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## Spotted sea trout (*Cynoscion nebulosus*) and pinfish (*Lagodon rhomboides*) dietary analysis according to habitat type

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SPOTTED SEA TROUT (*CYNOSCION NEBULOSUS*) AND PINFISH (*LAGODON  
RHOMBOIDES*) DIETARY ANALYSIS ACCORDING TO HABITAT TYPE

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  
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## ABSTRACT

The diets of a transitory fish (spotted sea trout, *Cynoscion nebulosus*) and a fish with presumed greater site-fidelity (pinfish, *Lagodon rhomboides*) were analyzed with respect to habitat. Sampling occurred in Barataria Bay, Louisiana between May 2003 and May 2004. Spotted sea trout were caught using gillnets, and pinfish were caught using baited fish traps. Each gear was used on three habitats: mud bottom, oyster shell, and marsh edge. In addition, sampling for spotted sea trout was repeated at three locations along a salinity gradient. A total of 175 spotted sea trout stomachs and 137 pinfish stomachs were examined.

Seventeen prey species were identified in the guts of spotted sea trout, and thirteen species were identified for pinfish. Fifty-three percent of spotted sea trout stomachs were empty, and 37% of pinfish stomachs were empty. Unidentifiable fish and brown shrimp (*Farfantepenaeus aztecus*) were consistently the most important prey items for spotted sea trout in weight, frequency, and caloric value. Plant material, detritus, and small amounts of fish and shrimp, were consistently most important for pinfish by weight, frequency, and caloric value. The diet of spotted sea trout caught adjacent to marsh edge had the least dietary overlap with fish from other habitats, especially during winter and spring. The diet of pinfish caught over shell had the least dietary overlap with conspecifics from other habitats. Dietary breadth for spotted sea trout was similar across habitats and lowest in winter. Dietary breadth for pinfish was lowest on the shell habitat and in fall. Neither habitat, season, water quality (*i.e.*, dissolved oxygen, temperature, salinity), nor any other effect could be shown to be statistically significant in

distinguishing the diet of either fish using Analysis of Variance (ANOVA) and Analysis of Similarities (ANOSIM) statistical techniques.

Diets of these two fish were not notably diverse, but the dietary breadth for both fish species was high, suggesting that of those prey items consumed, there was no strong preference among the prey. This may be attributable to the transitory nature of the fish and the ubiquity of some of the prey types across habitat types.

## CHAPTER 1. INTRODUCTION

Coastal estuaries have long been understood to be important nursery grounds for the young of many marine organisms, including many commercially important species of finfish. These unique environments serve as home to fish that are life-long residents, as well as those who utilize the estuary for just a part of their life cycle (Weinstein *et al.* 1979, Rakocinski *et al.* 1992, Peterson and Turner 1994).

The confluence of fresh water and seawater found at the estuary creates a salinity gradient, which Attrill (2002) suggests is more important to the diversity and distribution of fish than are individual fish salinity tolerances. Furthermore, it may be that so-called estuarine organisms do not exist, but are the product of two merging salinity-defined ecoclines (freshwater species at their downstream limit, and marine species at their upstream limit) (Attrill and Rundle, 2002). Regardless of the system definition, estuarine primary and secondary production are very important to both proximal and distal processes.

Barataria Bay is a large (4100 km<sup>2</sup>) estuary on Louisiana's productive Gulf of Mexico coast and is a well studied and rapidly changing system. With the exception of dredged channels and natural bayous, the bay is very shallow (2.8 meters on average), with small diurnal tides (Waldrop 1959). Nine distinct vegetation types have been described along the salinity gradient by Visser *et al.* (1998). The marshes and bottom sediments have been shown to be a net sink for carbon, with brackish and intermediate waters accumulating the most carbon in the form of deteriorating above-ground biomass (Feijtel *et al.* 1985, Childers and Day 1990). Nutrients exported from the marshes are

flushed in a pulsed fashion at the beginning of a tidal cycle, while imported nutrients arrive more consistently and are related to upstream sources (Childers and Day, 1990).

The marshes that surround Barataria Bay are disappearing, however, due to a number of natural and anthropogenic processes, including marsh subsidence, accelerating rise in sea level, canal dredging, and dramatic decreases in fresh sediment supply caused by restraining the Mississippi River with levees (Evers *et al.* 1992, Bass and Turner 1997). Land loss in coastal Louisiana has been as high as 100 km<sup>2</sup> per year, and as much as 2% total area per year (Delaune *et al.* 1990). Modeling by Reyes *et al.* (2000) predicts that more than a quarter (1,105 km<sup>2</sup>) of the total land area will be lost by 2018 without human intervention.

Inland and freshwater marshes are disappearing at a greater rate than salt marshes, due in part to the former's accretionary reliance on unstable organic materials. Salt marshes are also receding quickly, though, as tides fail to deliver sediment past the marsh-water interface, allowing the marsh interior to subside and ponds to form (Delaune *et al.* 1990, Hatton *et al.* 1993). Convincingly, Childers and Day (1991) calculated that the land loss between 1967 and 1987 in Barataria Bay resulted in a 27% reduction in the supply of inorganic and organic nutrients from the depleted marshes to the estuarine water column. River diversions and the breaking of levees, intended to supply more sediments to the marshes, are hypothesized to be the most successful way to combat this problem (Reyes *et al.* 2000, Day *et al.* 2000, Martin *et al.* 2000).

This reduction in regenerated nutrients directly affects the volume of primary production in the bay, and most certainly indirectly affects fish production. In 1996, the U.S. Congress passed the Sustainable Fisheries Act, formerly the Magnuson Fishery

Conservation and Management Act, which included an Essential Fisheries Habitat (EFH) mandate requiring all regional fisheries plans to be amended to include information on the habitat requirements of the fish species being managed. Sequential levels of research are now prerequisite to making management decisions: these include estimates of fish abundance, life history, behavior, and recruitment with respect to the habitats that are used (Fluharty 2000). Fisheries scientists soon discovered a lack of knowledge regarding the habits and habitats of many fishes. Conducting the necessary research on fish EFH becomes even more critical in rapidly changing environments like Barataria Bay. The three primary aquatic habitats available to fish in Barataria Bay are mud bottom, marsh edge, and shell habitat.

### **1.1 SHELL HABITAT**

The primary shell habitat in Barataria Bay is oyster shell (*Crassostrea virginica*). Oysters have been a very important fishery in Barataria Bay for well over a century. Between 1958 and 1990, oyster lease acreage in the Barataria-Terrebonne basin increased by 180%, and between 1989 and 1994 the industry made over 18 million dollars per year (Melancon *et al.* 1998). Because it is a take-and-replace industry, the exact nature of individual reefs is always changing, but they may still provide notable habitat to fishes. Avre (1960) reported that oyster plantings did, indeed, attract and concentrate fishes in a formerly “unproductive” area, hypothesizing that the new reef provided better refuge and foraging opportunities than the surrounding bottom with no relief. Lenihan *et al.* (2001) showed that shallow oyster reefs served as refuge to high densities of fish fleeing hypoxic bottom water, but that this behavior was not induced by hypoxia alone.

Harding (2001) found strong seasonal and diel patterns in patchy zooplankton abundances around an oyster reef in a tributary of the Chesapeake Bay. He concluded that the historical decline in oysters in the Chesapeake led to a decline in zooplankton abundance, which in turn further reduced oyster recruitment and benthic complexity. In addition, Harding and Mann (2001) determined that while they could not prove oyster reefs to be EFH, reefs provided a higher quality habitat when compared to sand bars by exhibiting higher production and more optimal growth and survival among fishes.

The great majority of oyster reefs in the Barataria Basin are subtidal. Lehnert and Allen (2002) found that subtidal oysters supported greater abundances of fish than intertidal oyster reefs. This conflicts with Meyer and Townsend's (2000) findings that show intertidal reefs to be just as important to production as subtidal reefs. Regardless, it is obvious that oysters impact the water column and are important to other species, consuming nearly  $45 \text{ Kcal } m^{-2} d^{-1}$  of zooplankton, while blanketing interstitial spaces between shells with filtered detritus ready for consumption by other trophic levels (Dame and Patten 1981, Meyer and Townsend 2000).

The question remains, however, that even if oyster reefs are important to fish production, are there enough of them to help sustain fish populations as the environment changes? Mud bottom and marsh edge are simply much more abundant than oyster reefs on the Louisiana coast, even as the marshes decline.

## **1.2 MARSH EDGE HABITAT**

Baltz *et al.* (1993, 1998) suggested that even as the marshes become more fragmented, the total length of marsh edge increases, temporarily benefiting some species before inland, solid marsh begins to disappear. Baltz (1998) defined the microhabitat

niches that several estuarine fishes occupy in Barataria Bay in response to environmental variables and interactions with other species. They found that the 15 most abundant species were concentrated at the marsh-water interface, and that these were mostly larval and juvenile fish, further defining the importance of marsh edge as a nursery area.

Jones *et al.* (2002) found further evidence for resource partitioning in Barataria Bay waters, particularly between closely related fish species. The partitioning between species was mostly determined by interannual variation of water surface elevation, temperature, and salinity. Population density, individual growth, survival and secondary production were seen to be determined by this environmental variation.

Marsh studies conducted by Minello (1992) revealed that large fish predators directly exploit the marsh edge nurseries by moving into the marsh on a flood tide to feed. Minello (1992) also found that infaunal and epifaunal production at an artificially created marsh were lower than infaunal and epifaunal production at a natural marsh five years after creation. However, after 1-2 growing seasons, fishes and some decapods were at equivalent or higher abundances than at natural marshes. It is clear then, that marsh edge may play an important role in sustaining fish populations.

### **1.3 MUD BOTTOM HABITAT**

Shell and marsh nekton habitats have been studied extensively along the Louisiana coast and elsewhere in U.S., but there has been little mention of potential linkages between soft substrate mud habitat and fish assemblages or fish diets. In a review of European research in estuaries ranging from 36 - 68° N latitude, however, Pihl *et al.* (2002) found that subtidal and intertidal soft substrate habitats appeared to be very important to fish when compared to a number of other microhabitats including: salt



marsh, biogenic reefs (shell), tidal freshwater areas, reed beds, intertidal and subtidal hard substrate, and subtidal sea grass beds. They found that mud habitats consistently had the highest mean number of commercial and non-commercial fishes, and that mud habitats ranked highest according to a habitat importance index that combined species richness and the number of use functions for each fish species. Furthermore, subtidal mud habitats comprised greater than 50% of the total surface area of European estuaries, followed by intertidal mud habitats at 30%. So, while more complex habitats may provide better refuge and nursery areas for young fish, mud habitats may be providing more foraging opportunities because of greater surface area, less competition, and higher densities of benthic macroinvertebrates (Pihl *et al.* 2002). It should be acknowledged, however, that the mud habitats referenced by Pihl *et al.* (2002) are more easily and frequently sampled than other habitats. Regardless, it is clear that mud habitats should not be disregarded when assessing the functional ecology of fishes, especially in Louisiana estuaries as more and more marshlands subside and give way to open water and shallow, mud-bottom habitats.

#### **1.4 SPOTTED SEA TROUT LIFE-HISTORY AND DIET**

Spotted sea trout (*Cynoscion nebulosus*) are very important recreational fish species in Louisiana. The National Marine Fisheries Service estimates the 2002 total catch of Louisiana spotted sea trout at 9,131,830 fish (NMFS, personal communication). Spotted sea trout exhibit bimodal spawning within the estuary, peaking in May, and then again in late July/early August. Photoperiod, temperature and salinity have been implicated as the environmental catalysts for spawning (Hein and Shepard 1979). Hildebrand and Cable (1934) collected spotted sea trout larvae 8-10 km offshore of the

North Carolina coast, Daniels (1977) collected spotted sea trout larvae 6-15 km offshore of the Louisiana coast, and Collins and Finucane (1984) collected spotted sea trout larvae 1-39 km offshore of the Florida Everglades. In each case, though, the larvae were less than 5.0 mm in length, considerably smaller than their estuarine counterparts. Optimal salinities are between 25-35 ppt for larvae, but adult-mediated acclimation makes survival in hypo-saline conditions possible (Banks et al. 1991).

Tabb (1964) reported that spotted sea trout rarely move more than 30 miles, and because most estuaries are isolated by miles of exposed seashore, stocks may be isolated. Recent studies confirmed the existence of subpopulations, or estuary-specific populations, of spotted sea trout (Gold *et al.* 2003, Wiley and Chapman 2003, Bedee *et al.* 2003). Differences in age and growth trends, as well as reproductive behavior, among estuarine subpopulations can be attributed to a few factors: genetic isolation of stocks associated with geographical barriers, differences in habitat or water quality during one or more life stages, and differences in fishing pressure (Gold *et al.* 2003, Wiley and Chapman 2003, Bedee *et al.* 2003). Because of their tolerance of the dynamic environmental conditions in estuaries, spotted sea trout are practically unhindered by predation and competition. But because of the isolation of stocks, they may be vulnerable to the environmental decline of the estuary itself, thus serving as an appropriate indicator species (Bortone 2003).

Spotted sea trout have been found to eat copepods until they reach a standard length of 15 mm (McMichael and Peters 1989), eating more fish than crustaceans as they grow longer than 25 cm (Overstreet and Heard 1982, Hettler 1989). Seasonal shifts also were evident (more fish in the spring and summer, shrimp in the fall, and polychaetes and

crustaceans in the winter) (Overstreet and Heard 1982). In mesocosm experiments, Zimmerman and Minello (1984) show that spotted sea trout ate mostly in the lower half of their potential prey size range, and showed selection by size and behavior of available prey (avoidance, burrowing, and schooling all decreased selection).

Both Chester and Thayer (1990) and Hettler (1989) use food habits and other levels of information to identify seagrass as EFH for spotted sea trout, while Baltz *et al.* (1998) does the same to implicate marsh edge as EFH. Baltz *et al.* (1998) contends that growth is a function of location, with prey choice becoming important during ontogenic shifts. They also report that refugia and foraging sites in Barataria Bay, and throughout Louisiana, overlap.

Minello (1999) reports similar results, finding most juvenile spotted sea trout at submerged aquatic marsh edge, seagrass, and sometimes mixed-vegetation marsh edge. Because it may be shown that all available habitats should be considered EFH for some species, Minello (1999) suggests identifying Habitat Areas of Particular Concern (HAPC) for extra emphasis. He also makes it very clear that densities alone are inadequate for EFH identification, and that EFH must be identified according to the function of the habitat (*i.e.*, habitat-influenced diet).

## **1.5 PINFISH LIFE-HISTORY AND DIET**

Pinfish, *Lagodon rhomboides*, are not nearly as important to Louisiana recreational fishing as spotted sea trout; the National Marine Fisheries Service estimates the 2002 total catch of Louisiana pinfish at 246,596 fish (NMFS, personal communication). With that said, pinfish are extremely abundant and, presumably, have a large impact on the overall energy budget of the estuarine biological

system. Pinfish have also been shown to be voracious foragers, heavily impacting local populations of amphipods and other small crustaceans (Kjelson and Johnson 1976, Montgomery and Targett 1992, Thompson 2000). Because of concerns over the validity of site-specific diet data for transitory fish like spotted sea trout, and because pinfish are believed to demonstrate greater site-fidelity, an examination of habitat-influenced diet for pinfish were included in this study.

Pinfish as young as one year, and as small as 90 mm, leave the estuary each fall to spawn offshore (Hansen 1969), where they are known to school by size-class (Nelson 2002). Adults return to the estuary as soon as November and December, and larvae are transported into the estuary via on-shore currents shortly thereafter. Few fish live to return to the estuary a third year (Hansen 1969, Nelson 1998). Pinfish were previously assumed to spawn exclusively offshore, but Nelson (2002) demonstrated the possibility of some inshore spawning between late fall and early spring in Florida Gulf of Mexico estuaries.

Pinfish move very little during summer from preferred feeding grounds, congregating, but not schooling, around submerged aquatic structure (Hansen 1969, Stoner 1979). Irlandi and Crawford (1997) showed pinfish to rarely move away from intertidal marsh habitat unless bordered by seagrass or shell rubble (*i.e.*, oyster). Minello (1999) reported that pinfish are the dominant fish at most areas of submerged aquatic marsh edge. Jordan *et al.* (1996) found a 760% increase in fish abundance at seagrass beds over that of sand flats, mostly due to pinfish assemblages. Jordan *et al.* (1996) investigated further, showing experimentally that in the absence of predators, pinfish use each habitat equally, even swimming high in the water column. But in the presence of

predators, pinfish exhibit behaviorally mediated predator avoidance, staying away from open sand and open water.

Many investigators have reported a shift in pinfish diet with growth from juvenile to subadult (Hansen 1969, Czapla 1991, Montgomery and Targett 1992, Pike and Lindquist 1994, Thompson 2000). Although pinfish show no cellulase activity in the stomach, and have short guts typical of carnivores, most pinfish begin eating submerged plant material by summer and fall. Earlier in the year, and particularly when they are offshore in winter where submerged vegetation is unavailable, they mostly prey on amphipods, copepods (calanoid), shrimp, and small fish (Czapla 1991).

Growth is generally much faster for smaller size-classes, and slower for all size-classes in winter (Cameron 1969, Thompson 2000). Kjelson and Johnson (1976) showed that pinfish consume much more amphipod biomass than ration estimates based upon metabolic needs suggest, and Thompson (2000) used a bioenergetics model to show that they are capable of consuming amphipods at 20 times the rate that which amphipods are actually present *in situ*. This intense grazing pressure greatly decreases prey availability (it may even help explain the adaptive behavior of epifaunal amphipods), perhaps causing a facultative, not ontogenetic, diet switch to a mixed diet of animal and plant material (Stoner 1982).

Thompson (2000) found plant material less than or equal to 18% of the total diet; Hansen (1969) reports up to 40.6%, and Czapla (1991) finds plant material as high as 50% of the diet. In energetics experiments, Montgomery and Targett (1992) show that pinfish do indeed have the capacity to increase overall consumption and evacuation rate while eating plant material, thus maintaining growth rates and protein consumption.

They contend that there is no sizable difference between this mixed diet and a purely animal diet, in terms of caloric value, and that it may actually be beneficial to the fish because of lower energetic cost of capture. Low gastric pH apparently helps the pinfish to lyse plant cell walls, thus assimilating a substantial portion of the organic matter.

## **1.6 OBJECTIVES**

Because the Barataria Bay salinity gradient, and habitats found along this gradient, are surely to change quickly in the coming years, it is very important to understand now the role of these habitats in their use by fishes. The functional influence on diet that these habitats assert is the subject of this research. Spotted sea trout and pinfish were thus caught over the three different habitats in Barataria Bay, Louisiana (shell, marsh edge, and mud bottom). Spotted sea trout were also caught at three different stations along the north-south salinity gradient in the estuary. The stomach contents of each species were quantitatively examined with respect to habitat, fish size, bay location, season, and other factors. Any variation in the relative importance of particular prey associated with each habitat was quantified according to a number of prey variables (*i.e.*, prey type, abundance, weight, frequency of occurrence, caloric value, and relative importance) and environmental variables (*i.e.*, salinity, temperature, dissolved oxygen).

## **1.7 HYPOTHESES**

H<sub>0</sub> 1: there is no difference in the diet of spotted sea trout according to habitat (oyster reef, marsh edge, and mud bottom) or environmental variables (dissolved oxygen, temperature, salinity).

H<sub>0</sub> 2: there is no difference in the diet of pinfish according to habitat (oyster reef, marsh edge, and mud bottom) or environmental variables (dissolved oxygen, temperature, salinity).

H<sub>1</sub> 1: there is a statistically significant difference in the diet of spotted sea trout according to habitat (oyster reef, marsh edge, and mud bottom) or environmental variables.

H<sub>1</sub> 2: there is a statistically significant difference in the diet of pinfish according to habitat (oyster reef, marsh edge, and mud bottom) environmental variables.

H<sub>1A</sub> 1: marsh edge habitat provides the most beneficial diet, in terms of caloric value, to estuarine spotted sea trout.

H<sub>1A</sub> 2: marsh edge habitat provides the most beneficial diet, in terms of caloric value, to estuarine pinfish.

## CHAPTER 2. METHODS

### 2.1 STUDY SITES

Spotted sea trout were collected at three sites in Barataria Bay between May 2003 and May 2004: *Fisherman's Point* in the northern/upper bay region (lat. 29°31'10", long. 90°08'75"), *Manila Village* in the mid-bay region (lat. 29°25'50", long. 89°58'25"), and *Grand Terre* (lat. 29°17'50", long. 89°55'25"), in the southern/lower bay region. In this manner, habitats along a salinity gradient were represented; mean salinity at *Fisherman's Point* was 4.9 ppt, mean salinity at *Manila Village* was 11.4 ppt, and mean salinity at *Grand Terre* was 19.6 ppt. Within each site, all three habitat types (mud, marsh edge, shell) were sampled. At the *Grand Terre* station, oyster reef is located at the separate, but nearby, *Queen Bess Island* (lat. 29°18'30", long. 89°57'30"). Mean salinity at *Queen Bess* was 19.3 ppt.

Because pinfish were added to the study after sampling had already begun, and because questions of pinfish site-fidelity and habitat use were deemed more important than the role of salinity, *Grand Terre* was chosen as the exclusive site for pinfish sampling.

### 2.2 DATA COLLECTION

#### 2.2.1 Field Sampling

Environmental conditions were measured at each site using a YSI® Model 85/25 probe. Dissolved Oxygen (mg/L), temperature (°C), and salinity (ppt) were recorded and logged.

A 45.7 m gill net was deployed over each habitat type to catch spotted sea trout. All other species of fishes caught in the gill nets were used in a related fish community



and habitat study not discussed in this thesis. The deployment of these nets was staggered in time such that each net was fished for one hour, subsequently cleared of all catch, and fished for an additional hour. The additional hour served as a replicate in time. All nets were set parallel to shore. Each net had five 7.6 m panels that were randomly assigned a series of mesh sizes (2.5, 3.8, 5.1, 6.4, 7.6 cm). The differing mesh sizes were size-selective, allowing for a more diverse species and life-stage catch.

Because pinfish did not regularly get caught in the gill nets, 2 small-mesh (2.2 cm) fish traps were baited with fresh blue crab and set over each habitat type at Grand Terre. These traps were retrieved and emptied at least twice in a total fishing time that was usually 24 hours.

All captured fish were bagged, labeled, and stored on ice in an on-board cooler. The fish were transferred first to a freezer at the LDWF *Lyle S. St. Amant* Grand Terre Laboratory, and then to a -80° C freezer at Louisiana State University. This prevented further digestion of gut contents.

### **2.2.2 Laboratory Analysis**

Fish were thawed and measured for standard length ( $\pm 0.1$  cm) and weight ( $\pm 0.0001$  grams). Stomachs were removed and individually stored in jars of 10% formalin. Because spotted sea trout are highly transient, and only a short amount of time elapsed between capture and freezing, only the stomachs were removed. By not including the contents of the intestines, the stomach contents are presumed to represent only the food eaten over the habitat on which they were captured. Because pinfish are less transient and more time could potentially elapse between capture and freezing, the contents of both the stomach and intestines were examined.

After two days of fixation in formalin, the stomachs were transferred to 70% ethanol for ease of examination. Gut contents were examined under a Olympus® 40x *SZX-ILL D100* microscope equipped with a polarizer for assistance in identifying minute bone and plant material. Prey were identified to the lowest possible taxonomic designation. Prey were then sorted by taxa, placed in aluminum weighing dishes, and dried in a Yamoto® *Dx 600* drying oven at 70° C for at least 24 hours. Lastly, the dry weight ( $\pm 0.0001$ g) of each taxa was obtained using a Precisa® *XB 220A* microbalance.

The unidentified material present in the guts of spotted sea trout collected from February 2004 through May 2004 was further divided into one of the following three categories: plant, detritus, or unidentifiable. Of the aforementioned three categories found in these months, 21.5% was plant, 24.7% was detritus, and 53.8% was unidentifiable material. All material from fish caught before February, 2004 that was recorded as unidentifiable was retroactively delineated by using these percentages.

The unidentified material present in the guts of pinfish collected in August 2004 was further divided into one of the following four categories: plant, soil and sand, detritus, or unidentifiable. Of the aforementioned three categories found in August 2004, 13.34% was plant, 7.72% was soil and sand, 39.89% was detritus, and 59.05% was unidentifiable material. All material from fish before August 2004 that was recorded as unidentifiable was retroactively delineated by using these percentages.

## **2.3 STATISTICAL ANALYSES**

### **2.3.1 Descriptive Statistics**

A number of useful descriptive statistics were calculated for pinfish and spotted sea trout, including a (1) vacuity index, (2) dietary overlap, and (3) dietary

breadth. The vacuity index (VI) was calculated for the total diet, as well as by habitat, using the formula (Labropoulou *et al.* 1997):

$$VI = \frac{\# \text{ of empty stomachs}}{\text{total \# stomachs}} * 100. \quad (1)$$

Dietary overlap based on dry weight was calculated between habitat types and seasons using Schoener's index (Wallace 1981), defined as:

$$\alpha = 1 - 0.5 \left( \sum_{i=1,n} |\rho_{xi} - \rho_{yi}| \right)$$

where :

$\alpha = 0 \rightarrow 1$  ; 1 = complete overlap;

$\rho_{xi}$  = the proportion of prey  $i$  in diet  
of fish over habitat (or fish in season)  $x$ ; and,

$\rho_{yi}$  = the proportion of prey  $i$  in diet  
of fish over habitat (or fish in season)  $y$ .

(2)

Schoener's index ranges from 0 (no overlap) to 1.0 (total overlap). Zaret and Rand (1971) suggest that a value greater than 0.6 is considered significant overlap. To determine dietary breadth, a measure of how narrow or broad a diet is, a Shannon Diversity Index (Brower *et al.* 1997) based on dry weight was calculated for total diet, habitat, and season, using the equation:

$$H' = - \sum p_i \log p_i,$$

where :

$p_i$  = the proportion of the  $i_{th}$  food item,  
based on dry weight.

(3)

To evaluate the amount of food in the guts the following ratio (referred to as dry weight prey / fish dry weight, or DW/FW), original to this study, was constructed for each individual fish, with means calculated for the total diet and diet by habitat:

$$DW/FW = \frac{1}{P} * \sum_{j=1}^P \frac{DW_j}{FW_j}, \quad (4)$$

where: j = stomach number;

P = number of stomachs in specific habitat;

DW<sub>j</sub> = total dry weight (g) of all prey in stomach j; and,

FW<sub>j</sub> = weight of the fish (g) corresponding to stomach j.

Prey items were organized into logical categories, and the relative contribution (*i.e.*, importance) of each category was determined by using the following descriptive statistic indices: (5) percent composition dry weight (%DW), (6) percent frequency of occurrence (%FO), (7) Index of Caloric Importance (ICI), and (8) the Prey Importance Index (PI). Each of these indices was calculated for the total diet, diet by habitat, and diet by season. Percent dry weight and percent frequency of occurrence (Bowen 1996) are defined as:

$$\%DW = \frac{1}{P} \sum_{j=1}^P \left( \frac{W_{ij}}{\sum_{i=1}^Q W_{ij}} \right) * 100, \quad (5)$$

where i = prey category;

j = stomach number;

P = number of stomachs with prey;

W<sub>i</sub> = dry weight (g) of prey category; and,

Q = number of prey categories.

$$\%FO = \frac{\# \text{ stomachs with prey category } x}{\# \text{ stomachs with any prey}} * 100. \quad (6)$$

The caloric densities (calories / g) of major prey items found in spotted sea trout and pinfish were derived from previous studies. The index of caloric importance (McCawley 2003) could then be calculated using the formula:

$$ICI = (\%DW * C) * \%FO, \text{ and} \quad (7)$$

$$\%ICI = \frac{ICI \text{ for prey category } x}{\text{sum of all ICI values}} * 100,$$

where: C = calories per gram dry weight of prey category.

The prey importance index as defined by Pope *et al.* (2001) also was calculated:

$$PI = \frac{1}{P} \sum_{j=1}^P \left( \frac{W_{ij}C_i}{\sum_{i=1}^Q W_{ij}C_i} \right) * 100 \quad (8)$$

where i = prey category;

j = stomach number;

$W_{ij}$  = dry weight (g) of prey category  $i$  in stomach  $j$  ;

$C_i$  = caloric density (calories per gram) of prey category  $i$  ;

P = number of stomachs with prey; and,

Q = number of prey categories.

Hyslop (1980) comments that gravimetric measures (and the use of formalin) tend to overemphasize the importance of heavy items, while frequency measures often overemphasize the importance of numerous, but insubstantial (by weight or caloric value), items. The index of caloric importance and the prey importance index

are perhaps more valuable because they incorporate both gravimetric and frequency measures, as well as caloric values that reflect potential to influence growth rate. All of these indices are included, however, to facilitate comparisons with other literature.

### 2.3.2 Parametric Statistics

Analysis of Variance (ANOVA) was used to test for differences in the overall diets of spotted sea trout, and differences in the overall diets of pinfish. Data were entered into the statistical software package Statview©, with each fish stomach acting as a replicate. Three levels of information for each fish were entered as real numbers: the total dry weight of prey in the stomach, the prey dry weight multiplied by the average caloric value (calories per gram) of the prey in the stomach, and the stomach fullness (9). Stomach fullness is defined by Duarte and Garcia (1999) as:

$$\text{Fullness}_j = \frac{(PW_j / FW_j)}{P / F_{\max}}, \quad (9)$$

where:  $j$  = fish number;

$PW_j$  = cumulative dry weight of prey in stomach of fish  $j$ ;

$FW_j$  = dry weight of fish  $j$ ; and,

$P/F_{\max}$  = greatest ratio of prey weight to fish weight.

A one-way ANOVA was then run for each of three effects (entered as coded integers): 1) habitat (shell, marsh, mud); 2) season (spring, summer, fall, winter); and, 3) bay location, a measure of relative salinity (lower bay = high salinity, middle bay = moderate salinity, upper bay = low salinity), for spotted sea trout. Similarly, ANOVA also was performed with pinfish diet data with the following main effects: 1) habitat (shell, marsh, mud); 2) season (summer, fall); and, 3) time of day (day, night). A two-

way ANOVA, taking into account possible interactions between main effects, could not be performed because there were too many singularities in the matrix. To account for experimental-wide error, a Bonferonni adjustment was made to the acceptable p-value produced by the ANOVA F-test. In ecological diet studies,  $\alpha = .10$  is generally acceptable (Hyslop 1980), but with the Bonferonni adjustment ( $\alpha / k$ , where  $k$  = the number of tests),  $\alpha = 0.10 / 3 = 0.03$ . Thus, if the habitat, season, bay location, or time of day p-values for prey dry weight, dry weight x caloric value, or stomach fullness are 0.03 or less, the null hypotheses is rejected, and there is an acceptably low probability that the measured differences could have happened by chance.

### **2.3.3 Non-parametric Statistics**

Non-parametric statistical analyses often are better for diet studies than traditional parametric analyses because diet data contain many zeros, and rarely meet the normality assumptions required in parametric statistics. For this reason, Analysis of Similarities (ANOSIM), a non-parametric multivariate test conducted at a significance level of 0.10, was used to analyze the diets of spotted sea trout and pinfish separately.

ANOSIM was conducted using the PRIMER-5 statistical software package. As in the parametric tests, each fish stomach is a replicate, but data in PRIMER are entered exclusively as percent composition by dry weight of all prey items. The raw data underwent a square root transformation to bring it closer to normality, and a matrix was then built using Bray-Curtis similarity coefficients. In this manner, the contents of each stomach are compared to every other stomach (Clarke and Gorley 2001).

For spotted sea trout, the following effects were subject to one-way ANOSIM tests: 1) habitat (shell, marsh, mud); 2) season (spring, summer, fall, winter); 3) site,

coded for specific site and habitat combinations (*i.e.*, Grand Terre / marsh = GTMA); 4) bay location, a measure of relative salinity (lower bay = high salinity, middle bay = moderate salinity, upper bay = low salinity); and, 5) fish size (coded 1, 2, 3, 4; based upon intervals of 10 cm). In addition, two-way crossed ANOSIM tests were performed with the habitat effect run in combination with other effects, thus allowing for analyses of interactions.

For pinfish, the following effects were subject to one-way ANOSIM tests:

1) habitat (shell, marsh, mud); 2) season (summer, fall); 3) time (day, night); and, 4. fish size (coded 1, 2, 3, 4; based on intervals of 3 cm). In addition, two-way crossed ANOSIM tests were performed with the habitat effect run in combination with other effects, thus allowing for analyses of interactions.

ANOSIM constructs a similarity matrix and produces an R-statistic which describes the extent of dissimilarity in diet composition among the specified effects. If there are no differences between groups, then between-group similarities and within-group similarities will be roughly equal. An R-statistic is never more than about 0.15 by chance, hence if  $R > 0.15$ , the null hypothesis that no differences between groups can be rejected at the 0.001 (or 0.1%) level. A Significance Level Sample Statistic is also produced, which calculates percent correspondence (Clarke and Gorley 2001). A 'Global R' and Significance Level (%) is calculated for the entire data set, and an R-statistic and Significance Level (%) is also calculated for each pair-wise comparison.

To see if the organization of the data matrix had any impact on the results of the ANOSIM tests, the analyses were run with several different matrix arrangements, including: 1) unaltered original data; 2) data on unidentified material delineated as



described in section 2.2.2 of this thesis; 3) data consolidated into a shorter list of prey types, summing all fish dry weights and excluding very rare items; and, 4) data excluding bait (pinfish only). The consolidated list of prey types for spotted sea trout consisted of the following: fish, fish artifacts (otoliths and crops), brown shrimp, fiddler crab, polychaete, plant, detritus, and unidentified material. The consolidated list of prey types for pinfish consisted of the following: fish, shrimp, crab, plant, soil, detritus, and unidentifiable material. Adjusted data (# 2 above) is referred to as 'baseline' data because it is the primary data set used in all initial calculations and statistical tests.

Multi-Dimensional Scaling (MDS) also was performed using PRIMER. Multi-Dimensional Scaling constructs a map, or configuration in a specified number of dimensions that visually displays the ranking of the similarity matrix with the greatest 'goodness of fit', or lowest stress. This provided a snapshot of the variability in the diets of both spotted sea trout and pinfish.

In instances where ANOSIM discovered significant dissimilarities, a number of post-ANOSIM tests were available in PRIMER. SIMPER, or 'similarity percentage', was performed on the original data, providing a ranking that shows which prey items contributed most by percentage to the similarity in a within group test, or the percentage of dissimilarity contributed to a between group test. BV Step uses the Spearman Rank Correlation in a forward and backward iteration that systematically calculates the variance explained by one prey category, then adding another and another, recalculating the variance each time. The output is a specific set of prey that explains the highest percentage of variability (Clarke and Gorley 2001).

Finally, PRIMER was used to test if water-quality influenced the overall diet matrix. The BIO-ENV program tests the biotic matrix already constructed in the previous ANOSIM analyses against an abiotic environmental matrix, containing in this case data on dissolved oxygen (mg/L), salinity (ppt), and temperature (°C). The biotic matrix is fourth-root transformed using Bray-Curtis similarities between samples, and the abiotic matrix is transformed using Normalized Euclidean Distances. Spearman Rank Correlation then compares the two matrices, resulting in the output of the best abiotic variable or suite of abiotic variables to account for the most pattern in the data, as seen earlier in the MDS plot. A Correlation Statistic ranging from 0 to 1.0 is reported, with 1.0 resulting when 100% of the pattern explained.

### CHAPTER 3. ENVIRONMENTAL RESULTS

Salinities at the three sampling stations were characteristic of their location within the Barataria Bay north-south salinity gradient. Mean salinity at *Fisherman's Point* in upper Barataria Bay was  $4.8 \pm 3.7$  ppt; mean salinity at *Manila Village* in the mid-bay region was  $11.4 \pm 5.0$  ppt; and, mean salinity at *Grand Terre* in lower Barataria Bay was  $19.6 \pm 5.0$  ppt. Salinities were highest during fall, and lowest during late winter and mid-summer (see Figure 1).

Dissolved oxygen values were similar among the three stations along the north-south salinity gradient, peaking during the winter months (see Figure 2). Mean dissolved oxygen at *Fisherman's Point* in upper Barataria Bay was  $7.2 \pm 1.6$  mg/L; mean dissolved oxygen at *Manila Village* in the mid-bay region was  $7.2 \pm 1.5$  mg/L; and, mean dissolved oxygen at *Grand Terre* in lower Barataria Bay was  $7.6 \pm 1.7$  mg/L.

Temperature values reflected a typical seasonal pattern declining in the winter months and rising during spring (see Figure 3). Mean temperature at *Fisherman's Point* in upper Barataria Bay was  $23.9 \pm 6.9$  °C; mean temperature at *Manila Village* in the mid-bay region was  $24.1 \pm 6.1$  °C; and, mean temperature at *Grand Terre* in lower Barataria Bay was  $24.7 \pm 6.0$  °C.

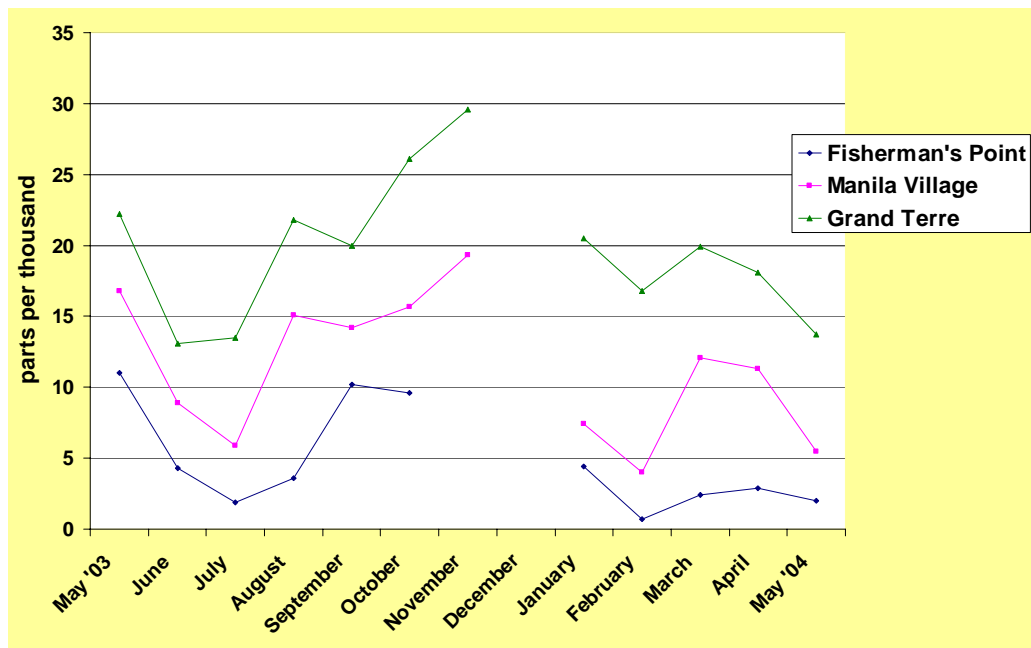


Figure 1. Monthly salinities (ppt) at each station along the north-south Barataria Bay salinity gradient between May of 2003 and May of 2004.

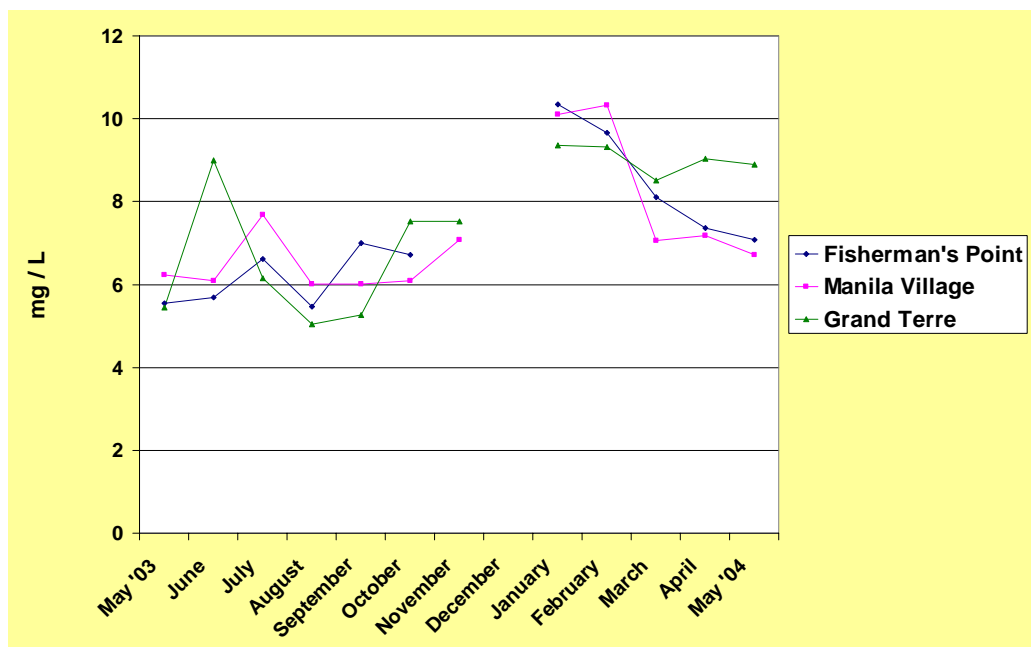


Figure 2. Monthly dissolved oxygen (mg/L) at each station along the north-south Barataria Bay gradient between May of 2003 and May of 2004.

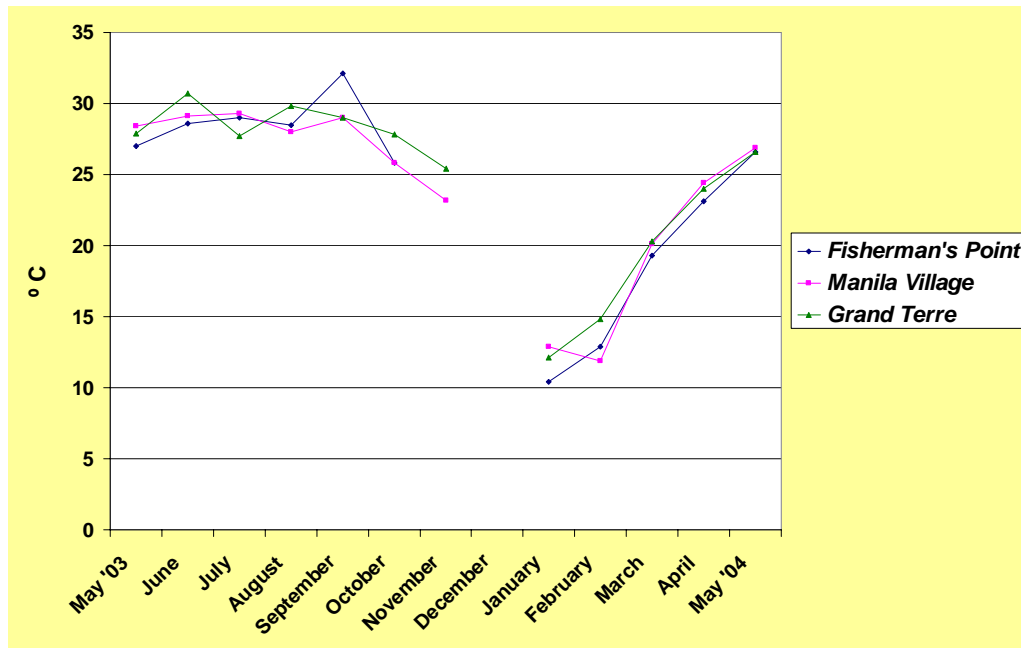


Figure 3. Monthly water temperatures (°C) at each station along the north-south Barataria Bay gradient between May of 2003 and May of 2004.

## **CHAPTER 4. SPOTTED SEA TROUT RESULTS**

### **4.1 CATCH DATA**

Monthly sampling was conducted for a total of twelve months, beginning in May of 2003 and ending in May of 2004. No samples were collected in December 2003 because of inclement weather. A total of 175 spotted sea trout stomachs were examined. The number of fish caught was highest in August, declining to low catches between October and March, rising again in April (see Figure 4). A total of 69 fish were caught over shell habitats, 42 on the marsh edge, and 64 over mud habitats. When catch data are broken down by habitat, the seasonal trend of high numbers in summer and low numbers in winter is evident (see Figure 5), with the exception of an unusually high catch of 26 catch on the mud habitats in August, 2003.

### **4.2 PREY TYPES AND CALORIC VALUES**

Seventeen prey types were identified in the guts of spotted sea trout. The vertebrate types included: unidentifiable fish, bay anchovy (*Anchoa mitchilli*), threadfin shad (*Dorosoma petenense*), Gulf menhaden (*Brevoortia patronus*), Atlantic croaker (*Micropogonias undulatus*), fish crops, and fish otoliths. Fish crops were combined with the unidentifiable fish prey category in descriptive statistics in this study. They account for a small fraction of this category, but should be excluded altogether from future analyses because they are digested much slower than other animal material and may not be representative of the prey consumed over any specific habitat. Invertebrate prey types included: brown shrimp (*Farfantepenaeus aztecus*), fiddler crab (genus *Uca*), amphipods (fam. *Gammarus* and *Melitidae*), Nereid polychaetes (fam. *Nereidae*), and a Nerite gastropod (fam. *Neritidae*). The brown shrimp prey type in this study is actually

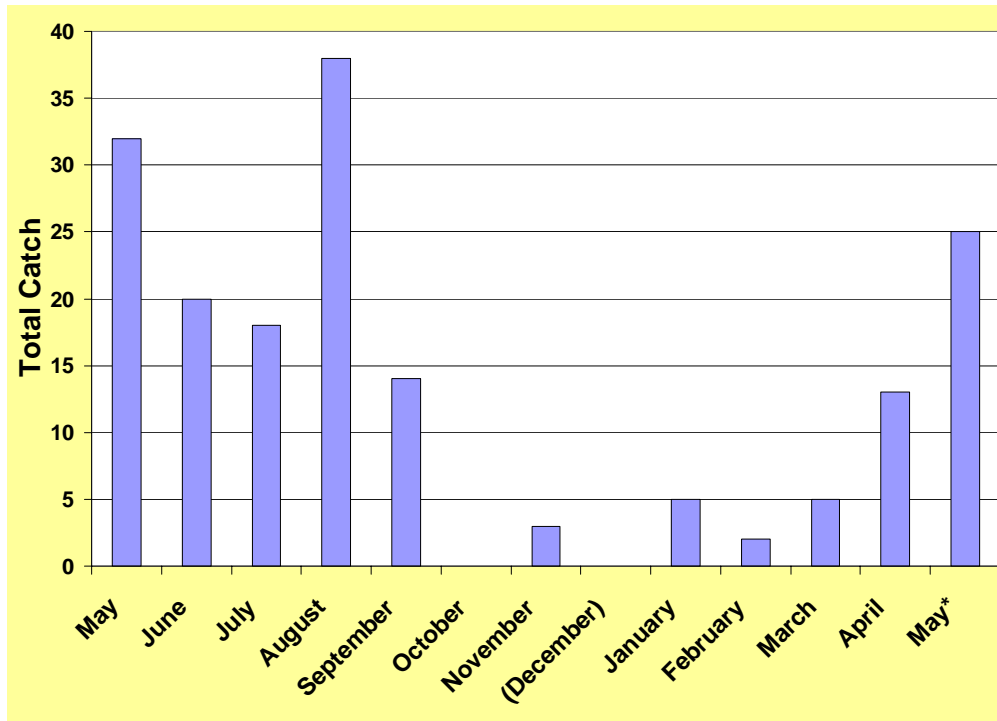


Figure 4. Total catch of spotted sea trout, May 2003 to May 2004. There was no sampling trip in December due to inclement weather.

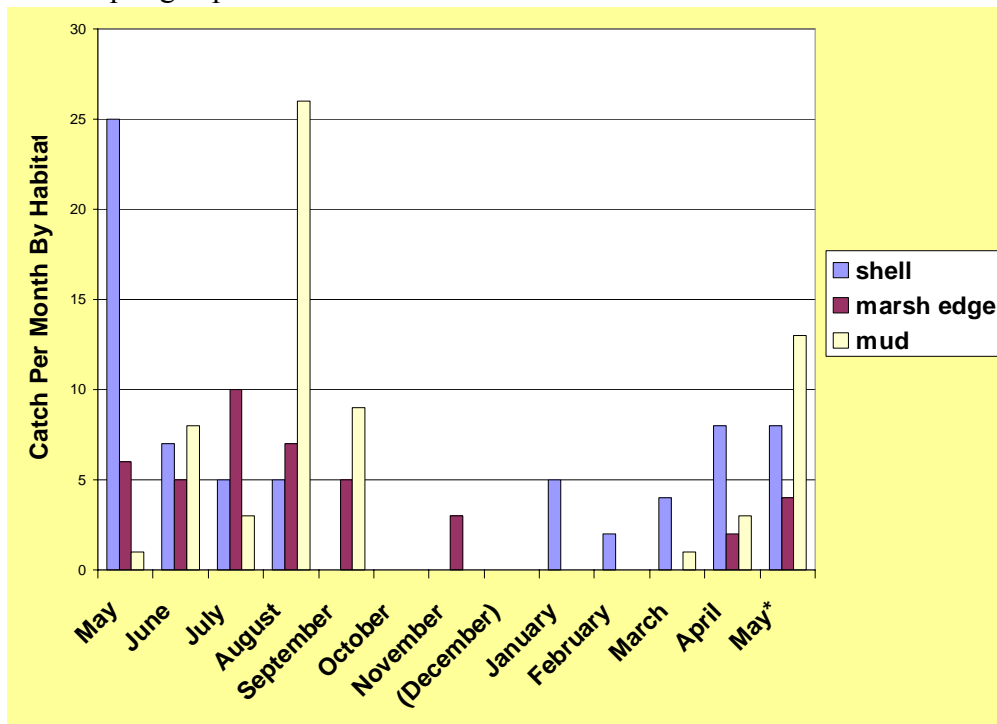


Figure 5. Total catch, by habitat, of spotted sea trout, May 2003 to May 2004.

the combination of identifiable brown shrimp and unidentifiable penaeid shrimp and will be renamed the ‘penaeid shrimp’ category in any future publication. The remaining prey types included: plant, seeds, detritus, and unidentifiable material. The plant material and seeds are both presumed to be entirely *Spartina alterniflora*. The caloric values of all prey types, with the exception of those categories that have no identifiable caloric value (fish otoliths and unidentifiable material), were found in the literature (see Table 1).

### **4.3 RESULTS BY DRY WEIGHT**

#### **4.3.1 Relative Contribution of Prey Types by Percent Dry Weight**

Percent dry weight, calculated for the overall spotted sea trout diet, revealed that fish and fish crops, combined into a single category, made up for the greatest portion by weight at 26.5%, followed by brown shrimp (22.3%), threadfin shad (12.8%), Atlantic croaker (9.1%), gulf menhaden (7.1%), and bay anchovy (5.8%). All other prey types made up less than 5% DW.

Broken down by habitat, the top three prey types by %DW over shell were fish and fish crops (41.6%), gulf menhaden (25.1%), and brown shrimp (17.0%). The top three prey types by %DW along the marsh edge were threadfin shad (42.4%), brown shrimp (27.1%), and fish and fish crops (12.1%). The top three prey types by %DW over mud were fish and fish crops (32.4%), brown shrimp (24.3%), and Atlantic croaker (21.9%) (Table 2).

When considered by season, the top three prey types by %DW were brown shrimp (34.0%), Atlantic croaker (21.9%), and fish and fish crops (17.7%) in spring; fish and fish crops (33.2%), threadfin shad (30.8%), and bay anchovy (14.4%) in summer;



Table 1. Caloric values of spotted sea trout prey items (calories per gram dry weight) and their sources in the literature.

Prey Types	Caloric Value (cal / g DW)	Source
Fish & Fish Crops	5,014	Wissing <i>et al.</i> 1973
Bay Anchovy	5,395	Wissing <i>et al.</i> 1973
Threadfin Shad	5,068	Eggleton <i>et al.</i> 2002
Gulf Menhaden	5,376	Thayer <i>et al.</i> 1973
Atlantic Croaker	4,638	Thayer <i>et al.</i> 1973
Brown Shrimp	4,749	McCawley, J. 2003
Fiddler Crab	2,405	Cummins & Wuycheck 1971
Amphipod	3,635	Wissing <i>et al.</i> 1973
Polychaete	4,587	Cummins & Wuycheck 1971
Gastropod	4,595	Cummins & Wuycheck 1971
Plant & Seeds	4,196	Cummins & Wuycheck 1971
Detritus	2,229	Cummins & Wuycheck 1971
Fish	5,014	Wissing <i>et al.</i> 1973
Invertebrates	3,820	Wissing <i>et al.</i> 1973
Plant & Seeds	4,196	Cummins & Wuycheck 1971
Detritus	2,229	Cummins & Wuycheck 1971

Table 2. Percent dry weight by habitat for spotted sea trout prey.

Prey Types	Total %DW	%DW Shell	%DW Marsh Edge	%DW Mud
Fish & Fish Crops	26.5	41.56	12.07	32.42
Bay Anchovy	5.8	6.1	5.7	10.7
Threadfin Shad	12.8	0	42.4	0
Gulf Menhaden	7.1	25.1	0	3
Atlantic Croaker	9.1	0	0	21.9
Brown Shrimp	22.3	17	27.1	24.3
Fiddler Crab	0.5	0	7.7	0.1
Amphipod	0.1	0.1	0	0.1
Polychaete	0.9	3.8	0	0
Gastropod	1	0	0	2.5
Plant & Seeds	3.1	4.8	2.2	2.3
Detritus	1.7	1.6	0	2.8

Table 3. Percent dry weight by season for spotted sea trout prey.

Prey Types	Total %DW	%DW Spring	%DW Summer	%DW Fall	%DW Winter
Fish & Fish Crops	26.5	17.7	33.2	60.4	46.1
Bay Anchovy	5.8	4.2	14.1	0	0
Threadfin Shad	12.8	0	30.8	0	0
Menhaden	7.1	17.0	0	0	0
Atlantic Croaker	9.1	21.9	0	0	0
Brown Shrimp	22.3	34.0	13.3	31.2	0
Fiddler Crab	0.5	0	1.2	0	0
Amphipod	0.1	0	0.2	0	0
Polychaete	0.9	0	0	0	53.4
Gastropod	1.0	0	2.5	0	0
Plant & Seeds	3.1	3.4	2.3	6.3	0.3
Detritus	1.7	1.8	2.6	2.1	0.2

fish and fish crops (60.4%), brown shrimp (31.2%), and plant / seeds (6.3%) in fall; and, polychaete (53.4%), fish / fish crops (46.1%), and detritus (0.2%) in winter (Table 3).

Unidentifiable fish and brown shrimp consistently made up the bulk of prey %DW by habitat and most seasons, augmented occasionally by heavy items that appeared only once or twice in the study (*i.e.*, threadfin shad, Atlantic croaker, polychaetes, and Gulf menhaden).

#### **4.3.2 Dietary Overlap**

Dietary overlap based upon prey dry weight for each habitat was calculated using Schoener's index and ranged from  $\alpha = 0.42$  to 0.63. Dietary overlap was highest for fish collected over shell vs. mud ( $\alpha = 0.63$ ) and lowest for fish collected over shell vs. marsh edge ( $\alpha = 0.42$ ). By season, dietary overlap ranged from  $\alpha = 0.18$  to 0.51. Overlap was highest for fish collected in summer vs. fall ( $\alpha = 0.51$ ) and lowest for fish collected in winter vs. spring ( $\alpha = 0.18$ ) (see Table 4).

#### **4.3.3 Dietary Breadth**

Dietary breadth, a measure of how relatively narrow or broad a diet is, was determined by using the Shannon Diversity Index and was 0.79 for all spotted sea trout combined based upon dry weight of prey. Broken down by habitat, dietary breadth ranged from 0.62 to 0.73. Dietary breadth was highest for fish collected over mud (0.73) and lowest for fish collected near the marsh edge (0.62). By season, dietary breadth ranged from 0.31 to 0.71. Dietary breadth was highest for fish collected in spring (0.71) and lowest for fish collected in winter (0.31) (see Table 5).

Table 4. Dietary overlap by habitat and season for spotted sea trout.

Habitat Types and Seasons	Schoener's Index ( $\alpha$ )
shell vs. marsh edge	0.42
shell vs. mud	0.63
marsh edge vs. mud	0.48
spring vs. summer	0.40
summer vs. fall	0.51
fall vs. winter	0.47
winter vs. spring	0.18

Table 5. Dietary breadth by habitat and season for spotted sea trout.

Habitat Types and Seasons	Shannon Diversity Index ( $H'$ )
Total	0.79
shell	0.66
marsh edge	0.62
mud	0.73
spring	0.71
summer	0.70
fall	0.40
winter	0.31

Table 6. Ratio of prey dry weight to spotted sea trout dry weight by habitat and season.

Habitat Types and Seasons	* multiply by $10^{-4}$
Total	6.3
shell	3.0
marsh edge	11.7
mud	6.4
spring	4.9
summer	5.7
fall	18.4
winter	3.2

#### **4.3.4 Proportion of Prey Dry Weight to Spotted Sea Trout Dry Weight**

The overall DW/FW ratio, a preliminary measure of the amount of food by ratio ranged from  $3.0 \times 10^{-4}$  to  $11.6 \times 10^{-4}$ . By habitat, DW/FW ranged from  $3.0 \times 10^{-4}$  to  $11.6 \times 10^{-4}$ . The greatest amount of food by weight was consumed at the marsh edge ( $11.6 \times 10^{-4}$ ), and the least amount of food by weight was consumed over shell ( $3.0 \times 10^{-4}$ ). By season, DW/FW ranged from  $3.2 \times 10^{-4}$  to  $18.4 \times 10^{-4}$ . The greatest amount of food by weight was consumed in fall ( $18.4 \times 10^{-4}$ ), and the least amount of food by weight was consumed in winter ( $3.2 \times 10^{-4}$ ) (see Table 6).

#### **4.3.5 Statistical Analysis of Prey Dry Weight and Stomach Fullness**

No statistical significance was found when testing the effects of habitat, season, and bay location on DW, DW\*CAL, and stomach fullness (one-way ANOVA,  $p > 0.03$ ) (see Table 7). As such the null hypothesis that there is no difference in DW, DW\*CAL, and stomach fullness according to the main effects could not be rejected.

#### **4.3.6 Statistical Analysis of Overall Spotted Sea Trout Diet**

No statistical significance was found when using ANOSIM to test the diet of spotted sea trout using baseline data against the main effects: habitat, season, site, bay location, and fish size. None of the R-values were greater than 0.15 (see Table 8), so the null hypothesis that there are no differences between groups could not be rejected at the 0.1% level. Because none of the one-way ANOSIM tests were significant, two-way ANOSIM tests for interactions and post-hoc tests (*i.e.*, BVStep and SIMPER) were not necessary. None of the changes in the data matrix [*i.e.*, 1) unaltered original data; and, 2) consolidated data] produced results that were inconsistent with results from baseline data.

Table 7. P-values for one-way ANOVA by habitat, season, and bay location for spotted sea trout dry weight prey (DW), DW x mean calories (cal / g) per stomach, and stomach fullness.

	Habitat	Season	Bay Location
DW	0.32	0.74	0.18
DW*CAL	0.44	0.11	0.83
Stomach Fullness	0.07	0.76	0.21

Table 8. One-way ANOSIM Global R-statistic values for spotted sea trout diet by habitat, season, site, bay location, and fish size.  $R > 0.15$  = significant at  $\alpha = 0.1$ .

Main Effects	ANOSIM (R)
Habitat	-0.002
Season	0.043
Site	-0.028
Bay Location	-0.026
Fish Size	0.061

The MDS plot showed that there was very little variation in the diet of spotted sea trout (see Figure 6). Plotted numbers correspond to the identification number of the individual fish and the low stress value of 0.01 means that this plot has the best possible ‘goodness-of-fit’. With the exception of a few outliers that contained rare prey items, all the values are clumped together, indicating that the overall contents of the stomachs were quite similar.

#### **4.3.7 Statistical Analysis of Spotted Sea Trout Diet and Water Quality**

The non-parametric statistical tool BIO-ENV was used to examine spotted sea trout diet and water quality by using Spearman Rank Correlation to compare the diet matrix constructed in the ANOSIM tests with a separate abiotic matrix containing data on dissolved oxygen (mg/L), salinity (ppt), and temperature (°C). BIO-ENV showed that the suite of variables that best ‘explained’ the pattern seen in the MDS plot included all three variables: dissolved oxygen, salinity, and temperature. However, the correlation factor that ranges between 0 and 1.0 was very low, at 0.08. Thus, no clear relationship between the prey dry weight diet matrix and environmental effects could be identified.

### **4.4 RESULTS BY FREQUENCY**

#### **4.4.1 Relative Contribution of Prey Types by Percent Frequency of Occurrence**

Percent frequency of occurrence, calculated for the overall spotted sea trout diet, revealed that fish and fish crops, combined into a single category, made up for the greatest percentage by frequency at 53.2%, followed by (combined into a single category) plant material and seeds (38.0%), brown shrimp (25.3%), detritus (21.5%), and bay anchovy (8.9%). All other prey types made up less than 5% FO.

*trout unadjusted*

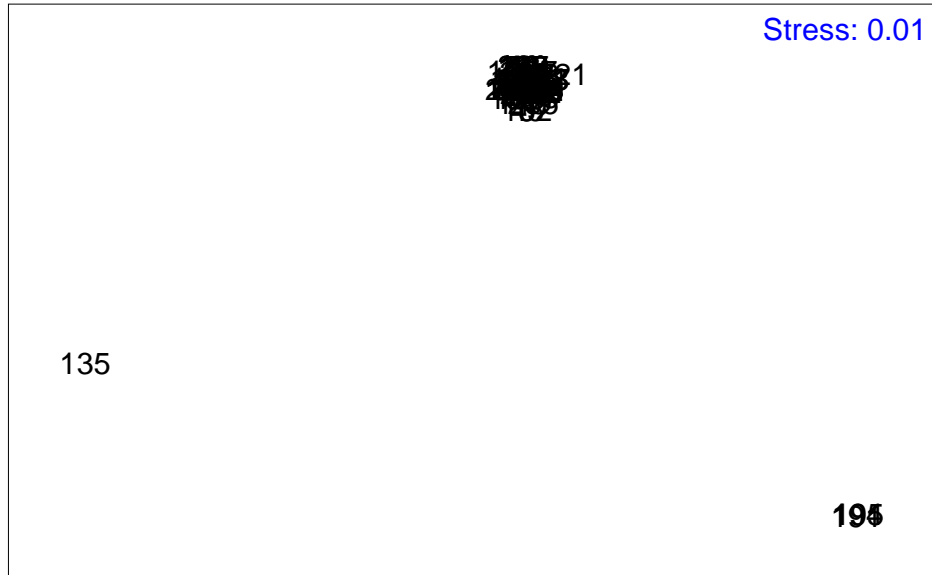


Figure 6. Multi-Dimensional Scaling plot for spotted sea trout.



Broken down by habitat, the top three prey types by %FO over shell were fish and fish crops (37.5%), plant and seeds (31.3%), and detritus (18.8%). The top three prey types by %FO along the marsh edge were plant and seeds (42.1%), fish and fish crops (36.8%), and brown shrimp (36.8%). The top three prey types by %FO over mud were fish and fish crops (53.6%), plant and seeds (46.4%), and detritus (42.9%) (Table 9).

When considered by season, the top three prey types by %FO were fish and fish crops (45.2%), brown shrimp (41.9%), and plant and seeds (25.8%) in spring; plant and seeds (56.3%), fish and fish crops (46.9%), and detritus (31.3%) in summer; plant and seeds (63.6%), fish and fish crops (45.5%), and detritus (35.4%) in fall; and, polychaete (60.0%), plant and seeds (40.0%), and fish and fish crops (20.0%) in winter (see Table 10). Unidentifiable fish, brown shrimp, plant material, and detritus were consistently the most frequently occurring prey items in the spotted sea trout diet.

#### **4.4.2 Vacuity Index**

The total vacuity index (VI) for spotted sea trout was 52.5%. Despite high vacuity, there was no obvious regurgitation as described in Treasurer (1988). Broken down by habitat, VI ranged from 12.4% to 20.9%. The lowest vacuity occurred along the marsh edge (12.4%), and the highest vacuity occurred over shell (20.9%). By season, VI ranged from 29.6% to 58.7%. The lowest vacuity occurred in winter (29.6%), and the highest vacuity occurred in summer (58.7%) (see Table 11).

### **4.5 RESULTS COMBINING WEIGHT, FREQUENCY, AND CALORIC VALUE**

#### **4.5.1 Relative Contribution of Prey Types by the Prey Importance Index**

The Prey Importance Index, calculated for the overall spotted sea trout diet, revealed that fish and fish crops, combined into a single category, made up for the

Table 9. Percent frequency of occurrence by habitat for spotted sea trout prey.

Prey Types	Total %FO	%FO Shell	%FO Marsh Edge	%FO Mud
Fish & Fish Crops	53.2	37.5	36.8	53.6
Bay Anchovy	8.9	6.3	5.3	110.7
Threadfin Shad	1.3	0	5.3	0
Menhaden	2.5	3.1	0	3.6
Atlantic Croaker	1.3	0	0	3.6
Brown Shrimp	25.3	25	36.8	21.4
Fiddler Crab	3.8	0	10.5	3.6
Amphipod	2.5	3.1	0	3.6
Polychaete	3.8	9.4	0	0
Gastropod	1.3	0	0	3.6
Plant & Seeds	38	31.3	42.1	46.4
Detritus	21.5	18.8	21.1	42.9

Table 10. Percent frequency of occurrence by season for spotted sea trout prey.

Prey Types	Total % FO	%FO Spring	%FO Summer	%FO Fall	%FO Winter
Fish & Fish Crops	53.2	45.2	46.9	45.5	20.0
Bay Anchovy	8.9	3.2	15.6	0	0
Threadfin Shad	1.3	0	3.1	0	0
Menhaden	2.5	6.5	0	0	0
Atlantic Croaker	1.3	3.2	0	0	0
Brown Shrimp	25.3	42.0	15.6	27.3	0
Fiddler Crab	3.8	0	6.3	0	0
Amphipod	2.5	0	6.3	0	0
Polychaete	3.8	0	0	0	60.0
Gastropod	1.3	0	6.3	0	0
Plant & Seeds	38.0	25.8	56.3	63.6	40.0
Detritus	21.5	19.4	31.3	36.4	20.0

Table 11. Vacuity Index by habitat and season for spotted sea trout.

Habitat Types and Seasons	%
Total	52.5
shell	20.9
marsh edge	12.4
mud	19.2
spring	58.7
summer	57.9
fall	35.3
winter	28.6

greatest portion of the index at 32.1%, followed by brown shrimp (26.2%), and plant material and seeds, when combined into a single category (15.4%). All other prey types made up less than 5% PI. Broken down by habitat, the top three prey types by PI over shell were fish and fish crops (33.5%), brown shrimp (27.4%), and plant and seeds (14.4%). The top three prey types by PI along the marsh edge were fish and fish crops (26.8%), brown shrimp (36.2%), and plant and seeds (13.4%). The top three prey types by PI over mud were fish and fish crops (34.0%), brown shrimp (18.7%), and plant and seeds (17.7%) (see Table 12).

When considered by season, the top three prey types by PI were brown shrimp (41.9%), fish and fish crops (23.9%), and plant and seeds (13.6%) in spring; fish and fish crops (34.3%), brown shrimp (16.6%), and bay anchovy (14.7%) in summer; fish and fish crops (34.8%), brown shrimp (33.2%), and plant and seeds (21.7%) in fall; and, polychaetes (59.1%), fish and fish crops (20.0%), and plant and seeds (12.7%) in winter (see Table 13).

Unidentifiable fish, brown shrimp, and plant material consistently made up the bulk of PI percentages by habitat and most seasons, augmented by a few seasonally occurring prey items (*i.e.*, bay anchovy and polychaetes). Although plant material does not make up a large proportion of the spotted sea trout diet by weight, it does occur frequently and has a relatively high caloric value (see Table 1). Thus plant material, probably ingested incidentally, is determined by the Prey Importance Index to be relatively important to the spotted sea trout diet.

Table 12. Prey Importance Index by habitat for spotted sea trout prey.

Prey Types	Total PI	PI - Shell	PI - Marsh Edge	PI - Mud
Fish & Fish Crops	32.1	33.5	26.8	33.5
Bay Anchovy	6.4	6	2.2	6
Threadfin Shad	1.3	0	5.6	0
Menhaden	2.4	3.5	0	3.5
Atlantic Croaker	0.7	0	0	0
Brown Shrimp	26.2	27.4	36.2	27.4
Fiddler Crab	2.5	0	10.3	0
Amphipod	0.03	0.05	0	0.05
Polychaete	3.9	10.2	0	10.2
Gastropod	1.3	0	0	0
Plant & Seeds	15.4	14.4	13.4	14.4
Detritus	6.3	4.2	2.7	4.2

Table 13. Prey Importance Index by season for spotted sea trout prey.

Prey Types	Total PI	PI Spring	PI Summer	PI Fall	PI Winter
Fish & Fish Crops	32.07	23.9	34.33	34.83	19.97
Bay Anchovy	6.43	1.44	14.73	0	0
Threadfin Shad	1.33	0	3.33	0	0
Menhaden	2.42	6.48	0	0	0
Atlantic Croaker	0.73	1.97	0	0	0
Brown Shrimp	26.24	41.89	16.55	33.21	0
Fiddler Crab	2.47	0	6.18	0	0
Amphipod	0.03	0	0.03	0	0
Polychaete	3.94	0	0	0	59.08
Gastropod	1.25	0	0.03	0	0
Plant & Seeds	15.39	17.2	13.63	21.66	12.65
Detritus	6.27	6.31	6.03	9.62	5.23

#### **4.5.2 Relative Contribution of Prey Types by the Index of Caloric Importance**

The Index of Caloric Importance, calculated for the overall spotted sea trout diet, revealed that fish and fish crops, combined into a single category, made up for the greatest portion by ICI at 65.1%, followed by brown shrimp (24.7%). All other prey types made up less than 5% ICI.

Broken down by habitat, the top three prey types by ICI over shell were fish and fish crops (69.0%), brown shrimp (17.9%), and detritus (15.3%). The top three prey types by ICI over marsh were brown shrimp (51.8%), fish and fish crops (24.4%), and threadfin shad (12.4%). The top three prey types by ICI over mud were fish and fish crops (67.1%), brown shrimp (19.0%), and bay anchovy (4.8%) (see Table 14).

When considered by season, the top three prey types by %FO were brown shrimp (55.5%), fish and fish crops (32.8%), and gulf menhaden (4.8%) in spring; fish and fish crops (69.2%), bay anchovy (10.6%), and brown shrimp (8.8%) in summer; fish and fish crops (70.0%), brown shrimp (20.6%), and plant and seeds (8.5%) in fall; and, polychaetes (75.8%), fish and fish crops (23.9%), and plant and seeds (0.3%) in winter (see Table 15).

Unidentifiable fish and brown shrimp consistently made up the bulk of prey types by ICI by habitat and most seasons, augmented by a few seasonally or sparsely occurring prey items (*i.e.*, threadfin shad, bay anchovy, gulf menhaden). The Index of Caloric Importance does not show plant material as being relatively important to the diet of spotted sea trout as did the Prey Importance Index, a similarly calculated descriptive statistical index.

Table 14. Index of Caloric Importance by habitat for spotted sea trout prey.

Prey Types	Total ICI	ICI Shell	ICI Marsh Edge	ICI Mud
Fish & Fish Crops	65.1	69	24.4	67.1
Bay Anchovy	2.5	1.8	1.8	4.8
Threadfin Shad	0.8	0	12.4	0
Menhaden	0.9	3.7	0	0.4
Atlantic Croaker	0.5	0	0	2.8
Brown Shrimp	24.7	17.9	51.8	19
Fiddler Crab	0.04	0	2.1	0
Amphipod	0.01	0.01	0	0.01
Polychaete	0.1	1.5	0	0
Gastropod	0.06	0	0	0.3
Plant & Seeds	4.6	5.5	6.8	3.5
Detritus	0.8	15.3	0.8	2.1

Table 15. Index of Caloric Importance by season for spotted sea trout prey.

Prey Types	Total ICI	ICI Spring	ICI Summer	ICI Fall	ICI Winter
Fish & Fish Crops	65.1	32.8	69.2	70.0	23.9
Bay Anchovy	2.5	0.6	10.6	0	0
Threadfin Shad	0.8	0	4.3	0	0
Menhaden	0.9	4.8	0	0	0
Atlantic Croaker	0.5	2.7	0	0	0
Brown Shrimp	24.7	55.5	8.8	20.6	0
Fiddler Crab	0.04	0	0.2	0	0
Amphipod	0.01	0	0.03	0	0
Polychaete	0.1	0	0	0	75.8
Gastropod	0.1	0	0.6	0	0
Plant & Seeds	4.6	3.0	4.7	8.5	0.3
Detritus	0.8	0.6	1.6	0.6	0.1

## CHAPTER 5. PINFISH RESULTS

### 5.1 CATCH DATA

Monthly sampling was conducted for a total of four months: July, August, and November 2003, and July of 2004. Fishing success was sporadic throughout the rest of the year because of migration of pinfish to and from offshore spawning habitats. A total of 137 pinfish stomachs were examined. The stomachs were almost exclusively from pinfish caught in the summer months; only seven fish were caught in November, so seasonal comparisons are of limited value. A total of 35 fish were caught over shell habitats, 33 on the marsh edge, and 69 over mud habitats. The highest abundances were on the mud habitats in August of 2003 and July of 2004 (see Figures 7 and 8).

### 5.2 PREY TYPES AND CALORIC VALUES

Thirteen prey types were identified in the guts of pinfish. The only vertebrate prey type was unidentifiable fish. The invertebrate prey types included: unidentifiable shrimp, unidentifiable crab, hermit crab (*Clibanarius vittatus*), fiddler crab (genus *Uca*), *Sphaeroma terebrans* (an isopod), and ribbed mussel (*Geukensia demissa*). The remaining prey types included: seeds, plant material, soil and sand consolidated into a single category, detritus, unknown material, and bait. The plant material and seeds are both presumed to be entirely *Spartina alterniflora*. The caloric values of all prey types, with the exception of those categories that have no caloric value (soil and sand, unidentifiable material), were found in the literature (see Table 16).

The bait was fresh blue crab (*Callinectes sapidus*) and was found in 12 stomachs. The bait appeared exclusively as blue crab gills, and was easily



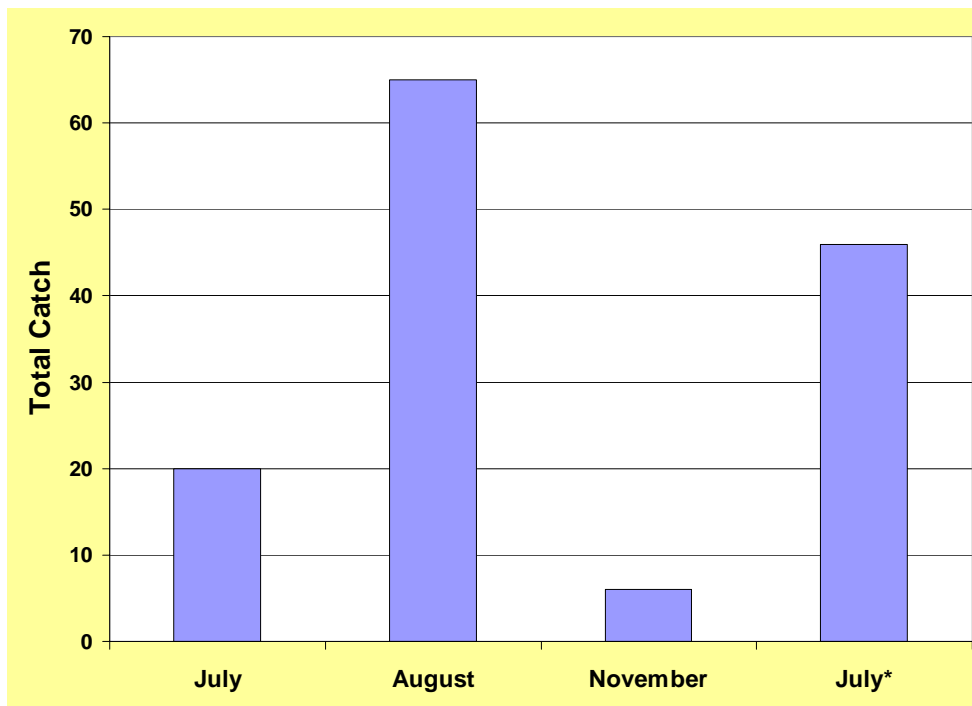


Figure 7. Total catch per month of pinfish, July 2003 to July 2004.

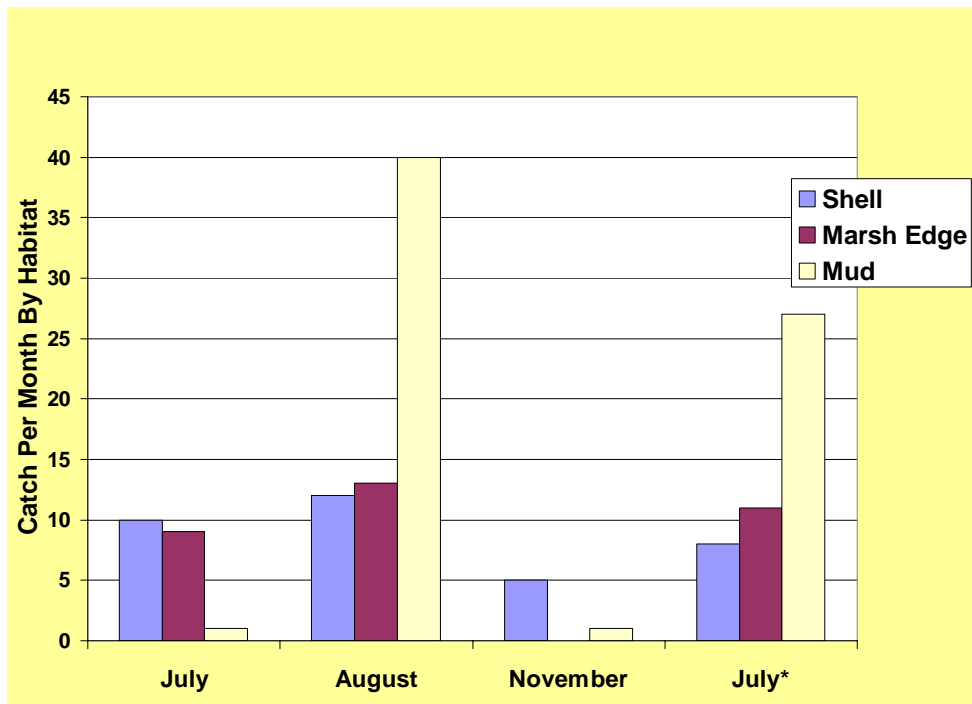


Figure 8. Total catch per month, by habitat, of pinfish, July 2003 to July 2004.

Table 16. Caloric values of the prey items for pinfish (calories per gram dry weight) and their sources in the literature.

Prey Types	Caloric Value (cal / g DW)	Source
Fish	5,014	Wissing <i>et al</i> 1973
Shrimp	4,870	Wissing <i>et al</i> 1973
Crab	3,823	Wissing <i>et al</i> 1973
Hermit Crab	3,712	McCawley, J. 2003
Fiddler Crab	2,405	Cummins & Wuycheck 1971
Sphaeroma	3,004	Cummins & Wuycheck 1971
Mussel	4,600	Cummins & Wuycheck 1971
Plant & Seeds	4,196	Cummins & Wuycheck 1971
Detritus	2,229	Cummins & Wuycheck 1971
Fish	5,014	Wissing <i>et al</i> 1973
Invertebrates	3,820	Wissing <i>et al</i> 1973
Plant & Seeds	4,196	Cummins & Wuycheck 1971
Detritus	2,229	Cummins & Wuycheck 1971

distinguishable from other crab material. Because bait is a relic of the process of fish trapping, and because pinfish have not been reported to prey on adult blue crab in the wild (Czapla 1991), the data on bait in the stomachs was excluded from statistical analysis.

The pinfish ranged from 6.8 cm to 12 cm in length. Pinfish between 6-9 cm ate 44.1% of the total plant material consumed, and pinfish between 9-12 cm ate 55.9% of the total plant material consumed.

### **5.3 RESULTS BY DRY WEIGHT**

#### **5.3.1 Relative Contribution of Prey Types by Percent Dry Weight**

Percent dry weight, calculated for the overall pinfish diet, revealed that detritus made up for the greatest portion by weight at 28.7%, followed by plant and seeds, combined into a single category (24.0%), shrimp (21.4%), hermit crab (17.5%), and fish (7.0%). All other prey types made up less than 5% DW.

Broken down by habitat, the top three prey types by %DW over shell were hermit crab (67.8%), detritus (15.7%), and plant and seeds (10.8%). The top three prey types by %DW along the marsh edge were shrimp (36.6%), plant and seeds (25.6%), and detritus (15.7%). The top three prey types by %DW over mud were detritus (28.8%), plant and seeds (26.7%), and shrimp (24.8%) (see Table 17). When considered by season, the top three prey types by %DW were detritus (32.5%), plant and seeds (26.6%), and shrimp (24.2%) in summer; and, hermit crab (68.6%), fish (20.4%), and crab (6.3%) in fall (see Table 18). Detritus, plant material, unidentifiable shrimp, and hermit crabs consistently made up the bulk of the prey dry weight over all habitats, augmented by fish and crab in fall.

### **5.3.2 Dietary Overlap**

Dietary overlap based upon prey dry weight for each habitat was calculated using Schoener's index and ranged from  $\alpha = 0.40$  to 0.79. Dietary overlap was highest for fish collected over marsh edge vs. mud ( $\alpha = 0.79$ ) and lowest for fish collected over shell vs. marsh edge ( $\alpha = 0.40$ ). By season, dietary overlap was  $\alpha = 0.20$  between summer and fall (see Table 19).

### **5.3.3 Dietary Breadth**

Dietary breadth, a measure of how relatively narrow or broad a diet is, was determined by using the Shannon Diversity Index and was 0.69 for all pinfish combined based upon dry weight of prey. Broken down by habitat, dietary breadth ranged from 0.41 to 0.67. Dietary breadth was highest for fish collected over marsh edge (0.67) and lowest for fish collected over shell (0.41). By season, dietary breadth was 0.65 in summer and 0.39 in fall (see Table 20).

### **5.3.4 Proportion of Prey Dry Weight to Pinfish Dry Weight**

The overall DW/FW ratio was  $12.6 * 10^{-4}$  for all pinfish collected during this study. By habitat, DW/FW ratio ranged from  $5.1 * 10^{-4}$  to  $18.8 * 10^{-4}$ . The greatest amount of food by weight was consumed at the marsh edge ( $18.8 * 10^{-4}$ ), and the least amount of food by weight was consumed over shell ( $5.1 * 10^{-4}$ ). By season, the DW/FW ratio was  $11.2 * 10^{-4}$  in summer and  $19.3 * 10^{-4}$  in fall (see Table 21).

### **5.3.5 Statistical Analysis of Prey Dry Weight and Stomach Fullness**

No statistical significance was found when testing the effects of season and time of day on DW, DW\*CAL, and stomach fullness (one-way ANOVA,  $p > 0.03$ ). As such

Table 17. Percent dry weight by habitat for pinfish prey.

Prey Types	Total %DW	%DW Shell	%DW Marsh Edge	%DW Mud
Fish	7	0	7.7	10.8
Shrimp	21.4	0	36.6	24.8
Crab	0.8	5.6	0	0
Hermit Crab	17.5	68	12.6	8.1
Fiddler Crab	0.1	0	0.2	0
Sphaeroma	0.4	0	1.4	0
Mussel	0.2	0	0	0.4
Plant / Seeds	24	10.8	25.9	27
Detritus	28.7	15.7	15.7	28.8

Table 18. Percent dry weight by season for pinfish prey.

Prey Types	Total % DW	%DW Summer	%DW Fall
Fish	7.0	5.2	20.4
Shrimp	21.4	24.2	0
Crab	0.8	0.1	6.3
Hermit Crab	17.5	10.7	68.6
Fiddler Crab	0.1	0.1	0
Sphaeroma	0.4	0.4	0
Mussel	0.2	0.2	0
Plant & Seeds	24.0	26.6	4.8
Detritus	28.7	32.5	0

Table 19. Dietary overlap by habitat and season for pinfish.

Habitat Types and Seasons	Schoener's Index ( $\alpha$ )
shell vs. marsh edge	0.4
shell vs. mud	0.41
marsh edge vs. mud	0.79
summer vs. fall	0.2

Table 20. Dietary breadth by habitat and season for pinfish.

Habitat Types and Seasons	Shannon Diversity Index ( $H'$ )
Total	0.69
shell	0.41
marsh edge	0.67
mud	0.66
summer	0.65
fall	0.39

Table 21. Ratio of prey dry weight to pinfish dry weight by habitat and season.

Habitat Types and Seasons	*multiply by $10^{-4}$
Total	12.6
shell	5.1
marsh edge	18.8
mud	13.2
summer	11.2
fall	19.3

the null hypothesis that there is no difference in DW, DW\*CAL, and stomach fullness according to the habitat and time of day effects could not be rejected.

No statistical significance was found when testing the effects of habitat on DW, DW\*CAL (one-way ANOVA,  $p > 0.03$ ). As such the null hypothesis that there is no difference in DW or DW\*CAL according to the habitat effect could not be rejected. However, the habitat effect was significant for stomach fullness with a p-value of 0.01. A post-ANOVA Fisher's PLSD test found significant pair-wise p-values for shell vs. mud ( $p = 0.02$ ) and marsh edge vs. mud ( $p = 0.01$ ). Because the habitat effect produces a stomach fullness p-value less than 0.03, the null hypothesis that there is no difference in stomach fullness for habitat groupings can be rejected (see Table 22).

#### **5.3.6 Statistical Analysis of Overall Pinfish Diet**

No statistical significance was found when using ANOSIM to test the diet of pinfish using baseline data against the main effects: habitat, time of day, and fish size. None of the corresponding R-values were greater than 0.15, so the null hypothesis that there are no differences between the habitat, time of day, and fish size groupings could not be rejected at the 0.1% level. Because none of the one-way ANOSIM tests were significant, two-way ANOSIM tests for interactions and post-hoc tests (*i.e.*, BVStep and SIMPER) were not necessary. None of the changes in the data matrix [*i.e.*, 1) unaltered original data; 2) consolidated data; and, 3) data excluding bait] produced results that were not consistent with results produced by using baseline data.

Statistical significance was found when using ANOSIM to test the diet of pinfish using baseline against the season effect, with an R-statistic of 0.297. Because the season R-

Table 22. P-values in one-way ANOVA results by habitat, season, and time of day for pinfish dry weight prey (DW), DW x mean calories (cal / g) per stomach, and stomach fullness.

	Habitat	Season	Time of Day
DW	0.79	0.28	0.05
DW*CAL	0.73	0.34	0.04
Stomach Fullness	0.01	0.87	0.21

Table 23. One-way ANOSIM Global R-statistic values for pinfish diet by habitat, season, time of day, and fish size.  $R > 0.15$  = significant at  $\alpha = 0.1$ .

Main Effects	ANOSIM (R)
Habitat	0.084
Season	0.297
Time of Day	0.121
Fish Size	0.033



statistic is greater than 0.15, the null hypotheses that there is no difference in season groups can be rejected at the 0.1% level (see Table 23). The SIMPER analysis showed that plant material accounted for the most dissimilarity in diet between seasons (20.53% dissimilarity), followed by unidentified material (14.9%), hermit crab (13.47%), unidentified fish (13.25%), unidentified crab (12.42%), and detritus (9.51%). It should be cautioned, however, that seasonal data for pinfish is of limited value because it is based upon only two seasons (summer and fall) and the fall season is based upon a very small number of samples.

The MDS plot showed that while there was some clumping in the upper-middle portion of the plot, there appeared to be more visual variation in the diets of the pinfish than that seen in the spotted sea trout MDS plot (see Figure 19). The plotted numbers correspond to the identification number of the individual fish and the stress value of 0.01 means that this plot has the best possible ‘goodness-of-fit’.

### **5.3.7 Statistical Analysis of Pinfish Diet and Water Quality**

The non-parametric statistical tool BIO-ENV also was used to examine pinfish diet and water quality by using Spearman Rank Correlation to compare the diet matrix constructed in the ANOSIM tests with a separate abiotic matrix containing data on dissolved oxygen (mg/L), salinity (ppt), and temperature (°C). BIO-ENV showed that the suite of variables that best ‘explained’ the pattern seen in the MDS plot included all three variables: dissolved oxygen, salinity, and temperature. However, the correlation factor that ranges between 0 and 1.0 was low, at 0.128. Thus, no strong relationship between the prey dry weight diet matrix and environmental effects could be identified.

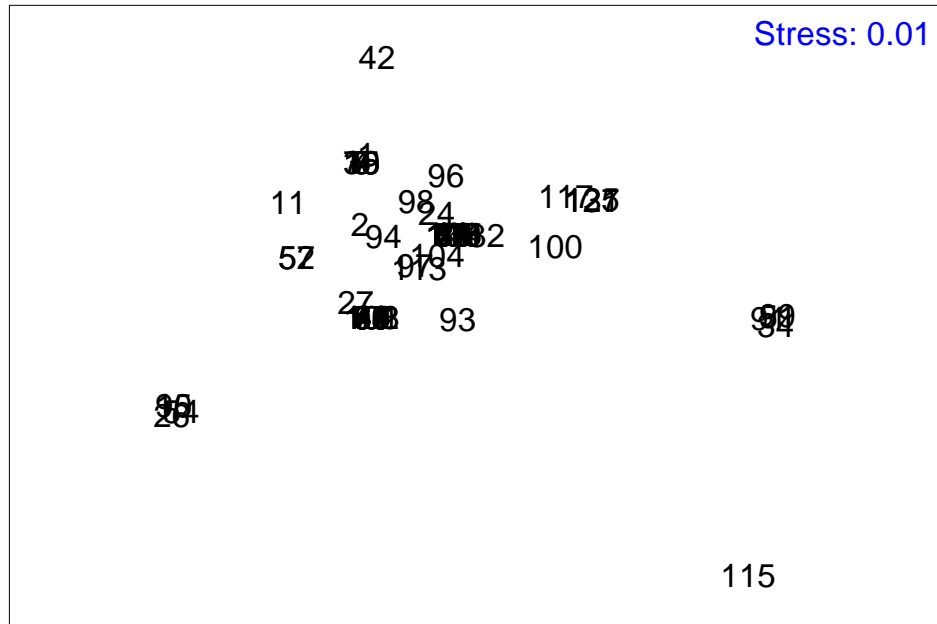


Figure 9. Multi-Dimensional Scaling plot for pinfish.

## **5.4 RESULTS BY FREQUENCY**

### **5.4.1 Relative Contribution of Prey Types by Percent Frequency of Occurrence**

Percent frequency of occurrence, calculated for the overall pinfish diet, revealed that plant material and seeds, combined into a single category, made up for the greatest percentage by frequency at 58.3%, followed by detritus (33.3%), shrimp (6.9%), and hermit crab (5.6%). All other prey types made up less than 5% FO.

Broken down by habitat, the top three prey types by %FO over shell were plant and seeds (41.7%), detritus (33.3%), and both the unidentifiable crab and hermit crab prey types (16.7%). The top three prey types by %FO along the marsh edge were plant and seeds (72.0%), detritus (32.0%), and both the fish and shrimp prey types (8.0%). The top three prey types by %FO over mud were plant and seeds (54.3%), detritus (34.3%), and shrimp (8.6%) (see Table 24).

When considered by season, the top three prey types by %FO were plant and seeds (50.6%), detritus (29.1%), and shrimp (6.3%) in summer; and, fish (25.0%), unidentifiable crab (25.0%), hermit crab (25.0%), and plant and seeds (25.0%) in fall (see Table 25). Detritus and plant material consistently made up the bulk of prey by frequency by habitat and season, while hermit crab, shrimp and fish prey types also were important.

### **5.4.2 Vacuity Index**

The total vacuity index (VI) for pinfish was 37.2%. Despite the high vacuity, there was no obvious regurgitation as described in Treasurer (1988). Broken down by habitat, VI ranged from 5.1% to 22.6%. The lowest vacuity occurred along the

Table 24. Percent frequency of occurrence by habitat for pinfish prey.

Prey Types	Total %FO	%FO Shell	%FO Marsh Edge	%FO Mud
Fish	4.2	0	8	5.7
Shrimp	6.9	0	8	8.6
Crab	2.8	16.7	0	0
Hermit Crab	5.6	16.7	4	2.9
Fiddler Crab	1.4	0	4	0
Sphaeroma	1.4	0	4	0
Mussel	2.8	0	0	5.7
Plant & Seeds	58.3	41.7	72	54.3
Detritus	33.3	33.3	32	34.3

Table 25. Percent frequency of occurrence by season for pinfish prey.

Prey Types	Total % FO	%FO Summer	%FO Fall
Fish	4.2	3.8	25
Shrimp	6.9	6.3	0
Crab	2.8	1.3	25
Hermit Crab	5.6	3.8	25
Fiddler Crab	1.4	1.3	0
Sphaeroma	1.4	1.3	0
Mussel	2.8	2.5	0
Plant & Seeds	58.3	50.6	25
Detritus	33.3	29.1	0

marsh edge (5.1%), and the highest vacuity occurred over mud (22.6%). By season, vacuity in summer was 45.0% and vacuity in fall was 20.0% (see Table 26).

## **5.5 RESULTS COMBINING WEIGHT, FREQUENCY, AND CALORIC VALUE**

### **5.5.1 Relative Contribution of Prey Types by the Prey Importance Index**

The Prey Importance Index, calculated for the overall pinfish diet, revealed that plant material and seeds, combined into a single category, made up for the greatest portion of the index at 44.1%, followed by detritus (29.2%), shrimp (7.9%), hermit crab (6.4%), and fish (5.9%). All other prey types made up less than 5% PI.

Broken down by habitat, the top three prey types by PI over shell were detritus (32.4%), plant and seeds (27.6%), and both unidentifiable crab and hermit crab categories (20.0%). The top three prey types by PI along the marsh edge were plant and seeds (50.4%), detritus (25.6%), and shrimp (8.3%). The top three prey types by PI over mud were plant and seeds (44.6%), detritus (31.1%), and shrimp (10.3%) (see Table 27). When considered by season, the top three prey types by PI were plant and seeds (44.5%), detritus (33.0%), and shrimp (7.6%) in summer; and, fish (25.0%), unidentifiable crab (25.0%), hermit crab (25.0%), and plant and seeds (25.0%) in fall (see Table 28).

Detritus and plant material consistently made up the bulk of prey identified as relatively important by habitat by using the PI index, while hermit crab, shrimp and fish prey types also were important, but occurred less consistently.

### **5.5.2 Relative Contribution of Prey Types by the Index of Caloric Importance**

The Index of Caloric Importance, calculated for the overall pinfish diet, revealed that plant material and seeds, combined into a single prey category, made up for the

Table 26. Vacuity Index by habitat and season for pinfish.

Habitat Types and Seasons	%
Total	37.2
shell	9.5
marsh edge	5.1
mud	37.2
summer	45.0
fall	20.0

Table 27. Prey Importance Index by habitat for pinfish prey.

Prey Types	Total PI	PI - Shell	PI - Marsh Edge	PI - Mud
Fish	5.9	0	7.2	6.9
Shrimp	7.9	0	8.3	10.3
Crab	3.2	20	0	0
Hermit Crab	6.4	20	4.2	3.4
Fiddler Crab	0.1	0	0.2	0
Sphaeroma	1.6	0	4.2	0
Mussel	1.7	0	0	3.7
Plant & Seeds	44.1	27.6	50.4	44.6
Detritus	29.2	32.4	25.6	31.1

Table 28. Prey Importance Index by season for pinfish prey.

	Total PI	PI Summer	PI Fall
Fish	5.9	4.1	25
Shrimp	7.9	7.6	0
Crab	3.2	1.5	25
Hermit Crab	6.4	4.6	25
Fiddler Crab	0.1	0.1	0
Sphaeroma	1.6	1.5	0
Mussel	1.7	1.6	0
Plant & Seeds	44.1	44.5	25
Detritus	29.2	33.0	0

greatest portion by of the index at 63.5%, followed by detritus (23.1%) and shrimp (7.8%). All other prey types made up less than 5% ICI.

Broken down by habitat, the top three prey items by ICI over shell were hermit crab (55.2%), plant and seeds (24.8%), and detritus (15.3%). The top three prey items by ICI along the marsh edge were plant and seeds (71.9%), shrimp (13.1%), and detritus (10.3%). The top three prey items by ICI over mud were plant and seeds (62.8%), detritus (22.5%), and shrimp (10.6%) (see Table 29). When considered by season, the top three prey types by ICI were plant and seeds (64.4%), detritus (24.1%), and shrimp (8.5%) in summer; and, hermit crab (63.5%), fish (25.5%), and crab (6.0%) in fall (see Table 30). Detritus and plant material consistently made up the bulk of prey identified as relatively important by habitat by using the ICI index, while hermit crab, shrimp and fish prey types also were important, but occurred less consistently.



Table 29. Index of Caloric Importance by habitat for pinfish prey.

Prey Types	Total ICI	ICI Shell	ICI Marsh Edge	ICI Mud
Fish	1.6	0	2.8	3.2
Shrimp	7.8	0	13.1	10.6
Crab	0.1	4.7	0	0
Hermit Crab	3.9	55.2	1.7	0.9
Fiddler Crab	0	0	0.02	0
Sphaeroma	0.02	0	0.2	0
Mussel	0.02	0	0	0.1
Plant & Seeds	63.5	24.8	71.9	62.8
Detritus	23.1	15.3	10.3	22.5
Fish	1.6	0	2.8	3.2
Invertebrates	11.9	59.9	15.0	11.6
Plant & Seeds	63.5	24.8	71.9	62.8
Detritus	23.1	15.3	10.3	22.5

Table 30. Index of Caloric Importance by season for pinfish prey.

Prey Types	Total ICI	ICI Summer	ICI Fall
Fish	1.6	1.1	25.5
Shrimp	7.8	8.5	0
Crab	0.1	0.01	6.0
Hermit Crab	3.9	1.7	63.5
Fiddler Crab	0	0	0
Sphaeroma	0.02	0.02	0
Mussel	0.02	0.03	0
Plant & Seeds	63.5	64.4	5.1
Detritus	23.1	24.1	0

## **CHAPTER 6. DISCUSSION**

### **6.1 SPOTTED SEA TROUT CONCLUSIONS**

Three generalities emerge concerning the homogeneity of diet observed in the spotted sea trout collected during this study: 1) spotted sea trout were selecting for a small suite of preferred prey, regardless of habitat or any other effect; 2) some of the prey themselves may have contributed to the lack of variance because they are presumed to be ubiquitous throughout the estuarine environment; and, 3) spotted sea trout were searching for these prey, further confounding any effort to establish a dietary link to habitat. These conclusions will be discussed below.

#### **6.1.1 Spotted Sea Trout Prey Selection**

The number of prey items consumed by spotted sea trout in this study was low (17), perhaps because the spotted sea trout sampled mostly were adult fish. Several studies report that larval spotted sea trout eat a diversity of small items (*i.e.*, copepods, larval bivalves, larval gastropods) and juvenile spotted sea trout shift to a diet of mysids, fish and shrimp. But as the spotted sea trout reach adulthood, their diet increasingly consists of fewer items, mostly fish and large shrimp (Darnell 1958, Springer and Woodburn 1960, Carr and Adams 1976, McMichael and Peters 1989, Hettler 1989).

Generally, the number of prey items consumed by spotted sea trout in this study was low when compared to research on the diet of spotted sea trout caught over sea grass habitat (*i.e.*, Hettler 1989, McMichael and Peters 1989, Llanso *et al.* 1998). Most seagrass is always flooded and highly structured, offering more foraging and refugia opportunities than marsh edge habitats, which may not always be flooded and accessible to fish if the tides are especially low or wind is blowing water off of the marsh. While

marsh edge may have high biomass, it does not always have high biodiversity when compared to sea grass or other fish habitats (Pihl *et al.* 2002).

When interpreting the descriptive indices calculated to determine the relative importance of specific prey items, I found that spotted sea trout diet was, in general, consistent across habitat types. In all of the indices (%DW, %FO, PI, ICI) unidentifiable fish and brown shrimp were found to be highest in relative importance. Plant material and detritus also were found to be relatively important by using %FO and PI. Plant material, however, may be incidentally ingested, perhaps resulting from missed attempts at more mobile prey. The idea that the spotted sea trout were eating similar prey items, regardless of habitat, was supported by the lack of statistical significance when running ANOVA and ANOSIM tests of habitat groupings. Furthermore, no statistically significant relationship between the diet of spotted sea trout and season, site, bay location, and fish size could be demonstrated.

Spotted sea trout in this study included polychaetes in their winter diet, a seasonal trend that was similarly reported by Overstreet and Heard (1982), McMichael and Peters (1989), and Llanso *et al.* (1998). This shift is most likely due to changing prey availability, rather than a response to environmental conditions. Using ANOVA and ANOSIM statistical tests, no significant relationship between spotted sea trout diet and seasonally changing water quality variables (dissolved oxygen, salinity, and temperature) was seen, but it is well documented that there are fewer fish and shrimp prey in the estuary during the fall and winter months (Tabb 1961, Dietz 1976). With less prey available, it is logical that dietary breadth is lowest in winter.

Given that spotted sea trout are living within their range of salinity tolerances, the lack of an identifiable statistical relationship between salinity, or relative salinity (*i.e.*, bay location), with the diet of spotted sea trout is supported by research reported by Wakeman and Ramsey (1985). In sampling seven separate study areas along the Louisiana coast, and one in Texas, they reported that while condition coefficients factors of spotted sea trout varied significantly according to study area, they did not appear to be affected by the salinity gradient present in each individual microgeographic study area.

Percent vacuity for the spotted sea trout in this study was high, but percent vacuity reported for spotted sea trout in the scientific literature ranges widely, from less than 2% in Llanso *et al.* (1998) to as high as 60% in Hettler (1989). This variability may be attributable to the variety of sampling techniques used, different localities, and different life-stages sampled. Overstreet and Heard (1982) suggested that spotted sea trout may be foraging less during spawning times. This may have contributed to the high spring and summer vacuity in this study, but this is speculation because no data on spawning condition were gathered.

With respect to habitat-specific vacuity, there was a smaller percentage of empty stomachs in those fish captured along the marsh edge then elsewhere, implying that more food is being encountered along the marsh edge. My interpretation of the DW/FW ratio seems to confirm these conclusions, with more food by weight found in those fish captured along the marsh edge. When examining the dietary overlap between habitat types, the marsh edge again is somewhat unique, with the least amount of overlap with shell, and then, mud habitats. This trend is limited, however, to the descriptive statistics; interpretation of results from parametric and non-parametric statistical analysis could find

no relationship between the diet of spotted sea trout and a number of factors, including habitat type.

Finally, although the list of spotted sea trout prey was fairly short, dietary breadth was still high overall (0.79), suggesting that of those prey items consumed, there was no strong preference among the prey. A number of studies also report that spotted sea trout diets are flexible, even reporting them as generalist-feeders (Darnell 1958, Tabb 1961, Hettler 1989, Springer and Woodburn 1960, and Llanso *et al.* 1998).

### **6.1.2 Prey Habitats and Site Fidelity**

Ultimately, the remarkably low variability in the diet of spotted sea trout, as seen in Figure 5 may not only be due to selectivity, but also the ubiquity of the prey items themselves. Nereid polychaetes and all of the fish prey in Table 1 can be found almost anywhere in the estuarine system. Other prey items, like amphipods, gastropods, and shrimp, are typically associated with a specific habitat type and should thus only appear in the gut contents of fish caught on that habitat (Heard 1979, Ruppert and Barnes 1994). As seen in this study, however, correspondence between prey items and habitat was low.

In previous studies fiddler crabs are associated with marsh edge and *Melitidae* amphipods are associated with shell (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994). Indeed, in this study fiddler crabs and *Melitidae* amphipods were found in the stomachs of trout caught on their associated habitats. However, the following prey items from this study were not found in the gut contents of fish caught on the habitats with which the prey are associated: 1) *Gammarus* amphipods are associated with the marsh edge (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994), but were found in a fish caught on the mud; 2) brown shrimp are associated with the marsh edge

(Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994), but were found in fish caught on all three habitat types; 3) *Nereid* gastropods are associated with the marsh edge (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994), but were found in a fish caught on the mud; and, 4) plant and seed material should have certainly been found mostly in fish taken along the marsh edge, but was, in fact, lowest by dry weight along the marsh edge. These results are counter-intuitive, suggesting to me that the spotted sea trout are too transitory to relate to a specific habitat or micro-habitat, traveling far and often to search for their preferred prey.

## **6.2 PINFISH CONCLUSIONS**

Two generalities emerge concerning the homogeneity of diet observed in the pinfish of this study: 1) pinfish collected in this study were selecting for a small suite of preferred food items (relying heavily on plant and detritus), regardless of habitat or any other effect; and, 2) although pinfish did seem to exhibit more site-fidelity than spotted sea trout, some of the prey on which they feed are very common across multiple habitats. These conclusions will be discussed below.

### **6.2.1 Pinfish Prey Selection**

None of the pinfish prey types reported in this study were unusual; many studies report pinfish eating a wide variety of items, including: small decapods, small fish, plant material, and detritus (Kjelson and Johnson 1976, Montgomery and Targett 1992, Pike and Lindquist 1994). There was an unusual absence of amphipods in the diet of pinfish from this study, whereas most previous studies report that amphipods are a principle component of the pinfish diet (Kjelson and Johnson 1976, Montgomery and Targett 1992, Pike and Lindquist 1994).

When interpreting the descriptive indices calculated to determine the relative importance of specific prey items, it was found that pinfish diet was, in general, consistent across habitat types. In all of the indices (%DW, %FO, PI, ICI) plant material and detritus were found to be highest in relative importance, with hermit crab and shrimp secondary. In addition, hermit crab was most important in association with the shell habitat. The idea that the pinfish were eating similar prey items, regardless of habitat, was supported by the lack of statistical significance when running ANOVA and ANOSIM tests of habitat groupings, the only exception being the significant relationship between pinfish stomach fullness and habitat. Furthermore, no statistically significant relationship between the diet of pinfish and season, time of day, and fish size could be demonstrated.

There appeared to be a superficial seasonal trend in the diets of pinfish, shifting from a diet of primarily plant, detritus, and shrimp in the summer to a diet of plant, fish, and hermit crab in the fall. This shift is most likely due to changing prey availability, rather than a response to environmental conditions, because using ANOVA and ANOSIM statistical tests, no significant relationship between pinfish diet and seasonally changing water quality variables (dissolved oxygen, salinity, and temperature) was seen. Again, pinfish seasonal analyses must be viewed with caution because of small sample sizes in fall.

Percent vacuity index for pinfish in this study (37.2%) was considerably higher than that reported in other studies; Czapla (1991) reported 2% VI and Thompson (2000) reported 3% VI. High vacuity may have resulted from the some pinfish being captured soon after the fish traps were set in the water; as much as 12 hours could have elapsed

between capture and processing, resulting in evacuation of gut contents. When examining vacuity further, it was shown to be lowest in those fish caught along the marsh edge, implying that more food is being encountered there. My interpretation of the DW/FW ratio seems to confirm this, with more food by weight found in those fish captured along the marsh edge. These results were similar to spotted sea trout findings, but unlike the spotted sea trout, the shell (not the marsh edge) exhibited the lowest overlap (first with marsh, and then mud). Shell also had the lowest calculated dietary breadth. So for pinfish, the marsh edge may provide more food, and the shell habitat may have the least prey diversity. This trend is limited, however, to the descriptive statistics; interpretation of results from parametric and non-parametric statistical analysis could find no relationship between the diet of pinfish and a number of factors, including habitat.

Of special interest to researchers of pinfish diet is the proportion of plant in their diet, and the roughly 24% dry weight reported here compares to 18% reported by Thompson (2000), 50% reported by Czapla (1991), and 40.6% reported by Stoner (1976). This study and the above cited reports show that pinfish never became totally herbivorous. Some studies have reported an ontogenetic diet shift in pinfish greater than 9 cm to a diet consisting of more plant material than animal material (Thompson 2000). I found that nearly 45% of the total plant material consumed was eaten by pinfish between 6-9 cm, which does not support an ontogenetic shift at 9 cm. Knowing that pinfish graze on zooplankton at a much higher rate than metabolically necessary (Kjelson and Johnson 1976, Montgomery and Targett 1992, Thompson 2000), it is entirely possible that local populations of zooplankton, particularly amphipods, were



grazed down locally or existed in patchy abundances in other areas of Barataria Bay that I did not sample. This may help to explain the conspicuous absence of amphipods in the diet of these pinfish, but without having simultaneously sampled for amphipods, however, this remains conjecture.

Finally, although the list of pinfish prey was fairly short, dietary breadth was still high overall at 0.7. This is very similar to dietary breadth values reported in Thompson (2000), and it suggests that of those prey items consumed, there was no strong preference among the prey. A number of studies also find that pinfish are generalists with flexible diets (Czapla 1991, Pike and Lindquist 1994, Thompson 2000).

### **6.2.2 Prey Habitats and Site Fidelity**

Ultimately, the low variability in the diet of pinfish may not only be due to selectivity but also to the ubiquity of the prey items themselves, like the hermit crab and fish prey that can be found almost anywhere in the estuarine system (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994). Even though the pinfish prey results in this study were generally dominated by a few prey items that are common across all habitats, some of the other less common prey seemed to have some habitat specificity. For example, most shrimp are associated with the marsh edge or mud (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994) and no shrimp in this study were found in fish caught on the shell habitat. Fiddler crab and *Sphaeroma terebrans*, almost exclusively associated with marsh edge habitat (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994) were indeed found in the guts of pinfish captured along the marsh edge in this study. In addition, plant material, soil, and sand were lowest in quantity in those fish caught over shell, as should be expected. The only counterintuitive

result was the presence of mussel (typically found on shell reef or at the base of submerged marsh vegetation) (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994) in the diet of a fish caught over the mud bottom in this study (Felder 1973, Wood 1974, Heard 1979, Rupert and Barnes 1994). So, although there was no statistical dietary trend in habitat groupings, when all results are taken together, it appears that pinfish may indeed exhibit more site-fidelity than spotted sea trout.

### **6.3 SUGGESTIONS FOR FUTURE WORK**

When considering the original set of hypotheses, the null hypotheses could not be rejected: no statistical differences in the diet of spotted sea trout or pinfish caught over different habitats, and along the salinity gradient, could be detected. The last hypotheses, that marsh edge provided the most beneficial diet to either fish (in terms of caloric value), could only be weakly confirmed by descriptive statistics. This lack of both dietary variation and linkage between habitat and diet was shown to be largely attributable to the prey selectivity of the fish, the ubiquity of the species being preyed upon, and, in the case of spotted sea trout, the transitory nature of the fish.

Defining Essential Fish Habitat by diet alone may not be possible for these two fish, because habitat suitability can be defined by a whole suite of variables, not just prey availability. Furthermore, I am concerned that not all of the three habitats included in this study should be considered when evaluating EFH. Mud bottom and marsh edge account for large portions of the total bay surface area available to fish, but oyster reef is of inconsistent quality in Barataria Bay and accounts for a very small portion of the total bay surface area.

Natural oyster reefs are created by gregarious settlement, with multiple generations settling on one another, causing the reef to grow vertically (Coen and Luckenbach, 2000). However, oysters in Barataria Bay are planted ('culched') and harvested often (Melancon *et al.* 1998). This results in low relief and inconsistent density, for example: 1) planted shell habitat at *Queen Bess Island*, while dense, is entirely low-relief, broken-up, and non-living shell; 2) live oysters at *Manila Village* are patchy (varying from 865 ml displacement per m<sup>2</sup> to 4,950 ml displacement per m<sup>2</sup>) and low-relief because it is harvested regularly; and, 3) relic shell habitat at *Fisherman's Point* is sparse (only 10-25 ml displacement per m<sup>2</sup>), consisting of mostly small, broken-up pieces (Boswell, 2004). Thus, shell reef in Barataria Bay may not necessarily be functioning the same as biogenic reefs, and may not be totally distinguishable from the surrounding mud habitat.

It may be appropriate when assessing Essential Fish Habitat to exclude small, structured habitats like oyster reefs or artificial reefs, even when they exhibit unusually high fish densities. These habitats often do not account for a large percentage of the total home-range of the fish population and therefore may not have a significant impact on overall population survival and production. Perhaps other parameters beyond habitat type should be included when considering what is 'essential' for mobile fish like spotted sea trout. For example, Tabb (1958) included the following characteristics when defining suitable habitat for spotted sea trout: 1) large, shallow areas of quiet water; 2) absence of predators and competitors; 4) large areas of submerged aquatic vegetation; 4) stable temperature between 60-80°F; and, 5) adequate areas of grass flats with a depth of 3-6 meters that can be used as refuge from the winter cold. Furthermore, Karr and Chu

(1997) suggested 5 parameters to define the quality of the whole ecological system: water quality, habitat structure, flow regime, energy source, and biological interactions.

As the marshes in Barataria Bay continue to subside and disappear, many of the variables defining both habitat and system quality will no doubt change dramatically, and the survival requirements of nekton may not be met. Salinity and temperature regimes may shift, the total carrying capacity of the marshes may decrease, and populations of important estuarine prey items (particularly shrimp and forage fishes) may decline. Preferred habitats like marsh edge may go unoccupied because of lower population numbers. The physical integrity of marshes is especially important when buffering estuarine populations against climactic anomalies and hurricanes. With little to no recruitment from outside populations, estuarine fish like spotted sea trout may experience a localized reduction in year-class success due to the impacts of such storms, greatly reducing subsequent recruitment (Baltz *et al.* 2003, Day *et al.* 2000, Martin *et al.* 2000, Reyes *et al.* 2000).

Remarkably, fisheries along the Louisiana coast have shown considerable resilience, not yet exhibiting any catastrophic changes. Although some changes in trawling data for demersal assemblages may have taken place over the last 60 years, they are difficult to prove because many of the various human impacts on the coastal ecosystem are confounding, having all occurred in the same time period. Eutrophication, for example, may be offsetting the negative impacts of marsh degradation (Chesney *et al.* 2000). It is unlikely, though, that populations of marine fish and invertebrates will continue to prove resilient indefinitely.

With concerns like these, it is important to continue examining the dietary and

habitat needs of fish like spotted sea trout and pinfish, but with special attention to behavioral response to habitat change, an idea not addressed in this study. Underwood *et al.* (2004) warn that reporting the presence of a fish over a habitat, or an item in the guts of that fish, do not represent ‘preference’, or even selection, of that habitat or food. Rather, the fish is merely associated with that habitat or prey. To really quantify preference, the direct study of individual fish prey selection is necessary, or at least demonstration of a difference in the relative proportions of prey in the diet compared with the relative proportions available in the wild. Unfortunately, sampling the bioavailability of prey in the same manner that the fish encounters the prey is very difficult; bioavailability information is usually skewed because of the differences between sampling gear and methods. Perhaps the best strategy in quantifying prey preference is a carefully designed experimental analysis. This may be the only way to separate out the variables that lead to potential negative, neutral, and positive responses to choosing one prey over another (*i.e.*, ‘catchability’, ‘acceptability’ and the past history of the consumer) (Underwood *et al.* 2004).

To further analyze the dietary behavior of spotted sea trout and pinfish from each of these scientific approaches, the following research is suggested: 1) broaden the scope of the study to include early and late-juvenile fish; 2) account for differential digestion of detritus and slower consumption rates in winter; 3) carefully examine other capture-site characteristics besides substrate type, dissolved oxygen, temperature, and salinity; 4) obtain spatially explicit *in situ* prey abundance estimates; and, 5) design and perform experimental analyses that offer multiple habitat and prey combinations for individual fish.

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## **VITA**

Micah Russell was born in Corvallis, Oregon, on June 15, 1979, to Michael and Barbara Russell. During the fall of 2000, he was awarded a Research Experience for Undergraduates fellowship from the National Science Foundation and studied estuarine fish feeding ecology at the Dauphin Island Sea Lab in Alabama. In spring of 2002 he graduated from Pacific University in Forest Grove, Oregon, with a Bachelor of Science in environmental biology and a Bachelor of the Arts in piano performance. A graduate assistantship was awarded to Micah in the fall of 2002 to attend Louisiana State University in Baton Rouge, Louisiana, where he has continued studying estuarine fish ecology and piano performance.