

Statewide Research - Freshwater Fisheries

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### Scientific Names

White catfish .....	<i>Ameirus catus</i>
Brown bullhead .....	<i>A. nebulosus</i>
Channel catfish.....	<i>Ictalurus punctatus</i>
Flathead catfish .....	<i>Pylodictis olivaris</i>
Redbreast sunfish .....	<i>L. auritus</i>
Bluegill.....	<i>Lepomis macrochirus</i>
Redear .....	<i>L. microlophus</i>
Redeye Bass .....	<i>Micropterus coosae</i>
Smallmouth Bass .....	<i>M. dolomieu</i>
Largemouth bass .....	<i>M. salmoides</i>

## JOB PROGRESS REPORT

STATE: South Carolina

PROJECT NO: F-63

PROJECT TITLE: Fisheries Investigations in Lakes and Streams - Statewide

STUDY: Survey and Inventory  
surveys -

STUDY TITLE: Fishery

Statewide Fishery Research

JOB NO.: I  
and Growth of Flathead

JOB TITLE: Age  
Catfish

PERIOD COVERED: July 01, 1997 - June 30, 1998

### Introduction

Flathead catfish *Pylodictis olivaris* were introduced into South Carolina in 1964, when they were stocked into the Santee-Cooper Lake system (Lakes Marion and Moultrie) and Lake Thurmond. Flatheads have now spread to other systems. Thomason et al. (1993) first documented flathead catfish in the Edisto River in a 1988-90 survey. Allen and Thomason, (1996) noted a dramatic increase in the average size and abundance of flathead catfish in the Edisto River in 1994; fish of 30 to 50 pounds were common. Crochet and Sample (1996) conducted a survey in 1995 that confirmed the presence of substantial numbers of flathead catfish in the Great Pee Dee River.

When flathead catfish initially invade a new system, they can expand quickly and affect the existing fish community. In the Altamaha River, Georgia, Thomas (1993) documented a marked increase in the flathead catfish population with a corresponding decrease in bullheads *Ameiurus* sp.

Hard parts of catfish can provide reasonable estimates of the age and growth.



Sneed (1951) and Marzolf (1955) used the pectoral spine to determine the growth of channel catfish *Ictalurus punctatus*; these authors sectioned the spines at the distal end of the basal recess. Turner (1982) determined that sectioning the pectoral spines of flathead catfish at the distal end of the basal recess would result in age underestimations, due to the loss of annuli caused by growth of the spine's lumen; he recommended obtaining sections from the articulating process of the spine to avoid loss of annuli. Guier et al. (1981) and Quinn (1988), used pectoral spines and reported a faster than average growth rate of flathead catfish in newly invaded coastal river systems of the southeastern United States (i.e. the Cape Fear River, NC, and the Flint River, GA, respectively). Crumpton et al. (1985) determined that otoliths did not provide acceptable estimates of age and growth in three Ictaluridae, *Ameiurus nebulosus*, *A. catus*, and *Ictalurus punctatus*.

The objective of this study was to use pectoral spines to compare growth rates of flathead catfish in three distinct areas of South Carolina. Also, a small sample of otoliths was aged to determine their potential use in age and growth studies of flathead catfish. Results will provide a baseline for biological monitoring and help determine appropriate management and sampling strategies.

## **Methods**

Pectoral spines of flathead catfish were obtained from completed or ongoing survey efforts conducted in Little River and Long Cane Creek (tributaries of Lake Thurmond), Edisto, and Great Pee Dee River. Total length, weight, and date of capture of catfish were recorded for each collection. Spines had been removed in a variety of ways and some did not have the articulating process in place. Therefore, a decision was made to section all spines as close as possible to the distal end of the basal recess. Upon receipt of spines, they were dried for four to 10 hours in a

50°C. oven to facilitate removal of flesh. Spines were sectioned in approximately 2 mm slices with an Isomet saw. The first section was made as close as possible to the distal end of the basal recess; succeeding sections were removed from more distal areas of the spine. The distal side of the section was attached to a microscope slide with liquified (by prior heating) thermoplastic cement. Date of collection and a unique identification number were placed on each slide with a permanent marker. The proximal end of the spine section was then lightly sanded with 600 grit sandpaper to remove saw marks. A compound microscope was used to locate annuli.

Number of annuli was independently estimated by two trained readers. If disagreement occurred, a conference was held to attempt to reconcile differences. After this process was completed, a third reader interpreted the number of annuli. When disagreement existed, a final, estimate of age was reached, if possible, through a conference.

Once a final estimate of the number of annuli was obtained, the diameter of the lumen, each annulus, and the end of the spine was measured in both the dorsal-ventral (DV) and anterior-posterior (AP) plane using an ocular micrometer. A mean diameter was calculated by averaging the AP and DV diameters. Linear regression analysis was performed to assess the correlation between:

- a) total length (dependent variable) and AP, DV, and average spine diameter,
- b) total length (dependent variable) and average lumen diameter, and
- c) average spine diameter (dependent variable) and average lumen diameter.

Back-calculation of length at age was performed using the direct proportion method (LeCren 1947). Length at age was compared among the three sites. The distance from the last visible annulus to the spine's edge was inspected as a function of date of collection to estimate periods of

active growth. Also, the Von Bertalanfy growth function (Ricker 1975) was calculated to compare growth at the three sites.

Sagittal otoliths were removed from eight Edisto River, flathead catfish. Initially, otoliths from several catfish were sectioned and polished in the sagittal, transverse, and frontal plane to determine which, if any, produced the best sections for estimating annuli. These preliminary samples eliminated the sagittal section and the remaining pairs of otoliths were sectioned and polished in both the transverse and frontal plane. Otolith estimates of the number of annuli was compared to similar estimates from accompanying pectoral spines.

## **RESULTS AND DISCUSSION**

Catfish used for pectoral spine age assessment were collected in 1994-1996. Collection dates for each sampling site were:

Edisto River - 8/11/94 through 10/12/95,

Little River and Long Cane Creek - 5/24/95 through 7/3/95, and

Great Pee Dee River - 5/23/94 through 7/15/96.

A highly significant relationship existed between  $\log_{10}$  total length (mm) and  $\log_{10}$  weight (g) of flathead catfish. The relationship for all sites combined was:

$$\log_{10} \text{ weight} = -5.46 + \log_{10} \text{ total length}; R^2 = 0.99; N = 224.$$

There was reasonable agreement among the three spine readers. From the Great Pee Dee River, 98 spines were read. Reader #1 and reader #2 initially agreed on the number of annuli on 37

of the spines. After consultation, readers #1 and 2 agreed on the number of annuli on an additional 30 spines; 25 were not readable while six could not be agreed upon. Reader #3 agreed with 45 of the 67 spines agreed upon by readers #1 and 2. After consultation, 18 additional spines were agreed upon. From the Edisto River, 87 spines were read. Reader #1 and reader #2 initially agreed on the number of annuli on 40 of these spines. After consultation, readers #1 and 2 agreed on the number of annuli in an additional 43 spines; four were not readable. Reader #3 agreed with 74 of the 83 spines. After final consultation, 12 additional spines were agreed upon, leaving only one spine that was classified as unreadable. On Little River and Long Cane Creek spines, reader #1 and 2 agreed on 21 of 40 after the first read. After consultation, an estimate of annuli was obtained for all 40 spines. Reader #3 agreed with 29 of these 40. Marginal increment analysis suggested that growth starts in late spring and substantial growth occurs by mid-summer (Table 1).

There were highly significant ( $P = 0.01$ ) correlations between total length and spine diameter at all sampling locations demonstrating that spine diameter at an annulus can be used to back-calculate total length at that annulus (Table 2). Also, there were highly significant ( $P=0.01$ ) relationships at all sampling locations between a) total length and lumen diameter (Table 3) and b) spine and lumen diameter (Table 4), showing that the spine lumen increases in size proportionally with total fish length and spine diameter.

Table 1. Marginal increment analysis of pectoral spines from flathead catfish caught from the Edisto River in 1995. Ratio is the quotient of new spine growth in 1995 divided by prior spine growth. All fish were either 3 or 4 years old.

Time Period	Total Number	Number w/ no growth	Ratio (SD)
March - May	8	3	1.045 (0.056)
June - July	6	0	1.053 (0.023)
August - Sept.	3	0	1.173 (0.032)

Table 2. Correlation between total length (dependent variable) and pectoral spine diameter of flathead catfish at three, South Carolina sampling sites. A double asterisk denotes significance at  $P = 0.01$ .

Location	N	R-squared		
		DV <sup>1</sup> diameter	AP <sup>2</sup> diameter	Mean diameter
Edisto R.	85	0.94**	0.95**	0.95**
Little R./Long Cane Creek	29	0.72**	0.56**	0.72**
Great Pee Dee R.	63	0.95**	0.96**	0.96**
All sites combined	177	0.95**	0.95**	0.96**

<sup>1</sup> DV = dorsal-ventral

<sup>2</sup> AP = anterior-posterior

Table 3. Correlation between total length (dependent variable) and average lumen diameter of flathead catfish pectoral spines at three, South Carolina sampling sites. A double asterisk denotes significance at  $P = 0.01$ .

Location	N	R-squared
Edisto R.	85	0.87**
Little R. and Long Cane Creek	29	0.64**
Great Pee Dee R.	63	0.91**
All sites combined	177	0.88**

Table 4. Correlation between average diameter (dependent variable) and average lumen diameter of flathead catfish pectoral spines at three, South Carolina sampling sites. A double asterisk denotes significance at  $P = 0.01$ .

Location	N	R-squared
Edisto R.	85	0.88**

Little R. and Long Cane Creek	29	0.63**
Great Pee Dee R.	63	0.87**
All sites combined	177	0.85**

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Mean length at age was higher in Edisto River than in Great Pee Dee River (Table 5). Mean length at age was intermediate in Little River and Long Cane Creek, though small sample size made interpretation difficult. Von Bertalanfy growth functions (VBGF) also supported that highest growth was presently occurring in the Edisto River ( Table 6). The estimated parameters and descriptive statistics of the calculated VBGF were:

Site	N	R <sup>2</sup>	K	T <sub>0</sub>	L <sub>inf</sub> (mm)
Edisto R.	86	0.91	0.034	-1.31	4,730
Little/Long Cane	23	0.06	0.498	0.37	992
Pee Dee R.	62	0.79	0.151	-1.21	1,063
All sites	171	0.62	0.229	-0.84	1,053

Mean back-calculated length at age-1 for the various age cohorts suggested some annulus concealment by the spine's lumen as the catfish aged, especially in the Edisto River (Table 7). At all sites, estimated length at age-1 was less than 300 mm for all age-1+ catfish. However, estimated length at age-1 of five year old Edisto River catfish was 425 mm. In the Great Pee Dee River, there was not an obvious pattern of annulus concealment by age-5 (Table 7). While suspiciously fast growth in the age-5 Edisto cohort may be due to annulus concealment, it is also possible that early-invading Edisto cohorts experienced faster growth at age one than the most recent cohort.



Table 5. Mean length (TL, mm) at age estimates from pectoral spines of flathead catfish from 3 sites in South Carolina. Stderr denotes standard error.

Age	Location								
	Edisto River			Little R. and Long Cane			Great Pee Dee R.		
	N	TL	Stderr	N	TL	Stderr	N	TL	Stderr
1	6	260	16	-	-	-	18	255	13
2	10	360	26	1	510	-	3	419	34
3	9	550	43	2	731	6	4	539	41
4	10	769	32	3	789	85	5	560	17
5	5	970	23	4	883	42	5	642	30
6	2	1,153	17	1	864	-	3	670	26
7	1	1067	-	3	1,018	73	5	761	45
8	1	1,030	-	2	1,004	13	2	712	17
9	-	-	-	4	961	75	3	832	78
10	-	-	-	1	1,129	-	1	770	-
11	-	-	-	3	984	22	1	713	-
12	-	-	-	3	940	15	-	-	-
13	-	-	-				1	901	-
14	-	-	-	1	940	-	1	1,101	-
15	-	-	-	-	-	-	-	-	-

Table 6. Estimated length (TL, mm) at age of flathead catfish at two locations in South Carolina. Estimates were obtained with Von Bertalanfy growth functions; estimated parameters are provided in text.

Age	Edisto R.	G. Pee Dee R.
1	356	300
2	502	407
3	643	499
4	779	578
5	910	646
6	1,038	704
7	1,160	754

Table 7. Mean back-calculated length at first annulus of flathead catfish from three locations in South Carolina.

Location	Age of fish	N	Mean length (mm)	Standard error
Edisto R.	1	6	260	16
	2	10	244	17
	3	9	276	21
	4	10	354	25
	5	5	425	35
Little R./Long Cane	1	-	-	-
	2	1	253	-
	3	2	447	39
	4	2	286	11
	5	4	439	36
G. Pee Dee R.	1	18	255	13
	2	3	330	17
	3	4	311	16
	4	5	254	20
	5	5	239	38
All combined	1	24	256	10
	2	14	263	16
	3	15	308	20
	4	18	316	19

Otolith and pectoral spine age estimates were in very close agreement for 7 fish ranging in size from 8.2 to 21.4 kg (Table 8). Preliminary inspection of otoliths revealed pronounced annuli in the transverse and frontal plane; good results were not obtained from the sagittal plane. Compared to spines, otoliths took more time to process per sample, as sanding to the correct width was critical. Otolith preparation time would decrease as experience was obtained. The degree of agreement between otolith and spine estimates, increases confidence in the previously-reported spine aging estimates. However, In future studies, otolith use is recommended. This would help to identify any bias in age estimates due to lumen expansion or misinterpretation.

Flathead catfish have been established in the tributaries of Lake Thurmond and the Great Pee Dee River for a longer time than the Edisto River. The 1981 and 1982 year classes were represented in these samples by fourteen year old specimens from the tributaries of Lake Thurmond and the Great Pee Dee River, respectively. The oldest age class represented by Edisto collections was the 1986 year class.

The growth of Edisto River flatheads was exceptionally fast when compared to other populations. Quinn (1988) noted that the growth of flathead catfish in the Flint River, GA exceeded reports from 35 of 36 sites in the United States; the Edisto growth rate exceeds that noted in the Flint River. Exceptionally high growth in the Edisto is probably associated with its recent invasion of this system. Growth rates in the Great Pee Dee River suggest the Edisto growth rate may decline with time.

Table 8. Comparison of aging results for Edisto River flathead catfish from pectoral spines and otoliths. Samples were collected on August 21, 1996. Otoliths were cut in transverse (T) and frontal (F) sections. Underlined spine age estimates indicate the age agreed upon after a conference.

		Otolith		Spine		
		Estimated Age		Estimated Age		
		Reader 1		Reader 1	Reader 2	
TL, mm	Wt, kg.	Left	Right	Left	Right	
980	14.4	6+ (T)	6+ (T)	6+	6+	6+
960	15.9	9+ (F)	9+ (F)	5+	5+	5+
1160	21.4	9+ (T)	9+ (F)	8+	8+	8+
1055	16.5	10+ (T)	10+ (F)	<u>10±</u>	10+	9+
1040	18.7	8+ (T)	8+ (F)	7+	7+	<u>8±</u>
1110	12.8	6+ (T)	6+ (F)	5+	5+	5+
895	8.2	4+ (T)	4+ (F)	3+	4+	<u>3±</u>

## **Conclusions and Recommendations**

1. Growth estimates for flathead catfish in three locations were obtained using pectoral spines sectioned at or near the distal end of the basal recess. Estimates may have some bias due to lumen expansion. Future work should section the spine at the articulating process to avoid the lumen. Properly prepared otoliths can and should be used to verify aging estimates obtained from spines. Otolith aging tends to confirm that the obtained spine estimates are acceptable for defining major trends.
2. As the lumen has a predictable rate of expansion, a more detailed statistical analysis of the previously reported data set may be able to produce a 'statistically corrected' estimate of age. This analysis should be pursued.
3. The Edisto River has a very fast growing population which invaded this system in or about 1986. The Pee Dee River, which seems to have been colonized by 1981, has slower growth than the Edisto, but still above average when compared to Midwestern populations. More samples are needed from Lake Thurmond tributaries to reliably estimate growth.
4. Growth in flathead populations, especially newly established ones, should be monitored every 5-10 years to quantify changes in growth parameters.
5. Now that growth is defined, food habit data and bioenergetic modeling can estimate the total consumption required to produce the observed growth. Linked with a population estimate, this approach would provide insights into the impact of flathead catfish on the fish community.

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## JOB PROGRESS REPORT

STATE: South Carolina

PROJECT NUMBER: F- 63

PROJECT TITLE: Fisheries Investigations in Lakes and Streams - Statewide

STUDY: Survey and Inventory  
Statewide

STUDY TITLE: Fishery surveys -  
Fisheries

Research

JOB NO: IIA  
performance of two  
strains of largemouth bass in private ponds  
Introduction

JOB TITLE: Relative

Two subspecies of largemouth bass *Micropterus salmoides*, the Florida largemouth bass *M. s. floridanus* and the northern largemouth bass *M. s. salmoides*, exist and readily interbreed in both hatchery and reservoir environments (Isely et al., 1987, Gilliland and Whitaker 1989, Philipp and Witt 1991). The native range of the Florida subspecies (FLMB) is restricted to peninsular Florida. The northern subspecies (NLMB) is native to waters north along the Atlantic coast states from Maryland and west to the Mississippi (Philipp et al., 1983).

Genetic differences between the two subspecies are measurable at four diagnostic enzyme coding loci (Philipp et al., 1983). The differences at two loci, aspartate aminotransferase (sAAT-2\*) and isocitrate dehydrogenase (sIDHP-1\*), are fixed meaning one allele or combination of alleles is present only in populations of the Florida subspecies and the other only in populations of the northern subspecies. At a third locus, malate dehydrogenase (sMDH-B\*), Florida populations are fixed for a Florida allele, while northern populations may be fixed for a northern allele or possess a combination of northern and Florida alleles. At the fourth diagnostic locus, superoxide dismutase (sSOD-1\*), northern populations are fixed for the northern allele while



Florida populations possess a combination of the northern and Florida alleles. Alleles typical of the northern subspecies are sAAT-2\*100 and sAAT-2\*110, sIDHP-1\*100, sMDH-B\*100, and sSOD-1\*147. Alleles typical of the Florida subspecies are sAAT-2\*126 and sAAT-2\*139, sIDHP-1\*121, sMDH-B\*114, and sSOD-1\*100.

South Carolina is located in the broad hybrid zone between the ranges of the two pure subspecies. A statewide allozyme study of largemouth bass confirmed that South Carolina populations were hybrids (Bulak et al., 1995). This study also showed the existence of a geographic cline within South Carolina where the relative abundance of Florida alleles decreased from southeast to northwest. The relative frequency of alleles that are fixed for the Florida subspecies ranged from 98% in Lake Moultrie, a Coastal Plain reservoir, to 36% in Lake Wateree, a Piedmont reservoir. Bulak et al. (1995) suggested that natural selection played a role in maintaining this allelic cline.

Physiological and ecological differences among FLMB, NLMB, and their hybrids have been documented. A number of studies have shown a difference in the response of the FLMB, NLMB, and their hybrids to various temperature regimes (Fields et al., 1987, Charmichael et al., 1988). Other studies have shown differences in timing of spawning, growth rate, reproductive success and survival of the two subspecies (Philipp and Witt 1991, Maceina et al. 1988, Gilliland and Whitaker 1989, Isely et al. 1987).

The objective of this study was to examine performance differences between progeny of Lake Wateree and Lake Moultrie largemouth bass. Privately-owned farm ponds throughout South Carolina were used as study sites. Each pond was stocked with progeny from either Lake Wateree or Lake Moultrie. The objective will be achieved by measuring growth of stocked bass at age-1 and age-3, and by monitoring the long-term temporal change in genotypes of age-0+ bass.

## Materials and Methods

### [Pond Selection]

Ponds were selected prior to stocking. A list of all pond owners purchasing fish from the South Carolina Department of Natural Resources was obtained. Through a series of phone interviews and pond visits, study sites were chosen based on the following criteria:

- size 1-3 acres
- either new or properly renovated
- little potential for invasion by wild fish
- agreement with pond owner to allow access for data collection

Ponds had been stocked in October with bluegill *Lepomis macrochirus* and redear *L. microlophus* fingerlings. Pond owners were advised that largemouth bass fingerlings would be delivered to their pond and they should not stock the ponds with bass from any other source.

### [Broodfish collection and fingerling production]

Largemouth bass for experimental stockings were produced from adult bass collected from Lakes Moultrie and Wateree. Lake Moultrie broodfish were collected by electrofishing in March of 1993. Lake Wateree broodfish were collected by electrofishing in March of 1994. In 1994 and 1995 each group of broodfish was allowed to spawn. Resulting fry were collected and transferred to grow-out ponds where they were raised to a total length of approximately 25mm. Fry were harvested from as many schools as possible to maximize the number of parents contributing to the gene pool.

Size at stocking and frequencies of alleles characteristic of the NLMB and FLMB were determined for each stock of fingerlings. Forty fingerlings from each stock were weighed (gm), measured (TL mm) and preserved in 100% isopropyl alcohol for future reference. Two sets of

100 fingerlings from each strain were placed on dry ice and stored frozen for allozyme analysis. Horizontal starch gel electrophoresis was performed according to Norgren (1986). Gels were stained for the four allozymes diagnostic for the northern and Florida bass subspecies. Allele frequencies of fingerlings were compared to source lake populations using the G-test (Sokal and Rohlf, 1969).

Tissue samples were taken from anesthetized L. Wateree broodfish (Leitner and Isely, 1994) prior to spawning in 1995 and allozyme analysis was performed. The purpose of this was to identify and remove from the broodfish pool any individuals possessing a rare allele at the IDHP-1\* locus.

#### [Stocking]

One half of the ponds in each region were stocked with Moultrie fish and the other half with Wateree fish. Prior to the first day of stocking, ponds were chosen at random for stocking with the Lake Moultrie strain. As each pond was chosen, its closest neighbor was assigned the Wateree strain. This ensured a uniform distribution of each strain throughout each region. Only one strain (Moultrie or Wateree) was hauled per day and the truck was flushed and stocked with fresh fingerlings each morning. Largemouth bass were hand counted and stocked at the rate of 50 and 100 fingerlings per acre for unfertilized and fertilized ponds, respectively.

At stocking and during regular pond visits, pond owners were advised of steps they should take to best manage their ponds. Recommendations included stabilization of banks, control of aquatic weeds, liming, and sufficient fish harvest.

#### [Water quality monitoring]

Selected water quality parameters were analyzed from each pond to define productivity differences among ponds. Parameters measured were hardness and alkalinity, at appropriate

intervals, and pH, temperature and chlorophyll-a concentration throughout the growing season.

Hardness and alkalinity were measured using a standard Hach kit with a digital titrator.

Temperature and pH measurements were made using an Orion field pH meter equipped with a

Ross electrode. Chlorophyll-a was determined with a Turner Filter Fluorometer Model 111.

Prior to calculating chlorophyll-a concentrations, the fluorometer was calibrated. A series of known concentrations of chlorophyll-a were read at each of four sensitivity settings. Using the values obtained, calibration factors were derived to convert fluorometric readings of unknowns at each sensitivity setting to chlorophyll-a concentrations, as follows:

$$F_s = \frac{C_a}{R_s}, \quad \text{where}$$

$F_s$  = calibration factor for sensitivity setting S,

$R_s$  = fluorometer reading for sensitivity setting S,

$C_a$  = concentration of chlorophyll-a,  $\mu\text{g/L}$ .

In the field water samples for chlorophyll-a determination were taken from 0.3 m below the surface at three sample sites on each pond. Sample sites followed the pond's stream gradient with an upper or inflow site, a middle, and a lower or outflow site. Each sample was inverted to mix any particles that may have settled and 50 ml were measured for filtration. The filter paper, with the filtrate, was carefully rolled, blotted, and placed in a glass vial with 0.7 ml of 10% magnesium carbonate solution. Tubes were capped and stored in the dark on dry ice for transport to the lab, where they were stored frozen for later analysis. When samples had been frozen for at least 24 hours, 6.3 ml of acetone were added, yielding a 90% buffered acetone solution in the tube. Samples were placed in the refrigerator overnight for thawing. The freeze-thaw cycle ruptures the phytoplankton cells, releasing the chlorophyll pigments into solution (H. N. McKellar, pers.

comm.). Sample tubes were removed from the refrigerator, shaken and the solution was pipetted off and centrifuged at 3,000 g's for about 15 minutes for clarification. An amount, generally 0.1-4.0 ml, of each sample was carefully measured to the nearest 0.1 ml, removed to a cuvette, and diluted with 90% buffered acetone. This dilution was placed in the fluorometer for a reading. To account for pheophytin, the solution was then acidified with one normal HCl and allowed to sit for one minute before being read again. The formula used for determining chlorophyll-a concentration ( $\mu\text{g/L}$ ) was:

$$\text{Chl}_a = \frac{F_s (r) (R_b - R_a)}{V}$$

where,

$F_s$ =conversion factor from calibration,

$r=2 (R_b/R_a)$ , as determined with pure chlorophyll-a for the instrument),

$R_b$ =Reading before acidification,

$R_a$ =Reading after acidification, and

$V = \frac{\text{Volume of sample}}{\text{Volume of extract.}}$

Mean annual water quality parameters were computed for each pond. Mean pH, hardness, and alkalinity were the simple average of measurements taken throughout the sampling season ( $n=1-3$ ). Mean annual chlorophyll-a concentration was computed by first taking the mean of the three samples for each sampling event and then taking the average of these means for each pond.

[Fish collections]

Adult largemouth bass were collected by electrofishing from each pond at one and three years post stocking. Ponds stocked in 1994 were sampled for adult largemouth bass from 6/15-7/27/95 and from 6/12-8/21/97. Ponds stocked in 1995 were sampled from 6/11-6/19/96 and from

6/1-6/26/98. At one year post stocking, where possible, we collected 10% of the number stocked with a minimum of 20. All fish were weighed, measured, and returned to the pond. Scales or otoliths were collected from fish that were suspiciously large or small, for age verification. Fish that were older than age one were noted and not included in further analysis. Growth rate for each fish was computed as:

$$\text{growth rate} = \frac{\text{length at harvest} - \text{length at stocking}}{\text{days since stocking.}}$$

Mean growth rate at age-1 of largemouth bass was computed for each study pond.

At 3 years post stocking, bass were collected from 1994 stocked ponds by electrofishing and angling. Electrofishing was used on the initial sampling visit to each pond. All fish collected were weighed, measured and fin clipped to avoid resampling. A length-frequency histogram was constructed in the field so that apparent age classes could be visualized. Scales were taken for age estimation from some fish from each size group, and from all fish that appeared to be older than age-1. An estimate of how many age-3 bass were collected from each pond was derived from length frequency data and an initial look at scale samples. Ponds where it did not appear at least 4, age-3 largemouth bass were collected were resampled using a combination of angling and electrofishing effort. Age was estimated from scales by two independent readers. Mean growth rate for age-3 bass was computed.

From 1995-1997 (1-3 years post stocking) young of the year (yoy) largemouth bass were collected annually from each pond for allozyme analysis. A beach seine was pulled along the edges of the pond until at least 20 yoy were collected. These fish were measured, wrapped in tinfoil and immediately placed on dry ice. They were transported to the lab where they were

stored frozen for analysis at the previously discussed four enzyme coding loci.

[Statistical analysis]

Water quality variables pH, hardness, alkalinity and chlorophyll-a concentration were tested for normal distribution using PROC UNIVARIATE (SAS, 1987). Variables that were not normally distributed were log transformed. For 1994 data only, hardness, alkalinity and pH were evaluated as predictors of chlorophyll-a concentration with linear regression analysis.

Because of expected variation among ponds, atypical ponds were identified and not included in analysis of growth. These included ponds where introductions of wild fish or poor water quality had an effect on forage availability.

PROC MIXED (SAS, 1996) was used to identify factors that were significant predictors of largemouth bass growth rate. The effects study site (pond), region, strain, the interaction of region and strain, and each water quality variable (non-log transformed) were tested.

In evaluating growth at age-1, of the four water quality variables tested only pH contributed significantly to the model. All other water quality variables were excluded from the model. The LSMEANS statement (SAS, 1987) was used to compute the mean growth rates for each region and type adjusted by the mean value of the significant covariate pH. The adjusted mean growth rates were tested for differences between region and type. Additional tests were performed with mean pH set at 6 and 8 to insure that the relationship was essentially the same at all pH values.

In evaluating growth at age-3, no water quality variable contributed significantly to the model. All four water quality variables were removed from the model and Proc Mixed was used to test region, strain, and their interaction as predictors of growth rate.

Allele frequencies of juveniles were calculated for each pond at each of the four diagnostic

loci. Allele frequencies for each pond were compared to those of parental stocks using the G-test (Sokal and Rohlf, 1969). A trend showing an increase or decrease in Florida type alleles for any group would be an indication of selection.

## Results

### [Pond selection and stocking]

Twenty four ponds were stocked in 1994. Of 12 Coastal Plain ponds, 7 were stocked with the Wateree and 5 with the Moultrie strain. Of 12 Piedmont ponds, 6 were stocked with the Wateree and 6 with Moultrie strain.

Thirteen ponds were stocked from May 19 - May 23, 1995. Of six Coastal Plain ponds, four were stocked with Moultrie and two with Wateree strain. Of seven Piedmont ponds, four were stocked with Wateree and three with Moultrie strain. A stocking summary is provided in Table 1.

Table 1. Number of ponds and total acres stocked with distinct strains of largemouth bass in 1994 and 1995.

<u>Region</u>		<u>Strain</u>	<u>Number</u>
<u>Ponds</u>	<u>Total Acres</u>		
Piedmont	Wateree	10	19.0
	Moultrie	9	12.3



Coastal Plain	Wateree	9	16.2
	Moultrie	9	12.5

---

Moultrie and Wateree strains were of similar size at stocking in both 1994 and 1995. In 1994, Moultrie fingerlings (N=41) averaged 26 mm TL (sd=3.3) and 0.2 grams while Wateree fingerlings (N=39) averaged 34 mm TL (sd=1.8) and 0.4 g (sd=0.08). In 1995, Moultrie fingerlings (N=44) averaged 32 mm TL (sd=3.9) and 0.3 g (sd=0.19) while Wateree fingerlings (N=40) averaged 25 mm TL (sd=2.7) and 0.12 g. Standard deviations for weight for 1995 Wateree and 1994 Moultrie stocks could not be calculated because some fingerlings were weighed in batches.

Allele frequencies of stocked fingerlings were generally consistent with source populations (Tables 2 and 3). Lake Moultrie fingerlings were not significantly different ( $P=0.05$ ) from wild stock at any of the four loci examined in either 1994 or 1995. Lake Wateree fingerlings stocked in 1994 were significantly different from wild stock at sMDH-B\* ( $P \leq 0.05$ ) and at sIDHP-2\* ( $P \leq 0.001$ ). At sMDH-B\*, the stocked fingerlings possessed the northern allele in significantly higher numbers than the wild stock. Analysis at sIDHP-2\* indicated that stocked fingerlings possessed a rare allele, sIDHP-2\*142, in significantly higher numbers than wild stock.

Table 2. Allele frequencies for Wateree strain largemouth bass fingerlings used to stock study ponds in 1994 and 1995, with survey data of allele frequencies for L. Wateree where stocks originated, and subsequent F<sub>1</sub> and F<sub>2</sub> generations. Alleles, or allele pairs, listed first are fixed (sAAT-2\*, sIDHP-2\*) or dominant in the Northern subspecies. Alleles listed second are fixed or dominant in the Florida subspecies. An \* indicates a significant difference from survey data at P=0.05 and a \*\* at P=0.001. A + indicates a significant difference from original stocks at P=0.05 (filial generations from 1994 fingerlings are compared to survey data at sIDHP-1\*).

<u>Locus/Allele</u>	<u>Survey data</u>		<u>1994 fing.</u>	<u>F<sub>1</sub></u>	<u>F<sub>2</sub></u>	
<u>1995 fing.</u>	<u>F<sub>1</sub></u>					
	N=122	N=100	N=240	N=262	N=100	N=56
<u>sAAT-2*</u>						
100, 110	0.66	0.65	0.71	0.65	0.44*	0.34
126, 139	0.34	0.35	0.29	0.35	0.54*	0.66
<u>sIDHP-1*</u>						
100	0.47	0.18**	0.44	0.45	0.66*	0.45+
121	0.52	0.50**	0.56	0.55	0.34*	0.55+
142	0.01	0.32**	0.00	0.00	0.00*	0.00
<u>sMDH-B*</u>						
100	0.60	0.73*	0.61+	0.64+	0.60	0.48

114	0.40	0.27*	0.39+	0.36+	0.40	0.52
<u>sSOD-1*</u>						
147	0.57	0.58	0.56	0.59	0.64	0.65
100	0.43	0.42	0.44	0.41	0.36	0.35

Table 3. Allele frequencies for Moultrie strain largemouth bass fingerlings used to stock study ponds in 1994 and 1995, with survey data of allele frequencies for L. Moultrie where stocks originated, and subsequent F<sub>1</sub> and F<sub>2</sub> generations. Alleles, or allele pairs, listed first are fixed (sAAT-2\*, sIDHP-2\*) or dominant in the Northern subspecies. Alleles listed second are fixed or dominant in the Florida subspecies. An \* indicates a significant ( $P \leq 0.05$ ) difference from survey data. A + indicates a significant ( $P \leq 0.05$ ) difference from original stocks.

<u>Locus/Allele</u> <u>1995 fing.</u>	<u>Survey data</u>		<u>1994 fing.</u>	<u>F<sub>1</sub></u>	<u>F<sub>2</sub></u>	
	<u>F<sub>1</sub></u>					
	N=116	N=52	N=156	N=181	N=100	N=84
<u>sAAT-2*</u>						
100, 110	0.10	0.19	0.00+	0.07+	0.14	0.07
126, 139	0.90	0.81	1.00+	0.93+	0.86	0.93
<u>sIDHP-1*</u>						
100	0.02	0.00	0.00	0.05+	0.02	0.00

121	0.98	1.00	1.00	0.95+	0.98	1.00
142	0.00	0.00	0.00	0.00+	0.00	0.00
<u>sMDH-B*</u>						
100	0.00	0.00	0.00	0.00	0.00	0.00
114	1.00	1.00	1.00	1.00	1.00	1.00
<u>sSOD-1*</u>						
147	0.19	0.14	0.29+	0.17	0.13	0.25+
100	0.81	0.86	0.71+	0.83	0.87	0.75+

Lake Wateree fingerlings from 1995 were significantly different ( $P=0.05$ ) from Lake Wateree wild stock at sAAT-2\* and at sIDHP-2\*. At sAAT-2\* the stocked fingerlings possessed the Florida alleles in significantly higher numbers than the wild stock. At sIDHP-2\* the stocked fingerlings possessed the northern allele in significantly higher numbers than the wild Lake Wateree stock.

Ponds were sampled for water quality parameters three times during the 1994 growing season and twice during the 1995-1997 growing seasons. Samples were taken in June, August and September/October in 1994, in June and August in 1995, in June/July and October in 1996, and in July and October in 1997. A wide range of water quality conditions were encountered from pond to pond (Table 4 and 5). Mean values for pH for 88% of ponds were between 6.5 and 9, the range at which fish grow best (Crochet, 1992). Fifty seven percent of ponds averaged 20 mg/l or higher for both hardness and alkalinity, the minimum concentration considered to provide adequate buffering capacity and support a healthy phytoplankton community (Crochet, 1992). High variance for hardness and alkalinity at certain ponds is due to the liming of those ponds to increase hardness and alkalinity during the course of sampling.

Age-1 largemouth bass were collected from 26 of 27 ponds, sampled from 6/15 to 7/27/95 and from 12 of 13 ponds sampled from 6/11 to 6/19/96. Mean growth rates were calculated for each pond by region and strain stocked (Table 6 and 7).

Largemouth bass age-1 to age-3 were collected from 23 of 24 ponds sampled in 1997. Age estimates were determined for 184 fish and 36 age-3's were identified.

Table 4. Water quality parameters measured on 1994 stocked study ponds, Summer 1994-Fall 1996. Values are mean values

for the three year course of sampling. Standard deviations are in parenthesis. Individual ponds are grouped by strain stocked (M = Moultrie, W = Wateree) and region (C = Coastal Plain, P = Piedmont) .

<u>Pond Name</u>	<u>chl-a(μ g/l)(SD)</u>	<u>pH(SD)</u>	<u>hardness(SD)</u>	
<u>alkalinity(SD)</u>				
<u>M/C</u>				
Mulberry	2.2 (0.8)	5.3(3.8)	36.2(35.4)	16.8 (14.3)
Price	4.6( 1.6)	8.1(0.82)	34.7 (5.1)	25.2 (7.2 )
Gollihugh	7.2(3.0)	8.9(0.73)	42.4(9.1)	40.7(22.8)
<u>M/P</u>				
Adams	2.7 (0.8)	7.9(1.2)	13.0(7.6)	11.2 (9.6)
Kirby	3.8 (1.4)	7.7(1.0)	9.3 (3.2)	9.7 (1.9)
Cline	3.1 (10.3)	6.6(2.8)	11.4 (2.8)	7.8 (1.7)
Lockridge	4.9 (1.0)	7.7(0.9)	15.7 (5.1)	18.2 (5.5)
Beer, G	7.1 (1.3)	7.3(0.6)	10.2 (2.9)	13.6 (0.4)
<u>W/C</u>				
Gift	2.9 (0.7)	7.9(0.8)	54.0( 9.8 )	45.0 ( 8.7 )
Shelley	3.6 (1.3)	7.1(0.7)	3.2 (1.4)	3.3 (1.8)
Carrol	4.9 (2.0)	6.4(4.4)	50.8 (12.1)	35.6 ( 7.0 )
Britton	7.0 (3.2)	7.5(1.8)	16.5 (7.1)	21.8 (13.6 )
New	6.3 (2.1)	6.9(3.1)	15.1 (4.8)	11.4 (5.9)
Chelsea	8.8(3.5)	7.7(1.6)	41.5 (31.8)	27.4 ( 5.1 )
<u>W/P</u>				
Childress, C.	3.2 (0.4)	7.2(0.4)	22.2 (3.3 )	24.7 (1.9 )
Coble	4.0 (0.7)	8.4(1.1)	20.9 (1.4 )	24.4 ( 3.0 )
Meeks	4.6 (1.1)	8.2(0.9)	14.9(2.1)	15.0 (1.7)
Thackston	5.5 (1.4)	6.9(3.2)	34.6 (6.3 )	31.8 ( 5.9 )
Beer, D.	9.5 (4.5)	8.8(1.1)	35.7 (4.1)	22.5(10.5)
Benfield	9.4 (4.0)	8.3(1.0)	38.2 (9.3)	37.3 (8.0 )

Table 4 continued

<u>Pond Name</u>	<u>chl-a(<math>\mu</math> g/l)(SD)</u>	<u>pH(SD)</u>	<u>hardness(SD)</u>	<u>alkalinity(SD)</u>
<u>Others</u>				
Childress, K. <sub>d</sub>	5.7 (1.5)	7.1(0.2)	42.9 (0.8)	41.0 (5.3 )
Davis <sub>d</sub>	10.4(3.2)	9.8(0.2)	33.6(13.6)	( - )( -- )
Harrison <sub>d</sub>	2.9 ( -- )	7.7 ( -- )	54.1( -- )	53.0 ( -- )
Minchey <sub>d</sub>	4.8 (2.1)	8.1(0.8)	44.6 (17.2)	32.5 (19.1 )
Turner <sub>d</sub>	4.2 (1.0)	7.2(0.2)	35.7(1.3)	41.6(1.60)
Bennet <sub>+</sub>	3.9 (1.3)	8.6(0.8)	21.3 (7.5)	23.7 (7.4 )
English <sub>+</sub>	5.7 (2.7)	7.8(3.4)	82.5 (50.2)	72.5 (30.7 )
Helmly <sub>+</sub>	4.4 (2.0)	6.1(4.2)	56.8(12.7)	90.2 (43.5)
Sims <sub>+</sub>	5.0 (1.6)	5.8(2.9)	32.4(9.1)	19.1( 3.9 )

<sub>+</sub> = ponds not included in growth analysis, but retained for future study

<sub>d</sub> = Ponds removed from study

Table 5. Water quality parameters measured on 1995 stocked study ponds, Summer 1995-Fall 1997. Values are mean values for the three year course of sampling. Standard deviations are in parenthesis. Ponds are grouped by strain stocked (M = Moultrie, W = Wateree) and region ( C = Coastal Plain, P = Piedmont).

<u>Pond Owner</u>	<u>chl-a (mg/L)(Std)</u>	<u>pH(Std)</u>	<u>hardness(Std)</u>
<u>alkalinity(Std)</u>			
<u>M/C*</u>			
Hughes	7.3 (3.0)	8.3 (1.1)	24.3 (1.3) (3.7)
Shields	5.2 (1.2)	6.8 (3.3)	172.8 (33.4) (24.4)
Platts	6.4 (3.2)	6.9 (3.5)	15.5(4.6)
McCants	3.6 (0.8)	8.0 (0.9)	84.4 (6.2) (7.2)
74.0 (14.6)			44.1
<u>M/P</u>			
Workman	7.8 (2.8)	8.0 (1.2)	16.9 (2.6) (4.8)
Freeland	6.9 (3.6)	7.8(0.9)	9.8 (1.8) (2.5)
Patterson	8.3 (3.2)	7.3(0.4)	15.3 (2.7 ) (3.1)
19.2			
<u>W/C</u>			
Ravenel	3.0 (1.1)	7.8(0.3)	90.9 (12.4) (8.5)
Branton	3.2 (1.6)	6.8 (3.4)	119.3 (27.0) (15.8)
86.0			74.8
<u>W/P</u>			
Hanvey	4.6 (0.9)	7.1 (0.4)	28.1 (7.7) (3.8)
Harrison	3.6 (1.3)	8.2 (0.9)	30.0 (5.7) (5.6)
Holland	3.5 (1.2)	7.7(0.7)	34.3 (5.6) (7.3)
McGee	6.0 (5.5)	8.6(1.0)	32.8 (10.3)
20.8			25.4
			25.0
			30.1





Table 6. Mean growth rate for age-1 largemouth bass collected in 1995. Individual ponds are grouped by strain stocked (M = Moultrie, W = Wateree) and region (C = Coastal Plain, P = Piedmont).

<u>Pond Name</u>	<u>Growth (mm/d)</u>	<u>Standard</u> <u>deviation</u>	<u>No. of adults</u>
<u>M/C</u>			
Mulberry	0.59	.04	17
Price	0.65	.07	15
Gollihugh	0.66	.03	18
<u>M/P</u>			
Cline	0.45	.09	14
Adams	0.48	.03	9
Beer, G	0.54	.03	14
Kirby	0.57	.07	20
Lockridge	0.58	.09	6
<u>W/C</u>			
Britton	0.46	.08	11
New	0.62	.03	21
Chelsea	0.65	.04	19
Carrol	0.67	.03	20
Gift	0.72	.05	3
Shelley	0.74	.03	3
<u>W/P</u>			
Childress, C	0.44	.03	21
Benfield	0.52	.03	19
Meeks	0.55	.02	19
Thackston	0.58	.03	29
Beer, D.	0.59	.02	15
Coble	0.64	.05	18

Table 6  
continued

Growth

<u>Others</u>	<u>(mm/day)</u>		<u>Standard</u> <u>Deviation</u>	<u>No.</u> <u>Adults</u>
Helmly <sub>+</sub>	0.30		.02	8
Bennet <sub>d</sub>	0.36		.08	12
English <sub>+</sub>	0.42	.05	6	
Sims <sub>+</sub>	0.44	.10	15	
Childress, K. <sub>d</sub>	0.49	.16	19	
Minchey <sub>d</sub>	0.73	.06	13	

<sub>d</sub> = ponds removed from the study

<sub>+</sub> = ponds not included in growth analysis but retained for future study

Table 7. Mean growth rate for age-1 largemouth bass collected in 1996. Individual ponds are grouped by strain stocked (M = Moultrie, W = Wateree) and region (C = Coastal, P = Piedmont).

<u>Pond Owner</u>	Standard		<u>Deviation</u>
	<u>Growth (mm/day)</u>		<u>No. Adults</u>
<u>M/C*</u>			
Hughes	0.70	0.07	19
Shields	0.59	0.02	20
Platts	0.48	0.04	19
McCants	0.66	0.16	7
<u>M/P</u>			
Workman	0.55	0.12	26
Freeland	0.62	0.12	19
Patterson	0.53	0.03	21
<u>W/C</u>			
Ravenel	0.78	0.06	8
Branton	-	-	0
<u>W/P</u>			
Hanvey	0.60	0.04	17
Harrison	0.57	0.07	30
Holland	0.38	0.07	11
McGee	0.59	0.07	19

Juvenile largemouth bass (n=1901) were collected with beach seines from 19 of 27 ponds in 1995, from 32 of 37 ponds in 1996, from 28 of 36 ponds in 1997, and from 10 of 11 ponds in 1998. Number collected per pond ranged from 10 to 33 for all but 6 ponds from which less than 10 fingerlings were collected. Average total length of fingerlings ranged from 29 mm to 134 mm for each pond sampled. Fingerlings were stored frozen for allozyme analysis. Analysis was completed for fingerlings collected from 1995 and 1996. Analysis of 1997 and 1998 fingerlings is pending.

Raw data for chlorophyll-a concentrations and alkalinity values were not normally distributed. A  $\log_{10}$  transformation resulted in a normal distribution for both and transformed data was used for these two variables in linear regression analysis.

Chlorophyll-a data from 1994 was significantly ( $p=.05$ ) related to pH, alkalinity, and hardness. The equations produced by the linear regression analysis were:

a.  $\log_{10}(\text{chlorophyll-a}) = 0.17 * \text{pH} + 0.14$  ;  $R^2 = 0.17$

b.  $\log_{10}(\text{chlorophyll-a}) = 0.40 * \log_{10}(\text{alk}) + 0.86$  ;  $R^2 = 0.19$

c.  $\log_{10}(\text{chlorophyll-a}) = 0.44 * \log_{10}(\text{hard}) + 0.82$  ;  $R^2 = 0.10$ . While all equations are significant, relatively low R-squared values indicate other factors are affecting chlorophyll-a concentration in the study ponds.

Data from five atypical ponds were removed from the data set. Largemouth bass from two of the ponds, Helmly and English, were stunted due to limited or zero bream reproduction and therefore minimal forage availability. Three other ponds, Bennet, Minchey, and K. Childress, were removed because introduced fish had severely impacted forage availability to stocked largemouth bass.

The growth difference between regions was significant ( $P = 0.05$ ). Largemouth bass stocked in Coastal Plain ponds grew faster, 0.61 mm per day ( $sd=0.11, N=215$ ), than those stocked in Piedmont ponds (0.55 mm per day,  $sd=0.09, N=324$ ).

Mean growth for all age 1 largemouth bass collected in 1995 and 1996 was 0.57 mm per day ( $sd=0.09, N=539$ ). Growth was computed for each fish at 386-474 days post stocking. Analysis showed that region and pH were significant predictors of growth rate. The test of least squares means showed no significant difference between growth rates of the two strains or between the interactions of strain and region.

Mean growth for all age-3 largemouth bass collected in 1997 was 0.29 mm/day ( $sd=0.04, N=36$ ). Growth was computed for each fish at 1128-1197 days post stocking. Analysis showed no significant difference in growth rate due to the interaction between region and strain. There was a significant difference ( $p=0.05$ ) between regions, with fish in the Coastal Plain growing more (0.30 mm per day,  $sd=0.03, N=20$ ) than fish in the Piedmont (0.28 mm per day,  $sd=0.04, N=16$ ). The difference between strains was significant at  $P = 0.10$  with fish of the Wateree type growing faster (0.30 mm per day,  $sd=0.03, N=21$ ) than fish of the Moultrie type (0.28 mm per day,  $sd=0.04, N=15$ ).

Allozyme analysis was completed and allele frequencies computed for fingerlings collected in 1995 and 1996. This data is included in Table 2 (Wateree strain) and Table 3 (Moultrie strain). This includes  $F_1$  and  $F_2$  generations from 1994 stocked ponds and  $F_1$  generations from 1995 stocked ponds. G-test comparisons showed significant deviations ( $P=0.05$ ) from Wateree strain parental stocks at one locus each for both the  $F_1$  and  $F_2$  generations from 1994 stocks, and the  $F_1$  generation from 1995 stocks. (Because of the potential discrepancy in sIDHP-1\* data for the 1994 Wateree strain, comparisons at that locus were made using allele frequencies for the Lake

Wateree largemouth bass population.) Significant ( $P=0.05$ ) deviations from Moultrie strain parental stocks were present at two loci each for both the  $F_1$  and  $F_2$  generations from 1994 stocks, and at one locus for the  $F_1$  generation from 1995 stocks.

### Discussion

Differences in growth for fish stocked in the Coastal Plain vs. those stocked in the Piedmont followed the same trend from age-1 - age-3. Throughout the study fish exhibited significantly greater growth in the Coastal Plain, a milder climate with a longer growing season.

High pond to pond variation may affect our ability to detect growth differences between largemouth bass strains. Growth at age-3 of the Wateree strain was significantly greater than the Moultrie strain at  $p=0.10$ . This followed the same trend as non-significant growth differences at age-1. However, small sample sizes of age-3 largemouth bass make it difficult to use this information in drawing inferences about the general population. Only  $N=36$  age-3's were collected from 16 of 23 ponds sampled.

A study design where ponds were stocked with equal numbers of both strains would have minimized the effect of pond to pond variation, increasing our power to detect growth differences due to strain. This approach was not chosen because of difficulty in marking the fingerlings. A larger sample size of all age-3 largemouth bass also would have added to the power of our data set. Unanticipated difficulty in collecting 3 year olds could have been avoided by total sampling (i.e. draining and rotenone renovation) of each pond. This was not considered due to the private ownership of each pond site.

Changes in allele frequencies of largemouth bass fingerlings over time will provide direct information on what genotypes are most successful in each region. As successive generations are added to the database, our power in detecting a shift in allele frequencies due to selection will

grow.

Reported genetic data for 1994 fingerlings of the Wateree strain may be incorrect and therefore was not used in comparing filial generations with original stocks. The presence of a rare allele, sIDHP-2\*142, in the 1994 stocking of Lake Wateree bass was of greatest concern. No juveniles produced from that stock in 1995-1996 have been found to possess this rare allele. The survey of Wateree parental stocks also found no individuals that possessed the rare allele. Experts will be consulted for help in determining how this potential discrepancy should be treated in further data analysis. Genetic comparison of Wateree fingerlings produced in both 1994 and 1995 illustrates the need to ensure as many parents as possible are contributing to hatchery stocks.

#### Recommendations

Continue study. Complete age estimates for adults collected in 1998 and repeat analysis of differences in growth at age-3. Complete genetic analysis of 1997 and 1998 progeny from each pond and compare to original stocks.

Implement standard hatchery procedures that are aimed at maximizing genetic diversity and minimizing unnatural selection. These would include maximizing the number of parents contributing to each year class produced, and avoiding inbreeding events by regular collection of wild broodstock and not adding hatchery produced fish to the broodfish pool.

Prepared by: Jean Leitner

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## JOB PROGRESS REPORT

STATE: South Carolina

PROJECT NUMBER: F- 63

PROJECT TITLE: Fisheries Investigations in Lakes and Streams - Statewide

STUDY: Survey and Inventory  
Statewide

STUDY TITLE: Fishery surveys -  
Fisheries

Research

JOB NO: IIB  
performance of two  
strains of largemouth bass in State Lakes  
Introduction

JOB TITLE: Relative

Two subspecies of largemouth bass *Micropterus salmoides*, the Florida largemouth bass *M. s. floridanus* and the northern largemouth bass *M. s. salmoides*, exist and readily interbreed in both hatchery and reservoir environments (Isely et al., 1987, Gilliland and Whitaker 1989, Philipp and Witt 1991). The native range of the Florida subspecies (FLMB) is restricted to peninsular Florida. The northern subspecies (NLMB) is native to waters north along the Atlantic coast states from Maryland and west to the Mississippi (Philipp et al., 1983).

South Carolina is located in the broad hybrid zone between the ranges of the two subspecies. A statewide allozyme study of largemouth bass confirmed that South Carolina populations were hybrids (Bulak et al., 1995). This study also showed the existence of a geographic cline within South Carolina where the relative abundance of alleles typical of the Florida subspecies decreased from southeast to northwest. The relative frequency of alleles that are fixed for the Florida subspecies ranged from 98% in Lake Moultrie, a Coastal Plain reservoir, to 36% in Lake Wateree, a Piedmont reservoir. It was suggested that natural selection played a role in maintaining this allelic cline.

Physiological and ecological differences among FLMB, NLMB, and their hybrids have

been documented. A number of studies have shown a difference in the response of the FLMB, NLMB, and their hybrids to various temperature regimes (Fields et al., 1987, Charmichael et al., 1988). Other studies have shown differences in timing of spawning, growth rate, reproductive success and survival of the two subspecies (Philipp and Witt 1991, Maceina et al. 1988, Gilliland and Whitaker 1989, Isely et al. 1987).

The objective of this study was to examine performance differences between Lake Wateree and Lake Moultrie genetic strains of largemouth bass found in South Carolina. Two newly renovated state owned lakes, Wallace and Sunrise, were stocked with largemouth bass fingerlings from each strain. Strains were produced on separate hatcheries from broodfish collected from Lakes Wateree and Moultrie. Each strain received either a single or double oxytetracycline mark prior to stocking. Lakes Wallace and Sunrise were stocked with equal proportions of each strain. The objective will be achieved by measuring growth of stocked bass at age-1 and age-3 and by monitoring the long term temporal change in juvenile genotypes.

### Methods

Sunrise Lake, a 20 acre lake in Lancaster County, and Lake Richard B. Wallace, a 280 acre lake in Marlboro County, were renovated during the summer of 1996. Largemouth bass for experimental stockings were produced from adult bass collected from Lakes Moultrie and Wateree. Lake Moultrie broodfish were collected by electrofishing in March of 1993 and were housed separately from other stocks at Cheraw State Fish Hatchery. Lake Wateree broodfish were collected in early Spring of 1997 and transported to Cohen Campbell Fisheries Center where they were stocked directly into a spawning pond separate from other stocks. Each group of broodfish was allowed to spawn. Resulting fry were harvested from as many schools as possible to maximize the number of parents contributing to the gene pool, and were grown out to

fingerlings.

Prior to stocking fingerlings from each strain were marked by immersion for 6 hours in a 500 ppm solution of oxytetracycline. Moultrie strain largemouth bass were double marked, first on 4/16/97 as fry, and then on 5/5/97 as fingerlings. Wateree strain largemouth bass were single marked as fingerlings on 4/25/97.

Each lake was stocked with equal numbers of each strain at the rate of 100 fish per acre in April and May of 1997. Lake Wallace was stocked with 28,000 and Sunrise Lake with 2000 largemouth bass. (Lakes were stocked in October 1996 with a combination of bluegill *Lepomis macrochirus* and redear *L. microlophus* fingerlings at the rate of 1000 per acre.) Wateree strain fingerlings were stocked on 4/25/97. Moultrie strain fingerlings were stocked on 5/5/97. Total lengths were recorded for a sample of 100 fingerlings from each strain at time of stocking. One hundred additional fingerlings from each strain were transported to the Berry's Mill Hatchery near Traveler's Rest and held in separate ponds for use in mark evaluation and genetic analysis.

Ponds at Berry's Mill were harvested on 11/6/97 and sagittal otoliths, liver, and muscle tissue were collected from each individual. Known single and double marked otoliths were randomly coded and given to an experienced reader for evaluation. Otoliths were mounted, sectioned and polished to the core. Presence or absence of a mark on the otolith was determined with a fluorescent compound microscope.

Liver and muscle tissues were stored at -80°C for genetic analysis. Horizontal starch gel electrophoresis was performed according to Norgren (1986). Gels were stained for four enzymes which are diagnostic for the Florida and northern subspecies of largemouth bass. These are aspartate aminotransferase (sAAT-2\*), isocitrate dehydrogenase (sIDHP-1\*) and superoxide

dismutase (sSOD-1\*) from liver tissue, and malate dehydrogenase (sMDH-B\*) from muscle tissue. Alleles typical of the northern subspecies are sAAT-2\*100 and sAAT-2\*110, IDHP-1\*100, sMDH-B\*100, and sSOD-1\*147. Alleles typical of the Florida subspecies are sAAT-2\*126 and sAAT-2\*139, sIDHP-1\*121, sMDH-B\*114, and sSOD-1\*100. A genetic baseline was determined for Lakes Moultrie and Wateree using data from an initial statewide survey (Bulak et al., 1995) and data collected from large and small fish for a related performance study. Allele frequencies of each stock was compared to baseline genetic data for source populations using the G-test (Sokal and Rohlf, 1969).

Lakes were sampled in the Spring and Summer of 1998 for collection of juveniles and age-1 adults. Adults were collected by electrofishing from Lake Wallace on March 31 and April 4, and from Sunrise Lake on May 22. Total length and weight were recorded for each individual. Sagittal otoliths were collected from each largemouth bass and stored in the dark until processed for mark determination. Liver and muscle tissues were collected from each individual and stored at -80°C until processed for genetic analysis. Seining for juveniles was conducted on Lake Wallace May 19 and on Sunrise Lake May 22. A variety of areas and habitats were sampled.

## Results

Size at stocking was similar for the Moultrie and Wateree strains. Moultrie strain fingerlings averaged 24.4 mm total length (n = 102, std = 2.6). Wateree strain fingerlings averaged 23.3 mm total length (n = 92, std = 6.2).

Mark evaluations were completed on a set of 68 otoliths. Because of questionable origin made evident by genetic analysis, 8 sets of otoliths were thrown out. Of 27 Wateree strain fish 100% were correctly identified. Of 33 Moultrie strain fish 91% were correctly identified.

Genetic analysis was completed for hatchery fingerlings of each strain, and comparisons made with historic data from wild stocks (Table 1.). Fingerlings of the Wateree strain were similar to the wild Wateree stock at three of four loci. However, at the sIDHP-1\* locus the Wateree strain fingerlings possessed significantly ( $p=0.05$ ) more of the sIDHP-1\*100 allele which is typical of the northern subspecies. Fingerlings of the Moultrie strain differed markedly from wild lake Moultrie stock at three of the four loci examined. They possessed significantly more of the sAAT-2\*100,110 alleles, the sIDHP-1\*100 allele, and the sMDH-B\*100 allele, all typical of the northern subspecies.. Fingerlings of the Moultrie strain possessed sMDH-B\*100 at a frequency of 20% although broodstock from Lake Moultrie were known to be fixed for sMDH-B\*114.

Table 1. Allele frequencies (proportions) for largemouth bass used to stock study lakes, with historic data for reservoirs where stocks originated. A + indicates allele frequencies significantly different from survey data.

<u>Lake Moultrie</u>		<u>Lake Wateree</u>		
<u>Locus/Allele</u>	<u>Historic Data</u>	<u>1997 Stocking</u>	<u>Historic Data</u>	<u>1997 Stocking</u>
<u>sAAT-2*</u>				
100, 110	146 (0.66)	26 (0.69)	47 (0.10)	16 (0.23) +
126, 139	74 (0.34)	12 (0.31)	443 (0.90)	54 (0.77) +
<u>sIDHP-1*</u>				
100	116 (0.48)	37 (0.69) +	11 (0.02)	12 (0.16) +
121	124 (0.52)	17 (0.31) +	455 (0.98)	64 (0.84) +
<u>sMDH-B*</u>				
100	141 (0.61)	39 (0.70)	0 (0.00)	16 (0.20) +
114	91 (0.39)	17 (0.30)	494 (1.00)	64 (0.80) +
<u>sSOD-1*</u>				
147	143 (0.57)	29 (0.54)	82 (0.19)	17 (0.24)
100	107 (0.43)	25 (0.46)	344 (0.81)	55 (0.76)

Largemouth bass adults were collected by electrofishing from Lake Wallace on 4/31/98 and 5/22/98. Fish averaged 274.1 mm total length ( $n = 104$ ,  $std = 28.2$ ) and weighed an average of 359.3 g ( $n = 104$ ,  $std = 123.5$ ). Largemouth bass adults were collected from Sunrise Lake on 5/22/98. These fish averaged 235.7 mm total length ( $n = 92$ ,  $std = 17.3$ ) and weighed an average of 171.7 g ( $n = 92$ ,  $std = 49.8$ ). Despite efforts to sample a variety of areas and habitats, no juvenile largemouth bass were collected from either lake.

### Discussion

The marked genetic difference between Moultrie strain fingerlings and Lake Moultrie broodfish is a concern, especially at the sMDH-B\* locus. It indicates that not all of the fingerlings stocked as Moultrie strain were produced from Lake Moultrie broodfish.

When they were collected in 1993 all Lake Moultrie broodfish underwent liver and muscle biopsies. Tissues were analyzed so that the alleles expressed at each loci for every fish was known. None of 112 fish biopsied possessed the sMDH-B\*100 allele. Eight out of 40 Moultrie strain fingerlings were homozygous for sMDH-B\*100 meaning they inherited that allele from both parents. All other fingerlings were homozygous for sMDH-B\*114. The presence of the northern allele and lack of heterozygotes indicate that the fish possessing the northern allele were spawned in a different pond and from a group of parents other than the Lake Moultrie broodfish.

Fish possessing the sMDH-B\*100 allele also possessed a different oxytetracycline mark from other Moultrie fingerlings. Moultrie fingerlings were marked twice, first as fry when harvested from the spawning pond, and then as fingerlings when taken from the hatchery for stocking. All eight of the fish homozygous for sMDH-B\*100 had only the later mark.

There are three possible explanations for the presence of the fish homozygous for sMDH-B\*100. The first is that the Moultrie strain fingerlings were contaminated on the



hatchery. This would have occurred sometime after the marking of fry but prior to the second marking, with the source of contamination either in the grow out pond or the fish house.

A second explanation is that the Moultrie strain fish were contaminated in the holding pond at Berry's Mill with fish of the single marked Wateree strain. The two strains were housed in adjacent ponds separated by an earthen dike. A third explanation is that the samples collected from Berry's Mill were mishandled and some Wateree strain fish were improperly coded as Moultrie strain. The probability that 8 fish chosen at random from the Wateree strain will all be homozygous for sMDH-B\*100 is  $P = 0.002$ .

If the Moultrie strain fingerlings were in fact contaminated prior to stocking, the effects on the experiment can be assessed. Our experimental design called for the lakes to be stocked with equal proportions of each strain. Performance would be assessed by measuring growth of stocked fish at age-1 and age-3, and by the long term monitoring of allele frequencies of subsequent year classes.

In fact, the lakes were stocked with 50% Wateree strain fingerlings, 30% Moultrie strain fingerlings, and 20% fingerlings of unknown origin. Because the fingerlings of unknown origin are single marked, in future collections they will be indistinguishable from fish of the Wateree strain. Growth assessments of the Wateree strain will include those fish of unknown origin. Assessment of reproductive success of the Moultrie and Wateree strains by following changes in allele frequencies of subsequent generations will be difficult because of the unbalanced stocking, and the inability to quantify the contribution of the unknowns.

While these factors negatively impact our ability to draw conclusions regarding the performance of the Moultrie and Wateree strains, valuable information can still be obtained. Genetically the 8 unknown fish are similar to the Wateree strain. Though as a group they possess

more northern alleles, individually they are not distinguishable from a Wateree strain fish.

Growth can still be compared between the Moultrie strain and the more northern, single marked fish.

Largemouth bass in Sunrise Lake grew much slower in their first year than those in Lake Wallace. While no water quality measurements were taken a visual inspection of the two lakes indicated they were managed quite differently. Lake Wallace appeared to have received more than adequate fertilizer applications; it was deep green with no visibility below the surface in some areas. Sunrise Lake was very clear throughout. If fertilizer applications were made at Sunrise Lake they were not effective. Both of these lakes were stocked at the fertilized rate of 1000 bream/100 bass per acre.

#### Recommendations

Continue study. Consult with hatchery personnel to determine if the source of contamination of the Moultrie strain can be identified. Process otoliths collected from 1998 age-1 largemouth bass. Compare growth at age-1. Ensure that all state lakes are managed optimally with regard to liming and fertilization regimes.

Prepared by: Jean Leitner

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## JOB COMPLETION REPORT

STATE: South Carolina

PROJECT NUMBER: F- 63

PROJECT TITLE: Fisheries Investigations in Lakes and Streams - Statewide

STUDY: Survey and Inventory  
Statewide

STUDY TITLE: Fishery surveys -  
Fisheries

Research

JOB NO: IIC  
evaluation of trophy

JOB TITLE: Genetic  
largemouth bass

### INTRODUCTION

Two subspecies of largemouth bass *Micropterus salmoides*, the Florida largemouth bass *M. s. floridanus*, and the northern largemouth bass *M. s. salmoides*, exist and readily interbreed in both hatchery and reservoir environments (Isely et al., 1987, Gilliland and Whitaker 1989, Philipp and Whit 1991). These subspecies readily interbreed in a natural zone of intergradation along the Atlantic coast from northern Florida through North Carolina (Philipp et al. 1983). The native range of the Florida subspecies (FLMB) is restricted to peninsular Florida. The northern subspecies (NLMB) is native to waters north along the Atlantic coast states from Maryland and west to the Mississippi (Philipp et al., 1983).

Many studies have compared the performance of the two subspecies and their hybrids. Differences in growth and survival rates (Philipp and Whitt 1991), timing of spawning (Isely et al. 1987, Maceina et al. 1988), oxygen uptake, and responses to controlled temperature regimes (Fields et al., 1987, Carmichael et al., 1988) were documented. Maceina et al. (1988) stocked a Texas lake with both subspecies and found that while the northern subspecies attained larger size at age-1, by age-3 the Florida subspecies possessed a size based fecundity advantage. In Illinois Philipp and Whitt (1991) compared each subspecies and their F<sub>1</sub> hybrids and found that the largemouth bass of the northern subspecies native to the area exhibited faster growth and better overwinter survival than the non-native Florida subspecies. The F<sub>1</sub> hybrids were intermediate between the two subspecies with respect to both growth and survival.

Genetic differences between the two subspecies are measurable at four diagnostic enzyme

loci (Philipp et al., 1983). The differences at two loci, aspartate aminotransferase (sAAT-2\*) and isocitrate dehydrogenase (sIDHP-1\*), are fixed meaning one allele or combination of alleles is present only in populations of the Florida subspecies and the other only in populations of the northern subspecies. At a third locus, malate dehydrogenase (sMDH-B\*), Florida populations are fixed for a Florida allele, while northern populations may be fixed for a northern allele or possess a combination of northern and Florida alleles. At the fourth diagnostic locus, superoxide dismutase (sSOD-1\*), northern populations are fixed for the northern allele while Florida populations possess a combination of the northern and Florida alleles. The northern and Florida alleles at each enzyme locus are listed in Table 1.

South Carolina is located in the broad hybrid zone between the ranges of the two pure subspecies. A statewide allozyme study of largemouth bass showed the existence of a geographic cline within South Carolina where the relative abundance of alleles typical of the Florida subspecies decreased from southeast to northwest (Bulak et al., 1995). The relative frequency of alleles that are fixed for the Florida subspecies at sAAT-2\* and sIDHP-1\* ranged from 98% in Lake Moultrie, a Coastal Plain reservoir, to 36% in Lake Wateree, a Piedmont reservoir. It was suggested that natural selection, acting to maximize the fitness, or reproductive efficiency, of individuals within each population, played a role in maintaining this cline. Rate of growth is a life history characteristic that is responsive to natural selection.

The objectives of this study were to compare the allele frequencies of a) large and small size classes, and b) young and old age classes within three South Carolina largemouth bass populations. Growth rates of the three populations were also compared.

## **METHODS**

Three populations were chosen from the original survey based on their genetic makeup and geographic location. Lake Moultrie is in the Coastal Plain and possessed the highest percentage (98%) of Florida alleles. Lake Murray, in the central region of the state, possessed about 76% Florida alleles. Lake Wateree, a Piedmont reservoir, possessed about 36% Florida alleles.

Largemouth bass in the small size class were generally less than 1.2 Kg. They were collected by electrofishing from all three populations from March 1993 to November 1993 (Table 2). Fish were either kept alive or held on ice for up to six hours. At the lab length and weight

were recorded for each fish. Liver and muscle tissue were dissected and immediately frozen and stored for genetic analysis. Sagittal otoliths were extracted and stored for aging.

Table 1. Northern and Florida alleles at four enzyme loci diagnostic for the northern and Florida subspecies of largemouth bass.

	<u>Locus</u>			
	<u>sAAT-2*</u>	<u>sIDHP-1*</u>	<u>sMDH-B*</u>	<u>sSOD-1*</u>
Northern allele(s)		sIDHP-1*100		sSOD-1*147
	sAAT-2*100		sMDH-B*100	
	sAAT-2*110			
Florida allele(s)		sIDHP-1*121		sSOD-1*100
	sAAT-2*126		sMDH-B*114	
	sAAT-2*139			

Table 2. Number of largemouth bass collected by method for each population.

<u>Population</u>	<u>Method</u>	<u>No. collected</u>
Moultrie	electrofishing	201
	taxidermist	57

Murray	electrofishing	87
	taxidermist	34
Wateree	electrofishing	138
	taxidermist	0

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Largemouth bass in the large size class were obtained from taxidermists and were generally 3.17 kg or greater. Participating taxidermists recorded length, weight, and date caught for these fish from Lakes Moultrie and Murray. They collected liver, muscle, and sagittal otoliths. Frozen samples and accompanying data were picked up periodically. Initial results from Lake Wateree taxidermists indicated that fish > 3.17 kg were rarely turned in. For this reason, the weight minimum for large fish was dropped to 1.60 kg on Lake Wateree. Fish in that size class were collected by electrofishing in March of 1994.

The data base for the comparison of large and small fish was augmented with information from fish collected for two previous efforts. Length, weight, and genetic data were obtained from the initial 1991 survey for all three populations, and from Lake Moultrie fish that were collected in 1993 to form a broodstock of known genotype (Bulak et al. 1995). Otoliths were not collected from either of these groups, so these fish were not included in the comparison of age classes.

All otoliths collected were either aged whole, or sectioned and polished. Annual growth rings were counted to determine age estimates. Otoliths were read by two independent readers. When age disagreement occurred, otoliths were read again. Otoliths for which the age could not be agreed on were not included in analysis.

Allele frequencies at the four diagnostic enzyme loci were determined for each individual using a horizontal starch gel electrophoresis system and histological stains as described and cited by Norgren (1986). Some individuals in the data set (N=17) possessed a rare allele, sIDHP-1\*142. These individuals were not included in analysis involving the IDHP-1\* locus. Differences in allele frequencies at each of the four loci, as a function of size (all specimens in

database) and age (aged members only) were evaluated for each population using the G-test, a  $\log(e)$  transformation of the chi-squared test (Sokal and Rohlf 1969). To account for the multiple G-tests run on each data set, the sequential Bonferroni test (Holm 1979) was applied to G-test results.

Growth differences among populations were evaluated by calculating Von Bertalanffy growth equations (Ricker 1975) for each population. Mean length at age and the accompanying standard error were also calculated (aged members only).

## RESULTS

A total of 516 largemouth bass were collected. Table 3 shows number collected by size class and number aged for each population.

Table 3. Number of largemouth bass collected by size class, and number aged, for each population.

<u>Population</u>	<u>Size class (kg)</u>	<u>No. collected</u>	<u>No. aged</u>
Moultrie	$\leq 1.20$	77	55
	$\geq 3.18$	55	55
Murray	$\leq 1.20$	82	51
	$\geq 3.18$	26	26
Wateree	$< 1.60$	108	69
	$\geq 1.60$	30	29



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Growth differences were found among the three study reservoirs. Largemouth bass from the Lake Moultrie population grew faster than those from Lake Murray or Lake Wateree (Figure

1). The following Von Bertalanfy growth equations were derived for each population:

Wateree: Total length, mm =  $580 (1 - e^{-0.191(t + 2.67)})$  ;  $R^2 = .66$ , N = 98

Murray: Total length, mm =  $702 (1 - e^{-0.139(t + 2.86)})$  ;  $R^2 = .79$ , N = 86

Moultrie: Total length, mm =  $759 (1 - e^{-0.177(t + 1.47)})$  ;  $R^2 = .85$ , N = 114

where, t = age(years).

Figure 1. Comparison of Von Bertalanfy growth function for three largemouth bass populations in South Carolina.

Allele frequencies were calculated for each reservoir (Table 4). As anticipated, Lake Moultrie had the greatest percentage of Florida alleles, 94%, at the two fixed loci sAAT\*-2 and IDHP\*-1. Lake Murray had 79% Florida alleles while Lake Wateree had 43% Florida alleles at those two loci.

One of 11 G-test analysis of allele frequencies as a function of size (Table 5) was significant ( $P=0.05$ ). For Lake Murray, comparison of allele frequencies at sSOD-1\* showed that fish in the large size class possessed the sSOD-1\*100 allele, which is present only in the Florida subspecies, in greater proportion than fish in the small size class. However, application of the sequential Bonferonni test showed that this individual G-test result was not significant when all 11 tests were taken into account. One of 11 G-test analysis of allele frequencies as a function of age (Table 6) was also significant ( $P=0.05$ ). For Lake Wateree, fish  $\geq$  age 7 possessed the Florida allele sIDHP-1\*121 in significantly greater proportion. Again, application of the sequential Bonferonni test showed that this G-test result was not significant when all 11 tests were taken into account.

Table 4. Largemouth bass allele frequencies at four enzyme specific loci. Alleles denoted by 'n' are either fixed (sIDHP\*-1 and sAAT\*-2) or dominant in the northern subspecies; alleles denoted by 'f' are either fixed or dominant in Florida subspecies. The sIDHP\*-1 allele denoted by 'r' is a rare allele not dominant in either subspecies.

<u>Frequencies (N)</u> <u>Locus/Allele</u>	<u>Allele</u>		
	<u>Moultrie</u> (N=258)	<u>Murray</u> (N=121)	<u>Wateree</u> (N=138)
<u>sAAT*-2</u>			
100 (n)	0.08	0.17	0.55
114 (n)	0.02 (N=245)	0.06(N=109)	0.11 (N=110)
126 (f)	0.24	0.29	0.14
139 (f)	0.66	0.48	0.20
<u>sIDHP*-1</u>			
100 (n)	0.02	0.18	0.47
114 (f)	0.96 (N=241)	0.79(N=113)	0.52 (N=122)
142 (r)	0.02	0.03	0.01
<u>sMDH*-B</u>			
100 (n)	0.00 (N=247)	0.10(N=102)	0.61 (N=116)
114 (f)	1.00	0.90	0.39
<u>sSOD*-1</u>			
100 (f)	0.81 (N=213)	0.59(N=105)	0.43 (N=125)
147 (n)	0.19	0.41	0.57

Table 5. Frequencies of northern (N) and Florida (F) alleles at four enzyme loci for two size classes of largemouth bass from Lakes Wateree, Moultrie and Murray with accompanying G-test statistic (G). A  $\chi^2$  value of 3.84, one degree of freedom is significant at  $P=0.05$  (\*).

<u>sMDH*-B</u>	<u>Size</u> <u>(Kg)</u>	<u>sSOD*-1</u>		<u>sAAT*-2</u>		<u>sIDHP*-1</u>			
		<u>N</u> <u>G</u>	<u>F</u>	<u>N</u> <u>G</u>	<u>F</u>	<u>N</u> <u>G</u>	<u>F</u>	<u>N</u> <u>G</u>	<u>F</u>
Wateree	<1.60	104	58	91	97	111	77	110	80
			1.32		0.00		1.28		0.16
	≥1.60	42	16	25	27	30	14	33	27
Moultrie	≤1.20	33	265	7	285	0	316	56	242
			0.50		0.65		0.00		0.73
	≥3.18	9	95	4	96	0	94	6	38
Murray	≤1.20	31	99	24	112	11	111	68	74
			0.01		2.72		1.26		4.71*
	≥3.18	12	40	13	31	7	39	8	22

Table 6. Frequencies of northern (N) and Florida (F) alleles at four enzyme loci for different age classes of largemouth bass. Largemouth bass  $\leq$  age 3 are compared with fish  $\geq$  age 7, by reservoir. Accompanying G-test statistics are listed (G). A  $X^2$  value of 3.84, one degree of freedom, is significant at  $P=0.05$  (\*).

Reservoir	Age	sAAT*-2			sIDHP*-1			sMDH*-B			sSOD*-1		
		N	F	G	N	F	G	N	F	G	N	F	G
Wateree	$\leq 3$	56	40		62	40		62	36		51	47	
				1.61						0.03			0.03
						4.50*							
	$\geq 7$	16	6		7	13		11	7		11	11	
											11		
Moultrie	$\leq 3$	11	73		1	95		0	96		68	16	
				3.17						NA			0.09
						1.25							
	$\geq 7$	5	87		3	85		0	82		30	6	
Murray	$\leq 3$	16	60		14	62		5	73		31	45	
				0.21						3.67			1.54
						1.30							
	$\geq 7$	16	50		15	41		10	50		13	31	

## **DISCUSSION**

Largemouth bass grew at different rates in each of the three study reservoirs. Especially interesting is the slower growth of the Lake Wateree population. Lake Wateree is one of the most productive reservoirs in South Carolina. Largemouth bass grow more slowly there than in Lake Moultrie or Murray, despite good habitat and an abundance of prey. Genetic or growing season differences among the three populations may account for the observed variation in growth rates. The Lake Moultrie population is in a warmer climate, and possesses more Florida alleles than the Lake Wateree population. The Lake Murray population is intermediate between the two with respect to both genetics and climate.

Analysis of allele frequencies as a function of age and of size in each population revealed one difference at  $P=0.05$  for each data set. However, it is important to take into account the number of tests run simultaneously. In so doing we find no difference between the trophy and smaller/younger segments of any of the three populations studied. Attainable size and longevity are two of many measures of a fishes performance. If certain fish in each study population possess a genetic trait, or combination of genetic traits, that give them an advantage over others with regard to attaining trophy size, it was not measurable by our data set.

## **RECOMMENDATIONS**

This study provided no direct evidence that 'trophy' bass were genetically distinct from smaller or younger members of the same population. However, differences may still exist. We looked at only four genetic markers, and only at the enzyme level. Any time that hatchery produced stocks are employed to augment a population the genetic character of those stocks should be considered. Hatchery stocks should at least be similar to the genetic baseline of the receiving population, and preferably produced from brood fish collected from that population. This will help preserve genetic integrity of individual populations and ensure that any natural selection acting on individual populations, measurable or not, is not undermined.

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## STUDY COMPLETION REPORT

STATE: South Carolina PROJECT NUMBER: F-63

PROJECT TITLE: Fisheries Investigations in Lakes and Streams - Statewide

SECTION TITLE: Survey and Inventory

STUDY III: Largemouth Bass Management Guidelines Development

### Summary

During the project period July 1, 1997 - June 30, 1998 recent literature dealing with black bass management was reviewed and summarized. Fishery resource agencies in 15 Southeastern states were contacted to obtain their current black bass management strategies; responses were compiled. Historical data from South Carolina fisheries district were obtained and evaluated. Data analysis subroutines to characterize length/weight and age and growth data were prepared. Workshops on the standardization of spring electrofishing and the development of a conceptual model to provide an objective analytical framework for black bass management were held in Eastover. A standardized sampling protocol was developed, evaluated, and revised based on the evaluation. Datasets generated using the protocol were analyzed. A dynamic population model for Lake Thurmond was produced.

### Introduction

The importance of largemouth bass to sport fishing in South Carolina is well known. A survey of freshwater fishermen commissioned by the South Carolina Wildlife and Marine Resources Department (SCWMRD), predecessor of the South Carolina Department of Natural Resources (SCDNR), in 1990 found that 28% of all fishermen fished for largemouth bass (Logan, 1990). Of fishermen who targeted a particular species, 37% fished for largemouth bass. According to a national survey conducted by the U.S. Department of the Interior et al. (1993), approximately

50% of resident and non-resident fishermen in South Carolina fished for black bass, primarily largemouth bass, in 1991. Logan (1990) reported that 48% of survey respondents felt that SCWMRD should pay more attention to the management of largemouth bass, and significant numbers supported harvest restrictions as management options.

Considerable effort is expended annually by district fisheries biologists in South Carolina to monitor the status of largemouth bass populations in reservoirs and streams. Techniques for conducting angler creel surveys, spring electrofishing and summer/fall cove rotenone sampling were standardized to facilitate the analysis and interpretation of data. Kirk (1989) summarized a decision-making process regarding management options that could follow from evaluation of the harvest potential of largemouth bass, based on data generated from standardized surveys and sampling. However, there are no definitive guidelines that management biologists must follow when making management recommendations.

Birth, growth, and death are dynamic processes which operate continuously and interactively on populations of living organisms. Population structure, however it is measured or expressed, is the cumulative result of these processes (each actually a rate function) at any point in time. Structural indices (age structure, length structure, relative condition) provide snap-shots which help to characterize the status of a population, but rate functions (recruitment, growth, and mortality) are needed to assess the dynamics of a population.

Historical spring electrofishing in South Carolina consisted primarily of the collection of largemouth bass length and weight data. Such data was useful for the computation of two structural indices: length structure and relative condition. Inferences were often made about recruitment and mortality from length structure representations and about growth from relative condition representations. However, rate functions can be estimated meaningfully only if the time

step is known. Therefore, accurate and precise aging studies are essential elements of a sampling program.

In 1995 the Freshwater Fisheries Section of SCDNR approved a statewide management plan for black bass, including largemouth bass. Management goals were established to provide continuity and guidance to department personnel and the public, while the need for site-specific management authority was recognized. One goal common to all four species was to develop, maintain, and enhance the biological databases needed to make sound management decisions. Presently this agency does not have a centralized database management system in place for freshwater fisheries.

SCDNR recognizes the importance of collecting, maintaining, assessing, and archiving biological data from fish populations of recreational, commercial, or ecological value to the state. Such databases can be used to define management options appropriate in different situations, depending on the results of structured and objective assessment of a population. Having such guidelines would promote uniform, consistent assessments of largemouth bass populations, and could enhance public understanding of and support for the process of managing the fishery.

The objective of the present study is to develop a quantifiable protocol for identifying and ranking management options within a system through compilation, analysis, and interpretation of existing largemouth bass population data.

## Materials and Methods

### *Literature Review*

State fisheries chiefs in 15 agencies belonging to the Southeastern Association of Fish and Wildlife Agencies (SEAFWA) were asked to provide copies of black bass management plans and/or strategies developed for use in their states, along with any other information that might be

helpful in compiling a summary of black bass management practices in the Southeast. Responses were evaluated to determine what states have developed statewide black bass management plans, which maintain statewide fisheries databases, which agencies have regulatory authority over the black bass fishery (and at what level), and which have written protocols or guidelines that reservoir management biologists must follow. States which have written guidelines for the preparation of reservoir management plans or reports, and those which follow standardized sampling procedures, bass population assessment methods, or decision-making criteria were noted. Responses were summarized and tabulated for review.

The current body of published literature on black bass management, including state reports, when available, was reviewed. Articles which dealt with promising quantitative approaches to management were of particular interest. Those which were considered to be most useful in advancing the objectives of the present study were summarized and included in an annotated bibliography.

#### *Database Compilation*

Each district was asked to provide one or more digital files of spring electrofishing data to show which data were captured and how they were structured and archived. After preliminary inspection of files, data were extracted into permanent SAS<sup>®</sup> datasets with standardized variable names to facilitate further analysis.

A standardized data-entry program written in Paradox<sup>®</sup> was distributed to fisheries districts in 1997 as part of a new spring electrofishing sampling protocol (SSP), described in a later section of this report. A run-time version of Paradox<sup>®</sup> was also distributed so that districts that did not otherwise use that database software could use the data-entry program. The program allowed fish and environmental data entered in a local database to be exported as digital text files on a floppy

disk and sent to the SCDNR Freshwater Fisheries Research Project in Eastover for incorporation into a centralized database. Data thus obtained from the districts were extracted, summarized, and analyzed using SAS<sup>®</sup> and Excel<sup>®</sup>.

### *Data Analysis*

A SAS<sup>®</sup> program was developed to compute age-frequency distributions in a population from an age-length key of a subsample of that population, based on an example in DeVries and Frie (1996). Modifications in the program permitted classification variables such as sex and lake zone to be taken into account when evaluating age-frequency distributions. The term "age-frequency distribution" is used in this report instead of "length-frequency distribution by age group", which might be more descriptive of the variables involved, because the former term has a history in fisheries literature (DeVries and Frie, 1996).

Largemouth bass data collected by the districts in accordance with the SSP included ages obtained from subsampled otoliths in 25-mm groups from 175 to 474 mm total length (TL). Age-length keys were developed from the subsamples and age-frequency distributions were computed for each population. Age-frequencies were assigned using age-length keys developed from 1997 samples. Fish <175 mm total length were assumed to be age-1.

Catch per unit effort (CPUE) of age-1 largemouth bass collected from each sampled reservoir during spring electrofishing in 1997 was used as an index of recruitment for that reservoir. This method provided a relative, rather than an absolute, estimate. Differences in age-1 CPUE between zones within a lake provided a measure of the inherent variability of recruitment. Historic data was evaluated to determine if variability in age-1 CPUE between years could be estimated for one or more reservoirs. Because effort data were not routinely included with sampling data prior to the development of the SSP, it was not possible to calculate historic age-1

CPUE values for most reservoirs, even if aging data existed. Spring electrofishing effort data were available for District 2 reservoirs beginning in 1994, however. Since sampling strategies followed in District 2 before and after 1997 were similar, except for the imposition of zones by the SSP, whole-lake CPUE by age computed for Lake Thurmond largemouth bass sampled in 1994-1996 could be compared to the value computed in 1997.

Mean and standard deviation of length at age were computed from the age-frequency distribution for each cohort (Steele and Torrie, 1960). If the age-frequency distribution for a cohort was truncated because of the 474-mm upper length limit of fish aged, the mean was considered biased. Means were computed by zone to see if there were zonal differences in growth within reservoirs.

Von Bertalanffy growth parameters for largemouth bass were estimated for each reservoir, and for zones within reservoirs, from the subset of aged fish. A SAS<sup>®</sup> program using PROC NLIN was written to generate the estimates. To improve the fit of the model and increase the sample size, unaged fish <175 mm were assumed to be age-1. Following the recommendation of Beamesderfer and North (1995),  $t_0$  (time when length was zero) was standardized at -0.024 year.

Mortality estimates were computed by regression analysis of  $\log_e$ -transformed CPUE-at-age data. Two analytical methods, based on different recruitment assumptions, were used to obtain estimates. In the first method, annual recruitment of largemouth bass in a reservoir was assumed constant, so a single regression was performed that integrated CPUE-at-age for all available sample years. This method produced a value representing the mean instantaneous total mortality rate  $Z$  of the included age classes during the years sampled. Total annual mortality  $A$  was then calculated as  $1-S$ , where  $S$  (total annual survival) =  $e^{-Z}$ . In the second method, annual recruitment was assumed to be variable. Regression analysis of CPUE on age was performed on

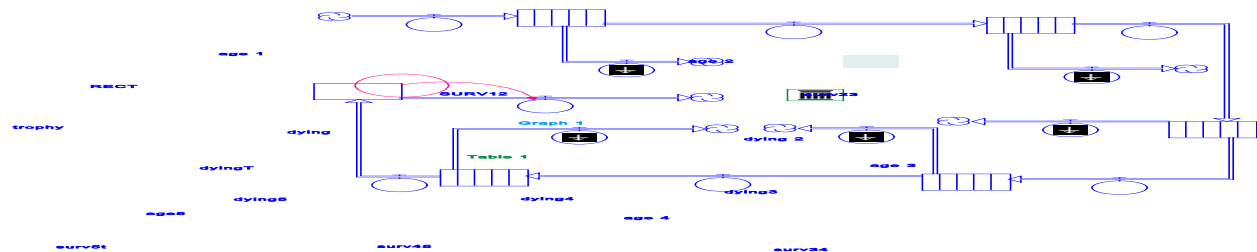
spawning cohorts for which data were available for the appropriate age classes.

### *Data Interpretation*

Workshops were held with district biologists in 1997 and 1998 to discuss black bass management objectives and sampling strategies, in order to develop and finalize an acceptable sampling protocol for spring electrofishing. The SSP developed for use in spring 1997, was modified on the basis of input received during the year. The modified version was distributed for use in 1998.

A population model was constructed using STELLA<sup>®</sup> (Stella II Authoring Version 3.07 for Windows, High Performance Systems, Inc., Hanover, NH) software as a tool for evaluating the interactions of recruitment, growth, and mortality and their influence on population size structure. The model was used interactively by management biologists during a workshop held in February, 1998, to evaluate the proposed modeling strategy. A flow diagram of the model is shown in Figure 1. Recruitment (RECT in Figure 1) was the model input, randomly assigned each annual time step from a normal distribution defined by the mean and associated variance of recruitment over the period of record. Annual mortality *A* was the test variable. Model output was a simulation of the abundance of age-classes 1 through 5, at given levels of total annual mortality.

Figure 1. Flow diagram of the STELLA<sup>®</sup> model used interactively to evaluate the proposed largemouth bass management strategy. Mortality acted on recruitment (RECT) in four multi-year simulations. Output was expressed as mean abundance of age classes 1-5. Abundance values were exported to a SAS program for individual assignments of length.



Values were passed to a SAS program which randomly assigned lengths to individuals in the simulated population from a normal length distribution based on observed mean length at age and the associated variance of a sampled population. The model was tested using Lake Thurmond population data. Each SAS output of population structure (i.e. length frequency) was compared to the observed length frequency of the Lake Thurmond population in 1997 using the Kolmogorov-Smirnov test ( $P = 0.01$ ). Size of the simulated populations was held approximately the same as the Lake Thurmond sample size in all runs to facilitate statistical comparison among groups.

## Results and Discussion

### *Literature Review*

A survey of 15 state natural resource agencies belonging to the Southeastern Association of Fish and Wildlife Agencies provided information about the current status of black bass management in the southeast. The broad spectrum of responses to questions regarding the existence of statewide management plans and databases, as well as guidelines or protocols for writing reservoir management plans, conducting sampling, assessing populations, writing reports,



and making management decisions indicated that the philosophy of management is still in flux but moving toward standardization in some areas. Survey results were summarized and are attached to this document as Appendix A (Part I). South Carolina was included for comparison. A notable finding was that only South Carolina among responding states does not delegate to its natural resource agency the authority to regulate the harvest of black bass through creel or length limits, either statewide or on a site by site basis (Table A-1).

Available current literature from published and unpublished sources indicated that key areas of concern with respect to black bass management include data collection and analysis, sampling methods, age and growth analysis, length-weight relationships, population dynamics, harvest potential, and harvest restrictions. Important findings in each of these areas were compiled and summarized. Beamesderfer and North (1995) synthesized a large body of North American black bass data and produced simulations that allowed them to predict the sensitivity of population characteristics such as yield, harvest, and relative abundance to changes in exploitation rate and length limits. Productivity of the population was a key factor in determining that sensitivity. An annotated literature review is attached as Appendix A (Part II).

#### *Database Compilation*

Twenty-eight spring electrofishing datasets compiled from 1986 through 1996 were obtained from five fisheries districts and evaluated. They represented one to seven years of sampling on 10 reservoirs (Table 1). Species and total length (mm) were the only variables included consistently in those datasets, though most usually included weight (g), collection date and a sample site identifier. Age and sex of subsampled fish were recorded from three reservoirs in 1995 and one reservoir in 1996. Sampling effort was not recorded prior to 1997, limiting the utility of older datasets in the analyses which follow. The existence of additional datasets was inferred

from annual progress and completion reports prepared by district personnel; however, not all datasets were available for review.

Datasets derived from samples collected using a standardized sampling protocol (SSP) developed in 1997 were obtained and evaluated. These included datasets from seven reservoirs in four districts in 1997 and seven reservoirs in three districts in 1998 (Table 1). Data recorded under the new protocol included information on individual fish [species, total length, weight, age (if needed for growth determination), and sex (if fish were sacrificed)], as well as general environmental information about the collection (zone, sample site, collection date, water temperature, conductivity, Secchi disk transparency, lake level, and sampling effort). Environmental data was not consistently collected between districts or years (Table 1).

Reservoirs with the most extensive databases included Lakes Thurmond and Secession in

Table 1. Black bass data sources evaluated for inclusion in a statewide database, by fisheries management district, reservoir, and year. Presence of species, total length, weight, age, and sex information in a dataset is signified by “+”. Other variables recorded include zone, sample site, individual fish identifier (ID), date (or month day year [MDY] separately), water temperature (Temp), Secchi disk transparency, specific conductivity (Cond), lake level, and sampling effort.

Dist	Reservoir	Year	Species	TL	Wt	Age	Sex	Data	Other
2	Hartwell	1994	+	+	+			Site, ID, MDY, Effort	
2	Hartwell	1996	+	+	+			Site, ID, MDY, Effort	
2	Broadway	1995	+	+	+			ID, MDY	
2	Broadway	1996	+	+	+			ID, MDY, Effort	
2	Broadway	1997	+	+	+			Site, ID, Date, Temp, Secchi, Cond, Effort	
2	Russell	1994	+	+	+			Site, ID, MDY, Effort	
2	Russell	1995	+	+	+	+	+	Site, ID, MDY, Effort	
2	Russell	1996	+	+	+			Site, ID, MDY, Effort	
2	Russell	1998	+	+	+	+	+	Zone, Site, ID, Date, Temp, Cond, Effort	
2	Secession	1994	+	+	+			Site, ID, MDY, Effort	
2	Secession	1995	+	+	+	+	+	Site, ID, MDY, Effort	
2	Secession	1996	+	+	+			Site, ID, MDY, Effort	
2	Secession	1997	+	+	+	+	+	Site, ID, Date, Temp, Secchi, Cond, Effort	
2	Secession	1998	+	+	+	+	+	Site, ID, Date, Temp, Secchi, Cond, Effort	
2	Thurmond	1994	+	+	+			Site, ID, MDY, Effort	
2	Thurmond	1995	+	+	+	+	+	Site, ID, MDY, Effort	
2	Thurmond	1996	+	+	+			Site, ID, MDY, Effort	
2	Thurmond	1997	+	+	+	+	+	Zone, Site, ID, Date, Temp, Cond, Effort	
2	Thurmond	1998	+	+	+	+	+	Zone, Site, ID, Date, Temp, Cond, Effort	
3	Greenwood	1986	+	+					
3	Greenwood	1987	+	+	+				
3	Greenwood	1990	+	+	+				
3	Greenwood	1996	+	+	+	+	+		
3	Greenwood	1997	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Temp, Secchi, Effort	
3	Greenwood	1998	+	+	+	+	+	Zone, Site, ID, Date, Temp, Secchi, Effort	
3	Murray	1998	+	+	+	+	+	Zone, Site, ID, Date, Temp, Secchi, Effort	
4	Monticello	1997	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Temp, Secchi, Cond, Effort	
4	Wylie	1995	+	+	+			Site, MY	
5	Moultrie	1991	+	+	+			Site, MDY	
5	Moultrie	1992	+	+	+			Site, MDY	

Data

Dist	Reservoir	Year	Species	TL	Wt	Age	Sex	Other
5	Moultrie	1997	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Temp, Secchi, Effort
5	Moultrie	1998	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Effort
5	Marion	1991	+	+	+			Site, MDY
5	Marion	1997	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Temp, Secchi, Effort
5	Marion	1998	+	+	+	+	+	Zone, Site, ID, Date, Lk Level, Effort
6	Brown	1990	+	+	+			Season
6	Brown	1991	+	+	+			Season
6	Brown	1992	+	+	+			Season
6	Brown	1993	+	+	+			Season
6	Brown	1994	+	+	+			Season
6	Brown	1995	+	+	+			Season
6	Brown	1996	+	+	+			Season
6	Brown	1997	+	+	+	0		Season
6	Brown	1998	+	+	+			Season

District 2, Lake Greenwood in District 3, and Lakes Moultrie and Marion in District 5. The long-term database for Lake Brown in District 6 included aging information in 1997 but the standardized sampling protocol was not followed. After preliminary inspection of each file, data were extracted into permanent SAS datasets with standardized variable names to facilitate further analysis.

#### *Data Analysis*

Age-frequency tables were produced for all reservoir populations for which data were available. An example (Lake Thurmond, 1997) is included in Table 2. By SSP guidelines, otoliths were collected from fish between 175 and 474 mm TL. However, in Lake Thurmond no fish in the 46.2 cm group (450-474 mm TL) were collected; the largest fish from which otoliths were taken was 448 mm TL. As a result, only ages 1 and 2 were fully represented in the age-length key.

Table 2. An age-length key and the length-frequency distribution by age group produced from it using 1997 Lake Champlain Bass Age Data provided by Thurmond District II. 1

AGE-LENGTH KEY												
Number of aged fish per length based on input data												
CM_GRP	N_AGED	AGE0	AGE1	AGE2	AGE3	AGE4	AGE5	AGE6	AGE7	AGE8	AGE9	AGE10
18.7	9	.	9	.	.	.	.	.	.	.	.	.
21.2	14	.	14	.	.	.	.	.	.	.	.	.
23.7	9	.	9	.	.	.	.	.	.	.	.	.
26.2	13	.	13	.	.	.	.	.	.	.	.	.
28.7	9	.	7	2	.	.	.	.	.	.	.	.
31.2	8	.	5	2	1	.	.	.	.	.	.	.
33.7	10	.	2	7	1	.	.	.	.	.	.	.
36.2	11	.	.	7	2	2	.	.	.	.	.	.
38.7	10	.	.	1	6	3	.	.	.	.	.	.
41.2	12	.	.	3	7	1	1	.	.	.	.	.
43.7	9	.	.	.	3	2	3	.	1	.	.	.
=====												
	114	0	59	22	20	8	4	0	1	0	0	0

AGE-FREQUENCY DISTRIBUTION  
From DeVries & Frie. 1996. Fisheries Techniques, 2nd Edition

CM_GRP	COUNT	AGE0	AGE1	AGE2	AGE3	AGE4	AGE5	AGE6	AGE7	AGE8	AGE9	AGE10
11.2	3	.	.	.	.	.	.	.	.	.	.	.
13.7	18	.	.	.	.	.	.	.	.	.	.	.
16.2	29	.	.	.	.	.	.	.	.	.	.	.
18.7	39	.	39	.	.	.	.	.	.	.	.	.
21.2	33	.	33	.	.	.	.	.	.	.	.	.
23.7	30	.	30	.	.	.	.	.	.	.	.	.
26.2	48	.	48	.	.	.	.	.	.	.	.	.
28.7	44	.	34	10	.	.	.	.	.	.	.	.
31.2	37	.	23	9	5	.	.	.	.	.	.	.
33.7	37	.	7	26	4	.	.	.	.	.	.	.
36.2	25	.	.	16	5	5	.	.	.	.	.	.
38.7	20	.	.	2	12	6	.	.	.	.	.	.
41.2	16	.	.	4	9	1	1	.	.	.	.	.
43.7	9	.	.	.	3	2	3	.	1	.	.	.
48.7	3	.	.	.	.	.	.	.	.	.	.	.
51.2	3	.	.	.	.	.	.	.	.	.	.	.
53.7	1	.	.	.	.	.	.	.	.	.	.	.
=====												
	395	0	214	67	38	14	4	0	1	0	0	0

The age-frequency distribution of the population should be interpreted accordingly. For the Lake Champlain population, the assumption that fish <175 mm TL were age-1 appears to be valid (Table 2).

Recruitment of largemouth bass varied within and between reservoirs and between years. Mean relative recruitment ranged from 41.0 age-1 fish per hour in Lake Thurmond to 3.1 per hour in Lake Moultrie (Table 3). Zonal differences in recruitment of largemouth bass were apparent in all reservoirs sampled in 1997 but the magnitude of the difference between zones was greatest on the Santee-Cooper lakes, even though recruitment there was generally depressed in all zones (Table 3). Relative recruitment on Lake Moultrie was nearly nine times greater at Black's Camp than it was at Angel's Cove. Mean relative recruitment for Lake Thurmond from 1994-1997 was 62.3 (standard deviation = 15.2), but was nearly twice as high in 1995 as it was in 1997 (Table 3). The numbers suggest a recent downturn in recruitment in the reservoir. Continued monitoring will determine whether this is part of a trend or simply expected annual variation.

Mean length at age and the associated standard deviation (s) were calculated for each age cohort. Largemouth bass were uniformly longest at each age class in Lake Monticello and generally shortest in Lake Greenwood (Table 4). Mean length of age-1 fish ranged from 16.3 cm (s = 4.77) in Lake Marion to 25.0 cm (s = 2.89) in Lake Monticello.

Because the SSP only required that fish up to 474 mm TL be subsampled for otolith-based age determination, estimates of mean length of certain age groups were probably biased. In Lake Thurmond, age class three may have been affected, while in lakes Marion and Moultrie, age class four was probably affected. Since fish up to 550 mm were aged in lakes Greenwood and Monticello in 1997, possible length at age bias in those reservoirs would be of concern primarily

Table 3. Catch per unit effort (no./hr) by age group of largemouth bass collected within zones in five South Carolina reservoirs by electrofishing in spring 1997. Zonal values were averaged to compute whole lake values in 1997. CPUE values for Lake Thurmond 1994-1996 were calculated by dividing total catch by total effort for each age group.

Reservoir	Zone	Year	Age Group					Combined
			1	2	3	4	5	
Marion	Crappie Neck	1997	6.7	1.3	10.7	2.7	0.0	42.0
	Goat Island		4.4	3.7	11.7	16.8	1.5	64.4
	Jacks Creek		20.8	8.5	6.2	4.6	0.0	40.8
	Whole Lake		10.6	4.5	9.5	8.0	0.5	49.1
Moultrie	Angels Cove	1997	0.6	3.5	12.7	6.9	2.9	44.0
	Blacks Camp		5.3	5.3	6.0	13.3	0.7	57.3
	East Dike		2.0	8.7	5.3	5.3	2.0	30.7
	Hatchery		4.6	4.6	10.2	8.3	13.9	77.8
	Whole Lake		3.1	5.5	8.6	8.5	4.9	52.5
Greenwood	Lower Lake	1997	14.4	7.2	4.4	3.2	1.2	31.2
	Mid Lake		41.1	9.0	11.5	4.1	1.6	69.9
	Upper Lake		51.6	23.1	4.4	5.5	0.0	90.1
	Whole Lake		35.7	13.1	6.8	4.3	0.9	63.7
Monticello	Lower Lake	1997	16.2	7.8	5.2	1.9	1.9	47.9
	Mid Lake		7.7	2.6	2.6	0.0	0.0	16.7
	Upper Lake		9.9	7.3	6.8	2.1	1.6	35.9
	Whole Lake		11.3	5.9	4.9	1.3	1.2	33.5
Thurmond	Lower Lake	1997	47.2	5.9	3.8	1.7	—	60.8
	Mid Lake		52.3	28.6	11.0	3.9	1.1	97.0
	Upper Lake		23.4	6.2	2.6	1.0	1.0	39.4
	Whole Lake		41.0	13.6	5.8	2.2	0.7	65.7
Thurmond	Whole Lake	1996	60.1	10.1	4.3	1.7	0.1	78.5
	Whole Lake	1995	79.9	15.3	6.1	2.1	0.4	106.0
	Whole Lake	1994	66.0	12.8	8.0	3.1	0.6	92.8

Table 4. Mean total length (cm) and standard deviation, by age class from age-frequency tables, of largemouth bass collected within zones in five South Carolina reservoirs by electrofishing in spring 1997. Data collected from Lake Thurmond during 1994-1996 included for comparison.

Reservoir	Zone	Year	Age Group				
			1	2	3	4	5
Marion	Crappie Neck	1997	14.7 (4.74)	32.5 (1.77)	37.1 (2.87)	38.7 (2.88)	
	Goat Island		22.5 (6.07)	31.7 (2.09)	37.1 (1.80)	41.0 (2.12)	
	Jacks Creek		15.5 (3.38)	32.1 (3.01)	38.7 (1.89)	40.4 (3.03)	
	Whole Lake		16.3 (4.77)	32.0 (2.57)	37.7 (2.47)	40.6 (2.73)	
Moultrie	Angels Cove	1997	27.7 (—)	31.2 (4.18)	36.5 (2.81)	42.7 (2.92)	
	Blacks Camp		17.5 (2.67)	27.8 (2.66)	35.4 (3.31)	38.8 (2.36)	
	East Dike		19.5 (1.44)	33.5 (4.50)	37.1 (2.66)	40.0 (2.32)	
	Hatchery		23.7 (1.77)	32.7 (1.37)	35.7 (3.12)	39.5 (3.95)	
	Whole Lake		20.5 (3.72)	32.6 (4.64)	36.5 (2.90)	40.4 (3.16)	
Greenwood	Lower Lake	1997	16.3 (3.58)	30.6 (3.98)	36.4 (3.05)	39.6 (3.77)	
	Mid Lake		17.0 (5.19)	31.0 (2.36)	35.7 (2.00)	40.2 (3.35)	
	Upper Lake		19.7 (2.84)	28.1 (2.49)	29.3 (1.25)	40.2 (1.37)	
	Whole Lake		17.8 (4.28)	29.8 (3.20)	35.5 (3.27)	40.1 (3.12)	42.9 (4.09)
Monticello	Lower Lake	1997	24.8 (3.34)	35.3 (4.40)	40.1 (2.57)	44.3 (1.25)	
	Mid Lake		26.6 (1.89)	36.7 (1.58)	42.3 (3.34)		
	Upper Lake		24.1 (2.57)	36.0 (1.39)	39.1 (3.09)	45.8 (3.33)	
	Whole Lake		25.0 (2.89)	35.2 (3.75)	40.7 (3.60)	45.4 (2.46)	
Thurmond	Lower Lake	1997	22.8 (6.12)	37.8 (3.34)	39.0 (2.63)		
	Mid Lake		22.3 (4.75)	32.3 (2.77)	36.3 (4.02)		
	Upper Lake		22.9 (4.68)	34.5 (1.22)	41.7 (1.12)		
	Whole Lake		23.0 (5.55)	33.8 (3.21)	37.8 (3.73)		
Thurmond		1996	19.9 (6.53)	33.8 (2.75)	36.7 (3.27)		
		1995	23.4 (5.94)	32.5 (3.43)	36.7 (4.01)		
		1994	21.5 (5.29)	34.3 (3.22)	38.1 (3.34)		



for older age classes.

Von Bertalanffy growth parameters derived for lakes Thurmond and Greenwood using available length and age data were compromised by the limited data for older fish. For the Lake Thurmond population the asymptotic length  $L_{\infty}$  was only 474 mm and the growth coefficient  $k$  was 0.56. For the Lake Greenwood population  $L_{\infty}$  was 496 mm and  $k$  was 0.43. Zonal differences in growth through age-4 were seen in Lake Thurmond but not in Lake Greenwood (Figure 2). Predicted lengths of fish in the upper zone of Lake Thurmond were greater at each age than those in the middle and lower zones.

CPUE data collected from Lake Thurmond were used to produce mortality estimates because four consecutive years (1994-97) of data were available. Age structure for each year's CPUE data was derived from the 1997 age-length key. Age-1 fish were omitted because they appeared to be over-represented relative to older fish. Fish older than age-4 were omitted because their numbers could not be accurately estimated. Total mortality estimates for Lake Thurmond largemouth bass ranged from 0.50 to 0.62 (Table 5). Bettross et al. (1994) reported an average annual exploitation rate of 0.23 from Lake Thurmond in 1991-1993.

#### *Data Interpretation*

A standardized spring electrofishing protocol was developed in collaboration with management biologists and implemented during the 1997 sampling season. Modifications were made to the protocol prior to the 1998 sampling season. The purpose of the protocol was to ensure that representative samples of black bass were obtained across the state, so that differences within and between populations could be detected when they existed. Data generated using the protocol were readily imported into a statewide black bass database, then summarized and

Figure 2. Growth curves of largemouth bass through age-4 in two South Carolina reservoirs, by zone, using data collected in 1997. Lengths at age were predicted from the von Bertalanffy growth function, with  $t_0$  set at -0.024 (Beamesderfer and North, 1995).

Lake Thurmond

Lake Greenwood analyzed with

Table 5. Estimated instantaneous (Z) and annual (A) mortality of two age ranges of largemouth bass in Lake Thurmond based on alternative assumptions of constant recruitment (all observations included) and variable recruitment (specific cohorts included). Catch per unit effort by age-class was estimated for four consecutive years of sampling data by applying the 1997 age-length key to each year's length frequency distribution. Linear regression of the natural log-transformed CPUE on age was used to determine Z and its associated  $R^2$ .

Recruitment	Age Range	Observations Included	Z	$R^2$	A
Constant	2-4	All (N=12)	0.84	0.91	0.57
Variable	2-4	1992 & 1993 cohorts (N=6)	0.98	0.97	0.62
Constant	2-3	All (N=8)	0.70	0.77	0.50
Variable	2-3	1992, 1993 & 1994 cohorts (N=6)	0.83	0.86	0.56

computer programs developed to extract key population metrics. The South Carolina Standardized Sampling Plan is attached to this report as Appendix B.

Total annual mortality (A) was the only variable evaluated in STELLA simulations.

Recruitment and length-at-age estimates were held constant. Values of A tested were:

Simulation	A, age-1 to age-2	A, age-2 to age-5
1	0.81	0.50
2	0.81	0.60
3	0.75	0.50

Using simulation 1 output, seven of 20 simulated populations produced length frequencies that were statistically different ( $P=0.01$ ) from the observed Lake Thurmond population. Compared to the simulated populations, the observed always had a higher median length and usually had a higher 75<sup>th</sup> percentile length. Using simulation 2 output, ten of 20 simulated populations produced length frequencies that were statistically different ( $P=0.01$ ) from the observed population.

Compared to the simulated populations, the observed always had a higher median length and 75<sup>th</sup> percentile length. Simulation 3 resulted from a decision based on the results of simulations 1 and 2 to slightly lower age-1 to age-2 mortality. This produced 1 of 20 simulated populations with length frequencies that were statistically different ( $P=0.01$ ) from the observed. Five simulated populations had the same median and nine had the same 75<sup>th</sup> percentile length as the observed population. The potential effects of proposed changes in management strategies on largemouth bass population structure observed during spring electrofishing in Lake Thurmond can now be

effectively evaluated with a dynamic model, using the parameters given.

### Recommendations

1. Develop reservoir-specific black bass management models which can be integrated into the tool bags of management biologists. Existing user-friendly models (e.g. MOCPOP, LSIM, and GFSIM) will be evaluated for this purpose.
6. Revise the standardized sampling plan, removing the upper size limit on samples collected for aging. The present limit truncates length-frequency distributions for older age classes and results in biased growth curves.
7. Ensure that the guidelines of the SSP are followed.
4. Ensure that sampling efficiency remains the same from year to year so that a long-term database can be constructed. Alternatively, provide a way to control for changes in sampling efficiency within the analytical protocol.
5. Proceed with evaluation of 1998 spring electrofishing sampling data.

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## Appendix A: Largemouth Bass Management Strategies

### Part I: A Survey of Southeastern State Natural Resource Agencies

Fisheries chiefs in the agencies belonging to the Southeastern Association of Fish and Wildlife Agencies (SEAFWA) were contacted to provide copies of black bass management plans and/or strategies developed for use in their states, along with any other information that might be helpful in compiling a summary of black bass management practices in the Southeast. Responses were evaluated to determine which states have developed statewide black bass management plans, which maintain statewide fisheries databases, which agencies have regulatory authority over the black bass fishery (and at what level), and which have written protocols or guidelines that reservoir management biologists must follow. This latter criterion was subdivided to obtain specific information regarding reservoir management plans, sampling procedures, bass population assessment, reports, and decision making.

Information on black bass management plans and/or strategies was received from fisheries chiefs or their designees in all 15 states agencies (besides South Carolina) in SEAFWA. Some of the responses were thorough, others provided only sketchy information of limited utility. Results of the survey are summarized in Table A-1. Appropriate responses for South Carolina are included for comparison.

Seven of 15 states, including South Carolina, have a statewide black bass management plan in place or under development (Table A-1). The others manage largemouth bass strictly at the local or regional level. Plans which were available for review are summarized in the next section. Eight of 13 states presently maintain or are developing statewide fisheries databases. Natural resource agencies in 14 of 15 states have the authority to establish harvest restrictions for black bass, either statewide or on a site by site basis. This authority is usually implemented by an agency's advisory board or commission. Maryland has regulatory authority over largemouth bass

Table A-1. Summary of responses from natural resource agencies in 16 southeastern states regarding the management of black bass, including whether or not a statewide black bass management strategy was available for review. "Development" indicates a topic was planned but not yet fully implemented. Responses which didn't provide sufficient information about a topic are indicated by "?".

State	Standardized Protocols or Guidelines for Reservoir Management							
	Black Bass Management Plan	Statewide Black Bass Database	Regulatory Authority	Management Plans	Black Bass Sampling	Bass Population Assessment	Reports	Decision - Making
AL	No	?	Yes	Yes	Yes	No	Yes	No
AR	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes
FL	No	Development	Yes	No	Development	No	Examples	No
GA	No	Yes	Yes	No	Yes	No	No	No
KY	No	Yes	Yes	?	Yes	?	?	No
LA	Yes	?	Yes	Yes	Yes	Not defined	Yes	?
MD	No	No	Yes	No	No	No	No	No
MS	?	No	Yes	?	Yes	?	?	?
MO	No	No	Yes	Yes	Yes	Available	Yes	No
NC	Yes	No	Yes	No	Development	?	No	?
OK	Yes	Yes	Yes	Development	Yes	Yes	Development	?
SC	Yes	Development	No	No	Yes	No	No	No
TN	Development	Development	Yes	No	Development	No	Semi	No
TX	Yes	Yes	Yes	Yes	Yes	No	No	No
VA	No	No	Yes	No	No	No	No	No
WV	No	?	?	?	?	?	?	?





in "inland" waters but not in "tidal" waters (L. Fewlass, MD DNR, personal communication). Only South Carolina among responding states does not delegate to its natural resource agency the authority to establish length and creel regulations for black bass.

Thirteen of 15 states have or are developing standardized procedures for sampling largemouth bass populations in lakes and reservoirs. Standardized reservoir sampling plans of seven southeastern states were reviewed and referenced (Alabama Department of Conservation and Natural Resources, undated; Arkansas Game & Fish Commission, 1989; Georgia Department of Natural Resources, 1991; Kentucky Department of Fish and Wildlife Resources, 1993; Louisiana Department of Wildlife and Fisheries, 1994; Missouri Department of Conservation, 1987; Oklahoma Department of Wildlife Conservation, undated). Most of the plans are comprehensive, covering all of the gear types typically used to assess fish populations or communities.

Five of 13 states have or are developing guidelines for their agency biologists to follow when preparing reservoir management plans. Seven of 13 states have some form of standardized format to be followed in writing reports. Alabama and Missouri both provide comprehensive outlines detailing specific information to be included in annual reports. An advantage of such a system is that it makes it easy to compare the results of management activities across time and reservoirs. Arkansas (Bass Management Task Force, 1991) and Oklahoma provide guidelines for assessing bass populations on the basis of sampling results and/or other factors. Missouri developed guidelines (Kruse, 1988), but they are not widely used (P. Pitts, MO DC, personal communication). The Arkansas Largemouth Bass Management Plan includes minimum length and slot limit criteria which can be used as guidelines for decision-making with respect to harvest restrictions or other largemouth bass management options. No other states attempted to establish

decision-making criteria (Table A-1).

Six states have statewide largemouth (or black) bass management plans in place, copies of which were obtained and reviewed. The six plans represent the spectrum of approaches to such an endeavor, from basic to complex.

Oklahoma's plan (Oklahoma Department of Wildlife Conservation, undated) is relatively basic. It consists of three goals. Goal 1 pertains to raising public awareness of black bass management, while Goals 2 and 3 pertain to enhancing recreational opportunities by sustaining, improving, or enhancing largemouth and smallmouth bass populations, respectively. Several objectives and strategies are identified as a way of measuring progress toward attaining each goal.

The Arkansas plan (Bass Management Task Force, 1991) devolves from a single goal: "Provide diverse bass fishing opportunities to satisfy the variety of angler wants." To attain this rather general goal, four objectives are established which become the framework for a complex plan which defines criteria for establishing harvest restrictions and trophy lakes, evaluating pros and cons of tournament fishing, and identifying research, resource, and program needs.

The Louisiana plan (Arnoldi, et al, 1990) takes an approach somewhat similar to Arkansas', building on a single goal ("To provide the best possible fishing experience for anglers fishing in Louisiana waters by increasing the probability that anglers have the opportunity to catch quality bass") implemented with eight objectives. Several objectives are quantifiable, specifying, for example, target angler catch rates of largemouth bass in the quality-trophy length categories. Others have to do with access and habitat issues. One important objective in the Louisiana plan seeks to establish a reliable index of bass populations and use it to test response of bass populations to regulations. Unfortunately, the recommendations for implementing the plan do not address how such an index would be developed, other than listing physical and biological parameters (as

opposed to criteria) to be considered for quality and trophy lakes.

North Carolina's plan (North Carolina Wildlife Resources Commission, 1993) defines six goals in support of a single unacknowledged but overarching goal ("The WRC will maintain black bass species representation, population densities and size diversity sufficient to support and promote bass fishing in North Carolina..."). Each of the supporting goals relates to a specific aspect of black bass management, including habitat protection, angler expectations, size and creel limits, geographic and species differences, stocking, and research needs. From two to six strategies are defined to address each goal. The North Carolina plan is logically developed and cohesively written.

Texas developed its plan as a non-technical program report published in booklet form for distribution to the public (Texas Parks and Wildlife Department, 1995). Nevertheless, the plan incorporates most of the elements of the more conventionally formatted black bass management plans developed by other states. A section on management tools describes the rationale behind the application of harvest restrictions, both statewide and local.

The South Carolina plan (South Carolina Department of Natural Resources, 1995) summarizes the status of the four *Micropterus* species known to occur in the states's waters, then separately describes up to six management goals for each. The plan recognizes the importance of site-specific, as opposed to statewide, management of populations, given that all water bodies are different. However, because SCDNR does not have regulatory authority with respect to harvest restrictions, management options, whether site-specific or statewide, are limited.

## Appendix A: Largemouth Bass Management Strategies

### Part II: A Survey and Review of Current Literature

The current body of literature on black bass management, including articles published within the past 10 years in journals and proceedings, state reports, when available, and to some extent popular magazines, was reviewed. Citations relevant to the development of black bass management guidelines, arranged by major topic and sub-topic, are discussed and included herein.

### Data Collection and Analysis

#### *Sampling Fish Populations, including Largemouth Bass*

Burkhardt and Gutreuter (1995) demonstrated the importance of standardizing electrofishing by standardizing power (wattage) to reduce catch variation. Harden and Connor (1992) found differences in electrofishing efficiencies between crews (and between fish-size categories for the same crew) and recommended that population estimates based on electrofishing results be treated cautiously. More samples may be needed to provide adequate precision, or a lower standard of precision should be used. Hill and Willis (1994) determined that electrofishing in high conductivity water may be more efficient using pulsed AC rather than DC. Hall (1986) used a mark-recapture study to show a positive linear relationship between electrofishing catch per hour and largemouth bass density. Coble (1992) combined data from Hall (1986) with his own study and produced a regression equation relating largemouth bass population density to electrofishing catch per unit effort. McInerny and Cross (1996) compared day and night electrofishing for largemouth bass during spring and fall in Minnesota lakes and determined that all four sampling periods provided accurate data on size-structure of fish  $\geq 200$  mm total length, but only catch comparisons from the same time period were meaningful. Daytime catch per unit effort of stock-size largemouth bass was negatively affected by water clarity. For long-term

monitoring, electrofishing should be done during the same season and diel period. Dumont and Dennis (1997) compared day and night electrofishing for three species of fish and found that, for stock-size largemouth bass, day vs. night catch was similar in the spring, different in the fall when significantly more fish were collected at night. Miranda et al. (1996) evaluated the precision of electrofishing sample duration for estimating relative abundance of largemouth bass and provided guidelines for optimizing sample duration. Gilliland (1985) evaluated standardized sampling procedures used by Oklahoma Department of Wildlife Conservation and determined that either a sample size of 150 largemouth bass or an effort of 5 hours of electrofishing during spring and fall sampling was adequate to describe the population structure. Van Horn et al. (1991) demonstrated that intra-seasonal variation in electrofishing CPUE can lead to misinterpretation of results when making management recommendations based on single-pass sampling. Weithman et al. (1979) described a sequential electrofishing sampling technique that minimized sample size (and therefore sampling effort) needed to make a decision about proportional stock density (PSD) of a largemouth bass population. Gablehouse and Willis (1986) looked at angler catch data to see if it was useful in assessing size structure and density of largemouth bass populations. They determined that biases in catch resulting from fisherman intention and technique had to be interpreted before such data could be used reliably. Maceina et al. (1993) used a catch-depletion method of electrofishing in vegetated areas surrounded by block nets to estimate the population of age-0 largemouth bass. Because CV's for mean fish density were similar to those obtained using rotenone, they suggested the method provided a useful alternative to rotenone. Jackson and Noble (1995) demonstrated size differences in vulnerability of age-0 largemouth bass to three gear types: boat-mounted electrofisher, back-pack electrofisher, and bag seine. They cautioned against generalizing about recruitment processes in a population on the basis of samples collected using a

single gear type.

### *Analysis*

Bivin et al. (1989) described a personal computer-based system for collecting, analyzing, and reporting electrofishing and rotenone sample data using customized programs and standard dBase data files. The system simplified data processing and analysis, resulted in faster turn-around of data for field biologists, and facilitated development and maintenance of a statewide data base. Maceina (1997) found that residuals associated with catch-curve regressions could represent variable recruitment in fish populations and described an analytical approach that would eliminate the need to measure recruitment indices each year.

### Age and Growth Analysis

DeVries and Frie (1996) updated a chapter by Jearld (1983) on age determination and added important discussion of growth analysis, including techniques. Their list of current references is extensive. Gutreuter (1987) evaluated back-calculation equations as estimators of growth and determined that proportional equations (those that take into account individual lengths and scale radii) were more efficient than regression equations. He pointed out that, for routine analyses of growth, using only the most recent annuli avoids the confusion of Lee's phenomenon. Gutreuter and Childress (1990) looked for indirect indicators of growth in largemouth bass and crappie. They tested several condition indices in multiple regression models but found none provided a useful or reliable estimate of growth. Howells et al. (1994) used scales and otoliths from known-age 6, 7, and 8 year old largemouth bass to test the ability of experienced readers to correctly age older fish. While otoliths gave better results than scales, fewer than 50% were



correctly aged by any reader. The percentage of otoliths correctly aged  $\pm 1$  year nearly doubled. Lengths back-calculated using scales exhibited Lee's phenomenon; those using otoliths did not exhibit Lee's phenomenon but gave inconsistent results. Morrow (1990) compared scales and otoliths for aging and back-calculating lengths of largemouth bass. Scale and otolith ages agreed only 72% of the time, and agreement percentage was positively correlated with growth rate. Otoliths usually produced smaller back-calculated lengths than scales. Hoyer et al. (1985) validated annulus formation in Florida strain largemouth bass through age 5. They also found that sectioning otolith produced more accurate aging results, particularly in slower growing fish, as early as age 2, because outer annuli were obscured in whole mounts. Back-calculations of fish length from sectioned and correctly-aged whole otoliths were equally accurate. Maceina and Murphy (1989) found differences in otolith morphometry between Florida and northern strain largemouth bass which could result in bias if not taken into account when back-calculating lengths, particularly in mixed-stock populations. Miller and Storck (1984) found differences in growth rates between early and late-spawned largemouth bass juveniles. Slow growth in late-spawned fish was attributed to their inability to make the transition from an invertebrate to a fish diet. Time of spawning may be an important factor affecting year class strength. Bettoli et al. (1992) found that most largemouth bass became piscivorous at a smaller size after littoral-zone submersed vegetation was removed by grass carp. Faster first-year growth was noted in every year class produced after vegetation was eliminated. Miranda and Durocher (1986) looked at the effects of environmental factors on growth of largemouth bass in Texas reservoirs and determined that the best predictors of growth were reservoir age, average depth, standing stock, and prey-predator ratio. McCauley and Kilgore (1990) found that temperature (accumulated day-degrees over 10°C)

was significantly correlated with growth of largemouth bass, and suggested that more than half the variability in growth may be due to environmental temperature. Schramm et al. (1992) evaluated back-calculation procedures using Florida strain largemouth bass otoliths. The direct proportion method produced the most reliable estimates of fish length at earlier ages, with no evidence of Lee's phenomenon occurring.

### Length-Weight Relationships

Anderson and Neumann (1996) updated and considerably expanded an earlier chapter on the same topic by Anderson and Gutreuter (1983). Cone (1989) argued that commonly used length-weight indices are flawed because they are based on invalid assumptions and can lead to erroneous conclusions. He recommended using least squares regression parameters as a more accurate method for examining length-weight relationships in fish populations. Several researchers (Springer et al., 1990) responded to Cone's (1989) criticism with a debate over the merits of the various indices of condition, relative weight in particular.

### Population Dynamics

Beamesderfer and North (1995) summarized published growth and natural mortality data from hundreds of largemouth and smallmouth bass populations in North America. They found age at quality length (an index of growth rate) and natural mortality of largemouth bass, and growth rates of smallmouth bass, were significantly correlated with latitude and temperature indices. They also used modeling software to estimate effects of fishing under various exploitation and length-restriction scenarios on bass populations with constant recruitment, simulating low, average, and high productivity in those populations. Results varied with population productivity,

but the authors concluded that productive populations provided the most management flexibility because all key population and fishery characteristics were sensitive to exploitation rate and length limit. Bettross et al. (1994) used age and growth estimates from scales, mortality estimates from a tagging study, and age frequency distributions from spring electrofishing to evaluate existing harvest regulations for largemouth bass in three major impoundments on the Savannah River. Using an equilibrium yield model, they concluded that exploitation of quality and preferred size classes of largemouth bass in Clarks Hill Lake (Lake Thurmond) could be high enough to warrant a change to a 16" size limit. Because exploitation rates on lakes Russell and Hartwell were moderate to low, those lakes were not considered candidates for larger size limits. Davies et al. (1982) compared recruitment dynamics of largemouth bass in farm ponds and reservoirs, then modeled recruitment as a function of prey availability. They suggested that management strategies should hinge on maintaining an adequate size structure within prey populations through bass predation so that each species has an opportunity to reproduce, survive and grow. Miranda and Hubbard (1994) demonstrated that predation may be a significant source of over-winter mortality for smaller-sized age-0 largemouth bass. Perry et al. (1995) conducted bioenergetics simulations with largemouth bass population data from a no-harvest reservoir and showed that catch-and-release regulations have the potential to limit growth and reduce the average size of otherwise harvestable fish. Increasing fishing mortality would increase availability of forage and result in weight increases of survivors.

### Harvest Potential

Kirk (1989) summarized methods developed for sampling reservoirs by electrofishing and

rotenone to collect representative samples of the largemouth bass population and the prey community. Age analysis of largemouth bass from four South Carolina reservoirs indicated that fish up to 9 in total length were recruits (age 1). Total mortality in most populations was high. A flow diagram was developed to assess harvest potential of a reservoir and facilitate management decision-making. Following the flow diagram by answering questions about the largemouth bass population and the fishery led to one of three management strategies: minimum size limit, slot limit, and no harvest restrictions. A problem with the diagram and its general application is that the questions were framed in qualitative terms such as “adequate numbers”, “good size structure”, “adequate growth” that are not defined and may mean different things in to biologists working in different reservoirs. Buynak and Mitchell (1993) compared electrofishing and angler catch rates within years and between successive years in a single reservoir to see if spring electrofishing results could be used to predict angler success. Reliable predictive models would help define harvest potential of largemouth bass in reservoirs and would provide managers with a way to evaluate the results of management activities. While some significant relationships were found, results from eight consecutive years of data collection and analysis were inconsistent. Kruse (1988) developed guidelines for characterizing largemouth bass populations based on scores assigned to measures of abundance, size structure, and recruitment derived from spring electrofishing. He attempted to include an angling component in the assessment but was unable to find a consistent and repeatable relationship between electrofishing catch rates and angler success. Maceina et al. (1995) described a catch-depletion method using spring electrofishing to estimate the abundance of harvestable largemouth bass in reservoirs. They suggested the technique could replace cove rotenone studies. McInerny and Degan (1993) found a strong linear relationship between catch rates of largemouth bass in Lake Wylie, South Carolina, using a stratified random

shoreline electrofishing protocol and population density estimates obtained with mark-recapture methods.

### Harvest Restrictions

Harvest restrictions (i.e. creel limits and/or size limits) have become increasingly popular with fisheries managers in recent years as tools to perform a variety of functions, among them restricting access to a fishery, allocating harvest among anglers, restructuring size distribution in a population, controlling excess production, and manipulating the prey base. Whether these tools work as intended or not is an open question. Published descriptions of applications tend to report success stories rather than failures. Part of the reason for this is that it is difficult to quantify, with statistical confidence, index parameters that show cause and effect relationships in lake or reservoir fish populations. Novinger (1984) laid the groundwork for studies of the effects of size limits on their target populations by calling for well-designed study plans with clear objectives, taking into account the influence of recruitment, growth, and mortality, as well as angler expectations and compliance, on the outcome. Storey and Ott (1992) assessed the effects of a catch-and-release regulation on the largemouth bass population of a newly opened reservoir in Texas. Electrofishing and creel surveys showed a decline in proportion of fish > 356 and a downward shift in PSD in two years, though no illegal harvesting was observed. Changes were attributed to increased mortality resulting from heavy fishing pressure. Ager (1989) reported that increasing the minimum legal size of largemouth bass from 305 to 406 mm increased forage in the reservoir by increasing gizzard shad recruitment. Sampling indicated that largemouth bass

population increased in abundance and its size structure shifted to larger fish, while the size structure of gizzard shad shifted downward. Terre and Zerr (1992) evaluated the effects of a statewide 356 mm minimum length limit for largemouth bass on 28 Texas reservoirs using electrofishing sample data two years after imposition of the limit. They detected increased densities of adult largemouth bass, and a shift in size structure toward larger fish. Van Horn, et al. (1981) reported mixed results from the imposition of a 45-cm minimum size limit on four reservoirs in North Carolina. The proportion of quality-size largemouth bass in the population increased in two reservoirs but not in the other two. Anthony and Orth (1986) used computer simulations to demonstrate the effects of minimum-length and slot limits on largemouth bass population structure. Large minimum-length and slot limits both increased the availability of quality-size fish in the simulated population. Buynak et al. (1991a) found that the abundance of largemouth bass and the forage community increased after the minimum-size limit was raised from 12 to 14 inches, but could not separate the effects from those caused by an extended drought during the time the size-limit changes were put into effect. Buynak et al. (1991b) evaluated differential harvest regulation changes for largemouth bass and spotted bass in the same reservoir. Minimum size of largemouth bass was increased from 12 to 15 inches, while the size limit on spotted bass was removed. Angler exploitation of largemouth bass decreased, while that of spotted bass increased. Conversely, electrofishing catch rates for all sizes of largemouth bass increased while those for spotted bass greater than 9 inches decreased. Cofer (1993) reported that a 356–558 mm slot limit on a designated trophy bass lake resulted in significantly higher electrofishing catch rates of largemouth bass >355 mm and >508 mm, but did not affect catch rates of trophy bass (>558 mm). Dean et al. (1991) found that introducing a 356-457 mm slot limit with a 5 fish daily bag in place of a 254 mm minimum length/10 fish daily bag limit in two Texas reservoirs

effectively restructured the largemouth bass populations. Creel surveys and electrofishing indicated that the proportion of fish >356 mm in both reservoirs increased in the years following introduction of the slot limit. Mean weight of largemouth bass harvested nearly tripled after seven years. Kraai (1993) found that raising minimum length limit for smallmouth bass from 254 to 356 mm in a Texas reservoir led to an abundance of slow-growing smaller fish, and recommended replacing the minimum length limit with a slot limit. Mitchell and Sellers (1989) found that raising the minimum length limit for largemouth bass in a Texas reservoir from 254 to 406 mm and decreasing the creel limit from 10 fish to 3 resulted in the population structure shifting to a dominance of larger, older fish as angler catch rates increased while harvest declined. Kurzawski and Durocher (1993) evaluated a 381–533 mm slot limit on a newly-opened Texas Reservoir. They concluded that the high slot limit prevented initial overharvest and helped maintain a population structure dominated by larger individuals. Martin (1995) assessed slot limits for largemouth bass in two New Jersey ponds. Despite the fact that an angler education program was an integral part of the study, she found that harvest of fish below the protected size was too low to restructure the population, and concluded that angler cooperation is essential for interactive management programs to work. Wynne et al. (1993) evaluated a 305–406-mm slot limit for largemouth bass in a North Carolina reservoir and found that it successfully restructured the size distribution of the population as measured by electrofishing and by angler catch records. Dean and Wright (1992) devised a system based on electrofishing catch per unit effort by length group and relative weight to help managers visually assess largemouth bass population data to determine which harvest restrictions if any were appropriate. Dent (1986) assessed sampling methods and parameters necessary to evaluate harvest restrictions. He determined that a comprehensive methodology was required because growth, mortality, exploitation rates, and relative abundance,

as well as angler catch rates and harvest were all important in analyzing the effectiveness of size and catch limits. Wilde (1997) summarized the results of 91 studies of largemouth bass fishery responses to length limits. Minimum length limits, in general, increased population size but failed to increase the proportion of larger fish in the population. Angler catch rates did improve, however, though harvest did not. Slot length limits successfully restructured bass populations, increasing the proportions of larger fish, but failed to increase angler catch rate or harvest. Studies tended to be limited by inadequate creel data and minimal duration.



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## Appendix B: Standardized Sampling Plan

## South Carolina Largemouth Bass Sampling Plan - Reservoirs

### Spring Electrofishing

- A. Objective: to obtain good estimates of the following population parameters
  - 1. Length structure
  - 2. Growth rate/age structure
  - 3. Relative condition
- B. Methods
  - 1. Obtaining a representative sample of the population
    - a. Large reservoirs (spatially complex or too big to effectively sample the entire shoreline)
      - 1. Sampling design: 3 zone  $\times$  3 site matrix (recommended minimum)
        - a. Define at least three zones representative of the spatial heterogeneity of the reservoir (e.g. upper, middle, and lower).
        - b. Within each zone, randomly select at least three primary sample sites and an excess of secondary sites (to be used if target numbers of fish are not met at the primary sites). Sample sites are defined as areas of shoreline with accessible habitat, and should accommodate 30 min of electrofishing. A numbered grid overlay of a reservoir map may facilitate randomization.
        - c. Increase the number of zones as needed to account for habitat variability within the reservoir system. Increase the number of sample sites as needed to obtain the required sample size of fish within each zone (see Target numbers of fish, below).
      - 2. Target numbers of fish
        - a. Objective: length structure/relative condition
          - 240 fish/reservoir (for 3  $\times$  3 design; increase by 80 for each additional zone included)
          - 80 fish/zone
          - 30 fish/sample site (maximum number that count toward the 80 fish/zone target; if more are collected during a 30 min sample, they can be measured, weighed, and recorded)
        - b. Objective: growth rate/age structure
          - 10 fish/25-mm length group/reservoir (for 3  $\times$  3 design; increase by 4 for each additional zone sampled)
          - 4 fish/25-mm length group/zone (maximum)
    - b. Small reservoirs (not spatially complex, small enough to effectively sample the entire shoreline)
      - 1. Sampling design: 1 zone  $\times$  3 site matrix (recommended minimum)
        - a. Treat the entire reservoir as a single zone.
        - b. Divide the shoreline into at least three sample sites, characterized by habitat features if discernable. Sample sites should accommodate 30 min of electrofishing without overlap.
      - 2. Target numbers of fish

- a. Objective: length structure/relative condition  
100 fish/reservoir  
40 fish/sample site (maximum number that count toward the 100 fish/reservoir target; if more are collected during a 30 min sample, they can be measured, weighed, and recorded)
    - b. Objective: growth rate/age structure  
10 fish/25-mm length group/reservoir (maximum)
  - c. Sampling considerations
    1. Water temperature: 15-20°C.
    2. Time of day: day sampling recommended; night sampling appropriate if Secchi disk transparency  $\geq 3$  m or there is a history of night sampling.
    3. Effort
      - a. Sample each primary site for 30 min of actual electrofishing time (pedal time) or actual time until the target number of fish is collected.
      - b. Collect all largemouth bass. Other species should be collected only if specific objectives to sample them have been defined in the study plan.
      - c. Add secondary sites in the order selected until a reasonable effort has been made to capture the target number of fish. Determining "reasonable effort" is at the discretion of the supervising biologist.
      - d. The crew leader decides whether to use continuous or intermittent pedal.
  - d. Age and growth
    1. Obtain otolith-based growth information for 3 consecutive years at least once every 10 years, more often if a reservoir is changing rapidly or if additional information on the status of the population is needed.
    2. Include fish 175-574 mm total length (TL). Fish longer than 574 mm may be included at the discretion of the supervising biologist.
2. Data collection
  - a. Work up fish after each 30 minute sample (sooner if fish are stressed).
    1. Measure (mm TL) and weigh (g) each fish in the field and record on Fish Data Form. If otoliths are being collected, follow the procedures in Age and Growth: Field below; otherwise, return the fish to water alive.
    2. Age and Growth: Field
      - a. The Otolith Tally Sheet, attached, will help the crew leader keep track of the number of fish from which otoliths have been taken, by length group. When a fish is measured, check the Tally Sheet to see if its otoliths are needed. If fish in its length group are needed, check it off on the Tally Sheet and remove the otoliths as described below. Once the target number of fish from a length group has been met for a zone or reservoir, excess fish in that length group can be returned to the water. [If fish will not be processed in the field, tag each with an ID number, noted on the Fish Data Form, before placing on ice].
      - b. Remove both sagittal otoliths from each fish to be aged and store dry in vials [avoid scale envelopes, which do not protect otoliths from damage].
      - c. Record an ID number on both the vial and the Fish Data Form.
      - d. Substitute scales for otoliths only if prior sampling from a reservoir has

demonstrated scale ages are accurate for the size range of fish included. If there are no existing data, verify scale ages with a subsample of otoliths representing 50% of each 25-mm length group of fish aged by scales.

3. Sex: fish sacrificed for aging should also be sexed, if possible.
  3. Data recording
    - a. Record data separately for each site on attached forms.
      1. Environmental Data Form: date, fisheries district, drainage system, reservoir name, lake level (m above/below full pool), zone identifier (large reservoirs), sample site identifier (GPS location if available), water temperature (°C, 0.5 m below surface), Secchi disk visibility (0.1 m), conductivity (µmho/cm), start and finish time (24 hr clock), electrofisher settings [AC/DC, voltage, pulse width, frequency, current output (amps), pedal operation (continuous, intermittent, combination) and actual electrofishing time (pedal time)], collectors names, and a general description of the habitat sampled.
      2. Fish Data Form: date, reservoir name, and sample site identifier (GPS location) for each sample site at the top of the form.
  4. Age and growth: lab procedures
    - a. Whole mounts are acceptable for most LMB up to 9 or 10 years, unless annuli are obscure or measurements are needed for back-calculation. In those cases, section the right sagittal otolith transversely, then mount and polish according to accepted methods. Whether whole or sectioned, otoliths should be read independently by two readers. Resolve differences between readers by mutual agreement, if possible. If agreement cannot be reached, prepare and independently read the left otolith. If agreement still cannot be reached, omit the fish from further consideration of age.
    - b. Back-calculate lengths, if necessary, depending on specific study objectives. Standardized procedures are still under consideration and development.
    - c. Archive whole or sectioned otoliths.
    - d. Age-frequency distribution may be computed from an age-length key (see DeVries, D.R., and R.V. Frie. 1996. Determination of age and growth. Chapter 16 in Murphy, B.R. and D.W. Willis, editors. Fisheries Techniques, Second Edition. American Fisheries Society, Bethesda, MD). A SAS program to perform this computation is available from Eastover upon request.
- C. Database management
1. Enter data in a standardized format using the Paradox data entry program provided. Environmental data and individual fish data are entered in separate linked files. Each district will produce one environmental data file and one fish data file each year, regardless of the number of reservoirs sampled
  2. Print, proof, and correct each dataset; export proofed datasets onto 3½" floppies and send to Eastover for processing and archiving. Datasets may also be sent as attachments to e-mail if that medium is available.

## Spring Electrofishing Sampling Strategy: Summary

Large Reservoir: 3 zones × 3 sample sites per zone matrix (minimum)

Target Number of LMB

Objective	Sample Site	Zone	Reservoir
Length structure/relative condition	30	80	240
Growth rate/age structure		4 per 25-mm length group	10 per 25-mm length group

Add more zones as needed to account for habitat variability in the reservoir

Add more sites as needed to reach target sample size in a zone

Small Reservoir: 1 zone × 3 sample site matrix (minimum)

Target Number of LMB

Objective	Sample Site	Reservoir
Length structure/relative condition	40	100
Growth rate/age structure		10 per 25-mm length group

Add more sites as needed to reach target sample size (if possible)

South Carolina Department of Natural Resources  
Largemouth Bass Electrofishing Environmental Data Form

Date: \_\_\_\_\_ Fisheries District: \_\_\_\_\_ Drainage: \_\_\_\_\_

Reservoir: \_\_\_\_\_ Lake level (m above/below full pool): \_\_\_\_\_

Zone: \_\_\_\_\_ Sample site ID: \_\_\_\_\_

Water temp (°C): \_\_\_\_\_ Secchi disk (m): \_\_\_\_\_ Conductivity (µmho/cm): \_\_\_\_\_

Time start (24 hr): \_\_\_\_\_ Time end (24 hr): \_\_\_\_\_

Electrofisher settings:

AC/DC \_\_\_\_\_ Voltage \_\_\_\_\_ Pulse width \_\_\_\_\_ Frequency \_\_\_\_\_

Output (amps) \_\_\_\_\_ Pedal operation\* \_\_\_\_\_ Pedal time (sec) \_\_\_\_\_

\*1=Continuous 2=Intermittent 3=Combination

Collectors: \_\_\_\_\_

Habitat description:

## South Carolina Department of Natural Resources Largemouth Bass Electrofishing Fish Data Form

Date: \_\_\_\_\_ Reservoir: \_\_\_\_\_

Zone: \_\_\_\_\_ Sample Site ID: \_\_\_\_\_

[illegible][illegible]

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South Carolina Department of Natural Resources  
Largemouth Bass Electrofishing Otolith Tally Sheet

Date: \_\_\_\_\_ Reservoir: \_\_\_\_\_

[illegible]



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