WestVirginiaUniversity
THE RESEARCH REPOSITORY @ WVU

Graduate Theses, Dissertations, and Problem Reports

2012

Estimation of striped bass (Morone saxatilis) diets using fatty acid signature analysis

Edward J. McGinley
West Virginia University

Follow this and additional works at: https://researchrepository.wvu.edu/etd

Recommended Citation

McGinley, Edward J., "Estimation of striped bass (Morone saxatilis) diets using fatty acid signature analysis" (2012). *Graduate Theses, Dissertations, and Problem Reports*. 3560.
https://researchrepository.wvu.edu/etd/3560

Estimation of striped bass (*Morone saxatilis*) diets using fatty acid signature analysis

Edward J. McGinley

Dissertation submitted to the
Davis College of Agriculture, Natural Resources, and Design
at West Virginia University
in partial fulfillment of the requirements
for the degree of

Doctor of Philosophy
in
Forest Resource Science

Approved by

Kyle J. Hartman, Ph.D., Chair
Ashok Deshpande, Ph.D.
John Jacobs, Ph.D.
Patricia Mazik, Ph.D.
Stuart A. Welsh, Ph.D.

Division of Forestry and Natural Resources

Mogantown, West Virginia
2012

Abstract

Estimation of striped bass (*Morone saxatilis*) diets using fatty acid signature analysis

Edward J. McGinley

Accurate estimates of diets are essential for the management of fisheries, especially when this information is used to construct food webs for a system. Traditionally, these studies have relied on examining the stomachs contents through direct observation of sacrificed fish, or using instruments to "flush" the items from the stomach. These methods only provide information on the recent feeding history. Fatty acid analysis is a biochemical technique that offers promise for examining diets in fish over a longer time scale than just the last few prey species consumed. The goal of this dissertation was to examine the feasibility and efficacy of using fatty acid signature analysis to record striped bass (*Morone saxatilis*) diets.

I examined how collection location (Upper or Lower Chesapeake bay), species, and season affected the fatty acid signature (a compilation of all fatty acids present) and lipid content of four common striped bass prey items: Atlantic menhaden (*Brevoortia tyrannus*), bay anchovy (*Anchoa mitchilli*), spot (*Leiostomus xanthurus*), and blue crab (*Callinectes sapidus*). Demersal species (spot and blue crab) were separated from pelagic species (menhaden and bay anchovy) based upon their fatty acid signature. Spot and blue crab were also grouped by season, with summer blue crab and spot distinct from fall blue crab and spot. Blue crab and spot within a season had very similar fatty acid signatures. Anchovy and menhaden did not show the same type of seasonal grouping as the demersal species. Anchovy and menhaden had the highest lipid content followed by spot, and blue crab had the lowest lipid content. Collection location did not appear to play a role in structuring the fatty acid signature. These results necessitate the collection of prey species at the same time as collection of predators for fatty acid signature analysis.

Fatty acids are deposited in tissues based upon the needs of that particular tissue. I assessed the diet history of striped bass based on two different tissues, adipose and liver tissue. Striped bass were held in flow through tanks at the NOAA Fisheries James J. Howard Laboratory in Sandy Hook, New Jersey, with water being pumped directly from the Sandy Hook Bay. Fish were fed a diet of spot for six weeks, at which point the spot diet was switched to a diet consisting of menhaden. Lipid levels in both tissues increased after the diet was switched to menhaden, a prey that had approximately twice the amount of lipid. The entire fatty acid signature did not change to mimic the prey as reported in a study in which the authors demonstrated that the fatty acid signature of cod (*Gadus morhua*) significantly changed to a squid signature in approximately three weeks. However, in the present study, certain marker fatty acids specific to the prey were able to distinguish the diet switch from spot to menhaden. The change in marker fatty acids and lipid levels was evident after a period of 31 days. Both liver tissue and adipose tissue demonstrated the change in diet, but adipose tissue may offer a more surgically feasible and non-lethal sample in striped bass.

The effect of striped bass size on fatty acid incorporation was analyzed for three different size classes; small (150 – 200 mm), medium (300 – 380 mm) and large (fish greater than 460 mm). Fish were housed in flow through tanks at the NOAA Laboratory in Oxford, Maryland, with water being pumped directly from the Tred Avon River. Striped bass were fed a diet of spot for four weeks, at which time the diet was switched to menhaden for four weeks. Lipid levels for

these fish indicated that there was little to no deposition of lipids throughout the experimental feeding of menhaden. Fatty acid signatures also indicated that the entire fatty acid signatures, nor marker fatty acids, were able to determine the diet switch. Based upon these findings and negligible growth, the most likely cause was a lack of consumption by striped bass. Due to high turbidity, feeding was difficult to observe.

One of the most promising aspects of fatty acids analysis is the ability to estimate the proportional contribution of different prey items to the diet using prey fatty acids and their respective lipid levels. The statistical program, quantitative fatty acid signature analysis (QFASA), can perform this type of analysis, and also takes into account the effect of predator metabolism of each fatty acid by using calibration coefficients. I tested this model using striped bass fed diets containing mixtures of spot and menhaden and a control diet of just menhaden. In this experiment, the striped bass were fed spot for six weeks before the menhaden feeding experiment began to allow sufficient time for the fatty acids to become homogenized within the striped bass tissues. The model correctly quantified the contribution of spot after six weeks, but it was unable to correctly assess the inputs from the mixed diets or menhaden diet alone. Recent studies have shown that fatty acids may take 12 – 14 weeks to stabilize in fish, which is twice as long as this experiment ran. Most of the work performed with QFASA has tested the model for homeotherms, e.g. marine mammals and seabirds. The fact that fish are poikilotherms may necessitate the duration of fatty acid incorporation to be on the scale of several months rather than weeks. Poikilotherms regulate the internal body temperature based upon ambient water temperatures, while homeotherms require a constant energy source to maintain a set body temperature. Fatty acids may be mobilized quicker and have a shorter retention time in homeotherm tissues. However, this situation is improbable for a generalist fish species like striped bass that will consume a variety of prey items and have the potential to be highly mobile.

Lastly, I tested the QFASA model on wild caught striped bass to determine the possibility of using this model on wild fish with prey items caught at the same time. Fish were caught during the fall when they are most likely to be consuming a high proportion of menhaden. Percent biomass of the stomach contents for these fish was compared to previous studies that collected similar sized fish (age-3) during the same season. The stomach contents of fish for this experiment, and fish from previous studies, showed that menhaden made the bulk of the diet (greater than 70%). The QFASA estimated the contribution of menhaden to be minimal (less than 2%). It is possible that striped bass were not feeding on menhaden for a long enough duration for the fatty acid signatures of menhaden to become predominant.

Dedication

I would like to dedicate this dissertation to my wife, Emily. She was always willing to help in both the field and laboratory without complaint, and was able to keep her sense of humor throughout the trying process. I would still be working on samples in the lab and writing my dissertation if not for her. I would also like to dedicate this to my loving parents, John and Mary Ellen. They have always supported my endeavors and told me I could grow up to be anything I wanted. With their help, I have achieved my dreams.

Acknowledgments

Table of Contents

List of Tables

Chapter 5

Chapter 6

List of Figures

Chapter 4

Chapter 5

Chapter 6

Chapter 1 – Literature Review

Striped bass ecology and exploitation

Striped bass (*Morone saxitilis*) are a large anadromous fish that spawn in the Chesapeake Bay tributaries between April and May and then return to the coastal waters afterwards (Chapoton and Sykes 1961). The Chesapeake Bay is not the only spawning grounds, but may be responsible for a majority of the coastal stock (Kohlenstein 1981). Eggs hatch in about five to seven days, with larvae consuming their yolk sac for the next several days (Setzler-Hamilton and Hall 1991). Age-3 striped bass and younger may move minimally away from the natal estuaries (Mansueti 1961). Juveniles feed upon a diet of mostly invertebrates, but they also consume larvae of other fish (Boynton et al. 1981). Age-1 striped bass still consume a diet of invertebrates, but start to feed on fish as the year continues, and by age-2 these fish are primarily piscivores (Hartman and Brandt 1995).

Striped bass males reach sexual maturity at age-2 while only a few females reach maturity at the same age. Females may not spawn annually until they reach age-7 (Maryland DNR 1987). Once sexual maturity is reached, some striped bass migrate to coastal waters to join the migratory population. Some fish may remain in the Bay until they reach anywhere between three to eight years with older year classes representing a smaller and smaller proportion of the resident population (Rugolo and Jones 1989).

The large numbers and size of striped bass have supported a commercial and recreational fishery since colonial times (CBEF 2006). However, starting in the 1970s, annual recruitment of striped bass began to plummet and the population began a drastic decline (ASMFC 1981). The U.S. Congress passed legislation that declared any state in violation of the plan prepared and adopted by Atlantic States Marine Fisheries Commission (ASMFC) would have a moratorium

placed on their striped bass fishery (Atlantic Striped Bass Conservation Act 1984). In 1985, Maryland imposed a self moratorium, and Virginia followed suit in 1989. These measures, along with stocking efforts (Secor and Houde 1998) appeared to adequately address the problem as the population began to rebound. The three year average of Maryland's juvenile index from 1987-1989 exceeded 8.0, a criterion needed to reopen the fishery (Richards and Rago 1999). Amendment 4 to the ASMFC Plan was adopted in 1989, and it allowed states to reopen a tightly controlled fishery on striped bass. Another caveat of the amendment was that states had to monitor their striped bass populations (ASMFC 1989). Several factors, including an improved juvenile index and improved spawning stocks led to a declaration by the ASMFC that striped bass could be considered a recovered species (ASMFC 1995). Continuing research and monitoring of striped bass populations is needed to prevent a similar situation from occurring again.

Diets of striped bass undergo an ontogenetic shift as striped bass grow. Juveniles are opportunistic feeders, and their diets are comprised of insect larvae, polychaetes, larval fish, and amphipods (Setlzer-Hamilton and Hall 1991). The location of juvenile striped bass will influence their diet, with fish in freshwater portions of the Potomac Estuary feeding on oligochaetes and insects and fish in higher salinities were feeding more on amphipods, mysids, and fish larvae (Boynton et al. 1981). Striped bass become more dependent on fish species as a prey source as they grow, and are primarily piscivorous by age-2 (Hartman and Brandt 1995).

The diets of piscivorous striped bass have been well studied (Gardinier and Hoff 1982, Hartman and Brandt 1995, Overton et al. 2000, Rudershausen et al. 2005). These fish prey upon a variety of items such as Atlantic menhaden (*Brevoortia tyrannus*), bay anchovy (*Anchoa mitchilli*), spot (*Leiostomus xanthurus*), other clupeids, and blue crab (a decapod; *Callinectes*

sapidus). Atlantic menhaden can make up a large proportion of the diet (Hartman and Brandt 1995, Overton et al. 2000).

The diets of striped bass may have undergone a shift in the Chesapeake Bay unrelated to life stage. The mean daily consumption by striped bass on menhaden in 2000 was consistently less for all age groups than observed in 1993 (Overton et al. 2000). Striped bass mean daily consumption of blue crabs was also higher in 2000 than estimated for striped bass in 1993 (Overton et al. 2000). The decreased presence of menhaden in the diet also coincided with a change in the weight-at-length and weight-at-age measurements for striped bass suggesting that striped bass were not sufficiently substituting other prey items for lost menhaden (Uphoff 2003). Based upon calculations, striped bass demand began to exceed prey supply in the late 1990s (Uphoff 2003) due to a seven fold increase in the population between 1982 and 1994 (NEFSC 1998).

Weakfish (*Cynoscion regalis*) and bluefish (*Pomatomus saltatrix*) compete with striped bass for bay anchovy, menhaden, and spot (Merriner 1975, Hartman and Brandt 1995, Parthree et al. 2006). The weakfish population was at a low level in the late 1980s until recruitment increased from a low point in 1989 and the population rebounded to high numbers in the 1990s (NESFC 2000). Bluefish (*Pomatomus saltatrix*) consume menhaden, spot, and anchovies (Hartman and Brandt 1995, Harding and Mann 2001, Scharf et al. 2004) and are therefore considered a competitor of striped bass. The biomass of this population had a historic high in 1981, and then decreased by roughly 84% over the next 14 years. Recruitment increased in 1994 and 1996 and in 1999 was the highest since 1989 (Beal et al. 2000). As striped bass were increasing during the mid to late 1990s, two other major competitors also saw their numbers increase which would lead to more pressure on the already fragile prey base.

Environmental stressors may have led to emaciated striped bass with visible lesions collected between 1997 and 2000 (Uphoff 2003). Overton et al. (2003) found an infection rate of 53% for striped bass in the Chesapeake Bay. Infected fish were defined as having granulomas originating from *Mycobacterium* spp. that were associated with organs or external lesions. This study also documented the condition of the fish; striped bass with no granulomas or external lesions had the highest Fulton condition factor (0.99) and fish with both granulomas and external lesions had the lowest condition (0.81). Studies have demonstrated that more than one species of bacteria of the genus *Mycobacterium* may be responsible for the outbreak seen in striped bass (Heckert et al. 2001, Rhodes et al. 2004). What is not clear is whether striped bass are in poor condition because of the bacterial infection, or if they are in poor condition prior to contracting the infection. It is important to understand the life history of the primary prey of striped, Atlantic menhaden, in order to make appropriate management decisions

Atlantic menhaden ecology and exploitation

Atlantic menhaden breed throughout inshore waters along the Atlantic and at around one month the young are carried by currents into various estuaries, including the Chesapeake Bay (Massmann et al. 1962). Larvae metamorphose into juveniles and concentrate in large schools throughout the summer. These fish remain in the Chesapeake Bay until fall, at which point they migrate out and overwinter along southern coastal waters (Lippson 1991).

Atlantic menhaden have been an important commercial fishery since the 1950s, and each year hundreds of thousands metric tons are harvested (NOAA 2009). Menhaden can reach ages of 10-12 (Ahrenholz 1991), however the size structure has become truncated due to overfishing with few fish obtaining ages greater than six (Ahrenholz 1987). Vaughan and Smith (1988)

reported that the fishery was beginning to rely more heavily on younger fish (pre-spawners age-2 and younger), and the trend has continued to present day. The percentage of age-3+ fish present in the catch from 2004-2008 was never more than 27%, and the bulk of the fish were age-2 (NOAA 2009). A cap on the harvest of menhaden was instituted, but the landings from 2006-2008 have fell below the cap by an average of 30%, which means the cap could be ineffective in preventing overfishing of the stock (CBEF 2009).

Diet analysis

Stomach content analysis

Historically the method for determining the diet of fish was to collect samples, sacrifice the fish, and observe what was present in the stomach (i.e. Ewers and Boesel 1935, Moffet and Hunt 1945). A method was also developed by Seaburg (1957) in which the stomach contents were "flushed" out and the fish did not need to be sacrificed. These data were usually analyzed using metrics such as frequency of occurrence, percent composition by number, or percent composition by weight (Bowen 1996). These metrics by themselves often emphasize large and possibly infrequent prey items or small and possibly numerous items (George and Hadley 1979, Hyslop 1980). Combinations of these metrics have been used in order to try and reduce the bias involved (George and Hadley 1979).

There are some inherent biases that are associated with the analysis of stomach contents (Iverson et al. 2004). The identification of prey items in the stomach is easier if the meal has just been eaten, and little digestion has taken place. However, if it has been some time since the meal was eaten, the whole prey item may no longer be intact, forcing the identification of the prey to be made from what materials are left, i.e. chitinous hard parts or scales (Bowen at al. 1993). The

rate of digestion for tissues is not homogeneous, and this could also bias the conclusion of the

predator's diet. The stomach contents of a fish are also assumed to be representative of the

forage base in the area. A voracious piscivore, e.g. bluefish, may feed randomly on encountered

prey items (Juanes et al. 1993), which may lead to a misrepresentation of what prey base is

available to a predator. Opportunistic predators like striped bass may encounter an unusual prey

item during feeding, or encounter prey not regularly consumed. Due to these factors, the

published diet of any given predator may be incorrect as the information used may not represent

the entire picture.

Alternative strategies for diet analyses

Stable isotope analysis

Stable isotopes are present in natural materials, can be measured with great precision and

isotopic concentrations change in predictable ways as elements cycle through the biosphere.

Isotopic values of carbon show modest increases with each trophic level while values of nitrogen

increase through each successive trophic level (Peterson and Fry 1987), and can lead to a

determination of trophic level (Hecky and Hesslein 1995, Vander Zanden et al. 1997).

The use of stable isotope analysis to determine origins of materials in the environment

requires that the samples are isotopically different. The fractionation of isotopes refers to the

pathway that the "heavy" and "light" isotopes take in various reactions. These pathways need to

be known for stable isotope analysis to be applied in a system (Lajtha and Michener 1994).

There may be too many sources of isotopes, i.e. prey species, to resolve the diet of a predator.

This occurs when prey isotopic signatures are not significantly different from one another. One

solution is to group prey species (Phillips et al. 2005) such as fish or krill diets of penguins

(Tierney et al. 2008) or littoral, pelagic, or profundal energy sources in a lake (Lepak et al. 2006). Another possible solution is to obtain a range of feasible contributions from each source, and then combine related sources a posteriori (Phillips et al. 2005). The combination of sources in stable isotope analysis leads to decreased taxonomic resolution, but offers a longer time frame in which to infer diets (Tierney et al. 2008).

Fatty acid analysis

Fatty acids are a class of lipids that are comprised of a carboxylic acid group and a relatively long chain of carbon and hydrogen atoms. These compounds can be saturated (no double bonds) or contain one to six double bonds between the carbons atoms and they can perform a variety of functions (Gurr and Hardwood 1991). Eicosanoids are molecules that are synthesized from 20 carbon length (C20) polyunsaturated fatty acids and are involved in blood clotting, immune responses, renal function, and neural function (Tocher 2003). Sargent et al. (1999) found that certain fatty acids obtained exclusively from the diet in salmon were responsible for the parr-smolt transformation.

Fatty acids are deposited largely unmodified from prey to predator (Iverson 1997a). For example, adult menhaden persist on a diet of larger phytoplankton (Friedland et al. 1989) which is deficient in the fatty acids C20 and C22:1, and consequently the signature of the menhaden are lacking in these two compounds (Arts et al. 2001). An organism that does not require a large supply of energy immediately can store fatty acids in molecules known as triacylglycerols. Triacylglycerols are a major class of neutral lipid in which the fatty acid is esterified to the alcohol group of the glycerol (Tocher 2003). Unused protein and carbohydrates can be converted to fats, but the process is not reversible (Gurr and Hardwood 1991). The triacylglycerols are

stored in a variety of locations including the mesenteric adipose tissue and the adipose tissue between myosepta (Henderson and Tocher 1987).

"Essential fatty acids" cannot be biosynthesized and must be obtained from the diet (Tocher 2003). They range between 12-24 carbons in length (usually even in number) and may undergo moderate changes, such as chain lengthening and desaturation (Tocher 2003) or remain completely unchanged (Budge et al. 2006). Some fatty acids are biosynthesized by the organism and also contribute to the fatty acid reservoir. The combination of these three components leads to a "fatty acid signature" (Iverson 1993) or fingerprint that is unique to that species. For example, juvenile cobia (*Rachycentron canadum*) started to exhibit a significant change in their fatty acids after 3 days of diet change, and greater than 90% of the PUFAs were conserved after an average of 12 days (Turner and Rooker 2005).

The fatty acids can be tracked as they move their way through the food web (Iverson 1997a), and diet can be estimated from the fatty acids present and their proportions with respect to other fatty acids (Iverson 2004). Fatty acids may be present in the prey in a form that is not readily available to the predator. Wax esters are a neutral lipid in which the fatty acid is esterified to a fatty alcohol group (Gurr and Hardwood 1991). The predator removes the fatty alcohol, oxidizes it to a fatty acid, and its fate will be the same as other fatty acids ingested, i.e. incorporation into triacylglycerols and phosphoglycerides (Sargent et al. 1979, Tocher 2003).

Studies have also shown that a broader temporal scale can be inferred from analyzing the fatty acids of an organism. Often, it is possible to determine diet items eaten over the past several weeks, versus the last meal (Iverson 1997b). Digestion rates and the presence of gastric enzyme resistant body parts are no longer an issue because researchers are investigating trace marker

products of food resources, and not the food resource itself to determine diet. This could lead to elimination of biases present when trying to determine diet from examination of the stomach.

Exact abundances of prey items cannot be inferred, but it is possible to estimate the proportions of various prey species in the diet. Very mobile species such as striped bass would make an ideal candidate for this technique as they feed both in coastal waters and in the Chesapeake Bay as adults. Fatty acid signature analysis may also be able to delineate between resident striped bass, or blackbacks, and those migrating upstream to spawn, or greenbacks (Gallagher et al. 1998). They key for this technique is establishing a library of prey fatty acid signatures that can be compared to striped bass tissues. Despite variation within species, relatively few fatty acids may be needed to distinguish between the prey types (Recks and Seaborn 2008). Because preferred prey of striped bass in the Chesapeake Bay has been well documented; the next step becomes determining how these molecules are incorporated by striped bass. Fatty acid analysis may overcome the deficiencies of stomach content analysis (removal of unidentifiable items from analyses and short term time frame) and stable isotope analysis (low taxonomic resolution) to provide detailed information for a longer time frame than the last meal.

Literature cited

Ahrenholz, D.W. 1991. Population biology and life history of the North American menhadens, *Brevoortia* spp. Marine Fisheries Review 53:3-19.

Ahrenholz, D.W., W.R. Nelson, and S.P. Epperly. 1987. Population and fishery characteristics of Atlantic menhaden, *Brevoortia tyrannus*. Fishery Bulletin 85:569-600.

Arts, M.T., R.G. Ackman, and B.J. Holub. 2001. "Essential fatty acids" in aquatic ecosystems: a crucial link between diet and human health and evolution. Canadian Journal of Fisheries and Aquatic Sciences 58:122-137.

ASMFC (Atlantic States Marine Fisheries Commission). 1981. Interstate fisheries management plan for the striped bass. Fisheries Management Report No. 1, Washington, D.C.

ASMFC (Atlantic States Marine Fisheries Commission). 1989. Supplement to the striped bass FMP-Amendment #4. ASMFC, Fisheries Management Report 15, Washington, D.C.

ASMFC (Atlantic States Marine Fisheries Commission). 1995. Amendment #5 to the interstate fishery management plan for Atlantic striped bass. ASMFC. Fisheries Management Report 24, Washington, D.C.

Atlantic Striped Bass Conservation Act of 1984. 16 U.S.C. § 5151-5158.

Beal, R. 2000. Review of the Atlantic States Marine Fisheries Commission fishery management plan for bluefish (*Pomatomus saltatrix*). Atlantic States Marine Fisheries Commission.

Bowen, S.H. 1996. Quantitative description of the diet. In: Fisheries Techniques (eds. Murphy, B.R. and Willis, D.W.), pp. 513-532. American Fisheries Society, Bethesda, MD.

Bowen, W.D., J.W. Lawson, and B. Beck. 1993. Seasonal and geographic variation in the species composition and size of prey consumed by grey seals (*Halichoerus grypus*) on the Scotian shelf. Canadian Journal of Fisheries and Aquatic Sciences 50:1768-1778.

Boynton, W.R., H.H. Zion, T.T. Polgar, 1981. Importance of juvenile striped bass diets in the

    Potomac estuary. Transactions of the American Fishery Society 110:56-63.

Budge, S.M., S.J. Iverson, H.N. Koopman. 2006. Studying trophic ecology in marine ecosystems

    using fatty acids: a primer on analysis and interpretation. Marine Mammal Science

    22:759-801.

CBEF (Chesapeake Bay Ecological Foundation). 2009. Ecological depletion of Atlantic

    menhaden effects on Atlantic coast striped bass: First year-round diet study of large

    Chesapeake Bay striped bass.

Chapoton, R.B. and J. E. Sykes. 1961. Atlantic coast migration of large striped bass as evidenced

    by fisheries and tagging. Transactions of the American Fisheries Society 90:13-20.

Ewers, L.A. and M.W. Boesel. 1935. The food of some buckeye lake fishes. Transactions of the

    American Fisheries Society 65:57-70.

Friedland, K.D., D.W. Ahrenholz, and J.F. Guthrie. 1989. Influence of plankton on distribution

    patterns of filter-feeder *Brevoortia tyrannus* (Pisces: Clupeidae). Marine Ecology

    Progress Series 54:1-11.

Gallagher, M.L., L. Paramore, D. Alves, and R.A. Rulifson. 1998. Comparison of phospholipid

    and fatty acid composition of wild and cultured striped bass eggs. Journal of Fish Biology

    52:1218-1228.

Gardinier, M.N. and T.B. Hoff. 1982. Diet of striped bass in the Hudson River estuary. New

    York Fish and Game Journal 29:152-165.

George, E.L. and W.F. Hadley. 1979. Food habitat partitioning between rock bass (*Ambloplites

    rupestris*) and smallmouth bass (*Micropterus dolomieui*) young of the year. Transactions

    of the American Fisheries Society 108:253-261.

Gurr, M.I. and J.L. Hardwood. 1991. Lipid biochemistry: an introduction. 4th ed. Chapman and Hall. London, England.

Harding, J.M. and R. Mann. 2001. Diet and habitat use by bluefish, *Pomatomus saltatrix*, in a Chesapeake Bay estuary. Environmental Biology of Fishes 60:401-409.

Hartman, K.J. and S.B. Brandt. 1995. Trophic resource partitioning, diets, and growth of sympatric estuarine predators. Transactions of the American Fishery Society 124:520-537.

Heckert, R.A., S. Elankumaran, A. Milani, and A. Baya. 2001. Detection of a new *Mycobacterium* species in wild striped bass in the Chesapeake Bay. Journal of Clinical Microbiology 39:71-715.

Hecky, R.E. and R.H. Hesslein. 1995. Contributions of benthic algae to lake webs as revealed by stable isotope analysis. Journal of the North American Benthological Society 14:631-653.

Henderson, R.J. and D.R. Tocher. The lipid composition and biochemistry of freshwater fish. Progress in Lipid research 26:281-347.

Hyslop, E.J. 1980. Stomach contents analysis – a review of methods and their application. Journal of fish Biology 17:411-429.

Iverson, S.J. 1993. Milk secretion in marine mammals in relation to foraging: can milk fatty acids predict diet? Symposium of the Zoological Society of London 66:263-291.

Iverson, S.J. C. Field, W.D. Bowen, and W. Blanchard. 2004. Quantitative fatty acid signature analysis: a new method of estimating predator diets. Ecological Monographs 74:211-235.

Iverson, S.J., K.J. Frost, and L.F. Lowry. 1997a. Fatty acid signatures reveal fine scale structure of foraging distribution of harbor seals and their prey in Prince William Sound, Alaska. Marine Ecology Progress Series 151:255-271.

Iveson, S.J., J.P.Y. Arnold, and I.L. Boyd. 1997b. Milk fatty acid signatures indicate both major and minor shifts in the diet of lactating Antarctic fur seals. Canadian Journal of Zoology 75:1881-97.

Juanes, F., R.E. Marks, K.A. McKown, and D.O. Conover. 1993. Predation by age-o bluefish on age-0 anadromous fishes in Hudson River estuary. Transactions of the American Fisheries Society 122:348-356.

Kohlenstein, L.C. 1981. On the proportion of the Chesapeake Bay stock of striped bass that migrates into the coastal fishery. Transactions of the American Fishery Society 110:168-179.

Lajtha, K. and R.H. Michener. 1994. Stable isotopes in ecology and environmental science. Blackwell Scientific Publications. London, England.

Lepak, J.M., C.E. Kraft, and B.C. Weidel. 2006. Rapid food web recovery in response to removal of an introduced apex predator. Canadian Journal of Fisheries and Aquatic Sciences 63:569-575.

Lippson, R.L. 1991. Atlantic menhaden (*Brevoortia tyrannus*). in Habitat requirements for Chesapeake Bay living resources. Funderbunk, S.L., S.J. Jordan, J.A. Mihursky, D. Riley, eds. Chesapeake Research Consortium, Inc. Solomons, MD.

Mansueti, R.J. 1961. Age, growth, and movements of the striped bass, *Roccus saxitilis*, taken in size selective fishing gear in Maryland. Chesapeake Science 2:9-36.

Maryland DNR (Department of Natural Resources). 1987. Third annual status report on striped bass 1987. Tidewater Administration, Fisheries Division, Annapolis, MD.

Massmann, W.H., J.J. Norcross, and E.B. Joseph. 1962. Atlantic menhaden larvae in Virginia coastal waters. Chesapeake Science 3:42-45.

Merriner, J.V. 1975. Food habits of the weakfish, *Cynoscion regalis*, in North Carolina waters. Chesapeake Science 16:74-76.

Moffet, J.W. and B. P. Hunt. 1945. Winter feeding of bluegills, *Lepomis macrochirus* Rafinesque, and yellow perch, *Perca flavescens* (Mitchill) in Cedar Lake, Washtenaw County, Michigan. Transactions of the American Fisheries Society 73:231-242.

NEFSC (Northeast Fisheries Science Center). 1998. Report of the 26th regional stock assessment workshop (26th SAW): Stock assessment review committee (SARC) consensus summary of assessments. NEFSC Reference Document 98-03, Woods Hole, MA, USA, pp. 142-160.

NEFSC (Northeast Fisheries Science Center). 2000. A report of the 30th Northeast regional stock assessment workshop (30th SAW): Public review workshop. Woods Hole, MA, USA, 58 pgs.

NOAA (National Oceanic and Atmospheric Administration). 2009. Forecast for the 2009 Gulf and Atlantic menhaden purse-seine fisheries and review of the 2008 fishing season. Sustainable Fisheries Branch, NMFS Beaufort, NC.

Overton, A.S., E.B. May, J. Griffin, F.J. Margraf. 2000. A bioenergetics approach for determining the effect of increased striped bass population on its prey and health in the Chesapeake Bay. Report to: Maryland Department of Natural Resources. Maryland Cooperative Fish and Wildlife Research Unit, University of Maryland Eastern Shore, Princess Anne, MD.

Parthree, D.J., C.F. Bonzek, R.J. Latour. Final report: Chesapeake Bay trophic interactions laboratory series. Virginia Institute of Marine Science, Gloucester Point, VA.

Peterson, B.J. and B. Fry. 1987. Stable isotopes in ecosystem studies. Annual Review of Ecology and Systematics 18:293-320.

Phillips, D.L., S.D. Newsome, J.W. Gregg. 2005. Combining sources in stable isotope mixing models: alternative methods. Oecologia 144:520-527.

Recks, M.J. and G.T. Seaborn. 2008. Variation in fatty acid composition among nine forage species from a southeastern US estuarine and nearshore coastal ecosystem. Fish Physiology and Biochemistry 34:275-287.

Rhodes, M.W., H. Kator, I. Kaattari, D. Gauthier, W. Vogelbein, C.A. Ottinger. 2004. Isolation and characterization of mycobacteria from striped bass *Morone saxatilis* from the Chesapeake Bay. Diseases of Aquatic Organisms 61:41-51.

Richards, R.A. and P.J. Rago. 1999. A case history of effective fishery management: Chesapeake Bay striped bass. Transactions of the American Fisheries Society 19:356-375.

Rudershausen, P.J., J.E. Tuomikoski, and J.A. Buckel. 2005. Prey selectivity and diet of striped bass in western Albemarle Sound, North Carolina. Transactions of the American Society 134:1059-1074.

Rugolo, L.J. and P.W. Jones. 1989. A recruitment based interseason control model for Chesapeake Bay striped bass. Maryland Department of Natural Resources, Annapolis, MD.

Sargent, J., G. Bell, L. McEvoy, D. Tocher, A. Estevez. 1999. Recent developments in the essential fatty acid nutrition of fish. Aquaculture 177:191-199.

Sargent, J.R., R. McIntosh, A. Bauermeister, and J.H.S. Blaxter. 1979. Assimilation of the wax esters of marine zooplankton by herring (*Clupea harengus*) and rainbow trout (*Salmo gairdnerii*). Marine Biology 51:203-207.

Scharf, F.S., J.P. Manderson, M.C. Fabrizio, J.P. Pessutti, J.E. Rosendale, R.J. Chant, A.J. Bejda. 2004. Seasonal and inter-annual patterns of distribution and diet of bluefish within a middle Atlantic bight estuary in relation to abiotic and biotic factors. Estuaries 27:426-436.

Seaburg, K.G. 1957. A stomach sampler for live fish. The Progressive Fish-Culturist 19:137-139.

Secor, D.H. and E.D. Houde. 1998. Use of larval stocking in restoration of Chesapeake Bay striped bass. ICES Journal of Marine Science 55:228-239.

Setzler-Hamilton, E.M. and L. Hall Jr. 1991. In Habitat requirements for Chesapeake Bay living resources. Eds. Funderburk, S.L., S.J. Jordan, J.A. Mihursky, D. Riley. Chesapeake Research Consortium. Solomons, MD.

Tierney, M., P.D. Nichols, K.E. Wheatley, M.A. Hindell. 2008. Blood fatty acids indicate inter- and intra-annual variation in the diet of Adelie penguins: comparison with stomach content and stable isotope analysis. Journal of Experimental Marine Biology and Ecology 367:65-74.

Tocher, D.R. 2003. Metabolism and functions of lipids and fatty acids in teleost fish. Reviews in Fisheries Science 11:107-184.

Turner, J.P. and J.R. Rooker. 2005. Effect of dietary fatty acids on the body tissues of larval and juvenile cobia and their prey. Journal of experimental Marine Biology and Ecology 322:13-27.

Uphoff Jr., J.H. 2003. Predator-prey analysis of striped bass and Atlantic menhaden in upper Chesapeake Bay. Fisheries Management and Ecology 10:313-322.

Vander Zanden, M.J., G. Cabana, J.B. Rasmussen. 1997. Comparing trophic position of freshwater fish calculated using stable nitrogen isotope ratios (δ^{15}N) and literature dietary data. Canadian Journal of Fisheries and Aquatic Sciences 54:1142-1158.

Vaughan, D.S. and J.W. Smith. 1988. Stock assessment of the Atlantic menhaden, *Brevoortia tyrannus*, fishery. NOAA Technical Report NMFS 63. 26p.

Chapter 2 - Spatial and temporal patterns in fatty acids signatures of

striped bass prey taxa[1]

Abstract

The Chesapeake Bay striped bass *Morone saxatilis* population was declared recovered from overfishing in the late 1990's. However, the new burgeoning population has exerted a large predation pressure on other species that also support important commercial fisheries. In the context of Ecosystem Based Fisheries Management, accurate estimates of diet are needed to determine: 1) estimates of relative prey consumption and 2) the occurrence of dietary shifts. Fatty acid analysis is an alternative to traditional diet estimators which could possibly remove inherent biases related to stomach analyses and more accurately estimate feeding history. In this study, we created a library of fatty acids and lipid values for common striped bass prey items to determine the effects of season, location, and life stage. We targeted four commonly consumed striped bass prey in the Chesapeake Bay: Atlantic menhaden *Brevoortia tyrannus*, bay anchovy *Anchoa mitchilli*, spot *Leiostomus xanthurus*, and blue crab *Callinectes sapidus*. The prey samples were collected in spring, summer, and fall in multiple locations throughout the Bay in order to capture the seasonal and spatial variation in fatty acid signatures contained within each prey group. Results indicate that pelagic feeders, e.g. menhaden and anchovy, differ in their fatty acid signature compared to demersal feeders, e.g. spot and blue crab, when separated by classification and regression trees (CART) and discriminant function analysis (DFA). Spot and blue crab generally have lower lipid content, making them possibly less nutritious alternatives for striped bass. Menhaden and anchovy have similar signatures, and these signatures change very little with respect to season and location while blue crab and spot exhibit a strong seasonal signature. Blue crab and spot have similar signatures within a season. Menhaden exhibit differences in abundances of certain fatty acids when juveniles are compared to adults. Our

19

results suggest fatty acid signatures of prey taxa vary by season and location, and these

parameters need to be incorporated into food web modeling.

Introduction

Striped bass *Morone saxatilis* are an abundant piscivore that have supported a

commercial and recreational fishery since colonial times (Chesapeake Bay Fisheries Ecosystem

Advisory Panel 2006). The fishing pressure along the entirety of its range eventually manifested

itself in a population decline observed in the 1970s and 1980s (Boreman and Austin 1985).

Through legislation and stocking efforts (Secor and Houde 1998), the population began to

rebound. Striped bass numbers peaked in 2004 with an estimated abundance of 70.8 million fish

up from a low in 1982 of 8.8 million fish.

A recovered striped bass population could lead to increased prey demand (Hartman

2003). Altering one aspect of the food web will have ramifications on all other species that are

connected. Atlantic menhaden *Brevoortia tyrannus* not only constitute a large proportion of

striped bass diets, they also contribute to a reduction fishery along the Atlantic coast. These fish

can make up >50% of the diets of age-1+ striped bass, depending on the month (Hartman and

Brandt 1995). The Atlantic menhaden fishery has started to rely more heavily on younger fish,

including pre-spawners (age-2 and below, Vaughn and Smith 1988). Removal of key striped bass

prey items by humans has led to a shortage for striped bass (Uphoff 2003). Griffin and Margraf

(2003) found that large striped bass in the 1990s relied more on bay anchovy and age-0 clupeids

compared to the 1950s, when Atlantic menhaden was the primary prey for large striped bass.

This lack of menhaden may have led to a higher mean daily consumption of blue crabs in

Chesapeake Bay striped bass from 1993 to 2000 (Overton et al. 2000). This pattern also occurred

in the Hudson River, where it was estimated that striped bass consumption exceeded alosid supply (Hartman 2003). An increase in consumption beyond what the preferred prey can support would necessitate striped bass consuming other species. A diet shift may be indicative of an insufficient prey base to support the striped bass population.

Interactions among species are complex, and are not fully accounted for in single-species approaches to management. The concept of ecosystem based fisheries management has become increasingly important. This framework tries to incorporate species interactions like food web dynamics into the system's model rather than trying to manage each species separately (Brodziak and Link 2002). Links among species and energy flow are examples of reference points that have been postulated as aspects that offer possibilities for modeling (Link 2005). In order to measure these reference points, accurate estimates of feeding history are needed.

Traditional analysis of diet via stomach content analysis is relatively easy and inexpensive (Hyslop 1980). Stomach flushing (Seaburg 1957) involves injecting water into the stomach of the fish to retrieve diet items. While this method does provide information about the diet, gastric evacuation rates of the prey are not accounted for, and may bias the results (Hyslop 1980, Iverson et al. 2004). Identification of prey items in the stomach is easier if the meal has just been eaten, and little digestion has taken place. Once the digestive processes have been initiated, correct identification of prey becomes more difficult. Prey items will remain in the gut for a shorter duration at elevated temperatures (Elliot 1972, He and Wurtsbaugh 1993) and prey species with chitinous material that resists digestion will be more readily enumerated (Gannon 1976). Larger prey size, especially whole fish, will affect the rate of digestion due to the small surface area to volume ratio (He and Wurtsbaugh 1993). Therefore, stomach content analysis

becomes a snapshot of instantaneous foraging unless corrections for evacuation rates are included.

An alternative to analyzing stomach contents is fatty acid analysis. Fatty acids are deposited in predator tissues with little modification (Iverson 1997a). An organism that does not require a large supply of energy immediately can store fatty acids in molecules known as acylglycerols (Gurr and Hardwood 1991). The fatty acids can be tracked as they are cycled through the food web (Iverson 1997a, Beck et al. 2005, Turner and Rooker 2005, Beck et al. 2007), and diet composition can be estimated from the fatty acids present and their proportions with respect to other fatty acids (Iverson et al. 2004).

Studies have also shown that a broader temporal scale can be inferred from analyzing the fatty acids of an organism. Often, it is possible to determine diet items eaten over the past several weeks, versus the last meal (Iverson 1997b). Digestion rates and the presence of gastric enzyme resistant body parts are no longer an issue because researchers are investigating end products of food resources, and not the food resource itself to determine diet. This could lead to elimination of biases when trying to determine diet from examination of the stomach.

Fatty acid analysis does have problems that need to be addressed for the technique to be a valuable resource. For one, exact abundances of prey items cannot be inferred, but it is possible to estimate the proportions of various prey species in the diet using fatty acid specific statistical techniques (quantitative fatty acid signature analysis; Iverson et al. 2004). Another downside of fatty acid analysis is that no fatty acid is completely unique to a species (Dalsgaard et al. 2003) and several fatty acids may be needed to distinguish between species (Recks and Seaborn 2008). Therefore, it is difficult to assign fatty acid trophic markers (FATMs) for food webs. A way to alleviate this problem is to design laboratory experiments that examine the process of lipid

integration by a predator for a specific group of prey, e.g. striped bass in the Chesapeake Bay (see Kirsch et al. 1998).

To address the issue of clarifying the diet of striped bass in the Chesapeake Bay, FATMs must be established specifically for this system. In order to complete this task, the fatty acid signature of prey striped bass encounter must be analyzed for their fatty acid signatures throughout the bay, and throughout multiple seasons to determine how fatty acids and lipid levels change in time and space.

The specific goals of this project were to determine 1) if fatty acid signatures and lipid levels of common striped bass prey were different from one another within a season and region; and 2) if there was intra-species variation in prey taxa fatty acid signature and lipid levels based upon season or region.

Materials and Methods

Sample collection

In order to assess spatial and temporal variation, Atlantic menhaden and bay anchovy *Anchoa mitchilli* were collected during spring (2008), summer (2008), and fall (2009) in regions in the Chesapeake Bay with differing salinities (2-8 parts per thousand [ppt] in the upper bay, and 10-16 ppt in the lower bay, Fig. 1). Spot *Leiostomus xanthurus* and blue crab *Callinectes sapidus* were collected during summer and fall (2008) in the upper bay (Table 1). Sufficient numbers of spot and blue crab were not collected from the lower bay in any season or from the upper bay in spring to perform fatty acid analysis. Samples from the upper Chesapeake were collected by trawl net and beach seines in collaboration with the National Oceanic and Atmospheric Administration (NOAA) lab in Oxford, Maryland. Adult menhaden were obtained

from a pound net fisherman in Cambridge, Maryland. The large size of adult menhaden necessitated removal of heads and caudal fins to homogenize the tissue. Therefore, the values obtained for this sample group reflect analysis on the body minus the head and caudal fin. All other individuals analyzed were whole prey species. Species in the lower Chesapeake were collected by the Virginia Institute of Marine Science (VIMS) with bottom trawls (Tuckey and Fabrizio 2011). Once collected, species collected from a location were separately vacuum sealed and frozen at -20°C.

Sample preparation

Prey samples were thawed at the time of analysis, weighed to the nearest 0.1 gram, and total length was measured to the nearest millimeter. Each sample consisted of a homogenate of three to five grams of fish or blue crab and, in the case of small individuals several organisms were combined until the weight range was reached. A sample group refers to a collection of individual samples of a single taxon, collected from a single location in one season.

Small organisms were ground whole with a chemical drying agent, diatomaceous earth (Dionex, Sunnyvale, CA). Larger prey items were homogenized, and a three to five gram sample was ground with the drying agent. Lipid was extracted with methylene chloride/methanol (3:1; v:v) under nitrogen using an Accelerated Solvent Extractor (ASE 300, Dionex, Sunnyvale, CA) according to Folch et al. (1957). Samples were extracted at 100°C and 1500 psi (USEPA 2007). Three samples of tissue were extracted at both 100° and 120°C to compare the yield in total lipid extracted (Schafer 1998) and create a correction factor.

Back extraction with 0.88% potassium chloride solution in deionized water was performed to remove non-lipid materials from extracts according to Folch et al. (1957). The extraction process can be repeated for a sample to maximize the yield, and a comparison between

one and two extractions was performed for two sample groups (spring anchovy and menhaden from the upper bay). The lipid levels obtained in the second extraction were used to create a corrected lipid content for all other samples which were extracted only once. This step also allowed verification that the correct fatty acid signature can be obtained without extracting all lipids. Total lipid extracted was determined gravimetrically for each sample and reported as a percent of wet weight.

Extracted lipid species containing fatty acids as various derivatives were transesterified using a solution of sulfuric acid and methanol. In this process, aclyglycerols are broken apart to their constituents: glycerol and fatty acids methyl esters (FAMEs). Free fatty acids present in the lipid mixture are also converted to FAMEs (Christie 2003). Sample extracts were cleaned with silica gel and anhydrous sodium sulfate to remove polar components and water, which may degrade the performance of the gas chromatograph column. The silica column was conditioned with hexane and the sample was added along with hexane and diethyl ether, 95:5, v/v (Christie 2003). The mixture was concentrated to 5 ml and diluted to a final total lipid concentration of 100 µg/ml. Finished samples were run on a gas chromatograph (Agilent 6890N Network GC System, Santa Clara, CA) / mass spectrometer (GC/MS, Agilent, 5973 Network Mass selective detector, Santa Clara, CA) in select ion monitoring mode.

The GC/MS was used to identify and quantify fatty acids methyl esters in each sample. Fatty acid methyl esters were injected into a fused silica capillary column, DB-23 60m length X 0.250mm OD X 0.25µm film thickness (Agilent, Santa Clara, CA) using an autosampler (Agilent 7683 Series Auto Sampler) with hydrogen as a carrier gas. The temperature program was as follows: hold at an initial temperature of 50°C for 2 min, hold for 1 min at 150°C after ramping at 20°C·min^{-1}, ramp to 215°C at 1.25^{o}C·min^{-1}. The fatty acid retention times were compared to

known standards from Nu-Chek Prep (GLC MIX 85, 411, 642, 632, Elysian, MN). These four standard mixtures were combined along with stearidonic acid methyl ester (C18:4n3, Cayman Chemical, Ann Arbor, MI) to make a total FAME standard solution of 12.25 μg/ml. Matreya 25 FAME mixture (Pleasant Gap, PA) was used to obtain retention times for C20:1n7. Restek cis/trans FAME mixture (Bellefonte, PA) was used to obtain retention times for C18:1n9 trans and C18:2n6 trans. These standards and an internal standard of C13:0 FAME (100 μg/ml) were added to quantify the amount of each fatty acid methyl ester present. The GC/MS tracks the select ion signature of each compound and was used to verify the identity of each peak. There were peaks which consisted of more than one fatty acid and it was impossible to determine which fatty acid was creating the peaks. Therefore, the term fatty acid "complex" was assigned to this peak. The complex refers to which fatty acid(s) could be responsible for the peak, e.g. C22:5n3 and C22:6n3.

Quality control

Silver ion chromatography

An aliquot of solution from several sample groups was analyzed using silver ion chromatography and compared to the Resek cis/trans FAME standard (FAMES: C18:1n9 trans, C18:2n6 cis, C18:1n9 cis, C18:1n12 cis, C18:1n12 trans, C18:0, C18:1n7 trans, C18:1n7 cis) to verify the elution order of cis and trans monomers (Christie 1989). The step was performed on fatty acid methyl esters. The aliquot was washed with seven different solutions; three containing hexane and acetone (96:4, 90:10, and 25:75 by volume), and four solutions containing acetone and acetonitrile (95:5, 93:7, 88:12, and 85:15 by volume). These solutions allow different monomers to be eluted separately, and the samples are run on the GC/MS.

Fatty acid precursors

We used thin layer chromatography (TLC) to determine if fatty acid precursors (plasmalogens and wax esters) were present in the prey species (Harding et al. 1975). Silica coated glass plates (Whatman, Piscataway, NJ) were soaked in toluene and allowed to dry overnight. An aliquot of 200 μl was pipetted onto the plates. Toluene was absorbed onto the plates until the top of the sample aliquot was reached. Plates were developed in toluene and allowed to dry. Bands were sprayed with 2-7-dichloroflourescin dye to identify bands of lipid classes. Each band was scraped separately, dissolved in methylene chloride, and shaken for one hour. Samples were moved to a clean test tube, oxidized using Jones's reagent, transesterified, and run on the GC/MS (Touchstone 1995).

Samples were oxidized using Jones's reagent, a solution of chromium trioxide in dilute sulfuric acid and acetone (Budge and Iverson 2003). The resulting solution was transesterified as described above, and run on the GC/MS along with the portion that did not undergo oxidation. A standard of the compound C16:0 dimethyl acetal (DMA) was obtained and run as the only fatty acid precursor standard on the GC/MS. The ion signature of C16:0 DMA was used as the basis to indentify other dimethyl acetals.

Instrument tune and maintenance

The Autotune function was performed on the GC/MS prior to every batch run (20-26 samples). This function measures the relative abundance of three mass ions, 69.00, 219.00, and 502.00. Once the relative abundance of mass ion 219.00 dropped below 90.00, the mass spectrometer source was cleaned. Also prior to each batch run, the injector septum was changed. The column, injector inlet, and seal were changed as needed during the course of this experiment. After each column change, the column was allowed to condition before any samples

were run. Standards were run through the instrument several times to ensure the correct retention times were being recorded.

Identification of cetoleic acid

Analyses and identification of cetoleic acid methyl ester and gadoleic acid methyl ester in herring liver were performed by Ashok Deshpande and Bruce Dockum of NOAA Fisheries, Sandy Hook Laboratory in collaboration with Sue Budge of Dalhousie University. Herring are known to contain the compound C22:1n11 (cetoleic acid), therefore, a sample of herring liver was obtained from a NOAA-NEFSC Groundfish Survey during the course of this experiment and analyzed. Cetoleic acid is an important fatty acid in marine systems (Cooper et al. 2006), and therefore this sample was obtained to determine if this fatty acid was prevalent in the Chesapeake Bay food web.

Statistical analyses

We used an *a priori* model building strategy (generalized linear model in program R [R Development Core Team 2010]) to determine the effects of season, species, and weight of the sample on the lipid content (as percent of wet weight) of each sample. Bay location was not included in the model because spot and blue crab were only collected in the upper bay. Weight was deemed the most important variable as lipid levels will vary with the size of the prey species. Species was also determined to be an important variable as different organisms will have varying levels of lipid. These two variables were included in most of the models. Season was also included because species will have different lipid levels throughout the year. Due to the sample size of prey species and the necessity to include second- and third-order interactions, not all combinations of models were included in the analysis. Models included: 1) a global of all three predictor variables and all second-order and third-order interaction terms, 2) predictor

variables and a single second-order interaction term, 3) a single predictor variable, and 4) a null model that included no predictor variables (Table 2). The Akaike information criterion for small sample size (AIC_C; Burnham and Anderson 2002) was used to select the best model. A plot of residuals demonstrated that the data violated the assumption of normality. Both the continuous variables weight and lipid content were natural log transformed which corrected the violation of normality. All model building and selection was performed in R (R Development Core Team 2010).

Differences in fatty acid signature were analyzed with different multivariate techniques or methods to determine which provided the best separation among groups. The first multivariate technique used was classification and regression trees (CART). CART partitions the data recursively into groups that are increasingly homogenous. The end result is a dichotomous key that allows the researcher to follow the variables responsible for the group structure (McCune and Grace 2002). The stopping point is determined by either reaching a minimum deviance of a final node or the minimum number of observations has been met (Iverson et al. 2002). Due to size restrictions, the dataset could not be separated into a learning and predictive subset. Instead, the data was separated into 10 equal parts within R (R Development Core Team 2010), and the model was built with 90% of the data, and the remaining 10% became the test set. This cross validation technique was run 50 times in R to produce the error and misclassification rate. Only, fatty acids that had a mean value of 0.4% or higher of the total fatty acids identified were used in order to remove any observations that may be erroneous. Sample groups that grouped close together at the bottom of the dendrogram were assumed to have similar fatty acid signatures.

The second technique, discriminant function analysis (DFA), maximizes among group variation compared to within group variation (McCune and Grace 2002). Wilk's lambda (λ) was

used to indicate the power of the analysis, with smaller values indicating greater success. The analysis provided a classification matrix, and the predicted group membership provided a basis for the misclassification rate of the technique. The variables used in DFA must be 1 less than the number of organisms in a sample group, therefore, DFA was performed using the 5 fatty acids that exhibited the highest amount of variation, and was also performed using 5 composite metrics: sum of saturated fatty acids, sum of monounsaturated fatty acids, sum of omega 6 fatty acids, sum of omega 3 fatty acids, and the sum of all co-eluted compounds. The mean of the ordination scores was calculated for each sample group, and was plotted for each DFA. This technique was then compared to CART to determine which method provided the better classification scheme for the sample groups in this study.

The difference in mass percent of fatty acids was compared among adult menhaden and two groups of juveniles. Patterns in fatty acid abundance based upon fish total length were examined using regression. All statistical tests were performed using the R statistical package (R Development Core Team 2010). The ANOVA performed on fatty acid abundances between menhaden juveniles and adults had a Bonferonni correction applied to the alpha level for the four group comparisons ($\alpha = 0.0125$).

Results

Method optimization

Silver ion chromatography

The first fraction consisted of saturated fatty acids (C18:0) and trans monoenes (C18:1n12t, C18:1n9t, and C18:1n7t); the second fraction consisted of cis monoenes (C18:1n

12c, C18:1n9c, and C18:1n7c); and the third fraction consisted of cis-cis dienes (C18:2n6c). The retention times for cis and trans monomers were obtained and used for peak identification.

Fatty acid precursors

Jones's oxidation indicated the fatty acid precursor C16:0 DMA was contributing 0.01 – 1.26% of the total fatty acid signature and was included in all analyses for fatty acids. Values for C17:0 DMA and C18:0 DMA were never higher than 0.05% and were not included. The attempt to use TLC was unsuccessful in this experiment. Chromatograms of the transesterified bands revealed no peaks.

Cetoleic and gadoleic acid

Identification of cetoleic acid FAME (C22:1n11) was achieved using herring liver samples. The retention time was compared to prey species run in this experiment. Cetoleic acid was identified in three sample groups: bay anchovy from the lower Chesapeake in spring (average 0.37% total mass) and fall (average 0.25% total mass), and from menhaden collected from the Choptank River in summer (average 1.73% total mass). Cetoleic acid was also identified in individuals from other samples groups, but the average for the group was less than 0.05% total mass. Because identification of cetoleic acid was based the retention time from one herring liver, and was not run with each sample group, this compound was not included in the analysis.

Gadoleic acid (C20:1n11) is very similar in structure to C20:1n12, a compound included in our Nu-Chek standard solution. Peaks were identified as C20:1n12, which may be of minimal importance for marine organisms. However, no commercial standard is available for gadoleic acid and the peak is labeled as both compounds (C20:1n11/12, Budge, personal communication).

Lipid correction

Corrections were applied to the total lipid data to approximate the lipid values as closely as possible. The second extraction of a sample at $100^{\circ}C$ revealed a recovery of approximately 14.7% of the total lipid recovered in the first extraction (g lipid per g fish; n = 16). All samples extracted were corrected based upon this amount. Samples run on the ASE at temperatures of $100^{\circ}C$ and $120^{\circ}C$ yielded an average of 16.74% more lipid when extracted at $120^{\circ}C$ (g lipid/g of fish; n = 3). All prey samples were corrected to this amount because samples were run at $100^{\circ}C$.

Lipid content

The average lipid content ranged from 1.18% – 15.10% for all sample groups tested (Table 1). Blue crab had the lowest average lipid content of any species (1.18 – 1.49%), followed by spot (4.49 – 6.05%), Atlantic menhaden (4.95 – 12.46%), and bay anchovy (5.89 – 15.10%). The best model to predict wet weight lipid content included the predictor variables of species, season, and sample weight, along with the interaction term between species and sample weight. This model had a ΔAIC_c value of 0.71 and had 11 parameters (Table 2). Two other models had low ΔAIC_c values of 3.19 and 3.86. While these models had low Akaike weights (0.14 and 0.10) they still were able to explain some of the variation. Based upon parsimony and the graphical output, which indicated that some interaction was occurring in the model (Fig. 2), the model described above was chosen as the most appropriate. Parameter estimates for the three models with the highest weight are provided in Tables 3 – 5.

All three predictor variables were important for determining the lipid content. Anchovy and blue crab both decreased in lipid percent as the weight of the sample increased, regardless of the season. Menhaden decreased in lipid percent as weight increased in the spring, showed no

relationship in summer, and increased in lipid percent as sample weight increased in the fall. Spot showed no relationship in summer, and decreased in lipid as weight increased in the fall.

Fatty acids

A total of 36 fatty acids and fatty acid complexes (peaks consisting of 2 indistinguishable fatty acids) were identified and quantified from the sample groups. The most abundant fatty acids present across all species were (in elution order on a DB-23 column): C14:0, C15:0, C16:0, C16:1n7, C17:0, C18:0, C18:1n9, C18:1n7, C18:2n6, C20:4n6, C20:5n3, the complex C22:5n3 + C22:6n3, and C24:1n9 (Figs. 3 and 4). The sums of their concentrations ranged from 80-93% of total fatty acids present. Some fatty acids had higher relative concentrations in certain prey species. Stearidonic acid (C18:4n3) was higher in menhaden and bay anchovy than demersal species. Spot and blue crab had higher abundances of C20:1n11/12, C20:1n9, and C20:1n7 compared to anchovy and menhaden.

Among species variation in fatty acid signatures

CART analysis was able to correctly classify 92.8% of the individual samples (refers to a sample of a single prey taxa or a composite of prey taxa to reach the weight range of three to five grams) in 17 sample groups (n = 142 of 153, Table 6). A sample group refers to a collection of individual samples of a single taxon, collected from a single location in one season. Both sample groups of blue crab had no misclassifications (0%). The fall group of spot had no misclassifications while the summer group had only one misclassification (5.9%). Six anchovy sample groups had a maximum misclassification of one within a sample group, and overall three misclassifications were made (5.5%). Menhaden were more difficult to classify. A total of seven menhaden specimens were misclassified (11.7%). Two menhaden sample groups had two individual samples misclassified, and one group had three.

Blue crab and spot were separated from anchovy and menhaden in terms of fatty acid signature (Fig. 5). There was a seasonal aspect to the grouping, with blue crab and spot from the same season grouped together. All samples of blue crab and spot were collected from the upper Bay so the influence of Bay location could not be evaluated. A lower abundance of C20:4n6 was indicative of fall blue crab and spot (Fig. 5: O and Q) while summer blue crab and spot (Fig. 5: N and P) had a higher abundance of C20:1n7. Anchovy and menhaden sample groups were separated from blue crab and spot, but overlapped with each other on the CART dendrogram. Anchovy displayed some signs of being grouped by season and location, e.g. samples collected in the summer and fall of the lower bay (Fig. 5: E and C) and samples collected in spring from the upper and lower bay (Fig. 5: A and B) were similar in fatty acid signature. Menhaden adults collected in the summer and menhaden juveniles collected in the fall from the upper bay were also similar in fatty acid signature (Fig. 5: L and M). The patterns structuring menhaden signatures seem limited to this coupling. Overall, there appears to be some separation of menhaden and anchovy signal, but there is also overlap in their fatty acid signatures.

The DFA using the five fatty acids with highest variation and five fatty acid metrics had lower classification rates when compared to CART (79.7 and 73.9% respectively, Table 3). Both methods were able to correctly classify spot, but had much higher misclassification rates for anchovy, menhaden, and blue crab.

Even with the higher misclassification rates, there was still separation among blue crab/spot and anchovy/menhaden (Figs. 6 and 7). The DFA using five fatty acids had better separation, and demonstrated that spot and blue crab are higher in C18:1n9 when compared to menhaden and anchovy, and have a higher abundance of C14:0 and C16:1n7 than anchovy (Fig. 6). There was some overlap of menhaden and anchovy, but separation between the two species

was evident. Anchovy have a higher proportion of C16:0 and the complex of C22:5n3 + C22:6n3, while menhaden have a higher proportion of C14:0 and C16:1n7.

The DFA that used fatty acid metrics did not exhibit the same separation seen with CART or DFA for the five fatty acids (Fig. 7). Blue crab and spot were separated from anchovy and menhaden by having higher abundances of monounsaturated fatty acids, omega-6, and omega-3 fatty acids. Menhaden and anchovy overlapped and no discernible patterns were evident.

Ontogenetic shifts in fatty acid signatures

Several of the 36 fatty acids analyzed exhibited differences when compared to the size of the menhaden (Fig. 8). There were significant differences in the mass percent of certain fatty acids between adult and juvenile menhaden from the upper bay in summer. Adult menhaden had significantly higher levels of C18:1n7 (ANOVA, $F_{2,23} = 17.95$, $p < 0.001$) and C20:5n3 ($F_{2,23} = 56.01$, $p < 0.001$). Juvenile menhaden had significantly higher levels of C16:0 ($F_{2,23} = 18.01$, $p < 0.001$) and C18:1n9 ($F_{2,23} = 36.47$, $p < 0.001$). The two sample groups of juvenile menhaden were not significantly different from one another for any of the four fatty acids listed above.

<center>**Discussion**</center>

Fatty acid analysis holds promise for accurately determining forage history in striped bass. The classification rate ranged between 73.9 and 92.8 depending on the technique. Both CART and DFA of 5 fatty acids and 5 fatty acid metrics were able to show patterns among the species groups. Further, we showed that there are discrete differences among the total lipid content of prey species encountered by striped bass which vary by season and ontogeny. This work is extremely important in the context of Ecosystem Based Fisheries Management and

<center>35</center>

modeling efforts. This study offers a library of fatty acids that can be used as trophic markers for striped bass forage species in multiple seasons and locations, along with the spatial and temporal variation in lipid of prey that are often neglected when modeling consumption.

The main factors influencing total lipid content of prey items appear to be species, season, and weight of the sample. Blue crab had the lowest lipid content of the four species in all seasons tested. The values seen in this experiment are consistent with other values found in the literature for corresponding seasons (Farragut 1965, Gokodlu and Yerlikaya 2003). Spot had higher lipid content than blue crab. The values we observed for spot in the Chesapeake Bay were lower than those reported in the literature for summer and similar to values reported for fall. Lipid content in our study was 4.04% for summer samples and 5.86% for fall samples while Waters (1982) reported levels of 8.69% for summer and 5.45% for fall. Our study used small age-0 spot ranging in size from 4 – 14g, versus a range of 113 – 340g for Waters (1982) with a similar extraction technique of organic solvents. Values reported by Waters (1982) reflect lipid content of fillets versus whole organisms as used in this experiment. Whole body lipids tend to be higher than fillets (Lanari et al. 1999, Chaiyapechara et al. 2003), therefore, the difference seen is not attributable to sampling technique, but rather may reflect a natural ontogenetic change in the lipid storage capabilities of spot.

The lipid levels of menhaden in our experiment ranged from 4.95 – 12.59% (mean = 10.41%). These fish represented a broad spectrum of sizes from 1.57 – 140.80g fish, although size did not appear to affect the percent lipid seen in this species. Dubrow et al. (1976) found that menhaden lipid levels ranged from 2.46 – 20.75% (mean = 12.08%) depending on season. Menhaden sampled in our experiment represented a broad size range, 78.7 – 455.4g. There was a

seasonal component, with lowest levels present in spring, and increasing through the summer and fall.

Seasonal change in lipid is expected in temperate fish that experience cold water temperatures throughout the winter. Fish can exit winter months in poor condition due to relying on lipid reserves for energy. Once these reserves have been exhausted, mortality is likely to occur (Biro et al. 2004). Similar results have been found in other species, e.g. Atlantic menhaden (Dubrow et al. 1976), gilthead sea bream *Sparus aurata* (Grigorakis et al. 2002), and alewife *Alosa pseudoharengus* (Flath and Diana 1985), although there appears to be substantial variation. Therefore it is imperative for an individual to deposit lipid reserves during the warmer months in order to make it through winter.

As the weight of the sample increases, the lipid content decreases for some species tested. This value is the relative proportion of wet weight that is comprised of lipid. As a fish gets larger, the absolute amount of lipid it contains will be larger when compared to small fish, but the proportion appears to decrease in anchovy and blue crab for each season and for menhaden and spot in certain seasons. The trend of lipid increase or decrease is species dependent (Thompson et al. 1991, Anthony et al. 2000). Blue crab, menhaden, and anchovy were sampled across a large size range, however, the number of samples was relatively small, and therefore more samples are needed to verify this pattern. As for spot, only juvenile fish were sampled but the trend of decreasing lipid appears to be present. The size of spot collected in our experiment represents the size of fish that will be consumed by juvenile striped bass. A larger prey item would still be preferable for striped bass because of the larger absolute value of lipid available for the predator.

Although the four prey species share common fatty acids, differences among the prey species were observed. The fatty acids that were different separated demersal feeding species, e.g. spot (Homer and Mihursky 1991) and blue crab (van Heukelem 1991), from planktivores, e.g. menhaden (Ahrenholz 1991) and bay anchovy (Hartman et al. 2004). Demersal fish have higher proportions of C20 monounsaturated isomers, which were much less prevalent in the planktivores. This lack of 20:1 isomers can be attributed to the dearth of these fatty acids in the phytoplankton consumed by menhaden (Arts et al. 2001). Other possible sources include benthic copepods (Arts et al. 2001) and polychaete worms associated with muddy substrate (Luis and Passos 1995). The Chesapeake Bay mesozooplankton community is dominated by copepods, specifically two calenoid copepods (Brownlee and Jacobs 1987), yet the C20:1 isomers are still at lower levels than for demersal species. The stomach contents were not removed from the prey samples which could affect their fatty acid signature. However these prey items are eaten whole by striped bass. Including the stomach contents of prey provides a more realistic representation of fatty acid signatures that will be encountered by striped bass. Therefore, it seems that these isomers form a larger part of the demersal rather than the pelagic food web.

The multivariate tools helped to provide insight into the structure behind fatty acid signatures. Among the three techniques evaluated, CART provided the best separation and lowest misclassification rate for the sample groups. The first and second nodes in CART were the demersal species, and as stated before, a C20 isomer was important in the branching pattern of these sample groups. The other fatty acid responsible for separating demersal and pelagic species was arachidonic acid (C20:4n6) which separates summer and fall demersal species. Linoleic acid (C18:2n6) is synthesized by phototrophic organisms (Desvilettes et al. 1997), and in turn is converted to arachidonic acid by higher organisms (Sargent et al. 1999). Phytoplankton

can settle out of the water column, and there is an opportunity for demersal organisms to consume fatty acids that would normally be present within the pelagic food web. Budge and Parrish (1998) found that very little of the organic material settling out was buried within the sediments. This suggests that demersal organisms have a high degree of efficiency in utilizing organic material before it can be incorporated into the sediments and is removed from the benthic food web. Further work needs to be done to compare the ability of demersal and pelagic species to desaturate and elongate fatty acids like linoleic acid (C18:2n6).

The demersal species consisted of blue crab and spot, and were grouped by season and not by species. Because both species may be viewed as opportunistic demersal feeders, it is not surprising to see such a high degree of overlap in their fatty acid signatures. The fact that the species are split according to season may indicate a seasonal shift in prey availability for the two species. There are some fatty acids that are higher in blue crab and spot in one season versus the other (C20:0, and C22:0), which may suggest that the fatty acid signature of demersal prey shifts from summer to fall. Hines et al. (1990) found that spot and blue crab had similar diets within a season, and that both species experienced a diet shift from spring to fall. Our results indicate that these two species have similar diets, and do experience a seasonal shift even though all four sample groups were collected from different systems in the upper Chesapeake Bay.

Among the planktivore sample groups (menhaden and anchovy), there was less distinction provided by season or location within the bay. While the CART dendrogram does separate menhaden and anchovy (Fig. 5), there is an anchovy grouping, a group of overlap, and a group of menhaden. The menhaden are not grouped in couplets as are the other species, suggesting that the fatty acid signature of each sample groups is unique. There is not enough overlap among menhaden sample groups to place them on the same couplet.

The difference seen in menhaden and anchovy signatures from benthic feeders is most likely the result of the plankton signatures themselves. Menhaden feed on zooplankton as larvae, and start to develop the ability to utilize phytoplankton as juveniles (June and Carlson 1971). Menhaden switch to a diet comprised mainly of phytoplankton as late stage juveniles and adults, but retain the ability to feed upon zooplankton (Lippson 1991). Anchovy feed on a diet of zooplankton throughout their lives (Johnson et al. 1990, Hartman et al. 2004).

Phytoplankton can have different fatty acid signatures (Napolitano et al. 1997). As zooplankton graze on the phytoplankton, another level of complexity is added because zooplankton will incorporate phytoplankton signatures and some modification may take place (Sargent and Falk-Petersen 1981). The fact that menhaden signatures are a mix of zooplankton and phytoplankton may explain why there is some similarity to anchovy and not a distinct signature for menhaden.

Adult menhaden were assumed to have a different fatty acid signature compared to smaller conspecifics, due to differences in feeding. This should be manifested in a separation of adult and juvenile menhaden in the CART output. According to the CART analysis, it is possible to distinguish adult versus juvenile menhaden. Of the 36 fatty acids and complexes identified, several fatty acids were significantly different in abundance between adult and juvenile menhaden. A small number of fatty acids appear capable of separating sample groups. Napolitano et al. (1997) demonstrated that only a change in the abundance of three to four fatty acids marked the difference among three different species of phytoplankton seen in Argentina. In both our study and that of Napolitano et al. (1997), the spectrum of fatty acids was similar in groups studied. It was the difference in abundance of fatty acids rather than a difference in fatty acids present that separated groups.

A more limited technique, DFA, was compared to CART to determine if a small number of fatty acids, or fatty acid metrics could provide the same information. This technique can only use n-1 variables for the model. The misclassification rates were much higher for DFA (20.3 and 26.1) compared to CART; however, the graphical output is still able to show separation among the groups. The DFA based upon the five fatty acids with highest variation separated the demersal species from the planktivorous species by oleic acid (C18:1n9) rather than arachidonic acid or a C20:1 isomer as in CART analysis. Anchovy and menhaden are categorized by having less oleic acid. Menhaden, however, have a higher abundance of myristic acid (C14:0) and palmitoleic acid (C16:1n7), while anchovy are more rich in palmitic acid (C16:0) and a combination of DPA (C22:5n3) and DHA (C22:6n3). These fatty acids were chosen because they exhibited the highest variation among all fatty acids. The higher values of C14:0 and C16:0 in menhaden versus demersal species corresponds to the literature (Gruger et al. 1964, Lytle and Lytle 1994), which suggests these fatty acids may indicate a planktivore. This technique is less effective in modeling the fatty acid signatures of striped bass prey items, because the misclassification rate is higher, and information is being left out because small sample groups necessitate fewer variables in the model.

Omega-3 fatty acids are found in high levels in marine environments because phytoplankton are able to synthesize certain fatty acids (e.g. DHA), which are then transferred up the food web through zooplankton and fish. Certain demersal polychaetes can also be high in omega-3 fatty acids, along with palmitic and oleic acids (Garcia-Alonso et al. 2008). It should be noted that co-eluted fatty acids for this DFA refer to eicosapentaenoic acid (EPA) and docohexsaenoic acid (DHA), two fatty acids which do make up a large proportion of the omega-3 category. Even though spot and blue crab are categorized by omega 3 fatty acids, menhaden

and anchovy still have omega-3 fatty acids at relatively high levels (7.09-27.44%) because of EPA and DHA. The groupings seen for this technique still separate demersal and pelagic species, but there seems to be more overlap between the anchovy and menhaden. It would be advisable to avoid DFA because information is left out in making the metrics.

Fatty acid analysis is a valuable technique in distinguishing among species (Gallagher et al. 1991, Lytle and Lytle 1994, Budge et al. 2002) or among seasons (Bandarra et al. 2001, Zlatanos and Laskaridis 2007). In the present study, the four species were correctly classified 92.8% of the time despite variation within a sample group. The second goal of this study was to determine if season or location influenced the fatty acid signature. The demersal species, spot and blue crab, were collected from the upper bay and demonstrated a clear seasonal pattern in fatty acid signature. Planktivorous fish, menhaden and anchovy, did not exhibit the same such pattern differences. Season and location appear to have less of an influence on these two species. It is also apparent that the fatty acid signature of Atlantic menhaden changes as these fish grow. Overall, there are differences in lipid values and fatty acid abundances among seasons and locations. These differences need to be taken into account when constructing food web models. The information obtained in this study offers a suite of fatty acids and lipid values that will provide fatty acid trophic markers for modeling forage history of top predators located in the Chesapeake Bay.

Literature cited

Ahrenholz, D. W. 1991. Population biology and life history of the North American menhadens, Brevoortia spp. Marine Fisheries Review 53:3-19.

Anthony, J.A., D.D. Roby, and K.R. Turco. 2000. Lipid content and energy density of forage fishes from the northern Gulf of Alaska. Journal of Experimental Marine Biology and Ecology 248:53-78.

Arts, M. T., R.G. Ackman, and B.J. Holub. 2001. "Essential fatty acids" in aquatic ecosystems: a crucial link between diet and human health and evolution. Canadian Journal of Fisheries and Aquatic Sciences 58:122-137.

Bandarra, N., I. Batista, M.L. Nunes, and J.M. Empis. 2001. Seasonal variation in the chemical composition of horse-mackerel (*Trachurus trachurus*). European Food Resources and Technology 212:535-539.

Beck, C. A., L.D. Rea, S.J. Iverson, J.M. Kennish, K.W. Pitcher, and B.S. Fadely. 2007. Blubber fatty acid profiles reveal regional, age-class and sex difference the diet of young Stellar sea lions in Alaska. Marine Ecology Progress Series 338:269-280.

Beck, C. A., S.J. Iverson, and W.D. Bowen. 2005. Blubber fatty acids of gray seals reveal sex differences in the diet of a size-dimorphic marine carnivore. Canadian Journal of Zoology 83:377-388.

Biro, P.A., A.E. Morton, J.R. Post, and E.A. Parkinson. 2004. Over-winter lipid depletion and mortality of age-0 rainbow trout (*Oncorhynchus mykiss*). Canadian Journal of Fisheries and Aquatic Sciences 61:1513-1519.

Boreman, J. and H.M. Austin. 1985. Production and harvest of anadromous striped bass stocks along the Atlantic coast. Transactions of the American Fisheries Society 114:3-7

Brodziak, J. and J. Link. 2002. Ecosystem-based fishery management: what is it and how can we do it? Bulletin of Marine Science 70:589-611.

Brownlee, D.C. and F. Jacobs. 1987. Mesozooplankton and microzooplankton in the Chesapeake Bay. In: Majumdar, Pages 217-269 *in* S.K., L.W. Hall, H.W. Austin, editors. Contaminant problems and Management of living Chesapeake Bay resources, vol. 527. The Pennsylvania Academy of Science, Philadelphia, PA.

Budge, S. M., S.J. Iverson, W.D. Bowen, and R.G. Ackman. 2002. Among- and within-species variability in fatty acid signatures of marine fish and invertebrates on the Scotian shelf, Georges Bank, and southern Gulf of St. Lawrence. Canadian Journal of Fisheries and Aquatic Sciences 59:886-898.

Budge, S. M., and C.C. Parrish. 1998. Lipid biogeochemistry of plankton, settling matter and sediments in Trinity Bay, Newfoundland. II. Fatty acids. Organic Geochemistry 29:1547-1559.

Burnham, K.P., and D.R. Anderson. 2002. Model selection and multi-model inference. 2nd ed. Springer, Berlin, Germany

Chaiyapechara, S., M.T. Casten, R.W. Hardy, F.M. Dong. 2003. Fish performance, fillet characteristics, and health assessment index of rainbow trout (*Oncorhynchus mykiss*) fed diets containing adequate and high concentrations of lipid and vitamin E. Aquaculture 219:715-738.

Chesapeake Bay Fisheries Ecosystem Advisory Panel (National Oceanic and Atmospheric Administration Chesapeake Bay Office). 2006. Fisheries ecosystem planning for Chesapeake Bay. American Fisheries Society, Trends in Fisheries Science and Management 3, Bethesda, Maryland.

Christie, W.M. 2003. Lipid Analysis. PJ Barnes and Associates. Bridgewater, England.

Dalsgaard, J. M. S. J., G. Kattner, D. Muller-Navarra, and W. Hagen. 2003. Fatty acid trophic

markers in the pelagic marine environment. Advances in Marine Biology 46:225-340.

Desvilettes, C.H., G. Bourdier. C.H. Ambland, and B. Barth. 1997. Use of fatty acids for the

assessment of zooplankton grazing on bacteria, protozoans, and microalgae. Freshwater

Biology 38:629-637.

Dubrow, D., M. Hale, and A. Bimbo. 1976. Seasonal variations in chemical composition and

protein quality of menhaden. Marine Fisheries Review 38:12-16.

Elliot, J.M. 1972. Rates of gastric evacuation in brown trout, *Salmo trutta* L. Freshwater Biology

2:1-18.

Farragut, R. 1965. Proximate composition of Chesapeake Bay blue crab (*Callinectes sapidus*).

Journal of Food Science 30:538-544.

Flath, L.E. and J.S. Diana. 1985. Seasonal energy dynamics of the alewife in southeastern Lake

Michigan. Transactions of the American Fisheries Society 114:328-337.

Folch, J., M. Lees, and G.H.S. Stanley. 1957. A simple method for the isolation and purification

of total lipids from animal tissues. Journal of Biological Chemistry 226:497-509.

Gallagher, M. L., M.L. Harrell, and R.A. Rulifson. 1991. Variation in lipid and fatty acid

contents of Atlantic croakers, striped mullet, and summer flounder. Transactions of the

American Fisheries Society 120:614-619.

Gannon, J.E. 1976. The effects of differential digestion rates of zooplankton by alewife, *Alosa

pseudoharengus*, on determinations of selective feeding. Transactions of the American

Fisheries Society 105:89-95.

Garcia-Alonso, J., C.T. Muller, and J.D. Hardege. 2008. Influence of food regimes and seasonality on fatty acid composition in the ragworm. Aquatic Biology 4:7-13.

Gokodlu, N. and P. Yerlikaya. 2003. Determination of proximate composition and mineral contents of blue crab (*Callinectes sapidus*) and swim crab (*Portunus pelagicus*) caught of the Gulf of Antalya. Food Chemistry 80:495-498.

Griffin, J. C. and F. J. Margraf. 2003. The diet of Chesapeake Bay striped bass in the late 1950s. Fisheries Management and Ecology. 10:323-328.

Grigorakis, K., M.N. Alexis, K.D.A. Taylor, and M. Hole. 2002. Comparison of wild and cultured gilthead sea bream (*Sparus aurata*); composition, appearance and seasonal variation. International Journal of Food Science and Technology 37:477-484.

Gruger, E.H., R.W. Nelson, and M.E. Stansby. 1964. Fatty acid composition from 21 species of marine fish, freshwater fish, and shellfish. Journal of the American Oil Chemists Society 41:662-667.

Gurr, M.I. and J.L. Hardwood. 1991. Lipid biochemistry: an introduction. 4th ed. Chapman and Hall. London, England.

Hartman, K.J. and S.B. Brandt. 1995. Trophic resource partitioning, diets, and growth of sympatric estuarine predators. Transactions of the American Fishery Society 124:520-537.

Hartman, K. J. 2003. Population-level consumption by Atlantic coastal striped bass and the influence of population recovery upon prey communities. Fisheries Management & Ecology 10:281-288.

Hartman, K. J., J. Howell, and J.A. Sweka. 2004. Diet and daily ration of bay anchovy in the Hudson River, New York. Transactions of the American Fisheries Society 133:762-771.

He, E. and W.A. Wurtsbaugh. 1993. An empirical model of gastric evacuation rates for fish and an analysis of digestion in piscivorous brown trout. Transactions of the American Fisheries Society 122:717-730.

Hines, A., A.M. Haddon, and L.A. Wiechert. 1990. Guild structure and foraging impact of blue crabs and epibenthic fish in a subestuary of Chesapeake Bay. Marine Ecology Progress Series 67:105-126.

Homer, M. L. and J. A. Mihursky. 1991. Spot (*Leiostomus xanthurus*). Pages 11-17 *in* S. L. Funderburk, S.J. Jordan, J.A. Mihursky, and D. Riley, editors. Habitat requirements for Chesapeake Bay living resources. Chesapeake Research Consortium, Inc., Solomons, MD.

Hyslop, E.J. 1980. Stomach contents analysis – a review of methods and their application. Journal of Fish Biology 17:411-429.

Iverson, S.J., K.J. Frost, and L.F. Lowry. 1997a. Fatty acid signatures reveal fine scale structure of foraging distribution of harbor seals and their prey in Prince William Sound, Alaska. Marine Ecology Progress Series 151:255-271.

Iveson, S.J., J.P.Y. Arnold, and I.L. Boyd. 1997b. Milk fatty acid signatures indicate both major and minor shifts in the diet of lactating Antarctic fur seals. Canadian Journal of Zoology 75:1881-97.

Iverson, S. J., K.J. Frost, and S.L.C. Lang. 2002. Lipid content and fatty acid composition of forage fish and invertebrates in Prince William Sound, Alaska: factors contributing to among and within species variability. Marine Ecology Progress Series 241:161-181.

Iverson, S.J. C. Field, W.D. Bowen, and W. Blanchard. 2004. Quantitative fatty acid signature analysis: a new method of estimating predator diets. Ecological Monographs 74:211-235.

Johnson, W.S., D.M. Allen, M.V. Ogburn, and S.E. Stancyk. 1990. Short-term responses of adult bay anchovies *Anchoa mitchilli* to estuarine zooplankton availability. Marine Ecology Progress Series 64:55-68.

June, F.C. and F.T. Carlson. 1971. Food of young Atlantic menhaden, *Brevoortia tyrannus*, in relation to metamorphosis. Fishery Bulletin 68:493-512.

Kirsch, P. E., S.J. Iverson, W.D. Bowen, S.R. Kerr, and R.G. Ackman. 1998. Dietary effects on the fatty acid signature of whole Atlantic cod (Gadus morhua). Canadian Journal of Fisheries and Aquatic Sciences 55:1378-1386.

Lanari, D., B.M. Poli, R. Ballestrazzi, P. Lupi, E. D'Agaro, and M. Mecatti. 1999. The effects of dietary fat and NFE levels on growing European sea bass (*Dicentrachus labrax* L.). Growth rate, body and fillet composition, carcass traits and nutrient retention efficiency. Aquaculture 179-351-364.

Link, J.S. 2005. Translating ecosystem indicators into decision criteria. ICES Journal of Marine Science 62:569 - 576

Lippson, R. L. 1991. Atlantic menhaden (*Brevoortia tyrannus*). Pages 7.1-7.6 *in* S. L. Funderburk, S.J. Jordan, J.A. Mihursky, and D. Riley, editors. Habitat requirements for Chesapeake Bay living resources. Chesapeake Research Consortium, Solomons, MD.

Luis, O.J. and A.M. Passos 1995. Seasonal changes in lipid content and composition of the polychaete *Nereis (Hediste) diversicolor*. Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology 111: 579-586.

Lytle, J. S., and T. F. Lytle. 1994. Fatty acid pattern differences among individuals of two estuarine fishes (*Leiostomus xanthurus* and *Mugil cephalus*). Gulf Research Reports 9:49-56.

McCune, B. and J.B. Grace. 2002. Analysis of ecological communities. MjM Software, Gleneden Beach, OR.

Napolitano, G.E., R.J. Pollero, A.M. Gayoso, B.A. MacDonald, and R.J. Thompson. 1997. Fatty acids as trophic markers of phytoplankton blooms in the Bahia Blanca estuary (Buenos Aires, Argentina) and in Trinity Bay (Newfoundland, Canada). Biochemical Systematics and Ecology 25:739-755.

Overton, A. S., E.B. May, J. Griffin, and F.J. Margraf. 2000. A bioenergetics approach for determining the effect of increased striped bass population on its prey and health in the Chesapeake Bay. Maryland Department of Natural Resources, Annapolis.

R Development Core Team (2008). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Recks, M.J. and G.T. Seaborn. 2008. Variation in fatty acid composition among nine forage species from a southeastern US estuarine and nearshore coastal ecosystem. Fish Physiology and Biochemistry 34:275-287.

Sargent, J, G. Bell, L. McEvoy, D. Tocher, and A. Estevez 1999. Recent developments in the essential fatty acid nutrition of fish. Aquaculture 177:191-199.

Sargent, J.R. and S. Falk-Petersen. 1981. Ecological investigations on the zooplankton community in Balsfjorden, northern Norway: lipids and fatty acids in *Meganyctiphanes norvegica*, *Thysanoessa raschi*, and *T. inermis* during mid-winter. Marine Biology 62:131-137.

Schafer, K. 1998. Accelerated solvent extraction of lipids for determining the fatty acid composition of biological material. Analytica Chimica Acta 358:69-77.

Seaburg, K. G. 1957. A stomach sampler for live fish. The Progressive Fish Culturist:137-139.

Secor, D.H. and E.D. Houde. 1998. Use of larval stocking in restoration of Chesapeake Bay

    striped bass. ICES Journal of Marine Science 55:228-239.

Thompson, J.M, E.P. Bergensen, C.A. Carlson, and L.R. Kaeding. 1991. Role of size, condition,

    and lipid content in the overwinter survival of age-0 Colorado squawfish. Transactions of

    the American Fisheries Society 120:346-353.

Turner, J. P., and J. R. Rooker. 2005. Effect of dietary fatty acids on the body tissues of larval

    and juvenile cobia and their prey. Journal of Experimental Marine Biology and Ecology

    322:13-27.

Uphoff Jr., J.H. 2003. Predator-prey analysis of striped bass and Atlantic menhaden in upper

    Chesapeake Bay. Fisheries Management and Ecology 10:313-322.

USEPA (United States Environmental Protection Agency). 2007. Pressurized fluid extraction

    (PFE). USEPA SW-846. Method 3545A, Update IV. Washington, D.C.

van Heukelem, W. F. 1991. Blue crab (*Callinectes sapidus*). Pages 6.1-6.24 *in* S. L. Funderburk,

    S.J. Jordan, J.A. Mihursky, and D. Riley, editors. Habitat requirements of Chesapeake

    Bay living resources. Chesapeake Research Consortium, Inc., Solomons, MD.

Vaughn, D.S. and J.W. Smith. 1988. Stock assessment of the Atlantic menhaden, *Brevoortia*

    tyrannus, fishery. NOAA Technical Report NMFS 63. 26p.

Waters, M. 1982. Chemical composition and frozen storage stability of spot, *Leiostomus*

    xanthurus. Marine Fisheries Review 44:14-22.

Zlatanos, S. and K. Laskaridis. 2007. Seasonal variation in the fatty acid composition of three

    Mediterranean fish – sardine (*Sardina pilchardus*), anchovy (*Engraulis encrasicholus*)

    and picarel (*Spicara smaris*). Food Chemistry 103:725-728.

Table 1. The library of prey species analyzed for fatty acid analysis. The values for length, weight, and lipid content represent the mean ± 1

standard error of the mean. Lipid content is based upon gram of lipid per gram of fish.

Species	Season	Bay location	River system	Sample size	Length ± SEM (mm)	Weight ± SEM (g)	Lipid content ± SEM (% ww)
Atlantic menhaden	Spring	Upper	Choptank River	8	78.00 ± 1.69	4.64 ± 0.33	4.95 ± 0.22
	Spring	Lower	York River	36 $^\Delta$	51.30 ± 1.25	1.57 ± 0.10	8.65 ± 1.16
	Summer	Upper	Corsica River	10	97.5 ± 0.69	8.37 ± 0.15	12.59 ± 1.94
	Summer	Lower	James River	10	143.34 ± 3.93	39.57 ± 2.25	12.07 ± 1.84
	Fall	Upper	Choptank River	9	127.56 ± 2.84	24.33 ± 1.98	12.46 ± 1.17
Atlantic menhaden adult	Summer	Upper	Nanticoke River	10	Ŧ 233.08 ± 10.53	Ŧ 140.80 ± 21.49	Ŧ 11.75 ± 1.91
Bay anchovy	Spring	Upper	Patuxent River	8	59.5 ± 1.84	1.37 ± 0.09	10.80 ± 0.99
	Spring	Lower	Main stem Chesapeake	10	56.61 ± 1.94	1.97 ± 0.17	8.21 ± 1.23
	Summer	Upper	Magothy River	15	73.41 ± 1.74	2.76 ± 0.20	14.93 ± 3.90
	Summer	Lower	Main stem Chesapeake	23 $^\Delta$	65.25 ± 1.43	2.57 ± 0.46	5.89 ± 0.44
	Fall	Upper	Trappe River	35 $^\Delta$	49.61 ± 1.70	1.47 ± 0.31	15.10 ± 2.98
	Fall	Lower	Main stem Chesapeake	34 $^\Delta$	46.71 ± 1.20	1.30 ± 0.15	13.34 ± 3.78
Blue crab	Summer	Upper	Magothy River	12	*46.71 ± 2.57	7.21 ± 1.45	1.49 ± 0.28
	Fall	Upper	Chester River	10	*120.6 ± 4.59	89.1 ± 7.41	1.18 ± 0.17
Spot	Summer	Upper	Rhode River	10	84.67 ± 2.12	7.97 ± 0.58	4.49 ± 0.34
	Fall	Upper	Chester River	11	79.55 ± 2.85	6.05 ± 0.82	6.05 ± 0.75

Ŧ Lengths and weights are measurements with head and tail present. The lipid content for this sample group was calculated based on the body with no head or tail.

*Length refers to carapace width, point to point.

Δ Several individuals were used in one sample due to low weight.

Table 2. Summary of model selection statistics for lipid percent in samples; K represents the number of parameters in the model; ΔAIC_c represents the difference between the AIC_c value for that model and the set's minimum AIC_c value; W represents the Akaike weight of each model.

Model	K	ΔAIC_C	W
We+Sp+Se+We*Sp[A]	11	0	0.71
We+Sp+Se+We*Sp+We*Se+Sp*Se	17	3.19	0.14
We+Sp+Se	8	3.86	0.10
We+Sp+Se+We*Sp+We*Se+Sp*Se+We*Se*Sp	21	5.41	0.05
We+Sp	6	14.24	0
We	3	156.82	0
Null model	2	160.99	0

[A] Model terms are sample weight (We), species (Sp: Atlantic menhaden, bay anchovy, spot, and blue crab), and season (Se: spring, summer and fall).

Table 3. Parameter estimates from the AIC model that included all factors (weight, species, and season) along with the second order interaction term of weight by season. The standard error (SE) of the estimate is given along with the p value. The weight of the model was 0.71.

Parameter	Estimate	SE	p value
Intercept	3.06	0.29	**0.0001**
Log weight	-0.47	0.21	**0.02**
Blue crab	-2.27	0.40	**0.0001**
Menhaden	-0.57	0.28	**0.04**
Spot	-0.62	0.67	0.36
Spring	-0.75	0.19	**0.0001**
Summer	-0.20	0.11	0.07
Log weight*Blue crab	0.29	0.23	0.2
Log weight*Menhaden	0.49	0.19	**0.01**
Log weight*Spot	0.08	0.38	0.84

Table 4. Parameter estimates from the AIC model that included all factors (weight, species, and season) along with the second order interaction terms of weight by species, weight by season, and species by season. The standard error (SE) of the estimate is given along with the p value. The weight of the model was 0.14.

Parameter	Estimate	SE	p value
Intercept	2.80	0.65	**0.0001**
Log weight	-0.27	0.47	0.57
Blue crab	1.61	1.60	0.31
Menhaden	1.40	0.94	0.14
Spot	0.09	0.79	0.90
Spring	-0.56	0.62	0.37
Summer	-1.10	0.55	**0.05**
Log weight*Blue crab	-0.71	0.42	0.09
Log weight*Menhaden	-0.28	0.34	0.42
Log weight*Spot	-0.42	0.51	0.41
Log weight*Spring	0.06	0.45	0.89
Log weight*Summer	0.54	0.39	0.16
Blue crab*Spring	NA	NA	NA
Menhaden*Spring	-1.20	0.84	0.15
Spot*Spring	NA	NA	NA
Blue crab*Summer	-2.31	1.27	0.07
Menhaden*Summer	-0.71	0.74	0.34
Spot*Summer	-0.03	0.36	0.93

Table 5. Parameter estimates from the AIC model that included all factors (weight, species, and

season). The standard error (SE) of the estimate is given along with the p value. The weight of

the model was 0.10.

Parameter	Estimate	SE	p value
Intercept	2.57	0.14	**0.0001**
Log weight	-0.09	0.06	0.13
Blue crab	-2.08	0.17	**0.0001**
Menhaden	0.16	0.15	0.29
Spot	-0.73	0.14	**0.0001**
Spring	-0.60	0.16	**0.0002**
Summer	-0.16	0.10	0.13

Table 6. Results from the classification and regression trees (CART) and discriminant function analysis (DFA) in this experiment. The discriminant function analysis was used on the 5 FA with the highest variation and metrics consisting of saturated, monounsaturated, omega 3, omega 6, and co-eluted FAs. Sample size in each group is denoted by n, and the number correctly classified is listed under CART and the two DFA categories.

		Number correctly classified		
Sample	n	CART (24 FA)	DFA (5 FA)	DFA (5 FA sums)
Bay Anchovy				
Spring Upper	8	8	3	4
Spring Lower	8	7	8	8
Summer Upper	9	8	6	2
Summer Lower	10	10	9	9
Fall Upper	10	10	9	8
Fall Lower	10	9	9	6
Menhaden				
Spring Upper	8	6	8	8
Spring Lower	10	10	10	10
Summer Upper (Choptank River)	8	6	7	8
Summer Upper (Corsica River)	9	9	7	8
Summer Upper (Adult-Nanticoke River)	9	9	4	2
Summer Lower	10	10	9	7
Fall Upper	6	3	1	0
Blue Crab				
Summer Upper	11	11	10	8
Fall Upper	10	10	6	9
Spot				
Summer Upper	6	5	5	5
Fall Upper	11	11	11	11
Total correct		142/153	122/153	113/153
% Correctly classified		92.8	79.7	73.9

Figure 1. Map of collection sites in the Chesapeake Bay. The black line represents a division

      between bay locations. Above the line, salinities ranged from 2-8 parts per thousand

      (ppt), and below the line samples salinities ranged from 10-16 ppt. The larger map is a

      representation of the box in the inset.

Figure 2. Natural log lipid percent (of wet weight) of samples collected in the a) spring, b)

      summer, and c) fall.

Figure 3. Major fatty acids in a) anchovy; and b) menhaden signatures. Solid bars represent

      samples collected from the upper Bay and patterned bars represent samples collected

      from the lower Bay.

Figure 4. Major fatty acids in a) spot; and b) blue crab signatures. Solid bars represent samples

      collected from the upper Bay and patterned bars represent samples collected from the

      lower Bay.

Figure 5. Classification and regression tree (CART) output for prey species classification. The

      frequency refers to which individual was classified at that terminal node. Letters in the

      legend correspond to letters at the end of each branch of the dendrogram.

Figure 6. Discriminant function analysis of the group mean centroids for the 5 fatty acids with

      the highest variation. The first 2 functions explained 70.5% of the variation. Arrows on

      the axes represent increases in that particular fatty acid in the direction noted.

Figure 7. Discriminant function analysis of the group mean centroids for the 5 composite sums of

fatty acids. The first 2 functions explained 71.5% of the variation. Arrows on the axes

represent increases in that particular fatty acid metric in the direction noted.

Figure 8. Abundance of 4 fatty acids that exhibit differences based upon the size of menhaden

analyzed (n=26). The adult menhaden length refers to total length, with head and tail

included.

Figure 1.

Figure 2.

Natural log lipid percent (% wet weight)

Natural log wet weight (g)

a.

b.

c.

Anchovy
Menhaden
Spot
Blue crab
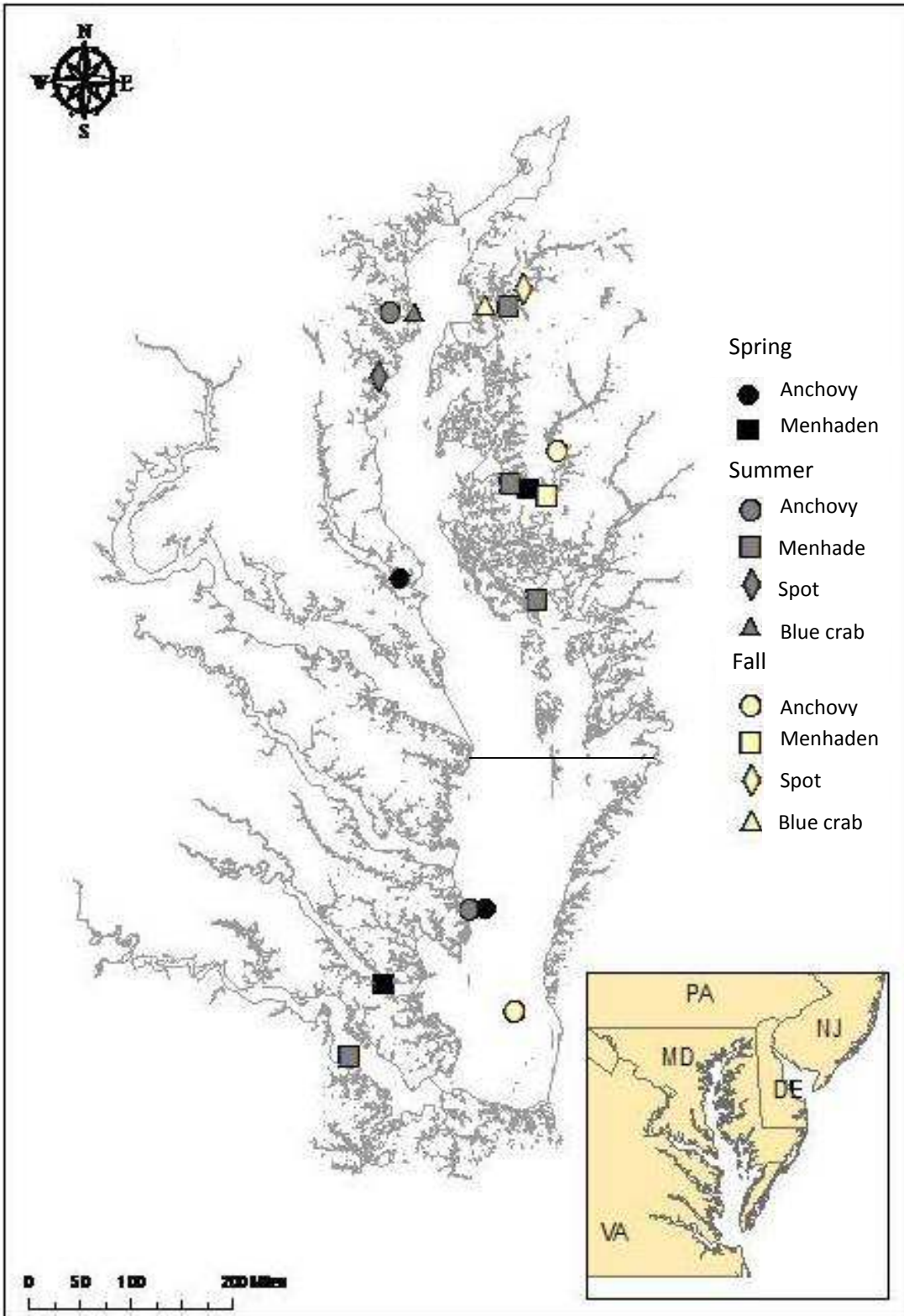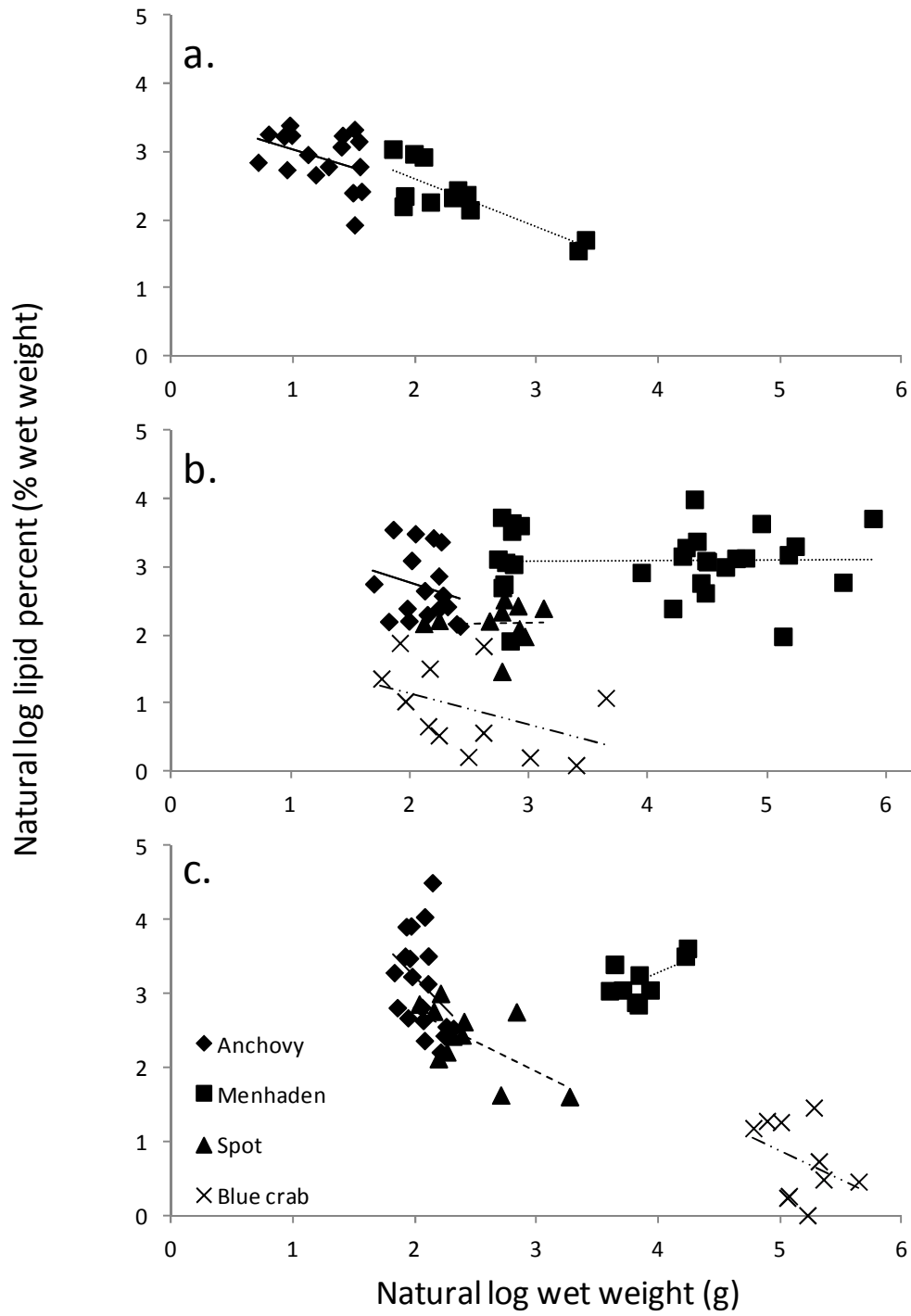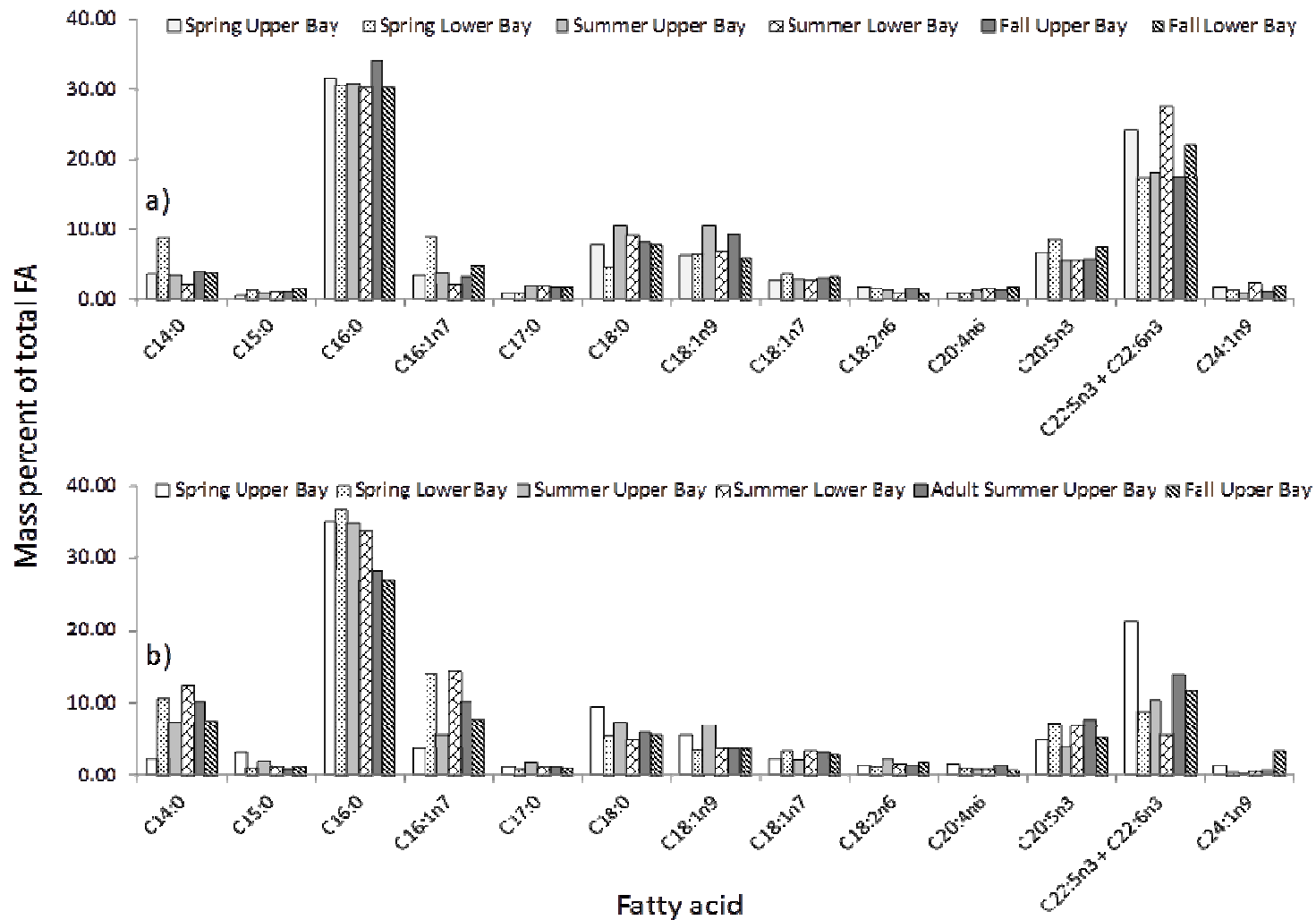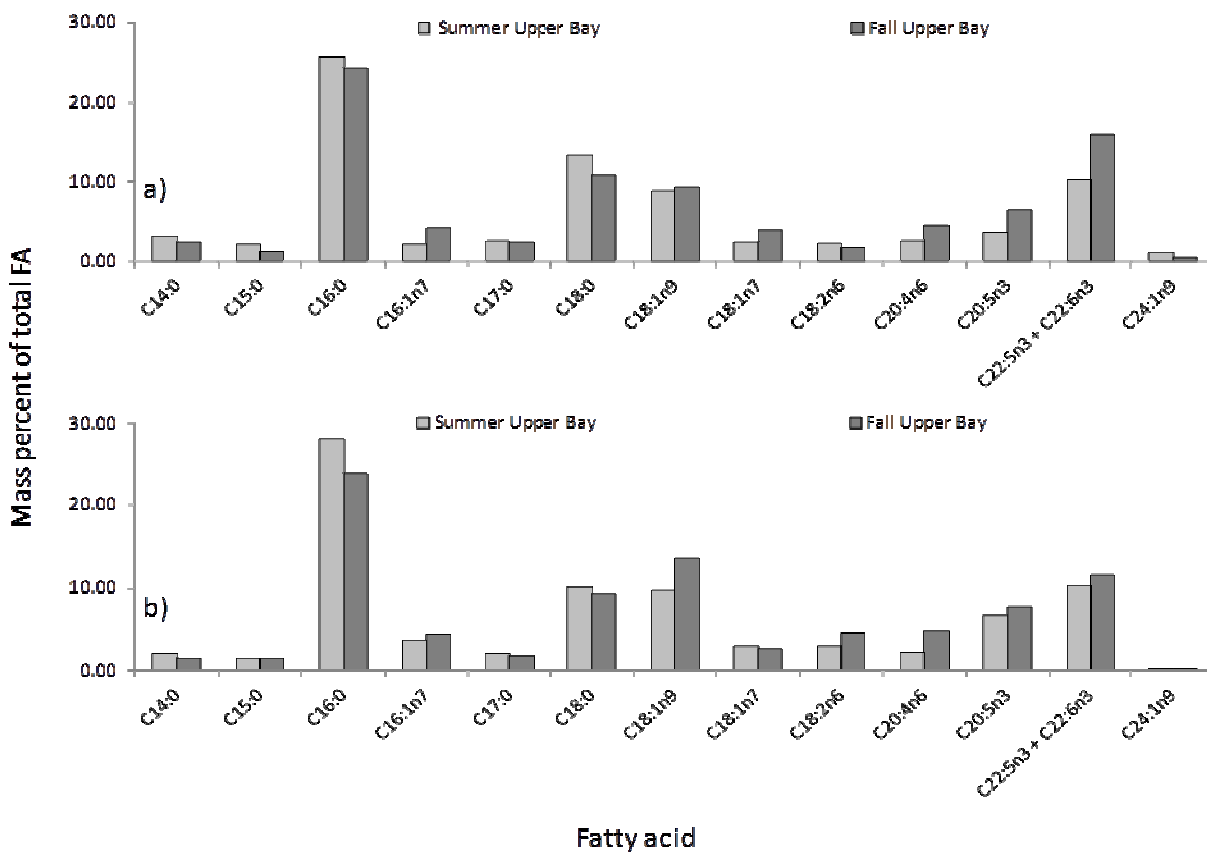
Figure 3.

Figure 4.

Figure 5.

Legend

A=Anchovy, spring, lower bay
B=Anchovy, spring, upper bay
C=Anchovy, summer, lower bay
D=Anchovy, summer upper bay
E=Anchovy, fall, lower bay
F=Anchovy, fall, upper bay
G=Menhaden, spring, lower bay
H=Menhaden spring, upper bay
 I=Menhaden, summer, lower bay

J=Menhaden, summer, upper bay
K=Menhaden, summer, upper bay
L=Menhaden (Adult), summer, upper bay
M=Menhaden, fall, upper bay
N=Blue crab, summer upper bay
O=Blue crab, fall, upper bay
P=Spot, summer, upper bay
Q=Spot, fall, upper bay

Freq.
H=8/8

Freq.
E=1/12
G=10/12
J=1/12

Freq.
N=11/11

Freq.
O=10/11
P=1/11

Freq.
E=9/9

Freq.
C=10/11
J=1/11

Freq.
A=7/7

Freq.
B=8/8

Freq.
E=1/11
F=10/11

Freq.
J=6/6

Freq.
M=3/3

Freq.
L=9/9

Freq.
D=8/8

Freq.
D=1/12
I=10/12
K=1/12

Freq.
M=1/10
K=9/10

Freq.
M=1/7
N=1/7
P=5/7

Freq.
Q=11/11

64
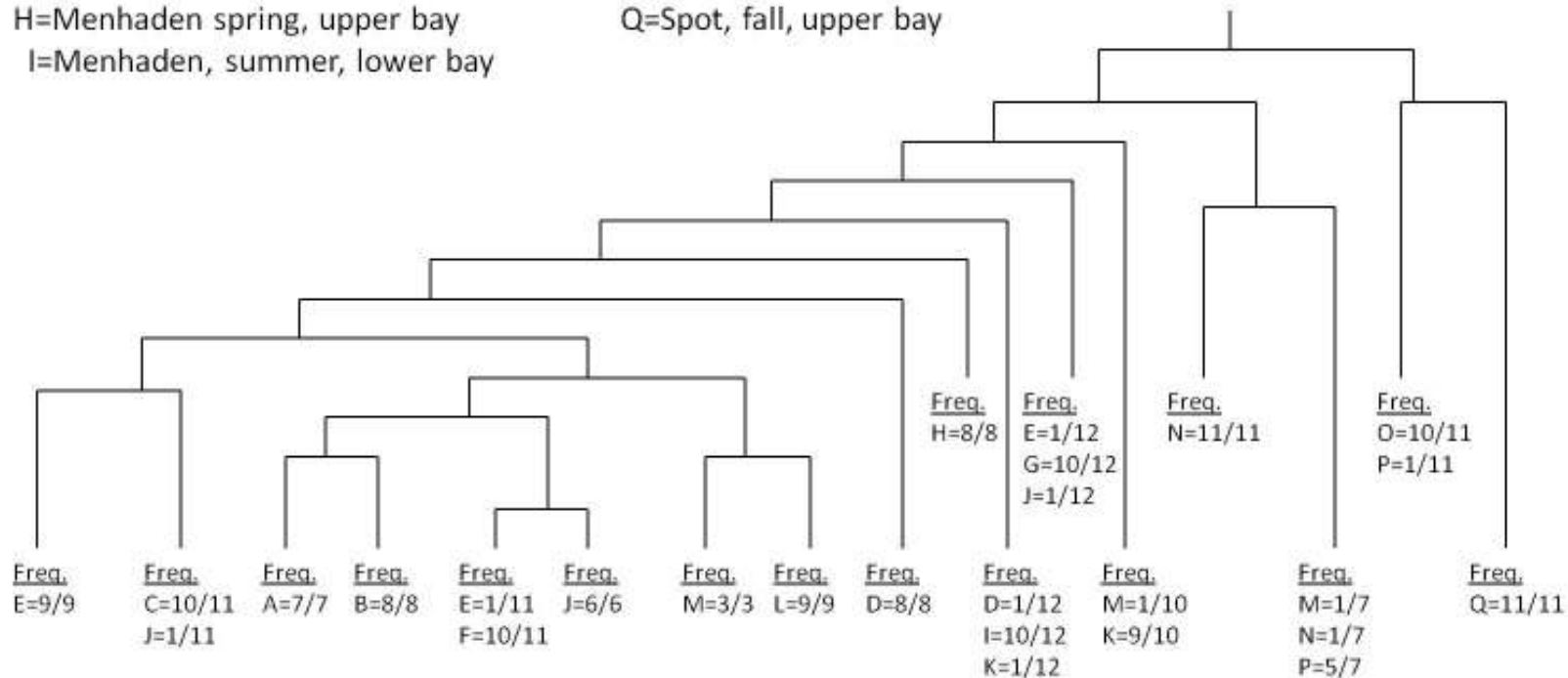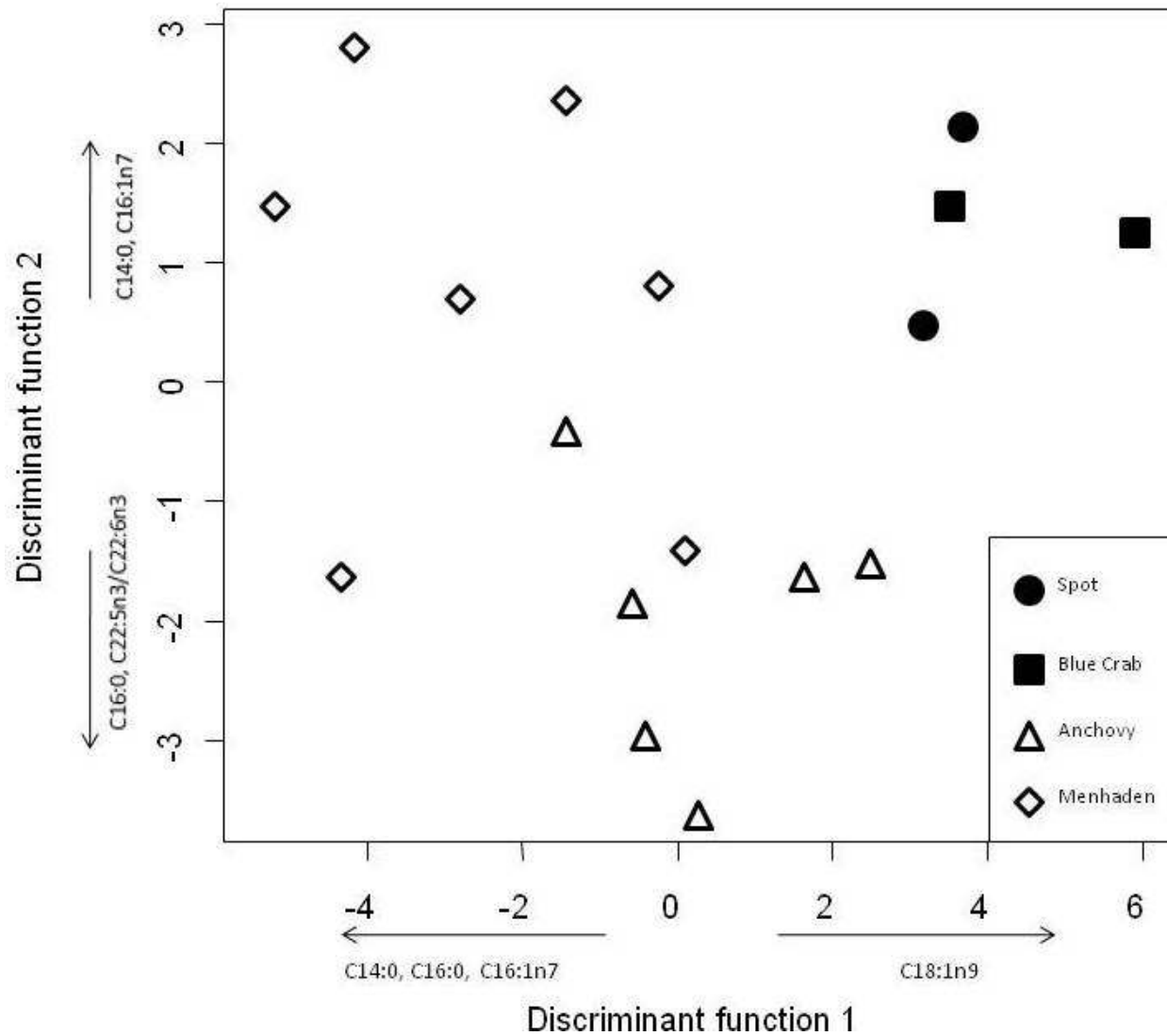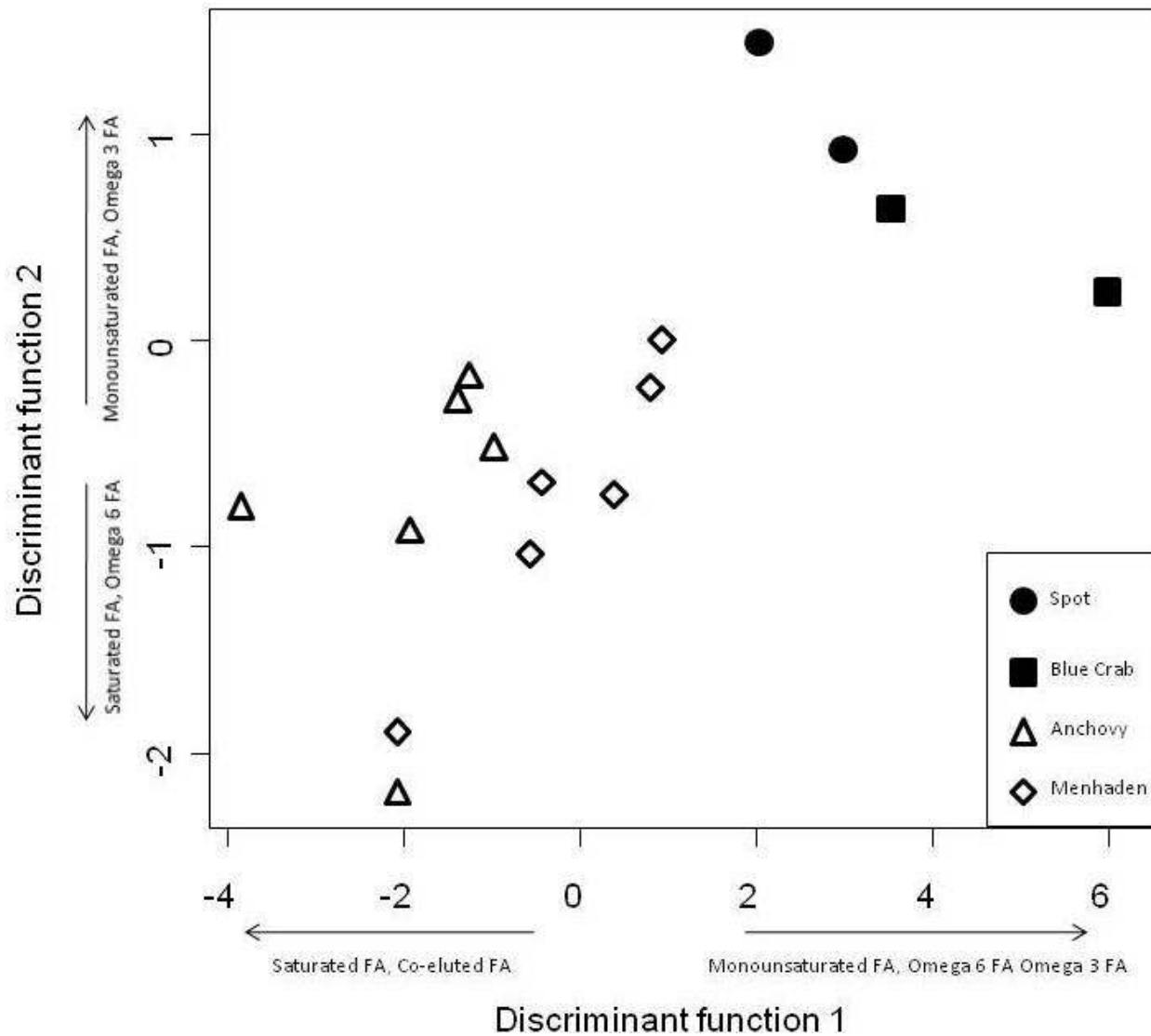
Figure 6.

Figure 7.

Figure 8.
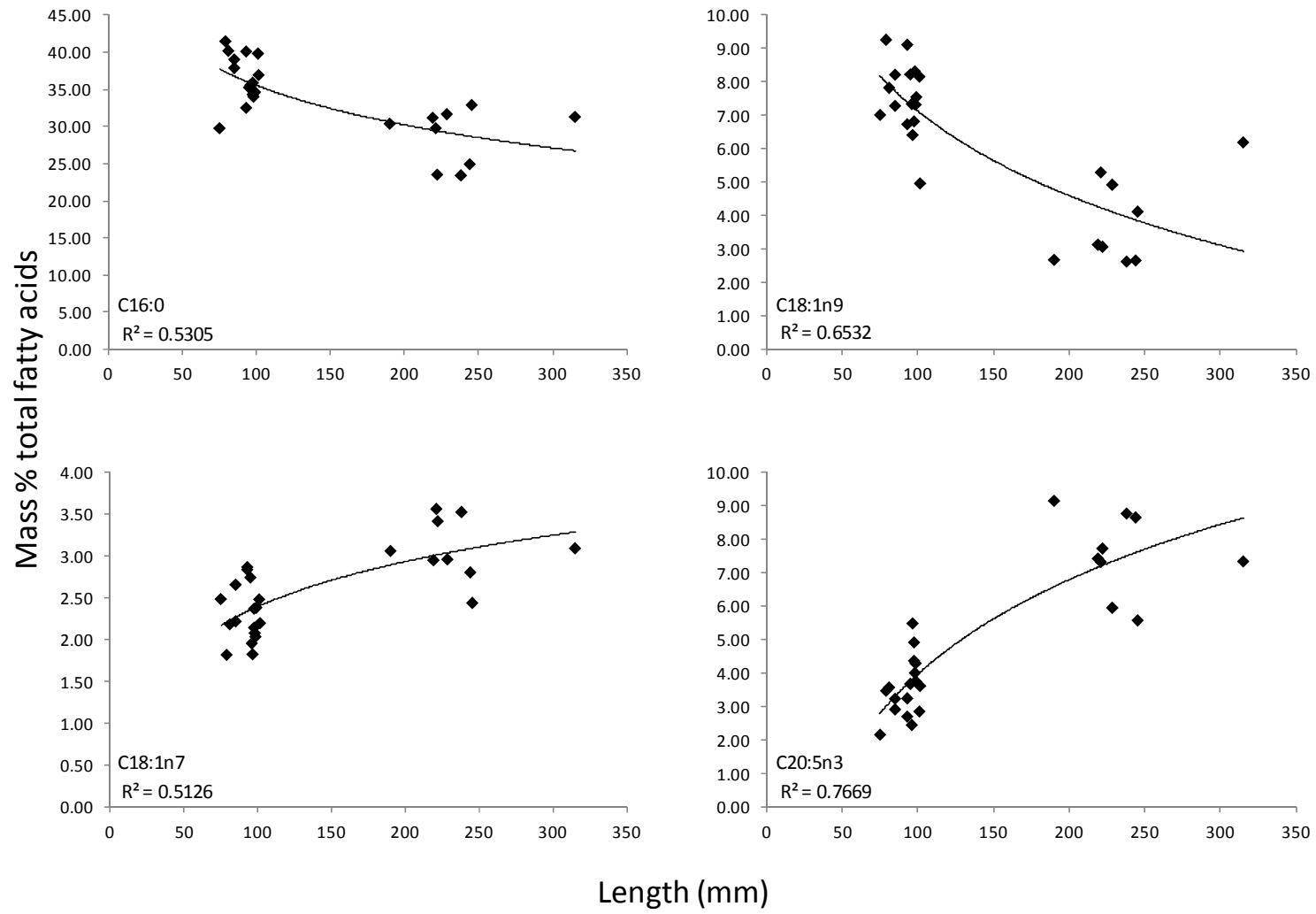
Chapter 3 - A comparison of liver and belly flap for evaluating striped bass diet using fatty acid signatures[1]

Abstract

The process of fatty acid conservation from prey to predator has been well studied in aquatic systems. Predators incorporate prey fatty acids, often with little to no modification into their own tissues. This fact has been the basis for trophic studies in many different systems because it removes the bias of examining stomach contents. However, there has been little research to determine which tissues in carnivorous fish offer the clearest picture of feeding history. The goal of this study was to compare fatty acid incorporation between liver and adipose tissue in striped bass *Morone saxatilis*. Striped bass were fed a diet of bay anchovy *Anchoa mitchilli* for four weeks to establish a baseline signature. Experimental feeding consisted of spot *Leiostomus xanthurus* for six weeks followed by a diet of Atlantic menhaden *Brevoortia tyrannus* for six weeks. Liver samples were consistently lower in lipids than belly flap regardless of the diet. Both tissues increased in lipid once the diet was switched to menhaden. Fatty acid signatures were different between the two prey types, and this difference was evident in both striped bass tissues tested. Proportions of discrete fatty acids, rather than the entire fatty acid profile, indicated the change in diet from spot to menhaden and were evident in striped bass tissues after a period of 31 days. Both tissues provide information on fatty acid incorporation and make good candidates for tracking diets; however belly flap tissue offers an easier and nonlethal sampling opportunity.

Keywords: Fatty acid trophic markers, lipid, Atlantic menhaden, spot

Introduction

The feeding history of an organism is an important component of its overall ecology, and as such, has been widely studied in fish (Adams 1976, Currin et al. 1984). Striped bass *Morone saxatilis* (Walbaum 1792) specifically have been well studied due to their position as a top predator and an important recreational and commercial fishery (Schafer 1970, Hartman and Brandt 1995). Because any estimate of feeding history includes some biases, the results obtained can be skewed to varying degrees based upon the amount of error inherent in the method.

Traditionally, the method for analyzing stomach contents has been direct observation, whether by sacrificing the fish or manually removing the contents (Seaburg 1957). Estimations of diet were based on various metrics including (but not limited to): percent composition by weight or number and frequency of occurrence. From these metrics, indices of preference have been constructed based upon prey taxa prevalence in the stomach and the environment (Bowen 1996). Due to confounding factors such as the presence of chitinous material, prey size, and water temperature (Elliot 1972, Gannon 1976, He and Wurtsbaugh 1993), the rates of digestion for prey taxa are not identical. Therefore, estimations of diet and preference are likely biased, and a more accurate method is needed.

One alternative is fatty acid analysis, which is a method that focuses on biochemical markers for estimating diet history. Fatty acids are long chains of carbon and hydrogen atoms with a carboxylic acid functional group. Within organisms, excess energy in the form of fatty acids is stored as compounds known as acylglycerols. The most common form of energy storage involves three fatty acids esterified to a glycerol backbone (Gurr and Hardwood 1991). Fatty acids can be tracked as they are cycled through the food web (Iverson 1997a), and the diet can be estimated from the fatty acids present and their relative proportions with respect to other

fatty acids (Iverson et al. 2004). Studies have also shown that a broader temporal history can be inferred from analyzing the fatty acids of an organism. Often, it is possible to determine diet items eaten over the past several weeks, versus the last meal (Iverson 1997b). These compounds are deposited from prey to predator largely unmodified (Iverson 1997a), providing a good candidate for eliminating the biases associated with analyzing stomach contents.

The fatty acid profile of the predator should begin to match that of the prey after a few weeks (Kirsch et al. 1998). Fatty acids are transported via lipoproteins within the lymphatic system (Tocher 2003) to storage sites after re-esterification into triacylglycerols (Iverson 2009). This process may not deliver fatty acids in equal quantities to every tissue. This process may be based more on the needs of the tissue for a particular fatty acid and differences in fatty acid delivery to specific tissues has been observed in fish (Montero et al. 2001, Njinkoue et al. 2002). It is imperative to understand the process of fatty acid incorporation in tissues that are intimately involved in fatty acid processing and storage.

Fatty acid analysis does have problems that need to be addressed for the technique to be a valuable resource. No fatty acid is completely unique to a species (Dalsgaard et al. 2003) and several fatty acids may be needed to distinguish between species (Recks and Seaborn 2008). This shortcoming requires several fatty acids present in the signature to be used to distinguish among prey items. A way to alleviate this problem is to design laboratory experiments that examine the process of lipid integration of a specific prey, and to examine how the signature changes when the diet is switched.

In order to determine the efficacy of fatty acid analysis for estimating striped bass diets, the pattern of incorporation must be determined first. Therefore, the aims of this study were to: 1) determine the rate at which striped bass tissues incorporated prey fatty acids; 2) examine if the

entire prey fatty acid signature was incorporated into striped bass tissues; and 3) determine whether the liver or belly flap tissues were reliably mimicking the fatty acid signature of the prey. These two tissues were chosen because they hold little commercial value and represent areas largely responsible for storage (adipose) and processing (liver) of lipids.

Materials and Methods

Experimental design

Striped bass (Chesapeake Bay strain; total length 325 ± 6.21 mm) were obtained from West Virginia University's Aquaculture Extension Unit and were transported to the James J. Howard Marine Sciences Laboratory in Sandy Hook, New Jersey. Fish were randomly distributed among three 120 gallon tanks. The tanks were part of a flow through system with water pumped directly from the Sandy Hook Bay. Temperature ranged from 20.4 - 26.8^{o}C, and salinity ranged from 23.0 - 26.4 parts per thousand (ppt) during the feeding experiment. Striped bass were fed a diet of bay anchovy *Anchoa mitchilli* (Valenciennes 1848) every other day for four weeks prior to the experiment to establish a baseline fatty acid signature (Kirsch et al. 1998). At the start of the experiment (day 0), fish were fed a diet of spot *Leiostomus xanthurus* (Lacepede 1802) *ad libitum* every other day to approximate ½ satiation. Four fish from each tank were sacrificed every two weeks starting on day 0. On day 40, the diet was switched to Atlantic menhaden *Brevoortia tyrannus* (Latrobe 1802) and four fish were sacrificed on days 71 and 88. Fish were weighed (g) and measured (total length in mm) and vacuum sealed with a FoodSaver vacuum sealer (Sunbeam Products, Inc., Niles, IL) and frozen at -20°C until they could be analyzed.

Sample preparation

  Striped bass were thawed at the time of analysis, and tissue samples were weighed to the nearest 0.1 gram. The liver and a portion of the belly flap were removed. The belly flap (with skin intact) follows a line marked by the termination of the ribs and initiation of the peritoneum of the body wall (Jacobs et al. 2008).

  All tissues were ground with a chemical drying agent, diatomaceous earth (Dionex, Sunnyvale, CA). Lipid was extracted with methylene chloride/methanol (3:1; v:v) under nitrogen using an Accelerated Solvent Extractor (ASE 300, Dionex, Sunnyvale, CA) according to Folch et al. (1957). Samples were extracted at $100^{o}C$ and 1500 psi (USEPA 2007). Three samples of tissue were extracted at both $100^{o}C$ and $120^{o}C$ to compare the yield in total lipid extracted (Schafer 1998) and create a correction factor. Total lipid extracted was determined gravimetrically for each sample and reported as a percent of wet weight.

  Extracted lipid species containing fatty acids as various derivatives were transesterified using a solution of sulfuric acid and methanol (Christie 2003). Fatty acid methyl esters were run on a gas chromatograph (Agilent 6890N Network GC System, Santa Clara, CA) / mass spectrometer (GC/MS, Agilent, 5973 Network Mass selective detector, Santa Clara, CA). The chromatograph was equipped with a fused silica capillary column, DB-23 60m length X 0.250mm OD X 0.25µm film thickness (Agilent, Santa Clara, CA) with hydrogen as a carrier gas. The temperature program was as follows: hold at an initial temperature of $50^{o}C$ for 2 min, hold for 1 min at $150^{o}C$ after ramping at $20^{o}C \cdot min^{-1}$, ramp to $215^{o}C$ at $1.25^{o}C \cdot min^{-1}$. The fatty acid retention times were compared to known standards from Nu-Chek Prep (Elysian, MN), Cayman Chemical (Ann Arbor, MI), Matreya (Pleasant Gap, PA) and Restek (Bellefonte, PA). There were peaks which consisted of more than one fatty acid and it was impossible to determine

which fatty acid was creating the peaks. Therefore, the term "fatty acid complex" was assigned to this peak. The complex refers to which fatty acid(s) could be responsible for the peak, e.g. C22:5n3 and C22:6n3.

Statistical analyses

To compare the effect of the prey species on growth, analysis of variance (ANOVA) was used to determine if there were differences in the weight of striped bass among all sample days (R Development Core Team 2010, Vienna, Austria). Residuals of the non-transformed data were normally distributed ($p > 0.05$) and therefore no transformation was performed. A Tukey's HSD post hoc test was performed to identify where the differences occurred.

Fatty acid methyl esters were expressed as the percent of the total fatty acid concentrations. These values were used to determine differences in the prey species. These values were then compared to tissues samples throughout the experiment to determine if striped bass tissues reflected prey values.

A permutational multivariate analysis of variance (PERMANOVA, Anderson 2005) was performed on the tissue samples and the two prey types, spot and menhaden, to determine if there were significant differences among the groups (R Development Core Team 2010, Vienna, Austria). This technique does not require variables constrained to a normal distribution and instead it creates a distribution based on permutations of the data.

Nonmetric multidimensional scaling (NMDS) was also performed on the data (R Development Core Team 2010, Vienna, Austria). This ordination technique is an iterative search for the best position of samples in multidimensional space, and uses the stress value as an indicator for the best solution. A distance matrix was calculated for the fatty acids, and the

samples are ordinated based on the distances calculated (McCune and Grace 2002). This technique will allow visual confirmation of the PERMANOVA results.

<div align="center">

Results

</div>

Lipid content and growth

The lipid content of liver tissue ranged from 2.14 – 13.45% (n=45) and from 5.63 – 25.67% (n=46) for belly flap samples. Belly flap had consistently higher levels of lipid when compared to liver samples within a sampling day (Fig. 1). Although the average wet weight of fish increased throughout the experiment (Fig. 2), the lipid content of both tissues increased dramatically after the diet was switched to Atlantic menhaden (day 71 and 88), indicating the diet was the primary source of variation. The slope of weight gain in striped bass within this experiment increased starting on day 28, which was prior to the diet switch to menhaden (Fig. 2).

Fatty acids

A total of 39 fatty acids, and one fatty acid complex were identified and quantified in this experiment. The 11 fatty acids: C14:0, C16:0, C16:1n7, C18:0, C18:1n9, C18:1n7c, C18:2n6, C20:4n6, C20:5n3, C22:5n3, and C22:6n3, accounted for 85-90% of all fatty acids seen in the liver and adipose tissue samples (Fig. 3). These fatty acids accounted for 83% of the total fatty acids in the spot samples (Fig. 3). These fatty acids were also abundant in livers and belly flaps removed from striped bass along with the menhaden used as the diet. The percent of total for these fatty acids ranged from 85-89% of the fatty acids identified in striped bass tissues on days 71 and 88. However, in menhaden, the value was 77.54% and fatty acids others than those listed above were important, including C18:4n3 and C18:3n3.

Fatty acid groupings (saturated, monounsaturated, omega-3, and omega-6) fluctuated throughout the experiment, regardless of the feed. When striped bass were fed spot, both liver and belly flap saturated fatty acids were at lower levels than those seen in spot. Monounsaturated fatty acids were at much higher levels in the striped bass than in spot (26-36% and 22% respectively). Omega-3 fatty acids levels were similar between liver and spot, while belly flap levels increased throughout the experiment. Both tissues experienced an initial increase in omega 6 fatty acids from day 0 to 12. Starting on day 28, the proportion of these fatty acids decreased.

The diet switch from spot to menhaden also caused changes in the groups of fatty acids in striped bass tissues. The saturated fatty acids decreased in the liver, while increasing in the belly flap. Monounsaturated fatty acids were at much higher levels in striped bass tissues than spot. In both tissues, the omega-3 fatty abundance increased and omega 6 abundance levels decreased.

The PERMANOVA revealed a significant interaction between the sampling day and the type of tissue ($F_{5,122} = 6.76$, $p < 0.001$). Therefore, comparisons were made based upon subsets of the data (Table I). Liver and belly flap were significantly different in fatty acid signature from one another on every sampling day (6 comparisons, $p < 0.01$). Both tissues types were always significantly different than the diet of spot (8 comparisons, $p < 0.001$) or menhaden (4 comparisons, $p < 0.01$).

The patterns of change within a tissue were different between the liver and the belly flap (Table I). The liver fatty acid signature was not significantly different among days 0, 12, and 28. The signature is also not significantly different between days 28 and 40, while day 40 is different than day 0 and 12. This suggests that day 28 is a transition signature and that change is occurring. All liver samples collected during feeding of spot were significantly different than liver samples collected when striped bass were fed menhaden. For the belly flap, day 0 was

significantly different than all other sample days. Days 12, 28, and 40 were not different from each other. After the diet was switched to menhaden, the belly flap samples were different than all samples collected when striped bass were fed spot. The fatty acid signature of liver and belly flap samples collected on day 71 and 88 were not different from each other, but were significantly different than all other sampling days.

The nonmetric multidimensional scaling (NMDS) demonstrates that the fatty acid signatures of the striped bass samples are changing during the experiment (Fig. 4, stress 10.42). Belly flap and liver samples exhibit different patterns from each other. However, the within tissue pattern is similar in plots of axis 1 vs. 2 and 1vs. 3. The belly flap signature changes as the experiment progresses. The belly flap changes to closer approximate spot during the first six weeks of the experiment, and when the diet is switched, the signature changes and starts to mimic that of menhaden. The signature of the liver samples is not as clear. The signature starts to change to closely approximate spot from day 0 – 12. Signatures on day 28 and day 40 start to move away from the spot signature and back to the day 0 signature. Once the diet is switched to menhaden, the signature changes to approximate the fatty acid signature seen on day 0, and then becomes distinct from all other samples, including the diet item.

Because the overall signature of striped bass tissues and prey were always significantly different, individual fatty acids were compared between menhaden and spot to establish unique marker fatty acids. Menhaden and spot contained the same component fatty acids, but the abundances differed between the two prey types (Fig. 5a). Striped bass tissues were then analyzed to determine if prey marker fatty acids were present (Figs. 5b and 5c). Although the tissue experienced fluctuations while being fed spot, the difference is clearly visible when the diet was switched to menhaden. The fatty acids C15:0, C18:3n3, C18:4n3, C19:0, C20:1n12,

C20:1n7, and C20:2n6 exhibited differing values when spot and menhaden tissues were compared. These differences were then transferred to the striped bass tissues, and helped distinguish the diet switch. The fatty acid C19:0 was particularly useful because it was absent from menhaden samples. This fatty acid was removed from striped bass tissues within three weeks of the diet change. Some fatty acids are stored differently between the two tissue types. The liver exhibited lower levels of C15:0 than observed in spot, while the belly flap showed higher levels of C18:3n3 than seen in menhaden.

Discussion

Menhaden and spot have distinct and measurable affects on striped bass tissues. The fatty acid signature of striped bass liver and belly flap samples exhibited fatty acids indicative of a prey switch, from spot to menhaden, after a period of three weeks. Studies have shown that the fatty acids seen in the predator will mimic the fatty acids seen in the prey (Seaborn et al. 2000, Morias et al. 2001, Turner and Rooker 2005, Martinez et al. 2009). This change in fatty acid signature from one diet item to another has been quantified within a period of three weeks for cod *Gadus morhua* Linnaeus 1758 (Kirsch et al. 1998). Although change occurred in the fatty acids of striped bass throughout the experiment, regardless of the diet, certain fatty acids exhibited a distinct change within 31 days of when the diet was switched to menhaden.

Changes in lipid levels were evident in both tissues, and both tissues increased the percentage of lipid after the switch from spot to menhaden. Kirsch et al. (1998) found cod fed a low lipid diet item, squid (2.0% dry weight), did not increase the total fat levels. In our experiment, the lipid level of spot was 6.05 % (wet weight) while the lipid level of menhaden was 11.75 % (wet weight), and this change in prey lipid level was easily seen in striped bass

tissues. Lipid storage depots vary among teleost fish, with possible locations including mesenteric fat, skeletal muscle, and liver (Sheridan 1988). While the pattern of lipid deposition is extremely similar between liver and belly flap in striped bass (both tissues increased lipid levels when fed menhaden), more lipid is stored in the belly flap than the liver.

Diet of striped bass appeared to be the main cause of lipid change within tissues tested. The lipid levels within the liver and belly flap appear to be relatively stable even while the fish is growing. The amount of lipid present in the tissue increases dramatically when the diet is switched to a high lipid prey which is a common result seen in fish (Refstie et al. 2001, Boujard et al. 2004). All energy not used immediately will go into storage for processes like metabolism and reproduction. This shows that the diet of striped bass and other top predators has a major impact on physiological processes. A less nutritious diet will lead to lower energy reserves, which in turn could lead to reduced growth, reduced reproductive output, and possibly mortality. Therefore, tracking the value of the diet becomes an important step along with tracking the diet itself.

The wet weight of striped bass increased dramatically starting on day 28. This increase cannot be assigned to the diet switch from spot to menhaden, because the switch did not take place until day 40. Variables such as temperature and salinity did not change over the course of the experiment, and do not explain the difference seen. Striped bass were observed to be feeding for the duration of the experiment, and therefore were assumed to have been acclimated to the tank system. Water was constantly flowing through the tanks to limit build-up of waste products. Although the density of striped bass in the tanks was reduced as fish were sampled, the density of fish remained below 0.5 lbs•g^{-1} (Lasordo et al. 1998); and was 0.1 lbs•g^{-1} after day 28, when

the increase in growth was seen. Therefore, some factor other than density and water quality seems to have led to a change in weight gain in striped bass in this experiment.

The major fatty acids seen in both tissues were similar but there appears to be a difference in where certain fatty acids are located at increased concentrations. Even as the experiment progressed and fatty acid inputs were changing due to a new prey source, some patterns remained intact. For example, C14:0 and C16:1n7 made a larger contributions to the belly flap fatty acid signature, while C20:4n6 and C22:6n3 had a higher mass percent in liver samples until the diet was switched to menhaden. The levels of C22:6n3 are similar in both spot and menhaden, but different values are present in the tissues depending on the prey type. The transport and storage of fatty acids may influence the tissue signature. Fatty acids are transported via lipoproteins within the lymphatic system (Tocher 2003) to storage sites after re-esterification into triacylglycerols (Iverson 2009). This process may not deliver fatty acids in equal quantities to every tissue; and this process may be based more on the needs of the tissue for a particular fatty acid.

Fatty acids will remain in storage tissue until needed for energy but the oxidation process is not uniform among all fatty acids. Selective metabolism of fatty acids is influenced by molecular structure, a process documented in rats (Leyton et al. 1987, Raclot and Groscolas 1993), humans (Delany et al. 2000), and rabbits (Connor et al. 1996). Although these studies were not focused on fish, the mechanisms are likely to be similar. This difference in oxidation may be responsible for the difference in abundance of certain fatty acids between the tissue types. Results from the literature that compared liver and muscle were not always consistent with the results from this experiment. Abundance values were similar between liver and muscle tissues in largemouth bass *Micropterus salmoides* (Lacepede 1802; Subhadra et al. 2006) and

white seabream *Diplodus sargus* (Valenciennes 1830; Cejas et al. 2004). The gilthead sea bream *Sparus aurata* (Linnaeus 1758) did show differences similar to the values seen in this experiment when liver and muscle tissues were compared (Montero et al. 2001). The values obtained in this experiment suggest that certain fatty acids in adipose tissue, i.e. C22:6n3, may be oxidized more readily than other fatty acids and fatty acids such as C18:1n9 may not be as beneficial for energy as they are accumulated above the values seen in both prey items.

There was a clear pattern of change seen in each tissue from the start of the experiment through the terminus. It was expected that quantified fatty acids would change to match the fatty acid pattern seen in the prey (Pollierer et al. 2010). Studies suggest that predator signatures should mimic prey species around three weeks (Kirsch et al. 1998).This did not occur, as significant differences were always obtained when spot signatures were compared to both tissues sampled in striped bass. Despite significant differences in overall fatty acid signature, there are marker fatty acids which can help to identify a prey source within a period of about 31 days. The NMDS results indicate that there is a constant change occurring within both tissues regardless of prey type. When individual fatty acids are analyzed, we see that there are indeed differences within the tissues when the prey is changed. The fatty acid C19:0 provides the best example. Although this fatty acid is only present in small amounts in spot (0.58%), these trace amounts are still seen in the striped bass tissue. This fatty acid is not present within menhaden, and we see this fatty acid removed from striped bass tissues within 31 days of fish being fed menhaden. This pattern also emerges when other fatty acids are analyzed but does not hold true for every fatty acid consumed.

Although neither tissue fully approximated the signature of the diet item, the possibility of identifying diet items from striped bass tissues does appear feasible. The entire fatty acid

signature is not necessarily a good indicator of diet, although it does provide information on the change in signature in general. Both the liver and the belly flap exhibited changes in the fatty acid signature as the diet was changed. Although either tissue appears to be a candidate for fatty acid signature analysis, only the belly flap offers the possibility of non-lethal sampling. A technique would need to be developed to sample a small portion of the tissue without allowing for infection. This type of sampling has been performed on marine mammals where a sample of lipid is obtained by removing a tissue plug from the blubber layer (Bradshaw et al. 2003, Walton and Pomeroy 2003, Wetzel and Reynolds 2004). Laparoscopic surgery has been performed on shortnose sturgeon *Acipenser brevirostrum* (Lesueur 1818), Atlantic sturgeon *A. oxyrinchus oxyrinchus* (Mitchill 1815; Matsche et al. 2011) and on tench *Tinca tinca* (Linnaeus 1758; Macri et al. 2011) to sample reproductive tissues. This minimally invasive technique provides a method for internal tissue sampling and is non-lethal. Laparoscopic surgery may be adapted to sample fish tissues to track fatty acid changes, but may not be cost-effective.

Striped bass in the wild will not consume only one prey item. Thus, it becomes imperative to develop models that allow for multiple prey species within the diet. An important first step is to provide an accurate method for identifying the process of fatty acid incorporation into striped bass tissues, and how the signature changes during a diet shift. This study has shown that individual prey items contain fatty acid trophic markers (FATMs), which can be used to distinguish among items consumed. This information can be used to model the proportions of each diet item consumed (Iverson et al. 2004), and in turn, allow for more accurate estimates of diet.

Acknowledgments

Literature cited

Adams, S. M. (1976). Feeding ecology of eelgrass fish communities. *Transactions of the American Fisheries Society* **105**, 514-519.

Anderson, M.J. (2005). PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance. Department of Statistics, University of Auckland

Bowen, S.H. (1996). Quantitative description of the diet. In *Fisheries Techniques* (Murphy, B.R. and Willis, D.W., eds.), pp. 513-532. Bethesda, MD: American Fisheries Society

Boujard, T., A. Gelineau, G. Corraze, G. Dutto, E. Gasset, & S. Kaushik. (2004). Regulation of feed intake, growth, nutrient, and energy utilization in European sea bass (*Dicentrarchus labrax*) fed high fat diets. *Aquaculture* **231**, 529-545.

Bradshaw, C. J. A., M.A. Hindell, N.J. Best, K.L. Phillips, G. Wilson, & P.D. Nichols. (2003). You are what you eat: describing the foraging ecology of southern elephant seals (*Mirounga leonina*) using blubber fatty acids. *Proceedings of the Royal Society of London B* **270**, 1283-1292.

Cejas, J.R., E. Almansa, S. Jěrez, A. Bolaňos, M. Samper, & A. Lorenzo. (2004). Lipid and fatty acid composition of muscle and liver from wild and captive mature female broodstocks of white sea bream, *Diplodus sargus. Comparative Biochemistry and Physiology Part B* **138**, 91-102.

Christie, W.M. (2003). *Lipid Analysis*. Bridgewater, England: PJ Barnes and Associates.

Connor, W.E., D.S. Lin, & C. Colvis. (1996). Differential mobilization of fatty acids from adipose tissue. *Journal of Lipid Research* **37**, 290-298.

Currin, B. M., J.P. Reed, & J.M. Miller. (1984). Growth, production, food consumption, and mortality of juvenile spot and croaker: a comparison of tidal and nontidal nursery areas. *Estuaries* **7**, 451-459.

Dalsgaard, J. M. S. J., G. Kattner, D. Muller-Navarra, and W. Hagen. (2003). Fatty acid trophic markers in the pelagic marine environment. *Advances in Marine Biology* **46**, 225-340.

Delany, J.P., M.M. Windhauser, C.M. Champagne, & G.A. Bray. (2000). Differential oxidation of individual dietary fatty acids in humans. *American Journal of Clinical Nutrition* **72**, 905-911.

Elliot, J.M. (1972). Rates of gastric evacuation in brown trout, *Salmo trutta* L. *Freshwater Biology* **2**, 1-18.

Folch, J., M. Lees, & G.H. Sloane Stanley. (1957). A simple method for the isolation and purification of total lipids from animal tissues. *The Journal of Biological Chemistry* **276**, 497-509.

Gannon, J.E. (1976). The effects of differential digestion rates of zooplankton by alewife, *Alosa pseudoharengus*, on determinations of selective feeding. *Transactions of the American Fisheries Society* **105**, 89-95.

Gurr, M.I. & J.L. Hardwood. (1991). *Lipid biochemistry: an introduction*. 4th edn. London, England: Chapman and Hall.

Hartman, K.J. & S.B. Brandt. (1995). Trophic resource partitioning, diets, and growth of sympatric estuarine predators. *Transactions of the American Fishery Society* **124**, 520-537.

He, E. & W.A. Wurtsbaugh. (1993). An empirical model of gastric evacuation rates for fish and an analysis of digestion in piscivorous brown trout. *Transactions of the American Fisheries Society* **122**, 717-730.

Iverson, S. J. (2009). Tracing aquatic food webs using fatty acids: from qualitative indicators to quantitative determination. In *Lipids in aquatic ecosystems* (Arts, M.T., M.T. Brett, & M.J. Kainz eds). pp 281-307. New York, NY: Springer Science.

Iverson, S.J., J.P.Y. Arnold, & I.L. Boyd. (1997b). Milk fatty acid signatures indicate both major and minor shifts in the diet of lactating Antarctic fur seals. *Canadian Journal of Zoology* **75**, 1881-97.

Iverson, S.J., K.J. Frost, & L.F. Lowry. (1997a). Fatty acid signatures reveal fine scale structure of foraging distribution of harbor seals and their prey in Prince William Sound, Alaska. *Marine Ecology Progress Series* **151**, 255-271.

Iverson, S.J. C. Field, W.D. Bowen, & W. Blanchard. (2004). Quantitative fatty acid signature analysis: a new method of estimating predator diets. *Ecological Monographs* **74**, 211-235.

Jacobs, J. M., M.R. Rhodes, B. Martin, D. McIntosh, W.F. Van Heukelem, & R.M. Harrell. (2008). Tissue-specific sampling for estimation of striped bass whole-body proximate composition. *Transactions of the American Fisheries Society* **137**, 386-392.

Jobling, M. (1994). *Fish Bioenergetics*. London, England: Chapman and Hall.

Kirsch, P. E., S.J. Iverson, W.D. Bowen, S.R. Kerr, & R.G. Ackman. (1998). Dietary effects on the fatty acid signature of whole Atlantic cod (Gadus morhua). *Canadian Journal of Fisheries and Aquatic Sciences* **55**, 1378-1386.

Lasordo, T.M., M.P. Masser, and J. Rakocy. (1998). Recirculating aquaculture tank production systems: an overview of critical considerations. Southern Regional Aquaculture Center Publication No. 451. 6pgs.

Leyton, J., P.J. Drury, & M.A. Crawford. (1987). Differential oxidation of saturated and unsaturated fatty acids in vivo in the rat. *British Journal of Nutrition* **57**, 383-393.

Macri, F., G. Rapisarda, G. Marino, M. De Majo, & G. Aiudi. (2011). Use of laparoscopy for the evaluation of the reproductive status of tench (*Tinca tinca*). *Reproduction in Domestic Animals* **46**, 130-133.

Martinez, I., I.B. Standal, D.E. Axelson, B. Finstad, & M. Aursand. (2009). Identification of the farm origin of salmon by fatty acid and HR ^{13}C NMR profiling. *Food Chemistry* **116**, 766-773.

Matsche, M.A., R.S. Bakal, & K.M. Rosemary. (2011). Use of laparoscopy to determine sex and reproductive status of shortnose (*Acipenser brevirostrum*) and Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*). *Journal of Applied Ichthyology* **27**, 627-636.

McCune, B. & J.B. Grace. (2002). *Analysis of Ecological Communities*. Gleneden Beach, OR: MjM Software Design.

Montero, D., L.E. Robaina, J. Socorro, J.M. Vergara, L. Tort, & M.S. Izquierdo. (2001). Alteration of liver and muscle fatty acid composition in gilthead seabream (*Sparus aurata*) juveniles held at high stocking density and fed an essential fatty acid deficient diet. *Fish Physiology and Biochemistry* **24**, 63-72.

Morias, S., J.G. Bell, D.A. Robertson, W.J. Roy, & P.C. Morris. (2001). Protein/lipid ratios in extruded diets for Atlantic cod (*Gadus morhua* L.): effects on growth, feed utilization, muscle composition, and liver histology. *Aquaculture* **203**, 101-119.

Njinkoue, J.M., G. Barnathan, J. Miralles, E.M. Gaydou, & A. Samb. (2002). Lipids and fatty

acids in muscle, liver, and skin of three edible fish from the Senegalese coast: *Sardinella

maderensis*, *Sardinella aurita*, and *Cephalopholis taeniops*. *Comparative Biochemistry

and Physiology Part B: Biochemistry and Molecular Biology* **131**, 395-402.

Pollierer, M.M., S. Scheu, & D. Haubert. (2010). Taking it to the next level: trophic transfer of

marker fatty acids from basal resources to predators. *Soil Biology and Biochemistry* **42**,

919-925.

Raclot, T. & R. Groscolas. (1993). Differential mobilization of white adipose tissue fatty acids

according to chain length, unsaturation, and positional isomerism. *Journal of Lipid

Research* **34**, 1515-1526.

Recks, M.J. and G.T. Seaborn. (2008). Variation in fatty acid composition among nine forage

species from a southeastern US estuarine and nearshore coastal ecosystem. *Fish

Physiology and Biochemistry* **34**, 275-287.

Refstie, S., T. Storebakken, G. Baeverfjord, & A.J. Roem. (2001). Long-term protein and lipid

growth of Atlantic salmon (*Salmo salar*) fed diets with partial replacement o fish meal by

soy protein products at medium or high lipid level. *Aquaculture* **193**, 91-106.

Schafer, R. H. (1970). Feeding habits of striped bass from the surf waters of Long Island. *New

York Fish and Game Journal* **17**, 1-17.

Seaborn, G. T. M. L. J., & T.I.J. Smith. (2000). Differentiation between cultured hybrid striped

bass and wild striped bass and hybrid bass using fatty acid profiles. *North American

Journal of Fisheries Management* **20**, 618-626.

Seaburg, K. G. (1957). A stomach sampler for live fish. *The Progressive Fish Culturist* **19**, 137-

139.

Sheridan, M.A. (1988). Lipid dynamics in fish: aspects of absorption, transportation, deposition, and mobilization. *Comparative Biochemistry and Physiology Part B* **90**, 679-690.

Subhadra, B., R. Lochmann, S. Rawles, & R. Chen. (2006). Effect of dietary lipid source on the growth, tissue composition and hematological parameters of largemouth bass (*Micropterus salmoides*). *Aquaculture* **255**, 210-222.

Tocher, D. R. (2003). Metabolism and functions of lipids and fatty acids in teleost fish. *Reviews in Fisheries Science* **11**, 107-184.

Turner, J. P., & J. R. Rooker. (2005). Effect of dietary fatty acids on the body tissues of larval and juvenile cobia and their prey. *Journal of Experimental Marine Biology and Ecology* **322**, 13-27.

USEPA (United States Environmental Protection Agency). (2007). Pressurized fluid extraction (PFE). USEPA SW-846. Method 3545A, Update IV. Washington, D.C.

Walton, M., & P. Pomeroy. (2003). Use of blubber fatty acid profiles to detect inter-annual variations in the diet of grey seals *Halichoerus grypus*. *Marine Ecology Progress Series* **248**, 257-266.

Wetzel, D. L., & I. J.E. Reynolds. (2004). Definitive identification of fatty acid constituents in marine mammal tissues. *Canadian Journal of Fisheries and Aquatic Sciences* **61**, 554-560.

Electronic References

R Development Core Team. 2010. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Table I. Results from the PERMANOVA performed on the fatty acid data from each sample. The entire fatty acid signature from each group was compare to all other groups. Asterisks represent a significant difference in overall signature.

	Spot	Liv 0	Liv 12	Liv 28	Liv 40	Liv 71	Liv 88	BF 0	BF 12	BF 28	BF 40	BF 71	BF 88
Spot													
Liv 0	***												
Liv 12	***	ns											
Liv 28	***	ns	ns										
Liv 40	***	**	***	ns									
Liv 71	***	***	***	*	***								
Liv 88	***	***	***	*	**	ns							
BF 0	***	***											
BF 12	***		***					***					
BF 28	***			***				***	ns				
BF 40	***				***			***	ns	ns			
BF 71	***					***		***	***	***	*		
BF 88	***						**	***	***	***	**	ns	
Menhaden	***	***	***	***	***	**	***	***	***	***	***	**	**

* $p < 0.05$

** $p < 0.01$

*** $P < 0.001$

ns = not significant
A blank space indicates comparison was not tested

Figure 1. Fat contents of the liver (■) and belly flap (♦) of striped bass analyzed. Spot was fed starting on day 0 until day 40, and the diet was switched to menhaden on day 40 as indicated by the vertical line.

Figure 2. Average wet weight of striped bass from each sampling day. Spot was fed starting on day 0 until day 40, and the diet was switched to menhaden on day 40 as indicated by the vertical line.

Figure 3. Average mass percent of the major fatty acids detected in menhaden (▢) and spot (▬) and striped bass tissues: a) liver samples, and b) belly flap samples on days 0 (⊠), 12 (⊟), 28 (⬚), 40 (⬚), 71 (▦), and 88 (◨). Error bars represent 1 standard error of the mean.

Figure 4. Non-metric multidimensional scaling (NMDS) plot of striped bass tissue samples and the prey items, spot and menhaden. A plot of a) axes 1 and 2 and b) axes 1 and 3 was needed to reduce the stress value (10.42). The alphanumeric code refers to the tissue (Liv = liver and BF = belly flap) and the day on which the sample was collected (i.e. 0 = day 0, 40 = day 40, etc.). The two prey species are spot and menhaden (MH).

Figure 5. Patterns in abundance of select fatty acids. Abundance is based upon mass percent of total fatty acids. The diet was changed from spot to menhaden on day 40. Solid bars represent lipid masses during the time when striped bass were fed spot, while pattern bars represent lipid masses after switching the diet to menhaden. Part a) spot (solid

bars) and menhaden (patterned bars) fatty acids, b) liver fatty acids for days 0, 12, 28, 40, 71, and 88 and c) belly flap fatty acids for days 0, 12, 28, 40, 71, and 88.

Figure 1.
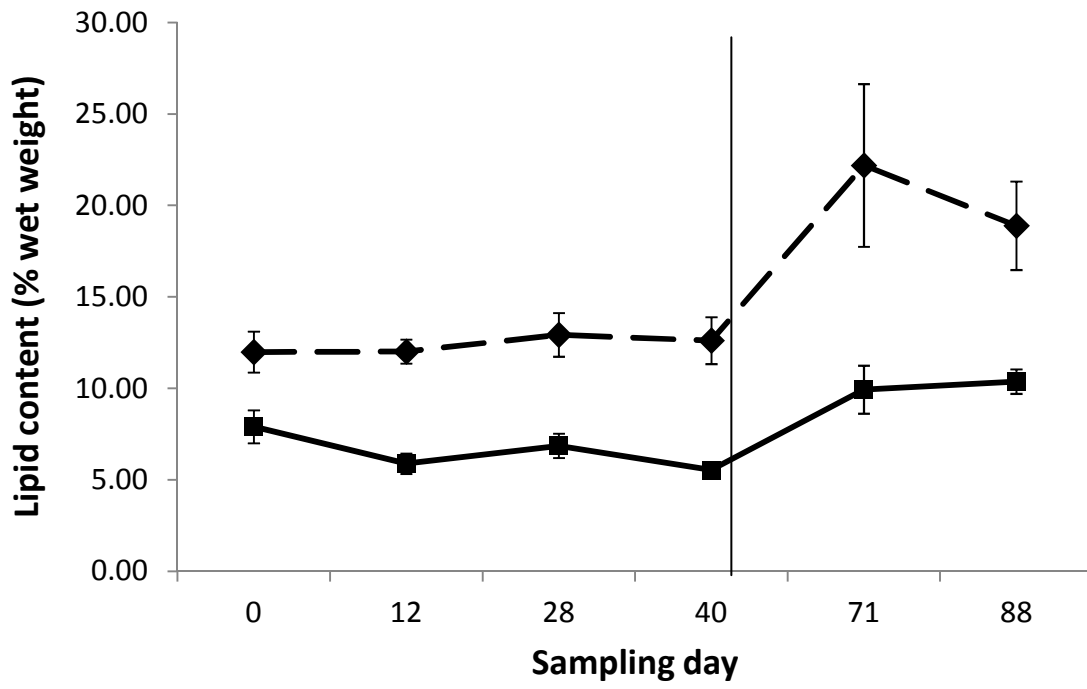
Figure 2.
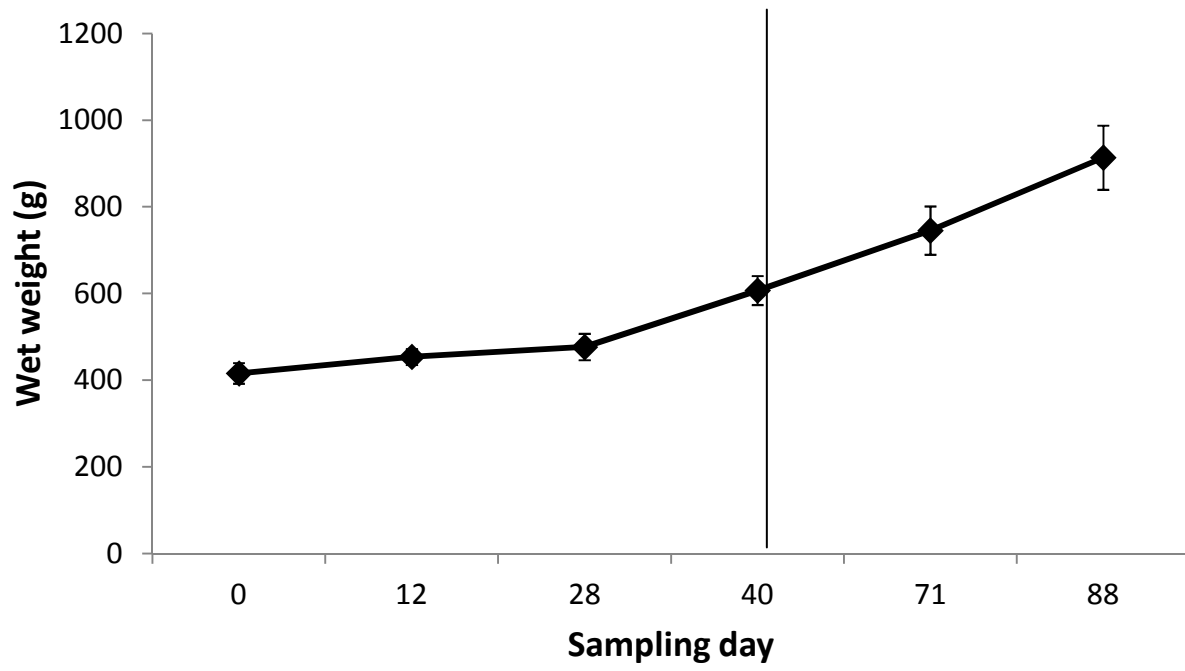
Figure 3.
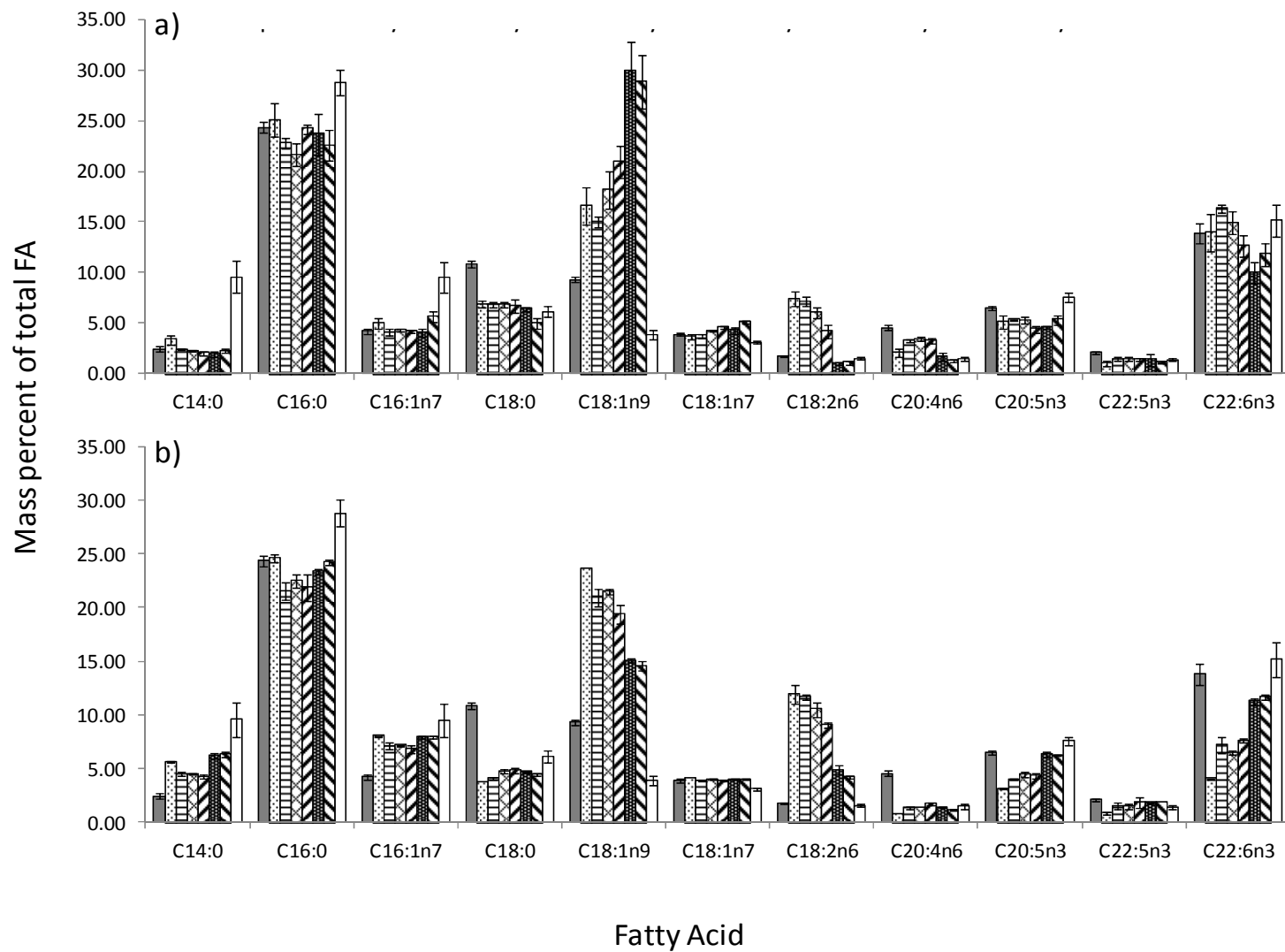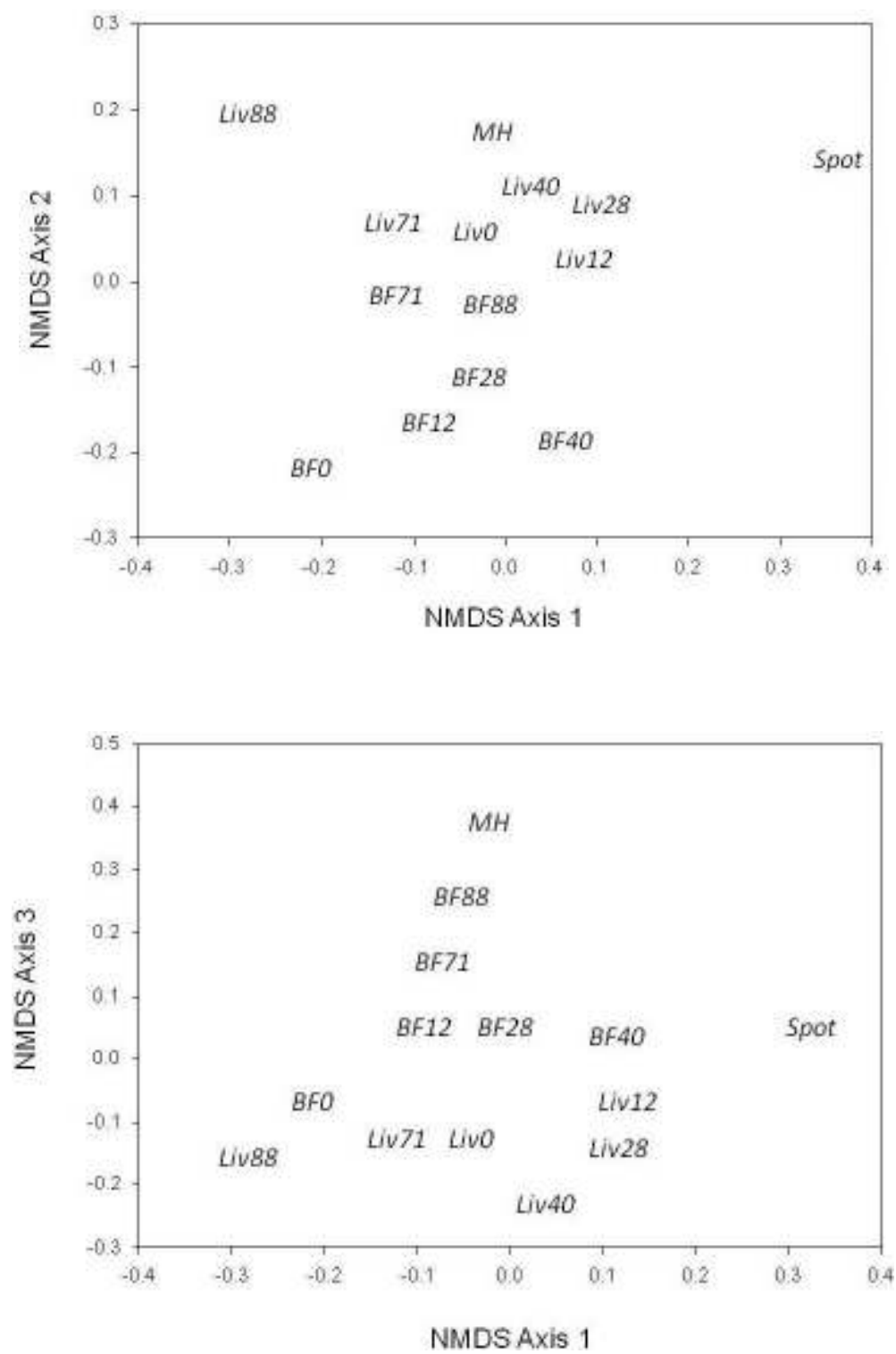
Figure 4.

Figure 5.
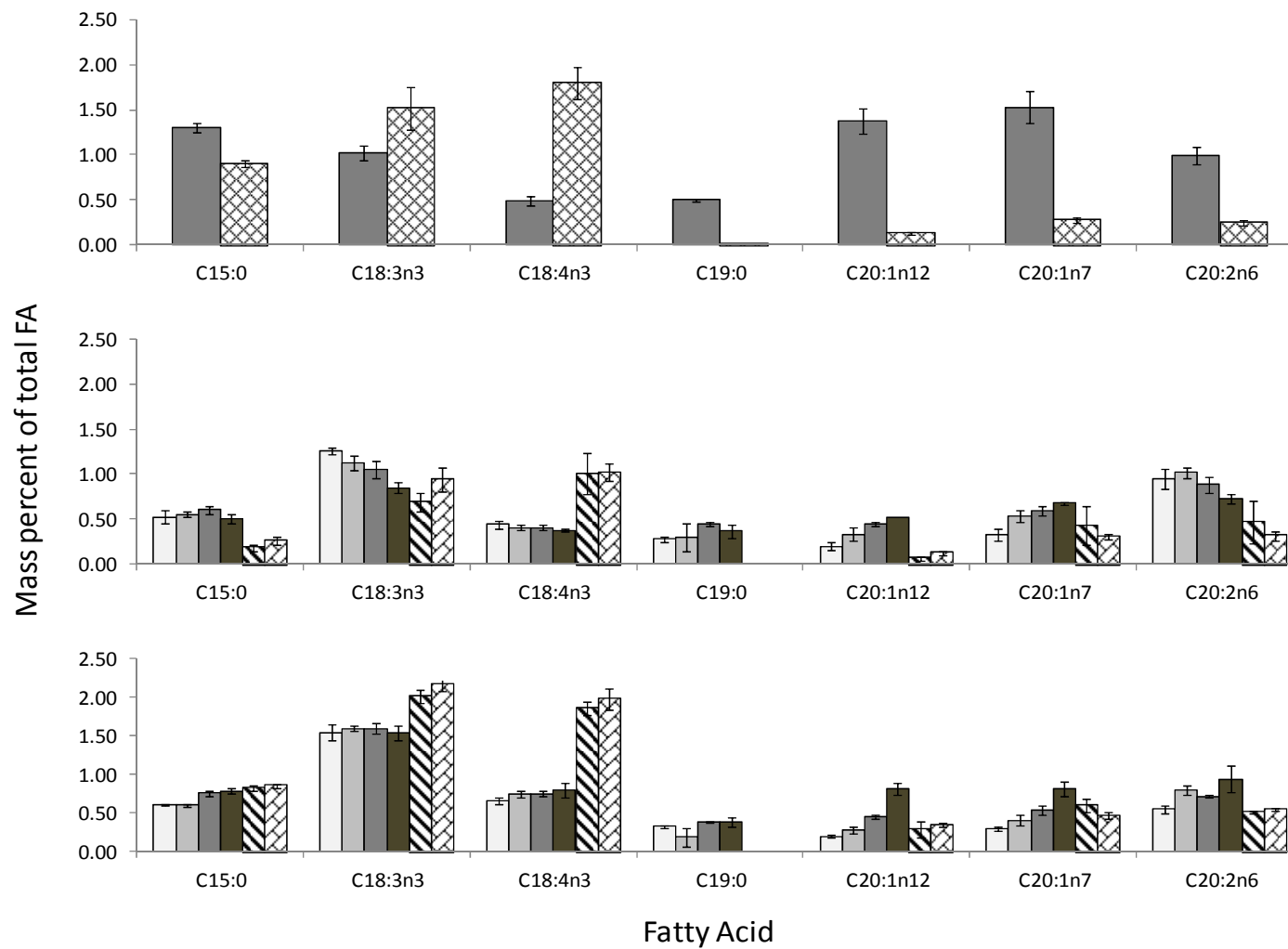
Chapter 4 - The effect of striped bass size on fatty acid signature incorporation

Abstract

Fatty acids are a group of molecules that are transferred from prey to predator and deposited with little to no modification. Analysis of fatty acids has been used to elucidate the trophic web within aquatic systems. However, little research has been conducted to determine the effects of organism size on how fatty acids are incorporated within a species. Striped bass (*Morone* saxatilis) of three different sizes were fed a baseline diet of spot (*Leiostomus xanthurus*) for a period of four weeks then fed an experimental prey of Atlantic menhaden (*Brevoortia tyrannus*) for four weeks. All feeding was done every other day to satiation. Indicator fatty acids that distinguished menhaden from spot were unable to demonstrate a signature shift away from baseline within any of the striped bass size groups. A lack of feeding due to low temperatures for part of the experiment and a failure of larger fish to feed at adequate levels are most likely the causes for a lack of signature change. This experiment helps to demonstrate that fatty acid signature analysis may not provide an accurate record of recent diet when fish are feeding at low levels.

Introduction

The size of a fish will influence not only what is consumed, but how the diet is processed. Larger fish have larger mouths than their smaller conspecifics, allowing them to consume a broader range of prey items. Anadromous striped bass (*Morone saxatilis*) in the Chesapeake Bay provide a good example of different diet regimes for different life stages. Juveniles will consume prey indicative of the salinity regime, e.g. oligochaetes and insect larvae are more important in freshwater; and amphipods, mysids, and fish larvae are more important in higher salinity waters (Boynton et al. 1981). Striped bass will become primarily piscivorous by age 2 (Hartman and

Brandt 1995a). Once the meal has been consumed, the energy derived is devoted to metabolic costs, including growth, and the remainder will be devoted to energy storage. Smaller fish tend to put more energy towards growth to escape predation while larger individuals will put more energy towards storage for use in stressful times (Jobling 1994).

Traditional methods for estimating diets in fish have involved sacrificing the fish or flushing the stomach with water and enumerating the prey items in the stomach (Seaburg 1957). Estimations of diet were based on various metrics including (but not limited to): percent composition by weight or number and frequency of occurrence. From these metrics, indices of preference have been constructed based upon prey taxa prevalence in the stomach and the environment (Bowen 1996). Due to confounding factors such as the presence of chitinous material, prey size, and water temperature (Elliot 1972, Gannon 1976, He and Wurtsbaugh 1993), the rates of digestion for prey taxa are not identical. Therefore, estimations of diet and preference are likely biased, and a more accurate method is needed.

The main constituents of lipids in fish are fatty acids, which hold promise as a chemical marker for tracking diets (Gurr and Hardwood 1991). These fatty acids are directly obtained through the diet and are deposited within the organism with little to no modification (Iverson 1997). They can be tracked as they are cycled through the food web (Iverson 1997), and prey taxa consumed can be identified from the fatty acids present and their relative proportions with respect to other fatty acids (Iverson 2004). Fatty acid analysis offers a good candidate for tracking feeding history for fishes.

One factor that may complicate diet studies is the nutritional needs of various age classes. Juvenile striped bass must consume enough energy to make it through the winter and at the same time grow quickly enough to escape predation by other species. In general, smaller fish put more

energy towards growth rather than lipid deposition when compared with adult fish (Jobling 1994). Therefore, the fatty acids deposited in the lipid reserves of this species could vary between different age classes of striped bass. Iverson et al. (2002) showed that length of fish was an important indicator of the mass percent of C20:1n11 (gadoleic acid). Therefore, the question of how different size striped bass incorporate fatty acids needs to be clarified.

Objectives

Different sized fish allocate energy differentially to somatic and gonadal tissue. To evaluate how this may impact fatty acid signature incorporation, we conducted lab experiments with striped bass of three different sizes. Fish were fed the same prey type at the same relative ration (satiation every other day). By standardizing the feeding regime, the process of fatty acid storage across fish size can be determined.

Materials and Methods

Sample collection and tank experiment

Forty seven striped bass were placed into 800 gallon tanks based upon their length: 16 fish between 150 – 200 mm (Delmarva Aquatics: Chesapeake-Delaware strain), 16 fish between 300 – 380 mm, and 15 fish greater than 460 mm. The medium and large sizes of striped bass were collected from Chesapeake Bay tributaries near Oxford, Maryland by hook and line. These fish were transported by boat in a live well to the NOAA/NCCOS/Oxford laboratory in Oxford, Maryland. The tanks were flow through with water being pumped from the Tred Avon River. Water temperature was measured daily (Fig. 1). Fish in each tank were fed a diet of spot (*Leiostomus xanthurus*) *ad libitum* every other day for four weeks to approximate 50% C_{max} and

homogenize the fatty acid signature among all sizes (Fig. 2). After four weeks, the striped bass were fed Atlantic menhaden (*Brevoortia tyrannus*) *ad libitum* every other day for four weeks. Four striped bass were sacrificed from each tank on day 0 and 13 with an overdose of tricaine methanesulfonate (MS-222; Western Chemical, Ferndale, WA), weighed (g) and measured (total length in mm), and vacuum sealed with a FoodSaver vacuum sealer (Sunbeam Products, Inc., Niles, IL) and frozen at -20°C until they could be analyzed. Remaining fish in each tank were sampled on day 28 (eight small fish, eight medium fish, and seven large fish).

Sample preparation

Striped bass were thawed at the time of analysis, and tissue samples were weighed to the nearest 0.1 gram. One sample consisted of three to five grams of adipose tissue located beneath the ribs on the ventral surface, hereafter referred to as belly flap (Jacobs et al. 2008). This tissue has been shown to provide an accurate record of diet in striped bass (Chapter 3). All samples were ground with a chemical drying agent, diatomaceous earth (Dionex, Sunnyvale, CA), and had a known amount of internal standard (C13:0 triacylglycerol) added to determine the efficiency of the extraction process.

Lipids were extracted under nitrogen using an Accelerated Solvent Extractor (ASE 300, Dionex, Sunnyvale, CA) with a mixture of 3:1 (v/v) dichloromethane:methanol and 0.1% butylated hydroxytoluene (BHT) as an antioxidant. Samples were extracted at 100°C and 1500 psi (United States Environmental Protection Agency 2007). Samples were then back-extracted using a 0.88% potassium chloride solution in deionized water to remove non-lipid materials from extracts (Folch et al. 1957). Total lipid extracted was determined gravimetrically for each sample and reported as a percent of wet weight.

Extracted lipid species containing fatty acids as various derivatives were transesterified using a solution of sulfuric acid and methanol. In this process, aclyglycerols are broken apart to their constituents: glycerol and fatty acids methyl esters (FAMEs). Free fatty acids present in the lipid mixture are also converted to FAMEs using this process (Christie 2003). Sample extracts were cleaned with silica gel and anhydrous sodium sulfate to remove polar components and water, which may degrade the performance of the gas chromatograph column. The silica column was conditioned with hexane and the sample was added along with hexane and diethyl ether, 95:5, v/v (Christie 2003). The mixture was concentrated to 5 ml and diluted to a final total lipid concentration of 100 μg/ml. Finished samples were run on a gas chromatograph (Agilent 6890N Network GC System, Santa Clara, CA) / mass spectrometer (GC/MS, Agilent, 5973 Network Mass selective detector, Santa Clara, CA) in select ion monitoring mode.

The GC/MS was used to identify and quantify fatty acid methyl esters in each sample. FAMEs were injected into a fused capillary column, DB-23 60m length X 0.250mm OD X 0.25μm film thickness (Agilent, Santa Clara, CA) using an autosampler (Agilent 7683 Series Auto Sampler). Hydrogen was used as a carrier gas. The temperature program was as follows: hold at an initial temp of $50^{o}C$ for 2 min, hold at $150\,^{o}C$ for 1 min after ramping at $20\,^{o}C \cdot min^{-1}$, ramp at $1.25\,^{o}C \cdot min^{-1}$ until $215\,^{o}C$. The fatty acid retention times were compared to known standards from Nu-Chek Prep (GLC MIX 85, 411, 642, 632, Elysian, MN). These four standard mixtures were combined along with stearidonic acid (C18:4n3, Cayman Chemical, Ann Arbor, MI) to make a total FAME standard solution of 12.25 μg/ml. Matreya 25 FAME mixture (Pleasant Gap, PA) was used to obtain retention times for C18:1n9 trans and C18:2n6 trans. Restek cis/trans FAME mixture (Bellefonte, PA) was used to obtain retention times for C20:1n7. These standards and an internal standard of C13:0 (100 μg/ml) were added to quantify the

amount of each fatty acid methyl ester present. The GC/MS tracks the select ion signature of each compound and was used to verify the identity of each peak. There were peaks which consisted of more than one fatty acid and it was impossible to determine which fatty acid(s) constituted the peaks. Therefore, a term fatty acid "complex" was assigned to this peak. The complex refers to which fatty acids could be responsible for the peak, e.g. $C22:3n3$ and $C22:5n6$.

Statistical analyses

We used an *a priori* model building (generalized linear model in program R [R Development Core Team 2010]) strategy to determine the effects of sampling day, fish weight, and water temperature on the lipid content (as percent of wet weight) of each sample. Models included: 1) a global of all three predictor variables and all interaction terms (including the three interaction term), 2) predictor variables and a single interaction term, 3) a single predictor variable, and 4) a null model that included no predictor variables (Table 1). The Akaike information criterion for small sample size (AIC_C; Burnham and Anderson 2002) was used to select the best model. A plot of residuals demonstrated that the data violated the assumption of normality. Both the continuous variables weight and lipid content were natural log transformed which corrected the violation of normality. All model building and selection was performed in R (R Development Core Team 2010).

Indicator fatty acids were identified for the prey types using the relative mass percent of the total. These values were then compared to tissues samples throughout the experiment to determine if striped bass tissues reflected prey values. This technique distinguished the diet switch from spot to menhaden in an earlier chapter (Chapter 3).

Nonmetric multidimensional scaling (NMDS) was also performed on the data (R Development Core Team 2010). This ordination technique is an iterative search for the best

position of samples in multidimensional space, and uses the stress value as an indicator for the best solution. A distance matrix was calculated for the fatty acids, and the samples are ordinated based on the distances calculated (McCune and Grace 2002).

Results

Lipid content

Lipid content (percent wet weight) in striped bass ranged from 2.64 – 24.66% (n = 42, Fig. 3). Lipid levels decreased from day 0 to 13 for the medium and large striped bass, while the small fish showed an increased in lipid content during this same interval. All three size classes showed an increase in lipid content from day 13 to day 28. Large striped bass had the biggest change, with slight increases in both small and medium striped bass. The regression model with the most weight for explaining lipid content was the null model meaning no factor measured adequately predicted lipid levels (Table 1). Temperature and weight were each better models than sampling day.

A difference in weight within a striped bass size class was difficult to discern as the experiment progressed (Fig. 4). Small and large striped bass showed a small increase in weight from day 0 to 13, followed by slight decrease to day 28. Medium striped bass remained at constant weight from day 0 to 13, and decreased slightly from day 13 to 28.

Fatty acids

A total of 40 fatty acids and one fatty acid complex (C22:3n3 and C22:5n6) were identified in striped bass lipids. The 12 fatty acids C14:0, C16:0, C16:1n7, C18:0, C18:1n9, C18:1n7c, C18:2n6, C20:4n6, C20:5n3, C22:0, C22:5n3, and C22:6n3 accounted for 76.05 – 88.41% of the mass percent of fatty acids. These fatty acids accounted for 84.72% of the mass

percent of fatty acids present in menhaden tissues fed to striped bass (Table 1). These fatty acids decreased in abundance from day 0 to day 28 for all size classes; 87.82 to 76.05% for small fish, 88.41 to 83.76% for medium fish, and 87.09 to 77.27% for large striped bass.

The nonmetric multidimensional scaling indicated that no discernible change was evident in striped bass belly flap tissue as they were fed menhaden (Fig. 5). The samples from each size class were clustered in one area regardless of the sampling day. Samples moving away from the cluster appear to move neither towards menhaden or spot. Two samples, a small striped bass from day 28 and a large fish from day 13, are extreme outliers compared to the rest of the samples.

Relative abundances of certain fatty acids have the ability to distinguish a diet switch in striped bass from a diet of spot to one of menhaden (Chapter 3). When the striped bass tissues were analyzed for these marker fatty acids, no pattern of fatty acid incorporation was apparent (Fig. 6). Many of these indicator fatty acids did not change between the sampling days for all three size classes of striped bass. The fatty acid complex C22:3n3 and C22:5n6 changed its relative abundance for medium striped bass in the opposite direction that the diet fatty acids would indicate. This same fatty acid complex did follow a decreasing pattern in large striped bass.

Discussion

Energy that is yielded from the diet is used for metabolic processes and the remainder is stored within the organism. These storage molecules will contain fatty acid trophic markers (FATMs; Graeve et al. 1994, Dalsgaard et al. 2003) indicative of feeding history, and they should change based upon the prey being consumed (Iverson 2009). Fatty acids within striped

bass belly flap tissue should have recorded the diet change from spot to menhaden during our experiment. It is apparent that other factors, e.g. reduced feeding or temperature, prevented definitive conclusions from being made in this experiment. However, we still were able to gain valuable insight into fatty acid incorporation within varying sizes of striped bass.

Lipid stores accumulate once the energetic needs of the organism have been met first. The Atlantic menhaden feed had higher lipid content (11.75 % wet weight) than spot (6.05% wet weight) which should have resulted in lipid content increases in striped bass tissues (Refstie et al. 2001). Decreases in lipid content were evident after the experiment had progressed for a period of 13 days for two size classes of fish. Removal of lipid stores would suggest that feeding is reduced or absent, and the fish is relying on stored lipids for metabolic costs (Maddock and Burton 1994). Fish that are fed ad libitum should have stable lipid levels, if not show an increase. Jacobs et al. (2008) fed striped bass ad libitum three times a week while keeping the temperature constant at $19^{o}C$, and whole body lipid levels increased on each subsequent sampling period (21 and 42 days). Fish that had a decrease in lipids were fish that were starved for the duration of the experiment. The European seabass (*Dicentrarchus labrax*), also a member of the family Moronidae, exhibited progressive declines in lipids as starvation set in (Stirling 1976). These studies indicate fish that are not feeding will show a concomitant decrease in lipid levels within the body. The results seen in our experiment demonstrate decreases in lipid levels for part of the experiment, which means striped bass were relying on stored lipids. The increase after day 13 suggests that fish began to replace the depleted lipid stores.

According to Hartman and Brandt (1995b), water temperature affects the specific growth rate of striped bass fed at maximum consumption, regardless of size, although larger fish seem to have a broader range of temperatures at which they experience optimal growth. Cox and Coutant

found that juvenile striped bass fed at 60% ration and 16°C still experienced positive growth rates. The incidence of lower growth suggests that temperature may not have been the reason for lack of lipid accumulation and weight gain. Rather, the reduced or absence of feeding may be responsible.

Previous studies of fatty acid signature incorporation have suggested that the predator fatty acids should begin to mimic that of the prey (Seaborn et al. 2000, Turner and Rooker 2005). The change of signature has been shown to occur within two to three weeks (Kirsch et al. 1998). Several fatty acids were at similar levels as they were within spot tissues on day 0 (Fig. 2), but not all fatty acids. This suggests that the signature of the prey was never completely expressed within the predator tissue. We have demonstrated that the entire fatty acid signature of striped bass tissues did not change under the conditions tested, but rather marker fatty acids are indicative of a prey switch (Chapter 3). However, in this experiment, when looking at those marker fatty acids that distinguish between the two prey types, there is not a clear pattern of change in the proportions of these fatty acids in any size of striped bass. There is an exception with two fatty acids, C22:3n3 + C22:5n6 in large fish and C20:2n6 in medium fish. It is difficult to ascertain what constitutes normal fluctuations within each size group without knowing the degree to which individual fish were eating and how temperature affected the fish.

Water visibility within the tanks was low due to unfiltered water being pumped in directly from the Tred Avon River. The initiation and cessation of feeding was difficult to determine due to this factor. Because some of these fish were wild caught, they may never have taken to feeding while housed in the tanks, although it is possible for hook and line captured fish to be habituated to feeding in captivity, e.g. white bass *Morone chrysops* (Kohler et al. 1994). This difficulty highlights a problem that may be prevalent within the bay ecosystem. Wild fish

often will experience degraded conditions or periods of low temperature, in which feeding becomes diminished. Unlike marine mammals which have a large blubber layer, and hence a large fatty acid deposit to call upon, piscivorous fish lipid supplies are most likely not as extensive. This would not be an issue if all fatty acids were used equally. However, during periods of starvation, fatty acids will be differentially mobilized depending on the tissue (Jezierska et al. 1982) which could explain the decreases in striped bass fatty acids that were abundant in menhaden tissues. As the lipid supplies are relied upon more heavily, the signature will become skewed farther away from the original signature.

While the experiment did not yield discernible patterns of fatty acid incorporation, it does offer an opportunity for further research. All three size classes of striped bass were exposed to the same temperature, and provided with the same proportion of food, but they did not respond in a similar manner. Medium and large size classes lost lipid from day 13 to 28 while small fish were able to maintain a slight increase throughout the experiment.

The practice of using fatty acids in food web studies has been used successfully in other studies, including fish. The limiting step of this technique appears to be that fish need to be feeding at sufficiently high rates in order for the fatty acid signature of the prey taxa to become incorporated into predator tissue. Periods of low feeding will cause signatures to be skewed away from the prey, leading to erroneous conclusions about the diet. It is suggested that this technique only be used during bouts of known feeding until more research can be done to determine correction factors for lowered feeding due to stress and low temperatures.

Acknowledgments

Literature cited

Boynton, R.W., T.T. Polgar, and H. Zion. 1981. Importance of juvenile striped bass food habits in the Potomac Estuary. Transactions of the American Fisheries Society 110:56-63.

Burnham, K.P., and D.R. Anderson. 2002. Model selection and multi-model inference. 2nd ed. Springer, Berlin, Germany. 496pp.

Christie, W.M. 2003. Lipid Analysis. PJ Barnes and Associates. Bridgewater, England.

Cox, D. K., and C. C. Coutant. 1981. Growth dynamics of juvenile striped bass as functions of temperature and ration. Transactions of the American Fisheries Society 110:226-238.

Dalsgaard, J. M. S. J., G. Kattner, D. Muller-Navarra, and W. Hagen. 2003. Fatty acid trophic markers in the pelagic marine environment. Advances in Marine Biology 46.

Folch, J., M. Lees, and G.H. Sloane Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. The Journal of Biological Chemistry:497-509.

Graeve, M., G. Kattner, and W. Hagen. 1994. Diet-induced changes in the fatty acid composition of Arctic herbivorous copepods: experimental evidence of trophic markers. Journal of Experimental Marine Biology and Ecology 182:97-110.

Gurr, M.I. and J.L. Hardwood. 1991. Lipid biochemistry: an introduction. 4th ed. Chapman and Hall. London, England.

Hartman, K. J., and S. B. Brandt. 1995a. Trophic resource partioning, diets, and growth of sympatric estuarine predators. Transactions of the American Fisheries Society 124:520-537.

Hartman, K. J., and S. B. Brandt. 1995b. Comparative energetics and the development of
bioenergetics models for sympatric estuarine piscivores. Canadian Journal of Fisheries
and Aquatic Sciences 52:1647-1666.

Iverson, S. J. 2009. Tracing aquatic food webs using fatty acids: from qualitative indicators to
quantitative determination. Pages 281-307 *in* M. T. Arts, editor. Lipids in aquatic
ecosystems. Springer Science.

Iverson, S.J., K.J. Frost, and L.L. Lowry. 1997. Fatty acid signatures reveal fine scale structure
of foraging distribution of harbor seals and their prey in Prince William Sound, Alaska.
Marine Ecology Progress Series 151: 255-271.

Iverson, S. J., K.J. Frost, and S.L.C. Lang. 2002. Fat content and fatty acid composition of forage
fish and invertebrates in Prince William Sound, Alaska: factors contributing to among
and within species variability. Marine Ecology Progress Series 241:161-181.

Iverson, S. J., C. Field, W.D. Bowen, and W. Blanchard. 2004. Quantitative fatty acid signature
analysis: a new method of estimating predator diets. Ecological Monographs 74:211-235.

Jacobs, J. M., M.R. Rhodes, B. Martin, D. McIntosh, W.F. Van Heukelem, and R.M. Harrell.
2008. Tissue-specific sampling for estimation of striped bass whole-body proximate
composition. Transactions of the American Fisheries Society 137:386-392.

Jezierska, B., J.R. Hazel, and S.D. Gerking. 1982. Lipid mobilization during starvation in the
rainbow trout, *Salmo gairdneri*, with attention to fatty acids. Journal of Fish Biology
21:681-692.

Jobling, M. 1994. Fish Bioenergetics. Chapman and Hall. London, England.

Jones, P.W., J.S. Wilson, R.P. Morgan, H.R. Lunsford, and J. Lawson. 1977. Potomac River

fisheries study – striped bass spawning stock assessment – interpretative report for 1974

through 1976. Chesapeake Biological Lab. UMCEES Ref. No. 77-55 CBL.

Kirsch, P. E., S.J. Iverson, W.D. Bowen, S.R. Kerr, and R.G. Ackman. 1998. Dietary effects on

the fatty acid signature of whole Atlantic cod (Gadus morhua). Canadian Journal of

Fisheries and Aquatic Sciences 55:1378-1386.

Kohler, C.C., R.J. Sheehan, C. Habicht, J.A. Malison, and T.B. Kayes. 1994. Habituation to

captivity and controlled spawning of white bass. Transactions of the American Fisheries

Society 123:964-974.

Maddock, D.M. and M.P.M. Burton. 1994. Some effects of starvation on the lipid and skeletal

muscle layers of the winter flounder, *Pleuronectes americanus*. Canadian Journal of

Zoology 72:1672-1679.

Maryland Department of Natural Resources (MD DNR). 1987. Maryland striped bass research.

Final report: Project AFC – 15 Segment 1. September 1987. Tidewater Administration,

Fisheries Division, Annapolis, MD.

McCune, B. and J.B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design,

Gleneden Beach, OR.

R Development Core Team (2008). R: A language and environment for statistical computing. R

Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL

http://www.R-project.org.

Refstie, S. T. Storebakken, G. Baeverfjord, and A.J. Roem. 2001. Long-term protein and lipid

growth of Atlantic salmon (*Salmo salar*) fed diets with partial replacement of fish meal

by soy protein products at medium or high lipid level. Aquaculture 193:91-106.

Seaborn, G. T. M. L. J., and T.I.J. Smith. 2000. Differentiation between cultured hybrid striped

  bass and wild striped bass and hybrid bass using fatty acid profiles. North American

  Journal of Fisheries Management 20:618-626.

Setzler-Hamilton, E. M., and L. H. H. Jr. 1991. Striped bass (*Morone saxitilis*). Pages 13.11-

  13.27 *in* S. L. Funderburk, S.J. Jordan, J.A. Mihursky, and D. Riley, editor. Habitat

  requirements for Chesapeake Bay living resources. Chesapeake Research Consortium,

  Solomons, MD.

Stirling, H.P. 1976. Effects of experimental feeding and starvation on the proximate composition

  of the European bass *Dicentrarchus labrax*. Marine Biology 35:85-91.

Turner, J. P., and J. R. Rooker. 2005. Effect of dietary fatty acids on the body tissues of larval

  and juvenile cobia and their prey. Journal of Experimental Marine Biology and Ecology

  322:13-27.

Uphoff Jr., J. H. 2003. Predator-prey analysis of striped bass and Atlantic menhaden in upper

  Chesapeake Bay. Fisheries Management and Ecology 10:313-322.

United States Environmental Protection Agency (USEPA). 2007. Pressurized fluid extraction

  (PFE). USEPA SW-846. Method 3545A, Update IV. Washington, D.C.

Table 1. Summary of model selection statistics for lipid percent in samples; K represents the number of parameters in the model; ΔAIC_c represents the difference between the AIC_c value for that model and the set's minimum AIC_c value; W represents the Akaike weight of each model.

Model	K	Δ AICc	W
Null	2	0	0.5
T[A]	3	2.24	0.16
We	3	2.3	0.16
D	4	3.21	0.1
D+T	5	5.53	0.03
D+We	5	5.73	0.03
D+We+T	6	8.24	0.01
D+We+T+D*We	8	13.52	0
D+We+T+D*We+D*T+W*T	11	22.03	0
D+We+T+D*We+D*T+We*T+D*We*T	13	28.49	0

[A] Model terms are temperature (T), weight (We), and sampling day (D).

Table 2. Mean fatty acid concentrations expressed as a mass percent of all fatty acids present ± 1 standard error of the mean; nd = not detected. The fatty acids shown are for the prey item, spot, and the tissues collected from striped bass days 0 through 40. Unless specified, all unsaturated fatty acids have cis configuration.

Fatty acid	Menhaden	Small			Medium			Large		
		Day 0	Day 13	Day 28	Day 0	Day 13	Day 28	Day 0	Day 13	Day 28
C12:0	0.26 ± 0.04	0.15 ± 0.04	0.06 ± 0.03	0.44 ± 0.33	0.06 ± 0.02	0.06 ± 0.03	0.10 ± 0.03	0.07 ± 0.04	0.18 ± 0.13	0.10 ± 0.02
C13:0	0.40 ± 0.07	0.40 ± 0.03	0.11 ± 0.07	0.38 ± 0.24	0.26 ± 0.08	0.16 ± 0.11	0.53 ± 0.19	0.53 ± 0.33	0.09 ± 0.09	5.74 ± 5.39
C14:0	7.30 ± 0.51	4.36 ± 0.28	4.43 ± 0.14	3.08 ± 0.62	3.68 ± 0.08	3.99 ± 0.12	3.40 ± 0.29	4.12 ± 0.27	3.97 ± 0.47	3.31 ± 0.81
C14:1n5	0.05 ± 0.02	0.13 ± 0.02	0.07 ± 0.04	0.08 ± 0.03	0.14 ± 0.03	0.19 ± 0.03	0.19 ± 0.06	0.09 ± 0.05	0.46 ± 0.25	0.14 ± 0.03
C15:0	2.07 ± 0.3	1.37 ± 0.11	1.39 ± 0.12	1.25 ± 0.14	1.19 ± 0.05	1.32 ± 0.05	1.23 ± 0.18	1.30 ± 0.08	1.44 ± 0.18	0.87 ± 0.20
C16: 0 DMA	0.51 ± 0.1	0.39 ± 0.03	0.69 ± 0.31	0.45 ± 0.12	0.36 ± 0.09	0.41 ± 0.05	0.53 ± 0.10	0.41 ± 0.07	0.64 ± 0.22	0.60 ± 0.20
C16:0	34.82 ± 0.42	26.52 ± 0.23	27.14± 1.55	23.83 ± 3.07	27.11 ± 0.46	27.77 ± 0.67	26.27 ± 1.71	26.94± 1.15	25.44 ± 1.88	23.54 ± 3.02
C16:1n7	5.51 ± 0.16	7.00 ± 0.37	6.79 ± 0.14	5.06 ± 0.65	6.59 ± 0.23	6.62 ± 0.23	6.16 ± 0.56	7.55 ± 0.32	5.44 ± 0.88	6.38 ± 1.34
C17:0	1.81 ± 0.16	1.27 ± 0.06	1.48 ± 0.16	1.19 ± 0.09	1.13 ± 0.03	1.30 ± 0.05	1.13 ± 0.11	1.18 ± 0.07	2.00 ± 0.60	1.11 ± 0.14
C18:0	7.31 ± 0.37	6.40 ± 0.26	7.24 ± 0.34	6.70 ± 0.87	6.09 ± 0.33	6.55 ± 0.21	6.79 ± 0.32	6.59 ± 0.44	9.10 ± 1.22	6.20 ± 0.28
C18:1n12t	0.15 ± 0.05	0.88 ± 0.11	0.82 ± 0.23	0.49 ± 0.19	0.62 ± 0.11	0.52 ± 0.02	0.62 ± 0.03	0.67 ± 0.04	0.47 ± 0.17	0.46 ± 0.09
C18:1n9t	0.14 ± 0.05	nd	nd	0.33 ± 0.12	0.19 ± 0.11	0.09 ± 0.09	0.19 ± 0.07	0.10 ± 0.10	nd	1.38 ± 1.05
C18:1n7t	nd	0.04 ± 0.04	0.14 ± 0.14	nd	nd	0.03 ± 0.03	0.04 ± 0.02	nd	nd	0.19 ± 0.19
C18:1n9	6.98 ± 0.31	14.34 ± 0.18	15.70± 0.54	11.92 ± 1.73	15.78 ± 1.20	16.25 ± 0.40	15.94 ± 1.16	17.32± 0.79	12.90 ± 1.29	15.61 ± 2.44
C18:1n7	2.21 ± 0.10	4.22 ± 0.17	3.53 ± 0.06	4.67 ± 1.44	3.88 ± 0.12	3.55 ± 0.04	3.92 ± 0.25	4.07 ± 0.16	3.91 ± 0.52	3.49 ± 0.71
C18:2n6t	nd	nd	0.39 ± 0.39	0.60 ± 0.46	nd	nd	nd	nd	nd	nd
C18:2n6	2.42 ± 0.25	2.52 ± 0.30	2.37 ± 0.35	1.54 ± 0.17	1.57 ± 0.11	1.93 ± 0.16	1.52 ± 0.29	1.75 ± 0.43	2.46 ± 1.31	1.86 ± 0.37
C18:3n6	0.42 ± 0.20	nd	0.06 ± 0.04	0.24 ± 0.10	0.03 ± 0.03	0.12 ± 0.04	0.11 ± 0.04	0.03 ± 0.03	0.05 ± 0.05	0.29 ± 0.15
C18:3n3	1.15 ± 0.18	0.92 ± 0.07	0.97 ± 0.04	0.69 ± 0.14	0.88 ± 0.04	0.96 ± 0.09	0.81 ± 0.12	1.00 ± 0.14	0.80 ± 0.36	0.71 ± 0.18
C18:4n3	2.81 ± 0.19	0.69 ± 0.07	0.82 ± 0.29	1.31 ± 0.50	0.53 ± 0.02	0.72 ± 0.07	0.62 ± 0.13	0.56 ± 0.05	0.73 ± 0.29	0.98 ± 0.32
C19:2n6	0.24 ± 0.22	nd	0.02 ± 0.02	0.40 ± 0.30	nd	0.01 ± 0.01	0.06 ± 0.02	nd	0.58 ± 0.43	0.69 ± 0.63
C20:0	1.14 ± 0.19	1.14 ± 0.01	1.43 ± 0.10	2.05 ± 0.69	1.17 ± 0.03	1.29 ± 0.07	1.28 ± 0.07	1.32 ± 0.02	1.95 ± 0.45	1.46 ± 0.28
C20:1n15	nd	0.02 ± 0.02	0.22 ± 0.20	0.36 ± 0.23	nd	0.05 ± 0.03	0.12 ± 0.06	nd	nd	0.45 ± 0.39
C20:1n12	0.01 ± 0.01	nd	nd	1.07 ± 0.80	nd	nd	nd	nd	nd	nd
C20:1n9	0.01 ± 0.01	1.34 ± 0.11	1.85 ± 0.08	2.06 ± 0.04	1.38 ± 0.03	1.69 ± 0.03	1.89 ± 0.13	1.70 ± 0.08	1.65 ± 0.56	1.78 ± 0.38
C20:1n7	0.03 ± 0.02	1.38 ± 0.16	1.73 ± 0.13	1.23 ± 0.15	1.13 ± 0.06	1.38 ± 0.07	1.36 ± 0.11	1.36 ± 0.16	1.29 ± 0.47	1.42 ± 0.17

Table 2 continued.

Fatty acid	Menhaden	Small			Medium			Large		
		Day 0	Day 13	Day 28	Day 0	Day 13	Day 28	Day 0	Day 13	Day 28
C20:2n6	0.01 ± 0.01	0.66 ± 0.05	1.10 ± 0.34	0.66 ± 0.24	0.59 ± 0.00	0.76 ± 0.04	0.77 ± 0.07	0.64 ± 0.05	0.62 ± 0.21	0.73 ± 0.09
C20:3n6	0.08 ± 0.04	0.09 ± 0.03	0.07 ± 0.04	0.91 ± 0.40	0.12 ± 0.02	0.28 ± 0.05	0.17 ± 0.05	0.06 ± 0.04	1.01 ± 0.82	0.17 ± 0.06
C20:4n6	1.04 ± 0.12	2.36 ± 0.08	2.27 ± 0.06	1.48 ± 0.34	2.11 ± 0.19	2.05 ± 0.12	2.33 ± 0.19	2.30 ± 0.08	1.78 ± 0.60	1.65 ± 0.36
C20:3n3	nd	0.39 ± 0.04	0.34 ± 0.13	0.43 ± 0.19	0.38 ± 0.03	0.38 ± 0.01	0.39 ± 0.03	0.29 ± 0.04	0.47 ± 0.18	0.75 ± 0.48
C20:5n3	7.18 ± 0.34	5.27 ± 0.22	3.79 ± 0.22	2.66 ± 0.71	4.91 ± 0.18	3.79 ± 0.41	3.60 ± 0.21	4.17 ± 0.28	2.29 ± 0.82	3.47 ± 0.51
C22:0	0.15 ± 0.05	1.77 ± 0.13	1.68 ± 0.08	3.18 ± 1.15	2.00 ± 0.09	1.74 ± 0.06	1.94 ± 0.26	1.72 ± 0.06	1.40 ± 0.47	2.04 ± 0.29
C22:1n9	nd	0.16 ± 0.03	0.28 ± 0.14	1.36 ± 0.47	0.23 ± 0.01	0.29 ± 0.07	0.48 ± 0.17	nd	0.70 ± 0.30	0.32 ± 0.05
C22:2n6	0.22 ± 0.09	nd	0.11 ± 0.11	0.64 ± 0.27	0.01 ± 0.01	0.17 ± 0.09	0.17 ± 0.10	nd	0.77 ± 0.71	0.35 ± 0.24
C22:4n6	nd	0.63 ± 0.02	0.47 ± 0.16	1.69 ± 0.57	0.45 ± 0.13	0.76 ± 0.18	0.90 ± 0.27	0.79 ± 0.11	2.55 ± 1.79	0.80 ± 0.19
C22:3n3 + C22:5n6	0.14 ± 0.04	0.73 ± 0.05	0.13 ± 0.13	1.53 ± 0.69	0.53 ± 0.15	1.10 ± 0.34	1.46 ± 0.61	0.66 ± 0.05	0.83 ± 0.36	0.54 ± 0.11
C22:5n3	1.31 ± 0.09	1.96 ± 0.03	2.02 ± 0.17	3.39 ± 1.24	2.44 ± 0.06	1.87 ± 0.22	2.41 ± 0.60	1.77 ± 0.58	3.36 ± 2.02	1.41 ± 0.46
C22:6n3	8.64 ± 0.88	11.09 ± 0.58	7.87 ± 0.68	8.54 ± 2.19	12.25 ± 1.35	8.86 ± 0.49	9.48 ± 2.22	8.77 ± 0.60	6.62 ± 1.61	8.31 ± 1.62
C24:1n9	0.25 ± 0.08	0.31 ± 0.02	0.30 ± 0.10	0.76 ± 0.42	0.43 ± 0.08	0.70 ± 0.30	1.03 ± 0.85	0.16 ± 0.06	1.07 ± 0.65	0.37 ± 0.08

Figure 1. Water temperature for the experimental tanks throughout the experiment. The dashed line represents the day on which the diet was switched from the baseline prey of spot to that of the experimental prey of menhaden.

Figure 2. Baseline fatty acid signature of all three size classes of striped bass after four weeks of being fed spot.

Figure 3. Lipid content of the three size classes of striped bass analyzed. Spot was fed as a baseline prey for four weeks prior to the start of the experiment. Starting on day zero, menhaden was used as the experimental prey. Error bars represent 1 standard error of the mean.

Figure 4. Wet weight of striped bass from each sampling day. Error bars represent 1 standard error of the mean.

Figure 5. Nonmetric multidimensional scaling of fatty acid signatures for striped bass, the baseline prey, spot, and the experimental prey, menhaden. Size class of fish is denoted as Sm for small, Med for medium, and Lg for large. The sampling days for each size class are denoted as 0 is day 0, 13 is day 13, and 28 is day 28. MH refers to menhaden fed as the experimental prey.

Figure 6. Patterns in abundance of select fatty acids for the three sizes of striped bass and the

prey species fed during this experiment. Abundance is based upon mass percent of total

fatty acids.

Figure 1.
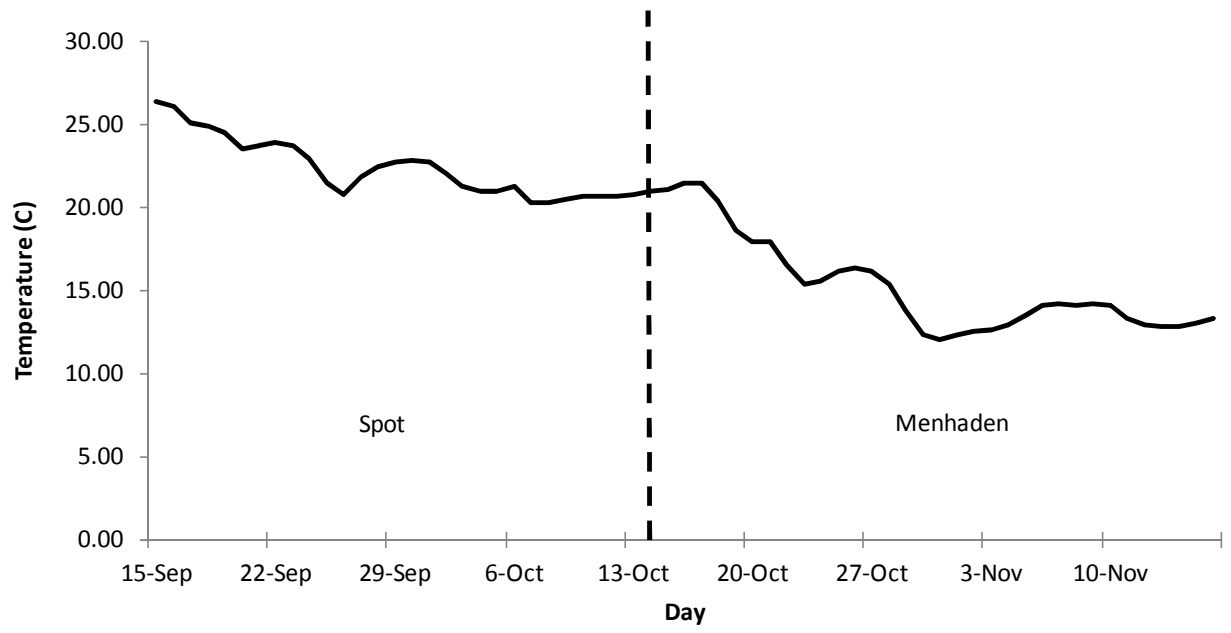
Figure 2.
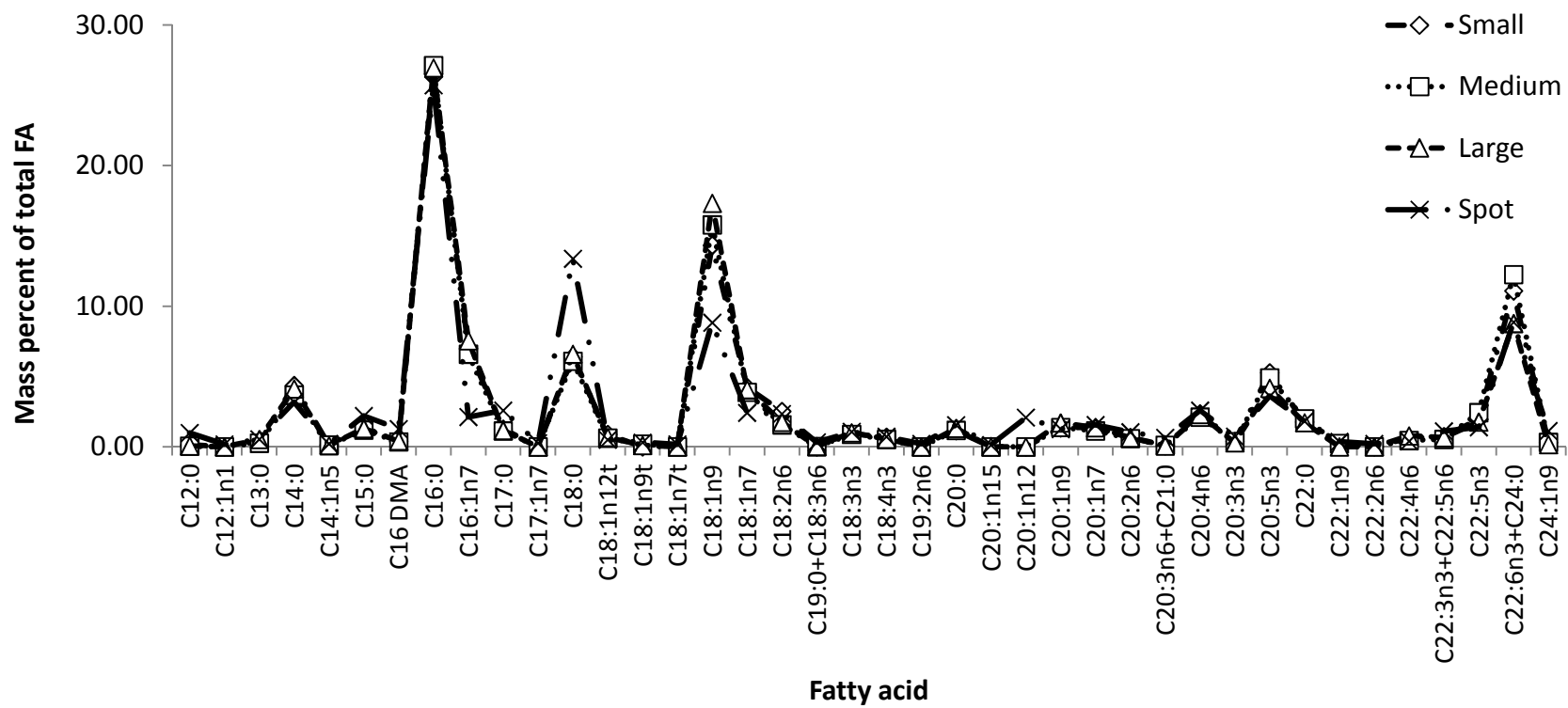
Figure 3.
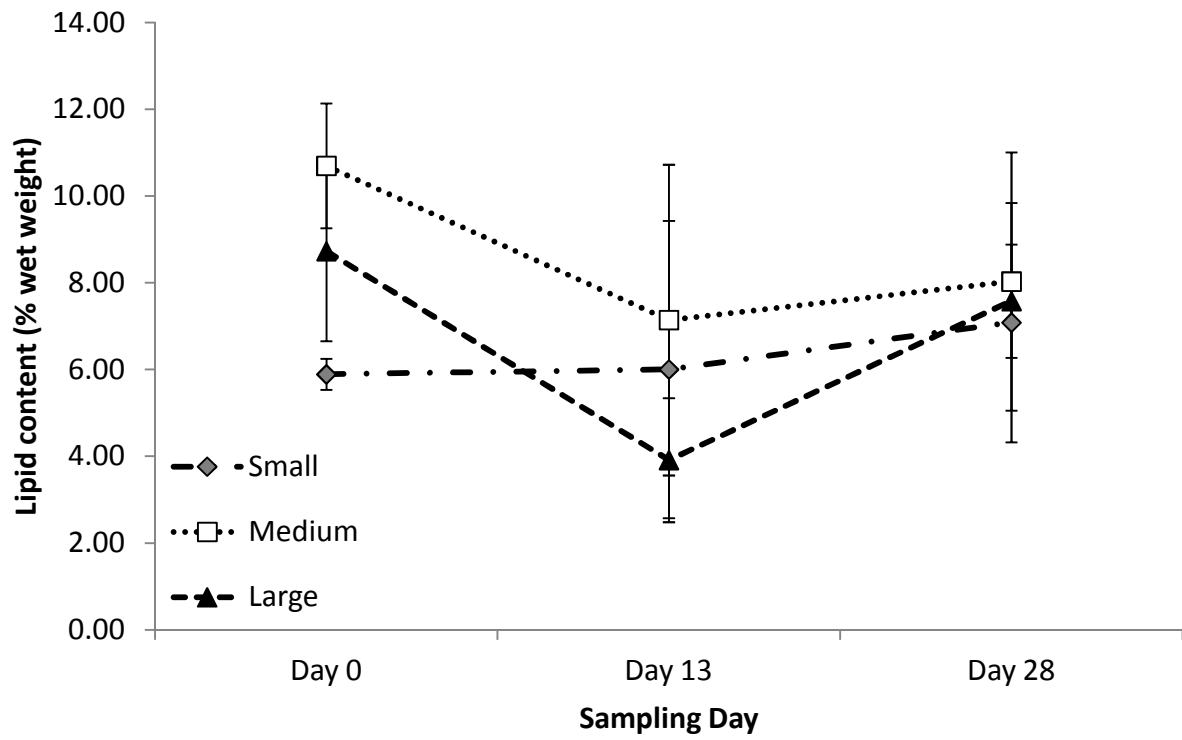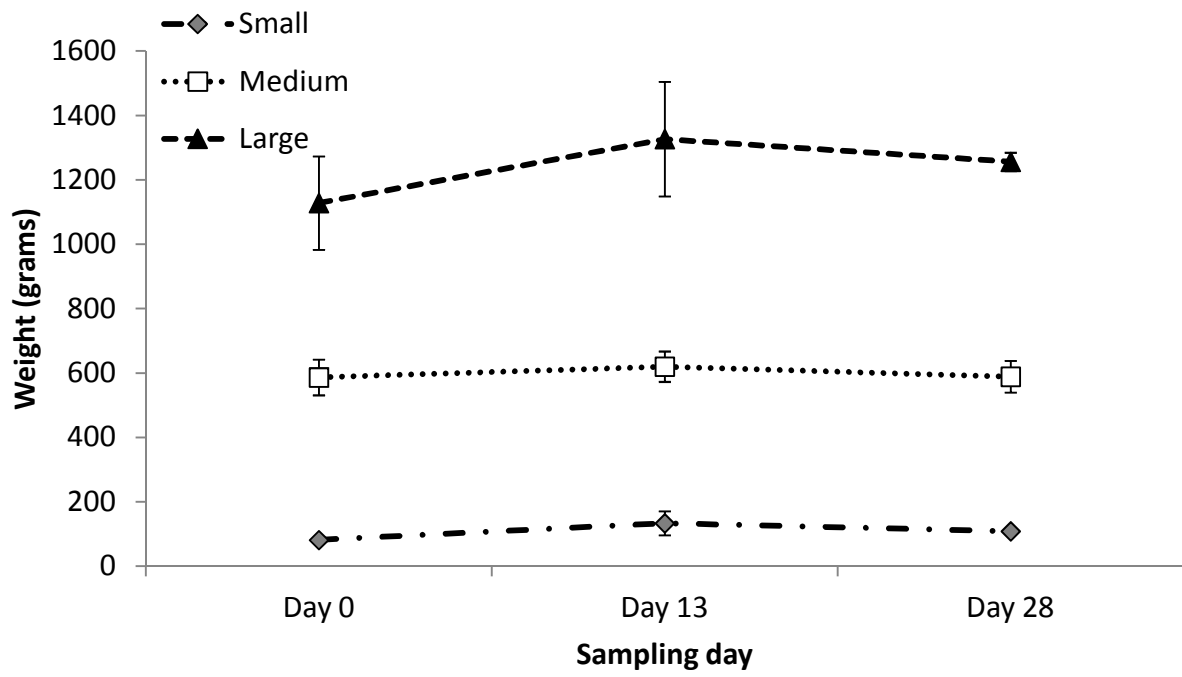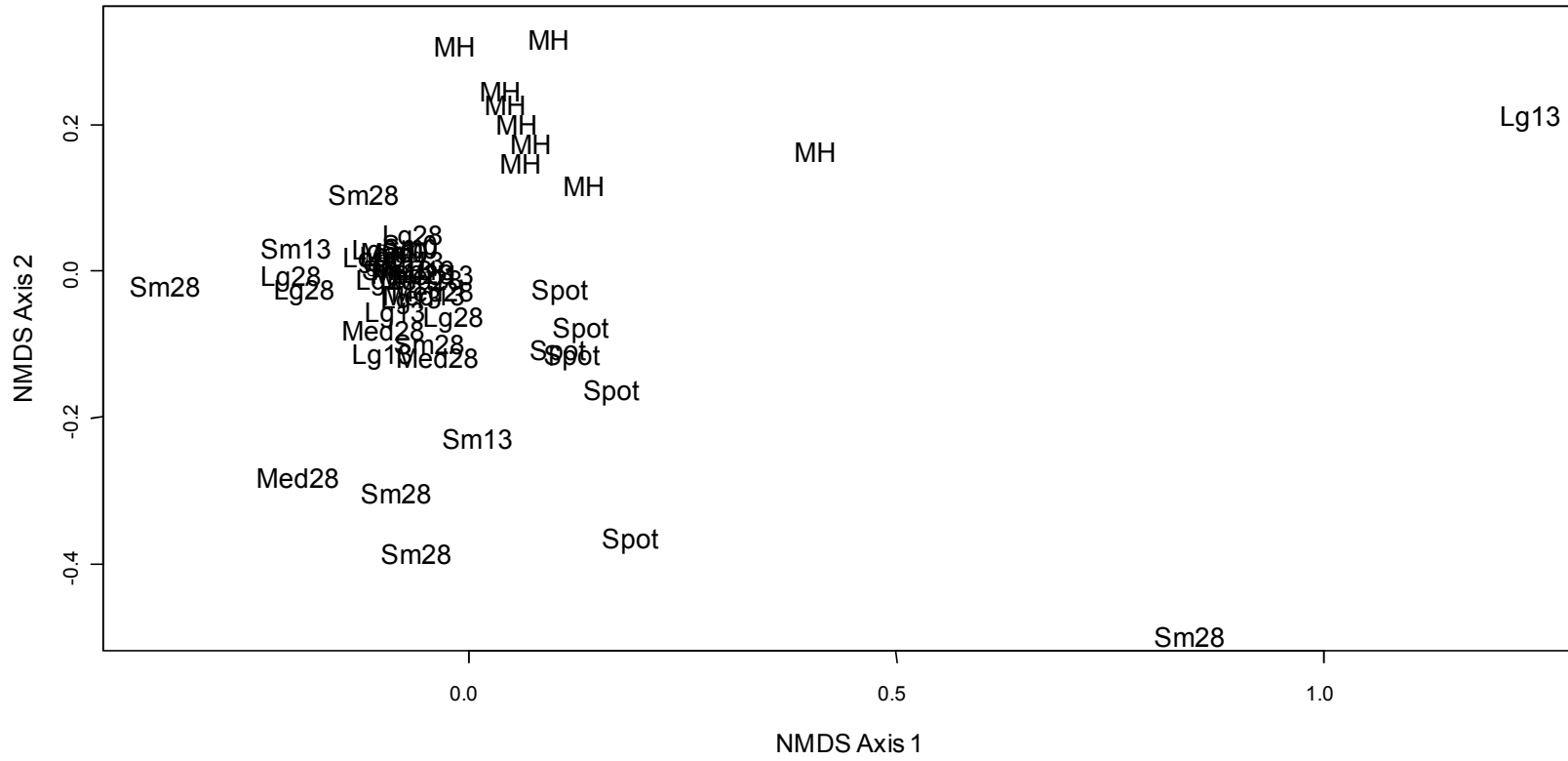
Figure 4.

Figure 5.

Figure 6.
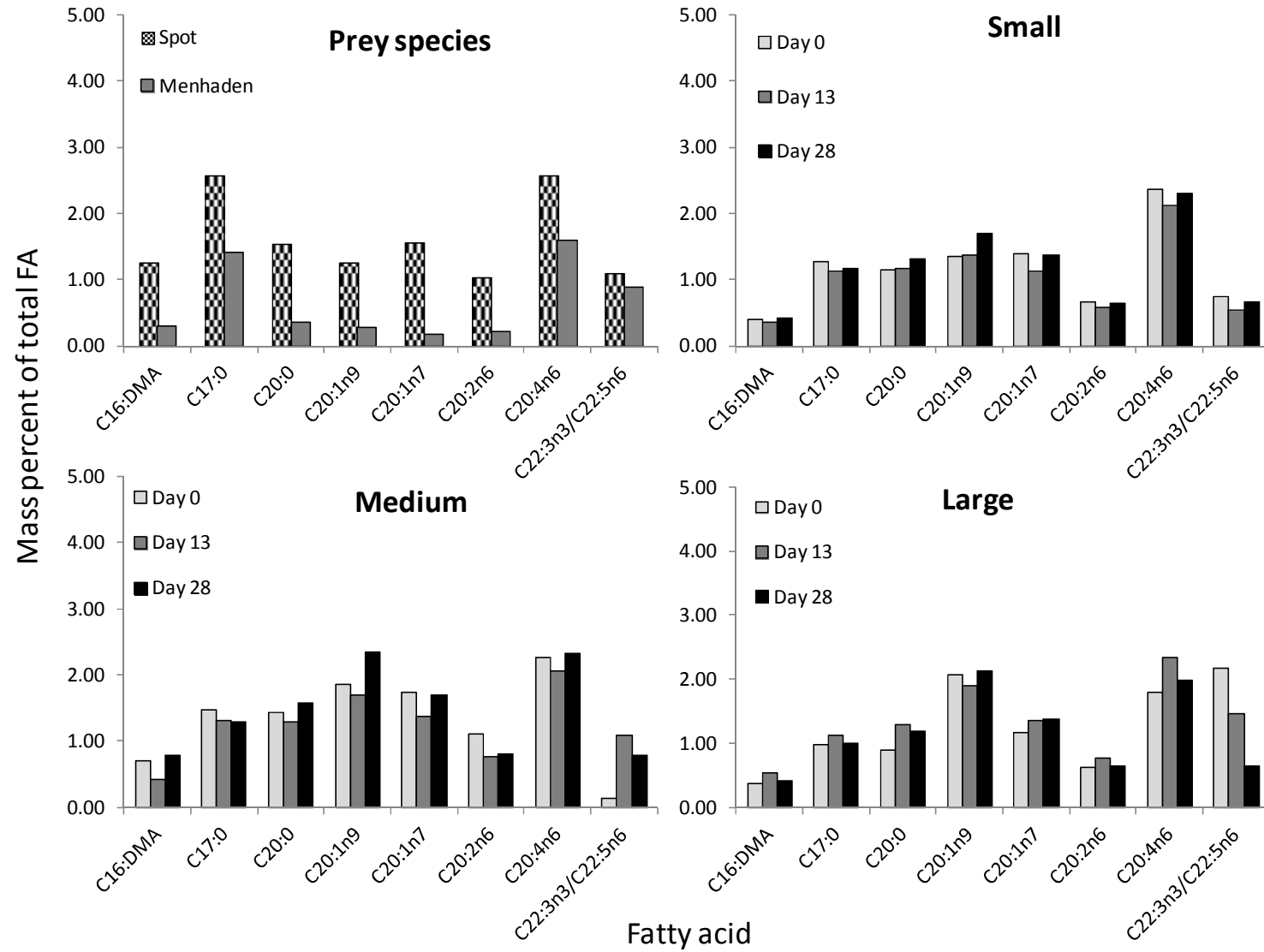
Chapter 5 - Evaluation of quantitative fatty acid signature analysis (QFASA) to estimate

diets of striped bass

Abstract

The process of studying diets has relied on analysis of prey items present in the stomach. Due to unequal digestion rates, temperature effects, and changes in feeding patterns, there may be significant error or bias associated with analyzing stomach contents. Alternative methods, e.g. stable isotopes and fatty acids, rely on the chemical signatures associated with the prey items rather than the prey items themselves. Fatty acid analysis specifically has the potential to estimate the proportional contribution of prey species by using a technique known as quantitative fatty acid analysis (QFASA). This model estimates what combination of prey results in the fatty acid signature present in predator tissues. The goal of this study was to evaluate the utility of using the QFASA model to estimate the contribution of spot (*Leiostomus xanthurus*) and Atlantic menhaden (*Brevoortia tyrannus*) fed to striped bass (*Morone saxatilis*) under controlled laboratory conditions. All striped bass were fed spot for six weeks to calculate calibration coefficients and establish a baseline signature. Calibration coefficients are values that correct for predator metabolism of fatty acids derived from the prey. Starting at day 0, three experimental diets (100% menhaden; 70% menhaden 30% spot; and 30% menhaden 70% spot) were fed to striped bass for six weeks. Liver and belly flap were removed from striped bass on day 0, 21, and 48 and analyzed for fatty acids. The QFASA model underestimated the contribution of menhaden throughout the experiment, but performed better with a truncated fatty acid dataset consisting of fatty acids derived mainly from the diet. Estimated contributions of menhaden averaged 18 – 20% for fish fed the 100% menhaden diet, 10 – 20% menhaden for fish fed the 70% menhaden 30% spot diet, and 5 – 20% menhaden for fish fed the 30% menhaden 70% spot diet. Striped bass grew better and had higher lipid levels in the liver and belly flap tissues in striped bass fed menhaden than either one of the diet mixtures. At this point, it appears that

QFASA is not a viable technique to use to estimate striped bass feeding history. Most previous QFASA studies have focused on homeotherms, e.g. seabirds and marine mammals, and therefore fish may represent a more difficult species to model feeding history.

Introduction

Construction of food webs often relies on estimates of feeding history derived from stomach contents. However, analyzing stomach contents has well documented biases associated with it, e.g. variation in retention times of hard material (Hyslop 1980), temperature effects on digestion (He and Wurtsbaugh 1993), shearing of prey items leading to incomplete ingestion of the entire prey item (Scharf et al. 1997), and inconsistent methodology limiting comparisons among studies (Cortes 1997). These samples represent an estimation that goes back only as far as when these prey items were consumed (Iverson et al. 2004) and no information can be inferred over a longer period of time.

Fatty acids have been used extensively in marine settings for a variety of purposes: to identify farm raised versus wild fish (Ackman and Takeuchi 1986), to assess the health of hatchery reared fish (Brett et al. 1997), and for formulation of beneficial diets for hatchery fish (Sargent et al. 1997). The most promising aspect of studying fatty acids is the ability to track diets of organisms.

Recent advances in trophic ecology have tried to use fatty acid analysis, stable isotopes, or both in an effort to provide information on feeding history over a longer time period (Smith et al. 1996, Hooker et al. 2001. Ruess et al. 2005, Herman et al. 2005). Fatty acids are stored as triacylglycerols and represent a storage depot of energy for an organism (Gurr and Hardwood 1991). When an organism ingests organic material, the fatty acids that comprise the prey species

are transferred to the predator with little or no modification, allowing these molecules to be used as tracers of the diet (Iverson 2009).

Studies focused on estimating which prey item is important in the diet by comparing the fatty acid signature of the prey item to the predator in question have been conducted for fish (Ratanayake and Ackman 1979, Kuusipalo 2000, Jobling 2003) and marine mammals (Bradshaw et al. 2003, Beck et al. 2005). Metabolism of the predator will prevent its fatty acid signature from exactly matching that of the prey (Budge et al. 2006); however, the key components are often identifiable (Dalsgaard 2003).

The qualitative estimation of what is being consumed may not always be straightforward. Wild organisms are often opportunistic and will consume a variety of prey items (Hartman and Brandt 1995). Therefore the fatty acids present in the predator will represent a mixture of all the prey items. Quantitative fatty acid signature analysis (QFASA) is a multivariate optimization model that estimates proportions of prey in a predator's diet based upon prey and predator fatty acid signatures and lipid content of the prey (Iverson et al. 2004). The metabolism of the predator is taken into account with calibration coefficients, values that are designed to correct for predator deposition and mobilization of various fatty acids (Iverson et al. 2004). To date, studies have focused on using marine mammals (Nordstrom et al. 2008, Tucker et al. 2009, Meynier et al. 2010) with few studies focusing on birds (Iverson et al. 2007) or fish (Budge et al. 2011).

Striped bass (*Morone saxatilis*) is one of the first fish species to have their diets estimated using this model. Striped bass are large anadromous fish that historically represented an important commercial and recreational fishery in the Chesapeake Bay (Chesapeake Bay Fisheries Ecosystem Advisory Panel 2006). During the 1970s and 1980s striped bass in Chesapeake Bay experienced a well documented decline (ASMFC 1981). Through a moratorium

placed on the fishery, this species rebounded and was declared a recovered species in the 1990s (ASMFC 1995). Starting in the early 2000s, striped bass were found emaciated and infected with lesions linked to mycobacteriosis (Overton et al. 2003). One theory suggested that the recovered population may have outgrown its prey base (Uphoff 2003). When resources are not present for an organism to meet its metabolic demands, it will be more susceptible to disease. Therefore, an accurate method is needed to estimate feeding history of striped bass to determine if a dietary shift has occurred.

The purpose of this study was to evaluate the QFASA model for striped bass. Specifically, our objectives were: 1) to establish calibration coefficients (corrections for predator metabolism) for striped bass liver and belly flap tissues; 2) to use the QFASA model to estimate the contribution of two prey taxa (spot [*Leiostomus xanthurus*] and menhaden [*Brevoortia tyrannus*]) to the signatures of striped bass fed a known mixture of these two prey items; and 3) to use the QFASA model to estimate the proportion of menhaden to the signatures of striped bass after the diet was completely switched from spot to menhaden.

Materials and Methods

Feeding study

Striped bass (total length 325 ± 6.21 mm) were obtained from West Virginia University's Aquaculture Extension Unit and were transported to the James J. Howard Marine Sciences Laboratory in Sandy Hook, New Jersey. Eighty fish were randomly distributed among three 120 gallon tanks. The tanks were part of a flow through system with water pumped directly from the Sandy Hook Bay. Temperature ranged from 20.4 - 26.8°C, and salinity ranged from 23-26.4 parts per thousand (ppt) during the feeding experiment.

Fish were fed a diet of spot (*Leiostomus xanthurus*; collected from the Chester River, Maryland in fall) every other day for six weeks prior to the experiment to establish calibration coefficients of predator metabolism for striped bass (see below) and to establish a baseline fatty acid signature (Kirsch et al. 1998). At the start of the experiment (day 0), tank one was fed a diet of menhaden (*Brevoortia tyrannus*; collected from the Naticoke River, Maryland in the summer), tank two was fed a mixture of menhaden and spot (70:30 w:w), and tank three was fed a mixture of menhaden and spot (30/70 w:w). Individual spot and menhaden were weighed together prior to homogenizing based upon the 70:30 or 30:70 treatments. The combination of fish was then placed in a food processor to mix the fish tissues. The homogenate was combined into pellets that could be consumed whole by striped bass. The pellets were stored in a -20°C walk-in freezer in air tight bags until feeding. Ten samples of each homogenate were kept for lipid and fatty acid analysis.

Four striped bass were sacrificed from each tank on days 21 and 48. Fish were weighed (g) and measured (total length in mm), and vacuum sealed with a FoodSaver vacuum sealer (Sunbeam Products, Inc., Niles, IL) and frozen at -20°C until they could be analyzed.

Sample preparation

Striped bass were thawed at the time of analysis and tissue samples were weighed to the nearest 0.1 gram. The liver and a portion of the belly flap were removed. The belly flap (with skin intact) follows a line marked by the termination of the ribs and initiation of the peritoneum of the body wall (Jacobs et al. 2008). All tissues were ground with a chemical drying agent, diatomaceous earth (Dionex, Sunnyvale, CA) and a known amount of internal standard (C13:0 triacylglycerol) was added to determine the efficiency of the extraction process. A sample with just the internal standard (the method blank) was also analyzed.

Lipids were extracted under nitrogen using an Accelerated Solvent Extractor (ASE 300, Dionex, Sunnyvale, CA) with a mixture of 3:1 (v/v) dichloromethane:methanol and 0.1% butylated hydroxytoluene (BHT) as an antioxidant. Samples were extracted once at 100°C and 1500 psi (USEPA 2007). Samples were back-extracted using a 0.88% potassium chloride solution in deionized water to remove non-lipid materials from the extracts (Folch et al. 1957). Total lipid extracted was determined gravimetrically for each sample and reported as a percent of wet weight.

Extracted lipid species containing fatty acids as various derivatives were transesterified using a solution of sulfuric acid and methanol. In this process, aclyglycerols are broken apart to their constituents: glycerol and fatty acids methyl esters (FAMEs). Free fatty acids present in the lipid mixture are also converted to FAMEs (Christie 2003). Sample extracts were cleaned with silica gel and anhydrous sodium sulfate to remove polar components and water, which may degrade the performance of the gas chromatograph column. The silica column was conditioned with hexane and the sample was added along with hexane and diethyl ether, 95:5, v/v (Christie 2003). The mixture was concentrated to 5 ml and diluted to a final total lipid concentration of 100 μg/ml. Finished samples were run on a gas chromatograph (Agilent 6890N Network GC System, Santa Clara, CA) / mass spectrometer (GC/MS, Agilent, 5973 Network Mass selective detector, Santa Clara, CA) in select ion monitoring mode.

The GC/MS was used to identify and quantify fatty acid methyl esters in each sample. FAMEs were injected into a fused capillary column, DB-23 60m length X 0.250mm OD X 0.25μm film thickness (Agilent, Santa Clara, CA) using an autosampler (Agilent 7683 Series Auto Sampler). Hydrogen was used as a carrier gas. The temperature program was as follows: hold at an initial temperature of 50°C for 2 min, hold for 1 min at 150°C after ramping at

$20^{\circ}C \cdot min^{-1}$, ramp to $215^{\circ}C$ at $1.25^{\circ}C \cdot min^{-1}$. The fatty acid retention times were compared to known standards from Nu-Chek Prep (GLC MIX 85, 411, 642, 632, Elysian, MN). These four standard mixtures were combined along with stearidonic acid (C18:4n3, Cayman Chemical, Ann Arbor, MI) to make a total FAME standard solution of 12.25 µg/ml. Matreya 25 FAME mixture (Pleasant Gap, PA) was used to obtain retention times for C20:1n7. Restek cis/trans FAME mixture (Bellefonte, PA) was used to obtain retention times for C18:1n9 trans and C18:2n6 trans. These standards and an internal standard of C13:0 FAME (100 µg/ml) were added to quantify the amount of each fatty acid methyl ester present. The GC/MS tracks the select ion signature of each compound and was used to verify the identity of each peak. There were peaks which consisted of more than one fatty acid and it was impossible to determine which fatty acid(s) constituted the peaks. Therefore, a term fatty acid "complex" was assigned to this peak. The complex refers to which fatty acids could be responsible for the peak, e.g. C22:3n3 and C22:5n6.

Calibration coefficients and fatty acid subsets

Calibration coefficients were calculated by dividing the proportion of a FA in a predator by the value of that same FA in the prey (Iverson et al. 2004). During the baseline feeding, all three tanks of striped bass were fed spot for a period of 6 weeks. At the end of the six weeks, 12 striped bass were sacrificed and the fatty acid signature of the liver and belly flap was analyzed. Eleven samples of spot were also analyzed for their fatty acid signature, resulting in 132 calibration coefficients for each fatty acid in both the liver and belly flap. The six highest and lowest values were removed before the mean was calculated (10% trimmed mean; Iverson et al. 2004).

A subset of the fatty acids is used for the QFASA model, which excludes fatty acids that may arise from biosynthesis, or fatty acids inconsistently detected. In our experiment, we used an

extended fatty acid subset including all fatty acids that had a proportion > 0.1% (n=30; Table 1).

A dietary subset was also constructed to mimic the dataset presented in Iverson et al. (2004).

This subset consisted of fatty acids derived primarily from the diet (n=16; Table 1). Once fatty

acids were removed from the dataset, the proportions were renormalized to 100%.

Diet estimation

The diet of striped bass was estimated using the QFASA model (Iverson et al. 2004).

The model uses the prey FA signatures and tries to estimate which mixture of prey comes closest

to matching the predator's actual signature. The best mixture of prey FAs is then weighted by the

fat content of the prey species. Species that have a higher fat content will have more FAs to

contribute to the signature seen in the predator, and the weighting algorithm takes this into

account. The model uses an optimization procedure which aims to minimize the distance

between the predicted and observed predator signature, the Kullback-Liebler (KL) distance. The

optimization uses a quasi-Newton algorithm with a Broyden-Fletcher-Goldfarb-Shanno formula

and was carried out with a package developed at Massey University, New Zealand (Fatty acid

solution – Dr. R. Sheriff and A/Prof P.C.H. Morel).

Statistical analyses

Fatty acid methyl esters were expressed as the percent of the total fatty acid

concentrations. These values were used to determine differences in the prey species and were

then compared to tissues samples throughout the experiment to determine if striped bass tissues

reflected prey values.

Nonmetric multidimensional scaling (NMDS) was also performed on the data (R

Development Core Team 2010, Vienna, Austria). This ordination technique is an iterative search

for the best position of samples in multidimensional space, and uses the stress value as an

indicator for the best solution. A distance matrix was calculated for the fatty acids, and the samples were ordinated based on the distances calculated (McCune and Grace 2002).

Results

Mixed diet feed

The fatty acids of spot and menhaden were markedly different, however, the fatty acids present in the two mixed diets (70MH:30SP and 30MH:70MH) did not represent a continuum between spot and menhaden as intended (Table 2). The fatty acids detected in the diet mixtures should have intermediate values when compared to the contributing tissues of spot and menhaden, however there were several instances of values for the mixed diets being out of the range, e.g. C18:3n3 and C20:3n3. There are also several instances in which the 70MH:30SP diet has proportions more similar to spot than menhaden, e.g. C14:0, C16:1n7, C18:0, and C18:1n7. The same type of pattern holds true for 30MH:70SP, e.g. C14:0, C16:0, C18:0, and C18:4n3. Despite these anomalies, the NMDS plot revealed that both mixtures were intermediate between spot and menhaden, with the 70MH:30SP mixture being more similar to menhaden and the 30MH:70SP being more similar to spot (Fig.1).

Growth

The lengths and weights, as well as the lipid content of both the liver and belly flap samples changed depending on the feeding regime (Table 3). Striped bass fed menhaden alone obtained larger sizes, based on length and weight, than striped bass fed either of the mixtures. The lipid content of both the liver and belly flap of striped bass fed menhaden increased above the starting lipid levels. The results for the mixed diets were variable. Striped bass fed the 70MH:30SP mixture obtained a larger final length, weight, and belly flap lipid content. Striped

bass fed the 30MH:70SP mixture obtained larger final liver lipid content. Overall, the standard errors were high for many of the measurements due to low sample size (n=4 samples per group).

Fatty acid composition

The fatty acid signature of striped bass fed the various diets did not show distinct variation in their signatures (Fig.1). There was a general pattern of signatures moving towards those of the various diets as the experiment progressed. However, there was a large degree of overlap of the three diets in both the liver and belly flap tissues and a separation from the initial diet of spot. The NMDS also indicates that the striped bass fed spot for six weeks prior to the start of the experimental feeding were more dissimilar versus spot than the striped bass fed experimental diets. Liver samples were more similar to each other than belly flap samples in terms of their respective fatty acid signatures regardless of the diet (100% menhaden or either mixed diet; Fig. 1).

Individual striped bass fatty acids did not show a consistent pattern of change from the initial diet to that of the 70MH:30SP mixture (Fig. 2) or the 30MH:70SP mixture (Fig. 3). Certain fatty acids were often higher in striped bass tissues from day 0 through day 48, despite the initial and mixture diets having lower proportions, e.g. C18:2n6 and C18:1n9 (Fig. 2b), C18:1n9 (Fig. 3a), and C18:1n9 (Fig. 3b). Certain striped bass fatty acids remained at the level of the initial diet, e.g. C16:1n7 (Fig. 2a) and C14:0 (Fig. 3a). Other fatty acids remained at levels of the mixture diet and showed no change, e.g. C20:4n6 (Fig. 2b) and C18:0 (Fig. 3a).

QAFSA model

Calibration coefficients were calculated for both striped bass liver and belly flap tissues (Table 3). There were apparent differences in the metabolism and storage of fatty acids in liver and belly flap tissues, therefore calibration coefficients were only used for the tissue in which

they were calculated. Two different sets of fatty acid datasets were used in the QFASA model (Table 3) and these were adopted from Iverson et al. (2004).

Striped bass samples fed the baseline diet of spot were analyzed with the QFASA model to determine the duration needed before the diet consisted of 100% spot (Fig. 4). The anchovy signature was still present when the baseline feeding was initiated. The proportional contribution of spot increased throughout the six weeks until the signature of striped bass tissues consisted of 95-98% spot fatty acids.

Samples from both tissues sampled on day 0, 21, and 48 during the experiment for each diet were run with the QAFSA model and appropriate calibration coefficients. The model was run once with the full fatty acid dataset (Fig. 5) and the dietary dataset (Fig. 6).

Liver and belly flap samples run using the full dataset yielded slightly different results, although both instances were not able to correctly identify the diet mixture fed. On day 0, the model indicated that the spot signature accounted for 95-98% of the prey consumed by striped bass. Samples from striped bass switched to a diet of menhaden estimated the contribution of menhaden to be small, decreasing slightly from 20% on day 21 to 18% on day 48 (Fig. 5b). The contribution of menhaden to liver signatures was 10% on day 21 and 20% on day 48 (Fig. 5a). The contribution of menhaden estimated by QFASA was incorrect for both tissues fed the 70MH:30SP and the 30MH:70SP diets. All results estimated menhaden contributing ~5 – 20% of the diet, with lower values for the 30MH:70SP diet. In both mixed diet treatments, the contribution of menhaden decreased from day 21 to day 48.

Running the QFASA model with the dietary dataset yielded better results than the full dataset, but the estimated contribution of menhaden was lower than the actual. Results from liver samples from striped bass fed menhaden estimated the contribution of menhaden to be 50% on

day 21 and 80% on day 48 (Fig. 6a). Estimates from belly flap samples indicated the contribution of menhaden to be 45% on day 21 and 40% on day 48 (Fig 6b). The proportion of menhaden in striped bass fed the 70MH:30SP yielded 10% on day 21 and 18% on day 48 for liver, while belly flap samples had 10% menhaden in their signatures on both day 21 and 48. The proportion of menhaden in striped bass fed 30MH:70SP was 15% on day 21 and 10% on day 48 for liver, while belly flap samples had 20% menhaden on day 21 and 8% on day 48.

Discussion

The QAFSA model performed poorly in regards to all captive feeding trials in this study. The model had problems with estimating the contribution of menhaden alone to the fatty acid signatures of liver and belly flap tissues from striped bass fed this prey species for six weeks. Model estimates derived from the liver yielded more accurate results as the liver represents a processing organ for fatty acids recently consumed while adipose tissue is a long term storage site for fatty acids. The model also consistently underestimated the proportion of menhaden fatty acids in the signature of striped bass fed a known mix of spot and menhaden.

In order to run the QFASA model, inputs of prey fatty acids, predator fatty acids, and calibration coefficients are needed. Calibration coefficients are based on the incorporation of a specific prey fatty acid into a predator's tissue (Iverson et al. 2004) and need to be estimated by captive feeding experiments. Captive feeding trials can also be used to test the model before applying it to wild populations; such studies have lasted for 42 days (Iverson et al. 2007, Nordstrom et al. 2008) and 50 days (Wang et al. 2010). Based upon these studies and the fact that predator tissues should begin to mimic prey species after a period of approximately three weeks (Kirsch et al. 1998), we fed striped bass for a period of six weeks to calculate calibration

coefficients. We also ran the captive feeding trials that the QFASA model was based on for another six weeks. However, the data indicated that the switch from spot to menhaden was not complete after the period of six weeks. Liver tissue showed an increase in the proportion of menhaden from day 21 - 48 for both datasets (although more pronounced in the dietary dataset), but the highest value obtained was ~80%. There is little research to suggest the appropriate duration for a captive feeding study of fish for QAFSA validation. Despite the fact that a fish predator will mimic its prey signature after a period of three weeks, Atlantic salmon (*Salmo salar*) required 12 weeks for the proportions of fatty acids in the muscle and belly flap to completely change substantially and remain stable (Budge et al. 2011). The comparisons among taxonomic groups may confound the issue. Fish are poikilothermic while seabirds and mammals are homeothermic, necessitating the use of more energy for homeostasis in homeotherms, and in turn using more fatty acids. Therefore, an experiment lasting six weeks for a fish species may be enough to distinguish fatty acid signatures in a predator tissue qualitatively, but not quantitatively.

The model results overestimated the contribution of spot to the striped bass signature. The mixed diets should have represented a continuum of fatty acids between the two components (Budge et al. 2011). However, the values for several fatty acids were outside the range of spot and menhaden. The fat content of the 70MH:30SP diet mixture had a lower fat content than the 30MH:70SP mixture, possibly indicating a disproportionate contribution of spot in the diet formulated to be higher in menhaden. This combination of fatty acids resulted in the model recognizing spot as more prevalent than it actually was. In this study, prey species were weighed, homogenized, and frozen prior to feeding. Previous studies have combined fish and krill oil to a standard lipid content (14% wet weight) and pelleted using an industrial steam pellet mill (Budge

et al. 2011), or combined fish species, e.g. silverside (*Menida menida*), rainbow smelt (*Osmerus mordax*), and herring (*Clupea harengus*) with no information about how the prey species were combined (Iverson et al. 2007). The lack of a standardized procedure for mixed diet formulation may have led to heterogeneous diet mixtures being fed to striped bass. This fact along with the length of the experiment could have led to the poor performance of the QFASA model.

Striped bass fed a diet of menhaden alone led to larger fish and a higher lipid content of the liver and belly flap tissues than either of the mixed diets. The lipid content of the menhaden diet (11.75%) was higher than either the 70MH:30SP (6.56%) or 30MH:70SP (7.32%) mixtures. Due to the possibility of non-uniform mixtures in both the mixed diet treatments, it is difficult to reach a conclusion on the effect of a lowered ratio of menhaden in the diet on growth parameters and lipid deposition. However, it is obvious that the removal of a preferred prey (menhaden) leads to reduced growth and lowered lipid stores, although high standard errors due to low sample size prevent definitive conclusions. This becomes an important biological ramification in relation to the possible overfishing of menhaden (NOAA 2009). Studies have shown that the proportion of menhaden in the diet has decreased since the recovery of the striped bass population (Overton et al. 2000). More research is needed to assess the long term effects of diet mixtures of high- and lower-lipid prey on the growth and health of striped bass.

Based upon the findings of Budge et al. (2011) on the incorporation of prey fatty acids, including diet mixtures, fish may need longer time periods (12 weeks) in which to reach a plateau in terms of fatty acid turnover; however, this situation may not be realistic for wild fish populations. Studies that have examined striped bass diets using stomach contents have shown that there are several prey items that comprise the diets (Hartman and Brandt 1995, Overton et al. 2000, Rudershausen et al. 2005). This suggests that there are new prey signatures being added to

striped bass tissues constantly, precluding the dominance of just one prey signature. Although much work has been done with marine mammals (Beck et al. 2007, Thiemann et al. 2007, Thiemann et al. 2008, Meynier et al. 2010), this technique is relatively new to fisheries management. QFASA models may prove to be useful for fish populations, but more research needs to be done to verify that the model is capable of estimating diets without the focal species needing to feed on one prey item for an extended period of time.

Literature cited

Ackman, R.G. and T. Takeuchi. 1986. Comparison of fatty acids and lipids of smolting hatchery-fed and wild Atlantic salmon *Salmo salar*. Lipids 21: 117-120.

ASMFC (Atlantic States Marine Fisheries Commission). 1981. Interstate fisheries management plan for the striped bass. Fisheries Management Report No. 1, Washington, D.C.

ASMFC (Atlantic States Marine Fisheries Commission). 1995. Amendment #5 to the interstate fishery management plan for Atlantic striped bass. ASMFC. Fisheries Management Report 24, Washington, D.C.

Beck, C. A., S.J. Iverson, and W.D. Bowen. 2005. Blubber fatty acids of gray seals reveal sex differences in the diet of a size-dimorphic marine carnivore. Canadian Journal of Zoology 83:377-388.

Beck, C.A., S.J. Iverson, W.D. Bowen, and W. Blanchard. 2007. Sex differences in grey seal diet reflect seasonal variation in foraging behaviour and reproductive expenditure: evidence from quantitative fatty acid signature analysis. Journal of Animal Ecology 76:490-502.

Bradshaw, C. J. A., M.A. Hindell, N.J. Best, K.L. Phillips, G. Wilson, and P.D. Nichols. 2003. You are what you eat: describing the foraging ecology of southern elephant seals (*Mirounga leonina*) using blubber fatty acids. Proceedings of the Royal Society of London B 270:1283-1292.

Brett, M. and D. Muller-Navarra. 1997. The role of highly unsaturated fatty acids in aquatic foodweb processes. Freshwater Biology, 38: 483–499.

Budge, S.M., S.J. Iverson, and H.N. Koopman. 2006. Studying trophic ecology in marine ecosystems using fatty acids: a primer on analysis and interpretation. Marine Mammal Science 22:759-801.

Budge, S.M., S.N. Penney, S.P. Lall. 2011. Response of tissue lipids to diet variation in Atlantic salmon (*Salmo salar*): implications for estimating diets with fatty acid analysis. Journal of Experimental Marine Biology and Ecology 409:267-274.

Chesapeake Bay Fisheries Ecosystem Advisory Panel (National Oceanic and Atmospheric Administration Chesapeake Bay Office). 2006. Fisheries ecosystem planning for Chesapeake Bay. American Fisheries Society, Trends in Fisheries Science and Management 3, Bethesda, Maryland.

Christie, W.M. 2003. Lipid Analysis. PJ Barnes and Associates. Bridgewater, England.

Cortes, E. 1997. A critical review of methods of studying fish feeding based on analysis of stomach contents: application to elasmobranch fishes. Canadian Journal of Fisheries and Aquatic Sciences 54:726-738.

Dalsgaard, J., M. St. John, G. Kattner, D. Muller-Navarra, and W. Hagen. 2003. Fatty acid trophic markers in the pelagic marine environment. Advances in Marine Biology 46:224-340.

Folch, J., M. Lees, and G.H.S. Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. Journal of Biological Chemistry 226:497-509.

Gurr, M.I. & J.L. Hardwood. 1991. *Lipid biochemistry: an introduction*. 4th edn. London, England: Chapman and Hall.

Hartman, K.J. and S.B. Brandt. 1995. Trophic resource partitioning, diets, and growth of sympatric estuarine predators. Transactions of the American Fishery Society 124:520-537.

He, E. & W.A. Wurtsbaugh. 1993. An empirical model of gastric evacuation rates for fish and an analysis of digestion in piscivorous brown trout. Transactions of the American Fisheries Society 122, 717-730.

Herman, D.P., D.G. Burrow, P.R. Wade, J.W. Durban, C.O. Matkin, R.G. Leduc, L.G. Barrett-Lennard, and M.M. Krahn. 2005. Feeding ecology of eastern North Pacific killer whales *Orcinus orca* from fatty acid, stable isotope, and organochlorine analyses of blubber biopsies. Marine Ecology Progress Series 302:275-291.

Hooker, S.K., S.J. Iverson, P. Ostrom, and S.C. Smith. 2001. Diet of northern bottlenose whales inferred from fatty acid and stable isotope analyses of biopsy samples. Canadian Journal of Zoology 79:1442-1454.

Hyslop, E.J. 1980. Stomach contents analysis – a review of methods and their application. Journal of Fish Biology 17:411-429.

Iverson, S. J. 2009. Tracing aquatic food webs using fatty acids: from qualitative indicators to quantitative determination. In *Lipids in aquatic ecosystems* (Arts, M.T., M.T. Brett, & M.J. Kainz eds). pp 281-307. New York, NY: Springer Science.

Iverson, S.J., A.M. Springer, and A. Kitaysky. 2007. Seabirds as indicators of food web structure and ecosystem variability: qualitative and quantitative diet analyses using fatty acids. Marine Ecology Progress Series 352:235-244.

Iverson, S.J., C. Field, W.D. Bowen, and W. Blanchard. 2004. Quantitative fatty acid signature analysis: a new method of estimating predator diets. Ecological Monographs 74:211-235.

Jacobs, J. M., M.R. Rhodes, B. Martin, D. McIntosh, W.F. Van Heukelem, and R.M. Harrell. 2008. Tissue-specific sampling for estimation of striped bass whole-body proximate composition. Transactions of the American Fisheries Society 137:386-392.

Jobling, M. 2003. Do changes in Atlantic salmon, *Salmo salar* L., fillet fatty acids following a dietary switch represent wash-out or dilution? Test of a dilution model and its application. Aquaculture Research 34:1215-1221.

Kirsch, P.E., S.J. Iverson, W.D. Bowen, S.R. Kerr, and R.G. Ackman. 1998. Dietary effects on the fatty acid signature of whole Atlantic cod (*Gadus morhua*). Canadian Journal of Fisheries and Aquatic Sciences 55:1378-1386.

Kuusipalo, L. 2000. Muscle fatty acids as indicators of niche and habitat in Malawian cichlids. Limnology and Oceanography 45:996-1000.

McCune, B. and J.B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design, Gleneden Beach, OR.

Meynier, L., P.C.H. Morel, B.L. Chilvers, D.D.S. Mackenzie, and P.J. Duignan. 2010. Quantitative fatty acid signature analysis on New Zealand sea lions: model sensitivity and diet estimates. Journal of Mammalogy 91:1484-1495.

NOAA (National Oceanic and Atmospheric Administration). 2009. Forecast for the 2009 Gulf and Atlantic menhaden purse-seine fisheries and review of the 2008 fishing season. Sustainable Fisheries Branch, NMFS Beaufort, NC.

Nordstrom, C.A., L.J. Wilson, S.J. Iverson, and D.J. Tollit. 2008. Evaluating fatty acid signature analysis (QFASA) using harbor seals *Phoca vitulina richardsi* in captive feeding studies. Marine Ecology Progress Series 360:245-263.

Overton, A.S., E.B. May, J. Griffin, F.J. Margraf. 2000. A bioenergetics approach for determining the effect of increased striped bass population on its prey and health in the Chesapeake Bay. Report to: Maryland Department of Natural Resources. Maryland

Cooperative Fish and Wildlife Research Unit, University of Maryland Eastern Shore, Princess Anne, MD.

Overton, A.S., F.J. Margraf, C.A. Weedon, L.H. Pieper, E.B. May. 2003. The prevalence of mycobacterial infections in striped bass in Chesapeake Bay. Fisheries Management and Ecology 10:301-308.

R Development Core Team. 2010. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Ratanayake, W. M., and R. G. Ackman. 1979. Fatty alcohols in capelin, herring, and mackerel oils and muscle lipids: I. Fatty alcohol details dietary copepod fat with certain fish depot fats. Lipids 14:795-803.

Rudershausen, P.J., J.E. Tuomikoski, and J.A. Buckel. 2005. Prey selectivity and diet of striped bass in western Albemarle Sound, North Carolina. Transactions of the American Society 134:1059-1074.

Ruess, L., A. Tiunov, D. Haubert, H.H. Richnow, M.H. Haggblom, and S. Scheu. 2005. Carbon stable isotope fractionation and trophic transfer of fatty acids in fungal based soil food chains. Soil Biology and Biochemistry 37: 945-953.

Sargent, J.R., L.A. McEvoy, and J.G. Bell. 1997. Requirements, presentation, and sources of polyunsaturated acids in marine fish larval feeds. Aquaculture 155:117-127.

Scharf, F.S., J.A. Buckel, F. Juanes, and D.O. Conover. 1997. Estimating piscine prey size from partial remains: testing for shifts in foraging mode by juvenile bluefish. Environmental Biology of Fishes 49:377-388.

Smith, R.J., K.A. Hobson, H.N. Koopman, and D.M. Lavigne. 1996. Distinguishing between populations of fresh- and salt-water harbor seals (*Phoca vitulina*) using stable-isotope ratios and fatty acid profiles. Canadian Journal of Fisheries and Aquatic Sciences 53:272-279.

Thiemann, G., S.J. Iverson, and I. Sterling. 2008. Polar bear diets and arctic marine food webs: insights from fatty acid analysis. Ecological Monographs 78:591-613.

Thiemann, G., S.M. Budge, S.J. Iverson, and I. Sterling. 2007. Unusual fatty acid biomarkers reveal age- and sex-specific foraging in polar bears (*Ursus maritimus*). Canadian Journal of Zoology85:505-517.

Tucker, S., W.D. Bowen, S.J. Iverson, W. Blanchard, and G.B. Stenson. 2009. Sources of variation in diets of harp and hooded seals estimated from quantitative fatty acid signature analysis (QFASA). Marine Ecology Progress Series 384:287-302.

Uphoff Jr., J.H. 2003. Predator-prey analysis of striped bass and Atlantic menhaden in upper Chesapeake Bay. Fisheries Management and Ecology 10:313-322.

USEPA (United States Environmental Protection Agency). 2007. Pressurized fluid extraction (PFE). USEPA SW-846. Method 3545A, Update IV. Washington, D.C.

Wang, S.W., T.E. Hollmen, and S.J. Iverson. 2010. Validating quantitative fatty acid signature analysis to estimate diets of spectacled and Stellar's eiders (*Somateria fischeri* and *Polysticta setlleri*). Journal of Comparative Physiology B 180:125-139.

Table 1. Calibration coefficients calculated for fatty acids in the liver and belly flap samples. Fatty acid datasets represent the variables included in the QFASA models.

| | Calibration coefficients | | Fatty acid datasets | |
Fatty acid	Liver	Belly flap	Full	Dietary
C14:0	0.91	1.87	X	
C15:0	0.40	0.60	X	
C16:0	0.99	0.88	X	
C16:1n7	1.01	1.62	X	
C17:0	0.54	0.41	X	
C18:0	0.65	0.44	X	
C18:1n9t	1.00	0.61	X	
C18:1n9	2.23	2.06	X	
C18:1n7c	1.22	1.02	X	
C18:2n6	2.42	5.26	X	X
C18:3n6	1.32	1.84	X	X
C18:3n3+C19:0	0.88	1.64	X	X
C18:4n3	0.86	1.90	X	X
C20:0	0.54	0.97	X	
C20:1n12	0.43	0.68	X	
C20:1n9	2.88	1.82	X	X
C20:1n7	0.51	0.65	X	X
C20:2n6	0.81	1.10	X	X
C20:3n6+C21:0	0.84	1.27	X	X
C20:4n6	0.72	0.39	X	X
C20:3n3	0.56	1.11	X	X
C20:5n3	0.67	0.67	X	X
C22:0	0.38	0.95	X	
C22:1n9	1.15	1.38	X	X
C22:2n6	0.92	2.39	X	X
C22:4n6	0.42	0.61	X	X
C22:3n3+C22:5n6	0.51	0.66	X	X
C22:5n3	0.68	0.94	X	
C24:0+C22:6n3	0.93	0.57	X	X
C24:1n9	0.90	1.44	X	

Table 2. Mean fatty acid concentrations expressed as a mass percent of all fatty acids present ± 1 standard error of the mean; nd = not detected. The fatty acids shown are for the menhaden and spot, the two prey taxa that comprised the mixed diets: 70% menhaden and 30% spot (70MH:30SP) and 30% menhaden and 70% spot (30MH:70SP).

	Menhaden (n=10)	70MH:30SP (n=8)	30MH:70SP (n=7)	Spot (n=11)
Fat content	11.75 ± 1.91	6.56 ± 0.48	7.32 ± 0.84	6.05 ± 0.18
Fatty acid				
C12:0	0.11 ± 0.01	0.11 ± 0.00	0.12 ± 0.01	0.05 ± 0.04
C12:1n1	0.35 ± 0.05	0.02 ± 0.00	0.04 ± 0.01	0.02 ± 0.03
C13:0	0.18 ± 0.03	0.24 ± 0.01	0.21 ± 0.04	0.26 ± 0.26
C14:0	10.28 ± 1.53	5.81 ± 0.17	8.34 ± 0.98	2.42 ± 0.81
C14:1n5	0.05 ± 0.01	0.09 ± 0.01	0.11 ± 0.01	0.03 ± 0.01
C15:0	0.90 ± 0.04	1.69 ± 0.05	1.18 ± 0.13	1.30 ± 0.17
C16:0	28.26 ± 1.21	26.39 ± 1.53	27.87 ± 2.08	24.36 ± 1.69
C16:1n7	10.25 ± 1.41	6.97 ± 0.13	7.53 ± 0.81	4.21 ± 0.81
C17:0	1.22 ± 0.08	2.56 ± 0.09	1.74 ± 0.11	2.48 ± 0.42
C18:0	5.91 ± 0.51	9.03 ± 0.30	6.98 ± 0.42	10.83 ± 1.00
C18:1n12t	0.23 ± 0.05	nd	0.13 ± 0.05	0.06 ± 0.14
C18:1n9t	0.02 ± 0.02	nd	0.04 ± 0.04	0.06 ± 0.14
C18:1n9	3.82 ± 0.47	10.46 ± 0.42	7.52 ± 0.28	9.30 ± 0.80
C18:1n7	3.17 ± 0.10	3.82 ± 0.11	3.47 ± 0.16	3.88 ± 0.52
C18:2n6t	nd	nd	0.01 ± 0.01	0.03 ± 0.06
C18:2n6	1.54 ± 0.13	1.75 ± 0.05	1.63 ± 0.06	1.75 ± 0.21
C18:3n6	0.47 ± 0.03	0.18 ± 0.02	0.27 ± 0.05	0.13 ± 0.02
C19:0	nd	0.52 ± 0.02	0.45 ± 0.08	0.50 ± 0.07
C18:3n3	1.51 ± 0.25	1.71 ± 0.13	2.19 ± 0.26	1.02 ± 0.27
C18:4n3	1.87 ± 0.17	1.27 ± 0.09	1.73 ± 0.13	0.49 ± 0.16
C19:2n6	nd	0.03 ± 0.01	0.12 ± 0.10	0.05 ± 0.01
C20:0	0.25 ± 0.02	0.47 ± 0.03	0.46 ± 0.07	0.47 ± 0.16
C20:1n15	0.02 ± 0.01	0.04 ± 0.01	0.02 ± 0.01	0.04 ± 0.03
C20:1n12	0.13 ± 0.02	2.08 ± 0.11	0.80 ± 0.10	1.38 ± 0.48
C20:1n9	0.39 ± 0.09	1.07 ± 0.04	1.22 ± 0.19	0.90 ± 0.24
C20:1n7	0.28 ± 0.03	1.96 ± 0.11	0.98 ± 0.11	1.53 ± 0.59
C20:2n6	0.24 ± 0.03	0.67 ± 0.11	0.60 ± 0.14	0.99 ± 0.32
C20:3n6	0.27 ± 0.03	0.17 ± 0.03	0.27 ± 0.11	0.17 ± 0.07

Table 1 continued.

Fatty acid	Menhaden	70MH30SP	30MH70SP	Spot
C20:4n6	1.45 ± 0.22	1.68 ± 0.07	1.04 ± 0.13	4.56 ± 0.75
C20:3n3	0.30 ± 0.05	0.50 ± 0.08	0.53 ± 0.14	0.44 ± 0.06
C20:5n3	7.80 ± 0.35	5.23 ± 0.20	6.13 ± 0.34	6.52 ± 0.65
C22:0	0.52 ± 0.11	0.39 ± 0.07	0.71 ± 0.29	0.38 ± 0.09
C22:1n9	0.22 ± 0.08	0.25 ± 0.07	0.69 ± 0.37	0.28 ± 0.08
C22:2n6	0.11 ± 0.07	0.12 ± 0.07	0.09 ± 0.06	0.15 ± 0.09
C22:4n6	0.36 ± 0.12	0.63 ± 0.12	0.68 ± 0.33	1.15 ± 0.29
C22:3n3+C22:5n6	1.38 ± 0.20	0.94 ± 0.14	1.15 ± 0.40	1.34 ± 0.48
C22:5n3	1.44 ± 0.18	1.66 ± 0.17	1.72 ± 0.36	2.10 ± 0.38
C24:0+C22:6n3	13.87 ± 1.02	8.88 ± 0.40	10.09 ± 0.82	13.83 ± 3.30
C24:1n9	0.74 ± 0.16	0.62 ± 0.14	1.13 ± 0.32	0.56 ± 0.27

Table 3. Length, weight, and lipid levels of liver and belly flap samples for each diet fed during the experiment. Values represent the mean and 1 standard error of the mean.

Week	Diet	Length (mm)	Weight (g)	Lipid content Liver (% WW)	Belly flap (% WW)
0	-	352.83 ± 6.59	607.00 ± 33.50	5.54 ± 0.34	12.62 ± 1.28
21	Menhaden	360.75 ± 7.43	745.25 ± 55.68	9.93 ± 1.31	22.19 ± 4.45
48	Menhaden	390.50 ± 6.89	913.50 ± 74.15	10.37 ± 0.67	18.89 ± 2.42
21	70MH:30SP	355.50 ± 9.15	593.00 ± 50.40	5.64 ± 0.45	8.10 ± 0.59
48	70MH:30SP	369.25 ± 43.20	801.25 ± 120.31	5.14 ± 0.12	11.36 ± 2.01
21	30MH:70SP	358.00 ± 27.00	629.50 ± 133.50	6.49 ± 0.24	10.10 ± 0.41
48	30MH:70SP	361.25 ± 6.98	744.28 ± 64.02	6.43 ± 0.66	9.47 ± 2.40

Figure 1. NMDS plot of fatty acids identified in the diets, liver, and belly flap tissue in striped

bass during the experiment. Fish were fed a diet of spot for six weeks and sampled on

day 0. Striped bass were fed three diets: menhaden in tank 1 (T1), 70% menhaden:30%

spot in tank 2 (T2), and 30% spot:70% menhaden in tank 3 (T3). These fish were

sampled after a period of 6 weeks.

Figure 2. Mean values of select fatty acids from striped bass fed a mixed diet of 70% menhaden

30% spot for a) liver samples and b) belly flap samples. Error bars represent 1 standard

error of the mean.

Figure 3. Mean values of select fatty acids from striped bass fed a mixed diet of 30% menhaden

70% spot for a) liver samples and b) belly flap samples. Error bars represent 1 standard

error of the mean.

Figure 4. Proportional contributions of spot and anchovy to the fatty acid signature of striped

bass during the baseline feeding of spot as calculated by the QFASA model. Anchovy

was used as the feed during the acclimation of striped bass to the tank system. The

dietary dataset of fatty acids variables was used to evaluate contributions.

Figure 5. Proportional contributions of spot and menhaden to the fatty acid signature of striped

   bass as calculated by the QFASA model. The full dataset of fatty acids variables was

   used to evaluate contributions. Day refers to when the striped bass was sacrificed, MH

   is the 100% menhaden diet, 70MH:30SP is the 70% menhaden 30% spot diet, and

   30MH:70SP is the 30% menhaden 70% spot diet.

Figure 6. Proportional contributions of spot and menhaden to the fatty acid signature of striped

   bass as calculated by the QFASA model. The dietary dataset of fatty acids variables

   was used to evaluate contributions. Day refers to when the striped bass was sacrificed,

   MH is the 100% menhaden diet, 70MH:30SP is the 70% menhaden 30% spot diet, and

   30MH:70SP is the 30% menhaden 70% spot diet.

Figure 1.

Figure 2.

Figure 3.

Figure 4.

Figure 5.

Figure 6.

a) Liver

b) Belly flap

Sample day and diet
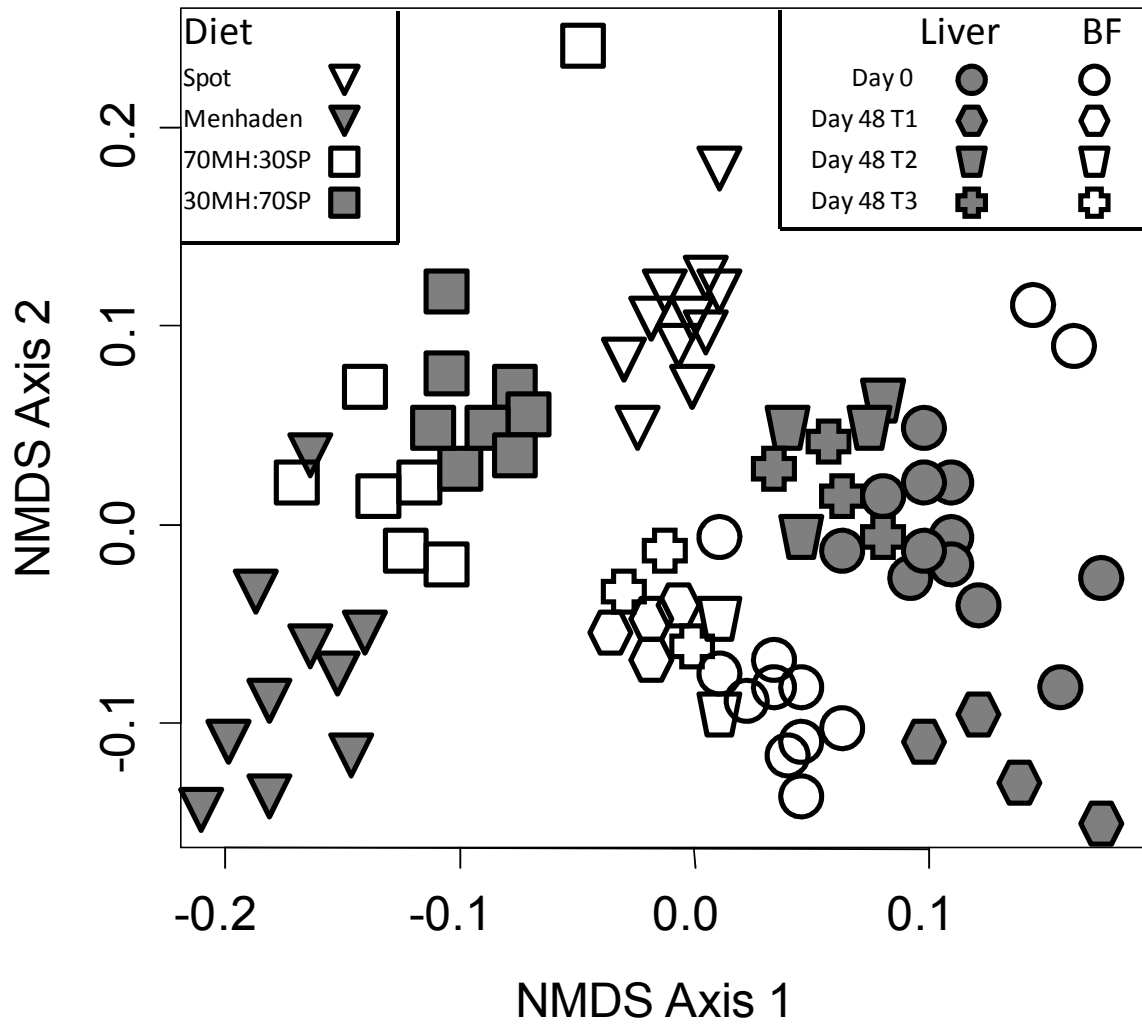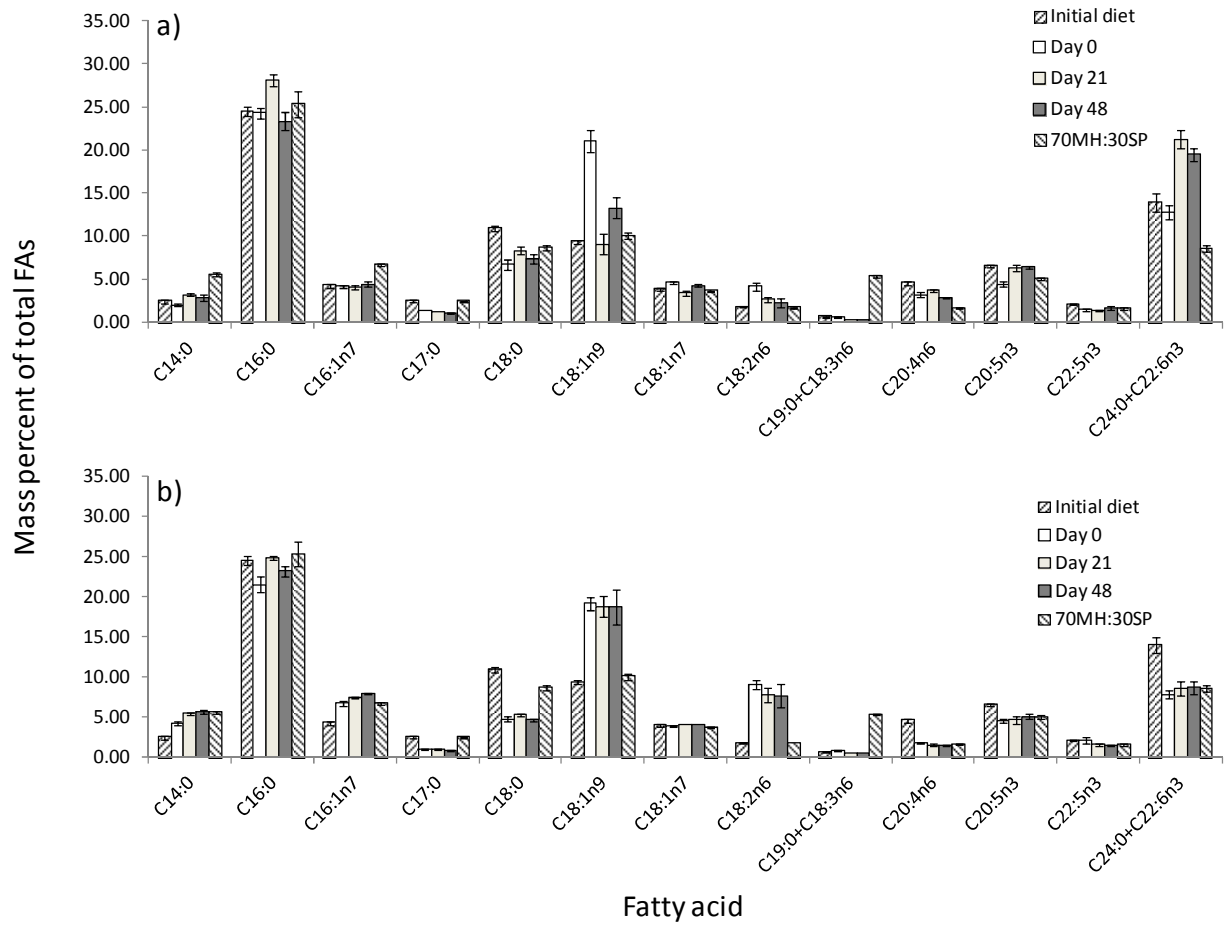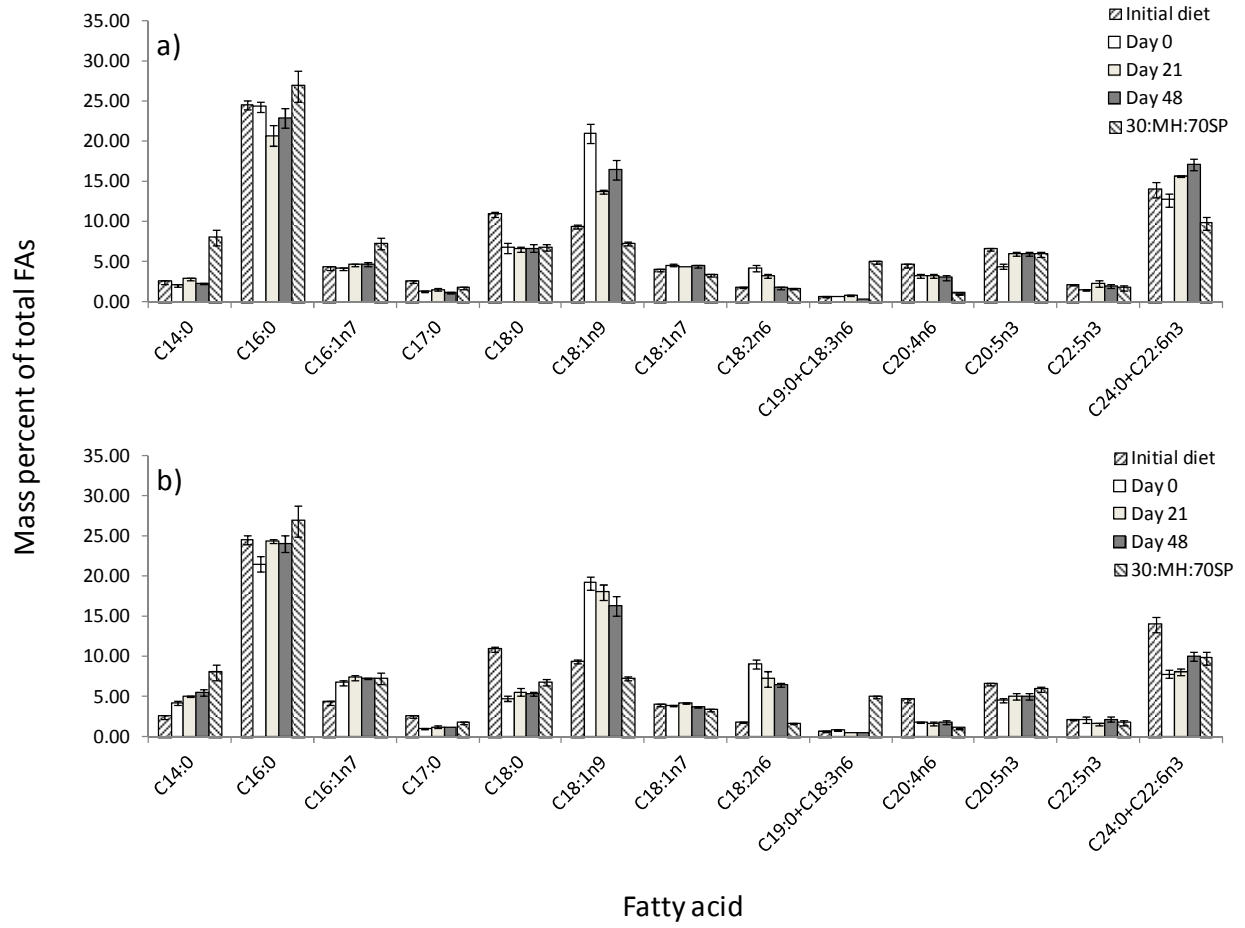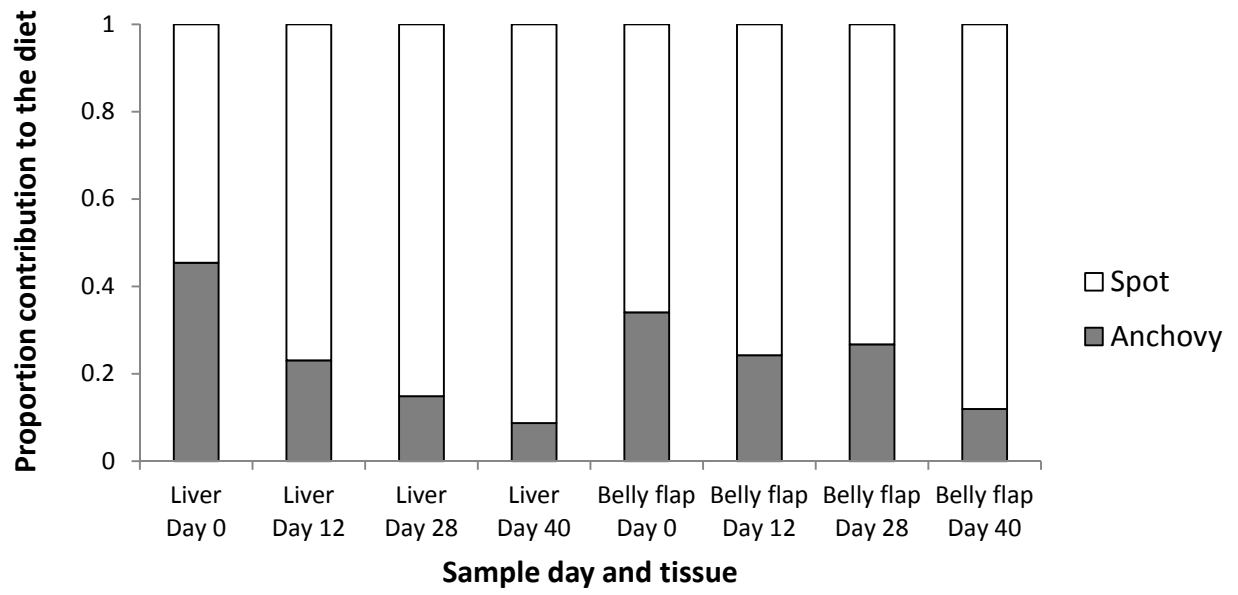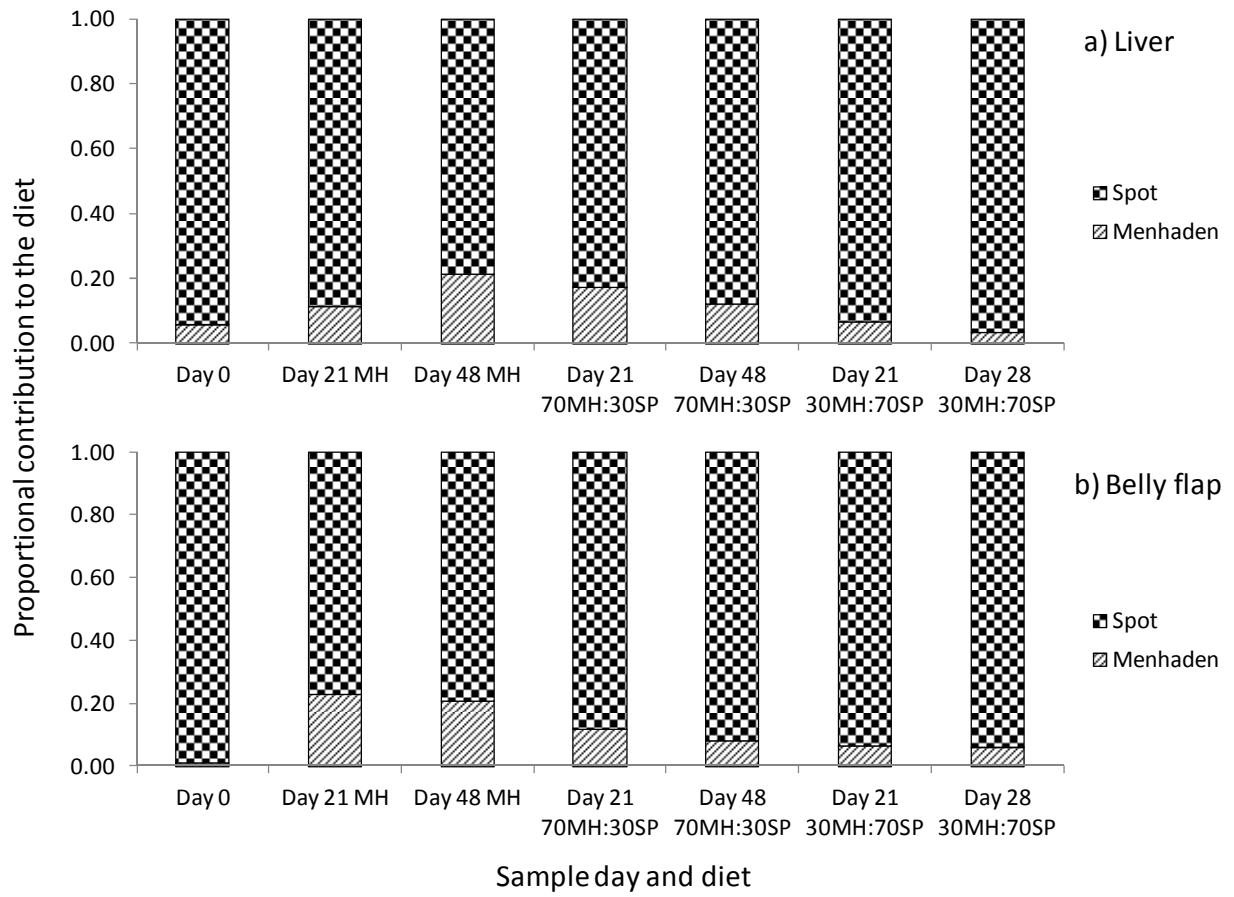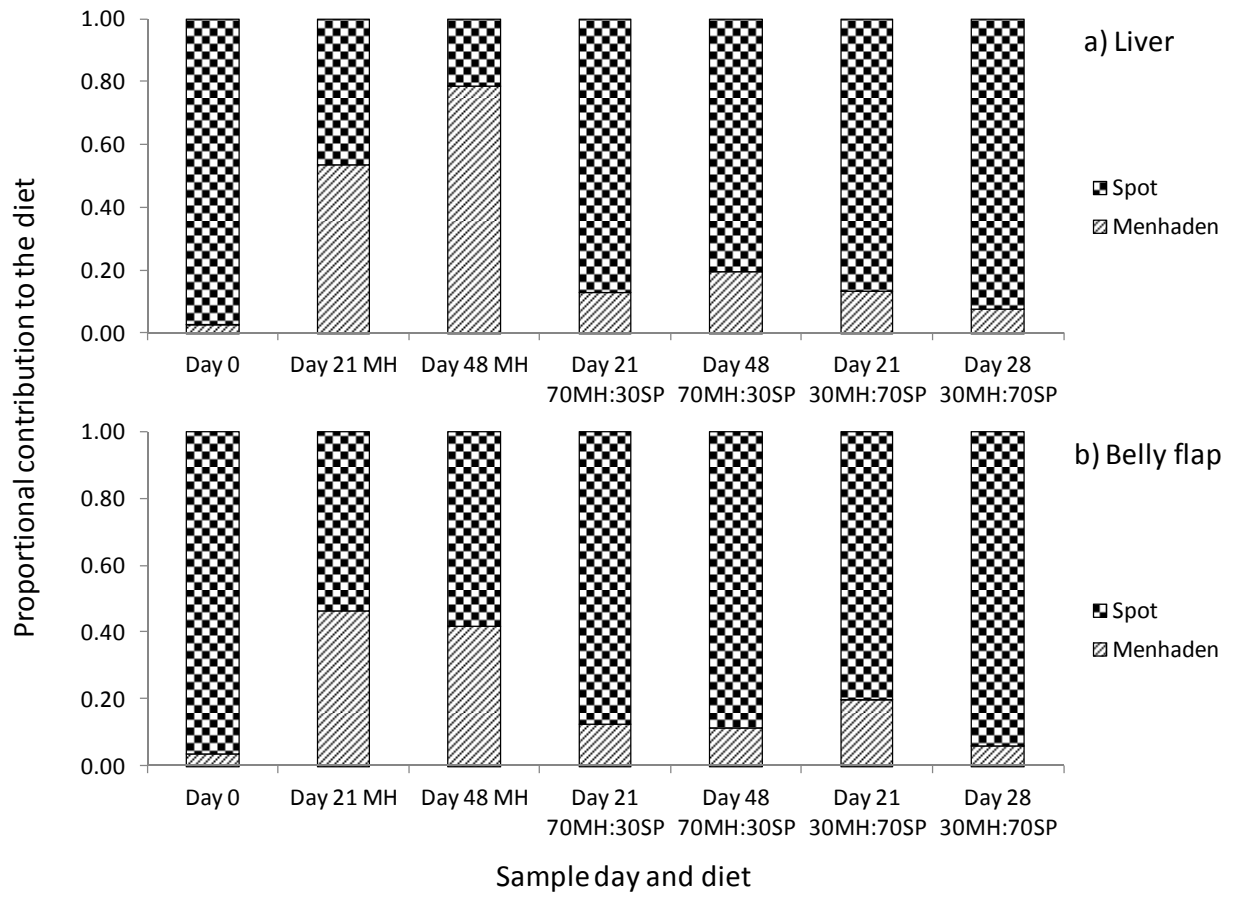
Chapter 6 - Field validation of quantitative fatty acid signature analysis (QFASA) for

Chesapeake Bay striped bass

Abstract

Food web models are constructed based upon information gathered from analyzing stomach contents of the candidate species. These methods have well known biases, e.g. shorter retention time for prey items in the gut at elevated temperatures, over estimation of prey with digestion resistant hard parts, and metrics skewed towards the last meal. A biochemical alternative, fatty acid (FA) analysis, has been shown in previous studies to give a more accurate estimation of the feeding history. Recently, a quantitative model based on FAs has been developed. Quantitative FA signature analysis (QFASA) can estimate proportions of prey items consumed. The goal of this study was to corroborate the findings of the QFASA model and stomach contents for wild caught striped bass. Seventy-eight striped bass (*Morone saxatilis*) were caught in the Choptank River during October 2009. During the fall, the QFASA model should have the highest chance of success due to high and continued feeding on Atlantic menhaden (*Brevoortia tyrannus*), which provides a simple natural condition on which to test the model. These fish were analyzed for FAs and prey species caught in the upper Chesapeake Bay in fall were used as the possible prey items. Stomach contents revealed that striped bass were indeed feeding predominantly on menhaden, however, the QFASA model using the dietary dataset (16 FA) and the extended dataset (30 FA) determined that bay anchovy (*Anchoa mitchilli*) and spot (*Leiostomus xanthurus*) were the major constituents of the diet. Studies using QFASA on homeotherms (marine mammals and sea birds) have produced reliable and verifiable prey structure in the wild. However, the results of the present study indicate that the QFASA model needs to be further optimized for fish species in a laboratory setting before it can employed in the field.

Introduction

The interactions between predator and prey and the subsequent construction of food webs are important themes in the ecology of fishes (Baird and Ulanowicz 1989). These food webs provide links among the various species modeled, and are often based on estimates of the diet from the recovery of prey items in the stomachs of the predator (e.g., Berg 1979, Durbin et al. 1983, Blanco et al. 2003, Harvey et al. 2003). The effects of predation become increasingly important when the prey species are also commercially harvested (Yodzis 2001). Although the idea of connectedness of species is not a new one (Paine 1980), it is only recently that this concept has been incorporated into management strategies, e.g. ecosystem based fisheries management (Pikitch et al. 2004). These models attempt to include species interactions rather than modeling each species as an individual unit (Brodziak and Link 2002). However, these models are all based upon estimates of diet from stomach contents analysis, which has well known biases (Pierce and Boyle 1991).

An analysis of stomach contents often provides information on the recent feeding history. The rate of digestion is not equal for all items consumed. Hard parts associated with prey items may be more resistant to digestion (Hyslop 1980), leading to an overestimation of these particular prey species. The digestive capacity of a predator is not uniform and is influenced by temperature. Warmer temperatures will lead to higher metabolic activities and reduce the time in which prey items are retained in the gut (He and Wurtsbaugh 1993). These biases have the potential to insert a large degree of error when calculating diets for fish, and this error will be carried through the construction of food web models.

Alternative methods for estimating diet include the use of fatty acids (FAs; Desvilettes et al. 1997, Kharlamenko et al. 1995, Alfaro et al. 2006) or stable isotopes (Hansson et al. 1997,

Pinnegar and Polumin 2000, Sotiropoulos et al. 2004) as trophic markers. FAs are a large

constituent of lipids and undergo little to no modification when incorporated into a predator's

tissue (Tocher 2003). Therefore, it can be argued that the FAs present in a predator should mimic

those of the prey species consumed (Kirsch et al. 1998). Studies that have attempted to use FA

analysis with fish have focused on using prey FA signatures (a compilation of all FAs identified

within an individual) to estimate what species a predator was consuming (Budge et al. 2002,

Kainz et al. 2004, Alfaro et al. 2006). This qualitative estimate of diet is limited because

proportions of different prey groups consumed cannot be inferred.

A model developed recently, quantitative FA signature analysis (QFASA; Iverson et al.

2004), allows the proportion of each diet item to be estimated by comparing the signatures of

prey versus the signature of the predator. Predator FA metabolism is accounted for with

calibration coefficients (CC), which are calculated in captive feeding trials (Iverson et al. 2004).

These values account for the differential mobilization and use of fatty acids in a predator. The

values will be one if there is an exact match between prey abundance and predator abundance of

that fatty acid, less than one if that fatty acid is more prevalent in prey tissues and greater than

one if the fatty acid is more prevalent in the predator's tissues. This model has been used for

marine mammals (Thiemann et al. 2008, Tucker et al. 2009, Meynier et al. 2010) and sea birds

(Iverson et al. 2007), with little research performed on fish species (Budge et al. 2011).

Constructing food web models in the Chesapeake Bay has been performed, but

difficulties are abundant. Striped bass (*Morone saxatilis*), for example, are apex predators that

spawn in the bay. The spawning population returns to the coastal stocks after breeding, while

immature fish will remain in the bay for the first two to eight years of life, depending on the sex

of the fish (Setzler-Hamilton and Hall 1991). The Chesapeake Bay is categorized as a continuum

of salinity with oligohaline (0 – 6 parts of thousand [ppt]), mesohaline (6-18 ppt), and polyhaline (>18 ppt) regions (Stroup and Lynn 1963). Salinity structures the fish community in each of these regions which leads to different community compositions (Jung and Houde 2003). The effects of season and salinity make modeling food webs for the entire Chesapeake Bay troublesome. Baird and Ulanowicz (1989), for example, were only able to model species interactions for the mesohaline region of the bay in summer.

Accurate estimates for an apex predator like striped bass are necessary to properly manage the population. This fish has been an important commercial and recreational resource (Chesapeake Bay Fisheries Ecosystem Advisory Panel 2006), but due to overfishing, the population began a rapid decline in the 1970s (ASMFC 1981). This reduction in population resulted in a moratorium being placed on the species in Maryland and Virginia during the 1980s, which led to a population recovery in the 1990s (ASMFC 1995), and stock assessments have shown the population is stable. However, there have been observations of emaciated fish with external lesions and internal granulomas (Baya 1998, Uphoff 2003). These symptoms are linked to the infectious bacteria mycobacteriosis (Heckert et al. 2001, Rhodes et al. 2004). Theories suggest that striped bass might be competing for their preferred prey, menhaden, with other fish species and humans. Hartman (2003) demonstrated that striped bass feeding demand in the Hudson River has exceeded the supply of prey species. A similar situation may be occurring in the Chesapeake Bay which could lead to an increased susceptibility to bacterial infections. Malnutrition in fish can lead to suppressed immune systems, which in turn make the fish susceptible to infectious agents like *Mycobacterium* spp. (Jacobs et al. 2009).

The goal of this study was to use the QFASA model on wild caught Chesapeake Bay striped bass to estimate the contribution of prey species to the diet. To reach this goal, our

specific objectives were: 1) to estimate striped bass diets using stomach contents, and 2) to compare the stomach contents to the results of the QFASA model to determine if and to what proportion spot (*Leiostomus xanthurus*), menhaden, bay anchovy (*Anchoa mitchilli*), and blue crab (*Callinectes sapidus*) collected in the same season as the striped bass contributed to the striped bass FA signature in the belly flap tissue.

Materials and Methods

Sample collection and preparation

Seventy-eight striped bass were collected in October 2009 by hook and line, with the help of a local fisherman (J. Price), in the vicinity of the Choptank River, Maryland. Fish were vacuum sealed and stored at $-20^{o}C$ once returned to the lab. Striped bass were thawed at the time of analysis, weighed to the nearest 0.1 gram, and measured to the nearest millimeter (total length). A sample of the belly flap was removed for lipid extraction and FA analysis. The belly flap (with skin intact) follows a line marked by the termination of the ribs and initiation of the peritoneum of the body wall (Jacobs et al. 2008). The belly flap represents a depot of fat storage and a tissue with little commercial value, making it an ideal candidate for estimating diet with FAs. All tissues were ground with a chemical drying agent, diatomaceous earth (Dionex, Sunnyvale, CA) and had a known amount of internal standard (C13:0 triacylglycerol) added to determine the efficiency of the extraction process.

Lipids were extracted under nitrogen using an Accelerated Solvent Extractor (ASE 300, Dionex, Sunnyvale, CA) with a mixture of 3:1 (v/v) dichloromethane:methanol and 0.1% butylated hydroxytoluene (BHT) as an antioxidant. Samples were extracted once at $100^{o}C$ and 1500 psi (USEPA 2007). Samples were back-extracted using a 0.88% potassium chloride

solution in deionized water to remove non-lipid materials from the extracts (Folch et al. 1957). Total lipid extracted was determined gravimetrically for each sample and reported as a percent of wet weight.

Extracted lipid species containing FAs as various derivatives were transesterified using a solution of sulfuric acid and methanol. In this process, acylglycerols are broken apart to their constituents: glycerol and FAs methyl esters (FAMEs). Free FAs present in the lipid mixture are also converted to FAMEs (Christie 2003). Sample extracts were cleaned with silica gel and anhydrous sodium sulfate to remove polar components and water, which may degrade the performance of the gas chromatograph column. The silica column was conditioned with hexane and the sample was added along with hexane and diethyl ether, 95:5, v/v (Christie 2003). The mixture was concentrated to 5 ml and diluted to a final total lipid concentration of 100 μg/ml. Finished samples were run on a gas chromatograph (Agilent 6890N Network GC System, Santa Clara, CA) / mass spectrometer (GC/MS, Agilent, 5973 Network Mass selective detector, Santa Clara, CA) in select ion monitoring mode.

The GC/MS was used to identify and quantify FA methyl esters in each sample. FAMEs were injected into a fused capillary column, DB-23 60m length X 0.250mm OD X 0.25μm film thickness (Agilent, Santa Clara, CA) using an autosampler (Agilent 7683 Series Auto Sampler). Hydrogen was used as a carrier gas. The temperature program was as follows: hold at an initial temp of 50^oC for 2 min, hold at 150^oC for 1 min after ramping at $20^oC \cdot min^{-1}$, ramp at $1.25^oC \cdot min^{-1}$ until 215^oC. The FA retention times were compared to known standards from Nu-Chek Prep (GLC MIX 85, 411, 642, 632, Elysian, MN). These four standard mixtures were combined along with stearidonic acid (C18:4n3, Cayman Chemical, Ann Arbor, MI) to make a total FAME standard solution of 12.25 μg/ml. Matreya 25 FAME mixture (Pleasant Gap, PA)

was used to obtain retention times for C20:1n7. Restek cis/trans FAME mixture (Bellefonte, PA) was used to obtain retention times for C18:1n9 trans and C18:2n6 trans. These standards and an internal standard of C13:0 FAME (100 µg/ml) were added to quantify the amount of each FA methyl ester present. The GC/MS tracks the select ion signature of each compound and was used to verify the identity of each peak. There were peaks which consisted of more than one FA and it was impossible to determine which FA(s) constituted the peaks. Therefore, the term "FA complex" was assigned to this peak. The complex refers to which FAs could be responsible for the peak, e.g. C22:3n3 and C22:5n6.

Statistical analyses

Nonmetric multidimensional scaling (NMDS) was performed on the data (R Development Core Team 2010, Vienna, Austria). This ordination technique is an iterative search for the best position of samples in multidimensional space, and uses the stress value as an indicator for the best solution. A distance matrix was calculated for the FAs, and the samples are ordinated based on the distances calculated (McCune and Grace 2002).

Calibration coefficients and FA subsets

Calibration coefficients were calculated during a captive feeding study with striped bass and spot. The values account for the metabolism of a FA by striped bass, as different FAs may have different fates when they are digested. Twelve striped bass were sacrificed after being fed spot for six weeks, and the FA signature of the belly flap was analyzed. Eleven samples of spot were also analyzed for their FA signature. The coefficients were obtained by dividing the proportion of a FA in striped bass by the proportion of that same FA in spot (Iverson et al. 2004). Each striped bass is compared to each prey item, resulting in 132 calibration coefficients for each

FA. The six highest and lowest values were removed before the mean was calculated to remove any extreme outliers (10% trimmed mean; Iverson et al. 2004).

A subset of the FAs is used for the QFASA model. In our experiment, two FA datasets were created. The first, an extended FA dataset, included all FAs that had a proportion > 0.1% (n=30). The second dataset, a dietary dataset, consisted of FAs derived primarily from the diet (n=16). For a list of these calibration coefficients and fatty acid subsets, see Chapter 5: Table 1. Once FAs the datasets were constructed, the proportions were renormalized to 100%.

Diet estimation

Stomach contents of striped bass were examined and a percent of occurrence for each prey taxa was calculated. Vertebrae of digested fish were counted in order to classify the fish species (K. Hartman; personal communication). The diet of striped bass was estimated using the QFASA model (Iverson et al. 2004). The model uses the prey FA signatures and tries to estimate which mixture of prey comes closest to matching the predator's actual signature. The best mixture of prey FAs is weighted by the fat content of the prey species, as fish with higher lipid levels will contribute a larger concentration of FAs. The model uses an optimization procedure which aims to minimize the distance between the predicted and observed predator signature, the Kullback-Liebler (KL) distance. The optimization uses a quasi-Newton algorithm with a Broyden-Fletcher-Goldfarb-Shanno formula and was carried out with a package developed at Massey University, New Zealand (FA solution – Dr. R. Sheriff and A/Prof P.C.H. Morel).

Results

The prey analyzed in this experiment differed in their fat content, with menhaden and bay anchovy having higher lipid levels than blue crab and spot. Striped bass belly flap tissue had an average lipid content of 5.24% by wet weight (Table 1).

The complete FA signature for striped bass and the prey items are given in Table 2. There were marked differences among the prey species in regards to the proportions of individual FAs (Fig. 2). For example, blue crab had approximately twice the relative amount of C18:2n6, while menhaden had the highest levels of C16:1n7 and C24:1n9. There were also differences related to whether the prey taxa were benthic or pelagic, e.g. menhaden and bay anchovy (pelagic) had higher levels of C14:0 and C16:0 compared to spot and blue crab (benthic).

Diet estimation

The percent biomass of prey in the stomachs was calculated for striped bass in this experiment. Of the 78 striped bass (505.91 ± 5.27 mm; Table 1) analyzed, 36 had empty stomachs. The diet of striped bass specimens that had full stomachs consisted of menhaden (93.12%), blue crab (1.25%) and unidentified fish (5.63%; not included in Fig. 3). Unidentified fish consisted of vertebral columns that were incomplete, preventing positive identification. Diet estimations (as percent biomass of prey) from Hartman and Brandt (1995) and Overton et al. (2000) are provided for age three striped bass in the middle Chesapeake during the fall (September – October) for comparison.

A qualitative approach was taken with the FA signatures from each taxa, and ordinated using NMDS (Fig. 2). Based upon the spacing of species and the 95% confidence interval around them, spot contributed the highest proportion to striped bass FA signatures, followed by bay

169

anchovy. There was a minimal degree of overlap between menhaden and striped bass, and blue crab and striped bass.

Quantitative estimates of diet based on stomach contents and the QFASA model were not in agreement (Fig. 3). According to the model, spot contributed the highest proportion to the diet, followed by bay anchovy. Menhaden and blue crab made minor contributions to the diet. The results from the 16 FA dataset and 30 FA dataset yielded different results. The truncated dataset estimated the contribution of spot and bay anchovy to by 59.22 and 37.61%, respectively; while the full dataset estimated the contribution of spot and bay anchovy to be 80.66 and 16.34%, respectively. The results from both models had high KL distances: 30.32 for the 16 FA model and 27.32 for the 30 FA model.

Discussion

The results of this study indicate that the QFASA model was unable to correctly characterize the diets of striped bass collected from the Chesapeake Bay. FA incorporation into a predator's signature takes several weeks (Kirsch et al. 1998). Striped bass were sampled in October based upon the findings of previous studies in which striped bass were feeding heavily on menhaden between August and December (Hartman and Brandt 1995, Overton et al. 2000). This suggested that by collecting striped bass in October, sufficient time would be allowed for the menhaden signature to become incorporated into striped bass tissues. The stomach contents reinforced this notion due to the higher number of menhaden present in the diet.

Lipid levels in the wild caught striped bass indicate that these fish have been feeding for an extended period. Jacobs et al. (2008) found that age-1 striped bass (mean length = 241.8mm) had belly flap lipid levels of 8.67% by wet weight for fed fish and 3.72% by wet weight for

starved fish after a period of 42 days. Larger fish tend to store more lipid than smaller fish, and the striped bass in our experiment were around age-3 fish. A lipid level of 5.24% by wet weight would point to a situation where striped bass have been feeding below satiation, but were not being starved.

Striped bass in this experiment were caught within days of each other in order to limit the variation in the feeding regime. The stomach contents of the striped bass collected indicated that the predominant prey item was Atlantic menhaden. Hartman and Brandt (1995) and Overton et al. (2000) found comparable diets for similar sized fish in the fall. While other prey items were found in the diets for our study, the unidentified group may have been menhaden whose spinal columns were separated during the digestive process.

The switch to a new prey item in fish is detectable after a period of three weeks (Kirsch et al. 1998) but the signature may not be completely stable until 12 to 14 weeks (Jobling 2003, Budge et al. 2011). The instability of the FAs in the signature of fish may prevent quantitative estimates of diet using QFASA. The diet of a fish may need to be constant for a period of months rather than weeks or days in order for the QFASA model to be utilized. This situation is not likely, as striped bass are known to change their feeding throughout the year (Manooch 1973, Overton et al. 2009). However, based upon the findings of this study, striped bass may have been feeding on spot and bay anchovy for the past several weeks, prior to the sampling period.

Some striped bass are known to move to different areas over small temporal scales, e.g. one to four weeks (Mansueti 1961). These different areas may be a different river system in which prey species are the same, but the FA signature of the prey is different (Chapter 2). The movements of striped bass would prevent researchers from knowing from which system prey species originated, possibly leading to using an erroneous dataset of FAs to represent potential

171

prey species. While this would still be an issue if FA signatures were incorporated over a period of three weeks, it becomes a serious obstacle to overcome if FAs need 12 to 14 weeks to stabilize in fish tissues.

The results of the QFASA model indicated that spot and bay anchovy were the predominant prey of striped bass; with blue crab and menhaden making small contributions to the diet. The proportional contribution of spot was less in the model that used the dietary FA dataset (16 FA) versus the full dataset (30 FA). Studies have shown that the dataset based only on FAs obtained from the diet yields better results (Iverson et al. 2004, Beck et al. 2007). We have also found that even when the model was incorrect, better results were obtained from this truncated dataset (Chapter 5).

Studies have shown that the FA signature of fish species is affected by season (Bandarra et al. 2001) and location (Recks and Seaborn 2008). Therefore, prey samples analyzed were collected in the same season (often in the same month) as the striped bass. However, due to logistical constraints, prey samples were collected from areas in the upper bay as close to the Choptank River as possible, but not necessarily from the same system. No discernible pattern in FA signature was detected for anchovy and menhaden in the upper bay versus the lower bay (Chapter 2), and it becomes difficult to determine whether collecting samples from areas different than collection of striped bass affected the results seen in this experiment. It would be advisable for future studies to collect all species at the same time in the same location (if possible) to try and remove any spatial error.

The process by which the FA profile changes in striped bass could provide insight into the shortcomings of the QFASA model. A dilution model worked well with Atlantic salmon (*Salmo salar*) to document the change of FAs from one diet to another (Jobling 2003). In this

model, the FAs from the original diet become diluted or lowered in prevalence as the new diet is consumed. FA signatures from both diets would be present in the tissues until FAs from the new diet have finally diluted the concentrations of fatty acids from the previous diet. Striped bass in this study may not have been feeding on menhaden long enough to allow for the dilution of spot signatures.

The FA model QFASA does not appear feasible for estimating the diets of striped bass in the Chesapeake Bay at this time. The results indicate the diet was dominated by a prey that was not present in the stomachs. There is a possibility that the FA signature did not have adequate time to stabilize and reflect the current diet. In order for this technique to be viable in estimating diets of wild fish, there must be a minimal lag time between the corroboration of FA incorporation and stomach contents. Further laboratory studies are needed to determine whether FAs present in striped bass represent an amalgam of all prey taxa, or if the previous signature is washed out completely replaced by the new signature. More work also needs to be done to determine which tissue represents the best candidate for the QFASA model. The belly flap is a long term storage depot, and it may be difficult to overwrite the fatty acid signature. Other areas of the fish that contain mobile fatty acids, e.g. the liver of the blood, may represent a better alternative to further test this model.

Literature cited

Alfaro, A.C., F. Thomas, L. Sergent, and M. Duxbury. 2006. Identification of trophic interactions within an estuarine food web (northern New Zealand) using FA biomarkers and stable isotopes. Estuarine, Coastal and Shelf Science 70:271-286.

ASMFC (Atlantic States Marine Fisheries Commission). 1981. Interstate fisheries management plan for the striped bass. Fisheries Management Report No. 1, Washington, D.C.

ASMFC (Atlantic States Marine Fisheries Commission). 1995. Amendment #5 to the interstate fishery management plan for Atlantic striped bass. ASMFC. Fisheries Management Report 24, Washington, D.C.

Baird, D., and R. E. Ulanowicz. 1989. The seasonal dynamics of the Chesapeake Bay ecosystem. Ecological Monographs 59:329-364.

Bandarra, N., I. Batista, M.L. Nunes, and J.M. Empis. 2001. Seasonal variation in the chemical composition of horse-mackerel (*Trachurus trachurus*). European Food Resources and Technology 212:535-539.

Baya, A.M. 1998. Gram positive bacteria and skin lesions in Chesapeake Bay fish. Proc Workshop Causes and Effects of Ulcerative Lesions in Fish, Easton, MD. Maryland Department of Natural Resources, Oxford, MD. p 11.

Beck, C.A., S.J. Iverson, W.D. Bowen, and W. Blanchard. 2007. Sex differences in grey seal diet reflect seasonal variation in foraging behavior and reproductive expenditure: evidence from quantitative FA signature analysis. Journal of Animal Ecology 76:490-502.

Berg, J. 1979. Discussion of methods of investigating the food of fishes, with reference to a preliminary study of the prey of *Gobiusculus flavescens* (Gobiidae). Marine Biology 50:263-273.

Blanco, S., S. Romo, M.J. Villena, and S. Martinez. 2003. Fish communities and food web interactions in some shallow Mediterranean lakes. Hydrobiologia 506-509:473-480.

Brodziak, J. and J. Link. 2002. Ecosystem-based fishery management: what is it and how can we do it? Bulletin of Marine Science 70:589-611.

Budge, S. M., Iverson, S. J., Bowen, W. D and Ackman, R. G. 2002. Among- and within-species variability in the FA signatures of marine fish and invertebrates on the Scotian Shelf, Georges Bank, and southern Gulf of St. Lawrence. Canadian Journal of Fisheries and Aquatic Sciences 59, 886-898.

Budge, S.M., S.N. Penney, S.P. Lall. 2011. Response of tissue lipids to diet variation in Atlantic salmon (*Salmo salar*): implications for estimating diets with FA analysis. Journal of Experimental Marine Biology and Ecology 409:267-274.

Chesapeake Bay Fisheries Ecosystem Advisory Panel (National Oceanic and Atmospheric Administration Chesapeake Bay Office). 2006. Fisheries ecosystem planning for Chesapeake Bay. American Fisheries Society, Trends in Fisheries Science and Management 3, Bethesda, Maryland.

Christie, W.M. 2003. Lipid Analysis. PJ Barnes and Associates. Bridgewater, England.

Desvilettes, C., G. Bourdier, C. Amblard, and B. Barth. 1997. Use of FAs for the assessment of zooplankton grazing on bacteria, protozoans, and microalgae. Freshwater Biology 38:629-637.

Durbin, E.G., A.G. Durbin, R.W. Langton, and R.E. Bowman. 1983. Stomach contents of silver hake, *Merluccius bilinearis*, and Atlantic cod, *Gadus morhua*, and estimation of their daily rations. Fishery Bulletin 81:437-454.

Folch, J., M. Lees, and G.H. Sloane Stanley. 1957. A simple method for the isolation and
purification of total lipids from animal tissues. The Journal of Biological Chemistry:497-
509.

Hansson, S., J.E. Hobbie, R. Elmgren, U. Larsson, B. Fry, and S. Johansson. 1997. The stable
nitrogen isotope ratio as a marker of food-web interactions and fish migration. Ecology
78:2249-2257.

Hartman, K.J. 2003. Population-level consumption by Atlantic coastal striped bass and the
influence of population recovery upon prey communities. Fisheries Management and
Ecology 10:281-288.

Hartman, K. J., and S. B. Brandt. 1995. Trophic resource partioning, diets, and growth of
sympatric estuarine predators. Transactions of the American Fisheries Society 124:520-
537.

Harvey, C.J., S.P. Cox, T.E. Essington, S. Hansson, and J.F. Kitchell. 2003. An ecosystem model
of food web and fisheries interactions in the Baltic Sea. ICES Journal of Marine Science
60:939-950.

He, E. and W.A. Wurtsbaugh. 1993. An empirical model of gastric evacuation rates for fish and
an analysis of digestion in piscivorous brown trout. Transactions of the American
Fisheries Society 122:717-730.

Heckert, R.A., S. Elankumaran, A. Milani, and A. Baya. 2001. Detection of a new
Mycobacterium species in wild striped bass in the Chesapeake Bay. Journal of Clinical
Microbiology 39:710-715.

Hyslop, E.J. 1980. Stomach contents analysis – a review of methods and their application.
Journal of Fish Biology 17:411-429.

Iverson, S.J. C. Field, W.D. Bowen, and W. Blanchard. 2004. Quantitative FA signature analysis: a new method of estimating predator diets. Ecological Monographs 74:211-235.

Iverson, S.J., A.M. Springer, and A. Kitaysky. 2007. Seabirds as indicators of food web structure and ecosystem variability: qualitative and quantitative diet analyses using FAs. Marine Ecology Progress Series 352:235-244.

Jacobs, J. M., M.R. Rhodes, B. Martin, D. McIntosh, W.F. Van Heukelem, and R.M. Harrell. 2008. Tissue-specific sampling for estimation of striped bass whole-body proximate composition. Transactions of the American Fisheries Society 137:386-392.

Jacobs, J.M., M.R. Rhodes, A. Baya, R. Reimschuessel, H. Townsend, and R.M. Harrell. 2009. Influence of nutritional state on the progression and severity of mycobacteriosis in striped bass *Morone saxatilis*. Diseases of Aquatic Organisms 87:183-197.

Jobling, M. 2003. Do changes in Atlantic salmon, *Salmo salar* L., fillet FAs following a dietary switch represent wash-out or dilution? Test of a dilution model and its application. Aquaculture Research 34:1215-1221.

Jung, S. and E.D. Houde. 2003. Spatial and temporal variabilities of pelagic fish community structure and distribution in Chesapeake Bay, USA. Estuarine, Coastal, and Shelf Science 58:335-351.

Kainz, M., M.T. Arts, and A. Mazumder. 2004. Essential FAs in the planktonic food web and their ecological role for higher trophic levels. Limnology and Oceanography 49:1784-1793.

Kharlamenko, V.I., N.V. Zhukova, S.V. Khotimchenko, V.I. Svetashev, and G.M. Kamenev. 1995. FAs as markers of food sources in a shallow-water hydrothermal ecosystem

(Kraternaya Bight, Yankich Island, Kurile Islands). Marine Ecology Progress Series 120:231-241.

Kirsch, P. E., S.J. Iverson, W.D. Bowen, S.R. Kerr, and R.G. Ackman. 1998. Dietary effects on the FA signature of whole Atlantic cod (Gadus morhua). Canadian Journal of Fisheries and Aquatic Sciences 55:1378-1386.

Manooch III, C. S. 1973. Food habits of yearling and adult striped bass, *Morone saxatilis* (Walbaum), from Albemarle Sound, North Carolina. Chesapeake Science 14:73-86.

Mansueti, R. J. 1961. Age, growth, and movements of the striped bass, *Roccus saxatilis*, taken in size selective fishing gear in Maryland. Chesapeake Science 2:9-36.

McCune, B. & J.B. Grace. 2002. *Analysis of Ecological Communities*. Gleneden Beach, OR: MjM Software Design.

Meynier, L., P.C.H. Morel, B.L. Chilvers, D.D.S. Mackenzie, and P.J. Duignan. 2010. Quantitative FA signature analysis on New Zealand sea lions: model sensitivity and diet estimates. Journal of Mammalogy 91:1484-1495.

Overton, A. S., E.B. May, J. Griffin, and F.J. Margraf. 2000. A bioenergetics approach for determining the effect of increased striped bass population on its prey and health in the Chesapeake Bay. Maryland Department of Natural Resources, Annapolis.

Overton, A. S., F.J. Margraf, and E.B. May. 2009. Spatial and temporal patterns in the diet of striped bass in Chesapeake Bay. Transactions of the American Fisheries Society 138:915-926.

Paine, R.T. 1980. Food webs: linkage, interaction strength and community infrastructure. Journal of Animal Ecology 49:666-685.

Pierce, G.J. and P.R. Boyle. 1991. A review of methods for diet analysis in piscivorous marine

    mammals. Oceanography Marine Biology: An Annual Review 29:409-486.

Pikitch, E.K., C. Santora, E.A. Babcock, A. Bakun, R. Bonfil, D.O. Conover, P. Dayton, P.

    Doukakis, D. Fluharty, B. Heneman, E.D. Houde, J. Link, P.A. Livingston, M. Mangel,

    M.K. McAllister, J. Pope, and K.J. Sainsbury. 2004. Ecosystem-based fishery

    management. Science 305:346-347.

Pinnegar, J.K. and N.V.C. Polumin. 2000. Contributions of stable-isotope data to elucidating

    food webs of Mediterranean rocky littoral fishes. Oecologia 122:399-409.

R Development Core Team. 2010. R: A language and environment for statistical computing. R

    Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL

    http://www.R-project.org.

Recks, M. A., and G. T. Seaborn. 2008. Variation in FA composition among nine forage species

    from a southeastern US estuarine and nearshore coastal ecosystem. Fish Physiology and

    Biochemistry 34:275-287.

Rhodes, M.W., H. Kator, I. Kaattari, D. Gauthier, W. Vogelbein, and C.A. Ottinger. 2004.

    Isolation and characterization of mycobacteria from striped bass *Morone saxatilis* from

    the Chesapeake Bay. Diseases of Aquatic Organisms 61:41-51.

Setzler-Hamilton, E. M., and L. H. Hall Jr. 1991. Striped bass (*Morone saxitilis*). Pages 13.11-

    13.27 *in* S. L. Funderburk, S.J. Jordan, J.A. Mihursky, and D. Riley, editor. Habitat

    requirements for Chesapeake Bay living resources. Chesapeake Research Consortium,

    Solomons, MD.

Sotiropoulos, M.A., W.M. Tonn, and L.I. Wassenaar. 2004. Effects of lipid extraction on stable carbon and nitrogen isotope analyses of fish tissues: potential consequences for food webs. Ecology of Freshwater Fish 13:155-160.

Stroup, E.D. and R.J. Lynn. 1963. Atlas of salinity and temperature distributions in Chesapeake Bay 1951 – 1961 and seasonal averages 1949 – 1961. The Chesapeake Bay Institute, Graphical Summary Report, Ref. 63-1. Johns Hopkins University, Baltimore, Maryland, USA.

Thiemann, G., S.J. Iverson, and I. Sterling. 2008. Polar bear diets and arctic marine food webs: insights from FA analysis. Ecological Monographs 78:591-613.

Tocher, D.R. 2003. Metabolism and functions of lipids and FAs in teleost fish. Reviews in Fisheries Science 11:107-184.

Tucker, S., W.D. Bowen, S.J. Iverson, W. Blanchard, and G.B. Stenson. 2009. Sources of variation in diets of harp and hooded seals estimated from quantitative FA signature analysis (QFASA). Marine Ecology Progress Series 384:287-302.

USEPA (United States Environmental Protection Agency). 2007. Pressurized fluid extraction (PFE). USEPA SW-846. Method 3545A, Update IV. Washington, D.C.

Uphoff Jr., J.H. 2003. Predator-prey analysis of striped bass and Atlantic menhaden in upper Chesapeake Bay. Fisheries Management and Ecology 10:313-322.

Yodzis, P. 2001. Must top predators be culled for the sake of fisheries? Trends in Ecology and Evolution 16:78-84.

Table 1. Length, weight, and fat content of striped bass and the four prey species collected in the upper bay during the fall (September – October). Values represent the mean ± 1 standard error of the mean, N refers to the samples size of each group.

Species	N	Length (mm)	Weight (g)	Fat content (% WW)
Striped bass	78	505.91 ± 5.27	1271.29 ± 39.08	5.24 ± 0.54
Menhaden	9	127.56 ± 2.84	23.33 ± 1.98	12.46 ± 1.17
Bay anchovy	9	50.12 ± 1.87	3.81 ± 0.15	15.10 ± 2.98
Spot	11	79.55 ± 2.85	6.04 ± 0.82	6.05 ± 0.75
Blue crab	10	120.60 ± 4.59	89.10 ± 7.41	1.18 ± 0.17

Table 2. Mean FA concentrations expressed as a mass percent of all FAs present ± 1 standard error of the mean; nd = not detected. FAs shown are for striped bass and the four prey species analyzed in the QFASA model. All species were collected in the upper bay in fall (September – October). FAs listed are in the cis configuration unless otherwise noted.

Fatty acid	Striped bass	Menhaden	Bay anchovy	Spot	Blue crab
C12:0	0.15 ± 0.04	0.19 ± 0.05	0.09 ± 0.03	0.05 ± 0.01	0.09 ± 0.01
C12:1n1	0.04 ± 0.01	0.02 ± 0.02	nd	0.02 ± 0.03	0.07 ± 0.05
C13:0	0.13 ± 0.02	0.60 ± 0.20	0.50 ± 0.03	0.26 ± 0.08	0.13 ± 0.04
C14:0	3.46 ± 0.18	7.91 ± 0.75	3.60 ± 0.19	2.42 ± 0.24	1.49 ± 0.13
C14:1n5	0.11 ± 0.02	0.29 ± 0.22	0.04 ± 0.02	0.03 ± 0.001	0.04 ± 0.01
C15:0	0.81 ± 0.02	1.39 ± 0.13	1.4 ± 0.07	1.30 ± 0.05	1.41 ± 0.06
C15:1n5	nd	0.01 ± 0.01	0.33 ±0.03	nd	0.24 ± 0.03
C16 DMA	nd	0.42 ± 0.12	0.40 ± 0.02	nd	0.01 ± 0.01
C16:0	27.74 ± 0.52	29.63 ± 2.45	30.10 ± 0.49	24.36 ± 0.51	23.81 ± 0.69
C16:1n7	5.50 ± 0.23	7.79 ± 1.96	4.71 ± 0.35	4.21 ± 0.24	4.40 ± 0.24
C17:0	1.16 ± 0.07	1.00 ± 0.07	1.64 ± 0.05	2.48 ± 0.13	1.76 ± 0.12
C17:1n7	0.02 ± 0.02	0.13 ± 0.08	nd	nd	nd
C18:0	6.89 ± 0.19	6.25 ± 0.37	7.74 ± 0.14	10.83 ± 0.30	9.19 ± 0.44
C18:1n12t	nd	0.19 ± 0.05	0.19 ± 0.09	0.06 ± 0.14	0.55 ± 0.11
C18:1n9t	0.26 ± 0.02	0.31 ± 0.16	0.03 ± 0.02	0.06 ± 0.04	0.23 ± 0.04
C18:1n9	10.79 ± 0.55	4.19 ± 0.30	5.97 ± 0.19	9.30 ± 0.24	13.55 ± 1.02
C18:1n7	3.57 ± 0.09	2.90 ± 0.12	3.06 ± 0.04	3.88 ± 0.16	2.61 ± 0.09
C18:2n6	1.27 ± 0.09	1.65 ± 0.17	0.95 ± 0.05	1.75 ± 0.06	4.45 ± 0.48
C18:3n6 + C19:0	0.46 ± 0.05	0.47 ± 0.10	0.35 ± 0.01	0.63 ± 0.03	0.39 ± 0.03
C18:3n3	1.01 ± 0.08	2.08 ± 0.15	0.96 ± 0.10	1.02 ± 0.08	0.55 ± 0.02
C18:4n3	1.13 ± 0.08	2.43 ± 0.38	1.09 ± 0.11	0.49 ± 0.05	0.39 ± 0.04
C19:2n6	0.21 ± 0.04	0.14 ± 0.11	0.01 ± 0.01	0.05 ± 0.001	nd
C20:0	0.32 ± 0.04	1.43 ± 0.54	0.44 ± 0.02	0.47 ± 0.05	0.38 ± 0.04
C20:1n15	0.07 ± 0.02	0.30 ± 0.13	0.02 ± 0.01	0.04 ± 0.01	0.02 ± 0.01
C20:1n12	0.71 ± 0.05	0.54 ± 0.23	0.19 ± 0.02	1.38 ± 0.14	2.64 ± 0.48
C20:1n9	1.58 ± 0.07	0.33 ± 0.09	0.28 ± 0.02	0.90 ± 0.07	1.23 ± 0.05
C20:1n7	0.79 ± 0.07	0.69 ± 0.32	0.29 ± 0.02	1.53 ± 0.18	3.33 ± 0.43
C20:2n6	0.64 ± 0.09	0.28 ± 0.09	0.34 ± 0.03	0.99 ± 0.10	1.25 ± 0.08
C20:3n6 + C21:0	0.27 ± 0.05	0.68 ± 0.21	0.19 ± 0.02	0.19 ± 0.04	0.31 ± 0.04
C20:4n6	2.67 ± 0.12	0.80 ± 0.04	1.67 ± 0.07	4.56 ± 0.23	4.75 ± 0.35
C20:3n3	0.37 ± 0.07	0.68 ± 0.36	0.23 ± 0.03	0.44 ± 0.02	0.30 ± 0.07
C20:5n3	5.78 ± 0.21	5.36 ± 0.84	7.39 ± 0.12	6.52 ± 0.20	7.68 ± 0.54
C22:0	0.54 ± 0.15	1.79 ± 0.34	0.59 ± 0.03	0.38 ± 0.03	0.42 ± 0.07
C22:1n9	0.50 ± 0.06	0.67 ± 0.33	0.08 ± 0.01	0.28 ± 0.03	0.11 ± 0.05
C22:2n6	0.17 ± 0.06	0.57 ± 0.33	nd	0.15 ± 0.09	0.08 ± 0.19

Table 2 continued.

Fatty acid	Striped bass	Menhaden	Bay anchovy	Spot	Blue crab
C22:4n6	0.68 ± 0.06	0.63 ± 0.37	0.44 ± 0.10	1.15 ± 0.09	0.47 ± 0.15
C22:3n3 + C22:5n6	1.14 ± 0.11	1.35 ± 0.42	0.88 ± 0.07	1.34 ± 0.14	1.08 ± 0.21
C22:5n3	1.97 ± 0.11	0.33 ± 0.33	1.92 ± 0.14	2.10 ± 0.11	0.84 ± 0.14
C22:6n3 + C24:0	16.24 ± 0.62	10.14 ± 0.37	19.98 ± 0.64	13.83 ± 1.00	10.67 ± 0.44
C24:1n9	0.85 ± 0.08	2.50 ± 1.18	1.85 ± 0.16	0.56 ± 0.08	0.06 ± 0.04

Figure 1. FA proportions for the four prey taxa examined in this study. Values represent the

mean of each FA; errors bars represent one standard error of the mean.

Figure 2. NMDS of FA signatures from striped bass and the four prey taxa collected in the fall

(September – October). Symbols represent the group centroid mean of each taxon, and

ellipses are the 95% confidence intervals around the mean.

Figure 3. Proportional contribution of the four prey taxa to the diets of striped bass. QFASA

model results from a 16 FA dataset and a 30 FA dataset are based on the contribution of

each prey taxa fatty signature to striped bass FA signatures in the belly flap. Values for

stomachs are the percent biomass of each prey taxa in striped bass samples in this

experiment. Values from Hartman and Brandt (1995) and Overton et al. (2000) are

contribution of each prey taxa based upon percent biomass to the diets of age 3 striped

bass in the middle portion of the Chesapeake Bay during the fall (September –

October).

Figure 1.
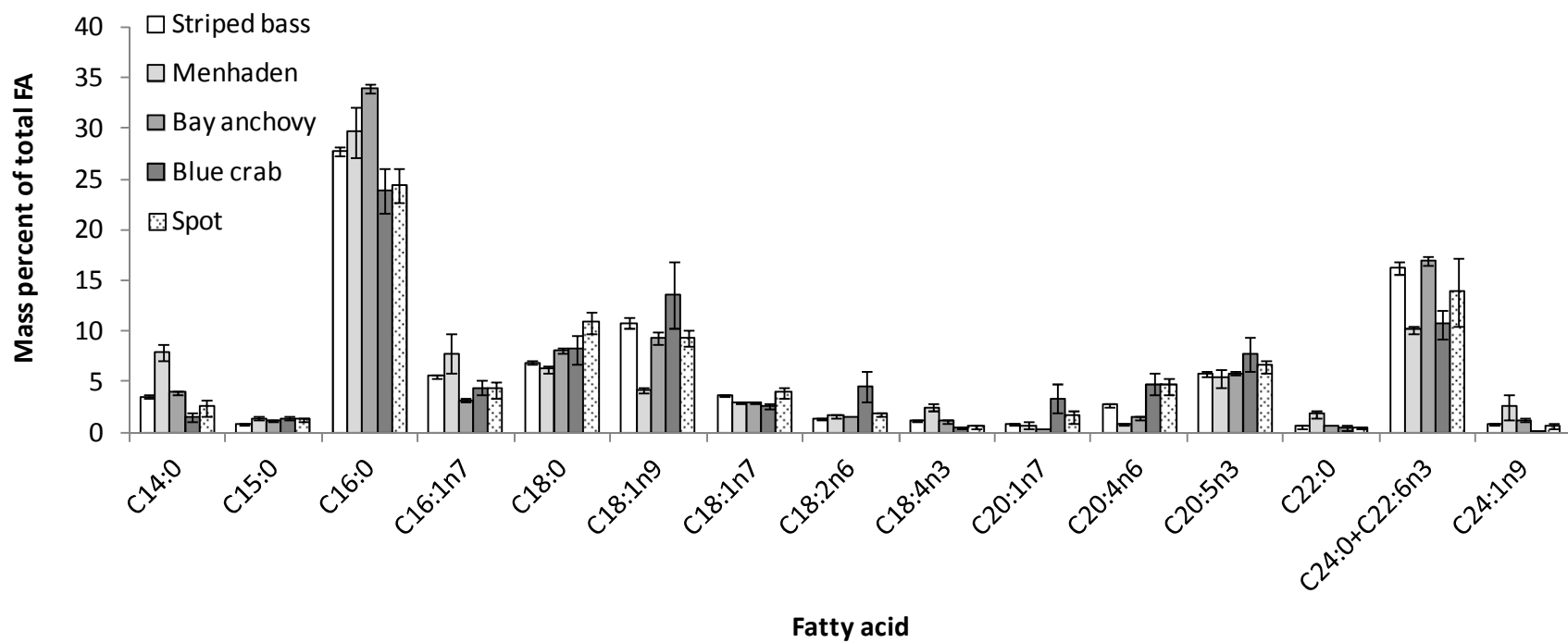
Figure 2.
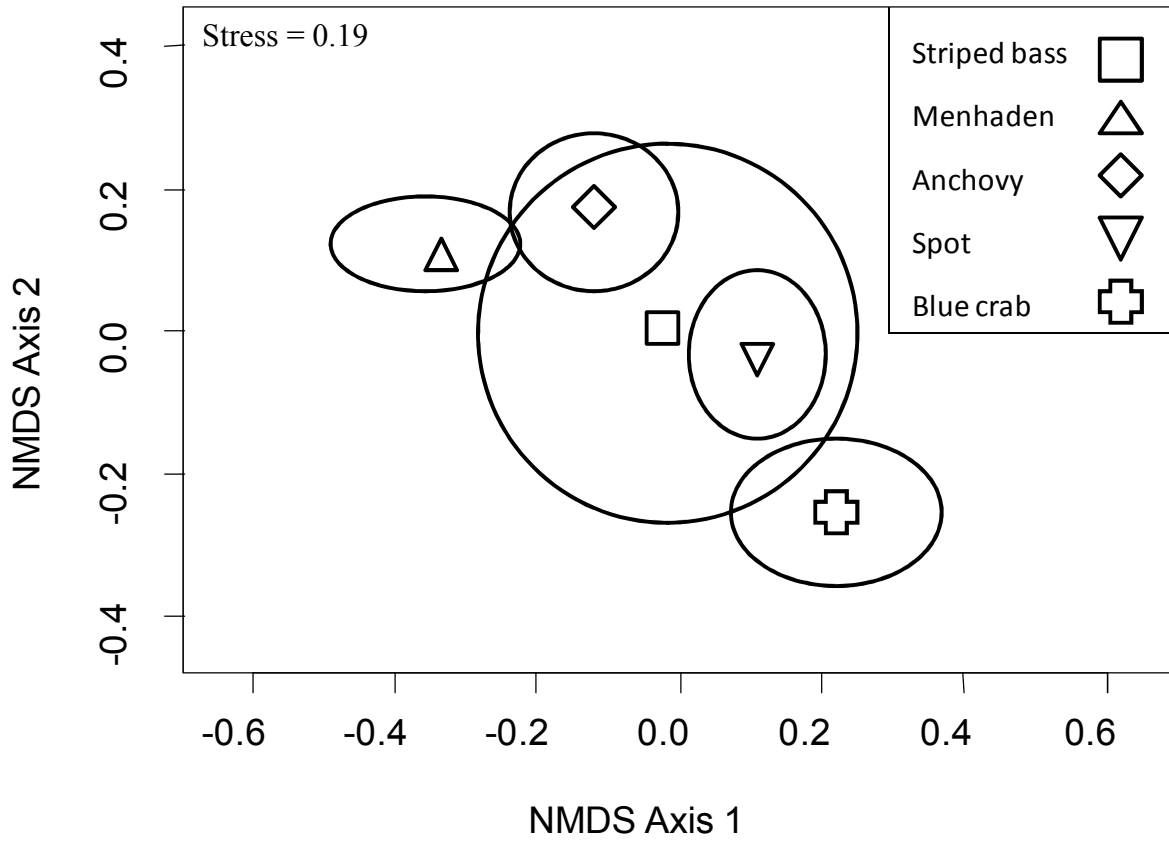
Figure 3.
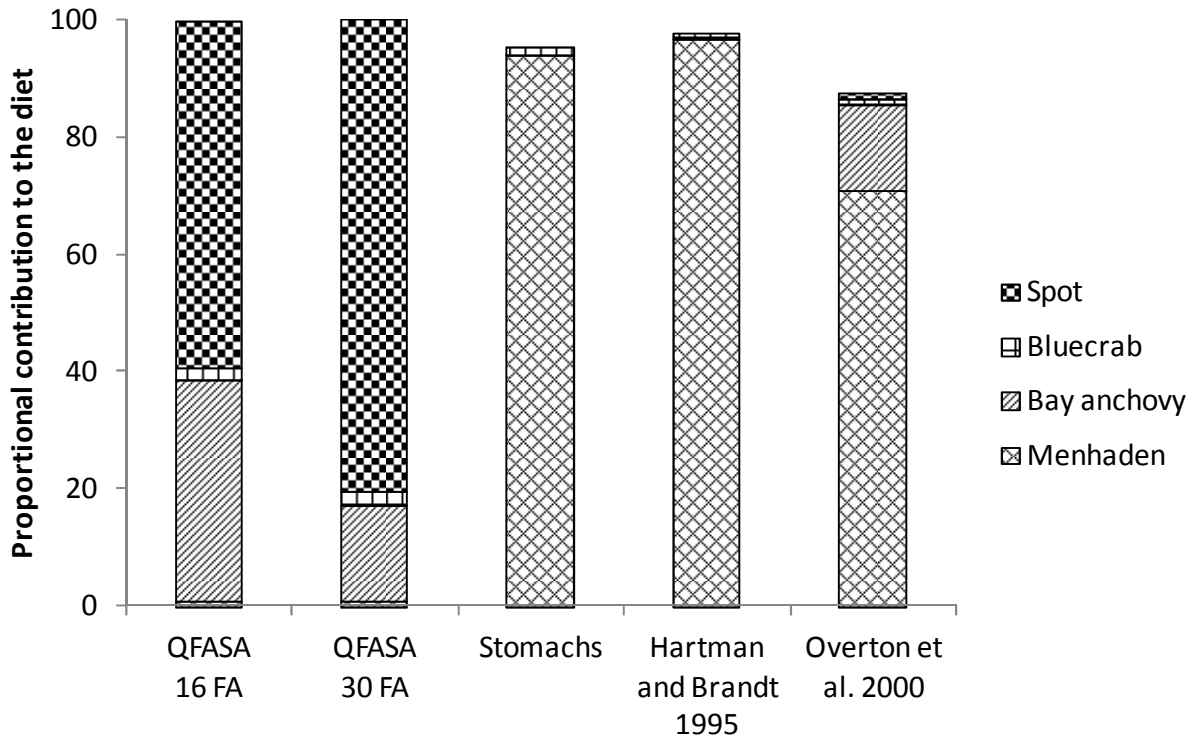
Chapter 7: Dissertation summary

The goal of this project was to determine the efficacy of using fatty acids to track striped bass diets. To achieve this goal, I had several objectives. First, I established a library of FA signatures and lipid contents for commonly consumed striped bass prey species in multiple seasons and locations in the Chesapeake Bay. My findings indicated that demersal prey species, spot (*Leiostomus xanthurus*) and blue crab (*Callinectes sapidus*), and pelagic prey species, Atlantic menhaden (*Brevoortia tyrannus*) and bay anchovy (*Anchoa mitchilli*) differed in their fatty acid signatures. Spot and blue crab were readily differentiated from each other and were grouped according to season and not species. Bay anchovy and menhaden did not exhibit clear patterns in terms of season or location in the bay.

Lipid content for striped bass prey items indicated that there were patterns with regards to season and species. Bay anchovy and menhaden consistently had the highest lipid content, spot had the next highest lipid content, and blue crab had the lowest lipid levels. Seasonal impacts on lipid content for menhaden, bay anchovy, and spot yielded an increase throughout the year, while blue crab lipid levels decreased from summer to fall.

These results highlight the importance of sampling prey species at the same time as predator species being studied. Fatty acids proportions and lipid levels will vary throughout the year and this fluctuation prevents using data on prey species collected in a different season than a predator.

The role that different tissues play in the deposition and storage of fatty acids in fish has been an area lacking in research. I tested both liver and belly flap (adipose tissue) from striped bass and documented how the fatty acid signature changed as the diet was switched from spot to menhaden. Striped bass were fed spot for a period of six weeks and menhaden for a period of six

weeks. Fish were serially sacrificed and had their fatty acid signatures recorded. There was a definitive change in the lipid levels once striped bass were changed to a diet of menhaden. The entire fatty acid signature in striped bass liver and belly flap did not change to match the prey as the literature suggests. However, certain marker fatty acids were able to distinguish the diet of striped bass.

Therefore, it becomes imperative to have very detailed information regarding the fatty acid signature of prey species in question. The fatty acids tested in this experiment (a suite of approximately 40) were sufficient to delineate the change in diet. Prey items that have very similar diets may require more information on fatty acids, e.g. branched fatty acids. A period of six weeks is not long enough for the entire striped bass signature to match that of the prey, but it is sufficient time for certain fatty acids to become present in levels that can point to the current diet being eaten.

Another factor that may influence the process of fatty acid deposition is predator size. To test the effect of this variable, I obtained striped bass of three different sizes and fed them the same diet to satiation. It became apparent when analyzing this data that feeding was reduced or absent entirely for all size classes of striped bass. Water temperature or stress from handling may have caused striped bass to fail to consume the offered prey, preventing analysis of how size affects fatty acid signature incorporation. No growth was recorded, lipid levels remained the same or decreased, and fatty acid markers that worked in the previous experiment were unable to highlight the diet switch. Size may be an important factor in influencing fatty acid incorporation but no conclusions could be made from this experiment.

One of the most promising areas of research that involve fatty acids is the ability to estimate the proportion of different prey items consumed. The model, quantitative fatty acid

190

signature analysis (QFASA), requires information on prey fatty acids and lipid levels, predator fatty acids, and information on how predators metabolize different fatty acids (calibration coefficients). Most studies have focused on homeotherms such as seabirds and marine mammals, and as such, this study represented one of the first attempts to try and apply the model to a poikilotherm.

To test the QFASA model, striped bass were fed a baseline diet of spot before being fed mixtures of spot and menhaden. As a control, another set of striped bass were fed a diet of solely menhaden. The model was able to correctly identify spot as the diet item after the baseline feeding regime. However, in all instances (menhaden alone, and spot and menhaden mixtures), the model consistently overestimated the contribution of spot to the diet. Problems may have arisen from the formulation of the diet due to lipid levels. However, menhaden is higher in lipid than spot, and any overestimation should have been skewed to the higher lipid prey: menhaden. A recent study that tried to also apply the model to poikilotherms (salmon) found that stabilization of fatty acids may take as long as 12 – 14 weeks in fish (Budge et al. 2011). This time frame begins to present issues of striped bass remaining in one area of this long. As I have stated, prey species need to be collected at the time of predator capture. Striped bass are mobile predators and sampling prey becomes problematic over such an extended time period. The second issue becomes striped bass consuming the same diet over this time period. As more species are included into the diet at different proportions, the signature is constantly changed. Results begin to become difficult to interpret and unreliable.

As a concurrent experiment with the QFASA model development, I wanted to capture wild striped bass to determine the degree of corroboration between the model and items information collected from striped bass stomachs. Wild striped bass were collected with hook

and line in the fall from the Choptank River. This time of year represents a season in which reliance on menhaden as a primary food source is extremely high. Therefore, the fatty acids from menhaden should have had ample time to become deposited within striped bass tissues. Data from the stomachs and other studies conducted on similarly aged striped bass (age-3) during the same season indicated that menhaden was the dominant prey item. However, results from the QFASA model pointed to spot and bay anchovy making large contributions to the diet, with very little contribution coming from menhaden.

Fatty acid analysis has been used as a tool to garner more information about the diet of an organism. At this time, it is my recommendation that quantitative methods for establishing feeding history with fatty acids are not ready to be employed in the fisheries field. The results from the model did not match the known diet fed to striped bass. A longer study should be conducted to determine how long striped bass need to be fed the same diet before the QFASA model can estimate diet. While qualitative estimates have been shown to work in this experiment, the same level of information can be gathered from looking at stomach contents for a fraction of the cost. Until the problems with QFASA are adequately researched for poikilotherms, other methods should be employed to estimate the diet.

Literature cited

Budge, S.M., S.N. Penney, S.P. Lall. 2011. Response of tissue lipids to diet variation in Atlantic

salmon (*Salmo salar*): implications for estimating diets with FA analysis. Journal of

Experimental Marine Biology and Ecology 409:267-274.

Chapter 2 Raw Data

Values for length (mm), weight (g), lipid content (% wet weight) and fatty acids routinely identified for the prey species analyzed in Chapter 2 of this experiment. Average total length is presented when several prey species were used in the sample. Samples that are designated with an A and B were samples that were extracted twice on the Accelerated Solvent Extractor (ASE).

Table A1. Bay anchovy collected from the upper bay in spring.

| | Sample number | | | | | | | | | | | |
	1A	1B	2A & B	3 A	3 B	4A & B	5A	5B	6A & B	7A	7B	8A & B
Total length (mm)	53.00	-	65.50	63.5	-	59.5	58.5	-	64	61	-	51.0
Weight (g)	1.01	-	1.52	1.62	-	1.31	1.33	-	1.80	1.25	-	1.10
Lipid content (%)	8.58	-	9.60	7.12	-	14.75	12.72	-	8.07	12.58	-	12.96
C12:0	0.10	1.37	nd	0.10	1.06	0.14	0.08	nd	0.08	0.05	1.63	0.06
C13:0	nd	nd	nd	nd	nd	nd	0.17	nd	0.06	0.12	nd	nd
C14:0	1.80	3.49	2.49	3.73	6.63	3.97	5.00	6.56	3.78	3.85	nd	4.04
C14:1n5	nd	3.08	nd	0.04	3.03	nd	nd	2.69	0.04	0.02	6.55	0.06
C15:0	0.50	0.82	0.55	0.76	2.11	0.88	0.93	nd	0.65	0.69	1.85	0.06
C15:1n5	0.06	nd	nd	0.03	nd	nd	nd	nd	0.01	0.01	nd	0.04
C16:0 DMA	0.58	1.15	0.53	0.54	2.61	0.50	0.40	nd	0.39	0.38	nd	0.55
C16:0	31.48	32.71	29.63	32.91	30.94	30.86	32.66	37.45	30.77	32.14	32.56	30.84
C16:1n7	2.24	2.40	2.52	4.67	nd	4.21	4.13	5.80	3.29	2.69	2.28	3.95
C17:0	0.84	1.07	1.17	0.85	1.01	1.17	1.20	nd	0.97	1.14	1.01	1.04
C17:1n7	nd	nd	0.30	0.17	nd	0.35	0.17	nd	0.10	0.08	nd	nd
C18:0	7.56	12.56	7.99	7.55	11.64	7.51	6.69	12.28	7.28	8.95	17.69	8.30
C18:1n9t	nd	nd	nd	0.14	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	6.43	6.64	6.76	7.42	8.84	6.72	5.56	7.11	5.94	5.30	6.41	6.61
C18:1n7	2.12	2.87	2.64	3.29	4.09	2.48	2.49	nd	2.31	2.01	2.06	3.06
C18:2n6	1.44	1.93	1.84	1.80	2.56	2.12	1.91	nd	1.38	1.32	2.55	1.62
C19:0 + C18:3n6	0.16	nd	nd	0.28	nd	0.26	0.24	nd	0.20	0.20	nd	0.26
C18:3n3	1.09	1.03	1.51	1.81	1.72	1.99	2.03	nd	1.12	1.07	1.11	1.14

Table A1 continued. Bay anchovy collected from the upper bay in spring.

	Sample number
	1A	1B	2A & B	3 A	3 B	4A & B	5A	5B	6A & B	7A	7B	8A & B
C18:4n3	0.96	0.83	1.61	1.31	1.07	2.16	2.50	nd	1.37	1.29	1.53	1.38
C19:2n6	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:0	0.21	nd	0.32	0.41	nd	0.35	0.33	nd	0.35	0.35	nd	0.36
C20:1n12	0.20	nd	nd	0.23	nd	0.35	0.45	nd	0.28	0.26	nd	0.50
C20:0	0.21	nd	0.32	0.41	nd	0.35	0.33	nd	0.35	0.35	nd	0.36
C20:1n12	0.20	nd	nd	0.23	nd	0.35	0.45	nd	0.28	0.26	nd	0.50
C20:1n9	0.25	nd	0.36	0.35	nd	0.52	0.37	nd	0.26	0.20	nd	0.37
C20:1n7	0.12	nd	nd	0.30	nd	0.48	0.55	nd	0.33	0.24	nd	0.67
C20:2n6	0.46	nd	0.50	0.60	nd	0.62	0.59	nd	0.46	0.43	nd	0.59
C20:3n6 + C21:0	nd	nd	nd	0.14	nd	nd	nd	nd	0.08	0.07	nd	0.11
C20:4n6	0.99	0.91	1.22	1.52	1.11	1.14	0.67	nd	0.93	0.75	0.56	0.96
C20:3n3	0.25	nd	nd	0.27	nd	nd	0.23	nd	0.20	0.18	nd	0.23
C20:5n3	6.03	5.33	7.26	6.42	5.74	6.93	7.45	7.99	7.04	6.16	4.71	5.84
C22:0	0.45	nd	0.40	0.59	nd	0.70	0.60	nd	0.66	0.70	nd	0.52
C22:1n9	nd	nd	nd	0.07	nd	nd	nd	nd	0.08	0.07	nd	0.07
C22:2n6	nd	nd	nd	nd	nd	nd	nd	nd	0.02	nd	nd	nd
C23:0	0.23	nd	nd	0.19	nd	0.23	0.15	nd	0.17	0.17	nd	0.30
C22:4n6	nd	nd	nd	nd	nd	0.13	0.12	nd	0.10	0.10	nd	0.12
C22:5n6	0.59	nd	0.72	0.92	0.82	0.49	0.38	nd	0.50	0.44	nd	0.51
C22:5n3	0.48	nd	0.46	0.61	0.93	0.54	0.62	nd	0.47	0.52	0.70	0.60
C24:0	0.91	1.55	0.89	0.98	0.95	1.08	0.68	nd	0.95	0.71	0.78	1.20
C22:6n3	28.50	21.62	26.76	16.68	13.12	19.42	18.52	20.11	25.32	24.66	15.26	22.17
C24:1n9	1.57	0.61	1.57	1.12	nd	1.70	1.49	nd	2.05	1.80	0.74	1.86

Table A2. Bay anchovy collected from the lower bay in spring.

	Sample number							
	1	2	4	5	6	7	8	9
Total length (mm)	60	58	64	61	59	58	57	51
Weight (g)	2.31	2.33	2.23	2.02	2.23	2.36	2.20	1.28
Lipid content (%)	11.68	8.04	13.88	12.69	3.43	5.58	5.49	7.67
C12:0	0.13	0.18	0.14	0.15	nd	1.25	0.18	1.07
C12:1n1	nd	nd	nd	nd	nd	0.07	nd	nd
C13:0	0.27	0.16	0.14	0.24	nd	0.17	0.20	0.31
C14:0	8.36	8.35	10.14	9.60	7.55	7.72	8.83	8.41
C14:1n5	0.10	0.07	0.08	0.14	nd	0.08	0.12	0.10
C15:0	1.09	1.25	1.21	1.31	1.12	1.29	1.12	1.19
C16:0 DMA	0.29	0.34	0.30	0.39	nd	0.35	0.29	0.36
C16:0	27.96	30.34	31.50	27.78	34.37	29.72	30.20	30.60
C16:1n7	8.58	7.96	8.69	11.39	8.03	8.32	9.62	7.83
C17:0	0.83	0.82	0.80	0.82	0.64	1.10	0.86	1.19
C18:0	4.08	3.57	4.12	3.58	6.82	5.25	4.25	4.98
C18:1n12t	0.46	0.21	0.16	nd	nd	0.21	nd	nd
C18:1n9	6.37	5.82	5.65	6.40	7.06	6.93	6.36	6.12
C18:1n7	3.21	3.14	3.06	3.61	4.47	3.68	3.27	3.64
C18:2n6	1.20	1.59	1.32	1.29	1.71	1.50	1.38	1.38
C19:0 + C18:3n6	0.25	0.20	0.23	0.27	nd	0.29	0.26	0.27
C18:3n3	0.98	1.02	0.95	0.82	0.81	0.85	1.01	0.75
C18:4n3	1.39	1.46	1.47	1.42	1.34	1.02	1.29	1.22
C20:0	0.32	0.29	0.35	0.39	nd	0.35	0.28	0.30
C20:1n12	0.31	0.14	0.26	0.34	nd	0.42	0.35	0.53
C20:1n9	0.49	0.61	0.52	0.88	nd	0.68	0.74	0.74
C20:1n7	0.64	0.42	0.55	0.55	nd	0.67	0.68	0.84
C20:2n6	0.32	0.42	0.38	0.33	nd	0.49	0.44	0.45
C20:3n6 + C21:0	nd	nd	0.10	nd	nd	0.10	0.12	0.16

Table A2 continued. Bay anchovy collected from the lower bay in spring.

	Sample number							
	1	2	4	5	6	7	8	
C20:4n6	1.00	0.64	0.66	1.20	1.11	1.13	0.85	0.70
C20:3n3	nd	nd	0.12	nd	nd	0.09	0.10	0.13
C20:5n3	10.06	8.71	8.08	8.93	7.91	7.72	8.49	7.32
C22:0	0.49	0.35	0.45	nd	nd	0.41	0.22	0.47
C22:1n11	0.28	0.53	0.43	nd	nd	0.56	0.52	0.67
C22:1n9	nd	nd	0.13	nd	nd	nd	0.13	0.13
C22:2n6	nd	nd	0.01	nd	nd	nd	nd	nd
C22:4n6	nd	nd	0.11	0.19	nd	0.13	0.11	0.15
C22:5n6	0.52	0.32	nd	0.52	nd	nd	0.36	0.33
C22:5n3	0.53	0.50	0.43	0.59	nd	0.49	0.47	0.49
C24:0	0.47	0.45	0.30	0.36	nd	0.59	0.53	0.57
C22:6n3	17.99	18.92	15.75	15.55	15.82	15.15	15.26	15.47
C24:1n9	1.05	1.22	1.40	0.96	1.24	1.20	1.08	1.15

Table A3. Bay anchovy collected from the upper bay in summer.

	Sample number								
	1	2	3	4	5	6	7	8	9
Average total length (mm)	81.30	63.03	69.20	85.55	81.50	77.40	68.78	70.18	72.50
Weight (g)	3.72	3.84	2.71	3.64	3.19	3.07	4.77	4.47	5.18
Lipid content (%)	11.00	16.30	7.80	4.51	17.29	4.47	14.44	15.29	43.30
C12:0	0.13	0.15	0.28	0.29	0.45	0.10	0.18	0.51	0.43
C13:0	nd	0.33	0.47	0.44	0.93	0.77	0.35	0.29	nd
C14:0	5.89	3.68	2.70	2.53	2.32	1.59	3.35	3.99	3.75
C14:1n5	0.82	nd	nd	nd	nd	nd	nd	nd	nd
C15:0	0.69	1.01	1.01	1.07	1.03	0.74	0.95	1.39	1.24
C15:1n5	nd	nd	0.11	0.08	nd	0.11	nd	nd	nd
C16:0 DMA	1.23	0.49	0.61	0.63	0.69	0.92	0.57	0.60	0.06
C16:0	32.05	30.27	28.79	31.17	31.62	30.24	28.62	31.66	32.37
C16:1n7	14.04	2.82	1.66	2.02	1.91	2.12	2.52	2.62	2.79
C17:0	0.90	1.59	1.98	2.26	2.03	1.91	2.13	2.01	2.35
C17:1n7	0.38	0.53	nd	nd	nd	nd	nd	nd	nd
C18:0	8.54	8.57	11.22	11.26	12.26	10.54	10.79	10.13	10.87
C18:1n12t	nd	0.73	nd	nd	nd	nd	nd	nd	nd
C18:1n9	17.77	9.27	8.23	8.94	10.33	9.37	8.35	9.46	10.95
C18:1n7	5.17	2.59	2.67	2.72	2.57	2.74	2.09	2.52	2.76
C18:2n6	0.53	1.66	1.54	1.22	1.11	1.36	1.80	1.52	1.54
C19:0 + C18:3n6	nd	0.37	0.37	0.40	0.56	0.35	0.41	0.36	0.53
C18:3n3	nd	1.09	0.73	0.64	0.57	0.47	0.89	0.97	0.74
C18:4n3	nd	1.48	0.76	0.43	0.67	0.32	1.09	1.03	0.96
C20:0	0.23	nd	0.48	0.54	0.62	0.42	0.51	0.54	0.67
C20:1n15	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:1n12	1.02	0.87	0.89	0.54	0.65	0.52	1.20	0.93	0.95
C20:1n9	3.73	0.42	0.46	0.45	0.35	0.38	0.58	0.54	nd

Table A3 continued. Bay anchovy collected from the upper bay in summer.

	Sample number								
	1	2	3	4	5	6	7	8	9
C20:1n7	0.33	0.24	0.25	0.43	0.42	0.49	0.58	0.26	0.40
C20:2n6	0.25	0.69	1.02	0.69	0.77	0.73	0.98	0.93	1.27
C20:3n6 + C21:0	nd	nd	0.18	0.18	nd	nd	nd	nd	nd
C20:4n6	nd	0.94	1.59	2.14	1.83	1.77	1.27	1.29	1.00
C20:3n3	nd	0.22	0.37	0.38	0.38	0.26	0.46	0.31	nd
C20:5n3	0.36	7.17	6.94	5.95	4.27	5.52	6.65	6.77	6.96
C22:0	nd	0.51	0.50	0.58	0.46	0.47	0.48	0.40	nd
C22:1n11	2.80	nd	nd	nd	nd	nd	nd	nd	nd
C22:1n9	0.75	nd	nd	nd	nd	nd	nd	nd	nd
C22:4n6	nd	0.24	0.33	0.32	nd	0.30	0.41	0.34	nd
C22:5n6	nd	0.51	0.51	0.56	0.77	0.45	0.27	0.47	nd
C22:5n3	0.28	1.99	1.31	0.71	0.81	0.58	1.20	0.93	0.38
C24:0	0.11	0.93	0.89	1.22	1.68	1.29	1.16	0.74	0.97
C22:6n3	1.48	17.79	20.16	17.82	16.87	21.47	19.13	15.69	16.06
C24:1n9	0.53	0.87	0.98	1.40	1.08	1.71	1.03	0.79	nd

Table A4. Bay anchovy collected from the lower bay in summer.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Average total length (mm)	66.13	88.70	60.98	61.95	63.53	63.35	65.28	63.85	70.70	60.62
Weight (g)	3.58	5.43	4.68	4.85	4.83	4.15	5.59	5.03	4.69	4.25
Lipid content (%)	5.44	4.35	8.76	6.57	6.55	7.05	4.21	5.60	5.43	4.95
C12:0	0.08	0.07	0.12	0.06	nd	0.19	0.13	0.15	0.00	0.03
C12:1n1	0.03	0.05	nd	0.03	nd	0.04	nd	0.02	0.02	0.03
C13:0	0.57	0.51	0.42	0.44	0.61	0.43	0.35	0.36	0.38	0.53
C14:0	1.42	0.99	2.53	2.19	1.39	2.77	1.95	3.14	2.42	1.88
C14:1n5	0.02	nd	0.02	0.04	nd	0.02	0.04	0.03	nd	0.03
C15:0	1.08	0.79	1.31	1.08	0.89	1.36	1.18	0.96	1.15	1.03
C15:1n5	0.36	0.38	0.34	0.34	0.46	0.23	0.29	0.22	0.26	0.38
C16:0 DMA	0.55	0.46	0.43	0.50	0.50	0.48	0.46	0.38	0.38	0.41
C16:0	29.82	28.68	29.10	29.88	27.87	32.97	31.68	30.45	29.24	31.33
C16:1n7	1.88	1.64	1.91	1.76	1.69	2.32	1.92	2.82	1.70	1.66
C17:0	1.80	1.53	1.94	1.77	1.89	1.93	1.79	1.48	1.72	1.65
C18:0	8.92	7.86	9.55	9.28	9.58	9.73	9.02	8.05	8.82	9.52
C18:1n12t	nd	nd	nd	nd	nd	nd	0.22	nd	nd	nd
C18:1n9	7.48	7.30	6.32	6.35	6.96	7.35	6.76	6.84	5.47	6.66
C18:1n7	2.83	2.82	2.53	2.49	2.63	2.69	2.50	2.62	2.31	2.46
C18:1n9t	nd	nd	nd	nd	0.17	nd	nd	nd	nd	nd
C18:2n6	0.87	0.89	0.93	0.90	0.92	1.05	0.86	1.08	0.98	0.86
C19:0 + C18:3n6	0.30	0.25	0.28	0.27	0.34	0.35	0.28	0.29	0.28	0.28
C18:3n3	0.49	0.45	0.63	0.66	0.52	0.61	0.57	0.73	0.73	0.60
C18:4n3	0.23	0.23	0.47	0.55	0.35	0.39	0.34	0.60	0.74	0.39
C20:0	0.35	0.27	0.37	0.32	0.36	0.43	0.35	0.37	0.40	0.35
C20:1n15	0.05	0.01	0.03	nd	nd	0.02	0.01	0.02	0.01	nd
C20:1n12	0.23	0.12	0.31	0.25	0.22	0.37	0.23	0.35	0.21	0.17

Table A4 continued. Bay anchovy collected from the lower bay in summer.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
C20:1n9	0.45	0.37	0.41	0.36	0.34	0.44	0.35	0.40	0.28	0.28
C20:1n7	0.31	0.24	0.40	0.29	0.37	0.40	0.29	0.47	0.26	0.25
C20:2n6	0.46	0.39	0.45	0.43	0.42	0.45	0.41	0.44	0.39	0.39
C20:3n6 + C21:0	0.19	0.14	0.17	0.02	0.15	0.17	0.17	0.14	0.16	0.15
C20:4n6	nd	2.36	1.66	1.53	1.36	1.83	1.60	1.61	1.35	1.51
C20:3n3	0.20	0.27	0.22	0.19	0.18	0.23	0.19	0.20	0.19	0.18
C20:5n3	6.12	7.39	5.40	5.15	4.44	5.49	5.17	6.12	5.37	4.83
C22:0	0.58	0.44	0.53	0.50	0.50	0.47	0.42	0.45	0.60	0.53
C22:1n9	0.14	0.10	0.18	0.15	nd	0.17	0.12	0.13	0.23	0.13
C22:2n6	0.05	nd	nd	0.02	0.03	0.02	0.01	0.02	0.04	0.01
C23:0	0.07	0.05	0.13	0.08	0.10	0.06	0.11	0.09	0.15	0.09
C22:4n6	0.20	0.23	0.51	0.25	0.75	0.32	0.77	0.27	0.59	0.40
C22:3n3	0.74	0.91	1.13	0.91	1.61	0.67	1.15	0.96	1.41	1.07
C22:5n3	0.76	0.68	0.80	0.57	0.68	0.71	0.57	0.78	0.60	0.48
C24:0	1.42	1.30	1.46	1.40	2.14	0.96	1.33	1.33	1.46	1.32
C22:6n3	26.77	27.70	24.77	26.78	26.43	20.26	24.32	23.43	27.11	26.15
C24:1n9	2.18	2.11	2.23	2.19	3.11	1.56	2.06	2.18	2.59	2.38

Table A5. Bay anchovy collected from the upper bay in fall.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Average total length (mm)	46.83	46.63	75.60	64.95	48.78	54.88	57.93	56.48	41.31	47.88
Weight (g)	3.54	4.09	3.87	3.97	3.41	3.17	3.57	3.96	4.73	4.56
Lipid content (%)	25.03	16.70	8.36	28.33	24.79	8.30	12.64	5.34	6.43	1.65
C12:0	0.45	0.20	0.50	0.51	0.17	0.17	0.45	0.27	0.33	0.44
C12:1n1	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C13:0	0.30	0.21	0.28	0.21	0.19	0.19	0.27	0.23	0.38	0.29
C14:0	3.75	5.36	3.23	3.85	4.38	4.40	3.45	2.55	4.30	3.88
C14:1n5	nd	nd	nd	nd	nd	0.04	0.05	nd	nd	nd
C15:0	1.27	1.09	1.43	1.23	1.06	1.07	1.11	1.05	1.02	1.30
C15:1n5	nd	nd	nd	0.04	0.05	0.06	0.03	0.07	nd	nd
C16:0 DMA	0.49	0.51	0.64	0.53	0.51	0.52	0.48	0.74	0.52	0.53
C16:0	34.17	34.02	32.78	35.00	32.88	33.06	37.53	33.53	33.93	33.33
C16:1n7	2.56	3.02	4.41	3.12	2.94	2.96	3.61	2.46	2.78	3.22
C17:0	1.70	1.46	2.46	1.88	1.44	1.45	1.55	1.50	1.40	1.65
C17:1n7	0.53	0.13	0.81	0.64	0.49	nd	nd	nd	nd	nd
C18:0	7.88	8.30	6.85	7.50	8.12	8.16	7.11	9.03	10.06	8.02
C18:1n12t	0.49	0.41	0.74	0.24	nd	nd	nd	nd	nd	nd
C18:1n9	8.81	6.91	11.89	10.85	7.18	7.22	11.73	9.82	7.94	10.56
C18:1n7	3.01	2.53	3.34	3.15	2.67	2.69	3.20	2.92	3.11	3.20
C18:2n6	1.43	1.60	1.50	1.47	1.62	1.63	1.17	1.24	1.33	1.52
C19:0 + C18:3n6	0.32	0.27	0.36	0.34	0.29	0.29	0.31	0.35	0.37	0.36
C18:3n3	1.34	1.93	0.90	0.93	1.64	1.65	0.88	0.78	1.70	1.14
C18:4n3	0.97	1.59	0.45	0.77	1.36	1.37	0.71	0.59	2.15	1.05
C19:2n6	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:0	0.53	0.51	0.46	0.55	0.47	0.47	0.57	0.53	0.45	0.57
C20:1n12	0.29	0.34	0.54	0.33	0.32	0.32	0.34	0.35	0.32	0.36

Table A5 continued. Bay anchovy collected from the upper bay in fall.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
C20:1n9	0.51	nd	0.59	0.57	0.39	0.39	0.55	0.52	0.43	0.43
C20:1n7	0.33	nd	0.33	0.36	0.33	0.33	0.36	0.45	0.30	0.34
C20:2n6	0.54	0.45	0.64	0.58	0.46	0.46	0.53	0.63	0.34	0.46
C20:3n6 + C21:0	0.17	0.13	nd	0.18	0.14	0.14	0.16	0.16	nd	0.14
C20:4n6	1.46	0.83	2.01	1.88	0.97	0.98	1.57	2.22	0.81	1.53
C20:3n3	0.35	0.24	0.44	0.39	0.23	0.23	0.28	0.34	0.33	0.32
C20:5n3	6.23	5.80	5.62	6.13	6.17	6.21	5.49	5.84	5.00	5.73
C22:0	0.59	0.74	0.32	0.45	0.69	0.69	0.57	0.54	0.39	0.56
C22:1n9	nd	0.10	nd	0.06	0.07	0.09	nd	0.09	nd	nd
C23:0	nd	nd	nd	nd	0.13	nd	nd	0.10	nd	nd
C22:4n6	0.13	0.13	0.35	0.13	0.15	0.15	0.17	0.25	nd	0.18
C22:5n6	0.83	0.51	0.68	0.74	0.48	0.48	0.75	0.83	0.23	0.70
C22:5n3	0.43	0.40	0.95	0.50	0.46	0.46	0.52	0.47	0.56	0.60
C24:0	0.80	0.84	0.74	0.78	1.03	1.04	0.71	1.11	1.03	0.93
C22:6n3	16.05	17.58	13.12	13.30	18.96	19.07	13.07	17.19	16.77	15.48
C24:1n9	1.26	1.50	0.66	0.80	1.58	1.58	0.71	1.24	1.72	1.19

Table A6. Bay anchovy collected from the lower bay in fall.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Total length (mm)	55	67	60	46	62	52	52	62	51	55
Weight (g)	3.51	4.52	4.23	4.65	5.02	3.46	3.92	3.38	4.07	3.09
Lipid content (%)	16.20	4.55	44.97	5.70	6.28	7.26	6.98	16.65	11.49	13.37
C12:0	0.15	0.26	nd	nd	0.14	0.09	0.13	nd	0.03	0.07
C12:1n1	nd	nd	nd	nd	nd	0.06	0.06	nd	0.06	0.09
C13:0	0.48	0.37	0.45	0.46	0.37	0.51	0.45	0.68	0.55	0.66
C14:0	3.57	3.43	3.87	4.64	3.33	3.16	3.99	2.95	2.76	4.20
C14:1n5	0.04	nd	nd	nd	0.03	0.03	0.04	0.13	0.02	0.15
C15:0	1.19	1.22	1.59	1.72	1.07	1.30	1.53	1.51	1.17	1.66
C15:1n5	0.31	0.30	0.46	0.27	0.20	0.29	0.20	0.52	0.35	0.35
C16:0 DMA	0.36	0.39	0.47	0.55	0.35	0.38	0.37	0.38	0.32	0.38
C16:0	29.22	28.24	28.29	29.85	32.49	31.47	32.26	28.99	28.79	30.58
C16:1n7	4.06	3.47	5.43	6.81	3.54	4.39	5.71	3.87	4.12	5.56
C17:0	1.70	1.93	1.76	1.65	1.57	1.49	1.51	1.76	1.44	1.56
C18:0	7.96	7.77	7.92	7.32	7.98	7.51	6.95	8.51	7.83	7.43
C18:1n12t	nd	0.30	nd	0.14	0.25	nd	nd	nd	nd	0.19
C18:1n9	5.88	6.77	6.90	5.23	6.05	5.50	5.07	6.32	5.93	5.82
C18:1n7	3.05	2.92	3.25	3.10	2.93	3.06	2.95	3.12	2.87	3.28
C18:1n9t	nd	nd	nd	0.14	nd	nd	nd	0.18	nd	nd
C18:2n6	0.78	0.81	0.96	1.21	0.80	0.93	1.08	0.90	0.88	1.16
C19:0 + C18:3n6	0.34	0.37	0.42	0.43	0.34	0.33	0.33	0.35	0.31	0.31
C18:3n3	0.81	0.60	0.88	1.43	0.69	0.95	1.26	0.67	0.85	1.41
C18:4n3	1.13	0.85	0.93	1.78	0.75	1.08	1.41	0.65	1.02	1.29
C19:2n6	nd	nd	nd	nd	nd	nd	nd	nd	0.05	nd
C20:0	0.48	0.49	0.54	0.46	0.43	0.37	0.40	0.46	0.33	0.47
C20:1n15	nd	nd	nd	0.07	0.02	nd	0.01	nd	nd	0.10

Table A6 continued. Bay anchovy collected from the lower bay in fall.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
C20:1n12	0.25	0.18	0.25	0.27	0.17	0.13	0.16	0.17	0.11	0.18
C20:1n9	0.30	0.29	0.36	0.27	0.26	0.22	0.20	0.36	0.21	0.31
C20:1n7	0.34	0.22	0.29	0.39	0.27	0.26	0.27	0.21	0.25	0.41
C20:2n6	0.35	0.36	0.41	0.21	0.35	0.31	0.30	0.52	0.31	0.28
C20:3n6 + C21:0	0.17	0.16	0.27	0.26	0.15	0.15	0.16	0.29	0.14	0.14
C20:4n6	1.62	1.59	2.08	1.29	1.77	1.85	1.40	1.78	1.67	1.57
C20:3n3	0.16	0.16	0.40	0.36	0.16	0.18	0.18	0.24	0.19	0.22
C20:5n3	7.31	6.64	7.13	7.51	7.36	8.04	7.12	7.60	7.23	7.71
C22:0	0.64	0.64	0.63	0.66	0.54	0.55	0.54	0.76	0.42	0.52
C22:1n11	0.07	0.25	0.43	0.56	0.02	0.08	0.06	0.63	0.27	0.12
C22:1n9	0.10	nd	0.13	0.12	0.08	0.07	0.08	nd	0.09	0.10
C22:2n6	0.08	0.09	0.08	nd	0.03	0.02	0.03	nd	nd	nd
C23:0	0.24	0.21	0.22	0.16	0.14	0.10	0.09	0.15	0.15	0.14
C22:4n6	0.57	0.59	1.23	0.20	0.26	0.25	0.27	0.18	0.54	0.32
C22:3n3	0.92	1.33	1.19	0.72	0.67	0.73	0.79	0.62	1.00	0.84
C22:5n3	0.85	1.08	0.63	0.61	0.59	0.59	0.55	0.52	0.73	0.57
C24:0	1.50	1.71	1.25	0.93	0.90	1.07	0.97	1.30	1.58	1.12
C22:6n3	20.73	21.20	17.63	16.89	21.25	20.82	19.51	20.92	22.98	17.30
C24:1n9	2.29	2.83	1.29	1.32	1.71	1.67	1.62	1.80	2.47	1.43

Table A7. Blue crab collected from the upper bay in summer.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	51	43	57	42	45	67	37	41	40	52	44	38
Weight (g)	6.81	6.80	14.81	4.28	4.68	19.01	2.89	4.33	3.53	10.05	5.99	3.37
Lipid content (%)	3.13	0.88	0.54	0.96	0.84	1.46	1.94	2.24	1.39	0.61	0.61	3.27
C12:0	0.14	0.05	0.12	0.11	0.02	0.15	0.08	0.22	0.03	0.04	nd	nd
C12:1n1	nd	nd	nd	0.09	0.01	nd	0.01	nd	nd	nd	0.08	nd
C13:0	0.12	0.34	0.21	0.53	0.22	0.19	0.17	0.29	0.68	0.60	0.94	1.04
C14:0	2.70	2.02	2.38	1.70	1.32	3.04	2.36	1.89	2.08	2.15	1.18	2.22
C14:1n5	0.04	0.01	0.02	nd	0.02	0.15	0.02	0.05	0.05	0.16	0.08	0.23
C15:0	1.29	1.15	1.12	1.62	1.57	1.73	1.75	1.30	1.36	1.59	1.54	2.28
C16:0 DMA	0.79	1.26	0.83	0.84	0.97	0.50	0.70	0.64	0.80	0.63	1.28	1.27
C16:0	33.42	31.41	33.09	25.42	24.81	24.00	33.96	26.64	28.12	25.85	21.92	27.23
C16:1n7	5.30	2.96	3.51	2.90	1.93	8.07	4.16	3.33	2.79	3.38	1.53	3.90
C17:0	1.32	2.51	1.87	2.05	2.32	1.17	2.03	1.91	1.82	1.96	2.81	2.79
C17:1n7	nd	nd	nd	0.15	nd	nd	nd	nd	nd	nd	nd	nd
C18:0	7.15	12.93	9.40	10.04	12.52	5.51	10.03	9.08	10.08	10.07	13.23	11.53
C18:1n12t	0.81	1.06	0.68	0.90	0.87	0.99	1.04	1.37	1.00	1.13	0.60	1.19
C18:1n9t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.29	nd
C18:1n7t	0.12	nd	0.08	0.14	0.08	0.14	0.16	0.15	0.11	nd	nd	nd
C18:1n9	11.41	8.70	11.60	9.19	8.89	10.38	10.97	11.20	9.31	7.09	7.99	8.82
C18:1n7	2.92	4.09	4.85	2.13	2.68	2.21	2.34	3.02	2.60	2.27	2.63	2.48
C18:2n6t	nd	nd	nd	0.09	nd	nd	nd	nd	nd	nd	nd	nd
C18:2n6	2.58	2.33	3.00	2.54	2.87	3.12	3.06	2.49	3.20	3.11	2.59	3.50
C18:3n6	0.15	0.11	0.13	nd	0.09	0.17	0.06	0.14	0.11	0.23	0.16	nd
C19:0	nd	nd	nd	nd	nd	nd	0.03	nd	nd	nd	nd	nd
C18:3n3	0.87	1.15	1.47	0.76	1.26	1.00	0.89	0.97	1.36	1.87	0.91	1.33

Table A7 continued. Blue crab collected from the upper bay in summer.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C18:4n3	1.17	0.42	0.62	0.78	0.25	1.33	0.44	0.72	0.82	0.79	0.27	nd
C19:2n6	0.03	0.01	nd	0.11	nd	0.09	0.06	nd	0.05	nd	nd	nd
C20:0	1.29	1.39	1.46	1.99	1.50	1.61	1.64	1.63	1.59	1.90	1.64	2.31
C20:1n15	nd	0.10	0.10	0.18	0.11	0.18	0.13	0.07	0.08	0.14	nd	0.42
C20:1n12	2.82	1.15	1.63	4.63	1.43	4.22	4.21	2.34	2.26	3.47	1.68	3.42
C20:1n9	1.17	1.06	1.49	1.43	1.06	1.20	1.34	1.12	1.04	1.04	1.10	1.94
C20:1n7	2.69	1.14	0.92	4.63	1.31	4.35	4.33	1.84	2.14	3.57	1.42	2.93
C20:2n6	1.02	1.36	1.63	1.70	1.94	1.30	1.45	1.82	1.62	1.79	2.16	2.48
C20:3n6	0.10	0.13	0.14	0.18	0.17	0.18	0.09	0.15	0.14	0.27	0.30	0.27
C21:0	0.15	0.17	0.22	0.28	0.13	0.21	0.22	0.20	0.19	0.24	0.27	1.32
C20:4n6	1.59	2.82	2.25	2.29	3.17	1.65	1.35	2.04	2.11	1.97	3.07	1.98
C20:3n3	0.26	0.36	0.50	0.34	0.40	0.37	0.25	0.30	0.42	0.40	0.41	0.55
C20:5n3	6.52	8.69	6.63	5.97	10.02	4.58	4.35	6.74	7.50	5.84	8.75	4.47
C22:0	1.21	1.15	1.20	1.28	1.31	1.19	0.93	2.06	1.74	1.74	1.30	1.86
C22:1n11	nd	nd	nd	0.23	nd	nd	nd	nd	nd	0.11	nd	0.28
C22:1n9	0.14	0.15	0.16	nd	0.24	0.28	0.22	0.20	0.17	0.26	0.37	nd
C22:2n6	0.02	0.04	0.03	0.75	nd	nd	0.04	nd	0.03	0.31	nd	nd
C22:4n6	0.12	0.36	0.18	nd	0.56	0.13	0.10	0.60	0.30	0.35	0.85	nd
C22:3n3 + C22:5n6	0.37	0.20	0.27	1.22	0.57	0.37	0.16	0.55	0.45	0.54	0.50	0.56
C22:5n3	0.45	0.92	0.45	1.78	3.40	3.58	0.43	1.86	1.31	3.06	5.65	0.82
C22:6n3 + C24:0	7.84	6.19	5.61	9.05	9.99	10.25	4.29	10.92	10.40	9.86	9.80	4.60
C24:1n9	0.07	0.11	0.16	nd	nd	0.36	0.14	0.14	0.10	0.22	0.54	nd

Table A8. Blue crab collected from the upper bay in fall.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Total length (mm)	114	119	154	116	99	111	115	126	130	122
Weight (g)	101.9	78.5	140.8	59.9	66.4	74.6	79.6	97.8	105.5	92.4
Lipid content (%)	1.04	0.64	0.79	1.63	1.80	1.77	0.65	2.15	0.82	0.50
C12:0	0.07	0.05	0.10	0.09	0.07	0.12	0.06	0.17	0.08	0.06
C12:1n1	0.08	nd	0.10	0.12	0.08	0.07	0.09	0.16	nd	nd
C13:0	0.30	nd	0.28	0.20	0.12	0.13	nd	0.23	nd	nd
C14:0	1.37	1.29	1.42	1.51	1.39	2.19	1.17	2.25	1.32	1.02
C14:1n5	0.05	0.03	0.03	0.08	0.04	0.06	0.02	0.04	0.07	0.01
C15:0	1.55	1.50	1.41	1.28	1.29	1.88	1.28	1.38	1.31	1.27
C15:1n5	0.32	0.41	0.23	0.22	0.20	0.24	0.30	0.22	0.29	nd
C16:0 DMA	nd	0.07	nd	nd	nd	nd	nd	nd	nd	nd
C16:0	23.08	23.58	23.50	24.00	20.04	26.99	25.42	26.80	23.24	21.48
C16:1n7	4.55	3.88	4.59	5.70	3.90	5.16	3.66	4.13	5.07	3.37
C17:0	1.33	2.09	2.23	1.11	1.39	1.70	2.07	1.80	1.64	2.22
C18:0	7.11	10.19	7.83	6.39	6.29	7.87	9.11	8.12	8.71	10.26
C18:1n12t	0.51	0.70	0.58	0.44	0.57	0.55	0.44	0.76	0.49	0.42
C18:1n9t	0.22	nd	0.18	0.47	0.19	0.35	0.17	0.08	0.26	0.37
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	0.08	nd	nd
C18:1n9	18.48	14.03	10.98	19.15	11.07	11.42	11.51	13.00	15.62	10.22
C18:1n7	2.90	2.56	2.29	2.96	2.23	2.33	2.52	2.94	2.77	2.55
C18:2n6	6.00	3.20	2.85	7.51	5.09	3.72	3.44	4.42	5.20	3.06
C18:3n6	0.38	0.39	0.35	0.31	0.62	0.35	0.35	0.39	0.44	0.36
C18:3n3	0.47	0.47	0.54	0.52	0.64	0.52	0.53	0.56	0.58	0.66
C18:4n3	0.38	0.21	0.50	0.32	0.60	0.46	0.29	0.54	0.30	0.36
C19:2n6	0.02	0.01	nd	nd	nd	nd	nd	nd	nd	nd
C20:0	0.29	0.37	0.32	0.22	0.68	0.34	0.39	0.34	0.38	0.43

Table A8 continued. Blue crab collected from the upper bay in fall.

					Sample number					
	1	2	3	4	5	6	7	8	9	10
C20:1n15	nd	0.03	nd	0.06	nd	0.08	nd	nd	0.04	nd
C20:1n12	2.87	0.00	4.66	2.33	3.50	3.87	3.10	3.25	nd	2.83
C20:1n9	1.15	1.52	1.22	0.97	1.33	1.23	1.15	1.09	1.19	1.41
C20:1n7	3.02	nd	4.99	2.43	3.92	4.26	3.41	3.64	4.23	3.44
C20:2n6	1.08	1.64	1.09	1.01	1.57	1.08	1.14	1.06	1.32	1.54
C20:3n6	0.29	0.32	0.28	0.24	0.67	0.22	0.27	0.25	0.28	0.26
C21:0	nd	nd	nd	nd	nd	0.01	nd	nd	0.02	nd
C20:4n6	4.19	6.43	4.93	3.39	4.41	3.83	5.45	3.43	4.97	6.44
C20:3n3	0.17	0.25	0.24	0.18	0.97	0.23	0.29	0.27	0.20	0.24
C20:5n3	6.32	9.25	8.00	6.09	6.25	6.44	9.43	6.63	7.38	11.05
C22:0	0.29	0.36	0.32	0.32	1.02	0.37	0.37	0.29	0.38	0.47
C22:1n9	0.05	0.08	0.06	0.05	0.60	0.05	0.05	0.03	0.06	0.06
C22:2n6	0.02	0.02	nd	0.01	0.61	0.02	0.02	0.01	0.03	0.03
C22:4n6	0.33	0.45	0.28	0.30	1.85	0.30	0.36	0.28	0.33	0.27
C22:3n3 + C22:5n6	0.83	1.36	0.84	0.62	2.90	0.97	0.80	0.68	0.99	0.84
C22:5n3	0.64	1.22	0.65	0.83	1.68	nd	0.69	0.96	0.72	0.99
C22:6n3 + C24:0	9.30	12.03	12.13	8.28	12.21	10.60	10.63	9.38	10.08	12.04
C24:1n9	nd	nd	nd	0.28	nd	nd	nd	0.34	nd	nd

Table A9. Menhaden collected from the upper bay in spring.

	Sample number										
	1A	1B	2A & B	3A	3B	4A	5A & B	6A	7A	7B	8A & B
Total length (mm)	71	-	76	78	-	73	86	81	80	-	79
Weight (g)	3.33		4.16	4.65		3.43	5.76	5.20	5.20		4.99
Lipid content (%)	4.47	-	4.80	-	-	-	4.26	5.72	-	5.35	5.11
C12:0	nd	nd	0.04	0.04	0.67	0.18	0.05	0.06	nd	0.05	0.07
C12:1n1	nd	nd	nd	nd	nd	nd	nd	0.03	nd	nd	nd
C13:0	nd	nd	nd	nd	nd	0.09	nd	0.21	nd	nd	nd
C14:0	1.89	3.27	2.50	1.64	3.07	3.46	1.46	3.96	4.13	2.19	2.28
C14:1n5	nd	3.35	nd	nd	2.04	nd	nd	nd	0.80	nd	nd
C15:0	1.51	2.63	1.25	1.21	1.70	11.78	1.24	4.75	4.10	1.48	2.17
C15:1n5	nd	nd	0.08	0.08	nd	nd	0.08	0.07	nd	0.15	0.08
C16:0 DMA	0.67	nd	0.48	0.56	1.57	0.27	0.61	0.49	1.13	0.80	0.56
C16:0	36.53	41.52	36.58	35.80	30.61	33.65	34.32	32.09	29.48	36.06	34.34
C16:1n7	2.66	3.77	3.50	3.42	3.34	3.87	3.23	5.26	5.13	3.99	4.02
C17:0	1.20	nd	1.48	0.99	1.93	1.00	1.25	1.72	2.26	1.52	1.50
C17:1n7	nd	nd	0.47	0.44	nd	nd	nd	nd	nd	nd	nd
C18:0	10.27	16.34	9.99	8.75	12.97	9.37	9.46	7.40	12.66	9.75	9.50
C18:1n12t	nd	nd	nd	nd	nd	0.29	nd	nd	nd	nd	nd
C18:1n9t	nd	nd	nd	nd	nd	0.12	nd	nd	nd	nd	nd
C18:1n9	6.42	7.61	5.82	4.76	6.22	6.69	4.96	4.17	5.35	5.80	5.08
C18:1n7	2.48	1.95	2.73	2.33	2.91	1.93	2.55	2.46	2.26	2.53	2.44
C18:2n6	1.29	nd	1.74	1.21	1.47	1.16	1.28	1.42	nd	1.59	1.51
C19:0 + C18:3n6	0.31	nd	0.30	0.24	0.33	0.22	0.30	0.46	nd	0.37	0.37
C18:3n3	0.98	nd	1.72	0.93	1.01	1.07	1.08	1.14	0.72	0.97	nd
C18:4n3	0.63	nd	1.05	0.59	0.68	0.86	0.49	1.18	1.03	0.47	0.84
C20:0	0.21	nd	0.22	0.18	0.59	0.21	0.19	0.23	nd	0.24	nd
C20:1n12	nd	nd	0.37	nd	nd	0.24	0.07	0.16	nd	nd	nd

Table A9 continued. Menhaden collected from the upper bay in spring.

	1A	1B	2A & B	3A	3B	4A	5A & B	6A	7A	7B	8A & B
					Sample number						
C20:1n9	nd	nd	0.39	0.11	nd	0.25	nd	0.18	1.40	0.15	0.15
C20:1n7	nd	nd	0.18	0.13	nd	0.09	0.16	0.30	nd	0.15	0.12
C20:2n6	0.30	nd	0.30	0.33	0.58	0.20	0.43	0.35	nd	0.28	0.49
C20:3n6 + C21:0	nd	nd	0.11	0.14	nd	0.09	0.14	0.17	nd	0.19	0.23
C20:4n6	1.68	nd	1.22	1.72	1.92	0.95	2.14	1.94	3.66	1.76	2.14
C20:3n3	0.22	nd	0.18	0.27	nd	0.19	0.28	0.24	nd	0.24	0.30
C20:5n3	4.68	3.94	4.85	4.45	3.45	3.28	5.29	6.17	5.47	5.09	5.17
C22:0	nd	nd	0.44	0.42	nd	0.29	0.43	0.49	nd	0.40	0.49
C22:1n11	nd	nd	nd	nd	nd	0.07	nd	nd	nd	nd	0.17
C22:1n9	nd	nd	nd	0.04	nd	0.05	nd	0.08	nd	nd	nd
C22:2n6	nd	nd	nd	nd	nd	0.01	nd	nd	nd	nd	nd
C23:0	0.57	nd	0.17	0.16	nd	0.14	0.15	0.40	nd	0.13	0.18
C22:4n6	nd	nd	nd	nd	nd	0.06	0.14	0.16	nd	0.12	nd
C22:3n3	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C22:5n6	0.69	nd	0.47	0.64	0.41	0.40	0.82	0.67	nd	0.56	0.57
C22:5n3	0.61	nd	0.64	0.52	0.58	0.38	0.59	0.61	nd	0.55	0.63
C24:0	1.28	nd	0.46	0.68	0.81	0.40	0.78	0.46	2.75	0.09	0.72
C22:6n3	20.24	15.61	18.89	24.09	17.79	15.55	24.37	18.08	16.68	20.71	22.49
C24:1n9	1.62	nd	1.39	1.72	1.17	1.12	1.66	1.24	0.97	1.65	1.37

Table A10. Menhaden collected from the lower bay in spring.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Average total length (mm)	57.03	58.88	48.28	47.98	47.58	45.25	43.82	45.86	55.83	54.46
Weight (g)	3.93	3.63	3.21	3.37	3.06	3.77	3.23	3.56	14.88	13.99
Lipid content (%)	9.28	9.70		5.23	10.39					
C12:0	0.12	0.15	0.15	0.15	0.16	0.35	0.09	nd	0.30	0.20
C13:0	nd	nd	nd	0.17	0.26	nd	0.33	nd	nd	nd
C14:0	8.58	12.81	10.85	12.94	11.21	10.38	7.26	8.14	11.80	11.19
C14:1n5	nd	nd	nd	0.07	nd	nd	nd	nd	nd	nd
C15:0	0.97	1.06	0.85	0.97	0.95	1.54	0.80	1.16	1.07	0.97
C15:1n5	nd	nd	nd	0.04	nd	nd	nd	nd	nd	nd
C16:0 DMA	0.44	0.28	0.35	0.30	0.32	nd	0.50	0.64	0.43	0.37
C16:0	38.95	38.27	37.29	38.11	34.29	37.00	34.34	35.32	36.26	35.00
C16:1n7	13.05	16.80	15.18	16.06	14.50	14.34	9.61	12.11	15.55	13.91
C17:0	1.27	0.85	0.69	0.62	0.65	1.64	0.84	0.49	0.40	0.41
C17:1n7	nd	nd	nd	0.23	0.27	nd	nd	nd	nd	nd
C18:0	6.42	4.36	4.94	4.03	4.24	6.76	6.82	6.76	4.57	4.88
C18:1n9	3.56	3.10	3.31	3.09	2.86	4.53	3.79	4.21	2.87	3.15
C18:1n7	3.58	2.96	3.29	3.14	3.37	4.26	3.49	3.53	2.87	3.15
C18:2n6	1.33	1.30	1.25	1.13	1.20	1.05	0.94	1.03	1.25	1.31
C19:0 + C18:3n6	0.38	0.37	0.38	0.30	0.37	nd	0.41	nd	0.45	0.46
C18:3n3	0.90	1.67	1.15	1.60	1.34	0.79	0.61	1.03	1.51	1.22
C18:4n3	2.52	2.66	2.49	2.53	3.10	1.92	2.06	2.44	2.90	3.20
C20:0	nd	0.16	0.18	0.16	0.18	nd	0.24	nd	0.21	nd
C20:1n12	nd	nd	nd	0.13	nd	nd	nd	nd	nd	nd
C20:1n9	nd	nd	nd	0.10	nd	nd	nd	nd	nd	nd
C20:1n7	nd	nd	0.12	0.13	0.08	nd	nd	nd	nd	nd

Table A10 continued. Menhaden collected from the lower bay in spring.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
C20:2n6	nd	nd	nd	0.08	nd	nd	nd	nd	nd	nd
C20:3n6 + C21:0	nd	0.25	0.22	0.10	0.26	nd	nd	nd	nd	nd
C20:4n6	1.11	0.65	0.98	0.60	0.86	0.93	1.70	1.67	0.81	1.08
C20:5n3	6.61	5.90	6.80	6.02	8.37	5.95	8.71	8.13	7.41	7.88
C22:0	nd	nd	0.24	0.23	0.31	nd	0.38	nd	0.33	nd
C22:5n6	nd	0.17	0.18	0.14	0.30	nd	0.38	nd	0.23	nd
C22:5n3	0.53	0.77	0.67	0.47	0.75	0.82	0.91	0.53	0.43	0.96
C24:0	0.67	0.36	0.66	0.47	0.56	0.54	0.86	0.79	0.50	0.81
C22:6n3	8.43	4.89	7.50	5.65	8.97	7.19	14.24	11.87	7.84	9.85
C24:1n9	0.60	0.22	0.30	0.23	0.26	nd	0.70	0.15	nd	nd

Table A11. Menhaden collected from the upper bay in summer (Choptank River).

	Sample number							
	1	2	3	4	5	6	7	8
Total length (mm)	79	95	81	93	85	101	85	75
Weight (g)	5.00	8.44	4.87	7.66	5.89	9.83	7.42	5.42
Lipid content (%)	0.53	0.05	0.24	0.60	0.18	0.24	0.47	0.79
C12:0	nd	0.07	nd	nd	0.12	nd	nd	nd
C12:1n1	nd	0.03	nd	nd	0.07	nd	nd	nd
C13:0	nd	nd	nd	nd	0.55	0.57	1.23	nd
C14:0	2.54	1.85	3.54	2.78	3.21	4.42	3.31	1.63
C14:1n5	nd	0.02	0.06	0.14	0.09	0.19	0.14	0.07
C15:0	1.02	0.86	1.40	1.19	1.26	1.59	1.23	0.78
C15:1n5	0.18	0.58	0.59	0.47	0.48	0.33	0.37	0.37
C16:0 DMA	0.27	0.31	0.25	0.23	0.22	0.32	0.25	0.44
C16:0	41.45	35.24	39.06	38.16	37.47	38.93	37.72	29.77
C16:1n7	2.41	3.18	4.51	3.33	3.88	5.39	3.51	3.51
C17:0	1.18	1.21	1.69	1.37	1.29	1.70	1.37	1.29
C18:0	10.17	10.40	9.99	10.32	9.52	9.12	9.67	9.01
C18:1n12t	nd	nd	0.26	nd	nd	nd	0.20	nd
C18:1n9	9.26	8.22	7.60	8.67	7.00	7.97	8.18	7.01
C18:1n7	1.82	2.75	2.13	2.70	2.55	2.43	2.21	2.49
C18:2n6t	nd	nd	0.08	nd	nd	nd	nd	nd
C18:1n9t	0.49	0.41	0.27	0.60	0.46	0.61	0.31	0.81
C18:2n6	nd	1.83	1.74	1.88	1.81	1.77	2.10	1.60
C19:0 + C18:3n6	0.30	0.31	0.29	0.28	0.49	0.39	0.38	0.93
C18:3n3	0.91	0.71	1.43	0.81	1.04	1.14	1.22	1.32
C18:4n3	0.16	0.96	1.03	0.91	1.05	1.19	1.23	0.77
C19:2n6	nd	nd	0.14	0.10	0.21	nd	nd	nd
C20:0	0.35	0.31	nd	0.45	0.34	0.47	0.38	0.44
C20:1n15	nd	0.09	0.25	nd	0.11	nd	0.11	0.33

Table A11 continued. Menhaden collected from the upper bay in summer (Choptank River).

	Sample number							
	1	2	3	4	5	6	7	8
C20:1n12	nd	0.13	0.16	nd	nd	0.11	0.36	0.13
C20:1n9	0.11	0.22	0.23	0.40	0.28	0.26	0.35	0.28
C20:1n7	nd	0.22	0.14	0.16	0.12	0.23	0.28	0.33
C20:2n6	0.17	0.18	0.40	0.20	0.38	nd	0.20	0.21
C20:3n6 + C21:0	0.10	0.13	0.05	0.04	0.25	0.36	0.23	0.27
C20:4n6	1.66	2.06	1.63	1.29	1.52	1.33	1.22	1.74
C20:3n3	0.29	0.18	0.15	0.11	0.26	0.15	0.19	0.27
C20:5n3	3.48	3.68	3.48	2.57	3.11	2.80	2.91	2.16
C22:0	0.72	0.74	0.75	0.56	0.58	0.87	1.12	0.67
C22:1n11	nd	nd	2.79	4.81	3.69	2.18	0.35	nd
C22:1n9	0.12	0.05	0.08	0.13	0.08	0.15	0.11	0.18
C22:2n6	0.08	0.01	0.06	0.03	nd	0.06	0.08	nd
C23:0	0.37	0.23	0.07	0.04	0.14	0.35	0.44	0.62
C22:4n6	0.28	0.29	0.74	0.17	0.58	0.22	0.23	1.03
C22:3n3	0.45	0.76	1.21	0.63	1.01	0.55	0.31	2.03
C22:5n3	0.47	0.39	0.26	0.25	0.34	nd	nd	1.78
C24:0	1.99	1.03	1.04	1.12	1.26	1.24	1.58	3.06
C22:6n3	16.08	18.61	9.84	12.08	12.26	10.23	12.85	19.31
C24:1n9	1.14	1.76	0.60	1.02	0.92	0.36	2.05	3.36

Table A12. Menhaden collected from the upper bay in summer (Corsica River).

	Sample number								
	1	2	3	4	5	6	7	8	9
Total length (mm)	98	97	98	99	96	101	96	97	93
Weight (g)	7.68	8.21	7.97	8.51	7.92	8.62	8.08	8.77	8.65
Lipid content (%)	11.20	10.69	7.41	3.37	20.69	16.93	7.75	10.40	19.08
C12:0	0.25	0.34	0.44	0.16	0.28	0.30	0.33	0.28	nd
C13:0	0.35	0.36	0.41	0.42	0.37	0.64	0.72	0.33	nd
C14:0	7.84	8.48	6.62	5.41	7.99	9.15	8.07	7.73	4.39
C14:1n5	0.04	0.05	nd	0.03	0.15	nd	nd	0.16	nd
C15:0	1.40	2.57	1.42	1.57	1.34	3.87	1.95	1.48	3.04
C16:0 DMA	0.60	0.61	0.76	0.83	0.03	0.41	0.83	0.55	nd
C16:0	34.53	34.28	34.00	34.62	35.05	36.92	35.57	35.87	32.50
C16:1n7	4.44	6.62	3.45	4.54	8.07	9.24	3.04	5.05	5.11
C17:0	1.66	1.66	1.78	1.39	1.19	1.77	2.52	1.69	2.68
C17:1n7	nd	nd	nd	nd	nd	0.31	nd	nd	nd
C18:0	6.36	6.72	7.81	8.17	5.67	6.56	9.18	7.11	8.21
C18:1n12t	0.18	nd	0.23	0.26	nd	nd	0.43	0.21	nd
C18:1n9t	nd	0.23	0.23	0.23	nd	0.20	0.38	nd	nd
C18:1n9	7.32	6.81	8.31	7.55	6.41	4.96	7.34	7.35	6.73
C18:1n7	2.04	2.14	2.08	2.38	1.83	2.20	1.96	2.37	2.87
C18:2n6	1.91	2.26	2.32	2.12	2.68	1.78	1.79	2.72	4.16
C18:3n6 + C19:0	0.19	0.23	0.13	0.20	0.37	0.37	nd	0.27	2.00
C18:3n3	0.73	0.87	0.92	0.81	0.92	2.03	1.09	0.82	2.12
C18:4n3	3.51	2.97	2.90	1.74	3.59	2.89	2.24	2.91	2.56
C19:2n6	0.03	nd	nd	nd	nd	nd	nd	0.07	2.02
C20:0	0.79	0.88	0.85	0.87	0.94	0.97	1.19	1.18	2.58
C20:1n15	0.06	nd	nd	nd	nd	nd	nd	0.16	nd

Table A12 continued. Menhaden collected from the lower bay in summer (Corsica River).

	Sample number								
	1	2	3	4	5	6	7	8	9
C20:1n12	0.14	0.17	0.23	0.22	nd	0.31	nd	0.22	nd
C20:1n9	0.35	0.42	0.45	0.37	0.36	0.58	0.73	0.45	2.08
C20:1n7	0.14	0.15	0.22	0.18	0.17	0.36	0.35	0.26	nd
C20:2n6	0.17	0.21	0.27	0.17	0.29	0.34	0.35	0.27	nd
C20:3n6 + C21:0	0.13	0.13	0.29	0.13	0.10	0.63	1.06	0.16	nd
C20:4n6	0.83	1.04	1.09	1.23	0.77	0.77	1.22	0.75	0.90
C20:3n3	0.10	0.12	0.33	0.10	0.10	0.39	0.45	0.11	1.58
C20:5n3	4.30	4.93	4.01	3.75	5.49	3.62	2.46	4.37	3.25
C22:0	2.17	1.62	2.18	2.13	1.72	1.13	1.96	1.97	2.52
C22:1n9	0.32	0.10	0.25	0.26	0.08	0.15	0.51	0.11	1.68
C22:2n6	0.17	0.02	0.07	0.12	nd	0.26	0.47	0.07	0.83
C23:0	0.28	0.05	nd	0.19	nd	0.64	0.46	nd	0.47
C22:4n6	0.47	0.14	0.32	0.47	0.32	0.06	0.19	0.17	nd
C22:3n3 + C22:5n6	0.76	0.54	0.54	0.86	0.73	nd	0.80	0.44	1.31
C22:5n3	1.44	0.60	0.85	1.60	1.23	0.81	1.32	0.96	0.95
C24:0 + C22:6n3	13.43	11.52	13.81	14.31	11.76	5.38	9.05	11.19	3.45
C24:1n9	0.55	0.14	0.41	0.65	nd	nd	nd	0.19	nd

Table A13. Adult menhaden collected from the upper bay in summer.

	Sample number								
	1	2	3	4	5	6	7	8	9
Total length (mm)	222	190	238	315	208	244	219	221	228
Weight (g)	110.2	68.2	140.2	292.3	85.0	144.7	105.1	103.9	222.1
Lipid content (%)	18.89	6.82	13.54	20.32	11.31	11.93		11.40	7.97
C12:0	0.07	0.12	0.12	0.10	0.11	0.09	0.08	0.14	0.10
C12:1n1	0.34	0.34	0.22	0.50	0.58	0.24	0.40	0.33	0.18
C13:0	0.35	0.20	0.21	0.25	0.15	0.15	nd	0.19	0.31
C14:0	3.69	11.16	10.46	16.11	4.78	17.02	5.76	10.73	6.20
C14:1n5	0.02	0.07	0.04	0.07	0.04	0.07	0.02	0.06	0.04
C15:0	0.90	1.03	1.04	0.89	0.80	0.78	1.01	0.79	0.88
C15:1n5	nd	nd	nd	nd	nd	nd	nd	0.06	0.14
C16:0 DMA	nd	nd	nd	0.02	nd	nd	nd	0.03	0.05
C16:0	32.89	23.52	30.37	23.42	31.28	24.92	31.17	29.74	31.65
C16:1n7	3.37	12.01	12.53	15.10	3.10	14.05	7.99	11.54	5.70
C17:0	1.53	1.08	1.25	1.03	1.45	1.03	1.59	0.89	1.44
C18:0	7.76	4.82	5.07	4.66	7.52	4.68	7.51	4.82	8.19
C18:1n12t	0.37	0.46	0.26	0.28	nd	0.24	0.37	0.27	nd
C18:1n9t	nd	nd	nd	nd	0.19	nd	nd	nd	nd
C18:1n9	4.12	3.07	2.68	2.63	6.19	2.66	3.13	5.29	4.92
C18:1n7	2.44	3.42	3.06	3.53	3.09	2.81	2.95	3.56	2.96
C18:2n6	1.45	1.39	1.17	1.25	1.84	1.27	1.42	2.27	1.73
C18:3n6	0.31	0.50	0.49	0.63	0.31	0.58	0.45	0.42	0.39
C18:3n3	1.53	1.17	1.23	1.22	3.30	1.27	0.97	1.15	1.81
C18:4n3	1.20	1.76	1.75	2.24	2.52	2.48	0.95	1.57	1.70
C20:0	0.23	0.27	0.20	0.23	0.37	0.22	0.21	0.20	0.28
C20:1n15	0.02	0.04	nd	0.04	0.03	nd	nd	0.01	0.01

Table A13 continued. Adult menhaden collected from the upper bay in summer.

	Sample number								
	1	2	3	4	5	6	7	8	9
C20:1n12	0.12	0.23	0.13	0.12	0.16	0.08	0.14	0.08	0.10
C20:1n9	0.30	0.42	0.21	0.21	0.95	0.18	0.24	0.52	0.35
C20:1n7	0.23	0.48	0.21	0.24	0.37	0.19	0.27	0.22	0.26
C20:2n6	0.29	0.33	0.14	0.20	0.40	0.17	0.22	0.21	0.25
C20:3n6	0.17	0.44	0.24	0.31	0.18	0.26	0.34	0.25	0.18
C20:4n6	1.43	1.30	2.03	1.35	0.60	1.16	2.78	1.02	1.41
C20:3n3	0.26	0.55	0.21	0.25	0.50	0.25	0.15	0.23	0.26
C20:5n3	5.58	7.73	9.15	8.78	7.34	8.66	7.43	7.32	5.96
C22:0	0.64	0.66	0.40	0.39	1.30	0.43	0.43	0.34	0.30
C22:1n11	nd	nd	nd	nd	nd	nd	nd	0.06	nd
C22:1n9	0.10	0.63	0.08	0.08	0.63	0.08	0.06	0.13	0.09
C22:2n6	0.02	0.24	nd	0.02	0.58	0.02	0.02	nd	nd
C22:4n6	0.25	1.20	0.27	0.23	0.33	0.17	0.46	nd	0.24
C22:3n3 + C22:5n6	0.86	2.72	1.05	1.36	0.91	1.15	1.71	1.00	1.18
C22:5n3	1.10	2.69	1.24	1.22	1.57	1.13	1.42	1.16	1.11
C24:0 + C22:6n3	25.29	12.30	12.14	10.66	15.29	11.09	17.99	12.92	18.59
C24:1n9	0.76	1.64	0.34	0.40	1.23	0.44	0.36	0.49	1.05

Table A14. Menhaden collected from lower bay in summer.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
Total length (mm)	139	147	150	161	128	151	139	137	142	146
Weight (g)	36.11	39.95	44.04	51.83	25.59	44.56	33.33	37.28	42.20	40.79
Lipid content (%)	2.68	-	3.84	1.37	-	-	4.32	4.77	1.55	5.92
C12:0	nd	nd	0.10	0.14	0.09	0.09	0.07	0.11	nd	nd
C12:1n1	nd	0.09	0.04	nd	0.03	0.05	0.05	0.02	nd	nd
C13:0	0.22	0.19	0.20	0.23	0.19	0.21	0.32	0.23	0.17	0.17
C14:0	10.44	10.51	12.41	12.01	16.81	13.37	12.69	10.69	12.69	11.01
C14:1n5	0.17	0.21	0.14	0.16	0.12	0.11	0.11	0.12	0.21	0.17
C15:0	1.19	1.36	1.17	1.35	0.91	1.35	1.28	1.14	1.15	1.39
C15:1n5	0.16	0.14	0.11	0.25	0.10	0.10	0.15	nd	0.21	0.23
C16:0 DMA	0.28	0.28	0.22	0.28	0.14	0.18	0.20	0.23	0.22	0.38
C16:0	34.05	30.92	36.02	32.27	33.14	33.32	37.41	33.16	33.62	33.07
C16:1n7	16.20	15.67	12.89	14.31	12.52	13.61	12.44	13.60	15.38	17.70
C17:0	1.47	1.72	1.28	1.49	0.55	0.93	1.01	1.15	1.22	1.72
C17:1n7	0.45	0.50	nd	0.46	nd	nd	nd	nd	nd	0.40
C18:0	5.55	4.91	4.88	5.24	3.99	4.10	4.60	4.30	4.66	5.01
C18:1n12t	nd	nd	nd	nd	nd	nd	nd	nd	0.14	nd
C18:1n9	4.52	4.48	3.43	4.75	2.95	3.22	2.83	3.09	3.52	5.06
C18:1n7	4.25	4.00	3.10	3.67	2.21	2.63	2.67	2.97	3.10	4.29
C18:1n9t	nd	0.26	nd	nd	nd	nd	nd	nd	nd	nd
C18:2n6	1.91	1.91	1.45	1.88	1.18	1.31	1.57	1.59	1.52	2.01
C19:0 + C18:3n6	nd	0.64	0.36	0.49	0.38	0.40	0.36	0.42	0.47	0.70
C18:3n3	2.36	2.86	0.94	1.84	0.52	0.77	0.76	1.24	0.96	3.25
C18:4n3	1.30	1.36	2.03	1.96	2.21	2.60	2.18	2.94	2.92	1.53
C19:2n6	nd	0.09	nd	0.09	nd	0.11	nd	nd	0.13	0.18
C20:0	0.29	0.26	0.28	0.29	0.20	0.19	0.22	0.30	0.32	0.29
C20:1n15	0.05	nd	0.05	0.06	0.07	0.03	0.06	0.05	0.11	0.12

Table A14 continued. Menhaden collected from lower bay in summer.

	Sample number									
	1	2	3	4	5	6	7	8	9	10
C20:1n12	0.21	nd	0.14	0.25	0.12	0.19	0.13	0.16	0.14	0.25
C20:1n9	0.43	0.49	0.27	0.50	0.19	0.24	0.16	0.46	0.45	0.52
C20:1n7	0.31	0.30	0.19	0.21	0.16	0.23	0.14	0.16	0.38	0.28
C20:2n6	0.19	0.26	0.15	0.16	0.12	0.10	0.15	0.19	0.13	0.32
C20:3n6 + C21:0	0.57	0.40	0.28	0.27	0.17	0.17	0.19	0.19	0.32	0.50
C20:4n6	1.33	1.09	0.09	0.91	1.08	0.82	0.83	0.90	0.76	0.99
C20:3n3	0.26	0.33	0.09	0.21	0.07	0.09	0.10	0.11	0.16	0.32
C20:5n3	5.07	4.84	7.97	6.40	8.98	7.65	6.94	7.84	7.19	4.94
C22:0	0.54	0.31	0.51	0.46	0.56	0.48	0.41	0.47	0.37	nd
C22:1n11	0.03	nd	nd	nd	nd	nd	nd	nd	0.19	nd
C22:1n9	0.09	0.14	nd	0.14	0.06	0.07	0.04	0.11	0.07	0.21
C22:2n6	nd	nd	0.01	0.05	nd	0.10	0.03	0.10	0.10	0.23
C23:0	0.13	nd	nd	0.09	nd	0.14	0.11	0.17	0.13	nd
C22:4n6	0.28	1.61	0.19	0.12	0.29	0.42	0.19	0.44	0.12	0.16
C22:3n3 + C22:5n6	0.64	0.84	0.41	0.37	0.55	0.57	0.23	1.11	0.21	nd
C22:5n3	1.71	1.81	0.84	1.30	1.75	1.23	0.78	1.26	0.71	0.42
C24:0	nd	1.11	0.48	nd	nd	1.07	0.50	0.50	0.32	0.36
C22:6n3	3.03	3.77	6.77	5.13	6.89	6.92	7.58	7.59	5.21	1.81
C24:1n9	0.29	0.35	0.53	0.24	0.69	0.83	0.52	0.91	0.32	nd

Table A15. Menhaden collected from the upper bay in fall.

	Sample number					
	1	2	3	4	5	6
Total length (mm)	126	119	124	128	129	142
Weight (g)	22.92	23.11	18.86	25.25	22.46	33.75
Lipid content (%)	8.66	12.93	14.92	10.55	8.93	16.67
C12:0	nd	0.17	0.32	nd	0.25	0.19
C12:1n1	nd	0.10	nd	nd	nd	nd
C13:0	1.30	0.49	0.73	4.44	0.30	0.17
C14:0	5.41	8.94	7.50	3.21	7.86	9.83
C14:1n5	1.18	nd	0.08	1.91	0.09	0.09
C15:0	1.88	1.29	1.23	nd	1.36	1.18
C15:1n5	nd	nd	nd	nd	nd	0.04
C16:0 DMA	nd	0.45	0.74	1.63	0.56	0.35
C16:0	22.14	26.69	31.53	6.56	36.66	31.14
C16:1n7	nd	9.19	9.26	3.01	10.16	10.32
C17:0	1.26	0.99	0.98	nd	0.90	0.88
C17:1n7	nd	0.39	0.24	2.30	nd	nd
C18:0	7.60	5.67	5.84	3.10	6.48	5.66
C18:1n12t	nd	0.29	0.25	nd	0.19	0.21
C18:1n9t	0.93	0.12	0.26	2.64	0.22	nd
C18:1n9	4.09	3.70	5.07	nd	4.65	3.43
C18:1n7	3.10	2.65	2.68	1.75	3.26	2.83
C18:2n6t	nd	0.11	nd	1.33	nd	nd
C18:2n6	1.00	1.57	1.92	2.85	1.81	1.94
C18:3n6	0.73	0.45	0.19	1.34	0.18	0.28
C19:0	0.50	nd	nd	2.09	nd	nd
C18:3n3	2.53	1.65	2.25	1.93	2.09	1.86
C18:4n3	1.80	2.03	2.05	0.56	2.36	3.91
C19:2n6	0.56	0.10	nd	1.59	0.04	Nd

Table A15 continued. Menhaden collected from the upper bay in fall.

	Sample number					
	1	2	3	4	5	6
C20:0	3.59	0.88	0.81	1.15	0.85	1.03
C20:1n15	0.79	0.37	0.16	1.63	0.06	0.12
C20:1n12	1.43	0.48	0.28	nd	0.21	0.29
C20:1n9	nd	0.34	0.58	0.64	0.35	0.39
C20:1n7	1.95	0.34	0.49	nd	0.32	0.34
C20:2n6	nd	0.52	0.36	3.08	0.15	0.39
C20:3n6	1.18	0.27	nd	1.30	0.23	0.16
C21:0	1.17	nd	0.14	nd	0.11	0.13
C20:4n6	0.68	0.87	0.88	1.23	0.82	0.78
C20:3n3	2.14	0.34	0.39	0.98	0.23	0.31
C20:5n3	3.21	4.77	4.88	1.92	5.59	8.33
C22:0	3.13	1.49	1.58	4.04	1.17	1.57
C22:1n9	1.93	0.57	0.49	nd	0.11	0.27
C22:2n6	1.72	nd	0.91	4.25	0.15	0.08
C22:4n6	1.99	nd	0.85	nd	0.18	0.12
C22:3n3 + C22:5n6	2.38	2.33	1.07	8.70	0.48	0.49
C22:5n3	nd	1.64	nd	nd	nd	nd
C22:6n3 + C24:0	10.02	9.66	11.38	20.29	9.20	10.44
C24:1n9	6.67	3.39	1.63	8.56	0.38	0.45

Table A16. Spot collected from the upper bay in summer.

	Sample number					
	1	2	3	4	5	6
Total length (mm)	90	71	86	90	93	72
Weight (g)	9.63	4.12	7.93	9.08	11.25	4.69
Lipid content (%)	3.59	4.35	5.16	5.65	5.44	4.54
C12:0	0.31	0.19	nd	0.53	0.44	4.46
C12:1n1	nd	nd	nd	0.19	nd	0.88
C13:0	nd	0.42	1.14	1.09	0.57	nd
C14:0	3.26	2.55	2.59	3.55	3.94	3.20
C14:1n5	0.15	0.08	0.35	0.25	0.45	nd
C15:0	2.21	2.35	2.35	2.22	2.19	1.88
C16:0 DMA	1.49	1.10	1.18	1.35	1.10	1.36
C16:0	27.33	25.62	21.60	25.47	26.22	28.00
C16:1n7	1.99	2.65	1.88	2.14	2.48	1.60
C17:0	2.55	2.78	2.77	2.15	2.48	2.78
C17:1n7	0.20	nd	nd	nd	0.87	0.51
C18:0	14.23	13.61	13.15	12.35	12.32	14.59
C18:1n12t	0.60	1.01	nd	0.47	0.37	0.57
C18:1n9t	0.35	nd	0.73	0.53	nd	0.26
C18:1n7t	nd	nd	0.40	0.38	nd	0.27
C18:1n9	9.42	8.80	9.34	8.91	8.82	7.71
C18:1n7	2.28	2.82	2.56	3.00	2.00	1.85
C18:2n6	2.56	1.71	2.76	2.85	2.12	2.07
C18:3n6 + C19:0	0.26	0.17	0.62	0.46	0.43	nd
C18:3n3	0.92	0.86	1.71	1.12	1.01	0.81
C18:4n3	0.66	0.58	0.60	0.85	0.93	0.70
C19:2n6	0.48	0.13	0.39	0.27	nd	nd

Table A16 continued. Spot collected from the upper bay in summer.

	Sample number					
	1	2	3	4	5	6
C20:0	1.29	1.60	1.90	1.50	1.60	1.41
C20:1n15	0.24	nd	0.51	nd	nd	nd
C20:1n12	1.52	2.07	2.41	1.74	3.00	1.76
C20:1n9	1.15	1.29	1.67	1.04	1.35	1.10
C20:1n7	1.29	1.64	1.63	1.36	2.14	1.30
C20:2n6	1.31	0.27	0.94	1.14	1.72	0.88
C20:3n6 + C21:0	0.85	0.18	2.00	nd	0.53	0.32
C20:4n6	2.54	3.03	3.57	3.07	2.06	1.22
C20:3n3	0.81	0.33	1.38	1.02	0.48	0.36
C20:5n3	3.32	5.02	3.33	4.38	4.35	1.25
C22:0	1.95	1.95	2.31	2.12	1.47	1.12
C22:1n9	0.23	0.22	0.45	0.32	nd	0.92
C22:2n6	nd	0.26	nd	nd	nd	1.01
C22:4n6	0.37	0.79	0.88	nd	0.38	nd
C22:3n3 + C22:5n6	1.05	0.98	1.31	2.10	1.19	nd
C22:5n3	1.19	1.49	0.92	1.46	1.04	2.24
C22:6n3 + C24:0	8.77	11.14	8.68	7.82	9.30	7.46
C24:1n9	0.87	0.31	nd	0.80	0.65	4.16

Table A17. Spot collected from the upper bay in fall.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
Total length (mm)	81	103	75	71	87	76	86	74	75	76	71
Weight (g)	5.48	13.04	3.79	4.44	8.43	5.39	7.41	4.52	5.00	4.76	4.27
Lipid content (%)	6.87	2.50	8.71	4.17	7.84	5.74	2.55	10.06	5.65	4.57	7.90
C12:0	0.07	0.06	0.12	nd	0.03	0.04	0.02	0.08	0.06	nd	0.07
C12:1n1	0.09	nd	0.02	nd	nd	0.02	0.01	0.02	0.01	nd	0.02
C13:0	0.65	0.18	0.22	nd	0.24	nd	nd	0.39	0.74	nd	0.40
C14:0	2.62	4.00	2.93	1.04	2.40	1.93	1.77	3.34	2.23	1.88	2.52
C14:1n5	0.03	0.03	0.06	nd	0.04	0.03	0.02	0.04	0.02	0.02	0.03
C15:0	1.20	1.37	1.34	0.96	1.38	1.09	1.30	1.37	1.35	1.39	1.58
C16:0	23.94	27.47	25.82	22.23	23.14	24.60	23.58	26.64	24.26	22.23	23.98
C16:1n7	4.15	4.63	4.22	2.39	4.70	3.80	4.13	5.77	4.06	4.00	4.48
C17:0	2.22	2.16	2.62	1.99	2.28	2.30	2.51	2.86	2.55	2.26	3.52
C18:0	9.24	10.99	9.97	11.15	10.33	11.70	12.06	9.97	11.55	9.95	12.25
C18:1n12t	nd	nd	0.39	0.29	nd	nd	nd	nd	nd	nd	nd
C18:1n9t	nd	nd	0.39	0.29	nd	nd	nd	nd	nd	nd	nd
C18:1n9	9.75	8.67	10.58	8.88	8.55	10.15	8.69	10.56	8.84	8.95	8.65
C18:1n7	3.56	3.39	4.01	3.13	3.95	3.79	3.87	4.08	3.94	3.74	5.20
C18:2n6t	0.14	nd	nd	nd	nd	nd	nd	0.15	nd	nd	nd
C18:2n6	1.71	1.69	1.96	1.44	1.57	1.93	1.73	2.10	1.64	1.51	1.95
C18:3n6	0.10	0.13	0.16	0.09	0.12	0.15	0.11	0.17	0.12	0.12	0.14
C19:0	0.38	0.58	0.50	0.41	0.57	0.52	0.51	0.51	0.48	0.46	0.57
C18:3n3	1.12	1.07	1.40	0.61	0.97	0.91	0.85	1.54	1.02	0.70	1.00
C18:4n3	0.53	0.48	0.73	0.20	0.44	0.36	0.40	0.65	0.61	0.37	0.65
C19:2n6	0.05	0.06	0.05	0.05	0.05	0.06	0.04	0.05	0.09	0.06	0.03

Table A17 continued. Spot collected from the upper bay in fall.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
C20:0	0.59	0.81	0.52	0.25	0.49	0.42	0.32	0.49	0.42	0.29	0.52
C20:1n15	nd	nd	nd	nd	0.07	0.06	0.06	0.05	0.06	0.06	0.04
C20:1n12	1.44	0.99	1.10	0.52	2.35	1.44	1.79	1.09	1.25	1.47	1.70
C20:1n9	1.43	0.95	0.90	0.44	1.10	0.90	0.79	0.87	0.79	0.81	0.89
C20:1n7	2.09	1.16	1.25	0.47	2.76	1.42	1.77	1.24	1.27	1.53	1.86
C20:2n6	1.82	0.79	0.88	0.58	1.12	0.86	1.03	0.82	0.95	0.87	1.14
C20:3n6	0.36	0.15	0.13	0.14	0.19	0.16	0.17	0.12	0.15	0.19	0.13
C21:0	0.26	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:4n6	4.36	4.12	3.81	5.40	3.82	4.93	4.93	3.33	4.60	5.84	4.97
C20:3n3	0.48	0.38	0.48	0.42	0.59	0.36	0.40	0.42	0.47	0.41	0.40
C20:5n3	5.57	5.95	5.91	6.99	6.51	6.83	7.38	6.63	7.04	7.22	5.64
C22:0	0.53	0.46	0.48	0.40	0.42	0.32	0.27	0.38	0.35	0.31	0.28
C22:1n9	0.37	0.20	0.40	0.30	0.35	0.23	0.22	0.20	0.25	0.35	0.15
C22:2n6	0.19	0.06	0.24	0.21	0.36	0.12	0.07	0.11	0.09	0.09	0.10
C22:4n6	1.46	1.55	1.03	1.35	1.43	1.16	1.25	0.61	0.90	1.01	0.88
C22:3n3 + C22:5n6	1.45	1.87	1.26	2.48	1.47	1.06	1.02	0.81	1.19	1.24	0.89
C22:5n3	2.59	2.01	2.15	2.60	2.50	2.20	2.27	1.55	1.72	1.81	1.68
C24:0 + C22:6n3	12.68	11.28	11.45	21.41	12.57	13.67	14.21	10.69	14.39	18.35	11.44
C24:1n9	0.77	0.31	0.57	0.90	1.12	0.48	0.42	0.30	0.52	0.50	0.28

Chapter 3 Raw Data

Values for length (mm), weight (g), lipid content (% wet weight) and fatty acids routinely identified for striped bass liver and belly flap samples analyzed in Chapter 3 of this experiment.

Table B1. Striped bass liver samples from day 0.

	Sample number								
	1	2	3	4	5	6	7	8	9
Total length (mm)	344	344	314	303	373	313	322	303	300
Weight (g)	491.4	493.4	357.6	326.2	594.0	353.1	382.4	325.6	358.8
Lipid content (%)	6.47	9.76	7.20	6.40	5.09	9.59	4.94	10.94	13.45
C12:0	0.09	nd	0.06	0.04	0.06	nd	0.05	0.06	0.04
C12:1n1	0.03	0.05	0.02	0.02	0.02	0.00	0.02	0.03	0.02
C13:0	nd	nd	nd	0.09	0.34	nd	nd	nd	nd
C14:0	3.70	4.50	3.34	2.86	2.05	2.22	3.28	4.75	3.30
C14:1n5	0.06	0.08	0.07	0.08	0.05	0.05	0.07	0.13	0.09
C15:0	0.62	0.71	0.62	0.47	0.30	0.40	0.54	0.61	0.43
C16:0	29.55	26.93	25.49	23.91	18.94	22.45	23.98	31.89	22.61
C16:1n7	5.05	5.77	4.57	4.69	3.34	3.89	4.92	6.45	5.79
C17:0	0.99	0.83	0.94	0.87	0.73	0.78	1.08	1.02	0.69
C18:0	8.62	6.36	7.40	6.68	5.49	7.04	6.84	7.84	5.71
C18:1n9t	0.17	0.15	0.21	0.21	0.31	0.29	0.21	0.26	0.23
C18:1n9	13.66	13.63	13.02	18.96	15.99	14.23	13.97	21.57	23.80
C18:1n7	3.39	3.05	3.27	4.07	3.36	3.49	3.78	4.24	4.49
C18:2n6	nd	11.33	7.85	8.29	5.60	7.39	8.99	8.01	9.24
C18:3n6	0.18	0.21	0.16	0.19	0.32	0.25	0.20	0.12	0.28
C19:0	0.25	0.23	0.23	0.22	0.40	0.35	0.30	0.24	0.29
C18:3n3	1.28	1.31	1.08	1.15	1.30	1.17	1.27	1.27	1.48
C18:4n3	0.33	0.42	0.39	0.44	0.65	0.46	0.48	0.24	0.57
C19:2n6	0.07	0.05	0.07	nd	0.23	0.19	0.10	nd	nd
C20:0	0.19	0.15	0.15	0.14	0.56	0.32	0.20	0.23	0.17

Table B1 continued. Striped bass liver samples from day 0.

	Sample number								
	1	2	3	4	5	6	7	8	9
C20:1n15	nd	nd	0.01	0.04	0.25	0.11	0.03	0.02	0.05
C20:1n12	0.18	0.13	0.08	0.16	0.51	0.23	0.15	0.24	0.13
C20:1n9	1.16	0.96	1.23	1.61	1.91	1.36	1.34	2.24	1.83
C20:1n7	0.32	0.22	0.18	0.35	0.71	0.31	0.26	0.33	0.26
C20:2n6	1.07	0.98	0.79	0.83	1.64	0.95	0.97	0.60	0.69
C20:3n6	0.12	0.14	0.13	0.14	0.48	0.33	0.20	0.08	0.14
C21:0	nd	nd	0.01	nd	nd	nd	nd	nd	0.01
C20:4n6	3.18	1.60	2.52	1.87	3.12	2.64	2.18	0.55	1.15
C20:3n3	0.18	0.22	0.15	0.15	0.78	0.57	0.13	0.08	0.16
C20:5n3	5.45	5.53	5.45	6.55	5.91	5.80	5.18	2.45	3.91
C22:0	0.08	0.82	0.09	0.44	0.80	0.52	0.15	0.45	0.16
C22:1n9	nd	nd	0.16	0.22	0.85	nd	0.19	nd	0.18
C22:2n6	0.04	0.22	0.05	0.04	1.06	0.53	0.09	0.05	0.05
C22:4n6	0.14	nd	0.13	0.13	0.61	0.37	0.17	0.08	0.11
C22:3n3 + C22:5n6	0.38	0.30	0.29	0.24	1.51	1.10	0.36	0.12	0.22
C22:5n3	0.76	0.80	0.71	0.63	2.74	1.54	0.83	0.28	0.74
C24:0 + C22:6n3	18.32	11.98	18.67	12.88	15.08	17.48	17.03	3.18	10.63
C24:1n9	0.37	0.35	0.42	0.32	2.02	1.21	0.48	0.29	0.38

Table B2. Striped bass belly flap samples from day 0.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	344	344	314	339	303	373	326	313	322	322	303	300
Weight (g)	491.4	493.4	357.6	475.4	326.2	594.0	431.4	353.1	400.5	382.4	325.6	358.8
Lipid content (%)	5.63	12.02	15.48	10.84	20.10	-	10.41	9.68	14.10	10.07	10.94	12.57
C12:0	0.09	0.07	0.11	0.09	0.08	0.10	0.11	0.08	nd	0.08	0.06	0.15
C12:1n1	0.02	0.02	0.03	0.03	0.03	0.04	0.03	0.02	0.03	0.03	0.03	0.07
C13:0	0.29	0.18	0.23	0.13	0.11	0.12	0.24	0.37	0.18	0.17	0.13	nd
C14:0	5.97	4.81	5.77	5.65	5.14	5.72	6.21	5.26	6.15	5.83	5.54	5.33
C14:1n5	0.19	0.23	0.24	0.24	0.24	0.25	0.29	0.18	0.26	0.25	0.27	0.22
C15:0	0.64	0.46	0.63	0.55	0.57	0.66	0.62	0.56	0.68	0.69	0.58	0.62
C15:1n5	nd	nd	nd	nd	nd	nd	nd	0.01	nd	nd	nd	nd
C16:0	29.24	20.38	22.64	24.65	23.89	24.88	26.28	23.57	27.92	23.15	24.56	25.14
C16:1n7	7.70	7.05	7.91	7.99	7.76	7.99	8.72	7.81	8.63	8.28	8.08	8.18
C17:0	0.72	0.58	0.62	0.69	0.62	0.68	0.70	0.74	0.77	0.56	0.65	0.63
C18:0	4.25	3.03	3.58	3.68	3.72	3.44	3.49	4.05	3.77	3.27	3.78	4.30
C18:1n9t	0.28	0.21	0.16	0.16	0.19	0.16	0.18	0.18	0.17	0.17	0.17	0.20
C18:1n9	23.64	21.31	22.21	22.92	24.58	22.50	24.72	24.58	25.60	22.59	24.59	24.16
C18:1n7	4.12	3.76	3.77	3.87	4.10	3.90	4.25	4.14	4.42	3.93	4.23	4.45
C18:2n6	nd	12.63	12.50	13.63	13.73	13.10	12.79	13.80	10.46	14.60	12.88	10.76
C18:3n6	0.32	0.38	0.28	0.32	0.33	0.32	0.28	0.27	0.23	0.32	0.32	0.30
C19:0	0.39	0.40	0.28	0.26	0.29	0.26	0.31	0.30	0.32	0.54	0.32	0.30
C18:3n3	nd	1.94	1.73	1.66	1.82	1.74	1.43	1.49	1.09	1.92	1.70	1.60
C18:4n3	0.78	0.82	0.85	0.70	0.73	0.79	0.51	nd	0.38	0.89	0.62	0.80
C19:2n6	nd	nd	0.11	0.11	nd	nd	0.05	0.02	nd	0.06	nd	0.19
C20:0	0.44	0.67	0.29	0.24	0.20	0.21	0.18	0.19	0.20	0.21	0.22	0.62
C20:1n15	0.14	0.13	nd	0.04	0.05	nd	0.03	0.04	0.01	0.06	0.02	0.19
C20:1n12	0.38	0.39	0.16	0.16	0.16	0.15	0.14	0.18	0.20	0.18	0.18	0.10
C20:1n9	1.75	1.70	1.31	1.33	1.29	1.18	1.38	1.46	1.66	1.23	1.52	1.47

Table B2 continued. Striped bass belly flap samples day 0.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C20:1n7	0.62	0.48	0.31	0.23	0.18	0.25	0.17	0.22	0.34	0.23	0.22	0.31
C20:2n6	1.09	1.22	0.51	0.63	nd	0.60	nd	0.68	0.53	0.61	0.61	0.67
C20:3n6	0.51	0.92	0.25	0.18	0.14	0.18	0.17	0.16	0.10	0.21	0.45	0.28
C21:0	nd	nd	nd	nd	nd	0.02	nd	nd	nd	nd	0.07	nd
C20:4n6	0.97	0.94	0.80	0.61	0.58	0.61	0.38	0.60	0.27	0.61	0.92	0.69
C20:3n3	0.35	0.67	0.27	0.10	0.12	0.13	0.11	0.13	0.08	0.13	0.36	0.12
C20:5n3	3.35	3.51	5.28	2.98	3.28	3.27	2.09	2.82	1.93	3.26	2.36	2.77
C22:0	0.67	0.53	0.64	0.16	0.21	0.11	0.09	0.13	0.38	0.11	0.13	0.30
C22:1n9	0.90	0.94	0.30	0.15	0.17	0.14	0.18	0.22	0.25	0.14	0.19	0.27
C22:2n6	0.63	0.48	0.06	0.06	0.06	0.07	0.04	0.06	0.07	0.06	0.06	0.15
C23:0	nd	nd	nd	0.08	nd	nd	nd	nd	nd	nd	nd	nd
C22:4n6	0.57	0.65	0.16	0.13	0.11	0.17	0.11	0.14	0.09	0.14	0.13	0.25
C22:3n3 + C22:5n6	0.97	1.03	0.28	0.25	0.27	0.30	0.22	0.24	0.12	0.23	0.15	0.28
C22:5n3	1.49	2.00	0.76	0.71	0.70	0.77	0.59	0.73	0.43	0.78	0.58	0.50
C24:0 + C22:6n3	5.16	4.37	4.81	4.39	4.33	4.95	2.66	4.26	2.02	4.28	3.13	3.36
C24:1n9	1.40	1.09	0.16	0.21	0.21	0.25	0.27	0.29	0.26	0.21	0.21	0.28

Table B3. Striped bass liver samples from day 12.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
Total length (mm)	341	322	342	354	355	339	323	350	323	347	317
Weight (g)	487.3	363.8	461.3	509.5	518.8	435.5	398.4	504.2	371.5	467.4	380.1
Lipid content (%)	6.81	4.26	9.81	7.59	2.14	9.32	5.77	6.34	6.03	7.82	5.72
C12:0	0.01	0.04	0.02	0.04	0.03	0.03	0.02	0.02	0.02	0.02	0.02
C12:1n1	nd	0.02	nd	0.01	0.02	nd	0.01	0.16	0.18	0.16	0.07
C14:0	1.71	2.76	2.59	2.06	2.37	2.76	2.61	2.37	2.27	3.23	1.60
C14:1n5	0.04	0.04	0.06	0.04	0.05	0.09	0.05	0.06	0.04	0.07	0.03
C15:0	0.45	0.75	0.58	0.43	0.67	0.53	0.64	0.51	0.57	0.57	0.41
C15:1n5	nd	nd	nd	nd	nd	nd	nd	0.02	0.01	0.01	0.04
C16:0 DMA	nd	nd	nd	nd	nd	nd	nd	0.03	0.02	0.02	0.04
C16:0	20.15	23.58	24.73	18.79	23.92	22.35	22.75	24.54	27.46	24.98	17.06
C16:1n7	3.71	3.95	4.25	3.61	3.86	5.62	4.84	4.01	3.11	5.24	2.49
C17:0	1.13	1.39	1.35	1.04	1.47	0.96	1.28	0.96	1.04	0.84	0.85
C18:0	6.30	7.53	8.09	6.40	8.26	5.28	7.39	6.61	8.00	5.92	5.09
C18:1n12t	nd	nd	nd	nd	nd	nd	nd	0.35	0.36	0.25	0.36
C18:1n9t	0.47	0.19	0.30	0.40	0.23	0.29	0.29	0.24	0.23	0.20	0.25
C18:1n9	17.90	10.99	13.07	18.46	12.59	20.96	14.49	17.46	12.70	17.11	9.41
C18:1n7	4.31	3.43	3.54	4.23	3.57	4.65	4.11	3.95	3.23	3.82	2.66
C18:2n6	6.40	7.57	8.20	7.66	6.89	8.17	8.58	5.99	5.16	9.04	4.95
C18:3n6	0.24	0.18	0.20	0.30	0.19	0.24	0.22	0.37	0.36	0.37	0.74
C19:0	0.47	0.37	0.47	0.65	0.40	0.33	0.56	nd	nd	nd	nd
C18:3n3	1.09	1.03	1.18	1.64	0.92	1.25	1.33	0.83	0.72	1.11	1.21
C18:4n3	0.42	0.32	0.32	0.58	0.31	0.41	0.32	0.37	0.29	0.36	0.77
C19:2n6	0.13	nd	0.05	nd	0.09	0.05	0.04	nd	nd	nd	nd
C20:0	0.31	0.16	0.15	0.42	0.16	0.14	0.14	0.17	0.17	0.14	0.93
C20:1n15	0.15	0.01	nd	0.15	nd	0.03	0.04	nd	nd	nd	nd

Table B3 continued. Striped bass liver samples from day 12.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
C20:1n12	0.44	0.25	0.20	0.51	nd	0.30	0.24	0.33	0.24	0.24	0.88
C20:1n9	1.94	1.02	1.25	2.21	0.99	2.12	1.27	1.62	1.54	1.65	1.71
C20:1n7	0.60	0.38	0.34	0.74	0.33	0.49	0.40	0.48	0.41	0.38	1.30
C20:2n6	1.12	0.92	0.92	1.36	0.89	0.86	0.90	0.74	0.81	0.93	1.73
C20:3n6	0.33	0.17	0.16	0.29	0.11	0.14	0.15	0.15	0.12	0.14	0.97
C21:0	nd	nd	nd	nd	0.01	nd	nd	nd	nd	nd	nd
C20:4n6	3.17	4.03	3.50	2.71	3.64	2.09	3.27	3.03	3.55	2.53	4.42
C20:3n3	0.45	0.25	0.19	0.57	0.23	0.27	0.23	0.20	0.18	0.18	1.26
C20:5n3	5.59	5.73	5.17	4.99	6.95	4.36	5.09	5.16	5.21	4.19	6.09
C22:0	0.34	0.10	0.14	0.40	0.52	0.21	0.12	0.28	0.28	0.23	1.37
C22:1n9	nd	nd	nd	0.64	nd	0.34	nd	0.13	0.18	0.14	1.34
C22:2n6	0.39	0.11	0.12	0.69	0.07	0.09	0.10	0.03	0.02	0.03	1.86
C22:4n6	0.52	0.29	0.22	0.59	0.26	0.21	0.23	0.46	0.41	0.26	2.31
C22:3n3 + C22:5n6	1.33	0.68	0.43	1.21	0.47	0.41	0.44	0.53	0.50	0.39	3.71
C22:5n3	1.84	1.47	0.98	2.02	1.16	0.92	0.93	1.32	1.11	1.06	3.37
C24:0 + C22:6n3	15.34	19.81	16.70	13.26	17.84	12.62	16.51	16.01	18.97	13.77	18.70
C24:1n9	1.22	0.47	0.53	0.87	0.50	0.43	0.40	0.50	0.53	0.40	nd

Table B4. Striped bass belly flap samples from day 12.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
Total length (mm)	341	322	342	355	339	323	365	350	323	347	317
Weight (g)	487.3	363.8	461.3	509.5	518.8	435.5	398.4	504.2	371.5	467.4	380.1
Lipid content (%)	13.92	9.81	16.16	14.03	11.42	11.45	8.92	13.12	11.91	8.62	10.97
C12:0	0.05	0.10	0.03	0.08	0.08	0.02	0.05	0.07	0.10	0.08	0.08
C12:1n1	nd	0.05	nd	0.04	nd	0.16	0.49	nd	1.22	1.06	1.09
C13:0	0.12	0.17	nd	0.06	0.22	nd	nd	nd	nd	nd	nd
C14:0	4.37	6.11	3.30	3.78	4.45	3.23	4.93	4.65	4.48	4.81	5.04
C14:1n5	0.32	0.25	0.21	0.17	0.23	0.07	0.22	0.25	0.25	0.26	0.23
C15:0	0.64	0.85	0.35	0.55	0.58	0.57	0.59	0.62	0.68	0.59	0.56
C15:1n5	nd	nd	nd	nd	0.01	0.01	0.05	0.06	nd	0.06	nd
C16:0 DMA	nd	nd	nd	nd	nd	0.02	0.03	0.05	0.12	nd	nd
C16:0	21.42	22.86	14.94	16.38	23.11	24.98	22.89	22.86	23.24	22.19	21.90
C16:1n7	7.57	7.88	5.40	5.70	8.14	5.24	7.57	7.59	7.17	7.89	7.84
C17:0	0.75	0.73	0.49	0.59	0.71	0.84	0.59	0.58	0.63	0.52	0.54
C18:0	4.39	3.95	3.16	3.07	4.01	5.92	4.00	4.20	4.67	3.81	3.92
C18:1n12t	nd	nd	nd	nd	nd	0.25	0.15	0.20	0.15	0.15	0.12
C18:1n9t	0.37	0.16	0.28	0.22	0.24	0.20	0.17	0.15	0.20	0.19	0.20
C18:1n9	22.18	20.50	17.19	18.38	22.27	17.11	23.02	21.17	21.66	23.83	23.07
C18:1n7	4.14	3.80	3.28	3.52	4.22	3.82	3.95	3.77	3.87	3.98	3.89
C18:2n6t	0.20	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:2n6	12.50	12.99	10.05	10.33	12.15	9.04	12.81	11.11	11.65	12.36	13.19
C18:3n6	0.36	0.30	0.42	0.43	0.31	0.37	0.38	0.41	0.38	0.34	0.39
C19:0	0.45	0.43	0.42	0.46	0.28	nd	nd	nd	nd	nd	nd
C18:3n3	1.43	1.68	1.91	1.90	1.67	1.11	1.54	1.57	1.48	1.55	1.68
C18:4n3	0.61	0.83	0.98	0.97	0.69	0.36	0.71	0.86	0.73	0.71	0.74
C19:2n6	nd	nd	0.63	nd	nd	nd	nd	nd	0.03	nd	nd

Table B4 continued. Striped bass belly flap samples from day 12.

	Sample number										
	1	2	3	4	5	6	7	8	9	10	11
C20:0	0.41	0.19	0.81	0.79	0.19	0.14	0.22	0.21	0.23	0.22	0.23
C20:1n15	0.21	nd	0.42	0.26	0.03	nd	0.02	nd	nd	nd	nd
C20:1n12	0.12	0.21	0.57	0.66	0.25	0.24	0.25	0.31	nd	0.22	0.22
C20:1n9	1.43	1.12	1.72	1.67	1.37	1.65	1.42	1.14	1.00	1.65	1.29
C20:1n7	0.46	0.25	0.88	0.75	0.27	0.38	0.31	0.32	0.27	0.28	0.27
C20:2n6	0.80	0.60	1.09	1.26	0.66	0.93	0.70	0.58	0.66	0.78	0.72
C20:3n6	0.32	0.23	1.50	0.77	0.21	0.14	0.25	0.25	0.33	0.25	0.29
C21:0	nd	0.07	nd	0.42	0.02	nd	nd	nd	nd	0.07	nd
C20:4n6	1.15	0.99	2.16	1.52	1.01	2.53	0.89	1.35	1.14	1.04	0.98
C20:3n3	0.29	0.18	1.25	1.00	0.23	0.18	0.21	0.23	0.29	0.24	0.20
C20:5n3	4.06	3.76	3.87	4.04	3.82	4.19	3.64	4.89	3.91	3.60	3.81
C22:0	0.38	0.14	1.72	1.21	0.13	0.23	0.22	0.21	nd	0.18	0.24
C22:1n9	0.27	0.16	1.15	1.15	0.14	0.14	0.16	0.13	0.16	0.23	0.16
C22:2n6	nd	0.10	0.99	0.80	0.04	0.03	0.09	0.08	nd	nd	nd
C22:4n6	0.32	0.24	1.82	1.38	0.17	0.26	0.35	0.52	0.25	0.45	0.19
C22:3n3 + C22:5n6	0.45	0.30	2.54	1.90	0.42	0.39	0.47	0.58	0.51	0.28	0.53
C22:5n3	1.32	1.19	3.61	3.04	0.87	1.06	1.00	1.16	1.00	0.89	0.86
C24:0 + C22:6n3	5.75	6.31	7.85	8.72	6.38	13.77	5.34	7.57	7.26	4.96	5.10
C24:1n9	0.43	0.31	2.98	2.01	0.40	0.40	0.34	0.31	0.26	0.30	0.44

Table B5. Striped bass liver samples from day 28.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	315	362	331	339	319	348	332	309	317	368	291	352
Weight (g)	423.3	612.7	464.1	519.1	413.4	530.8	444.2	367.7	362.8	656.2	334.9	595.5
Lipid content (%)	4.70	6.58	6.30	5.51	10.63	4.75	9.41	10.91	5.19	7.35	4.45	6.52
C12:0	0.01	0.02	0.02	0.03	0.01	0.01	0.03	0.03	nd	nd	nd	0.01
C12:1n1	nd	0.01	nd	0.02	nd	nd	nd	nd	nd	0.02	nd	nd
C13:0	nd	0.32	0.11	0.20	nd	nd	0.10	0.13	nd	0.07	nd	0.12
C14:0	1.77	2.47	2.38	2.31	1.68	1.85	3.44	2.14	1.62	2.42	1.80	2.25
C14:1n5	0.07	0.08	0.10	0.09	0.09	0.05	0.15	0.09	0.03	0.08	0.04	0.09
C15:0	0.48	0.60	0.77	0.62	0.29	0.65	0.67	0.40	0.65	0.61	0.82	0.55
C15:1n5	nd	nd	nd	nd	0.01	nd	nd	nd	nd	nd	nd	nd
C16:0	22.39	19.20	20.38	23.39	25.18	21.28	21.11	27.45	20.16	17.43	22.80	19.24
C16:1n7	3.66	5.34	4.35	3.94	4.31	3.17	5.75	4.42	2.70	4.95	3.07	5.33
C17:0	1.34	1.10	1.46	1.58	0.81	1.69	1.20	1.03	1.65	1.31	1.93	1.19
C18:0	6.31	5.01	7.42	7.45	5.55	8.09	6.05	6.23	7.87	6.27	9.62	5.36
C18:1n9t	0.42	0.24	0.25	0.25	0.44	0.30	0.23	0.39	0.30	0.27	0.25	0.28
C18:1n9	21.69	19.84	14.13	15.93	28.64	10.95	20.67	27.39	9.34	18.20	11.16	24.02
C18:1n7	4.79	4.44	4.08	3.96	4.58	3.53	4.27	4.75	3.09	4.73	3.76	5.22
C18:2n6	3.25	9.63	7.93	5.12	1.34	5.49	9.42	3.07	4.38	9.03	3.56	7.71
C18:3n6	0.17	0.25	0.25	0.21	0.19	0.20	0.26	0.22	0.17	0.29	0.13	0.29
C19:0	0.37	0.39	0.42	0.43	0.34	0.52	0.42	0.41	0.54	0.47	0.47	0.47
C18:3n3	0.62	1.49	1.05	0.77	0.41	0.99	1.31	0.75	0.80	1.74	0.79	1.49
C18:4n3	0.32	0.48	0.47	0.35	0.24	0.29	0.52	0.35	0.37	0.58	0.26	0.59
C19:2n6	0.07	0.09	nd	0.07	0.11	0.06	0.09	0.08	0.11	0.15	0.07	0.08
C20:0	0.20	0.17	0.17	0.22	0.20	0.20	0.18	0.23	0.34	0.20	0.19	0.25
C20:1n15	0.05	0.07	0.03	0.05	0.07	0.05	0.03	0.08	0.13	0.09	0.03	0.07
C20:1n12	0.57	0.43	0.34	0.46	0.42	0.31	0.39	0.51	0.41	0.55	0.40	0.66

Table B5 continued. Striped bass liver samples from day 28.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C20:1n9	2.36	2.24	1.55	1.86	2.88	1.32	1.54	2.47	1.12	1.79	1.24	2.22
C20:1n7	0.84	0.63	0.48	0.63	0.50	0.44	0.49	0.61	0.56	0.72	0.56	0.88
C20:2n6	0.72	1.15	1.05	0.85	0.41	0.90	0.79	0.67	1.03	1.09	0.83	0.98
C20:3n6	0.09	0.22	0.19	0.17	0.09	0.11	0.14	0.09	0.39	0.25	0.11	0.23
C21:0	0.01	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.01	nd
C20:4n6	3.30	2.82	3.94	4.25	2.33	5.05	2.62	2.02	5.27	2.55	4.97	2.21
C20:3n3	0.27	0.24	0.24	0.23	0.21	0.24	0.20	0.16	0.62	0.39	0.27	0.23
C20:5n3	5.25	5.02	5.66	5.93	3.98	7.06	4.68	3.20	7.09	4.98	6.33	4.28
C22:0	0.19	0.19	0.18	0.19	0.20	0.31	0.11	0.07	0.44	0.27	0.12	0.08
C22:1n9	0.51	0.40	0.21	0.23	0.57	nd	0.20	0.25	nd	0.33	0.14	0.30
C22:2n6	0.08	0.23	0.06	0.05	0.16	0.11	0.07	0.03	0.32	0.40	0.05	0.06
C22:4n6	0.39	0.44	0.35	0.35	0.35	0.41	0.26	0.27	0.61	0.46	0.42	0.36
C22:3n3 + C22:5n6	0.63	0.86	0.66	0.67	0.83	0.68	0.46	0.43	1.77	0.80	0.73	0.56
C22:5n3	1.36	1.74	1.48	1.41	1.16	1.35	1.12	0.79	2.21	1.96	1.49	1.39
C24:0 + C22:6n3	15.18	11.81	17.35	15.39	10.64	21.81	10.66	8.56	21.75	14.07	21.29	10.64
C24:1n9	0.30	0.34	0.48	0.31	0.77	0.53	0.36	0.23	2.16	0.48	0.29	0.33

Table B6. Striped bass belly flap samples from day 28.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	315	362	331	339	319	348	332	309	317	368	291	352
Weight (g)	423.3	612.7	464.1	519.1	413.4	530.8	444.2	367.7	362.8	656.2	334.9	595.5
Lipid content (%)	15.50	10.48	12.47	12.60	9.26	8.65	22.09	12.22	-	11.92	9.89	17.13
C12:0	0.03	0.08	0.07	0.08	0.08	0.05	0.06	0.06	0.06	0.06	0.03	nd
C12:1n1	nd	nd	nd	0.03	nd	nd	nd	nd	0.02	0.08	nd	nd
C13:0	0.43	0.41	0.13	0.43	0.41	0.26	0.45	0.23	0.22	0.68	0.57	nd
C14:0	3.60	3.95	4.80	4.52	3.93	5.04	4.94	3.87	4.41	4.18	4.33	4.57
C14:1n5	0.23	0.32	0.22	0.20	0.16	0.23	0.28	0.20	0.23	0.28	0.18	0.29
C15:0	0.74	0.49	0.58	0.85	0.86	0.68	0.70	0.84	0.72	0.58	0.91	0.84
C15:1n5	0.03	nd	nd	nd	nd	nd	0.02	nd	nd	nd	nd	nd
C16:0	19.42	19.78	21.94	22.27	24.89	21.62	20.91	23.22	23.52	20.75	23.83	21.79
C16:1n7	6.24	7.30	7.65	6.90	6.17	7.31	7.65	6.54	7.26	7.52	6.50	7.76
C17:0	0.95	0.67	0.90	1.11	1.48	0.88	0.72	1.25	0.82	0.85	1.20	1.03
C17:1n7	nd	0.16	nd	nd	nd	0.04	nd	nd	nd	nd	nd	nd
C18:0	4.34	4.03	4.29	4.91	5.96	4.45	4.25	4.90	4.81	3.93	5.93	4.12
C18:1n9t	0.21	0.21	0.23	0.24	0.26	0.22	0.27	0.23	0.21	0.21	0.21	0.14
C18:1n9	17.80	21.61	23.71	19.96	20.60	20.83	24.63	18.77	22.48	22.55	18.99	22.40
C18:1n7	3.71	3.59	3.95	4.03	4.28	3.76	4.36	3.77	3.90	3.95	3.99	3.80
C18:2n6t	0.37	nd	nd	nd	nd	0.20	nd	nd	nd	0.16	nd	nd
C18:2n6	8.37	12.29	13.78	10.75	4.70	11.38	12.98	7.17	12.36	11.65	8.79	11.40
C18:3n6	0.17	0.33	0.45	0.31	0.19	0.23	0.34	0.17	0.27	0.49	0.22	0.43
C19:0	0.38	0.23	0.48	0.31	0.41	0.36	0.33	0.35	0.34	0.42	0.34	0.46
C18:3n3	1.28	1.46	2.26	1.63	1.30	1.53	1.72	1.12	1.41	1.92	1.33	1.69
C18:4n3	0.65	0.60	1.10	0.75	0.49	0.74	0.75	0.62	0.54	0.92	0.64	0.95
C19:2n6	0.09	0.09	nd	0.14	0.07	0.21	nd	0.12	nd	0.11	0.14	0.17
C20:0	0.21	0.18	0.18	0.25	0.32	0.22	0.22	0.24	0.23	0.46	0.29	0.42

238

Table B6 continued. Striped bass belly flap samples from day 28.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C20:1n15	0.05	nd	0.06	0.09	0.02	0.03	0.06	0.03	nd	0.08	0.06	0.22
C20:1n12	0.63	0.22	nd	0.68	0.80	0.43	nd	0.59	0.46	0.23	0.71	0.58
C20:1n9	1.17	1.31	1.34	1.37	1.64	1.17	1.30	1.20	1.37	1.32	1.21	1.46
C20:1n7	0.69	0.07	0.16	0.60	1.01	0.42	0.31	0.62	0.55	0.23	0.73	0.69
C20:2n6	0.59	0.75	0.68	0.82	0.70	0.77	0.58	0.67	0.61	0.67	0.78	0.85
C20:3n6	0.25	0.38	0.30	0.21	0.31	0.32	0.29	0.31	0.17	0.24	0.20	0.28
C21:0	nd	nd	nd	nd	nd	nd	0.02	nd	nd	0.11	nd	nd
C20:4n6	1.34	1.18	1.13	1.35	1.58	1.26	0.79	1.54	1.03	1.26	1.65	1.20
C20:3n3	0.82	0.20	0.21	0.40	0.30	0.21	nd	0.45	0.25	0.31	0.32	0.35
C20:5n3	7.37	4.15	3.66	5.32	4.52	3.94	5.25	4.57	3.43	4.97	4.38	4.06
C22:0	1.09	0.57	0.21	0.29	0.27	0.30	0.21	0.39	0.24	0.27	0.17	0.18
C22:1n9	0.70	0.68	nd	0.33	0.20	0.11	0.22	0.35	0.23	nd	0.14	0.19
C22:2n6	0.45	nd	nd	0.14	0.09	0.03	0.06	0.32	0.04	0.12	0.07	0.11
C22:4n6	1.03	0.69	0.19	0.33	0.49	0.31	0.25	1.04	0.28	nd	0.34	0.25
C22:3n3 + C22:5n6	1.10	0.71	0.30	0.66	0.72	0.51	0.34	1.03	0.49	0.56	0.54	0.49
C22:5n3	2.77	2.48	0.88	1.39	1.78	1.73	0.81	2.96	1.04	1.19	1.38	1.00
C24:0 + C22:6n3	8.85	7.87	4.08	5.95	8.63	7.69	3.63	8.87	5.63	6.21	8.49	5.68
C24:1n9	1.87	0.94	0.08	0.42	0.41	0.52	0.30	1.39	0.39	0.44	0.42	0.15

Table B7. Striped bass liver samples from day 40.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	360	330	344	366	371	350	382	356	379	355	299	342
Weight (g)	698.8	526.4	644.2	707.2	704.3	581.6	705.3	612.9	689.1	604.8	319.9	495.2
Lipid content (%)	5.67	6.16	5.48	4.70	5.44	6.78	3.82	5.36	4.50	8.29	5.56	4.68
C12:0	0.02	0.02	0.03	0.00	0.01	0.02	0.03	0.02	0.02	0.01	0.01	0.01
C12:1n1	nd	nd	nd	nd	nd	nd	0.12	0.10	nd	0.01	nd	0.04
C13:0	nd	0.07	0.09	nd	nd	0.16	0.07	0.08	0.27	0.06	nd	0.30
C14:0	1.98	1.63	1.97	1.99	1.94	2.11	2.63	2.59	1.83	1.94	1.50	2.07
C14:1n5	0.11	0.05	0.06	0.06	0.05	0.07	0.09	0.09	0.06	0.07	0.04	0.08
C15:0	0.33	0.29	0.54	0.48	0.54	0.50	0.64	0.61	0.60	0.42	0.56	0.56
C15:1n5	nd	nd	nd	0.01	nd	nd	0.03	0.03	nd	nd	nd	0.00
C16:0 DMA	nd	nd	nd	nd	nd	nd	0.04	0.04	nd	nd	nd	nd
C16:0	25.22	23.85	24.02	25.46	22.82	21.75	28.61	25.52	21.27	23.95	26.26	21.59
C16:1n7	5.37	3.47	4.15	3.71	4.24	4.83	4.65	4.18	3.65	4.52	3.40	3.86
C17:0	0.88	1.16	1.57	1.11	1.45	1.26	1.20	1.19	1.25	1.11	1.60	1.56
C18:0	6.46	7.05	7.47	7.65	7.54	6.47	nd	7.66	7.19	7.06	8.02	7.84
C18:1n9t	0.43	0.42	0.41	0.39	0.45	0.27	0.29	nd	0.33	0.40	0.31	0.25
C18:1n9	30.50	22.41	21.88	21.47	20.88	21.06	20.06	17.85	14.15	26.02	18.53	16.24
C18:1n7	5.01	4.14	5.01	4.38	5.09	4.86	4.58	4.10	4.04	5.05	4.38	4.25
C18:2n6t	0.15	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:2n6	3.30	2.14	3.57	3.06	4.58	5.45	5.82	4.99	6.40	3.78	1.83	4.80
C18:3n6	0.21	0.23	0.20	0.16	0.22	0.22	0.43	0.42	0.19	0.22	0.14	0.20
C19:0	0.54	0.48	0.52	0.34	0.46	0.38	nd	nd	0.39	0.40	0.43	0.44
C18:3n3	0.72	0.81	0.86	0.70	1.02	1.21	0.88	0.75	0.91	0.90	0.54	0.89
C18:4n3	0.35	0.42	0.36	0.29	0.43	0.39	0.39	0.40	0.36	0.42	0.31	0.41
C19:2n6	0.10	nd	0.07	0.07	0.11	nd	0.03	nd	0.10	0.08	0.05	0.04
C20:0	0.19	0.55	0.24	0.18	0.31	0.16	0.20	0.20	0.18	0.26	0.21	0.19

Table B7 continued. Striped bass liver samples from day 40.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C20:1n15	0.04	0.12	0.04	0.04	0.07	0.03	nd	nd	0.04	0.06	0.03	0.03
C20:1n12	0.49	0.62	0.58	0.42	0.73	0.50	0.44	0.44	0.40	0.61	0.61	0.46
C20:1n9	2.81	2.30	3.00	2.25	3.25	2.97	2.55	1.84	1.76	2.72	2.25	2.03
C20:1n7	0.55	0.76	0.83	0.64	0.87	0.68	0.65	0.59	0.52	0.72	0.72	0.63
C20:2n6	0.49	0.94	0.78	0.61	0.87	0.93	nd	0.80	1.01	0.78	0.65	0.85
C20:3n6	0.09	0.28	0.09	0.13	0.15	0.14	0.16	0.18	0.14	0.09	0.10	0.15
C20:4n6	1.86	3.17	2.93	3.44	2.78	2.52	3.48	3.59	5.05	2.18	3.89	3.70
C20:3n3	0.13	0.44	0.21	0.20	0.26	0.25	0.22	0.23	0.37	0.21	0.22	0.24
C20:5n3	2.99	4.28	3.96	4.61	4.07	3.94	4.67	4.40	6.55	3.68	4.64	5.23
C22:0	0.08	0.36	0.12	0.09	0.09	0.12	0.16	0.16	0.21	0.10	0.14	0.17
C22:1n9	0.30	0.87	0.27	0.33	0.32	0.53	0.25	0.18	nd	0.34	0.24	0.34
C22:2n6	0.03	0.85	0.07	0.11	0.09	0.19	0.05	0.05	0.07	0.04	nd	0.25
C22:4n6	0.26	0.62	0.37	0.40	0.39	0.48	0.76	0.75	0.38	0.31	0.51	0.50
C22:3n3 + C22:5n6	0.38	1.14	0.52	0.57	0.50	0.86	0.62	0.63	0.65	0.40	0.88	0.80
C22:5n3	0.70	1.98	1.16	1.16	1.18	1.73	1.49	1.52	1.59	1.08	1.64	1.95
C24:0 + C22:6n3	6.78	11.43	11.65	13.12	12.02	12.30	13.22	13.31	17.63	9.75	14.11	16.52
C24:1n9	0.15	0.64	0.40	0.36	0.21	0.65	0.51	0.51	0.44	0.23	1.27	0.51

Table B8. Striped bass belly flap samples from day 40.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	360	330	344	366	371	350	382	356	379	355	299	342
Weight (g)	698.8	526.4	644.2	707.2	704.3	581.6	705.3	612.9	689.1	604.8	319.9	495.2
Lipid content (%)	20.50	14.84	16.47	17.95	7.95	10.98	16.72	10.73	8.84	10.15	7.27	8.99
C12:0	0.04	0.02	0.06	0.07	nd	nd	0.14	0.08	0.06	0.02	0.06	0.15
C12:1n1	nd	nd	nd	nd	nd	0.93	nd	nd	nd	nd	nd	nd
C13:0	0.10	0.14	0.13	0.16	0.09	nd	0.23	0.16	0.25	0.32	nd	nd
C14:0	2.81	3.04	4.54	4.83	4.72	4.35	4.41	4.21	3.85	3.93	3.68	5.24
C14:1n5	0.16	0.15	0.21	0.25	0.24	0.50	0.22	0.21	0.18	0.18	0.15	0.37
C15:0	0.50	0.59	0.79	0.86	0.69	0.88	0.71	0.81	0.58	0.76	1.11	0.83
C15:1n5	nd	nd	nd	nd	0.07	0.15	0.11	0.09	nd	nd	nd	0.15
C16:0 DMA	nd	nd	nd	nd	nd	0.23	0.05	0.06	nd	nd	nd	0.06
C16:0	15.07	15.48	22.89	22.31	21.11	21.11	22.33	23.62	20.41	23.80	25.88	21.96
C16:1n7	4.72	4.87	7.24	7.23	6.67	7.57	7.13	7.23	6.73	6.84	5.86	7.59
C17:0	0.69	0.95	1.09	0.96	1.01	0.85	0.81	1.06	0.91	1.24	1.43	0.74
C18:0	3.41	3.68	5.03	4.99	4.95	4.17	4.44	4.99	4.41	5.38	6.90	4.28
C18:1n12t	nd	nd	nd	nd	0.31	nd	0.23	nd	nd	nd	0.27	0.18
C18:1n9t	0.25	0.29	0.28	0.21	nd	0.30	0.19	0.24	0.19	0.24	nd	0.23
C18:1n12	nd	nd	nd	nd	nd	0.14	nd	nd	nd	nd	nd	nd
C18:1n9	14.26	13.51	20.89	20.29	19.49	19.49	20.37	19.80	20.89	21.12	16.23	21.35
C18:1n7	3.00	3.16	4.35	3.85	4.07	3.69	3.79	4.01	3.81	4.24	3.87	3.87
C18:2n6t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.09	nd
C18:2n6	7.54	6.44	10.37	9.21	9.53	9.24	9.63	9.02	11.57	9.26	4.27	10.90
C18:3n6	0.54	0.35	0.36	0.30	0.56	nd	0.39	0.45	0.30	0.28	0.27	0.41
C19:0	0.62	0.67	0.50	0.25	0.58	0.86	nd	nd	0.37	0.40	0.45	0.06
C18:3n3	2.07	1.60	1.91	1.49	1.68	1.53	1.52	1.33	1.70	1.72	1.11	1.31
C18:4n3	1.32	1.08	0.98	0.69	0.92	0.83	0.79	0.63	0.73	0.77	0.80	0.54

Table B8 continued. Striped bass belly flap samples from day 40.

					Sample number							
	1	2	3	4	5	6	7	8	9	10	11	12
C19:2n6	nd	nd	0.21	0.10	nd	nd	0.08	nd	nd	0.15	0.24	nd
C20:0	0.74	0.76	0.28	0.28	0.30	0.67	0.29	0.23	0.36	0.42	0.44	0.30
C20:1n15	0.51	nd	nd	0.18	nd	0.45	nd	0.03	0.07	0.21	nd	0.10
C20:1n12	1.15	1.52	0.66	0.68	0.85	0.90	0.69	0.60	0.53	0.95	1.14	0.45
C20:1n9	1.96	1.64	1.35	1.29	1.97	1.55	1.89	1.28	1.57	1.42	1.49	1.34
C20:1n7	1.67	1.24	0.71	0.65	1.00	0.94	0.63	0.67	0.57	0.61	1.24	0.61
C20:2n6	2.11	1.70	0.81	0.70	1.06	1.52	0.92	0.70	0.80	0.70	0.67	0.71
C20:3n6	1.26	0.71	0.24	0.38	0.43	0.49	0.30	0.23	0.24	0.17	0.16	0.30
C21:0	0.43	nd	0.06	0.11	nd	0.16	nd	0.02	0.01	0.02	nd	0.10
C20:4n6	2.08	2.20	1.24	1.50	1.87	1.97	1.64	1.84	1.23	1.31	2.17	1.47
C20:3n3	1.84	1.12	0.45	0.28	0.31	0.81	0.21	0.28	0.30	0.42	0.26	0.17
C20:5n3	5.85	5.41	4.01	4.51	4.27	4.44	4.22	4.58	3.37	4.28	4.90	3.45
C22:0	0.85	1.35	0.28	0.37	0.38	0.30	0.21	0.26	0.28	0.16	0.16	0.25
C22:1n9	1.57	1.15	0.22	0.28	0.43	0.44	0.12	0.16	0.28	0.19	0.11	0.27
C22:2n6	1.05	1.83	0.06	nd	0.19	0.22	0.08	0.16	0.51	0.02	0.20	0.09
C23:0	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.11
C22:4n6	1.61	2.15	0.41	0.71	0.26	nd	0.54	0.45	0.86	0.35	0.80	0.70
C22:3n3 + C22:5n6	2.51	2.82	0.42	0.56	0.70	0.62	0.54	0.33	1.22	0.47	0.64	0.49
C22:5n3	4.85	4.48	1.32	1.60	1.38	1.34	1.81	1.48	2.37	1.18	1.89	1.39
C24:0 + C22:6n3	9.34	10.47	5.31	7.13	7.58	6.16	7.79	8.35	7.30	6.23	10.65	6.41
C24:1n9	1.55	3.42	0.34	0.73	0.32	0.24	0.54	0.35	1.20	0.25	0.42	1.06

Table B9. Striped bass liver samples from day 71.

	Sample number			
	1	2	3	4
Total length (mm)	371	360	340	372
Weight (g)	827.1	778.4	581.3	795.5
Lipid content (%)	12.54	7.30	8.08	11.81
C12:0	0.03	0.02	0.03	0.02
C12:1n1	nd	nd	0.26	nd
C14:0	1.83	1.37	2.69	1.93
C14:1n5	0.08	0.06	0.09	0.08
C15:0	0.17	0.12	0.28	0.16
C15:1n5	0.01	0.05	nd	0.01
C16:0 DMA	0.01	0.05	0.02	0.02
C16:0	23.34	18.24	28.06	25.11
C16:1n7	4.38	2.96	4.58	4.28
C17:0	0.42	0.50	0.63	0.39
C18:0	6.31	6.04	6.14	7.05
C18:1n12t	0.13	nd	0.14	nd
C18:1n9t	0.33	0.41	0.27	0.31
C18:1n9	35.43	26.65	23.77	33.97
C18:1n7	4.78	3.40	4.47	4.40
C18:2n6	0.85	1.27	1.31	0.81
C18:3n6	0.20	0.62	0.23	0.20
C18:3n3	0.57	1.01	0.60	0.60
C18:4n3	0.74	1.69	0.74	0.84
C20:0	0.12	0.84	0.14	0.13
C20:1n15	nd	0.16	nd	nd
C20:1n12	0.08	nd	0.09	0.09

Table B9 continued. Striped bass liver samples from day 71.

	Sample number			
	1	2	3	4
C20:1n9	2.39	2.43	2.34	2.62
C20:1n7	0.20	1.07	0.22	0.21
C20:2n6	0.19	1.16	0.32	0.20
C20:3n6	0.09	1.05	0.12	0.09
C20:4n6	1.17	2.66	1.71	1.15
C20:3n3	0.13	0.74	0.14	0.12
C20:5n3	4.65	4.58	4.96	4.18
C22:0	0.21	1.20	0.25	0.18
C22:1n9	0.16	1.34	0.16	0.16
C22:2n6	0.02	0.59	0.03	0.01
C22:4n6	nd	0.84	0.17	nd
C22:3n3 + C22:5n6	0.51	2.89	0.51	0.57
C22:5n3	0.74	3.17	0.93	0.76
C24:0 + C22:6n3	9.40	8.31	13.16	9.00
C24:1n9	0.33	2.51	0.45	0.32

Table B10. Striped bass belly flap samples from day 71.

	Sample number			
	1	2	3	4
Total length (mm)	371	360	340	372
Weight (g)	827.1	778.4	581.3	795.5
Lipid content (%)	23.32	34.14	13.67	17.62
C12:0	0.13	0.14	0.09	0.11
C12:1n1	nd	nd	0.87	nd
C14:0	5.86	5.86	6.91	6.02
C14:1n5	0.24	0.17	0.21	0.19
C15:0	0.76	0.89	0.85	0.79
C15:1n5	0.17	0.20	0.06	nd
C16:0 DMA	0.09	nd	nd	nd
C16:0	23.35	22.74	24.10	23.33
C16:1n7	8.04	7.82	8.15	7.74
C17:0	0.88	0.98	0.90	0.99
C18:0	4.66	4.72	4.45	4.83
C18:1n12t	0.30	0.25	0.16	0.21
C18:1n9t	0.11	0.26	0.19	0.24
C18:1n9	15.06	15.59	14.77	14.89
C18:1n7	3.79	4.18	3.91	4.03
C18:2n6	5.01	4.21	5.98	4.27
C18:3n6	0.50	0.58	0.43	0.33
C18:3n3	2.07	2.21	1.80	1.93
C18:4n3	1.92	1.94	1.60	1.94
C19:2n6	nd	0.14	nd	nd
C20:0	0.39	0.35	0.22	0.36
C20:1n15	nd	0.05	0.02	nd
C20:1n12	nd	0.40	0.24	0.49
C20:1n9	1.29	1.28	1.24	1.35

Table B10 continued. Striped bass belly flap samples from day 71.

	Sample number			
	1	2	3	4
C20:1n7	0.50	0.72	0.41	0.76
C20:2n6	0.46	0.55	0.52	0.51
C20:3n6	0.28	0.50	0.29	0.35
C21:0	0.13	nd	nd	nd
C20:4n6	1.45	1.59	1.10	1.33
C20:3n3	0.42	0.25	0.36	0.51
C20:5n3	6.69	6.43	5.63	6.41
C22:0	0.23	0.39	0.35	0.43
C22:1n9	nd	0.17	0.16	0.19
C22:2n6	0.07	0.10	0.08	nd
C22:4n6	nd	nd	0.32	0.55
C22:3n3 + C22:5n6	0.90	0.89	0.82	1.04
C22:5n3	1.73	1.94	1.68	1.82
C24:0 + C22:6n3	12.01	11.04	10.57	11.44
C24:1n9	0.46	0.47	0.57	0.61

Table B11. Striped bass liver samples from day 88.

	Sample number			
	1	2	3	4
Total length (mm)	407	394	374	387
Weight (g)	1006.3	1010.8	696.1	942.6
Lipid content (%)	9.90	11.72	8.72	11.14
C12:0	0.03	0.03	nd	0.03
C12:1n1	nd	nd	0.04	nd
C13:0	nd	nd	0.06	nd
C14:0	2.22	1.96	2.94	1.86
C14:1n5	0.10	0.11	0.13	0.08
C15:0	0.26	0.22	0.38	0.17
C15:1n5	nd	nd	nd	0.01
C16:0 DMA	0.02	0.02	nd	0.01
C16:0	26.09	23.97	18.88	21.36
C16:1n7	5.14	5.34	7.13	4.97
C17:0	0.55	0.44	0.54	0.37
C18:0	5.73	5.61	3.58	4.90
C18:1n12t	0.15	0.13	nd	0.15
C18:1n9t	0.33	0.44	0.27	0.28
C18:1n9	24.87	30.45	24.60	35.60
C18:1n7	4.90	4.83	5.58	4.99
C18:2n6	1.06	0.96	1.73	0.74
C18:3n6	0.25	0.22	0.31	0.18
C18:3n3	0.93	0.89	1.29	0.64
C18:4n3	0.93	1.01	1.29	0.86
C20:0	0.18	0.19	0.16	0.16
C20:1n12	0.10	0.12	0.17	0.10
C20:1n9	3.52	3.91	3.43	3.12
C20:1n7	0.29	0.28	0.41	0.25

Table B11 continued. Striped bass liver samples from day 88.

	Sample number			
	1	2	3	4
C20:2n6	0.30	0.28	0.46	0.23
C20:3n6	0.09	0.09	0.13	0.10
C21:0	nd	nd	0.01	nd
C20:4n6	1.23	1.12	1.55	1.14
C20:3n3	0.20	0.18	0.31	0.14
C20:5n3	5.30	4.94	6.58	4.50
C22:0	0.32	0.32	0.30	0.17
C22:1n9	0.25	0.26	0.23	0.16
C22:2n6	0.02	0.03	0.02	0.03
C22:3n3 + C22:5n6	0.69	0.54	0.77	0.58
C22:5n3	1.11	0.84	1.38	1.05
C24:0 + C22:6n3	12.36	9.84	14.76	10.29
C24:1n9	0.48	0.44	0.58	0.76

Table B12. Striped bass belly flap samples from day 88.

	Sample number			
	1	2	3	4
Total length (mm)	407	394	374	387
Weight (g)	1006.3	1010.8	696.1	942.6
Lipid content (%)	18.62	25.67	14.40	16.88
C12:0	0.09	0.16	0.10	0.11
C12:1n1	0.62	nd	nd	nd
C13:0	0.09	0.11	nd	0.13
C14:0	6.71	6.13	5.65	6.59
C14:1n5	0.22	0.25	0.20	0.19
C15:0	0.83	0.86	0.80	0.90
C15:1n5	nd	0.06	nd	0.01
C16:0 DMA	0.03	0.08	nd	nd
C16:0	24.48	23.93	23.64	24.66
C16:1n7	7.77	7.98	7.79	8.15
C17:0	0.85	0.87	0.89	1.03
C18:0	4.34	4.24	4.78	4.58
C18:1n12t	0.14	0.20	0.19	0.20
C18:1n9t	0.19	0.20	0.23	nd
C18:1n7t	0.08	nd	0.08	nd
C18:1n9	13.48	15.36	15.08	14.24
C18:1n7	3.86	3.89	3.98	4.15
C18:2n6	3.88	4.23	4.49	4.19
C18:3n6	0.56	0.35	0.37	0.43
C18:3n3	2.31	2.30	1.91	2.17
C18:4n3	2.11	2.22	1.59	2.00
C20:0	0.31	0.27	0.27	0.28
C20:1n15	nd	0.06	0.06	0.06
C20:1n12	0.32	0.31	0.40	0.36

Table B12 continued. Striped bass belly flap samples from day 88.

	Sample number			
	1	2	3	4
C20:1n9	1.37	1.41	1.37	1.30
C20:1n7	0.36	0.45	0.50	0.55
C20:2n6	0.54	0.53	0.61	0.50
C20:3n6	0.20	0.25	0.29	0.30
C21:0	0.07	nd	nd	nd
C20:4n6	1.06	1.07	1.32	1.01
C20:3n3	0.50	0.46	0.37	0.39
C20:5n3	6.31	6.09	6.31	6.27
C22:0	0.37	0.47	0.36	0.19
C22:1n9	0.17	0.18	0.16	nd
C22:2n6	0.07	nd	0.05	0.14
C22:4n6	0.42	0.38	0.59	0.47
C22:3n3 + C22:5n6	0.98	1.07	0.99	0.83
C22:5n3	1.80	1.84	1.86	1.81
C24:0 + C22:6n3	11.97	11.08	12.16	11.24
C24:1n9	0.56	0.67	0.57	0.57

Chapter 4 Raw Data

Values for length (mm), weight (g), lipid content (% wet weight) and fatty acids routinely identified for striped bass belly flap samples analyzed in Chapter 4 of this experiment. Small, medium and large refers to the size class of striped bass.

Table C1. Striped bass belly flap samples from day 0.

| | Sample number | | | | | | | | | | | |
| | Small | | | | Medium | | | | Large | | | |
	1	2	3	4	1	2	3	4	1	2	3	4
Total length (mm)	163	176	167	164	312	302	282	318	379	388	368	420
Weight (g)	74	92	82	78	528	560	508	750	996	1108	870	1538
Lipid content (%)	4.75	6.10	5.83	6.31	1.98	8.16	15.93	15.68	13.49	2.43	5.58	12.58
C12:0	0.06	0.24	0.13	0.15	0.10	0.10	0.02	0.01	0.15	nd	0.11	nd
C13:0	0.39	0.41	0.48	0.34	0.49	0.22	0.14	0.19	0.35	1.49	0.27	nd
C14:0	3.90	5.15	4.35	4.04	3.52	3.58	3.89	3.75	4.54	4.59	3.43	3.94
C14:1n5	0.15	0.10	0.17	0.12	0.07	0.12	0.18	0.18	0.20	nd	0.15	nd
C15:0	1.19	1.55	1.56	1.19	1.04	1.20	1.27	1.24	1.39	1.49	1.13	1.19
C16:0 DMA	0.44	0.31	0.37	0.44	0.64	0.25	0.28	0.26	0.24	0.56	0.37	0.49
C16:0	26.62	26.37	27.08	26.00	27.05	27.91	25.85	27.63	25.70	30.21	26.81	25.05
C16:1n7	6.54	7.90	7.29	6.28	6.12	6.30	7.04	6.91	8.07	6.91	7.10	8.13
C17:0	1.41	1.31	1.27	1.11	1.10	1.18	1.06	1.19	1.20	1.37	1.04	1.08
C18:0	7.04	5.84	6.13	6.61	7.02	5.67	5.57	6.10	6.07	7.86	5.93	6.51
C18:1n12t	1.03	0.96	0.99	0.56	0.96	0.51	0.48	0.54	0.60	0.77	0.60	0.69
C18:1n9t	nd	nd	nd	nd	nd	0.40	nd	0.38	nd	nd	0.41	nd
C18:1n7t	0.16	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	14.81	13.94	14.27	14.32	12.25	16.28	17.30	17.30	17.44	15.13	17.86	18.84
C18:1n7	4.68	4.29	3.99	3.94	3.58	3.80	4.03	4.11	4.08	3.63	4.19	4.37
C18:2n6	2.72	2.23	1.87	3.27	1.34	1.43	1.81	1.69	1.55	1.12	1.32	3.01
C18:3n6	nd	nd	nd	nd	nd	0.11	nd	nd	nd	nd	0.11	nd
C18:3n3	0.89	1.11	0.91	0.76	0.77	0.87	0.97	0.93	0.93	0.77	0.90	1.42
C18:4n3	0.50	0.80	0.63	0.81	0.47	0.59	0.52	0.54	0.60	0.47	0.47	0.68

Table C1 continued. Striped bass belly flap samples from day 0.

	Sample number											
	Small				Medium				Large			
	1	2	3	4	1	2	3	4	1	2	3	4
C20:0	1.18	1.12	1.14	1.13	1.17	1.09	1.23	1.20	1.32	1.36	1.34	1.24
C20:1n15	nd	nd	0.08	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:2n6	0.75	0.71	0.53	0.65	0.60	0.59	0.58	0.59	0.75	0.68	0.55	0.57
C20:3n6	0.13	0.12	0.10	nd	0.10	0.10	0.16	0.13	0.13	nd	0.12	nd
C20:4n6	2.36	2.28	2.60	2.21	2.66	1.84	2.01	1.94	2.40	2.22	2.45	2.13
C20:3n3	0.45	0.31	0.45	0.35	0.33	0.35	0.47	0.36	0.38	0.27	0.20	0.32
C20:5n3	4.61	5.58	5.41	5.49	5.29	4.44	4.99	4.90	4.85	3.80	4.42	3.62
C22:0	1.43	1.81	1.77	2.07	2.25	1.99	1.84	1.90	1.78	1.60	1.87	1.63
C22:1n9	0.22	0.19	0.18	0.07	0.24	0.26	0.23	0.22	nd	nd	nd	nd
C22:2n6	nd	nd	nd	nd	nd	0.06	nd	nd	nd	nd	nd	nd
C22:4n6	0.59	0.69	0.61	0.60	0.06	0.55	0.57	0.63	0.87	0.51	0.72	1.04
C22:3n3 + C22:5n6	0.62	0.87	0.68	0.76	0.08	0.71	0.62	0.71	0.62	0.58	0.82	0.62
C22:5n3	1.95	2.02	2.00	1.88	2.44	2.55	2.47	2.29	0.07	2.11	2.22	2.67
C22:6n3 + C24:0	10.59	10.13	10.85	12.78	15.96	12.14	11.30	9.59	9.86	7.56	9.74	7.94
C24:1n9	0.35	0.30	0.32	0.28	0.60	0.44	0.45	0.23	0.26	0.22	0.18	nd

Table C2. Striped bass belly flap samples from day 13.

	Sample number											
	Small				Medium				Large			
	1	2	3	4	1	2	3	4	1	2	3	4
Total length (mm)	170	168	166	216	299	312	331	304	455	413	365	407
Weight (g)	92.1	100.6	96.4	246.4	504.5	664.7	722.6	590.9	1814.2	1344.0	994.2	1154.1
Lipid content (%)	5.92	2.74	3.47	11.88	11.08	7.44	4.98	5.07	7.55	2.49	3.98	1.64
C12:0	0.12	0.12	nd	nd	nd	0.11	nd	0.12	0.17	nd	0.54	nd
C13:0	0.21	0.24	nd	nd	nd	0.19	0.44	nd	0.34	nd	nd	nd
C14:0	4.13	4.31	4.51	4.77	4.23	3.72	4.18	3.85	5.27	4.01	3.14	3.46
C14:1n5	0.15	0.11	nd	nd	0.26	0.15	0.20	0.16	0.49	1.15	nd	0.21
C15:0	1.40	1.26	1.19	1.72	1.46	1.23	1.28	1.32	1.66	1.78	1.30	1.01
C16:0 DMA	0.33	0.32	0.50	1.62	0.49	0.47	0.25	0.44	0.66	nd	0.99	0.90
C16:0	27.63	30.01	28.18	22.75	26.62	27.90	26.95	29.61	23.95	21.26	26.44	30.10
C16:1n7	7.06	6.88	6.83	6.41	7.14	6.84	6.39	6.11	8.01	4.24	4.32	5.18
C17:0	1.40	1.25	1.30	1.96	1.30	1.17	1.40	1.35	1.39	3.77	1.67	1.17
C18:0	7.12	7.46	8.00	6.39	6.26	6.17	6.69	7.10	6.41	11.86	7.87	10.26
C18:1n12t	0.49	0.49	0.83	1.46	0.55	0.45	0.54	0.55	0.51	nd	0.60	0.77
C18:1n9t	nd	nd	nd	nd	0.37	nd	nd	nd	nd	nd	nd	nd
C18:1n7t	nd	nd	nd	0.56	nd	0.13	nd	nd	nd	nd	nd	nd
C18:1n9	15.85	14.89	17.16	14.89	16.67	16.76	15.04	16.51	15.14	9.25	13.07	14.12
C18:1n7	3.62	3.44	3.64	3.41	3.56	3.49	3.51	3.66	3.69	4.65	2.52	4.77
C18:2n6t	nd	nd	nd	1.54	nd	nd	nd	nd	nd	nd	nd	nd
C18:2n6	1.95	1.80	3.37	2.37	1.96	1.89	1.54	2.34	1.64	6.37	0.96	0.87
C18:3n6	0.11	0.14	nd	nd	0.11	0.16	0.20	nd	0.18	nd	nd	nd
C18:3n3	1.07	0.93	0.99	0.90	0.88	1.01	0.77	1.18	1.21	nd	1.58	0.43
C18:4n3	0.60	0.62	0.38	1.66	0.91	0.68	0.55	0.74	1.18	nd	1.22	0.51
C19:2n6	0.06	nd	nd	nd	nd	0.06	nd	nd	0.24	1.87	nd	0.20
C20:0	1.38	1.23	1.71	1.40	1.40	1.26	1.10	1.40	1.85	3.26	1.41	1.29
C20:1n15	nd	0.09	nd	0.81	nd	0.09	0.09	nd	nd	nd	nd	nd

254

Table C2 continued. Striped bass belly flap samples from day 13.

	Sample number											
	Small				Medium				Large			
	1	2	3	4	1	2	3	4	1	2	3	4
C20:1n9	1.82	1.64	1.98	1.95	1.76	1.64	1.65	1.74	2.28	nd	1.92	2.40
C20:1n7	2.07	1.43	1.67	1.75	1.39	1.29	1.58	1.25	2.17	nd	1.76	1.23
C20:2n6	0.75	0.59	0.96	2.11	0.87	0.67	0.76	0.75	0.91	nd	0.88	0.68
C20:3n6	0.16	0.14	nd	nd	0.22	0.30	0.20	0.42	0.17	3.44	0.42	nd
C21:0	nd	nd	nd	nd	nd	nd	nd	0.27	nd	4.05	nd	nd
C20:4n6	2.42	2.20	2.29	2.14	2.34	2.16	1.83	1.87	2.20	nd	2.48	2.43
C20:3n3	0.47	0.31	nd	0.58	0.38	0.37	0.38	0.40	0.70	nd	0.80	0.37
C20:5n3	4.09	3.80	3.16	4.13	4.59	4.31	2.82	3.42	3.91	nd	2.67	2.58
C22:0	1.70	1.67	1.48	1.87	1.84	1.85	1.62	1.65	1.93	nd	1.85	1.82
C22:1n9	0.27	0.19	nd	0.67	0.21	0.12	0.43	0.40	0.42	1.58	0.37	0.41
C22:2n6	nd	nd	nd	0.46	0.10	0.05	0.45	0.09	0.11	2.88	nd	0.09
C23:0	0.02	nd	nd	0.47	0.16	0.03	0.32	0.23	nd	nd	nd	nd
C22:4n6	0.66	0.53	0.67	nd	0.62	0.55	1.29	0.58	nd	7.84	1.62	0.76
C22:3n3 + C22:5n6	nd	nd	0.51	nd	0.68	0.69	2.12	0.89	0.66	1.75	nd	0.92
C22:5n3	1.99	2.11	1.58	2.39	1.87	1.83	2.41	1.35	2.96	nd	9.13	1.35
C24:0 + C22:6n3	8.51	9.44	7.11	6.43	8.29	9.93	9.43	7.81	6.71	2.04	8.46	9.25
C24:1n9	0.38	0.38	nd	0.45	0.50	0.29	1.58	0.44	0.87	2.94	nd	0.45

Table C3.Small and medium striped bass belly flap samples from day 28.

	Sample number											
	Small						Medium					
	1	2	3	4	5	6	1	2	3	4	5	6
Total length (mm)	175	174	174	182	176	174	308	350	318	318	270	310
Weight (g)	106.2	96.4	116.1	118.8	110.7	102.7	648.0	702.5	690.4	530.3	384.8	578.4
Lipid content (%)	9.34	12.12	3.54	5.47	5.08	2.71	24.66	1.37	6.38	1.72	3.91	10.13
C12:0	nd	0.09	2.08	nd	0.21	0.24	nd	0.05	0.15	0.10	0.20	0.10
C12:1n1	nd	nd	1.12	nd	nd	0.20	nd	nd	nd	nd	nd	nd
C13:0	nd	1.15	nd	nd	1.14	nd	0.81	nd	0.75	1.15	nd	0.49
C14:0	5.00	4.38	0.85	2.25	2.47	3.56	3.95	2.12	3.88	3.47	3.05	3.93
C14:1n5	nd	0.17	nd	0.20	0.04	0.11	0.46	0.09	0.10	0.17	0.12	0.17
C15:0	1.90	1.19	1.27	1.05	0.90	1.16	1.89	0.52	1.18	1.19	1.24	1.38
C15:1n5	nd	nd	nd	nd	nd	0.25	nd	nd	nd	nd	nd	nd
C16:0 DMA	nd	0.23	0.72	0.45	0.52	0.81	0.30	0.79	0.41	0.67	0.76	0.26
C16:0	19.45	31.91	31.47	15.17	16.97	27.99	26.23	18.56	28.95	30.47	25.37	28.04
C16:1n7	3.64	7.54	4.66	3.85	4.19	6.48	7.39	3.73	7.36	6.01	5.79	6.66
C17:0	1.25	1.15	1.52	1.19	0.81	1.20	1.22	0.62	1.21	1.22	1.13	1.40
C17:1n7	nd	nd	nd	0.15	nd	0.19	nd	nd	nd	nd	nd	nd
C18:0	7.23	7.20	9.73	3.85	4.65	7.55	6.90	6.05	6.56	8.28	6.67	6.28
C18:1n12t	nd	0.37	1.34	0.41	0.23	0.59	0.57	0.65	0.53	0.61	0.62	0.75
C18:1n9t	0.69	0.47	nd	nd	0.29	0.52	nd	0.22	0.44	0.27	0.23	nd
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.11	nd	0.12
C18:1n9	7.98	18.70	11.71	7.86	10.32	14.94	20.04	11.31	17.33	15.66	15.55	15.73
C18:1n7	4.15	3.89	11.71	2.19	2.47	3.64	5.07	3.81	3.94	3.81	3.23	3.68
C18:2n6t	2.87	nd	0.35	0.22	nd	0.19	nd	nd	nd	nd	nd	nd
C18:2n6	1.78	1.61	0.97	1.50	1.24	2.13	2.59	0.40	1.46	1.42	1.59	1.68
C18:3n6	nd	0.14	0.36	0.62	0.32	nd	0.21	nd	0.16	0.12	nd	0.15
C19:0	nd	nd	nd	nd	nd	0.44	nd	nd	nd	nd	nd	nd
C18:3n3	nd	0.73	0.80	0.90	0.86	0.85	1.03	0.26	0.89	0.75	0.96	0.99

Table C3 continued. Small and medium striped bass belly flap samples from day 28.

	Sample number											
	Small						Medium					
	1	2	3	4	5	6	1	2	3	4	5	6
C18:4n3	3.77	0.61	0.95	0.68	0.82	1.00	1.10	0.16	0.62	0.35	0.72	0.75
C19:2n6	1.84	0.06	nd	0.50	nd	nd	nd	0.07	0.07	0.09	0.11	0.04
C20:0	5.41	1.03	1.81	1.64	0.77	1.64	1.55	1.04	1.21	1.39	1.26	1.26
C20:1n15	1.45	0.07	nd	0.38	0.26	nd	0.39	0.15	0.09	nd	nd	0.10
C20:1n12	4.88	nd	nd	1.55	nd	nd	nd	nd	nd	nd	nd	nd
C20:1n9	2.14	2.17	2.08	1.90	1.97	2.09	2.15	2.34	1.87	1.67	1.46	1.87
C20:1n7	0.58	1.00	1.47	1.38	1.32	1.62	1.84	0.98	1.37	1.31	1.30	1.35
C20:2n6	0.61	0.04	nd	1.47	1.19	0.65	1.08	0.63	0.69	0.59	0.86	0.76
C20:3n6	2.64	0.11	nd	1.41	0.86	0.46	nd	0.37	0.12	0.19	0.21	0.15
C21:0	1.03	0.05	0.26	0.87	0.26	nd	nd	nd	nd	nd	0.30	nd
C20:4n6	nd	1.32	1.28	1.97	2.25	2.09	2.29	2.13	2.08	2.50	3.16	1.84
C20:3n3	nd	0.25	nd	1.13	0.89	0.31	0.36	0.49	0.28	0.39	0.45	0.35
C20:5n3	nd	2.56	1.57	4.18	4.73	2.89	3.49	3.74	3.50	2.71	4.13	4.03
C22:0	8.67	1.06	1.82	3.45	2.48	1.62	1.48	3.12	1.49	1.57	2.17	1.81
C22:1n9	2.97	0.27	0.40	1.96	2.19	0.37	0.40	1.32	0.23	0.24	0.44	0.25
C22:2n6	0.91	0.06	0.86	nd	1.74	0.26	0.64	nd	0.20	0.07	0.12	nd
C23:0	nd	0.06	nd	3.24	nd	nd	nd	nd	nd	nd	0.28	nd
C22:4n6	1.56	0.39	0.85	3.83	2.91	0.61	0.74	2.22	0.56	0.48	0.90	0.51
C22:3n3 + C22:5n6	nd	0.40	0.65	3.42	3.94	0.74	1.24	4.46	0.55	0.76	1.07	0.67
C22:5n3	2.37	1.10	1.04	7.27	7.27	1.28	2.06	5.34	1.64	1.23	2.04	2.16
C24:0 + C22:6n3	2.33	6.17	4.32	15.93	13.78	8.71	0.54	16.99	7.92	8.99	12.50	9.93
C24:1n9	0.92	0.31	nd	nd	2.74	0.61	nd	5.26	0.22	0.30	nd	0.37

Table C4. Large striped bass belly flap samples from day 28.

| | Sample number | | | | | |
| | Large | | | | | |
	1	2	3	4	5	6
Total length (mm)	409	382	407	391	430	410
Weight (g)	1340.1	1230.3	1192.9	1168.6	1310.8	1296.0
Lipid content (%)	11.44	8.20	2.10	19.95	0.72	3.07
C12:0	0.15	0.12	nd	0.16	0.07	0.08
C12:1n1	nd	nd	nd	nd	nd	nd
C13:0	0.52	0.41	32.71	0.34	0.34	0.13
C14:0	3.75	5.93	nd	4.41	3.20	2.55
C14:1n5	0.15	0.22	nd	0.15	0.20	0.10
C15:0	1.12	1.27	nd	1.27	0.73	0.81
C15:1n5	nd	nd	nd	0.07	nd	nd
C16:0 DMA	0.30	0.33	1.56	0.33	0.48	0.60
C16:0	27.20	26.69	8.49	25.45	26.30	27.13
C16:1n7	7.72	8.70	nd	7.63	8.44	5.78
C17:0	1.07	1.13	1.69	1.20	0.69	0.87
C17:1n7	0.09	nd	nd	nd	nd	nd
C18:0	6.87	5.94	4.99	6.34	6.38	6.69
C18:1n12t	0.58	0.46	nd	0.59	0.58	0.57
C18:1n9t	0.39	0.26	6.62	0.33	0.35	0.33
C18:1n7t	nd	nd	1.15	nd	nd	nd
C18:1n9	22.26	14.06	5.23	17.10	20.23	14.80
C18:1n7	4.61	3.97	nd	4.28	4.43	3.64
C18:2n6t	nd	nd	nd	nd	nd	nd
C18:2n6	1.92	1.85	3.53	1.71	0.98	1.20
C18:3n6	0.12	0.17	1.06	0.20	0.08	0.11
C19:0	nd	nd	2.47	0.07	nd	nd
C18:3n3	0.82	1.20	nd	1.13	0.46	0.67

Table C4 continued. Large striped bass belly flap samples from day 28.

	Sample number					
	Large					
	1	2	3	4	5	6
C18:4n3	0.36	1.23	2.33	1.14	0.23	0.60
C19:2n6	0.02	nd	3.86	0.10	0.07	0.09
C20:0	1.20	1.15	2.83	1.35	1.07	1.12
C20:1n15	0.08	0.10	2.42	nd	0.10	nd
C20:1n12	nd	nd	nd	nd	nd	nd
C20:1n9	2.47	1.64	nd	2.28	2.50	1.78
C20:1n7	1.94	1.08	1.62	1.44	1.64	0.81
C20:2n6	0.75	0.59	1.13	0.74	0.61	0.54
C20:3n6	0.17	0.17	nd	0.46	0.12	0.11
C21:0	nd	nd	nd	nd	nd	nd
C20:4n6	1.51	1.75	nd	1.78	2.34	2.50
C20:3n3	0.27	0.29	3.14	0.39	0.15	0.24
C20:5n3	2.19	4.91	2.33	3.85	2.66	4.90
C22:0	1.18	1.98	3.31	1.84	1.70	2.22
C22:1n9	0.29	0.26	0.58	0.31	0.29	0.21
C22:2n6	0.07	0.08	1.52	0.24	0.10	0.08
C23:0	nd	nd	nd	nd	nd	0.01
C22:4n6	0.61	0.50	1.74	0.73	0.75	0.47
C22:3n3 + C22:5n6	0.59	0.58	nd	0.66	0.65	0.77
C22:5n3	1.56	0.05	nd	1.98	2.51	2.33
C24:0 + C22:6n3	4.90	10.45	3.68	7.95	8.17	14.70
C24:1n9	0.35	0.50	nd	0.56	0.40	0.44

Chapter 5 Raw Data

Values for length (mm), weight (g), lipid content (% wet weight) and fatty acids routinely identified for striped bass liver and belly flap samples analyzed in Chapter 5 of this experiment. Mixed diets shown are a homogenous mixture of spot and menhaden samples and do not have a length measurement.

Table D1. Menhaden and spot (70:30 by weight) diet mixture.

	Sample number							
	1	2	3	4	6	7	8	9
Weight (g)	6.34	4.80	5.76	5.44	5.72	5.38	4.55	4.37
Lipid content (%)	6.58	6.29	9.76	6.18	6.48	6.14	5.67	5.41
C12:0	0.24	0.11	0.10	0.13	0.12	0.12	0.10	0.10
C12:1n1	0.04	0.02	0.02	0.03	0.00	0.03	0.02	0.02
C13:0	0.44	0.30	0.22	0.24	0.29	0.22	0.24	0.20
C14:0	10.12	5.62	5.47	5.41	5.56	6.02	6.89	5.81
C14:1n5	0.21	0.06	0.12	0.08	0.08	0.07	0.11	0.08
C15:0	3.32	1.44	1.62	1.77	1.84	1.57	1.82	1.64
C16:0	40.14	31.52	30.30	30.53	29.25	22.75	22.23	22.05
C16:1n7	12.57	6.30	7.23	6.74	7.01	6.97	7.61	6.82
C17:0	4.97	2.23	2.42	2.52	2.82	2.24	2.70	2.79
C18:0	17.61	8.83	8.24	9.12	9.31	7.41	9.86	9.58
C18:1n9	20.44	10.10	11.37	10.18	10.70	8.03	10.04	11.76
C18:1n7	7.38	3.75	3.61	3.77	3.67	3.33	4.30	3.96
C18:2n6	3.35	1.71	1.65	1.77	1.64	1.66	1.66	2.04
C18:3n6	0.31	0.12	0.15	0.16	0.14	0.27	0.21	0.18
C19:0	0.96	0.39	0.47	0.50	0.53	0.60	0.55	0.56
C18:3n3	3.30	2.12	1.35	1.85	1.28	1.66	1.35	2.20
C18:4n3	2.60	1.64	1.09	1.25	0.88	1.35	0.98	1.47
C19:2n6	0.00	0.04	0.06	0.00	0.00	0.00	0.00	0.10
C20:0	0.82	0.43	0.39	0.43	0.47	0.65	0.47	0.49
C20:1n15	0.12	0.00	0.06	0.03	0.05	0.07	0.00	0.04

Table D1 continued. Menhaden and spot (70:30 by weight) diet mixture.

	Sample number							
	1	2	3	4	6	7	8	9
C20:1n12	3.99	1.44	2.32	2.03	2.43	1.98	2.08	2.14
C20:1n9	2.06	0.92	0.98	1.04	0.95	1.11	1.22	1.18
C20:1n7	3.69	1.26	2.10	1.93	2.16	2.19	1.92	2.07
C20:2n6	1.42	0.61	0.69	0.75	0.00	1.10	0.75	0.71
C20:3n6	0.25	0.13	0.14	0.13	0.13	0.37	0.17	0.14
C20:4n6	2.96	1.46	1.48	1.53	1.86	2.02	1.69	1.72
C20:3n3	0.90	0.46	0.38	0.40	0.41	1.04	0.39	0.45
C20:5n3	9.77	4.77	4.57	4.57	5.09	6.11	5.79	5.45
C22:0	0.59	0.23	0.26	0.29	0.34	0.84	0.37	0.43
C22:1n9	0.32	0.13	0.19	0.14	0.15	0.71	0.21	0.29
C22:2n6	0.11	0.04	0.00	0.03	0.05	0.60	0.06	0.09
C22:4n6	1.00	0.35	0.47	0.49	0.66	1.43	0.55	0.51
C22:3n3 + C22:5n6	1.46	0.66	0.66	0.71	1.13	1.87	0.94	0.77
C22:5n3	2.90	1.27	1.32	1.29	1.48	2.73	1.94	1.60
C24:0 + C22:6n3	16.89	8.99	8.05	7.78	7.13	9.33	10.24	10.08
C24:1n9	0.88	0.56	0.44	0.42	0.41	1.56	0.56	0.48

Table D2. Menhaden and spot (30:70 by weight) diet mixture.

	Sample number						
	1	2	3	4	5	6	7
Weight (g)	4.39	3.62	4.44	4.27	3.96	4.31	4.49
Lipid content (%)	10.67	10.09	4.81	4.74	8.57	8.19	5.94
C12:0	0.10	0.11	0.16	0.10	0.14	0.12	0.10
C12:1n1	0.03	0.07	0.05	0.02	0.04	0.04	0.03
C13:0	0.13	0.40	0.27	0.13	0.22	0.18	0.12
C14:0	8.63	3.12	10.62	8.73	10.38	7.07	9.79
C14:1n5	0.10	0.08	0.14	0.13	0.11	0.08	0.11
C15:0	1.09	0.57	1.44	1.11	1.71	1.14	1.19
C15:1n5	0.00	0.00	0.00	0.00	0.02	0.00	0.00
C16:0	26.08	18.44	32.97	32.30	23.69	32.98	28.60
C16:1n7	7.18	3.76	8.08	7.62	10.66	6.50	8.89
C17:0	1.58	1.32	1.93	1.67	2.16	2.01	1.52
C18:0	6.19	5.23	8.14	6.80	8.06	7.92	6.55
C18:1n12t	0.00	0.17	0.27	0.24	0.00	0.25	0.00
C18:1n9t	0.00	0.29	0.00	0.00	0.00	0.00	0.00
C18:1n9	6.40	7.16	7.95	8.40	7.69	8.23	6.80
C18:1n7	3.51	2.69	3.38	3.84	4.03	3.26	3.62
C18:2n6t	0.00	0.00	0.00	0.00	0.00	0.08	0.00
C18:2n6	1.72	1.47	1.60	1.52	1.71	1.88	1.48
C18:3n6	0.29	0.53	0.20	0.21	0.24	0.18	0.25
C19:0	0.44	0.92	0.35	0.33	0.40	0.37	0.30
C18:3n3	2.33	3.25	1.64	1.76	1.67	2.98	1.67
C18:4n3	2.18	1.60	1.49	1.40	1.47	2.26	1.73
C19:2n6	0.00	0.73	0.13	0.00	0.00	0.00	0.00
C20:0	0.58	0.79	0.54	0.30	0.37	0.36	0.30
C20:1n15	0.00	0.00	0.05	0.02	0.03	0.03	0.04
C20:1n12	1.05	1.06	0.68	0.48	1.10	0.57	0.67

262

Table D2 continued. Menhaden and spot (30:70 by weight) diet mixture.

	Sample number						
	1	2	3	4	5	6	7
C20:1n9	1.13	2.31	1.03	1.18	0.86	1.10	0.94
C20:1n7	1.07	1.43	0.93	0.66	1.21	0.69	0.84
C20:2n6	0.90	1.34	0.44	0.33	0.44	0.40	0.35
C20:3n6	0.30	0.94	0.11	0.12	0.14	0.10	0.18
C21:0	0.00	0.00	0.01	0.00	0.00	0.00	0.00
C20:4n6	1.05	1.62	0.74	0.69	1.35	0.75	1.06
C20:3n3	0.63	1.34	0.32	0.27	0.32	0.43	0.36
C20:5n3	6.55	6.46	5.08	6.62	6.49	4.64	7.09
C22:0	0.80	2.38	0.52	0.27	0.33	0.29	0.35
C22:1n9	0.70	2.86	0.37	0.21	0.23	0.16	0.31
C22:2n6	0.46	0.00	0.00	0.04	0.03	0.04	0.08
C22:4n6	1.19	2.46	0.16	0.15	0.29	0.17	0.32
C22:3n3 + C22:5n6	1.78	3.30	0.41	0.48	0.73	0.63	0.75
C22:5n3	2.23	3.61	0.78	1.49	1.23	1.16	1.53
C24:0 + C22:6n3	9.46	13.61	6.39	9.70	9.93	10.12	11.45
C24:1n9	2.14	2.53	0.64	0.64	0.51	0.81	0.63

Table D3. Liver and belly flap samples from striped bass fed the 70% menhaden 30% spot mixture from day 71.

	Liver				Belly flap			
	1	2	3	4	1	2	3	4
Length (mm)	329	360	371	362	-	-	-	-
Weight (g)	478.2	626.7	714.0	554.9	-	-	-	-
Lipid content (%)	6.42	5.14	8.63	9.30	7.93	8.63	9.30	6.55
C12:0	0.02	0.01	0.02	0.01	0.07	0.08	0.08	0.06
C12:1n1	0.00	0.00	0.00	0.13	0.21	0.34	0.76	0.21
C13:0	0.08	0.00	0.07	0.07	0.16	0.11	0.00	0.00
C14:0	3.07	2.79	3.28	3.47	5.40	5.26	5.15	5.64
C14:1n5	0.05	0.04	0.06	0.03	0.20	0.18	0.19	0.21
C15:0	0.66	0.69	0.70	0.75	0.86	0.88	0.79	0.77
C15:1n5	0.02	0.03	0.03	0.02	0.09	0.00	0.07	0.04
C16:0 DMA	0.04	0.04	0.05	0.04	0.06	0.03	0.05	0.03
C16:0	26.43	29.14	27.06	29.25	25.33	24.08	24.31	24.60
C16:1n7	4.54	3.32	4.36	4.00	7.24	7.13	7.37	7.66
C17:0	1.19	1.22	1.14	1.19	1.07	1.12	0.90	0.78
C18:0	7.52	9.12	7.63	8.94	5.46	5.69	4.98	4.75
C18:1n12t	0.25	0.00	0.26	0.00	0.00	0.00	0.24	0.19
C18:1n9t	0.24	0.21	0.18	0.00	0.21	0.00	0.18	0.18
C18:1n9	11.59	8.11	10.07	6.44	16.92	16.37	18.87	22.03
C18:1n7	4.14	3.18	3.30	2.94	4.15	4.14	3.97	4.02
C18:2n6	2.52	2.07	3.35	2.63	6.55	5.98	8.15	9.84
C18:3n6	0.40	0.38	0.36	0.37	0.41	0.43	0.44	0.39
C18:3n3	0.84	0.65	0.80	0.77	1.36	1.32	1.45	1.28
C18:4n3	0.49	0.37	0.46	0.39	0.84	0.89	0.85	0.64
C20:0	0.17	0.16	0.14	0.13	0.27	0.29	0.24	0.23
C20:1n15	0.00	0.00	0.00	0.00	0.01	0.00	0.02	0.01
C20:1n12	0.17	0.14	0.18	0.12	0.47	0.69	0.00	0.00
C20:1n9	1.61	1.20	1.41	0.71	1.37	1.40	1.45	1.46

Table D3 continued. Liver and belly flap samples from striped bass fed the 70% menhaden 30% spot mixture from day 71.

	Liver				Belly flap			
	1	2	3	4	1	2	3	4
C20:1n7	0.34	0.30	0.35	0.28	0.60	0.00	0.57	0.52
C20:2n6	0.53	0.50	0.62	0.54	0.59	0.67	0.66	0.66
C20:3n6	0.14	0.13	0.17	0.13	0.22	0.28	0.23	0.23
C20:4n6	3.20	4.06	3.85	3.66	1.63	1.83	1.39	1.21
C20:3n3	0.23	0.22	0.23	0.23	0.25	0.38	0.23	0.16
C20:5n3	6.14	6.85	6.89	5.27	4.78	5.50	4.77	3.26
C22:0	0.18	0.22	0.21	0.18	0.15	0.20	0.16	0.13
C22:1n9	0.18	0.15	0.15	0.10	0.19	0.13	0.09	0.12
C22:2n6	0.03	0.02	0.03	0.02	0.17	0.98	0.10	0.02
C22:4n6	0.51	0.49	0.54	0.31	0.49	0.75	0.59	0.44
C22:3n3 + C22:5n6	0.67	0.66	0.63	0.69	0.69	0.00	0.66	0.47
C22:5n3	1.44	1.23	1.40	1.32	1.60	1.98	1.43	1.23
C24:0 + C22:6n3	19.58	21.61	19.34	24.11	9.50	10.30	8.15	6.14
C24:1n9	0.78	0.69	0.69	0.76	0.45	0.59	0.42	0.39

Table D4. Liver and belly flap samples from striped bass fed the 30% menhaden 70% spot mixture from day 71.

	Liver		Belly flap		
	1	2	1	2	3
Length (mm)	331	385	-	-	-
Weight (g)	496.2	763.3	-	-	-
Lipid content (%)	6.25	6.73	9.70	10.51	-
C12:0	0.02	0.01	0.05	0.05	0.06
C12:1n1	0.01	0.00	0.00	0.03	0.00
C13:0	0.14	0.07	0.14	0.21	0.00
C14:0	3.02	2.68	4.89	5.02	5.16
C14:1n5	0.10	0.05	0.13	0.24	0.22
C15:0	0.75	0.57	1.08	0.80	0.88
C15:1n5	0.00	0.00	0.06	0.00	0.00
C16:0 DMA	0.00	0.00	0.03	0.00	0.05
C16:0	21.86	19.37	24.08	24.60	24.00
C16:1n7	4.76	4.42	6.81	7.82	7.30
C17:0	1.64	1.32	1.49	1.01	0.98
C18:0	6.82	6.20	6.39	4.88	5.29
C18:1n12t	0.00	0.00	0.00	0.00	0.29
C18:1n9t	0.33	0.32	0.12	0.20	0.22
C18:1n9	13.34	13.87	16.24	19.66	17.82
C18:1n7	4.43	4.40	4.40	4.18	4.07
C18:2n6	2.98	3.45	5.65	9.01	6.88
C18:3n6	0.23	0.30	0.19	0.24	0.42
C19:0	0.48	0.58	0.47	0.35	0.00
C18:3n3	1.09	1.52	1.42	1.61	1.55
C18:4n3	0.78	0.97	1.03	0.84	1.12
C19:2n6	0.11	0.20	0.11	0.00	0.00
C20:0	0.31	0.44	0.41	0.23	0.25
C20:1n15	0.09	0.00	0.08	0.02	0.00

Table D4 continued. Liver and belly flap samples from striped bass fed the 30% menhaden 70% spot mixture from day 71.

	Liver		Belly flap		
	1	2	1	2	3
C20:1n12	0.52	0.58	0.99	0.51	0.59
C20:1n9	2.08	2.04	1.68	1.26	1.35
C20:1n7	0.69	0.81	1.08	0.57	0.00
C20:2n6	0.91	1.31	0.99	0.68	0.77
C20:3n6	0.21	0.29	0.32	0.23	0.24
C20:4n6	3.46	2.96	1.87	1.23	1.82
C20:3n3	0.57	0.86	0.41	0.24	0.30
C20:5n3	6.26	5.75	4.82	4.31	5.63
C22:0	0.33	0.35	0.34	0.13	0.17
C22:1n9	0.70	0.68	0.36	0.15	0.13
C22:2n6	0.52	0.81	0.13	0.06	0.08
C22:4n6	0.56	0.69	0.69	0.29	0.89
C22:3n3 + C22:5n6	1.41	2.29	0.60	0.46	0.76
C22:5n3	1.93	2.64	1.73	1.30	1.67
C24:0 + C22:6n3	15.53	15.66	8.20	7.23	8.61
C24:1n9	1.04	1.54	0.50	0.33	0.43

Table D5. Liver and belly flap samples from striped bass fed the 70% menhaden 30% spot mixture from day 88.

	Liver				Belly flap			
	1	2	3	4	1	2	3	4
Length (mm)	312	399	406	360	-	-	-	-
Weight (g)	670.2	851.9	943.7	741.4	-	-	-	-
Lipid content (%)	5.02	5.48	4.90	5.15	10.98	6.21	12.31	15.93
C12:0	0.02	0.03	0.03	0.02	0.09		0.08	
C12:1n1	0.00	0.00	0.22	0.00	0.46		0.36	
C13:0	0.04	0.03	0.00	0.07	0.08		0.14	
C14:0	2.31	3.33	3.42	2.27	5.77		5.28	
C14:1n5	0.04	0.04	0.08	0.05	0.23		0.25	
C15:0	0.46	0.67	0.61	0.47	0.89		0.76	
C16:0 DMA	0.01	0.03	0.03	0.03	0.04		0.04	
C16:0	26.37	22.79	22.21	21.56	23.56		22.25	
C16:1n7	3.81	4.25	4.98	4.53	7.90		7.74	
C17:0	1.03	1.24	1.08	0.93	0.95		0.72	
C18:0	8.52	8.04	6.94	5.95	4.75		4.43	
C18:1n12t	0.00	0.28	0.29	0.00	0.24		0.24	
C18:1n9t	0.58	0.41	0.19	0.23	0.19		0.16	
C18:1n9	14.61	9.65	13.56	15.17	16.27		20.58	
C18:1n7	4.02	4.05	4.55	4.43	3.89		3.98	
C18:2n6	1.30	2.65	3.57	1.69	6.06		9.06	
C18:3n6	0.33	0.43	0.39	0.34	0.40		0.41	
C18:3n3	0.71	1.05	1.06	0.86	1.57		1.61	
C18:4n3	0.62	0.79	0.85	0.73	1.13		0.90	
C19:2n6	0.00	0.00	0.00	0.00	0.00		0.02	
C20:0	0.25	0.23	0.24	0.22	0.65		0.26	
C20:1n15	0.00	0.00	0.02	0.00	0.09		0.03	
C20:1n12	0.30	0.31	0.29	0.33	0.82		0.40	
C20:1n9	2.58	1.64	2.41	3.04	1.56		1.36	

Table D5 continued. Liver and belly flap samples from striped bass fed the 70% menhaden 30% spot mixture from day 88.

	Liver				Belly flap			
	1	2	3	4	1	2	3	4
C20:1n7	0.45	0.47	0.54	0.59	0.86		0.56	
C20:2n6	0.44	0.63	0.67	0.57	0.70		0.66	
C20:3n6	0.13	0.16	0.17	0.16	0.32		0.25	
C21:0	0.00	0.00	0.00	0.00	0.12		0.03	
C20:4n6	2.61	3.16	2.78	2.90	1.56		1.35	
C20:3n3	0.24	0.32	0.25	0.28	0.34		0.31	
C20:5n3	6.11	6.62	6.30	6.61	5.31		4.64	
C22:0	0.58	0.66	0.43	0.44	0.30		0.28	
C22:1n9	0.22	0.21	0.27	0.35	0.14		0.12	
C22:2n6	0.04	0.06	0.05	0.17	0.03		0.06	
C22:4n6	0.41	0.56	0.48	0.80	0.55		0.54	
C22:3n3 + C22:5n6	0.62	0.92	0.71	1.33	0.81		0.57	
C22:5n3	1.18	1.86	1.54	2.01	1.56		1.43	
C24:0 + C22:6n3	18.33	21.36	18.17	19.67	9.32		7.79	
C24:1n9	0.74	1.06	0.61	1.19	0.46		0.35	

Table D6. Liver and belly flap samples from striped bass fed the 30% menhaden 70% spot mixture from day 88.

	Liver				Belly flap		
	1	2	3	4	1	2	3
Length (mm)	373	341	365	366	-	-	-
Weight (g)	790	592.6	893.7	702.1	-	-	-
Lipid content (%)	6.71	5.15	8.16	5.69	7.64	14.22	6.54
C12:0	0.02	0.03	0.02	0.03	0.11	0.07	0.10
C12:1n1	0.13	0.00	0.00	0.00	0.72	0.00	0.92
C13:0	0.00	0.06	0.05	0.07	0.40	0.06	0.30
C14:0	1.90	2.54	2.21	2.44	5.65	4.65	5.78
C14:1n5	0.06	0.07	0.07	0.06	0.17	0.19	0.20
C15:0	0.52	0.71	0.58	0.73	1.09	0.88	1.02
C15:1n5	0.02	0.02	0.00	0.00	0.00	0.00	0.00
C16:0 DMA	0.02	0.02	0.02	0.03	0.05	0.00	0.04
C16:0	25.87	23.04	20.29	22.12	25.28	21.84	23.70
C16:1n7	4.29	4.95	5.26	4.03	7.25	6.93	7.29
C17:0	1.02	1.22	0.94	1.37	1.09	1.04	1.06
C17:1n7	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C18:0	7.23	6.83	5.13	7.33	5.49	4.89	5.47
C18:1n12t	0.48	0.00	0.00	0.00	0.48	0.45	0.48
C18:1n9t	0.25	0.36	0.00	0.26	0.00	0.00	0.00
C18:1n9	17.84	15.27	18.93	13.55	14.90	18.56	14.64
C18:1n7	4.31	4.33	4.86	4.26	3.54	3.75	3.64
C18:2n6	1.32	1.88	1.96	1.86	6.11	6.91	6.03
C18:3n6	0.35	0.41	0.34	0.42	0.43	0.42	0.43
C18:3n3	0.66	0.86	0.87	0.84	1.29	1.46	1.50
C18:4n3	0.49	0.63	0.64	0.57	0.94	1.00	1.15
C19:2n6	0.00	0.00	0.00	0.00	0.04	0.00	0.00
C20:0	0.21	0.25	0.25	0.31	0.22	0.26	0.27
C20:1n15	0.00	0.00	0.00	0.00	0.02	0.00	0.03

Table D6 continued. Liver and belly flap samples from striped bass fed the 30% menhaden 70% spot mixture from day 88.

Day 88 Tank 3	Liver				Belly flap		
	1	2	3	4	1	2	3
C20:1n12	0.49	0.70	0.66	0.69	0.67	0.79	0.69
C20:1n9	2.89	2.37	3.69	2.31	1.07	1.49	1.14
C20:1n7	0.67	0.84	0.91	0.87	0.72	0.86	0.82
C20:2n6	0.53	0.64	0.69	0.66	0.63	0.78	0.65
C20:3n6	0.14	0.18	0.18	0.19	0.30	0.26	0.26
C20:4n6	2.63	2.93	2.53	3.95	1.97	1.37	1.96
C20:3n3	0.24	0.35	0.33	0.37	0.27	0.34	0.30
C20:5n3	5.53	5.90	5.69	6.80	5.11	4.12	5.43
C22:0	0.29	0.54	0.41	0.41	0.24	0.43	0.30
C22:1n9	0.17	0.28	0.28	0.17	0.08	0.14	0.16
C22:2n6	0.03	0.18	0.07	0.02	0.00	0.15	0.10
C22:4n6	0.61	0.94	0.71	0.64	0.50	1.09	0.62
C22:3n3 + C22:5n6	0.78	1.41	1.22	0.93	0.76	1.60	0.98
C22:5n3	1.47	2.45	2.05	1.87	1.69	2.84	1.81
C24:0 + C22:6n3	16.05	15.80	17.18	19.22	10.29	8.81	10.34
C24:1n9	0.50	0.99	0.96	0.60	0.43	1.56	0.42

Chapter 6 Raw Data

Values for length (mm), weight (g), lipid content (% wet weight) and fatty acids routinely identified for striped bass belly flap samples analyzed in Chapter 6 of this experiment.

Table E1. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
Total length (mm)	496	426	580	504	554	515	490	504	456	539	542	545
Weight (g)	1088.2	762.4	1880.3	1098.0	1408.2	1256.6	1200.8	1054.7	772.2	1422.2	1482.9	1586.0
Lipid content (%)	4.29	2.05	8.87	2.37	-	3.09	9.45	3.15	2.35	22.05	2.21	3.27
C12:0	0.13	0.06	0.13	0.12	3.06	0.03	0.11	0.14	0.30	0.05	0.06	0.03
C12:1n1	nd	0.04	0.05	0.22	0.84	nd	nd	nd	nd	0.03	0.08	0.02
C13:0	nd	nd	0.37	nd	1.34	nd	nd	nd	0.17	nd	nd	nd
C14:0	5.67	3.53	9.20	4.85	2.82	2.04	2.51	6.28	4.08	2.40	2.29	1.55
C14:1n5	0.04	0.09	0.27	nd	1.84	0.02	0.04	0.14	0.25	0.01	0.08	0.01
C15:0	1.06	0.70	1.22	1.13	1.33	0.54	0.76	1.05	1.06	0.60	0.76	0.88
C15:1n5	nd	nd	nd	nd	0.89	nd	0.01	nd	0.00	nd	nd	nd
C16:0	27.10	30.12	27.01	36.35	3.81	32.88	31.37	30.12	27.08	34.16	32.23	30.41
C16:1n7	7.37	5.27	9.70	5.42	1.90	3.03	4.10	6.17	10.40	3.26	5.52	3.06
C17:0	1.05	1.24	1.36	1.04	2.13	0.54	1.29	1.12	1.03	1.11	1.21	1.36
C17:1n7	nd	nd	nd	nd	1.21	nd	nd	nd	nd	nd	nd	nd
C18:0	6.38	6.35	4.07	8.47	2.73	8.18	8.57	7.82	4.61	9.27	7.28	9.94
C18:1n9t	0.38	0.35	0.22	0.23	0.99	0.22	0.32	nd	0.41	0.19	0.31	0.22
C18:1n7t	nd	nd	nd	nd	0.69	nd	nd	nd	nd	nd	nd	nd
C18:1n9	6.52	12.43	11.13	8.01	1.97	6.24	9.34	13.61	21.21	5.72	14.45	5.63
C18:1n7	3.88	4.52	4.62	3.35	1.17	3.14	3.78	4.69	5.46	2.73	4.25	2.91
C18:2n6	1.99	1.30	1.49	0.89	4.46	0.97	0.87	1.12	1.18	1.08	0.78	1.59
C18:3n6	0.21	0.13	0.18	0.06	4.07	0.06	0.07	0.09	0.11	0.07	0.04	0.09
C19:0	0.28	0.22	0.21	0.29	nd	0.18	0.20	0.26	0.24	0.16	0.19	0.22
C18:3n3	1.58	0.69	1.65	0.38	5.88	0.43	0.37	0.50	0.59	0.70	0.27	1.22

Table E1 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	1	2	3	4	5	6	7	8	9	10	11	12
C18:4n3	2.48	0.84	1.57	0.39	1.10	0.62	0.33	0.70	0.44	1.01	0.14	0.24
C19:2n6	0.32	0.12	nd	0.16	2.63	0.13	0.13	0.04	0.03	0.18	0.13	0.18
C20:0	0.22	0.11	0.17	0.25	1.67	0.09	0.22	0.33	0.30	0.12	0.17	0.13
C20:1n15	0.02	0.01	0.01	0.05	0.77	0.02	0.05	nd	nd	0.01	0.02	0.08
C20:1n12	0.49	0.46	0.47	1.14	0.83	0.36	1.04	2.20	1.54	0.17	1.27	0.21
C20:1n9	1.00	1.33	0.99	1.48	1.74	1.04	1.88	2.98	2.52	0.70	1.94	0.81
C20:1n7	0.43	0.53	0.57	0.72	nd	0.32	1.12	1.96	2.50	0.19	1.08	0.23
C20:2n6	0.41	0.34	0.40	0.48	4.13	0.32	0.46	0.63	0.74	0.31	0.48	0.44
C20:3n6	0.10	0.09	0.24	0.14	3.98	0.09	0.09	0.11	0.13	0.10	0.10	0.31
C21:0	0.01	0.01	nd	0.04	nd	0.02	0.01	0.01	nd	0.01	nd	0.00
C20:4n6	1.64	1.61	1.16	2.72	3.59	2.73	3.02	1.73	1.45	3.31	3.04	6.07
C20:3n3	0.25	0.18	0.41	0.24	5.61	0.16	0.14	0.16	0.19	0.19	0.18	0.27
C20:5n3	5.51	5.41	5.68	4.31	3.05	5.14	5.23	2.99	3.25	6.87	3.93	7.28
C22:0	0.27	0.13	0.46	0.15	1.69	0.07	0.11	0.08	0.11	0.05	0.09	0.08
C22:1n9	0.18	0.10	0.93	0.19	1.96	0.14	0.13	0.14	0.32	nd	0.18	0.09
C22:2n6	0.04	0.02	0.03	nd	4.15	0.04	nd	nd	0.05	0.01	nd	0.02
C22:4n6	0.30	0.26	0.32	0.49	2.98	0.43	0.45	0.61	0.40	0.29	0.83	0.39
C22:3n3 + C22:5n6	0.68	0.63	0.47	0.59	7.01	1.06	0.92	0.52	0.46	0.83	0.80	2.24
C22:5n3	1.65	1.56	2.02	1.40	3.10	1.39	1.38	1.49	1.41	1.21	2.05	1.73
C22:6n3 + C24:0	19.58	18.18	10.63	13.66	5.24	26.35	18.98	9.62	5.78	22.21	13.24	19.33
C24:1n9	0.80	1.04	0.60	0.57	1.65	0.98	0.59	0.57	0.19	0.71	0.56	0.73

Table E2. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	13	14	15	16	17	18	19	20	21	22	23	24
Total length (mm)	521	419	557	440	576	457	587	545	439	607	482	521
Weight (g)	1136.7	608.4	1702.3	736.3	1688.8	816.1	1868.2	1572.4	904.2	1940.6	1102.9	1238.0
Lipid content (%)	2.39	2.17	8.65	1.90	-	3.06	4.96	7.34	18.75	5.23	4.32	1.12
C12:0	nd	0.12	0.07	0.87	0.06	0.12	0.09	0.43	0.14	0.08	0.04	0.04
C12:1n1	0.03	nd	0.03	0.51	0.03	nd	nd	nd	0.12	nd	nd	nd
C13:0	nd	0.38	nd	nd	nd	nd	nd	0.33	0.40	0.29	nd	nd
C14:0	1.20	6.63	2.60	2.99	3.14	4.43	3.72	2.61	5.58	4.13	1.24	1.58
C14:1n5	0.02	0.15	0.02	0.16	0.04	0.16	0.18	0.14	0.18	0.12	0.01	0.03
C15:0	0.48	0.89	0.87	0.51	0.75	0.89	0.86	1.21	0.87	0.66	0.45	0.51
C15:1n5	nd	0.01	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C16:0	31.74	25.49	32.52	22.22	34.21	29.44	31.01	24.28	26.68	31.32	26.26	29.43
C16:1n7	2.47	8.36	3.86	2.67	3.56	7.18	8.86	8.06	8.12	6.43	2.15	2.97
C17:0	0.97	0.77	1.26	0.64	0.89	0.91	1.35	1.29	1.06	1.17	0.92	0.71
C17:1n7	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:0	9.44	5.18	8.36	10.47	9.09	6.54	5.85	5.22	5.36	6.11	8.02	9.57
C18:1n9t	0.20	0.26	0.23	0.73	0.27	0.28	0.83	0.37	0.31	0.35	0.23	0.40
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	6.19	12.15	9.04	8.40	8.73	16.00	18.19	20.15	15.41	14.56	5.40	9.94
C18:1n7	2.30	4.39	3.22	2.26	2.90	4.33	5.33	5.18	4.84	3.53	1.92	3.50
C18:2n6	0.81	1.31	0.94	6.74	0.82	0.93	2.28	1.60	1.40	1.31	0.93	1.01
C18:3n6	0.03	0.14	0.05	1.08	0.06	0.07	0.26	0.16	0.17	0.14	0.06	0.11
C19:0	0.19	0.17	0.24	0.99	0.23	0.25	0.34	0.33	0.26	0.18	0.19	0.33
C18:3n3	0.36	1.11	0.44	2.77	0.42	0.51	0.60	0.82	0.92	1.03	0.45	0.40
C18:4n3	0.32	1.21	0.28	3.54	0.42	0.44	0.54	0.34	1.28	1.44	0.42	0.39
C19:2n6	0.17	0.10	0.13	1.41	0.11	0.12	0.09	0.07	0.07	0.11	0.18	0.23
C20:0	0.10	0.17	0.21	3.12	0.26	0.29	0.10	0.27	0.28	0.17	0.16	0.20
C20:1n15	0.00	0.02	0.04	0.79	0.02	0.06	nd	0.05	0.05	0.02	0.06	0.03

Table E2 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	13	14	15	16	17	18	19	20	21	22	23	24
C20:1n12	0.09	0.59	0.93	nd	0.93	1.36	nd	1.02	1.17	0.18	0.22	0.74
C20:1n9	0.61	1.56	1.34	3.82	1.65	2.39	1.24	1.92	2.66	1.11	0.61	1.47
C20:1n7	0.13	0.85	0.73	3.41	0.63	1.42	0.72	1.46	1.06	0.22	0.26	0.50
C20:2n6	0.32	0.46	0.33	5.75	0.35	0.49	0.46	0.73	0.55	0.28	0.48	0.34
C20:3n6	0.07	0.12	0.09	nd	0.09	0.10	0.11	0.16	0.16	0.10	0.38	0.10
C21:0	0.01	nd	nd	nd	nd	0.01	0.01	0.14	0.05	0.03	0.16	0.02
C20:4n6	4.42	0.65	4.19	5.48	4.12	2.72	1.84	2.52	1.30	1.84	4.42	3.84
C20:3n3	0.13	0.16	0.13	0.20	0.15	0.14	0.22	0.18	0.16	0.15	0.36	0.11
C20:5n3	6.38	4.83	4.16	1.62	4.32	2.75	3.25	4.62	3.54	5.05	5.65	6.03
C22:0	0.04	0.19	0.11	nd	0.12	0.15	0.05	0.19	0.14	0.17	0.58	0.13
C22:1n9	nd	0.59	0.10	nd	0.32	0.87	0.12	0.43	0.37	0.22	0.53	0.21
C22:2n6	0.03	0.35	0.03	nd	nd	0.32	0.02	0.11	nd	0.07	1.23	nd
C22:4n6	0.36	0.59	0.73	nd	0.94	0.70	0.26	0.56	0.50	0.30	1.86	0.52
C22:3n3 + C22:5n6	1.05	1.35	1.08	0.13	1.17	0.84	0.50	0.55	0.67	0.65	2.97	0.99
C22:5n3	0.99	3.10	1.43	0.47	1.40	1.65	1.57	2.39	2.48	1.13	2.98	1.61
C24:0 + C22:6n3	27.58	13.92	19.73	6.00	17.10	10.63	9.00	9.88	11.09	14.80	25.57	21.32
C24:1n9	0.76	1.70	0.49	0.24	0.71	0.50	0.19	0.24	0.62	0.54	2.64	0.67

Table E3. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	25	26	27	28	29	30	31	32	33	34	35	36
Total length (mm)	490	565	486	468	537	510	540	530	524	539	570	606
Weight (g)	914.4	1876.7	1132.2	1054.1	1620.0	1372.9	1512	1340	1474	1710	1986	2032
Lipid content (%)	2.50	14.39	7.81	2.82	7.81	32.51	4.16	0.56	2.01	1.45	1.12	2.25
C12:0	0.11	0.07	0.01	0.09	0.39	0.07	0.10	0.06	0.04	0.04	0.05	0.05
C12:1n1	0.05	nd	nd	nd	nd	0.02	0.01	nd	0.01	nd	nd	0.01
C13:0	nd	nd	nd	nd	0.24	nd	nd	nd	nd	nd	nd	nd
C14:0	4.07	5.13	1.16	3.40	4.76	2.23	5.29	3.24	2.73	1.66	2.91	2.87
C14:1n5	0.10	0.05	0.01	0.06	0.27	0.02	0.07	0.03	0.03	0.03	0.16	0.02
C15:0	0.83	0.80	0.60	0.78	1.41	0.68	0.84	0.93	0.59	0.89	0.55	0.65
C15:1n5	nd	0.03	nd	0.00	0.12	nd	nd	nd	nd	nd	nd	nd
C16:0	25.75	28.39	31.47	22.49	23.14	32.69	28.91	32.75	28.15	26.39	30.44	29.74
C16:1n7	8.62	8.06	2.56	6.02	8.99	3.95	7.08	5.05	4.38	3.58	7.49	4.08
C17:0	0.99	1.28	0.94	0.88	1.01	1.01	1.14	1.14	0.66	1.08	0.68	0.75
C17:1n7	nd	nd	nd	nd	0.15	nd	nd	nd	nd	nd	nd	nd
C18:0	5.13	7.69	8.59	5.00	5.02	8.60	6.57	7.49	8.06	5.42	5.73	8.83
C18:1n9t	0.38	nd	0.24	0.26	0.50	0.27	0.29	0.26	0.27	0.27	0.44	0.21
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	14.89	7.21	8.15	7.77	20.91	7.02	9.72	7.70	7.26	6.41	20.41	6.21
C18:1n7	4.14	3.70	2.90	3.18	4.68	3.33	3.74	3.42	3.15	2.52	4.65	2.73
C18:2n6	1.66	1.09	0.95	1.17	1.89	0.85	1.30	1.03	0.93	nd	0.84	1.10
C18:3n6	0.17	0.18	0.06	0.16	0.24	0.06	0.14	0.10	0.12	0.12	0.07	0.07
C19:0	0.19	0.23	0.32	0.32	0.34	0.23	0.20	0.25	0.27	0.36	0.21	0.23
C18:3n3	2.09	1.01	0.50	1.29	1.29	0.74	1.28	0.76	0.86	0.62	0.35	0.96
C18:4n3	2.18	1.26	0.50	1.38	1.60	0.65	2.49	1.21	1.08	0.54	0.35	1.81
C19:2n6	nd	0.15	0.16	0.33	0.06	0.18	0.14	0.11	0.25	0.22	0.08	0.13
C20:0	0.21	0.20	0.13	0.49	0.59	0.18	0.20	0.18	0.26	0.34	0.19	0.18
C20:1n15	0.05	0.04	0.04	0.34	nd	0.05	0.04	0.05	0.10	0.13	0.05	0.03

Table E3 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	25	26	27	28	29	30	31	32	33	34	35	36
C20:1n12	0.28	0.20	0.26	0.78	1.84	0.47	0.67	0.58	0.33	0.85	1.34	0.25
C20:1n9	1.29	1.14	0.86	1.44	2.28	1.29	1.58	1.17	1.08	1.56	2.13	0.81
C20:1n7	0.53	0.36	0.27	1.08	2.48	0.44	0.57	0.57	0.42	0.94	1.24	0.30
C20:2n6	0.47	0.37	0.19	1.02	0.79	0.39	0.42	0.38	0.52	1.03	0.42	0.39
C20:3n6	0.15	0.18	0.14	0.46	0.24	0.09	0.09	0.18	0.32	0.61	0.13	0.12
C21:0	0.03	nd	nd	nd	nd	0.02	nd	nd	nd	nd	nd	nd
C20:4n6	1.82	2.26	4.11	2.25	1.40	2.62	1.81	2.57	2.79	4.45	2.30	2.56
C20:3n3	0.34	0.20	0.18	0.70	0.24	0.15	0.17	0.13	0.46	0.69	0.13	0.16
C20:5n3	6.97	7.62	6.70	6.78	3.38	5.72	6.87	6.34	6.05	5.82	3.44	7.15
C22:0	0.15	0.22	0.09	0.87	0.17	0.05	0.14	0.13	0.35	0.92	0.07	0.07
C22:1n9	nd	0.57	0.09	0.80	0.62	0.30	0.23	0.15	1.01	2.22	0.19	0.28
C22:2n6	0.10	0.20	0.11	0.82	nd	0.03	nd	0.03	nd	0.43	nd	0.01
C22:4n6	0.26	0.39	1.05	1.69	0.27	0.44	0.39	0.48	0.94	2.64	0.66	0.27
C22:3n3 + C22:5n6	0.56	0.80	1.41	2.15	0.75	0.90	0.64	0.80	2.06	2.67	0.63	0.87
C22:5n3	1.26	1.37	2.80	5.83	1.40	1.33	1.42	1.78	2.00	4.51	2.35	1.11
C24:0 + C22:6n3	13.84	16.73	21.96	15.56	6.14	22.26	14.88	18.29	20.81	17.35	8.89	24.38
C24:1n9	0.36	0.45	0.50	2.26	0.42	0.69	0.56	0.67	1.63	2.67	0.46	0.62

Table E4. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	37	38	39	40	41	42	43	44	45	46	47	48
Total length (mm)	440	457	514	544	557	554	540	513	473	519	497	522
Weight (g)	822	956	1260	1742	1770	1670	1534	1558	1106	1374	1192	1448
Lipid content (%)	1.88	6.38	4.68	5.55	2.45	2.02	3.51	7.55	3.48	3.31	3.52	1.10
C12:0	0.07	0.06	0.12	0.11	0.04	0.07	0.18	0.05	0.09	0.12	0.10	0.06
C12:1n1	nd	0.04	nd	0.01	0.06	0.04	0.06	0.03	0.02	0.02	0.03	0.04
C13:0	nd	nd	nd	nd	nd	nd	0.54	nd	0.37	nd	nd	nd
C14:0	4.24	3.21	5.57	2.32	1.74	3.40	4.40	2.03	3.40	2.27	1.66	1.83
C14:1n5	0.10	0.08	0.08	0.03	0.03	0.05	0.18	0.02	0.06	0.02	0.02	0.03
C15:0	0.77	0.66	0.98	0.66	0.63	0.88	1.13	0.61	0.78	0.78	0.84	0.61
C15:1n5	0.01	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C16:0	28.62	28.27	29.01	29.76	20.56	27.47	25.73	18.69	26.88	30.52	29.80	29.80
C16:1n7	5.76	4.66	6.87	4.37	4.93	5.76	7.09	3.11	5.91	3.37	3.77	3.12
C17:0	0.83	1.13	0.94	1.01	6.20	0.96	1.78	0.45	0.84	0.79	1.40	1.70
C17:1n7	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:0	6.92	7.84	6.58	7.59	5.84	7.40	4.45	5.31	6.85	9.67	8.53	8.76
C18:1n9t	0.32	0.36	0.26	0.18	nd	0.30	0.17	nd	0.19	nd	0.21	0.19
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	12.06	12.76	11.26	6.36	5.57	6.87	13.27	4.79	8.29	7.26	8.72	9.64
C18:1n7	3.67	4.19	3.94	2.78	3.33	3.41	3.84	2.39	3.30	3.44	2.75	3.07
C18:2n6	1.11	0.89	1.25	0.95	0.77	0.93	1.30	0.87	1.26	0.90	1.06	0.99
C18:3n6	0.12	0.10	0.14	0.09	0.04	0.16	0.25	0.28	0.09	0.17	0.05	0.06
C19:0	0.19	0.23	0.24	0.18	0.14	0.28	0.26	0.28	0.23	0.25	0.25	0.24
C18:3n3	1.15	0.69	1.21	0.77	1.08	1.31	1.73	0.95	1.16	0.86	0.49	0.50
C18:4n3	2.06	0.66	2.05	1.13	0.65	1.11	1.85	1.24	1.93	0.84	0.45	0.74
C19:2n6	0.13	0.11	0.12	0.17	0.19	0.20	0.14	0.54	0.17	0.21	0.14	0.36
C20:0	0.25	0.27	0.32	0.12	0.10	0.22	0.34	0.81	nd	0.28	0.16	0.38
C20:1n15	nd	0.07	0.04	0.02	0.18	0.04	0.04	nd	nd	0.03	0.00	0.02

Table E4 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	37	38	39	40	41	42	43	44	45	46	47	48
C20:1n12	0.76	1.27	0.92	0.25	0.34	0.37	0.57	1.15	0.46	0.33	0.33	0.66
C20:1n9	1.91	2.90	2.22	0.82	0.91	1.13	1.58	1.41	1.00	1.16	1.04	1.44
C20:1n7	0.55	1.19	0.63	0.31	0.51	0.35	0.66	0.88	0.54	0.36	0.67	0.68
C20:2n6	0.39	0.61	0.51	0.39	0.54	0.26	0.49	0.97	0.45	0.59	0.57	0.58
C20:3n6	0.13	0.14	0.23	0.15	0.22	0.14	0.17	0.66	0.18	0.14	0.21	0.10
C21:0	nd	nd	nd	nd	nd	0.10	nd	nd	nd	0.10	nd	0.16
C20:4n6	1.46	1.75	1.83	3.58	3.72	2.82	2.02	3.19	2.42	2.56	4.57	3.17
C20:3n3	0.18	0.21	0.29	0.17	0.35	1.20	0.30	0.85	0.29	0.23	0.23	0.24
C20:5n3	5.40	4.14	5.18	7.48	6.25	6.88	6.27	8.42	7.78	5.64	6.69	4.86
C22:0	0.22	0.35	0.15	0.22	0.25	0.17	0.31	1.63	0.40	0.41	0.14	2.36
C22:1n9	0.67	0.97	0.28	0.14	0.31	0.20	0.49	2.03	0.49	0.37	0.15	0.71
C22:2n6	0.10	0.17	nd	0.02	0.11	nd	0.11	0.79	0.05	nd	0.02	0.18
C22:4n6	0.37	0.50	0.61	0.46	1.00	0.55	0.38	2.17	0.36	0.24	0.81	0.41
C22:3n3 + C22:5n6	0.59	0.69	0.72	1.39	1.52	0.87	0.76	3.00	0.84	0.85	1.08	1.04
C22:5n3	1.51	1.45	1.71	1.64	2.69	1.56	2.05	4.65	1.76	1.30	2.00	1.39
C24:0 + C22:6n3	16.77	16.88	13.33	23.83	25.72	21.28	14.60	23.60	20.68	23.15	20.38	19.27
C24:1n9	0.64	0.52	0.40	0.50	3.49	1.26	0.51	2.08	0.48	0.74	0.69	0.63

Table E5. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	49	50	51	52	53	54	55	56	57	58	59	60
Total length (mm)	532	560	481	465	550	568	519	476	460	542	525	552
Weight (g)	1328	1822	1200	854	1460	1794	1256	1026	996	1482	1446	1818
Lipid content (%)	3.49	4.58	9.92	2.24	2.46	6.46	2.96	3.22	3.74	2.70	7.95	3.19
C12:0	0.12	0.12	0.13	0.09	0.08	0.03	0.10	0.06	0.11	0.10	0.10	0.13
C12:1n1	0.02	nd	nd	0.04	nd	nd	0.04	0.02	0.04	0.03	nd	0.03
C13:0	0.43	0.29	nd	nd	0.34	nd	nd	nd	nd	0.24	0.33	0.55
C14:0	3.80	8.75	3.06	2.47	4.37	1.87	2.64	2.43	2.67	6.17	5.42	4.46
C14:1n5	0.08	0.28	0.03	0.03	0.09	0.02	0.03	0.04	0.05	0.08	0.08	0.04
C15:0	1.06	1.20	0.82	0.68	0.86	0.72	0.84	0.85	0.86	0.96	0.98	0.74
C15:1n5	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C16:0	29.31	26.13	29.13	21.60	22.08	20.45	35.02	29.11	27.46	27.92	30.59	27.86
C16:1n7	5.91	9.23	4.66	3.52	6.12	3.30	3.67	3.59	4.48	7.77	7.16	5.91
C17:0	0.96	1.47	0.74	1.41	1.31	1.37	1.40	1.41	1.29	1.31	1.36	1.40
C17:1n7	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:0	7.54	4.65	8.36	5.92	5.13	6.55	8.53	7.79	7.34	6.25	6.47	7.39
C18:1n9t	0.22	nd	0.17	0.18	0.27	0.26	0.25	nd	0.19	0.12	nd	0.14
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	11.17	9.43	5.82	5.78	10.36	5.26	7.02	8.80	10.13	8.76	9.62	6.46
C18:1n7	3.39	4.15	2.67	2.70	3.32	2.57	2.58	2.83	3.34	3.64	3.57	3.29
C18:2n6	1.24	1.69	1.09	1.11	1.42	0.97	1.19	0.84	0.86	1.30	1.43	1.27
C18:3n6	0.10	0.21	0.11	0.19	0.26	0.18	0.09	0.07	0.09	0.16	0.14	0.14
C19:0	0.27	0.21	0.22	0.33	0.29	0.39	0.23	0.25	0.21	0.22	0.22	0.19
C18:3n3	0.99	2.07	0.86	0.99	1.51	1.34	1.11	0.63	0.73	1.44	1.45	1.33
C18:4n3	1.29	1.95	1.27	1.49	1.94	0.94	1.21	0.66	1.06	2.53	2.14	2.85
C19:2n6	0.15	nd	0.22	0.60	0.30	0.39	0.15	0.12	0.18	0.15	0.14	0.23
C20:0	0.34	0.27	0.38	0.47	0.48	0.62	0.22	0.24	0.18	0.28	0.25	0.20
C20:1n15	0.03	nd	nd	0.43	0.30	0.24	0.04	0.02	0.03	0.05	0.04	0.03

Table E5 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	49	50	51	52	53	54	55	56	57	58	59	60
C20:1n12	1.12	0.34	0.29	0.79	0.85	0.71	0.18	0.55	0.76	0.73	0.64	0.39
C20:1n9	1.69	1.17	0.94	1.60	1.83	1.24	0.88	1.49	1.73	1.47	1.41	0.95
C20:1n7	1.24	0.53	0.31	0.96	0.83	0.76	0.19	0.48	0.62	0.65	0.48	0.31
C20:2n6	0.58	0.44	0.37	0.71	0.90	0.96	0.31	0.60	0.40	0.39	0.43	0.44
C20:3n6	0.12	0.19	0.16	0.72	0.39	0.73	0.13	0.25	0.18	0.10	0.10	0.91
C21:0	0.06	nd	nd	0.40	0.19	nd	nd	nd	nd	0.04	0.04	nd
C20:4n6	2.58	1.04	4.55	3.41	2.05	4.04	2.80	3.56	2.84	1.46	1.81	3.78
C20:3n3	0.23	0.33	0.18	0.58	0.91	0.83	0.18	0.33	0.23	0.23	0.26	0.81
C20:5n3	5.66	6.29	7.92	6.28	5.60	7.80	5.31	6.78	6.78	7.15	5.85	7.18
C22:0	0.49	nd	0.25	1.44	1.02	1.34	0.15	0.47	0.80	0.52	0.13	0.14
C22:1n9	0.44	0.12	0.21	1.01	1.18	1.21	0.12	0.40	0.54	0.48	0.21	0.19
C22:2n6	0.03	0.04	0.06	0.58	0.92	0.36	0.03	0.10	0.06	0.05	nd	nd
C22:4n6	0.33	0.20	0.27	1.54	0.91	1.33	0.30	0.66	0.65	0.34	0.38	0.43
C22:3n3 + C22:5n6	0.72	0.63	1.26	3.19	2.18	3.51	0.87	1.07	0.91	0.64	0.66	0.71
C22:5n3	1.42	2.19	1.92	4.20	3.40	4.32	0.92	1.81	2.16	1.64	1.54	1.36
C24:0 + C22:6n3	14.40	13.86	20.81	19.19	13.64	20.35	20.86	21.12	19.61	14.00	14.06	17.16
C24:1n9	0.48	0.52	0.74	3.36	2.39	3.03	0.43	0.56	0.44	0.60	0.51	0.59

Table E6. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	61	62	63	64	65	66	67	68	69	70	71	72
Total length (mm)	511	488	515	557	406	432	434	491	536	449	527	511
Weight (g)	1170	1266	1392	1440	594	864	820	1250	1524	1232	1526	1194
Lipid content (%)	4.40	2.87	2.22	2.92	3.54	6.11	9.19	11.13	10.07	5.33	3.49	2.94
C12:0	0.09	0.07	0.04	0.07	0.23	0.19	0.07	0.09	0.10	0.07	0.07	0.06
C12:1n1	0.03	0.02	0.01	0.04	0.03	0.02	0.03	0.02	0.03	0.03	0.04	0.03
C13:0	0.43	0.33	nd	nd	0.30	0.21	0.24	0.13	0.26	nd	0.42	nd
C14:0	4.95	4.36	1.91	2.34	2.34	2.86	3.32	3.04	5.05	2.84	4.06	2.44
C14:1n5	0.08	0.12	0.04	0.04	0.15	0.18	0.16	0.16	0.12	0.08	0.04	0.08
C15:0	0.95	0.72	0.58	0.57	0.86	0.80	1.02	0.64	0.81	0.59	0.71	0.86
C15:1n5	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C16:0	27.81	30.14	29.21	29.09	25.30	26.99	30.17	21.18	26.17	22.15	28.00	29.21
C16:1n7	7.01	6.78	3.18	3.96	6.21	7.66	5.51	7.11	6.88	4.81	6.25	5.47
C17:0	0.97	1.34	0.78	0.76	1.14	1.04	1.22	0.97	0.77	1.05	1.24	1.19
C17:1n7	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:0	7.48	6.99	9.14	8.67	4.64	5.40	4.95	4.55	5.94	5.19	7.47	7.31
C18:1n9t	nd	0.39	0.23	0.20	0.26	0.25	0.24	0.30	0.14	0.25	0.22	0.26
C18:1n7t	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C18:1n9	9.12	14.84	8.69	8.71	20.15	24.03	21.81	18.03	11.17	9.16	7.20	16.60
C18:1n7	4.28	4.32	3.43	2.74	4.36	4.53	4.24	5.51	3.85	3.53	3.52	3.95
C18:2n6	1.33	1.17	0.82	0.89	1.44	1.20	2.01	1.10	1.49	1.18	1.66	0.86
C18:3n6	0.17	0.15	0.06	0.08	0.13	0.09	0.15	0.21	0.15	0.18	0.19	0.07
C19:0	0.25	0.24	0.27	0.23	0.37	0.22	0.26	0.35	0.20	0.25	0.23	0.21
C18:3n3	1.14	0.97	0.45	0.60	0.86	0.48	1.37	0.73	1.41	0.94	1.35	0.60
C18:4n3	2.00	1.39	0.36	0.55	0.50	0.17	1.03	0.65	2.36	1.01	2.08	0.61
C19:2n6	0.14	0.14	0.19	0.12	0.03	0.04	0.06	0.30	0.12	0.30	0.19	0.07
C20:0	0.26	0.21	0.15	0.18	0.35	0.29	0.26	0.40	0.23	0.38	0.25	0.21
C20:1n15	0.05	0.04	0.02	0.03	nd	0.02	nd	0.23	0.03	0.11	0.07	0.06

Table E6 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number											
	61	62	63	64	65	66	67	68	69	70	71	72
C20:1n12	1.01	0.88	0.84	0.56	0.93	1.15	0.33	1.59	0.87	0.82	0.31	0.83
C20:1n9	1.63	2.83	1.80	0.93	2.04	1.99	1.62	2.98	2.02	2.03	1.09	2.36
C20:1n7	0.76	0.78	0.55	0.49	1.25	1.75	0.41	1.74	0.60	0.89	0.33	0.82
C20:2n6	0.46	0.34	0.37	0.32	0.72	0.55	0.46	1.56	0.51	0.97	0.41	0.46
C20:3n6	0.12	0.09	0.09	0.11	0.13	0.12	0.13	0.36	0.13	0.71	0.12	0.10
C21:0	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
C20:4n6	1.54	1.26	3.85	3.23	2.07	1.72	1.56	1.88	2.23	3.23	2.19	2.00
C20:3n3	0.23	0.18	0.15	0.12	0.24	0.17	0.28	0.80	0.26	0.60	0.20	0.06
C20:5n3	6.22	4.11	5.32	7.72	8.17	5.80	2.35	5.32	6.29	7.81	7.07	5.20
C22:0	0.14	0.11	0.10	1.41	1.47	1.31	0.10	0.58	0.26	0.77	0.13	0.35
C22:1n9	0.23	0.24	0.19	0.55	0.61	0.59	0.25	0.87	0.31	1.09	0.18	0.37
C22:2n6	nd	nd	nd	nd	0.10	0.05	0.04	0.34	nd	nd	0.04	0.03
C22:4n6	0.45	0.42	0.63	0.51	0.47	0.38	0.42	1.66	0.53	1.69	0.32	0.50
C22:3n3 + C22:5n6	0.71	0.53	1.19	0.99	0.57	0.37	0.70	1.81	0.80	1.95	0.91	0.58
C22:5n3	1.67	1.38	1.52	1.65	1.90	1.53	1.65	3.31	1.94	3.30	1.37	1.80
C24:0 + C22:6n3	15.67	11.76	23.06	21.03	9.48	5.71	11.20	8.35	15.45	18.38	19.49	13.94
C24:1n9	0.63	0.33	0.79	0.49	0.20	0.11	0.36	1.13	0.53	1.64	0.58	0.46

Table E7. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number				
	73	74	75	76	77
Total length (mm)	470	503	545	454	573
Weight (g)	1036	1020	1560	958	1100
Lipid content (%)	6.20	1.68	3.09	6.36	8.87
C12:0	0.06	0.05	0.10	0.20	0.15
C12:1n1	nd	0.03	0.04	0.11	0.04
C13:0	nd	0.26	nd	0.45	0.30
C14:0	2.33	2.28	3.88	2.65	6.17
C14:1n5	0.04	0.08	0.11	0.11	0.15
C15:0	0.60	0.69	0.85	0.66	0.91
C15:1n5	nd	nd	nd	nd	nd
C16:0	28.22	22.14	32.27	21.57	25.51
C16:1n7	3.87	5.85	6.58	3.91	7.86
C17:0	1.18	1.10	1.28	0.94	1.11
C17:1n7	nd	nd	nd	nd	nd
C18:0	8.02	4.60	7.06	5.08	6.20
C18:1n9t	0.21	0.33	0.25	0.33	0.15
C18:1n7t	nd	nd	nd	nd	nd
C18:1n9	9.75	12.53	15.61	11.33	12.04
C18:1n7	3.95	3.61	3.66	2.31	4.31
C18:2n6	0.92	1.08	1.22	0.77	1.47
C18:3n6	0.07	0.21	0.11	0.22	0.18
C19:0	0.22	0.40	0.25	0.30	0.26
C18:3n3	0.72	0.64	1.06	0.99	1.46
C18:4n3	0.52	0.82	1.00	0.81	2.38
C19:2n6	0.13	0.25	0.12	0.14	0.11
C20:0	0.18	0.79	0.32	0.31	0.28
C20:1n15	0.03	0.33	nd	nd	0.04

Table E7 continued. Striped bass collected from the Choptank River for field validation of the QFASA model.

	Sample number				
	73	74	75	76	77
C20:1n12	0.77	1.30	0.58	0.38	1.48
C20:1n9	2.00	1.96	1.40	1.20	2.32
C20:1n7	0.55	1.53	0.73	0.75	1.12
C20:2n6	0.40	1.47	0.47	0.35	0.55
C20:3n6	0.15	0.36	0.15	nd	0.12
C21:0	0.03	nd	nd	nd	nd
C20:4n6	3.09	2.86	1.89	2.45	1.28
C20:3n3	0.29	0.99	0.19	0.24	0.19
C20:5n3	6.18	5.83	4.40	15.32	6.21
C22:0	0.34	1.04	0.21	11.51	0.45
C22:1n9	0.39	1.40	0.17	3.01	0.35
C22:2n6	nd	0.47	0.07	nd	0.08
C22:4n6	0.59	2.05	0.30	0.96	0.74
C22:3n3 + C22:5n6	0.96	2.35	0.61	0.70	0.59
C22:5n3	1.56	3.98	1.20	1.07	2.22
C24:0 + C22:6n3	20.97	12.32	11.40	8.60	10.82
C24:1n9	0.71	2.03	0.47	0.28	0.41

Methods Appendix

The following information represents the methods used at the NOAA Fisheries James J. Howard Marine Sciences Laboratory in Sandy Hook, New Jersey. Some methods described were attempted but were not used in the final preparation of samples due to an inability to obtain proper results or to save time and money. All glassware and metal tools, e.g. spatulas, were cleaned three times with acetone and three times with methylene chloride dispensed from PTFE spray bottles prior to any sample addition. Air in test tubes containing fatty acids in methylene chloride or hexane was flushed under a gentle stream of nitrogen prior to storage in the freezer to prevent oxidation. All liquid and solid hazardous waste was disposed of in accordance with safety protocols of the laboratory.

Grinding the samples

Samples were removed from the freezer at the time of analysis and weighed to the nearest 0.1 g. Accelerated solvent extractor (ASE) stainless steel cells (34 ml) were prepped and filled with diatomaceous earth. A sample consisted of three to five grams of the sample. The diatomaceous earth was added to the mortar along with the sample. The sample was ground using a pestle until the sample was thoroughly mixed with the diatomaceous earth. The entire contents of the mortar were then added to the ASE cell. A known amount of C23:0 triacylglycerol standard (typically 60 to 100 μg) was added to each cell and to a cell that only contained diatomaceous earth. Samples that exceeded three to five grams in weight were homogenized in a blender first, and then three to five grams of sample was removed, and the above procedure was followed.

Running samples on the Accelerated Solvent Extractor (ASE)

Cells were placed onto the ASE along with glass collecting bottles. A solution of 3:1 by volume of methylene chloride and methanol was prepared and 0.01% of butylated hydroxytoluene (BHT) was added as an antioxidant. The ASE and the nitrogen supply were turned on, and the line was rinsed with the methylene chloride:methanol solution four times. Once this process was complete, the program was initiated. This program heated each cell to $100^{\circ}C$ and applied a pressure of 1500 psi to each cell along with adding the organic solution. The first extraction provided 90.8% of the lipids that were sufficient to perform the quantitative FAME analysis. The second extraction provided 9.2% of the lipids extracted in the first extraction step. All subsequent extractions involved only one ASE extraction. The total lipid data was corrected for the amount of lipid still remaining in the sample after the first extraction.

Backwashing samples

Once the extraction process was complete, the collection bottles were transferred to a prepped 125 ml separatory funnel. The following backwash procedure was used assuming the volume of the extract was approximately 80 ml. The extract was combined with 20 ml of a 0.88% saline solution (potassium chloride and deionized water) and the solution was swirled, frequently venting the stopcock on the funnel to release the built up gas. The two layers were allowed to completely separate. Two distinct layers formed in the funnel, with the lower layer consisting primarily of a mixture of methylene chloride and the fatty acids and their derivatives. The upper layer is primarily a mixture of water and methanol. The lower layer was drained and collected in a prepped collection bottle. The upper layer that remained in the funnel was drained off as waste. The methylene chloride layer was poured into the separatory funnel and 10 ml of

the potassium chloride solution and 10 ml of methanol added to the separatory funnel. The funnel was swirled again and the stopper was removed periodically to release any built up gas. The lower layer was collected in a prepped collection bottle and the upper was discarded as waste. The lower layer was combined with anhydrous sodium sulfate and stored in the freezer overnight.

Sample evaporation and lipid determination

The sample that was backwashed was removed from the freezer the following day and transferred to a Turbovap tube to concentrate the sample to about 10 ml. The sample was transferred to a 10 ml volumetric flask, and the volume was adjusted to 10 ml with methylene chloride. As necessary the sample was run under nitrogen until the proper volume was obtained.

To determine the total organics extracted, aluminum pans were prepped, labeled, and weighed. An aliquot of one to two ml was then placed into each pan. The sample was allowed to evaporate for a period of several hours, at which time, the pan was weighed again. The final weight was subtracted from the original to calculate the total organics extracted, which was a surrogate for the lipid concentration for our samples.

Thin layer chromatography

Thin layer chromatography (TLC) was used in an attempt to separate the various fatty acid precursor species from the fatty acids and from each other. TLC plates were placed in the muffle furnace for one hour at 110°C. After the plates were cooled, they were placed in the developing chamber that contained toluene, and remained in the chamber until the toluene rose

drawn to the top of the plate by capillary action. Toluene from the plates was first allowed to air out in the hood and then the plates were allowed to dry in a dessicator overnight.

Approximately 200 µl, which equates to 20 to 30 µg of sample, was applied to the bottom of the plate. The plate was then placed in a beaker and the toluene was drawn to the top of the band and then evaporated. The process of drawing toluene to the top of the band was repeated two more times to focus the band to a narrow band.

The plate was then placed in the developing chamber and toluene was allowed to be drawn to within one inch of the top of the plate. Plates were then removed from the chamber and allowed to dry in the hood. Plates were sprayed with a fluorescent dye (2-7-dichloroflourescin). Plates were illuminated with ultraviolet (UV) light and bands were marked with a razor. Bands were then scraped with the razor blade onto aluminum foil sheets prepared with acetone and methylene chloride. Scrapings were gathered from each band separately and placed into a test tube with methylene chloride. Test tubes were placed on the inverter for an hour. Solvent was removed and placed into a prepared test tube with the solid material being discarded as hazardous waste.

Despite several attempts using TLC to separate the fatty acid precursors for oxidation using Jones reagent, no fatty acid precursors were present in the bands. Therefore, this method was discontinued and not used for any of the samples. Instead, fatty acid precursor standards were obtained, e.g. C16:0 dimethyl acetal (DMA), C17:0 DMA, C18:0 DMA, and compared to the output from the mass spectrometer to determine if fatty acid precursors were present in the samples.

Oxidation of fatty acid precursors

The oxidation step will reduce the proportion of polyunsaturated fatty acids (PUFAs) in the mixture if FAMEs are not separated from fatty alcohols prior to the oxidation step. Thin layer chromatography (TLC) normally separates these compounds; however, we did not achieve separation of the bands when TLC was attempted. Instead, an aliquot of sample was analyzed on the GC/MS, and an aliquot was oxidized. Because PUFAs are rarely present in wax esters, the resulting chromatograms could be compared to determine the contribution of fatty alcohols to the overall signature.

Approximately one to two ml, which equates to 40 to 60 μg of sample, was placed into a test tube. The sample was evaporated with nitrogen and had 250 μl of Jones reagent (13.5 g CrO_3 + 6.4 ml H_2SO_4 with enough distilled water to prepare 50-ml of Jones reagent) and two ml of acetone added. The test tube was vortexed for one minute and allowed to stand for 10 minutes. After 10 minutes, one ml of deionized water and 2 ml of hexane were added and the test tube was vortexed for one minute. The contents of the test tube were added to a separatory funnel, and the test tube was washed with hexane two more times to obtain as much of the sample in the test tube as possible. Once the sample separated into two distinct layers in the separatory funnel, the lower layer was drained into the original test tube and the top layer was drained into a new test tube. Hexane was added to the original test tube, and the process of draining the layers was repeated as above two more times. Anhydrous sodium sulfate was added to the new test tube containing the hexane and sample and allowed to sit in the freezer overnight. The following day, samples were removed from the anhydrous sodium sulfate and were derivatized.

Derivatization with boron triflouride (BF$_3$)

Approximately two millimeters of a 20 to 30 µg/ml solution was used for the Derivatization process and evaporated to dryness. A 0.5 molar solution of sodium hydroxide (NaOH) and methanol was prepared. The sample was combined with 1.5 ml of the methanolic NaOH solution in a test tube and placed on the heater at 100°C for five minutes. The test tube was removed from heat and allowed to cool in a beaker of water. The prepared aliquot of boron triflouride and methanol (BF$_3$) along with 250-µl 2,2-dimethoxypropane was added to each test tube and vortex for 1 minute. Three millimeters of saturated sodium chloride solution was added to the sample and was placed on the inverter for one minute. Samples were removed from the inverter and placed onto a centrifuge for two minutes at 1300 rpm. The liquid portion of the sample was removed from the solid collected at the bottom and place into a new test tube with anhydrous sodium sulfate for 20 minutes. The liquid portion was removed from the anhydrous sodium sulfate and placed into a new test tube. This method was also conducted using BF$_3$ without NaOH at 60°C and 100°C.

Derivatization with Hilditch reagent

Hilditch reagent was used to derivatize most of the samples in this study. This method uses sulfuric acid (H$_2$SO$_4$) and methanol to perform the transesterification step. Due to the simplicity, effectiveness, and lower costs associated with this method, it was deemed to be a more favorable technique.

Methanol was placed in a 100ml volumetric flask and had two to three grams of sodium sulfate added to the flask. Methanol was allowed to dry overnight at room temperature. On the day of the experiment, methanol was placed into a clean Erlenmeyer flask along with 1.5 ml

concentrated H_2SO_4. The flask was swirled to evenly distribute the acid. This solution remained viable for one week and excess solution was stored in a stoppered flask at room temperature.

Two ml of sample was evaporated to dryness under a steady stream of nitrogen in a clean 10 ml screw top test tube. Thee ml of Hilditch reagent was added and the test tube was vortexed for 10 seconds. Test tubes were placed on the heat block at 62 C for 1 hr. Samples were cooled to room temperature (time: approximately 30 min; alternatively run cool tap water over the tubes for 1 min). Three ml of hexane was added and the sample was vortexed. Three ml saturated NaCl was added and the test tube was gently swirled. Lastly, the test tube was centrifuged at 1300 rpm for two min. The top layer was removed and placed in a second tube that contained approximately 0.5 g anhydrous sodium sulfate. The aqueous bottom layer was washed two more times with one ml hexane, each time removing the top layer and adding it to the new test tube. The pooled FAME mixture was allowed to sit at room temperature for 20 min on anhydrous sodium sulfate. The maximum amount of Hexane solution of fatty acid methyl esters was carefully decanted from anhydrous sodium sulfate, and it was placed into a clean test tube.

Silver ion chromatography

Silver ion chromatography was performed in order to verify the elution order of cis and trans fatty acids on the gas chromatograph - mass spectrometer. The only difference between some monounsaturated fatty acids is the orientation of the double bond, e.g. oleic acid (C18:1n9c) and elaidic acid (C18:1n9t). Several solutions need to be prepared prior to the initiation of silver ion chromatography (these solutions are prepared by volume and the volume in parentheses is the elution volume needed for each sample): 1) 96:4 hexane:acetone (six ml), 2) 75:25 hexane:acetone (4 ml), 3) 25:75 hexane:acetone (5 ml), 4) 95:5 acetone:acetonitrile (5 ml),

5) 93:7 acetone:acetonitrile (5 ml), 6) 88:12 acteone:acetonitrile (5 ml), and 7) 85:15 85:15 acteone:acetonitrile (5 ml).

A Supelco vacuum manifold for SPE cartridges is needed for this process. The flow rate using the vacuum should be about 5ml/minute. The silver ion cartridge is attached to the top port of the manifold. A test tube was attached to the manifold and five ml of acetone was drawn through the manifold. This process was repeated with five ml of hexane drawn through the cartridge. A prepped test tube was attached to the bottom of the manifold to collect the sample as it is drawn through the manifold. The sample was added, and the first mixture was drawn through the cartridge after the sample was added. When the sample was done eluting, the vacuum was turned off and two ml of the solvent mixture was added and the vacuum was turned back on. The test tube was removed and labeled with which fraction with which it was combined. This was repeated for each solution added to the cartridge so that the sample had seven different mixtures added to the cartridge and collected in seven different test tubes. These samples were run on the gas chromatograph - mass spectrometer to determine which fatty acids were present and to verify their elution times and order.

Column cleanup

Each sample will require 20 ml of solvent mixture containing 95 hexane: 5 diethyl ether (v:v). Exeter glass chromatography/drying columns (22 mm x 275 mm) were prepped for each sample prior to the column cleanup. Silica gel was the adsorbent required for the cleanup process, and it was activated at $700^{o}C$ in a muffle furnace for 24 hours. Once all materials were prepared, a glass wool plug was inserted into the column, followed by a two cm layer of silica gel, and lastly a one cm layer of anhydrous sodium sulfate was added. The column was

conditioned with 20 ml of hexane prior to sample addition. A Turbovap tube was then placed under the glass column to collect the cleaned sample. The sample was then added, and eluted with 20 ml of the hexane:diethyl ether solution. The sample was then concentrated to a smaller known volume using the same procedure as the backwashing step.

GLC standard mixture

In this experiment, a group of standards were combined to create a GLC standard mixture of target fatty acid methyl esters. Known standards from Nu-Chek Prep (GLC MIX 85, 411, 642, 632, Elysian, MN) were combined along with stearidonic acid (C18:4n3, Cayman Chemical, Ann Arbor, MI) to make a total FAME standard solution of 12.25 µg/ml. Matreya 25 FAME mixture (Pleasant Gap, PA) was used to obtain the retention time for C20:1n7. Restek cis/trans FAME mixture (Bellefonte, PA) was used to obtain retention times for C18:1n9 trans and C18:2n6 trans.

Running samples on the gas chromatograph/mass spectrometer (GC/MS)

The GC/MS was used to identify and quantify fatty acid methyl esters in each sample. FAMEs were injected into a fused capillary column, DB-23 60m length X 0.250mm OD X 0.25µm film thickness (Agilent, Santa Clara, CA) using an autosampler (Agilent 7683 Series Auto Sampler). Hydrogen was used as a carrier gas. The temperature program was as follows: hold at an initial temp of $50^{o}C$ for 2 min, hold at $150^{o}C$ after ramping at $20^{o}C \cdot min^{-1}$, hold at $215^{o}C$ after ramping at $1.25^{o}C \cdot min^{-1}$.

The GC/MS tracks the select ion signature of each compound and was used to verify the identity of each peak. There were peaks which consisted of more than one fatty acid and it was

impossible to determine which fatty acid(s) constituted the peaks. Therefore, a term fatty acid "complex" was assigned to this peak. The complex refers to which fatty acids could be responsible for the peak, e.g. C22:3n3 and C22:5n6.

The goal for the sample concentration to be run on the GC/MS was approximately 100 µg/ml. After the sample was concentrated after column cleanup, it was diluted as needed to reach this concentration and added to a prepped GC vial. Prior to the analyses of the samples, calibration standards were run to verify the elution order on the column. A group of samples was run in batches of approximately 15 to 20. After four samples were run, a GLC standard mix was run to verify that elution times were not shifting during the course of this batch run. The chromatograms were obtained from the MS using the Agilent ChemStation software, and the peaks present were compared to the known standards for identification. The area under the peaks was quantified by this program allowing proportions of each fatty present to be calculated, which became the fatty acid signature of each sample.

The Autotune function was performed on the GC/MS prior to every batch run (20-26 samples). This function measures the relative abundance of three mass ions, 69.00, 219.00, and 502.00. Once the relative abundance of mass ion 219.00 dropped below 90.00, the mass spectrometer source was cleaned. Also prior to each batch run, the injector septum was changed. The column, injector inlet, and seal were changed as needed during the course of this experiment. After each column change, the column was allowed to condition before any samples were run. Standards were run through the instrument several times to ensure the correct retention times were being recorded.

Curriculum vitae

Edward McGinley

236 S. H_IGH_ S_T._ A_PT._ 2, M_ORGANTOWN_, WV 26501
P_HONE_: 484-358-5389 • _EMAIL_: _ED_MCGINLEY_11@_YAHOO_._COM_

Education

Ph.D. Candidate, (ABD) Wildlife and Fisheries   Started May 2008
Division of Forestry and Natural Resources   Expected Graduation: August 2012
West Virginia University, Morgantown, WV
Dissertation: *Estimation of striped bass diets using fatty acid analysis*
GPA: 3.88

M.S. Wildlife and Fisheries   Graduated: May 2008
Department of Biology
Frostburg State University, Frostburg, MD
     Thesis: *The effects of sedimentation on the seasonal feeding of mottled sculpin*
     GPA: 3.88

B.S. Biology   Graduated: May 2004
Department of Biology
Saint Francis University, Loretto, PA
     GPA: 3.26

Research Experience

West Virginia University   May 2008 – present
Graduate student, Department of Wildlife and Fisheries
- Dr. Kyle Hartman, 310A Percival Hall, PO Box 6125, Morgantown, WV 26506, 304-293-4797
  Use fatty acid signature analysis to determine striped bass diets. Run tank experiments to establish the effects of size and mixed diets upon the fatty acid signature. Evaluate the timeline for striped bass incorporation of a new fatty acid signature.

Research Assistant   July 2012
- Dr. Kyle Hartman
  In July 2012, I will be joining Dr. Hartman for a research expedition to Mongolia. The goal of our trip is to measure bioenergetics parameters for several fish species in Lake Hovsgol. This area is under extreme thermal stress from climate change and offers an undisturbed system to measure how increases in air temperature affects fish metabolism.

Frostburg State University   September 2005 – May 2008
Graduate student, Department of Biology
- Drs. Richard Raesly and William Seddon, 201 Compton Science Center, 101 Braddock Road Frostburg, MD 21532, 301-687-4166
  Studied diets of mottled sculpin in western Maryland headwater streams. Quantified the effects of sediment loading on diets throughout the year.

Field Technician: May 2006-August 2006
- Gabriel Strain, 322 Percival Hall, Morgantown, WV 26506
  Assisted in comparing methods for sampling salamanders in southern Maryland watersheds. Duties included electrofishing, trap assemblage, and identification of salamanders.

Rutgers University   May 2006 – August 2006
Field Technician: Haskins Shellfish Research Laboratory
- Greg DeBrosse, Haskin Shellfish Research Laboratory, 6959 Miller Avenue, Port Norris, NJ 08349, 609-463-0633
  Rearing of adult Asiatic oyster species, and maintenance of system housing them. Maintenance and care for adult Atlantic oyster raised on tidal flats of the Delaware Bay, NJ.

Professional Experience

Maryland Sea Grant Ecosystem based fisheries management: Food Web Team
Quantitative support staff   March 2009 – present
- Maryland Sea Grant assembled this group in order to modify current fisheries management plans by implementing a multispecies approach. The specific goal of the food web team is to establish benchmarks to assess the health of striped bass in the Chesapeake Bay. These benchmarks are based upon data relating to striped bass, and also to their prey items including Atlantic menhaden and plankton.

Teaching Experience

West Virginia University
Morgantown, WV: Department of Wildlife and Fisheries
Lecturer -Responsible for developing and teaching the class
- Wildlife Management 493N – Environmental Physiology   Fall 2011

Graduate student instructor –Assisted in grading and teaching the class
- Wildlife Management 314 – Marine Ecology, Dr. Kyle Hartman   May 2011
- University 194X – Finned Friends in Society, Dr. Kyle Hartman   Fall 2009

Frostburg State University
Frostburg, MD: Department of Biology
Graduate student instructor - Responsible for teaching the lab and assisted in teaching the lecture
- Biology 201 – Anatomy and Physiology I, Dr. David Morton   Fall 2005-2007
- Biology 202 – Anatomy and Physiology II, Dr. David Morton   Spring 2006-2008

Presentations

Invited seminars
- McGinley, E.J. November 2010. *Organized chaos: my life as a grad student.* Frostburg State University Career Day, Frostburg, MD.
- McGinley, E.J., R. Raesly, and W. Seddon. October 2008. *The effects of sedimentation on the seasonal feeding of mottled sculpin.* James J. Howard Marine Science Lab, Sandy Hook, NJ.

Submitted professional presentations
- McGinley, E.J. and K.J. Hartman, January 2012. *A comparison of striped bass liver and belly flap fatty acid signatures.* American Fisheries Society Southern Division Meeting: Biloxi, MI
- McGinley, E.J. and K.J. Hartman., September 2011. *Which Tissues Provide the Best Results In Evaluating Striped Bass Diet Using Fatty Acid Signatures?* American Fisheries Society national meeting: Seattle, WA.
- McGinley, E.J., K.J. Hartman, and J. Jacobs. November 2010. *Spatial and temporal patterns in select striped bass prey items.* American Fisheries Society student colloquium: Virginia Tech University, Blacksburg, VA
- McGinley, E.J., K.J. Hartman, and J. Jacobs. September 2010. *The effect of size on striped bass fatty acid signature incorporation.* American Fisheries Society national meeting: Pittsburgh, PA
- McGinley, E.J., K.J. Hartman, J. Jacobs, and J. Councilman. March 2010. *Spatial and temporal patterns in select striped bass prey items.* American Fisheries Society Tidewater Chapter meeting: Annapolis, MD.
- McGinley, E.J., K.J. Hartman, and J. Jacobs. March 2010. *Spatial and temporal patterns in select striped bass prey items.* West Virginia University Davis College of Agriculture, Natural Resources and Design and West Virginia Agriculture & Forestry Experiment Station Graduate Research Day: Morgantown, WV.
- McGinley, E.J., K.J. Hartman, and J. Jacobs. August 2009. *Spatial and temporal patterns in select striped bass prey items.* American Fisheries Society national meeting: Nashville, TN.
- McGinley, E.J, R. Raesly, and W. Seddon. February 2008. *The effects of sedimentation on the seasonal feeding of mottled sculpin.* American Fisheries Society Southern Division meeting: Wheeling, WV.

Submitted posters
- Porto, M., A. Anderson, E.J. McGinley, K.J. Hartman, and E. Merriam. September 2011. *The development of a brown bullhead bioenergetics model.* Seattle, WA.
- McGinley, E.J. April 2011. *Spatial and temporal patterns in select striped bass prey items.* West Virginia University Davis College of Agriculture, Natural Resources and Design and West Virginia Agriculture & Forestry Experiment Station Graduate Research Day: Morgantown, WV.
- McGinley, E.J., R. Raesly, and W. Seddon. July 2007. *The effects of sedimentation on the seasonal feeding of mottled sculpin.* American Society of Ichthyologists and Herpetologists: St. Louis, MO

Publications

Articles
- McGinley, E.J., R. Raesly, and W. Seddon. In revision. *The effects of sedimentation on the seasonal feeding of mottled sculpin.* American Midland Naturalist
- McGinley, E.J., K.J. Hartman, A. Deshpande, and J. Jacobs. In review. *Spatial and temporal patterns in select striped bass prey items.* Northeast Fisheries Science Center
- McGinley, E.J., K.J. Hartman, A. Deshpande, and J. Jacobs. In review. *A comparison between liver and adipose tissue fatty acid incorporation in striped bass.* Northeast Fisheries Science Center

Master's thesis
- McGinley, E.J., R. Raesly, and W. Seddon. 2008. *The effects of sedimentation on the seasonal feeding of mottled sculpin.* Thesis, Frostburg State University

Professional memberships and Leadership

American Fisheries Society
- Parent society 2007 – present, Southern Division representative for the Student Subsection on Education (2008)
- West Virginia State Chapter 2008 – present
- West Virginia University student chapter 2008 – present, President (2009)
- Estuaries Section 2009 – present
- Tidewater Chapter 2010 – present

Awards

- 2009 American Fisheries Society Estuaries Section Travel Award ($500)

Laboratory techniques

- Accelerated solvent extractor (ASE)
- Gas chromatograph and mass spectrometer (GC/MS)

Certifications

- PADI Open water SCUBA
- CPR/First AID

Computer skills

- Proficient in Microsoft Word, Excel, and PowerPoint
- Statistical package R
- Working knowledge of Arc-GIS
- Proficient in GC/MS software package ChemStation

References

- Dr. Kyle Hartman, West Virginia University, 310A Percival Hall, PO Box 6125, Morgantown, WV 26506, 304-293-4797, Hartman@wvu.edu

- Dr. Patricia Mazik, USGS Coop Office, West Virginia University, 333 Percival Hall, PO Box 6108, Morgantown, WV, 26506, 304-293-4943, pmazik@wvu.edu

- Dr. John Jacobs, NOAA/NOS/Cooperative Oxford Laboratory, Oxford, MD 21654 410-226-5193, John.Jacobs@noaa.gov