

Abstract

Evaluating spatial and temporal overlap between larval alosines and potential zooplankton prey in lower Roanoke River and Albemarle Sound, North Carolina

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Spatial and temporal overlap between zooplankton and larval American shad (*Alosa sapidissima*), river herring (alewife *A. pseudoharengus* and blueback herring *A. aestivalis*), and hickory shad (*A. mediocris*) was evaluated in lower Roanoke River and Albemarle Sound, North Carolina. Zooplankton abundances in this system have historically been lower than those found in other coastal river systems. It was hypothesized food limitation during the early life history of alosines was contributing to recruitment failure. Zooplankton and ichthyoplankton samples were collected concurrently March through June 2008-09 at 19 stations, within three areas: River, Delta, and Sound. Significant spatial and temporal differences were observed for alosine abundances. Abundances (number/100m³ ± SD) were significantly higher in 2009 (30.8 ± 149.8), than in 2008 (4.1 ± 20.9). Across both years, River (21.0 ± 127.6) alosine abundances were significantly higher than those in Delta (7.4 ± 35.4) and Sound (4.6 ± 24.8). Zooplankton abundances were higher than observed in previous studies and did not differ significantly between years. Zooplankton abundances exhibited the opposite spatial trend of alosines with significantly higher abundances (number/m³ ± SD) observed in the Sound (16,547 ± 14,678) than in the River (4,934 ± 3,806) and Delta (4,647 ± 2,846). Differences in zooplankton composition were evaluated using analysis of similarity. Composition in the Sound significantly differed

from the River and Delta. Canonical correspondence analysis explored the relationship between zooplankton and the environment and found that some differences in composition could be explained by salinity preferences of zooplankton taxa. Zooplankton size distribution was evaluated and the most common taxa segregated into two groups based on size. Rotifers and copepod nauplii comprised the small size group and Daphniidae, Bosminidae, calanoid copepods, and cyclopoid copepods composed the larger size class. Mouth gape models were developed for each alosine species and used to estimate maximum prey size at first feeding. At first feeding, alewives, blueback herring, and hickory shad are primarily able to consume copepod nauplii and rotifers. Larval American shad are larger and have a wider potential prey breadth including Bosminidae, cyclopoid copepods, copepod nauplii and rotifers, at first feeding. During both years, there was a high amount of overlap between larval alosines and size-appropriate zooplankton, suggesting larval alosines in this system are not food limited.

EVALUATING SPATIAL AND TEMPORAL OVERLAP BETWEEN LARVAL ALOSINES
AND POTENTIAL ZOOPLANKTON PREY IN LOWER ROANOKE RIVER AND
ALBEMARLE SOUND, NORTH CAROLINA

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CHAPTER 1: INTRODUCTION

Roanoke River and Albemarle Sound

Roanoke River basin is the largest basin of any North Carolina estuary, encompassing 25,035 km² (Konrad 1998; NCDENR 2000). Roanoke River originates in the Blue Ridge Mountains of Virginia and flows southeast, extending approximately 660 km between its headwaters to where it empties into Albemarle Sound, North Carolina (Konrad 1998; Pearsall et al. 2005). Roanoke and Chowan Rivers are the two main tributaries emptying into Albemarle Sound. Roanoke River accounts for over 50% of the freshwater input into Albemarle Sound while the Chowan River accounts for approximately 25% (Gray and Copeland 1989). Roanoke River is one of the largest alluvial rivers on the East Coast of the United States. The lower region below the fall line is surrounded by bottomland hardwood floodplain forests and is the largest and least fragmented ecosystem of this type in the mid-Atlantic (NCDENR 2000; Pearsall et al. 2005).

Between 1951 and 1996, seven dams were built to provide both flood control and hydroelectric power generation. Roanoke Rapids Dam is located the furthest downstream at river kilometer (RKM) 220 and directly regulates the flow of lower portions of the river (Konrad 1998; Pearsall et al. 2005). The U.S. Army Corp of Engineers and North Carolina regulatory agencies have placed restrictions on when maximum discharge or peaking for hydroelectric power generation can occur. From April 1st to June 15th, the peak spawning period for striped bass, the amount of allowable maximum discharge is reduced and water releases are scheduled to meet flow targets for spawning (Manooch and Rulifson, 1989; Pearsall et al. 2005). Peak flow is also restricted during warm weather, as regulated by the betterment plan. This plan was

instituted after a large fish kill in the lower Roanoke River in the summer of 2005. Discharge from the dam was reduced dramatically and resulted in a large amount of drainage of hypoxic water from riparian wetlands into the river. Discharge is now regulated to maintain high dissolved oxygen levels (NCDENR 2000). Releases are gradually reduced to prevent hypoxic swamp water drainage in the main stem of the river. During non-flood control operations, restrictions are lifted and maximum discharge occurs during times of peak energy consumption (Pearsall et al. 2005).

Albemarle Sound is a shallow estuary with mean depth < 5 m and is part of the Albemarle-Pamlico Estuarine System (APES), which is composed of broad, shallow, drowned river valleys. APES is the second largest estuary and the largest lagoonal estuary in the United States. Pamlico Sound and Albemarle Sound are the two main basins in this system. Albemarle Sound is the northern most basin and is separated from Pamlico Sound by Croatan and Roanoke Sounds (Gray and Copeland 1989). The Outer Banks form a barrier separating Albemarle Sound from the Atlantic Ocean. Oregon Inlet is located south of Albemarle Sound and is the only connection between Albemarle Sound and the Atlantic Ocean. This limited saltwater intrusion combined with high freshwater input from several rivers results in Albemarle Sound having salinity values < 5 ppt. (Copeland et al. 1983; Pearsall et al. 2005). The Outer Banks also protects Albemarle Sound from gravitational tides, with water circulation being primarily wind driven (Copeland et al. 1983).

Alosine biology and life history

Blueback herring (*Alosa aestivalis*), alewives (*A. pseudoharengus*), American shad (*A. sapidissima*) and hickory shad (*A. mediocris*) are schooling fish belonging to the family

Clupeidae. “Alosines” will be used when collectively referring to all four species. As adults, alosines are characterized as being silver with dark blue or green backs, lacking hard spines, and having compressed bodies that form sharp keels along their ventral midline (Robins and Ray 1986). Juveniles and adults are found in a variety of habitats, including coastal ocean waters, rivers, and freshwater portions of estuaries. Alosine eggs and larvae are found only in systems with significant levels of freshwater input (Able and Fahay 1998).

Alosines are distributed along the east coast of North America. Historical blueback herring distribution ranges from Cape Breton, Nova Scotia south to St. Johns River, Florida. Occasional reports have surfaced of blueback herring being found as far south as Halifax River, Florida (Greene et al. 2009). Landlocked populations have been reported in Clayton Lake, Virginia (Klauda et al. 1991). They are most abundant in the Mid and South Atlantic Bights (Loesch and Lund 1977; Able and Fahay 1998). Historical alewife distribution extends from the Gulf of St. Lawrence to South Carolina. Recent surveys have suggested that alewives are no longer found south of North Carolina (Greene et al. 2009). There are also populations in the Great Lakes and completely landlocked populations in New York. They are most abundant in the region between the Gulf of Maine and Chesapeake Bay. In areas of overlap between alewife and blueback herring distribution, alewives are more abundant in the northern portion, while blueback herring are dominant in the southern region (Greene et al. 2009). Alewives and blueback herring are often collectively referred to as river herring, because of their similar appearances and overlapping distributions (Rulifson 1994). American shad distribution ranges from Gulf of St. Lawrence to St. Johns River, Florida (Winslow 1994; Able and Fahay 1998). They were introduced on the Pacific coast of North America and currently range from Cook Inlet, Alaska to Baja California. Landlocked populations occur on the west coast, but have not

been reported on the Atlantic coast (Greene et al. 2009). Hickory shad distribution extends from Bay of Fundy south to St. Johns River, Florida. They are rarely observed north of Cape Cod, Massachusetts (Able and Fahay 1998; Batsavage and Rulifson 1998).

Maturation rates differ among alosine species and within species geographically along a north south cline. Alewife and blueback herring males mature between ages 3 and 4 while the females mature later between 4 and 6 years (Able and Fahay 1998). American shad males reach maturity between ages 3 and 5 years and females between ages 4 and 6 years (Able and Fahay 1998; Bilkovic et al. 2002). The Albemarle Sound acts as a transition zone for American shad. In this system, American shad life history patterns are more similar to northern stocks than those found in the south. American shad in the Albemarle Sound mature at later ages than stocks in other North Carolina systems (ASMFC 2007; Greene et al. 2009). No geographical differences have been observed in hickory shad maturation rates. In Roanoke River, North Carolina and St. Johns River, Florida, hickory shad males and females mature between 2-5 years (Batsavage and Rulifson 1998; Harris et. al. 2007). The distribution and early life histories for each alosine species is summarized in Table 1.1.

All four species are anadromous and migrate inshore to spawn beginning late winter-early spring. Alewives, blueback herring, and American shad return to their natal rivers to spawn, but there is no evidence to support this with hickory shad (Green et al. 2009). Optimum water temperature range for blueback herring spawning is between 21-24°C, but they have been documented spawning in temperatures as low as 14°C. Spawning occurs in both lotic and lentic waters over hard substrates, but they avoid areas with standing water (Walsh et al. 2005; Greene et al. 2009). Alewives begin spawning in lentic waters when temperature is between 13 and

15°C. They spawn over a variety of substrates including gravel, sand, detritus, and submerged vegetation (O'Connell and Angermeier 1997; Able and Fahay 1998; Walsh et al. 2005). In areas where alewives and blueback herring have overlapping distributions, alewives begin spawning in late February; 3-4 weeks earlier than blueback herring which begin spawning in late March (O'Connell and Angermeier 1997; Able and Fahay 1998). In the sympatric range, blueback herring and alewives utilize different spawning habitats. Blueback herring do not migrate as far upstream as alewives. Blueback herring spawn predominately in the main-stream flow, while alewives select shorebank eddies and deep pools for spawning. (Able and Fahay 1998; Greene et al. 2009). American shad spawn when water temperatures are between 12 and 20°C, typically between March and early June (Able and Fahay 1998; Bilkovic et al. 2002). Hickory shad spawning occurs in water temperatures ranging from 8 to 22°C, but peaks when water temperatures are between 12 to 19°C. In Roanoke River, North Carolina, hickory shad were observed spawning in water < 1 m deep with moderate to high velocity, over substrates containing cobble, gravel, and sand (Greene et al. 2009; Harris and Hightower 2010).

During their early life histories, alewives use the low salinity waters of their natal rivers and estuaries as nursery habitat. Egg and larval development occur while they are in these rivers and estuaries. As juveniles, alewives, blueback herring, and American shad initially stay in these nursery areas then migrate to the Atlantic Ocean during autumn of their first year. Hickory shad juveniles exhibit a variety of migration behaviors; some remain in estuarine waters until fall, while others migrate directly to ocean waters and spend no time in freshwater nurseries (Able and Fahay 1998, Greene et al. 2009).

Fishery resources

Managing and restoring alosine stocks are currently of high importance to many state and federal U.S. and Canadian agencies (Greene et al. 2009). Alosines have historically been important in Albemarle Sound, North Carolina. American shad and river herring once supported a variety of fisheries, including large drift gill net, stake gill net, anchored gill net, pound net, haul seine, bow net, fish wheel, and hook and line (Winslow 1994). They were one of the first fishing industries in North Carolina and became established by the 1770's. Their oily flesh allowed for the fish to be salt preserved without refrigeration and shipped to far away markets including the British West Indies, Azores, Canary Islands, southern Europe, and throughout the colonies. The market for these fish expanded as advances in transportation were made. When ice became available during the late 1800's, the number of markets being charged premium prices for fresh American shad increased (Hightower et al. 1996). Hickory shad were not as commercially important as American shad and river herring and have only supported minor fisheries, because the quality of their meat is considered inferior to American shad and river herring (Greene et al. 2009).

The U.S. Fishery Commission began recording landing data in 1887. Coast-wide peak American shad landings are identified as occurring in 1897, with recorded landings of 4 million kg. Limburg and Waldman (2009) found that American shad landings were higher and peaked earlier in 1832, when data fish house records are included. Regardless of which baseline is used, it is agreed there has been a dramatic decline and their landings, are now below 1 million kg (Hightower et al. 1996; ASMFC 2007). A coast-wide American shad stock assessment documented stocks are at all time lows and do not appear to be recovering. In Albemarle Sound, they are a species of concern (ASMFC 2007; NCDMF 2010). In North Carolina, river herring

landings remained relatively stable ranging from 6-7 million kg until their peak in the 1970's, when landings exceeded 8 million kg. Since the 1970's, there has been a steady decrease with landings falling below 1 million kg (Rulifson 1994; Winslow 1994; Hightower et al. 1996). River herring stocks in Albemarle Sound are depleted (NCDMF 2010). Hickory shad status in North Carolina is currently unknown, but coast wide landings suggest populations are viable. There was a slight increase in North Carolina landings in 2009, but landings are still slightly below the 10 year average (Taylor et al. 2009; NCDMF 2010).

Currently in North Carolina, American shad and hickory shad are managed under the ASMFC shad and herring fishery management plan. In 2004, the American shad ocean-intercept fishery was closed. American shad and hickory shad fisheries are opened and closed by proclamation from January 1st to April 14th each year. There is no commercial quota for these species; harvest is regulated through restrictions on fishing gears (NCDMF 2008). In an effort to revive American shad stocks, larvae are stocked in several coastal states, with over 8 million larva stocked in the upper reaches of Roanoke River in 2008. (Taylor et al. 2009). In 2007, a statewide moratorium on all river herring harvest was accepted by N.C. Marine Fisheries Commission (NCDMF 2007). Despite these efforts, current American shad and river herring stocks are well below historic levels of abundance and biomass (Taylor et al. 2009).

Early life history

Year class strength and recruitment of fish is strongly correlated to survival during egg and larval development. The availability of food resources is considered one of the more influential factors affecting survival in the larval phase (Cushing 1990; Leggett and DeBlois 1994). In 1914, Hjort developed the critical period hypothesis, which directly links larval

survival with feeding success. Under this hypothesis, there is a critical period, defined as the period of time after yolk sac absorption when a larva is transitioning from endogenous to exogenous feeding. Year class strength is dependent on successful first feeding during this critical period. If there is not a temporal overlap between larval fish and their prey, many larvae will not have a successful first feeding and will starve. There will be a high mortality rate and year class strength will suffer (Fig. 1.1). Hjort (1914) hypothesized that larval production was timed to follow phytoplankton production to increase feeding success (Leggett and DeBlois 1994; Houde 2008).

Cushing's match/mismatch hypothesis builds upon Hjort's critical period hypothesis. The match/mismatch hypothesis expands the importance of prey availability and feeding success to include the entire larval period, not just for the first feeding (Cushing 1990). This hypothesis suggests fish spawning occurs at fixed times in both spring and autumn to overlap with peak plankton production. This hypothesis also highlights the importance of larval fish distributions overlapping with zooplankton distributions both temporally and spatially. If larval fish and zooplankton distributions coincide in both time and space, then Cushing defines this as a match. If there is any level of separation, temporally and/or spatially between larvae and zooplankton, then a mismatch has occurred. Larval growth and survival is expected to be higher when a match is occurring, and this leads to an increase in year class strength and recruitment in juvenile and adult populations (Cushing 1990; Leggett and DeBlois 1994).

Zooplankton

The term "plankton" is derived from the Greek word *planoas*, which means to wander. It is used to refer to suspended organisms, with limited locomotion abilities (Johnson and Allen

2005; Miller and Kendall 2009). The planktonic community consists of both primary producers and heterotrophic consumers. Phytoplankton refers to photosynthetic protists and bacteria that act as primary producers, while zooplankton refers to the consumers, consisting of protozoa and animals (Johnson and Allen 2005).

Life history is a common approach for describing zooplankton. Zooplankton that spend their entire life as plankton are called holoplankton. Examples of holoplankton include cladocerans, rotifers, copepods, and jellyfish. Meroplankton are organisms that spend part of their lives in the plankton and then either settle in the benthos or enter the nekton community, including the larval stages of fish, decapods, and bivalves (Lenz 2000; Johnson and Allen 2005).

Several abiotic parameters, such as water temperature, salinity, depth, and current velocity have an impact on zooplankton diversity and distribution on both spatial and temporal scales. In rivers, current velocity, in conjunction with discharge rates from dams has a strong impact on zooplankton. Zooplankton abundance can be diluted by swift currents and high discharge rates. Conversely, if currents are weak and water residence time is high, zooplankton populations can be replenished as zooplankton are transported downstream (Hynes 1970; Akopian et al. 1999; Obertegger et al. 2007; Dickerson et al. 2010). Currents also play a role in zooplankton production in riverine systems. Production is hypothesized to occur in areas of slow moving water, such as backwaters, side channels, and reservoirs created by dams (Hynes 1970).

Salinity is considered the most influential factor affecting zooplankton structure in estuaries (Lenz 2000; Johnson and Allen 2005). Salinity tolerances are species dependent, with some species able to tolerate wide salinity ranges, while others have narrow ranges.

Zooplankton diversity typically decreases as salinity decreases (Johnson and Allen 2005; Hwang

et al. 2010). In freshwater and oligohaline systems, zooplankton composition is usually dominated by rotifers, cladocerans, and copepods. Rotifers dominate numerically, while copepods dominate in terms of biomass (Pace and Orcutt 1981; Thorp and Covich 2001).

Thermal preferences differ among zooplankton taxa and water temperature is considered to be equally important in both rivers and estuaries. Several studies have documented seasonal differences in dominant zooplankton taxa that correspond to differences in water temperature (Hynes 1970; Soetaert and Rijswijk 1993; Kimmel and Roman 2004). While individual parameters, such as water temperature, salinity, and current velocity have a strong influence on zooplankton communities, they do not act in isolation and several environmental factors can act in combination to control zooplankton dynamics (Hynes 1970; Johnson and Allen 2005). Graham and Bollens (2010) observed seasonal differences in zooplankton community structure related to water temperature, but upwelling and freshwater input were also influencing zooplankton populations.

In aquatic systems, zooplankton act as a link between primary producers and higher level consumers (Lenz 2000; Johnson and Allen 2005). Zooplankton feed on phytoplankton and then transfer energy up the food chain as they are consumed (Lenz 2000). Zooplankton are consumed by a variety of aquatic animals, including other zooplankton, planktivorous fishes, benthic filter feeders, and baleen whales (Lenz 2000; Johnson and Allen 2005). Larval fish are one of the main groups that prey upon zooplankton (Johnson and Allen 2005). Zooplankton are the main food source for larval fish as they undergo yolk sac absorption and transition to feeding exogenously (Yufera and Darias 2007; Miller and Kendal 2009).

In larval fish, mouth gape is a limiting factor at the onset of exogenous feeding, restricting the prey size that can be consumed (DeVries et al. 1998; Yufera and Darias 2007). Mouth gape becomes less limiting as fish grow. There is a positive relationship between fish length and mouth gape (DeVries et al. 1998; Puvanendran et al. 2004). Studies have suggested that the width when the mouth is open at a 90° angle is the maximum functional mouth gape. Optimal prey sizes are generally within 30- 50% of the mouth gape (Bremigan and Stein 1994; Turingan et al. 2005; Riley et al. 2009).

Zooplankton abundances in Roanoke River and Albemarle Sound have historically been much lower than those found in other North Carolina river systems. A long term study conducted from 1984-1991 by Rulifson et al. (1993) and a study by Coggins (2005) in 2003 documented abundances that were between 1-2 orders of magnitude lower than other systems (Table 1.2). American shad, river herring, and hickory shad all spawn in Roanoke River and their larvae use this system as nursery habitat (Greene et al. 2009; Harris and Hightower 2010). In this system, American shad are currently listed as a species of concern, river herring stocks are depleted and current hickory shad landings are below the 10 year average (ASMFC 2007; Greene et al. 2009; NCDMF 2010). One possible explanation for failure of these stocks to rebound could be high levels of larval mortality caused by food limitation. Zooplankton abundances are low in this system, increasing the probability of a spatial and/or temporal disconnect between zooplankton and larval alosines.

Research objectives

The purpose of this thesis is to evaluate foraging potential of larval American shad, blueback herring, alewives, and hickory shad in three areas in the lower Roanoke River and

Albemarle Sound, North Carolina. Two main research objectives were addressed to evaluate larval alosine foraging potential and each objective is discussed in separate chapters. Chapter II focuses on zooplankton abundance and composition. The overlap between larval alosines and zooplankton is discussed in Chapter III. Chapter IV summarizes the main findings of this thesis.

In Chapter II, zooplankton abundance and composition in three areas within lower Roanoke River and Albemarle Sound are described. Spatial differences in abundance were evaluated using analysis of variance (ANOVA). PRIMER-E was used to conduct analysis of similarity (ANOSIM) and similarity percentages (SIMPER) analysis of zooplankton composition. ANOSIM was used to evaluate spatial differences in zooplankton composition and the results are visualized using a non-metric multidimensional scaling (NMDS) ordination plot. SIMPER analysis allows for identification of key taxa driving dissimilarities between areas. Multiple analysis of variance (MANOVA) evaluated if differences among areas could be observed when both environmental data and zooplankton abundance and composition data are included in analysis. Canonical correspondence analysis (CCA) evaluated how environmental parameters drive zooplankton patterns in each area. Results from this study were compared to previous work in Roanoke River and Albemarle Sound and long term patterns in zooplankton abundance were evaluated.

Chapter III focuses on the potential of larval alosines to prey upon zooplankton. Abundance and distribution of larval alosines was described. Mouth gape models were developed for each alosine species and from these, optimal prey sizes were estimated. Analysis of covariance (ANCOVA) was used to test if there are differences in mouth gape among the species. Size distribution was modeled for the most numerous zooplankton taxa. Using

abundance data from Chapter II, spatial and temporal overlap between larval alosines and size appropriate zooplankton prey was evaluated. Cross correlations were conducted to evaluate the relationship between larval alosines and zooplankton.

Chapter IV summarizes and highlights the main findings of this research.

Table 1.1. Life history traits for alosines in North America.

	Species			
	Alewife	American shad	Blueback herring	Hickory shad
Distribution	Gulf of St. Lawrence - South Carolina	Gulf of St. Lawrence - St. Johns River, Florida	Nova Scotia - St. Johns River, Florida	Bay of Fundy - St. Johns River, Florida
Maturation	Males: 3-4 years Females: 4-5 years	Males: 3-5 years Females: 4-6 years	Males: 3-4 years Females: 4-5 years	Males: 2-5 years Females: 2-5 years
Spawning dates	Late March - April	March - June	Late April - early May	Late April - early June
Spawning temperature range	13 - 15°C	12 - 20°C	21 - 24°C, but as low as 14°C	8 - 22°C, peaks between 12 - 19°C
Spawning location	Shorebank eddies and deep pools	Main stream	Main stream over hard substrates	Open water, over substrates containing cobble, gravel, and sand
Total length at hatching	3.5 mm	7 - 10 mm	3.1 - 4.2 mm	5.2 - 6.6 mm
Total length at yolk-sac absorption	6.0 mm	9 - 12 mm	6.0 mm	7.0 mm

Table 1.2. Zooplankton abundance (number/m³) in several North Carolina coastal river systems.

Study	System	Sampling period	Mesh size (µm)	Abundance (number/m ³)
Mallin (1991)	Neuse River	May-Dec.	76	32,877
Fulton (1984)	Newport River	All year	76	21,900
Thayer et al. (1974)	Newport River	All year	156	6,200
Birkhead et al. (1979)	Cape Fear River	All year	156	7,450
Winslow et al. (1985)	Chowan River	All year	70	3,423
Rulifson et al. (1993)	Roanoke River	All year	250	327
	Roanoke Delta	All year	250	696
	Albemarle Sound	All year	250	532
Coggins (2005)	Roanoke River	June, Sept., & Nov.	90	892

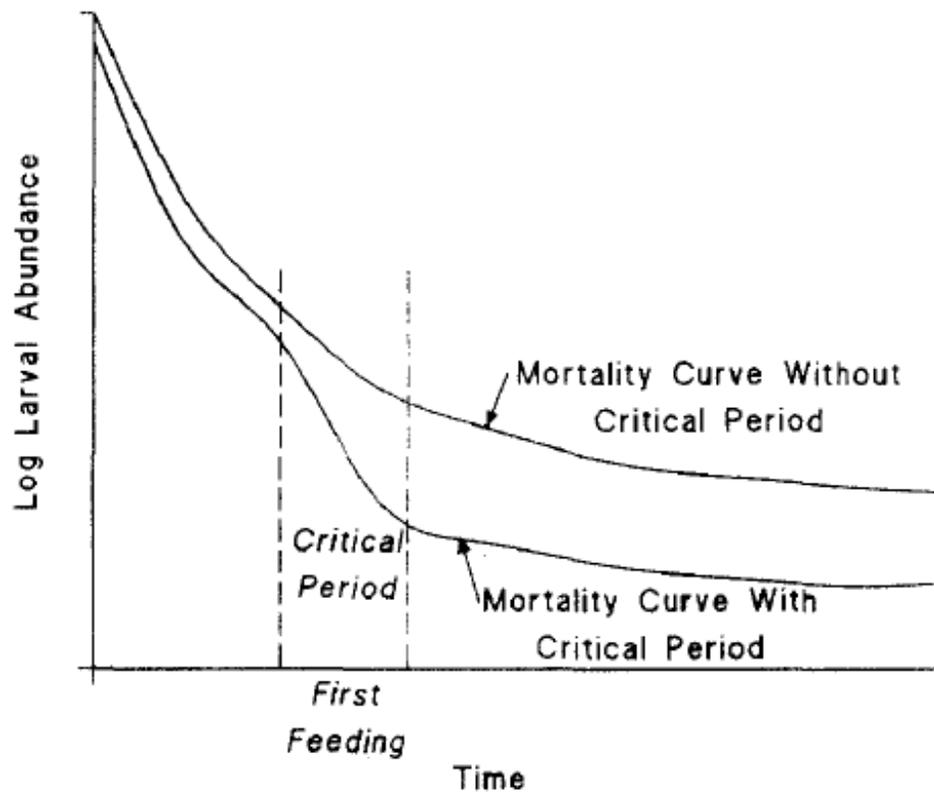


Fig. 1.1. Representation of Hjort's Critical Period Hypothesis (Hjort 1914) showing the hypothesized link between survival during first feeding and year-class strength.

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CHAPTER 2: SPATIAL AND TEMPORAL TRENDS IN ZOOPLANKTON ABUNDANCE
AND COMPOSITION IN LOWER ROANOKE RIVER AND ALBEMARLE SOUND,
NORTH CAROLINA

Abstract

Zooplankton abundance and composition were evaluated in lower Roanoke River and Albemarle Sound, North Carolina. In this system zooplankton abundances have historically been lower than other North Carolina coastal river systems. Samples were collected weekly at 19 stations using a vertical net haul technique, during March-June in 2008 and 2009. The stations were located within three areas identified as River, Delta, and Sound. River is the area furthest upstream with seven stations between RKM 9.5 – 22, scattered throughout the main stem of Roanoke River and its tributaries and distributaries. Delta is the transitional region where the Roanoke, Middle, and Cashie Rivers converge at the Highway 45 Bridge, before diverging and flowing into Albemarle Sound. Sound has six stations in Batchelor Bay, the western portion of Albemarle Sound. Zooplankton abundances (number/m³ ± SD) were not significantly different ($t(95) = -1.47, p = 0.144$) between 2008 (7,214 ± 8,048) and 2009 (9,774 ± 11,967). Spatial differences were observed with abundances in the Sound (16,546 ± 14,678) being significantly higher ($F(2,94) = 12.98, p < 0.001$) than those in both River (4,934 ± 3,806) and Delta (4,647 ± 2,846). Zooplankton composition was dominated by rotifers, cladocerans, and copepods, which accounted for 96% of zooplankton, but the percentage of each varied spatially and temporally. Abundances in this study were significantly higher than those reported in earlier studies.

Differences in zooplankton composition were also observed between this study and previous studies.

Introduction

In aquatic systems, zooplankton act as a link between primary producers and higher level consumers (Lenz 2000; Johnson and Allen 2005). Zooplankton feed upon phytoplankton, aquatic primary producers, and transfer energy up the food chain as they are consumed (Lenz 2000). Zooplankton are consumed by a variety of aquatic animals, including other zooplankton, planktivorous fishes, benthic filter feeders, and baleen whales (Lenz 2000; Johnson and Allen 2005). Larval fish are one of the main groups that prey upon zooplankton (Johnson and Allen 2005). Zooplankton are the main prey resource for larval fish undergoing yolk sac absorption and transitioning to feeding exogenously (Yufera and Darias 2007; Miller and Kendal 2009).

Several abiotic parameters, such as water temperature, salinity, depth, and current velocity have an impact on zooplankton diversity and distribution on both spatial and temporal scales. In rivers, current velocity, in conjunction with discharge rates from dams is believed to have a strong impact on zooplankton. Zooplankton abundances can be diluted by swift currents and high discharge rates. Conversely, if currents are slow and water residence time is high, populations can be replenished as zooplankton are transported downstream (Hynes 1970; Akopian et al. 1999; Obertegger et al. 2007; Dickerson et al. 200). Current velocity also has a role in zooplankton production in riverine systems. Production occurs in areas of slow moving water, such as backwaters, side channels, and reservoirs created by dams (Hynes 1970).

Salinity is considered the most influential factor affecting zooplankton in estuaries (Lenz 2000; Johnson and Allen 2005). Salinity tolerances are species dependent, with some species

able to tolerate wide salinity ranges, while others have narrow ranges. Zooplankton diversity decreases as salinity decreases (Johnson and Allen 2005; Hwang et al. 2010). In freshwater and oligohaline systems, zooplankton composition is usually dominated by rotifers, cladocerans, and copepods. Rotifers dominate numerically, while copepods dominate in terms of biomass (Pace and Orcutt 1981; Thorp and Covich 2001).

Thermal preferences differ among zooplankton taxa and water temperature is considered to be equally important in both rivers and estuaries. Studies encompassing multiple seasons have documented seasonal differences in dominant zooplankton taxa that corresponds to changes in water temperature (Hynes 1970; Soetaert and Rijswijk 1993; Kimmel and Roman 2004). While individual environmental parameters can exert a strong influence on zooplankton communities, several factors can act in combination to control zooplankton dynamics (Hynes 1970; Johnson and Allen 2005). Graham and Bollens (2010) observed seasonal differences in zooplankton community structure related to water temperature, but upwelling and freshwater input were also influencing zooplankton populations.

Zooplankton abundances in Roanoke River and Albemarle Sound have historically been much lower than those found in other North Carolina river systems (Fig 2.1). A long-term study (1984-1991) conducted by Rulifson et al. (1993) and another study by Coggins (2005) in 2003 documented average abundances that did not exceed 900 number/m³ and were 1-2 orders of magnitude lower than other systems (Table 2.1). American shad, river herring (alewife and blueback herring), and hickory shad all spawn in the Roanoke River and their larvae use this system as nursery habitat (Greene et al. 2009; Harris and Hightower 2010). In this system, American shad are currently listed as a species of concern, river herring stocks are depleted and

current hickory shad landings are below the 10 year average (ASMFC 2007; Greene et al. 2009; NCDMF 2010). High levels of starvation during the larval stage may make it difficult for these stocks to recover. Zooplankton abundances are low in this system, increasing the probability of a spatial and/or temporal disconnect between zooplankton and larval alosines.

In this chapter, spatial and temporal variation in zooplankton abundance and composition is evaluated within three areas in lower Roanoke River and Albemarle Sound, North Carolina. Environmental data was analyzed separately and in combination with zooplankton data. Zooplankton abundances from this study were compared to those from Rulifson et al. (1993) and Coggins (2005) to evaluate long term trends in this system. Zooplankton abundance and composition results are used in Chapter 3 to evaluate foraging potential of larval alosines in the three study areas and to determine if all areas are suited to serve as nursery habitat.

Methods

Roanoke River and Albemarle Sound

Roanoke River basin is the largest basin of any North Carolina estuary, encompassing 25,035 km² (Konrad 1998; NCDENR 2000). Roanoke River originates in the Blue Ridge Mountains of Virginia and flows southeast, extending approximately 660 km between its headwaters to where it empties into Albemarle Sound, North Carolina (Konrad 1998; Pearsall et al. 2005). Roanoke and Chowan Rivers are the two main tributaries emptying into Albemarle Sound. Roanoke River accounts for over 50% of the freshwater input into Albemarle Sound (Gray and Copeland 1983). It is one of the largest alluvial rivers on the East Coast. The lower region below the fall line is surrounded by bottomland hardwood floodplain forests and is the

largest and least fragmented ecosystem of this type in the mid-Atlantic (NCDENR 2000; Pearsall et al. 2005).

Albemarle Sound is a shallow estuary with mean depths < 5 m and is part of Albemarle-Pamlico Estuarine System (APES). This system is made up of broad, shallow, drowned river valleys. APES is the second largest estuary and the largest lagoonal estuary in the United States. Pamlico Sound and Albemarle Sound are the two main basins in this system. Albemarle Sound is the northern most basin and is separated from Pamlico Sound by Croatan and Roanoke Sounds (Gray and Copeland 2002). The Outer Banks form a barrier separating Albemarle Sound from the Atlantic Ocean. Oregon Inlet is located south of Albemarle Sound and acts as the only source of saltwater intrusion. This limited saltwater intrusion combined with high freshwater input from several rivers results in Albemarle Sound having salinity values < 5 ppt. (Copeland et al. 1983; Pearsall et al. 2005). The Outer Banks also protects Albemarle Sound from gravitational tides, with water circulation being primarily wind driven (Copeland et al. 1983).

Field collection and data processing

Samples were collected from three areas within the lower Roanoke River and Albemarle Sound, North Carolina. The three sampling areas were classified as River, Delta, and Sound and contained a total of 19 stations. River is the area furthest upstream with seven stations between RKM 9.5 – 22, scattered throughout the main stem of the river and its tributaries and distributaries. Delta is the transitional region between River and Sound where the Roanoke, Middle, and Cashie Rivers converge at the Highway 45 Bridge, before diverging and flowing into the Albemarle Sound. There are two stations in the Roanoke, Middle, and Cashie Rivers, for a total of six stations. The Delta station furthest upstream is located in the Roanoke at RKM

5. Sound has six stations in Batchelor Bay, the western portion of Albemarle Sound. The stations extend 2 – 4 km from the mouths of the Roanoke and Middle Rivers (Fig. 2.2).

Zooplankton samples were collected March through June 2008-09. Sampling was conducted at weekly intervals, and began at sunset. Zooplankton samples were collected using a 3:1 conical net with a 0.5 m opening and 90 μm nitex mesh. The plankton net was deployed using a vertical net haul technique where the net is lowered to the bottom and then pulled vertically through the water column. A preliminary study comparing the catch efficiency of vertical hauls, surface tows, and using a bilge pump to filter water through the net showed no significant difference in species abundance or composition when using vertical hauls or the pumping method, while abundances were significantly lower ($F(2, 45) = 21.49, n = 48, p < 0.001$) using surface tows (K. Riley, ECU, unpublished data). The contents of the net were washed down and condensed into the sample jar and preserved with 5% buffered formalin. In 2009, samples were not collected in the Delta during calendar week 20 and during weeks 12, 16, and 25 in the Sound because of mechanical issues with the boat and inclement weather.

Environmental parameters were recorded at each station during each sampling event. Air temperature ($^{\circ}\text{C}$), wind speed (m/s), and direction were measured using a Skymate Model Sm-18. Surface and bottom water temperatures ($^{\circ}\text{C}$), salinity, conductivity (μS), and dissolved oxygen concentration (mg/L), were measured using a YSI Model 85 Multiparameter Water Quality Meter. A Hanna Model HI 98128 pH meter was used to measure surface pH. Current velocity (m/s) and direction were measured one meter below the surface using a Marsh-McBirney FLO-MATE Portable Velocity Flow Meter, Model 2000.

Daily water discharge rates were obtained from Roanoke Rapids Dam water monitoring gage, located 4.5 km downstream of the dam and 215 km upstream from Albemarle Sound. The gage records hourly discharge rates and river height data and is maintained by U.S. Geological Survey and Dominion Power Company. Precipitation and daily air temperatures (°C) were obtained from a 10-m weather station located at Tidewater Research Station in Plymouth, North Carolina. The State Climate Office of North Carolina maintains and operates the weather station. Data are maintained by the National Climatic Data Center.

For each sampling date, three samples from each area were randomly selected for processing. Subsamples were taken using a Hensen-Stempel pipette. Individuals were counted and identified to the lowest possible taxon using an Olympus Model SZX-ILLD100 stereomicroscope. Zooplankton were identified using taxonomic keys found in Thorp and Covich (2001) and Balcer et al. (1984). Abundance (number/m³) was estimated by dividing total number of zooplankton per sample by the volume of water filtered.

During comparisons of zooplankton abundance and composition from this study and from Rulifson et al. (1993) rotifers and copepod nauplii are excluded from analysis. Rulifson et al. (1993) collected zooplankton using a 250 µm mesh net. Rotifers and copepod nauplii are typically < 200 µm in size and are not efficiently collected in a 250 µm mesh net (Thorp and Covich 2001).

Statistical analyses

An independent samples *t*-test evaluated if zooplankton abundances and environmental parameters differed between sampling years. Differences between abundances from this study and previous studies (Rulifson et al. 1993; Coggins 2005) were also compared using independent

samples *t*-test. Spatial and monthly differences between abundances and abiotic factors were evaluated using a one-way analysis of variance (ANOVA). If the ANOVA was significant, differences were further examined using the Ryan-Einot-Gabriel-Welch (REGWQ) post-hoc test, which holds family wise alpha at 0.05. A one-way multiple analysis of variance (MANOVA) tested whether the three sampling areas were significantly different when both environmental and zooplankton data were included in the analysis. In addition to testing for significant differences among groups, MANOVA also provides a value, Λ , measuring how large the differences are among groups. Λ ranges from 0 to 1, with 0 indicating strong differences (Tabachnick and Fidell 2007). To protect against multicollinearity, a Pearson correlation matrix was conducted using all environmental variables. If a pair of variables had an $r \geq 0.9$, one of the variables was deleted from MANOVA and canonical correspondence analysis (CCA). Unless otherwise noted, all statistical analyses were conducted using SAS 9.2 (SAS Institute, Cary, North Carolina).

Primer-E v6 (Primer-E Ltd, Plymouth, UK) evaluated spatial differences in community structure. Prior to analysis, zooplankton data were fourth-root transformed, with rare species being down-weighted. Data were fourth-root transformed because that transformation down-weights the impact of the most abundant species, while still allowing mid-range species to exert some influence in the calculation of similarity indices (Clarke and Warwick 2001). Non-metric multi-dimensional scaling (NMDS) was used as a visual representation showing similarities within areas based on differences in Bray-Curtis dissimilarity values. The closer two points are located on the ordination plot, the more similar those two points are. One-way analysis of similarity (ANOSIM) tested if there were significant differences in community structure among the three areas. The test statistic for ANOSIM, R , usually ranges from 0 to 1, with 0 indicating

little similarity among groups. Similarity percentages (SIMPER) were used to analyze which taxa are driving dissimilarity among areas. SIMPER procedure decomposes Bray-Curtis dissimilarity values and transforms them into percentage contributions from each taxon. SIMPER also allows for the identification of discriminating taxa, those which consistently contribute to dissimilarity between two areas (Clarke and Warwick 2001; Clarke and Gorley 2006).

Canonical correspondence analysis evaluated the relationship between environmental parameters and zooplankton abundance and composition. Prior to analysis, both data sets were evaluated for normality by assessing kurtosis, skewness, and Shapiro-Wilk values for each parameter. All parameters met the criteria for a normal distribution except for zooplankton abundance and salinity, which were $\log_{10}(n + 1)$ transformed prior to analysis. Precipitation was excluded since data was only available for the entire sampling region and not for each discrete area. CCA was performed using CANOCO 4.5 (ter Braak and Smilauer 2002) and the corresponding biplots were created using CANODRAW for Windows. In a biplot the strength of the vector is reflected in the length of the line. A small angle between vectors indicates a positive correlation, while angles approaching 180° have a strong negative correlation. Angles near 90° are not correlated. The origin of the synthetic gradients (axes) represents the global average for each vector (ter Braak and Verdonschot 1995; Rahel and Jackson 2007).

Results

Environmental data

Environmental data ($n = 97$) are summarized in Table 2.2. Monthly patterns were observed for dissolved oxygen, air and water temperatures. Dissolved oxygen followed the

expected seasonal pattern of being highest in March and decreasing throughout the sampling season. Hypoxic conditions (< 2.0 mg/L) occurred infrequently in late May and June accounting for 1% of bottom dissolved oxygen readings. Dissolved oxygen also varied between years and was significantly higher ($\mu \pm SD$) in 2009 (7.9 ± 2.0) than 2008 (6.5 ± 1.5). Air and water temperatures followed the opposite pattern of dissolved oxygen and were lowest in March and increased through June. Significant yearly differences were observed for pH and wind speed (Table 2.3). In 2008 (7.5 ± 0.2), pH was significantly higher than in 2009 (6.7 ± 0.4). Wind speed was also higher in 2008 (5.2 ± 5.1) than in 2009 (1.7 ± 1.1).

Salinity, current velocity, and wind speed were significantly higher in the Sound (Table 2.4). Mean salinity was 0.1 ± 0.0 in both the River and Delta. Salinity was significantly higher ($F(2,94) = 35.69, n = 97, p < 0.001$) in the Sound, ranging from 0.1 – 2.8 ppt, with a mean value of 0.5 ± 0.6 . The majority of salinity values did not exceed 1.0 ppt. Current velocity was similar in River and Delta, ranging from 0.0 – 0.83 m/s, with a mean velocity in both areas of 0.1 ± 0.1 . Current velocity was significantly higher ($F(2,94) = 3.98, n = 97, p = 0.022$) in the Sound (0.2 ± 0.1), ranging from 0.0 – 1.2 m/s, and currents were most frequently from the west. Generally winds were < 10.0 m/s and were typically from the south or southwest. Mean wind speed in the Sound (6.4 ± 5.5) was significantly higher ($F(2,94) = 13.69, n = 97, p < 0.001$) than River (1.7 ± 2.2) and Delta (2.8 ± 2.8) areas. Mean depth at River (4.8 ± 0.7) stations was significantly higher ($F(2,94) = 85.44, n = 97, p < 0.001$) than Delta (3.2 ± 0.6) and Sound (3.3 ± 0.3) stations (Table 2.4).

Water discharge from Roanoke Rapids Dam exhibited different patterns in 2008 and 2009. In 2008, daily discharge rates from Roanoke Rapids Dam ranged from 64 – 416 m³/s.

Discharge rates peaked from mid-April to June 1st and exceeded 220 m³/s. In response to heavy rains, discharge rates were higher in 2009. Flows peaked twice during 2009, once in late March and again during mid-late June. Discharge rates were > 220 m³/s for 90% of the sampling period and with peaks > 500 m³/s.

Zooplankton abundance and taxonomic composition

Zooplankton abundances were highly variable. Mean zooplankton abundances (number/m³ ± SD) were not significantly different ($t(95) = -1.47, n = 97, p = 0.144$) between 2008 (7,214 ± 8,048) and 2009 (9,774 ± 11,967). Month had a significant effect on zooplankton abundances ($F(3) = 4.93, n = 97, p = 0.003$). When abundances were combined for all areas, abundances in March (13,104 ± 12,654) were significantly higher than those in April (6,848 ± 7,309), May (6,054 ± 7,309), and June (8,118 ± 8,875).

Area also had a significant ($F(2,94) = 12.98, n = 97, p < 0.001$) effect on zooplankton abundances. Sound (16,547 ± 14,678) had significantly higher abundances than the River (4,934 ± 3,806) and Delta (4,647 ± 2,846) areas (Fig. 2.3). No clear temporal patterns emerge in each area, so the years were evaluated separately. Abundances in the Sound were the most variable with the widest range in abundances. Even though overall abundances were significantly higher in the Sound, the lowest observed abundance in any area was in the Sound at week 20 in 2008, with an abundance of 935 ± 496. Highest abundance occurred in week 12, with 33,384 ± 47,621. In 2009, lowest abundances were once again in the later part of the sampling season, occurring at week 21 with abundances of 2,710 ± 466. Peak abundances in the Sound occurred the following week and were the largest observed during the study at 51,816 ± 52,092. Temporal patterns were more consistent in the River. Highest abundances were observed in week 13 in 2008

(13,562 ± 10,797) and 2009 (19,751 ± 25,719). In both years, lowest values were observed in early summer. In 2008, lowest abundances occurred in week 22 (1,380 ± 20) and during week 23 (1,261 ± 1,430) in 2009. In the Delta, the two years exhibited opposite patterns. In 2008, zooplankton peaked in late June (week 26) with an abundance of 10,672 ± 7,901 and was lowest in March during week 13 with an abundance of 997 ± 598. During 2009, zooplankton abundance was highest in week 11 (12,727 ± 4,235) and lowest in week 24 with an abundance of 1,802 ± 940 (Fig. 2.4).

Zooplankton communities were dominated by five taxa: calanoid copepods, cyclopoid copepods, copepod nauplii, rotifers, and cladocerans (Fig. 2.5). Calanoid and cyclopoid taxa include both copepodite and adult life stages. Several families of cladocerans were identified in this study, including Daphniidae, Bosminiidae, Sididae, Chydoridae, and Leptodoridae. These five taxa account for a minimum of 96% of the composition for each area across both years. Some of the less common taxa included ostracods, gammarid amphipods, and harpacticoid copepods. A complete list of all taxa is in Table 2.5.

Zooplankton community structure varied temporally and spatially. Temporal differences occurred on both monthly and yearly scales. In 2008, monthly changes in composition were observed. In the River, rotifers were dominant in March representing over 60% of zooplankton. Rotifers were less abundant in April, and cladocerans were the dominant taxa representing 37% of zooplankton. In May (47%) and June (36%) rotifers were dominant. From March through May calanoid copepods were not common in the River, but in June, there was an increase in abundance (32%) and they were almost as abundant as rotifers. In the Delta, calanoid copepods (65%) were dominant in March, but had low abundances April-June. In April and May, copepod

nauplii and rotifers had similar abundances and were the most common taxa. Rotifers accounted for 32% in April and 33% of the composition in May. Copepod nauplii represented 33% in April and 32% in May. Rotifers were the dominant taxa in June (48%). In the Sound, copepod nauplii were the dominant taxa for all of 2008. In 2009, zooplankton communities did not follow the same temporal patterns observed in 2008. One of the biggest differences was the increased dominance of rotifers. Rotifers were the dominant taxa, except during March in the River and Sound and always accounted for at least 35% of the zooplankton (Fig 2.6).

A one-way ANOSIM indicated weak (Global $R = 0.298$) but significant ($p = 0.1\%$) differences in zooplankton composition among the areas (Fig. 2.7). Post-hoc comparisons revealed that River and Delta were not significantly different ($R = 0.054$, $p = 1.3\%$), while the Sound was significantly different from both River ($R = 0.527$, $p = 0.1\%$) and Delta ($R = 0.357$, $p = 0.1\%$). SIMPER analysis comparing the Sound to both the River and Delta showed the level of dissimilarity among comparisons. Bray-Curtis average dissimilarity for the Sound-River was 36.9 and 34.0 for Sound-Delta comparisons. Calanoid copepods contributed the most to differences between the Sound and other areas. Copepod nauplii and rotifers were also important to dissimilarity among the areas, ranking 2nd and 3rd highest contributors. Their order differed between Sound-River and Sound-Delta comparisons. In the Sound-River, Ostracoda and Chydoridae were identified as discriminating taxa. Harpacticoid copepods were the discriminating taxa in Sound-Delta comparisons (Table 2.6).

Relationship between zooplankton and environment

Air temperature and conductivity were both highly correlated ($r \geq 0.9$) to other parameters and were excluded from MANOVA and CCA analyses. A one-way MANOVA

indicated that strong and significant ($\Lambda = 0.11$, $\eta^2 = 0.89$, $p < 0.001$) differences exist among areas. Area explained 89% of the variance in the environmental and zooplankton data. These results indicated that the three sampling areas can be distinguished from each other when both environmental and zooplankton data are analyzed together.

CCA revealed differences in relationships among abiotic parameters and their level of influence in each area, and also many shared patterns (Fig. 2.8). In all areas, water temperature was one of the most influential vectors. No other vector was among the most influential in more than one area. In the River, depth exerted a strong effect. Dissolved oxygen was the most influential parameter in the Delta. In the Sound, current velocity, salinity, and water temperature were all strong and equal in their degree of influence. Temperature and dissolved oxygen were negatively correlated in all areas. The influence of salinity was not consistent across the areas. The level of salinity influence was lowest in the River and strongest in the Sound. In the River and Delta, salinity and flow velocity were negatively correlated. No correlation for those vectors was observed in the Sound.

There were some spatial differences in how the most abundant taxa were influenced by environmental parameters. In all areas, Rotifer abundance was positively affected by slightly above average water temperatures. Abundances were higher with below average current velocity, in the River, and with above average current velocity in the Delta. Rotifer abundances were also higher in the Delta and Sound when dissolved oxygen levels slightly below average. Copepod nauplii are positioned near rotifers on all biplots and exhibited similar patterns. The most common cladocerans, Bosminidae and Daphniidae were all positively influenced by low salinity levels. In all areas, Bosminidae were also more abundant with above average water

velocity. In the River and Delta, Daphniidae exhibited opposite trends for temperature and dissolved oxygen. In the River, lower temperature and higher dissolved oxygen was favorable. Calanoid and cyclopoid copepods exhibited different patterns. Calanoid copepods were positively correlated with salinity while cyclopoid copepods were negatively affected by salinity in all areas. Calanoid copepod abundance varied spatially to water temperature and depth. Cyclopoid copepods were positively influenced by water velocity in both the River and Delta (Fig. 2.8).

Long-term zooplankton patterns

Zooplankton abundances found by Rulifson et al. (1993) and Coggins (2005) were significantly lower than those observed in this study (Table 2.7). Excluding copepod nauplii and rotifers for comparison with Rulifson et al. (1993), did result in a large reduction in abundance values. Even with the removal of those taxa, abundances from this study were between two to seven times higher than abundances from Rulifson et al. (1993). Using the same mesh size (90 μ m), there was a large difference between abundances in this study and Coggins (2005). In the River, Coggins (2005) had an average zooplankton abundance of 892 ± 775 and in this study average abundance was $4,934 \pm 3,806$.

Spatial abundance trends differ between this study and Rulifson et al. (1993). In this study, highest abundances were found in the Sound, and they were significantly higher than those observed in the River and Delta, which had similar abundances. In Rulifson et al. (1993), highest abundances were found in the Delta, and they were twice as high as those found in the River, which had the lowest abundances. Sound abundances were 25% higher than abundances in the River (Table 2.7).

Zooplankton communities between this study and Rulifson et al. (1993) were similar in the River and Delta, but differed in the Sound. In the River and Delta, for both studies, Bosminidae, Daphniidae, and cyclopoid copepods were the most abundant taxa. Calanoid copepods were more prevalent in this study, representing 13% and 28% of the composition in the River and Delta, respectively. Most years, calanoid copepods represented less than 7% of the composition in the River and Delta, in Rulifson et al. (1993). In 1986 and 1988, there was a slight increase to 11% and 15%, respectively. More pronounced differences between the two studies are found when comparing zooplankton composition in the Sound. In Rulifson et al. (1993) zooplankton community was dominated by Bosminidae, Daphniidae, and unknown cladoceran species. Throughout the multiyear study, cladocerans always accounted for $\geq 40\%$ of zooplankton in the Sound. Cyclopoid copepods were also abundant in the Sound, and represented 15-36% of Sound composition. Calanoid copepods composed $\leq 10\%$ of zooplankton community, except in 1986 when they made up 36% of the community. In contrast, zooplankton composition in this study was dominated by calanoid copepods, which made up 76% of the Sound community. Bosminidae (11%), cyclopoid copepods (6%), harpacticoid copepods (3%), and Daphniidae (2%) were the only other taxa to represent $> 1\%$ of the composition.

Major differences were observed between River community structure in this study and in Coggins (2005). In this study, the main River taxa were rotifers (41%), copepod nauplii (18%), Daphniidae (11%), Bosminidae (11%), and cyclopoid copepods (10%). The zooplankton community in Coggins (2005) was dramatically different. The main difference is the lack of rotifers, which accounted for 0% of the composition in Coggins (2005), compared to 41% in this study. The main River zooplankton taxa in Coggins (2005) were cyclopoid copepods (35%), Daphniidae (19%), copepod nauplii (13%), Bosminidae (12%), and Diptera (10%). In the two

studies, Daphniidae, Bosminidae, and copepod nauplii had similar prevalence in the zooplankton communities. Few insects were collected in this study and compose < 1% of the community, versus 10% in Coggins (2005).

Discussion

Zooplankton abundances, in all three areas, were significantly higher than those reported in previous work and are similar to those observed in other North Carolina coastal river systems (Table 2.1). In this time period, increases in zooplankton abundances were also observed in Chowan River, North Carolina (Leech et al. 2008). Many factors could be attributing to these differences in zooplankton abundance. Rulifson et al. (1993) measured phytoplankton concentration in Roanoke River and Albemarle Sound and found that concentrations were large enough to support much higher zooplankton abundances than what was observed. They hypothesized that various environmental parameters, such as daily river flow and seasonal temperatures were responsible for zooplankton patterns in this system. Values for dissolved oxygen, salinity, temperature, turbidity, and pH were similar between this study and Rulifson et al. (1993), and no major differences are observed.

Some differences in water quality are observed when data from state agencies are included. Water quality in the lower Roanoke River and Albemarle Sound has been classified as being of good quality in recent years (NCDENR 2010). In the mid 1990's, the betterment plan was created which regulates discharge from Roanoke Rapids with the goal of maintaining daily average dissolved oxygen concentrations of 5 mg/L and reducing hypoxic and anoxic events (NCDENR 2000). Ambient monitoring from 2005-2009 had no observations of daily averages outside of the standards set by NCDENR for dissolved oxygen (<5.0 mg/L), pH (< 6.0, > 9.0), or

turbidity (> 50 NTU) (NCDENR 2010). Water quality monitoring prior to the enactment of the betterment plan did not identify any long term problems with any of the water quality parameters, but did show periods of hypoxia with associated fish kills (NCDENR 1996). Anecdotal evidence suggests there are differences between water quality between this study and Rulifson et al. (1993) not being captured by solely investigating water quality values. In the earlier study, there were frequent problems with the nets clogging with detritus, which was not an issue during this study (R. Rulifson, ECU, personal communication). It is possible that factors causing hypoxic events during that time period and high levels of detritus in Rulifson et al. (1993) contributed to differences in zooplankton abundance between the two studies.

Flow velocity and high discharge rates from dams are considered to be important factors in regulating zooplankton abundance and composition (Hynes 1970). Identifying the influence of river velocity in all three studies is difficult and beyond the scope of this study. Zooplankton abundances are negatively correlated with velocity. Zooplankton production in riverine systems occurs in areas of slow moving water. High river flows and discharge rates negatively influence zooplankton abundances by having a wash out effect that dilutes populations (Hynes 1970).

Discharge rates and rankings for the past 100 years, ending in 2009, for the different years in all three studies are presented in Table 2.8. Coggins (2005) sampled in 2003 when discharge rates were highest ($660.8 \text{ m}^3/\text{s}$) and zooplankton abundances were low. This suggests that high flows were responsible for low abundances, but when zooplankton composition is included, there is no clear picture. In other studies where high flows and dam discharge have negatively influenced zooplankton abundance, rotifers were still dominant in these systems. Larger zooplankton taxa, such as cladocerans and copepods were negatively influenced by high

flow and had lower abundances (Cowell 1970; Obertegger et al. 2007). Even in studies where rotifers were negatively influenced by flow, they were still the numerically dominant taxa, but abundances were lower than in areas with low current velocity (Dickerson et al. 2010). In the spring samples, Rotifers were absent from Coggins (2005), suggesting that high flow is not the only parameter influencing zooplankton abundance in this study.

During the Rulifson et al. (1993) study, samples were collected over several years and discharge rates and flows were highly variable. In 1987, the 2nd highest discharge rates ($\mu = 566.2$ m/s) for the past 100 years were observed and this was also the year with the highest zooplankton abundance ($\mu = 606$ number/m³) in this study. During that study, abundances did not follow any clear pattern in respect to flow. Abundances in this study were significantly higher than those previously reported, and discharges were moderate to low in comparison to the past 100 years. It is possible flow and discharge partially explain these differences in abundance, but more research is needed to fully understand what factors are driving long term zooplankton trends in this system.

Significant differences in zooplankton composition and abundance were observed between the Sound and other study areas. SIMPER analysis showed that calanoid copepods were most responsible for the Sound being significantly different in composition from the River and Delta areas. Calanoid copepods were more abundant in the Sound and comprised a higher percentage of the zooplankton. Higher abundances of copepod nauplii and rotifers in the Sound also contributed to the Sound being significantly different. In the River and Delta, higher abundances of cyclopoid copepods, cladocerans, and ostracods also contributed to significant differences when compared to the Sound (Table 2.6). Many of these differences can be

explained by salinity differences among the areas. Salinity was significantly higher in the Sound where calanoid copepods had the highest abundance. CCA showed they were positively correlated with salinity, while taxa that were less common in the Sound, such as cladocerans and cyclopoid copepods were negatively correlated with salinity (Fig 2.8). While salinity was a strong influence in the Sound, it had lesser influence in the River and Delta. The River area is located the furthest upstream of the areas. In times of low flow, there have been estuarine influences from Albemarle Sound, but typically the River is far enough upstream to where those influences are rare and this is considered a true freshwater area (NCDENR 2000).

The results of the one-way MANOVA suggested the majority of variance ($\eta^2 = 89\%$) in zooplankton trends and the environment were explained by the effect of area. Significant differences in zooplankton abundance and composition were not observed between the River and Delta, despite the strongly significant MANOVA. Differences in zooplankton composition between the two areas were more pronounced in 2008 (Fig. 2.7). It is possible the surge of rotifers in 2009 masked differences in zooplankton composition between the River and Delta. In the Rulifson et al. (1993) study, Delta abundances were significantly higher than those in the River. Long term observation of zooplankton in these areas needs to be conducted, to see if with more years of data, these two areas can be classified as separate areas based on zooplankton patterns and not just location. Long term observation is also needed to address whether the increase in zooplankton abundance seen in this study is a permanent or a temporary fluctuation.

Table 2.1. Average zooplankton abundance (number/m³) in several North Carolina coastal river systems.

Study	System	State	Mesh size (μm)	Abundance (number/m ³)
Mallin (1991)	Neuse River	NC	76	32,877
Fulton (1984)	Newport River	NC	76	21,900
Thayer et al. (1974)	Newport River	NC	156	6,200
Birkhead et al. (1979)	Cape Fear River	NC	156	7,450
Winslow et al. (1985)	Chowan River	NC	70	3,423
Rulifson et al. (1993)	Roanoke River	NC	250	327
	Roanoke Delta	NC	250	696
	Albemarle Sound	NC	250	532
Coggins (2005)	Roanoke River	NC	90	892

Table 2.2. Average monthly values ($\mu \pm$ SD) for environmental parameters collected March-June 2008 and 2009 in lower Roanoke River and Albemarle Sound, North Carolina.

Area	Month	Air temp (°C)	Conductivity (μ S)	D.O. (mg/L)	Flow (m/s)	pH	Salinity (ppt)	Water temp (°C)	Wind speed (m/s)
River	March 08	13.1 \pm 3.0	134.2 \pm 99.1	8.2 \pm 1.1	0.1 \pm 0.1	7.4 \pm 0.1	0.1 \pm 0.0	13.2 \pm 1.8	4.6 \pm 5.7
	April 08	16.4 \pm 2.8	112.6 \pm 11.2	7.0 \pm 1.9	0.1 \pm 0.1	7.4 \pm 0.1	0.1 \pm 0.0	15.7 \pm 1.5	2.0 \pm 2.8
	May 08	18.8 \pm 2.5	119.6 \pm 8.4	5.7 \pm 0.9	0.2 \pm 0.2	7.5 \pm 0.1	0.1 \pm 0.0	20.2 \pm 1.0	2.7 \pm 3.9
	June 08	25.3 \pm 2.8	136.6 \pm 17.0	5.1 \pm 1.0	0.1 \pm 0.1	7.3 \pm 0.2	0.1 \pm 0.0	27.2 \pm 2.2	0.8 \pm 1.4
	March 09	11.6 \pm 4.1	77.4 \pm 16.2	10.3 \pm 1.2	0.2 \pm 0.1	6.6 \pm 0.6	0.1 \pm 0.0	9.6 \pm 2.3	1.1 \pm 1.1
	April 09	14.8 \pm 3.6	91.5 \pm 7.4	8.3 \pm 0.6	0.1 \pm 0.1	6.5 \pm 0.3	0.1 \pm 0.0	16.2 \pm 2.3	1.0 \pm 1.3
	May 09	20.5 \pm 2.9	107.1 \pm 6.8	7.0 \pm 0.9	0.1 \pm 0.1	6.5 \pm 0.2	0.1 \pm 0.0	21.8 \pm 1.5	1.0 \pm 1.0
	June 09	24.2 \pm 0.8	110.8 \pm 7.73	5.0 \pm 1.4	0.1 \pm 0.1	6.5 \pm 0.1	0.1 \pm 0.0	24.4 \pm 1.3	0.7 \pm 1.0
Delta	March 08	12.3 \pm 4.1	203.7 \pm 231.3	7.8 \pm 1.4	0.1 \pm 0.0	7.4 \pm 0.1	0.1 \pm 0.2	13.4 \pm 1.8	6.0 \pm 5.4
	April 08	15.7 \pm 2.1	139.9 \pm 59.8	6.4 \pm 1.6	0.1 \pm 0.1	7.5 \pm 0.1	0.1 \pm 0.0	16.0 \pm 1.6	2.9 \pm 5.7
	May 08	18.9 \pm 2.5	133.1 \pm 18.6	5.7 \pm 0.9	0.2 \pm 0.1	7.5 \pm 0.2	0.1 \pm 0.0	21.4 \pm 4.7	4.3 \pm 5.0
	June 08	25.1 \pm 2.3	155.5 \pm 19.4	5.0 \pm 0.8	0.2 \pm 0.2	7.3 \pm 0.2	0.1 \pm 0.0	28.8 \pm 5.7	1.3 \pm 1.3
	March 09	12.5 \pm 6.2	89.1 \pm 13.5	10.1 \pm 1.3	0.2 \pm 0.1	6.9 \pm 0.3	0.1 \pm 0.0	9.9 \pm 2.1	1.9 \pm 1.5
	April 09	14.2 \pm 4.0	96.1 \pm 11.7	7.8 \pm 0.4	0.1 \pm 0.1	6.5 \pm 0.3	0.1 \pm 0.0	16.3 \pm 2.4	1.7 \pm 1.6
	May 09	21.0 \pm 4.1	116.7 \pm 7.8	6.9 \pm 0.5	0.1 \pm 0.1	6.6 \pm 0.2	0.1 \pm 0.0	22.2 \pm 1.6	1.6 \pm 1.2
	June 09	24.8 \pm 1.3	114.8 \pm 29.5	4.9 \pm 1.3	0.1 \pm 0.1	6.5 \pm 0.2	0.1 \pm 0.0	25.6 \pm 0.9	1.3 \pm 1.3
Sound	March 08	12.1 \pm 3.3	1,779.8 \pm 1,140.4	8.8 \pm 1.7	0.1 \pm 0.0	7.8 \pm 0.3	1.3 \pm 0.7	12.6 \pm 1.6	12.0 \pm 2.2
	April 08	15.8 \pm 1.7	642.2 \pm 966.4	7.9 \pm 2.1	0.2 \pm 0.2	7.6 \pm 0.1	0.4 \pm 0.6	15.9 \pm 2.2	9.7 \pm 4.3
	May 08	19.7 \pm 1.5	408.0 \pm 490.0	6.4 \pm 0.8	0.1 \pm 0.1	7.6 \pm 0.1	0.3 \pm 0.3	20.5 \pm 1.0	10.7 \pm 8.8
	June 08	25.5 \pm 1.9	835.6 \pm 643.7	5.4 \pm 0.8	0.3 \pm 0.2	7.4 \pm 0.2	0.4 \pm 0.3	26.9 \pm 1.7	4.3 \pm 2.1
	March 09	12.1 \pm 6.5	1,464.7 \pm 842.3	10.9 \pm 0.8	0.3 \pm 0.3	7.0 \pm 0.4	1.2 \pm 0.7	8.9 \pm 2.5	2.3 \pm 1.7
	April 09	16.3 \pm 2.1	174.0 \pm 270.6	8.7 \pm 0.7	0.3 \pm 0.2	6.6 \pm 0.2	0.1 \pm 0.1	16.7 \pm 2.2	3.1 \pm 1.9
	May 09	20.9 \pm 4.6	791.0 \pm 984.5	8.6 \pm 1.0	0.3 \pm 0.1	6.9 \pm 0.3	0.4 \pm 0.5	22.0 \pm 2.4	3.0 \pm 0.7
	June 09	26.0 \pm 1.1	646.5 \pm 713.8	6.4 \pm 1.3	0.1 \pm 0.1	6.8 \pm 0.3	0.3 \pm 0.3	26.2 \pm 0.3	1.7 \pm 1.1

Table 2.3. Comparison between average values for environmental parameters in 2008 and 2009 in lower Roanoke River and Albemarle Sound, North Carolina. Values for 2008 and 2009 represent $\mu \pm SD$.

Parameter	2008	2009	<i>t</i>	<i>p</i>
Air temperature (°C)	18.3 ± 5.3	18.0 ± 6.0	0.30	0.765
Conductivity (µS)	389.9 ± 593.3	270.0 ± 479.9	1.09	0.280
Dissolved oxygen (mg/L)	6.5 ± 1.5	7.9 ± 2.0	-3.74	< 0.001
Flow (m/s)	0.1 ± 0.1	0.2 ± 0.1	-1.54	0.128
pH	7.5 ± 0.2	6.7 ± 0.4	16.11	< 0.001
Precipitation (mm)	2.5 ± 2.9	3.0 ± 2.0	-0.91	0.367
Salinity (ppt)	0.2 ± 0.4	0.1 ± 0.3	0.84	0.401
Water temperature (°C)	19.4 ± 5.8	18.1 ± 6.1	0.95	0.344
Wind (m/s)	5.2 ± 5.1	1.7 ± 1.3	4.49	< 0.001

Table 2.4. Mean values for environmental parameters in each sampling area in lower Roanoke River and Albemarle Sound, North Carolina. Means sharing a letter in their superscript are not significantly different at the 0.5 level according to a Ryan-Einot-Gabriel-Welch (REGWQ) procedure.

Environmental parameter	Area		
	River	Delta	Sound
Current velocity (m/s)	0.1 ^A	0.1 ^A	0.2 ^B
Depth (m)	4.8 ^A	3.2 ^B	3.3 ^B
DO (mg/L)	7.4 ^A	6.8 ^A	7.7 ^A
pH	7.0 ^A	7.0 ^A	7.3 ^B
Salinity	0.1 ^A	0.1 ^A	0.3 ^B
Water temp (°C)	18.5 ^A	19.1 ^A	18.8 ^A
Wind speed (m/s)	1.7 ^A	2.8 ^A	6.4 ^B

Table 2.5. Complete list of taxa collected March-June 2008 and 2009 in lower Roanoke River and Albemarle Sound, North Carolina. A (+) indicates that the taxa was collected from that area, while a (-) indicates absence. CCA abbreviation is the code used to identify the taxa on CCA biplots. A (.) indicates the taxa was rare and not used in the analysis.

Taxa	CCA abbreviation	Area		
		River	Delta	Sound
Bosminidae	Bos	+	+	+
Branchiopoda	.	+	-	-
Calanoida	Cal	+	+	+
Chydoridae	Chy	+	+	+
Coleoptera	.	+	+	+
Copepod nauplii	Naup	+	+	+
Crab megalopa	.	+	-	-
Cyclopoida	Cyc	+	+	+
Daphniidae	Daph	+	+	+
Diptera	Dip	+	+	+
Fish egg	.	+	+	+
Fish larvae	.	+	+	+
<i>Gammarus</i> spp.	Gam	+	+	+
Gastropoda	.	+	-	-
Harpacticoida	Har	+	+	+
Isopoda	.	-	-	+
Leptodoridae	.	-	+	+
Nematoda	.	+	+	+
Oligochaeta	.	+	+	+
Ostracoda	Ost	+	+	+
Rotifera	Rot	+	+	+
Sididae	Sid	+	+	+
Unknown	.	+	+	-

Table 2.6. SIMPER analysis evaluating dissimilarity between areas identified as significantly different using ANOSIM. Abundances are 4th root transformed. Average Bray-Curtis dissimilarity scores are listed as average dissimilarity. Diss/SD identifies how consistently a taxa contributes to dissimilarity. Values with an asterisk identify discriminating taxa. Contribution percentage is the amount of dissimilarity that can be attributed to a taxon.

Area comparison	Taxa	Average abundance		Average dissimilarity	Diss/SD	Contribution %	
		Sound	Comparison area				
Sound & River	Calanoida	5.8	2.9	4.8	1.4	12.9	
	Copepod nauplii	8.1	5.2	4.1	1.4	11.1	
	Rotifera	7.2	6.2	3.8	1.2	10.3	
	Daphniidae	2.2	4.1	2.8	1.3	7.6	
	Ostracoda	0.6	2.5	2.8	1.7*	7.6	
	Chydoridae	0.7	2.4	2.7	1.7*	7.4	
	Bosminidae	3.3	3.8	2.6	1.3	7.0	
	Cyclopoida	3.0	4.2	2.5	1.3	6.8	
	Harpacticoida	2.5	1.3	2.5	1.4	6.6	
	Gammarus	1.5	0.1	2.0	1.1	5.5	
	Diptera	0.7	1.8	1.9	1.4	5.3	
	Sididae	0.5	0.9	1.4	0.9	3.9	
	Sound & Delta	Calanoida	5.8	3.3	4.6	1.4	13.4
		Rotifera	7.2	6.0	3.9	1.2	11.5
Copepod nauplii		8.1	5.6	3.9	1.4	11.4	
Harpacticoida		2.5	1.0	2.7	1.5*	7.9	
Cyclopoida		3.0	4.4	2.6	1.3	7.5	
Chydoridae		0.7	2.0	2.5	1.4	7.2	
Bosminidae		3.3	3.7	2.4	1.3	7.1	
Ostracoda		0.6	1.8	2.1	1.4	6.1	
Daphniidae		2.2	3.3	2.1	1.2	6.1	
Gammarus		1.5	1.0	2.1	1.2	6.0	
Diptera	0.7	1.4	1.7	1.2	5.0		
Sididae	0.5	0.8	1.4	0.8	4.0		

Table 2.7. Comparison of mean zooplankton abundance ($\mu \pm SD$) between this study and previous studies in Roanoke River and Albemarle Sound, North Carolina. For comparison with Rulifson et al. (1993), rotifers and copepod nauplii were excluded from analysis.

Study	Area	Abundance (number/m ³)		<i>t</i>	<i>df</i>	<i>p</i>
		Study	Binion (2011)			
Rulifson (1993)	River	327 ± 50	1,998 ± 2,683	-3.63	33	0.001
	Delta	696 ± 5272	1,537 ± 1,454	-2.99	34	0.005
	Sound	532 ± 323	3,670 ± 3,878	-4.37	31	< 0.001
Coggins (2005)	River	891 ± 775	4,933 ± 3,806	5.15	16	< 0.001

Table 2.8. Comparison of Roanoke Rapids Dam discharge rates (m³/s) for all years sampled by Rulifson et al. (1993), Coggins (2005), and Binion (2011). Rankings are based on 100 years of data, ending in 2009. A rank of 1 indicates the fastest mean discharge rate.

Study	Year	Rank	Discharge (m ³ /s)		
			Mean	Min	Max
Rulifson et al. (1993)	1984	10	435.3	195.0	569.5
	1985	95	111.3	71.6	142.2
	1986	92	128.3	80.3	156.3
	1987	2	566.2	240.0	808.7
	1988	91	132.1	69.9	195.4
	1989	13	402.0	295.3	531.8
	1990	18	366.3	326.5	404.4
	1991	34	321.1	200.4	472.9
Coggins (2005)	2003	1	660.8	517.4	860.0
Binion et al. (2011)	2008	77	177.9	70.9	286.3
	2009	42	280.7	235.4	405.8

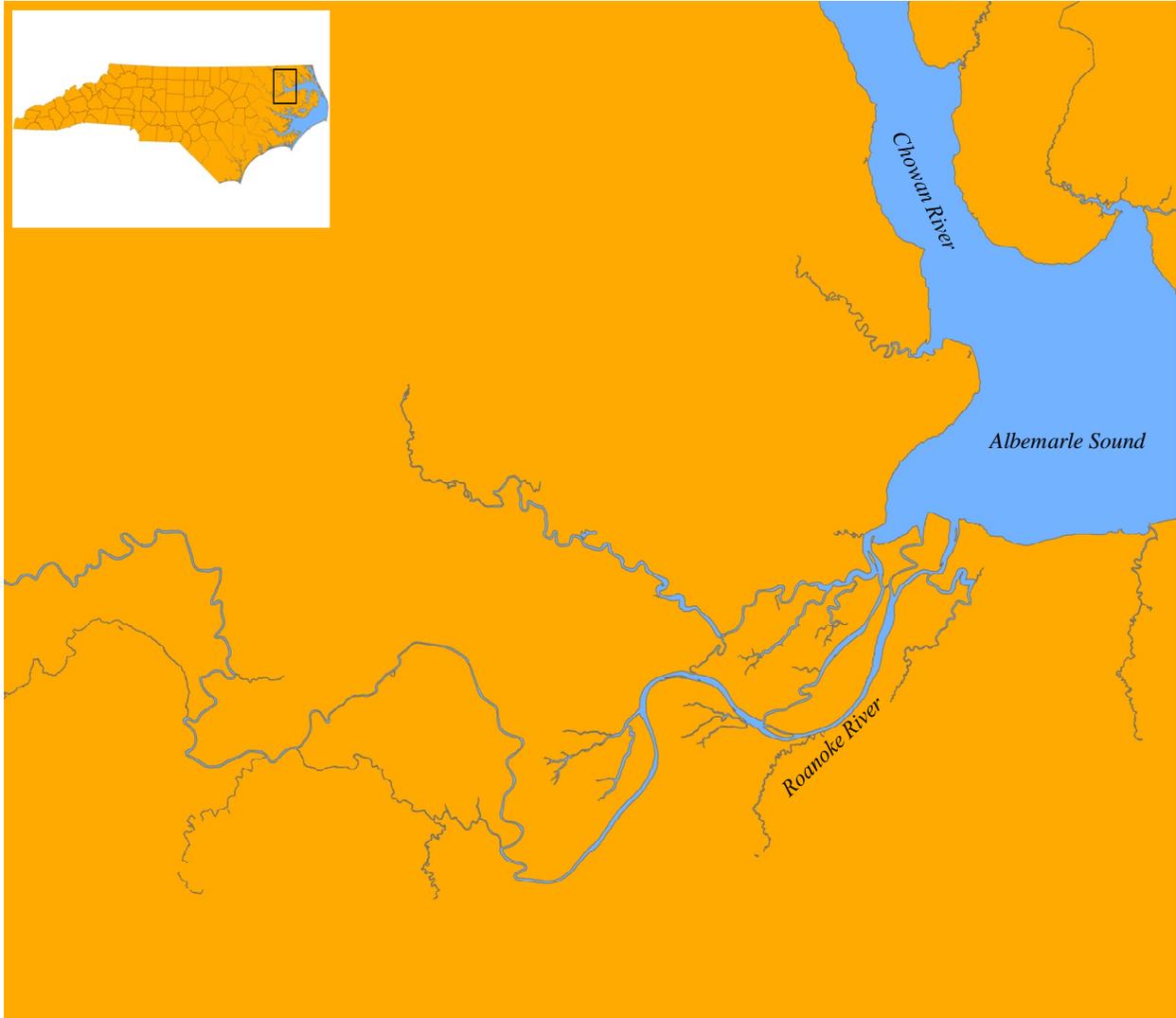


Fig 2.1. Roanoke River and Albemarle Sound, North Carolina located at 76°N, 36°W.

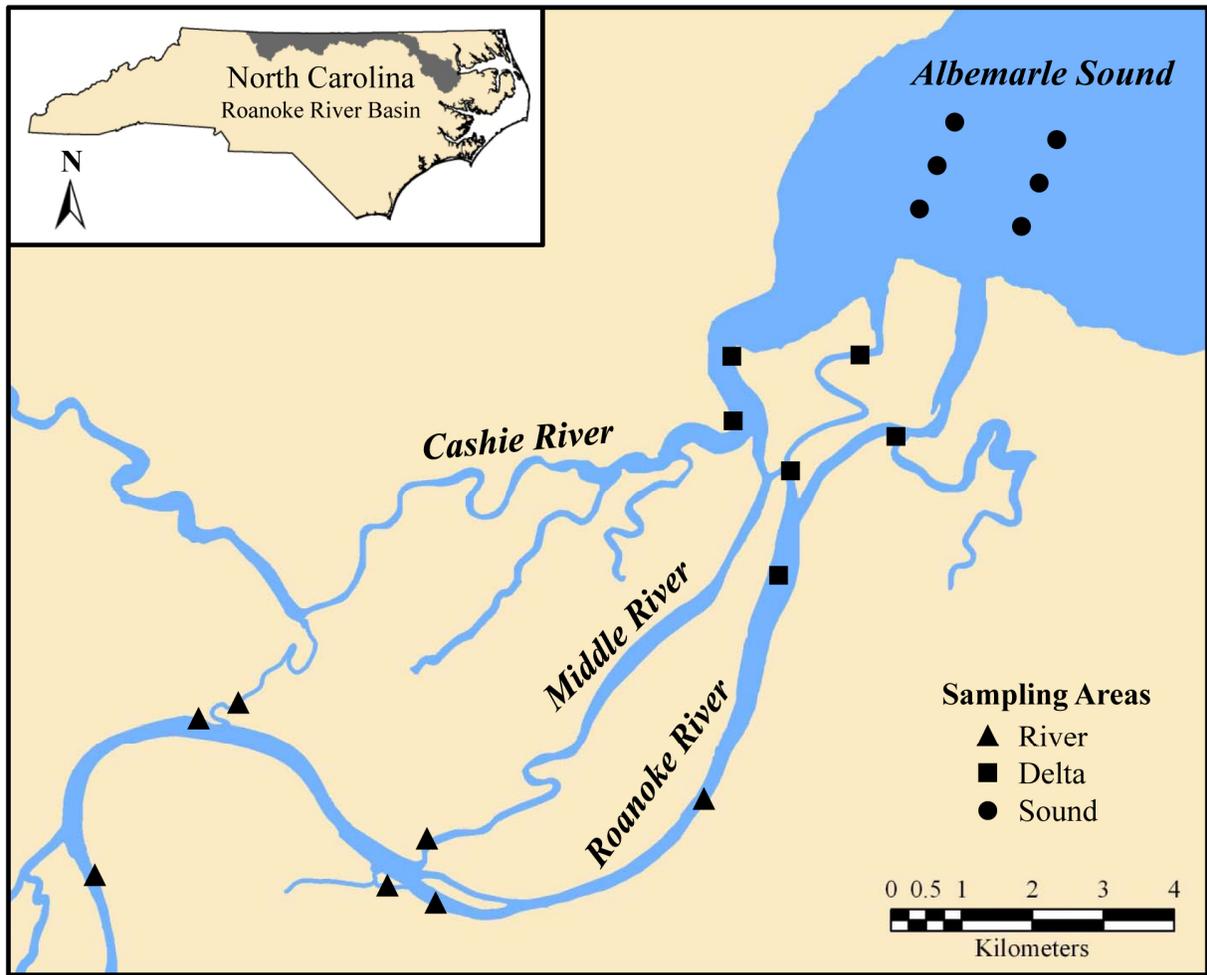


Fig 2.2. Location of the sampling stations and the division of the three sampling areas in lower Roanoke River and Albemarle Sound, North Carolina.

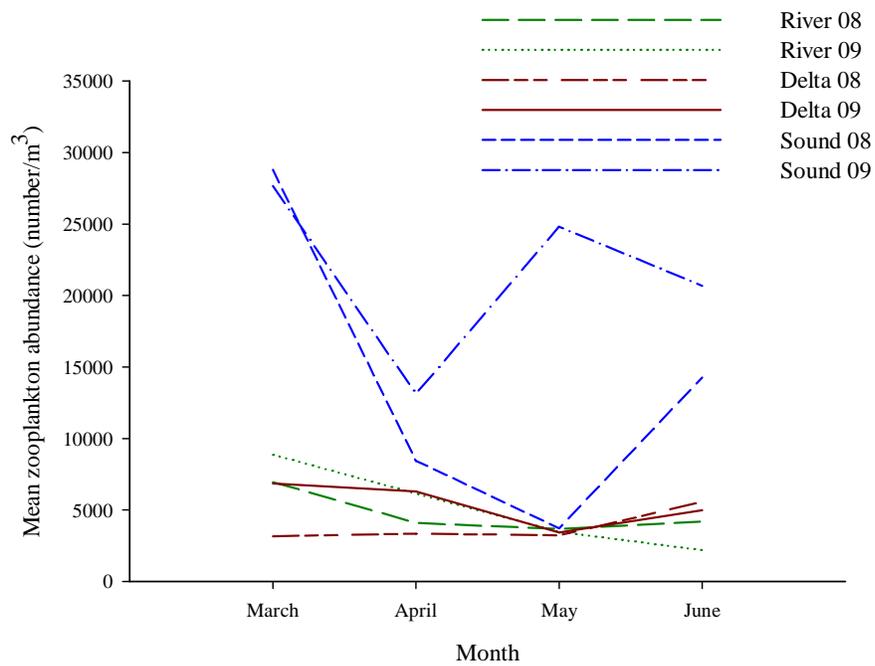


Fig 2.3. Mean monthly zooplankton abundance (number/m³) in each sampling area in Roanoke River and Albemarle Sound, North Carolina during 2008 and 2009.

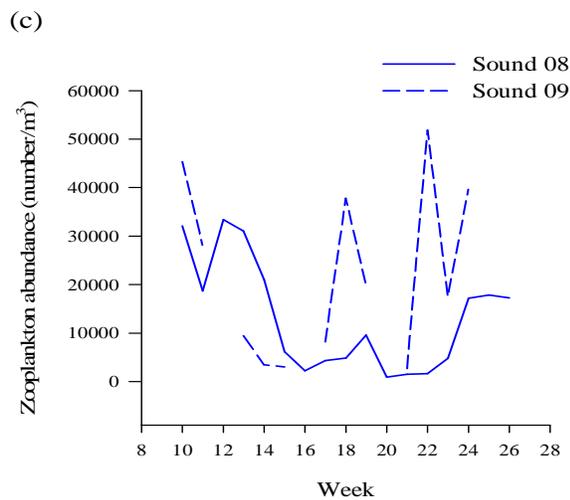
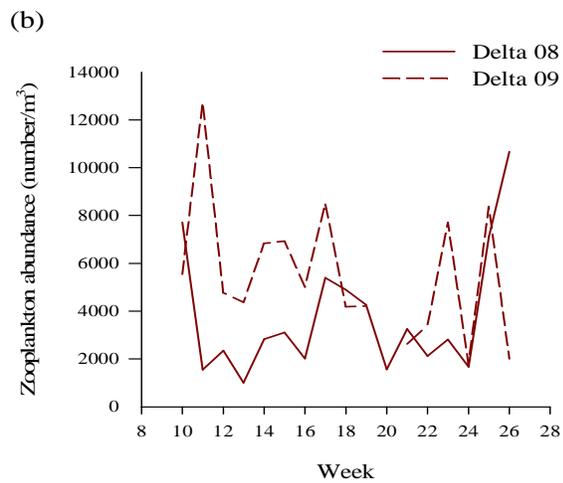
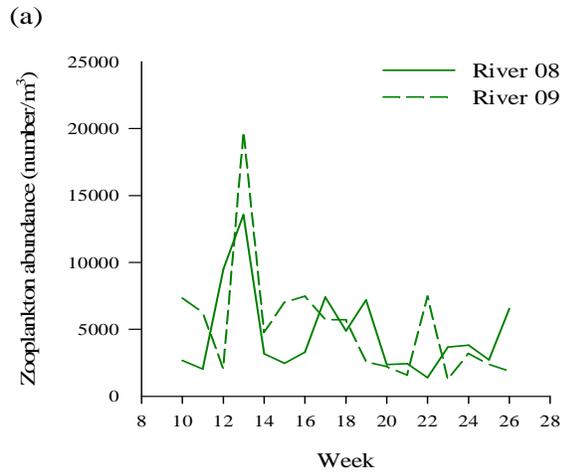


Fig. 2.4. Weekly zooplankton abundance (number/m³) in Roanoke River and Albemarle Sound, North Carolina in (a) River, (b) Delta, and (c) Sound. Note scale difference for Y axis.

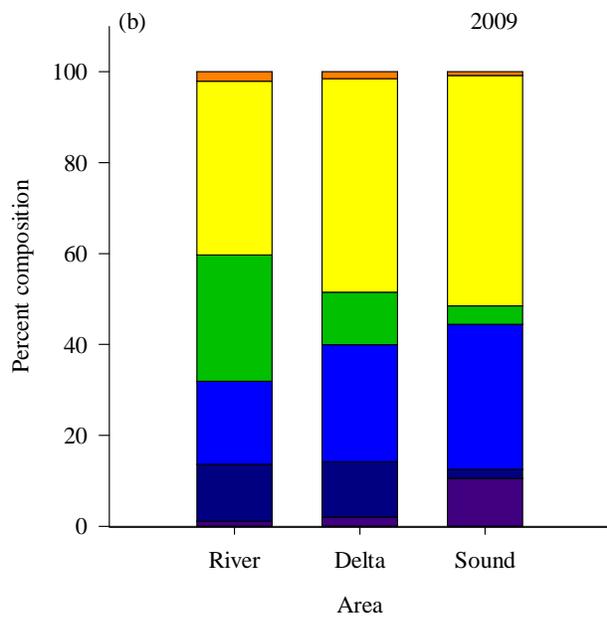
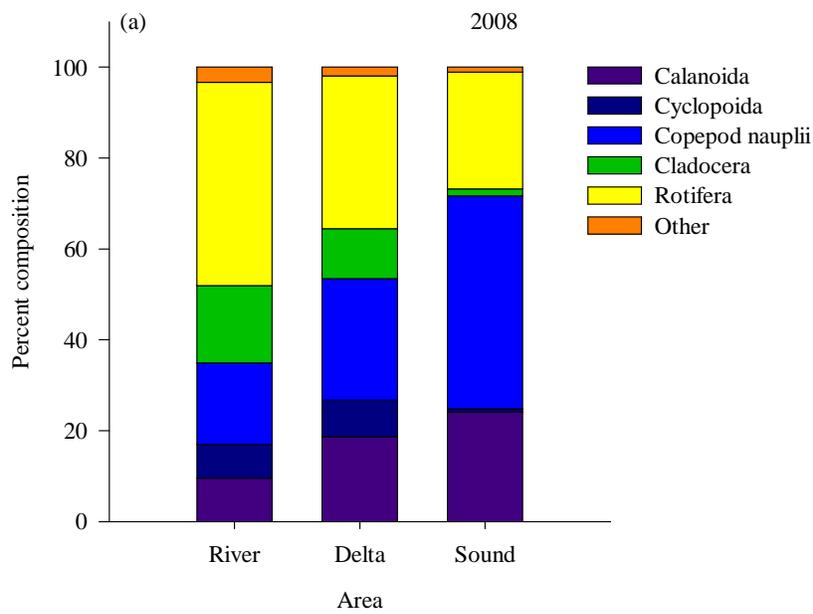


Fig 2.5. Overall zooplankton taxonomic composition by area for (a) 2008 and (b) 2009 in lower Roanoke River and Albemarle Sound, North Carolina.

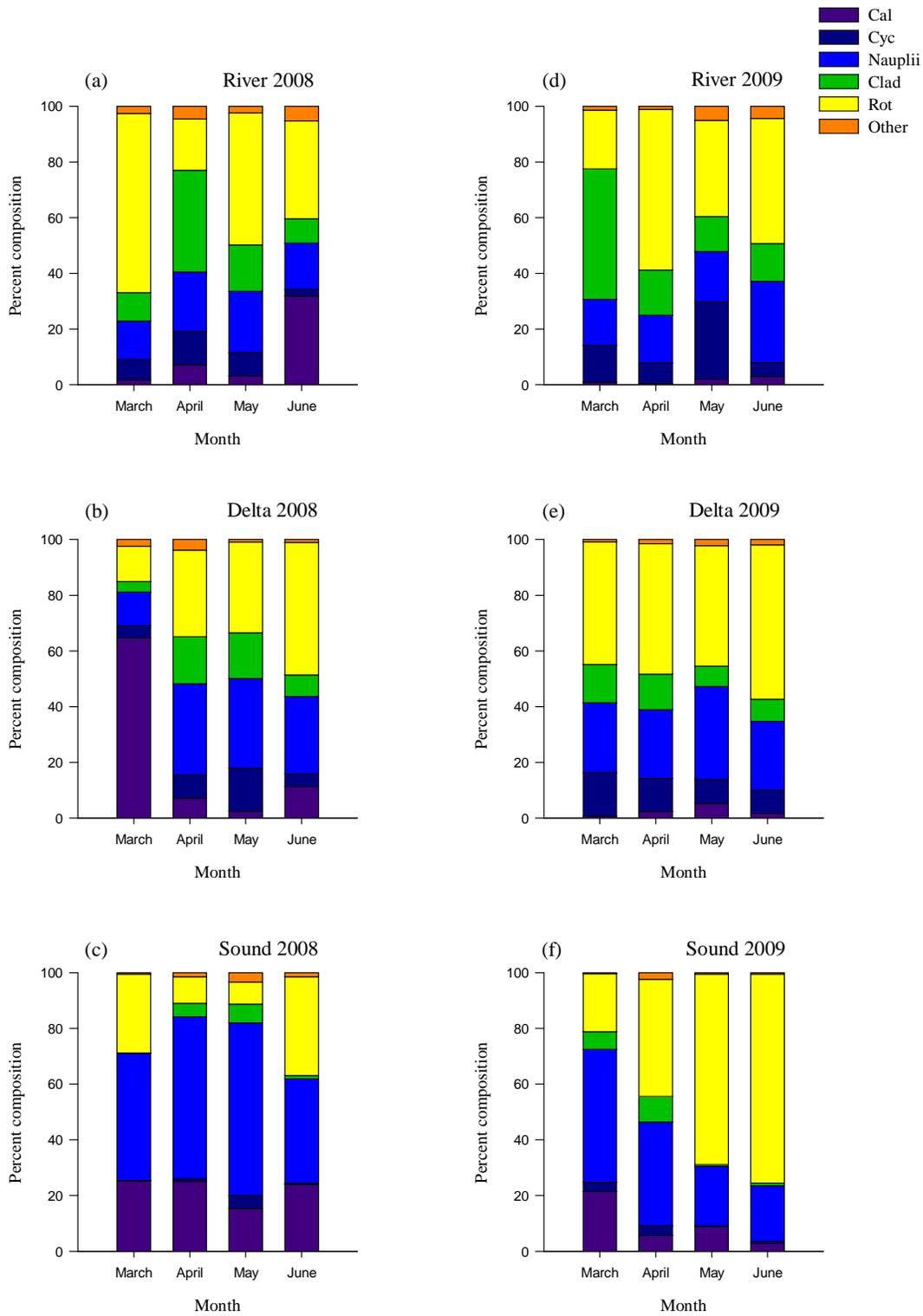


Fig. 2.6. Monthly zooplankton composition in 2008 for (a) River, (b) Delta, and (c) Sound and in 2009 for (d) River, (e) Delta, and (f) Sound.

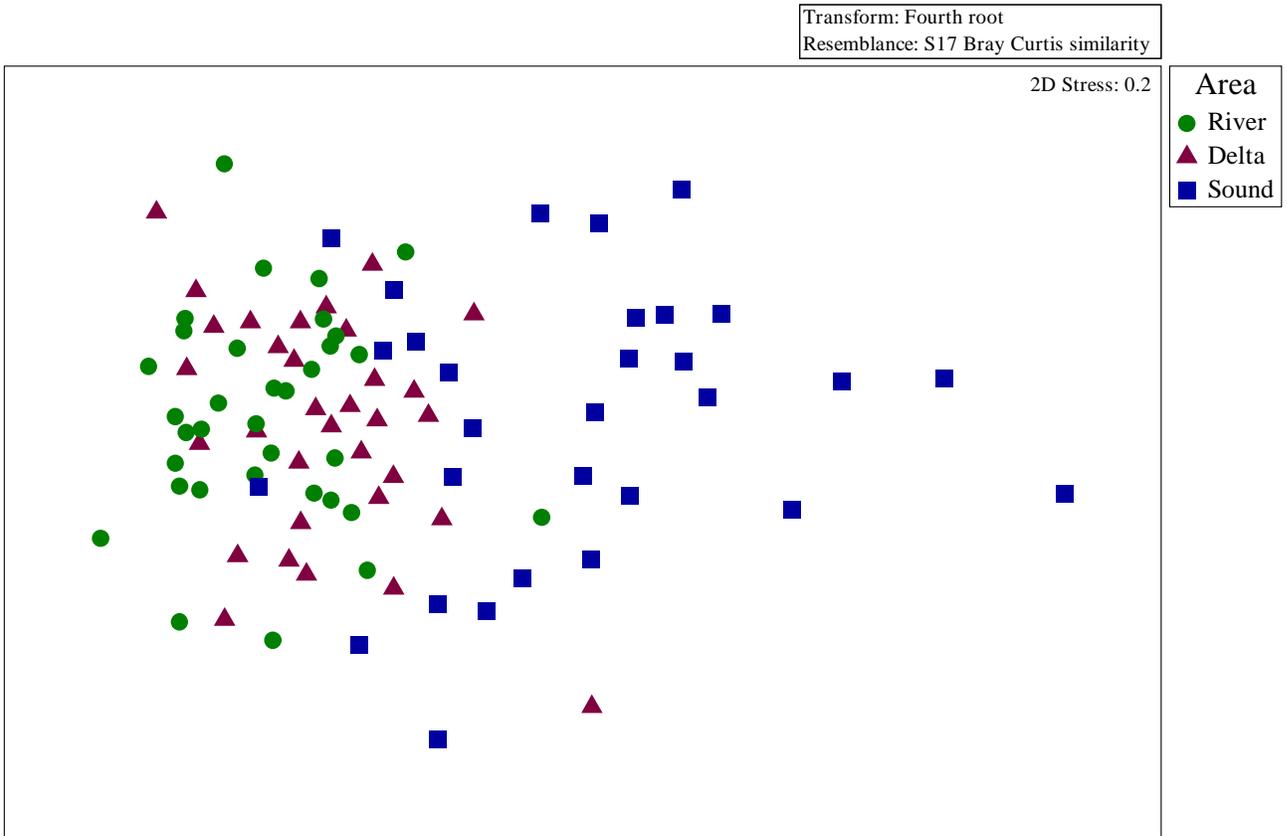
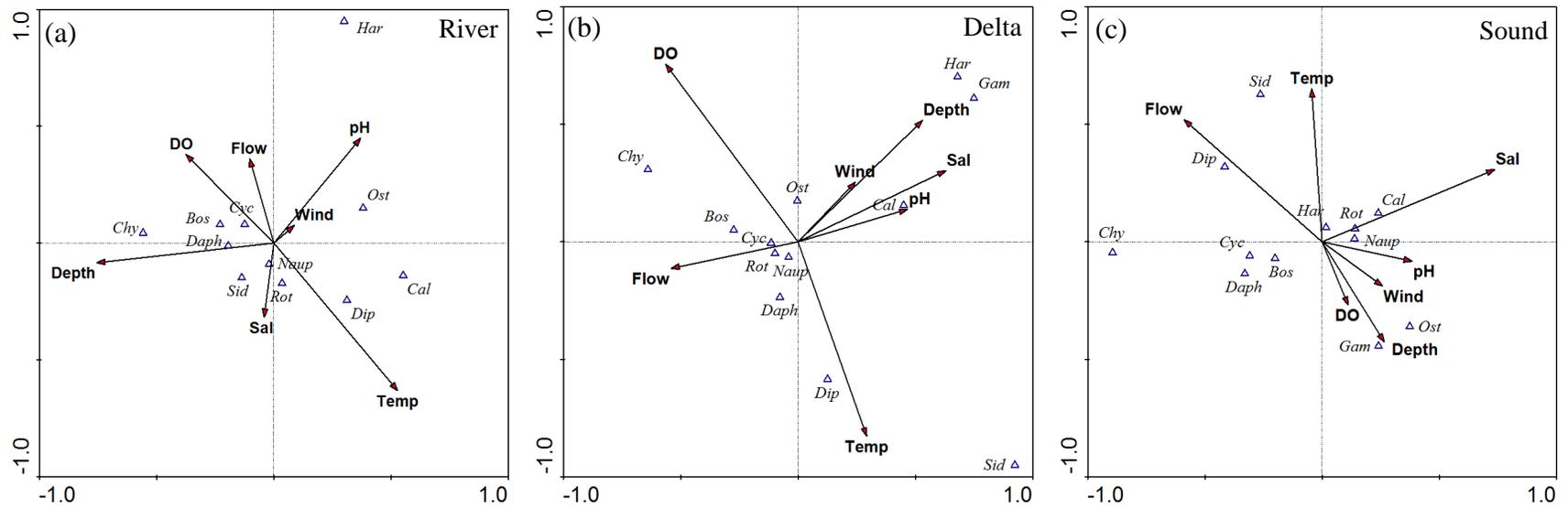


Fig. 2.7 NMDS ordination plot illustrating similarity among samples in respect to area, for zooplankton samples collected in Roanoke River and Albemarle Sound, North Carolina.



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Fig 2.8. CCA biplots for areas (a) River, (b) Delta, and (c) Sound in Roanoke River and Albemarle Sound, North Carolina. The abbreviation for each taxa is located in Table 2.5. Environmental parameters included in analysis are dissolved oxygen (DO), current velocity (Flow), wind velocity (Wind), water depth (Depth), Salinity (Sal), water temperature (Temp), and pH.

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CHAPTER 3: FORAGING POTENTIAL OF LARVAL ALOSINES IN LOWER ROANOKE
RIVER AND ALBEMARLE SOUND, NORTH CAROLINA

Abstract

Spatial and temporal overlap between zooplankton and larval American shad (*Alosa sapidissima*), river herring (alewife *A. pseudoharengus* and blueback herring *A. aestivalis*), and hickory shad (*A. mediocris*) was evaluated to determine if larval alosines are food limited in lower Roanoke River and Albemarle Sound, North Carolina. Zooplankton and ichthyoplankton samples were collected concurrently March through June 2008-09 at 19 stations, divided among three areas: River, Delta, and Sound. Significant spatial and temporal differences were observed for alosine abundances. Abundances (number/100m³ ± SD) were significantly higher in 2009 (30.8 ± 149.8), than in 2008 (4.0 ± 20.9). Across both years, River (21.0 ± 127.6) alosine abundances were significantly higher than those in Delta (7.4 ± 35.4) and Sound (4.6 ± 24.8). Zooplankton abundances exhibited the opposite spatial pattern with significantly higher abundances (number/m³ ± SD) in the Sound (16,547 ± 14,678) than in the River (4,934 ± 3,806) and Delta (4,647 ± 2,846). Zooplankton size distribution was evaluated and the most common taxa segregated into two groups based on size. Rotifers and copepod nauplii comprised the small size group and Daphniidae, Bosminidae, calanoid copepods, and cyclopoid copepods were in the larger size class. Mouth gape models were developed for each alosine species and used to estimate maximum prey size at first feeding. Alewives, blueback herring, and hickory shad are able to consume copepod nauplii and rotifers at first feeding. American shad larvae are larger and are able to feed on Bosminidae and cyclopoid copepods, in addition to copepod nauplii and

rotifers. During both years in all areas, there was a high amount of overlap between larval alosines and zooplankton, suggesting larval alosines in this system are not food limited.

Introduction

Year class strength and recruitment of fish are strongly related to survival during egg and larval development. The availability of food resources is considered one of the more influential factors affecting survival in the larval phase (Cushing 1990; Leggett and DeBlois 1994). In 1914, Hjort developed the critical period hypothesis, which directly links larval fish survival with feeding success. Under this hypothesis, there is a critical period, the period of time after yolk sac absorption when a larva is transitioning from endogenous to exogenous feeding. Year class strength is dependent on successful first feedings during this period. If there is not a temporal overlap between larval fish and their prey, then many larvae will not have a successful first feeding and starve. There will be a high mortality rate and year class strength will suffer. Hjort hypothesized that larval production was timed to follow phytoplankton production to increase feeding success (Leggett and DeBlois 1994; Houde 2008).

Cushing's match/mismatch hypothesis builds upon Hjort's critical period hypothesis. The match/mismatch hypothesis expands the importance of prey availability and feeding success to include the entire larval period, not just for the first feeding (Cushing 1990). This hypothesis suggests that fish spawning occurs at fixed times in both spring and autumn to overlap with peak plankton production. This hypothesis also highlights the importance of larval fish distribution overlapping with zooplankton distribution both temporally and spatially. If larval fish and zooplankton distribution overlap in both time and space, this is defined as a match. If there is any separation, temporally and/or spatially between larvae and zooplankton, then a mismatch has

occurred. Larval growth and survival is expected to be higher when a match is occurring and this increase in larval survival corresponds to an increase in year class strength and recruitment in juvenile and adult populations (Cushing 1990; Leggett and DeBlois 1994). Increases in feeding success and larval survival were observed in areas with high zooplankton and larval striped bass overlap in Chick and Van Den Avyle (1999) and Martino and Houde (2010).

In larval fish, mouth gape is a limiting factor at the onset of exogenous feeding, restricting the prey size that can be consumed (DeVries et al. 1998; Yufera and Darias 2007). Mouth gape becomes less limiting as fish grow. A positive relationship exists between fish length and mouth gape (DeVries et al. 1998; Puvanendran et al. 2004). Previous studies have suggested that maximum functional mouth gape for a larval fish is when the mouth is open at a 90° angle. Optimal prey sizes are generally within 30- 50% of the mouth gape (Bremigan and Stein 1994; Turingan et al. 2005; Riley et al. 2009).

American shad, river herring, and hickory shad all spawn in Roanoke River, North Carolina and their larvae use this area as nursery habitat. All of these species have experienced population declines (ASMFC 2007; Greene et al. 2009; Harris and Hightower 2010; NCDMF 2010). A coast-wide American shad stock assessment documented that stocks are at all time lows and do not appear to be recovering (ASMFC 2007). In Albemarle Sound, American shad are currently listed as a species of concern (ASMFC 2007; NCDMF 2010). River herring stocks in Albemarle Sound are depleted (NCDMF 2010). Hickory shad status in North Carolina is currently unknown, but coast wide landings suggest the populations are viable. In 2009, there was a slight increase in hickory shad landings, but they were still below the 10 year average (Greene et al. 2009; NCDMF 2010).

Currently in North Carolina, American shad and hickory shad are managed under the ASMFC shad and herring fishery management plan. In 2004, the American shad ocean-intercept fishery was closed. American shad and hickory shad fisheries are opened and closed by proclamation from January 1st to April 14th each year. There is no commercial quota for these species; harvest is regulated through restrictions on fishing gears (NCDMF 2008). In an effort to revive American shad stocks, larvae are stocked in several coastal states and in 2008, over 8 million larvae were stocked in the upper reaches of the Roanoke River (Taylor et al. 2009). In 2007, a statewide moratorium on all river herring harvest was accepted by N.C. Marine Fisheries Commission (NCDMF 2007). Despite these efforts, current American shad and river herring stocks are well below historic levels of abundance and biomass (Taylor et al. 2009).

The purpose of this study is to evaluate if the failure of alosine stocks to recover in Roanoke River and Albemarle Sound, North Carolina can be linked to match/mismatch regulation during the larval phase. Spatial and temporal distribution of American shad, alewives, blueback herring, and hickory shad were evaluated within three areas located in lower Roanoke River and Albemarle Sound. Mouth gape models were created to estimate optimal prey sizes for each species. Spatial and temporal overlap between larval alosines and size appropriate zooplankton prey were analyzed. Whether match/mismatch regulation is occurring was determined by evaluating the degree of overlap between larval alosine and potential prey populations.

Methods

Study area

Larval fish and ichthyoplankton samples were collected from three areas within the lower Roanoke River and Albemarle Sound, North Carolina. These areas were classified as River, Delta, and Sound and contained a total of 19 stations. River is the area furthest upstream with seven stations between RKM 9.5 – 22, scattered throughout the main stem of the river and its tributaries and distributaries. Delta is the transitional region between River and Sound where the Roanoke, Middle, and Cashie Rivers converge at the Highway 45 Bridge, before diverging and flowing into the Albemarle Sound. There are two stations each in the Roanoke, Middle, and Cashie rivers, for a total of six stations. The Delta station furthest upstream is located in the Roanoke at RKM 5. Sound has six stations in Batchelor Bay, the western portion of Albemarle Sound. The stations extend 2 – 4 km from the mouths of the Roanoke and Middle Rivers (Fig. 3.1).

Larval fish and zooplankton collection

Larval fish and zooplankton samples were collected concurrently March through June in 2008 and 2009. Sampling began at sunset. Ichthyoplankton were collected using paired surface pushnets mounted on the bow of the boat. Each net was housed in an aluminum frame with a 0.5 m square opening. Each larval fish net had a 5:1 ratio and was constructed from 505 μm nitex mesh. A Sea-Gear model MF315 flowmeter was mounted in the center of each net to estimate the amount of water filtered during each tow. The nets were pushed into the current for two minutes at a speed of 1.03 ± 0.11 m/s (Overton and Rulifson 2007). The contents of each net were condensed into a 1 L plastic collection jar. The contents of the left net were preserved with 95% ethanol while the contents of the right net were preserved with 5% buffered formalin. The

amount of ethanol used for preservation changed between the two sampling years. In 2008, the ratio of sample water to ethanol was approximately 70% sample water and 30% ethanol and there was a high amount of deterioration. In 2009, the amount of ethanol used for preservation was increased with a ratio of water to ethanol was closer to 5% sample water and 95% ethanol. To determine larval abundance, the catches between the two nets were averaged together.

Zooplankton samples were collected using a 3:1 conical net with a 0.5 m opening and 90 μm nitex mesh. The plankton net was deployed using a vertical net haul technique where the net is lowered to the bottom and then pulled vertically through the water column. The contents of the net were washed down and condensed into a sample jar and preserved with 5% buffered formalin. In 2009, samples were not collected in the Delta during calendar week 20 and during weeks 12, 16, and 20 in the Sound because of mechanical issues with the boat and severe weather.

Data processing and analyses

In the laboratory, larval alosines were identified to species, enumerated, and notochord length was measured to the nearest 0.1 mm. Alosine abundance data did not meet normality assumptions, even with data transformation and nonparametric tests were used for comparisons. Yearly differences in alosine abundance were evaluated using a Wilcoxon Rank-Sum Test. Kruskal-Wallis ANOVA evaluated abundance differences among species and sampling areas. If the ANOVA was significant, post-hoc comparisons were conducted using Wilcoxon Rank-Sum Tests with family wise alpha being controlled by a Dunn-Sidak adjustment. SAS 9.2 (SAS Institute, Cary, North Carolina) was used to conduct all statistical analyses.

For mouth gape analysis, only individuals preserved in formalin were used. Few American shad larvae were collected, so mouth gape analysis was conducted on larvae obtained from U.S. Fish and Wildlife Service Edenton National Fish Hatchery, Edenton, North Carolina. Larvae were separated into 1 mm size bins based on notochord length. In bins where there were > 20 fish, a minimum of 20 larvae in each bin was analyzed. In bins with ≤ 20 larvae, all possible fish were analyzed. All measurements were done using Image-Pro Discovery 4.5. For each fish, the upper and lower jaws were measured to the nearest 0.1 mm. The upper jaw was measured across the premaxillae and maxillae to the point of articulation with the dorsal process of the dentary. The lower jaw was measured along the length of the dentary to the point of articulation with the angular and maxillae. Mouth gape was calculated using the law of cosines for a mouth open at 90° angle (Riley et al. 2009). This angle is considered the maximum functional degree of opening for feeding in most larval species (Turingan et al. 2005; Riley et al. 2009). Mouth gape models for each species were created using linear regression analysis. A one-way analysis of covariance (ANCOVA) tested if mouth gapes differed significantly among species. Prey size estimates are based on the length when the yolk sac is absorbed and larvae begin to feed exogenously. Yolk sac absorption occurs when alewives and blueback herring are 6 mm, at 7 mm in hickory shad, and between 9-12 mm in American shad larvae (Lippson and Moran 1974).

For each sampling date, three zooplankton samples from each area were randomly selected for processing. Subsamples were taken using a Hensen-Stempel pipette. Individuals were counted and identified to the lowest possible taxon using an Olympus Model SZX-ILLD100 stereomicroscope. Zooplankton were identified using taxonomic keys found in Thorp and Covich (2001) and Balcer et al. (1984). Body length and widths were measured, to the

nearest 0.1 mm, using Image-Pro Discovery 4.5. Zooplankton abundance values estimated in Chapter 2 were used in this chapter to evaluate spatial temporal overlap between larval alosines and zooplankton.

Results

Alosine abundance and distribution

Differences in alosine abundance (number/100 m³ ± SD) and composition were observed between sampling years. In 2009 (30.8 ± 149.8) abundances were significantly higher ($p \leq 0.001$) than in 2008 (4.1 ± 20.9). Alewives (28.5%) were the most abundant alosine in 2008, followed closely by blueback herring (26.7%) and hickory shad (24.4%). In 2009, blueback herring were dominant, accounting for 64.8% of larval alosines collected. Hickory shad (23.8%) were the second most abundant species. American shad were the least common alosine for both years (Table 3.1).

Weekly patterns were different between the two years. Larval alosines were collected during all weeks of sampling in 2008, but were not present the first week of sampling in 2009. Blueback herring were the first species collected both years. Alewives were observed earlier in 2008, and were the 2nd species collected. In 2009, hickory shad were the 2nd species collected. American shad larvae were not common (Fig. 3.2).

Strong spatial differences in larval abundance were observed. Mean alosine abundances were significantly different ($p \leq 0.001$) among all three sampling areas. Highest abundances were observed in the River (21.0 ± 127.6) and were lower in the Delta (7.4 ± 35.5) and Sound (4.6 ± 24.8). This trend was consistent across both years and for all species. Average monthly abundances were always highest in the River, except for American shad in May 2008 (Fig. 3.3).

Clear patterns are not present when species abundance is analyzed on temporal and spatial scales. Alewife abundances were highest in the River, in March of both years. In 2008, alewife abundances peaked in March where in 2009 peak abundances were observed in April. In the Sound, alewives were present all months in 2008, but were absent in March 2009. Blueback herring abundances were consistent in 2008 within each area. There was an increase in blueback herring abundances in 2009 and abundances varied across the months. In all areas, abundances peaked in April and were at least twice as large as abundances observed in March. After peaks in April, there was a sharp decrease in abundances for the remaining months. In 2009, hickory shad patterns in abundance were similar to blueback herring. Hickory shad also had an increase in abundances in 2009 with peaks in April. In 2008, monthly hickory shad trends were similar in the River and Delta, with highest abundances in April (Fig. 3.3).

Alosine mouth gape analysis and prey size estimates

Larval alosine notochord lengths ranged from 3 – 14 mm, with 97% of larvae ≤ 7 mm. Over 90% of alewives, blueback herring, and American shad were between 4 -7 mm, with lengths > 8 mm rarely collected. American shad larvae had the narrowest length distribution and were typically larger, with all but one larva ≥ 7 mm (Fig. 3.4). Mean length ($\mu \pm SD$) was similar between alewives 4.7 ± 1.2 and blueback herring 4.6 ± 1.0 . Mean hickory shad length (6.5 ± 1.0) was larger than alewives and blueback herring. American shad larvae had the largest mean length (8.7 ± 2.1)

For all species, there was a strong linear relationship between mouth gape and notochord length (Fig 3.5). A one-way ANCOVA indicated there were significant differences, ($F(4,459) = 2115.0, p < 0.001$), in mouth gape sizes among species. American shad larvae had the largest

mean mouth gape ($\mu = 0.67$ mm), followed by alewives ($\mu = 0.57$ mm), blueback herring ($\mu = 0.56$ mm), and hickory shad ($\mu = 0.53$ mm). Alewife and blueback herring mouth gapes did not significantly differ from each other, while all other comparisons were significantly different.

For mouth gapes calculated at a 90° opening, estimated prey size at yolk sac absorption was similar among alewives, blueback herring, and hickory shad larvae and are wider for American shad larvae. At 6 mm, alewife and blueback herring have a $400\ \mu\text{m}$ mouth gape with an estimated maximum prey size of $200\ \mu\text{m}$. Hickory shad at 7 mm have a $430\ \mu\text{m}$ mouth gape and an estimated prey size of $215\ \mu\text{m}$. At 9 mm, American shad mouth gape is twice as wide at first feeding. Mouth gape is estimated at $820\ \mu\text{m}$ with a maximum prey size of $410\ \mu\text{m}$ (Fig. 3.5).

Zooplankton size distribution was estimated for the most abundant taxa in Chapter 2. These taxa included calanoid and cyclopoid copepods, copepod nauplii, rotifers, Daphniidae, and Bosminidae, which accounted for 98% of total abundance. The greatest variation in body length is seen with calanoid copepods, cyclopoid copepods, and Daphniidae, with differences in weekly length exceeding $400\ \mu\text{m}$. Bosminidae lengths showed some weekly variation with length differences $< 200\ \mu\text{m}$. Copepod nauplii body lengths did not vary greatly. Overall, there was little change in rotifer length, but there were a few weeks where a large increase was observed (Fig. 3.6).

Mean zooplankton body length and widths are plotted in Fig. 3.7. Based on these measurements, zooplankton taxa separated into two size classes. The smaller taxa included rotifers and copepod nauplii with remaining taxa comprising the larger size class. Along the length axis the two groups begin to separate at $200\ \mu\text{m}$ and at approximately $120\ \mu\text{m}$ across the

width axis. Based on mouth gape estimates, copepod nauplii and rotifers are size appropriate for alewives, blueback herring, and hickory shad larvae at first feeding. Bosminidae and cyclopoid copepods at the low end of their size distributions could also serve as potential prey for these species. American shad larvae have a wider potential prey breadth including rotifers, copepod nauplii, cyclopoid copepods, and Bosminidae. Smaller Daphniidae and calanoid copepods are also within American shad estimated prey size range.

Larval alosine and zooplankton spatial and temporal overlap

There was a high degree of spatial and temporal overlap between larval alosine and zooplankton abundances in all three sampling areas. Weekly mean alosine abundances were generally < 1 number/m³. Mean weekly zooplankton abundances, including all taxa, ranged from 934 – 51,815 number/m³. When abundances of the most suitable prey (copepod nauplii and rotifers), were evaluated separately, weekly abundances ranged from 283 – 51,034 number/m³. There is always overlap between larval alosines and size appropriate prey. Spatially, the highest overlap occurs in the Sound where zooplankton abundances were the highest and larval alosine abundances were the lowest. Higher weekly peaks occur in the River than in the Delta, but there is still overlap in zooplankton and larval alosines in all areas (Fig. 3.8).

Discussion

Differences in mouth gape in coexisting larvae allows for a reduction in interspecific competition (Crecco and Blake 1983; Bremigan and Stein 1994; Makrakis et al. 2008). In this study, alewife and blueback herring mouth gapes were not significantly different from each other, but all other comparisons with American shad and hickory shad were significant. Mouth

gape calculations for blueback herring and hickory shad are comparable to those calculated by Crecco and Blake (1983). When mouth gape size at yolk sac absorption was calculated, alewife, blueback herring, and hickory shad had similar mouth gapes and American shad mouth gapes were larger.

Temporal differences in spawning have been observed as a mechanism to reduce competition between alewife and blueback larvae in areas where their distributions overlap. Alewives typically begin spawning in late February or early March, while blueback herring begin spawning in late March (O'Connell and Angermeier 1997; Able and Fahay 1998). In both years of this study, blueback herring larvae were collected earlier than alewife larvae. This was also observed in the Tar-Pamlico River, North Carolina (Overton et al. in review). Temporal overlap was greater in 2008 between these two species. Blueback herring larvae were collected one week earlier, than alewife larvae, and both were collected the remaining weeks of the study. In 2009, alewives were not collected until week 15, following a peak in blueback herring abundance the previous week. Hickory shad larvae also experienced a temporal overlap with both blueback herring and alewives (Fig. 3.2). Temporal differences, on a weekly scale, were observed, and there were differences in when peak abundances occurred. Competition among larvae feeding exogenously may be reduced by temporal differences in spawning peaks and not by a complete separation in spawning activity. Larval American shad mouth gape is significantly larger than the other alosine species, and they do not have to compete for prey, but were rarely collected and least abundant. Food availability does not appear to be the driving force behind low American shad numbers.

Maximum larval fish mouth gape is typically estimated for 90 degrees, but some larval fish are capable of opening their mouths to 120 degrees (Riley et al. 2009). Hickory shad mouth morphology is different from the other alosine species. The lower jaw of hickory shad slopes at an angle $> 40^\circ$, while the other species lower jaw slopes at an angle $< 40^\circ$ (Walsh et al. 2005). This difference in morphology may allow hickory shad to open their mouths at larger angles, enabling them to consume larger prey at smaller sizes. This could be one of the reasons hickory shad populations have remained more stable than the other alosines. If hickory shad are capable of feeding at a larger mouth gape opening, this could act as a way to reduce competition between hickory shad and river herring. At first feeding, hickory shad may be capable of consuming larger prey items than predicted in this study, allowing them to feed on prey resources not available to river herring. If mouth gape for hickory shad is calculated at a 120° opening, optimal prey sizes at first feeding are at $300 \mu\text{m}$. This maximum prey size is between the estimated prey sizes for river herring and American shad larvae with a 90° opening. This would reduce competition between river herring and hickory shad. Feeding studies need to be conducted with hickory shad to determine if they are capable of opening their mouths at larger angles.

Zooplankton abundances were much higher in this study than those previously conducted and there was a high ratio of zooplankton to larval alosines (See Chapter 2). This high overlap was observed even when only copepod nauplii and rotifers were included in the analysis. Competition among larval alewives, blueback herring, and hickory shad could be possibly reduced by the large volume of available prey. Studies evaluating diet niche overlap found dissimilarities among larval fish with similar mouth gapes (Gaughan and Potter 1997; DeVries et al. 1998; Makrakis et al. 2008). When zooplankton abundances are high, competition is reduced

and larval fish with similar mouth gapes exhibit different feeding behaviors and selectivity because of by high prey availability (Gaughan and Potter 1997).

During both years in all areas, there was a high amount of overlap between zooplankton and larval alosines. Laboratory studies examining growth and survival are conducted with larval alosine and zooplankton abundances higher than those observed in this study. In Riley et al. (in review), larval American shad growth and survival were evaluated at three different prey abundances equivalent to 1,000, 50,000, and 500,000 number/m³. These abundances reflect all zooplankton and not just size appropriate prey. Larval American shad were stocked at abundances equivalent to 4,000 number/m³. Growth was significantly higher in treatments with the two highest abundances, but survival rates were similar in all three treatments. Except for one observation in the Sound, zooplankton abundances never exceeded 50,000, but alosine abundances were typically < 1 number/m³. In Johnson and Dropkin (1995), larval American shad were stocked at the equivalent of 8,000 number/m³ and *Artemia* nauplii were stocked at 500,000 and 1,000,000 number/m³. Growth was not different between the two treatments, but survival was higher in the treatment with larger nauplii abundances. For both studies, if zooplankton abundance is divided by larval shad abundance, the zooplankton to fish ratio is 125:1. Ratios this low were never observed during this study, even when calculated with rotifers and copepod nauplii only. This suggests larval alosines in this system are not food limited and all three areas are suitable to serve as larval alosine nursery habitat based on prey availability. Further research is needed to determine the amount of overlap between zooplankton and all planktivores in lower Roanoke River and Albemarle Sound, North Carolina.

Table 3.1: Comparison of larval alosine species composition in 2008 and 2009 in Roanoke River and Albemarle Sound, North Carolina.

Species	Frequency percent (%)	
	2008	2009
Alewife	28.5	11.1
<i>Alosa spp.</i>	18.3	0.2
American shad	2.1	0.2
Blueback herring	26.7	64.8
Hickory shad	24.4	23.8

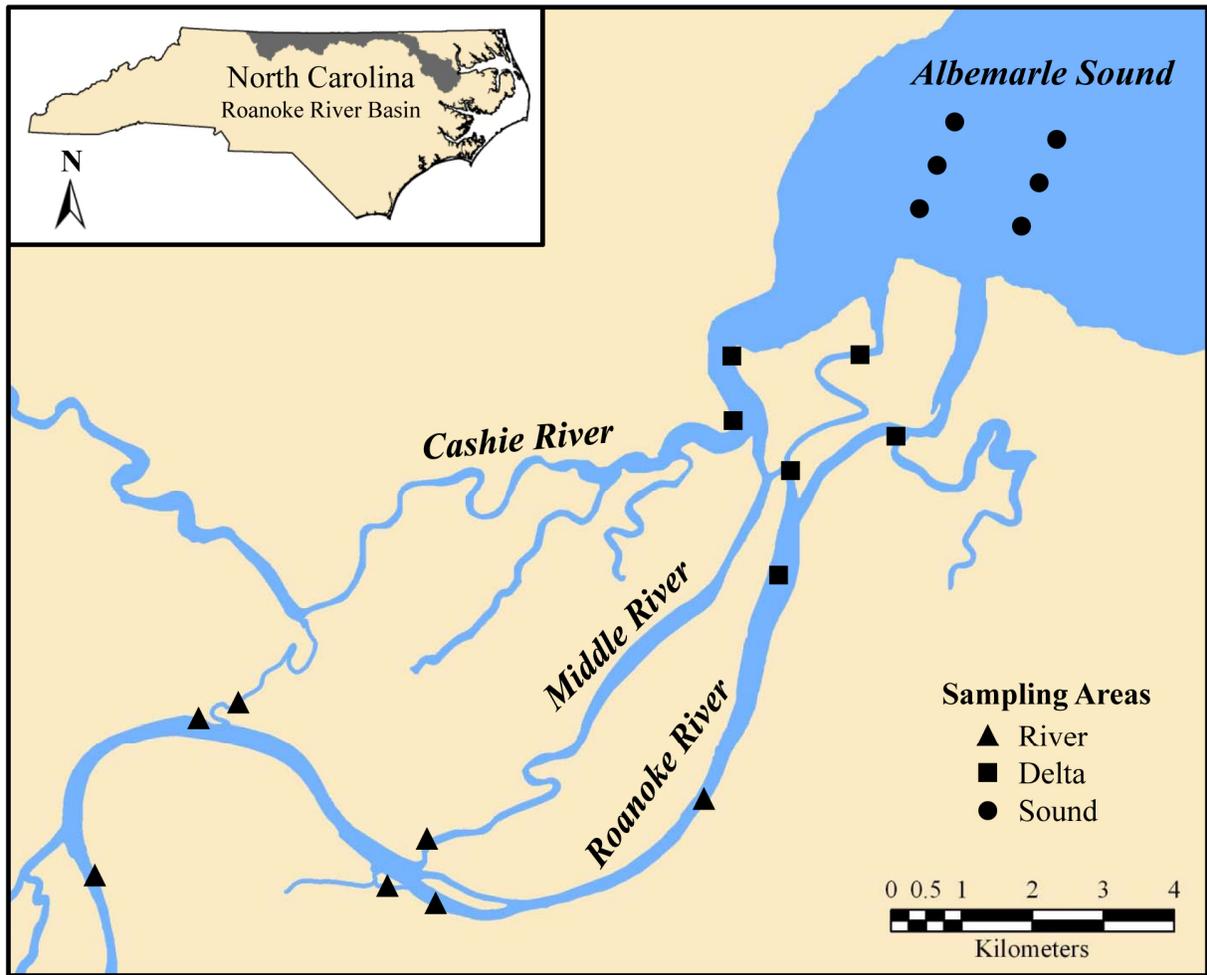


Fig 3.1. Location of the sampling stations and the division of the three sampling areas in lower Roanoke River and Albemarle Sound, North Carolina.

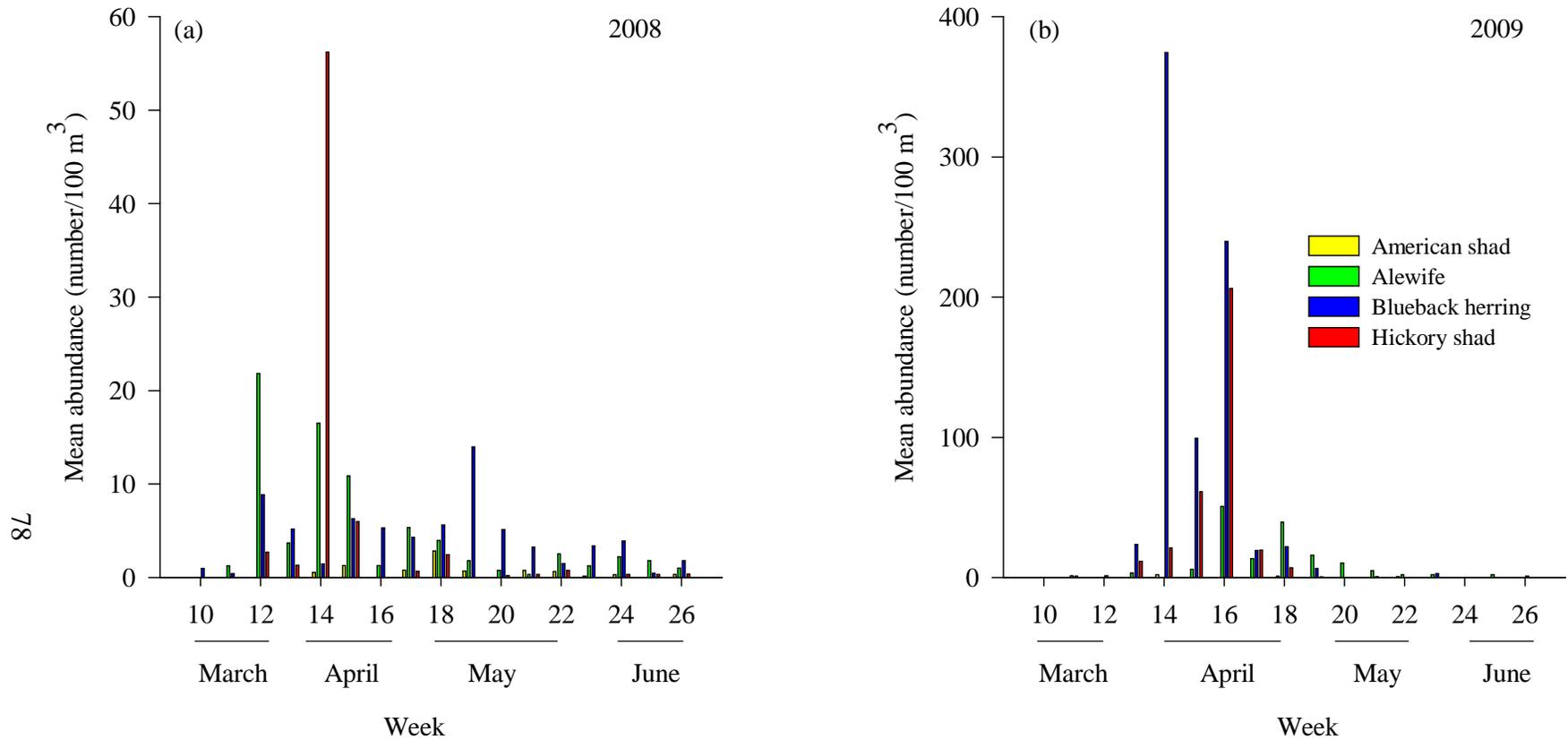


Fig. 3.2. Mean weekly abundance of larval American shad, alewives, blueback herring, and hickory shad in (a) 2008 and (b) 2009 in Roanoke River and Albemarle Sound, North Carolina. Note the scale change.

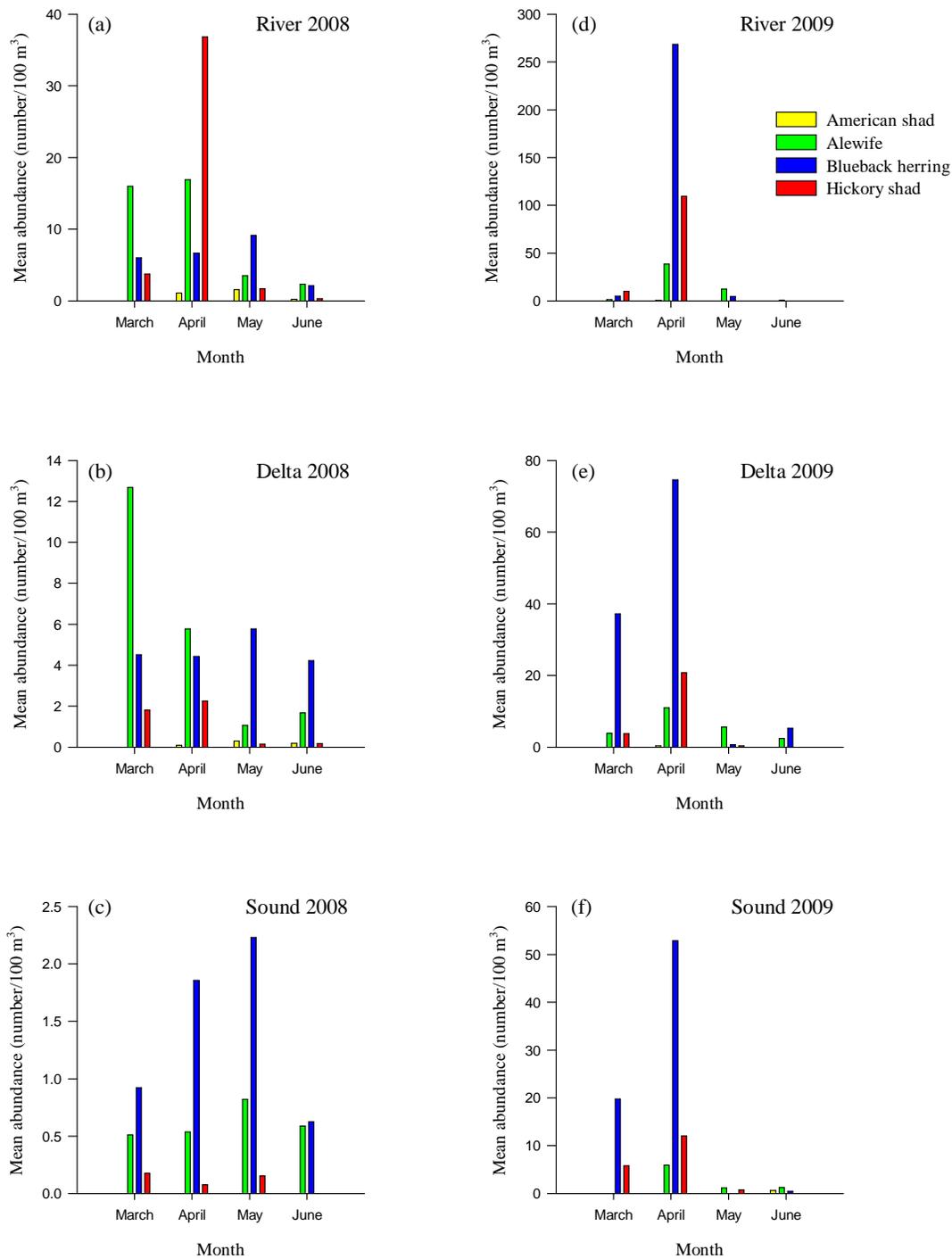


Fig 3.3. Comparison of alosine species composition in (a) River 2008, (b) Delta 2008, (c) Sound 2008, (d) River 2009, (e) Delta 2009, and (f) Sound 2009 in Roanoke River and Albemarle Sound, North Carolina. Note change in scale.

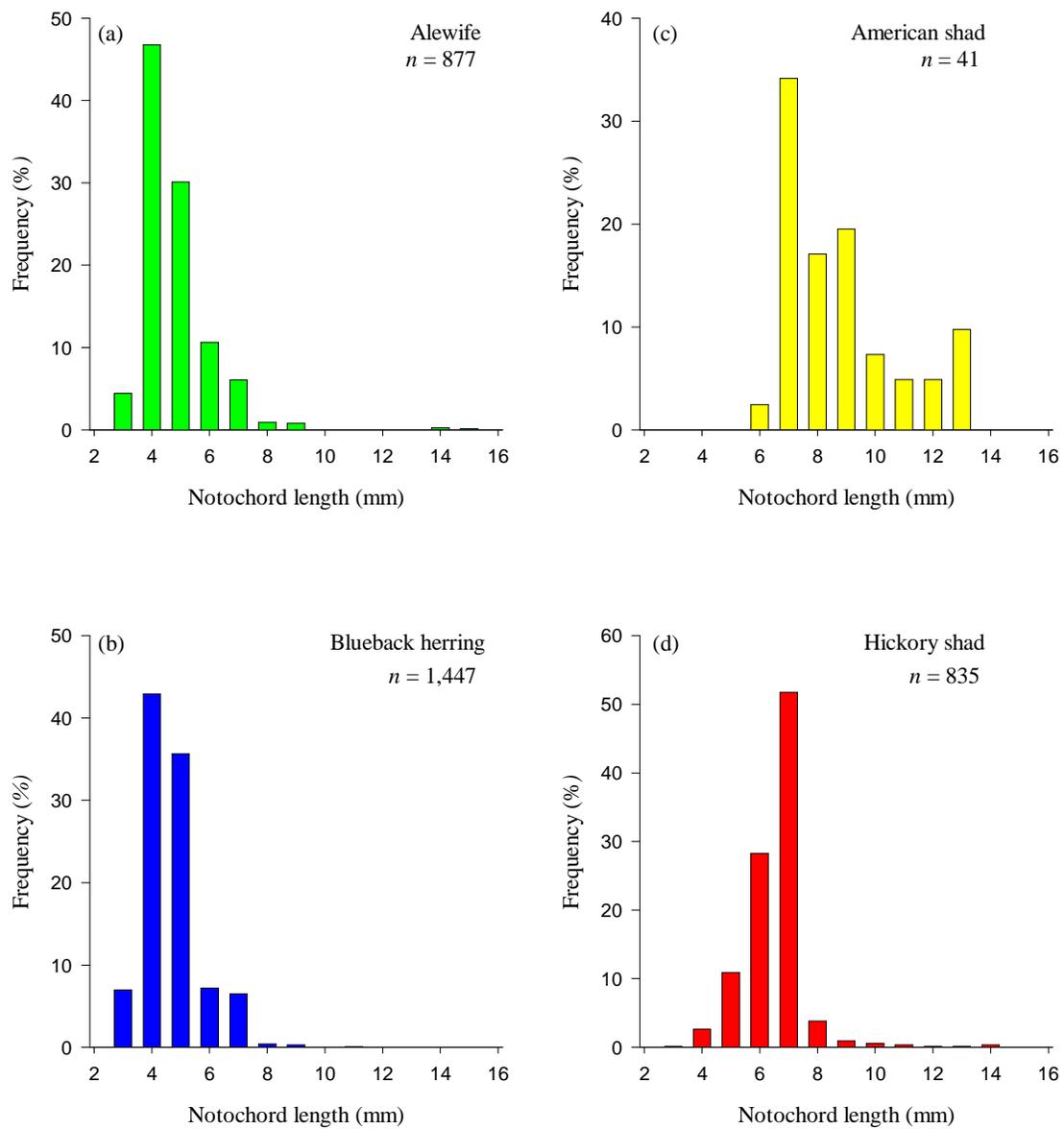


Fig 3.4. Frequency (%) distribution of each alosine species collected in Roanoke River and Albemarle Sound, North Carolina.

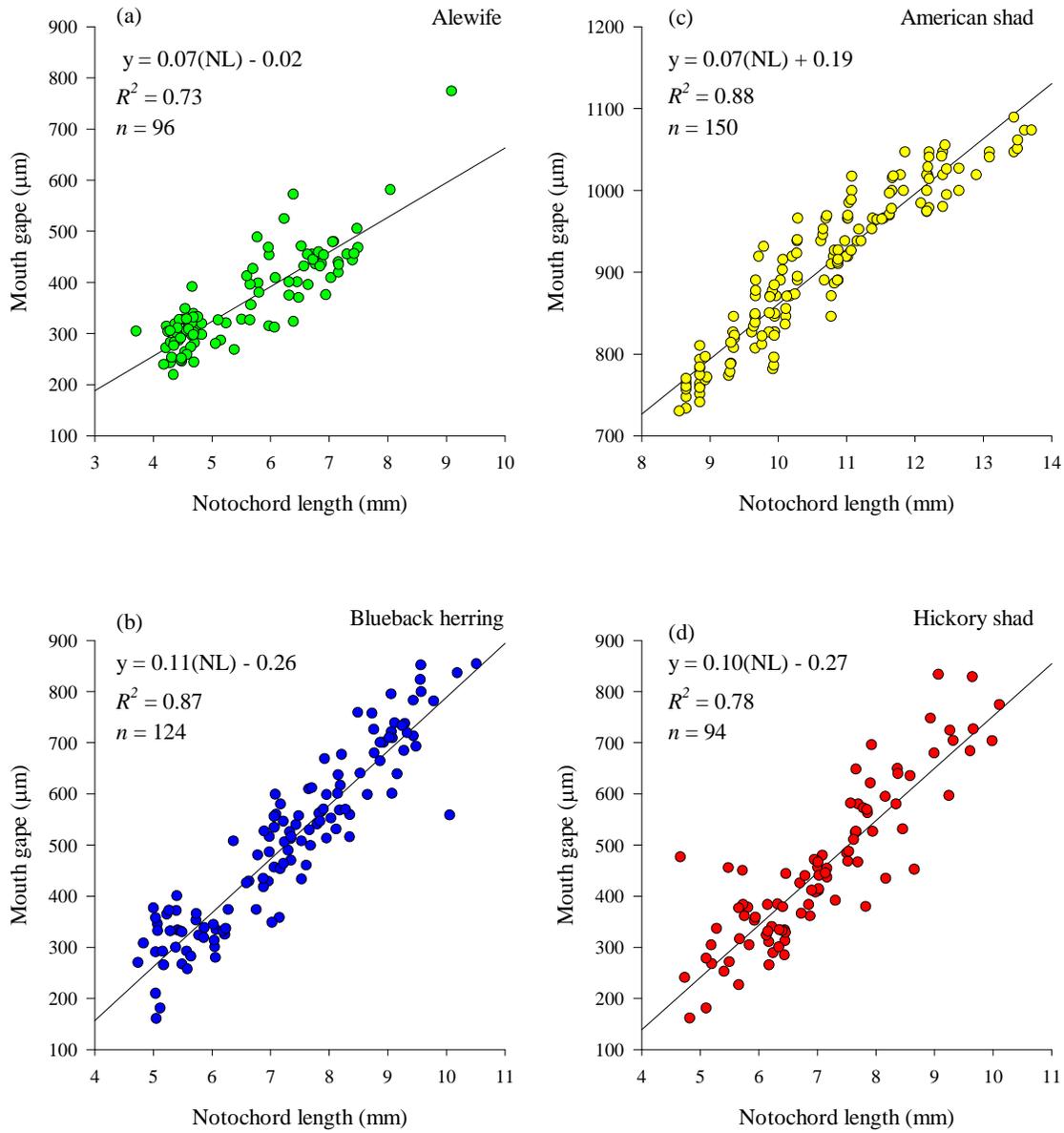


Fig. 3.5. Mouth gape regression models for (a) alewife, (b) blueback herring, (c) American shad, and (d) hickory shad. Models are calculated with the mouth open at 90°. Note change in scale.

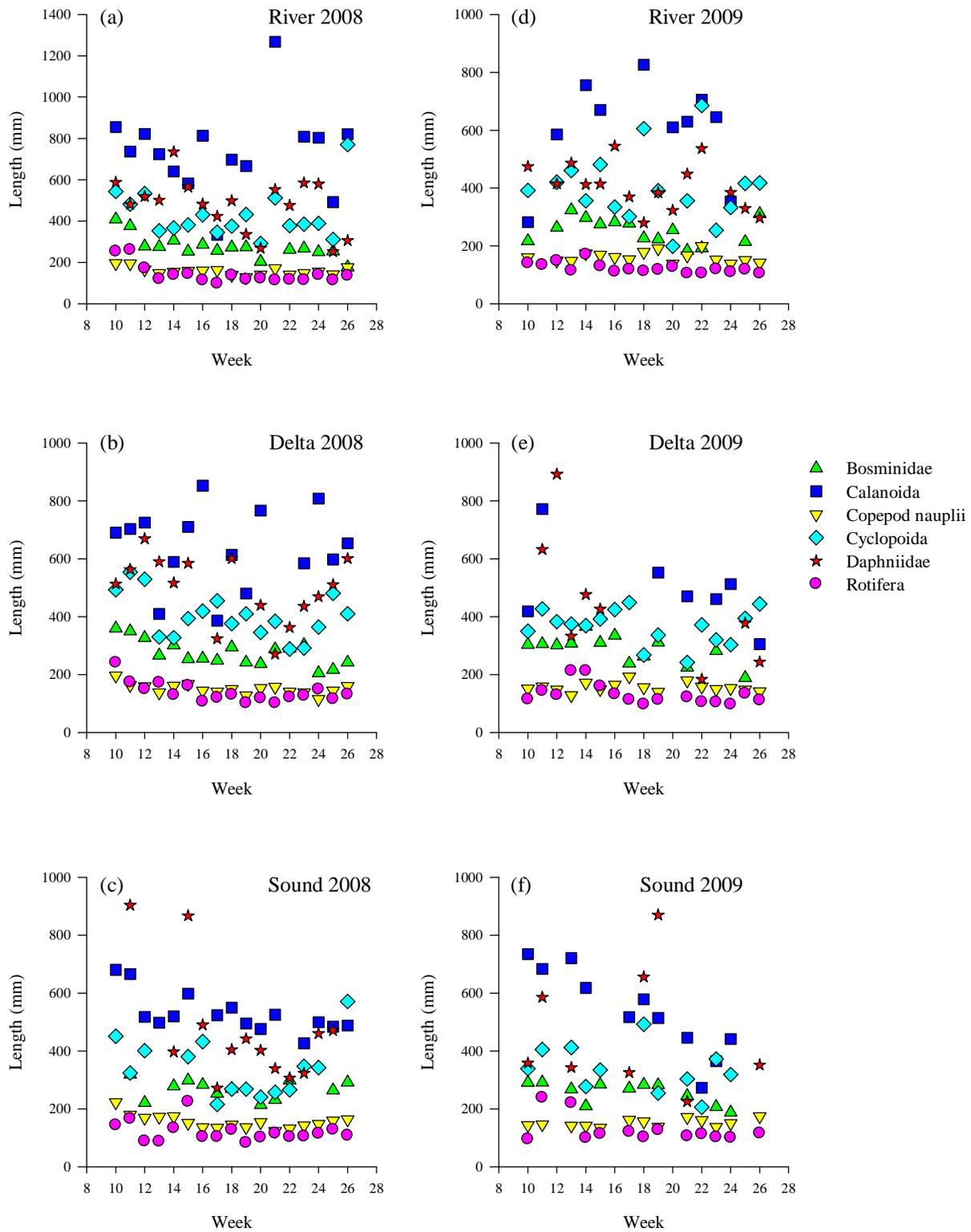


Fig. 3.6. Mean weekly length for the most common zooplankton taxa in (a) River 2008, (b) Delta 2008, (c) Sound 2008, (d) River 2009, (e) Delta 2009, and (f) Sound 2009.

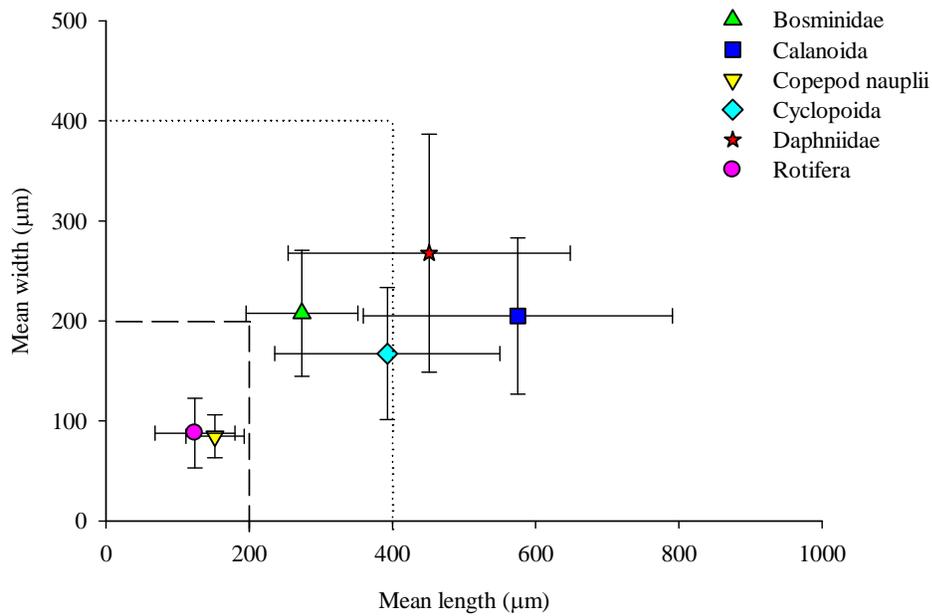


Fig. 3.7. Body lengths and widths for the most common zooplankton taxa collected in Roanoke River and Albemarle Sound, North Carolina. Values represent $\mu \pm$ standard deviation. The dashed lines represent maximum prey size for larval alewives, blueback herring, and hickory shad at first feeding. The dotted line represents maximum prey size for larval American shad at first feeding.

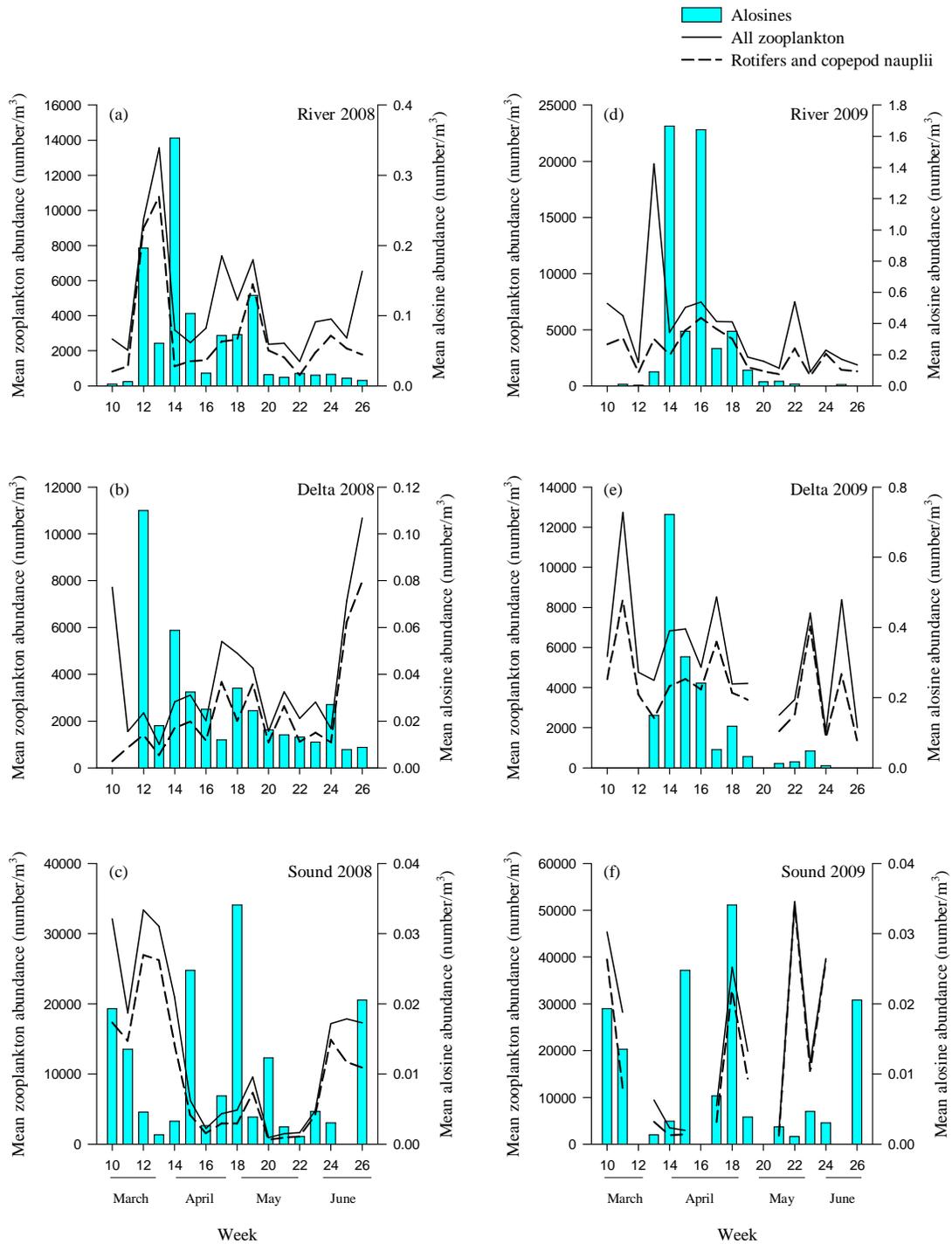


Fig. 3.8. Zooplankton and larval alosine spatiotemporal overlap in (a) River 2008, (b) Delta 2008, (c) Sound 2008, (d) River 2009, (e) Delta 2009, and (f) Sound 2009. Note the scale change.

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CHAPTER 4: DISCUSSION

Zooplankton abundance and composition

Zooplankton trends were analyzed within three areas in lower Roanoke River and Albemarle Sound where studies were previously conducted. A significant increase in abundance was observed in all three areas. Abundances from this study were more comparable to abundances documented in other North Carolina river systems. Zooplankton composition was affected by many environmental parameters, but salinity and temperature were two of the most influential.

Spatial differences in environmental parameters explained differences in zooplankton composition in the Sound when compared to River and Delta, but no clear evidence emerged to explain why abundances were higher in the Sound. It is possible that top down regulation is occurring within this system. Grazing by larval and juvenile fish could be causing lower abundances in the River and Delta areas (Bollens 1988). Throughout the study, larval fish abundances, for all species were higher in the River and Delta than in the Sound. In Lake Michigan, it was estimated that larval and juvenile alewives consumed 2-8% of zooplankton biomass daily (Hewett and Stewart 1989). In addition to alosines, many fish species utilize the Roanoke River and Delta region for spawning and nursery habitats. Striped bass (*Morone saxatilis*) and white perch (*M. americana*) are both spring spawners that use this area. The glass eel stage of American eel and several resident species in the families of Centrarchidae, Cyprinidae, and Ictaluridae are also found in this system. All of these species were collected during this study and during previous work by Overton and Rulifson (2007). Modeled population dynamics between larval fish and zooplankton communities demonstrated the ability

of larval fish grazing to reduce zooplankton abundance (Bollens 1988). Declining zooplankton abundance with increasing larval fish abundance was shown in several mesocosm experiments (Welker et al. 1994; Qin and Culver 1996). In the Roanoke River, alosine, striped bass, and white perch spawning begins in late March-early April. During this study, the majority of zooplankton peaks were observed before the onset of spawning by these fish.

The ability of larval fish to exert top down control on zooplankton abundance in open systems has been debated. Larval alosine abundances were typically < 1 number/m³. These abundances are considered too dilute to have a significant effect on zooplankton abundances that are typically at least one to two orders of magnitude larger (Cushing 1983). Diet analysis of larvae collected in Conception Bay, Newfoundland, Canada supported this hypothesis. Pepin and Penney (2000) estimated total zooplankton consumption by larval fish was $< 0.1\%$ of potential prey in that system.

Larval alosine abundance

Mouth gape analysis for larval alewives, blueback herring, and hickory indicated copepod nauplii and rotifers were suitable prey items at the onset of exogenous feeding. American shad larvae are larger at first feeding and able to consume a wider range of prey items. When spatial and temporal overlap between larval alosines and size appropriate prey was evaluated, a high degree of spatial and temporal overlap was observed. Larval alosines in this system do not appear to be food limited.

Low alosine abundances could be related to high levels of predation on larval alosines. In laboratory experiments, cyclopoid copepods were observed attacking and consuming both alewives and blueback herring (Binion, personal observation). While processing preserved

ichthyoplankton samples, larval alewives, blueback herring, and hickory shad were observed with cyclopoid copepods attached to their bodies. It is not possible to know whether this occurred prior to or during the collection when the zooplankton and fish were artificially concentrated, but it does support the idea of copepod predation on larval fish. Cooper (1996) also observed evidence of cyclopoid copepod predation on ichthyoplankton collected in Roanoke River. McGovern and Olney (1988) examined predation on larval striped bass in laboratory experiments. The authors documented high rates of predation by cyclopoid copepods and larval spottail (*Notropis hudsonius*) and satinfish (*N. analostanus*) shiners. Predation by juvenile striped bass, white perch, shiners, minnows, sunfish, and catfish were also observed. It is possible high predation rates on larval alosines are suppressing populations.

Future research needs

Long term zooplankton monitoring is needed within lower Roanoke River and Albemarle Sound, North Carolina to determine if the higher abundances observed in this study are the new baseline for this system or just an anomaly. With long term observation, the relationship between environmental parameters and zooplankton can be better understood as well as the factors driving zooplankton dynamics.

Alosine stocks need to continue to be monitored. By assessing adult populations for the next 1-2 years, it can be evaluated if high levels of prey availability in 2008-09 translated to increases in adult recruitment. More larval alosine studies are needed to figure out if there is a link between larval mortality and adult recruitment or if factors later in life are responsible for inability of stocks to recover.

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APPENDIX A: ANIMAL CARE AND USE



Animal Care and Use Committee
East Carolina University
212 Ed Warren Life Sciences Building
Greenville, NC 27834
252-744-2436 office • 252-744-2335 fax

May 4, 2007

Anthony Overton, Ph.D.
Department of Biology
Howell Science Complex
East Carolina University

Dear Dr. Overton:

Your Animal Use Protocol entitled, "Defining Essential Fish Habitat in Estuaries and Coastal River Systems: A Model-Based Approach," (AUP #D214) was reviewed by this institution's Animal Care and Use Committee on May 4, 2007. The following action was taken by the Committee:

"Approved as submitted"

A copy is enclosed for your laboratory files. Please be reminded that all animal procedures must be conducted as described in the approved Animal Use Protocol. Modifications of these procedures cannot be performed without prior approval of the ACUC. The Animal Welfare Act and Public Health Service Guidelines require the ACUC to suspend activities not in accordance with approved procedures and report such activities to the responsible University Official (Vice Chancellor for Health Sciences or Vice Chancellor for Academic Affairs) and appropriate federal Agencies.

Sincerely yours,

A handwritten signature in cursive script that reads "Robert G. Carroll, Ph.D."

Robert G. Carroll, Ph.D.
Chairman, Animal Care and Use Committee

RGC/jd

enclosure



Animal Care and Use Committee
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MEMORANDUM

TO: Samantha Binion
Department of Biology

FROM: Dorcas O'Rourke, D.V.M. 
University Veterinarian

SUBJECT: Certificate of Training
Training Date 1/15/09

DATE: January 30, 2009

This letter is provided to certify that you have completed training in humane methods of animal experimentation, proper handling of selected species of research animals, and methods for reporting deficiencies in animal care and treatment. The training was provided in accordance with U.S. Department of Agriculture (9 CFR 2.32) regulations and the Public Health Service Policy.

This training included information on ECU animal care organizational structure, regulatory requirements, IACUC procedures, program for veterinary and animal care, occupational health and safety program, and methods for reporting concerns. Information on biology and care, proper restraint and procedures, and allergies and zoonoses were also provided.

We suggest that you retain this letter in your training file for future reference.

BIOGRAPHICAL SKETCH

I was born in Flatwoods, Kentucky, and raised in Atlanta, Georgia. In 2003, I received a B.S. in Biology from Emory University, Atlanta, Georgia. As an undergraduate, I completed internships at Mote Marine Laboratory in Sarasota, Florida and Marine Biological Laboratory, Woods Hole, Massachusetts. These internships solidified my interests in fisheries research. After graduation, I returned to Mote and was a Staff Biologist II in the Center for Coastal Ecology. While at Mote, I studied the effects of power plant impingement, entrainment, and thermal discharge on zooplankton, larval and juvenile fish communities in Tampa Bay, Florida. I left Mote in 2008 to enter the graduate program at East Carolina University.