The Role of the Cape Fear River Discharge Plume in Fisheries Production: Aggregation and Trophic Enhancement

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ABSTRACT

Estuarine habitats have long been valued as critical nurseries for estuarine dependent species of finfish and shellfish. The role of nearshore coastal habitats, particularly river discharge plumes, in fisheries production is less clear. Past researchers have suggested that discharge plumes may enhance fisheries production by: 1) aggregating larval fishes and crustaceans, and 2) providing them with a trophic advantage compared to adjacent ocean habitats. I tested these hypotheses through sampling in the Cape Fear River discharge plume (CFR), North Carolina. Physicochemical conditions were described for plume and adjacent ocean habitats to compare their relative suitability for ichthyoplankton. Temperature and salinity were significantly different between ocean and plume habitats and between the surface and bottom for 2002. In contrast, temperature was not significantly different between ocean and plume habitats but salinity was for 2003. Monthly ichthyoplankton sampling was conducted at stations inside and outside the CFR plume during 2002 and 2003. Ichthyoplankton were sampled using 60cm plankton nets (705µm mesh) towed at the surface, 1 meter, and bottom depths during daylight hours on ebbing tides to: 1) compare larval concentrations and diversities, and 2) examine vertical distribution.

Thirty-three taxa of fishes from twenty-two families were collected in 2002 and eighteen taxa from eleven families were collected in 2003 with a total of 1,497 larval fishes collected. Dominant families for 2002 were Sciaenidae, Blenniidae, and Gobiidae. The dominant families differed slightly for 2003 and were Engraulidae, Gobiidae, and Sciaenidae. Seasonal transitions in taxonomic composition occurred in the fall and spring of both years. Total ichthyoplankton concentrations and diversities were generally
higher inside of the plume than at ocean stations. Within the plume, ichthyoplankton concentrations were higher at the bottom than at surface and 1 meter depths, suggesting that larvae may be utilizing selective tidal stream transport to facilitate estuarine ingress. Larval diversities for the ocean habitat were highest at the depth of 1 meter. Biochemical assays (cytochrome c oxidase and hexokinase activities) and otolith microstructural analysis were used as condition indices to examine Atlantic croaker (Micropogonias undulatus), spot (Leiostomus xanthurus) and brown shrimp (Farfantepenaeus aztecus) collected from plume versus estuarine habitats. No significant differences in enzyme activities or recent growth rates were observed, suggesting that the plume habitat may promote levels of physiological condition comparable to those of estuarine habitats. Future studies to examine the function of river discharge plumes in fisheries production should focus on habitat suitability and compare physiological condition of fishes and crustaceans in these habitats to surrounding waters. Improved understanding of the influence of discharge plumes is needed to determine whether these areas may be designated as Essential Fish Habitat.
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DEDICATION

This thesis is for my wife and family, whose support and understanding helped me accomplish my goal.
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INTRODUCTION

Fisheries Recruitment

Recruitment is essential to the viability of fish populations and fisheries (Ross 1997). The term recruitment refers to the amount of fish added to the exploitable stock each year due to reproduction, growth, and/or migration into the fishing area. Recruitment therefore has a controlling influence on adult population size (Jennings et al. 2001). Consequently, a major goal of fisheries biology over the last twenty years has been to better understand the process of recruitment and its causal mechanisms (Jennings et al. 2001). By having a better understanding of recruitment, fishery managers may be able to estimate fluctuations of population sizes in advance and adjust harvest levels accordingly (Grimes and Finucane 1991).

Nursery Habitats and Their Function

Habitat characteristics are known to influence recruitment through their effects on the feeding, growth and survival of young fishes (Diaz et al. 2003; Lankford and Targett 1994; Rooker et al. 1998). Certain habitats are recognized as critical for recruitment because they have a tendency to increase growth and survival for young fishes. For example estuaries are often considered to be critical nurseries that enhance recruitment of many commercially important fishes and shellfish species (Day et al. 1989). Estuaries occur globally in a variety of geographic regions and provide numerous complex habitats for fishes and decapods to utilize. Salt marshes for example are widely believed to function as nurseries because of their high production of vascular plant detritus and the structural refuge they provide individuals from predation (Boesch and Turner 1984). Seagrass beds may also provide refugia from predators. Heck and Crowder (1991)
conducted a study on predation of fishes in seagrass beds and noticed that predation was lower in structurally complex beds, suggesting that structural complexity may enhance recruitment by increasing survival for many species. Such protected inshore habitats are referred to as nursery habitats. Habitats such as these are recognized for having high primary and secondary productivity as well as supporting large abundances and diversities of fishes and invertebrates (Beck et al. 2001). These estuarine ecosystems are commonly regarded as nurseries because of their profound effect on diversity and productivity of macrofauna (Boesch and Turner 1984, NRC 1995, Butler and Jernakoff 1999).

A large percentage of commercially important fish species occurring along the U.S. Atlantic and Gulf coasts utilize estuaries during one or more life history stages (McHugh 1967; Day et al. 1989). Estuaries are nursing grounds for many juvenile species, in particular the families Sciaenidae, Clupeidae, and Bothidae (Weinstein 1979; Ross and Epperly 1985). In North Carolina the six most valuable commercial species (blue crabs, shrimps, southern flounder, Atlantic menhaden, summer flounder, Atlantic croaker) are all considered estuarine dependent because they utilize estuarine nurseries as postlarvae and juveniles. These estuarine areas are now referred to as Essential Fish Habitat (EFH), which designates all waters and substrate necessary to fish for spawning, breeding, feeding, and/or growth to maturity (Magnuson-Stevens Act).

Joseph (1973) identified three features of estuaries that make these habitats good nurseries for juvenile fishes: (1) physicochemical conditions, which are physiologically suitable for growth (e.g. warmer temperatures and intermediate salinities); (2) abundant prey resources; and (3) low predation risk. Presumably these mechanisms enhance
feeding and growth rates of juvenile fishes during the estuarine stage of their life cycle (Lankford and Targett 1994). Although fisheries biologists have long valued estuarine habitats and their role as nurseries (Ross 2003), the nursery value of alternative coastal habitats such as river discharge plumes is less clear.

River Discharge Plumes

River discharge plumes are coastal areas where freshwater empties onto the continental shelf and mixes with ocean waters (Grimes 2001). River discharge plumes are characterized by having frontal zones where plume and ocean waters mix and where isohalines are closely spaced and approach the surface (Grimes and Kingsford 1996). Also nested within the frontal zone are turbidity fronts, which are seaward projections of concentrated suspended particulate matter often represented by distinct color discontinuities (Garvine and Monk 1974). River discharge plumes vary in their spatial dimensions depending upon river size, flow discharge volume, tidal amplitude, wind, topography of the entrance, and nature of the currents over the continental shelf (Le Fevre 1986; Dustan and Pinckney 1989; Unclees and Stephens 1990; Gelfenbaum and Stumpf 1993; O’Donnel 1993; Sharples and Simpson 1993).

It is believed that river discharge plumes generate conditions that could enhance growth and survival of ichthyoplankton, which would have positive effects on recruitment (Grimes and Finucane 1991). Globally, most fisheries production is associated with coastal zones in the vicinity of major riverine discharge (Grimes 2001). For example, landings from coastal areas surrounding the Mississippi River delta account for 70-80% of the total annual commercial catch from the Gulf of Mexico (NMFS 1998),
which is second only to Alaska in terms of regional contributions to the total U.S. landings. This incurred production is attributed to three nutrient enrichment processes: coastal upwelling, tidal mixing, and land-based runoff from major river outflow (Caddy and Bakun 1994). River discharge influences surrounding coastal waters by creating a nutrient rich environment where physical dynamics (e.g., hydrodynamic convergence, water column stratification, transport, and retention of fish larvae) and biological processes (e.g., primary and secondary production and feeding, growth, and predation) may enhance larval survival and recruitment (Grimes 2001). Nutrient rich, turbid river water (where photosynthesis is light limited) is discharged and mixes with clear, nutrient poor ocean water (where photosynthesis is nutrient limited) and it is this process that stimulates biological production of phytoplankton (Grimes and Finucane 1991).

Hydrodynamic convergence is a key physical process that generates large stocks of phytoplankton, zooplankton, and fish larvae (Grimes and Kingsford 1996; Grimes and Finucane 1991; Govoni and Grimes 1992). Two groups of species are typically found at these locations: 1) estuarine-dependent species such as Atlantic menhaden (*Brevoortia tyrannus*), southern flounder (*Paralichthys lethostigma*), red drum (*Sciaenops ocellatus*), spot (*Leiostomus xanthurus*), weakfish (*Cynoscion regalis*) and Atlantic croaker (*Micropogonias undulatus*) and 2) coastal species such as king mackerel (*Scomberomorus cavalla*), Spanish mackerel (*Scomberomorus maculatus*), bluefish (*Pomatomus saltatrix*), and jacks such as blue runners (*Caranx crysos*) and crevalle jack (*Caranx hippos*). Because the life cycles of these species are entirely or mostly carried out in waters directly influenced by river discharge, alterations or variability in discharge
rates may affect critical demographic processes (e.g. growth, mortality, and recruitment) that regulate population size (Grimes 2001, Govoni 1997).

Interannual variability in freshwater discharge can be pronounced, but the implications for recruitment have received limited attention. Govoni (1997) reported an inverse association between river discharge and the annual abundance of juvenile gulf menhaden in the Mississippi River plume. He observed that if river discharge increased then recruitment of halfyear old gulf menhaden decreased. In contrast, a decrease in river discharge generally resulted in an increase in recruitment for halfyear old gulf menhaden. A decrease in larval survival in higher flow years may be attributed to an increase in the size of the river plume and its frontal zone. If the river plume increases in size it could translate to prolonged vulnerability to predation (Govoni 1997).

River plumes have a major influence on coastal waters and could potentially affect the numbers of many fish species that survive to recruitment (Grimes and Kingsford 1996; Sabates and Maso 1990; Grimes and Finucane 1991). For example, construction of the Aswan Dam in the 1960’s apparently caused the collapse of fisheries in the Nile Delta region because flow and the nutrients that fueled primary production were reduced (Nixon 2004). Since the 1980’s nutrient inputs have increased and fisheries landings now exceed pre-dam levels (Smetacek 1986; Nixon 2004). This case study is consistent with the hypothesis that river discharge enhances fishery production.

Since recruitment contributes the most to fish stock biomass, Grimes (2001) noted that by enhancing recruitment fishery production is influenced most. There are three critical factors that contribute directly to recruitment success: (1) feeding success; (2) predation; and (3) transport (Grimes and Finucane 1991). The larval stage is where
mortality rates are the highest and plume habitats could promote faster growth through this stage, which increases survival and could influence recruitment (Werner and Gilliam 1984). The prevailing view regarding river discharge plume function is referred to as the trophic advantage hypothesis (Grimes and Finucane 1991). This view contends that larvae within the plume (1) take advantage of abundant food resources and consume a superior diet, (2) grow faster, and (3) experience a shorter larval stage duration (Grimes and Finucane 1991). In addition to providing enhanced feeding conditions, the plume environment may also provide abiotic conditions that are more suitable for larval growth than adjacent coastal waters. For example, the lower salinities that would characterize a discharge plume could lower the osmoregulatory costs for larval and juvenile fishes, freeing more energy for growth (Lankford and Targett 1994; Moustakas et al. 2004).

Tandler et al. (1995) conducted a study with gilthead sea bream (Sparus aurata) and observed that larvae had higher survival and growth rates at salinities lower than full strength seawater. As a result of increased growth, fishes occupying plume habitats may experience a higher probability of recruiting to the adult population (Grimes 2001).

To manage fisheries effectively, it is important to identify and protect those habitats which contribute positively to recruitment. While there is some evidence for large plumes being beneficial, few studies have tested for these functions in smaller coastal plumes. If plumes aggregate larvae and their prey, and confer a trophic advantage, then these areas may warrant consideration and management as Essential Fish Habitat (Grimes 2001).
Cape Fear River Discharge Plume

The Cape Fear River (CFR) estuary is located in southeastern North Carolina and is characterized as a partially mixed drowned river valley type estuary (Dyer 1973). It is the only river in North Carolina that empties directly into Atlantic Ocean. The CFR discharge flows southward upon entering the ocean for most of the year, but during the summer months the river plume can be affected by strong southerly winds. Unlike the larger plumes associated with major river systems, the CFR plume is relatively small. Average tidal flow during ebb tide at the mouth of the Cape Fear River has been estimated at 5,664 m³ s⁻¹ (J.H. Carpenter, U.S. Nuclear Regulatory Commission, unpublished data). In contrast the Mississippi river plume discharges 735 km³ (Gunter 1979). It is an ideal plume system for studying fishery production because multiple sites within and outside of the plume can be sampled during a single ebbing tide.

Life Histories of Fishery Species

The life histories of most commercially important species along the southeastern U.S. coast are quite similar and involve interaction with river discharge plumes. Most of these species are considered estuarine-dependent. The family Sciaenidae, which includes red drum (*Sciaenops ocellatus*), spot (*Leiostomus xanthurus*), and Atlantic croaker (*Micropogonias undulatus*), and Paralichthyidae, including southern and summer flounder (*Paralichthys lethostigmata* and *Paralichthys dentatus*), generally spawn offshore. Their larvae are transported across the continental shelf, and must ingress through the plume environment before gaining entry to their nursery ground (Boehlert and Mundy 1988; Hales and Van Den Avyle 1989). Upon reaching adulthood they will migrate out
of the estuary towards their offshore breeding grounds where the cycle will repeat itself. Penaeid shrimp have a similar life history. Little research has been directed at studying the effects of discharge plumes on these species as they ingress through this environment.

Physiological Condition and Growth

Fluctuation of environmental conditions can have an impact on an individual’s physiological condition and growth (Lankford and Targett 1994). Many estuarine dependent species encounter three different habitats (ocean, riverine plume and estuary) during their journey from offshore spawning sites towards their juvenile nursery areas. These habitats may differ in their nursery value due to differences in salinity, food availability, refuge, predation, and other biotic and abiotic factors that influence growth and survival.

One approach to estimating the relative suitability of different habitats is to compare the physiological condition and/or growth of individuals that reside there. There are many methods for estimating the condition and growth of fishes.

Morphometrics, which is a series of morphological measurements, can be applied to larvae thru adult stage fish. One example of this method would be the study conducted by Powell and Chester (1985). They used six body measurements (standard length, head length, eye diameter, body depth at anus, body depth at cleithral symphysis and body depth at pectoral fin base), to describe changes in the body shape of larval spot during starvation and used these changes to classify spot larvae according to their nutritional condition. This method is inexpensive and measurements can be taken easily without special preparation (Theilacker 1978). Drawbacks of this method would be that lab-
raised and field-caught fish can differ in condition due to the effects of a controlled environment in a lab. Additionally, shrinkage caused by preservation of tissues can bias measurements.

Comparison of biochemical indicators (e.g. metabolic enzymes, RNA: DNA ratios, lipids) between individuals from different habitats can also provide information on physiological condition (Clemmesen 1994). There is a variety of enzymes that can be utilized for such studies from glycolytic enzymes to those found in specific tissues. Some enzymatic assays can provide insight as to whether an individual is starving or feeding well, based on enzyme activity values. It is assumed that higher activity values generally translate into better physiological condition (Pelletier et al. 1994; Pelletier et al. 1995), however enzyme activity can be high due to stress. Guderley et al. (1996) examined physiological status of Atlantic cod (Gadus morhua), comparing wild versus lab-raised fish to determine if fish were starving or feeding well. They reported that white muscle of starved cod had lower lactate dehydrogenase activity than that of fed cod. Guderley et al. (1996) also reported similar findings for intestinal cytochrome c oxidase, noting that activity was lower in starved versus fed fish.

Bulow (1987) suggested that the ratio between RNA and DNA content is an index of cellular metabolic intensity. DNA content is fixed within a cell, but the amount of RNA tends to vary depending on the amount of protein synthesis. This ratio has been used to estimate recent growth rates in fishes and has proven to be a useful indicator of physiological condition (Clemmesen and Doan 1996). Although often expensive and time consuming these indicators can be applied to large numbers of individuals and can provide rapid feedback.
Otoliths, which are the inner ear bones of teleost fishes, are useful structures for fish ecologists because these structures can provide information on an individual’s age and growth rate (Panella 1971). Otolith microstructural features include both daily and annual rings, which can be used to determine an individual’s age. Increment widths between these rings have been shown to correlate with somatic growth rate (Clemmesen and Doan 1996). Therefore, fish that are in good physiological condition (growing well) should display wider daily increment widths than fish that are starving or growing slowly (Clemmesen and Doan 1996). Otolith increment widths can be used to estimate daily or annual growth rates. Comparing daily increment widths of individuals from different habitats can reveal information on relative habitat value. This method is very time consuming and requires expensive equipment.

Objectives

The objectives of this study were to 1) characterize abiotic conditions (temperature, salinity, and dissolved oxygen) in the CFR plume versus adjacent coastal waters, 2) document the diversity and abundance of ichthyoplankton occurring within versus outside of the CFR plume, 3) to describe spatial and temporal variability in the larval assemblages within and adjacent to the CFR plume, and 4) determine if the CFR plume has positive effects on physiological condition by using biochemical indicators and otolith microstructural analysis of selected finfishes and decapods sampled from the CFR plume versus estuarine habitats.
METHODS

Physicochemical Parameters

Physicochemical parameters at sampling sites were measured following each plankton tow using an YSI model 85. Salinity, temperature, and dissolved oxygen were measured at the surface and bottom for each station sampled.

Larval Distribution and Abundance

The distribution and abundance of larval fishes and decapods in the vicinity of the Cape Fear River plume was examined through plankton sampling. Sampling was conducted once a month from January 2002 to December 2003, during the day corresponding with either the full or new moons and on an ebbing tide. Larvae were sampled at six stations corresponding to those used by the University of North Carolina at Wilmington Coastal Ocean Research Monitoring Program (CORMP) (Figure1). Two stations were located within the plume (stations 6 and 2), one station on the front (station F) when present, two stations outside of the plume (stations 4 and 7), and one in the estuary (station 1). These stations were fixed, but due to the spatial variability of the plume, alternative stations were sampled at times to insure that both ocean and plume habitats were represented in each month. When ocean stations (stations 4 and 7) appeared to be impacted by the plume, additional ocean stations were sampled (stations 8 and 9). The plume was determined using visual cues as well as physicochemical parameters, (salinity of the water). Three 60 cm plankton nets with 750µm mesh
(surface, 1 meter, and bottom) were fished simultaneously for ten minutes. Two tows of the 3 nets were collected for each station. General Oceanics flowmeters (model 2030R) were attached to the surface and bottom nets to calculate the volume of water sampled. The contents of the cod end were sieved through 950μm mesh to reduce volume, then immediately preserved in 95% ethanol. After 24 hours the contents were transferred to fresh 95% ethanol (Grimes and Finucane 1991). The samples were then separated into fish, crab, and shrimp larvae, identified to the lowest taxonomic level possible, and enumerated.

Habitat Suitability: Plume vs. Estuary

The suitability of plume versus estuarine habitats for juvenile fishes and shrimp was evaluated through synoptic trawl and seine sampling in 2003. Sampling was conducted between 1100-1400 hours on June 17, 2003. This one-time collection was designed to sample selected target species (Atlantic croaker *Micropogonias undulatus*, spot *Leiostomus xanthurus* and brown shrimp *Farfantepenaeus aztecus*) from two different habitats for biochemical assays and otolith microstructural analysis. This design controlled for possible variability in physiological condition due to time of day differences. These species were selected because they are commercially important, abundant, and use both estuarine and plume habitats during their life cycle. There is also interest in these species from a management perspective.

Plume samples were collected from CFR plume station 2 using an otter trawl. Dimensions of the trawl were 16’ head rope, 3/4 inch mesh wing with a 5mm mesh
inserted into the cod end. The net was towed for ten minutes and target species were fixed immediately in liquid nitrogen for laboratory analysis.

The estuarine samples were collected using a fifty-foot beach seine (3/8” wing mesh and 1/8” mesh bag) from the Fort Fisher boat basin, in the mesohaline portion of the CFR estuary. Target species were sorted and immediately flash frozen in liquid nitrogen.

Biochemical Assays

Biochemical assays were conducted to assess the physiological condition of selected resources species collected from plume versus estuarine habitats. Two species of fishes (spot, *L. xanthurus* and Atlantic croaker, *M. undulatus*), and one crustacean (brown shrimp, *F. aztecus*) were assayed. Two different enzymes were assayed for each habitat and species: cytochrome C oxidase (COX), which is part of the electron transport chain and hexokinase (HK), which is the first step of glycolysis.

Tissue and Enzyme Preparation

Sixteen individuals each of *M. undulatus* and *F. aztecus*, eight from the plume and eight from the estuary were used (Table 1). For *L. xanthurus*, sample sizes were n=3 and n=8 for plume and estuary sites, respectively. Epaxial muscle of the fish and abdominal muscle of the shrimp were removed for analysis. Tissue extracts were prepared by homogenizing and sonicating approximately 0.5-1 g of tissue in 19 volumes (fishes) and approximately 1 g of tissue in 6 volumes for the (shrimp) of 50mM imidazole-HCl, 50mM KCl, and 50mM dithiothreitol (DTT), and adjusted to a pH of 7.4. Homogenates were
then centrifuged at 13000xg for 20 minutes. The supernatant was weighed and stored at -80°C until assays were performed.

COX Assay

The COX assay is measured spectrophotometrically and based on the oxidation of cytochrome c (Foster et al. 1993). A mixture of 950μl of assay buffer (50mM Tris buffer, 600mM KCl), 20μl of enzyme dilution buffer, and 80μl of tissue homogenate were added to a 1ml cuvette. The mixture was then placed in a Pharmacia Ultrospec 4000 spectrophotometer equipped with a thermostatted cell holder and a refrigerated recirculating water bath was used to maintain the temperature at 20°C. My assay conditions followed Pelletier et al. (1994). The reaction was initiated by the addition of a cytochrome solution and the rate of COX activity was measured by the decline in absorbance at 550nm.

Hexokinase Assay

Hexokinase assays were performed in a buffer comprised of 0.0067g of glucose, 0.0152g magnesium chloride (MgCl₂), 0.0095ml of monothioglycerol, and 0.107g HEPES dissolved in 10ml of water. 450μl of the assay buffer, .4 mM of NADP, .4 mM of G6PH, and 300μl of sample homogenate were placed in a 1ml cuvette and referenced in the spectrophotometer. The reaction was initiated by placing 15μl of ATP into the cuvette and activity was determined from the increase in absorbance at 340nm.
Otolith Microstructural Analysis

Sagittal otoliths were removed from fishes and used to back calculate recent growth rates (days prior to catch) of individuals collected from estuarine versus plume environments. The otoliths were embedded in spur medium (3.3g of vinyl cyclohexene dioxide, 2.0g-diglycidyl ether, 8.7g-noneryl succinic anhydride, and 0.1g dimethylaminoethanol) and allowed to harden at 70°C for twelve hours (Secor et al. 1991). A transverse section of each otolith was then cut using a Buehler low speed isomet saw. Sections were mounted on glass slides, ground using 600 grit aluminum oxide wet sand paper and polished using 0.3μm alumina polishing powder. Otoliths were then read using an Olympus BH2 epifluorescent light microscope. Otolith sections were photographed at two different magnifications using a diagnostic spot RT KE camera. The first picture was taken at a magnification of 4x to capture the whole otolith. The second picture was taken at a magnification of 10x to get a detailed view of the area that was going to be measured (most recent increments from otolith margin). This detailed photograph of the area to be measured was roughly between the nucleus and the margin of the otolith. Daily increments were measured in microns using the software program Image Pro Plus version 4.1.

Data Analyses

Data on ichthyoplankton composition and abundance, enzyme activities and otolith microstructure were used to test two hypotheses regarding the function of the CFR plume habitat.

$H_1$: The CFR plume serves to aggregate larvae and juveniles.
**H₂**: The CFR plume environment functions as EFH by enhancing physiological condition of fishes and crustaceans based on daily increment growth and enzymatic activity.

Physicochemical parameters were analyzed using a three-way ANOVA to test for effects of month, habitat, and depth. A one-way ANOVA was used to compare ichthyoplankton concentrations at each station for each month in the years 2002 and 2003. A two-way ANOVA was used to analyze month and habitat effects for each month in 2002 and 2003. Shannon-Weiner diversity indices were calculated for each sample as:

\[ H = - \sum (P_i \ln P_i) \]

Diversity values were analyzed using Primer-E (Clarke and Warwick 2001) and a two-way ANOVA was conducted to test the effect of depth and habitat for all months sampled in 2002 and 2003. A two-way ANOVA was conducted for each habitat to test for significance of depth and habitat effects. Enzyme activity values were first examined using a linear regression to analyze whether body size affected enzyme activity. Mean length of *M. undulatus*, *L. xanthurus* and *F. aztecus* for each habitat was analyzed using a Student’s t-test. A linear regression was also performed to analyze whether body size affected otolith increment width. A Student’s t-test was conducted to analyze mean length of *M. undulatus* and *L. xanthurus* for each habitat. Correlations among biochemical and otolith indicators were tested using the Pearson correlation coefficient. All statistical analyses were performed using Statistica version 6.0 (Statsoft 2001).
RESULTS

Physicochemical Parameter

2002 was characterized as a dry year in contrast 2003 was characterized as a wet year due to the effects of an El Niño event. Discharge during 2002 ranged from 775.58 ft$^3$/sec in August to 10123.23 ft$^3$/sec in December (Appendix 1). In contrast discharged during 2003 ranged from 3084.67 ft$^3$/sec in November to 21652.67 ft$^3$/sec in April (USGS 2004).

2002

Temperature varied significantly across months with warmer values occurring during the summer months (Table 2). Plume and ocean habitats differed significantly with warmer temperatures occurring in the plume. There was a significant difference between surface and bottom temperatures with warmer values occurring at the surface (Table 2). There was no significant interaction between month*habitat, habitat*depth, and month*habitat*depth, but there was a significant interaction between month*depth with higher values occurring during the summer months.

Salinity during 2002 varied significantly across months with more saline values occurring during summer months (Table 2). There was a significant difference between the plume and ocean habitats with higher values occurring in the ocean. Surface and bottom depths differed significantly with the bottom being more saline than the surface.
There was no significant interaction between month*habitat, habitat*depth, and month*habitat*depth, but there was a significant interaction between month*depth with higher values occurring at the bottom during the summer months (Table 2). Salinities recorded at plume stations ranged from 31.8 ppt in March to 34.8 ppt in June (Figure 2). Salinities recorded at ocean stations were slightly higher ranging from 32.1 ppt in November to 35.2 ppt in June.

Dissolved oxygen during 2002 varied significantly across months with higher values occurring during the winter months in both habitats (Table 2). There was no significant difference between plume and ocean habitats. There was a significant difference between the surface and bottom with higher values occurring at the surface (Table 2). There was no significant interaction between month*habitat, habitat*depth, month*depth, and month*habitat*depth. Dissolved oxygen in the plume habitat followed the same trend as the ocean habitat (Figure 2). A minimum of 5.5 mg/l occurred in November and a maximum of 9.4 mg/l occurred in January. Dissolved oxygen content of both habitats appeared to drop from the beginning of the year to the end of the year.

2003

There is a dramatic contrast for 2003 compared with 2002. The effects of the increased precipitation were dramatic reducing the salinity of the ocean to below 35 ppt for the entire year (Figure 2). Differences between ocean and plume habitats were distinct for 2003.

Temperature varied significantly across months with higher values occurring during the summer months (Table 2). Plume and ocean habitats did not differ during
2003. Temperature was significantly higher at the surface than at the bottom. No interaction occurred between month*habitat, month*depth, habitat*depth, and month*habitat*depth (Table2). Water temperatures at plume and ocean habitats increased proportionately from February (~8.5°C) to early July (~28°C).

Salinity varied significantly across months with higher values occurring during the winter (Table2). Salinity differed significantly between the ocean and plume habitats with higher values occurring in the ocean. There was a significant interaction between month*habitat with higher values occurring in the plume during winter months (Table2). There was also a significant interaction between month*depth with higher salinity values occurring at the bottom for all months. There was no significant interaction between habitat*depth and month*habitat*depth (Table2). Plume salinities ranged from 23.7 ppt in April to 32.3 ppt in February (Figure 2). Precipitation was heavy in late winter and early spring of 2003. From April to July salinity increased to about ~30 ppt then began to fall late in July into August. Salinities again increased in late August and leveled off in September at ~32 ppt. Ocean salinities remained higher than plume salinities for all months except October. Ocean salinities were lower in 2003 ranging from 29.1ppt in April to 32 ppt in February.

Dissolved oxygen varied significantly across months with higher values occurring during the winter months (Table2). There was no significant effect of habitat or depth on dissolved oxygen. There was a significant interaction between month*habitat with higher values occurring in the plume during the winter months (Table2). There was no significant interaction between month*depth, habitat*depth, and month*habitat*depth. Dissolved oxygen content in the plume and ocean reached a minimum of 3.3 mg/l in
August and a maximum 8.9 mg/l for the plume occurred in February and 9.4 mg/l for the ocean.

Larval Distribution and Abundance

Thirty-three taxa of fishes from twenty-two families were collected in 2002 (Table 3) and eighteen taxa from eleven families were collected in 2003 (Table 4). A total of 1,497 larvae and early juveniles were collected over the two years with 826 being collected in 2002 and 671 collected in 2003. The majority of the fishes collected were estuarine-dependent species that are spawned on the continental shelf and immigrate into the estuary and spend their early life there (e.g. *Micropogonias undulatus*, *Leiostomus xanthurus*, *Lagodon rhomboides*). A minority of larvae and early juveniles collected were estuarine resident species that spend their adult life in the estuary and spawn in the spring and summer months (e.g. *Hypsoblennius hentzi*, *Monocanthis hispidus*, *Symphurus plaguisa*, *Gobiosoma bosc*). The most abundant family during the sampling period was Sciaenidae, which was collected in all three habitats and at all depths. Sciaenids comprised approximately 27% in 2002 and 33% in 2003 of all fishes collected (Tables 3 and 4). The families Blenniidae and Gobiidae were also relatively abundant, with blennies more prevalent in 2002 and gobies in 2003 (Tables 3 and 4).

Taxonomic Composition vs. Habitat

2002

In January 2002, Atlantic croaker (*Micropogonias undulatus*) was the dominant species at plume stations (75% of the total catch) (Figure 3A). At ocean stations pinfish
(Lagodon rhomboides), flounder (Paralichthyidae), and M. undulatus were equally abundant comprising approximately 30% of the catch. The plume frontal boundary was sampled during this month. Micropogonias undulatus and L. rhomboides each represented 43% of the catch whereas spot (Leiostomus xanthurus) comprised 40% of the catch. February was not sampled due to weather conditions.

In March 2002 a shift in the dominant taxa occurred (Figure 3A). Leiostomus xanthurus represented 75% of the catch at both plume and ocean stations. Leiostomus xanthurus and paralichthyds comprised 50% of the catch in the estuary and no frontal region was present for this month. April is not represented due to inclement weather conditions.

May 2002 appeared to be a transitional period in which species assemblages changed from winter-spawned to summer-spawned taxa (Figure 3B). Number of taxa increased dramatically for each habitat during May compared with previous months. The plume habitat was dominated by the family Engraulidae (37%) where as ocean stations were dominated by feather blennies (Hypsoblennius hentzi) (86%). Blennioids were also dominant at estuarine (29%) and frontal (75%) stations.

In June 2002 the family Gobiidae dominated in the plume (53%) while Blennioids dominated at ocean (46%) stations (Figure 3B). The dominant catch in the estuary was southern kingfish (Menticirrhus americanus) (60%) while the front was dominated by Gobioids (65%).

Gobioids once again dominated the plume habitat (48%) in August 2002 (Figure 3C). Ocean stations were dominated by unidentifiable pre-flexion larvae (20%) and the
family Sciaenidae (22%). The estuary was not sampled due to a strong current and no front was present.

In September 2002 Gobioids remained dominant at plume stations (33%), but ocean stations were dominated by blue runners (*Caranx crysos*) (33%) (Figure 3C). The estuary was not sampled due to a strong current and no front was present.

October 2002 was also a transitional month marked by a decline in numbers of taxa (Figure 3D). October marked a shift from summer to winter-spawners. Number of taxa decreased in the plume and estuary but the ocean remained relatively the same. Gobioids dominated the plume catch (90%) and were still present in the ocean (12%). Unidentifiable larvae represented (32%) of the catch in the ocean followed by the family Triglidae (20%). Gobioids also dominated the estuary representing 50% of the total catch.

In November 2002 gobies were still present in the plume, but only comprised 4% of the catch (Figure 3D). *Micropogonias undulatus* were the dominant species making up 92% of the catch. *Micropogonias undulatus* also comprised 100% of the catch in the ocean and the estuary. December 2002 was not represented due to inclement weather conditions.

2003

January 2003 is not represented due to inclement weather conditions. The plume habitat in February 2003 was dominated by *L. xanthurus* (53%), while the ocean was dominated by *L. rhomboides* (60%) (Figure 3E). *Lagodon rhomboides* also dominated
the estuary catch (87%) and the front was dominated by 40% \textit{L. rhomboides} and 40% \textit{L. xanthurus}. March was not sampled due to inclement weather conditions.

In April 2003 the plume was characterized as 36% unidentifiable larvae, 27% Paralichthyds, and 27% Gobioids (Figure 3E). No front was present this month. May and June 2003 were not sampled.

In July 2003 Gobioids dominated the plume catch (33%) whereas \textit{C. cryos} dominated the ocean catch (44%) (Figure 3F). No front was present and the estuary was not sampled.

Bay anchovies (\textit{Anchoa mitchilli}) dominated the plume (19%), ocean (74%), and estuarine (36%) stations during the month of August 2003 (Figure 3F). No front was detectable for the month.

In September 2003 the plume habitat was dominated by naked gobies (\textit{Gobiosoma bosc}) (67%) and the ocean was dominated by the family Syngnathidae (40%) (Figure 3G). The estuary was not sampled and no plume front was sampled.

October 2003 was a transitional month representing a shift in taxa from summer to fall/winter spawners. Number of taxa decreased in the ocean while remaining relatively unchanged in the plume. Almost 90% of the plume catch was \textit{M. undulatus} (Figure 3G). The family Gobiidae dominated the ocean comprising 60% of the total catch (Figure 3G). The estuary was characterized as 50% unidentifiable larvae and 50% were of the family Syngnathidae (Figure 3G). November 2003 was not sampled due to inclement weather conditions.

In December 2003 species diversity decreased at all habitats. \textit{Micropogonias undulatus} dominated the plume catch (84%) (Figure 3H). \textit{Lagodon rhomboides}
represented 100% of the ocean catch, while the estuary was split 50% *L. rhomboides* and 50% *M. undulatus*.

Total Larval Concentration: Habitat Effects and Seasonality

During 2002 total larval concentration was not significantly influenced by habitat but was significantly affected by month (habitat $F=3.03$, $p=0.05$: month $F=2.47$, $p=0.018$). Plume stations had higher mean larval concentrations than ocean stations for all months except May (Figure 6) and mean total larval concentrations were higher for station two (plume) four out of the seven months sampled in 2002 (Figure 4). Pairwise comparisons did not reveal significant differences between ocean and plume sites for any given month. There was a highly significant interaction between month and habitat (month*habitat $F=2.44$, $p=0.003$). Ocean and estuarine habitats displayed little seasonal variation with larval concentrations staying relatively similar across all months sampled. Seasonal variation was more pronounced within the plume habitat (Figure 6). Peaks in plume larval concentration occurred in January and August (2002) as well as February and August for 2003. During 2003 there was no significant effect of either habitat or month on total larval concentration (TLC) (month $F=0.000$, $p=1.00$; habitat $F=1.36$, $p=0.244$) (Figure 6). Station 2 displayed higher larval concentrations four out of the seven months sampled in 2003 (Figure 5).

Taxonomic Diversity

The diversity of larval fishes caught at the surface was not affected by month or habitat (month $F=1.21$, $p=0.311$; habitat $F=0.637$, $p=0.430$). There was no significant
interaction between month and habitat for the surface (month*habitat F=1.44, p=0.185). There was a trend for ocean diversity to be higher than plume diversity in the late summer and early fall (Figure 7). Also, ocean diversity values appeared to show more seasonal variation whereas plume diversity values remained relatively consistent for all months sampled in 2002 and 2003.

The diversity of larval fishes caught at the depth of 1 meter was significantly affected by month, but not by habitat (month F=2.48, p=0.014, habitat F=1.24, p=0.272). There was no interaction between month and habitat (month*habitat F=1.12, p=0.372). Seasonal variation in both habitats occurred during the same months each year (Figure 7). August and October 2002 for the ocean had the highest diversities, while the highest diversity for the plume occurred in May 2002. The ocean diversities for 2003 were somewhat lower for the same months compared to 2002.

Although the month effect was not statistically significant at the bottom depth, habitat had a significant effect at this depth (month F=1.88, p=0.063; habitat F=12.3, p<0.001). There was no significant interaction between month and habitat (month*habitat F=0.842, p=0.623). Bottom diversities were higher for the plume than ocean stations during 2003, but this habitat effect did not occur in 2002. There was high seasonal variation for the plume habitat, while ocean diversity was very low across the years 2002 and 2003 (Figure 7).

Depth Effects

There was a significant effect of depth on diversity, but not for habitat (depth F=8.42, p<0.001; habitat F=2.03, p=0.155) with the highest diversity occurring at 1 meter
for the ocean and at the bottom for the plume (Figure 8). A Tukey test revealed that
diversity at depth was higher at the bottom than at the surface (p<0.001). There was a
significant interaction between habitat and depth (habitat*depth F=7.71, p<0.001). A
Tukey test revealed that plume and ocean diversities were similar at surface and 1 meter
depths. However, bottom diversities were higher for the plume than for ocean stations
(p=0.002).

Biochemical Assays

A linear regression analysis revealed no significant effect of body size on COX
activity for *M. undulatus* or *L. xanthurus* found either in the plume or estuarine habitat
(Atlantic croaker plume F=0.161, p=0.702, estuary F=0.001, p=0.975, spot p=0.756,
estuary p=0.566) (Figure 9). Habitat did not affect COX enzyme activity for
*M. undulatus* (p=0.904). A Student’s t-test was performed for *L. xanthurus* to test for
habitat effect on enzyme activity and revealed no significance effect (p=0.434).

A linear regression analysis was conducted and revealed no significant effect of
body size on hexokinase activity for *M. undulatus* or *L. xanthurus* found either in the
plume or estuarine habitat (Atlantic croaker plume F=2.95, p=0.137, estuary F=0.121,
p=0.740; spot plume F=0.579, p=0.585, F=0.219 estuary p=0.656) (Figure 10).
Consequently mean values were calculated for each habitat and compared using a
Student’s t-test. The test revealed no significant effect of habitat on hexokinase activity
(p=0.074). Activity values were higher for the plume than the estuary (Figure 11)
although not significant. Comparison of mean enzyme activity level revealed no
significant effect of habitat on enzyme activity for *L. xanthurus* (p=0.601).
Farfantepenaeus aztecus were also used to analyze enzyme activity as an approach for estimating habitat suitability. Only hexokinase was assayed for this species. A linear regression analysis was conducted to determine if body size had an effect on HK activity and no significance was revealed (BS plume F=0.683, p=0.440, estuary F=0.181, p=0.685) (Figure 14) (Figure 12). A Student’s t-test was then used to determine if mean HK values differed between habitats. No significant effect was revealed (p=0.320).

Otolith Microstructural Analysis

A linear regression analysis was conducted to test whether body size had an effect on otolith increment width. No significant effect was revealed for L. xanthurus otoliths (plume F=0.447, p=0.625, estuary F=3.97, p=.103) (Figure 15). There was also no effect of body size on otolith increment width for M. undulatus (plume F=0.209, p=0.664, estuary F=1.88, p=0.219).

A student’s t-test was conducted to test whether habitat had an effect on otolith increment width. No significant effect differences were revealed for L. xanthurus (p=0.643). Mean length was slightly larger for the plume than the estuary (Figure 16). No significance was revealed for M. undulatus as well (p=0.456).

Correlations Among Biochemical Indices

Micropogonias undulatus otolith increment widths were not highly correlated with COX activity values, suggesting that COX activity may not respond strongly to variation in growth rate (Figure 12). There was a slight correlation between otolith
increment widths and HK values suggesting that HK activity may respond more linearly to variation in growth rates.

*Leiostomus xanthurus* otolith increment widths were not correlated with COX or HK activity values, suggesting that enzyme activity may not respond strongly to variation in growth rate (Figure 13). There was a slight correlation between COX and HK activity suggesting that enzyme activity may respond to each other in response to growth rate.

**DISCUSSION**

My research in the Cape Fear River Plume noted distinct variations in physicochemical parameters during 2002-2003. The CFR plume habitat appeared to be significantly affected by the presence of an El Niño weather event. Larval fish assemblages were distinctly different between ocean and plume habitats. Distinct seasonal assemblages were apparent for 2002 and 2003. There was not enough evidence from the biochemical indices to support the trophic advantage hypothesis. Further research is needed in this area to clarify whether there is a trophic enhancement provided to larvae within the CFR plume and to compare the plume to adjacent coastal waters. Ichthyoplankton concentrations appeared to be higher in the CFR plume than adjacent coastal waters, possibly suggesting larval aggregation.

There have been many studies conducted in the Mississippi River Plume as well as other plumes that have characterized the larval assemblages, water dynamics, and production processes associated with river discharge plumes (Govoni et al. 1989, Grimes and Finucane 1991, Ditty 1986, Reiss and McConaugha 1999, Thorrold and Mckinnon 1995). There is less known about the components of smaller river plumes on the fisheries
production process. This study was an attempt to examine similar questions in a relatively small plume, Cape Fear River, to determine whether processes reported for larger plume operate at smaller scales. The Cape Fear River plume is representative of many systems in the southeastern United States, including the Savannah River, St. John’s River, and the Wynah Bay.

Physicochemical Parameters

Over the course of this study, abiotic conditions such as salinity, temperature, and dissolved oxygen varied dramatically between 2002 and 2003. Conditions during 2002 were characterized as dry with low freshwater discharge due to below average rainfall. Throughout 2002 temperature and salinity in the plume habitat was significantly different than the ocean habitat. Both habitats displayed similar levels of seasonal temperature fluctuation. Seasonal fluctuations of the plume habitat in 2002 were minor and differed only slightly from that of the ocean habitat. In contrast, there was greater seasonal fluctuation of temperature, salinity and dissolved oxygen in the plume habitat during 2003. Temperature was not significantly different between plume and ocean habitats, but salinity was. The plume and ocean salinities were much lower in 2003 during the late winter and early spring months. Plume salinities fluctuated from April through late September when salinities finally began to level off. Southeastern North Carolina experienced higher rainfall amounts due to the effect of an El Niño event. This weather event helped change the dynamics of the plume from the previous year. During the summer months when the water temperatures should be at their peaks there was a distinct separation between the plume and ocean habitats.
Abiotic factors such as salinity, temperature and dissolved oxygen can have a significant impact on larvae. Physicochemical data from this study indicated that the plume habitat could fluctuate greatly from year to year depending on weather patterns and large-scale weather events. Abiotic fluctuations in the plume habitat may be important due to their potential influence on larval fish growth and/or survival. Indications from previous studies suggest that temperature and salinity can significantly affect the growth and/or survival of larval fishes (Moustakas et al. 2004, Watanabe 2003, Hoss et al. 1988, Holt et al. 1981). Effects of temperature and salinity on larval fish stage growth rates have been demonstrated on species found in the CFR plume (Atlantic croaker M. undulatus, spot L. xanthurus, and southern flounder P. lethostigma).

Laboratory studies (Moustakas et al. 2004, Henne and Watanabe 2003, Hoss et al. 1988) indicated that fluctuations in temperature and/or salinity can have dramatic effects on the growth and survival of fishes during their early life history stages. Larval M. undulatus spawn from late fall to early spring while L. xanthurus spawn from late winter to early spring. Peak recruitment of L. xanthurus occurs after peak recruitment of M. undulatus and generally after the coldest temperatures of the year (Hoss et al. 1988). Hoss et al. (1988) reported that $10^\circ$C or less was a critical temperature for growth and survival of L. xanthurus and survival decreased significantly at temperatures below $10^\circ$C. In contrast M. undulatus were able to survive at these cold temperatures. During peak recruitment of L. xanthurus temperatures in the plume and ocean habitats did not fall below $14^\circ$C for either year. These temperatures are higher than those reported by Hoss et al. (1988) as energetically stressful to these larval fishes. Thus, the Cape Fear River plume appears to provide favorable temperatures for those fall/winter spawners.
Salinity can have an impact on larval growth and survival as well (Holt et al. 1981, Henne and Watanabe 2003, Moustakas et al. 2004). Spawning of red drum (*Sciaenops ocellatus*) occurs in later summer through early fall (Holt et al. 1981). Salinities in the Cape Fear River plume during this time ranged from a minimum of 23.7 ppt in 2003 to a maximum of 34.8 ppt in 2002. Studies have indicated that larval *S. ocellatus* are tolerant of ranges in salinity. Holt et al. (1981) reported that salinity did not effect 2-week survival of larval *S. ocellatus*. Studies on other species have indicated contrasting results. For example, Henne and Watanabe (2003) and Moustakas et al. (2004) reported that growth and survival were significantly higher for larval *P. lethostigma* at salinities of 34 ppt than 24 ppt. During February and April when larval *P. lethostigma* occur in this area, salinity in 2003 ranged from 24 to 34 ppt. Salinity variations of this magnitude could potentially influence the growth rates of larval fishes in the vicinity of the Cape Fear River plume. Future studies should further investigate the recruitment implications of the seasonal and interannual salinity variations observed in this study.

Larval Distribution and Concentrations

Plankton sampling revealed that ichthyoplankton concentrations were generally higher within the plume than at ocean stations. Larval abundances in the plume habitat were ~2.5 times greater than abundances found in the ocean habitat. Additionally, concentrations within the plume were higher at the bottom, while concentrations in the ocean were higher at 1 meter. Numerically dominant taxon found in the plume were Sciaenids (25%) while Blennioids were most prevalent in the ocean habitat (10%).
Findings from my study are comparable to studies of larger plumes such as the Mississippi River plume and the Chesapeake Bay plume (Govoni et al. 1989, Grimes and Finucane 1991, Dity 1986, Reiss and McConaugha 1999, Thorrold and Mckinnon 1995). Work conducted in the Mississippi River plume found that larval fishes were more abundant in the vicinity of the river discharge plume than adjacent gulf waters (Govoni et al. 1989, Grimes and Finucane 1991, Dity 1986). Reiss and McConaugha (1999) noted that larval abundances were greater in the vicinity of the Chesapeake Bay plume than shelf waters as did Thorrold and Mckinnon’s (1995) study in a river plume off the Central Great Barrier Reef. Findings from the Cape Fear River plume also recorded higher larval fish abundances within the plume, but abundances were higher at the bottom in contrast to the work from Mississippi River plume. Results for the Cape Fear River plume suggest that smaller systems appear to have similar effects on larval fish assemblages as larger plumes.

During six of the fourteen months sampled, station two, a plume station, supported higher larval concentrations than all other stations sampled. This is interesting because of where this station is located. Station two is located between station one (estuarine station) and four (ocean station) on the edge of the shipping channel. These higher concentrations could be because larvae aggregate here as a means to ingress into the estuary. Larvae can be transported into the estuary via the saltwedge (Epifanio and Garvine 2001) created by lighter freshwater that rushes out and to the South, while heavier, more saline water occurs along the bottom. Larvae can also ingress into the estuary by a process called selective tidal stream transport (STST) in which larvae are high in the water column during flood tides and lower in the water during ebb tides.
(Boehlert and Mundy 1998 and Forward et al. 1999). Forward et al. (1999) found that in general *L. xanthurus, L. rhomboides, Paralichthys lethostigma* and *dentatus* (southern and summer flounder) were more abundant in neuston and oblique samples during flood tide at night in Shackleford Channel, which would indicate the presence of STST. It would be difficult to determine from this study whether or not larvae collected exhibited STST since only one tidal cycle was sampled.

Interannual Variability

This study provided an excellent opportunity to sample two dramatically different years. Year 1 (2002) was characterized as dry whereas year 2 (2003) was characterized as wet. It appears that higher precipitation from the El Niño event during 2003 affected larval assemblages and abundances for the plume and ocean habitats. Garcia et al. (2003) reported similar findings during the El Niño of 1997-1998. Garcia et al. (2003) reported that relative abundances of all fish groups were about five times lower during the El Niño then before or after. Change in flow of the Cape Fear River might explain some of this variability. An increase in river discharge could disperse larvae, increase the size of the plume, and subsequently reduce larval recruitment. Govoni (1997) reported that when Mississippi River discharge was high, recruitment of halfyear old *B. tyrannus* was low and when river discharge was low recruitment was higher for the species. Findings from the Cape Fear River plume study are comparable to those of Govoni’s (1997). Govoni (1997) speculated that decreased flows reduces plume size and could positively effect recruitment by shortening the time larvae spend out in open waters to predation. Increased river discharge decreased the taxonomic diversity of larval fishes
found overall in 2003. Taxon numbers caught decreased by nearly half of what it was in 2002. River discharge appeared to effect larval concentrations and taxonomic diversity when compared to previous studies.

Taxonomic Composition

Larval fishes collected in plume and ocean habitats were representative of the juvenile and adult fishes that are common in southeastern North Carolina. Many of the fish larvae caught were post flexion, indicating that they could have been spawned far offshore and were transitioning from their oceanic to estuarine habitat. This metamorphosis from larval to juvenile stages upon entry into the estuary is consistent with the findings of Witting et al. (1999) in Great Bay, NJ.

A distinct seasonal assemblage occurred throughout the course of this research. The transitional months appeared to be similar for 2002 and 2003. In both years October represented a shift from summer to fall/winter spawners, while late spring (May) represented another transitional period from winter to spring/summer spawners. The fall through spring months were dominated by estuarine dependent taxa and showed seasonal patterns (e.g. *M. undulatus, L. xanthurus, L. rhomboides*).

There appeared to be a correlation between temperature and species assemblages that is represented by a seasonal pattern. This seasonal pattern in taxonomic composition is comparable with other studies where seasonal temperature variation is greater than interannual variation. Witting et al. (1999) and Warlen and Burke (1990) each conducted studies where distinct taxa were representative of seasonal progression: for the months January through April *M. undulatus, L. xanthurus, Paralichthyidae, L.*
*rhomboides*, and *B. tyrannus* were the dominant taxa. During the same period data collected during this study followed similar seasonal trends. Concentrations of larval Paralichthyids and *B. tyrannus* were higher in Witting et al. (1999) and Warlen and Burke’s (1990) studies than in this one. This difference could be because of proximity to the Gulf Stream or because they’re sampling occurred at night during a flood tide. The absence of fall and winter spawning by resident estuarine fishes would explain why no larvae were collected over this period and is consistent with the work of Witting et al. (1999) and Warlen and Burke (1990). *Lagodon rhomboides* larvae were dominant in the plume habitat during 2002, but higher numbers were recorded in the estuarine habitat during 2003. Estuarine resident taxa within the families Gobiidae, Engraulidae, Blenniidae, Atherinidae, and Soleidae generally spawn when water temps are above \( \geq 16^\circ C \) (Dahlberg and Conyers 1973). Resident taxa first appeared in April 2003 and May 2002, which coincides with this temperature. Peaks in abundance and taxonomic diversity seemed to coincide with the seasonal increase in water temperature (Warlen and Burke 1990). What is revealed by these studies conducted in New Jersey and North Carolina estuaries, is a winter and spring assemblage consistent with patterns observed in this study for the Cape Fear River plume.

Grimes and Finucane (1991) and Govoni et al. (1989) conducted ichthyoplankton studies in the Mississippi River plume comparing plume versus ocean habitats. What is interesting is the diversity of taxon and assemblages between the two systems. Dominant taxa in their studies (Carangids, Engraulids, Sciaenids, Blennioids) were similar to the dominant taxa from the Cape Fear River plume; however, the taxonomic diversity and abundances were much greater for the Mississippi River plume than the Cape Fear River
plume. This may be a result of the fact that the Mississippi River plume is much larger than the Cape Fear River plume or the difference could be from gear type. Studies from the Mississippi River plume used 1 x 2 m nets while I used 60 cm neuston nets. The ocean collection for the Mississippi River plume was dominated by carangids, which contrast findings from the Cape Fear River plume study. Although Carangids were caught in ocean samples of the Cape Fear River plume, Blennioids dominated the catch in the ocean.

Taxonomic composition was generally similar during 2002 and 2003. Sciaenids were characteristic of the plume habitat during both years and were most abundant at the bottom. Grimes and Finucane (1991) recorded carangids as the dominant taxa in the plume and ocean habitat. This also contrasts the work of Reiss and McConaugha (1999) in the Chesapeake Bay plume. They recorded Carangids and Blennioids as characteristic taxa in their plume samples. These differences from the Cape Fear River plume could be due to differences in vertical sampling. These authors did not sample the bottom where Sciaenids may have been more abundant.

Ichthyoplankton Diversity

Diversity of larval fishes varied spatially, seasonally, and interannually. Shannon-Weiner diversity values were affected significantly by habitat and depth. Highest diversity occurred in the plume at the bottom, while 1 meter depth had the greatest diversity for the ocean habitat. Ocean diversities appeared to be higher in the late spring early fall months at the surface and 1 meter, while the opposite was true at the bottom. This may indicate a mixture of individuals from different seasonal spawning
events. Overall the Shannon-Weiner diversity peaked for the three depths sampled in June thru August for 2002 and 2003. Diversities were higher at the depth 1 meter and the bottom during this period. This peak in diversity is consistent with the work performed by Jung and Houde (2002) in the Chesapeake Bay. Quattrini (2002) observed slightly higher diversity values for shelf caught larvae than reported from the Cape Fear River study. This difference could be due the fact that she sampled larvae further offshore and there may be a greater mixture of larval fishes.

Interannually, diversity at the surface and 1 meter in 2003 contrasted with that in 2002. Diversity at the bottom appeared to be slightly higher in late spring early summer for 2002 compared to 2003. There was an interesting switch in diversity between the plume and ocean habitats. In November 2002 diversity was higher in the plume habitat, but in December 2003 diversity was slightly higher in the ocean habitat.

Physiological Condition

Physiological condition measures were used to test the trophic advantage hypothesis, which is one of the suggested advantages of the plume habitat. Powell et al. (1990) reported that L. xanthurus larvae inside of the Mississippi River plume had a lower percentage of starvation than those in the plume front. Powell et al. (1990) did not find consistent differences in the nutritional condition of ichthyoplankton sampled from the Mississippi River plume and adjacent coastal waters. Devries et al. (1990) noted that king mackerel (Scomberomorus cavalla) had slightly higher growth rates when associated with Mississippi River plume water than fish from other locations. In contrast, Allman and Grimes (1998) determined from daily growth increments that little tunny
(Euthynnus alletteratus) larvae collected from ocean waters off the Panama City coastline grew faster than conspecifics collected from the Mississippi River plume. Thus many studies conducted in the Mississippi River plume are inconclusive regarding the suitability of plume habitat for larval feeding and growth.

Enzyme assays and otolith increment widths were used to assess whether the Cape Fear River plume habitat was suitable for promoting growth and physiological condition of finfish and shellfish. Habitat comparisons were originally to be performed on fishes and crustaceans from the plume and ocean sites, but I was unable to collect identical species from both habitats. Consequently I collected identical species from the plume and middle estuary and used enzymes and otoliths increments to compare the suitability of these two habitats.

There was no significant effect of habitat on otolith increment width for *L. xanthurus*. Although these results suggest that individuals from plume and estuarine habitats were growing at comparable rates, sample sizes (particularly in the plume habitat) are low and should be increased for future studies.

No significant habitat differences for *L. xanthurus*, *M. undulatus*, and *F. aztecus* were apparent from either the COX or hexokinase assays. There is a strong trend for *M. undulatus* caught in the plume to have higher activity values of hexokinase than those found in the estuary. In contrast, activity values of hexokinase for *L. xanthurus* were slightly higher in the estuary than the plume. Hexokinase values for *F. aztecus* were more variable and no apparent habitat difference is evident. This suggests that the CFR plume may be similar to the estuarine habitat. Overall correlations were weak for both species of fish suggesting further work should be conducted to analyze growth indices.
No significant evidence was found from the Cape Fear River plume to support the trophic advantage hypothesis. Future attempts to measure habitat value should incorporate multiple alternative habitats and dates using a variety of indicators (biochemical, otolith, physiological) to better assess the role of plume habitats in fisheries recruitment.

Future Issues

Objectives of this research were to test two hypotheses, 1) the aggregation hypothesis, which predicts that river discharge plumes support higher concentrations of larval fishes than adjacent coastal waters and, 2) the trophic advantage hypothesis that contends that fish larvae associated with plume waters experience a trophic advantage. Continued monitoring of the Cape Fear River plume is required to produce a long-term record. Future monitoring could help to better define essential habitats for the many resource species that were shown to occur in the Cape Fear River Plume. It would also be informative to examine the residency time of larval fishes. This study was not designed to examine residency time in the plume and whether fish larvae spend a significant portion of their life history in the plume as opposed just moving through this habitat. I also believe that more information is needed on how larval abundances and distribution might change with a flood tide in the vicinity of the Cape Fear River plume. A long-term picture could also help to understand effects of climate variability on recruitment. How would large-scale storm events in southeastern North Carolina impact larvae? During the summer months coastal North Carolina experiences weather events associated with tropical storms and hurricanes. There is a need for more thorough tests of
the aggregation and trophic enhancement hypotheses to develop a clearer understanding of this dynamic habitat.
Table 1. Atlantic croaker (Micropogonias undulates), spot (Leiostomus xanthurus) and brown shrimp (Farfantepenaeus aztecus) tissue samples used for biochemical assays and otolith microstructural analysis. TL = total length; P = Plume, E = Estuary

<table>
<thead>
<tr>
<th>I.D. Code</th>
<th>Species</th>
<th>Site*</th>
<th>TL (mm)</th>
<th>I.D. Code</th>
<th>Species</th>
<th>Site</th>
<th>TL (mm)</th>
<th>I.D. Code</th>
<th>Species</th>
<th>Site</th>
<th>TL (mm)</th>
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<td>SP-1</td>
<td>Spot</td>
<td>P</td>
<td>85</td>
<td>BS-1</td>
<td>Brown Shrimp</td>
<td>P</td>
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<td>78</td>
<td>SP-2</td>
<td>Spot</td>
<td>P</td>
<td>85</td>
<td>BS-2</td>
<td>Brown Shrimp</td>
<td>P</td>
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Table 2. Results of ANOVA to evaluate the effects of month, depth and habitat on physicochemical parameters measured at plume and ocean sites during 2002 and 2003.

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<th>Dissolved Oxygen</th>
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<td>P</td>
<td>F</td>
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<tr>
<td>Habitat</td>
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<td>16.1</td>
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<td>16</td>
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<td>Month*Habitat</td>
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<td>0.304</td>
<td>0.7</td>
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<th>Effect</th>
<th>Temperature</th>
<th>Salinity</th>
<th>Dissolved Oxygen</th>
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Table 3. Abundance of ichthyoplankton sampled during 2002 from plume, ocean and estuarine habitats. Abundance and vertical distribution of ichthyoplankton are listed for each of three depths (surface, 1 meter, bottom). Spawning location and season as well as adult habitat are provided for each species collected (Able and Fahay 1998).

<table>
<thead>
<tr>
<th>Family</th>
<th>Species</th>
<th>Common Name</th>
<th>Plume</th>
<th>Ocean</th>
<th>Estuary</th>
<th>Surface</th>
<th>1 meter</th>
<th>Bottom</th>
<th>Location</th>
<th>Season</th>
<th>Adult Habitat</th>
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<td>Balistidae</td>
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<td>sprius</td>
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<td>x</td>
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<td>x</td>
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<td>sprius</td>
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Table 4. Abundance of ichthyoplankton sampled during 2003 from plume, ocean and estuarine habitats. Abundance and vertical distribution of ichthyoplankton are listed for each of three depths (surface, 1 meter, bottom). Spawning location and season as well as adult habitat are provided for each species collected (Able and Fahay 1998).

<table>
<thead>
<tr>
<th>Family/Species</th>
<th>Common Name</th>
<th>Habitat</th>
<th>Depth</th>
<th>Spawning</th>
<th>Spawning</th>
<th>Principle</th>
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<tr>
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<td>x</td>
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<td>72</td>
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<td>Prionotus sp.</td>
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<td>x</td>
<td>x</td>
<td>x</td>
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<td>x</td>
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<td>x</td>
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<td>x</td>
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<td>x</td>
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<td>Hogchoker</td>
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<td>Sparidae</td>
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<td>Hippocampus erectus</td>
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<td>x</td>
<td>3</td>
<td>x</td>
<td>x</td>
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</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td><strong>475</strong></td>
<td><strong>73</strong></td>
<td><strong>123</strong></td>
<td><strong>103</strong></td>
<td><strong>76</strong></td>
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</table>
Figure 1. Coastal Ocean Research and Monitoring Program (CORMP) stations in the vicinity of the Cape Fear River plume, North Carolina.
Figure 2. Temperature, salinity and dissolved oxygen for plume and ocean habitats measured at the surface and bottom for the period of January 2002 to December 2003.
Figure 3A. Taxonomic composition of monthly ichthyoplankton samples from plume, ocean and estuarine sites in the vicinity of the CFR discharge plume for January and March 2002. Nomenclature abbreviations are in Table 2.
Figure 38. Taxonomic composition of monthly ichtyoplankton samples from plume, ocean, and estuarine sites in the vicinity of the CFR discharge plume for May and June 2002. Nomenclature abbreviations are indicated in table 2.
Figure 3C. Taxonomic composition of monthly ichthyoplankton samples from the plume, ocean, and estuarine sites in the vicinity of the CFR discharge plume for August and September 2002. Nomenclature abbreviations are indicated in Table 2.
Figure 3D. Taxonomic composition of monthly ichthyoplankton samples from plume, ocean, and estuarine site in the vicinity of the CPR plume for October and November 2002. Nomenclature abbreviations are indicated in Table 2.
Figure 3E. Taxonomic composition of monthly ichthyoplankton samples from the plume, ocean, and estuarine sites in the vicinity of the CFR discharge plume for February and April 2003. Nomenclature abbreviations are indicated in Table 3.
Figure 3F. Taxonomic composition of monthly samples from the plume, ocean, and estuarine site in the vicinity of the CFR discharge plume for July and August 2003. Nomenclature abbreviations are located in Table 3.
Figure 3G. Taxonomic composition of monthly ichthyoplankton samples from plume, ocean, and estuarine sites in the vicinity of the CFR discharge plume for September and October 2003. Nomenclature abbreviations are indicated in table 3.
Figure 3H. Taxonomic composition of monthly ichthyoplankton samples from plume, ocean, and estuarine sites in the vicinity of the CFR discharge plume for December 2003. Nomenclature abbreviations are indicated in Table 3.
Figure 4. Total ichthyoplankton concentration for each station sampled (stations 2 and 6= plume; stations 4,7,8,9= ocean; station 1= estuary) during all months sampled in 2002.
Figure 5. Total ichthyoplankton concentration for each station sampled (stations 2, 6, x= plume; stations 4, 7= ocean; station 1= estuary) during all months sampled in 2003.
Figure 6. Mean total ichthyoplankton concentration for plume, ocean and estuarine habitats for 2002 and 2003.
Figure 7. Shannon-Weiner Diversity indices for ichthyoplankton assemblages sampled from ocean and plume stations at each of three depths (surface, 1 meter below surface, and bottom) for the period January 2002 to December 2003.
Figure 8. Mean Shannon-Weiner Diversity indices for ichthyoplankton assemblages sampled from ocean and plume habitats at each of three depths (surface, 1 meter below surface, and bottom) averaged for all months over the period January 2002 to December 2003.
Figure 9. The relationship between body size and cytochrome c oxidase (COX) activity values for individual Atlantic croaker (*Micropogonias undulatus*) top and spot (*Leiostomus xanthurus*) bottom, collected from plume versus estuarine habitats on June 17, 2003. P-values refer to t-tests conducted to evaluate habitat effects.
Figure 10. The relationship between body size and hexokinase activity values for individual Atlantic croaker (*Micropogonias undulatus*) top and spot (*Leiostomus undulatus*) bottom, collected from plume versus estuarine habitats on June 17, 2003. P-values refer to t-tests conducted to evaluate habitat effects.
Figure 11. Mean cytochrome c oxidase and hexokinase activity values for Atlantic croaker (*Micropogonias undulatus*) and spot (*Leiostomus xanthurus*) comparing plume and estuarine habitats.
Figure 12. Correlation among various indices (otolith increment widths, cytochrome c oxidase activity and hexokinase activity) used to assess the physiological condition of Atlantic croaker sampled from estuarine vs. plume habitats. Panels shown in the matrix diagonal indicate the relative frequency distribution of individuals for each condition index.
Figure 13. Correlation among various indices (otolith increment widths, cytochrome c oxidase activity, and hexokinase activity) used to assess the physiological condition of Atlantic croaker sampled from estuarine versus plume habitats. Panels shown in the matrix diagonal indicate the relative frequency distribution of individuals for each condition index.
Figure 14. The relationship between body size and hexokinase activity values for individual brown shrimp (*Farfantepenaeus aztecus*), collected from plume and estuarine habitats on June 17, 2003. P-value refers to t-test conducted to evaluate habitat effects.
Figure 15. The relationship between body size and average otolith increment widths of spot (*Leiostomus xanthurus*) (top) and Atlantic croaker (*Micropogonias undulatus*) (bottom), sampled from plume and estuarine habitats. P-values refer to t-test conducted to evaluate habitat effects.
Figure 16. Mean marginal otolith increment widths for spot (*Leiostomus xanthurus*) (left) and Atlantic croaker (*Micropogonias undulatus*) (right), collected from plume and estuarine habitats. P-values from t-tests comparing mean values from plume and estuary sites are \( p = 0.643 \) and \( p = 0.456 \) for spot and Atlantic croaker respectively.
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