

Maturation and Fecundity of the Neuse and Tar-Pamlico Rivers Striped Bass (*Morone saxatilis*)  
Stocks in Coastal North Carolina

By

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Abstract

The Albemarle/Roanoke stock makes up the largest contingent of Striped Bass (*Morone saxatilis*) in the state; however, other economically important populations exist elsewhere. The Central Southern Management Area (CSMA) is one such region. Striped Bass populations are sustained in the CSMA through stocking by the North Carolina Wildlife Resources Commission (NCWRC), but the goal of the fishery management plan is to establish a self-sustaining spawning population of Striped Bass in the region. In order to improve management of the CSMA Striped Bass population, maturation and fecundity estimates are needed for stock assessment models. Striped Bass were sampled on and near the spawning grounds in the Neuse and Tar/Pamlico rivers during the pre-spawn, spawning, and post-spawn period (February-June). Each fish was measured (fork length and total length, mm) and weighed (g). Otoliths were removed for ageing and otolith chemistry. Sex was determined, and gonads were removed and weighed to determine the GSI and phase of reproduction. LSI and K factors were used to assess the condition of fish. Otoliths were sectioned for age, and then examined by LA-ICPMS to determine changes in concentrations of Strontium in the first year of life to determine origin. The age at 50% maturity was 2.67 years; by Age 3 98.2% of female Striped Bass were sexually mature. CSMA Striped Bass matured 0.5 years earlier compared to the ASMA/RRMA

population. Fecundity ranged from 223,110 eggs for an Age-3 female to 3,273,206 eggs for an Age-10 female (Mean=769,048.54; SE=54,047.42; n=87). Fish in the CSMA produced more eggs than ASMA/RRMA Striped Bass when compared by age. Observed lengths at age were significantly different between the CSMA and ASMA/RRMA females ( $F=978.92$ ;  $DF=1$ ;  $p<0.0001$ ). Results of otolith microchemistry revealed that 92.7% of female Striped Bass in my study were of hatchery origin. Hatchery and wild fish did not vary significant by condition factors or age and growth; however, oocyte characteristics did vary significantly by origin. Wild fish produced oocytes of higher mass and larger in diameter compared to hatchery fish. Length at 50% maturity in the CSMA was estimated at 467.8 mm TL (18.4 inches TL) and fish were estimated to be 100% mature at 537.3 mm TL (21.1 inches TL). The current harvest restrictions for Striped Bass are an 18-inch TL minimum. In order for fishery managers to ensure all fish have the opportunity to enter the spawning stock at least once before being harvested, minimum harvest restrictions should be set at 21 inches TL. Both wild and hatchery adults are capable of fully contributing to the spawning run, yet the stock is not recovering. Early life mortality investigations into why the stock is not recovering should be undertaken.



**Maturation and Fecundity of the Neuse and Tar-Pamlico Rivers  
Striped Bass (*Morone saxatilis*) Stocks in Coastal North Carolina**

A Thesis

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## Introduction

Striped Bass (*Morone saxatilis*) is one of the most important commercial and recreational fish species on the eastern seaboard (NCDENR 2011). Striped Bass inhabit lakes, rivers, estuaries, and the Atlantic Ocean on the east coast of the United States, and have been introduced elsewhere (Rulifson and Laney 1999). Mid-Atlantic and northern populations of Striped Bass are anadromous, meaning they spawn in rivers and spend their adult lives moving between rivers, estuaries, and the ocean (Merriman 1941). When Striped Bass spawn, they migrate to the spawning grounds of their river of origin (Greene et al. 2009). Males gather around females (usually five or more males) and force the female to the surface; the female then broadcasts eggs into the water and the males release milt to fertilize the eggs. Successful spawning requires water to have suitable flows, oxygen levels, temperature, and salinity (Rulifson et al. 1982); alteration of river flows can negatively affect the success of spawning (Rulifson and Manooch 1993). After spawning, the fertilized eggs hatch into larvae; these larval fish are carried down river to nursery habitats where they metamorphose into juveniles. At approximately 30 mm total length (TL), fins are fully formed and juveniles resemble adult Striped Bass. Juveniles can remain in the nursery areas for up to two years (Hassler et al. 1981; Setzler et al. 1980). In Albemarle Sound most female Striped Bass are sexually mature at age four, but males mature as young as age three (Olsen and Rulifson 1992; Boyd 2011). Striped Bass are iteroparous and when they reach sexual maturity, they migrate upstream to spawn in their natal river. Large Striped Bass that reach the ocean can migrate as far north as Cape Cod, Massachusetts, during the summer and migrate south to the North Carolina and Virginia border during the winter (Merriman 1941; Chapoton and Sykes 1961; Setzler et al. 1980; Callihan et al. 2014). Due to the

highly migratory characteristics of Striped Bass, fishery managers have utilized mark-recapture and more recently otolith chemistry studies to determine migration patterns of stocks (Merriman 1941; Chapoton and Sykes 1961; Secor et al. 1995; Paramore and Rulifson 2001; Morris et al. 2003; Morris et al. 2005).

In North Carolina, Striped Bass inhabit lakes, river systems, and the coastal ocean environments. The largest coastal stock of Striped Bass is located in the Albemarle Sound Management Area (ASMA) and Roanoke River Management Area (RRMA). However, other smaller populations exist in North Carolina including the Tar-Pamlico, Neuse, and Cape Fear (CFR) rivers. These populations are managed collectively as one unit known as the Central Southern Management Area (CSMA) (Figure 1), which is managed jointly by the North Carolina Division of Marine Fisheries (NCDMF) and the North Carolina Wildlife Resources Commission (NCWRC). Striped Bass in the CSMA are considered endemic and riverine, and rarely migrate outside of their respective river system (Setzler et al. 1980); however, one fish tagged in the CFR was recaptured in Buzzards Bay, Massachusetts (NCDENR 2011).

The upper portions of the CSMA rivers are oligohaline with salinities less than 0.1 ppt. The lower portions of the rivers are mesohaline with salinities ranging from 0.1 ppt to 32 ppt in the CFR, and 0.1 ppt to 22 ppt in the Neuse and Tar/Pamlico (Roelofs and Bumpus 1953). The Tar-Pamlico River, Neuse River and CFR have dams that prevent passage of anadromous fish species from reaching historical spawning grounds (Smith and Hightower 2012). Dams are located in Rocky Mount, North Carolina on the Tar River; Raleigh, North Carolina on the Neuse River; and Riegelwood, Elizabethtown, and Fayetteville, North Carolina, on the CFR. These dams on the CFR were constructed to allow ship traffic to move upriver, but the dams are no longer necessary because no ships have traveled upriver within the last decade. These dams have

most likely contributed to declines in populations of anadromous species such as Atlantic Sturgeon (*Acipenser oxyrinchus oxyrinchus*), American Shad (*Alosa sapidissima*), and Striped Bass (Beasley and Hightower 2000; Burdick and Hightower 2006; Smith and Hightower 2012). The CSMA possesses an active spawning population of Striped Bass in each river, but shows little evidence of self-recruitment based on the low abundance of juvenile Striped Bass in the nursery areas (NCDENR 2011). This suggests that there is high mortality of eggs and larvae, and/or insufficient spawning stock biomass.

The NCRWC and NCDMF stock 100,000 phase II (4-8 inch juvenile) Striped Bass each year in each of the three main CSMA coastal rivers in hopes of sustaining the Striped Bass fishery. Dobbs (2013) found that phase II Striped Bass stocked into CSMA coastal rivers had extremely high survival rates; 88% of sampled fish were phase II hatchery-reared fish, and only 12% were wild fish, reinforcing the theory that there is high larval/juvenile mortality and/or insufficient spawning stock. Phase II hatchery-reared fish may have an unfair advantage compared to wild spawned fish because the hatchery-reared fish are not susceptible to the abiotic and biotic factors causing high mortality rates of the “wild” larval and juvenile Striped Bass in the Tar/Pamlico, Neuse, and CFR. Unsuccessful recruitment from the larval to the juvenile phase could be attributed to multiple factors such as habitat degradation or inadequate flow rates in the river that do not successfully transport larval Striped Bass to primary nursery areas (Rulifson and Manooch 1990; Smith and Hightower 2012).

One major concern of researchers and fisheries biologists is that the current stocking regime for Striped Bass is ineffective (Patrick and Stellwag 2001; NCDENR 2011). The Roanoke River has been used as a source of broodstock for the Tar-Pamlico, Neuse, and Cape Fear rivers since 1980 (Woodroffe 2012). The Roanoke River broodstock used to spawn

hatchery-raised Striped Bass may not be genetically or ecologically fit for surviving, successfully spawning, or producing offspring with viable eggs in the CSMA rivers (Rulifson and Laney 1999; Bergey et al. 2003). Genetically and ecologically unfit fish producing eggs not suited to the Neuse and Tar-Pamlico rivers would help explain why unsuccessful spawning activity is occurring. Such a scenario is suggested by little evidence of successful recruitment of wild age 0 fish to the forming year class. In 2010, the NCWRC instituted an endemic stocking program in CSMA rivers in order to increase the spawning population of Striped Bass. The endemic stocking program uses fish collected on the spawning grounds in the Tar/Pamlico, Neuse, and CFR to use for broodstock in the hopes that endemic broodfish will produce offspring with viable eggs that are suited for each individual river system. The CFR was the first river stocked with phase II juvenile fish of endemic parentage (i.e., progeny of adult wild fish from the Cape Fear River were stocked only in the CFR). In 2012 endemic stocking was initiated in the Tar/Pamlico and Neuse rivers. All three river systems continue to receive progeny from their respective broodfish parentage (Kevin Dockendorf, NCWRC, personal communication).

Fishery managers suspect that the populations currently dominated by hatchery-reared fish may be one cause of spawning failure in these systems. Because hatchery fish are stocked in late fall after recruitment of wild juveniles to the forming year class is complete, the larger hatchery fish may dominate habitats and possibly mature earlier than wild fish, essentially lowering the success rate of wild fish even more. One way to examine this hypothesis is through comparisons of reproduction between wild and hatchery-reared Striped Bass. If differences in reproduction are observed, managers can use this information to adjust harvest size limits. Maturation and fecundity assessments have not been conducted in the CSMA rivers to date.

The goal of this study was to determine if Striped Bass in the CSMA are maturing at a rate similar to the ASMA/RRMA population. Age at maturity, condition indices, oocyte characteristics, and fecundity of stocked hatchery fish (Roanoke broodstock) are compared to endemic wild fish. Any observed differences in fecundity and maturity could provide relevant information on poor recruitment within the CSMA.

## Objectives

The following objectives were used to assess Striped Bass reproductive biology:

- 1) Obtain female Striped Bass from the NCWRC electroshocking survey, and the NCDMF independent gillnet survey on the spawning grounds and main stem of the Neuse and Tar/Pamlico rivers (2013 and 2014) during the pre-spawn, spawning, and post-spawn period (February-June).
- 2) Analyze biological samples to determine age, Liver Somatic Index (LSI), gonadosomatic index (GSI), Fulton's condition factor (K), oocyte characteristics, age at maturity, and fecundity.
- 3) Determine the origin (hatchery, endemic wild fish, or transient from another stock) of Striped Bass collected on the spawning grounds and main stem of the Tar/Pamlico and Neuse rivers using Laser Ablation-Inductively Coupled Plasma Mass Spectrometry (LA-ICPMS).
- 4) Develop a maturation and fecundity schedule for the Tar/Pamlico and Neuse Rivers and compare the fecundity, maturation schedule, and condition indices of hatchery-reared and wild Striped Bass.

- 5) Compare results with Boyd (2011) and identify significant differences between the ASMA/RRMA and CSMA Striped Bass populations.
- 6) Provide recommendations for management.

## Methods

Striped Bass were collected by North Carolina Wildlife Resources Commission (NCWRC) staff and North Carolina Division of Marine Fisheries (NCDMF) staff in 2013 and 2014 as part of each agency's annual sampling programs. Striped Bass were already euthanized and dead when they were received by ECU researchers. Fish were collected on the spawning grounds and main stems of these rivers from February through June during the pre-spawn, spawning, and post-spawn periods. In order to maintain the covariance at less than 10% for the average fecundity of a population, a minimum sample of 50 or more females is required (Hunter et al. 1985); therefore, investigators set the target number of samples at 50 females per river system per spawning year. NCWRC biologists recommended using the minimum sample size of female Striped Bass because very few female Striped Bass arrive on the spawning grounds, and taking more than necessary could negatively affect the spawning success of the CSMA population (Kevin Dockendorf, NCWRC, personal communication). Fish were collected from different size classes to ensure all age classes would be represented (Table 1). All female Striped Bass collected were used for maturation and fecundity analysis and to develop the CSMA age at maturity schedule.

Each fish was measured (FL and TL, mm) and weighed (0.1 g), and gender verified. Gonads and livers were removed and weighed (0.1 g). Otoliths were removed with plastic forceps, cleaned with distilled water, and stored in 1.5-mL micro-centrifuge tubes for ageing and micro-chemical analysis.

Protocols for maturation and fecundity analyses followed those used by Boyd (2011). Ovaries were removed and fixed in cold 10% buffered formalin and stored in a 4°C refrigerator for 24-48 hours, rinsed in distilled water for one hour, and then preserved with 70% histological

grade ethyl alcohol and stored in 500-mL sample containers. The middle portion of one ovary for each fish was excised, placed in 70% ethyl alcohol, and shipped to North Carolina State University in Raleigh, NC, for histological preparation. The anterior, middle, and posterior sections of the ovary develop similarly (Olsen and Rulifson 1992; Paramore and Rulifson 2001). If both ovaries appeared similar, then the ovary to be used for histological analysis was chosen at random. A subsample of the remaining ovary was used for fecundity analysis.

Histological preparation included dehydrating samples with alcohol, clearing with xylene, and permeating and embedding in low melting point paraffin. Serial sections were then cut in 4-micron increments. Paraffin was removed and sections were stained using Hematoxylin and Eosin (H&E).

Terminology used to assess the various reproductive phases followed those used by Brown-Peterson et al. 2011 and Lowerre-Barbieri et al. 2011 (Table 2). Briefly, histological sections of individual fish were analyzed using an Olympus SZX16 research stereomicroscope and Image-Pro Plus 5.1 analysis software, and categorized into one of five phases. Fish were classified as immature (Figure 2), developing (Figure 3) (early developing subphase), spawning capable (Figure 4), actively spawning subphase (Figure 5), regressing (Figure 6), or regenerating (Figure 7). Fish in the immature phase have small ovaries with indistinct blood vessels, and only primary growth (PG) oocytes can be seen histologically. In the developing phase ovaries become enlarged and blood vessels become more distinct. Primary growth (PG), cortical alveolar (CA), primary vitellogenic (Vtg1), and secondary vitellogenic (Vtg2) can be present in the developing phase but fish in the developing phase are not ready to spawn. Gonads in the spawning capable phase are large with prominent blood vessels and macroscopically visible oocytes. Tertiary vitellogenic (Vtg3) oocytes are present as well as early stages of oocyte

maturation (OM) in the spawning capable phase. Oocytes undergoing late germinal vesicle migration (GVM), germinal vesicle breakdown (GVBD), hydration, or ovulation are considered to be in the actively spawning subphase of the spawning capable phase. The regressing phase is distinguished by flaccid ovaries with prominent blood vessels; this phase of reproduction was formerly referred to as “spent”, a term still commonly used by fisheries biologists. In the regressing phase, postovulatory follicle complex (POF) and atresia are present; also there may be some CA and/or Vtg1, Vtg2 oocytes present in the regressing phase. In the regenerating phase, fish are sexually mature but reproductively inactive, only oogonia and PG oocytes are present though a thick ovarian wall, and degenerating POFs may also be present.

Secondary growth characteristics were examined to determine maturity and phase of maturity. Female Striped Bass were considered to be immature if they contain only primary growth oocytes (Brown-Peterson et al. 2011), and were considered mature if the ovaries contain oocytes in the secondary growth phase (Berlinsky and Specker 1991; Brown-Peterson et al. 2011). The length and width of 100 oocytes showing signs of secondary growth (Vtg) were measured using Image-Pro Plus 5.1 to determine mean oocyte diameter in each fish; only oocytes sectioned through the nucleus were measured (Morris et al. 2011).

Methods for the fecundity analysis followed those described in Boyd (2011). Only fish found to be mature through maturation analysis were used for fecundity analysis. After preservation in 70% ethyl alcohol, two subsamples of ovarian tissue were removed from the middle portion of the ovary since there is no significant difference between sections of the ovary (Merriman 1941; Olsen and Rulifson 1992). The subsamples of ovarian tissue were placed in 0.2-mm and 0.4-mm sieves and held under flowing water until the oocytes were separated. Oocytes were then blotted dry, and two 1.0-g samples were taken and placed in a glass vial with

70% ethyl alcohol. Vials were shaken for approximately one minute to ensure separation of the oocytes, stained using red eosin phloxine, and then placed in a petri dish with a soapy solution to reduce surface tension. Petri dishes were illuminated from underneath to show a high contrast between the eggs and background, and images were captured using a Nikon D5100 digital camera mounted in a fixed position. Images were uploaded into Image-Pro software; the number of eggs were automatically counted using the color cube based function. The mean number of eggs per gram was used to calculate the number of eggs in each ovary by multiplying mean eggs per gram by total ovary weight (Cailliet et al. 1986).

The Liversomatic index (LSI), Gonadosomatic index (GSI), and Fulton's Condition Factor (K) condition indices were calculated in the same manner as that of Gentry (2006). LSI was calculated as a percentage of the total body weight using the formula,  $LSI = (W_{liver}/W_{total}) \times 100$ . LSI values indicate the state of immediate condition or well-being of the fish during spawning and feeding, and reflect changes in the glycogen content of the liver. GSI was calculated using the formula,  $GSI = W_{gonads}/W_{total} * 100\%$ . Changes in GSI show how much energy an individual fish is allocating for gonadal development. K factor was calculated using the equation,  $K = W/L^3 \times 100,000$ , where W is the total wet weight (g) and L is the total length (mm) of the fish. Changes in K factor show the relative condition as a function of mass to length of individual fish.

Otoliths were used to age each specimen. Otoliths are assumed to be more accurate compared to scales, which tend to underestimate the true age for fish 10 years and older (Welch et al. 1993; Paramore and Rulifson 2001; Boyd 2011), and overestimate true age for fish age 4 and younger (Boyd 2011). Otoliths were sectioned using a Hilquist model 800 thin-sectioning machine with diamond blade. Otoliths were polished, and then read under an Olympus SZX16

research stereomicroscope. A primary and secondary reader aged all otoliths; a third reader resolved any discrepancies in otolith age (Welch et al. 1993). Agreement between multiple readers is the standard method to assess age agreement or precision.

To determine natal origin, otoliths were analyzed by LA-ICPMS using similar methods as described by Mohan (2009) and Mohan et al. (2012). The sagittal otolith pairs were extracted using non-metallic forceps, scrubbed to remove surface tissue, cleaned with distilled deionized water, and stored in open 1.5-ml micro-centrifuge polypropylene vials to dry for a period of 24 hours. After drying, the 1.5-ml micro-centrifuge polypropylene vials remained closed until LA-ICPMS analysis. One otolith was randomly selected for chemical analysis, as there is no significant difference in mass or chemical composition between the left and right otoliths (Mohan 2009; Mohan et al. 2012). Samples were then analyzed at the University of Manitoba, Winnipeg, Canada. For analysis, otoliths were embedded in an epoxy resin (Buehler Epoxicure) and a 2-mm thick dorso-ventral transverse section, including the core, were cut using a diamond blade Isomet saw (Buehler 646) at low speed. The cut sections were then re-embedded in 25-mm diameter, Plexiglas ring mounts (typically four otoliths per mount). The orientation and identity of each section within each ring mount was photographed for sample reference. To expose the nucleus region and otolith core, sections were ground down using wet sandpaper and polished with Buehler diamond polishing suspensions (9  $\mu\text{m}$  and 0.05  $\mu\text{m}$ ), ultrasonically cleaned for 2 min, and then digitally photographed to create an illustrated reference for LA-ICPMS analysis.

Laser scans were initiated beyond the nucleus, and conducted through the core and continued along the longest axis of otolith growth to the outer edge. Isotope counts of elements Strontium ( $\text{Sr}^{88}$ ), Barium ( $\text{Ba}^{138}$ ), Magnesium ( $\text{Mg}^{25}$ ), and Manganese ( $\text{Mn}^{55}$ ) were converted to ppm using Microsoft Excel and plotted versus laser distance. Relatively low stable levels of  $\text{Sr}^{88}$

and high Ba<sup>138</sup> throughout the otolith indicate resident riverine fish (Secor and Piccoli 1996). Increased Sr levels, followed by sudden and dramatic changes in Sr to low levels, indicate entry from ocean or estuarine environments into fresh waters to spawn (Secor and Piccoli 1996).

Dobbs (2013) identified a unique elemental pattern that results from a fish being reared at the Edenton National Fish Hatchery. This unique elemental pattern (Figure 8) is retained throughout the life of the fish. This pattern appearing in the otolith nucleus (the area laid down prior to the first annulus) was used as a natural tag to determine if a fish is of hatchery origin or wild origin (Campana 1999; Dobbs 2013). Results from LA-ICPMS analysis were used to determine origin of the Striped Bass used in my study, which made it possible to compare differences in condition indices and maturity and fecundity between fish of wild and hatchery origin. Techniques similar to that of Dobbs (2013) were used to determine origin.

#### *Statistical Analysis*

All data were stored and formatted using Microsoft Excel (2011) and then later analyzed using Microsoft Excel (2011) and JMP pro 10. For statistical analysis an analysis of variance (ANOVA) was used to make comparisons of FL, TL, age, GSI, LSI, maturity, and fecundity among all sampled fish, hatchery origin, and wild endemic fish. These comparisons were selected to test for significant differences in condition indices, oocyte characteristics, maturity, and fecundity of wild and hatchery-reared fish. Linear regressions were used to determine fecundity by age and total length (mm). Logistic regressions were used to determine age at 50% maturity (A<sub>50%</sub>) and total length (mm) at 50% maturity (L<sub>50%</sub>). Analysis of Covariance (ANCOVA) was used to compare origin, river, and management areas that had two or more variables.

## Results

### *Data Collection*

A total of 165 female Striped Bass were collected between April 17, 2013, and June 30, 2014 from the spawning grounds and main stem of the Tar/Pamlico (n=91) and Neuse (n=74) rivers. Female Striped Bass were collected by the NCWRC spring electroshocking survey and NCDMF independent gill net survey. Striped bass were already euthanized and dead when ECU graduate students collected samples. Samples were collected on the Neuse River between Raleigh, NC, and New Bern, NC, and Tar River between Dunbar Bridge and Aurora, NC.

### *Age and Growth*

Striped Bass in the Tar/Pamlico and Neuse rivers ranged from 217 mm to 943 mm TL (Mean=568.45; SE=8.93; n=165) with most fish being in the 525-549 mm size class; total length was normally distributed (Table 3). Age ranged from 1-10; the majority of fish sampled were Age 3 and Age 4. Weight ranged from 0.1092 kg to 9.582 kg (Mean=2.292; SE=0.1155; n=165); weight was not normally distributed and was skewed to the right. A logistic regression model was used to fit length at age data (n=165) to produce the Von Bertalanffy growth model. The Von Bertalanffy equation (Figure 9) estimated for female Striped Bass was

$$L_t = 905.171(1 - e^{-0.21015(t - 0.92704)}).$$

The weight-length relationship was estimated using data from 165 female fish (Figure 10):

$$\text{Weight (g)} = -4519.463 + 11.55939 * TL \text{ (mm)} + 0.0184272 * (TL - 568.455)^2$$

### *Condition*

GSI, K factor, and LSI were calculated by river (Table 4). GSI in the CSMA ranged from 0.035 to 23.135 (Mean=6.76; SE=0.43; n=165). GSI for Tar/Pamlico fish ranged from 0.035 to 17.53 (Mean=6.01; SE=0.57; n=91) and for Neuse River fish ranged from 0.052 to 23.135

(Mean=7.67; SE=0.65; n=74). GSI varied significantly between the Neuse and Tar/Pamlico fish (T=-1.91903; DF=154.9805; chi square=3.9033; p=0.0482\*).

LSI in the CSMA ranged from 0.44 to 2.40 (Mean=1.31; SE=0.02; n=165). LSI for Neuse River fish ranged from 0.62 to 1.95 (Mean=1.27; SE=0.03; n=74) and for Tar/Pamlico fish ranged from 0.44 to 2.40 (Mean=1.34; SE=0.03; n=91). LSI did not vary significantly between the Neuse and Tar/Pamlico fish (T=1.59; DF=161.44; p=0.1143). LSI for fish from both river systems was normally distributed.

K factor in the CSMA ranged from 0.86 to 1.35 (Mean=1.11; SE=0.01; n=165). K factor for Neuse River fish ranged from 0.86 to 1.33 (Mean=1.11; SE=0.01; n=74), and ranged from 0.89 to 1.35 (Mean=1.10; SE=0.01; n=91) in Tar/Pamlico River fish. K factor did not vary significantly between the Neuse and Tar rivers (T=-0.163; DF=153.87; p=0.8709) and was normally distributed in both river systems.

### *Maturity*

Age at 50% maturity was estimated at 2.67 years using a logistic regression model, and by Age 3 98.2% of all females were mature (Figure 11). All Age 2 and younger fish were immature. Length at 50% maturity (Figure 12) for female CSMA Striped Bass (n=165) was estimated at 467.8 mm TL (18.4 inches TL) using a logistic regression model. Fish were estimated to be 100% mature at 537.3 mm TL (21.1 inches TL).

### *Oocyte Characteristics*

Oocyte diameter was measured using Image-Pro Plus 5.1 imaging software to calculate mean oocyte diameter. Only fish with vitellogenic oocytes (n=104) were used when calculating oocyte diameter. Mean oocyte diameter for the Tar/Pamlico and Neuse river populations ranged from 98.12  $\mu\text{m}$  to 913.33  $\mu\text{m}$  (mean=675.01; SE=12.43; n=104). Oocyte diameter was not

correlated with weight ( $R^2=0.017$ ), total length ( $R^2=0.026$ ), or age ( $R^2=0.018$ ). In Neuse River fish, diameter ranged from 428.32  $\mu\text{m}$  to 884.99  $\mu\text{m}$  (mean=689.76; SE=13.99; n=52) and in the Tar/Pamlico fish diameter ranged from 98.12  $\mu\text{m}$  to 913.33  $\mu\text{m}$  (mean=660.25; SE=20.48; n=52). A two-sample t-test was used to compare mean oocyte diameter between river systems; oocyte diameter did not differ significantly between the Neuse and Tar/Pamlico River fish ( $T=1.19$ ;  $DF=90.09$ ;  $p=0.2373$ ).

Mean number of eggs per one-gram sample ranged from 1,652 to 17,668 eggs per gram (mean=3,658.64; SE=284.75; n=87). Mean eggs per gram was not correlated with weight ( $R^2=0.004$ ), total length ( $R^2=0.009$ ), or age ( $R^2=0.007$ ). Eggs per gram ranged from 1,700 to 8,998.5 (mean=3,446.79; SE=240.66; n=43) for the Neuse and for the Tar/Pamlico eggs per gram ranged from 1,652 to 17,668 (mean=3,865.67; SE=513.17; n=44). A Kruskal Wallance/Wilcoxon test was used to compare mean eggs per gram between river systems; eggs per gram did not differ significantly between the Neuse and Tar/Pamlico River populations ( $T=0.739$ ;  $DF=60.97$ ; chi square=0.3947;  $p=0.5299$ ).

### *Fecundity*

Striped Bass in the spawning capable reproductive phase (n=87) were used to estimate fecundity to ensure no eggs had already been released, which is possible in the regressing or actively spawning phase. Fecundity varied significantly by 25-mm size class ( $F=37.9609$ ;  $DF=13,73$ ;  $p<0.0001$ ) as well as by age ( $F=62.9346$ ;  $DF=7,79$ ;  $p<0.0001$ ), with older and longer fish producing more eggs than younger and shorter fish. Fecundity ranged from 223,110 eggs for an age-3 female to 3,273,206 eggs for an age 10 female (Mean=769,048.54; SE=54,047.42; n=87). Fecundity was not distributed normally and was skewed to the right. The relationship

between age and fecundity (Figure 13) was linear ( $R^2 = 0.667$ ) and was estimated using the formula:

$$Fecundity = -312599.8 + 243791.21 * AGE.$$

The relationship between total length (mm) and fecundity (Figure 14) was also linear ( $R^2 = 0.777$ ) and was estimated using the formula:

$$Fecundity = -2221005 + 5004.7067 * Total\ length\ (mm).$$

#### *Hatchery vs. Wild origin*

Origin was determined by analyzing results of LA-ICPMS and using methods described by Dobbs (2013). Of the 165 fish sampled, 153 (92.7%) were of hatchery origin and 12 (7.3%) were of wild origin. Age and growth, condition, and fecundity were compared between Neuse and Tar fish of hatchery and wild origin. No significant differences were found when total length was compared by origin for each age class using an ANOVA. Results of an ANCOVA determined that there was no significant difference in total length (mm) when compared with age and origin ( $F=0.0785$ ;  $DF=1$ ;  $p=0.7797$ ). No significant differences were found when weight was compared by origin for each age class using an ANOVA.

All hatchery fish under 525 mm TL were excluded when comparing between hatchery and wild origin because there were no wild fish captured < 525 mm TL. Normal quantile plots were visually assessed for normality of variables being compared; if normal quantile plots were distributed normally then a two-sample t-test was used to compare variables between fish origin. If normal quantile plots were not distributed normally, then a non-parametric Kruskal Wallace/Wilcoxon test was used to compare variables to origin. Mean oocyte diameter, LSI, and K Factor were distributed normally, and mean eggs per gram, fecundity, and GSI were distributed non-normally. Mean eggs per gram varied significantly by origin ( $T=-2.92$ ;

DF=38.63; Chi square=3.8687; p=0.0492\*) and mean oocyte diameter also varied significantly by origin (T=2.852; DF=21.757; p=0.0093\*). However, fecundity (T=1.428; DF=9.216; chi square=0.2571; p=0.6121) and LSI (T=0.646; DF=12.471; p=0.5299) did not vary significantly by origin. GSI did not vary significantly (T=3.296; DF=22.985; chi square=3.7955; p=0.0514) by origin, but was nearly significant with a p-value of 0.0514 with the alpha threshold set at 0.05. K factor did not vary significantly by origin (T=-0.09307; DF=28.64602; p=0.9265).

*Tar/Neuse 2013 & 2014 vs. ASMA/RRMA 2009 & 2010*

CSMA Striped Bass mature earlier than the Albemarle/Roanoke Striped Bass stock. In 2013 and 2014 age at 50% maturity for the CSMA (n=165) Striped Bass population was 2.67 years while the 2009 and 2010 ASMA/RRMA (n=397) population was 3.2 years; the CSMA population is reaching age at 50% maturity 0.5 years earlier than the ASMA/RRMA (Figure 16).

CSMA Striped Bass are larger at 50% maturity than Albemarle/Roanoke River fish. Total length at 50% maturity for the CSMA Striped Bass population (n=165) was 467.84 mm and the ASMA/RRMA (n=391) population was 414.6 mm, with the ASMA/RRMA population reaching 50% maturity at 51.21 mm shorter than the CSMA population (Figure 17).

CSMA Striped Bass are more fecund at age compared to Albemarle/Roanoke population. The slope of the linear regression for fecundity in the CSMA is about the same compared to the ASMA/RRMA (Figure 18). However, the CSMA fish produce more eggs at age; this indicates that Striped Bass in the CSMA are producing more eggs at the same age compared to the ASMA/RRMA population.

Striped Bass in the CSMA populations grew at a faster rate and had a greater total length (mm) at age compared to the ASMA/RRMA population (Figure 19). Growth rates were

compared between the CSMA and ASMA/RRMA using the Von Bertalanffy growth model. The Von Bertalanffy equation was estimated for female Striped Bass in the CSMA using the formula:

$$L_t = 905.171(1 - e^{-0.21015(t - 0.92704)})$$

The Von Bertalanffy equation was estimated for female Striped Bass in the ASMA/RRMA using the formula:

$$L_t = 1052.10(1 - e^{-0.125(t - 1.0)})$$

In order to determine if growth rates were significantly different, observed lengths at age for the CSMA and ASMA/RRMA were compared using an analysis of covariance (ANCOVA).

Observed lengths at age were significantly different between the CSMA and ASMA/RRMA ( $F=978.92$ ;  $DF=1$ ;  $p<0.0001$ ). To validate differences in observed lengths at age, a non-parametric Kruskal Wallace/Wilcoxon was used to compare the mean lengths at age by management area, a non-parametric test was used because sample sizes were not equal and distributions were non-normal. Age 1 fish were not used since Boyd (2011) did not collect any age 1 fish. Observed lengths were significantly different from Age 2 to Age 5: at Age 2 (chi square=25.24;  $DF=1$ ;  $p<0.001$ ), Age 3 (chi square=74.40;  $DF=1$ ;  $p<0.001$ ), Age 4 (chi square=46.61;  $DF=1$ ;  $p<0.001$ ), and Age 5 (chi square=33.79;  $DF=1$ ;  $p<0.0001$ ). Observed lengths were not significantly different between fish of Age 6 through Age 10: at Age 6 (chi square=2.89;  $DF=1$ ;  $p=0.0893$ ), Age 7 (chi square=2.40;  $DF=1$ ;  $p=0.1213$ ), Age 8 (chi square=0.00;  $DF=1$ ;  $p=1.0000$ ), Age 9 (chi square=0.07;  $DF=1$ ;  $p=0.7963$ ), and Age 10 (chi square=3.75;  $DF=1$ ;  $p=0.0527$ ). Comparisons of observed lengths at Age 12 and Age 15 could not be made because no fish of those ages were collected in the CSMA.

## Discussion

Striped Bass were sampled during the pre-spawn, spawning, and post-spawn period to ensure all phases of reproduction were represented. Samples were collected by 75-mm size classes, similar to those used by Boyd (2011), to produce a representative subset of the Tar/Pamlico (n=91) and Neuse (n=74) river populations. Fish were sampled with a variety of different sampling gears to avoid gear biases. Striped Bass were collected at different frequencies for each size class to produce a dataset with a normal distribution of age and length (Figure 15). However, this sampling regime resulted in relatively few young (Ages 1 and 2) and small fish (<400 mm) despite concentrated sampling efforts. Young and small fish were encountered infrequently during sampling. The NCWRC also found few young and small fish in the CSMA during spring electroshocking surveys (Dycus et al. 2014). The lack of age 1 and age 2 fish collected in this study could be due to young fish being too small to be captured in the size selective sampling gear used, and is most likely not a reflection of overall abundances of age 1-2 fish. However, few age 1-2 fish have been collected in the CSMA, even when using a wide variety of sampling gears, such as electrofishing, beach seine, gill net, and rod and reel (Barwick et al. 2009; Dobbs 2013; Dycus et al. 2014; McCargo et al. 2014). Electrofishing was the primary gear used to sample Striped Bass in the CSMA in this study; Bohlin et al. (1989) has shown that electrofishing has the potential to select for larger fish when sampling. It is possible that the areas sampled were not suitable habitat for juvenile Striped Bass. Submerged Aquatic Vegetation (SAV) beds have been shown to be suitable habitats for juveniles in the Albemarle Sound and Chesapeake Bay (Setzler et al. 1980; NCDENR 2011), these areas were not specifically targeted during sampling and this may be another reason for the lack of juveniles.

### *Maturity*

Samples were collected from a wide range of size classes to ensure that immature and mature fish were sampled. Immature and mature fish are needed to determine an accurate age at maturity. However, even with a concerted effort, only 18 immature fish were collected out of the 165 total fish during this study. The relatively low number of immature fish in the dataset could be due to sampling bias with electrofishing, rod and reel, and gill net selecting for larger fish. This could also reflect the lack of young-aged fish present in the population. Beach seines may be a more effective method of collecting juvenile fish. Very few juvenile fish have been collected in juvenile abundance index surveys to show evidence for successful recruitment (Barwick et al. 2009). Estimated age at maturity in the Tar/Pamlico and Neuse river populations was lower compared to estimates in the literature of the Roanoke/ASMA and Chesapeake populations (Olsen and Rulifson 1992; Setzler et al. 1980). These results were somewhat expected; Dobbs (2013) found that GSI values increased significantly from age 2 to 3, this indicates that gonads are more developed at age 3 compared to age 2 fish and would presumably be mature. The differences in age at maturity in these populations could be due to variations between stocks, population size, environmental conditions, or other factors. It is important to compare changes in age at maturity directly between individual stocks to eliminate these confounding factors.

Age at maturity can be used as an indicator of stock size and status; however, there are often other factors involved that can cause shifts in age at maturity. Environmental conditions can play a major role in age at maturity; warmer water temperatures may cause Striped Bass to reach maturity at a younger age (Setzler et al. 1980). Striped Bass in northern populations such as the Chesapeake and Hudson populations typically reach maturity between ages four and six compared to southern populations such as the Roanoke, Neuse, and Tar/Pamlico reaching

maturity between ages three and five. However, environmental conditions, stock sizes and statuses, and population dynamics vary throughout the Atlantic Coast; these factors could influence age at maturity. Most studies support the idea that early maturation is caused by a compensatory response to declining population sizes (Trippel 1995). Compensatory responses are density-dependent mechanisms that result when population sizes of a particular species decrease and result in less intraspecific competition between individuals. The decreased competition among individuals for resources results in faster growth rates that allow fish to develop faster and reach maturity at a younger age (Trippel 1995). The high levels of commercial and recreational exploitation, coupled with the young age at maturity of these stocks, could indicate this compensatory response is occurring in these systems.

When drawing conclusions about the status and health of a stock, it is important to have multiple years of age at maturity estimates to account for the confounding factors discussed earlier (Trippel 1995). A maturation study has never been conducted in the Neuse and Tar/Pamlico river system; therefore, it is impossible to determine if maturation schedules have changed over time. This study will serve as a baseline for fishery managers to compare changes in age at maturity over time and draw conclusions about the status and health of these stocks.

### *Fecundity*

A maturation and fecundity study has never been conducted for the CSMA Striped Bass population; therefore it is unclear if fecundity has changed over time. The linear relationship between fecundity and age, total length, and weight observed in this study have been reported in several other maturation and fecundity studies (Setzler et al. 1980; Olsen and Rulifson 1992; Boyd 2011). However, Striped Bass in the Neuse and Tar-Pamlico rivers produce more eggs at each age compared to the ASMA/RRMA (Figure 18). The mean fecundity at age could be

higher in the Neuse and Tar-Pamlico rivers compared to the ASMA/RRMA because the Neuse and Tar-Pamlico fish mature faster and are larger at age compared to the ASMA/RRMA Striped Bass.

#### *Hatchery vs. Wild origin*

Hatchery-reared Striped Bass have been unintentionally but fortuitously marked by a unique elemental signature that results from a fish being reared in the ENFH phase II ponds. This elemental signature is retained throughout the life of the fish and origin can be determined by analyzing otolith chemistry in the first year of life. The hatchery signal is characterized by high levels of Strontium (>4000 ppm) in the first year of life followed by a precipitous decrease in Strontium just before the first annulus (Figure 8). Fish of both hatchery and wild origin were collected in the main stem and upstream areas of the Tar/Pamlico and Neuse rivers using a variety of sampling gears. A total of 76% of the hatchery fish were collected upstream, and 24% were collected downstream. Interestingly, fish of wild origin were collected equally (50%) between upstream and downstream locations, but apparently there are so few wild fish in this system that any conclusions about the disparity in distribution between wild and hatchery fish would be speculative at best. The two groups exhibited no differences in length or weight; however, no wild fish from age 0-3 were collected, so comparisons of immature wild and hatchery fish were not possible. The lack of young wild fish in these systems is very alarming; it is possible that the wild fish are being overrun by the large number of stocked hatchery fish in the CSMA. More research is needed to determine if hatchery fish and wild fish are using similar or different habitats.

Identifying the hatchery signature made it possible to compare age and growth, condition, maturity, and fecundity between fish of wild and hatchery origin. Age at maturity could not be

compared by origin because no immature wild fish were collected. Age and length at 50% maturity was determined for the CSMA population; however, it was impossible to make comparisons between fish of hatchery and wild origin because no immature wild fish were collected. In spite of this problem, the wild fish collected were 100% mature at age 4 (n=3). The absence of immature wild fish from is alarming and most likely due to the low numbers of wild fish in the CSMA, with only 7.3% of fish sampled being of wild origin. The NCWRC and NCDMF also report few immature wild fish in juvenile sampling programs in the Tar-Pamlico and Neuse rivers; these findings support the assertion of few immature wild fish in the CSMA (Barwick et al. 2009; NCDENR 2011).

Condition, fecundity, and oocyte characteristics were compared by origin in the Neuse and Tar/Pamlico rivers. Physical characteristics were compared by origin to determine if a fish's rearing environment played a major role in physical development later in life. Mean eggs per gram and oocyte diameter varied significantly by origin, with wild origin fish producing oocytes of greater diameter and fewer oocytes per gram. Hatchery fish produced a mean of 3,808 eggs per gram and wild fish produced a mean of 2,512 eggs per gram. Mean weight of hatchery oocytes was 0.2626 mg per oocyte and wild oocytes weighed 0.3981 mg per oocyte. GSI was nearly significantly different ( $p=0.0514$ ). Oocytes of greater size and weight produced by the wild fish may explain why GSI values were nearly significantly different, but fecundity was not significantly different; gonads of wild fish would have to be larger in order to accommodate the larger and heavier eggs for reproduction. It can be concluded that wild Striped Bass produced heavier eggs by having fewer oocytes per gram. Bergey et al. (2003) found that Striped Bass oocyte diameter varied significantly between watersheds on the Atlantic Coast. Bergey et al. (2003) reported correlations of egg characteristics with the relative energy of the watershed: eggs

of heavier density were characteristic of relatively high-energy watersheds such as the Roanoke and Savannah rivers, while lower density eggs were found in tidally influenced watersheds of Chesapeake Bay. Reduced oocyte diameter and weight can be caused by environmental factors such as poor feeding or diet, adverse temperature, light and water quality (Hoar and Randall 1988). However, wild and hatchery fish occupied the same habitats during the same collection years and showed similar K factors and LSI values; therefore, it can be inferred that, at least in this system, environmental factors are not the causes of variability in oocyte size observed between adult wild and hatchery fish. Variation in egg size might reflect the conditions in which fish were reared, however, it is unclear if the effect of being reared in the ENFH during the first year of life would have effects later in life on size and weight of vtg3 oocytes. Perhaps a better explanation for variation in oocyte characteristics could be an artifact of wild fish producing oocytes that are genetically suited for the Tar/Pamlico and Neuse River watersheds, and hatchery (Roanoke broodstock parentage) fish producing oocytes that are best suited for the Roanoke River. Bergey et al. (2003) found that oocyte characteristics varied significantly among watersheds on the Atlantic coast and oocytes were suited for individual river systems. Ever since stocking began in the CSMA, Roanoke broodstock was used to stock the Tar/Pamlico, Neuse, and Cape Fear rivers. “Cross-stocking” (placement of young hatchery-reared fish into watersheds not native to the parental broodstock) was utilized as a management tool until 2012 when NCWRC first began using “endemic” broodstock in the CSMA. However, with the decades of stocking Roanoke broodstock into the CSMA it is highly unlikely that the original, “natural strain” of wild endemic fish are actually being used for hatchery production since 92.7% of fish in the CSMA originate from the hatchery; i.e., Roanoke broodstock parentage. Continuing to stock fish of Roanoke origin into the CSMA in hopes that “endemic” fish will

successfully spawn and reproduce is most likely ineffective; at this juncture it may be too late to rebuild natural stocks. The years and years of stocking non-native fish into these watersheds may have negatively affected the genetics of these stocks to reproduce successfully in these unique watersheds. The negative effects of stocking have been well documented on Salmonidae (Araki et al. 2008; Chittenden et al. 2010; Melnychuk et al. 2014), but fewer studies on the effects of stocking Moronidae are present in the literature (Patrick et al. 2006).

Fishery managers use fecundity estimates to make inferences about changes in population health over time. Since fecundity has never been estimated in the CSMA, we cannot make inferences about changes within the stock over time. However, comparisons can be made between fish that were naturally spawned in the CSMA, and hatchery released fish. Fecundity did not vary significantly by origin. However, wild origin fish produced eggs that were greater in size and weight compared to hatchery origin fish. Monteleone and Houde (1990) found that oocytes of greater size and weight tended to produce larger and more successful offspring compared to fish that produced smaller and lighter eggs. These larger and more successful offspring would presumably be better competitors, most likely increasing their chances of survival (Monteleone and Houde 1990). Even though fecundity was not significantly different by origin, it can be inferred that wild origin fish produced eggs of higher quality. Egg quality may be of little concern if there are not enough adult fish of wild origin left in the population to result in even a modest level of recruitment to forming year classes. State agency surveys indicate little evidence of recruitment of wild fish in the CSMA (Barwick et al. 2009; NCDENR 2011). Possible alternate explanations could be intraspecific competition between hatchery and wild fish in the juvenile phase, or other environmental factors causing the high mortality of eggs/larvae, resulting in unsuccessful recruitment from the larval to the adult stage.

The failure in recruitment first begins in the hatchery, where hatchery-reared Striped Bass are fed high protein pelletized diets and grow to much larger sizes compared to wild fish of the same age (Kevin Dockendorf, NCWRC, personal communication), which have to forage in the wild and are susceptible to many abiotic and biotic factors that cause high mortality in larval and juvenile fish. Chittenden et al. (2010) found that salmon that were reared in the hatchery, regardless of their genetics, were larger (length and weight) compared to their wild counterparts.

These larger hatchery fish are then released into the CSMA where they occupy similar ecological niches and outcompete their wild origin counterparts. Chittenden et al. (2010) found that hatchery-reared salmon were quicker to eat than wild fish before and after a simulated predation event. Hasegawa et al. (2014) found similar results on the effects on intraspecific competition between hatchery-reared and wild Chum Salmon (*Oncorhynchus keta*). Hasegawa et al. (2014) found that wild Chum Salmon decreased foraging rate with increased density of hatchery-reared Chum Salmon. Hasegawa et al. (2014) also found that wild Chum Salmon showed mass loss, even when densities of hatchery and wild fish were equal. The rearing environment and feeding methods of the hatchery may condition fish to be more voracious predators; this conditioning may persist later in life and cause hatchery fish to be better competitors for food compared to wild fish, which may be more cautious to feed.

The hatchery fish that out-compete juvenile wild fish in the Neuse and Tar-Pamlico rivers then survive to maturity where they attempt to spawn during the spawning season. The hatchery fish that arrive on the spawning grounds release their eggs in an attempt to reproduce, but the smaller eggs that hatchery fish release may not be fit to survive in the Tar/Pamlico and Neuse rivers. This could explain why spawning activity is occurring in the CSMA, but successful recruitment is not occurring.

*Tar/Neuse 2013 & 2014 vs. ASMA/RRMA 2009 & 2010*

The Tar-Pamlico and Neuse river populations of Striped Bass are listed as concerned in the latest fishery management plan (NCDENR 2011). The concerned listing in the latest fishery management plan (NCDENR 2011) is primarily due to the lack of data, high confidence intervals, and lack of precision for total mortality (Z). However, total mortality was described as excessive for the Tar/Pamlico and Neuse rivers (NCDENR 2011). Boyd (2011) conducted a maturation and fecundity study of the Albemarle/Roanoke stock in 2009 and 2010; during this study the stock was listed as recovered. Methodologies used by Boyd (2011) were employed in this study to minimize any variability in fecundity and maturity estimates that could result from using different methods; direct and valid comparisons of results were then made between the Tar/Neuse (concerned) and Albemarle/Roanoke (recovered) populations. Maturity, fecundity, and growth rates were compared between the CSMA and ASMA/RRMA to make inferences about the status of the Tar-Pamlico and Neuse river Striped Bass populations.

CSMA fish were found to be 50% mature 0.5 years earlier than the ASMA/RRMA stock. A shift in life history traits such as age at maturity has been attributed to high exploitation rates in similar studies (Trippel 1995). The younger age at maturity caused by high exploitation rates in the CSMA is an indicator that the fishery is stressed. High total mortality in the CSMA may be having detrimental effects on the stock, and are most likely inhibiting the ability of the stock to rebuild. In order for fishery managers to effectively manage the CSMA Striped Bass stock, high total mortality must be addressed and mitigated. Greater length at maturity in the CSMA may also pose a problem for fishery managers to rebuild this stock by allowing fish that were previously thought to be mature to be harvested, when in fact they are immature. Currently, Striped Bass in the CSMA can first be harvested at 457.2 mm TL (18 inches TL); this study

found that female Striped Bass were 100% mature at 537.33 mm TL (21.12 inches TL). Fishery managers must account for this shift in maturation in order to set appropriate minimum size limits to effectively manage these populations.

Fecundity estimates from the CSMA and ASMA/RRMA populations were compared to determine differences in fecundity between a stock listed as concerned (CSMA) and a healthy/recovered stock (ASMA/RRMA). Stressed or reduced populations of fish typically produce more eggs at age and length compared to populations that are not stressed (Trippel 1995); this phenomenon has been shown in studies of Plaice (*Pleuronectes platessa*), Lake Trout (*Salvelinus namaycush*), and Lake Whitefish (*Coregonus clupeaformis*) (Trippel 1995). The Central Southern Management Area female Striped Bass produced more eggs at age compared to the ASMA/RRMA population, and therefore would be considered a stressed or reduced population using Trippel's (1995) criteria.

Growth rates in the CSMA were faster compared to the ASMA/RRMA. Faster growth rates in CSMA compared to the ASMA/RRMA could indicate the stock is stressed and has been negatively affected by high total mortality rates in the CSMA. Fish grow faster in stressed or reduced populations by having less intraspecific competition for resources; this phenomenon is explained by Trippel (1995) and Hasegawa (2014) as compensatory response caused by density dependent growth. However, the best explanation for faster growth rates in the CSMA could be attributed to the majority of CSMA fish being reared in the Edenton National Fish Hatchery under optimal feeding and environmental conditions. Previous studies by NCWRC staff confirmed these results of CSMA fish having greater lengths at age 2 and 3 compared to fish in the Roanoke River (Dycus et al. 2014; McCargo and Dockendorf 2014). Chittenden et al. (2010) reported similar results of faster growth rates in hatchery-reared salmon.

### *Conclusions and Recommendations*

The Central Southern Management Area Striped Bass population exhibits all the characteristics of a stressed stock. Early age at maturity, truncated age classes, increased fecundity at age, faster growth rates, and greater length at maturity are hallmark characteristics of a stressed stock. Individually, these factors could be considered as natural variation, but when all of these factors are found in the same population it becomes clear that Striped Bass stocks of the Central Southern Management Area are stressed and/or reduced. Managers of the Central Southern Management Area Striped Bass populations could address the high levels of fishing mortality and/or natural mortality that may be causing this population to be stressed.

Fishery managers in North Carolina work to manage coastal Striped Bass populations so fish can enter the spawning population before being harvested. Current management regulations for recreational fishermen require Striped Bass in the Tar/Pamlico and Neuse river coastal waters to be a minimum of 18 inches TL; in joint and inland waters Striped Bass must be a minimum of 18 inches TL, and no fish between 22 and 27 inches may be harvested. Minimum size limits for commercial fishermen in the Tar/Pamlico and Neuse river coastal waters are currently 18 inches TL with no protective slot limit. Results of this study indicate that female Striped Bass in the Tar/Pamlico and Neuse rivers reach a length at 50% maturity of 467.84 mm TL (18.42 inches TL) and are estimated to be 100% mature at 537.33 mm TL (21.12 inches TL). To ensure all fish have the opportunity to enter the spawning population at least once, the length at 100% maturity (21 inches TL) could be used as the minimum harvestable size limit.

Mean oocyte diameter and mean eggs per gram (oocyte weight) were significantly different between hatchery and wild fish, with wild fish producing oocytes that were larger and heavier. Larger and heavier oocytes produced by wild fish may be better suited for the Tar-

Pamlico and Neuse rivers compared to the smaller and lighter oocytes produced by hatchery origin (Roanoke parentage) fish. The differences in oocyte characteristics may be one explanation for why spawning activity is occurring in the CSMA but little to no recruitment has been observed. However, there may be other factors involved for the unsuccessful recruitment in the CSMA. It is unclear what is driving the differences in oocyte size between hatchery and wild fish. Genetic differences or differences in the rearing environment may be causing the differences in oocyte diameter. More research is needed to determine the role that oocyte size plays in successful recruitment of the CSMA Striped Bass population.

The high percentage (92.7%) of hatchery origin Striped Bass in the CSMA is a cause for concern for fishery managers and researchers. The high percentage of hatchery fish in the population indicates that little successful recruitment is occurring in these river systems. Currently, 100,000 phase II Striped Bass are stocked in each river system in the CSMA annually. The phase II fish stocked in these river systems reach greater lengths compared to their wild counterparts of the same age and may be outcompeting the wild spawned fish, and therefore decreasing the likelihood of survival for wild fish. Large numbers of stocked fish may be overrunning the natural population of wild fish and effectively preventing the recovery of the CSMA Striped Bass stock. Some studies on salmonids show that survivability increases when salmon are stocked as parr (1 year old) rather than smolts (2 years old) (Jokikokko et al. 2006). Stocking Striped Bass as phase I rather than phase II may allow fish to be better suited to the environment in the CSMA. More research is needed to look at the effects of being stocked as phase I rather than phase II fish. ENFH should continue to rear Striped Bass in the Castle Hayne aquifer water to serve as a cheap and easy tag for hatchery fish. Continuing to use the high

strontium waters of the Castle Hayne aquifer will allow researchers using otolith chemistry to track the wild and hatchery-reared populations over time.

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## Tables

Table 1: Number of female Striped Bass collected in each river system for the 2013 and 2014 sampling seasons.

Size Class (mm)	River			
	Neuse		Tar/Pamlico	
	2013	2014	2013	2014
200-224	0	1	0	0
225-249	0	2	0	0
250-274	0	0	1	0
275-299	0	0	0	2
300-324	0	0	0	0
325-349	0	0	0	0
350-374	0	0	0	0
375-399	0	0	1	1
400-424	0	0	0	3
425-449	0	0	0	3
450-474	1	0	0	2
475-499	0	0	1	14
500-524	5	2	0	12
525-549	5	7	3	13
550-574	6	7	1	4
575-599	4	2	0	1
600-624	8	4	3	5
625-649	2	8	3	1
650-674	1	1	2	3
675-699	0	0	2	3
700-724	1	0	0	2
725-749	2	1	0	1
750-774	0	0	0	2
775-799	0	0	0	1
800-824	0	0	0	0
825-849	0	0	0	0
850-874	0	0	0	0
875-899	0	1	0	0
900-924	0	1	0	1
925-949	0	2	0	0
<b>Total</b>	<b>35</b>	<b>39</b>	<b>17</b>	<b>74</b>

Table 2: Microscopic definitions of the female reproductive cycle described by Brown-Peterson et al. (2011). The terminology listed was used to determine the reproductive phase of female Striped Bass.

Phase	Microscopic
Immature	Only oogonia and PG oocytes present. No atresia or muscle bundles. Thin ovarian wall with little space between oocytes.
Developing	PG, CA, Vtg1, and Vtg2 oocytes present. No evidence of POFs or Vtg3 oocytes.
Spawning Capable	Vtg3 oocytes present. Atresia of Vitellogenic and/or hydrated oocytes may be present. Early Stages of OM can be present.
Actively Spawning	Oocytes undergoing late GVM, GVBD, hydration or ovulation.
Regressing	Atresia and POFs present. Some CA and/or Vitellogenic oocytes present.
Regenerating	Muscle bundles, enlarged blood vessels, thick ovarian wall, degenerating POFs may be present.

Table 3: Mean, standard error, and range by age for total length (TL mm) and weight (g) of female Striped Bass sampled (n=165) from the Neuse and Tar/Pamlico rivers from 2013 and 2014.

	Age	N	Mean	SE	Range
TL	1	6	253.33	13.76	217-299
	2	11	431.09	10.69	382-491
	3	57	520.35	3.34	462-574
	4	47	577.13	4.94	515-632
	5	21	642.1	5.64	586-693
	6	4	667.25	19.33	615-706
	7	6	693.67	23.74	613-748
	8	4	709.25	11.1	690-734
	9	6	800.83	31.74	715-912
	10	3	930.33	10.27	910-943
Weight	1	6	166.58	26.05	109.2-258
	2	11	866.06	71.82	582-1400
	3	57	1561.99	35.24	941.7-2155
	4	47	2168.11	56.49	1421.63-3042
	5	21	3094.84	102.91	2255-4200
	6	4	3262.39	299.73	2536.1-4000
	7	6	3770.92	396.14	2460.5-5000
	8	4	3817.59	208.48	3574.37-4441
	9	6	5716.53	783.91	3500-8160
	10	3	8842	382.8	8302-9582

Table 4: LSI, GSI, and K factor calculated for the Tar/Pamlico (n=91) and Neuse (n=74) rivers.

	Neuse				Tar/Pamlico			
	N	Mean	SE	Range	N	Mean	SE	Range
LSI	74	1.27	0.03	0.62-1.95	91	1.34	0.03	0.44-2.40
GSI	74	7.7	0.65	0.05-23.13	91	6.01	0.57	0.03-17.53
K factor	74	1.11	0.01	0.86-1.33	91	1.1	0.01	0.89-1.35

# Figures

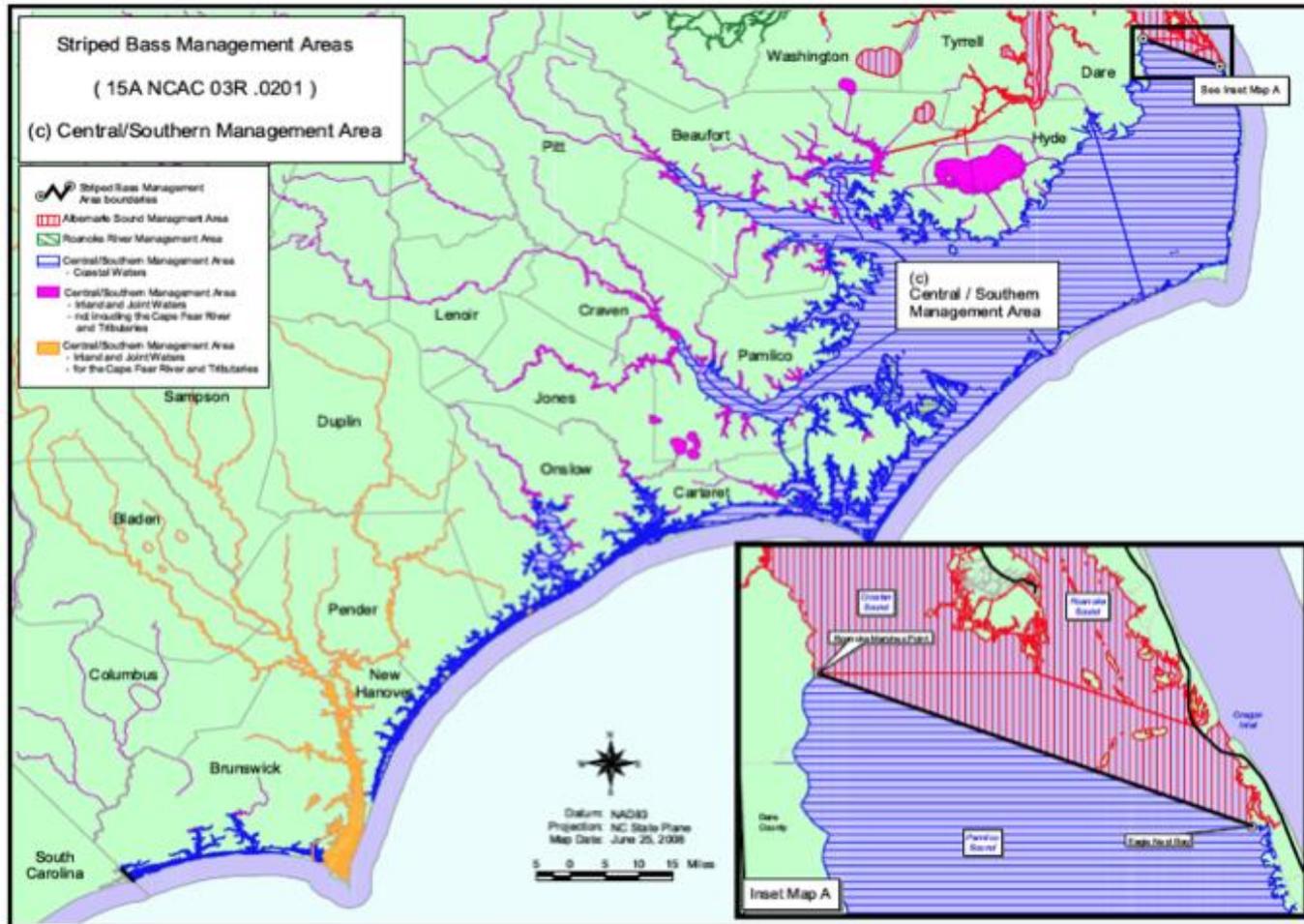


Figure 1: Map of the Central Southern Management Area. (NCDENR 2011)

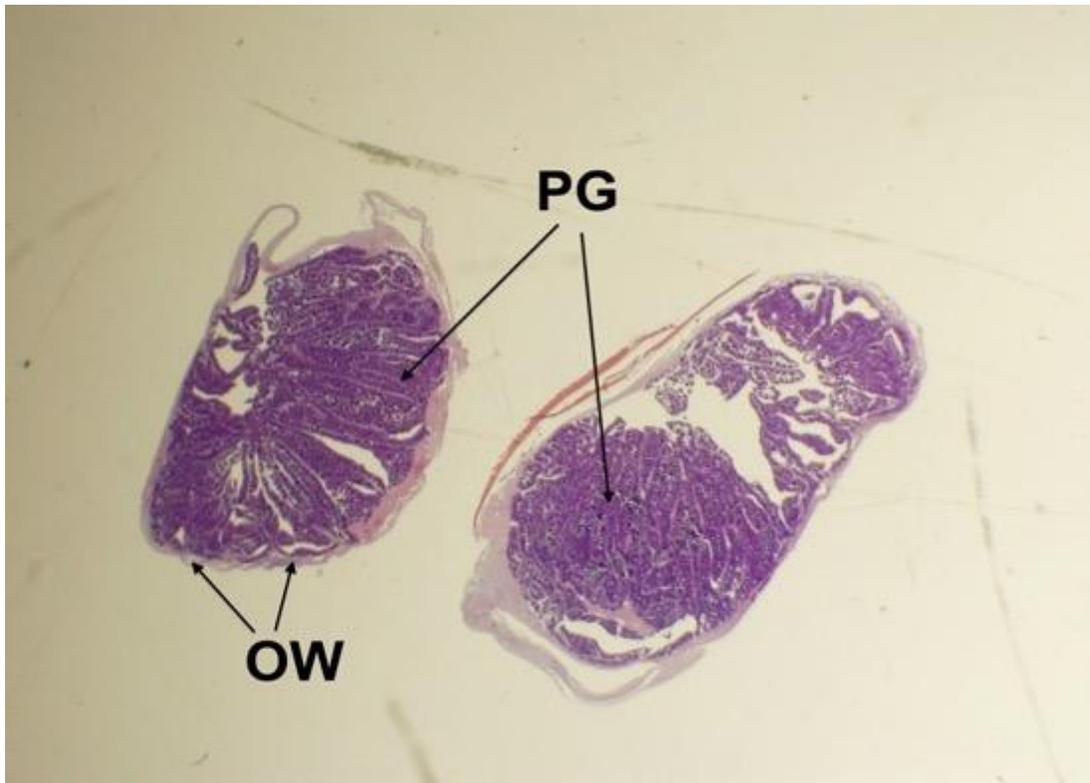


Figure 2: Gonads in the immature phase of the female reproductive cycle. Only primary growth oocytes (PG) are present in the immature phase. Gonads have thin ovarian walls (OW) in this reproductive phase.

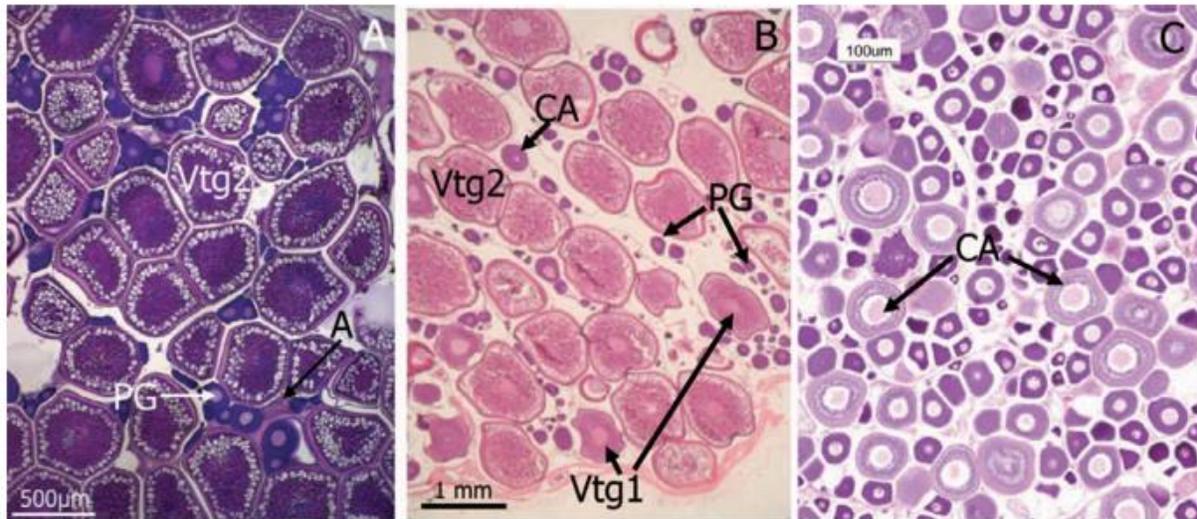


Figure 3: Gonads in the developing phase can have primary growth (PG) oocytes, first stage vitellogenic oocytes (Vtg1), secondary vitellogenic oocytes (Vtg2), and cortical alveoli (CA). No tertiary vitellogenic oocytes are present in the developing phase (taken from Brown-Peterson et al. 2011).

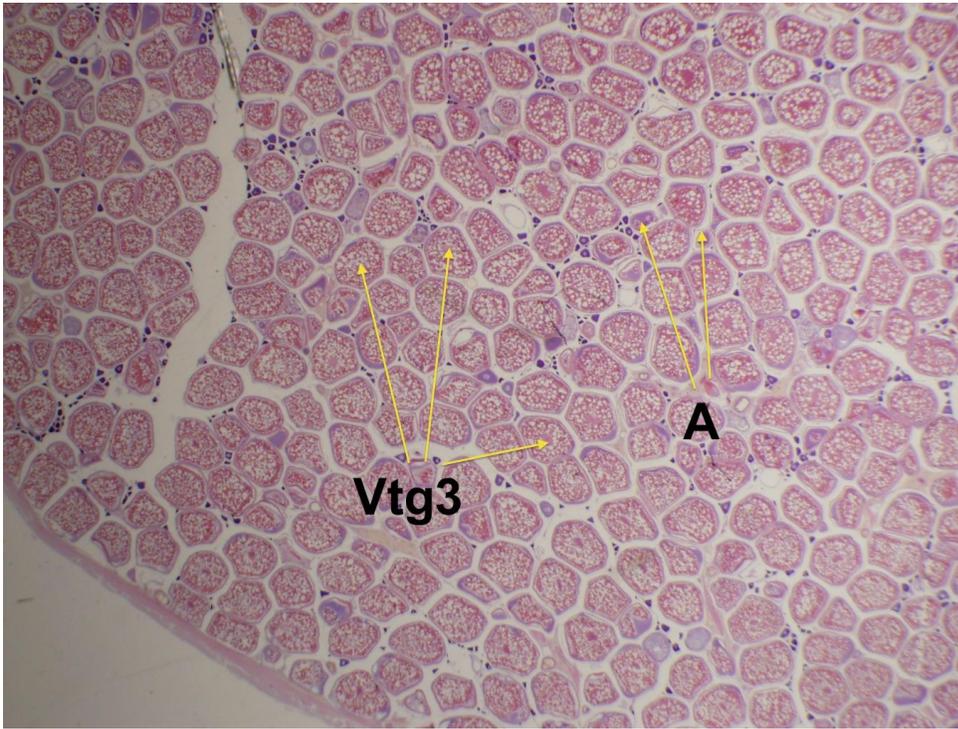


Figure 4: Female Striped Bass in the spawning capable reproductive phase. Tertiary vitellogenic oocytes (Vtg3), atresia (A), and primary growth (PG) oocytes are present in the spawning capable phase, and should spawn in the imminent season.

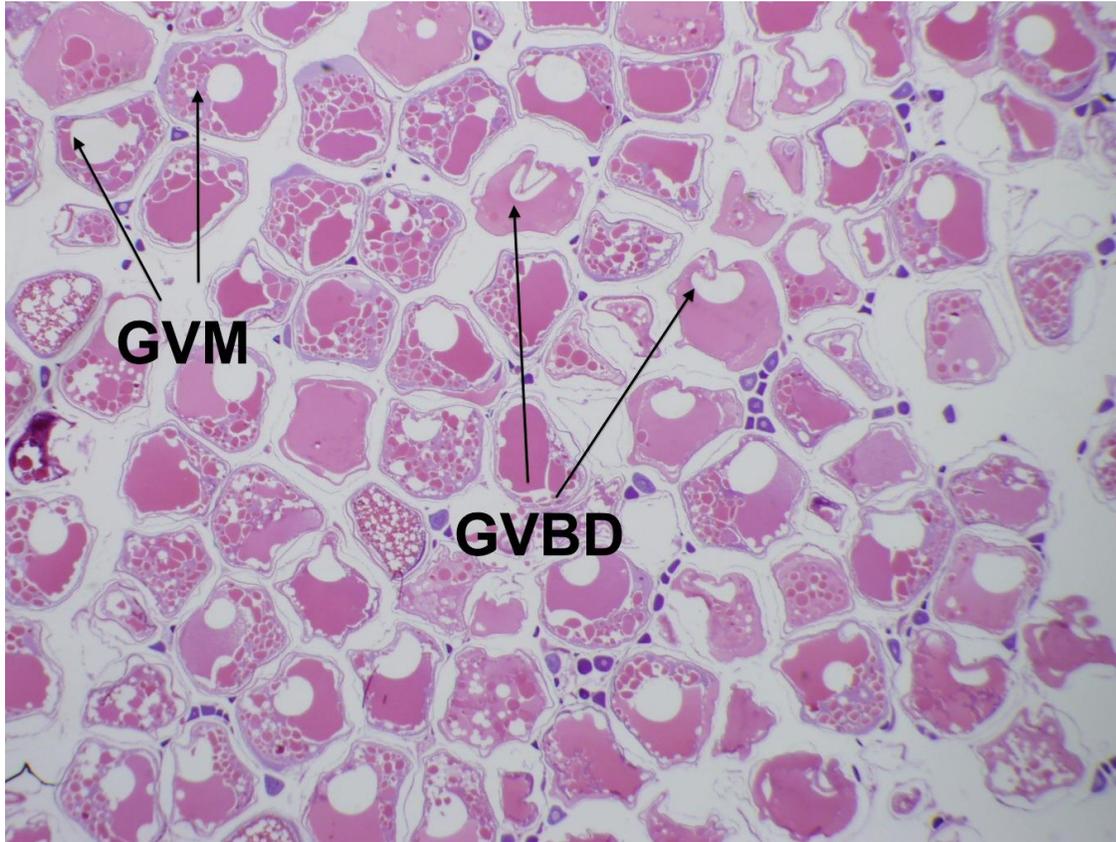


Figure 5: Female Striped Bass in the actively spawning phase have primary growth (PG) oocytes, late germinal vesicle migration (GVM), germinal vesicle breakdown (GVBD), and hydration.

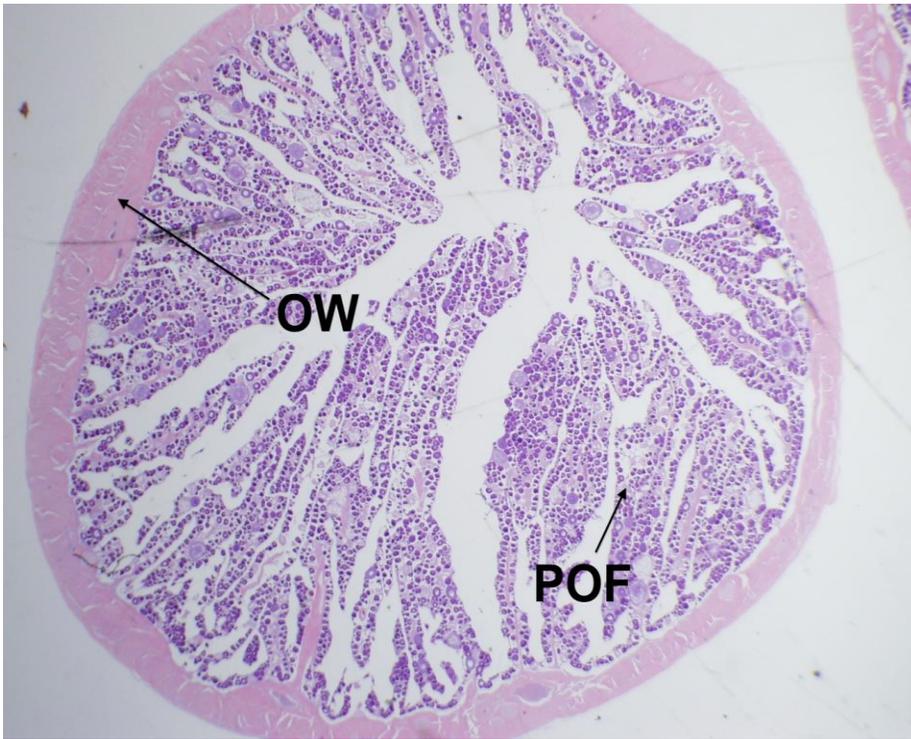


Figure 6: Female Striped Bass in the regressing phase have a thick ovarian wall (OW), post ovulatory follicles (POF), and primary growth oocytes (PG).

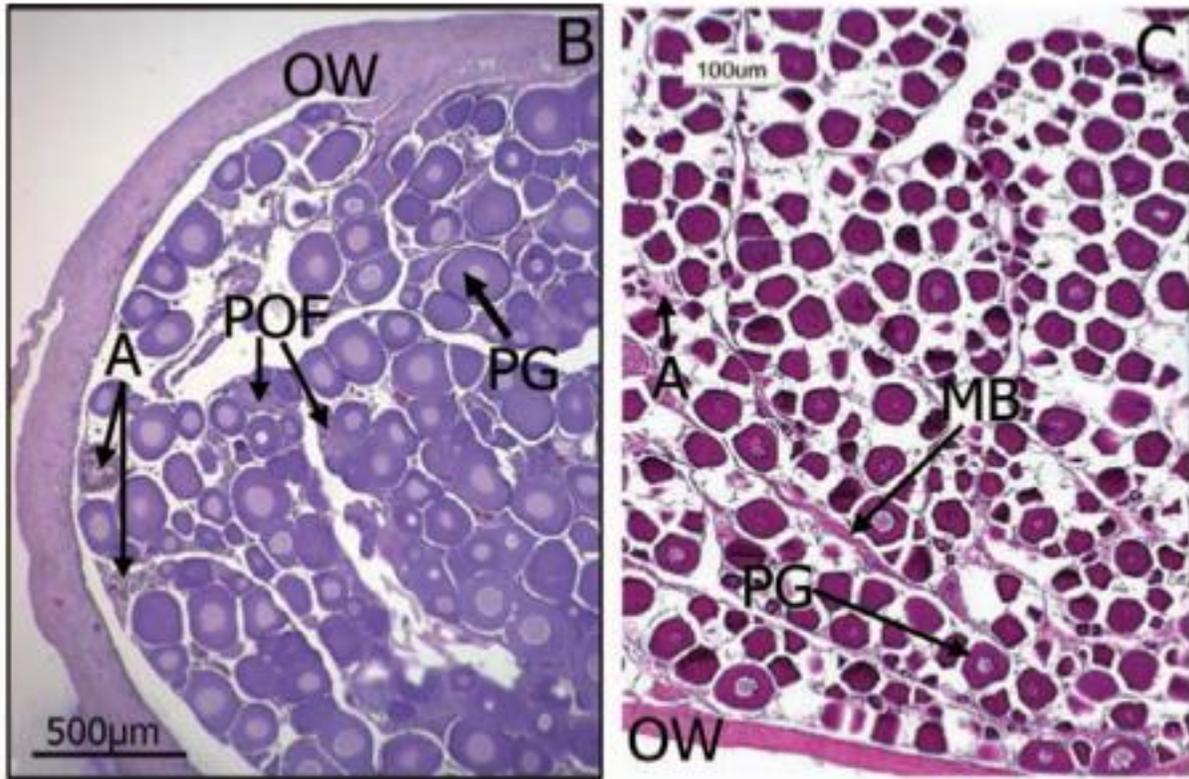


Figure 7: Female Striped Bass in the regenerating phase have a thick ovarian wall (OW), primary growth oocytes (PG), muscle bundles, and some atresia. Fish in the regenerating phase are sexually mature but reproductively inactive (taken from Brown-Peterson et al. 2011).

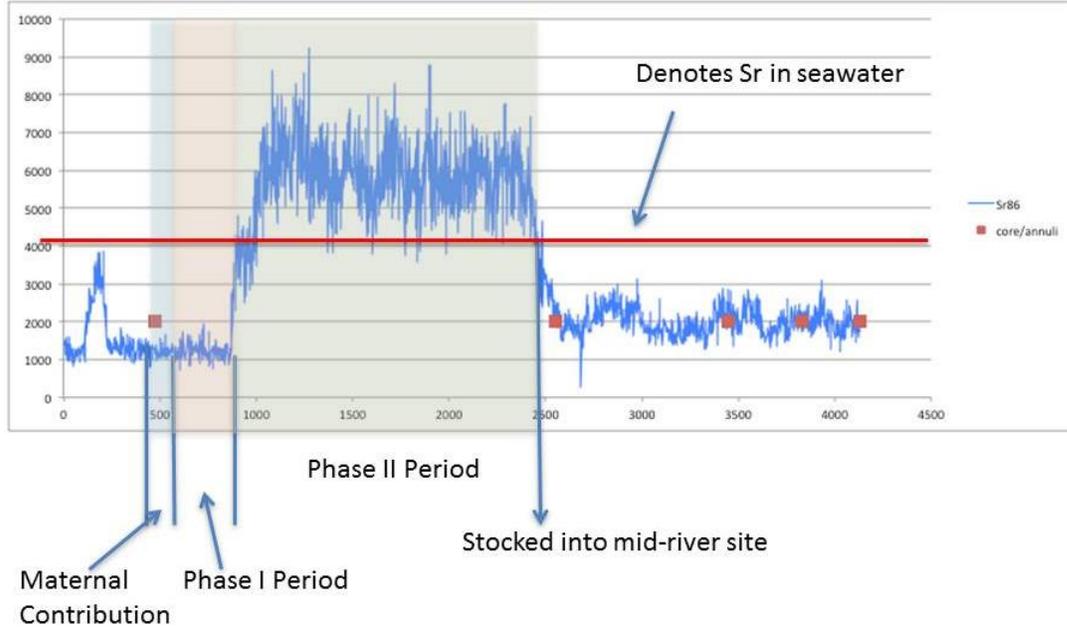


Figure 8: Prototypical hatchery fish strontium (Sr) signature, for comparison red squares represent otolith focus (farthest left) and annuli, left shaded area represents maternal contribution period, middle shaded area represents phase I period, right shaded area represents phase II period, and black line represents stocking event. The x axis represents distance scanned across the otolith, the y axis represents concentrations in parts per million of Sr.

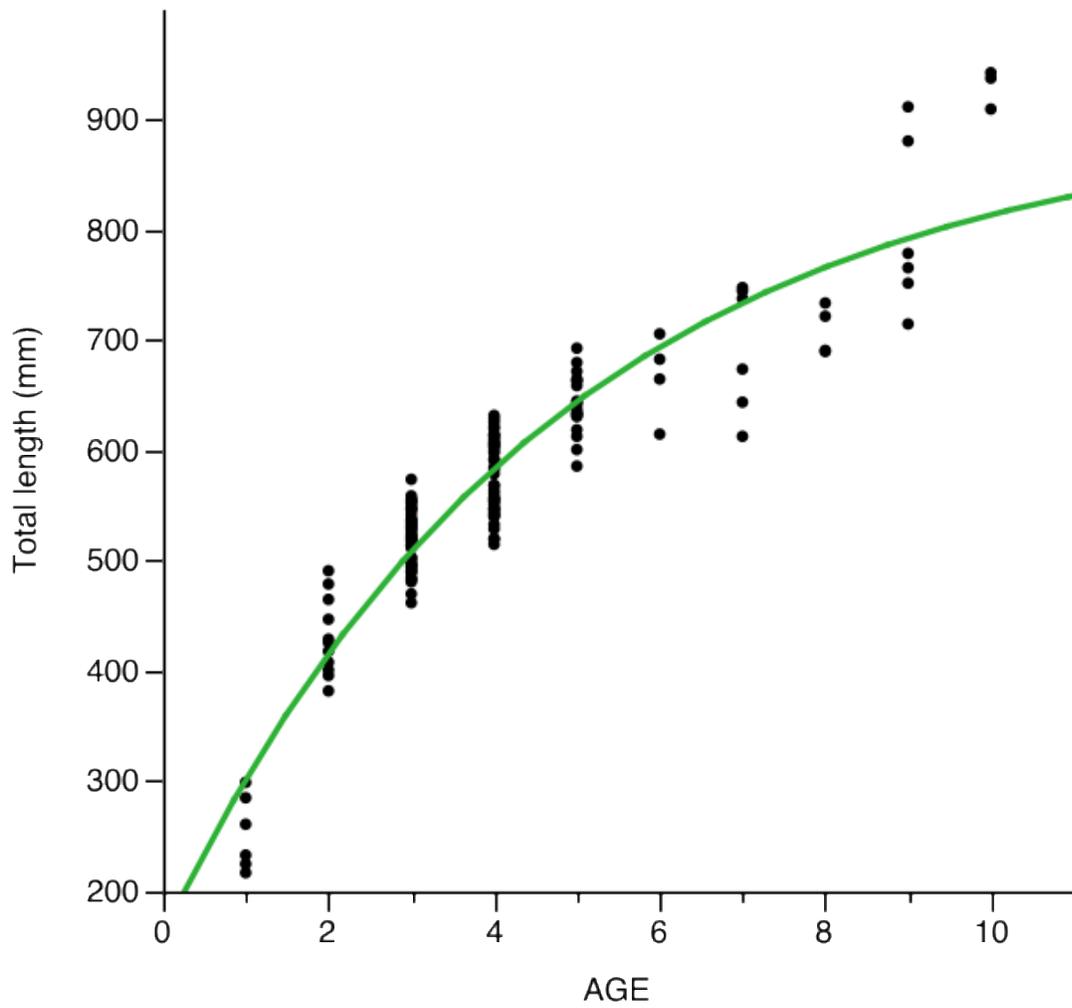


Figure 9: Logistic regression model was used to fit length at age data of female Striped Bass (n=165) to produce the Von Bertalanffy growth model ( $L_t = 905.171(1 - e^{-0.21015(t - 0.92704)})$ ).

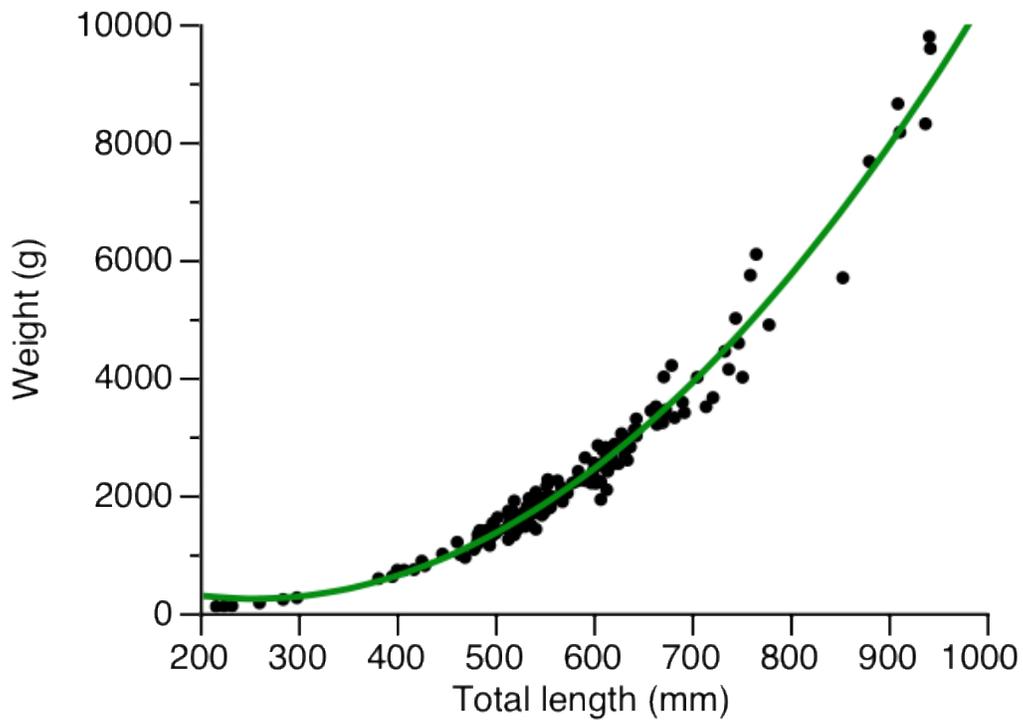


Figure 10: Weight-Length growth model of female Striped Bass for the Tar/Neuse Striped Bass population (n=165) estimated using the formula ( $Weight (g) = -4519.463 + 11.55939 * Total length (mm) + 0.0184272 * (Total length (mm) - 568.455)^2$ ).

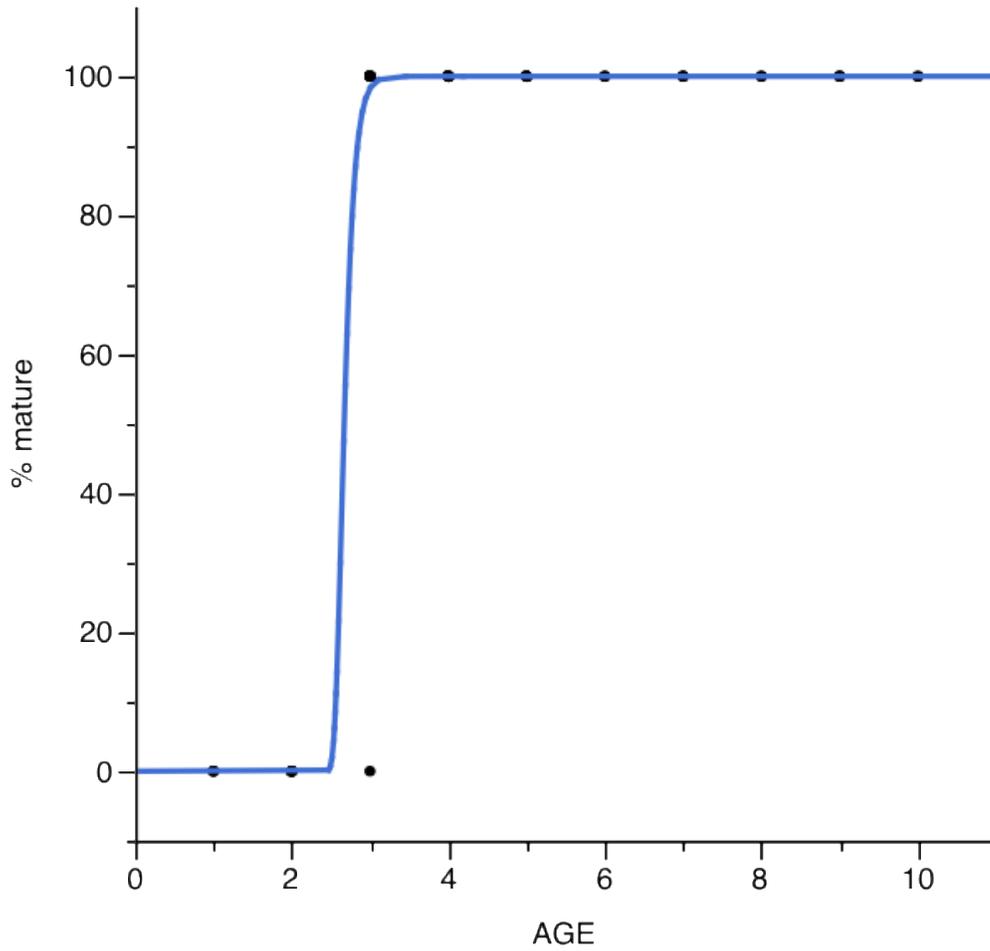


Figure 11: Age at maturity of female Striped Bass estimated using a logistic regression model for the Tar/Neuse Striped Bass population (n=165) was 2.67 years.

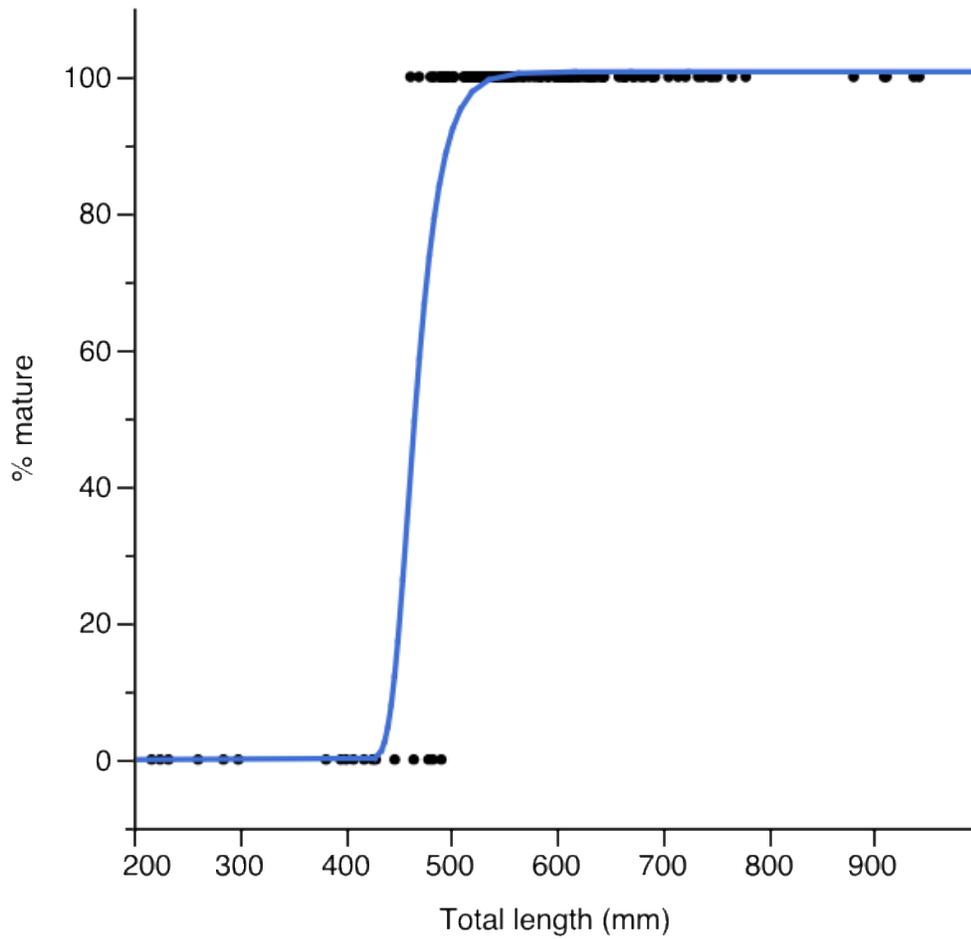


Figure 12: Length at maturity for the Tar/Pamlico and Neuse rivers Striped Bass population. Total length (mm) at 50% maturity estimated using a logistic regression model for female Tar/Pamlico and Neuse Striped Bass (n=165) was 467.84 mm TL (18.42 inches TL). Fish were estimated to be 100% mature at 537.33 mm TL (21.12 inches TL).

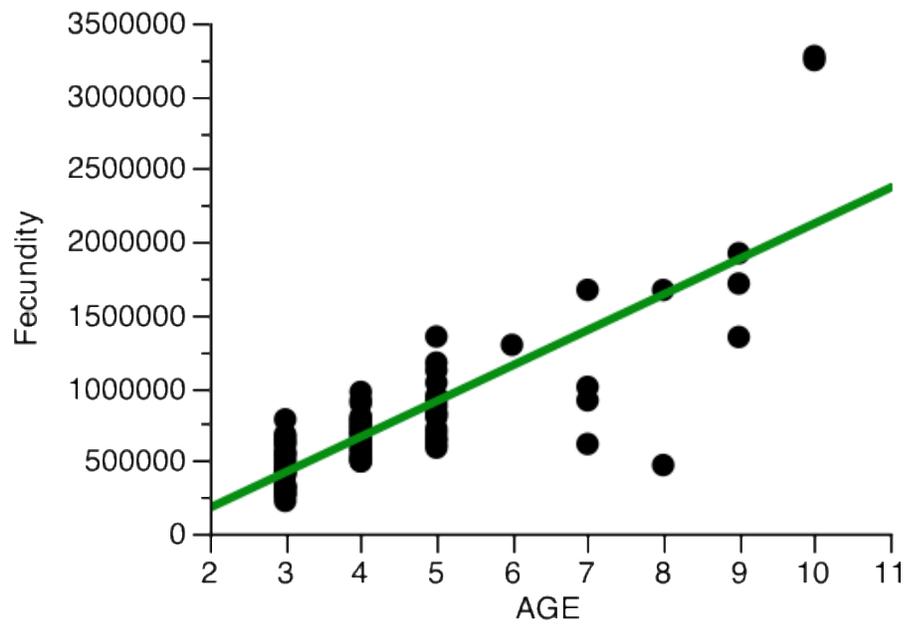


Figure 13: The relationship between age and fecundity was linear ( $R^2= 0.66734$  ;  $n=87$ ) and was estimated using the formula ( $Fecundity = -312599.8 + 243791.21*AGE$ )

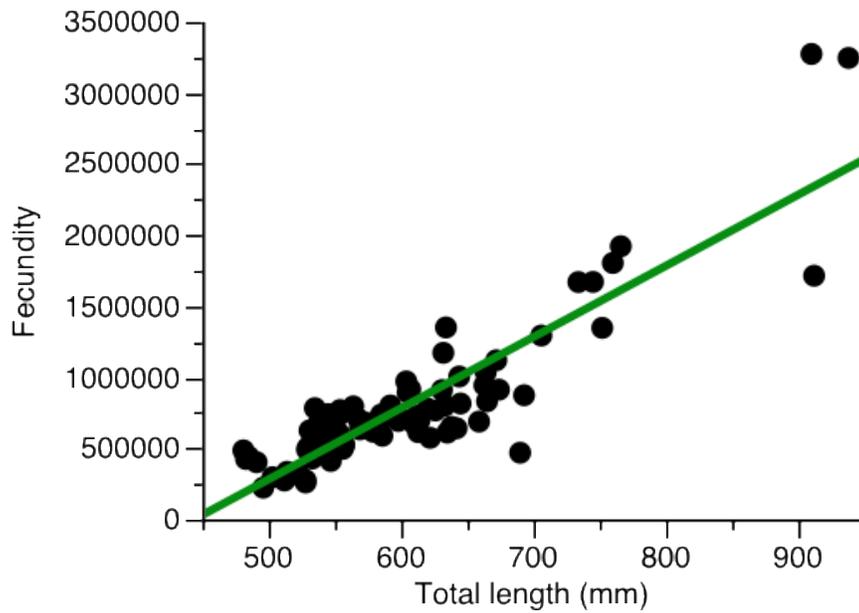


Figure 14: The relationship between total length (mm) and fecundity was linear ( $R^2=0.777$  ;  $n=87$ ) and was estimated using the formula ( $Fecundity = -2221005 + 5004.7067 * Total\ length\ (mm)$ ).

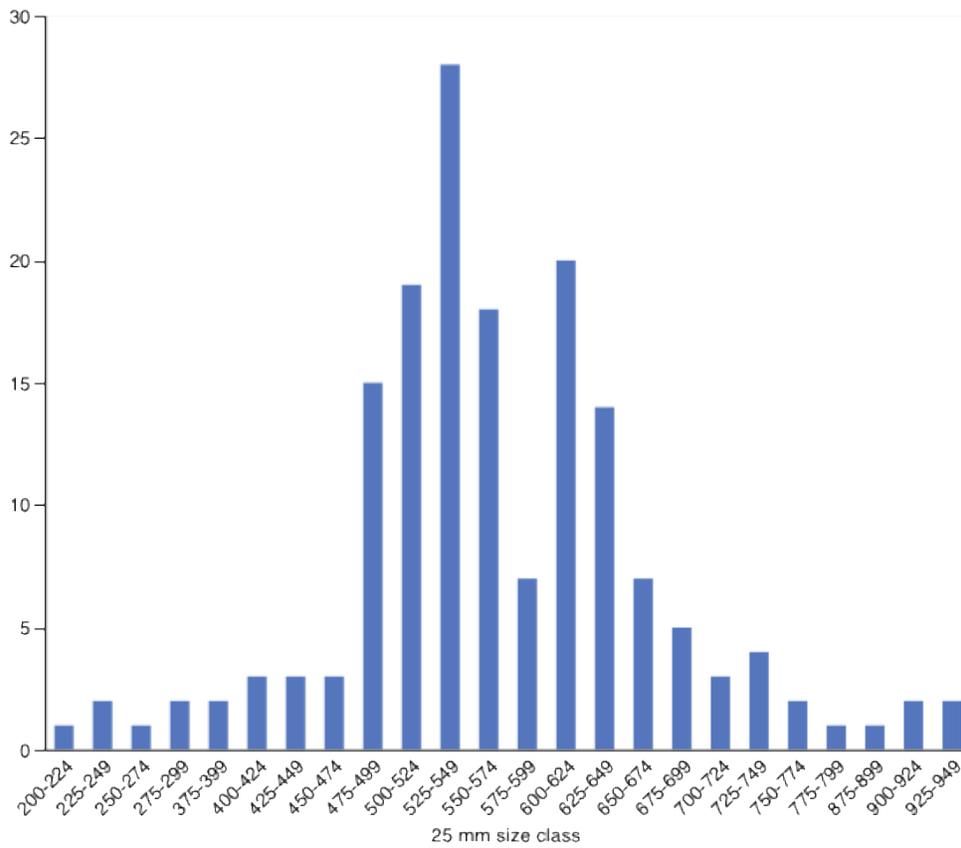


Figure 15: Frequency of female Striped Bass collected by 25 mm TL size classes.

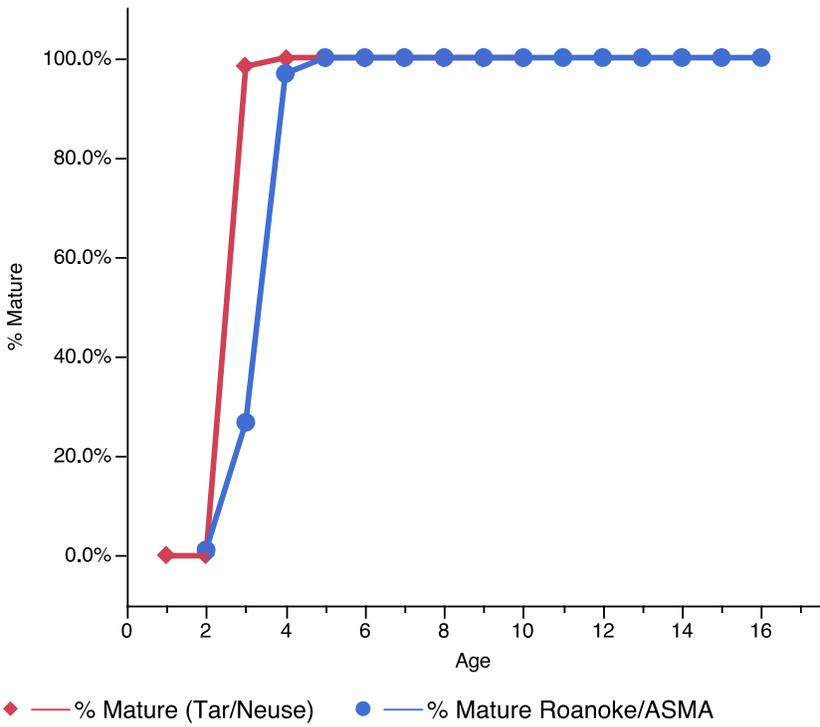


Figure 16: Overlay plot of estimated age at maturity for the Tar/Pamlico and Neuse (n=165) and Roanoke/ASMA (n=397) populations.

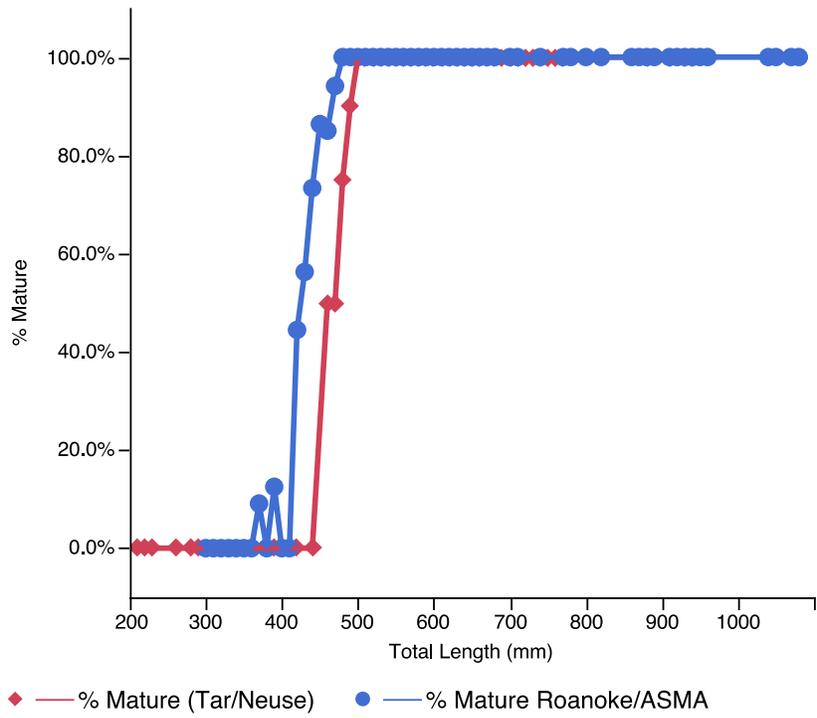


Figure 17: Overlay plot of estimated total length (mm) at maturity for the Tar/Pamlico and Neuse (n=165) and Roanoke/ASMA (n=391) populations.

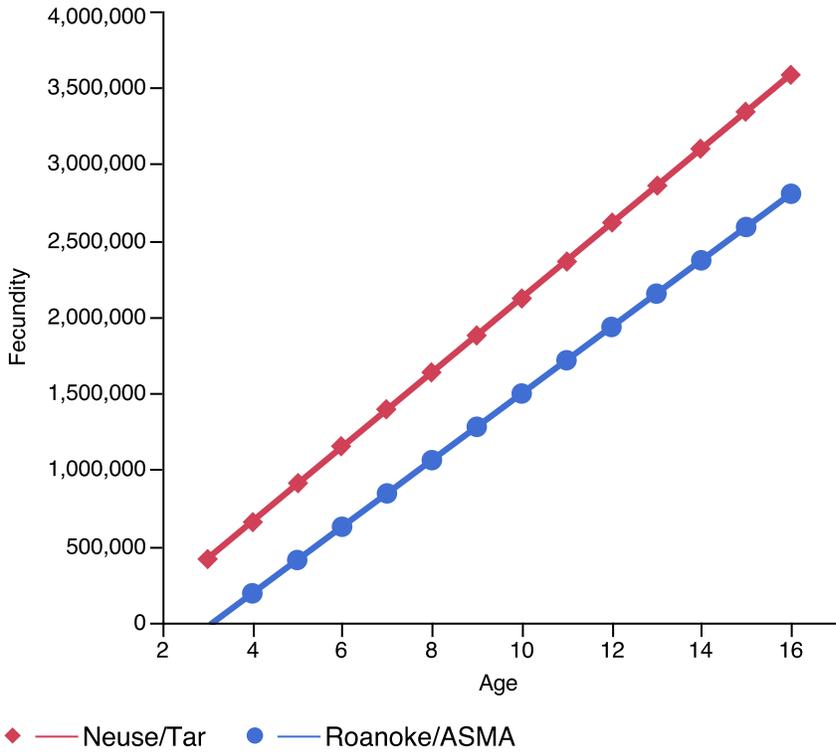


Figure 18: Overlay plot of estimated fecundity at age between the Neuse and Tar/Pamlico (n=87) and Roanoke/ASMA (n=80) populations.

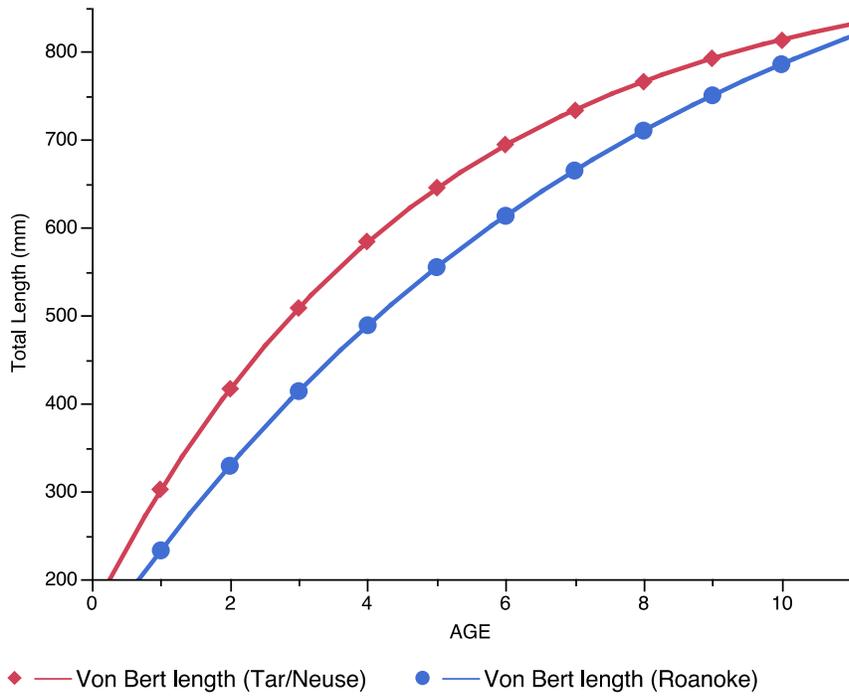


Figure 19: Overlay plot of predicted length-at-age using a Von Bertalanffy growth model for the Tar/Pamlico and Neuse (n=165) and Roanoke/ASMA (n=436) populations.

## Appendix



**Animal Care and  
Use Committee**

212 Ed Warren Life  
Sciences Building  
East Carolina University  
Greenville, NC 27834

252-744-2436 office  
252-744-2355 fax

June 12, 2013

North Carolina Wildlife  
Resources Commission

Dear Sir or Madam:

The vertebrate animal use described in the following application submitted to the North Carolina Wildlife Resources Commission was reviewed and is congruent with an IACUC-approved animal use protocol:

Title of Application: "Maturation and Fecundity of the CSMA Striped Bass Population:  
Is Hypoxia Causing Recruitment Failure?"

Name of Principal Investigator: Roger Rulifson, Ph.D./Evan Knight

Name of Institution: East Carolina University

Congruency Approval Date : June 12, 2013

Animal Use Protocol Expiration Date: (D291) June 12, 2016

This institution is fully accredited by AAALAC and has an Animal Welfare Assurance on file with the Office of Laboratory Animal Welfare. The Assurance Number is A3469-01.

Sincerely yours,

A handwritten signature in black ink that reads 'S. B. McRae'.

Susan McRae, Ph.D.  
Chair, Animal Care and Use Committee

SM/jd

cc: ECU Office of Sponsored Programs



**Animal Care and  
Use Committee**

212 Ed Warren Life  
Sciences Building  
East Carolina University  
Greenville, NC 27834

252-744-2436 office  
252-744-2355 fax

June 12, 2013

Roger Rulifson, Ph.D.  
Department of ICSP/Biology  
Flanagan Building  
East Carolina University

Dear Dr. Rulifson:

Your Animal Use Protocol entitled, "Maturation and Fecundity of CSMA Striped Bass (*Morone Saxatilis*)" (AUP #D291) was reviewed by this institution's Animal Care and Use Committee on 6/12/13. 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 black ink that reads 'S. B. McRae'.

Susan McRae, Ph.D.  
Chair, Animal Care and Use Committee

SM/jd

enclosure