TROPHIC ECOLOGY OF JUVENILE WEAKFISH (CYNOSCION REGALIS)

IN THE DELAWARE BAY USING STOMACH CONTENT

AND STABLE ISOTOPE ANALYSES

By

Brian Alberto Galvez

A THESIS

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This thesis is approved by the following members of the Final Oral Review Committee:

Dr. Gulnihal Ozbay, Committee Chairperson, Department of Agriculture and Natural Resources, Delaware State University

Dr. Stacy Smith, Committee Co-chairperson, Department of Agriculture and Natural Resources, Delaware State University

Dr. Kevina Vulinec, Committee Member, Department of Agriculture and Natural Resources, Delaware State University

Dr. Richard Barczewski, Committee Member, Department of Agriculture and Natural Resources, Delaware State University

Dr. Maurice Crawford, External Committee Member, Department of Natural Sciences, University of Maryland Eastern Shore

Dr. Howard Townsend, External Committee Member, Cooperative Oxford Laboratory, National Oceanic and Atmospheric Administration

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DEDICATION

I dedicate this work to the most important people in my life. To my parents, especially my dad, who fostered in me a sense of relentless curiosity to which I ascribe my love of the natural world. To my loving and beautiful wife, Quinn, whose constant love, support, patience, and hard work during my academic endeavors never ceased to amaze me. Thank you for believing in me and for inspiring in me the confidence that I needed to finish this work. I could not have done this without you.

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Brian Alberto Galvez

Faculty Advisor: Dr. Gulnihal Ozbay Co-Advisor: Dr. Stacy L. Smith

ABSTRACT

The Weakfish (*Cynoscion regalis*) is a commercially and recreationally valuable fish species that inhabits the Atlantic Coast of North America, where it primarily occurs from North Carolina to New York. The Weakfish fishery is depleted and has not rebounded despite fishing mortality decreases since 2011. The failure of the fishery to recover is currently attributed to elevated rates of natural mortality, the causes of which are not fully understood. Natural mortality is known to occur at higher rates in juvenile fish compared to adults. Because the diet of juvenile fishes is a factor of natural mortality and recruitment success, I studied the foraging habits of juvenile Weakfish in one of their primary spawning and nursery habitats, the Delaware Bay, in order to increase and update the understanding of their feeding habits prior to their emigration to offshore overwintering grounds.

To answer questions regarding sources of organic matter, prey availability, prey importance, and prey origin (marsh or pelagic derived), I applied stomach content and stable isotope analyses to juvenile Weakfish in 2017 and 2018 throughout most of their Delaware Bay residency (July through October), along both coasts of the Delaware Bay (Delaware and New Jersey nearshore areas), and along the salinity gradient (lower, middle, and upper bay). These methods were used to elucidate the mechanisms and factors controlling stable isotope values of juvenile Weakfish and to determine the prey species responsible for their tissue production and growth.

Juvenile Weakfish were caught from nearshore sampling stations sampled by Delaware and New Jersey Fish and Wildlife state agencies. The fish were separated into three size classes that represent a known ontogenetic shift in diet. Small Weakfish (0-60 mm SL) are mysid specialists, medium Weakfish (61-100 mm SL) begin to incorporate more forage fish into their diet, and large Weakfish (101-137 mm SL) are known to eat roughly equal proportions of forage fish and crustaceans by weight. Commonly consumed prey items, including mysid shrimp, gammarid amphipods, polychaete worms, isopods, sand shrimp, and Bay Anchovy, were also collected in 2018 for stable isotope analysis use in a Bayesian mixing model to infer the proportional contributions of individual prey items to juvenile Weakfish productivity. Generalized linear models (GLMs) were employed to analyze the relationships between carbon, nitrogen, and sulfur (CNS) stable isotopes and the factors of state (DE and NJ), bay location (lower, middle, and upper), and season (spring, summer, and fall). Results from stomach content analysis (SCA) were used as a measure of short-term diet and were also used as prior distributions in the Bayesian isotope mixing models using C and N stable isotopes.

Generalized linear models illuminated that CNS isotopic values varied between states, bay locations, seasons, and some sampling stations. Contrarily, GLMs also found that certain sample groups of Weakfish appeared to exhibit a degree of site fidelity, as isotopic values of Weakfish collected in individual sampling stations occasionally remained stable from summer to fall, demonstrated by slow and rapid turnover tissues of muscle and liver, respectively. SCA and mixing model results revealed seasonal, locational, and state relationships for both individual

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prey species and groups of prey species. During the summer, Weakfish of all size classes from the middle bay consistently consumed higher frequencies of the same prey items (using percent frequency as the SCA method) compared to Weakfish from the lower and upper bays as revealed by stomach content analysis and stable isotope mixing models. Mixing model results also found that Weakfish consistently relied on gammarid amphipods (mainly *Ampelisca* sp.) as the most important prey item (>60% assimilation in many cases) throughout their estuarine residency, contradictory to the findings of previous studies that found mysid shrimp to be the most important prey item of small and medium size classes (by weight).

The stomach content and stable isotope analyses applied to juvenile Weakfish indicate the overall variability in diet and organic matter sources over different spatiotemporal scales throughout the nearshore areas of the Delaware Bay. This variability highlights the importance of small-scale and locational (lower, middle, upper bay) primary and secondary productivity contributing to Weakfish production. Based on the findings presented in this study, future landmanagement decisions should consider the importance of small-scale and locational variability of optimal nursery habitats of juvenile Weakfish in the Delaware Bay.

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LIST OF ABBREVIATIONS

AICc	Corrected Akaike's Information Criterion
BMA	Benthic Microalgae
DE	Delaware
DDFW	Delaware Division of Fish and Wildlife
DIC	Dissolved Inorganic Carbon
DIN	Dissolved Inorganic Nitrogen
EA	Elemental Analyzer
EBFM	Ecosystem Based Fisheries Management
FMP	Fishery Management Plan
GLM	Generalized Linear Model
IRMS	Isotope Ratio Mass Spectrometer
IQR	Interquartile Range
mt	Metric Tons
NJ	New Jersey
NJFW	New Jersey Fish and Wildlife
SCA	Stomach Content Analysis
SIA	Stable Isotope Analysis
SIMM	Stable Isotope Mixing Model
SPOM	Suspended Particulate Organic Matter
TEF	Trophic Enrichment Factor
YOY	Young of the Year
%F	Percent Frequency of Occurrence

CHAPTER 1: INTRODUCTION

1.1 Background of the Weakfish Fishery

The Weakfish, *Cynoscion regalis* (Bloch and Schneider 1801), is a commercially and recreationally valuable species currently managed under Amendment 4 of the Weakfish Fishery Management Plan (FMP) by the Atlantic States Marine Fisheries Commission (ASMFC; Brust et al. 2016). The species is found on the Atlantic Coast of North America, where it ranges from Florida to Nova Scotia (Wilk 1979) but it is primarily found from Cape Hatteras, North Carolina to Long Island, New York (Hildebrand and Schroeder 1928). Considered to be a marine transient or estuary dependent species (Boutin 2008; Boutin and Targett 2019), Weakfish undergo a northwesterly spring migration from offshore wintering grounds to coastal bays, sounds, and estuaries to spawn, and subsequently make the return migration in the fall to the continental shelf located from the Chesapeake Bay to Cape Lookout, North Carolina (Shepherd and Grimes 1984; Mercer 1989). Once caught in excess of one-million pounds (Bigelow and Schroeder 1953), Weakfish have supported a fishery since at least the 1800s, and a highly valuable commercial fishery since the 1920s, with average landings ranging from 2,000 to 4,000 metric tons (mt) per year (Lowerre-Barbieri 1994; Brust et al. 2009, 2016).

In 1980, the commercial catch of Weakfish for the Mid-Atlantic region peaked at 16,000 mt valued at over \$7 million, but slowly dropped due to overfishing and eventually crashed to 3,000 mt in the early 1990s (Figure 1.1; Brust et al. 2016; NOAA 2017). The fishery slightly rebounded in the late 1990s after mandatory catch limits were imposed in 1995 but crashed again due to unknown causes and has yet to rebound (Brust et al. 2016). Weakfish landings in the Mid-Atlantic reached an all-time low in 2011 at 60 mt, and the fishery is currently less than 1% of what it was 30 years ago when it was at its peak (Figure 1.1; Brust et al. 2016, NOAA 2017).

Considered to be an important fish species of the Mid-Atlantic region, the decline of the Weakfish population has critically impacted recreational and commercial anglers, especially charter fishing captains (Captain J. Stewart, personal communication).



Figure 1.1. Commercial Weakfish landings (mt) for the Mid-Atlantic Region from 1950 to 2016 (NOAA).

1.2 Research Problem

According to the most recent stock assessment (Brust et al. 2016), the Weakfish population is currently depleted – not overfished – and the primary cause is attributed to high rates of natural mortality which have been occurring since the early 2000s. The definition of natural mortality is defined as the rate of death in a population due to natural causes such as lifespan, starvation, disease, and predation (Blackhart et al. 2006). While investigating the causes behind the elevated rates of natural mortality, managers from the 2009 stock assessment (Brust et al.) discovered a shift in the Weakfish diet (1990s through the early 2000s) from forage fish and large invertebrates to only smaller invertebrates. These researchers positively correlated an increased incidence of empty stomachs of juvenile Weakfish emigrating from nurseries to overwintering areas with an increase in total mortality, linking starvation with natural mortality. Additional stomach content analysis reports from Delaware Bay (2007-2012) by Delaware Division of Fish and Wildlife (DDFW) showed that 29-53% of Weakfish had empty stomachs during their estuarine residency (Clark 2007, 2008, 2009, 2010, 2011, 2012). The findings by Brust et al. (2009) and Clark (2007, 2008, 2009, 2010, 2011, 2012) emphasize the significant role that nursery habitats play relative to the survival of Weakfish. Nevertheless, the major problem that Weakfish face is the unknown cause(s) of elevated natural mortality rates (Brust et al. 2016). This unknown underscores the need to further investigate the diet and food availability of Weakfish in their primary spawning and nursery habitats such as the Delaware Bay (Hildebrand and Schroeder 1928; Colton et al. 1979; Szedlmayer et al. 1990; Brust et al. 2016).

1.3 Research Purpose

Throughout the 1990s and 2000s, the diet of juvenile and adult Weakfish was studied extensively in the Delaware Bay (Grecay and Targett 1996a; Lankford and Targett 1997; Paperno et al. 2000; Nemerson 2001; Nemerson and Able 2004; Able et al. 2009, 2018; Torre and Targett 2017; Boutin and Targett 2019). As the Weakfish population declined and natural mortality increased, research efforts of Weakfish diet in the Delaware Bay dwindled, with the last study conducted in 2013 (Torre and Targett 2017). Regardless of the number of previous studies, none of them characterized the diet of juvenile Weakfish on both sides of the bay (New Jersey and Delaware) and over the course of their estuarine residency (July to October) simultaneously. As listed in the 2016 Weakfish Stock Assessment (Brust et al. 2016), a high

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priority was placed on monitoring the diets of Weakfish over broad regional and spatial scales with "an emphasis on new studies within estuaries". Additionally, the fishery management priorities at the federal level emphasize an ecosystem-based fisheries management (EBFM) approach, calling for more information regarding the myriad variables that affect fish populations, especially in estuarine nursery habitats such as the Delaware Bay (NOAA 2017). These research and management priorities at both the regional and federal levels reveal the lack of extensive studies on the diet of Weakfish through time, space, and within estuaries, as well as the need for more up-to-date information regarding Weakfish diet as it relates to factors that influence natural mortality.

As Brust et al. (2016) pointed out, one of the main factors potentially influencing Weakfish natural mortality is competition. Specifically, Weakfish managers have speculated that interspecies competition with Bluefish (*Pomatomus saltatrix*), Spiny Dogfish (*Squalus acanthias*), and other piscivorous marine transient species is one of the primary drivers of increased Weakfish natural mortality. In this study, I focus on the aspect of juvenile Weakfish diet as it relates to food availability and trophic niche in their nearshore Delaware Bay nursery habitats. The overarching goal of this research is to directly answer the high priority research question of life history, biology, and habitat posed in the 2016 Weakfish Stock Assessment. This goal will be met by investigating the diet of different size class juvenile Weakfish in the Delaware Bay over time (summer to fall), and space (both New Jersey and Delaware nearshore nursery zones and along the estuarine salinity gradient from 0-60 km from the mouth of the bay toward the Delaware River).

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1.4 Experimental Approach

To answer the species-specific and ecosystem-based questions as defined by Brust et al. (2016) and NOAA (2017), I examined the diets of juvenile Weakfish over the spatiotemporal scales mentioned above using stomach content and stable isotope analyses. I used stomach content analysis (SCA) as both a measure of short-term diet, and as priors for a Bayesian isotope mixing model in which carbon and nitrogen stable isotopes are used to determine the proportion of diet items that contribute to juvenile Weakfish tissue assimilation. Second, I used a Bayesian mixing model for the known most frequently occurring prey item of juvenile Weakfish, the mysid shrimp (*Neomysis* sp.), in an attempt to identify the drivers governing their production in the Delaware Bay and establish a link to the primary production supporting juvenile Weakfish growth.

1.5 Research Hypotheses and Objectives

Previous research (see sections 2.2 and 2.4) and preliminary results for stable isotope and stomach content analyses of juvenile Weakfish suggest that Weakfish feed on a variety of prey items during their estuarine residency and multiple variables drive the cycling of organic matter in the Delaware Bay over time and space. These findings provide a baseline of information to which the results of this study can be compared. Using the online tool, Mental Modeler (Gray et al. 2013), Figure 1.2 serves as a crude hypothesis with regards to juvenile Weakfish diet, trophic level, and sources of organic matter in the Delaware Bay in terms of δ^{13} C as a measure of carbon source. The arrows in Figure 1.2 signify various connections. Starting with the bottom level, upper bay and lower/middle bay, the arrows indicate how bay locations affect the dominant marsh macrophyte and SPOM production. This relationship is mediated by salinity regime and the salinity tolerances of the plants, where *Phragmites* flourishes in oligohaline water and Spartina thrives in polyhaline water. The next level up, *Phragmites australis*, *Spartina alterniflora*, and SPOM, shows the connections between these basal food web sources and the predator prey relationships as they pertain to the Delaware Bay juvenile Weakfish food web. With regards to the δ^{13} C values of different food web components, the blue and red arrows indicate the δ^{13} C values of sources in terms of contributions of more enriched (+) or depleted (-) values, respectively. Generally, *Phragmites*-dominated systems are more depleted in δ^{13} C versus their *Spartina*-dominated counterparts.



Figure 1.2. A Delaware Bay food web based on preliminary results and previous studies. The hierarchical structure represents how bay location (salinity) affects the basal food web sources in the bay, where arrows represent food web connections. Beginning at the second level from the bottom, colors represent hypothetical influences with regards to δ^{13} C values (either enriched or depleted). See above paragraph and sections 2.1 and 2.2 for details.

This research focused on answering three questions:

1) What drives carbon, nitrogen, and sulfur stable isotope values of different size class Weakfish in terms of state (Delaware and New Jersey), season (summer and fall), and bay location (lower, middle, and upper)?

2) What is the diet of different size class juvenile Weakfish in the Delaware Bay over time (seasonally) and space (Delaware and New Jersey coasts for the lower, middle, and upper bay) as described by SCA and stable isotope mixing models?

3) What are the primary sources of organic matter that contribute to mysid shrimp production as described by stable isotope mixing models using marsh macrophytes *Spartina alterniflora*, *Phragmites australis*, suspended particulate organic matter (SPOM), and benthic microalgae (BMA) as source values?

The following chapters will serve as descriptors of this study. Beginning with Chapter 2: Literature Review, I introduce and review the following: 1) the study site, 2) past findings of Weakfish diet in the Delaware Bay, 3) SCA methods of fish and arguments for the method chosen for this study, 4) previous findings of stable isotope analysis in the Delaware Bay, 5) other studies using SCA coupled with stable isotope analysis, 6) stable isotope mixing models, 7) stable isotope turnover time, and 8) lipid correction in stable isotope studies. Chapter 3: Materials and Methods, describes the sample collection methodology, laboratory methods and statistical analyses for both stomach content and stable isotope data. Chapter 4: Results, breaks down the SCA results by year and size classes, followed by the mixing model results for Weakfish and mysid shrimp. Chapter 5: Discussion, Conclusions, and Future Research Recommendations, summarizes the interpretations of the results and the implications of this study for future management.

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CHAPTER 2: LITERATURE REVIEW

2.1 Study Site

The Delaware Bay is a drowned river valley estuary that serves as a nursery for a variety of marine transient and resident fishes (Nemerson 2001; Litvin and Weinstein 2004; Nemerson and Able 2004; Able 2005; Boutin 2008; Able et al. 2009; Boutin and Targett 2019). The bay has a pronounced spring bloom with high productivity and significant phytoplankton biomass accumulation that has historically occurred from March through mid-April (Pennock and Sharp 1986; Fogel et al. 1992; Able et al. 2009, 2018) and has been noted to occur between 30-50 km from the bay mouth (Yoshiyama and Sharp 2006; Sharp 2010).

The physicochemical conditions of the bay vary as you proceed toward the river, from near-oceanic conditions at the mouth of the estuary to near riverine conditions at the head (Nemerson and Able 2004). Its perimeter is nearly completely lined with salt marshes (Able et al. 2009, 2018), with a marked change in the species of marsh macrophytes along the salinity gradient – from *Spartina* spp. dominated marshes (mostly *Spartina alterniflora*) in the lower and middle bay to *Phragmites australis* dominated marshes in the upper bay (Figure 2.1; Litvin and Weinstein 2004; Able et al. 2009, 2018). Each side of the Delaware Bay is somewhat unique in terms of the rivers and creeks that flow into it. On the Delaware side, there are a multitude of tidal creeks and multiple rivers that drain into the bay. The New Jersey side is roughly equal to the Delaware side with regards to the number of marsh creeks but differs in that it has two significantly larger rivers, the Maurice and the Cohansey, that drain into the bay.



Figure 2.1. Map of the Delaware Bay. Highlighted areas in yellow represent the cover of the marsh macrophyte *Phragmites australis*. These zones were based from visual surveys and aerial photos. Map Credit: Smith and Katkowski (2011).

2.2 Past Findings of Weakfish Diet in the Delaware Bay

The East Coast of the United States is home to many estuarine systems that contain valuable nursery habitats for juvenile fishes, with the Delaware Bay being one of the largest (Lankford and Targett 1994; Grecay and Targett 1996a; Paperno et al. 2000, Boutin 2008). Larval and juvenile marine transient species such as Weakfish use estuaries extensively, benefitting "from a combination of abundant resource availability, low predation risk, and suitable physicochemical conditions" such as salinity and temperature (Lankford and Targett 1994; Beck et al. 2001; Nemerson and Able 2004; Vasconcelos et al. 2010). These features of estuaries and their associated nursery habitats have been observed to enhance the feeding and growth rates of juvenile fishes during their estuarine residency (Lankford and Targett 1994; Grecay and Targett 1996a; Beck et al. 2001; Boutin 2008; Nagelkerken et al. 2015; Boutin and Targett 2019). Houde (1989) contended that subtle differences in individual growth rates significantly affect the survival of larval and juvenile fishes. Lankford and Targett (1994) expanded upon Houde's idea, claiming that differences in growth rates among juvenile fishes are influenced by feeding and are significant factors of recruitment success, especially for juvenile Weakfish during their Delaware Bay residency. These findings point out the importance of estuaries to marine transient fishes such as Weakfish and illustrate the significance of the Delaware Bay nursery habitat for their growth and survival.

The Delaware Bay is one of the principal spawning and nursery habitats for Weakfish, where adults exhibit protracted spawning from mid-May through July (Hildebrand and Schroeder 1928; Colton et al. 1979; Szedlmayer et al. 1990). Larval Weakfish settle throughout the extensive salt marshes, nearshore areas, and open waters of Delaware Bay from late spring to fall and larger juveniles are found throughout the bay from July through October (Paperno et al. 2000; Litvin et al. 2011). Juvenile Weakfish feed on a variety of prey items: mysid shrimp (mainly *Neomysis americana*), Bay Anchovy (*Anchoa mitchilli*), Mummichog (*Fundulus heteroclitus*), sevenspine bay shrimp (*Crangon septemspinosa*), small crabs, razor clams, copepods, polychaete worms, larval horseshoe crabs (*Limulus polyphemus*), isopods (mainly *Edotia triloba*), and several species of gammarid amphipods (e.g. *Ampelisca* spp. and *Cerapus tubelaris*; Wilk 1979; Nemerson 2001; Nemerson and Able 2004; Boutin 2008; Willis et al. 2015; Boutin and Targett 2019, personal observation).

Within their marsh nursery habitats, the diet of juvenile Weakfish has been studied extensively. The species undergoes an ontogenetic shift in diet—feeding mainly on mysid shrimp (almost exclusively N. americana) when small (10-60 mm SL), to increasing proportions of fish when larger (60-100 mm SL), transitioning to an equal proportion of fish and crustaceans (by weight) when >100 mm SL (Nemerson 2001; Litvin and Weinstein 2004). In a field study, Grecay and Targett (1996a) discovered that the percent of mysids found in Weakfish stomachs was positively correlated with their size and condition in mid-salinity zones (15-20 ppt) of the Delaware Bay. An additional follow-up laboratory study by Lankford and Targett (1997) showed that the average energy intake rate of juvenile Weakfish was higher for individuals that fed on mysids rather than on other common shrimp species (e.g. C. septemspinosa), indicating that juvenile Weakfish converted a higher percentage of mysid energy into somatic growth. Nemerson (2001) described the seasonal prey consumption of juvenile Weakfish in the Delaware Bay and found that piscivory becomes important from July through September with consumption rates of approximately 20-40% fish by weight. These studies demonstrate the importance of mysids to the diet of juvenile Weakfish, confirm their ontogenetic shift in diet, and highlight the

need to understand the current state of prey availability as a function of diet and as a potential factor in natural mortality and/or recruitment success.

Despite the numerous studies focusing on juvenile Weakfish diet in the Delaware Bay, all of them used bulk SCA methods to determine diet. Although these methods are effective and informative measures of fish diet, their scope of inference is inherently limited to the specific days that the fish were collected, or at best, one or two days prior to collection. This project aims to widen the scope of inference compared to traditional SCA methods using SCA results as priors for a Bayesian stable isotope mixing model. Due to the planned use of the mixing model, I used a simple and effective measure of SCA that is still under debate by fisheries biologists (Baker et al. 2014).

2.3 Stomach Content Analysis Methods of Fish

Understanding the relationships that drive fish production is arguably the fundamental goal of fish ecology. Until the 1970s, SCA was the only method that fish biologists could use to determine the feeding habits of fish. The valuable role that SCA plays in fisheries biology and management has led to the creation of a variety of methods that have been developed, modified, and described (Hynes 1950; Hyslop 1980; Liao et al. 2001; Ainsworth et al. 2010; Brown et al. 2012; Moriarty et al. 2016).

Today, the most common indices used to describe the relative importance of prey taxa include percent by number (%N), weight (%W), volume (%V), and frequency (%F), as well as the index of relative importance (IRI) and the modified index of relative importance (MIRI; Liao et al. 2001). Considering this variety, there is still debate among fisheries biologists about which method best describes the dietary importance of specific prey taxa and which should be used as

the standard method in all SCA studies of fishes (Hynes 1950; Hyslop 1980; Cortes 1998; Brown et al. 2012; Baker et al. 2014; Buckland et al. 2017).

Hyslop (1980) noted the limitations of %N, %W, and %F, while others argued over the use of compound indices (e.g. IRI, MIRI) as meaningful methods to describe the importance of prey taxa to fish diets (Pitcher 1981; Cortes 1998; Liao et al. 2001; Brown et al. 2012). Percent number is limited due to its overemphasis of small prey, difficulty in identification of masticated prey, and its inherent lack in accounting for fish size. Percent weight overemphasizes single heavy items, and formalin, which is commonly used in fish stomach preservation and can increase the weight of diet items and thus overestimate their weight. Percent frequency is biased as it gives little indication of the relative amount or bulk of each food category present in the stomach. As these indices of prey importance have their limitations, researchers have suggested the use of compound indices (IRI and MIRI) to absorb biases associated with single indices (Pinkas et al. 1971; Hyslop 1980; Pitcher 1981; Cortes 1998). However, Brown et al. (2012) proved that the IRI overemphasizes frequently occurring prey and underemphasizes rare prey (Ortaz et al. 2006). On the other hand, Liao et al. (2001) suggested that MIRI acts similarly to %W. Clearly, every method of SCA has its limitations. Nevertheless, Hyslop's (1980) review paper was widely influential in that %W became the most popular index among fisheries scientists (Grecay and Targett 1996a; Nemerson 2001; Boutin 2008; Torre and Targett 2017; Able et al. 2018; Boutin and Targett 2019) to describe the dietary importance of prey taxa and the relationships between fish condition and prey availability (Liao et al. 2001; Baker et al. 2014).

Although %W has been the index of choice among fisheries scientists, Baker et al. (2014) raised an insightful argument against such metrics and proposed the use of %F as the only

reliable metric of prey importance. These researchers outlined two important arguments in support of this idea: 1) due to partial digestion, it is not physically possible to separate different prey items with an appropriate level of accuracy; and 2) the detailed stomach composition observed at a given point in time is the outcome of myriad immeasurable factors that interact to prevent the observed composition from providing an accurate representation of the actual composition of the prey consumed. In their first point, the authors explained that the level of prey digestion determines the difficulty in accurately separating prey taxa (Hyslop 1980), which varies depending on ingestion time, evacuation rates, temperature, and prey handling (Macdonald and Green 1983; Legler et al. 2010; Baker et al. 2014). Furthermore, prey items are frequently coated in mucous that is often physically impossible to separate, adding yet another source of bias in the calculation of %W of such prey items.

In their second point, Baker et al. (2014) pointed out that the actual composition of a fish stomach at a single point in time is the result of a broad range of immeasurable factors that are unrelated to the actual composition of the consumed diet. Competition, movement, prey location, anthropogenic stimuli, and other factors all play a role in the stomach contents of fish. Furthermore, prey handling and different digestion and evacuation rates of different prey types combine to provide %W data that are vague and difficult to interpret (Hyslop 1980; MacDonald et al. 1982; Baker et al. 2014). Conversely, %F data are precise and unambiguous because the values recorded simply represent the proportion of individuals containing a positively identified prey type. Baker et al. (2014) showed that when sample sizes are large enough, quantifying the diet by %F or %W provides similar results of dietary importance of prey taxa to fish. In other words, the greater the bulk of any consumed prey, the more likely it is to occur in the stomach of any individual (Royle and Nichols 2003). Percent frequency is thus a reliable metric in

determining the dietary importance of prey taxa in fish. Based upon these arguments presented by Baker et al. (2014), I used %F on juvenile Weakfish in this study rather than %W. Regardless of the number of SCA metrics that are employed today, the fundamental questions that drive the use of such techniques remain. Biologists are interested in the importance of prey taxa to predator growth and the relationships between prey importance and availability (Liao et al. 2001). Accurately quantifying these relationships is critical for effectively managing fisheries resources (Bowen 1996).

2.4 Stable Isotope Ecology

2.4.1 Stable Isotope Studies in the Delaware Bay

A motif in estuarine studies is that estuaries are highly dynamic and complex ecosystems with inputs, exchanges, and organic matter processing all playing a role in supporting abundant and diverse groups of fishes and invertebrates (Deegan and Garritt 1997; Beck et al. 2001; Bouillon et al. 2012; Litvin et al. 2018). To understand these dynamic estuarine processes, researchers have used stable isotope analysis (SIA). In the Delaware Bay, SIA has been applied to Weakfish to describe their movements, habitat use patterns, and utilization of estuarine organic matter (Wainright et al. 2000; Weinstein et al. 2000; Litvin and Weinstein 2004; Litvin et al. 2014). These studies have shown that juvenile Weakfish exhibit site fidelity but begin a saltatory behavior toward the bay mouth before emigrating from the estuary. Additionally, these studies postulated that locally produced detritus and organic matter from *Spartina spp*. and *Phragmites australis* contribute to the isotopic values of Weakfish. Other studies in the bay describe seasonal dynamics of dissolved inorganic and organic carbon (DIC and DOC δ^{13} C), dissolved inorganic and organic nitrogen (DIN and DON δ^{15} N), and the sources of local estuarine, terrigenous, and urban nutrient inputs (Fogel et al. 1992; Yoshiyama and Sharp 2006;

Sharp et al. 2009), revealing the multiple sources that contribute to the isotopic values of estuarine primary producers and thus consumers.

In the Delaware Bay and other Mid-Atlantic estuaries, these sources are characterized by the marsh macrophytes of the genus *Spartina* and species *Phragmites australis*, and benthic microalgae (BMA; Wainright et al. 2000; Weinstein et al. 2000; Currin et al. 2003; Litvin and Weinstein 2004). *Spartina* spp., especially *S. alterniflora*, are the native and dominant macrophytes in the Delaware Bay estuary. These plants are salt tolerant; thus, they occur in areas of high salinity and prolonged saltwater inundation. *P. australis* is an invasive marsh grass that cannot tolerate tidal waters with salinities much greater than 10‰. The salinity tolerances of these two plants create a gradient of marsh grasses driven by salinity (Section 2.1, Figure 2.1). This marsh grass gradient is also reflected isotopically due to the large differences in δ^{13} C caused by their differing photosynthetic pathways, with the C4 plant *Spartina* exhibiting more enriched δ^{13} C values (-14‰ to -12‰) relative to the more depleted values of the C3 plant *P. australis* (-25‰ to -28‰). The locally produced detritus and organic matter from these plants plays an important role in the isotopic values of consumers, allowing inference of which marsh macrophytes contribute to the food web of consumers along the salinity gradient.

The relative contribution of BMA is difficult to determine by δ^{13} C alone. δ^{34} S SIA is commonly employed in conjunction with δ^{13} C to separate the significance of BMA in estuarine food webs (Deegan and Garritt 1997; Wainright et al. 2000; Currin et al. 2003; Connolly et al. 2004; Litvin and Weinstein 2004; Bouillon et al. 2012). Using δ^{34} S of BMA in the Delaware Bay along with δ^{13} C and δ^{15} N of juvenile Weakfish, Litvin and Weinstein (2003, 2004) determined that BMA plays less of a role in the nutrition of Weakfish and found that their isotopic signatures were consistent with organic matter derived from SPOM and the dominant marsh macrophyte of the collection location. Furthermore, it has been suggested by Deegan and Garritt (1997) that δ^{34} S values >14‰ are indicative of a more pelagic-based food web, whereas δ^{34} S values <14‰ are indicative of a marsh-based food web.

The influences that end-members of phytoplankton, *Spartina* spp., *P. australis*, and BMA can have on the isotopic values of consumers is important to understand when interpreting the data collected from estuarine consumers.

2.5 Stable Isotope and Stomach Content Analyses in Studies of Fish Diet

As the use of stable isotopes in ecosystem and diet studies has increased, so has the coupling of SCA and SIA of fish. Multiple studies employ SIA in addition to SCA as means of increasing the resolution of the diet of a species of interest or determining its isotopic/trophic niche (Grey et al. 2002; Sherwood and Grabowski 2006; Sherwood et al. 2007; Hadwen et al. 2007; Buchheister and Latour 2011; Boyle et al. 2012; Kadye and Booth 2012; Jo et al. 2013; Pacella et al. 2013; Chung-Nan et al. 2015; Fuhrmann et al. 2017; Lee et al. 2019). Many of these studies utilize δ^{13} C as a measure of trophic niche, where depleted values are representative of a more pelagic based food web, and enriched values are indicative of a benthic food web. δ^{15} N is typically used as a measure of trophic level, with more enriched values signifying feeding at higher trophic levels.

Other studies have focused their efforts on creating isotopic food webs from the stable isotope values of the consumer's known and potential prey items. Gathering the isotope values of prey items elucidates the importance of prey species or groups that may not show themselves to be significant in the SCA results, leading to the use of stable isotope mixing models (SIMMs) to determine the relative proportions of prey items contributing to the tissue growth of fish. Although many of the coupled SIA/SCA studies employ a mixing model (Grey et al. 2002; Hadwen et al. 2007; Buchheister and Latour 2011; Boyle et al. 2012; Kadye and Booth 2012; Pacella et al. 2013; Chung-Nan et al. 2015; Lee et al. 2019), there are only a handful of published studies that use the most recently developed SIMM (MixSIAR) with fish as the consumer of interest (Gutmann Roberts et al. 2017; McClain-Counts et al. 2017; Samways et al. 2018; Latli et al. 2019; Lee et al. 2019). However, none of these researchers used SCA data as priors in the model, leaving open the question of how MixSIAR performs for diet partitioning using priors of known prey items of fish.

2.5.1 Mixing Models in Stable Isotope Ecology

Recently, SIMMs such as IsoSource (Phillips and Gregg 2003) have been developed to incorporate uncertainties in trophic enrichment factors (TEFs) and variability in the source and tracer data which are not limited by the ratio of sources to tracers (underdetermined system) (Phillips et al. 2014; Stock et al. 2018). Moore and Semmens (2008) introduced the Bayesian mixing model called MixSIR, which established a Bayesian likelihood framework for estimating source contributions and accounted for source and tracer variability (Stock et al. 2018). Since MixSIR was introduced, Bayesian mixing models have quickly advanced to become a flexible linear modeling framework (Parnell et al. 2013; Stock et al. 2018). More recently, Stock and Semmens (2018) created the open-source R software package, MixSIAR, which unifies several of the Bayesian SIMMs and is described in detail by Stock et al. (2018).

Within the Bayesian framework, the power in MixSIAR lies in its ability to incorporate informative prior distributions from stomach content data. Priors incorporated into the model can either be uninformative (uniform distribution) or informative (e.g. distribution from stomach content data). In this study, I used informative priors based upon stomach content data of Weakfish (see section 3.5.2.2 for details). After incorporating source and mixture data and priors, MixSIAR writes a custom JAGS (Just Another Gibbs Sampler) model file using Markov Chain Monte Carlo (MCMC) to produce diagnostics, posterior probability plots, and summary statistics of estimated median diet proportions including 95% credible intervals (Stock et al. 2018).

MixSIAR is a powerful tool for the stable isotope ecologist, but the user must be careful in creating the model and fitting the parameters. Additionally, mixing model users must account for the assumptions and limitations of the model used. TEFs must be specified, but this is typically derived from the literature and thus adds another source of uncertainty. Lastly, the influence of the prior can be a source of bias in the posterior distributions as its influence can increase with fewer data points, greater variance and less separation between source data. Despite the assumptions and limitations of MixSIAR and SIMMs, MixSIAR is an excellent tool for the stable isotope ecologist which incorporates multiple SIMMs in a single software package in R, and I use it in this study to partition the diet of juvenile Weakfish over time and space in the Delaware Bay. To my knowledge, this study is among the first to employ MixSIAR on fish and will also be one of the first to use SCA results of fish as informative priors in MixSIAR.

2.5.2 Stable Isotope Turnover

Stable isotope analysis is an excellent tool to elucidate multiple relationships between an organism of interest and the ecosystem that supports it. Nevertheless, many studies that couple SIA and SCA do not properly consider the turnover time of a fish's tissues, limiting the inference of what stable isotope values actually represent (Grey et al. 2002; Hadwen et al. 2007; Boyle et al. 2012; Kadye and Booth 2012; Chung-Nan et al. 2015; Lee et al. 2019). Thus, it is important to consider the differential turnover time of fish tissues to make more meaningful interpretations.

Isotopic turnover has been studied at length (Fry and Arnold 1982; Tieszen et al. 1983; Hesslein et al. 1993; Herzka and Holt 2000; MacAvoy et al. 2001; Bosley et al. 2002; Fry 2006; Logan et al. 2006; Phillips and Eldridge 2006; Buchheister and Latour 2010; Nelson 2011; Vander Zanden et al. 2015) and researchers have found that the interaction of growth and metabolism gives each tissue type a unique turnover rate. Metabolism-based turnover is the replacement or conversion of existing tissue using material from the recent diet whereas growthbased turnover is the dilution of existing mass by new mass created from recently consumed prey. Therefore, tissues that are metabolically active (e.g. liver) respond quicker to dietary changes, but other tissues such as muscle may take over one year to turn over (Vander Zanden et al. 2015). Variable turnover times of different tissues allows ecologists to infer the diet of a consumer over different time scales. In fish, the general relationship between growth and metabolic turnover times of different tissues is liver>blood>muscle, where liver exhibits the most rapid turnover time.

While many studies that use SCA and SIA to study fish diet consider the turnover time of tissues, many do not model turnover time due to lack of information regarding the species of interest (Cocheret de la Morinière et al. 2003; Sherwood et al. 2007; Boyle et al. 2012; Kadye and Booth 2012; Chung-Nan et al. 2015). Applying turnover rates to SIA data allows the researcher to interpret the data with increased credibility and more meaningful inference. In this study, I model turnover time of Weakfish muscle and liver tissues using a growth based turnover model intended for use in field studies (Buchheister and Latour 2010).

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2.5.3 Lipid Correction of $\delta^{13}C$

Within an organism, lipids are depleted in ¹³C compared to proteins and carbohydrates. In SIA, lipid content among organisms or different tissue types may bias δ^{13} C values (Kiljunen et al. 2006; Post et al. 2007; Logan et al. 2008; Reum 2011). The increasing use of SIA for ecological management issues and questions requires that researchers account for the lipid bias found within the tissues of their organisms of interest. This is typically done using a lipid-normalization method via a model that describes the relationship between the change in δ^{13} C following lipid removal and the original carbon:nitrogen (C:N) ratio of a sample (Kiljunen et al. 2006). Post et al. (2007) described the relationship between C:N ratios and δ^{13} C of aquatic animals. These researchers suggested that a C:N ratio between 3.5-7 be lipid normalized due to the presence of lipids that would significantly bias the δ^{13} C values of a sample. Post et al. (2007) also found that % carbon is a greater indicator of lipids in plants and suggested that plant tissues with greater than 40% carbon be lipid normalized.
CHAPTER 3: MATERIALS AND METHODS

3.1 Sampling Design

The sampling design for this project was a modified systematic stratified design derived from the Delaware Division of Fish and Wildlife (DDFW) and New Jersey Fish and Wildlife (NJFW) Delaware Bay juvenile finfish sampling stations. Thirty-six sampling sites located along the shorelines of the Delaware Bay were sampled by DDFW and NJFW juvenile finfish trawl surveys from May through October. To simplify the statistical design and because of expected changes in diet and isotopic values over time, months were pooled into seasons: May/June – spring, July/August – summer, and September/October – fall. Of the 36 stations, 11 are located in New Jersey and 25 are located in Delaware (Figure 3.1). The bay was separated into three parts along the salinity gradient (lower, middle, upper) using salinity data (averaged over 2014 through 2016) provided by both DDFW and NJFW from all Delaware Bay sampling sites (Figure 3.1), as well as 6 stations down the center of the bay not shown in Figure 3.1. The salinity zones were classified according the Venice Classification System (Anonymous 1958): upper (oligo-mesohaline; 5-12 ppt), middle (meso-polyhaline; 12-20 ppt), and lower (polyeuhaline; 20-34 ppt). The bay was separated in this manner due to the expected changes in isotopic values along the salinity gradient from conservative mixing and the Spartina-Phragmites gradient (Fry 2002; Litvin and Weinstein 2004). The Delaware Bay was then split in half to create six total strata representing the salinity gradient for both states: DE-upper, DEmiddle, DE-lower and NJ-upper, NJ-middle, and NJ-lower. The number of trawl stations within each stratum varied according to the salinity range of the three bay locations. For example, NJupper only consisted of one site due to the salinity range characterized by the upper bay and the relatively few stations in NJ compared to DE. All fish samples (except larval clupeids) were

collected by DDFW and NJFW trawls and were provided on an as-available basis, therefore, NJupper had limited data as Weakfish were not always caught at the single station that comprised this stratum.

Trawl sites selected by DDFW were initially chosen in 1977 from trawl sites used to monitor blue crab populations (Cole et al. 1978). These stations were randomly selected in grid squares by superimposing a 6,000-yard grid scheme on a navigational map of Delaware Bay. No documentation was found regarding how the stations in New Jersey were selected; however, I was informed that the NJ stations were selected as an extension of the map developed for DE (Hassel, personal communication).

As shown in Figure 3.1, DDFW has many more sampling stations than NJFW, resulting in more Weakfish being caught from the Delaware side of the bay. I attempted to account for the Delaware state bias by using fish caught from the southern, middle, and northern stations per each stratum. This accounted for within stratum variability along the salinity gradient and standardized the stations I used for collecting prey samples. When it was not possible to use samples from the southern, middle, and northern stations per stratum, I analyzed available specimens caught from adjacent stations to keep the best representation within a salinity zone. For both SIA and SCA, my goal was to collect at least 5 Weakfish per size class, per station (northern, middle, southern), per stratum, per month.

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Figure 3.1. Map of the Delaware Bay separated into three salinity zones and two states. Numbers represent the trawl locations sampled by DDFW and NJFW. The salinity gradient is based from salinity values collected from 2014-2016 from each station. Map credit: Devin Mendez.

3.2 Field Collections

3.2.1 Weakfish

In both 2017 and 2018, all Weakfish were collected by DDFW and NJFW juvenile finfish trawl surveys. Both state agencies used a 4.9 meter (m) otter trawl with a 3.8 centimeter (cm) stretch mesh cod-end lined with a 1.3cm stretch-mesh knotless liner. All tows were deployed against the tide for 10 minutes. Once caught, Weakfish were placed in labeled freezer bags and placed on ice. Upon return from the field, the specimens were placed in a -80° C freezer for storage. When possible, three size classes (small <60, medium 60-100, large 100-137 mm SL) of Weakfish were collected from each sampling station. Since Weakfish were provided on an as-available basis, not all size classes were caught in each month and from each station.

3.2.2 Weakfish Prey

From May to September of 2018, potential Weakfish prey items were caught for use in SIMMs. No prey samples were caught in 2017. Prey sampling was conducted on the Delaware side of the bay using Delaware State University's 5.5m flat bottom Olympic Jon boat at the southern, middle, and northern stations per stratum. However, due to boat issues, the center and northern stations of the DE-middle and all DE-upper strata were not sampled in August, and all the DE sites were not sampled in September. Prey sampling was conducted on the New Jersey side of the bay using the NJFW RV James W. Joseph. DDFW and NJFW provided Bay Anchovy and other small potential fish prey; however, larval fishes (mostly larval clupeids) and invertebrates were caught using a Wildco Bottom Sled with 500µm mesh net (52 X 38 X 13 cm), a plankton tow net (75 X 300cm with 333µm mesh and a 50 to 75cm reduction cowling), and a 0.5 liter van Veen sediment grab. Bay Anchovy were separated into three size classes for SIA (<30, 30-60, >60 mm SL). All plankton and benthic sled tows were towed against the tide at a

speed of 1-2 knots for 10 minutes when possible. However, due to fouling and water depth issues, tows were sometimes limited to 3-5 minutes and the plankton tow net was not always used. On the NJFW RV James W. Joseph, the plankton tow and otter trawl were deployed simultaneously, with the plankton tow being deployed off the starboard side outrigger. The tow line was outfitted with a custom-made planar board with weights attached to the bottom to allow the net to be submerged while the RV was in gear. The benthic sled was deployed on its own after the otter trawl was completed. The van Veen grab was used at the northern- and southern-most stations to collect infauna. All tows and sediment grab contents were placed in labeled freezer bags and on ice in the field and stored in a -80°C freezer for future analyses.

3.2.3 Marsh Macrophytes

The marsh macrophytes *S. alterniflora* and *P. australis* were collected in 2018 from the shorelines of both the NJ and DE sides of the bay from the central stations in each stratum. Samples were collected every other month beginning in May and June in DE and NJ, respectively. Three plants of each species were collected. *P. australis* was not present in the middle-DE station. In NJ, macrophytes for the lower bay were taken from two separate locations as *S. alterniflora* and *P. australis* were not present in the same locations that were accessible by road. Macrophytes from NJ-middle were taken from the shore property of the Haskins Shellfish Research Laboratory dock located on the Maurice River delta, and NJ-upper macrophytes were taken from the coastal town of Seabreeze, NJ. All plants were placed in labeled freezer bags, placed on ice, and stored in a -80°C freezer.

3.2.4 Suspended Particulate Organic Matter

Suspended particulate organic matter samples were collected by filling 500 mL Nalgene bottles with surface water. All SPOM samples were collected from the northern- and southernmost stations per stratum once per month from May to September. To account for variability in the SIA samples of SPOM, duplicate samples were collected for a total of four samples per stratum. All samples were placed on ice in the field and returned to the lab for same-day processing (see Section 3.4.4 for details).

3.2.5 Water Quality

Water quality parameters were collected using multiple YSITM Multiprobes (Yellow Springs Instruments, Yellow Springs, Ohio). Temperature, salinity, pH, and dissolved oxygen were measured at each sampling station. I used a YSITM Pro Plus in the field when conducting prey sampling on the DE side from May to August. DDFW and NJFW used a YSITM Model Pro 2030 and YSITM Model 85, respectively. All water quality data collected by DDFW was electronically shared with me.

3.3 Stomach Content Analysis

To measure the diet of juvenile Weakfish, SCA was performed using the percent frequency of occurrence method (%F) (Hyslop 1980; Baker et al. 2014; Buckland et al. 2017; Fuhrmann et al. 2017). Weakfish stomach contents were analyzed per size class, bay location, month, and state.

3.4 Laboratory Processing and Stable Isotope Analysis

Stable isotope analysis was conducted for all size classes of Weakfish and Bay Anchovy. Weakfish muscle and liver tissues were both analyzed for stable isotopes due to their differential turnover times. Stable isotope δ values were calculated using the formula: $\delta^{H}X$ [($R_{sample}/R_{standard}$ – 1)]*1000. The superscript H is the heavy isotope of the element X and R is the ratio of heavy to light isotope (e.g. ¹³C/¹²C) of the sample and the standard, respectively. All samples processed for SIA were dried to a constant weight in an oven for 48 hours at 60°C, ground to a fine powder using a mortar and pestle (when applicable), packaged into 4x6mm or 5x10mm aluminum capsules, and placed into a 96-well tray. One-hundred and twenty samples from 2017 were analyzed by the University of Maryland Center for Environmental Science Central Appalachians Stable Isotope Facility (CASIF) and the remainder were sent to the University of New Mexico Center for Stable Isotopes (UNM-CSI) for isotopic analysis.

CASIF used a Carlo Erba NC2500 elemental analyzer (EA) interfaced with a Thermo Delta V+ isotope ratio mass spectrometer (IRMS). The long-term error for this instrument for the QC standards is $\pm 0.12\%$ for δ^{13} C and $\pm 0.11\%$ for δ^{15} N. For δ^{13} C, standards from CASIF were verified for accuracy against the internationally known standards NBS 18, NBS 19, and L-SVEC. These were then calibrated against the international standard Vienna-Pee Dee Belemnite (V-PDB) scale. δ^{15} N samples were measured against the international standard of air.

UNM used a Costech 4010 EA coupled to a ThermoFisher Scientific Delta V Advantage mass spectrometer via a CONFLO IV interface for δ^{13} C, δ^{15} N, and δ^{34} S. Isotope ratios were reported using the standard δ notation relative to V-AIR and to V-PDB. Three internal laboratory standards were run at the beginning, during, and end of each analytical session. The standard error rates for δ^{13} C and δ^{15} N were ±0.05 and ±0.07, respectively. The laboratory standards were calibrated against IAEA N1, IAEA N2 and USGS 43 for δ^{15} N and NBS 21, NBS 22 and USGS 24 for δ^{13} C. For δ^{34} S, international standards NBS122, NBS123 and NBS127, and a laboratory standard (CP1) covering a range of δ^{34} S values from –4.6‰ to 20.3‰ were run at the beginning, during, and end of each analytical session. The standard error rates for δ^{34} S were calculated to be ±0.3‰ and were normalized to the standards listed above.

3.4.1 Fish

3.4.1.1 Weakfish

Weakfish were weighed, measured for standard and total length, and the stomachs were removed from a subsample of fish per station and size class. Stomachs were excised and preserved in a 10% formalin solution in exetainer vials for future SCA. For SIA preparation, scales on the dorsal region were removed with a blade, and muscle tissue was excised, rinsed with deionized water, placed into a labeled aluminum weigh boat, and dried. When available, livers from three Weakfish of the medium and large size classes were excised and treated in the exact manner as the muscle tissue for SIA preparation.

3.4.1.2 Prey Fish

The only prey fish species of Weakfish that were included in SIA were Bay Anchovy. For Bay Anchovy, scales and skin were scraped with a scalpel along the left lateral side of the body, and the muscle tissue was removed and prepared for SIA. When individual anchovies were too small to allow for sufficient muscle mass to be removed, the head, stomach, and fins were removed, and the rest of the fish was processed whole for SIA. All muscle tissue and processed whole fish was rinsed with DI water, dried, and processed for SIA.

3.4.2 Invertebrates

Two types of invertebrate prey items were processed for SIA for use in SIMMs: softbodied prey and hard-bodied prey (prey with a chitinous exoskeleton). The only soft bodied invertebrates processed for SIA were polychaete worms. Since polychaetes typically had full intestines, a blunt object was carefully pressed along the length of the animal from the anterior to the posterior to remove the stomach contents to prevent them from biasing the SIA results. Hard bodied prey items included mysid shrimp (mainly *N. americana*), gammarid amphipods (*C. tubelaris, Ampelisca* sp., and other unidentified gammarid spp.), isopods (mainly *E. triloba*), and sevenspine bay shrimp (*C. septemspinosa*). All hard-bodied invertebrates were acidified with a 1N HCl solution to remove carbonates (Pinnegar and Polunin 1999; Buchheister and Latour 2011). The acid solution was applied to dried/ground samples inside the mortar and was air-dried under a hood overnight prior to encapsulation.

3.4.3 Marsh Macrophytes

Following the protocols from Wainright et al. (2000), three individual plants of *S*. *alterniflora* and *P. australis* were processed in an equal manner. Five leaves were chosen from each plant as a composite sample and each were scraped with a scalpel to remove epiphytes. Due to difficulties powderizing leaf tissues with a mortar and pestle, leaves were dried for 72 hours at 60° C rather than the traditional 48 hours. This removed all moisture from the leaves and facilitated ease of grinding.

3.4.4 Suspended Particulate Organic Matter

Suspended particulate organic matter was processed in the laboratory directly after returning from the field. About 500 mL of water was filtered through pre-combusted 47 mm WhatmanTM GF/F using a low-pressure hand pump. Following the protocol of Levin and Currin (2012), GF/Fs were acidified with a 1N HCl solution to remove carbonates, rinsed with DI water, and dried. The dried contents on the GF/F were scraped off using a microspatula and placed into a 5x10mm aluminum capsule.

3.4.5 Stable Isotope Turnover Time of Weakfish

I used a growth-based turnover model (Buchheister and Latour 2010) to determine Weakfish specific turnover times:

$$t_{\alpha/100} = \frac{\ln(1 - \frac{\alpha}{100})}{kc}$$
(Eq. 1)

where $t_{\alpha/100}$ is the time that it takes for an α % turnover, *k* is the specific growth rate of the fish, and *c* is the turnover rate constant. If c = -1, growth is entirely responsible for turnover, whereas if c < -1, metabolism is contributing to turnover. This model estimates isotopic turnover based on growth and is measured in units of time (days).

Using the juvenile Weakfish growth coefficients k from Lankford and Targett (1994), the turnover constant c from Herzka and Holt (2000) and Buchheister and Latour (2010) for muscle and liver, respectively, I calculated the turnover times for muscle and liver for 50% and 99% percent turnover (Table 3.1, Figure 3.2) with Eq. 1. These values were used in interpreting the MixSIAR results as well as raw isotopic data.

Table 3.1. Projected time (in days) for (a) 99% and (b) 50% turnover of δ^{13} C and δ^{15} N of muscle and liver tissues based on *Eq*. 1 and estimated values of *k* and *c* (Lankford and Targett 1994; Herzka and Holt 2000).

a. 99%	Turnover tin	ne (days)	b. 50%	Turnover tin	ne (days)
	Muscle	Liver		Muscle	Liver
δ ¹³ C	68	12	δ ¹³ C	10	2
$\delta^{15}N$	81	10	$\delta^{15}N$	11	2



Figure 3.2. Projected time to (a) 99% and (b) 50% turnover of δ^{15} N for juvenile Weakfish (*Cynoscion regalis*) muscle (dotted line) and liver (continuous line) based on Eq. 1 and estimated literature values of *k* and *c*.

3.4.6 Trophic Enrichment Factor for MixSIAR

MixSIAR allows the user to set specific TEFs for each source and isotope. TEFs of ~1‰ for δ^{13} C and ~3.4‰ for δ^{15} N are widely used as the standard in aquatic systems (Minagawa and Wada 1984). In this study, I employed the standard values from Minagawa and Wada (1984) for muscle tissue with a standard deviation of 0.2 for both δ^{13} C and δ^{15} N. For liver tissue, I used TEFs with values closer to those found for summer flounder in Buchheister and Latour (2010). The liver TEFs used in MixSIAR were 2.8 for δ^{13} C and 2.2 for δ^{15} N both with standard deviation of 0.4.

3.4.7 Lipid Correction Factors

The δ^{13} C of animal tissues that are known to be enriched with lipids, such as Weakfish liver and some individual invertebrates, were corrected using a post-hoc correction factor for all δ^{13} C values from tissues with a C:N ratio greater than 3.5 as proposed by Post et al. (2007). The correction factor I applied to tissues with a C:N >3.5 was calculated as:

$$\delta^{13}$$
C corrected = δ^{13} C untreated - 3.32 + 0.99 × C:N (Eq. 2)

Post et al. (2007) also recommended that plants with >40% carbon be $\delta^{13}C$ corrected. I used the correction factor for plants with >40% carbon:

$$\delta^{13}C = -5.83 + 0.14 \times \text{\% carbon}$$
 (Eq. 3)

3.5 Statistical Analysis

3.5.1 Stomach Content Analysis

SCA results were analyzed using the %F (%frequency or presence/absence) calculated as:

$$F_i = \frac{N_i}{N} \times 100 \tag{Eq. 4}$$

where N_i is the number of stomachs containing prey item *i*, and *N* is the total number of stomachs analyzed. Frequency of occurrence of each size class was determined using Eq. 4 and was organized by season, bay location, and state. No classical statistical analyses were conducted on the SCA data. Only groups with a minimum sample size of 5 were reported. The %F results of each state/season/location combination for each size class were used to create informative priors to be used in MixSIAR.

3.5.2 Stable Isotope Analysis

All statistical analysis for SIA was performed in the R statistical environment (version 3.5; R Development Core Team 2018). The MASS package (Venables and Ripley 2002) was used for generalized linear modeling (GLM), and the MixSIAR package (Stock and Semmens 2016) was used for the Bayesian mixing models for diet partitioning of Weakfish and mysid shrimp. GLMs were compared to find the model with the most parsimonious fit for each size class, tissue (muscle and liver) and isotope (δ^{13} C, δ^{15} N, and δ^{34} S) combination using the AICcmodavg package (Mazerolle 2017).

3.5.2.1 Generalized Linear Modelling

Data normality was tested using the Shapiro-Wilks test. After several normality tests and data transformations failed to show that the data were normally distributed, I decided to use a GLM approach rather than using traditional non-parametric statistical tests. I used GLMs to

elucidate the drivers that influence δ^{13} C, δ^{15} N, and δ^{34} S values of each size class Weakfish caught in 2017 and 2018. Models were fit with a Gaussian distribution using the identity link function. A suite of models (Table 4.7) was created for each response variable of δ^{13} C and δ^{15} N (and δ^{34} S when available) using the factors of state, location (either nested or not nested within state), station (nested within location), season, month (nested within season), length, and weight. Candidate models were compared using corrected Akaike's Information Criterion (AICc) to determine which had the most parsimonious fit to the data (Burnham and Anderson 2004). The model with the lowest AICc or the lowest Δ_{AIC} was considered the best model from which ecological inferences were made. Models with a dispersion parameter calculated to be greater than 1.05 were considered to be overdispersed and were not included in the results.

3.5.2.2 MixSIAR

The fundamental mixing equation for all SIMMs is:

$$Y_{ij} = \sum_{k} \rho_k \mu_{jk}^s \tag{Eq. 5}$$

where Y_i is the mixture tracer value for each *j* tracer, μ_{jk}^s is the *k* source tracer mean, and ρ_k is the proportional contribution to the mixture (Stock et al. 2018). Here, tracer refers to a particular isotope (either δ^{13} C or δ^{15} N). The assumptions of this basic equation are 1) all source contributions to the mixture are known and quantified, 2) tracers are conserved through the mixing process, 3) source mixture and tracer values are fixed, 4) the ρ_k terms sum to unity (add up to 1), and 5) source tracer values differ (Phillips 2001; Stock et al. 2018). It should be noted that equations 5 and 6 only describe the fundamental equations of SIMMs. For a full description of the MixSIAR model equations including incorporation of priors, see Parnell et al. (2013). As in the traditional mixing model assumptions listed above, MixSIAR assumes that the *k* source means for the *j* tracers, μ_{jk}^{s} , are fixed and invariant. In this study, I used the "multiplicative error" term described in Stock et al. (2018):

$$Y_{ij} \sim N\left(\sum_{k} \rho_k \mu_{jk}^s, \sum_{k} \rho_k^2 \omega_{jk}^{s2} \times \varepsilon_j\right), \qquad (Eq.6)$$

where ω_{jk}^{s2} is the weighted combination of source variances and ε_j is the multiplicative error term for each tracer.

I used informative priors based off stomach content data of Weakfish. For example, if stomach content data of 20 Weakfish resulted in 30% frequency of mysid shrimp, 10% frequency of isopods, and 25% frequency of gammarids, an informative prior for a 3-source mixing model would be $\alpha = (30, 10, 25)$, where α_k corresponds to the *k* source sample size from the stomach contents. However, to avoid bias from "over-informativeness" from the sample size of the prior (n = 65 in this case), the α_k were rescaled such that:

$$\alpha_k = \frac{kn_k}{\sum n_k},\tag{Eq.7}$$

where $\sum \alpha_k = 3$, which is the number of sources in the example. The means are the same in both cases; however, the rescaled versus non-rescaled priors differ in how informative they are (Stock et al. 2018).

To determine the diet of each size class Weakfish from each state/season/location, a total of 45 models were created using SCA results as priors in each model. MixSIAR MCMC specifications were set to a standard chain length of 100,000 to allow for model convergence. Gelman-Rubin and Geweke diagnostics were checked for model convergence upon completion of each model run.

3.5.2.2.1 Weakfish

Mixing models were run for each size class juvenile Weakfish caught in 2018. Mixing models were not run for 2017 Weakfish due to the absence of prey samples caught in 2017. Using source data, isospace plots were created to ensure that mixture data fell within the convex hull (Phillips et al. 2014). If a source did not appear to contribute to the mixing system, it was removed as a source to simplify the model. Weakfish prey sources were either used as individual sources or pooled based on their importance in the diet. Mysid shrimp (*N. americana*), sand shrimp (*C. septimspinosa*), and polychaete worms were used as individual sources whereas Bay Anchovy (*A. mitchilli*) and larval clupeids were combined into one source labeled "Bay Anchovy", *E. triloba* and *S. laticauda* were combined into one source labeled "isopod", and *Ampelisca* sp., *C. tubelaris*, and UID gammarid amphipods were combined into another source labeled "gammarid". Alpha (α) priors for source values were included in each model using the %F SCA data from 2018 Weakfish (Tables 4.4 – 4.6).

3.5.2.2.2 Mysid Shrimp

Due to the importance of mysid shrimp in the diet of juvenile Weakfish, a mixing model was run using the end members of the Delaware Bay including *Spartina*, *Phragmites*, SPOM, and BMA. Except for BMA, all end-members were sampled in 2018 from each side of the bay from May to September. Source values of BMA were taken from Currin et al. (2003) that were collected from Delaware Bay marsh creeks. TEFs of 2 (SD 0.5) for δ^{13} C and 4.4 (SD 1) for δ^{15} N were used, as Johannsson et al. (2001) noted that 2.2 was the best value to use between a different species of mysid shrimp (*M. relicta*) and their prey. Since mysid shrimp generally do not feed directly on primary producers, the TEF between Delaware Bay end-members and mysid

shrimp was set to 3.5, approximately 1.5 times that between mysid shrimp and their prey. The same theory was applied to the TEF for δ^{13} C.

CHAPTER 4: RESULTS

4.1 Stomach Content Analysis

I analyzed a total of 1,028 juvenile Weakfish (with non-empty stomachs; SL = 21-137mm) from 36 stations for stomach contents during the 2-year study period. A notable shift in diet from crustacean to fish prey was observed with increased predator length. Although an increase in the frequency of fish prey was observed with an increase in size class, the frequency of crustacean prey remained high in the larger size classes in 2017 and 2018. Mysid shrimp and gammarid amphipods were the dominant prey items (found in up to 100% of the small and medium size classes), whereas individuals in the large size class had more diverse diets with higher %F observed for UID fish (likely Bay Anchovy), sand shrimp, and other arthropod species (e.g. small blue crabs, larval horseshoe crabs). The %F of all prey items remained relatively stable over both years, but mysid shrimp occurred in 79% for pooled size classes in 2017 (n = 348) and 91% in 2018 (n = 680; Tables 4.1 - 4.6). Overall, the contributions of individual prey species varied by season and state; however, a consistent decrease in gammarid amphipod consumption (mainly Ampelisca sp.) was noted from the summer to fall in 2017 and 2018 for both Delaware and New Jersey. This decrease in amphipod consumption was consistently related to an increase in mysid consumption over the same time period. This inverse seasonal relationship between mysids and gammarids is hereby mentioned as the "seasonal mysid-gammarid relationship". Additionally, all Weakfish caught from the middle bay consistently showed higher frequencies of prey items in their stomachs compared to Weakfish caught from the lower and upper bays. In the sections below, the SCA results of the three Weakfish size classes are separated into individual sections, and the results for each bay location are described in order from the lower to the upper bay. Due to the limited number of stations for NJ-upper, not enough Weakfish were caught to be analyzed from this stratum.

4.1.1 2017

4.1.1.1 Small Sized Weakfish (<60mm SL)

Small Weakfish were only caught from the middle and upper bays in 2017. In the middle bay, Weakfish were caught during both seasons from NJ but only during the summer from DE. In DE, mysids appeared in 73% of stomachs, gammarids appeared in 100% of stomachs, and isopods (mainly *E. triloba*) appeared in 45% of stomachs. In NJ, the seasonal mysid-gammarid relationship was observed; from summer to fall, mysids increased from 32% to 71% and gammarids decreased from 68% to 0%. Polychaetes also decreased from 47% to 0% in NJ from summer to fall.

In the upper bay, fish were caught during both seasons from DE. A weak seasonal mysidgammarid relationship was observed; from summer to fall, mysids increased from 93% to 100% and gammarids decreased from 15% to 0%. Sand shrimp increased over the same time period from 0% to 20%.

4.1.1.2 Medium Sized Weakfish (60-100mm SL)

Medium Weakfish from the lower bay were caught during both seasons in DE but only during the fall in NJ. In DE, the seasonal mysid-gammarid relationship was observed; from summer to fall, mysids increased from 77% to 83% and gammarids decreased from 85% to 58%. In NJ, mysids were observed in 100% of stomachs and other arthropod spp. (mainly crab megalopa) were observed in 60% of stomachs.

In the middle bay, medium Weakfish were caught during both seasons from both states. In DE, the seasonal mysid-gammarid relationship was observed; from summer to fall, mysids increased from 58% to 91% and gammarids decreased from 89% to 24%. Other seasonal relationships were also observed: from summer to fall, polychaetes and isopods decreased from 58% to 0% and 47% to 3%, respectively, and sand shrimp increased from 5% to 33%. In NJ, no seasonal mysid-gammarid relationship was observed, however, the same seasonal relationships observed in DE were also found in NJ: from summer to fall, polychaetes and isopods decreased from 57% to 0% and 43% to 0%, respectively.

In the upper bay, fish were caught during both seasons from DE. A weak seasonal mysidgammarid relationship was observed; from summer to fall, mysids increased from 78% to 81% and gammarids decreased from 26% to 19%. This was also observed in the upper bay with small Weakfish. Other prey items were more prevalent during the summer; however, their frequency of occurrence was much lower when compared to the lower and middle bays. Nevertheless, sand shrimp were observed to increase slightly from summer to fall (19% to 29%).

4.1.1.3 Large Sized Weakfish (100>137mm SL)

Large Weakfish were only caught from the lower and middle bays in 2017. In the lower bay, large Weakfish were caught during both seasons from DE, but none were caught in NJ. Large Weakfish exhibited a deviation from the other two size classes in terms of the summer to fall relationships of prey items. While there was a decrease in the frequency of gammarids from summer to fall, a decrease in the occurrence of mysids was also observed from 100% to 83%. However, crangon and UID fish increased from summer to fall from 17% to 28% and 0% to 28%, respectively.

In the middle bay, Weakfish were caught during both seasons in DE but only during the fall in NJ. In DE, the seasonal mysid-gammarid relationship was strong; from summer to fall, mysids occurred from 22% to 94% and gammarids occurred from 67% to 11%. However, larger

prey items of crangon and UID fish were also present during the summer at frequencies >22%.

During the fall in NJ, the occurrence of mysids was 100%, with a notable occurrence of 64% for

crangon and 36% for arthropod spp. (mainly crab megalopa).

Table 4.1. 2017 small Weakfish %F SCA table showing frequencies of occurrence for groupedprey items.

2017 Small Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Middle											
DE											
Summer	11	73%	100%	64%	0%	45%	0%	9%	9%	0%	0%
NJ											
Summer	19	32%	68%	47%	0%	21%	0%	0%	21%	0%	37%
Fall	24	71%	0%	0%	0%	0%	0%	0%	42%	0%	38%
Upper DE											
Summer	46	93%	15%	17%	0%	11%	0%	17%	20%	0%	4%
Fall	10	100%	0%	10%	20%	0%	0%	20%	0%	0%	0%
Total	110										

Table 4.2. 2017 medium Weakfish %F SCA table showing frequencies of occurrence for grouped prey items.

2017 Medium Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Lower											
DE											
Summer	13	77%	85%	8%	15%	8%	0%	15%	8%	0%	0%
Fall	12	83%	58%	0%	33%	0%	0%	8%	17%	0%	0%
NJ											
Fall	5	100%	0%	0%	0%	0%	0%	0%	60%	0%	0%
Middle											
DE											
Summer	19	58%	89%	58%	5%	47%	0%	26%	5%	0%	5%
Fall	33	91%	24%	0%	33%	3%	0%	21%	12%	0%	21%
NJ											
Summer	7	71%	14%	57%	0%	43%	0%	0%	14%	0%	57%
Fall	5	100%	20%	0%	20%	0%	0%	20%	60%	0%	20%
Upper											
DE											
Summer	27	78%	26%	19%	19%	7%	0%	11%	19%	0%	4%
Fall	21	81%	19%	5%	29%	14%	0%	0%	10%	0%	24%
Total	142										

2017 Large Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Lower											
DE											
Summer	12	100%	83%	8%	17%	8%	0%	0%	25%	8%	0%
Fall	29	83%	66%	3%	28%	21%	0%	28%	21%	0%	24%
Middle											
DE											
Summer	9	22%	67%	0%	33%	22%	0%	22%	33%	0%	56%
Fall	18	94%	11%	0%	33%	11%	0%	6%	11%	0%	22%
NJ											
Fall	11	100%	18%	9%	64%	27%	0%	18%	36%	0%	55%
Total	79										

Table 4.3. 2017 large Weakfish %F SCA table showing frequencies of occurrence for grouped prey items.

4.1.2 2018

4.1.2.1 Small Sized Weakfish (<60mm SL)

In the lower bay, small Weakfish were caught during both seasons in DE but only during the summer in NJ. In DE, a weak seasonal mysid-gammarid relationship was observed; from summer to fall, mysids increased from 87% to 100% and gammarids decreased from 67% to 54%. During the summer in NJ, the occurrence of mysids was 70% but gammarids only occurred in 22% of stomachs.

In the middle bay, Weakfish were caught during each season for both states. In both states, the seasonal mysid-gammarid relationship was observed, however, the frequency of gammarids during the summer was under 36% for both states. From summer to fall in DE, mysids increased from 65% to 100%, polychaetes decreased from 47% to 17%, and isopods (mainly *E. triloba*) decreased from 29% to 0%. From summer to fall in NJ, mysid shrimp consumption remained relatively stable from 88% to 82%. Additionally, unlike the small Weakfish from DE, small Weakfish from NJ ingested more polychaetes from summer to fall from 32% to 45%.

In the upper bay, small Weakfish were caught during both seasons from DE. Here, the frequency of mysid shrimp from summer to fall remained stable from 96% to 100%. Unlike the seasonal dynamics in other bay locations, gammarids, polychaetes, crangon, and UID fish all increased in occurrence from summer to fall.

4.1.2.2 Medium Sized Weakfish (60-100mm SL)

In the lower bay, Weakfish were caught during both seasons from both states. Fish from both states in the lower bay exhibited the seasonal mysid-gammarid relationship. From summer to fall in DE, mysids increased from 95% to 100% and gammarids decreased from 82% to 44%. Additionally, crangon increased over the same time period in DE from 3% to 26%. From summer to fall in NJ, mysids increased from 90% to 100% and gammarids decreased from 43% to 33%.

In the middle bay, Weakfish were caught during both seasons and from both states. A strong seasonal mysid-gammarid relationship was observed in both states, with notable %F rates of polychaetes in NJ (>60% during both seasons). From summer to fall in DE, mysids increased from 73% to 95% and gammarids decreased from 77% to 5%. From summer to fall in NJ, mysids increased from 78% to 94% and gammarids decreased from 80% to 19%. Sand shrimp also increased from summer to fall in NJ from 8% to 38%. Additionally, during the summer in NJ, UID fish/Bay Anchovy was observed at 38%.

In the upper bay, fish were caught during both seasons from DE. Here, a weak seasonal mysid-gammarid relationship was observed; from summer to fall, mysids increased from 90% to 98% and gammarids decreased from 26% to 19%.

4.1.2.3 Large Size Weakfish (100-137mm SL)

In the lower bay, fish were caught during each season from both states. A weak seasonal mysid-gammarid relationship was observed in both states with mysids occurring over 90% during both seasons. Only gammarid consumption in DE declined significantly from summer to fall from 79% to 11%. Crangon was observed to increase from summer to fall in both states, conforming to the general trend observed in other bay locations. UID fish was observed to occur in greater frequencies from large Weakfish caught in DE compared to those caught in NJ.

In the middle bay, fish were caught during each season from both states. The seasonal mysid-gammarid relationship was only observed in DE; from summer to fall, mysids increased from 60% to 100% and gammarids decreased from 60% to 6%. In NJ, gammarids also decreased over time from 85% to 21%, however, mysids remained relatively stable from 100% to 93%. Sand shrimp also exhibited a seasonal relationship in both states from 0% to 40% from summer to fall. UID fish/Bay Anchovy were observed to occur in a notable 85% of fish from NJ during the summer.

In the upper bay, fish were only caught in DE during the summer. Here, mysids occurred at a lower frequency than gammarids (40% vs. 60%), which is atypical from upper bay trends from other size classes. Sand shrimp was observed in 40% of stomachs, which breaks with the usual trend from other size classes that typically exhibit low frequencies of sand shrimp during the summer.

2018 Small Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Lower											
DE											
Summer	15	87%	67%	7%	0%	0%	0%	7%	27%	0%	0%
Fall	13	100%	54%	8%	0%	8%	0%	0%	15%	0%	0%
NJ											
Summer	27	70%	19%	0%	0%	7%	0%	11%	11%	0%	22%
Middle											
DE											
Summer	17	65%	35%	47%	0%	29%	0%	6%	0%	0%	6%
Fall	6	100%	0%	17%	0%	0%	0%	0%	0%	0%	0%
NJ											
Summer	41	88%	22%	32%	0%	10%	0%	2%	7%	0%	2%
Fall	11	82%	0%	45%	0%	18%	0%	18%	0%	0%	0%
Upper											
DE											
Summer	50	96%	16%	22%	0%	18%	0%	2%	4%	0%	2%
Fall	31	100%	26%	26%	13%	3%	0%	10%	3%	0%	0%
Total	211										

Table 4.4. 2018 small Weakfish %F SCA table showing frequencies of occurrence for grouped prey items.

Table 4.5. 2018 medium Weakfish %F SCA table showing frequencies of occurrence for grouped prey items.

2018 Medium Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Lower											
DE											
Summer	38	95%	82%	11%	3%	3%	0%	18%	3%	0%	0%
Fall	34	100%	44%	3%	26%	0%	0%	6%	15%	0%	0%
NJ											
Summer	30	90%	43%	3%	13%	7%	0%	13%	10%	0%	0%
Fall	21	100%	33%	0%	10%	5%	0%	0%	29%	5%	0%
Middle											
DE											
Summer	30	73%	77%	33%	20%	20%	0%	20%	10%	0%	0%
Fall	37	95%	5%	8%	16%	8%	0%	30%	3%	0%	0%
NJ											
Summer	40	78%	80%	63%	8%	18%	3%	35%	13%	0%	0%
Fall	16	94%	19%	69%	38%	19%	6%	13%	0%	0%	0%
Upper											
DE											
Summer	31	90%	26%	13%	13%	16%	0%	6%	0%	0%	0%
Fall	47	98%	19%	26%	21%	9%	2%	21%	0%	0%	2%
Total	324										

2018 Large Weakfish	n	Mysid spp.	Gammarid spp.	Polychaete spp.	Crangon	Isopod spp.	Bay anchovy	UID Fish	Other Arthropod spp.	Bivalve spp.	UID
Lower											
DE											
Summer	19	95%	79%	5%	11%	5%	0%	16%	11%	0%	5%
Fall	27	100%	11%	4%	22%	4%	4%	26%	4%	0%	0%
NJ											
Summer	16	94%	38%	0%	0%	6%	6%	6%	38%	0%	6%
Fall	23	100%	35%	0%	17%	13%	0%	13%	35%	0%	0%
Middle											
DE											
Summer	5	60%	60%	0%	0%	60%	0%	0%	0%	0%	20%
Fall	16	100%	6%	13%	50%	13%	0%	38%	13%	0%	6%
NJ											
Summer	13	100%	85%	77%	0%	8%	23%	62%	23%	0%	0%
Fall	14	93%	21%	79%	43%	29%	0%	0%	7%	0%	0%
Upper											
DE											
Summer	5	40%	60%	0%	40%	0%	0%	20%	0%	0%	20%
Total	138										

Table 4.6. 2018 large Weakfish %F SCA table showing frequencies of occurrence for grouped prey items.

4.2 Stable Isotope Analysis

4.2.1 Generalized Linear Models

For each size class Weakfish and isotope, I ran 4 model configurations (#1 - 4) for Weakfish muscle tissue and 2 to 3 model configurations (#2 - 4) were run for liver tissue of medium and large size class Weakfish (refer to Table 4.7 for model configuration list and symbol representations). In the following section, the results of the most parsimonious fit model for each size class, tissue, and isotope are presented. Sampling stations with the prefix "STA" refer to stations on the DE side of the bay and stations with the prefix "DBT" refer to stations on the NJ side of the bay. The model term "location" refers to the upper, middle, and lower bay locations. In total, eight models that had the most parsimonious fit to the data according to the AICc were overdispersed with dispersion parameters calculated to be greater than 1.05. For 2017 fish, medium Weakfish muscle δ^{13} C and δ^{34} S, and large Weakfish muscle δ^{13} C and δ^{15} N models were overdispersed. For 2018 fish, small Weakfish muscle δ^{13} C, medium Weakfish muscle δ^{13} C and δ^{15} N, and medium Weakfish liver δ^{15} N models were overdispersed. Due to the unreliability of these model results, the estimated model parameters are not listed below. Additionally, model parameters for 2017 large Weakfish liver δ^{34} S are not reported due to insufficient sample sizes by season.

Table 4.7. List of candidate models run for δ^{13} C, δ^{15} N, and δ^{34} S of each size class Weakfish. Variables to the right of a slash (/) are nested within the variable to the left, and stars (*) indicate interaction terms.

Model #	Model Configuration
1	state/location/station + length + season/month
2	state/location/station + weight + season/month
3	state/location/station*season/month
4	state*season/month + location/station

4.2.1.1 2017 Weakfish Muscle

4.2.1.1.1 Small Sized Weakfish $\delta^{13}C$ and $\delta^{15}N$

Model 4 had the lowest AICc for δ^{13} C of small Weakfish muscle, with the model terms of location/station having significant effects on delta values. Four location/station terms from NJ-middle had significant positive estimates (Table 4.8). These positive estimates show that the NJ-middle stations had more enriched δ^{13} C values compared to fish caught in NJ-lower stations.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-16.7	0.73	-22.99	< 0.001
Middle/DBT29A	2.07	0.84	2.45	< 0.05
Middle/DBT39	2.51	1.04	2.41	< 0.05
Middle/DBT30	2.05	0.93	2.21	< 0.05
Middle/DBT31	2.19	1	2.2	< 0.05

Table 4.8. Significant GLM estimates for 2017 small sized Weakfish muscle δ^{13} C values from Model 4.

Model 4 had the lowest AICc for δ^{15} N of small Weakfish, with model terms of season, location, and location/station all having significant effects on delta values (Table 4.9). The summer variable had the most significant estimate on δ^{15} N values, demonstrating that Weakfish caught in the summer were more enriched in δ^{15} N than Weakfish caught in the fall (Figure 4.1). There were two significant estimates by location: 1) upper, and 2) middle, both signaling that the middle and upper bay Weakfish exhibited more depleted δ^{15} N values than Weakfish caught from the lower bay. Lastly, three location/station terms were significant: 1) lower/DBT54, 2) middle/STA16, and 3) upper/STA10. These significant location/station terms revealed that individual stations displayed unique δ^{15} N values within their respective bay locations despite bay location estimates exhibiting opposite trends.

Coefficient	Estimate	Std. Error	t-value	P-value
L	14.6	0.40	22.66	-0.001
Intercept	14.6	0.49	22.66	<0.001
Summer	4.93	0.64	7.7	< 0.01
Upper	-4.4	0.88	-5.03	< 0.01
Middle	-4.49	1.06	-4.22	< 0.01
Lower/DBT54	-4.55	1.23	-3.6	< 0.01
Middle/STA16	3.99	1.17	3.41	< 0.01
Upper/STA10	1.61	0.54	3.03	< 0.01

Table 4.9. Significant GLM estimates for 2017 small sized Weakfish muscle δ^{15} N values from Model 4.



Figure 4.1. Boxplot of 2017 small sized Weakfish muscle comparing seasonal and locational δ^{15} N for pooled states. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR (Interquartile Range).

4.2.1.1.2 Large Sized Weakfish $\delta^{34}S$

For δ^{34} S of the large Weakfish muscle, data was not collected for more than one season, leaving only the state/location/station and state/location/station + length models available to run as a GLM. The model without length as a factor had the lowest AICc. The only term in the model that had a significant estimate was DE/lower/STA49 (Table 4.10), demonstrating that δ^{34} S values of large Weakfish muscle from STA49 were more enriched than Weakfish caught from other stations.

Table 4.10. Significant GLM estimate for 2017 large sized Weakfish muscle δ^{34} S values from the state/location/station model.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	17.45	0.48	36.21	< 0.001
DE/Lower/STA49	1.65	0.68	2.42	< 0.05

4.2.1.2 2017 Weakfish Liver

4.2.1.2.1 Medium Sized Weakfish $\delta^{13}C$ and $\delta^{15}N$

Model 2 had the lowest AICc for δ^{13} C of medium Weakfish liver, with one state/location term having a significant estimate: DE/upper (Table 4.11). The significant negative estimate of DE/upper conforms to the general association of more depleted δ^{13} C values in the upper bay compared to other bay locations in both states (Figure 4.2).

Table 4.11. Significant GLM estimates for 2017 medium sized Weakfish liver δ^{13} C values from Model 2.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-16.98	2.05	-8.26	< 0.001
DE/Upper	-5.48	1.7	-3.21	< 0.01

Model 2 had the lowest AICc for δ^{15} N, with state/location, season/month, and state/location/station having significant effects on delta values (Table 4.12). The term with the most significant estimate was DE/upper/STA07, indicating that δ^{15} N values of medium Weakfish liver were more enriched at STA07 than Weakfish caught from other stations in the upper bay. Two state/location terms were also significant: 1) NJ/middle, and 2) DE/upper (Figure 4.3). Finally, two season/month terms were highly significant: 1) summer/August, and 2) summer/July. Here, the differing estimates between individual months in the same seasons (July vs. August) demonstrate the monthly variability of δ^{15} N values of Weakfish liver even during the same season.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	12.89	1.12	11.53	< 0.001
DE/Upper/STA07	2.2	0.36	6.08	< 0.01
NJ/Middle	1.94	0.4	4.83	< 0.01
DE/Upper	-3.15	0.93	3.4	< 0.01
Summer/August	4.03	0.82	4.92	< 0.01
Summer/July	-3.11	0.66	-4.7	< 0.01

Table 4.12. Significant GLM estimates for 2017 medium sized Weakfish liver δ^{15} N values from Model 2.



Figure 4.2. Boxplot of 2017 medium sized Weakfish liver comparing state and locational δ^{13} C for pooled seasons. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.



Figure 4.3. Boxplot of 2017 medium sized Weakfish liver comparing state and locational $\delta^{15}N$ for pooled seasons. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.

4.2.1.2.2 Large Sized Weakfish $\delta^{13}C$ and $\delta^{15}N$

Model 2 had the lowest AICc for δ^{13} C of large Weakfish liver, with model terms of state, weight, and state/location all having significant estimates (Table 4.13). The terms of NJ and weight both showed significant positive estimates: 1) NJ, and 2) weight. These estimates suggest that Weakfish caught in NJ displayed more enriched δ^{13} C values compared to Weakfish caught in DE and that δ^{13} C values become slightly more enriched with increased weight. Two state/location terms were significant: 1) DE/upper, and 2) NJ/middle, showing that DE-upper showed more depleted δ^{13} C values than other locations in DE, and NJ-middle displayed more depleted δ^{13} C values than NJ-lower (Figure 4.4).

Table 4.13. Significant GLM estimates for 2017 large sized Weakfish liver δ^{13} C values from Model 2.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-19.68	1.2	-16.46	< 0.001
New Jersey	3.69	0.87	4.23	< 0.01
Weight	0.1	0.03	3.03	< 0.01
DE/Upper	-2.41	1.12	-2.15	< 0.05
NJ/Middle	-2.71	1.31	-2.07	< 0.05

Model 2 had the lowest AICc for δ^{15} N values, with the model term of season having significant estimates: 1) spring, and 2) summer (Table 4.14). These estimates suggest that Weakfish caught in the spring appeared to display more enriched δ^{15} N values while Weakfish caught in the summer displayed more depleted δ^{15} N values (Figure 4.5).

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Table 4.14. Significant GLM estimates for 2017 large sized Weakfish liver δ^{15} N values from Model 2.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	14.84	0.58	25.66	< 0.001
Spring	1.11	0.35	3.2	< 0.01
Summer	-0.7	0.32	-2.2	< 0.05



Figure 4.4. Boxplot of 2017 large sized Weakfish liver comparing state and locational δ^{13} C for pooled seasons. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.



Figure 4.5. Boxplot of 2017 large sized Weakfish liver comparing seasonal δ^{15} N for pooled states and locations. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.

4.2.1.3 2018 Weakfish Muscle

4.2.1.3.1 Small Sized Weakfish $\delta^{15}N$

Model 4 had the lowest AICc for δ^{15} N values, with state, location/station, and state*season/month terms all having significant estimates (Table 4.15). NJ had a significant negative estimate, indicating that small Weakfish from the state of NJ had depleted δ^{15} N values relative to DE. Eight location/station terms showed significant positive and negative estimates. The various significant estimates from multiple nested station terms suggest that Weakfish caught at individual stations had unique delta values relative to other stations within and between their respective bay locations and states. Finally, the two state*season/month terms that had
significant estimates display the δ^{15} N depletion from summer to fall in small Weakfish (Figure

4.6).

Table 4.15. Significant	GLM estimates f	for 2018 small	sized Weakfi	sh muscle δ ¹⁵ N	values from
Model 4.					

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	12.89	0.91	14.08	< 0.001
New Jersey	-3.46	0.87	-3.97	< 0.01
Middle/DBT15	4.7	1.26	3.73	< 0.01
Middle/DBT30	4.7	1.42	3.31	< 0.01
Middle/DBT47A	4.41	1.35	3.26	< 0.01
Lower/DBT54	3.65	1.38	2.65	< 0.05
Lower/DBT60	3.4	1.31	2.6	< 0.05
Upper/STA13	-2.87	0.75	-3.82	< 0.01
Middle/STA17	2.1	0.93	2.26	< 0.05
Middle/STA25	3.08	1.08	2.85	< 0.01
DE*Summer/July	2.34	0.71	3.31	< 0.01
DE*Fall/October	-2.7	0.64	-4.25	< 0.01



Figure 4.6. Boxplot of 2018 small sized Weakfish muscle comparing seasonal δ^{15} N by month for pooled states and locations. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.

4.2.1.3.2 Large Sized Weakfish $\delta^{13}C$ and $\delta^{15}N$

Model 2 had the lowest AICc for large Weakfish δ^{13} C, with season/month and state/location/station model terms both having significant effects (Table 4.16). One season/month parameter was found to be significant: fall/October, suggesting that large Weakfish caught in October displayed more depleted δ^{13} C values than Weakfish caught in September and the summer season. Two state/location/station terms were also significant: 1) DE/upper/STA08, and 2) DE/middle/STA20. These significant nested station parameters show the depletion of δ^{13} C along the salinity gradient from salt to freshwater.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-15.64	0.87	-17.91	< 0.001
Fall/October	-1.35	0.5	-2.71	< 0.01
DE/Upper/STA08	-5.19	1.13	-4.57	< 0.01
DE/Middle/STA20	-2.57	0.91	-2.82	< 0.01

Table 4.16. Significant GLM estimates for 2018 large sized Weakfish muscle δ^{13} C values from Model 2.

Model 2 had the lowest AICc for large Weakfish δ^{15} N values, with one parameter from

the state/location/station term having significance: DE/upper/STA08 (Table 4.17). This significant estimate shows that the upper bay station in DE of STA08 had significantly more enriched δ^{15} N values than fish caught from other stations in DE-upper.

Table 4.17. Significant GLM estimates for 2018 large sized Weakfish muscle δ^{15} N values from Model 2.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	14.28	0.48	29.54	< 0.001
DE/Upper/STA08	2.96	0.63	4.71	< 0.01

4.2.1.4 2018 Weakfish Liver

4.2.1.4.1 Medium Sized Weakfish $\delta^{13}C$

Model 2 had the lowest AICc for δ^{13} C values of medium Weakfish liver, with state/location and state/location/station model terms both having significant estimates (Table 4.18). DE/upper had a significant negative estimate, conforming to the commonly calculated negative estimates from upper bay Weakfish as evidence for depleted δ^{13} C values along the salinity gradient (Figure 4.7). Two state/location/station parameters were also shown to be significant: 1) DE/middle/STA20, and 2) DE/middle/STA17. The varying estimates for these two separate nested stations show the within-location variability of $\delta^{13}C$ values of medium

Weakfish liver.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-18.52	1.07	-17.36	< 0.001
DE/Upper	-3.57	0.96	-3.71	< 0.01
DE/Middle/STA20	-3.24	0.97	-3.34	< 0.01
DE/Middle/STA17	-1.77	0.85	-2.07	< 0.05

Table 4.18. Significant GLM estimates for 2018 medium sized Weakfish liver δ^{13} C values from Model 2.



Figure 4.7. Boxplot of 2018 medium sized Weakfish liver δ^{13} C of fish caught in DE comparing locations for pooled seasons. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.

Model 4 had the lowest AICc for δ^{13} C values of large Weakfish liver, with

4.2.1.4.2 Large Sized Weakfish $\delta^{13}C$ and $\delta^{15}N$

location/station and state*season/month both having significant estimates (Table 4.19). Two parameters for the location/state model term were significant: 1) upper/STA08, and 2) middle/STA20. These estimates indicate that the upper bay exhibits slightly more depleted δ^{13} C values than the fish from the middle bay. The NJ*fall/October also a had a significant negative estimate, suggesting that fish caught from NJ in October displayed more depleted δ^{13} C values than fish caught in NJ during September and during the summer (Figure 4.8).

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	-15.58	0.88	-17.74	< 0.001
Upper/STA08	-5.3	1.46	-3.63	< 0.01
Middle/STA20	-4.12	1.23	-3.36	< 0.01
NJ*Fall/October	-1.46	0.72	-2.02	< 0.05

Table 4.19. Significant GLM estimates for 2018 large sized Weakfish liver $\delta^{13}C$ values from Model 4.

Model 3 had the lowest AICc for δ^{15} N values of large Weakfish liver, with state, season, state/location/station, and state/location*season all having significant estimates (Table 4.20). NJ and summer as individual terms both had significant negative estimates: 1) NJ, and 2) summer, indicating that Weakfish caught in NJ exhibited slightly more depleted δ^{15} N values than their DE counterparts, and fish caught in the summer were more depleted in δ^{15} N than those caught in the fall (Figures 4.9 & 4.10). However, NJ/middle*summer had a significant positive estimate, suggesting that delta values of large Weakfish liver were more enriched during the summer in NJ-middle compared to fish caught during the summer in NJ-lower. Four state/location/station terms were significant: 1) DE/upper/STA08, 2) DE/middle/STA20, 3) NJ/middle/DBT39, and 4) NJ/middle/DBT47A. These significant estimates show that fish from the DE-upper station of STA08 displayed more enriched δ^{15} N values than Weakfish from the middle bay stations and demonstrate the within-location variability of δ^{15} N values of large Weakfish.

Coefficient	Estimate	Std. Error	t-value	P-value
Intercept	14.84	0.58	25.66	< 0.001
New Jersey	-1.6	0.49	-3.24	< 0.01
Summer	-1.3	0.57	-2.28	< 0.05
NJ/Middle*Summer	1	0.49	2.02	< 0.05
DE/Upper/STA08	3.48	0.73	4.78	< 0.01
DE/Middle/STA20	1.17	0.33	3.54	< 0.01
NJ/Middle/DBT39	-2.13	0.62	-3.46	< 0.01
NJ/Middle/DBT47A	1.65	0.49	-3.34	< 0.01

Table 4.20. Significant GLM estimates for 2018 large sized Weakfish liver δ^{15} N values from Model 3.



Figure 4.8. Boxplot of 2018 large sized Weakfish liver δ^{13} C of fish caught in NJ comparing months for pooled locations. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.



Figure 4.9. Boxplot of 2018 large sized Weakfish liver comparing seasonal δ^{15} N by state for pooled locations. Lines within boxes represent the median, diamonds represent the mean, and red stars represent outliers that are >1.5×IQR.



Figure 4.10. Interaction plot of mean δ^{15} N of 2018 large sized Weakfish liver by season and state for pooled locations.

4.2.2 MixSIAR

Summary statistics of MixSIAR results from the 46 individual models are reported below. Unless otherwise noted, the specific parameters for each size class were unchanged for the various state, season, and location combinations. Sources that were found to contribute >5% of the median diet proportion are reported along with their standard deviations (\pm). Sample sizes of sources that were used in the models were n = 207 for Bay Anchovy/larval clupeids, n = 23 for crangon, n = 11 for gammarid amphipods, n = 25 for isopods, n = 47 for mysid shrimp, and n = 19 for polychaete worms. Sources used were pooled over all study areas and time periods due to insufficient sample sizes of gammarids and polychaetes in certain bay location by season combinations. Any models that did not converge are noted, and any changes to model specifications to allow for convergence are explained.

4.2.2.1 2018 Weakfish Muscle

4.2.2.1.1 Small Sized Weakfish

Sources used for small Weakfish were gammarid amphipods, isopods, mysids,

polychaete worms, and Bay Anchovy. However, although Bay Anchovy/UID fish was found in the stomach of a minor percentage of small Weakfish, it was removed as a source after several models did not converge using this prey item as a source. Once Bay Anchovy was removed as a source, isospace plots were inspected, and subsequent model runs all converged. The TEFs of all sources were set as 1 (SD 0.2) for δ^{13} C and 3.4 (SD 0.2) for δ^{15} N. All small Weakfish mixing model runs used a MCMC chain length of 50,000, except for NJ/Fall/Middle, which used a chain length of 100,000 due to the failure of the model to converge at 50,000 iterations.

DE/Summer/Lower

Model estimations of median diet proportions were 37.1% (\pm 13.8%) for gammarids and 58.5% (\pm 15.4%) for mysids.

DE/Fall/Lower

Model estimations of median diet proportions were 56.3% ($\pm 12.3\%$) for gammarids and 37.3% ($\pm 11.4\%$) for mysids (Figure 4.11).



Figure 4.11. Posterior density plot of small sized Weakfish caught during the fall in DE-Lower. n = 9

DE/Summer/Middle

Model estimations of median diet proportions were 15.1% (\pm 13.5%) for gammarids, 53.5% (\pm 20.8%) for mysids, and 21.1% (\pm 19.5%) for polychaetes.

DE/Fall/Middle

Using the standard parameters and priors from DE/Fall/Middle fish, the first attempt of the model run did not converge. To give the model a better chance to converge, a model was run on the data using an uninformative prior. This adjustment allowed the model to converge with a chain length of 50,000. The resulting dietary proportions calculated by the model were 26.4% ($\pm 15.2\%$) for gammarids, 10.2% ($\pm 10.9\%$) for isopods, 31.3% (± 17.3) for mysids, and 25.2% ($\pm 20.4\%$) for polychaetes.

DE/Summer/Upper

Model estimations of median diet proportions were 81.2% (\pm 17.1%) for mysids and 6.7% (\pm 13.4%) for polychaetes.

DE/Fall/Upper

Model estimations of median diet proportions were 83.2% (\pm 19.6%) for gammarids and 12.7% (\pm 17.2%) for mysids.

NJ/Summer/Lower

Model estimations of median diet proportions were 67.4% (\pm 8.3%) for gammarids and 28.2% (\pm 8.2%) for mysids (Figure 4.12).



Figure 4.12. Posterior density plot of small sized Weakfish caught during the summer in the NJ-Lower. n = 12

NJ/Summer/Middle

Model estimations of median diet proportions were 56.6% (\pm 10.3%) for gammarids,

29.8% (± 10.4%) for mysids, and 9.2% (± 12.2%) for polychaetes.

NJ/Fall/Middle

Model estimations of median diet proportions were 33.3% (\pm 24.2%) for gammarids, 10.1% (\pm 20.5%) for isopods, 25.4% (\pm 12.8%) for mysids, and 19.4% (\pm 20.8%) for polychaetes.

Table 4.21. Comparison table of the available SCA (%F) and MixSIAR results (%) of small sized Weakfish from 2018. M-SIA represents stable isotope analysis of muscle tissue. Asterisks (*) represent models that converged only with uninformative priors.

		SUMMER			LOWER BAY			FALL		
DELAWARE	Musida	Commonida	Dolyahaataa	Isonoda		DELAWARE	Musida	Commonida	Dobuchaotos	Isonada
SCA (15)	1VIYSIUS	Gammarius	Polychaetes	Isopous		SCA(12)	100	Gammarius	Polychaetes	isopous
SCA (15)	8/	07	/	0		SCA (13)	100	54	8	8
M-SIA (5)	59	37	< 5	< 5		M-SIA (9)	37	56	< 5	< 5
NEW JERSEY										
SCA (27)	70	19	0	7						
M-SIA (12)	28	67	< 5	< 5						
					MIDDLE BAY					
DELAWARE						DELAWARE				
SCA (17)	65	35	47	29		SCA (6)	100	0	17	29
M-SIA (6)	54	15	21	< 5		M-SIA* (7)	31	26	25	10
NEW JERSEY						NEW JERSEY				
SCA (41)	85	22	32	10		SCA (11)	82	0	18	45
M-SIA (17)	30	57	9	< 5		M-SIA (9)	25	33	10	19
			-		UPPER BAY					
DELAWARE						DELAWARE				
SCA (50)	96	16	22	18		SCA (31)	100	26	26	3
M SIA (11)	90 01	10	22	10		M SIA (14)	12	20	20	5
M-SIA (11)	81	< 5	/	< 5		M-SIA (14)	13	83	< 5	< 5

SMALL WEAKFISH

4.2.2.1.2 Medium Sized Weakfish Muscle

Sources used for medium Weakfish were Bay Anchovy/larval clupeids, crangon, gammarid amphipods, isopods, mysids, and polychaetes. After isospace plots were inspected, the TEFs of all sources were set to 1 (SD 0.2) for δ^{13} C and 3.4 (SD 0.2) for δ^{15} N. Using these standard parameter settings, 9 out of 10 models converged without issues. All medium Weakfish mixing model runs used a MCMC chain length of 100,000.

DE/Summer/Lower

Model estimations of median diet proportions were $31.1\% (\pm 13.8\%)$ for gammarids and $57.6\% (\pm 17.4\%)$ for mysids (Figure 4.13).



Figure 4.13. Posterior density plot of medium sized Weakfish caught during the summer in the DE-Lower. n = 10

DE/Fall/Lower

Model estimations of median diet proportions were 33.4% (\pm 15.4%) for gammarids and 57.6% (\pm 17.5%) for mysids.

DE/Summer/Middle

Model estimations of median diet proportions were 26.7% (\pm 14%) for gammarids,

44.2% (\pm 19.9%) for mysids and 9.7% (\pm 13.3%) for polychaetes.

DE/Fall/Middle

Model estimations of median diet proportions were 76.5% (\pm 18.1%) for mysids and less than 5% for all other sources.

DE/Summer/Upper

Model estimations of median diet proportions were 8.3% (\pm 11.1%) for gammarids and 68.6% (\pm 17.4%) for mysids.

DE/Fall/Upper

Model estimations of median diet proportions were 71% (\pm 15.9%) for gammarids and 17% (\pm 13.1%) for mysids.

NJ/Summer/Lower

Model estimations of median diet proportions were 64.2% (\pm 9.9%) for gammarids and 28.7% (\pm 10.4%) for mysids (Figure 4.14).



Figure 4.14. Posterior density plot of medium sized Weakfish caught during the summer in the NJ-Lower. n = 16

NJ/Fall/Lower

Using the standard parameters and priors from NJ/Fall/Lower fish, the first attempt of the model run did not converge. To give the model a better chance to converge, a model was run without Bay Anchovy/larval clupeids as a source due to its absence in the diet and its enriched δ^{15} N values relative to other sources. This modification allowed the model to converge, with model estimations of median diet proportions of 52.1% (± 19.3%) for gammarids and 34.7% (± 14.8%) for mysid shrimp.

NJ/Summer/Middle

Model estimations of median diet proportions were 49.8% (\pm 13.1%) for gammarids, 21.1% (\pm 12.4%) for mysids, and 17.2% (\pm 14.5%) for polychaetes.

NJ/Fall/Middle

Model estimations of median diet proportions were 13.5% (\pm 13.1%) for gammarids, 8.9% (\pm 10.9%) for isopods, 31.1% (\pm 15.7%) for mysids, and 28.3% (\pm 17.9%) for polychaetes. 4.2.2.1.3 Large Sized Weakfish muscle

Sources used for large Weakfish were the same as those used for medium Weakfish. After isospace plots were inspected, the TEFs of all sources were set as 1.2 (SD 0.2) for δ^{13} C and 3.5 (SD 0.2) for δ^{15} N. Using these standard parameter settings, 6 out of 10 models converged without issues. All large Weakfish muscle mixing model runs used a MCMC chain length of 100,000.

DE/Summer/Lower

Model estimations of median diet proportions were 42.8% (\pm 14.1%) for gammarids and 44.3% (\pm 16%) for mysids.

DE/Fall/Lower

Model estimations of median diet proportions were 6.4% (\pm 6.9%) for Bay Anchovy/larval clupeids, 20.6% (\pm 16.2%) for gammarids, 18.2% (\pm 16.2%) for isopods, and 43.3% (\pm 14.8%) for mysids.

DE/Summer/Middle

Using the standard parameters and priors from DE/Summer/Middle fish, the first attempt of the model run did not converge. To give the model a better chance to converge, an uninformative prior was used and successfully allowed the model to converge. Model estimations of median diet proportions using an uninformative prior were 8.4% (\pm 9.2%) for Bay Anchovy/larval clupeids, 9.3% (\pm 9.6%) for crangon, 17.7% (\pm 14.2%) for gammarids, 6.6% (\pm 7.5%) for isopods, 23.3% (\pm 18.6%) for mysids, and 18.6% (\pm 16.6%) for polychaetes.

DE/Fall/Middle

Model estimations of median diet proportions were 5.5 % (\pm 5.6%) for crangon, 11.4% (\pm 15%) for gammarids, and 57.5% (\pm 20.4%) for mysids.

DE/Summer/Upper

Model estimations of median diet proportions were 7.6% ($\pm 11.4\%$) for Bay

Anchovy/larval clupeids, 23.2% (\pm 15.2%) for crangon, 33.2% (\pm 16.5%) for gammarids, and

25.7% (± 17.8%) for mysids (Figure 4.15).



Figure 4.15. Posterior density plot of large sized Weakfish caught during the summer in the DE-Upper. n = 5

NJ/Summer/Lower

Model estimations of median diet proportions were 51.3% (\pm 12%) for gammarids and 41.8% (\pm 12.1%) for mysids.

NJ/Fall/Lower

Model estimations of median diet proportions were 42.2% (\pm 12.9%) for gammarids, 5.3% (\pm 10.1%) for isopods, and 42.5% (\pm 13.3%) for mysids.

NJ/Summer/Middle

Model estimations of median diet proportions were 43.7% (\pm 14.6%) for gammarids,

23.2% (\pm 14.2%) for mysids, and 18.1% (\pm 15.1%) for polychaetes (Figure 4.16).



Figure 4.16. Posterior density plot of large sized Weakfish caught during the summer in the NJ-Middle. n = 7

NJ/Fall/Middle

Model estimations of median diet proportions were 20.8% (\pm 13.8%) for gammarids,

16% (\pm 12.7%) for isopods, and 48.9% (\pm 15.2%) for mysids.

4.2.2.2 2018 Weakfish Liver

4.2.2.2.1 Medium Sized Weakfish liver

Sources used for medium Weakfish liver were the same used for the medium and large size classes for muscle tissue. After isospace plots were inspected, the TEFs of all sources were set as 2.8 (SD 0.4) for δ^{13} C and 2.2 (SD 0.2) for δ^{15} N. Using these standard parameter settings, 8 out of 9 models converged without issues. All medium Weakfish liver mixing model runs used a MCMC chain length of 100,000.

DE/Summer/Lower

Model estimations of median diet proportions were 63.1% (\pm 16.8%) for gammarids and 29.3% (\pm 16.5%) for mysids (Figure 4.17).



Figure 4.17. Posterior density plot of medium sized Weakfish liver caught during the summer in the DE-Lower. n=8

DE/Fall/Lower

Model estimations of median diet proportions were 36.1% (\pm 16%) for gammarids and 53.3% (\pm 17.5%) for mysids.

DE/Summer/Middle

Model estimations of median diet proportions were 52.4% (\pm 19.7%) for gammarids, 24.3% (\pm 16.6%) for mysids, and 7.3% (\pm 11.9%) for polychaetes.

DE/Fall/Middle

Model estimations of median diet proportions were 6.4% (\pm 7.9%) for Bay Anchovy/larval clupeids and 72.7% (\pm 17.8%) for mysids.

DE/Summer/Upper

Model estimations of median diet proportions were 8.9% (\pm 11.3%) for gammarids and 62.1% (\pm 18.6%) for mysids.

DE/Fall/Upper

Using the standard parameters and priors from NJ/Fall/Lower fish, the first attempt of the model run did not converge. To give the model a better chance to converge, a flat/uninformative prior was used with only four sources. Bay Anchovy and crangon removed as sources because they did not appear to contribute to the mixing system after the isospace plot was inspected. These modifications allowed the model to converge, with model estimations of median diet proportions being 82.4% (\pm 16.6%) for gammarids, 5.1% (\pm 10.1%) for mysids, and 4.8% (\pm 9.4%) for polychaetes.

NJ/Summer/Lower

Model estimations of median diet proportions were 59.4% (\pm 18.4%) for gammarids and 32.3% (\pm 16.7%) for mysids.

NJ/Summer/Middle



Model estimations of median diet proportions were 75.8% (\pm 10.3%) for gammarids, 9.3% (\pm 7.8%) for mysids, and 7.1% (\pm 7.2%) for polychaetes (Figure 4.18).

Figure 4.18. Posterior density plot of medium sized Weakfish liver caught during the summer in the NJ-Middle. n=13

NJ/Fall/Middle

Model estimations of median diet proportions were 5.9% (\pm 8.2%) for crangon, 5.7% (\pm

13%) for gammarids, 42.6% (\pm 19%) for mysids, and 27.3% (\pm 17.8%) for polychaetes.

Table 4.22. Comparison table of the available SCA (%F) and MixSIAR results (%) of medium sized Weakfish from 2018. M-SIA and L-SIA represents stable isotope analysis of muscle tissue and liver tissue, respectively. The superscript dash (⁻) represents a model that converged with Bay Anchovy removed as a source and the superscript plus symbol (⁺) represents a model that converged with an uninformative prior and crangon and Bay Anchovy removed as sources. The source of Bay Anchovy/UID fish includes larval clupeids for MixSIAR results.

						<u>MEDI</u>	<u>UM WEAKFISH</u>						
			SUMMER			L	OWER BAY			FALL			
DELAWARE							DELAWARE						
	Mysids	Gammarids	Polychaetes	Isopods	Crangon	Bay Anchovy+	UID Fish	Mysids	Gammarids	Polychaetes	Isopods	Crangon	Bay Anchovy+UID Fish
SCA (38)	95	82	11	3	3	18	SCA (34)	100	44	3	0	26	6
M-SIA (10)	58	31	< 5	< 5	< 5	< 5	M-SIA (12)	58	33	< 5	< 5	< 5	< 5
L-SIA (8)	29	63	< 5	< 5	< 5	< 5	L-SIA (6)	53	36	< 5	< 5	5	< 5
NEW JERSEY							NEW JERSEY						
SCA (34)	90	43	3	7	13	13	SCA (21)	100	33	0	5	10	0
M-SIA (16)	29	64	< 5	< 5	< 5	< 5	M-SIA ⁻ (6)	35	52	< 5	< 5	< 5	< 5
L-SIA (8)	32	59	< 5	< 5	< 5	< 5	L-SIA	-	-	-	-	-	-
						Μ	IDDLE BAY						
DELAWARE							DELAWARE						
SCA (30)	73	77	33	20	20	20	SCA (37)	95	5	8	8	16	33
M-SIA (11)) 44	27	10	< 5	< 5	< 5	M-SIA (19)	77	< 5	< 5	< 5	< 5	< 5
L-SIA (6)	24	52	7	< 5	< 5	< 5	L-SIA (12)	72	< 5	< 5	< 5	< 5	6
NEW JERSEY							NEW JERSEY						
SCA (40)	78	80	63	18	8	38	SCA (16)	94	19	69	19	38	19
M-SIA (15)	21	50	17	< 5	< 5	< 5	M-SIA (11)	31	14	28	9	< 5	< 5
L-SIA (13)) -	-	-	-	-	-	L-SIA (8)	43	6	27	< 5	6	< 5
						U	PPER BAY						
DELAWARE							DELAWARE						
SCA (31)	90	26	13	16	13	6	SCA (47)	98	19	26	9	21	23
M-SIA (12)	69	8	< 5	< 5	< 5	< 5	M-SIA (18)	17	71	< 5	< 5	< 5	< 5
L-SIA (6)	62	9	< 5	< 5	< 5	< 5	$L-SIA^+(12)$	5	82	5	< 5	< 5	< 5
L 511 (0)	. 02	/	~ 5	~ 2	~ ~	~ 5	1.5111 (12)	2	02	5	~~	~ ~	

4.2.2.2.2 Large Sized Weakfish Liver

Sources used for large Weakfish liver were the same used for the medium and large size classes for muscle tissue. After isospace plots were inspected, the TEFs of all sources was set as 2.8 (SD 0.4) for δ^{13} C and 2.2 (SD 0.2) for δ^{15} N. Using these standard parameter settings, 6 out of 9 models converged without issues.

DE/Summer/Lower

Model estimations of median diet proportions were 55.4% (\pm 14.4%) for gammarids and 36.5% (\pm 14.5%) for mysids.

DE/Fall/Lower

Model estimations of median diet proportions were 5.2% (\pm 5.9%) for Bay Anchovy/larval clupeids, 31.4% (\pm 13.6%) for gammarids, and 51.4% (\pm 16.5%) for mysids (Figure 4.19).



Figure 4.19. Posterior density plot of large sized Weakfish liver caught during the fall in the DE-Lower. n=11

DE/Summer/Middle

Using the standard parameters and priors from DE/Summer/Middle fish, the first attempt of the model run did not converge. To give the model a better chance to converge, an uninformative prior was used which successfully allowed the model to converge. Model estimations of median diet proportions using the uninformative prior were 6.7% (\pm 8%) for Bay Anchovy/larval clupeids, 6.4% (\pm 7.1%) for crangon, 22.5% (\pm 16.1%) for gammarids, 14% (\pm 14.2%) for isopods, 16.9% (\pm 14.2%) for mysids, and 18.9% (\pm 15.2%) for polychaetes. *DE/Fall/Middle*

Model estimations of median diet proportions were 5.4% (\pm 5.3%) for crangon, 38.6% (\pm 19.1%) for gammarids, and 39.3% (\pm 18.4%) for mysids.

DE/Summer/Upper

Using the standard parameters and priors from DE/Summer/Upper fish, the first attempt of the model run did not converge. To give the model a better chance to converge, an uninformative prior was used and successfully allowed for model convergence. Model estimations of median diet proportions using a flat prior were 8.6% (\pm 11.5%) for Bay Anchovy/larval clupeids, 16.4% (\pm 13.6%) for crangon, 22.5% (\pm 16.2%) for gammarids, 8.2% (\pm 11%) for isopods, 14.6% (\pm 18.3%) for mysids, and 9.9% (\pm 12%) for polychaetes. *NJ/Summer/Lower*

Using the standard parameters and priors from NJ/Summer/Lower fish, the first attempt of the model run did not converge. To give the model a better chance to converge, an uninformative prior was used and successfully allowed the model to converge. Model estimations of median diet proportions from the uninformative prior were 64.9% (\pm 11.8%) for gammarids, 7.8% (\pm 8.5%) for isopods, 6.2% (\pm 7.2%) for mysids, and 7.2% (\pm 8.6%) for polychaetes.

NJ/Fall/Lower

Model estimations of median diet proportions were 55.7% (\pm 14%) for gammarids and 35.8% (\pm 14.1%) for mysids (Figure 4.20).



Figure 4.20. Posterior density plot of large sized Weakfish liver caught during the fall in the NJ-Lower. n=8

NJ/Summer/Middle

Model estimations of median diet proportions were 55.9% (\pm 15.7%) for gammarids,

17.8% (± 12.6%) for mysids, and 13.8% (± 14.1%) for polychaetes.

NJ/Fall/Middle

Model estimations of median diet proportions were 6.1% (\pm 7.1%) for crangon, 38.8% (\pm

18%) for mysids, and 31.5% (\pm 18.8%) for polychaetes.

Table 4.23. Comparison table of the available SCA (%F) and MixSIAR results (%) of large sized Weakfish from 2018. M-SIA and L-SIA represents stable isotope analysis of muscle tissue and liver tissue, respectively. Numbers in parentheses represent the sample sizes of each method. Asterisks (*) represent models that converged only with uninformative priors. The source of Bay Anchovy/UID fish includes larval clupeids for MixSIAR results.

			SUMMER			LOWER BA	AY			FALL			
DELAWARE							DELAWARE						
	Mysids	Gammarids	Polychaetes	Isopods	Crangon	Bay Anchovy+ UID Fish		Mysids	Gammarids	Polychaetes	Isopods	Crangon	Bay Anchovy+UID Fish
SCA (19)	95	79	5	5	11	16	SCA (27)	100	11	4	4	22	30
M-SIA (5)	44	43	< 5	< 5	< 5	< 5	M-SIA (13)	43	21	< 5	18	< 5	6
L-SIA (10)	37	55	< 5	< 5	< 5	< 5	L-SIA (11)	51	31	< 5	< 5	< 5	5
NEW JERSEY							NEW JERSEY						
SCA (16)	94	38	0	6	0	12	SCA (23)	100	35	0	13	17	13
M-SIA (10)	42	51	< 5	< 5	< 5	< 5	M-SIA (10)	43	42	< 5	5	< 5	< 5
L-SIA* (10)	6	65	7	8	< 5	< 5	L-SIA (8)	36	56	< 5	< 5	< 5	< 5
						MIDDLE B	AY						
DELAWARE							DELAWARE						
SCA (5)	60	60	0	60	0	0	SCA (16)	100	6	13	13	50	38
M-SIA (5)	23	18	19	7	9	8	M-SIA (13)	58	11	< 5	< 5	6	< 5
L-SIA (5)	17	23	19	14	6	7	L-SIA (12)	39	39	< 5	< 5	5	< 5
NEW JERSEY							NEW JERSEY						
SCA (13)	100	85	77	8	0	85	SCA (14)	93	21	79	29	43	7
M-SIA (7)	23	44	18	< 5	< 5	8	M-SIA (9)	49	21	< 5	16	< 5	< 5
L-SIA* (7)	18	56	14	< 5	< 5	< 5	L-SIA (7)	39	< 5	31	< 5	6	< 5
						UPPER BA	AY						
DELAWARE	10		0	0	10	20							
SCA (5)	40	60	0	0	40	20							
M-SIA (5)	26	33	< 5	< 5	23	8							
L-SIA* (4)	15	23	10	8	16	9							

LARGE WEAKFISH

4.2.2.3 Mysid Shrimp

Sources used for mysid shrimp were BMA, *Phragmites*, *Spartina*, and SPOM. After isospace plots were inspected, the TEFs of all sources was set as 1.5 (SD 0.5) for δ^{13} C and 3.5 (SD 0.5) for δ^{15} N. Using these standard parameter settings, the model converged without issues. *4.2.2.3.1 Delaware Bay Organic Matter Sources*

Model estimations of median diet proportions were 55.7% (\pm 13.8%) for BMA, 15.2% (\pm 8.2%) for *Phragmites*, 12.3% (\pm 7.2%) for *Spartina*, and 14.6% (\pm 10.2%) for SPOM. Although these results signify that BMA is a major contributor to mysid shrimp production, the results here are unreliable as consumer values did not fit within the convex hull (Figure 4.21). The consequences of sources forming a straight line rather than a polygon (after TEFs are added to the sources) is evidence that the model was nonsensical and thus should not be used in any aspect of ecological inference. The inclusion of δ^{34} S into the model is necessary for the model results to be realistic.



Figure 4.21. Isospace plot of mysid shrimp (individual dots) and the Delaware Bay endmembers of *P. australis*, SPOM, BMA (Currin et al. 2003), and *Spartina spp*.

CHAPTER 5: DISCUSSION, CONCLUSIONS, AND FUTURE RESEARCH RECOMMENDATIONS

5.1 Discussion

5.1.1 Drivers of Juvenile Weakfish Stable Isotope Values in the Delaware Bay

The results of the GLMs revealed that the isotopic values of juvenile Weakfish are influenced by multiple variables. The nested model terms elucidated that individual stations within certain bay locations and individual months within certain seasons contributed to differences in δ^{13} C, δ^{15} N, and δ^{34} S. Although there were no general trends between size classes, GLM results showed within-location variability between states for individual size classes, indicating that Weakfish from opposite sides of the bay exhibit differing delta values, possibly pointing towards differential basal food webs between states. Of the four models run for every size class, tissue, and isotope, models 2 and 4 consistently had the most parsimonious fit to the data as determined by the AICc values. The dichotomy between these models suggests that there are two differing mechanisms that best explain δ^{13} C and δ^{15} N values of juvenile Weakfish – state/location/station, season, and weight (model 2), and the state*season interaction plus location/station (model 4).

5.1.1.1 2017 Weakfish

5.1.1.1.1 Small Sized Weakfish (<60mm SL)

Small Weakfish δ^{13} C and δ^{15} N values were best described by model 4. For δ^{13} C, multiple significant estimates of location/station terms revealed that organic matter sources of small Weakfish are specific to certain locales (Litvin and Weinstein 2004) whereas δ^{15} N values of small Weakfish showed that season and bay location were better explanatory variables for this isotope. Assuming that all small Weakfish feed on the same trophic level, the increasingly

positive estimates of summer δ^{15} N values toward the head of the estuary could be indicative of either conservative mixing along the salinity gradient or increased nitrogen-loading from local ground-borne wastewater seepage (McClelland et al. 1997; Fry 2002; Litvin and Weinstein 2004; Sharp et al. 2009).

5.1.1.1.2 Medium Sized Weakfish (60-100mm SL)

Medium Weakfish $\delta^{15}N$ was best described by model 4. The best model terms that described $\delta^{15}N$ were state/location/station and season/month. The significant estimates from each model term are further evidence that $\delta^{15}N$ values vary over time and space within the estuary.

Using Weakfish liver as a determinant of short-term diet, GLM results of medium Weakfish liver illuminated that the data was best explained by model 2, indicating that the inclusion of weight as a model term succeeded in creating the most parsimonious fit of the data. However, weight was not shown to be significant for either model (both δ^{13} C and δ^{15} N). Regardless of its significance as a model parameter, the inclusion of weight into the model should be considered when inferring the drivers of Weakfish δ^{13} C values in the Delaware Bay. As the most significant model term of δ^{13} C for medium Weakfish liver, the positive estimate of summer/August revealed that these fish exhibit more enriched δ^{13} C values during the summer that are associated with benthic organic matter sources. Cross-validating this GLM result with the stomach contents of medium Weakfish caught during the summer in 2017 revealed that gammarid consumption was higher than any other prey source. As known detritus consumers, this significant positive estimate conforms to the expected values of a consumer that derives its organic matter from a benthic food web. The GLM results of $\delta^{15}N$ of medium Weakfish liver did not show any general relationship with any of the model variables except the location term of the upper bay. Similar to the finding of δ^{15} N values of small Weakfish, the upper bay displayed more enriched $\delta^{15}N$ values compared to those found in the middle and lower bays. In addition, the opposite estimates of summer/July (negative estimate) and summer/August (positive estimate) is evidence that $\delta^{15}N$ values in tissues with rapid turnover rates can change significantly in short periods of time.

5.1.1.1.3 Large Sized Weakfish (100-137mm SL)

GLM results for large Weakfish in 2017 elucidated that the data were overdispersed for δ^{13} C and δ^{15} N. The implications of this finding suggest that something other than the model terms drove the δ^{13} C and δ^{15} N values of large Weakfish in 2017. GLM residuals of δ^{34} S were not overdispersed, and the delta values were indicative of organic matter sources derived from a pelagic food web rather than a benthic food web.

For large Weakfish liver, GLM results showed that model 2 had the most parsimonious fit to the data for both isotopes. Model 2 results for δ^{13} C also show negative estimates for DE/upper and NJ/middle. The negative estimate of NJ/middle suggests that large Weakfish from this location are feeding on a more pelagic food web, or these fish could be reflecting isotopic values associated with conservative mixing due to the number of fish caught near the Maurice and Cohansey River deltas. For DE/upper, a possible explanation of the significant negative estimate can be attributed to the potential incorporation of *Phragmites* detritus due to the increased abundance of *Phragmites* in the upper bay (Wainright et al. 2000; Currin et al. 2003; Litvin and Weinstein 2004). *Phragmites* δ^{13} C values are highly depleted relative to other endmembers of the Delaware Bay (Figure 4.21) and have been known to be expressed in the delta values of juvenile Weakfish from the upper Delaware Bay (Litvin and Weinstein 2004). Additionally, the model term of weight showed that δ^{13} C values increase with increased weight of Weakfish, suggesting that Weakfish show a shift in trophic niche from a more pelagic based food-web toward a mixed benthic/pelagic based food-web with increasing growth.

5.1.1.2 2018 Weakfish

5.1.1.2.1 Small Sized Weakfish (<60mm SL)

 δ^{15} N values of small Weakfish caught in 2018 were best explained by model 4. Multiple significant estimates of model terms indicated that δ^{15} N values vary by state, station, and over each state during the same season/month. Results from model 4 for δ^{15} N showed a significant difference between DE and NJ, with NJ fish exhibiting more depleted values than DE fish throughout their estuarine residency. It is possible that this relationship is indicative of depleted baseline δ^{15} N values in NJ, or, since upper bay values typically display more enriched δ^{15} N values, the lack of samples in NJ-upper compared to DE-upper could be positively weighing mean δ^{15} N values of small Weakfish from the DE side. Regardless of the NJ/DE differences, the eight significant estimates of model 4 for 2018 small Weakfish δ^{15} N is evidence that there was no specific driver of δ^{15} N based upon the general model terms of state, location, and season. Rather, it appears that δ^{15} N of small Weakfish caught in 2018 displayed values that were specific to the locale in which they were caught.

5.1.1.2.2 Medium Sized Weakfish (60-100mm SL)

 δ^{13} C and δ^{15} N values of 2018 medium Weakfish muscle were best explained by model 2. Although there were significant model terms for both isotopes, the data for both δ^{13} C and δ^{15} N were overdispersed. Thus, the results of this model should not be used due to the dispersion parameters being greater than 1. This finding suggests that there are other variables not considered in the model that would better describe the drivers of δ^{13} C and δ^{15} N of medium Weakfish muscle in the Delaware Bay. Model 2 also had the most parsimonious fit to the data
for δ^{13} C and δ^{15} N of medium Weakfish liver. While the residuals for model 2 of δ^{15} N of medium Weakfish liver were also calculated to be overdispersed, the data for δ^{13} C was not, indicating that the inclusion of weight into the model helps to explain delta values of medium Weakfish. Although weight did not produce a significant estimate as a model term, DE/upper had a significant negative estimate and two state/location/station terms within DE-middle also provided significant negative estimates. These estimates are consistent with other GLM results from 2017 in that they conform to the previous findings of more depleted δ^{13} C values from saltwater to freshwater gradients associated with the *Spartina* to *Phragmites* gradient (Fry 2002; Currin et al. 2003) and variable δ^{13} C values from different locales (Litvin and Weinstein 2004). *5.1.1.2.3 Large Sized Weakfish (100-137mm SL)*

 δ^{13} C and δ^{15} N values of 2018 large Weakfish were best explained by model 2. Although not significant, the inclusion of weight to the model again indicates that weight contributes to Weakfish isotopic values. δ^{13} C of model 2 was best described by four stations that exhibited increasingly negative estimates from the farthest south station to the farthest north station. These results are further evidence of the δ^{13} C expression of the saltwater to freshwater and *Spartina* to *Phragmites* gradients. The other significant estimate was that of fall/October, which also produced a negative estimate. Unlike medium Weakfish caught in the fall who displayed a positive estimate, the negative estimate from large Weakfish caught during the fall could be the result of the slower turnover rate of large Weakfish muscle which could still be reflecting the depleted spring/summer phytoplankton end-member values. This estimate was quite weak, (-1.35, SE 0.5), which could indicate that δ^{13} C values are beginning to equilibrate to the more enriched values of phytoplankton arising from the period of slower growth (Pennock and Sharp 1986). Results from model 2 for δ^{15} N of 2018 large Weakfish only displayed significant estimates of two state/location/station terms that revealed positive estimates of two DE-upper stations, signifying the δ^{15} N enrichment proceeding from the mouth of the bay toward the Delaware River. Assuming that these Weakfish exhibited some degree of site fidelity, this finding suggests that the upper bay consistently shows more enriched δ^{15} N associated with both conservative mixing and anthropogenic nitrogen either from local sources (groundwater-borne wastewater) or the greater Wilmington and Philadelphia regions (Fry 2002; Litvin and Weinstein 2004).

The GLM results of 2018 large Weakfish liver revealed that models 4 and 3 best described δ^{13} C and δ^{15} N, respectively. Although these most parsimonious fit models were in contrast with those for muscle tissue, the significant model terms were similar for both tissues. NJ*fall/October showed a significant negative relationship, conforming to the estimate for fall/October for muscle tissues. The fact that both tissues exhibit similar estimates for the same time period suggests that large Weakfish muscle also exhibits rapid turnover time.

GLM results of δ^{15} N for large Weakfish liver produced multiple significant estimates that were shared with other size classes and tissues. The strongest estimates from model 3 revealed that DE-upper stations (and one DE-middle station) exhibit more enriched δ^{15} N values, again conforming to the δ^{15} N enrichment of fish caught in the upper bay. There were also strong estimates from NJ-middle stations that produced both positive and negative estimates of δ^{15} N. These differing estimates could be the result of within-location variability of δ^{15} N or the expression by different communities of Weakfish feeding on different trophic levels over the short-term as expressed by the rapid turnover rate of liver tissue (Buchheister and Latour 2011).

In summary, the findings from the GLM results of 2017 and 2018 Weakfish reveal that $\delta^{13}C,\,\delta^{15}N,$ and $\delta^{34}S$ are mediated by multiple interactions over time and space within the Delaware Bay. Differences of delta values of all three isotopes imply that stable isotope values of different size class juvenile Weakfish vary over each state (DE and NJ), season (summer and fall), month, location (lower, middle, upper) and station. I hypothesize that seasonal, locational, and between state variability of isotopic values are likely dictated via bottom-up effects in large areas as well as smaller locales as has been shown in other systems (Peterson and Fry 1987; Cloern et al. 2002; Buchheister and Latour 2011). The implications of these results provide fisheries scientists and ecosystem modelers with updated information regarding the local variability of primary productivity and juvenile Weakfish production in the Delaware Bay. Notably, the enriched δ^{15} N values of upper bay Weakfish appeared to provide a signal of anthropogenic nitrogen entering the ecosystem from upriver urban development and/or local wastewater seepage. The GLM results of multiple tissues with varying turnover rates suggest that juvenile Weakfish exhibit a relatively high degree of site fidelity during the same season and/or month as was observed for the large Weakfish muscle and liver tissues from Weakfish caught in October in New Jersey. Additionally, the finding that individual stations exhibited unique delta values is further evidence that juvenile Weakfish uptake local sources of organic matter relatively quickly and appear to remain in the same general area long enough to express the unique delta values of individual localities (e.g. stations). Lastly, although there was variability between and among different model parameters, juvenile Weakfish tissues also displayed expected delta values as a function of conservative mixing (Chanton and Lewis 1999; Fry 2002), the Spartina to Phragmites gradient (Litvin and Weinstein 2004), and seasonal phytoplankton availability in the Delaware Bay (Pennock and Sharp 1986; Fogel et al. 1992).

5.1.2 Spatiotemporal Diet of Juvenile Weakfish in the Delaware Bay Characterized by Stomach Content and Stable Isotope Analyses

For 2017 and 2018, the diet of the three different size class juvenile Weakfish examined in this study was found to change over time and space – seasonally, along the salinity gradient, and between states. Between year variability was found for paired states, locations, and seasons. However, this was expected as fish were only caught once per month from each station, significantly weighing the findings towards individual Weakfish diets during sampling days. Due to the expected differences of SCA results between years, only similarities between both years will be discussed.

Overall, the three size classes of Weakfish exhibited similar diets, with both methods showing a general reliance on crustacean prey rather than fish prey. While there was a general trend of increased fish consumption with increasing size classes, the expected trend of juvenile Weakfish being strictly mysid specialists when small to consuming equal proportions of fish and invertebrates when large was not found via either method, somewhat complying with the findings of Brust et al. (2009) who found that Weakfish diet shifted from large invertebrates and forage fish to small invertebrates only. SCA results from 2017 and 2018, and 2018 MixSIAR results revealed that the small and medium size classes fed mainly on mysid shrimp, gammarid amphipods, and polychaete worms, while other prey items occurred less frequently. Rather than an ontogenetic shift from mysids to equal proportions of fish and invertebrate prey, the diet results of this study show that an ontogenetic shift occurs from mysids, gammarids, and polychaetes to a slight increase in predation of fish prey, but mainly larger invertebrates such as crangon. However, larger Weakfish exhibited a continued reliance on smaller invertebrates – evidence of interspecies competition between the small and large size classes for small

invertebrates. While no attempt was made to calculate natural mortality, increased intraspecies competition can intensify the consequences of interspecies competition with Bluefish and other marine transient species (Brust et al. 2016).

Results from both years also revealed that the diet of all size classes for each season and state consisted of higher frequencies of prey species in the middle bay compared to fish caught from the lower and upper bays. Specifically, while mysids and gammarids were consistently important prey sources for all bay locations, the diet of middle bay Weakfish included higher frequencies of polychaete worms, isopods, crangon, other arthropod spp., and Bay Anchovy/UID fish. Additionally, SCA from both years elucidated several seasonal relationships of prey items for each state and bay location. From summer to fall, the %F of gammarid amphipods and polychaete worms decreased significantly. Contrarily, the %F of mysid shrimp and crangon increased from summer to fall. While this relationship was strongest in the middle bay over both years for each state, these findings point out the seasonal availability of prey items for juvenile Weakfish in the Delaware Bay.

With regards to between state variability within a certain bay location (e.g. comparison between DE and NJ for the lower bay), SCA results showed a general trend of higher %F of gammarids in DE when compared to NJ, whereas NJ Weakfish showed increased predation rates of other arthropod spp. (copepods, small crabs, crab megalopa, and larval horseshoe crabs) when compared to DE. However, close examination of these relationships for 2018 SCA and SIA results revealed an opposite trend – while %F of gammarids was consistently higher in DE than NJ via SCA, MixSIAR results consistently showed higher diet proportions of gammarids in Weakfish from NJ compared to DE, especially for the small and medium size classes.

The consumption of different prey items measured by %F and MixSIAR was found to vary over different bay locations of each state for certain seasons. Beginning with Delaware 2017 and 2018, mysid shrimp %F was generally highest during the summer in the upper bay. MixSIAR results from muscle tissue for the small and medium size classes also corroborated this relationship. This observation contrasts with the findings of Grecay and Targett (1996a) who found that Weakfish in the middle bay had a higher percentage of mysid shrimp in their gut (by weight) than any other prey item compared to the lower and upper bays. During the summer, gammarid consumption via %F and MixSIAR diet proportions was shown to be highest in the lower bay. During the same season, polychaete worm and isopod consumption consistently showed %F and MixSIAR diet proportions to be highest in the middle bay. I speculate that this observation is linked to the previously recorded spring bloom that typically occurs 30-50 km from the bay mouth (Pennock and Sharp 1986; Fogel et al. 1992; Able et al. 2009, 2018), which is at center and north of the middle bay location. Because phytoplankton support the base of the food web, its increased availability to primary consumers in the middle bay likely supports higher densities of small invertebrates at this location.

Prey items found in the stomachs of fish caught in New Jersey also exhibited locational variability in consumption rates and MixSIAR diet proportions of Weakfish. During the summer, gammarid amphipods were found to be highest in the lower bay. As found in DE, polychaete worms showed a strong relationship as shown by %F and MixSIAR diet proportions for being consumed the most in the middle bay. Lastly, during the fall, consumption rates of isopods were found to be highest in the middle bay, again conforming to the general trend of higher prey frequencies in the middle bay compared to the other bay locations.

While each method produced differing results regarding the importance of prey items of juvenile Weakfish over time and space, it is imperative to consider the limitations of each method, especially those of MixSIAR. As shown in Table 3.1, the time for isotopic turnover varies with the desired percent turnover, requiring the analyst to consider the significance of the MixSIAR results. Especially for the small Weakfish, where only muscle tissue was available, the median dietary proportion values of small Weakfish likely reflected the diet of the previous season or month. Even if the SCA and SIA results differed, they may both be accurate, as SCA represents diet on a matter of hours (Hyslop 1980) while SIA represents the diet over weeks to months (MacNeil et al. 2006; Buchheister and Latour 2010, 2011). On the other hand, the rapid turnover rate calculated for liver tissue theoretically represented the diet between 2 to 10 days prior for 50% and 99% turnover, respectively. Although the calculated turnover time is subject to error, the different rates of turnover between tissues used in this study are novel in that few studies implement species specific turnover rates. The turnover rates calculated here gives researchers more confidence in accepting the results found in this study. However, as noted in the MixSIAR results, the standard deviations were typically high, especially when the estimated median diet proportion was lower than 20% for a given source. This was likely due to the similarities between source values, as polychaetes and mysids occupied nearly equal isospace for δ^{13} C and δ^{15} N (Figure 5.1). Another limitation is the fact that I pooled sources over seasons, locations, and states. Some sources were mainly caught in DE or from a single bay location. Locational variability was shown for mysid shrimp using GLMs, thus the mixing models may be influenced by location specific delta values from various sources.



Figure 5.1. Isospace plot of small Weakfish sources demonstrating the proximity of mysid shrimp and polychaete worms. Individual dots represent values of TEF corrected small Weakfish from DE/Fall/Middle.

One of the most notable findings of this study was the apparent dependence of juvenile Weakfish on gammarid amphipods throughout the Delaware Bay and over the course of their estuarine residency. Contrary to previous studies (Grecay and Targett 1996a; Paperno et al. 1997; Nemerson 2001; Boutin and Targett 2019), both methods found that gammarid amphipods were particularly important prey items, especially during the summer months (July and August) when mysid shrimp are known to experience high natural mortality due to increased predation rates from other species (marine transient and resident species) and high water temperatures (Mayor and Chigbu 2018). While SCA showed high consumption rates of mysid shrimp over every size class, MixSIAR results for both muscle and liver tissue revealed that mysid shrimp rarely accounted for more than 50% of tissue assimilation. Gammarids, however, were shown to contribute >50% of assimilated diet on multiple occasions for both long and short terms as displayed by the slow turnover muscle tissue and rapid turnover liver tissue, respectively. Furthermore, differences in median diet proportions calculated via muscle and liver tissues were calculated, with high diet proportions of gammarids calculated from liver tissue in the fall showing the importance of this prey item throughout the estuarine residency of juvenile Weakfish in the Delaware Bay.

The results of this study that showed an ontogenetic shift in diet toward a more generalist diet of mainly small and larger invertebrates is potential evidence of strong intraspecies competition among different size class juvenile Weakfish over small invertebrates. Because diet contributes to growth and thus recruitment success (Houde 1989; Lankford and Targett 1994; Paperno et al. 2000), the implications of intraspecies competition between small and large juveniles (as well as small adults, personal observation) for small invertebrates can significantly reduce overall recruitment success of late hatching juvenile Weakfish. Furthermore, the within location/station site fidelity elucidated by GLMs of δ^{13} C for both years could be further evidence that nursery habitats of juvenile Weakfish are relatively small, possibly exacerbating the consequences associated with increased competition. The conclusion that Weakfish from the middle bay location, which supports the highest rates of primary productivity in the bay, may serve as the most suitable nursery region for juvenile Weakfish because of its role in supporting higher abundances of invertebrate prey.

5.2 Conclusions and Suggestions for Future Research

In summary, this study provides the first simultaneous analysis of juvenile Weakfish diet throughout the entire Delaware Bay (both DE and NJ nearshore zones) and over the course of their estuarine residency. The findings presented here directly answer the question posed in the 2016 Weakfish Stock Assessment to monitor Weakfish diets over broad regional and spatial scales. GLM results of carbon, nitrogen, and sulfur stable isotopes revealed two primary aspects regarding juvenile Weakfish movement and food web controls: 1) juvenile Weakfish exhibit a certain degree of site fidelity within individual size classes, and 2) their isotopic values are mediated by bottom-up controls via spatiotemporal primary productivity, conservative mixing, and possible anthropogenic nitrogen. The variability in delta values of the three stable isotopes could potentially be used as indicators of the local variability of nursery habitats described in Beck et al. (2001). These findings could have implications on future management decisions as the data show that juvenile Weakfish are affected by locally available organic matter and prey sources, suggesting that EBFM approaches should consider fine-scale variability when applying ecosystem-wide management decisions. Stomach content analysis and mixing model results also found variability in the diet of juvenile Weakfish for different prey items. The seasonal, state, and locational differences in their diet are further evidence of the local availability of various prey items. The results of this diet analysis show that the diet of juvenile Weakfish cannot be generalized throughout the Delaware Bay and over the course of their estuarine residency. Rather, more emphasis should be put on the locational and seasonal aspects of diet. While this study strictly analyzed the diet of juvenile Weakfish, I hypothesize that the same relationships can be found for other fish species in other temperate estuaries like the Delaware Bay. Finally, one of the most important findings of this study was the apparent importance of gammarid

amphipods to the Weakfish diet. Contrary to previous studies, this conclusion highlights a marked shift in the diet of juvenile Weakfish from mysids to gammarids in the Delaware Bay.

Future research should focus on monitoring the spatiotemporal variability of Weakfish prey items to determine if gammarid amphipods have established themselves as one of the dominant prey items of juvenile Weakfish in the bay. If the seasonal mysid-gammarid relationship continues, further investigation into the energy density between gammarids and mysids would provide valuable information regarding the overall survival, natural mortality, and recruitment success of juvenile Weakfish in the Delaware Bay. With regards to SIA, future efforts should focus on calculating species specific turnover rates for muscle and liver tissues. Additionally, since I was not able to determine the δ^{34} S values of important Weakfish prey items, future SIA studies in the Delaware Bay should focus on determining these unknown values. Establishing a bank of isotopic values for δ^{13} C, δ^{15} N, and δ^{34} S of a variety of organisms within the Delaware estuary will allow researchers to examine the relationships between pelagic, benthic, and marsh derived primary productivity and fisheries production. The apparent reliance that juvenile Weakfish have on small invertebrates such as mysid shrimp, gammarid amphipods, and polychaete worms underscores the importance of maintaining healthy habitats supporting invertebrate production contributing to essential fish habitat (EFH). Increased understanding of these relationships gives managers more hard evidence of the connectivity between anthropogenic development and the coastal zones that humans inhabit.

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