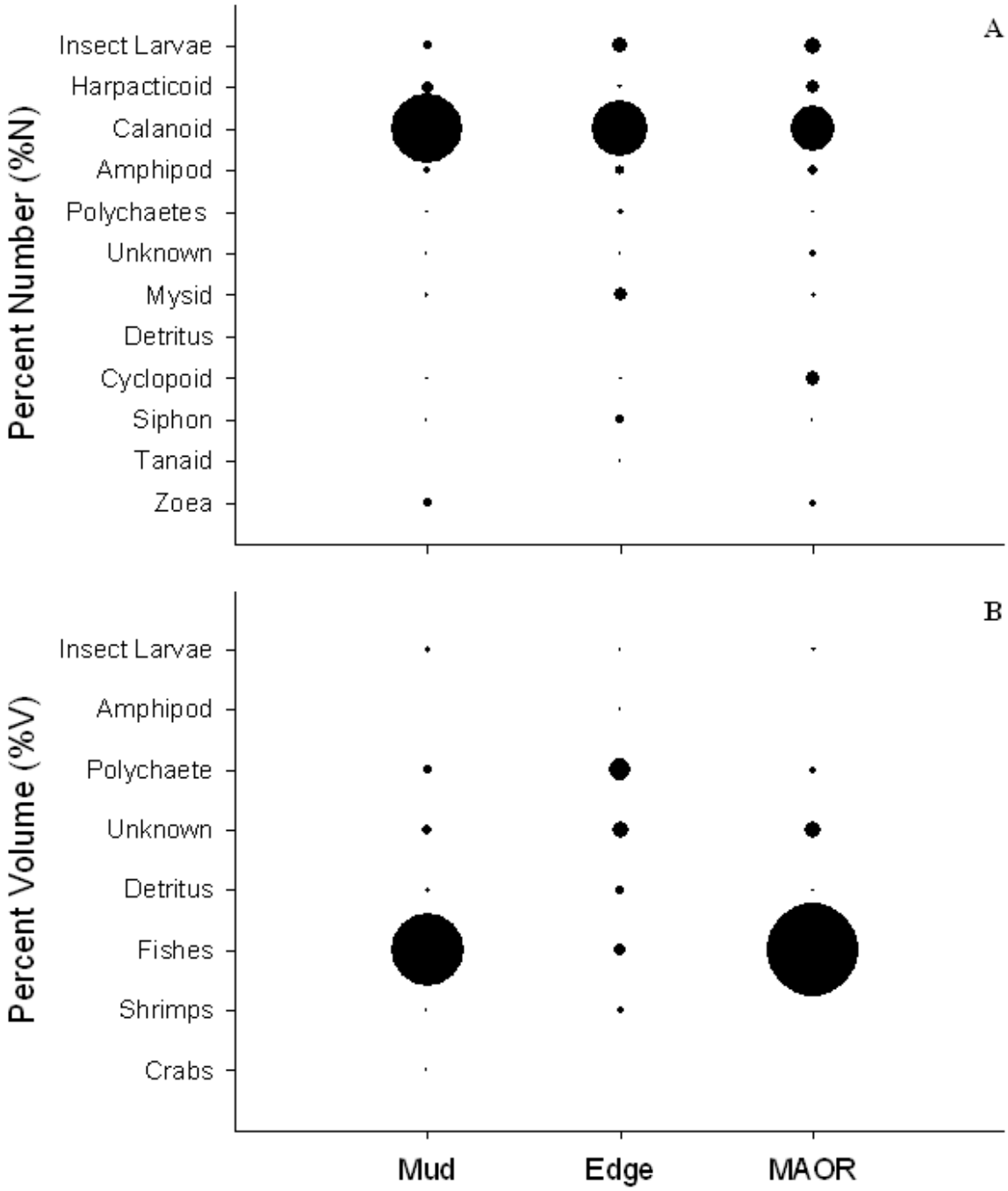

Figure 4.6. Percent number (A) and volume (B) contributions of important prey items to the diet of Atlantic croaker from each habitat type. Prey items are listed in order of their *%IRI* ranking from highest to lowest. Prey items with low percent values (≤ 1) were removed.

Table 4.5. The percent index of relative importance (*%IRI*) for each diet item found in Atlantic croaker stomachs. Percent IRI values listed for each month before (B) and after (A) MAOR addition.

Atlantic Croaker	March (B)	May (B)	July (B)	Sept. (B)	March (A)	May (A)	July (A)	Sept. (A)
Amphipods	13.6	0.0	10.3	--	32.6	3.7	0.0	--
Branchiurans	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Calanoids	22.2	72.8	1.0	--	0.0	70.6	0.4	--
Cyclopoids	1.1	0.8	0.0	--	0.0	2.1	0.0	--
Detritus	1.1	1.3	3.5	--	1.7	3.1	3.6	--
Eggs	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Gastropods	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Harpacticoids	16.2	5.2	1.2	--	12.2	2.2	1.7	--
Insect Larvae	38.7	0.3	0.0	--	36.6	3.2	26.7	--
Isopods	0.3	0.0	0.1	--	0.0	0.0	0.0	--
Mussels	0.0	0.0	0.0	--	0.2	0.0	0.0	--
Mysids	0.0	0.3	32.8	--	0.0	0.1	55.2	--
Nematodes	0.5	0.0	0.0	--	0.0	0.0	0.0	--
Nemerteans	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Oligochaetes	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Ostracods	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Plants	0.0	0.0	0.1	--	0.0	0.0	1.4	--
Sediment	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Clams	0.0	17.4	0.0	--	0.0	4.8	5.3	--
Stomatopods	0.0	0.0	0.0	--	0.0	0.0	0.0	--
Tanaids	0.0	0.0	0.0	--	0.3	0.4	0.2	--
Insects	0.3	0.0	0.0	--	0.2	0.5	0.0	--
Unknown	0.1	0.3	0.7	--	8.5	3.3	4.8	--
Zoea	0.0	0.3	0.0	--	0.0	4.2	0.0	--
Polychaetes	2.9	0.8	9.8	--	6.9	0.9	0.4	--
Crabs	0.3	0.1	40.1	--	0.0	0.5	0.0	--
Fishes	2.4	0.1	0.4	--	0.8	0.1	0.3	--
Shrimps	0.3	0.0	0.0	--	0.0	0.0	0.0	--

Table 4.6. Percent index of relative importance (*%IRI*) values for each diet item found in Atlantic croaker stomachs found in each habitat type.

Atlantic Croaker	Mud	Edge	MAOR
Amphipods	5.87	9.49	9.97
Branchiurans	0.00	0.01	0.16
Calanoids	52.48	36.17	32.72
Cyclopoids	0.81	0.66	4.66
Detritus	2.18	4.47	1.29
Eggs	0.00	0.04	0.00
Gastropods	0.00	0.00	0.00
Harpacticoids	10.72	2.90	12.99
Insect Larvae	12.21	21.34	24.22
Isopods	0.02	0.29	0.01
Mussels	0.01	0.00	0.05
Mysids	1.35	5.65	2.42
Nematodes	0.08	0.00	0.00
Nemerteans	0.00	0.00	0.00
Ostracods	0.00	0.00	0.00
Plants	0.01	0.09	0.01
Sediment	0.01	0.09	0.00
Clams	3.37	7.30	0.72
Stomatopods	0.00	0.02	0.00
Tanaids	0.05	0.61	0.66
Insects	0.28	0.00	0.60
Unknown	2.59	1.51	6.54
Zoea	0.96	2.21	1.25
Polychaetes	3.64	5.98	1.09
Crabs	2.66	0.21	0.00
Fishes	0.65	0.90	0.64
Shrimps	0.03	0.05	0.01

Table 4.7. SIMPER output of the items that explain >80% of the dissimilarity in Atlantic croaker diets in each habitat type (mud, unvegetated edge, and MAOR). Data were square root transformed.

Diet Item	Volume (mm^3)		Average Dissimilarity	% Contribution to Dissimilarity	% Cumulative Contribution
	Mud	MAOR			
Insect Larvae	0.57	0.57	13.26	15.68	34.48
Polychaetes	0.54	0.30	9.95	11.77	46.25
Amphipods	0.43	0.35	9.45	11.18	57.43
Fishes	0.50	0.71	7.22	8.54	65.97
Detritus	0.27	0.18	4.64	5.49	71.46
Calanoids	0.18	0.09	4.43	5.24	76.71
Mysids	0.15	0.12	4.36	5.15	81.86
Average dissimilarity = 84.56					
	Edge	MAOR			
Unknown	0.46	0.83	13.95	16.07	16.07
Insect Larvae	0.59	0.57	13.58	15.65	31.72
Polychaetes	0.79	0.30	12.48	14.37	46.09
Amphipods	0.41	0.35	9.06	10.43	56.52
Mysids	0.21	0.12	5.53	6.38	62.90
Fishes	0.15	0.71	5.53	6.37	69.27
Detritus	0.30	0.18	5.33	6.14	75.41
Calanoids	0.13	0.09	3.86	4.45	79.85
Clams	0.14	0.11	3.32	3.83	83.68
Average dissimilarity = 86.80					
	Mud	Edge			
Polychaetes	0.54	0.79	13.81	16.09	16.09
Insect Larvae	0.57	0.59	12.80	14.91	31.00
Unknown	0.67	0.46	12.12	14.12	45.12
Amphipods	0.43	0.41	9.59	11.17	56.29
Detritus	0.27	0.30	5.84	6.81	63.10
Mysids	0.15	0.21	5.60	6.52	69.62
Calanoids	0.18	0.13	4.79	5.58	75.19
Fishes	0.50	0.15	4.04	4.71	79.90
Clams	0.13	0.14	3.83	4.46	84.37
Average dissimilarity = 85.8					

Table 4.8. SIMPER output of the items that explain >90% (>80% for Atlantic croaker) of the dissimilarity in predator diets at impact locations before and after MAOR addition. Data were square root transformed.

Diet Item	Volume (mm^3)		Average Dissimilarity	% Contribution to Dissimilarity	% Cumulative Contribution
	MAOR (B)	MAOR (A)			
			Atlantic Croaker		
Insect Larvae	1.02	0.57	17.21	20.23	20.23
Unknown	0.13	0.83	11.85	13.93	34.16
Amphipods	0.49	0.35	11.09	13.04	47.20
Polychaetes	0.47	0.30	8.06	9.48	56.68
Siphons	0.42	0.11	7.91	9.30	65.97
Fishes	0.11	0.71	5.62	6.61	72.59
Calanoids	0.25	0.09	4.83	5.68	78.27
Detritus	0.16	0.18	3.98	4.68	82.95
Average dissimilarity = 85.05					
			Bay Whiff		
Mysids	2.12	1.07	23.81	31.04	31.04
Amphipods	0.61	0.52	11.44	14.91	45.96
Calanoids	0.65	0.10	9.29	12.11	58.07
Insect Larvae	0.07	0.64	7.96	10.38	68.45
Polychaetes	0.12	0.52	7.53	9.82	78.27
Crabs	0.57	0.00	5.22	6.81	85.08
Fishes	0.07	0.24	3.02	3.93	89.01
Unknown	0.06	0.14	2.24	2.92	91.94
Average dissimilarity = 76.70					
			Sand Seatrout		
Mysids	0.30	1.42	38.97	46.68	46.68
Fishes	1.18	1.25	14.14	16.94	63.62
Zoea	0.30	0.12	11.96	14.32	77.95
Calanoids	0.18	0.02	6.55	7.85	85.79
Unknown	0.28	0.00	4.72	5.66	91.45
Average dissimilarity = 83.48					
			Pinfish		
Detritus	3.79	0.18	30.45	34.06	34.06
Plants	0.00	4.63	27.07	30.28	64.34
Unknown	1.92	1.23	14.60	16.33	80.67
Mysids	0.98	0.58	7.20	8.05	88.72
Crabs	0.62	0.00	3.81	4.26	92.98
Average dissimilarity = 89.40					

Table 4.9. Energy density (ED; $J \cdot g^{-1}$) estimates used to calculate percent index of caloric importance (*%ICI*) values for important prey types (prey types < 0.1% *%IRI* were excluded) for each fish species by habitat type.

Atlantic Croaker	ED ($J \cdot g^{-1}$)	Mud	Edge	MAOR
Amphipods	15208.8	12.2	12.5	14.1
Calanoids	27723.2	18.5	15.5	15.4
Cyclopoids	27723.2	4.0	2.8	3.8
Fishes	20978.6	2.1	1.2	3.0
Harpacticoids	27723.2	22.1	15.5	19.6
Insect Larvae	20662.0	22.9	28.0	25.5
Mussels	20049.7	0.7	0.3	1.5
Mysids	31518.1	5.2	5.4	5.3
Plants	16233.9	0.6	2.0	0.7
Polychaetes	16731.8	6.3	9.4	3.5
Shrimps	19869.8	1.0	0.9	0.5
Clams	20049.7	2.1	1.7	1.5
Stomatopods	22551.8	0.2	0.7	0.0
Tanaids	12405.6	1.3	3.0	4.1
Zoea	14690.9	0.8	1.2	1.5
Bay Whiff	ED ($J \cdot g^{-1}$)	Mud	Edge	MAOR
Amphipods	15208.8	16.3	22.0	23.9
Calanoids	27723.2	23.5	16.7	15.3
Crabs	12321.9	2.9	2.9	0.0
Cyclopoids	27723.2	3.5	1.9	7.6
Fishes	20978.6	1.5	1.2	1.3
Insect Larvae	20662.0	3.2	1.3	14.2
Mysids	31518.1	40.7	43.1	26.7
Polychaetes	16731.8	5.3	7.0	10.3
Shrimps	19869.8	3.0	4.0	0.7
Sand Seatrout	ED ($J \cdot g^{-1}$)	Mud	Edge	MAOR
Amphipods	15208.8	18.4	15.5	0.0
Calanoids	27723.2	11.4	5.2	3.4
Crabs	12321.9	0.6	1.8	0.0
Fishes	20978.6	27.0	20.5	22.5
Mysids	31518.1	30.5	42.3	56.8
Shrimps	19869.8	3.5	8.6	3.2
Tanaids	12405.6	1.7	1.5	4.0
Zoea	14690.9	7.0	4.6	10.1
Pinfish	ED ($J \cdot g^{-1}$)	Mud	Edge	MAOR
Amphipods	15208.8	0.9	13.3	10.3
Calanoids	27723.2	0.0	2.6	7.0
Cyclopoids	27723.2	0.0	3.7	2.1
Fishes	20978.6	0.0	1.2	4.2
Insect Larvae	20662.0	13.5	23.9	22.2
Isopods	13765.4	0.0	2.5	0.0
Mysids	31518.1	32.8	2.5	7.8
Plants	16233.9	47.7	19.3	11.7
Polychaetes	16731.8	3.0	6.3	12.6
Tanaids	12405.6	2.3	24.8	22.1

Table 4.10. Overall diet and habitat-specific electivity indices (using Ivlev's electivity index) for diet items found in Atlantic croaker and bay whiff stomachs. Prey items listed represented ≥1.0% of the diet by *%IRI* indices.

Atlantic Croaker	Rank						**Bay Whiff**	Rank					
Organism	*p*	*r*	Overall	Mud	Edge	MAOR	Organism	*p*	*r*	Overall	Mud	Edge	MAOR
Amphipods	4	4	0.009	-0.018	-0.033	0.140	Amphipods	4	3	0.245	0.209	0.279	0.371
Anthomedusae	19	24	-1.000	-1.000	-1.000	-1.000	Anthomedusae	19	24	-1.000	-1.000	-1.000	-1.000
Bivalves	17	7	0.270	0.782	1.000	0.100	Bivalves	17	14	-0.366	0.491	--	-1.000
Branchiurans	22	18	0.068	-0.197	0.035	0.631	Branchiurans	22	24	-1.000	-1.000	-1.000	-1.000
Calanoids	3	1	0.130	0.111	0.150	0.293	Calanoids	3	1	0.301	0.278	0.358	0.271
Crabs	16	16	-0.134	-0.164	-0.610	-1.000	Crabs	16	8	0.272	0.227	0.196	-1.000
Cyclopoids	7	9	0.020	-0.032	0.045	0.362	Cyclopoids	7	4	0.271	0.181	0.095	0.590
Egg Mass	9	21	-0.734	--	0.790	--	Egg Mass	9	24	-1.000	--	-1.000	--
Fishes	14	12	0.046	-0.024	-0.066	0.126	Fishes	14	11	0.132	0.059	-0.016	0.046
Gastropods	10	22	-0.734	-0.708	-1.000	-1.000	Gastropods	10	24	-1.000	-1.000	-1.000	-1.000
Harpacticoids	2	3	-0.099	-0.123	-0.249	-0.046	Harpacticoids	2	10	-0.164	-0.200	-0.721	-0.259
Insects	21	17	-0.007	-0.042	-1.000	0.001	Insects	21	15	-1.000	-1.000	-0.085	-1.000
Insect Larvae	18	2	0.333	0.336	0.650	0.443	Insect Larvae	18	6	0.345	0.279	0.216	0.522
Isopods	8	14	-0.285	-0.388	-0.325	-0.461	Isopods	8	13	-0.194	-0.529	-0.104	-0.333
Mysids	11	6	0.188	0.093	0.668	0.601	Mysids	11	2	0.485	0.433	0.817	0.788
Nematodes	1	10	-0.365	-0.396	-0.863	-1.000	Nematodes	1	16	-1.000	-0.824	-1.000	-1.000
Ostracods	12	20	-0.495	-0.228	0.510	-1.000	Ostracods	12	24	-1.000	-1.000	-1.000	-1.000
Plants	24	15	1.000	1.000	1.000	1.000	Plants	25	24	--	--	--	--
Polychaetes	6	8	-0.008	-0.029	-0.052	0.003	Polychaetes	6	5	0.103	0.062	0.013	0.232
Shrimps	15	13	-0.060	-0.135	-0.116	-0.486	Shrimps	15	9	0.218	0.165	0.136	-0.361
Stomatopods	24	19	1.000	1.000	1.000	--	Stomatopods	25	24	--	--	--	--
Tanaids	5	11	-0.172	-0.270	-0.199	-0.265	Tanaids	5	12	-0.215	-0.463	-0.203	-0.402
Zoea	20	5	0.466	0.443	0.376	0.659	Zoea	20	7	0.497	0.463	0.393	0.622

Mysids, calanoids, and amphipods dominated the diet by *%N* and mysids and fish prey dominated the diet by *%V* (Figure 4.5). Despite a low diversity diet, bay whiff did not consume prey items in similar proportions in each month (Table 4.11). For example, *%IRI* values for mysids, the most important prey type in bay whiff diets, ranged from 1.3 - 94.9%.

Habitat-specific comparisons also indicated bay whiff diets were dominated by only a few prey types: mysids, polychaetes, insect larvae, fishes, crabs, calanoids, and amphipods, with mysids dominating the diet by *%N* and *%V* in all habitat types (Figure 4.7). In habitat-specific comparisons of *%IRI* values, mysids, calanoids, and amphipods comprised the majority of diets in mud and edge habitats, while amphipods, cyclopoids, insect larvae, and polychaetes comprised more of the diet at MAOR sites than in the other habitat types (Table 4.12). SIMPER analysis indicated mysids were the largest contributor to cumulative dissimlarity between habitat types, except between mud and MAOR where amphipods were the largest contributor to cumulative dissimilarity (Table 4.13). Comparisons among diets realized "before" and "after" at MAOR sites showed consumption of polychaetes, insect larvae, and fish prey increased at MAOR sites during the "after" period (Table 4.8). Estimates of habitat-specific prey quality (*%ICI*) indicated amphipods, calanoid copepods, and mysids contributed most to nutritional intake in bay whiff diets at all habitat types, with mysids contributing almost half the energetic intake at mud and edge sites (Table 4.12). The nutritional importance of mysids decreased at MAOR sites while insect larvae and polychaetes increased in value, with insect larvae and polychaetes each contributing >10% of the nutritional intake at MAOR sites.

Table 4.11. The percent index of relative importance (*%IRI*) for each diet item found in bay whiff stomachs. Percent *IRI* values listed for each month before (B) and after (A) MAOR addition.

Bay Whiff	March (B)	May (B)	July (B)	Sept. (B)	March (A)	May (A)	July (A)	Sept. (A)
Amphipods	49.1	0.2	0.2	26.3	46.8	29.2	0.0	0.0
Calanoids	27.5	66.5	0.2	3.0	0.0	17.6	0.0	0.0
Cyclopoids	0.3	0.0	0.0	0.0	3.3	11.3	0.0	0.0
Detritus	0.1	0.0	0.0	0.0	0.0	0.0	0.0	55.8
Harpacticoids	0.2	0.1	0.0	0.0	0.1	0.1	0.0	0.0
Insect Larvae	0.4	0.0	0.0	0.0	3.2	7.5	0.0	0.0
Isopods	0.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Mysids	1.3	25.3	80.6	63.7	2.9	19.1	94.9	15.1
Nematodes	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Clams	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Tanaids	0.0	0.0	0.0	0.1	0.0	0.3	0.0	0.0
Insects	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Unknown	0.0	0.0	0.0	0.0	0.0	4.0	0.0	0.0
Zoea	0.0	0.0	0.0	0.0	0.0	1.5	0.0	0.0
Polychaetes	6.2	0.7	0.0	0.0	43.7	8.4	0.0	0.0
Crabs	0.0	2.9	15.0	0.7	0.0	0.0	0.2	16.9
Fishes	0.6	4.3	1.6	6.2	0.0	0.5	2.8	0.0
Shrimps	13.5	0.0	2.3	0.0	0.0	0.3	2.2	12.3

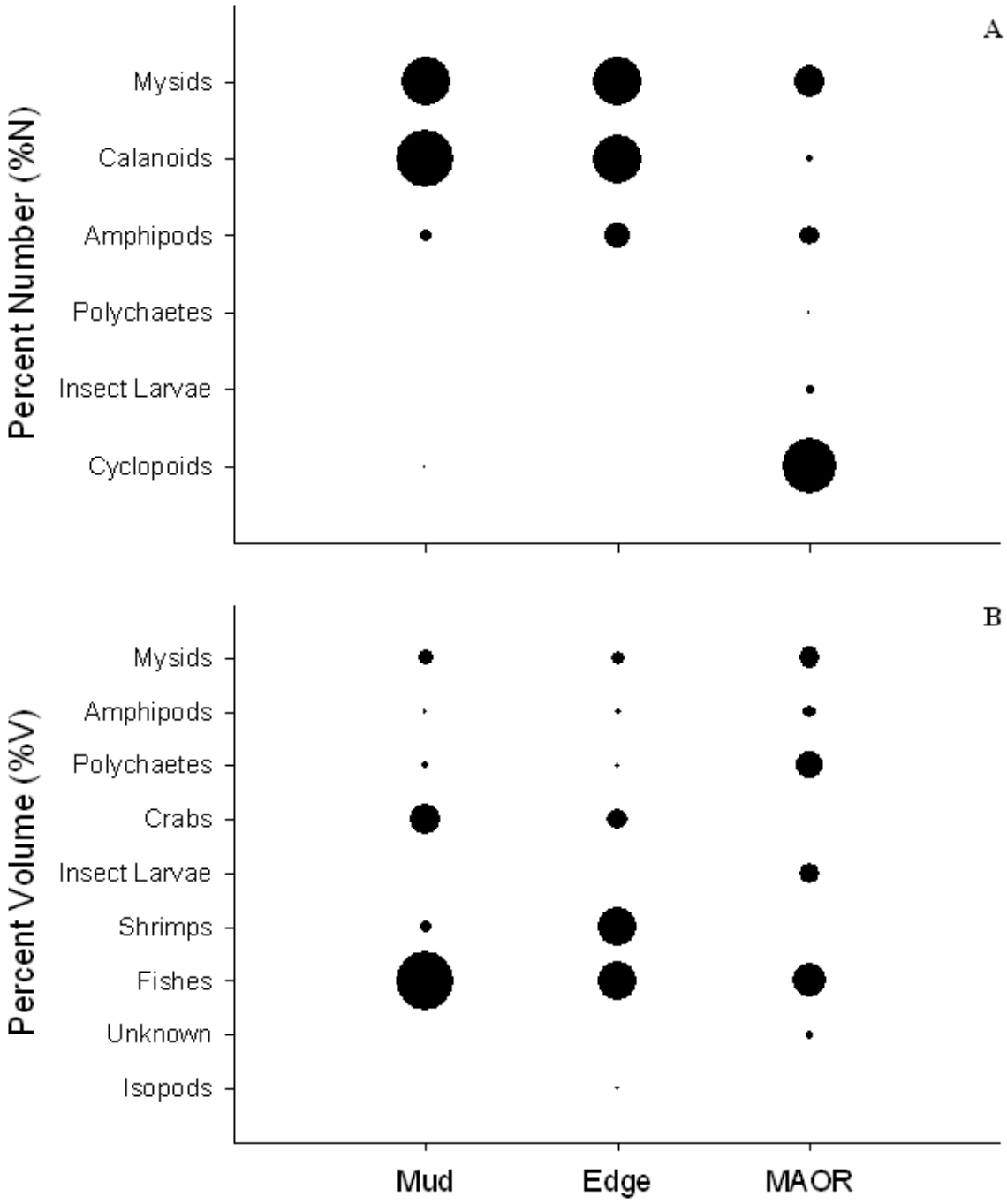

Figure 4.7. Percent number (A) and volume (B) contributions of important prey items to the diet of bay whiff from each habitat type. Prey items are listed in order of their *%IRI* ranking from highest to lowest. Prey items with low percent values (≤ 1) were removed.

Table 4.12. Percent index of relative importance (*%IRI*) values for each diet item found in bay whiff stomachs in each habitat type.

Bay Whiff	Mud	Edge	MAOR
Amphipods	4.93	12.47	16.19
Calanoids	28.38	18.00	2.98
Cyclopoids	0.38	0.06	16.59
Detritus	0.02	0.11	0.06
Harpacticoids	0.08	0.00	0.12
Insect Larvae	0.20	0.01	14.18
Isopods	0.00	0.28	0.02
Mysids	54.52	56.52	35.11
Nematodes	0.00	0.00	0.00
Clams	0.00	0.00	0.00
Tanaids	0.00	0.16	0.07
Insects	0.00	0.00	0.00
Unknown	0.15	0.01	1.25
Zoea	0.06	0.02	0.11
Polychaetes	1.26	1.38	10.92
Crabs	4.58	4.48	0.00
Fishes	4.19	2.11	2.37
Shrimps	1.25	4.39	0.03

Electivity indices indicated bay whiff strongly selected for (E>0.3) calanoids, insect larvae, mysids, and zoea; strongly selected against (E< -0.3) bivalves; and fully avoided (E=-1.0) anthomedusae, branchiurans, egg masses, gastropods, insects, nematodes, and ostracods (Table 4.10). Other prey types were consumed in proportion to their density in the environment (0.3>E>-0.3). Habitat-specific electivity indices indicated amphipods, cyclopoids, insect larvae, mysids, and zoea were strongly selected for by bay whiffs at MAOR sites.

- Sand Seatrout

The highest numbers, mean lenghts, and mean weights of sand seatrout were collected from mud sites (Tables 4.2 and 4.3). Mean lengths and weights of sand seatrout

Table 4.13. SIMPER output of the items that explain >90% of the dissimilarity of bay whiff diets in each habitat type (mud, unvegetated edge, and MAOR). Data were square root transformed.

Diet Item	Volume (mm^3)		Average Dissimilarity	% Contribution to Dissimilarity	% Cumulative Contribution
	Mud	MAOR			
Amphipods	0.51	0.52	10.17	13.50	45.85
Polychaetes	0.21	0.52	8.40	11.14	56.99
Insect Larvae	0.06	0.64	8.07	10.71	67.71
Calanoids	0.45	0.10	6.81	9.03	76.74
Fishes	0.34	0.24	4.41	5.85	82.59
Crabs	0.42	0.00	3.82	5.06	87.66
Unknown	0.07	0.14	2.38	3.16	90.82
Average dissimilarity = 75.35					
	Edge	MAOR			
Mysids	2.01	1.07	22.90	30.32	30.32
Amphipods	0.93	0.52	12.91	17.09	47.41
Polychaetes	0.26	0.52	8.50	11.26	58.67
Insect Larvae	0.02	0.64	7.63	10.10	68.77
Fishes	0.26	0.24	4.22	5.58	74.35
Calanoids	0.26	0.10	4.07	5.39	79.74
Crabs	0.41	0.00	4.05	5.37	85.10
Shrimps	0.40	0.02	3.75	4.97	90.07
Average dissimilarity = 75.53					
	Mud	Edge			
Mysids	2.21	2.01	22.43	33.12	33.12
Amphipods	0.51	0.93	12.42	18.34	51.45
Calanoids	0.45	0.26	7.15	10.56	62.01
Crabs	0.42	0.41	6.57	9.70	71.71
Polychaetes	0.21	0.26	5.20	7.67	79.38
Shrimps	0.17	0.40	4.73	6.99	86.37
Fishes	0.34	0.26	3.70	5.47	91.84
Average dissimilarity = 67.73					

were significantly smaller at MAOR sites ($p<0.05$) but were not significantly different between mud and edge sites ($p>0.05$; Tukey-adjusted LSmeans). Neither of the habitat-effect interaction terms (i.e., Period*Location or Period*Site) were significantly different nor were there any significant interaction terms that would indicate monthly diet shifts (i.e., Month*Location or Month*Site; $p> 0.01$; PERMANOVA). Overall, sand seatrout diets showed low diversity with mysids comprising almost 80% of the diet by *%IRI* (Table 4.4). Mysids dominated the prey consumed by *%N* and fish prey by *%V* (Figure 4.5). Mysids and fishes were consistently consumed in all months with mysids comprising between 62-81% of the total diet by *%IRI* during the months of July and September (Table 4.14).

Habitat-specific comparisons indicated the majority of sand seatrout diets were comprised of only seven prey types with mysids dominating diets by *%N* at all habitat types (Figure 4.8). Diets were least diverse at MAOR sites, but mysids and fish prey represented the majority of diet items in all habitat types by *%IRI* (Table 4.15). SIMPER analysis indicated mysids, and fish prey were the top two contributors to dissimilarity between all habitat types (Table 4.16), but mysid consumption increased and fish prey consumption decreased at MAOR sites (Table 4.15). Comparisons among diets realized "before" and "after" at MAOR sites indicated consumption of mysids and fish prey increased while consumption of zoea and calanoids decreased during the "after" period (Table 4.8). Estimates of habitat-specific prey quality (*%ICI*) were inconsistent across habitat types. Although amphipods, calanoids, mysids, and fish prey were major nutritional components of overall sand seatrout diets, mysids, fishes, and zoea were the predominant nutritional components of sand seatrout diets at MAOR sites (Table 4.9).

Table 4.14. The percent index of relative importance (*%IRI*) for each diet item found in sand seatrout stomachs. Percent *IRI* values listed for each month before (B) and after (A) MAOR addition.

Sand Seatrout	March (B)	May (B)	July (B)	Sept. (B)	March (A)	May (A)	July (A)	Sept. (A)
Amphipods	--	0.2	3.0	0.0	--	0.7	0.0	0.0
Calanoids	--	43.0	0.0	0.3	--	1.1	0.0	0.0
Cyclopoids	--	0.0	0.0	0.0	--	0.0	0.0	0.0
Detritus	--	0.3	0.0	1.7	--	0.5	0.0	0.0
Harpacticoids	--	0.0	0.0	0.0	--	0.1	0.0	0.0
Insect Larvae	--	0.0	0.0	0.0	--	0.1	0.0	0.0
Mysids	--	14.3	62.4	70.2	--	1.0	75.8	81.1
Clams	--	0.0	0.0	0.0	--	0.0	0.0	0.0
Stomatopods	--	0.0	0.1	0.0	--	0.0	0.0	0.0
Tanaids	--	0.1	0.0	0.0	--	0.2	0.0	0.0
Unknown	--	0.2	2.1	0.2	--	0.9	0.0	0.1
Zoea	--	16.3	0.0	16.3	--	43.8	0.1	0.0
Polychaetes	--	0.0	0.0	0.0	--	0.1	0.0	0.0
Crabs	--	0.0	0.8	0.0	--	0.0	0.0	0.0
Fishes	--	25.2	30.9	11.3	--	51.3	23.7	15.1
Shrimps	--	0.3	0.7	0.0	--	0.2	0.5	3.8

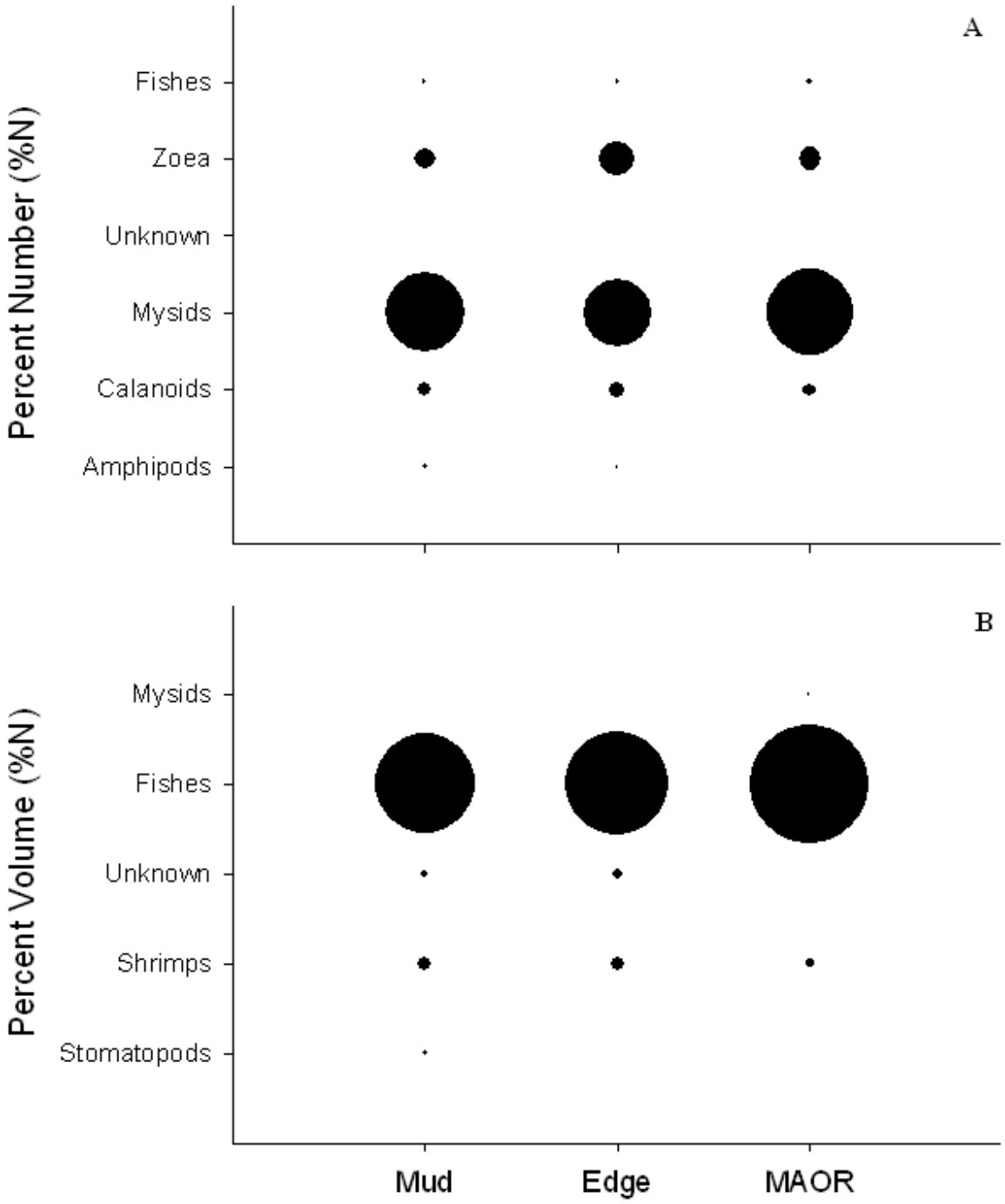

Figure 4.8. Percent number (A) and volume (B) contributions of important prey items to the diet of sand seatrout from each habitat type. Prey items are listed in order of their *%IRI* ranking from highest to lowest. Prey items with low percent values (≤ 1) were removed.

Table 4.15. Percent index of relative importance (*%IRI*) values for each diet item found in sand seatrout stomachs in each habitat type.

Sand Seatrout	Mud	Edge	MAOR
Amphipods	1.19	0.54	0.00
Calanoids	2.27	1.13	0.82
Cyclopoids	0.00	0.00	0.02
Detritus	0.07	0.09	0.00
Harpacticoids	0.01	0.00	0.00
Insect Larvae	0.00	0.00	0.00
Mysids	53.53	60.54	66.87
Clams	0.00	0.00	0.00
Stomatopods	0.04	0.00	0.00
Tanaids	0.03	0.03	0.08
Unknown	0.80	1.20	0.00
Zoea	3.85	3.68	3.61
Polychaetes	0.00	0.00	0.00
Crabs	0.15	0.42	0.00
Fishes	36.76	30.55	27.96
Shrimps	1.29	1.82	0.64

Electivity indices indicated sand seatrout strongly selected for (E>0.3) calanoids, crabs, fishes, mysids, shrimps and zoea; strongly selected against (E< -0.3) bivalves, cyclopoids and harpacticoids; and fully avoided (E=-1.0) anthomedusae, branchiurans, egg masses, gastropods, insects, isopods, nematodes and ostracods in the overall diet (Table 4.17). However, habitat-specific electivity values indicated crabs and shrimps were fully avoided by sand seatrout at MAOR sites, while positive selection indices for calanoids, fishes, mysids, and zoea increased (Table 4.17).

Table 4.16. SIMPER output of the items that explain >90% of the dissimilarity of sand seatrout diets in each habitat type (mud, unvegetated edge, and MAOR). Data were square root transformed.

Diet Item	Volume (mm^3)		Average Dissimilarity	% Contribution to Dissimilarity	% Cumulative Contribution
	Mud	MAOR			
Mysids	1.42	1.42	30.24	39.86	39.86
Fishes	1.89	1.25	20.96	27.63	67.49
Zoea	0.12	0.12	5.98	7.89	75.37
Unknown	0.31	0.00	4.33	5.70	81.08
Shrimps	0.27	0.18	3.59	4.73	85.81
Amphipods	0.22	0.00	2.89	3.81	89.61
Calanoids	0.07	0.02	2.45	3.22	92.84
Average dissimilarity = 75.87					
	Edge	MAOR			
Mysids	1.34	1.42	29.97	39.42	39.42
Fishes	2.09	1.25	19.59	25.76	65.18
Shrimps	0.48	0.18	6.46	8.50	73.68
Zoea	0.09	0.12	5.39	7.09	80.77
Unknown	0.46	0.00	4.46	5.86	86.63
Crabs	0.22	0.00	3.44	4.52	91.15
Average dissimilarity = 76.03					
	Mud	Edge			
Mysids	1.42	1.34	27.14	32.97	32.97
Fishes	1.89	2.09	21.77	26.45	59.43
Unknown	0.31	0.46	7.65	9.29	68.72
Shrimps	0.27	0.48	5.50	6.68	75.40
Zoea	0.12	0.09	4.87	5.91	81.31
Amphipods	0.22	0.13	4.16	5.05	86.36
Crabs	0.11	0.22	4.11	4.99	91.36
Average dissimilarity = 82.31					

Table 4.17. Overall diet and habitat-specific electivity indices (using Ivlev's electivity index) for diet items found in sand seatrout and pinfish stomachs. Prey items listed represented ≥1.0% of the diet by *%IRI* indices.

White Trout	Rank						**Pinfish**	Rank					
Organism	*p*	*r*	Overall	Mud	Edge	MAOR	Organism	*p*	*r*	Overall	Mud	Edge	MAOR
Amphipods	4	4	0.241	0.205	0.175	-1.000	Amphipods	4	5	0.146	-0.161	0.110	0.221
Anthomedusae	19	24	-1.000	-1.000	-1.000	-1.000	Anthomedusae	19	24	-1.000	-1.000	-1.000	-1.000
Bivalves	17	13	-0.386	0.369	--	-1.000	Bivalves	17	11	0.058	0.817	1.000	-1.000
Branchiurans	22	24	-1.000	-1.000	-1.000	-1.000	Branchiurans	22	14	0.358	0.315	0.555	-1.000
Calanoids	3	3	0.303	0.245	0.347	0.532	Calanoids	3	1	0.274	-0.191	0.294	0.412
Crabs	16	7	0.308	0.239	0.177	-1.000	Crabs	16	15	-0.534	-0.312	-1.000	-1.000
Cyclopoids	7	12	-0.381	-0.573	-1.000	0.017	Cyclopoids	7	6	0.213	0.064	0.310	-0.067
Egg Mass	9	24	-1.000	--	-1.000	--	Egg Mass	9	24	-1.000	--	-1.000	--
Fishes	14	5	0.450	0.367	0.541	0.593	Fishes	14	12	-0.057	-1.000	-0.153	0.201
Gastropods	10	24	-1.000	-1.000	-1.000	-1.000	Gastropods	10	24	-1.000	-1.000	-1.000	-1.000
Harpacticoids	2	10	-0.394	-0.420	-1.000	-1.000	Harpacticoids	2	10	-0.289	-0.251	-0.387	-0.611
Insects	21	24	-1.000	-1.000	-1.000	-1.000	Insects	21	16	-0.338	-0.067	-1.000	-1.000
Insect Larvae	18	11	-0.047	-0.027	-1.000	-1.000	Insect Larvae	18	7	0.384	0.564	0.630	0.433
Isopods	8	24	-1.000	-1.000	-1.000	-1.000	Isopods	8	9	-0.001	-0.137	-0.058	-1.000
Mysids	11	1	0.595	0.538	0.866	0.896	Mysids	11	3	0.342	0.520	0.612	0.742
Nematodes	1	24	-1.000	-1.000	-1.000	-1.000	Nematodes	1	17	-0.797	-1.000	-1.000	-0.679
Ostracods	12	24	-1.000	-1.000	-1.000	-1.000	Ostracods	12	18	-0.577	-1.000	0.420	-1.000
Plants	25	24	--	--	--	--	Plants	25	4	1.000	1.000	1.000	1.000
Polychaetes	6	9	-0.190	-0.190	-1.000	-1.000	Polychaetes	6	8	0.070	0.020	-0.102	0.358
Shrimps	15	6	0.336	0.267	0.282	-1.000	Shrimps	15	13	-0.064	-1.000	-0.028	-0.268
Stomatopods	25	24	--	--	--	--	Stomatopods	25	19	1.000	--	1.000	--
Tanaids	5	8	-0.072	-0.074	-0.532	-0.341	Tanaids	5	2	0.166	-0.038	0.070	0.175
Zoea	20	2	0.713	0.665	0.839	0.879	Zoea	20	24	-1.000	-1.000	-1.000	-1.000

- Pinfish

The highest number of pinfish were collected at edge sites, but the highest mean lengths and weights of pinfish were collected at mud sites (Tables 4.2 and 4.3). Mean lengths and weights of pinfish were significantly larger at mud sites ($p<0.05$; Tukey-adjusted LSmeans) but not significantly different between edge and MAOR sites. The habitat-effect interaction terms (i.e., Period*Location or Period*Site) were non-significant ($p>0.01$) but a significant monthly effect was observed in the Month*Site interaction term ($p<0.01$; PERMANOVA). In general, pinfish diets were diverse for *%N* but were dominated by plant material and detritus by *%V* (Figure 4.5). Plant material, tanaids, amphipods, and detritus comprised 65% of the overall diet by *%IRI* (Table 4.4). Invertebrates comprised large portions of the diet in March and May in both periods, with plant material increasing in dietary importance in July and September in the "after" period (Table 4.18).

Habitat-specific comparisons indicated pinfish diets were less diverse in mud habitat and much more diverse in both edge and MAOR habitats by *%IRI* (Table 4.19; Figure 4.9). Pinfish diets in edge and MAOR habitats consisted of nine prey types (≥1.0%) while mud sites consisted of only five prey types. For *%N*, pinfish diets were dominated by mysids and plant material at mud sites and calanoid copepods at edge sites, while several prey types were consumed in even proportions at MAOR sites (Figure 4.9). For *%V*, pinfish diets were dominated by plant material at mud sites, fish prey at MAOR sites, and several prey types at edge sites (Figure 4.9). For *%IRI*, dominant prey types were much more inconsistent between habitats with no prey type representing >10% of the habitat-specific diet in all habitat types (Table 4.19). SIMPER analysis

Table 4.18. The percent index of relative importance (*%IRI*) for each diet item found in pinfish stomachs. Percent *IRI* values listed for each month before (B) and after (A) MAOR addition.

Pinfish	March (B)	May (B)	July (B)	Sept. (B)	March (A)	May (A)	July (A)	Sept. (A)
Amphipods	46.4	0.1	--	--	20.1	14.6	0.0	0.0
Branchiurans	0.0	0.0	--	--	0.0	0.4	0.0	0.0
Calanoids	30.4	32.6	--	--	0.0	25.3	0.8	0.3
Cyclopoids	5.5	0.0	--	--	0.8	0.9	0.0	0.0
Detritus	0.0	65.4	--	--	0.1	15.6	3.4	0.7
Harpacticoids	0.0	0.0	--	--	0.7	0.0	0.0	0.1
Insect Larvae	1.5	0.0	--	--	2.6	2.4	5.0	0.1
Isopods	16.1	0.0	--	--	0.0	0.0	0.0	0.0
Mysids	0.0	0.2	--	--	0.1	0.1	17.3	10.4
Nematodes	0.0	0.0	--	--	0.0	0.0	0.0	0.0
Oligochaetes	0.0	0.0	--	--	0.0	0.0	0.0	0.0
Ostracods	0.0	0.0	--	--	0.0	0.0	0.0	0.0
Plants	0.0	0.0	--	--	0.1	5.5	69.0	41.5
Clams	0.0	0.0	--	--	0.0	0.1	0.0	0.0
Stomatopods	0.0	0.0	--	--	0.0	1.0	0.0	0.0
Tanaids	0.0	1.1	--	--	33.1	18.4	0.5	0.6
Insects	0.0	0.0	--	--	0.0	0.0	0.0	0.0
Unknown	0.0	0.5	--	--	6.2	9.3	3.0	44.4
Polychaetes	0.0	0.0	--	--	36.4	3.3	0.1	0.0
Crabs	0.0	0.0	--	--	0.0	0.0	0.0	0.0
Fishes	0.0	0.0	--	--	0.0	2.6	0.0	2.0
Shrimps	0.1	0.0	--	--	0.0	0.5	0.8	0.0

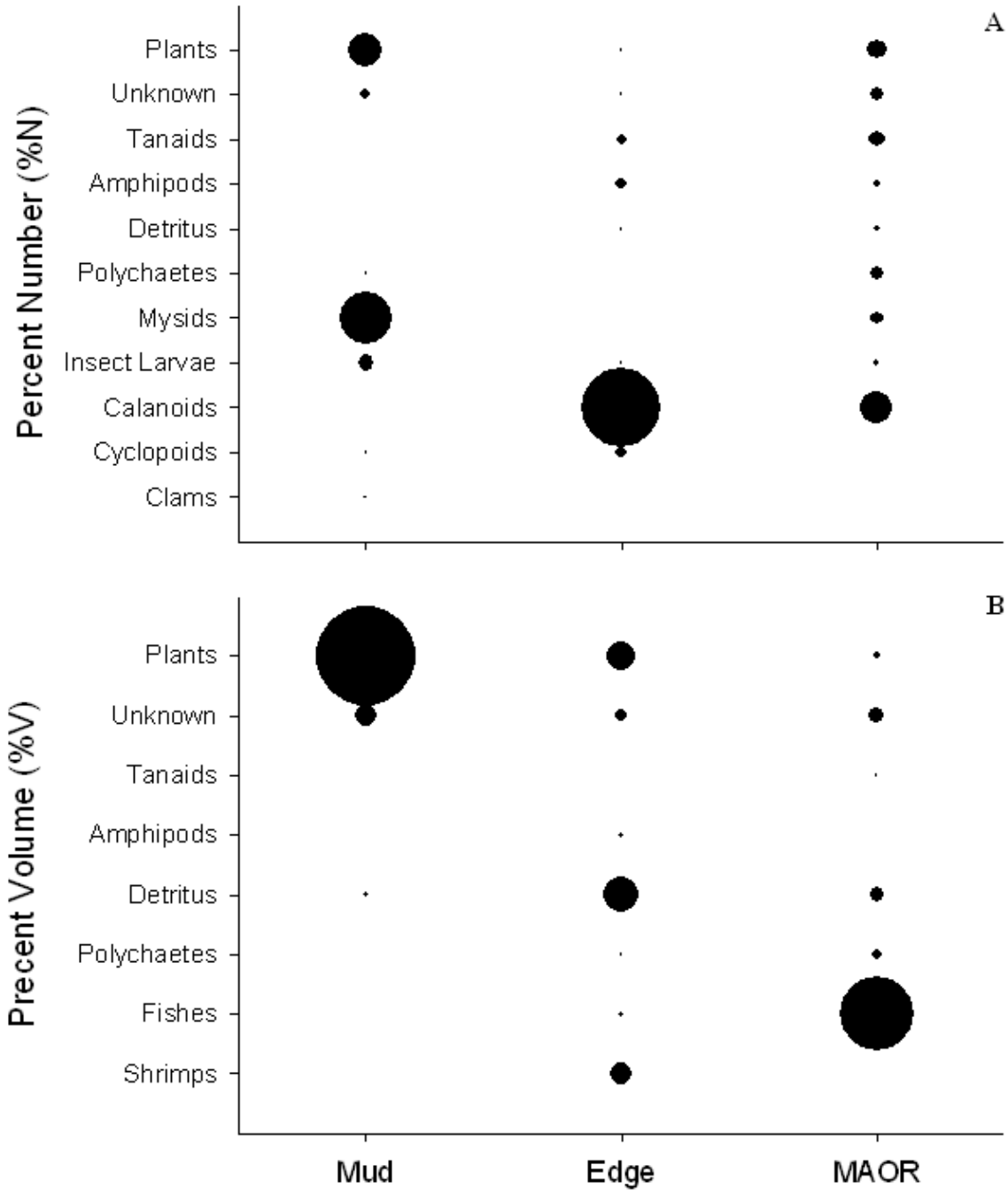

Figure 4.9. Percent number (A) and volume (B) contributions of important prey items to the diet of pinfish from each habitat type. Prey items are listed in order of their *%IRI* ranking from highest to lowest. Prey items with low percent values (≤ 1) were removed.

Table 4.19. Percent index of relative importance (*%IRI*) values for each diet item found in pinfish stomachs by habitat type.

Pinfish	Mud	Edge	MAOR
Amphipods	0.04	10.53	4.06
Branchiurans	0.05	0.08	0.00
Calanoids	0.05	30.00	13.00
Cyclopoids	0.04	2.31	0.08
Detritus	0.41	20.27	6.90
Harpacticoids	0.07	0.04	0.02
Insect Larvae	3.13	2.06	3.39
Isopods	0.04	0.36	0.00
Mysids	20.87	0.17	2.71
Nematodes	0.00	0.00	0.02
Ostracods	0.00	0.00	0.00
Plants	68.05	13.37	7.75
Clams	0.05	0.00	0.00
Stomatopods	0.00	0.24	0.00
Tanaids	0.07	9.72	15.63
Insects	0.00	0.00	0.00
Unknown	6.87	6.75	20.11
Polychaetes	0.24	1.37	14.80
Crabs	0.01	0.00	0.00
Fishes	0.00	0.31	11.52
Shrimps	0.00	2.42	0.02

indicated polychaete, tanaid, detritus, and amphipod consumption increased at MAOR sites compared to mud sites, and polychaete and fish prey consumption was higher at MOAR sites when compared to edge sites (Table 4.20). Comparisons among diets realized “before” and “after” at MAOR sites indicated detritus, plant material, mysids, and crabs contributed most to cumulative dissimilarity with plant material being the only prey type consumed more during the “after” period (Table 4.8). Estimates of habitat-specific prey quality *(%ICI*) indicated large portions of diets at mud sites were comprised

Table 4.20. SIMPER output of the items that explain >90% of the dissimilarity in pinfish diets in each habitat type (mud, unvegetated edge, and MAOR). Data were square root transformed.

Diet Item	Volume (mm^3)		Average Dissimilarity	% Contribution to Dissimilarity	% Cumulative Contribution
	Mud	MAOR			
Plants	4.53	0.78	31.89	36.39	36.39
Unknown	1.33	1.02	15.44	17.62	54.02
Polychaetes	0.23	0.84	8.86	10.11	64.13
Detritus	0.31	0.65	8.06	9.20	73.33
Mysids	0.57	0.19	6.18	7.05	80.38
Tanaids	0.04	0.50	4.34	4.95	85.33
Insect Larvae	0.24	0.21	3.54	4.04	89.36
Amphipods	0.05	0.27	2.86	3.27	92.63
Average dissimilarity = 87.62					
	Edge	MAOR			
Plants	1.35	0.78	14.62	17.15	17.15
Detritus	1.49	0.65	14.44	16.94	34.09
Unknown	0.83	1.02	13.25	15.55	49.64
Amphipods	1.15	0.27	11.52	13.51	63.15
Polychaetes	0.36	0.84	9.49	11.13	74.28
Tanaids	0.57	0.50	7.50	8.80	83.08
Fishes	0.14	0.57	3.77	4.43	87.51
Insect Larvae	0.16	0.21	3.09	3.63	91.13
Average dissimilarity = 85.23					
	Mud	Edge			
Plants	4.53	1.35	29.81	33.84	33.84
Unknown	1.33	0.83	12.76	14.48	48.32
Detritus	0.31	1.49	10.96	12.45	60.76
Amphipods	0.05	1.15	9.41	10.68	71.44
Tanaids	0.04	0.57	5.46	6.20	77.64
Mysids	0.57	0.06	5.15	5.84	83.48
Polychaetes	0.23	0.36	3.88	4.40	87.89
Insect Larvae	0.24	0.16	2.66	3.02	90.91
Average dissimilarity = 88.09					

of energetically poor prey items (i.e., amphipods, plant material, polychaetes, isopods, and tanaids) while diets at edge and MAOR sites were comprised mainly of energetically valuable prey items (i.e., copepods, fish prey, insect larvae and mysids; Table 4.9). At mud sites, plant material comprised almost 50% of the nutritional intake where as amphipods, insect larvae, plant material, polychaetes, and tanaids each contributed 10-22% of the nutritional intake at MAOR sites.

Electivity indices indicated pinfish fully selected for (E=1.0) plant material and stomatopods; strongly selected for (E>0.3) branchiurans, insect larvae, and mysids; strongly selected against (E< -0.3) crabs, insects, nematodes, and ostracods; and fully avoided (E=-1.0) anthomedusae, egg masses, gastropods, and zoea (Table 4.17). Habitat-specific electivity indices indicated that calanoids, insect larvae, mysids, and polychaetes were strongly selected for while amphipods, cyclopoids, fishes, shrimps, and tanaids were consumed in proportion to their relative densities in the environment.

4.3.3 Energy Density

- Atlantic Croaker

A total of 80 samples, comprised of 233 individuals, were inlcuded in the energy density analysis for Atlantic croaker. Maximum and minimum mean energy densities both were observed during the "after" period in March and July at 17329.3 and 14953.6 $J \cdot g^{-1}$, respectively, with an overall mean of 16161.1 $J \cdot g^{-1}$ (Figure 4.10). Mean energy density values declined from March to July in both periods. Energy density values were similar at MAOR sites before and after deployment (Figure 4.11) but were higher at MAOR sites compared with mud and edge sites (Figure 4.12). Energy densities were not

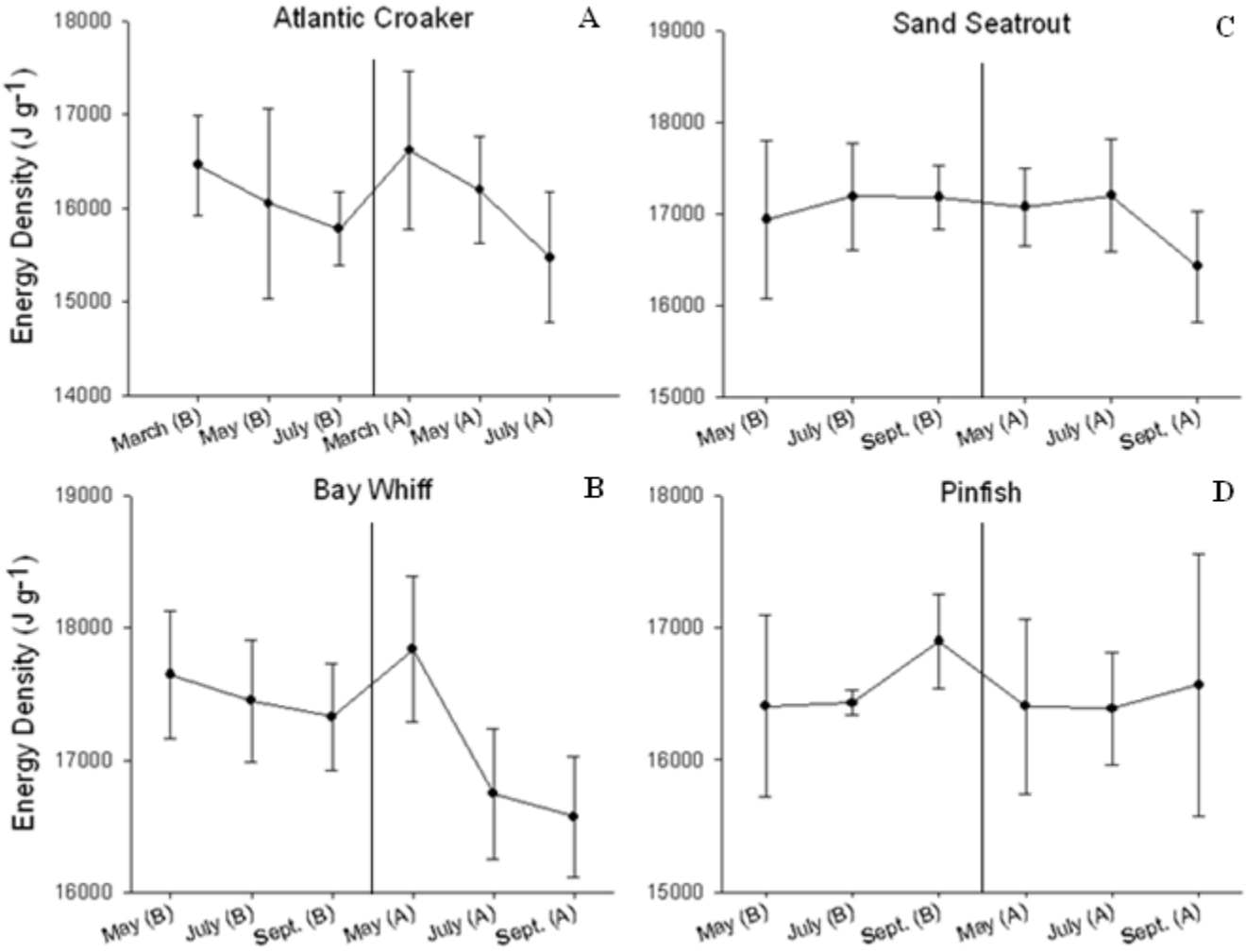

Figure 4.10. Line plot of the monthly mean energy density ($J \cdot g^{-1}$; dry weight; including standard error bars) for each fish species before and after MAOR deployment. Vertical line represents deployment of cobble during March 2010.

significant between habitats between years (Period*Habitat term; $p>0.05$; mixed-model ANOVA) but were highly significant between habitats between months and ponds (Month*Habitat and Month*Pond; $p<0.0001$; mixed-model ANOVA). Additionally, a strong month affect was observed as period, month and pond interaction terms were significant (Period*Month and Period*Month*Pond; $p<0.001$). Linear regression indicated a singificant negative relationship between energy density and length ($p=0.04$) but no signficant relationship for the habitat term or the Length*Habitat interaction term ($p>0.05$; ANCOVA). Linear regression of energy density versus length with 95% confidence intervals for Atlantic croaker is displayed in Figure 4.13.

- Bay Whiff

A total of 56 samples, conmprised of 106 individuals, were included in the energy density analysis for bay whiff. The maximum and minimum energy densitiy values were observed in May and September during the "after" period at 18269.0 and 16492.5 $J \cdot g^{-1}$, respectively. Mean energy density values were highest in May and decreased through September during both periods (Figure 4.10). Mean energy densities were similar at MAOR sites before and after deployment (Figure 4.11) and were also similar between habitat types (Figure 4.12). Energy densities were not significantly different for the Period*Habitat interaction term ($p>0.05$) and were not significantly different for the Month*Habitat interaction term ($p>0.05$). However, the interaction term Period*Month*Habitat was significant ($p=0.001$). The interaction term Period*Month was also significant, indicating monthly shifts in energy densities for bay whiff ($p=0.0002$). Linear regression indicated a singificant negative relationship between energy density and length ($p=0.005$), but neither the main effect for habitat nor the

Length*Habitat interaction were significant (p>0.05; ANCOVA). Linear regression of energy density on length with 95% confidence intervals is displayed in Figure 4.13.

- Sand Seatrout

A total of 44 samples, comprised of 84 individuals, were included in the energy density analysis for sand seatrout. Maximum and minimum energy density values were observed at MAOR habitats in July and September during the "after" period at 17584.5 and 16112.2 $J \cdot g^{-1}$, respectively. Mean monthly energy densities remained relatively constant around the overall mean of 17026.8 $J \cdot g^{-1}$ except in September during the "after" period when values declined sharply (Figure 4.10). Energy densities were only slightly lower at MAOR sites after deployment (Figure 4.11) but were highest at MAOR sites compared to other habitat types (Figure 4.12). A significant affect was detected for the habitat term (p=0.048) but not for the Period*Habitat interaction term (p<0.05). No month afftect was detected as energy densities of sand seatrout were not significant between periods and months (Period*Month term; p>0.05; mixed-model ANOVA). Linear regression indicated a singificant negative relationship between energy density and length (p=0.0001), habitat (p=0.005) and the interaction term (p=0.0005; ANCOVA) indicating trends in energy density with length were not similar between habitat types. Energy density remained stable within mud habitats, but declined sharply with length in both edge and MAOR habitat (Figure 4.14).

- Pinfish

A total of 28 samples, comprised of 57 individuals, were included in the energy density analysis for pinfish. Minimum energy density values (May 15999.6 $J \cdot g^{-1}$) and

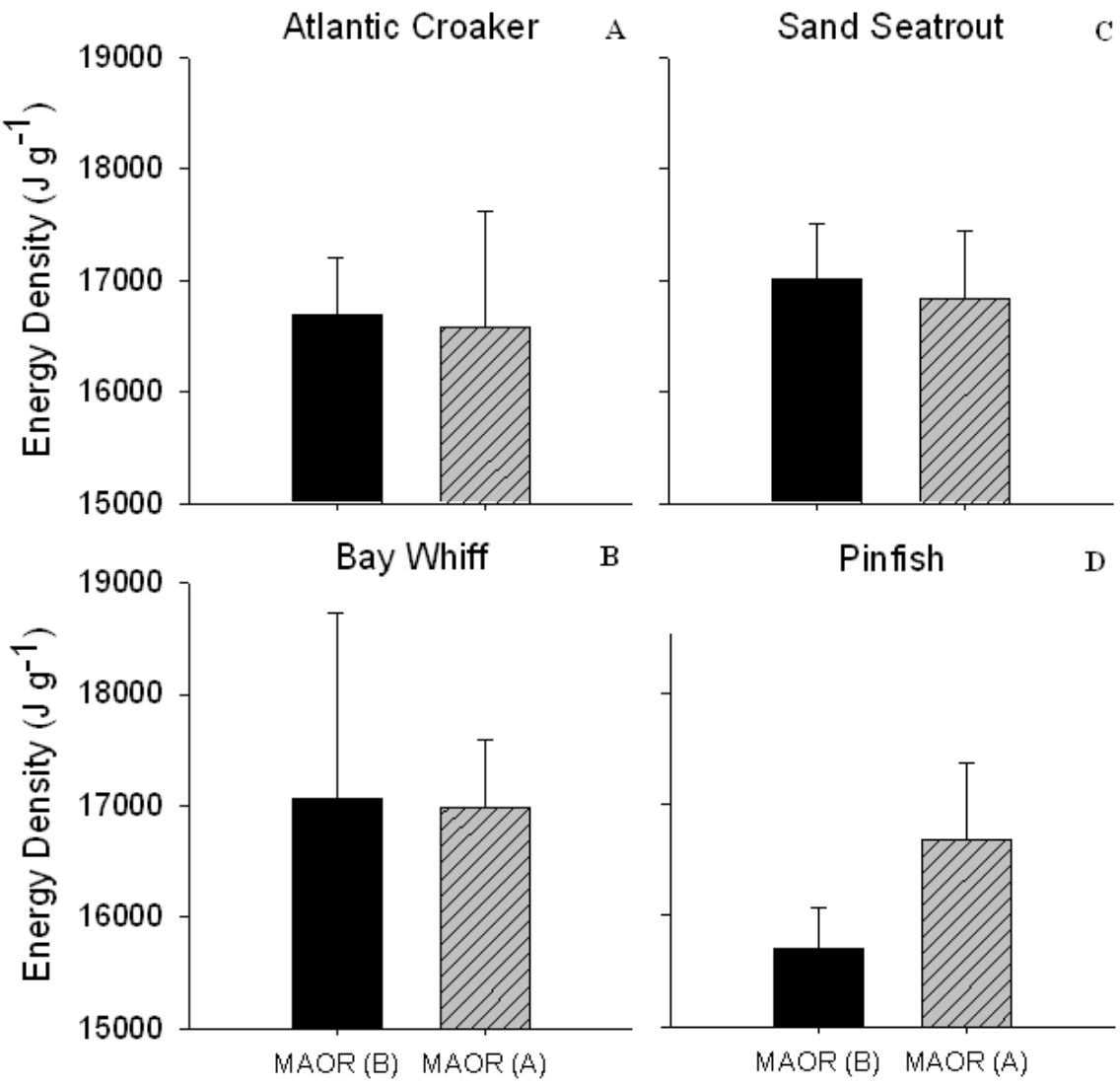

Figure 4.11. Vertical bar chart of the mean energy density ($J \cdot g^{-1}$; dry weight; including standard errors) for Atlantic croaker (A), bay whiff (B), sand seatrout (C), and pinfish (D) before (black) and after (grey) MAOR deployment.

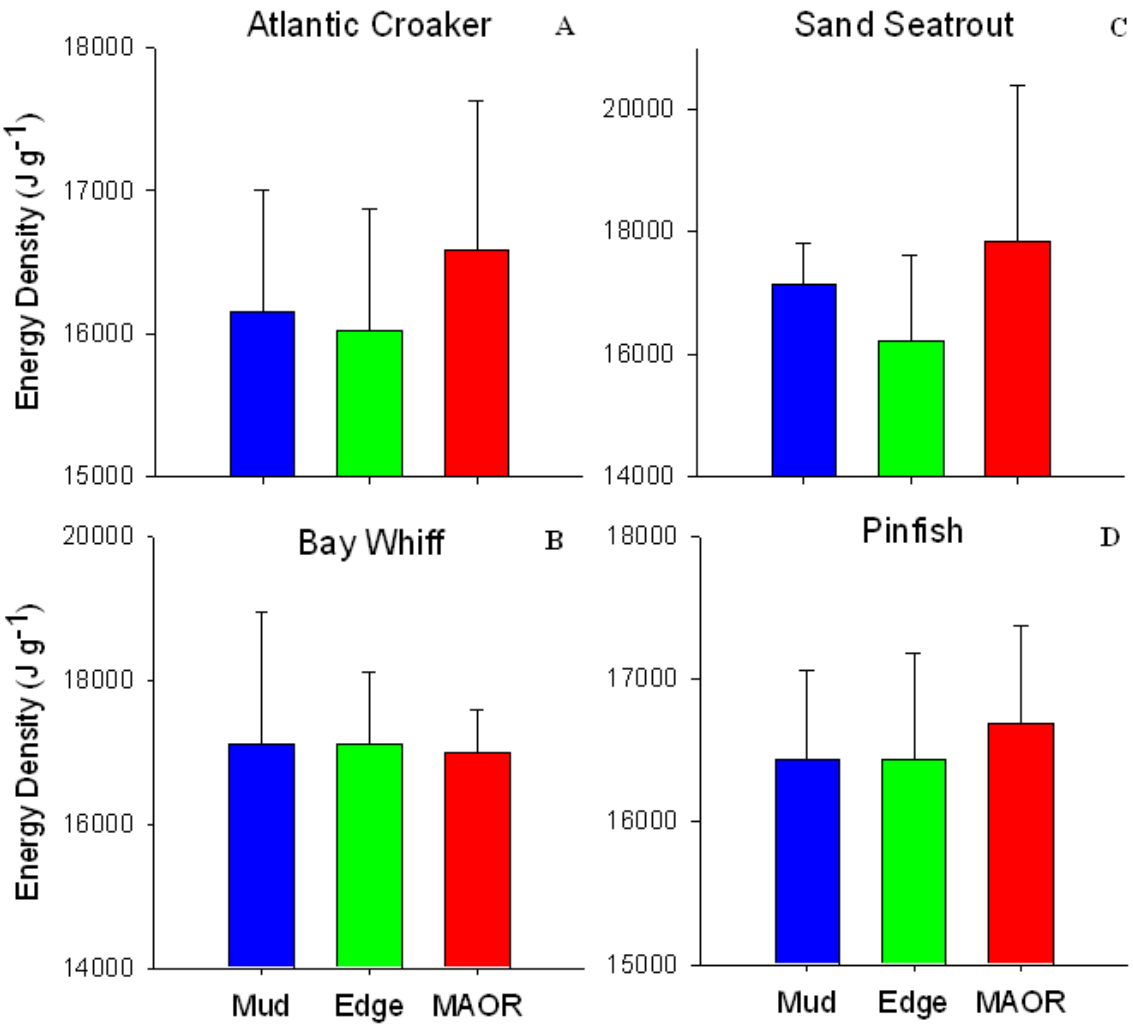

Figure 4.12. Vertical bar chart of the mean energy density ($J \cdot g^{-1}$; dry weight; including standard errors) for Atlantic croaker (A), bay whiff (B), sand seatrout (C), and pinfish (D) from mud (blue), edge (green), and MAOR (red) habitats.

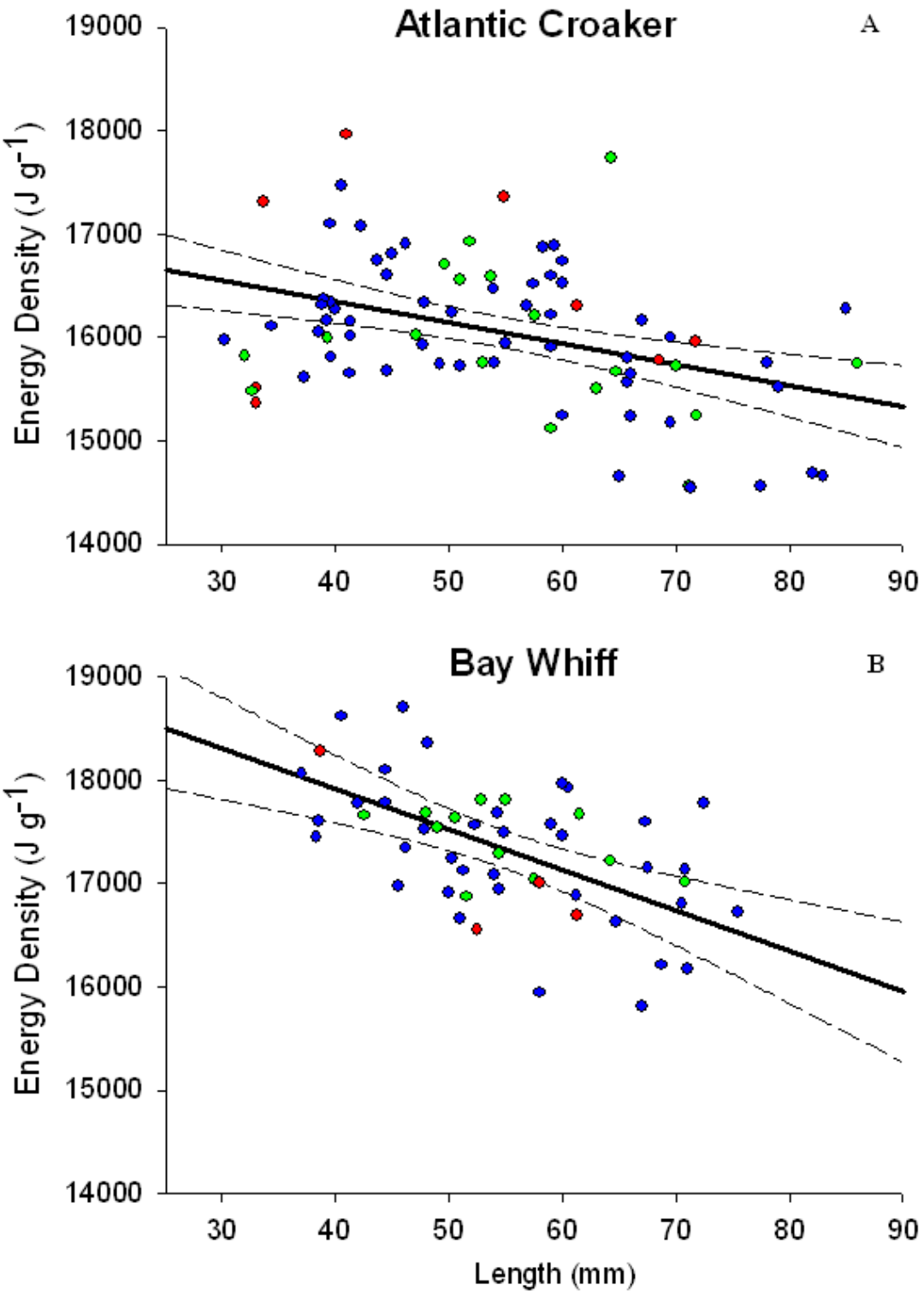

Figure 4.13. Linear regression of energy density ($J \cdot g^{-1}$; dry weight) on standard length (mm) for Atlantic croaker (A) and bay whiff (B) from mud (blue), edge (green), and MAOR (red) habitats. Habitat-specific energy densities were not significantly different.

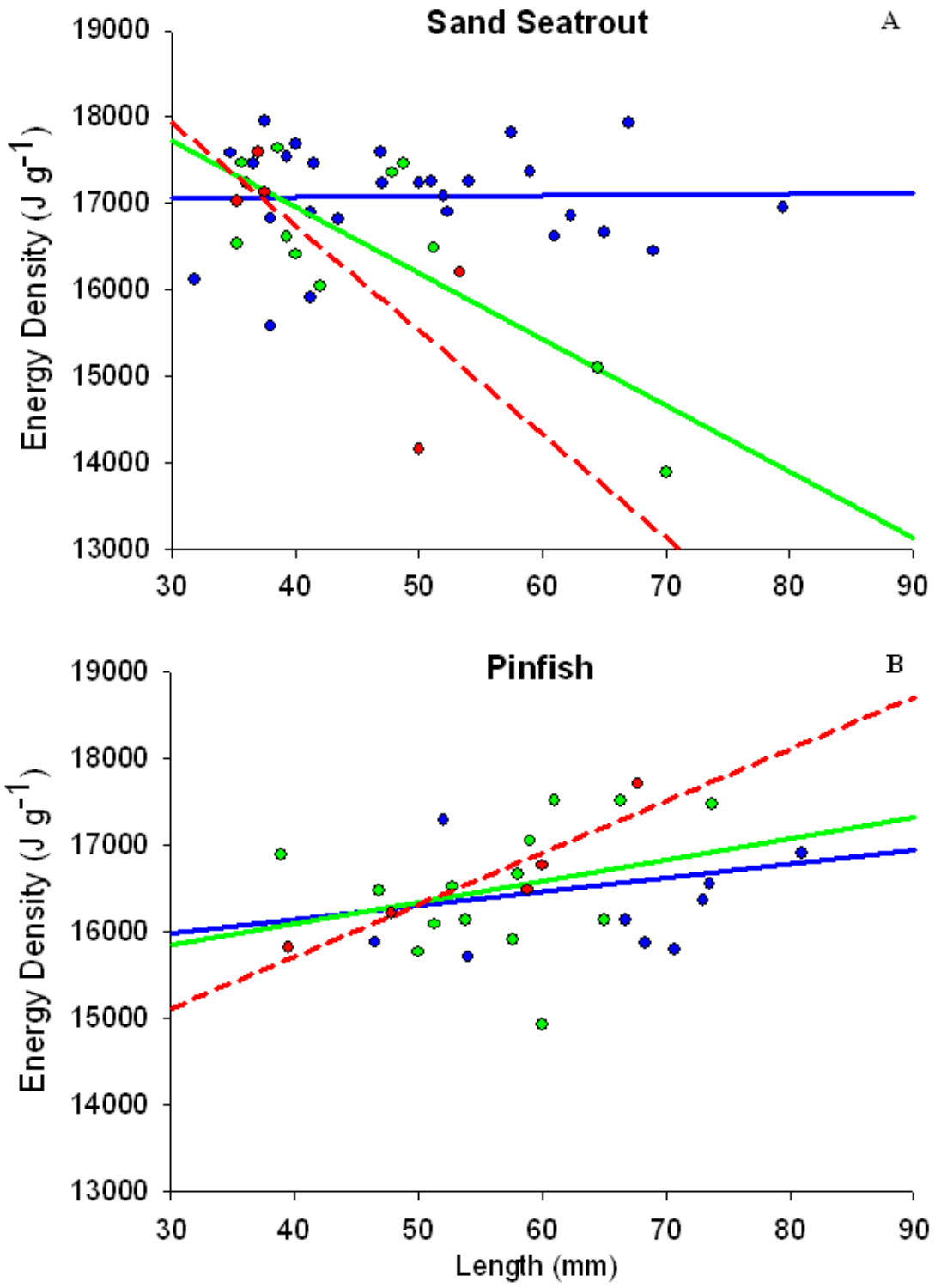

Figure 4.14. Linear regression of energy density ($J \cdot g^{-1}$; dry weight) on standard length (mm) for sand seatrout (A) and pinfish (B) from mud (blue), edge (green), and MAOR (red) habitats. Habitat specific energy densities were significantly different for sand seatrout but not for pinfish.

maximum values (September 17693.7 $J \cdot g^{-1}$) were observed at impact sites (i.e., MAOR sites during the second year) before deployment with an overall average of 16454.0 $J \cdot g^{-1}$ (Figure 4.10). Mean monthly energy densities increased from May through September in both periods. Energy densities of fish collected from MAOR sites were higher after MAOR deployment (Figure 4.11) and were higher in MAOR habitat compared with mud and edge habitats (Figure 4.12). Energy densities were significantly different between months ($p=0.0001$) and a significant affect of MAOR addition was also detected for pinfish (Period*Location term; $p=0.02$) but were not significantly different between habitats ($p>0.05$) in the mixed-model ANOVA. Linear regression indicated a singificant negative relationship between energy density and length ($p=0.005$), but neither the main effect for habitat nor the Length*Habitat interaction differed significantly ($p>0.05$; ANCOVA). Linear regression (ANCOVA) indicated that energy density increased with length in all three habitat types but was highest at the MAOR habitat (Figure 4.14).

4.4 Discussion

4.4.1 Sampling Design and Statistical Inference

- BACI Design

Previous literature has identified potential limitations of BACI experimental designs primarily attributable to type I errors, sampling designs that are incapable of accounting for ecological variance, and difficulties with interpretation of results (Hewitt et al., 2001; Stewart-Oaten et al., 1986; Stewart-Oaten and Bence, 2011). Of particular concern in executing viable statistical inferences using BACI designs are potential

violations of the assumptions: 1) interval or ratio scale response variables; 2) equal variance across time and space variable combinations; 3) independence of samples and associated error structures before and after within time and space combinations; and 4) approximate normal distributions for response variables in space (Hewitt, 2001; Schwarz, 2011). In addition, as both locations and sites were contained within each pond, some degree of pseudoreplication does exist within this experiment (Hurlburt, 1984).

I believe that the statistical design used for these analyses satisfy the concerns and objections associated with simple BACI designs for the following reasons: 1) a traditional "simple" temporal BACI design was not used but rather samples were collected during multiple months before and after perturbation; 2) a traditional "simple" spatial BACI design was not used but rather samples were collected from impact and control locations at spatial levels both larger and smaller than the level of impact (i.e., impact locations were sub-units of ponds and sites were sub-units of impact locations) with adequate replication; 3) the type I error rate was reduced by collecting multiple explanatory ecological variables (i.e., species) and including them into a single analysis (i.e., PERMANOVA); 4) many of the assumption violations typically associated with parametric analysis, such as ANOVA, when analyzing BACI data are not necessary in semi-nonparametric and nonparametric tests such as PERMANOVA and ANOSIM; and 5) comparisons between factors and their levels in PERMANOVA are made using dissimilarity matrices which utilize differences between temporal and spatial units simultaneously, as was recommended to control for autocorrelation by Stewart-Oaten et al., (1986).

- Environmental Parameters

As differences in the observed data can result from ecological impacts other than the impacts controlled in the experiment (Stewart-Oaten, 1986; Stewart-Oaten and Bence, 2001), environmental parameters were collected simultaneously with experimental data as a means to measure conditions that may have influenced observed results. When analyzed, significant differences between one or more ponds were observed for all four environmental variables suggesting experimental units were not under statistically similar conditions across space. However, I disregard these differences and attribute significant differences to type I error resulting from extremely high sample size, as the observed environmental conditions in marsh ponds were very similar. Ponds were paired in different geographical locations (approximately 1 km between pairs) but were in relatively close proximity within a pair (approximately 0.25 km between ponds within a pair). Thus, concerns regarding pseudoreplication (Hurlburt, 1984) should be satisfied through a relatively large distance between pairs, and concerns regarding the expectation of similar environmental conditions across experimental units (Hurlburt, 1984; Stewart-Oaten et al., 1986; Stewart-Oaten and Bence, 2001) should be satisfied by the relatively small distance between pairs of ponds and between ponds within a pair.

- Electivity Indices

As with all sampling gears, the sampling gears used in this experiment have inherent biases and selectivity for and against certain organisms, or were not targeted by the study desing. Thus, not every prey item within marsh ponds could be collected. Terrestrial forms of insects, plant material, and stomatopods were not effectively

collected in this experiment and electivity estimates for these prey types should be interpreted with caution. Insects were not targeted by the sampling gear and are considered incedental when present in lift tray or plankton net tows. Living plant material is affixed to or within a substrate and was not collected using lift trays or plankton nets. Stomatopods are mobile invertebrates that occupy burrows for refuge and thus would be expected to avoid plankton nets via burrows, as well as a hard structure such as a lift tray as it cannot be burrowed into effectively. The absence of these prey types from collections had little impact upon the calculation of electivity indices for other prey types as they represented very small portions of the diet of the four fishes used for diet analyses. Stomatopods likely have very low densities in marsh ponds and plant material is difficult to quanitfy for count data and *%N*, as it is often masticated during consumption by fishes.

- Energy Densities

It can be assumed that stomach contents reflect the abundant prey items available within a habitat type, but assuming that observed energy densities are a direct result of the conditions provided by the habitat type from which fishes were collected in this experiment is somewhat dubious. Changes in diet composition are not immediately reflected in energy composition due to the time required for metabolic turnover. In addition, the four fishes studied in this chapter are quite mobile relative to both pond and habitat size and could potentially utilize multiple habitats before the stomach contents from a single feeding period are digested. Therefore, data on fine-scale movement patterns of Atlantic croaker, bay whiff, sand seatrout, and pinfish were collected concomitantly with energy density data using passive integrated transponder (PIT) tags

continuously monitored by antenna arrays placed in both ponds that received MAOR treatments. Analyses of these data are not yet complete and could not be included in this thesis, but the results and analyses will be presented and discussed in a subsequent manuscript.

4.4.2 General Trends in Diet

This study identified similar diet compositions as found previously for Atlantic croaker (Hansen, 1969; Overstreet and Heard, 1978; Nemerson and Able, 2005; Simonsen, 2008), bay whiff (Toepfer and Fleeger, 1995), sand seatrout (Hein, 1999), and pinfish (Hansen, 1969; Stoner 1980b; Stoner, 1984). Despite the diversity of abundant prey types, fish diets were shown to be dominated by only a few taxa, with a single prey type comprising as much as 80% of the total diet. Opportunistic fishes (i.e., Atlantic croaker and pinfish) consumed a greater variety of prey items, including both pelagic and benthic forms, while more specialized fishes (i.e., bay whiff and sand seatrout) consumed primarily pelagic prey with much less variety. In Atlantic croaker dietary studies, Overstreet and Heard, (1978) identified 83 and 60 taxa in stomachs from Mississippi Sound and the near-shore Gulf of Mexico, respectively. Other studies have found diets generally consisting of annelids, molluscs, crustaceans, and fishes in variable proportions (Hansen, 1969; Overstreet and Heard 1978; Nemerson and Able, 2005; Simonsen, 2008). In another example involving bay whiff, Toepfer and Fleeger (1995) determined an asymptotic number of 12 prey items in stomachs from a Louisiana estuary, with diets consisting almost exclusively of mysids and calanoid copepods. Members of the genus *Cynoscion* (i.e., sand seatrout) typically feed on small invertebrates, transitioning to a more fish-dominated diet in late-juvenile and adult stages. Stomach content items and

relative proportions in this study almost perfectly mirrored previously reported diet descriptions for bay whiff (Toepfer and Fleeger, 1995) and sand seatrout (Hein, 1999). Despite an opportunistic feeding strategy and multiple ontogenetic shifts, pinfish diets within this study also matched previous findings (Stoner, 1980b). Up to five ontogenetic diet shifts have been reported for pinfish with a general transition from almost exclusively epibenthic meiofauna to a relatively high degree of herbivory in later life-stages (Stoner, 1980b). In addition, a wide variety of vegetation has been previously reported in pinfish diets including diatoms, filamentous algae and vascular plants (Hansen, 1969); these prey items can account for as much as approximately 30% of the diet (Stoner, 1980b). The proportion of pinfish diets comprised of plant material in my study was as high as 70% with the remaining portion comprised of various motile epibenthic invertebrates.

High frequency of occurrence of major prey types and a low percentage of empty stomachs suggests a variety of prey items were readily available in marsh ponds. Major prey types were present in diets throughout the year, but temporal shifts in diet composition were apparent for all the fishes I studied. Diet shifts were large in all fishes except sand seatrout, with a given prey type often comprising the majority of the diet in one month, and absent in previous or subsequent months. Despite strong shifts in the relative proportions of major prey items, bay whiff and sand seatrout diets consistently contained pelagic prey throughout most of the year. Bay whiff and sand seatrout diets were highly dependent upon mysids, which comprised as much as 95% of the total diet during some months. When preferred prey was not available, bay whiff and sand seatrout consumed a low variety of other prey types, but never in as high of proportions as mysids

and calanoid copepods. For Atlantic croaker and pinfish, both species fed opportunistically throughout the year. Earlier in the year, pinfish fed on a variety of epibenthic crustaceans and polychaetes but little consumption of plant material. Later in the year, plant material became a major component of pinfish diets in addition to invertebrate prey. Increased consumption of plant material with increasing fish size was presumably driven by ontogenetic diet shifts (Stoner, 1980b).

In general, mysids, pelagic copepods and zoea were positively selected; nematodes, gastropods, harpacticoids, and ostracods were negatively selected; and amphipods and polychaetes were consumed in proportion with their density in the environment. Pelagic mysids, copepods, and decapod larvae have been found to be the most energetically valuable prey items in inshore Gulf of Mexico waters, while benthic infauna and epifauna such as polychaetes, amphipods, and crabs were much less valuable calorically (Wissing et al., 1973). With caloric densities as high as 6600-7500 $cal \cdot g^{-1}$ (dry wt) pelagic meiofauna provide almost twice the energetic value of benthic infauna and epifauna (Wissing et al., 1973). My data suggest the four fish species studied in this experiment fed on lower quality prey types (amphipods, polychaetes and other benthic prey) in relative proportion to their density in the environment and selected for high quality food types, such as mysids and copepods, when available.

4.4.3 Habitat-Specific Diets

Variability in water level, thus access to marsh ponds, and the life stage of fishes in the ponds may have significantly altered the feeding ecology of fishes compared with other habitats (e.g., channels or open bays). The deployment of MAORs into shallow marsh ponds is a novel artificial habitat application so direct comparisons of habitat-

specific diet compositions with other marsh pond studies are not possible. That said, studies conducted in shallow open bays and channels generally agree with the habitat-specific diet compositions observed in this study. Simonsen (2008) observed generalistic feeding in Atlantic croaker and a predominance of piscivory in spotted seatrout at limestone-cobble reefs in a shallow, open bay in Louisiana (Simonsen, 2008). Simonsen (2008) found no significant habitat-specific differences for Atlantic croaker due to generalized feeding in all habitat types but did note the majority of prey items at reef sites were crustaceans. Other studies have indicated some habitat-specific specialization in sciaenid diets (i.e., spot, *Leiostomus xanthurus*), but this may only occur under infrequently occurring conditions, such as when hypoxic bottom waters make infaunal or burrowing invertebrates more vulnerable to predation (Pihl et al., 2002). In Simonsen (2008), shifts in spotted seatrout diets to predominantly fish prey at reef sites were attributed to increased prey availability. These marsh ponds did exhibit extremely high densities of planktivorous pelagic fishes (i.e., gulf menhaden) but sand seatrout did not consume fish prey in proportions similar to those of spotted seatrout observed in Simonsen (2008), who mostly collected larger specimens. A recent review of the ecology of sand seatrout identified soft bottom sand or mud as optimal habitat for young sand seatrout feeding predominantly on pelagic invertebrates, while hard-structures such as reefs serve as favorable habitats to adults and are associated with an ontogenetic diet shift to predominantly fish prey. No current study has evaluated pinfish diet compositions at either natural or artificial oyster reefs, however comparisons between sand and seagrass habitats suggest pinfish utilize complex habitats to reduce size-dependent predation, increase total food consumption, and maximize growth, but not to consume habitat-

specific prey resources (Levin et al., 1997; Harter and Heck, 2006). As living plant material were unavailable at MAOR sites it is unknown if pinfish diets would have changed similarly given available plant resources.

4.4.4 Role of Mimic Artificial Oyster Reefs in Marsh Ponds

Overall, MAORs do not appear to directly enhance the feeding ecology of the marsh pond fish community. Despite increases in diversity and number of some small epibenthic taxa in response to MAOR addition (Chapter 2), these data indicate MAOR-specific prey items were not effectively integrated into higher trophic levels. Increases in consumption of some MAOR-specific prey were observed, but pelagic prey dominated the diet composition of all four fishes. Of the six benthic, potential prey taxa that increased in density at MAOR after deployment, only insect larvae, polychaetes, and tanaids were important diet components of fishes at MOAR sites (tanaids in pinfish only). Bivalves, gastropods, and ostracods were rarely consumed by fishes and electivity indices indicated selection for these prey items was strongly negative for all but pinfish. The lack of increase in consumption of epibenthic meiofauna in Atlantic croaker, bay whiff, and sand seatrout diets suggest these prey were not effectively available or provided insufficient energetic return compared to other available prey types at MAOR sites.

Behavioral characteristics and ecology specific to each predator/prey type may explain the feeding interactions observed at MAOR sites. As surface dwellers, insect larvae are unlikely to be directly impacted by MAOR addition and, despite the statistical significance observed in Chapter 2, increased densities at MAOR sites are most likely artificial. Alternatively, the increase in vertical relief provided by MAORs could increase encounter rates or capture efficiency of insect larvae at MAOR sites and could explain

the proportionate increase of insect larvae in fish diets. Chapter 2 indicated polychaete densities also increased at MAOR sites but their primarily infaunal behavior and the complex structure of MAORs may have reduced capture efficiency by predators. There was potential for increases in polychaete densities combined with diurnal migrations to essentially create a "spill-over" effect at MAOR sites to fishes during nighttime feeding but there was little evidence to support this contention from these data. Swarms of polychaetes were observed swimming in the water column during some night-time sampling events. Such ephemeral, pelagic behavior could explain the occurrence of polychaetes in fish diets at MAOR sites without showing significant differences. Additional investigations into to the mechanisms controlling the incorporation of various food types into predator diets might provide some useful insight.

Differential prey quality and energetic return could also have contributed to the negative selection against benthic and epibenthic prey at MAORs, especially in combination with behavioral feeding ecology. Less energetically valuable (benthic) prey could have become even less desirable if MAORs decreased consumption efficiency of feeding fishes (Hughes, 1980). As benthic and epibenthic prey are quite common in Atlantic croaker diets (Hansen, 1969; Overstreet and Heard, 1978; Nemerson and Able, 2005; Simonsen, 2008), negative selection by Atlantic croaker suggests they could not be consumed effectively. Atlantic croaker mouth morphology, at the sizes observed in these marsh ponds (i.e., mean lengths ranging from 47.6 to 119 mm; Table 4.3), promotes capture success of pelagic invertebrates (or when benthic invertebrates move into the water column) while making grazing or picking epibenthic invertebrates from hard substrate surfaces inefficient (Schmitt and Holbrook, 1984; Sardina and Cazorla, 2005).

Similarly, sand seatrout are highly suited for piscivory due to their large gape (Hein, 1999), and bay whiff is an epibenthic, cryptic species passively waiting to ambush passing prey items (Toepfer and Fleeger, 1995). None of these species are specifically adapted to efficiently capturing epibenthic invertebrates, especially within complex, structural habitats. Of the prey types that increased in density in response to MAOR deployment, these were likely too low in number, too small, too large, or too inefficiently consumed to provide net energy gain to the fishes I studied (Hughes, 1980).

In contrast to the other three fish species, pinfish did demonstrate the ability to utilize MAOR-specific resources such as polychaetes, tanaids, and amphipods. Previous studies have indicated that electivity values for benthic and epibenthic prey items were not strongly positive in pinfish diets (Ivlev, 1961; Lechowicz, 1982). In my study however, electivity values were greater than zero for amphipods, polychaetes, and tanaids suggesting these prey types could be effectively consumed when encountered. In addition, pinfish mouth morphology may reduce capture efficiency of highly mobile free swimming prey, such as copepods and mysids, while increasing capture efficiency of epibenthic prey, such as amphipods and tanaids (Stoner, 1984). Although electivity values in pinfish diets in this study remained relatively high for pelagic prey, values were also higher for tanaids, polychaetes and amphipods at MAOR sites, all prey types that showed positive responses to MAOR addition in Chapter 2. These data provide evidence that MAORs may provide suitable habitat to younger pinfish consuming primarily epibenthic invertebrates while mud and edge habitats provide suitable habitat for larger pinfish consuming relatively large amounts of plant material (Stoner, 1982; Harter and Heck, 2006). High plant consumption and minimal positive selection for high quality

prey items suggests pinfish are able to obtain the majority of their nutritional requirements through abundant, low quality foods while opportunistically occupying dietary niches less-optimal to conspecifics or other fishes (Stoner, 1984).

4.4.5 Fish Condition and Comparative Habitat Value

Optimal foraging theory differentiates food consumption into a cost-benefit analysis between the energy consumed through searching, handling, and digestion, as well as avoiding predators, versus the energy gained through consumption and assimilation of a particular prey type (Hughes, 1980; Pyke, 1984). Therefore, two scenarios for the potential enhancement of feeding ecology in response to MAOR addition are plausible: 1) MAOR addition could directly increase the prey base by number or variety available to predator fishes, and therefore, increase total consumption; or 2) MAOR addition could increase the vulnerability of the prey base resulting in more efficient consumption and greater net energy return. More specifically fish could experience feeding enhancement, if MAORs allowed more efficient capture of desirable prey resources, and this effect may be observable through habitat-specific comparisons of total body condition (i.e., energy density). Atlantic croaker, sand seatrout, and pinfish all showed significant habitat-specific differences in energy density, with bay whiff energy densities being only slightly lower at MAOR sites. This suggests MAORs did provide quality habitat to all four fish species. However, habitat-specific regressions of energy density versus length were non-significant for Atlantic croaker and bay whiff and indicated only mud habitat was favorable for sand seatrout. The specific mechanisms driving these differences are unclear, but diet analyses suggest impacts to food resources at MAOR sites (scenario #1) were not a major factor. Habitat-specific regressions do

suggest the addition of MAORs may have negatively impacted young sand seatrout. The relatively small sizes of sand seatrout collected in this study (i.e., mean lengths ranging from 35.0 to 63.0 mm) are typical of young juveniles that depend upon open, mud-bottom habitat to consume small pelagic invertebrates (Hein, 1999). Older juveniles and sub-adult stages do display the ability to utilize hard structures for feeding, but likely recruited from marsh ponds to other habitats in deeper water prior to ontogenetic shifts toward structure-associated prey (Hein, 1999) as they were rarely collected in this study. Deployment of MAORs could have improved capture efficiency of preferred pelagic prey by all four fishes, but this contention is only speculative without additional experimentation.

Energy density analyses provide additional support for the contention that MAORs did enhance feeding ecology in pinfish by providing favorable habitat to younger juveniles. Pinfish energy densities were highest at MAOR sites compared with other habitat types and increased at MAOR sites after deployment. Regressions of energy content versus fish length indicated pinfish increased energy density with size whereas Atlantic croaker, bay whiff, and sand seatrout all declined in energy density as they grew. Habitat-specific regressions were not significant, but the regression of energy density versus length in pinfish from MAOR sites showed a higher rate (slope) of energy storage than pinfish from either mud or edge habitats. Opportunistic feeding and specific ontogenetic requirements may have allowed pinfish to utilize MAORs in such a way that decreased ontogenetic competition or improved resource utilization, thereby allowing for energy allocation to growth and storage rather than growth alone.

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CHAPTER 5: SUMMARY AND CONCLUSIONS

5.1 Summary

The objective of this study was to evaluate the potential for limestone cobble, deployed as mimic artificial oyster reefs (MAORs), to enhance the feeding ecology of the juvenile fish community in marsh ponds. Many studies have previously evaluated a wide variety of artificial reef types and applications but enhancement or success criteria are often based only upon the change in abundance or diversity at artificial reef sites without gathering concomitant data on utilization or vital rates. True enhancement requires the organism of study to exhibit increased vital rates such as recruitment, growth, or survival as enhancement based on numbers or diversity is equivocal at best. I sought to examine the impact of MAOR addition not only on juvenile fishes but on multiple dimensions of the marsh pond community as the ecological impact of reef addition is often quite variable and difficult to predict.

In Chapter 2, I evaluated the efficacy of a relatively novel technique for detecting differences in total-body fish condition between experimental treatments for the purposes of, for example, evaluating the impacts of environmental perturbations or comparisons of relative habitat quality. Bioelectric impedance analysis provides indirect, nondestructive estimates of compositional condition that are rapidly collected, repeatable, and independent of size (Kushner, 1992; Kyle et al., 2004). Previous research has shown impedance values derived from BIA measures to be very strongly correlated to proximate components and total body assessments of fish condition in multiple species (Cox and Hartman, 2005; Cox et al., 2010). In this study, however, BIA-derived condition

measures did not show a strong ability to distinguish between drastically different treatment groups, especially when compared to traditional condition measures. This study does not dispute the efficacy of BIA to evaluate fish condition, but rather provides insight into the potential complications and limitations of BIA when applied to fishes during particular ontogenetic life stages.

In Chapter 3, I evaluated the impact of MAOR addition on the abundant prey base potentially available to juvenile fishes in marsh ponds. This allowed identification of prey taxa that were vulnerable to habitat change through increases (i.e., MAOR addition) or decreases (i.e., replacement of natural habitat) in favorable or optimal habitat. As might be expected, the "replacement" of fine sediment with large cobble resulted in a decrease of resident infaunal meiofauna and total organism density was greatly reduced. Epibenthic invertebrates increased in density with six new taxa observed at MAOR sites after deployment. Macrofauna declined drastically from 23 to 8 species at MAOR sites and total organism density was also reduced. Four reef-associated species did exhibit increased densities and/or sizes at MAOR sites. These data suggest an overall negative impact of MAOR addition on the potential prey base of juvenile fishes in marsh ponds but provide some evidence for diversity enhancement of meiofauna and species-specific interactions in some reef-associated macrofauna.

In Chapter 4, I determined which food resources were most important to juvenile fishes in marsh pond food webs and evaluated the magnitude of the impact to the potential prey base to higher trophic levels. Potential impacts to marsh pond communities are certainly not limited to direct changes in diet composition, and thus I also examined fish condition for three purposes: 1) to assess the impact of MAOR addition from a

nutritional viewpoint in addition to direct impacts to fish diets; 2) to assess the relative quality of MAOR habitat versus other natural habitats; and 3) in the absence of diet-related differences, to assess the magnitude of alternative impacts, such as niche partitioning or predation refuge, that may also be attributable to MAOR addition, but were not directly targeted by this experimental design.

The impacts of MAORs to the potential prey base identified in Chapter 3 were not observed in the diet compositions of juvenile estuarine fishes. In general, pelagic prey items not associated with hard-structures were preferred by the species of fishes collected in this experiment and are likely representative of the marsh pond fish community in general, although exceptions exist (e.g., juvenile spot). Furthermore, diet composition at MAOR sites did not reflect changes in prey community observed in Chapter 3, as pelagic prey items again comprised the majority of fish diets in MAOR habitats. Analyses of fish condition indicated similar or increased energy densities in fishes collected at MAOR sites compared with other natural habitats. Pinfish, which are commonly associated with seagrass and other structured habitats, did show the ability to utilize MAOR associated-resources and may have used MAOR sites to improve feeding during specific ontogenetic stages.

5.2 Conclusions

Although multiple ecological viewpoints were investigated in this study, it is difficult to assess the degree of ecological functionality of MAORs in this marsh pond system. Natural community succession on artificial reefs and other hard structures typically involves a nonlinear increase followed by a gradual decline towards equilibrium

(Carter et al., 1985; Woodhead and Jacobson, 1985; Coen and Luckenbach, 2000). Despite relatively fast initial colonization, mature biological complexity typically occurs on a scale of years rather than months, and I did not expect a large increase in complexity to be observed at MAOR sites after only one year. There is certainly potential for MAORs to develop into fully functioning artificial oyster reefs, essentially equivalent to natural oyster reefs, given proper environmental conditions and time (Peterson et al., 2003). Limestone cobble has been shown to be a biologically suitable substrate for sessile invertebrate colonization, especially oyster spat (Haywood et al., 1999). However, due to the temporal limitations of this study, it is certain that MAORs had not reached maximum community complexity nor were they functioning at levels equivalent to natural oyster reefs. Visual monitoring of MAOR substrate during the study indicated MAORs displayed relatively low complexity of sessile invertebrates. Few sub-adult sessile colonizers (e.g., barnacles and oysters), no adult colonizers, and no plant or algal growth was observed on cobble stones. A typical equilibrated oyster reef would be comprised of a suite of flora and fauna (both colonial and mobile) represented by multiple life stages across multiple trophic levels (Peterson et al., 2003). Spawning of barnacles and oysters occurs in late winter/early spring and generally coincided with the deployment of MAORs (March). The lack of sessile colonization may have been attributable to a variety of factors: 1) limited inflow of water into secluded marsh ponds; 2) substrate surfaces may not have been immediately favorable to settling larvae, i.e., were not preconditioned by favorable growth of bacteria (Coen and Luckenbach, 2000); 3) high diversity and abundance of planktivorous (e.g., gulf menhaden, *Brevoortia patronus*) and invertivore (e.g., sheepshead, *Archosargus probatocephalus* and black drum, *Pogonias cromis*)

fishes (Brown and Swearingen, 1998); or proximity to major larval spawning areas. The lack of sessile organisms indicates the physical complexity of MAORs was effectively limited to the three dimensional arrangement of the stones themselves with little additional benefit from biological complexity.

The results of this study highlight the need to evaluate the impact of artificial habitats at the community-level by simultaneously incorporating utilization data across multiple trophic levels (Bohnsack et al., 1991; Svane and Petersen, 2001). Accurate and useful investigations of artificial habitat functionality should provide data regarding utilization, condition, and vital rates as assessments based on simple comparisons of community compositions alone provide equivocal evidence at best. Data from Chapter 3 suggest MOAR functionality was relatively high when examining meiofauna assemblage structure, but relatively low when examining the assemblage structure of macrofauna. Diet compositions indicated most of the fishes selected for study used few MAOR-associated resources. However, some resource utilization by pinfish and increased densities, lengths, and weights of naked gobies (*Gobiosoma bosc*), sheepshead, and gulf toadfish (*Opsanus beta*) indicated potential for MAORs to provide habitat or growth enhancement to a select group of specialized fishes. Alternatively, condition analyses suggested that MAORs provided good-quality habitat despite the immaturity of reef development. Condition analyses also suggested that sand seatrout may be negatively affected by the addition of MAORs as they have been shown to be dependent upon mud-bottom habitat at small sizes.

The relatively low utilization of MAOR habitats by the fishes I studied highlights the importance of and reliance upon mature and productive natural habitats by juvenile

fishes in Louisiana marshes. Despite the diversity of abundant prey types, juvenile estuarine fishes relied heavily upon key species to provide sufficient energy to meet metabolic demands. However, these data do not preclude the potential for successful enhancement of juvenile estuarine species using MAORs or alternative structures. Some evidence for positive effects on feeding ecology and fish condition was found to be attributable to MAORs, even during early stages of biological development. Reduced predation rates in shallow water refugia (marsh ponds) in combination with abundant high-quality prey provides important nursery habitat to juvenile estuarine fishes and facilitates successful transitions to adult habitat. Because nursery habitat is so important, additional investigations aimed at enhancing various biological or ecological aspects of juvenile fish life history are certainly warranted. However, the results of this study suggest that future research be directed towards enhancement of structure-associated fishes rather than the general marsh pond community.

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APPENDIX 1: GENERAL MODEL DESIGN FOR ALL STATISTICAL ANALYSES

Appendix 1. General model design for all statistical comparisons including response variables tested (y variables), model type, explanatory variables included in model (x variables), and additional comparisons (pairwise and similarity/dissimilarity comparisons).

Response Variables	Analysis Used	General Model Design	Pairwise Comparisons
Pond Comparisons			
Temperature water depth salinity pH	Mixed-Model ANOVA	y = period month pond period*pond month*pond	Tukey-Adjusted Lsmeans
Community Composition			
Meiofauna Taxa (1…n)	PERMANOVA	$y_1 \ldots y_n$= period month pond area site (all interaction terms)	ANOSIM
Small Macrofauna Species (1…n)	PERMANOVA	$y_1 \ldots y_n$= period month pond area site (all interaction terms)	ANOSIM
Diversity Indices (Shannon-Weaver Index)	Mixed-Model ANOVA	y = period month pond area (all interaction terms)	Tukey-Adjusted Lsmeans
y = meiofauna taxa or small macrofauna species of interest	Backward Elimination, Stepwise Regression	y = all environmental variables, meiofauna taxa, and small macrofauna species *only significant effect variables remain in final model	--
Diet Composition			
Prey Items (1…n) (Test of Main Effects)	PERMANOVA	$y_1 \ldots y_n$= period month pond area site (all interaction terms)	SIMPER Comparisons: Before/After Habitat Types
Fish Condition			
Energy Density (Tests for BACI Effects)	Mixed-Model ANOVA	y = period pond area (all interaction terms) *month included in repeated statement	Tukey-Adjusted Lsmeans
Energy Density (Habitat Comparisons)	Mixed-Model ANOVA	y = period pond habitat (all interaction terms) *month included in random statement	ANCOVA

APPENDIX 2: RELATIVE PROPORTIONS OF PREY ITEMS IN THE EVIRONMENT FOR CALCULATING ELECTIVITY INDICES

Appendix 2. Relative proportions of prey items collected from combined plankton tow and lift tray samples used to calculate habitat-specific electivity estimates. Proportions were calculated using log-transformed density estimates for each prey item.

Organism	Overall	Mud	Edge	MAOR
Amphipods	0.07	0.07	0.08	0.06
Anthomedusae	0.03	0.03	0.03	0.04
Bivalves	0.03	0.01	0.00	0.06
Branchiurans	0.02	0.02	0.01	0.00
Calanoids	0.07	0.08	0.08	0.06
Crabs	0.04	0.04	0.04	0.03
Cyclopoids	0.06	0.06	0.05	0.04
Egg Mass	-0.01	0.00	0.00	0.00
Fishes	0.04	0.05	0.04	0.04
Gastropods	0.05	0.05	0.05	0.08
Harpacticoids	0.09	0.10	0.11	0.10
Insects	0.02	0.03	0.02	0.01
Insect Larvae	0.03	0.04	0.02	0.04
Isopods	0.06	0.06	0.07	0.04
Mysids	0.05	0.05	0.02	0.02
Nematodes	0.11	0.12	0.15	0.12
Ostracods	0.04	0.01	0.01	0.07
Plants	0.00	0.00	0.00	0.00
Polychaetes	0.06	0.06	0.08	0.06
Shrimps	0.04	0.05	0.04	0.04
Stomatopods	0.00	0.00	0.00	0.00
Tanaids	0.07	0.06	0.09	0.09
Zoea	0.03	0.03	0.02	0.02
Total	1.00	1.00	1.00	1.00

APPENDIX 3: RELATIVE PROPORTIONS OF PREY TYPES IN STOMACH CONTENTS FOR CALCULATING ELECTIVITY INDICES

Appendix 3. Relative proportions of prey items in fish stomachs used to calculate electivity indices in each habitat type. Estimates are based on numbers of prey consumed (*N*) and do not include the "unknown" category.

Atlantic Croaker	Overall	Mud	Edge	MAOR
Amphipod	0.06	0.05	0.07	0.07
Anthomedusa	0.00	0.00	0.00	0.00
Bivalve	0.04	0.03	0.07	0.04
Branchiuran	0.00	0.00	0.00	0.00
Calanoid	0.51	0.55	0.44	0.35
Crabs	0.00	0.00	0.00	0.00
Cyclopoid	0.03	0.02	0.02	0.11
Egg Mass	0.00	0.00	0.00	0.00
Fishes	0.01	0.01	0.00	0.02
Gastropod	0.00	0.00	0.00	0.00
Harpacticoid	0.08	0.09	0.03	0.10
Insect	0.00	0.00	0.00	0.00
Insect Larva	0.08	0.07	0.12	0.13
Isopod	0.00	0.00	0.00	0.00
Mysid	0.04	0.03	0.11	0.04
Nematode	0.01	0.01	0.00	0.00
Ostracod	0.00	0.00	0.00	0.00
Plant	0.00	0.00	0.00	0.00
Polychaete	0.03	0.03	0.05	0.02
Shrimps	0.00	0.00	0.00	0.00
Stomatopod	0.00	0.00	0.00	0.00
Tenaid	0.01	0.00	0.02	0.02
Zoea	0.06	0.07	0.01	0.05
Total	0.97	0.97	0.97	0.94

Bay Whiff	Overall	Mud	Edge	MAOR
Amphipod	0.11	0.09	0.19	0.15
Anthomedusa	0.00	0.00	0.00	0.00
Bivalve	0.00	0.00	0.00	0.00
Branchiuran	0.00	0.00	0.00	0.00
Calanoid	0.40	0.44	0.38	0.05
Crabs	0.01	0.01	0.01	0.00
Cyclopoid	0.06	0.03	0.01	0.42
Egg Mass	0.00	0.00	0.00	0.00
Fishes	0.00	0.00	0.00	0.00
Gastropod	0.00	0.00	0.00	0.00
Harpacticoid	0.01	0.01	0.00	0.01
Insect	0.00	0.00	0.00	0.00
Insect Larva	0.01	0.01	0.00	0.07
Isopod	0.00	0.00	0.01	0.00
Mysid	0.37	0.38	0.38	0.24
Nematode	0.00	0.00	0.00	0.00
Ostracod	0.00	0.00	0.00	0.00
Plant	0.00	0.00	0.00	0.00
Polychaete	0.01	0.01	0.02	0.03
Shrimps	0.01	0.01	0.01	0.00
Stomatopod	0.00	0.00	0.00	0.00
Tenaid	0.00	0.00	0.01	0.00
Zoea	0.01	0.01	0.00	0.01
Total	1.00	1.00	1.00	0.99

Appendix 3. cont.

Sand Seatrout	Overall	Mud	Edge	MAOR
Amphipod	0.04	0.04	0.03	0.00
Anthomedusa	0.00	0.00	0.00	0.00
Bivalve	0.00	0.00	0.00	0.00
Branchiuran	0.00	0.00	0.00	0.00
Calanoid	0.09	0.09	0.09	0.10
Crabs	0.01	0.01	0.01	0.00
Cyclopoid	0.00	0.00	0.00	0.00
Egg Mass	0.00	0.00	0.00	0.00
Fishes	0.03	0.03	0.03	0.05
Gastropod	0.00	0.00	0.00	0.00
Harpacticoid	0.00	0.00	0.00	0.00
Insect	0.00	0.00	0.00	0.00
Insect Larva	0.00	0.00	0.00	0.00
Isopod	0.00	0.00	0.00	0.00
Mysid	0.63	0.65	0.54	0.67
Nematode	0.00	0.00	0.00	0.00
Ostracod	0.00	0.00	0.00	0.00
Plant	0.00	0.00	0.00	0.00
Polychaete	0.00	0.00	0.00	0.00
Shrimps	0.01	0.01	0.01	0.00
Stomatopod	0.00	0.00	0.00	0.00
Tenaid	0.00	0.00	0.00	0.00
Zoea	0.19	0.15	0.29	0.17
Total	1.00	1.00	1.00	1.00

Pinfish	Overall	Mud	Edge	MAOR
Amphipod	0.07	0.01	0.09	0.06
Anthomedusa	0.00	0.00	0.00	0.00
Bivalve	0.00	0.03	0.00	0.00
Branchiuran	0.00	0.01	0.00	0.00
Calanoid	0.53	0.01	0.64	0.29
Crabs	0.00	0.00	0.00	0.00
Cyclopoid	0.07	0.02	0.09	0.01
Egg Mass	0.00	0.00	0.00	0.00
Fishes	0.00	0.00	0.00	0.02
Gastropod	0.00	0.00	0.00	0.00
Harpacticoid	0.01	0.01	0.00	0.00
Insect	0.00	0.00	0.00	0.00
Insect Larva	0.04	0.14	0.03	0.05
Isopod	0.01	0.01	0.01	0.00
Mysid	0.08	0.44	0.01	0.12
Nematode	0.00	0.00	0.00	0.00
Ostracod	0.00	0.00	0.00	0.00
Plant	0.08	0.29	0.03	0.18
Polychaete	0.02	0.02	0.01	0.13
Shrimps	0.00	0.00	0.00	0.00
Stomatopod	0.00	0.00	0.00	0.00
Tenaid	0.08	0.01	0.08	0.15
Zoea	0.00	0.00	0.00	0.00
Total	1.00	1.00	1.00	1.00

APPENDIX 4: PERMISSION FROM JOURNAL "TRANSACTIONS OF THE AMERICAN FISHERIES SOCIETY" TO INCLUDE PUBLISHED MANUSCRIPT IN THESIS

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VITA

Steven Benjamin Garner was born in Panama City, Florida in 1985. He was more or less the only child of a very large family growing up and spent the vast majority of his time outside catching whatever swam, crawled, or flew if possible. If not playing baseball at the sandlot next door, catching animals from the ditch down the street, or bowling at the alley down the block, he was out fishing with his Dad and Grandfather. He never missed a day of school until the seventh grade when his grandfather took him fishing, rather than attending a half day session, and it was all downhill from there. He graduated from Bay High School in 2003, received his Bachelor of Science degree in marine biology from the University of West Florida in 2007, and began his graduate work here at LSU that same year under the supervision of Richard F. Shaw and James H. Cowan, Jr. in the Department of Oceanography and Coastal Sciences at the School of the Coast and Environment.