Hydro-peaking Impacts on Growth, Movement, Habitat Use and the Stress Response on Alabama Bass and Redeye Bass, in a Regulated Portion of the Tallapoosa River, Alabama.

by

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Abstract

Altered flow regimes caused by dam construction and operation can affect aquatic organisms in a variety of ways. The Tallapoosa River, in east-central Alabama, has been extensively impounded for flood control, navigation in the Alabama River, hydropower and water supply. None the less, the river still supports an important sport fishery. There has been previous research on the Tallapoosa River studying fish community responses to the altered flow regime. However, there has been minimal work on sportfish, including the black bass found within the river system. The objective of this research was to investigate the impacts of the altered flow regime on growth, movement, habitat use and the stress response on Alabama Bass *Micropterus henshalli* and Redeye Bass *Micropterus coosae*.

Dams and altered flow regimes may impact growth of aquatic organisms. Using incremental growth techniques, annual growth of Alabama Bass and Redeye Bass in the Tallapoosa River was evaluated in response to variation in flow regime. Age was the best explanatory variable that described growth in all models, although flow variables were included in more than half the models. Growth was higher for age-1 fish in years with less flow variation; however growth was similar among years for age-2 and age-3 fish. Overall growth rates for Alabama Bass and Redeye Bass were higher in the unregulated sites, than either regulated sites. Alabama Bass had higher growth rates than Redeye Bass at the Middle and Lower sites; however growth was similar between species the upper site. From this study it appeared that growth was not severely impacted by the altered flow regime.

Little is known about the movement and habitat use of Alabama Bass and Redeye Bass in the Tallapoosa River, specifically below R.L. Harris Dam, which operates as a hydropeaking facility. With the use of radio telemetry both species were tracked over 37 weeks to better understands movement and habitat use of these two species. Movement was strongly associated to season, with both species having the highest movement in the spring. No major difference was observed in movement based on the altered flow regime. However, shifts in habitat use were observed during the altered flows, which may be due to fish relocating to more suitable habitat or for better foraging.

Lastly, stressors, such as alteration in temperature, oxygen or hydrology, can induce acute or chronic stress, which in turn can impact the overall fitness of an organism. Cortisol response is a good indicator of acute stress and additional measurements of stress include leukocyte profiles, with neutrophils increasing and lymphocytes decreasing (N:L). The physiological stress response was studied in both Alabama Bass and Redeye Bass, to determine if the altered flow regime has any impact. Results showed that there is a trend for both baseline cortisol levels and N:L to be higher in the fish found at the disturbed location. Additionally, the percent change of cortisol was higher at the reference site. Results suggest that fish in the treatment site have an altered stress response that may be due to the non-natural flow regime.

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Introduction

Lotic ecosystems around the world have been manipulated and degraded due to the increase in world population and development. Many waterways also suffer from low water quality due to pollution and a variety of land uses. Additionally, water from these rivers has been diverted or impounded for irrigation, recreation, flood control and municipal uses. The World Dam Commission and the International Commission on Large Dams reported that, as of 2000, there were more than 45,000 dams >15 m worldwide, with 8,100 of these dams located in the United States (Gleick et al 2002). The total number of all sizes of dams in the world is unknown, but in 2009 the U.S. Army Corps of Engineer's National Inventory of Dams indicated there was a total of 82,642 dams in the United States, and 50% of these dams were less than 7.5 m in height (Ragon 2009).

There is abundant evidence of the adverse effects of dams on riverine ecosystems (Baxter 1977). Dams interrupt and alter lotic ecological processes by changing the flow of water, sediments, nutrients and biota (Ligon et al. 1995). Changes in sediment and channel morphology can impact the spawning grounds of fish (Jacobson and Galat 2008) and alter the habitat for both fish and macroinvertebrates (Teimann et al. 2004). Dams also impede or delay fish movement both upstream and downstream (Budy et al. 2002; Moser et al. 2002; Zigler et al. 2004), leading to habitat fragmentation (Dunham et al. 1997), and alter temperatures regimes which can impact fish populations by affecting growth or spawning success of native fish, and potentially benefiting non-native species (Edwards 1978; Bestgen and Williams 1994; Marchetti and Moyle 2001; Feyrer and Healey 2003; Holbrook et al. 2009).

Altered flow regimes caused by dam construction and operation can affect aquatic organisms in a variety of ways. Aquatic species have evolved life history strategies based on the natural flow regime; thus, altered flows may lead to lower recruitment; altered spawning and altered growth (summarized in Bunn and Arthington 2002). Additionally, disruption in the natural flow regime may alter food availability for all life stages of fishes below these impoundments (King et al. 2010). Dams often alter the timing and duration of the flow regime leading to reduced connectivity to floodplains and other habitats, as well as reduced effectiveness of fish passage structures (Calles and Greenberg 2009). The result of altered flow regimes may be a reduction in biomass of the macrophyte, macroinvertebrate and fish communities (De Jalon et al. 1994).

There are two major types of hydropower plants, base plants and peaking plants (Egré and Milewski 2002). Base plants are often found in areas where hydropower is abundant and operate to meet needs at base load over an extended period of time. Often these facilities have extra turbines and can operate at times of peak demand (Egré and Milewski 2002). Whereas, peaking plants operate only in peak mode and alternative sources of energy are used for non-peaking hours (Egré and Milewski 2002). Both types of facilities impact stream morphology, including wetted area, water depth, substrate composition, stream structure and heterogeneity (Scruton et al. 2003; Enders et al. 2008). Fishes below these plants, specifically peaking facilities, experience rapid changes in the quantity, quality and location of different habitats (Garcia et al. 2010). Bain et al. (1988) documented that highly variable and unpredictable flow modifications can cause a disturbance to fish due to the inaccessibility of certain habitats. This rapid change in habitat quality and quantity creates an unstable environment (Pert and Erman 1994). Although some habitats become available due to the higher flows, the rapidly falling water levels characterized by ending peaking operations disconnects these same habitats from the main channel, leading to fish stranding (Bradford 1997). Aarts et al. (2004) investigated how fish community structure was affected by river regulation and main channel connectivity to the associated floodplains. They found that there was a decrease in species richness and diversity with decreasing hydrological connectivity between river and floodplain (Aarts et al. 2004). Furthermore, Bowen et al. (1998) provided more evidence that both short-term and annual persistence of key habitats are important for maintaining diverse fish communities, especially nursery habitat for larval fish (Scheidegger and Bain 1995). Additionally, hydraulic refugia are critical for organisms to withstand the variations of physical variables based on site morphology (Valentin et al. 1996). Thus alteration of habitats in these river systems resulting from hydropeaking operations is having a major impact on aquatic organisms, especially fishes.

Altered flow regime may be detrimental to individual fish. Humphries and Lake (2000) found that larval recruitment was impacted by river regulation and that lower recruitment was a greater concern than a lack of fish spawning. The reduction of biomass of drift and benthic invertebrates created a reduction in prey available for drift-feeding fish (Moog 1993). Lagarrigue et al. (2002) showed that feeding patterns of Brown Trout *Salmo trutta* were strongly influenced when the difference between the natural and peaking flows were at the highest. Altered flow regimes may not only impact feeding patterns, but also overall energetic expenditures by fishes. Scruton et al. (2005) suggests that if fish holding their position in current have a high energetic cost when exposed to higher velocities, they will then make large movements to find a substitute habitat; however these energetic costs need to be further studied. It also has been documented with a laboratory study that there is a short-term stress response in juvenile Brown Trout to fluctuating flows, but habituation was observed shortly after (Flodmark et al. 2002). There are also energetic costs associated with the habituation of the stress response since changes need to be made behaviorally and/or physiologically (Scruton et al. 2005). However, very few studies have focused on the individual and sub-organismal responses to hydropower, including the energetic and physiological responses (Hasler et al. 2009).

Rivers and streams are not only important for navigation and hydropower, but also support economically important recreation activities. In 2006, more Americans participated in freshwater recreational fishing than marine recreational fishing (US DOI et al. 2008). These inland fisheries include recreational fishing on lakes, ponds, reservoirs, rivers and streams. Excluding the Great Lakes, more time and money was spent on fishing for black bass and panfish compared to other popular fisheries. In Alabama alone, $700 million was spent on recreational fishing in 2006, and black bass species were the highest targeted fish. Of the estimated 714,000 people that fished Alabama’s freshwater systems in 2006, 61% fished the rivers and streams spending close to $430 million. Thus, recreational fishing on the rivers and streams in the state are important to the economy of Alabama. Understanding the population dynamics of the recreational species is important to sustaining these resources.

The Tallapoosa River, in east-central Alabama, has been extensively impounded for flood control, navigation in the Alabama River, hydropower and water supply. However, the river still supports an important sport fishery for species such as Channel Catfish *Ictalurus punctatus*, Largemouth Bass *Micropterus salmoides,* Redbreast Sunfish *Lepomis auritus,* Redeye Bass (Coosa bass) *M. coosae*, and Alabama Bass *M. henshalli*. There has been previous research on the Tallapoosa River studying fish community responses to the altered flow regime (e.g., Kingsolving and Bain 1993; Travenichek et al. 1995; Freeman et al. 2001). However, there has been minimal work on sportfish (Sakaris 2006; Martin and Irwin 2010; Sakaris and Irwin 2010), especially the black bass found within the river system. More specifically there has been minimal work on sportfish below Harris Dam, which is a hydropeaking facility.

The two sportfish species that will be the focus of this research are Redeye Bass and Alabama Bass. Redeye Bass are native to the Mobile Basin and are distributed above the fall line and are typically found in small to medium sized upland streams and rarely are found in large rivers (Mette et al. 1996). Alabama Bass are also native to the Mobile Basin; however they are widely distributed throughout the system in both lentic and lotic habitats (Baker et al. 2008). These two black bass species are important native sportfish in the Tallapoosa River and understanding how they are affected by the current flow regime will aid in future management of the fishery. Thus, objectives of this research was to investigate the impacts of the altered flow regime on growth, movement, habitat use and the stress response of these two *Micropterus* species.

Study Site

Originating in northwest Georgia, the Tallapoosa River flows 421 km southwesterly across east-central Alabama to its confluence with the Coosa River, forming the Alabama River. The focus area of this research is the section of river downstream of Harris Dam to the headwaters of Martin Reservoir (Lake Martin), covering a distance of approximately 79.5 km. This portion of the river is located in the Piedmont Upland physiographic region of Alabama. This section of river is characterized by a physically stable channel, with low-gradient habitats and silt substrate as well as high-gradient shoal habitats dominated by bedrock and boulders. The flow is highly regulated by Harris Dam, which normally is operated in hydro-peaking mode, where water is released in pulses for 4-6 hours through one or two turbines (capacity of 226 m^3/sec) and power generation can occur once or twice a day, Monday thru Friday (Irwin and Freeman 2002). Dam operation results in extreme fluctuation in flow and stage, especially in the first 20 km downstream of the dam, creating highly variable habitats. Although continued adaptive management procedures are currently underway there are minimal regulations on the minimum flow, the magnitude, or the duration of water releases.

1. Hydro-peaking Effects on the Growth of Alabama Bass and Redeye Bass

Abstract. - Anthropogenic factors such as dam construction and hydropower generation can dramatically alter the flow regime of rivers and may impact growth of aquatic organisms. Using incremental growth techniques, annual growth of Alabama Bass *Micropterus henshalli* and Redeye Bass *Micropterus coosae* in the Tallapoosa River was evaluated in response to variation in flow regime. Fish were collected from Hillabee Creek and the Tallapoosa River above Harris Dam (unregulated sites) and at two sites downstream of the dam (regulated sites). Flow variables were created for each growth year and the best model that described growth for each species at each location was chosen using Akaike's Information Criterion. Additionally, growth increments at age 1, 2 and 3 were compared between a less-variable flow year and one of a higher variation. Lastly, an analysis of covariance was used to compare growth rates of both species at all sampling locations. Age was the best explanatory variable that described growth in all models although flow variables were included in more than half the models. Growth was higher for age-1 fish in years with less flow variation; however growth was similar among years for age-2 and age-3 fish. Overall growth rates for Alabama Bass and Redeye Bass were higher in the unregulated sites than either regulated site. Alabama Bass had higher growth rates than Redeye Bass at the middle and lower sites; however growth was similar between species at the upper site.

Introduction

Age and growth information is often used by biologists to investigate population structure and individual growth patterns. Fish have several anatomical structures (scales, otoliths, and bones) that have identifiable rings that represent growth and are a means for aging (Chambers and Miller 1995). When age information is coupled with fish length, growth history of year-classes can be determined using back-calculation (Frie 1982). This ability to analyze individual growth patterns is a great tool for fishery scientists (Chambers and Miller 1995). The information on past growth provides useful insight on how growth may be impacted based on environmental factors and specific management strategies (Maceina 1992; DeVries and Frie 1996). Additionally, growth influences reproduction (Roff 1983) which is an important factor in understanding the overall fitness of a fish and in population dynamics. Understanding the factors that affect growth can provide guidance for future management.

Hydroelectric dams alter the natural flow regime of rivers, especially when these dams are operated in peaking mode. Yet very few studies have evaluated growth of fishes and flow regimes, whether natural or non-natural. Weisberg and Burton (1993) assessed the growth of White Perch *Morone americana* before and after establishment of a minimum flow on the Susquehanna River, Maryland, and found that increasing the minimum flow increased first-year growth of White Perch. It was also shown in the Connecticut River that larval growth rates of American Shad *Alosa sapidissima* increased asymptotically with declining flows, but there was no relationship with juvenile growth rates (Crecco and Savoy 1985). Additionally in Norway, Jensen and Johnsen (2002) found that growth of Atlantic Salmon *Salmo salar* decreased in years with higher spring flows. A majority of the growth studies have focused on larval and juvenile life-stages, and an important rearing habitat for fish in this life stages are floodplains (Dudley 1974; Schlosser 1991; Sommer et al. 2001; Balcombe et al. 2007; Jeffres et al. 2008). Often, floodplain habitat is only connected to the stream during periods of higher flows, and the lack of connectivity could impact growth (Sommer et al. 2001). These studies provide some evidence that growth can be influenced by river hydrology. However, many of these studies have only evaluated specific life-stages and have not considered growth over a whole lifetime.

Ecologically speaking, a flow regime can be broken into five aspects: magnitude, frequency, duration, timing, and rate of change or flashiness (Poff and Ward 1989). These components can help define the type of river system and how certain anthropogenic factors affect it. The magnitude can be defined as the amount of water that passes a fixed location, and this can be analyzed by looking a minimum, maximum and mean flow (Poff et al. 1997). The definition for frequency is the number of times a certain magnitude is reached (Poff et al. 1997). Duration is the amount of time that is associated with a specified flow, and can be expressed relative to a certain event or overtime (Poff et al. 1997). Timing refers to the regularity in which a certain magnitude occurs (Poff et al. 1997). Lastly, flashiness is defined as how quickly a flow changes from one magnitude to another (Poff et al. 1997). One other component to a flow regime is the time of year that certain flows occur, and typically for southeastern U.S. streams, late-summer to fall is when the low flows typically occur. This seasonality of flows can provide important cues to the aquatic organisms within the systems.

Incremental growth analysis has been widely used in various fields, including marine fisheries but is less commonly used in freshwater fisheries. Rypel (2009) stated that many of the sclerochronology studies were focused on longer-lived fishes, which may explain the lack of studies on freshwater fish which are typically shorter-lived. Quist and Guy (2001) used this technique to assess the effects of habitat and community characteristics on growth of several prairie stream fishes in Kansas. Sammons and Maceina (2009) used incremental growth analysis to evaluate the effects of river flows on Redbreast Sunfish *Lepomis auritus* in Georgia rivers. Additionally, Rypel (2009) investigated the effects of climate on the growth of Largemouth Bass *Micropterus salmoides* in southeastern rivers and reservoirs. These studies demonstrated that growth of freshwater fishes were related to both flow and climate variables. However, little work has been conducted to evaluate how altered flow regimes from hydropeaking operations impacts growth of fishes. Thus the objective of this study was to evaluate growth of Alabama Bass and Redeye Bass in response to hydropeaking operations on the Tallapoosa River, Alabama. It is expected that the altered flow regime will have an impact on the growth of these fish.

Methods

From 2009-2011, I attempted to collect a total of 50 Redeye Bass and 100 Alabama Bass from three locations on the Tallapoosa River: Horseshoe Bend-Germany's Ferry (lower site), Wadley-Price Island (middle site) and the unregulated upper Tallapoosa (upper site) above Harris Dam (Figure 1-1). Due to difficulties in collecting Redeye Bass from the upper Tallapoosa River, additional fish were collected from Hillabee Creek (Hillabee), an unregulated tributary to the Tallapoosa River (Figure 1-1). These sites were chosen to allow examination of the effects of flow variation on age and growth across a gradient of flow variability, as the effects of Harris Dam on the flow regime lessen as the distance below the dam increases (Figure 1-2). The upper and Hillabee sites served as reference (unregulated) locations (Figure 1-1). Fish were collected from the middle and lower sites using a boom-mounted electrofishing boat during periodic sampling that occurred in October 2009 and May, August and October 2010 and 2011. Fish were collected at the upper site using the same gear in March and April, 2011 and fish from the Hillabee site were collected using hook and line sampling in July, 2011. In the laboratory, fish were measured (Total Length, mm), weighed (g) and otoliths were extracted and placed in vials.

Otoliths were broken through the nucleus and mounted onto slides using thermoplastic cement and then ground until a thin section was obtained (Maceina 1988). Otoliths were examined under an image-analysis system, and were measured from the focus to the outer edge; each annuli were measured in a similar manner and the total length at each annulus was calculated using the direct proportion method (DeVries and Frie 1996). Length-frequencies of the back-calculated ages were compared to the observed length-frequencies at each age to verify that back-calculated lengths were similar to actual lengths (Sammons and Maceina 2009). Growth increments for each growth year were determined using the equation:

$L_{inc} = L_n - L_{n-1}$; where,

L_{inc} = growth increments,

L = back-calculated length

n = growth year.

The start and end of a growth year was based on the timing of annuli formation. Based on observations of otoliths collected during the summer, the growth year for both species was defined as July 1-June 30.

River discharge information was obtained from four USGS (http://waterdata.usgs.gov/nwis/rt) gaging stations (02412000, 02414500, 02414715, and 02415000) that were located at or in close proximity to the sampling locations (Figure 1-1). All four of the gaging stations recorded datum every 30 minutes. Variables were calculated describing the five ecological aspects of the Tallapoosa River flow regime at each site to describe the flow regime characteristics (Table 1-1). The annual median discharge was calculated to evaluate magnitude and peaking flows were tallied for each growth year to evaluate frequency (based on Sakaris 2006). Duration was calculated by summing the number of days flow was above and below high and low flow periods, respectively. These two flow periods were selected from a flow duration curve generated using mean daily discharge for water years 2000-2010 at each station (Figure 1-3). The high flow point was selected by the Q5, where flows exceeded this point 5% of the time and represented extreme high flows. The low flow was the number of days the flow was below Q95, meaning that 95% of the time the flows will exceed this point, representing extreme low flows. The rate of change or flashiness was evaluated by calculating the Richards-Baker Flashiness Index (Baker 2004). This index ranges from 0-1, with 0 representing a stable habitat and 1 being extremely flashy. Additionally, the rate of flow change was described using the variance of discharge measured every 30 min by USGS gages. To incorporate a seasonal component, growth years were divided into four seasons: summer (July-September), fall (October-December), winter (January-March) and spring (April-June). Variance of discharge was calculated for each season and compared to the annual growth increments in an attempt to identify important periods for fish growth.

Relations between river flow variables and length increments were examined with a multiple regression analysis (Maceina 1992), using a general regression model:

$$L_{inc} = b_0 - b_1(AGE) + b_i(FLOW);$$

where, b_0, b_1,and b_i are the regression coefficients for the intercept and slope coefficients, and FLOW is one or more flow variables based on season, growth year, and a combination of season and growth year. For each site, candidate models were decided based on Akaike's Information Criteria (AIC) and the best model among these was chosen using the Variance Inflation and Condition indices (Burnham and Anderson 2002; SAS Institute 2004). Semi-partial correlation coefficients (SCORR1, SAS Institute 2004) and the squared partial correlation coefficients (PCORR2, SAS Institute 2004) were calculated to determine the amount of variation that variable accounted for in the best model.

To further assess impacts on growth based on flow variability, a year with high flow variation and a year with low flow variability were selected. Growth increments of both species age 1-3 fish were compared between years with low and high flow variability. Differences in growth increments among years were assessed using student's *t*-test for both species at all the sampling locations.

An analysis of covariance (ANCOVA) was used to evaluate differences in growth among sites for each species. Ages were adjusted by adding 0.083 (1/12) for every month beyond the end of the previous growth year (past June 30) the fish were collected. Ages were log transformed for all analyses. Independent variables in the ANCOVA were age and site, with length at age as the dependent variable. ANCOVAs tested for differences in the slopes between length at age and age among sites for each species (Zar 1984; Pope and Kruse 2007). If slopes were not different, an additional ANCOVA was run to test for differences in the adjusted mean length of each species among sites. This analysis was run for age 1-7 Alabama Bass and 1-5 Redeye Bass to minimize biases associated with low sample size of older fish. A similar analysis was run to test for differences in growth between species at each site (ages 1-4). Significance was set at $\alpha = 0.05$ for all tests.

Results

Overall, 361 Alabama Bass and 170 Redeye Bass were collected over all locations. Of the 531 otoliths, 516 were readable. Readable otoliths were obtained from 69 Alabama Bass and 18 Redeye Bass at the upper site, 147 Alabama Bass and 63 Redeye Bass from the middle site, and 133 Alabama Bass and 50 Redeye Bass from the lower site. An additional 36 Redeye Bass were collected from the Hillabee site, of which 34 yielded readable otoliths.

All candidate models explaining growth of Alabama Bass contained flow variables along with age at each site; however, Akaike weights were < 0.02 for all models at the upper and lower sites which is very low (Table 1-2). In contrast, Akaike weights for candidate models were more than 5-fold higher at the middle site. Most candidate models explaining growth of Redeye Bass also contained flow variables at all sites; however, the best model contained only age as an explanatory variable at both the upper and lower sites (Table 1-3). Similar to Alabama Bass, Akaike weights of models were highest at the middle site, but in general all Akaike weights were higher for Redeye Bass models than Alabama Bass models.

As expected, growth was inversely related to age and age accounted for 60-65% of the variation in the Alabama Bass models and 68-71% of the variation in the Redeye Bass models (Table 1-4). Due to high VIF the below variable was removed for Alabama Bass at the lower site and variance and above variables were removed from the Redeye Bass at the middle site. Overall no model had an environmental parameter that explained more than an additional 2% of the variation in the data (Table 1-4). However, three of the best models included environmental variables, and four of the best models included seasonal flow variables (Table 1-4).

All the best models for Alabama Bass included either an annual predictor or a seasonal predictor. The best models describing the factors that influence growth of Alabama Bass (Table 1-4) were:

Upper Site ($r^2 = 0.65$):

Growth Increments = 137.4917 – 15.0152(Age) – 0.4661(Peaks)

Middle Site ($r^2 = 0.66$):

Growth Increments = 134.3666 – 14.6317(Age) – 0.4063(Above) – 0.0595(Peaks) + 0.0025(Variance) – 0.0006(Summer)

Lower Site ($r^2 = 0.61$):

Growth Increments = 116.9735 – 14.9845(Age) + 0.0009(Fall) – 0.0004(Winter)

Only half of the best models explaining Redeye Bass growth included environmental variables (Table 1-4). The models at the upper and lower sites included only age as an explanatory variable. The best models explaining Redeye Bass growth at each site were:

Upper Site ($r^2 = 0.71$):

Growth Increments = 116.0749 – 23.1784(Age),

Middle Site ($r^2 = 0.74$):

Growth Increments = 154.1478 – 23.1412(Age) – 0.1289(Peaks) + 0.0008(Winter) – 0.0009(Summer)

Lower Site ($r^2 = 0.64$):

Growth Increments = 107.68 – 18.6227(Age)

Hillabee ($r^2 = 0.70$):

Growth Increments = 124.1703 – 22.0230(Age) – 0.0136(Fall)

Growth year 2009 was selected as the highly variable year and 2007 was selected for the low flow year for most comparisons (Table 1-5). Since most fish were collected at the lower site in the fall 2009, growth year 2004 was used for the highly variable year at that site (Table 1-5). There was a noticeable trend for Alabama Bass to experience faster growth in a low flow variability year. This difference was only significant for age-1 fish at the upper site (Figure 1-5; $|t| = 2.93$; $df = 25$; $p = 0.0072$) and age-3 fish at the middle site (Figure 5; $|t| = 2.56$; $df = 27$; $p = 0.0164$). Conversely, growth appeared faster in the high flow variation year for age-1 Alabama Bass at the middle site and age-2 Alabama Bass at the lower site although neither difference was significant ($|t| \geq 1.38$; $df \geq 26$; $p \geq 0.0842$). In contrast, mean growth of age-1 Redeye Bass was faster in the low flow variability year than the high flow variation year at all three sites on the Tallapoosa River (Figure 1-6; $|t| \geq 2.13$; $df \geq 5$; $p \leq 0.0399$). Additionally, growth of age-2 Redeye Bass at the middle site was also faster in the low flow variability year ($|t| = 2.45$; $df = 10$; $p = 0.0342$). However, growth increments were similar between years for all other age and site combinations.

Covariance analysis revealed the growth rate of Alabama Bass was faster at the upper site compared to the lower site ($|t| \leq 3.43$; $df = 2$; $p \geq 0.0007$), but was similar to the middle site ($|t| = 1.11$; $df = 2$; $p = 0.2694$). Growth of Alabama Bass in the middle site was faster than the lower site ($|t| = 2.18$; $df = 1$; $p = 0.0304$). However, Alabama Bass appeared to be larger at age-1 at the middle and lower sites than the upper site. Covariance analysis also revealed that the growth rate or Redeye Bass was faster at the upper site than all the other sites ($F = 4.51$; $df = 3{,}147$; $p = 0.0046$), but was similar among the other sites ($F = 0.93$; $df = 2{,}132$; $p = 0.3968$). Covariance analysis further revealed that the adjusted mean length of Redeye Bass was highest at Hillabee ($\bar{X}$ = 206 +/- 3.41 SE mm), next highest at the middle site ($\bar{X}$ = 188 +/- 2.59 SE mm), and lowest at the lower site ($\bar{X}$ = 177 +/- 2.95 mm; F = 19.93; *df* = 2,134; p < 0.0001). Alabama Bass had faster growth rates than Redeye Bass at the middle site (|*t*| = 3.09; *df* = 1; p = 0.0024), and the lower site (|*t*| = 2.28; *df* = 1; p = 0.0241), but growth rates were similar between species at the upper site (|*t*| = 1.85; *df* = 1; p = 0.0678). However, adjusted mean lengths of Alabama Bass (260 ± 4.38 SE mm) were larger than Redeye Bass (188 ± 8.06 SE mm) at the upper site (F = 61.17; *df* = 1,71; p < 0.0001).

Discussion

In this study it was found that the hydrologic regime had a minor effect on the growth of Alabama and Redeye Bass in the Tallapoosa River. In all models, age was the best predictor variable for explaining the variation in growth. This has been observed in previous studies (Maceina 1992; Sammons and Maceina 2009; Stocks et al. 2011), and was expected to be observed in this study as well. Typically, juvenile fish maintain higher growth rates until they reach sexual maturity in which growth begins to slow down and eventually reaches an asymptote (von Bertalanffy 1938). This growth pattern was observed for both species at all locations. However, flow variables were included in more than half the models, suggesting that the hydrologic regime did have some effect on growth of these species.

The three models that included peaks as a variable predicted that growth declined as frequency of peaks increased. Similarly, Sakaris (2006) found that growth of age-0 Channel Catfish *Ictaluris punctatus* and Flathead Catfish *Pylodictus olivaris* decreased as frequency of peaks increased in the Tallapoosa River. My study demonstrated that Alabama Bass growth was negatively influenced by peak flows at unregulated site. Additionally, Alabama Bass and Redeye Bass growth was impacted by flow variability at the middle site, which experienced the most dramatic flow variations due to the dam operation. However, the peak variable was not included in the models at the lower site. While this site does experience fluctuation in flow from hydropeaking operations, these effects become attenuated as distance from Harris Dam increases and channel width increases. Flow peaks are not as drastic at the lower site as they are at the middle site (although mean daily flows are higher at the lower site).

The remaining models for both Alabama Bass and Redeye Bass included both annual and seasonal predictors; 57% of the best models included a seasonal variable, indicating that the seasonal flow may have some importance to the growth of these fish. Most models that included a seasonal component identified flow in fall or winter as predictors of growth of both species. Growth of Alabama Bass increased with higher flow variation at the middle site and high variation in fall flows at the lower site. Likewise, growth of Redeye Bass increased with higher winter flow variation. Typically unregulated southeastern U.S. streams are at baseflow during the summer and fall (Linn 1997), therefore, increased discharge at a time when flows are normally at baseflow may allow for better foraging conditions. Similarly, high flow variation at the middle site may increase food availability for Alabama Bass and Redeye Bass by increasing drift and thus predation vulnerability of macroinvertebrates (Gore 1977; Beckett & Miller 1982). However, in general most models predicted that growth of Redeye Bass and Alabama Bass was negatively impacted by increased flows or flow variation. Overall, the annual and seasonal flow variables included in the models had very low predictive power and the candidate models had low Akaike weights. Additionally, no other variable explained more than 2% of the variation in the data after accounting for age and were not conclusive in establishing how the flow regime impacted growth. Because different variables impacted growth differently among species, site and season; it is possible that the true impacts of the hydrologic regime were not best defined by the variables chosen to examine growth.

In general, growth of age-1 fish of both species appeared to be greater in years with less flow variability. Flow variability is related to discharge, thus years of low variability are usually in years of low discharge. In streams where there is low flow variability, the environment is more stable (Poff and Allan 1995) and this may allow for prey to be more concentrated. In addition, this may allow for easier access to food and better predation allowing for greater growth. Schlosser (1985) found that juvenile abundances of sunfish and minnows increased in Illinois streams with low flow conditions, while abundance of darters and suckers were similar between low and high discharge years. If the abundance of different prey species increases with low flow, it's possible that predation may be enhanced. However, Power et al. (1996) explained the connection of flow regimes with riverine food webs and the lengths of food chains and that some variation in flow allows predators to have better access to prey without overharvesting. This suggests that there is an optimal level of variation in flow associated with the food chain and this should be further investigated in the Tallapoosa River.

Additionally, *Micropterus* species typically go through an ontogenetic shift in prey, shifting from zooplankton to insects and crayfish then finally becoming piscivores (Phillips et al. 1995; Long and Fisher 2000; Wheeler and Allen 2003). Schlosser (1982) observed higher growth rates of juvenile fish when there was higher primary productivity in headwater streams in east-central Illinois. Before a disturbance, 81% of the sampled locations in an Oklahoma stream had algae associated with them, which then decreased after the disturbance (Power and Stewart 1987). Thus, low disturbance or variability may increase primary production in regulated rivers, leading to higher growth in younger fishes. There appeared to be less effect of hydrology on growth of older fish in the Tallapoosa River, which may have been due to ontogenetic diet shifts or the effect of sexual maturity on growth.

It has also been established that low flow variability may increase spawning success and recruitment of fishes. Freeman et al. (2001) found that persistence of juvenile percids and cyprinids in a regulated portion of the Tallapoosa River was strongly correlated to the temporal variation in flows, based on adult spawning and juvenile rearing times. Smith et al. (2005)

observed that Smallmouth Bass recruitment success in an unregulated river was higher in years with moderate flow than in high-flow years. In Australia, Golden Perch *Macquaria ambigua* recruitment was found to be poor in flood years and strong in non-flood years (Mallen-Cooper and Stuart 2003). Also Nunn et al. (2003) documented that river discharge may impact year-class strength. Thus, recruitment of these black bass species in the Tallapoosa River may be impacted during high-flow years, which could result in a density dependence interaction with growth (Shelton 1981; Matthews et al. 2001). There appeared to be a longitudinal gradient in numbers of age-0 black bass observed during this study. Relative abundance of age-0 fish was noticeably lower at the middle site compared to the lower site, and the highest numbers of age-0 black bass were observed at the lower site. However, the fastest growth rates of Alabama Bass occurred at the middle site, whereas the slowest growth was observed at the lowest site. Thus, density dependence may have influenced some of the results of my study.

The hypothesis was that growth would be impacted at the sites with the most flow variation. The average growth of Alabama Bass and Redeye Bass was higher at the upper site which follows the hypothesis. However, there are some unexpected results from this study. Alabama Bass had growth rates at the middle site that were similar to the upper site, which were both significantly higher than the lower site. For Redeye Bass the growth rates were similar for the three remaining locations, but Hillabee had the highest mean growth of the three, followed by the middle site than the lower. Although Hillabee had higher mean growth, I expected that the growth rate of this site would be similar to the upper site. Additionally, I expected that growth would be faster at the lower site compared to the upper site, since there is less variation in the flow at the lower site. These results differ from what was found in Shoal Bass *Micropterus cataractae*, Spotted Bass *Micropterus punctulatus*, and Largemouth Bass in the Flint River, Georgia, which is a natural flowing river. These three species of black bass were found to have faster growth further downstream with growth decreasing the further upstream fish were collected (Sammons, unpublished data, Auburn University). It was also observed that the fish further downstream were more piscivorous suggesting that diet had an impact on growth (Sammons, unpublished data, Auburn University). Thus, the faster growth observed in the upper Tallapoosa River and Hillabee Creek for both these species suggested that there may have been impacts of the altered flow regime on growth that were not detected in my study. The faster growth at the middle site could be related to the density dependence effects already discussed, the possibility that foraging increased during pulses which increased drift prey (Cushman 1985), increased foraging with the increased flow variation (Power 1996), or possible temperature effects.

Harris dam releases hypolimnetic water and temperatures can fluctuate up to 10ºC with the flow variation (Irwin and Freeman 2002). Temperature is another well-known environmental variable that impacts growth of fishes (Beitinger and Fitzpatrick 1979; Imsland et al. 1996; Deegan et al. 1999). In warm water systems, cold water hypolimnetic releases can be detrimental to native species, specifically at early life stages of these fishes (Hickman and Hevel 1986; Clarkson et al. 2000). However, it would be expected that below the dam temperatures at the middle site would be warmer in the winter and cooler in the summer compared to the lower site. It has been established that black bass feeding efficiency is lower at cooler temperatures and although growth is similar at the moderate and higher temperatures, feed and dietary protein efficiency are higher at moderate temperatures compared to temperatures at the higher end of their range (Tidwell 2003). Temperature preferenda of Alabama Bass and Redeye Bass are unknown, but cooler summer water temperatures could increase growth since high summer water temperatures can results in severely curtailed growth (Nuemann et al. 1994). Diana (1984) also found that Largemouth Bass growth was higher in fish that were exposed to fluctuating temperatures, which may be similar to what fish are exposed to at the middle site. Unfortunately, due to the lack of sufficient temperature records at all the locations this variable was not analyzed in my study. I suggest that further research be completed on the impacts of the temperature on growth on different year classes of these two species.

Overall growth of Redeye Bass in my study was greater than that found by Catchings (1978) in Shoal and Little Shoal creeks, Alabama. My results were more similar to those found by Knight (2011) for Redeye Bass in Hillabee Creek. Knight (2011) noted that Catchings (1978) used scales to age these fish, whereas Knight used otoliths which may explain the differences in growth between the studies. Also, habitat, productivity, and food supplies may be better in the Tallapoosa River and Hillabee Creek compared to the two creeks in Catchings (1978) study. Smaller headwater streams usually rely on allochthonous input for nutrients and are not as productive compared to mid-size streams and larger rivers (Vannote et al. 1980), thus growth rates of fishes are likely higher in medium sized streams, assuming preferred temperatures are not exceeded. To better understand growth of this species research should be completed on different stream orders using the same age and growth method.

As noted by Sakaris (2006) for Channel Catfish, the effect of river hydrology on growth of these black bass species may be more complex that what can be described by linear regression analysis. It is also possible that new flow management strategies may have reduced the impact on growth of these species. In 2005, adaptive management flow pulses were implemented. Instead of releasing water all at once, pulses of water were released over a period of time based on the need. These pulses lessened the rise, reduced the peak, and the fall was less drastic. Yet I was unable to test the hypothesis that this management strategy may have helped lessen the impact on fish growth because there were no otolith samples from fish prior to 2005, when the pulse flows began. Several studies suggest that species that thrive in lentic environments may not be as heavily impacted by regulated streamflow (Bain et al. 1988; Poff and Allan 1995; Stewig and DeVries 2004). I hypothesized that Alabama Bass would not be greatly impacted by the variation in flow because they are a very adaptable species and that Redeye Bass would be impacted because they are a small obligate lotic species. However, this study did not provide strong evidence that growth of either species was heavily influenced by flow.

Management Implications

This study suggests that there is an impact on growth because of the dam, but impact is not directly correlated to the flow regime. Further investigation should be completed on why the two sites below the dam have different growth rates and why growth is faster above the dam. Growth at the middle site was faster compared to the lower site. Understanding if this is due to density dependence effects, temperature or foraging will provide managers with information that can aid in better management. Additionally, research on recruitment should be completed at all locations since it appears that recruitment at the middle site may be the biggest impact of the dam.

Currently the minimum flow requirement for this section of the Tallapoosa River is set at 46 cfs (E. Irwin, Auburn University, personal communication). Extreme low flows in the summer may result in low growth and higher activity (Murchie and Smokorowski 2004), suggesting that there is a minimum flow that allows higher productivity. Minimum flows may help lessen the variability of the flows by eliminating the extreme low flows and may provide a more stable environment. Travnichek et al. (1995) observed a beneficial response in the fish community after implementation of minimum flows on the lower Tallapoosa River below Thurlow Dam. Other studies have shown an increase of growth and abundance fish after implantation of minimum flows (Weisberg and Burton 1993; McKinney et al. 2001). Minimum flows prove to be beneficial to the aquatic biota and although currently there are management pulse flows, further assessing how implementation of higher minimum flows below Harris Dam could prove to be just as beneficial.

Table 1-1. List of hydrologic variables created to explain annual growth of Alabama Bass and Redeye Bass from three sites on the Tallapoosa River and one on Hillabee creek. Below, median, and flashiness are not included in the Wadley global model due to collinearity.

Site	Variable	Definition
Upper site	Above	Days river discharge was ≥ Q5 (44.43 m^3/s)
	Below	Days river discharge was ≤ Q95 (0.74 m^3/s)
	Flashiness	Richard-Bakers Index (0-1)
	Median	Median discharge (m^3/s)
	Peaks	Frequency of peaks greater than 6 m^3/s
	Variance	Variation in 30 min discharge
Middle site	Above	Days river discharge was ≥ Q5 (217.47 m^3/s)
	Peaks	Frequency of peaks greater than 14.2 m^3/s
	Variance	Variation in 30 min discharge
Lower site	Above	Days river discharge was ≥ Q5 (253.33 m^3/s)
	Below	Days river discharge was ≤ Q95 (7.59 m^3/s)
	Flashiness	Richard-Bakers Index (0-1)
	Median	Median discharge (m^3/s)
	Peaks	Frequency of peaks greater than 28.3 m^3/s
	Variance	Variation in 30 min discharge
Hillabee Creek	Above	Days river discharge was ≥ Q5 (22.93 m^3/s)
	Below	Days river discharge was ≤ Q95 (0.39 m^3/s)
	Flashiness	Richard-Bakers Index (0-1)
	Median	Median discharge (m^3/s)
	Peaks	Frequency of peaks greater than 4 m^3/s
	Variance	Variation in 30 min discharge

Table 1-2. The top five candidate models produced from the all model subset analysis for Alabama Bass growth and river hydrologic variables at each sampling location for the entire growth year and season. The model with the lowest AICc score was considered the best model among the candidate models.

Model	K_i	AIC_c	Δ_i	w_i	r^2
Upper					
Age Peaks	4	1922.94	0.00	0.0185	0.6469
Age Below Peaks	5	1923.57	0.63	0.0135	0.6493
Age Flashiness Peaks	5	1924.46	1.53	0.0086	0.6478
Age Peaks Spring	5	1924.64	1.71	0.0079	0.6475
Age Peaks Winter	5	1924.65	1.82	0.0079	0.6475
Middle					
Age Above Peaks Variance Spring	7	5692.98	0.00	0.1173	0.6591
Age Above Peaks Summer Fall Winter Spring	9	5693.53	0.54	0.0894	0.6611
Age Above Peaks Variance Summer Fall Winter	9	5693.75	0.76	0.0801	0.6610
Age Above Peaks Variance Fall Spring	8	5693.85	0.86	0.0760	0.6598
Age Above Peaks Variance Summer Spring	8	5693.80	1.82	0.0472	0.6593
Lower					
Age Fall Winter	5	4120.04	0.00	0.0198	0.6096
Age Peaks Fall	5	4120.36	0.32	0.0168	0.6093
Age Peaks Fall Winter	6	4120.41	0.37	0.0164	0.6110
Age Variance Fall	5	4120.74	0.71	0.0139	0.6090
Age Variance Fall Winter	6	4120.75	0.71	0.0139	0.6107

Table 1-3. The top five candidate models produced from the all model subset analysis for Redeye Bass growth and river hydrologic variables at each sampling location for the entire growth year and season. The model with the lowest AICc score was considered the best model among the candidate models.

Model	K_i	AIC_c	Δ_i	w_i	r^2
Upper					
Age	3	431.42	0.00	0.0494	0.7136
Age Flashiness	4	432.87	1.46	0.0228	0.7187
Age Summer	4	433.22	1.80	0.0200	0.7168
Age Spring	4	433.63	2.21	0.0163	0.7144
Age Below	4	433.71	2.34	0.0156	0.7140
Middle					
Age Peaks Winter Spring	6	1291.16	0.00	0.1402	0.7434
Age Peaks Winter	5	1291.88	0.72	0.0980	0.7384
Age Peaks Fall	5	1292.04	0.88	0.0902	0.7382
Age Peaks Summer	5	1292.10	0.93	0.0878	0.7381
Age Peaks Fall Spring	6	1292.19	1.03	0.0838	0.7417
Lower					
Age	3	999.93	0.00	0.0370	0.6418
Age Summer	4	1001.13	1.20	0.0204	0.6447
Age Variance Spring	5	1001.61	1.68	0.0160	0.6500
Age Spring	4	1001.62	1.69	0.0159	0.6432
Age Fall	4	1001.80	1.87	0.0146	0.6426
Hillabee					
Age Fall	4	769.04	0.00	0.0183	0.7033
Age Spring	4	769.31	0.27	0.0160	0.7023
Age Variance	4	769.60	0.57	0.0138	0.7013
Age Winter	4	769.99	0.95	0.0114	0.7000
Age Above Fall	5	770.42	1.38	0.0092	0.7053

Table 1-4. Overall best models explaining the relation of growth to annual and seasonal flow variables for Alabama Bass and Redeye Bass at each of the sampling locations. The squared partial regression coefficients (PCORR), the semi-partial regression coefficients (SCORR) and P-values for the model variables are included.

Location	Species	Variable	PCORR	SCORR	P-value	r^2
Upper	Alabama Bass	Age	0.6294	0.6310	<0.0001	0.6469
		Peaks	0.0175	0.0473	0.0016	
	Redeye Bass	Age			<0.0001	0.7136
Middle	Alabama Bass	Age	0.6517	0.6354	<0.0001	0.6592
		Above	0.0000	0.0025	0.2144	
		Peaks	0.0006	0.0044	0.0983	
		Variance	0.0019	0.0066	0.0439	
		Summer	0.0022	0.0063	0.0489	
	Redeye Bass	Age	0.7145	0.7003	<0.0001	0.7434
		Peak	0.0031	0.0331	0.0268	
		Winter	0.0073	0.0189	0.0954	
		Spring	0.0186	0.0677	0.0014	
Lower	Alabama Bass	Age	0.5976	0.5901	<0.0001	0.6096
		Fall	0.0062	0.0062	0.0985	
		Winter	0.0082	0.0211	0.0082	
	Redeye Bass	Age			<0.0001	0.6418
Hillabee	Redeye Bass	Age	0.6842	0.6985	<0.0001	0.7033
		Fall	0.0191	0.0604	0.0226	

Table 1-5. Minimum, maximum, mean and variance in the 30-min flow data from the growth years that represent high and low flow variability at the four sampling locations. The asterisks denote data that was collected in growth year 2004, otherwise the year of high variability was 2009 and low variability was 2007.

	Upper	Middle	Lower	Hillabee
High Variability				
Min Flow	1.72	3.08	4.41*	0.80
Max Flow	230.90	725.92	687.99*	298.78
Mean Flow	27.23	125.50	113.21*	15.33
Variance	1201.18	20994.99	11510.95*	777.86
Low Variability				
Min Flow	0.11	1.69	2.98	0.05
Max Flow	95.45	432.10	450.25	96.67
Mean Flow	6.02	27.09	31.96	3.14
Variance	85.32	3292.72	2429.44	32.79

Tren

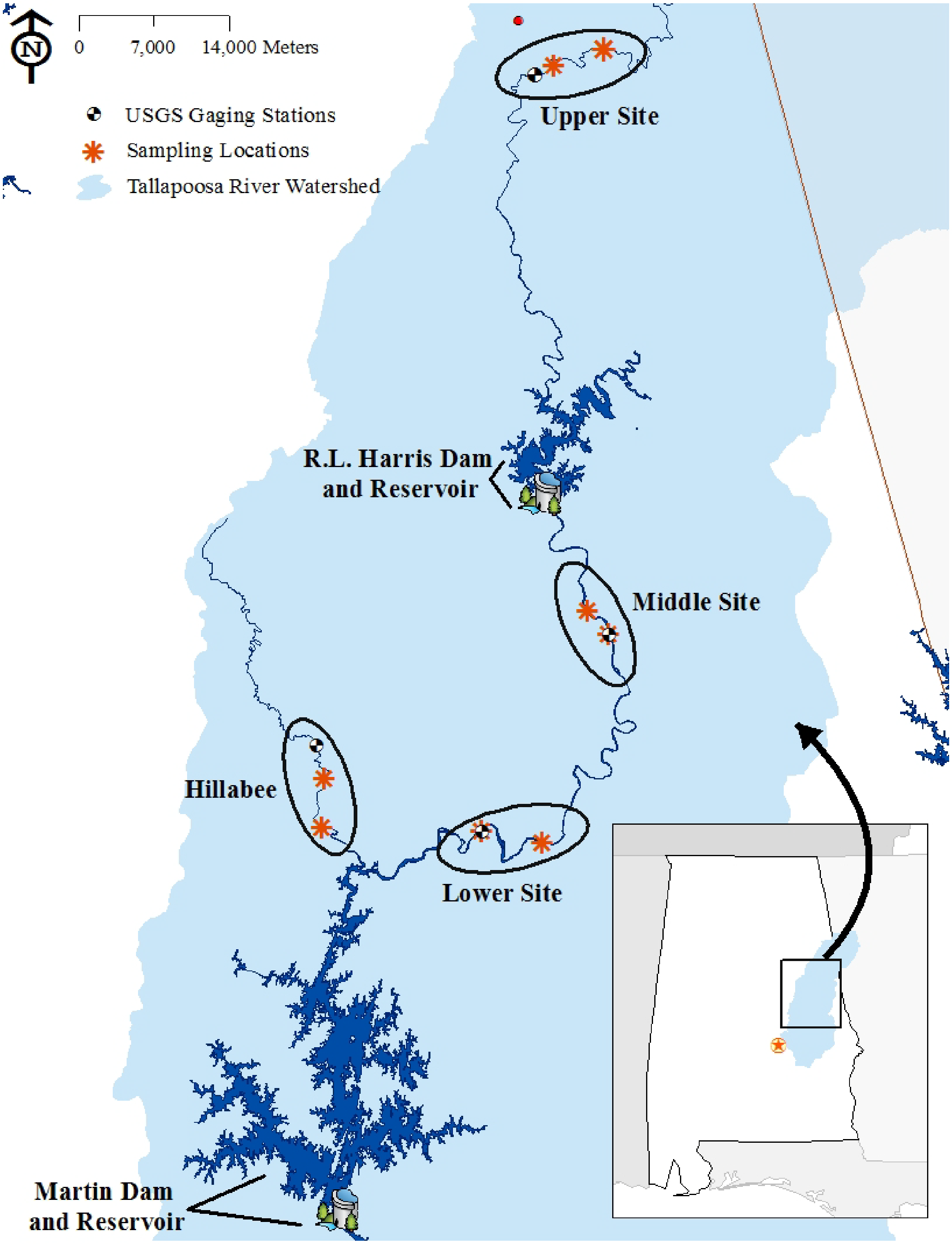

Figure 1-1. Map depicting the sampling locations and the USGS gaging stations in the Tallapoosa River watershed, Alabama.

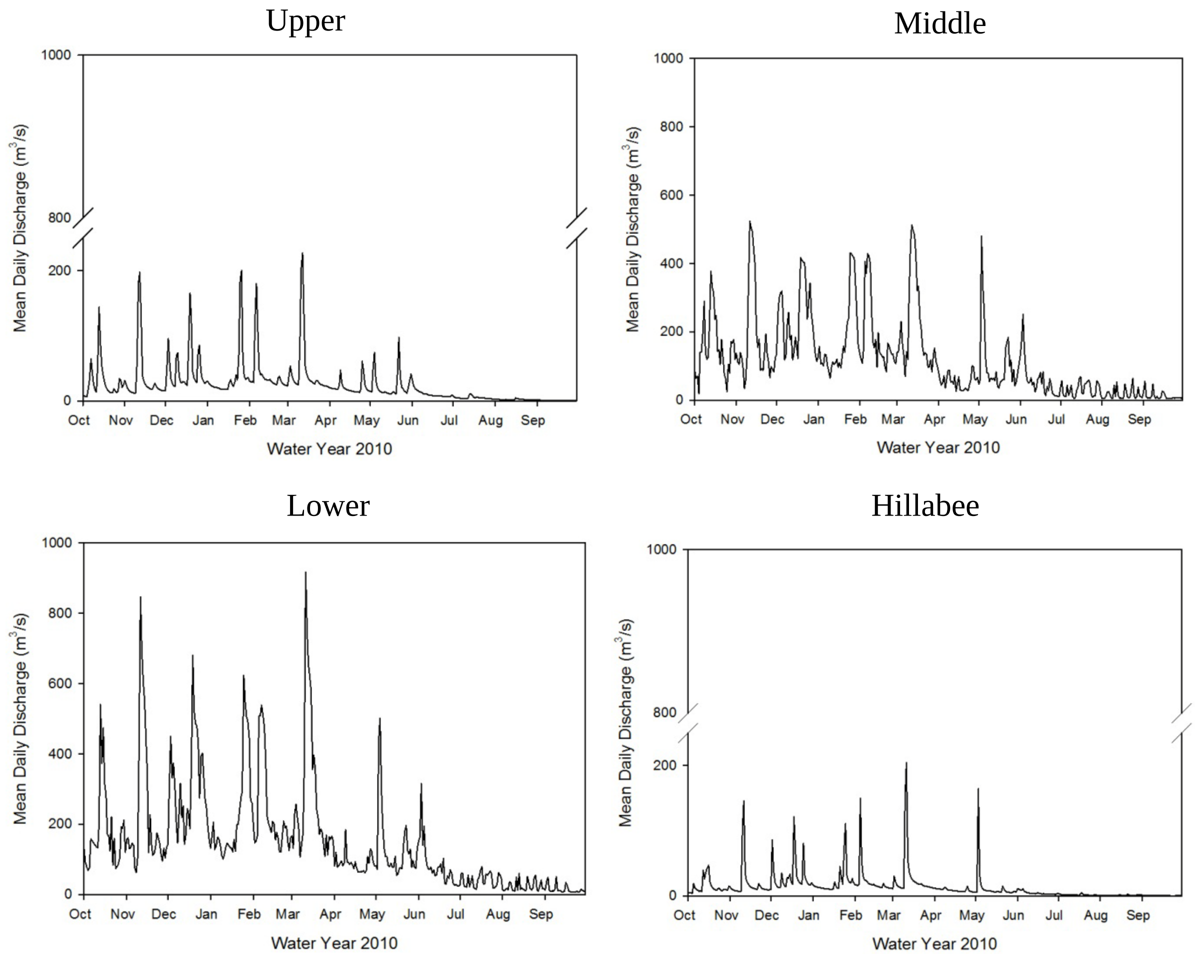

Figure 1-2. Hydrographs for water year (WY) 2010 at four USGS gaging stations (Upper, Middle, Lower and Hillabee).

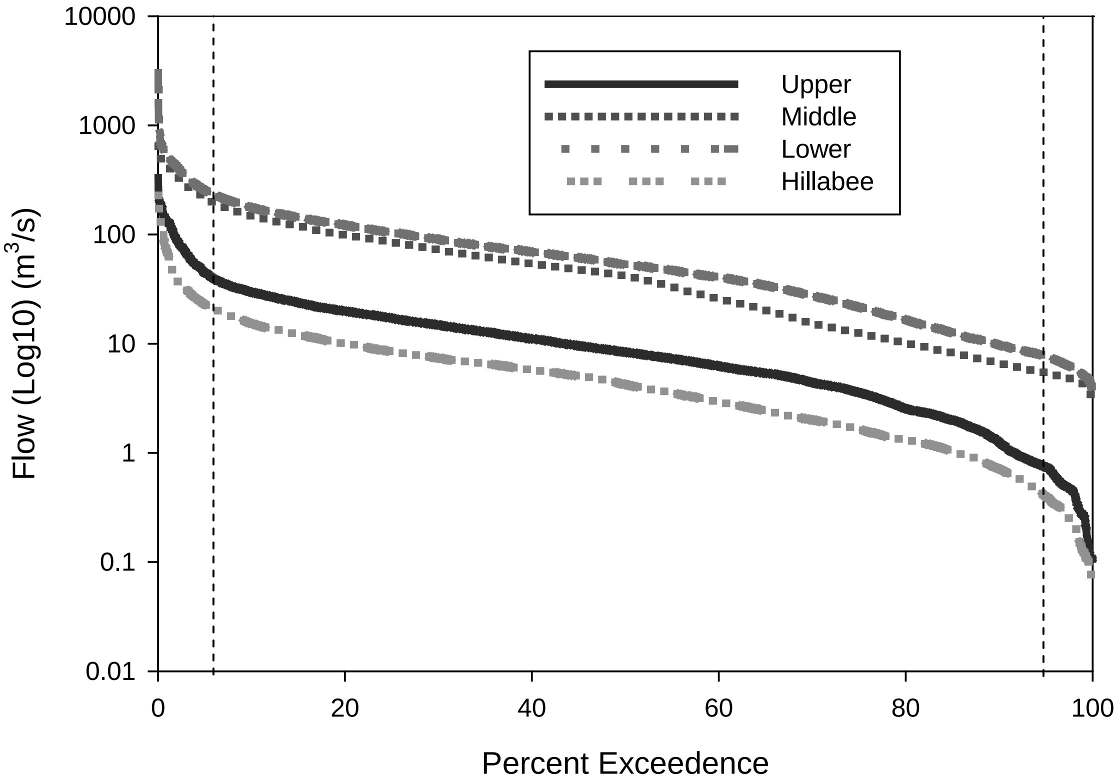

Figure 1-3. Flow duration curves based on mean daily discharges (m3/s) from WY 2000-2010 for the upper, middle, lower and Hillabee creek sites. The vertical dotted lines represent the Q5 and Q95, which are points where the flow will exceed them 5% or 95% of the time.

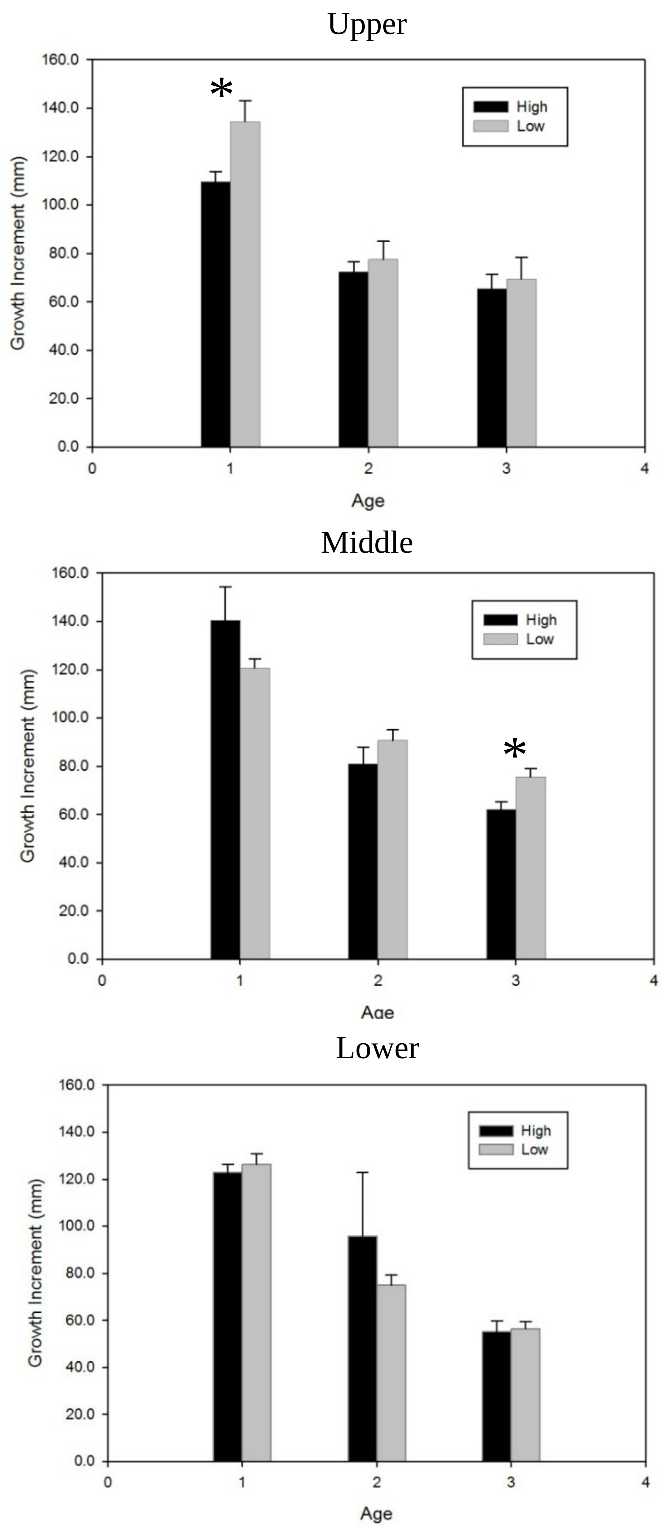

Figure 1-4. Mean growth increments for Alabama Bass in a year with high flow variability and a year with low flow variability. The asterisk represents data that are significantly different (P ≤ 0.05).

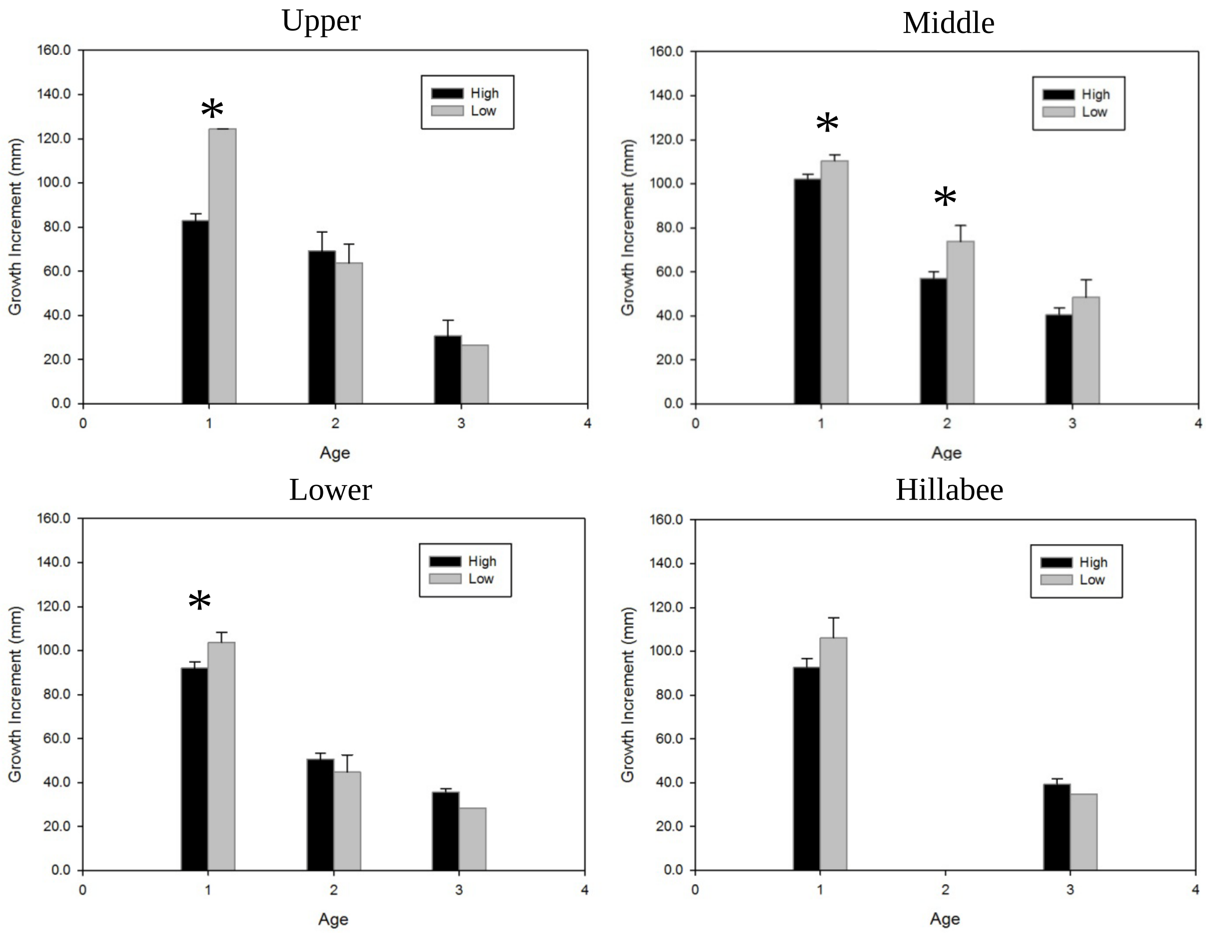

Figure 1-5. Mean growth increments for Redeye Bass in a year with high flow variability and a year with low flow variability. The asterisk represents data that are significantly different ($P \leq 0.05$).

2. Seasonal and Hydro-Peaking Operation Impacts on Movement and Habitat Use of Alabama Bass and Redeye Bass

Abstract. –Alabama Bass *Micropterus henshalli* and Redeye Bass *Micropterus coosae*, are two native game fish in the state of Alabama for which few studies have been conducted. Little is known about the movement and habitat use of these species in the Tallapoosa River, specifically below R.L. Harris Dam, which operates as a hydropeaking facility. With the use of radio telemetry 22 Alabama Bass and 20 Redeye Bass were tracked for 37 weeks, from December 2010 to September 2011. Starting January 31, 2011, 8-9 fish were tracked every 2 h over the course of 10 h. These diel tracks attempted to assess the movement and habitat use by the two species during different aspects of the hydrograph (base, rising, peak, and falling flows). All fish continued to be tracked every 3-4 weeks to ensure that fish had not moved out of the area, helped identify possible spawning areas for these fish and to gather locations to be used in establishing home ranges. When fish left the extended area, fish were located on float trips or during one aerial survey completed on June 20, 2011. Movement of both species was strongly associated to season, with the highest movement observed in the spring. Both Alabama Bass and Redeye bass movement did not appear to be affected by the altered flow regime or the different dam operations (peaking vs. pulsing flows). Total home range (95%) and core areas (50%) of both species were similar, however Redeye Bass total home range decreased as fish size increased. Shifts in substrate and cover habitat were observed by season, flow period and dam operations. Alabama Bass were typically found in fine sediment substrates but increasingly used more woody debris for cover from winter to summer. Alabama Bass habitat use differed among seasons during the different flow periods. Redeye Bass were typically found in rocky substrate

but less rocky cover and more woody debris in summer months. Similar to Alabama Bass the substrate and cover used by Redeye Bass during different flow periods was dependent on season. Both species used vegetative cover the most in spring. Although movement was not greatly impacted, the shifts in habitat during different flows should be further investigated to see if this a negative impact of the dam.

Introduction

Biologists often study movement of animals to better understand their behavior. Kahler et al. (2001) stated that movement might be one of the most important behavioral patterns because of the animal's ability to respond to conditions in order to increase growth, survival or reproductive success. Additionally, understanding what influences fish movement allows for the capability to predict how individual fish and/or populations respond to changing environmental factors (Albanese et al. 2004). Radio telemetry is widely used to establish movement behavior (extents and patterns) of fish (Tyus 1990; Guy et al. 1992; Moser et al. 2002) and developing home ranges for these fish (Tyus 1990; Vokoun and Rabeni 2005; Stormer and Maceina 2009). Studying fish behavior in riverine habitats can also help to better understand potential barriers to movement (Thompson and Rahel 1998; Warren Jr. and Pardew 1998), metapopulation dynamics (Schlosser 1995), predator-prey interactions (Gillam and Fraser 2001) and ontogenetic shifts in habitat use (Johnston and Kennon 2007). Movement of lotic fish species has been shown to depend on season or time of day, which can be associated with spawning or feeding behavior (Grabowski and Isely 2006; Parsley et al. 2008). Establishing the extents and possible impacts of seasonal and daily movements can be helpful in managing game species.

An important aspect of fish movement is determining home ranges, which include the core and overall area used. However, an animal will only have a home range if they exhibit site fidelity (McGrath 2005). Some of the first fisheries studies that evaluated home ranges concluded that riverine fish tended to be sedentary and had very restricted home ranges (Miller 1957; Gerking 1959); however, some more recent studies have established larger home ranges for many lotic species. Resident salmonid species may make large scale movements and remain in their home range (Gowan and Fausch 1996; Hilderbrand and Kershner 2000). Todd and Rabeni (1989) found that movement of Smallmouth Bass *Micropterus dolomieui* in a Missouri river ranged from 120-928 m/d, although there was a home pool that was used a majority of the time. Similarly, Largemouth Bass *Micropterus salmoides*, Spotted Bass *Micropterus punctulatus*, and Shoal Bass *Micropterus cataractae*, moved 8-20 km from their home location in the Flint River, Georgia (Goclowski 2010). Vokoun and Rabeni (2005) found that Flathead Catfish *Pylodictis olivaris* in two Missouri streams moved upstream and downstream of the core area, although many fish returned to the core area, and home ranges varied for individual fish regardless of the river, sex, or size. Even though there is a large variation in home range based on individuals, the knowledge of the average home range size and specific core areas may aid in the management of sportfish.

Movement of fish can be affect by availability or variety of habitat found in a particular system. In rivers, these habitats can be broken into mesohabitat types (run, riffle, pool, and glide) or microhabitats (depth, velocity, substrate, and cover) (Stormer 2007). Habitat selection and use can vary with life history stages (Schlosser 1991, Johnston and Kennon 2007), time of day (Brenden et al. 2006) or seasonally (Grabowski and Jennings 2009). Understanding habitat preferences of riverine fish can improve management as habitat degradation continues to persist.

Hydropeaking operations usually result in changes to the physical conditions of fish habitat and the availability of this habitat (De Vocht and Baras 2005). Valentin et al. (1996) found that hydraulic refugia are critical for organisms to withstand the variations of physical variables. Telemetry can be used to determine if fish behavior is affected by fluctuating flows. Young and Isely (2007) found that hydropower discharges affected the movement of adult

Striped Bass *Morone saxatilis* but these effects did not negatively impact the fish as long as the habitat was not degraded based on the operations.   By using radio telemetry, Brenden et al. (2006) found that river discharge affected the habitat used and selected by Muskellunge *Esox masquinongy* in the New River, Virginia. Further research on the movement and habitat use of fish during times of hydropeaking and at base flow is needed to increase understanding of how fish behavior and habitat use are affected by the altered flow regime.

Redeye Bass are native to the Mobile Basin and are distributed above the fall line, typically found in small to medium sized upland streams and rarely in large rivers (Mette et al. 1996). Alabama Bass are also native to the Mobile Basin and are widely distributed throughout the system in both lentic and lotic habitats (Baker et al. 2008). These two black bass species are important native sportfish in the Tallapoosa River; however, little information exists on the movement and habitat use of Alabama Bass and Redeye Bass. Alabama Bass are typically considered to be a habitat generalist (Baker et al. 2008); whereas, Redeye Bass can be defined as a habitat specialist (Boshung and Mayden 2004). Little data exists on the effects of hydropower peaking flows on the movement and habitat use of these two species within the Tallapoosa River below R.L. Harris Dam. Thus, the objectives of this study were to determine seasonal distribution, movement and habitat use patterns of Alabama Bass and Redeye Bass. Additionally, I assessed the effects of hydropower peaking operations on the movement and habitat use of these two black bass species. I expected movement to be greatest in the spring which would be associated to spawning migrations. I also expected that hydropower peaking flows would result in altered fish behavior and habitat use as they sought refuge from higher flows. Expected responses included increased use of large coarse substrate such as bedrock and use of flow refuges found along riverbanks or in off-channel areas.

Methods

In November 2010, 22 Alabama Bass and 20 Redeye Bass were collected from Price Island and Wadley, Alabama, areas of the Tallapoosa River (Figure 2-1) by electrofishing and angling. Radio tags (Advanced Telemetry Systems [ATS]) were surgically implanted into Alabama bass > 180 g with a 3.6-g tag with a unique frequency number that had a 335-d life expectancy (ATS Model F1580). Redeye Bass > 155 g were implanted with a 3.1-g tag with a unique frequency number that had a 257-d life expectancy (ATS Model F1570). Both of these minimum weights ensured that the tags were not > 2% of the body weight of the fish (Winter 1996), and the tagging procedures followed Maceina et al. (1999). Prior to surgery, each fish was anaesthetized in a solution of 150 mg/L tricane methanesulfonate. A small incision of 1 to 3 cm, depending on transmitter size, was made slightly to one side of the midventral line just posterior to the pelvic girdle. The antenna of the transmitter was pulled through the body wall, from inside, approximately 2 cm posterior to the incision using a cruciate needle. The transmitter was inserted into the body cavity and the incision was closed with 2 to 3 monofilament sutures. Betadine antiseptic was applied to the incision and the fish was placed in a holding tank for recovery. Fish were released at the site of capture after they had regained equilibrium.

Tracking of the fish began on December 1, 2010, ten days after the last fish was tagged, which allowed adequate time for the fish to recover from the surgery. Tracking continued weekly until January 31, 2011 when hydropeaking tracking began. All tracking was completed by September 26, 2011. The location of each fish was determined by moving along the stream in a boat until a signal was detected. Tracking would continue until the signal was strongest when the antenna was pointed at the water. At that point, the location of the fish was recorded using a global positioning system (GPS) unit (Lowrance *i*Finder). Water temperature and weather was recorded at the beginning of each day. Habitat characteristics were also collected, including river depth (m), water velocity (m/s), substrate and cover (Table 1; adapted from Gordon et al. 2004). The substrate was classified based on the composition of the streambed within a half meter radius of where the fish was located. The cover classification was based on the surrounding material that provided cover for the fish. River discharge data were obtained from the U.S. Geological Survey gaging station located at Wadley, Alabama (02414500; http://waterdata.usgs.gov/nwis/rt).

Beginning on January 31, 2011, weekly hydro-period tracking (diel) was conducted, rotating between the Wadley and Price Island locations (Figure 1). I attempted to track four Redeye Bass and four Alabama Bass on each sample date, but as fish redistributed themselves throughout the study reach, this sample size was not always achieved. Two tracking trips were attempted during each week to try to encompass the different hydropower peaking operations and different aspects of the hydrograph. Whenever possible, I attempted to track the same fish each survey period unless they moved out of the area. On each tracking trip, fish were located every 2 h over a 10-h period. Each observation during the diel track was classified into a flow category based on the following categories (Sammons 2011): base/low flow (flow low and steady), rising flow (flow increasing), peak flow (flow high and steady), and falling flow (flow decreasing). Diel tracking continued until tracking ceased on September 26, 2011. Locating all fish continued every 3-4 weeks to ensure that fish had not moved out of the area, to help identify possible spawning areas for these fish and to gather locations to be used in establishing home ranges. When fish left the diel tracking areas, they were located on float trips using a canoe. Additionally, one aerial survey was conducted on June 20, 2011 to ensure fish had not moved below the area surveyed by canoe or up tributary streams. A fish was classified as a mortality if the tag was recovered, if the tag was located on shore or there was little movement (> 0.05 m) within four tracks.

Daily movements of each fish were calculated based on the distance moved and the amount of time that lapsed between each location (Wilkerson and Fisher 1997; Sammons et al. 2003). Overall movement was calculated based on the number of days since the last observation:

$$\text{movement (MDPD)} = \frac{\text{distance from previous location (m)}}{\text{days between observations (d)}}$$

Diel movements were calculated based on the amount of time (h) that lapsed since the previous observation:

$$\text{movement (MDPH)} = \frac{\text{distance from previous location (m)}}{\text{days between observations (h)}}$$

All movement data were categorized into four seasonal groups based on water temperatures and time of year: winter (temperature < 11 °C; December 2010-Febuary 2011), spring (temperature 11-25 °C increasing; March-May 2011), summer (temperature > 25 °C; June-August 2011) and fall (temperature > 25 °C decreasing; September 2022). Additionally the distance from the shore and dam were calculated for each fish location. Movement upstream and downstream was calculated by taking the difference from the previous location's distance from the dam, a negative number meant the fish moved upstream to its next location and a positive number meant the fish's next movement was downstream. Diel movement data were further classified into two more categories; flow and operation. The flow categories were the same as previously described, and was selected based on the flow over the majority of the time that elapsed between observations. Operational data were broken into three groups based on the actual dam operations that happened while tracking occurred that day: peaking (hydro-peaking operations), pulsing (adaptive management pulses) and no operations (no operations).

Mixed model analysis (PROC mixed; SAS Institute 2004) with each tag as the random effect, was used to determine if there were seasonal differences (based on weekly observations) in movement, distance from shore (m), distance from the dam (m), upstream/downstream movement (m), depth and velocity used by the fish. This analysis was completed for both species and then used to compare species. Diel data were also analyzed using a mixed model analysis to determine if there were differences in movement (hourly), distance from shore, distance from dam, upstream/down-stream movement, depth and velocity, based on season and the flow categories and the hydropower operations (pulsing v. peaking) for each species. All movement data were transformed using the natural log. Differences detected in any of the mixed models were examined using a least squares comparison with a Bonferroni correction ($P = 0.05/n$).

Home ranges were calculated for fish with more than 15 observations, using the kernel density estimator (Seaman and Powell 1996; Rogers and White 2007) with the likelihood cross-validation as the smoothing parameter (Horne and Garton 2006). Site fidelity of each fish was tested using the Monte Carlo random walk test developed by Spencer et al. (1990) and modified by Hooge et al. (2001). This test was completed using the Geospatial Modeling Environment (GME; Beyer 2012), for ArcGIS 10 (Environmental Systems Research Institute, Redlands, California). Any fish that did not show site fidelity was excluded from the home range analysis (Spencer et al. 1990; Hooge et al. 2001). The overall home range was represented by the 95% density estimate and the more localized or core range was calculated by using a 50% density estimate (Hooge et al. 2001; Sammons et al. 2003). A student's *t*-test was used to compare the core and overall ranges between the two species. A linear regression was used to determine if there was a relationship between home range and fish size for each species.

Comparisons of microhabitat were used to see if there was a difference between: seasons, species, flow periods and hydropower operations. The frequency of substrate and cover use was compared among seasons and species. The frequency of substrate and cover use for each species was compared to the different flow periods and hydropower operations. All data were compared with goodness-of-fit χ^2 tests (SAS Institute 2004; Stormer 2007). Significance was set at α = 0.10 for all tests due to the relatively low number of fish tagged and the inherent variability of telemetry data in general (Sammons et al. 2003).

Results

Tracking Results and Fish Distribution

A total 37 weeks of tracking were completed from December 1, 2010 to September 26, 2011. This included 16 weekly tracks and 22 weeks of diel tracking (one week both a diel and weekly survey were conducted), totaling 33 days of diel tracks. At the start of the diel tracks my intention was to switch between the Price Island and Wadley locations, however only 5 days of tracking occurred at Price Island due to fish moving to areas inaccessible at lower flows or moving completely out of the study area. During weekly tracks, fish were located over a 41-km length of the Tallapoosa River (Figure 2-1). During the aerial survey of the whole river and associated tributary streams, none of the missing fish were found outside the area surveyed on weekly tracks. A total of 560 (322 Alabama Bass; 238 Redeye Bass) weekly observations and 1280 (734 Alabama Bass; 547 Redeye Bass) diel observations were made during my study. Fish were found 4 to 40 times during weekly tracks (counting only one observation for each diel survey) and the fish were found in 1 to 26 diel surveys (Table 2-2). In May, 2011, an angler returned two tags from harvested fish, one each from an Alabama Bass and a Redeye Bass. Of the remaining Alabama Bass, three more were found dead, four disappeared and were never relocated, and 14 were still alive at the end of the study (Table 2-2). Of the remaining Redeye Bass, nine were found dead, five disappeared and were never relocated, and five were alive at the end of the study. Four tags were recovered during surveys, and three of these tags were implanted into two more Redeye Bass and one more Alabama Bass. Of those fish, the Alabama Bass disappeared and was never relocated and both Redeye Bass were alive at the end of the study (Table 2-2). In total there were 12 mortalities, four tags in which we found and the other eight were classified based on location and movement. Overall, 45% of Redeye Bass and 14% of Alabama Bass died during the study.

Both Alabama Bass and Redeye Bass generally remained within 9 km of their tagging location, however, fish of both species moved 20 km upstream almost to R. L. Harris Dam. An Alabama Bass was located 15.5 km downstream of the Highway 22/77 Bridge (Figure 2-1). Longitudinal distribution of both species followed a noticeable seasonal pattern (Figure 2-2). During winter, most fish remained within 1 km of their tagging location, but some dispersion occurred in the spring by both species, with Redeye Bass generally displacing upstream and Alabama Bass displacing both upstream and downstream. Most of these fish returned to their tagging areas by summer and were found in that vicinity in the beginning of fall, although only one survey was completed in that season (Figure 2-2). Fall observations were removed from subsequent analyses because of the low sample size.

Seasonal Movement

Alabama Bass movement was greatest in the spring (144.31 +/- 1.66 SE m/d) and winter (120.90 +/- 1.60 SE m/d), and least in the summer (10.42 +/- 1.89 SE m/d) ($F = 6.19$; $df = 2,56$; $p = 0.0037$; Figure 2-3). Similarly, Redeye Bass movement was highest in the spring (56.87 +/- 2.03 SE m/d) than in the winter (5.42 +/- 1.76 SE m/d) and summer (2.24 +/- 3.08 SE m/d) ($F = 4.49$; df = 2, 40; $p = 0.0174$; Figure 2-3). Movement of Alabama Bass was higher than Redeye Bass in the winter ($F = 33.51$; $df = 1,317$; $p < 0.0001$), but movement was similar between the species in the other two seasons ($F \leq 1.62$; $p \geq 0.2058$; Figure 2-3).

Alabama Bass distance from shore ($F = 0.50$; $df = 2,57$; $p = 0.6071$), distance from the dam ($F = 1.53$; $df = 2,57$; $p = 0.2255$) and movement up and downstream ($F = 0.69$; $df = 2,57$; p

= 0.5061) were similar among seasons (Figure 2-4). Similarly, Redeye Bass distance from shore (F = 1.44; *df* = 2,46; p = 0.2464) and distance from dam (F = 2.37; *df* = 2,46; p = 0.1046) were similar among season. Fish exhibited similar upstream movement in spring and winter but was different in the summer (F = 11.03; *df* = 2,46; p < 0.0001; Figure 2-4). There were no differences between the species and the seasonal distance from shore (F ≤ 2.42; p ≥ 0.1322), seasonal distance from the dam in the summer and spring (F ≤ 1.76; p ≥ 0.1965) and movement upstream and downstream in the winter and spring (F ≤ 0.48; p ≥ 0.4944; Figure 2-4). However the species differed in the distance from the dam in the winter (F = 7.27; *df* = 1,40; p = 0.0102) and the upstream and longitudinal stream movement in the summer (F = 5.68; *df* = 1,26; p = 0.0248; Figure 2-4)

Home Range

A total of 29 fish (15 Alabama Bass, 14 Redeye Bass) had > 15 locations and therefore could be used for home-range analysis. All Redeye Bass and 14 of 15 Alabama Bass exhibited site fidelity and were retained for analysis. The average core area for Alabama Bass was 22 ha (0.82-111.18 ha) and 26.62 ha (5.90-68.43 ha) for Redeye Bass (Figure 2-5). A total of 36% of the Alabama Bass had more than 1 core area, compared to 64% of the Redeye Bass. Total home ranges averaged 81.39 ha (3.05-249.44 ha) and 85.63 ha (14.42- 231.77 ha) for Alabama Bass and Redeye Bass, respectively. The core area accounted for 9 to 45% of the total home range for Alabama Bass, and 20-45% of the total Redeye Bass home range (Figure 2-5). Sizes of both the core area (|*t*| = -0.49; *df* = 26; p = 0.6305) and total home range (|*t*| = -0.16; *df* = 26; p = 0.8721) were similar between the two species. There was no relationship between the length of Alabama Bass and the size of the core area (r = 0.08; p = 0.7677) or total home range (r = 0.11; p = 0.7095). There was a relationship between Redeye Bass size and core area (r = -0.50; p =

0.0678). There was a significant decrease in total home range area as Redeye Bass size increased ($r^2 = 0.30$; $p = 0.0428$; Figure 2-6).

Diel Movement

Alabama Bass hourly movement was greater in the winter than spring and summer ($F = 7.59$; $df = 2,31$; $p = 0.0021$; Figure 2-7). Redeye Bass hourly movement was also different by season, but unlike Alabama Bass, Redeye Bass had the least amount of movement in the winter (12.76 +/- 2.02 SE m/h) and movement was greater in spring and summer ($F = 33.09$; $df = 1,19$; $p < 0.0001$; Figure 2-7). Hourly movement of Alabama Bass was greater than Redeye Bass in the winter and spring ($F \geq 3.71$; $p \leq 0.0661$) but was similar in the summer ($F = 1.53$; $df = 1,13$; $p = 0.2378$; Figure 2-7).

Flow Period Movement

Diel locations were used to assess how hourly movement was impacted by different river stages (Figure 2-8). During many of the diel tracks, several Alabama Bass were observed using a tributary and occasionally a Redeye Bass was found close to another tributary. Alabama Bass were often observed in the tributary when flow was increased; however fish were found in the tributary a few times during base flow when the tributary was not dry (Figure 2-8). Alabama Bass and Redeye Bass hourly movement was similar across all flow periods in all seasons (Figure 2-9). However, movement of Alabama Bass was greater than Redeye Bass in spring during the rising ($F = 12.69$; $df = 1,21$; $p = 0.0018$), peak ($F = 3.10$; $df = 1,21$; $p = 0.0935$) and falling ($F = 7.50$; $df = 1,20$; $p = 0.0127$) flows, summer falling flows ($F = 4.31$; $df = 1,11$; $p = 0.0622$), and in winter during base ($F = 24.27$; $df = 1,19$; $p < 0.0001$), peak ($F = 8.49$; $df = 1,10$; $p = 0.0155$) and falling flow ($F = 15.72$; $df = 1,19$; $p = 0.0008$; Figure 2-9).

Alabama Bass were located farther from shore during peak flows than during falling flows in the winter; whereas distance from shore was similar between base and rising flows ($F = 3.02$; *df* = 3,35; $p = 0.0429$; Figure 2-10). In contrast, Alabama bass were found closer to shore during peak flows than during base, rising and falling flows in spring ($F = 0.0076$; *df* = 3,48; $p = 0.0076$). However, distance from shore of Alabama Bass was similar among flow periods in the summer (Figure 2-10). Similarly, distance from shore of Redeye Bass was similar among flow periods in the winter and summer ($F \leq 0.37$; $p \geq 0.7778$), but there was a difference between the flow periods in spring ($F = 2.29$; *df* = 3,32; $p = 0.0974$). Although there was as difference detected in spring for the flow periods, there were no difference among them. Redeye Bass were located farther from shore than Alabama Bass during peak flows in the spring ($F = 4.76$; *df* = 1,16; $p = 0.0444$), summer falling flows ($F = 3.97$; *df* = 1,11; $p = 0.0716$); but otherwise distance was similar between species in all other flow period and season combinations ($F \leq 1.98$; $p \geq 0.1966$). Flow period did not affect the distance from the dam for either species, but Alabama Bass were closer to the dam in summer base flows ($F = 3.31$; *df* = 1,13; $p = 0.0921$) and falling flows ($F = 5.37$; *df* = 1,11; $p = 0.0408$) compared to Redeye Bass. Flow periods did not impact whether a fish moved upstream or downstream from its previous location for either species and there were no difference between the species.

Dam Operations

Alabama Bass exhibited greater movement during peaking operations than pulsing flows in the spring ($F = 5.69$; *df* = 1,23; $p = 0.0257$), but movement was similar between flow regimes in the other seasons ($F \leq 0.67$; $p \geq 0.4235$; Figure 2-11). In contrast, movement of Redeye Bass was similar between peaking and pulsing operations in each season ($F \leq 0.57$; $p \geq 0.4654$). Alabama Bass exhibited greater movement than Redeye Bass during spring peaking operations

(F = 7.12; *df* = 1,22; p = 0.0141) and in winter during both peaking (F = 31.24; *df* = 1,17; p < 0.0001) and pulsing (F = 16.59; *df* = 1,15; p = 0.0010; Figure 2-11).

Alabama Bass were closer to shore (F = 15.09; *df* = 1,10; p = 0.0030) and further from the dam (F = 703.11; *df* = 1,10; p < 0.0001) during peaking operations than during pulsing operations in the winter and further from the dam during spring pulsing operations (F = 4.16; *df* = 1,22; p = 0.0536), although distance from shore and from the dam were similar between operations in the other seasons (F ≤ 2.89; p ≥ 0.1032). In the winter Alabama Bass were more likely to move downstream during peaking operation and upstream during pulsing operations (F = 3.39; *df* = 1,10; p = 0.0955). Redeye Bass were closer to the dam during winter pulsing (F = 421.37; *df* = 1,7; p < 0.0001) and spring peaking (F = 3.54; *df* = 1,12; p = 0.0842) operations. Otherwise the distance to shore, the distance from the dam and upstream/downstream movement was similar between the dam operations in the other seasons (F ≤ 3.17; p ≥ 0.1002). During winter pulsing operations Redeye Bass were closer to shore than Alabama Bass (F = 7.77; *df* = 1,6; p = 0.0317), and during summer peaking operations Alabama Bass were closer to the dam than Redeye Bass (F = 4.56; *df* = 1,10; p = 0.0585) otherwise behavior was similar among the operations in the other seasons (F ≤ 2.25; p ≥ 0.1649).

Habitat Use

The unknown category was used for 4% of the substrate observations in both weekly and diel surveys; and 16% and 12% of the cover observations in the weekly and diel surveys, respectively. Alabama Bass were mostly found in boulder, assorted rock, and fine substrate, but this varied across seasons ($\chi^2 \leq 24.37$; *df* = 6 p = 0.0004), with bedrock substrate being used more in the spring and summer (Figure 2-12). Substrate use by Redeye Bass also varied across seasons, with higher use of bedrock and lesser use of assorted rock during the winter (χ^2 =

113.10; *df* = 6; p < 0.0001). Although Alabama Bass used woody debris as cover a majority of the time in all seasons, use of this cover increased in the summer along with use of bedrock and boulders, with a concomitant decline in the use of vegetation and areas of no cover (χ^2 = 64.78; *df* = 13; p < 0.0001; Figure 2-13). Redeye Bass used bedrock more than woody debris as cover during the winter, but use of these two cover types was more similar in the summer; additionally, use of vegetative cover was highest in the spring (χ^2 = 50.58 *df* = 12; p < 0.0001). Alabama Bass were located in deeper water in winter compared to spring and summer (F = 24.09; *df* = 2,57; p < 0.0001; Figure 2-14). Stream velocities at fish locations were higher during the spring compared to summer for Alabama Bass (F = 2.54; *df* = 2,55; p = 0.0878). Redeye Bass were located deeper in winter and spring than in summer (F = 5.56; *df* = 2,44; p = 0.0070) and in faster water in spring than in winter or summer (F = 4.28; *df* = 2,44; p = 0.0200; Figure 2-14). Alabama Bass were found in deeper water than Redeye Bass in the winter (F = 29.81; *df* = 1,40; p < 0.0001; Figure 2-14), but were found in similar depths the other two seasons (F ≤ 1.75; p ≥ 0.1981). Redeye Bass were found in faster flows than Alabama Bass in the winter (F = 3.43; *df* = 1,40; p = 0.0715) and spring (F = 4.35; *df* 1,37; p = 0.0441), but flow velocity was similar between species in the summer (F = 0.00; *df* = 1,22; p = 0.9857).

Flow Periods and Habitat Use

Alabama Bass changed their substrate use during the different flow periods in winter (χ^2 = 2.66; *df* = 9; p = 0.0034; Figure 2-15) and spring (χ^2 = 27.70; *df* = 9; p = 0.0011; Figure 2-16), however there was no difference in summer (χ^2 = 8.90; *df* = 9; p = 0.4461; Figure 2-17). In the winter the substrate used shifted from more assorted rock during base flows to fine sediments during falling flow (χ^2 = 5.01; *df* = 2; p = 0.0819) and using all available substrates during peak flows to not using boulder substrates in falling flows (χ^2 = 11.72; *df* = 3; p = 0.0084). Otherwise all other flow combinations in the winter used similar substrate ($\chi^2 \leq 4.88$; $p \geq 0.1802$; Figure 2-15). In the spring base and rising flows were similar ($\chi^2 = 2.54$; *df* = 3; p = 0.4580). Areas with fine sediment were used more during peak flows compared to higher bedrock use during base ($\chi^2 = 15.08$; *df* = 3; p = 0.0018), rising ($\chi^2 = 7.69$; *df* = 3; p = 0.0526), and falling ($\chi^2 = 12.62$; *df* = 3; p = 0.0055) flows. Bedrock was used less during falling flows compared to base ($\chi^2 = 14.03$; *df* = 3; p = 0.0029) and rising ($\chi^2 = 8.46$; *df* = 3; p = 0.0374; Figure 2-16) flows. The cover that Alabama Bass used shifted during the different flow periods in all three seasons: winter ($\chi^2 = 27.76$; *df* = 12; p = 0.0060), spring ($\chi^2 = 104.89$; *df* = 18; $p < 0.0001$) and summer ($\chi^2 = 51.80$; *df* = 18; $p < 0.0001$). In the winter during base and rising flows ($\chi^2 = 5.50$; *df* = 4; p = 0.2399) and rising and peak flows ($\chi^2 = 1.57$; *df* = 3; p = 0.6652), Alabama Bass were located in areas with similar cover. However the fish chose areas with more bedrock in peak flows than during base flows ($\chi^2 = 9.34$; *df* = 4; p = 0.0530) and areas with more woody debris in falling flows compared to base ($\chi^2 = 11.18$; *df* = 4; p = 0.0246), rising ($\chi^2 = 10.03$; *df* = 4; p = 0.0398) and peak ($\chi^2 = 10.99$; *df* = 4; p = 0. 0267; Figure 2-15) flows. During the spring, Alabama Bass used vegetation as cover more during peak flows compared to any of the other flow periods: base ($\chi^2 = 61.17$; *df* = 5; $p < 0.0001$), rising ($\chi^2 = 38.07$; *df* = 5; $p < 0.0001$) and falling ($\chi^2 = 41.58$; *df* = 4; $p < 0.0001$). Although base and rising ($\chi^2 = 5.32$; *df* = 4; p = 0.2559) and base and falling flows were similar ($\chi^2 = 10.49$; *df* = 6; p = 0.1226), rising and falling differed ($\chi^2 = 13.81$; *df* = 4; p = 0.0318) with more vegetation used in the falling flows (Figure 2-16). Although woody debris was used as cover a majority of the time during all the flow periods in the summer, fish used undercut banks and vegetation during peak flows compared to base ($\chi^2 = 29.45$; *df* = 6; $p < 0.0001$), rising ($\chi^2 = 11.73$; *df* = 5; p = 0.0388) and falling ($\chi^2 = 16.79$; *df* = 6; p = 0.0101) flows. Cover used was similar in the remaining flow combinations ($\chi^2 \leq 8.57$; $p \geq 0.1277$; Figure 2-17).

Similar to Alabama Bass, Redeye Bass switched the substrates used during the different flow periods in winter ($\chi^2 = 19.49$; $df = 9$; $p = 0.0213$; Figure 2-18) and spring ($\chi^2 = 11.99$; $df = 6$; $p = 0.0623$; Figure 2-19), but not in the summer ($\chi^2 = 0.86$; $df = 6$; $p = 0.9903$; Figure 2-20). During winter peak flows, Redeye Bass used less bedrock and more assorted rock than during all the flow periods: base ($\chi^2 = 7.08$; $df = 2$; $p = 0.0291$), rising ($\chi^2 = 7.55$; $df = 2$; $p = 0.0229$) and falling ($\chi^2 = 4.52$; $df = 5$; $p = 0.0335$). Although bedrock use was high during falling flows, the use of assorted rock increased instead of using boulder substrate in base flows ($\chi^2 = 4.93$; $df = 2$; $p = 0.0846$) and fine substrate in rising flows ($\chi^2 = 4.90$; $df = 2$; $p = 0.0861$; Figure 2-18). During spring peak flows the similar pattern of use was observed, less bedrock and more assorted rocks and fine sediment areas were used compared to base ($\chi^2 = 10.37$; $df = 2$; $p = 0.0056$) and falling ($\chi^2 = 6.75$; $df = 2$; $p = 0.0342$). The other flow periods had similar patterns of substrate use ($\chi^2 \leq 4.08$; $p \geq 0.1301$; Figure 2-19). The cover that Redeye Bass used was similar during all flow periods in the winter ($\chi^2 = 2.99$; $df = 6$; $p = 0.8096$; Figure 2-18) and summer ($\chi^2 = 21.05$; $df = 18$; $p = 0.2768$; Figure 2-20), however it changed in the spring ($\chi^2 = 71.78$; $df = 15$; $p < 0.0001$; Figure 2-19). During spring peak flows more vegetation was used as cover compared to the other periods where there was higher bedrock use: base ($\chi^2 = 36.28$; $df = 4$; $p < 0.0001$), rising ($\chi^2 = 14.92$; $df = 4$; $p = 0.0049$), and falling ($\chi^2 = 36.91$; $df = 4$; $p < 0.0001$). Cover use was similar for the remaining flow combinations ($\chi^2 \leq 6.44$; $p \geq 0.2657$; Figure 2-19). Depths and velocities increased for both species in all seasons when the flow was in the rising and peak periods but there was not a correction applied for the increase of discharge so the depth and velocity analysis was removed.

Hydro-power Operation and Habitat Use

The substrate used by Alabama Bass changed in all the seasons between the two different dam operations (Figure 2-21). In the winter Alabama Bass used areas with more fine sediment during peaking flows and areas with more rocky habitats during pulsing flows ($\chi^2 = 14.88$; $df = 3$; $p = 0.0019$). During spring peaking flows Alabama Bass were found in locations with more fine sediment then bedrock ($\chi^2 = 19.44$; $df = 3$; $p = 0.0002$). In the summer Alabama Bass were found in areas with fine sediment during peaking flows ($\chi^2 = 18.95$; $df = 3$; $p < 0.0003$). Cover use for Alabama Bass was also different during the two dam operations in all the seasons (Figure 2-22). In the winter Alabama Bass were more often located in areas with woody cover and did not use bedrock for cover during peaking operations compared to the pulsing operations ($\chi^2 = 25.78$; $df = 4$; $p < 0.0001$). Although in the spring the use of woody cover was similar during both operations with more vegetative cover was used during peaking operations ($\chi^2 = 56.88$; $df = 6$; $p < 0.0001$). During summer peaking operations Alabama Bass were not found in areas with no cover and used more boulder cover than during peaking operations ($\chi^2 = 15.60$; $df = 6$; $p = 0.0161$).

The substrate that Redeye Bass used did not switch based on the dam operations in any of the seasons ($\chi^2 \leq 2.25$; $p \geq 0.5228$; Figure 2-23). Additionally, the cover used by Redeye Bass was not different between the two dam operations in the winter ($\chi^2 = 0.4225$; $df = 2$; $p = 0.8096$) or the summer ($\chi^2 = 9.2$; $df = 6$; $p = 0.1624$; Figure 2-24). However during peaking operations Redeye Bass were often found in bedrock, vegetative or woody areas; whereas during pulsing operations fish were located in all cover types except vegetative cover ($\chi^2 = 13.11$; $df = 3$; $p = 0.0224$).

Depth of Alabama Bass was similar between hydropower operations in winter and summer ($F \leq 0.17$; $p \geq 0.6846$), but were found in deeper water during peaking operations in the spring (F = 7.52; *df* = 1,176; p = 0.0067; Figure 2-25). Depth of Redeye Bass was similar between hydropower operations in each season ($F \leq 2.05$; $p \geq 0.1557$). Redeye Bass were found deeper than Alabama Bass during pulsing operations in the spring (F = 6.90; *df* = 1;127; p = 0.0097) and both peaking (F = 16.58; *df* = 1,93; p <0.0001) and pulsing (F = 5.55; *df* = 1, 164; p = 0.0197) operations in the summer. Alabama Bass were found in deeper water during winter pulsing operations (F = 3.86; *df* = 1,52; p =0.0549), otherwise depth was similar between the species in the other season and operation combinations ($F \leq 1.36$; $p \geq 0.2455$; Figure 2-25). Alabama Bass were found in higher velocities during spring peaking (F = 50.89; *df* = 1,176; p < 0.0001) and summer peaking (F = 4.69; *df* = 1,155; p = 0.0318) and winter pulsing (F = 3.90; *df* = 1,72; p= 0.0523; Figure 2-26) operations. Similarly, Redeye Bass were found in higher velocities during spring peaking (F = 14.65; *df* = 1,123; p = 0.0002) and summer peaking (F = 9.49; *df* = 1,102; p = 0.0027) operations, but velocity use was similar between operations in the winter (F = 0.10; *df* = 1,72; p = 0.7502; Figure 26). Redeye Bass were found in faster flow velocities than Alabama Bass during both spring pulsing (F = 4.40; *df* = 1,127; p = 0.0379) and peaking (F = 2.72; *df* = 1,172; p = 0.0979), but use was otherwise similar between species in all other season and operation combinations ($F \leq 2.38$; $p \geq 0.1261$).

Discussion

General Behavior and Habitat Use

Overall, a greater percentage of Alabama Bass survived compared to the percentage of Redeye Bass. Knight (2011) found that the tag retention of Redeye Bass was very high. At least 90% of the Redeye Bass mortalities in my study may have been caused by predation or fish may have died and then were eaten, but many tags were found in close proximity to what appeared to be animal dwellings. The fate of the fish that disappeared and never relocated was unknown; however. It is possible that the tag expired prematurely or that they were harvested by anglers and removed from the river, since no fish were located outside the survey area during the aerial survey.

Movement was greatest for both species during spring months, which has been commonly observed in other riverine *Micropterus* species, such as Shoal Bass (Stormer and Macenia 2009; Sammons 2011), Guadalupe Bass *Micropterus treculii* (Perkin et al. 2010), and Smallmouth Bass (Todd and Rabeni 1989). Spring movements are usually considered to be associated with spawning for most fish (Todd and Rabeni 1989; Pegg et al. 1997; Snedden et al. 1999; Palmer et al. 2005; Goclowski et al. In Press). The greatest movement of Alabama Bass and Redeye Bass in the Tallapoosa River occurred in April, and they spent several weeks in what appeared to be suitable habitat for spawning and then moved back to the general vicinity they occupied prior to migration. This time period was 1 to 2 months earlier than spawning times of Spotted Bass and Redeye Bass reported in other studies (Parsons 1954; Ryan et al. 1970), but was similar to spawning times of Shoal Bass, Largemouth Bass, and Spotted Bass in the Flint

River, Georgia (Goclowski 2010). Although flow pulses have been considered to be used by riverine fishes as spawning cues (Auer 1996; Jonsson and Jonsson 2009), migrations of Alabama Bass and Redeye Bass in April did not appear to coincide with any major changes in flow, therefore there may have been other cues for these species, including photoperiod and temperature. Although the suspected spawning movement in the Tallapoosa River was not related to flow, there was evidence of flow inducing movement. In March, heavy rains that caused resulted in a continual release using two turbines from Harris Dam for a week. An increase in movement was observed during these higher flows, but the fish did not move out of the central area till several weeks after this event. This suggests that movement may be influenced by flow, and could be related to fish trying to find more suitable habitat because of the higher flows (Albanese et al. 2004).

Movement of Redeye Bass was lower in the winter, which is likely a temperature-related effect. *Micropterus* species have been commonly found to display limited movement during the winter (Todd and Rabeni 1989; Karchesky and Bennett 2004; Hunter and Maceina 2008; Goclowski et al. In Press). Similar to other black bass, Redeye Bass in the Tallapoosa River selected an overwintering location and remained there until water temperatures warmed. However, Alabama Bass had significantly higher movement in the winter and spring than summer, which is contradictory to the results of Hunter and Maceina (2008) in Lake Martin, Alabama. Horton and Guy (2002) observed that Spotted Bass in a Kansas stream had higher movement in spring and fall than in the winter and summer. Similarly, Woodward and Noble (1999) found that while movement of Largemouth Bass in a North Carolina reservoir was lower in the winter, fish remained active throughout the season. Although movement was high for the Alabama Bass in the Tallapoosa River during the winter months, the fish did not move out of 8 km reach of river that fish were first collected. Considering hourly movement of Alabama Bass also remained high during the winter, these fish may have used multiple overwintering locations, similar to what was observed in Largemouth Bass in Idaho (Karchesky and Bennett 2004).

Even though Alabama Bass tended to move more than Redeye Bass, the species had similarly sized home ranges. I expected Alabama Bass would be more mobile and have a larger home range than Redeye Bass. However, in the spring both species moved > 20 km from their tagging locations, which was unexpected for Redeye Bass. Knight (2011) found that the core areas for Redeye Bass in a tributary to the Tallapoosa River ranged from 0.003 to 0.583 ha, and the total home range was 0.030-2.622 ha, which were much smaller than what I observed in this study. The fish in Knight's study (2011) tended to remain in shoal complex and minimal movement was observed outside of this area. It is possible that this was the best habitat available for these fish and there was no reason for movement to occur. Also, his study was conducted in the fall and results may have been different if the study occurred in the spring. Knight (2011) also found that the size of Redeye Bass home ranges decreased with increasing fish size, which was also observed in this study. Mean home range size for Alabama Bass was also higher than what Horton and Guy (2002) found with Spotted Bass in a Kansas creek. However, the Tallapoosa River was a larger system than those investigated in previous studies and home range size may be correlated to the size of the stream. In a small Alabama stream, Stormer and Maceina (2009) found that total home range of Shoal Bass averaged 0.47 ha in a 1.7-ha study area. In a larger system (54.4-ha), Sammons (2011) found Shoal Bass to have a slightly larger home range with approximately 40% of the fish having home ranges that were 5 to 10 ha. Observations were made of fish traveling 10-km upstream in a nearby tributary, suggesting that home ranges may be larger if fish were not restricted. In a study on a 200-km undammed reach of the Flint River, GA, (Goclowski, unpublished data, Auburn University) most Shoal Bass had no home ranges because of a lack of site fidelity and high movement rates. These results suggest that movement and home range size may increase with the size of the stream, which could explain why the home ranges of both Alabama Bass and Redeye Bass in the Tallapoosa River were larger than what was observed by similar species. Additionally, movement was strongly associated with season for both species. When fish moved out of the area in spring they returned to the core area by the summer. These results suggest that home range size was driven by the spring migrations. Core areas may be more important than the total home range, since the fish spend the majority of the time in these areas.

Similar to daily movement, hourly movement was different in the seasons. Hourly movement was greater for Alabama Bass in the winter compared to Redeye Bass; however it was similar in the other two seasons. It appears that besides winter the hourly movement for these species is similar but on a day-to-day basis Alabama Bass movement is greater. However, the core area and overall home ranges were similar for these species, which would suggest that overall movement is similar. It is possible that Alabama Bass move around in their core areas to forage more than Redeye Bass.

Although habitat differed among seasons, Alabama Bass used similar substrate throughout all the seasons and were found in both rocky and fine substrates. There was a shift in cover use for Alabama Bass, but more than 40% of the time in any season they were found in association with woody debris, which is similar to what has been found with Spotted Bass (Horton and Guy 2002; Goclowski 2010). Redeye Bass were often found in association with rocky substrates which was expected (Parsons 1954; Knight 2011), but the use of fine substrates increased in the summer. This shift in substrate use may have been in association with available habitat with the fact that there were more periods at base flows. Cover use also shifted in the summer for Redeye Bass, with woody debris being used more than other seasons. Both species were found in deeper water in winter, which is similar to other black bass studies (Todd and Rabeni 1989; Karchesky and Bennett 2004). The trends in velocities, were that Redeye Bass were found in areas with higher velocities in the winter and spring compared to Alabama Bass and Redeye Bass selected areas with higher velocities in the spring compared to the other seasons.

Effects of Hydropower Operations on Fish Behavior and Habitat Use

Hourly movement of both species did not appear to be impacted by the different flow periods. During most flow periods Alabama Bass exhibited greater movement then Redeye Bass. In the winter during base, peak and falling flows Alabama Bass movement was almost twice as much than Redeye Bass. During spring base flow, summer base, rising and peak flows Alabama Bass hourly movement rate was similar to Redeye Bass. This may suggest that either Redeye Bass are more sedentary species or that Alabama Bass are better adapted to the altered flow regime. Additionally in the spring Alabama Bass were found closer to shore during peak flows and although there were no differences in the distance from shore in spring for Redeye Bass, the fish were also observed closer to shore. Similar results were found for hydropeaking operations; with Alabama Bass having greater movement than Redeye Bass, especially in winter months when Redeye Bass appeared to have little movement. There were no specific trends in ether species relationship to the shore, dam or movement up and downstream. Although the actual total movement was not different during the different flow periods or the dam operations, it is possible that the reasons for movement were different, i.e. feeding (Sammons 2011) or searching for better habitat.

Similar to previous telemetry studies completed during hydropower operations, habitat shifts were observed for Alabama Bass and Redeye Bass during the flow periods for both species (Bunt et al 1999; Brenden et al. 2006). Alabama Bass used all available substrates that had vegetative/woody debris or boulders for cover during peak flow periods in winter and spring. Redeye Bass used more areas that were classified as assorted rock during winter and spring peak flows, but this could be an artifact of the classification of rocky habitat, which was unknown rock habitat and it was lumped into the assorted rock category. At higher flows it was harder to tell the substrate and it is possible that Redeye Bass were still using bedrock substrate. Also during peak flows in the spring both Alabama Bass and Redeye Bass were found to use vegetative cover more than woody debris (Alabama Bass) or bedrock (Redeye Bass). In a study in the Hudson River, 93% of Largemouth Bass nest were found in association with vegetative cover (Nack et al. 1993). It is possible that vegetative cover is important in spawning site selection for Alabama Bass and Redeye Bass.

During the dam operations the Alabama Bass used less bedrock substrates during peaking operations and Redeye Bass used more assorted rock habitat and fine substrate during the pulsing operations. Depth and velocity increased as the flow increased and this could be due to fish choosing deeper habitats, similar to what Pert and Erman (1994) found in Rainbow Trout *Oncorhynchus mykiss* in a flow-regulated river in California; however the increase of depths and velocity associated with the rise in flow may have compounded these results. Similarly, Alabama Bass and Redeye Bass in the Tallapoosa River were found in deeper water and higher velocity during peaking operations than during pulsing operations, but these results could have been affected by the rise in flow associated with peaking operations

Conclusion

Overall it appears that movement was influenced by season for both Alabama Bass and Redeye Bass, but flow periods and dam operations had little impact on the movement and habitat use of these species. Other studies have documented little impact on movement during hydropower operations on Shoal Bass (Sammons 2011) and Brown Trout *Salmo trutta* (Bunt et al. 1999). Unlike these studies I did not observed fish moving closer to bank during higher flows, which may suggest that there were flow refugia created by instream boulders or woody debris in this section of the Tallapoosa River. Redeye Bass had little movement during higher flows and were often associated with rocky cover.   These fish may use the eddies created by the rocky habitat that they already occupy, and do not have to move far to find more suitable habitat; whereas Alabama moved more in higher flows and this could be for foraging or seeking more suitable habitat (Albanese 2004).

Further research should be completed on the movement and habitat use of these two species. I suggest that a telemetry study be completed on unregulated tributaries to the Tallapoosa River to compare movements over the seasons and flow patterns. This will allow for a better understanding of movement characteristics in a natural system. This will also aid in understanding what impacts the altered flow regime and the different dam operations have on movement. Temperature monitoring should also be incorporated into a study, which may aid in finding the optimal temperatures for both species in a regulated and unregulated system. Incorporating habitat availability into the study will provide more insight on how much the altered flow regime is impacting the habitat used by these two species. Additionally, this study located possible spawning areas for both species. My study suggests that Redeye Bass spawned in areas close to vegetation, further upstream which is narrower than the more widely used area. Further research should be completed to gain a better understanding of where these fish are spawning and possible habitat requirements. Lastly an energetics study would provide more information on whether the flows are negatively impacting movement or growth (Chapter 1) of these fish.

Table 2-1. Classification of the substrate and cover categories used for microhabitat collection.

Substrate	Abbr.	Definition	Cover	Abbr.	Defintion
Bedrock	BR	Irregular and connected	Bedrock	BR	Irregular and connected
Boulder	BD	> 25 cm	Boulder	BD	> 25 cm
Assorted Rock	AR	< 25 cm or unidentifable rock	Assorted Rock	AR	< 25 cm or unidentifable rock
Fines	FN	Sand, Silt, Clay (<0.2 cm)	Woody Debris	WD	Combination of large wood (trees and logs) and small wood
Unknown	UNK	Unknown	Undercut Bank	UCB	Undercut banks
			Vegatation	VEG	Combination of submerged and overhanging vegatation
			No Cover	NC	No Cover
			Miscellanous	MISC	Non-natural cover
			Unknown	UNK	Unknown

Table 2-2. Species, length, weight, date tagged, last day located, number of locations and fate of the Alabama Bass and Redeye Bass tagged in the Tallapoosa River. Tag number with an astrisk represents tags that were found and re-used to tag another fish. Number of locations indicates the number of surveys the fish was located in. The number in parenthesis indicates the number of diel surveys the fish was located in.

Tag #	Species	TL (mm)	Wt (g)	Date Tagged	Date Last Located	No. Locations	Fate
001	Redeye	240	166	11/11/2010	6/21/2011	13	Unknown
010	Redeye	331	466	11/11/2010	5/26/2011	18 (5)	Unknown
041	Redeye	268	238	11/18/2010	5/10/2011	18 (5)	Harvasted
061	Redeye	255	203	11/18/2010	8/18/2011	16	Alive
081	Redeye	248	186	11/11/2010	1/4/2011	4	Mortality
101	Redeye	270	253	11/11/2010	2/23/2011	7	Mortality
120	Redeye	226	152	11/18/2010	2/21/2011	10 (2)	Mortality
140	Redeye	263	236	11/9/2010	9/26/2011	25 (9)	Alive
162	Redeye	280	269	11/18/2010	4/6/2011	10	Unknown
183	Redeye	243	165	11/11/2010	8/18/2011	21 (5)	Alive
202	Redeye	242	177	11/11/2010	4/6/2011	10	Mortality
220	Redeye	257	240	11/11/2010	1/24/2011	7	Morality
242	Redeye	236	164	11/18/2010	8/18/2011	16	Alive
261	Redeye	238	166	11/18/2010	8/1/2011	40 (26)	Mortality
281	Redeye	241	187	11/18/2010	7/12/2011	30 (16)	Mortality
302	Redeye	242	184	11/11/2010	4/28/2011	10	Unknown
311	Alabama	480	1242	11/10/2010	3/16/2011	10 (1)	Mortality
322	Redeye	261	250	11/10/2010	4/26/2011	16 (5)	Mortality
341	Redeye	239	156	11/10/2010	4/26/2011	16 (5)	Mortality
352	Alabama	476	1405	11/10/2010	7/18/2011	17 (4)	Unknown
361	Redeye	245	173	11/18/2010	6/21/2011	20 (6)	Unknown
381	Redeye	246	174	11/18/2010	8/17/2011	17	Alive
401	Alabama	435	1072	11/9/2010	8/17/2011	28 (13)	Alive
421	Alabama	247	190	11/9/2010	8/17/2011	14 (1)	Alive
440	Alabama	306	264	11/9/2010	8/17/2011	22 (6)	Alive
461	Alabama	447	1293	11/9/2010	8/17/2011	28 (17)	Alive
481	Alabama	494	1440	11/10/2010	8/17/2011	20 (4)	Alive
502	Alabama	350	465	11/9/2010	8/17/2011	29 (13)	Alive
522	Alabama	314	330	11/9/2010	9/26/2011	22 (6)	Alive
542	Alabama	262	191	11/9/2010	4/22/2011	11 (1)	Mortality
561	Alabama	342	488	11/9/2010	8/17/2011	18 (2)	Alive
580	Alabama	499	1391	11/9/2010	9/26/2011	34 (18)	Alive
600	Alabama	474	1352	11/9/2010	5/9/2011	14 (3)	Harvasted

Table 2-2. continued.

Tag #	Species	TL (mm)	Wt (g)	Date Tagged	Date Last Located	No. Locations	Fate
620	Alabama	353	487	11/9/2010	4/26/2011	15 (4)	Mortality
640	Alabama	472	1228	11/9/2010	8/17/2011	18 (2)	Alive
661	Alabama	483	1414	11/9/2010	8/17/2011	32 (16)	Alive
681	Alabama	465	1279	11/10/2010	5/10/2011	14 (3)	Unknown
701	Alabama	415	890	11/9/2010	5/10/2011	12	Unknown
721	Alabama	314	334	11/9/2010	8/17/2011	31 (15)	Alive
741	Alabama	432	1070	11/10/2010	8/17/2011	16 (2)	Alive
761	Alabama	282	221	11/9/2010	5/9/2011	13 (1)	Unknown
779	Alabama	492	1354	11/9/2010	8/17/2011	27 (12)	Alive
4110*	Redeye	264	230	6/14/2011	9/26/2011	14 (11)	Alive
1010*	Redeye	247	261	4/22/2011	9/26/2011	20 (15)	Alive
6200*	Alabama	320	350	5/13/2011	7/18/2011	5 (2)	Unknown

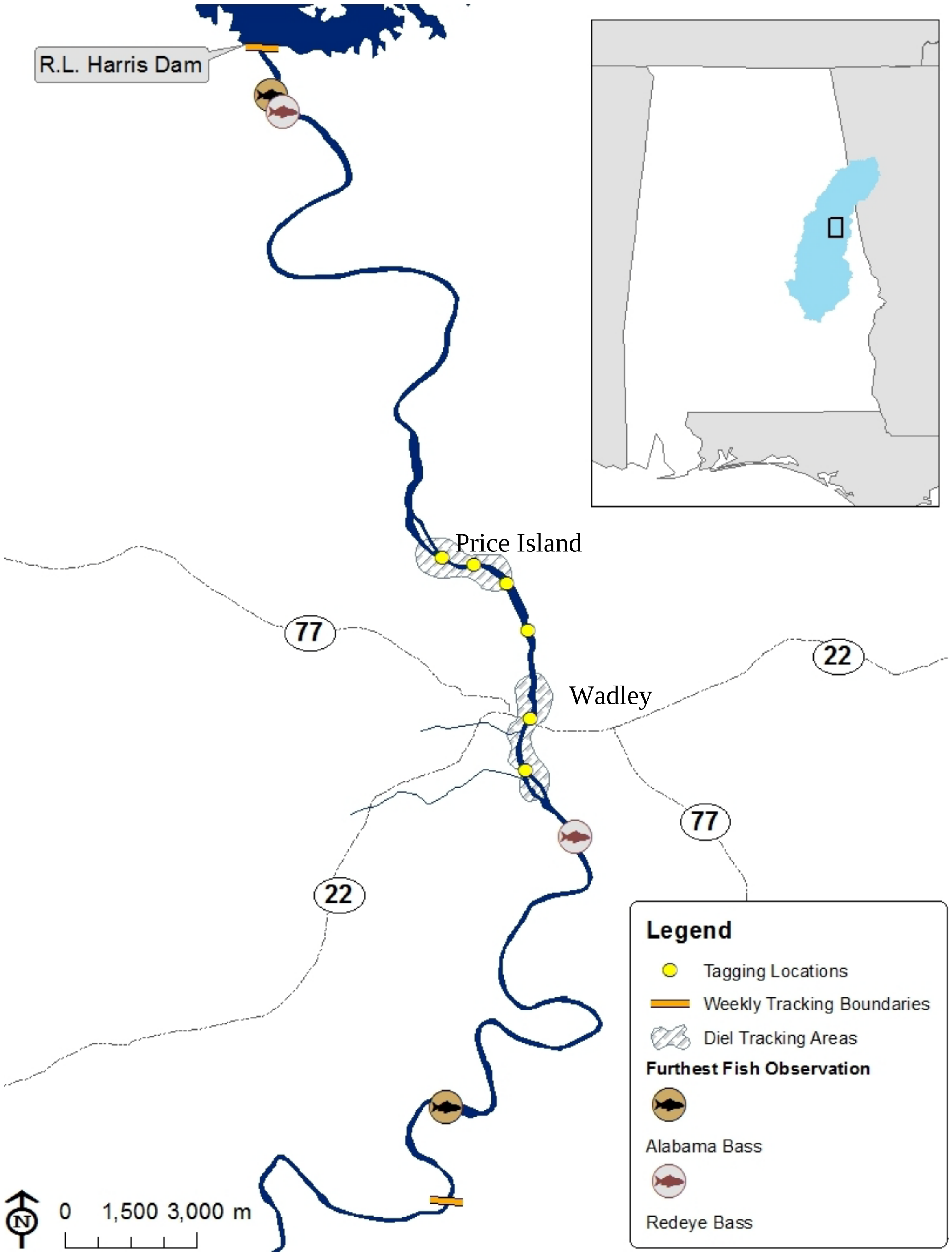

Figure 2-1. Map of the Tallapoosa River and the radio tagging locations, diel survey areas and the boundaries of the tracking surveys. The brown fish represents the furthest upstream and downstream observation for Alabama Bass and the gray represents the furthest upstream observation for Redeye Bass.

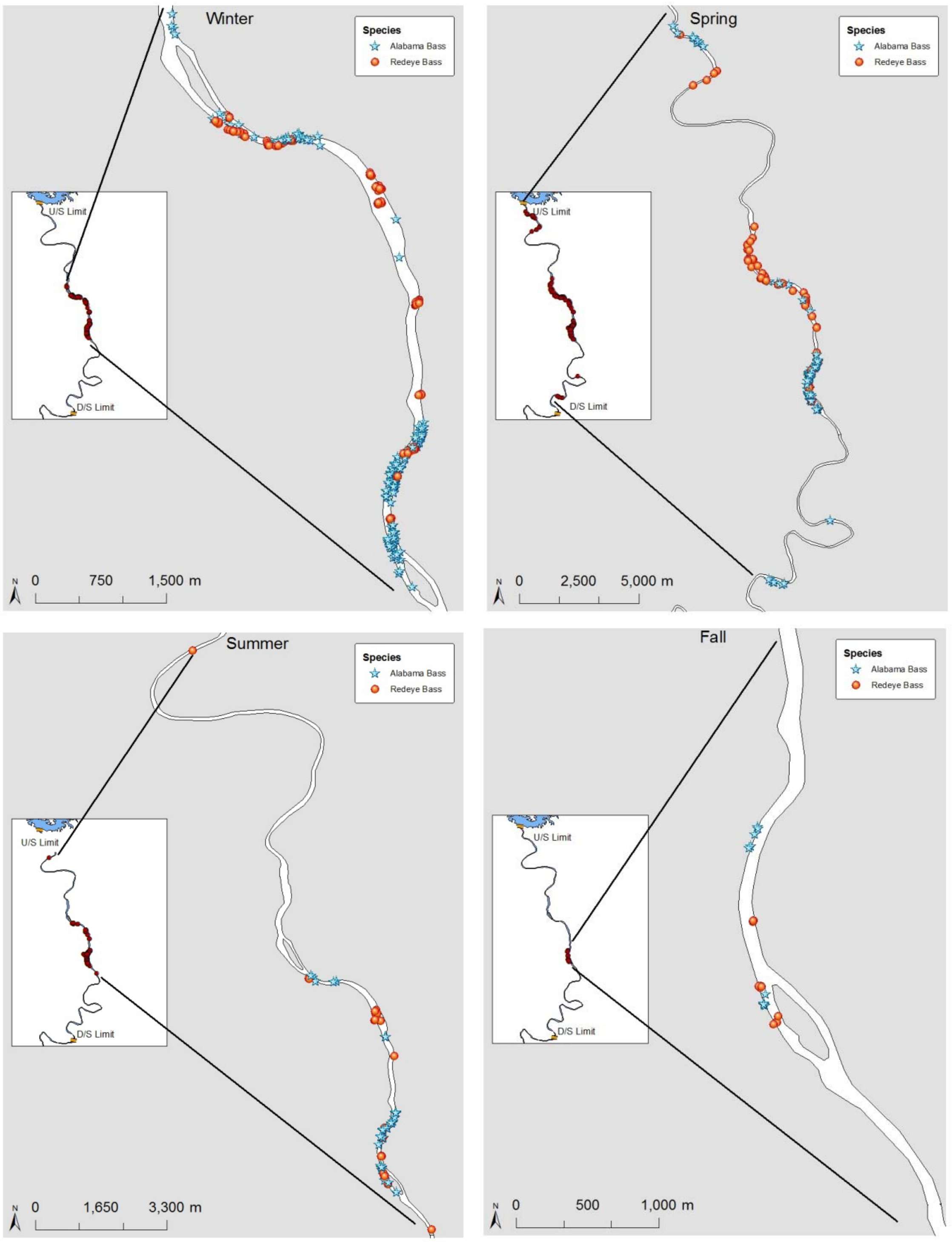

Figure 2-2. Seasonal locations of Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama.

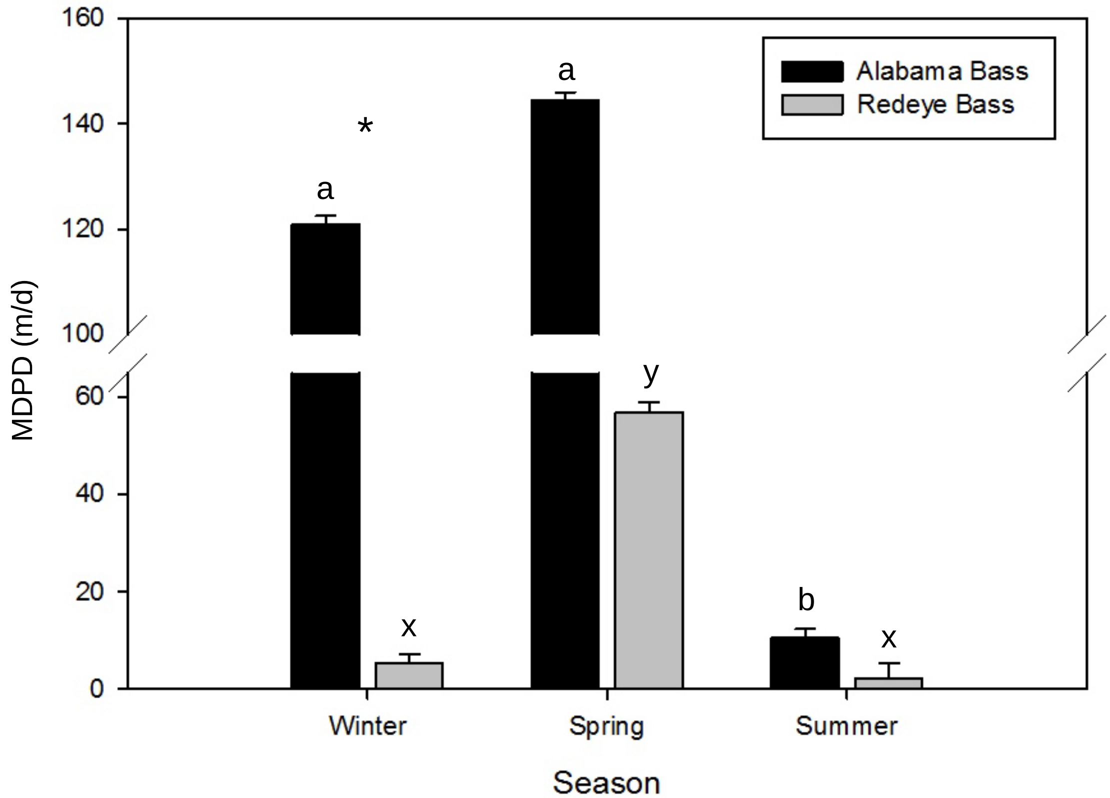

Figure 2-3. Mean daily movement in winter, spring and summer for Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$)

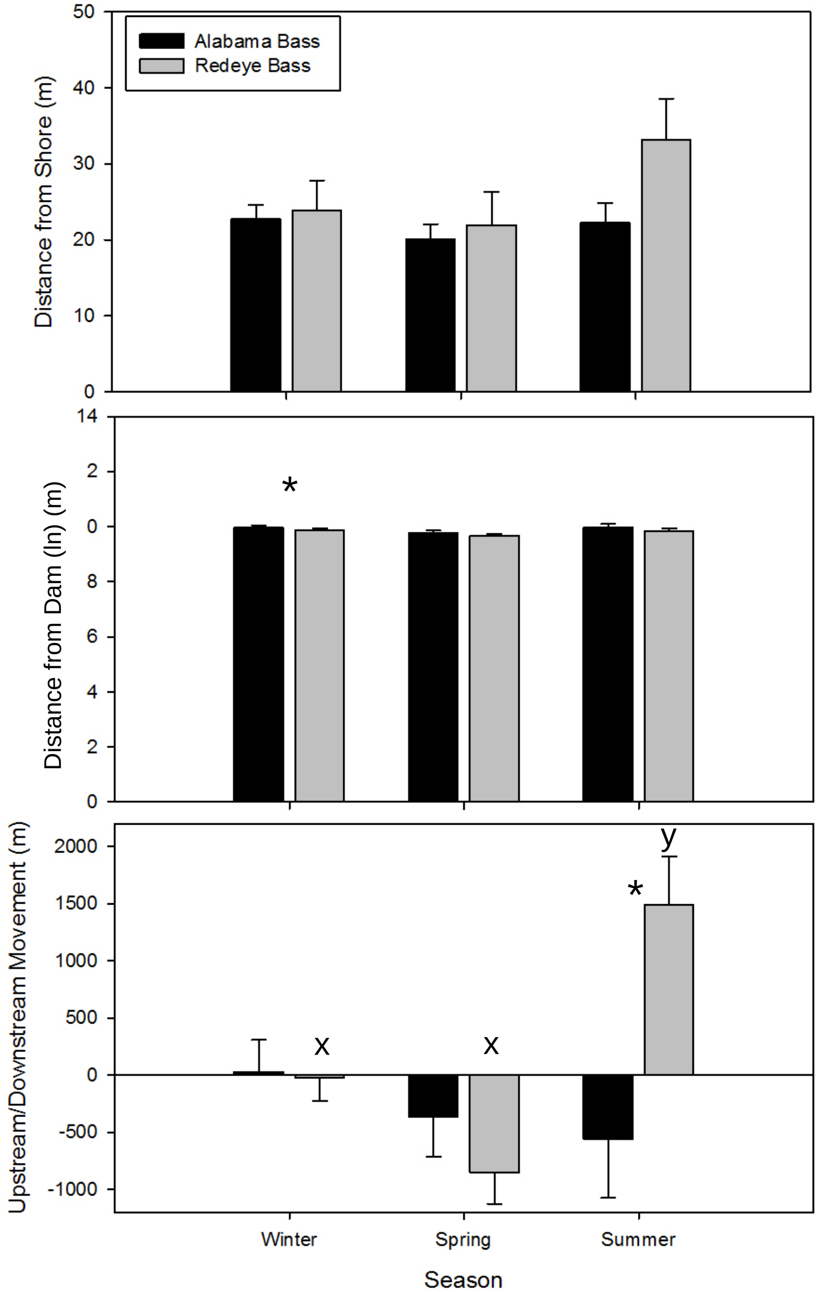

Figure 2-4. Alabama Bass and Redeye Bass distance from shore, distance from dam and upstream/downstream movement in the different seasons. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$) and no notation means there was no significant difference.

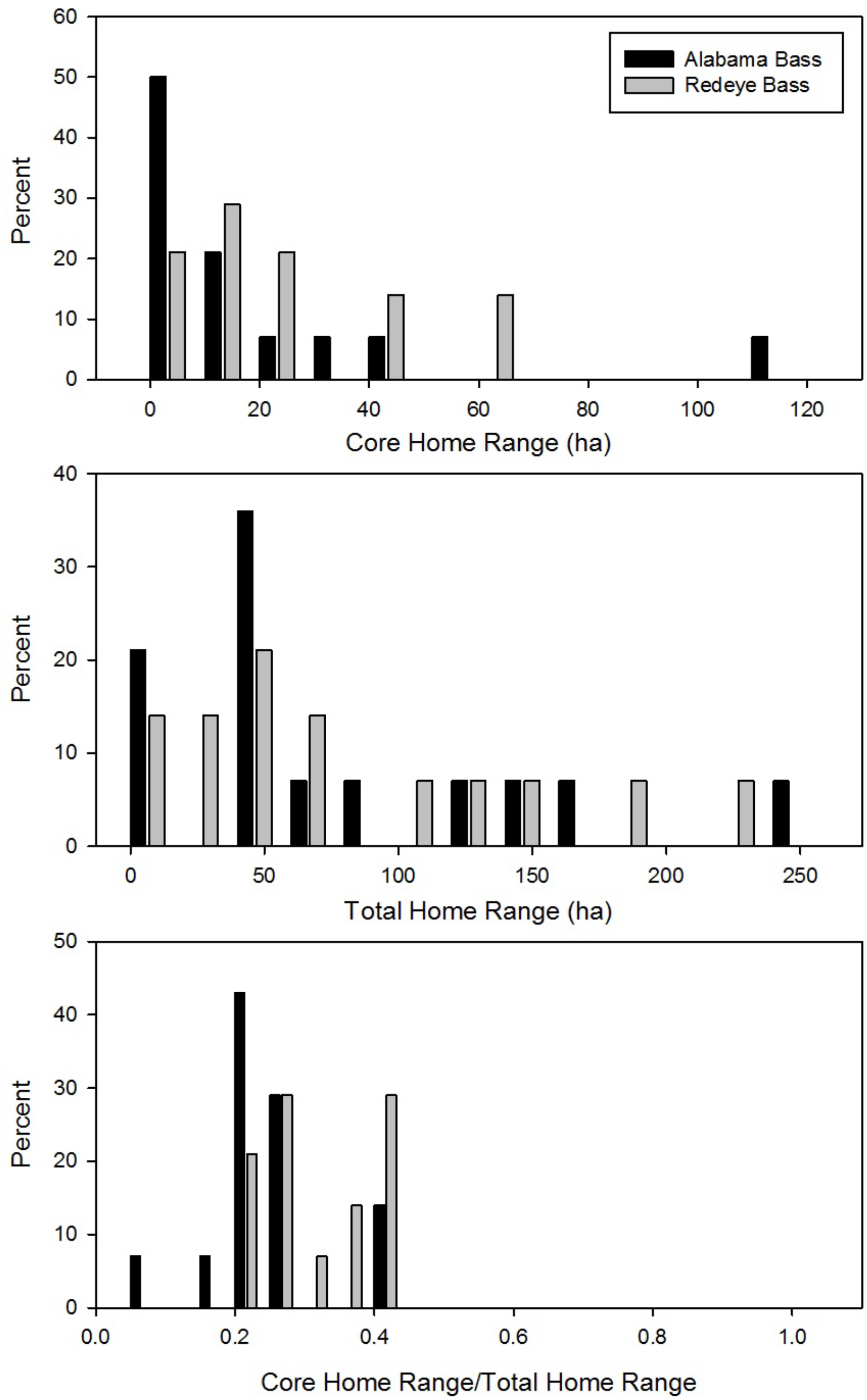

Figure 2-5. The percent of total home range, core home range and the proportion of core area to total home range for Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama.

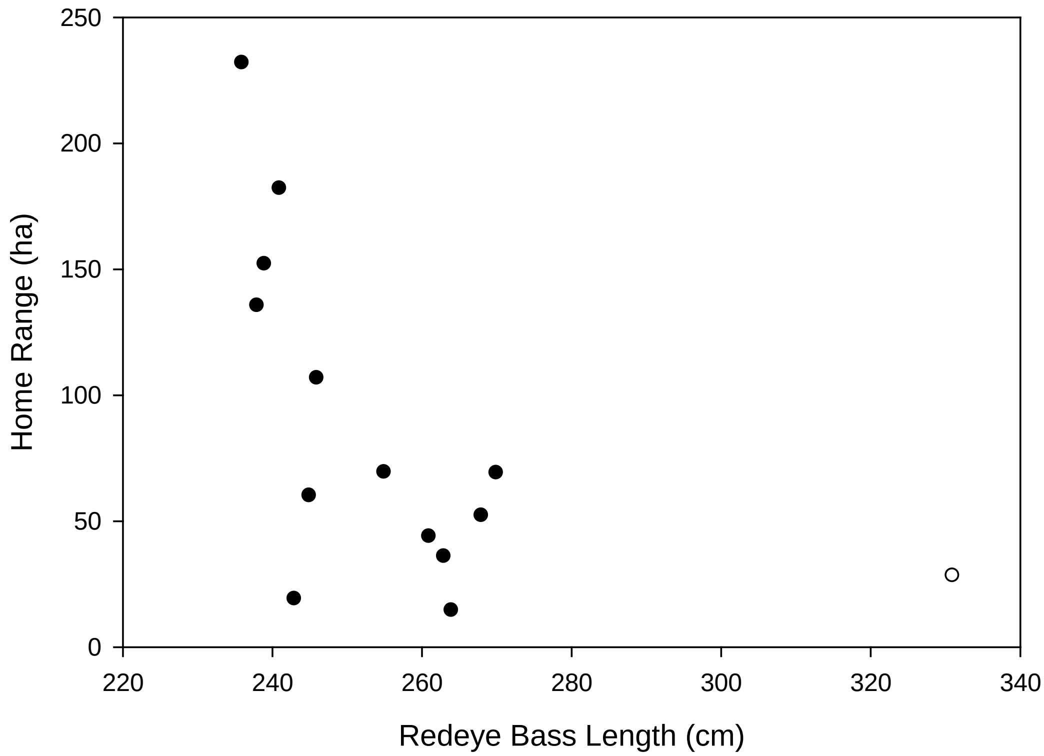

Figure 2-6. Redeye Bass total length (cm) and the total area of the overall home range for each fish.

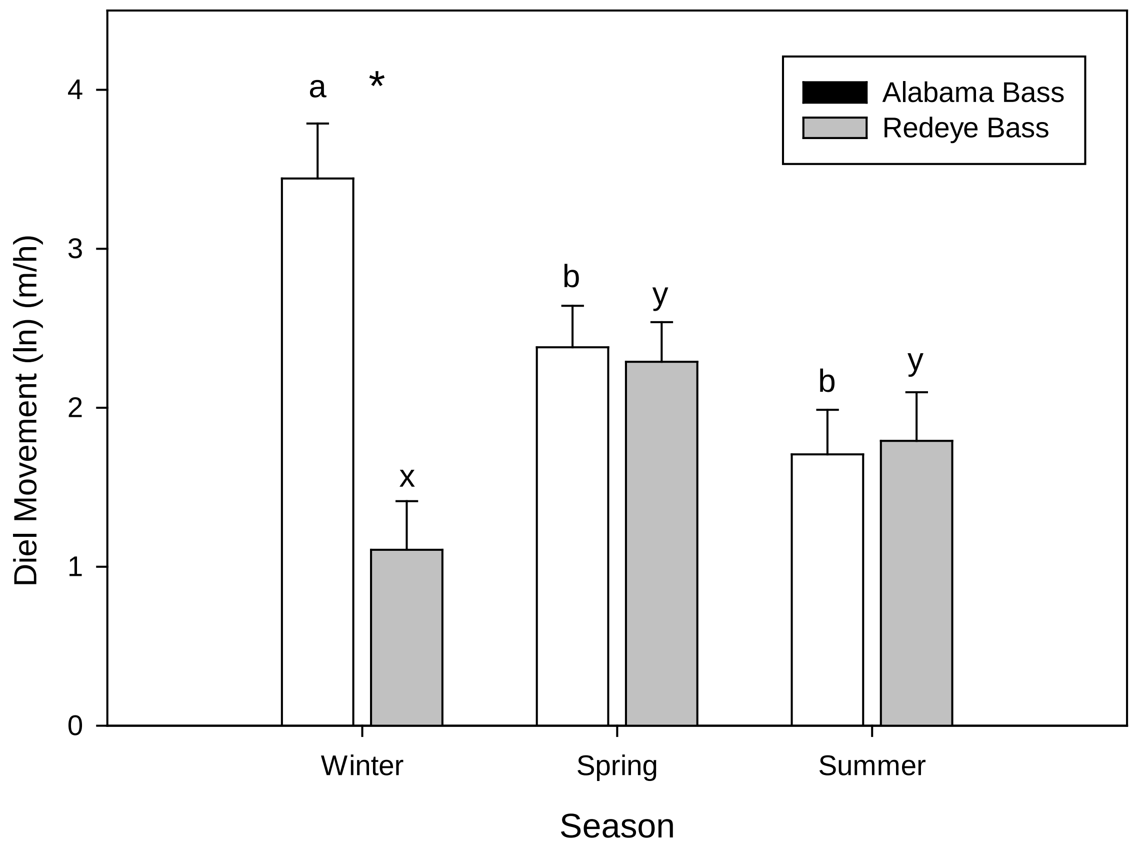

Figure 2-7. Mean diel movement in winter, spring and summer for Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$)

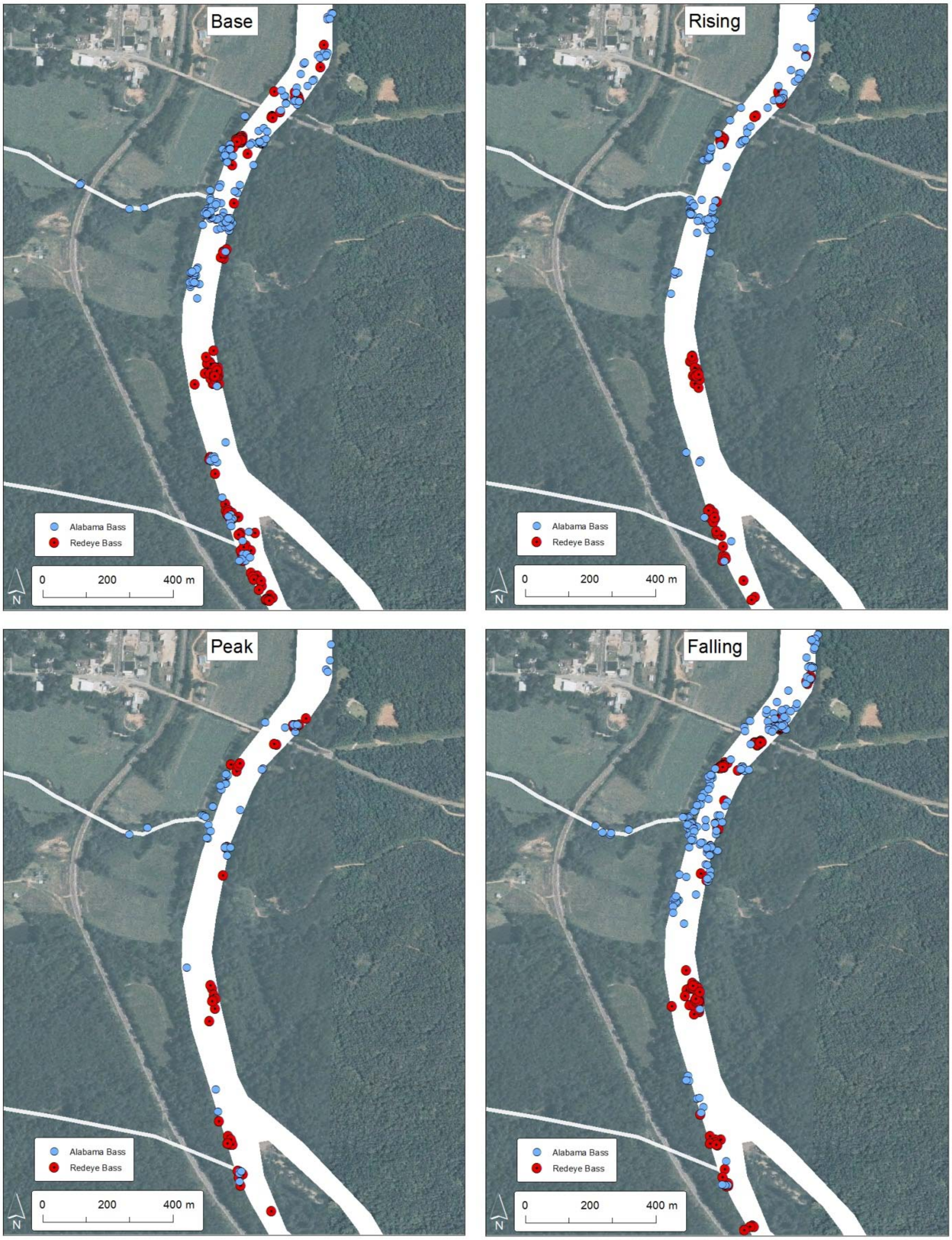

Figure 2-8. Locations of Alabama Bass and Redeye Bass during the four different flow periods (base, rising, peak and falling) in the Tallapoosa River, Alabama.

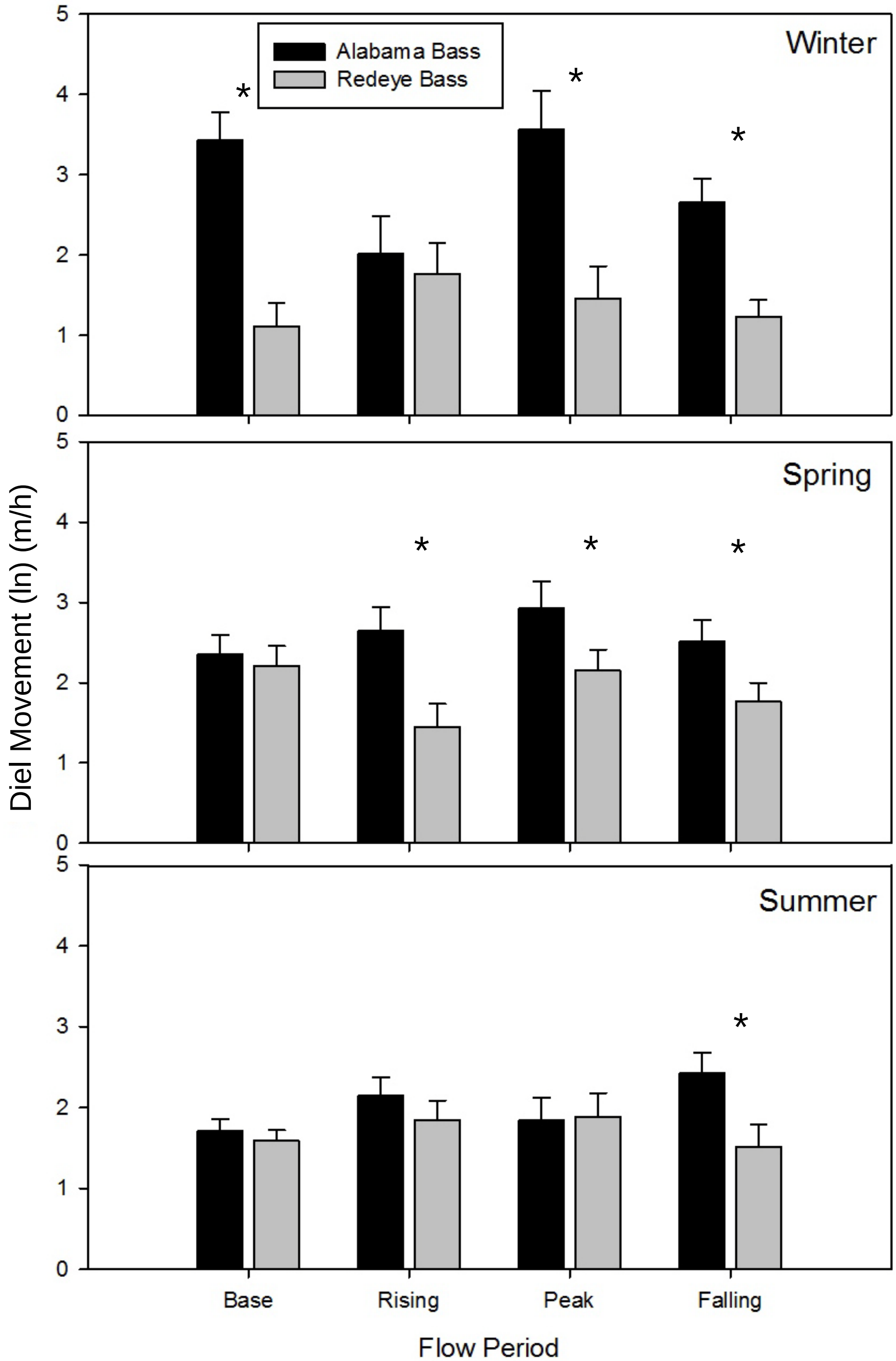

Figure 2-9. Diel movement of Alabama Bass and Redeye Bass during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$) and no notation means there was no significant difference.

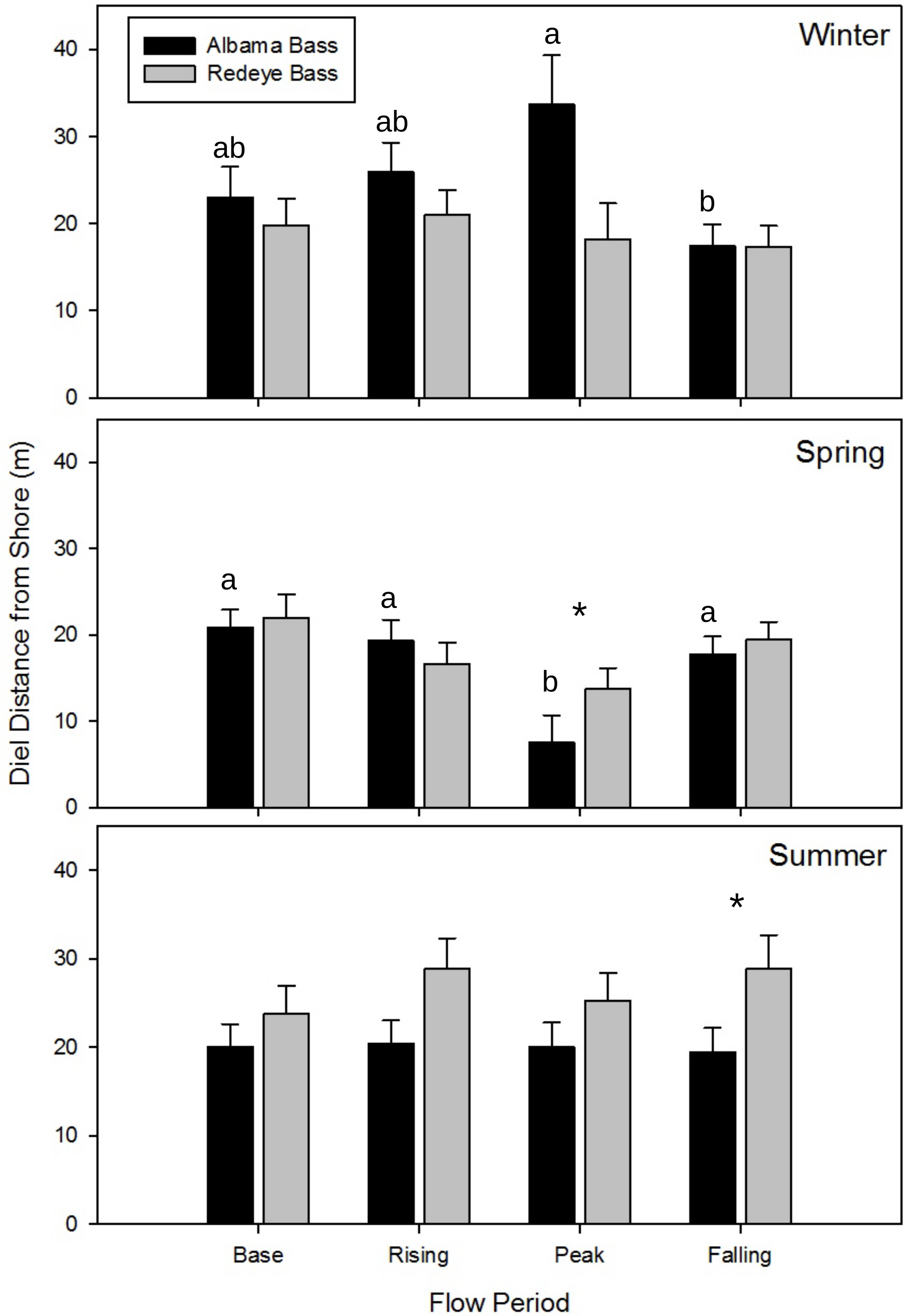

Figure 2-10. Diel distance from shore for Alabama Bass and Redeye Bass during the four different flow periods. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$) and no notation means there was no significant difference.

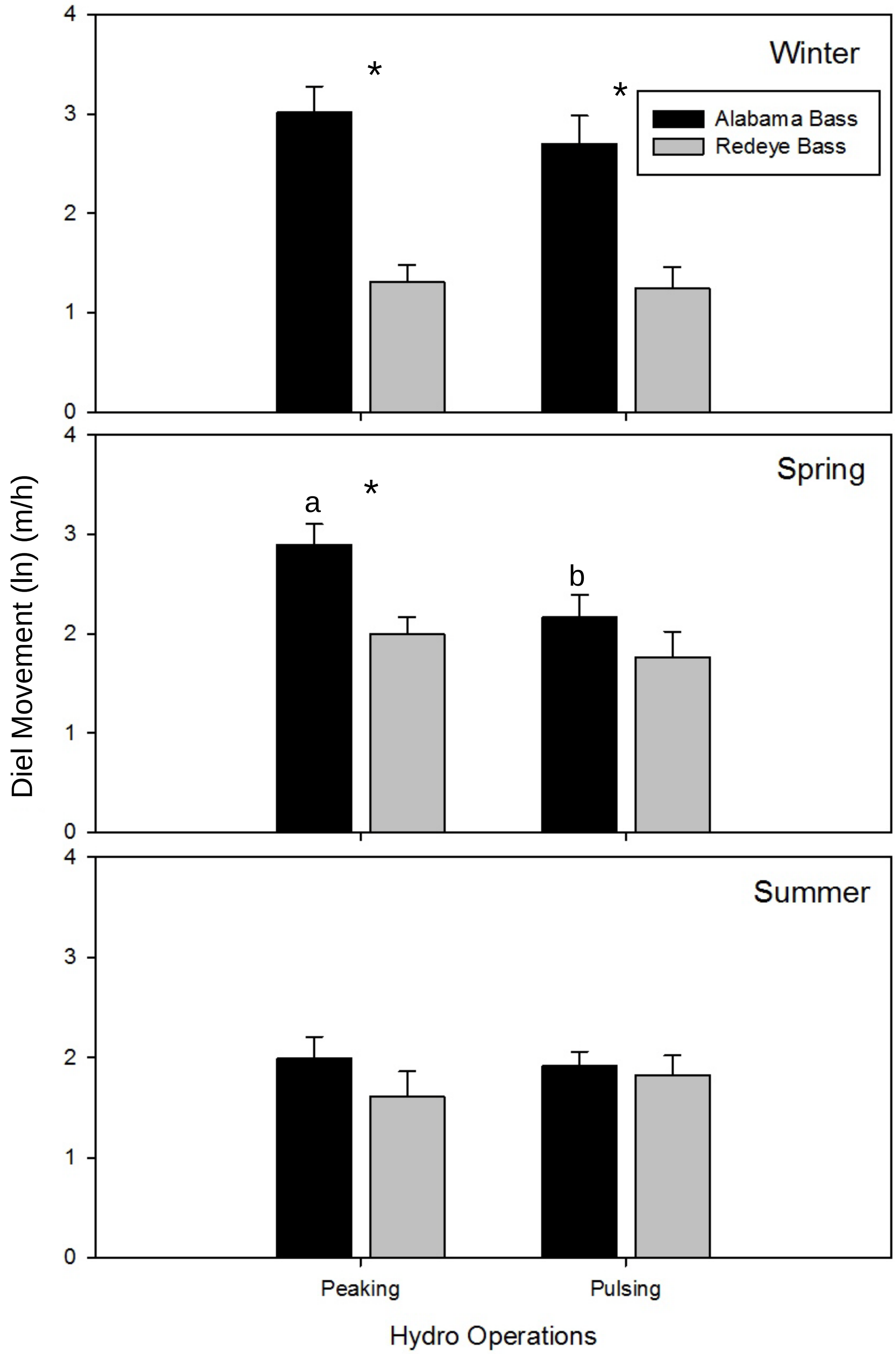

Figure 2-11. Diel movement of Alabama Bass and Redeye Bass during the different dam operations in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$) and no notation means there was no significant difference.

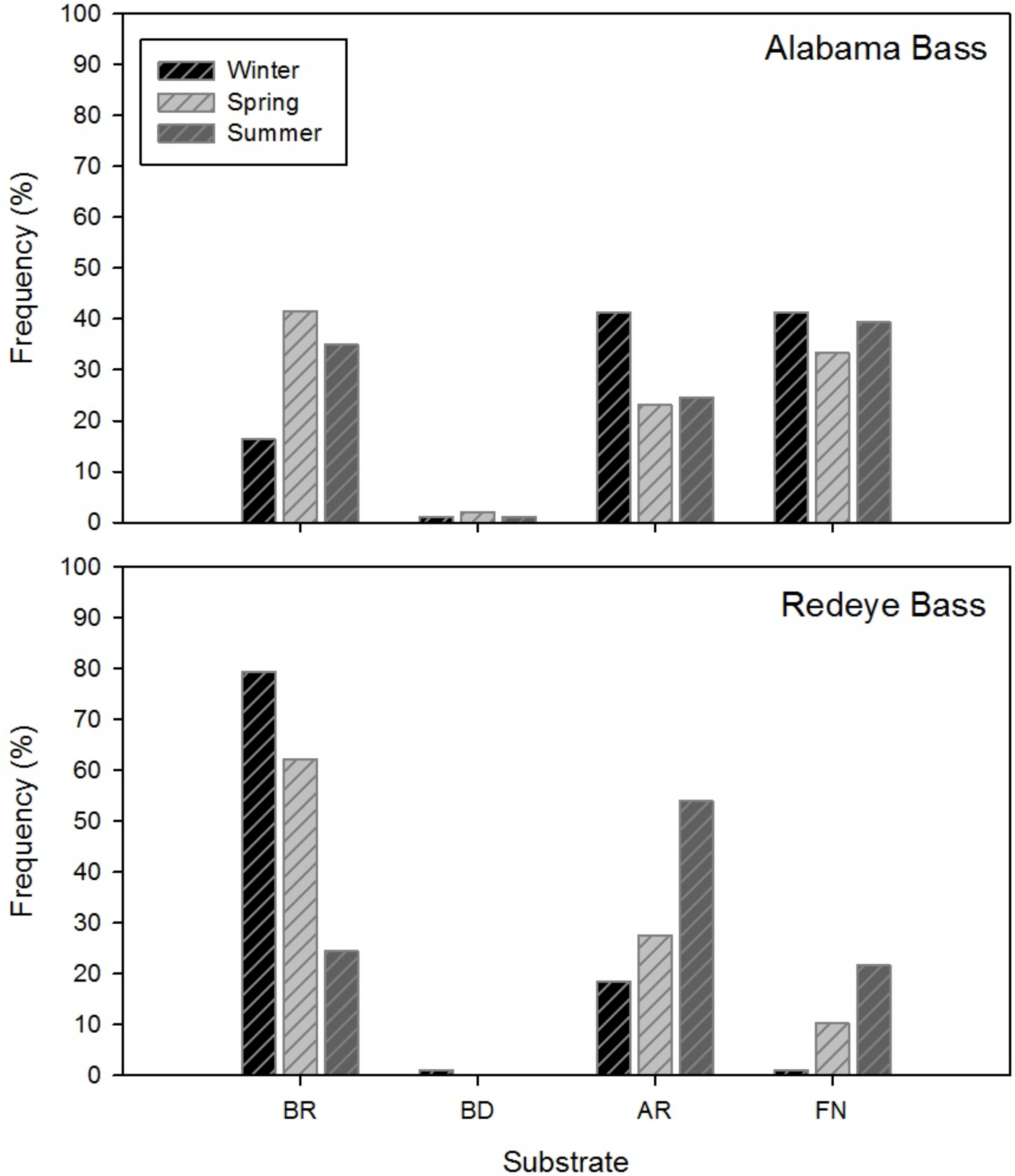

Figure 2-12. Seasonal substrate used by Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama.

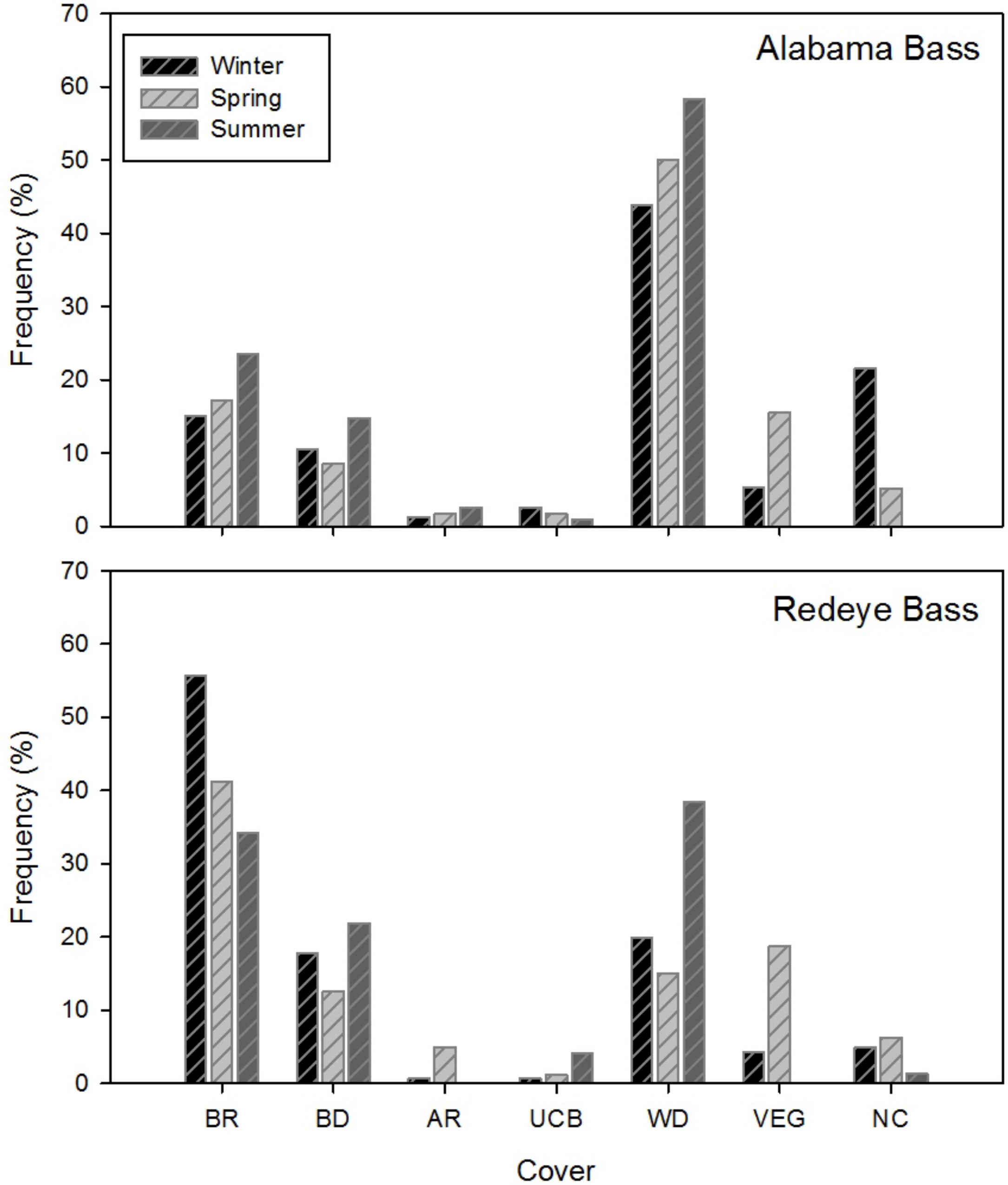

Figure 2-13. Seasonal cover used by Alabama Bass and Redeye Bass in the Tallapoosa River, Alabama.

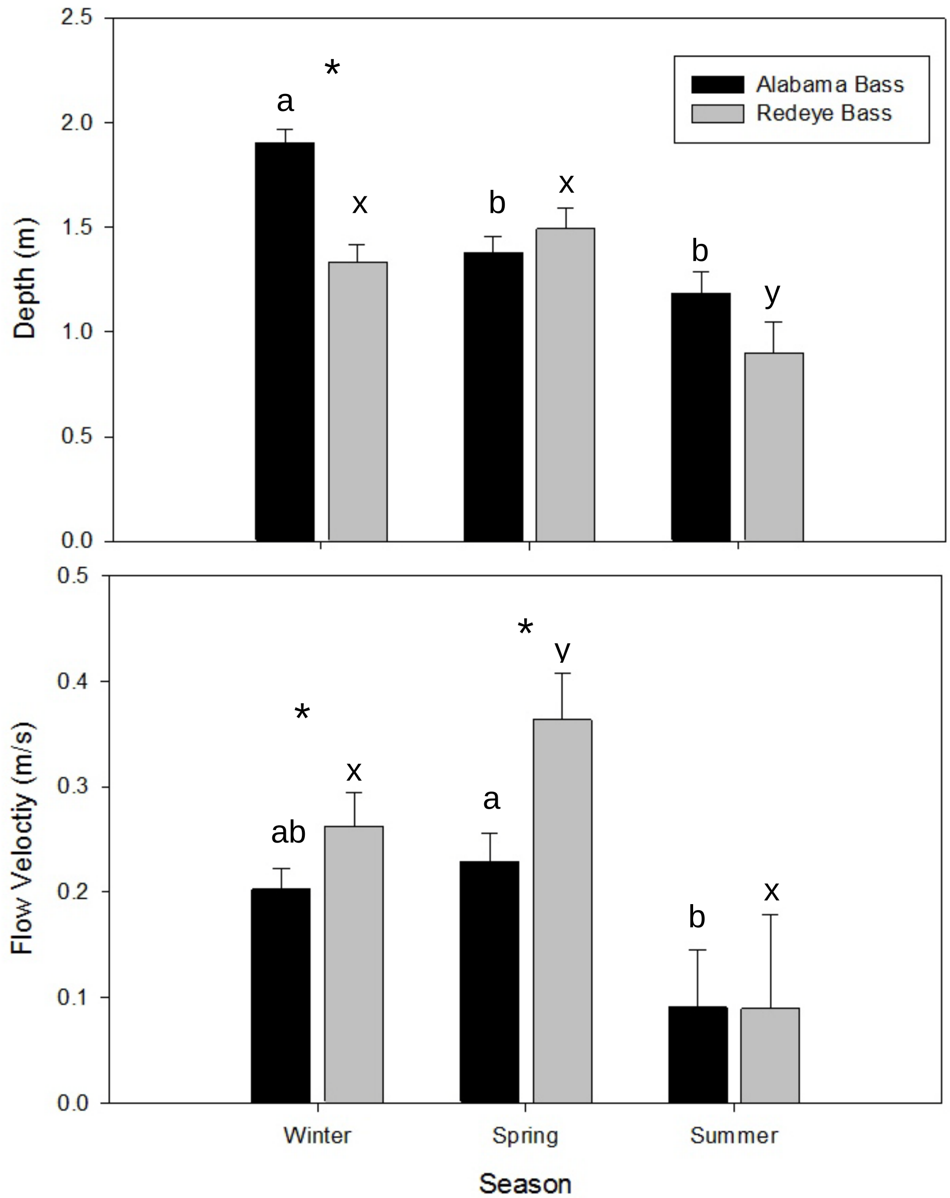

Figure 2-14. Seasonal depth and velocities at the locations where Alabama Bass and Redeye Bass were observed in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) within the species and the asterisk represents a difference between the species ($p < 0.10$) and no notation means there was no significant difference.

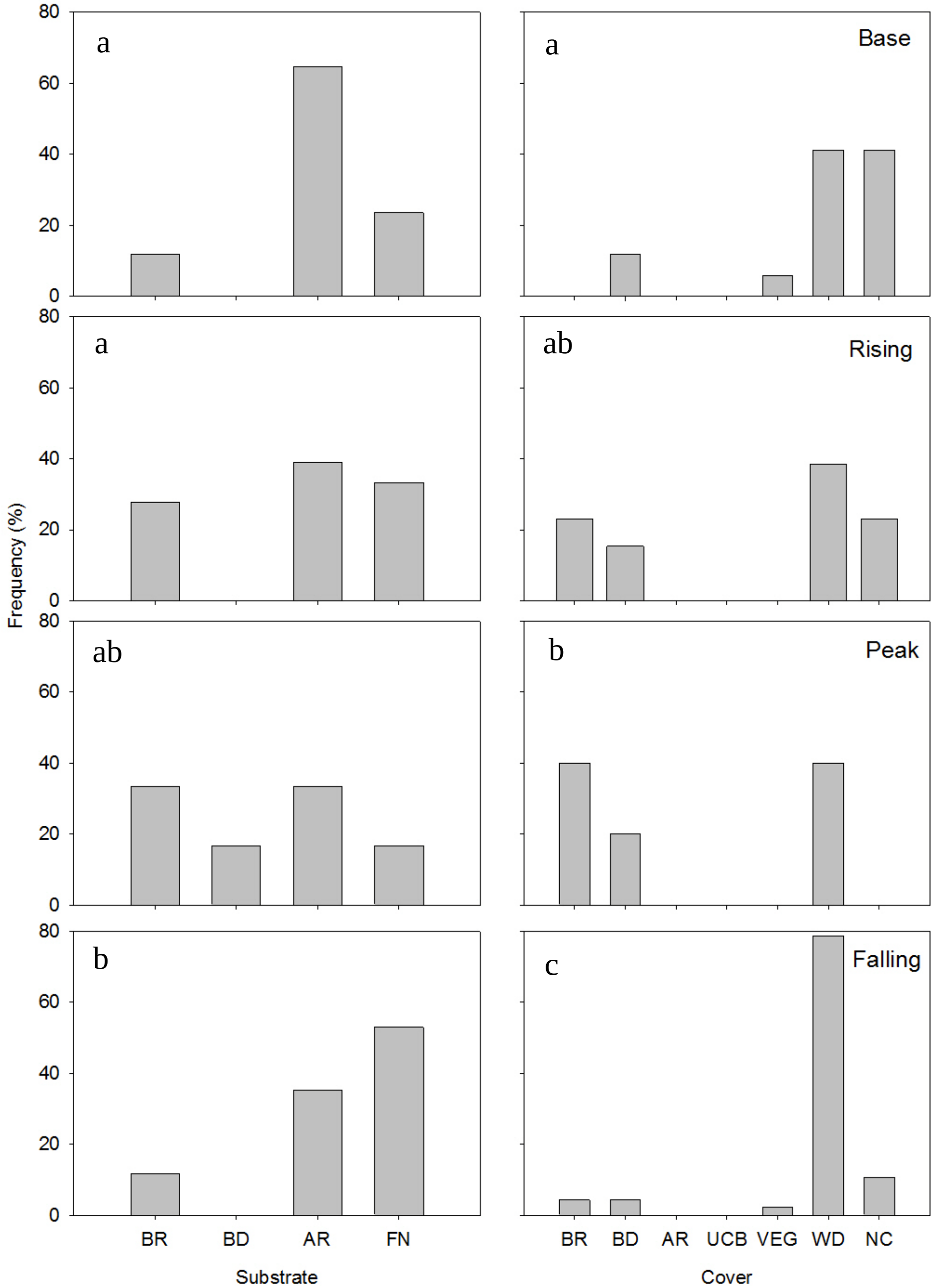

Figure 2-15. Substrate and cover used by Alabama Bass in winter 2010-2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference.

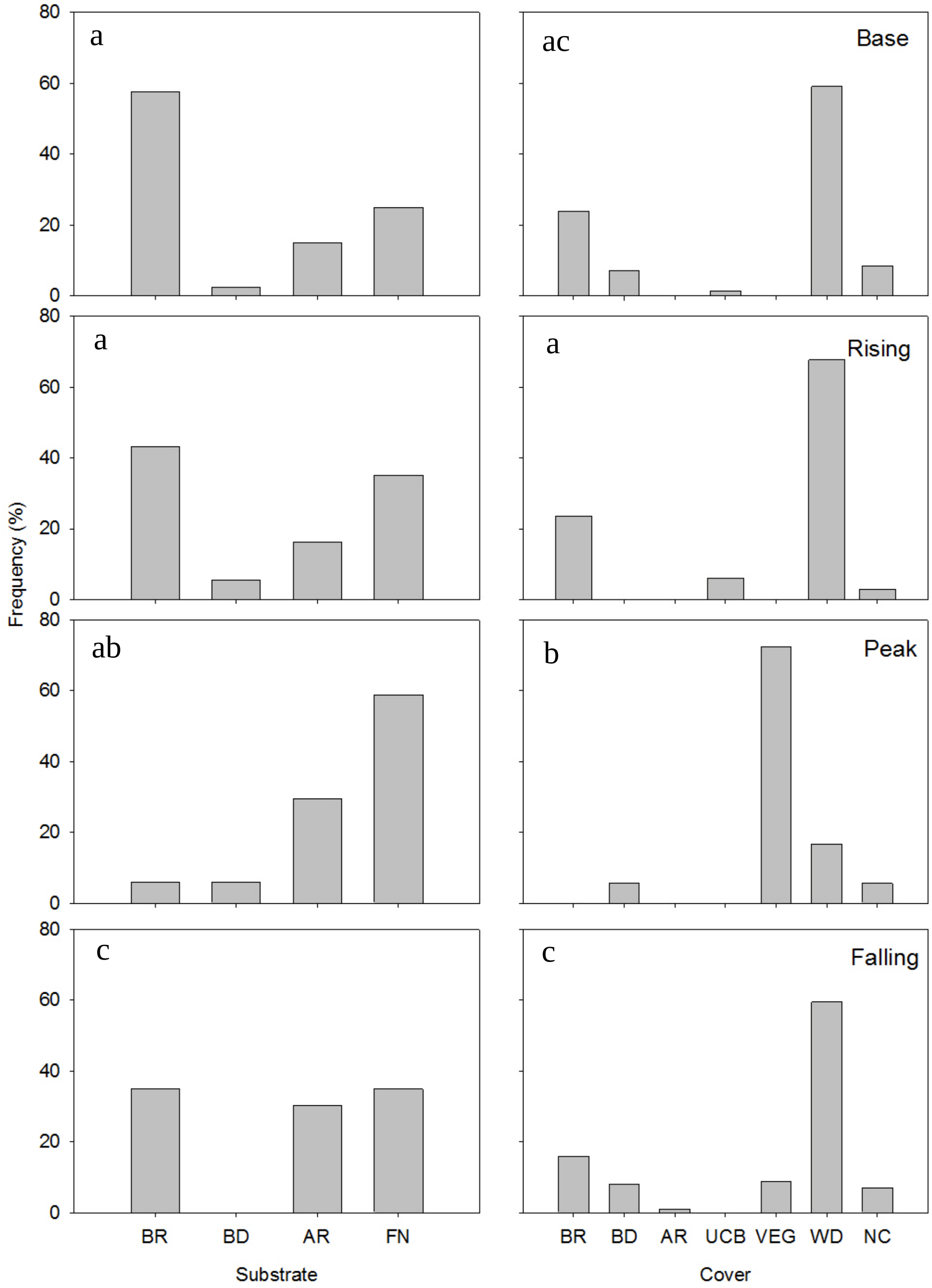

Figure 2-16. Substrate and cover used by Alabama Bass in spring 2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference

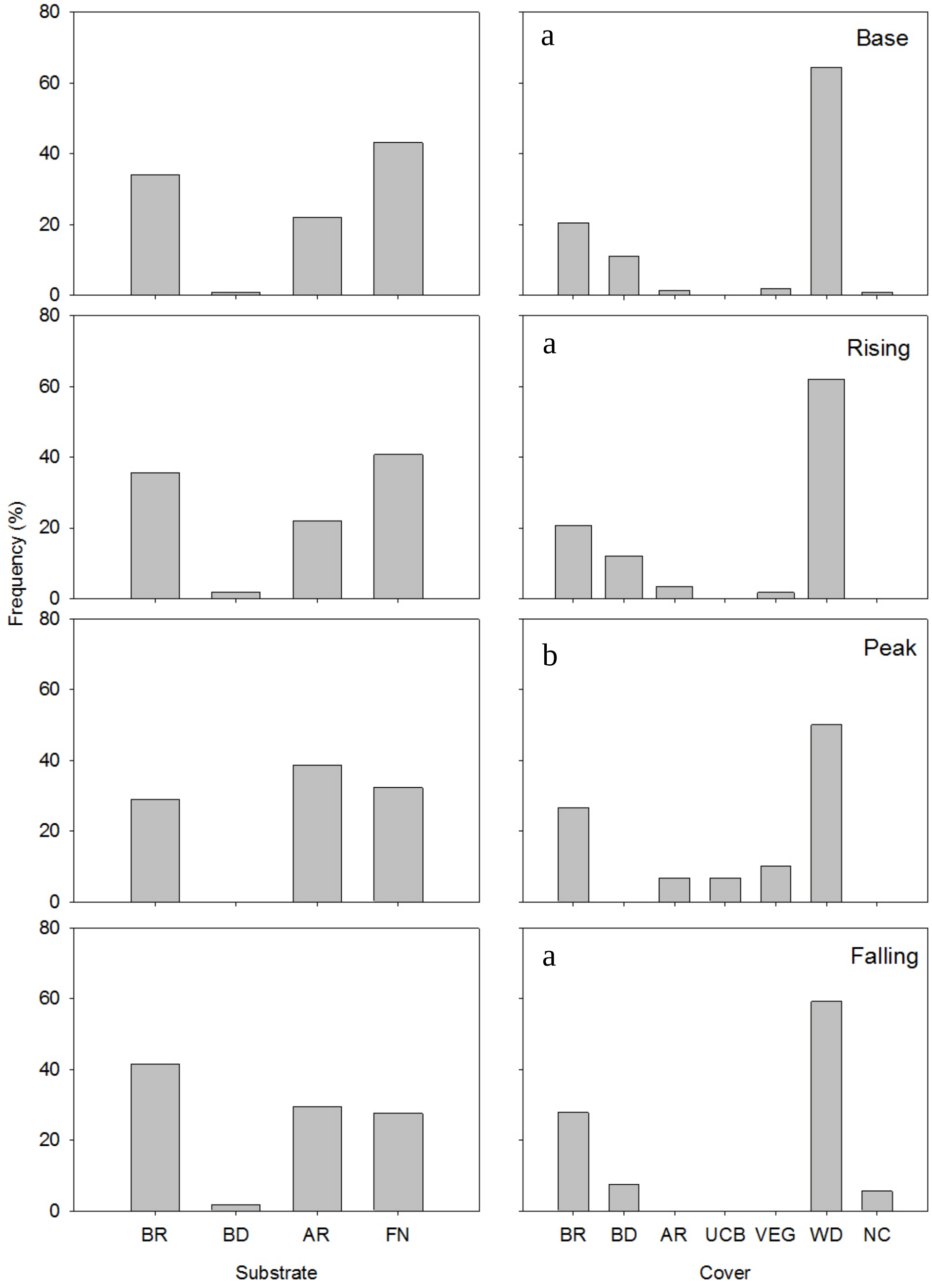

Figure 2-17. Substrate and cover used by Alabama Bass in summer 2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference.

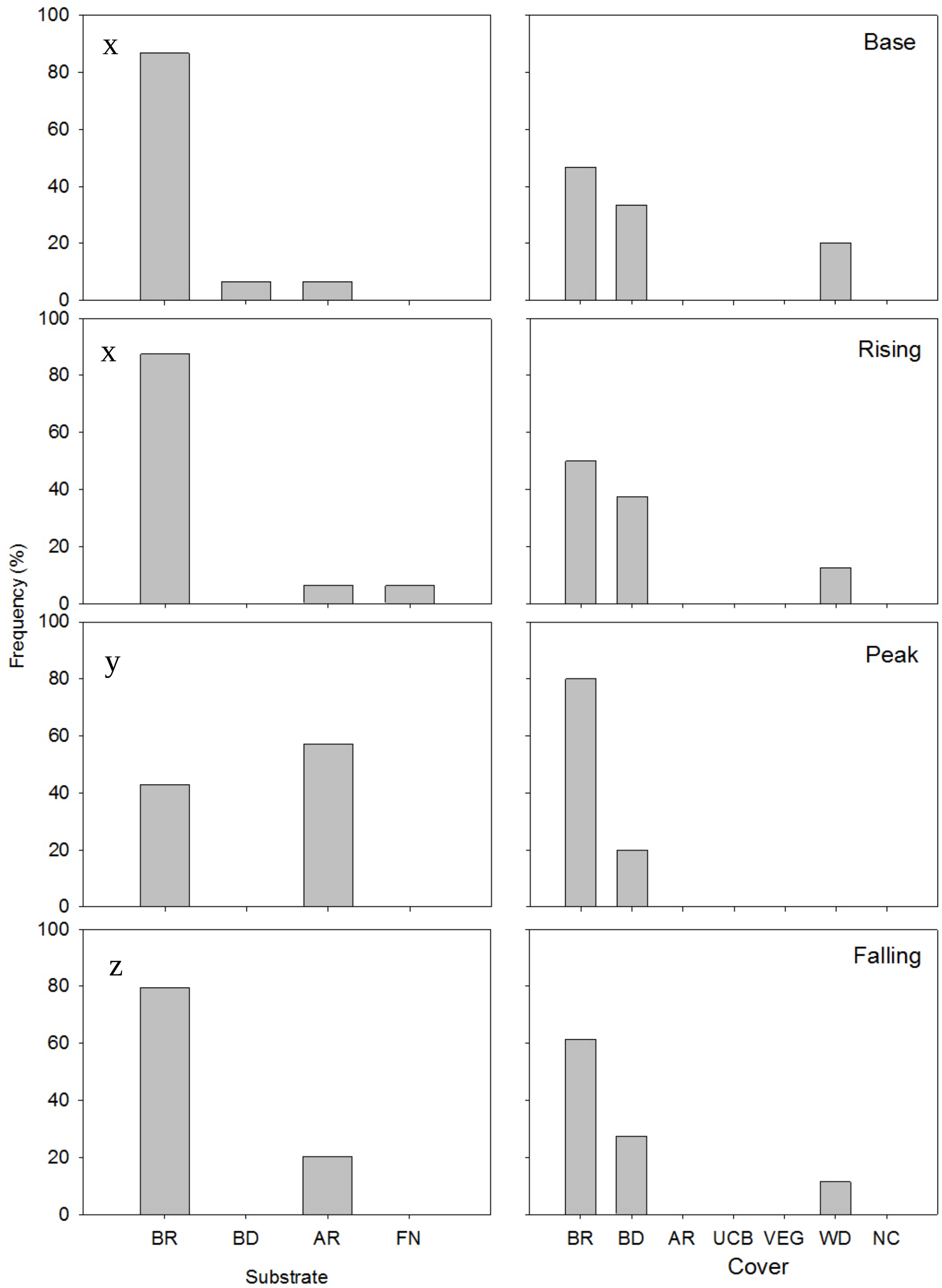

Figure 2-18. Substrate and cover used by Redeye Bass in winter 2010-2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference.

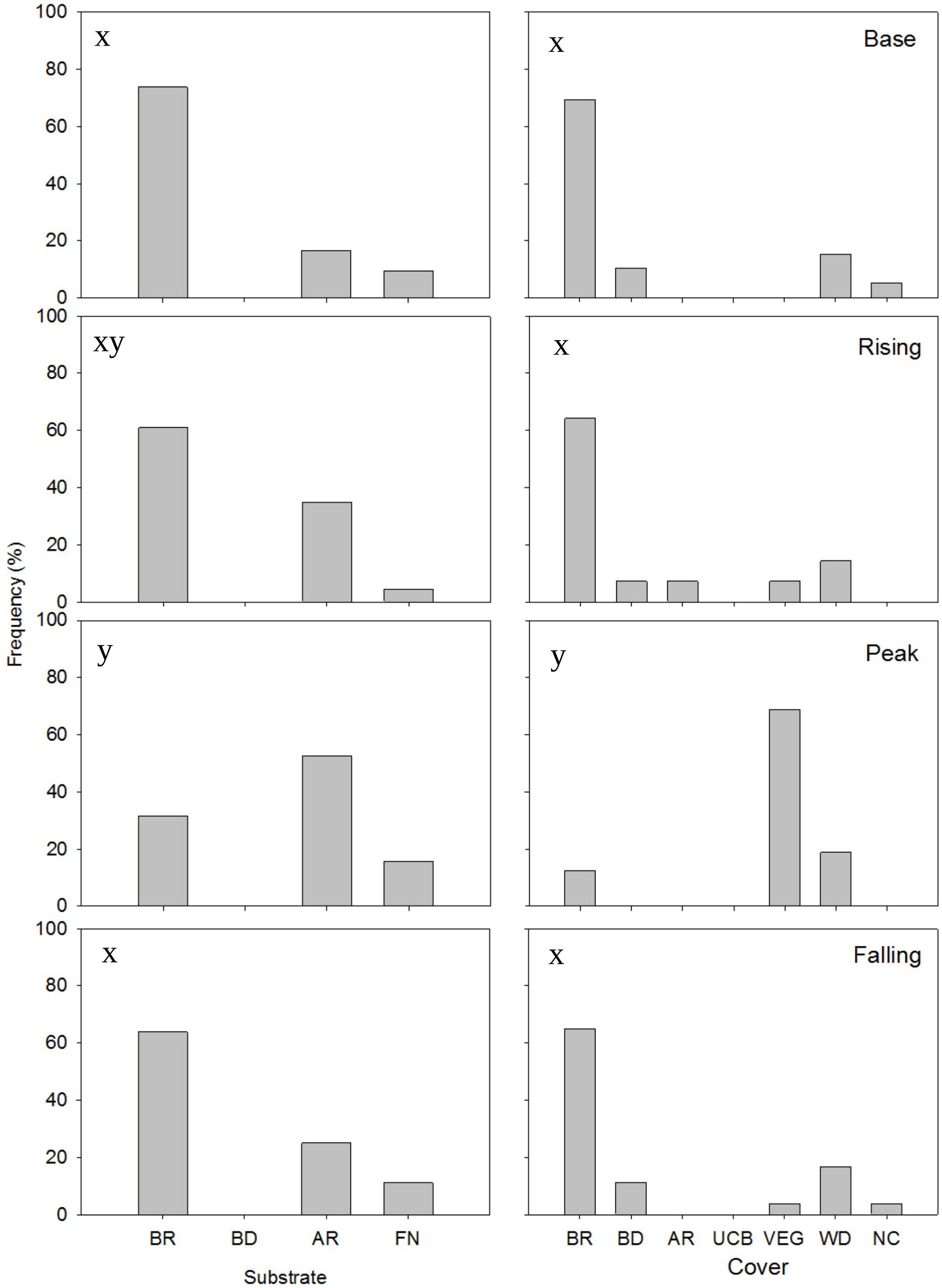

Figure 2-19. Substrate and cover used by Redeye Bass in spring 2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference.

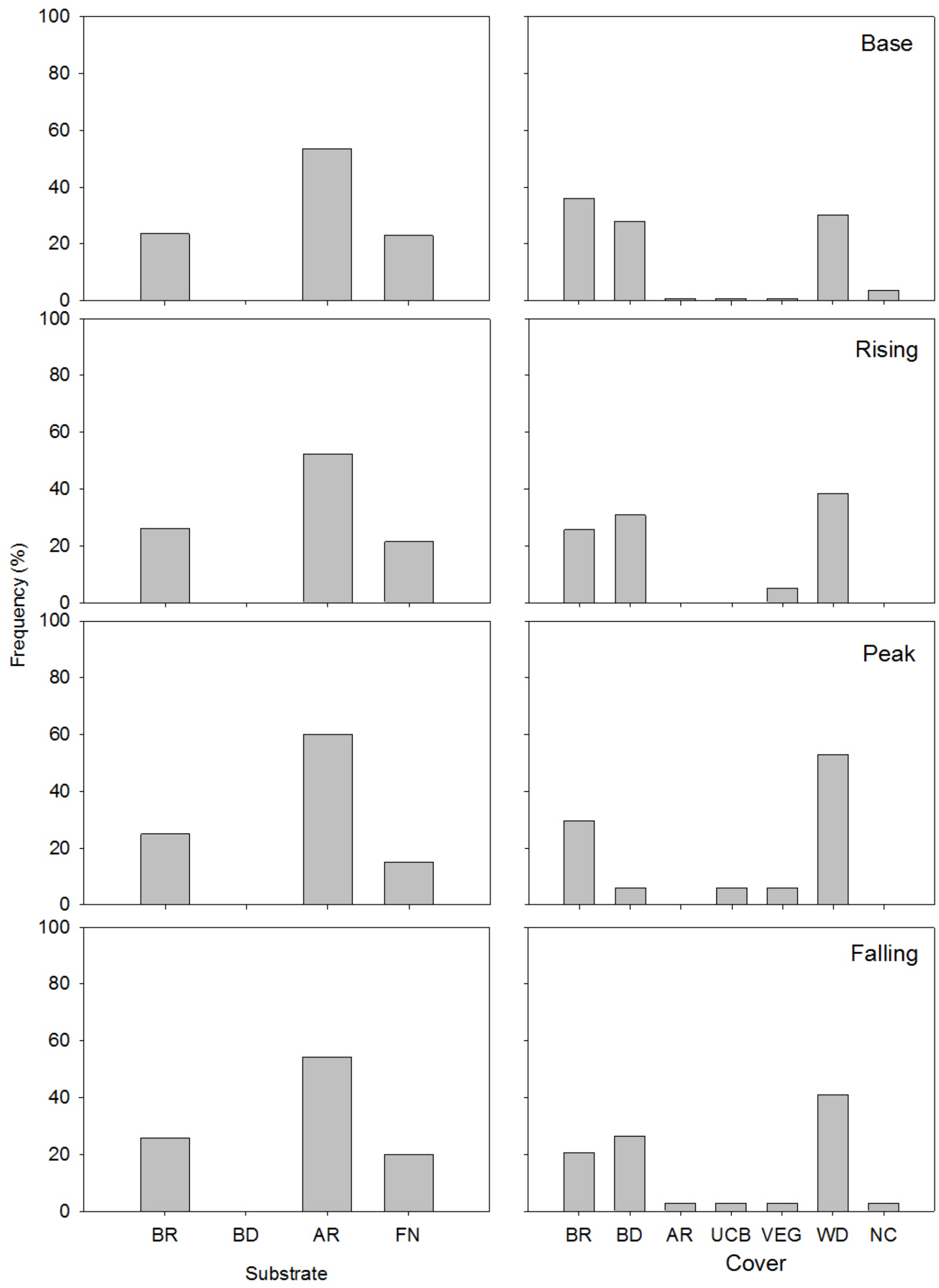

Figure 2-20. Substrate and cover used by Redeye Bass in summer 2010-2011 during the different flow periods in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.10$) between flow periods for substrate and cover and no notation means there was no significant difference.

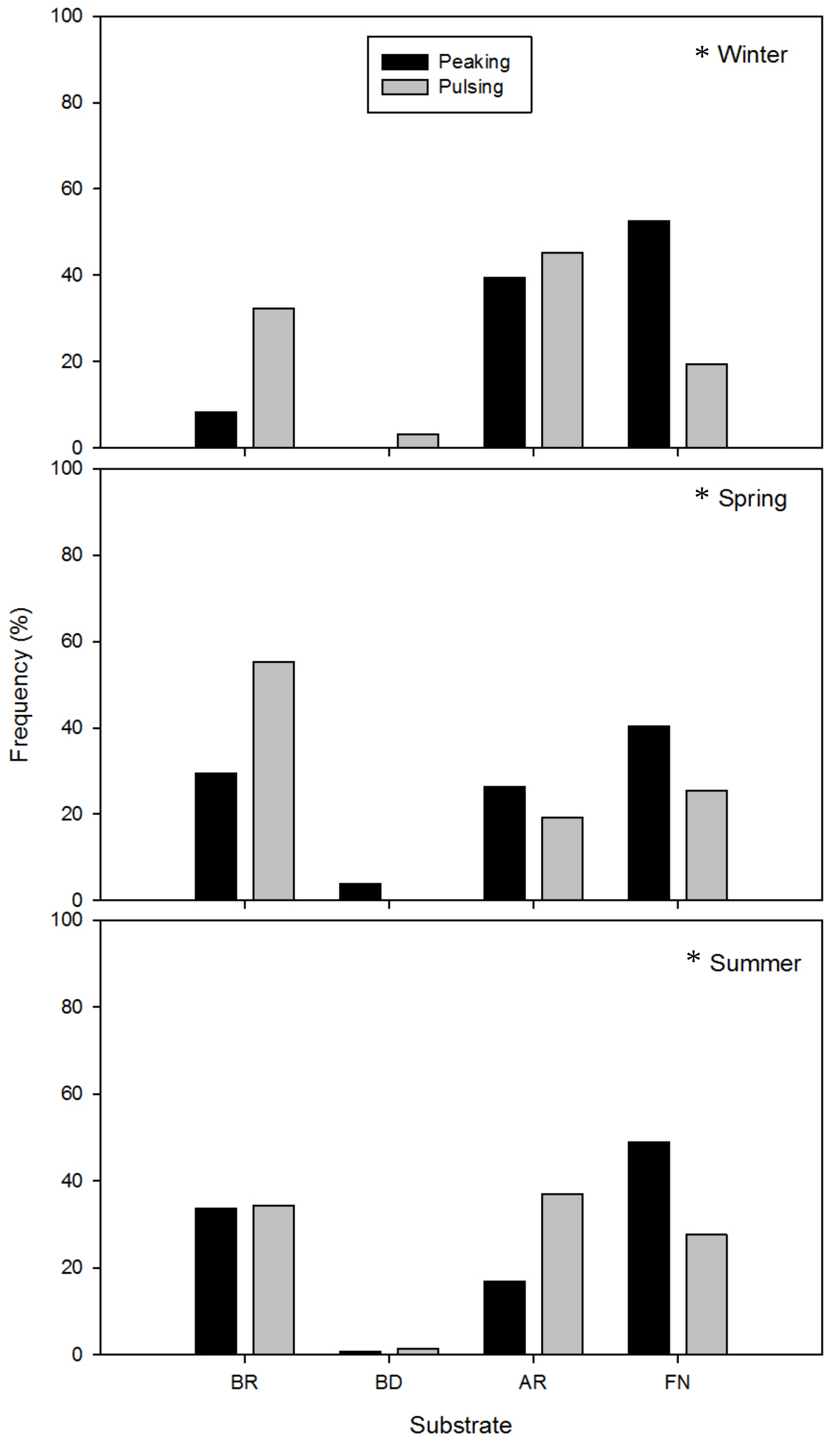

Figure 2-21. Substrate used by Alabama Bass during different dam operations in the Tallapoosa River, Alabama in all the seasons. An asterisk in the graph represents that substrate used during that season was different between the dam operations.

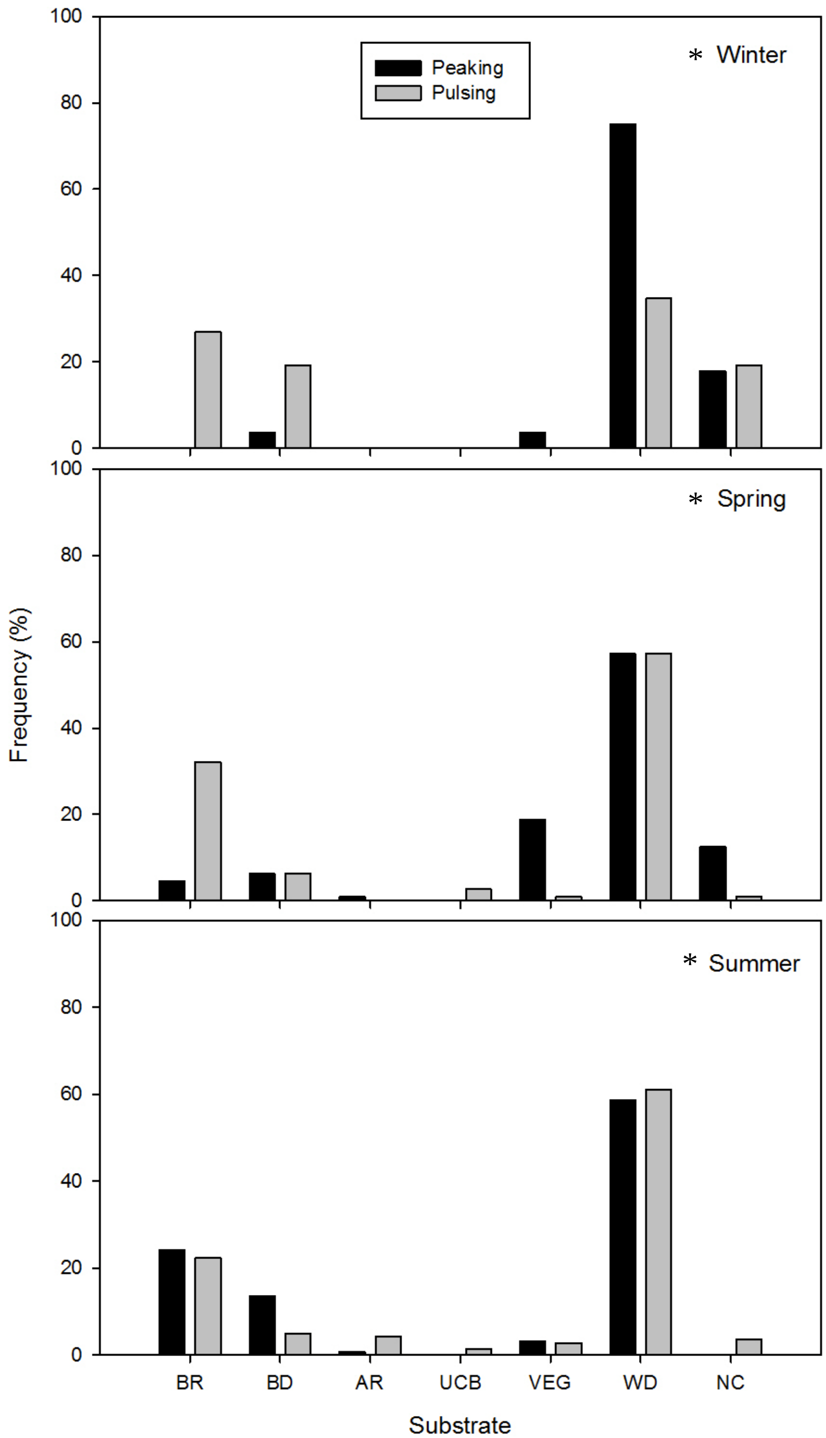

Figure 2-22. Cover used by Alabama Bass during different dam operations in the Tallapoosa River, Alabama in all the seasons. An asterisk in the graph represents that cover used during that season was different between the dam operations.

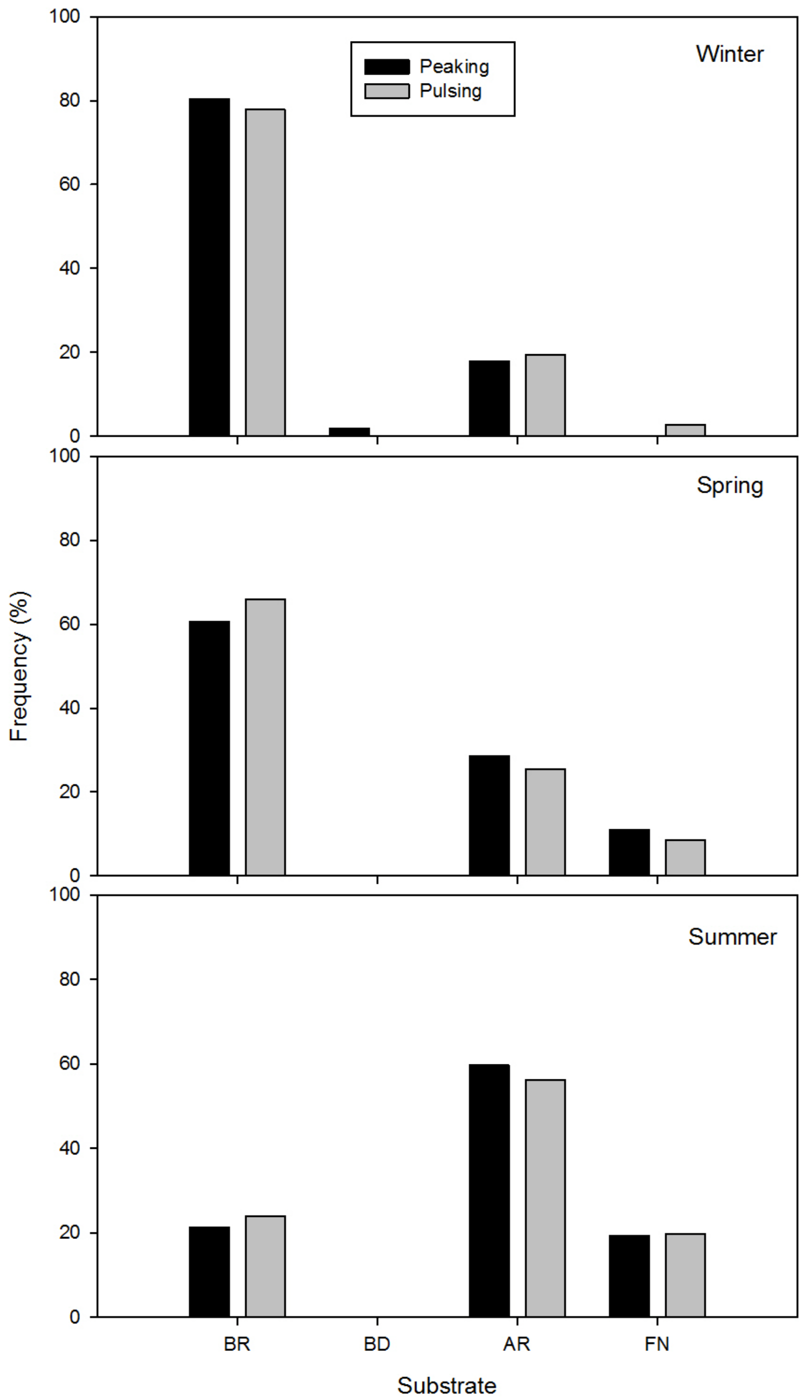

Figure 2-23. Substrate used by Redeye Bass during different dam operations in the Tallapoosa River, Alabama in all the seasons. An asterisk in the graph represents that substrate used during that season was different between the dam operations.

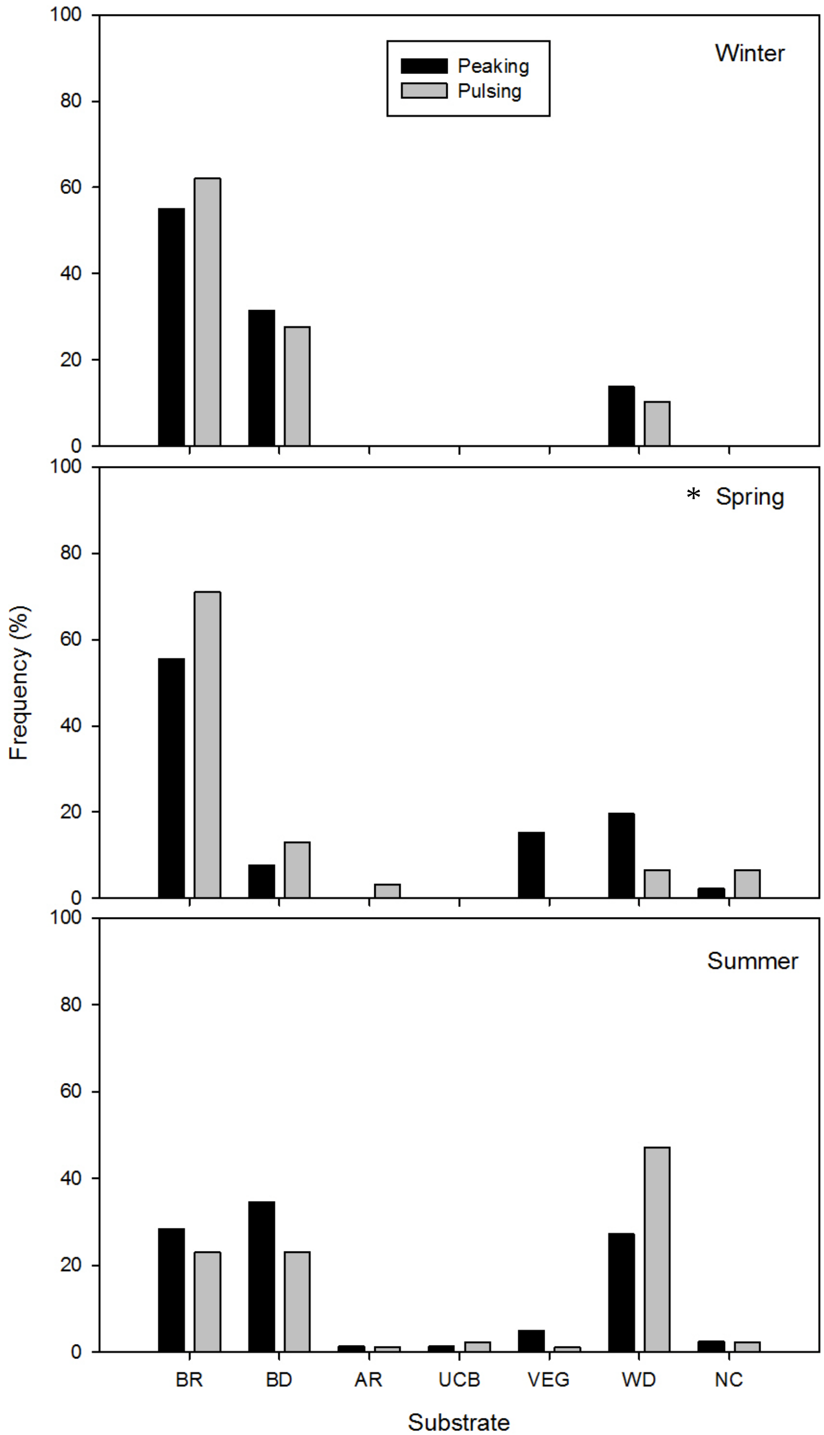

Figure 2-24. Cover used by Redeye Bass during different dam operations in the Tallapoosa River, Alabama in all the seasons. An asterisk in the graph represents that cover used during that season was different between the dam operations.

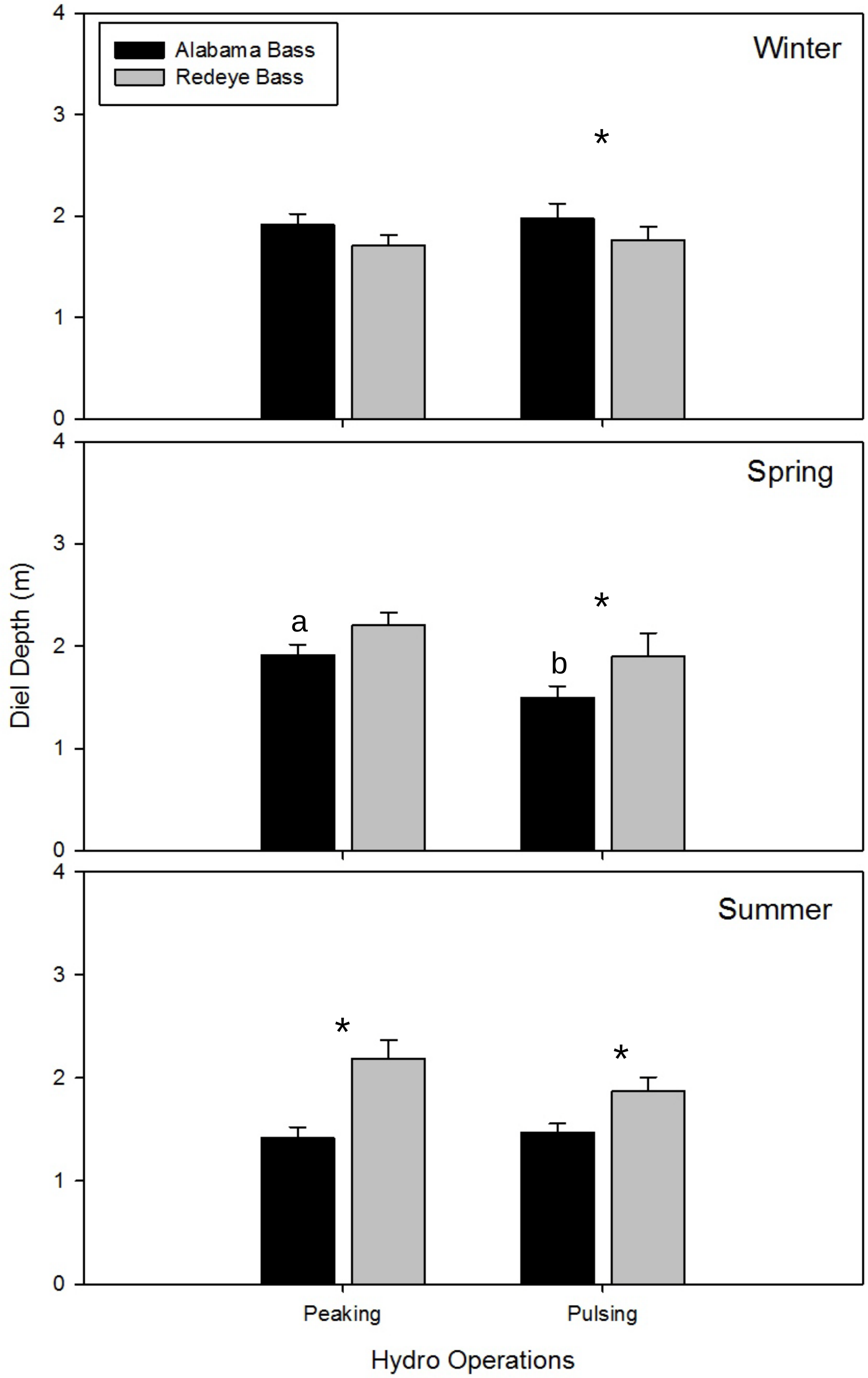

Figure 2-25. Water depth at the locations where Alabama Bass and Redeye Bass were observed during the different dam operations. Different letters represent a significant difference ($p < 0.05$) within the species and the asterisk represents a difference between the species ($p < 0.05$) and no notation means there was no significant difference.

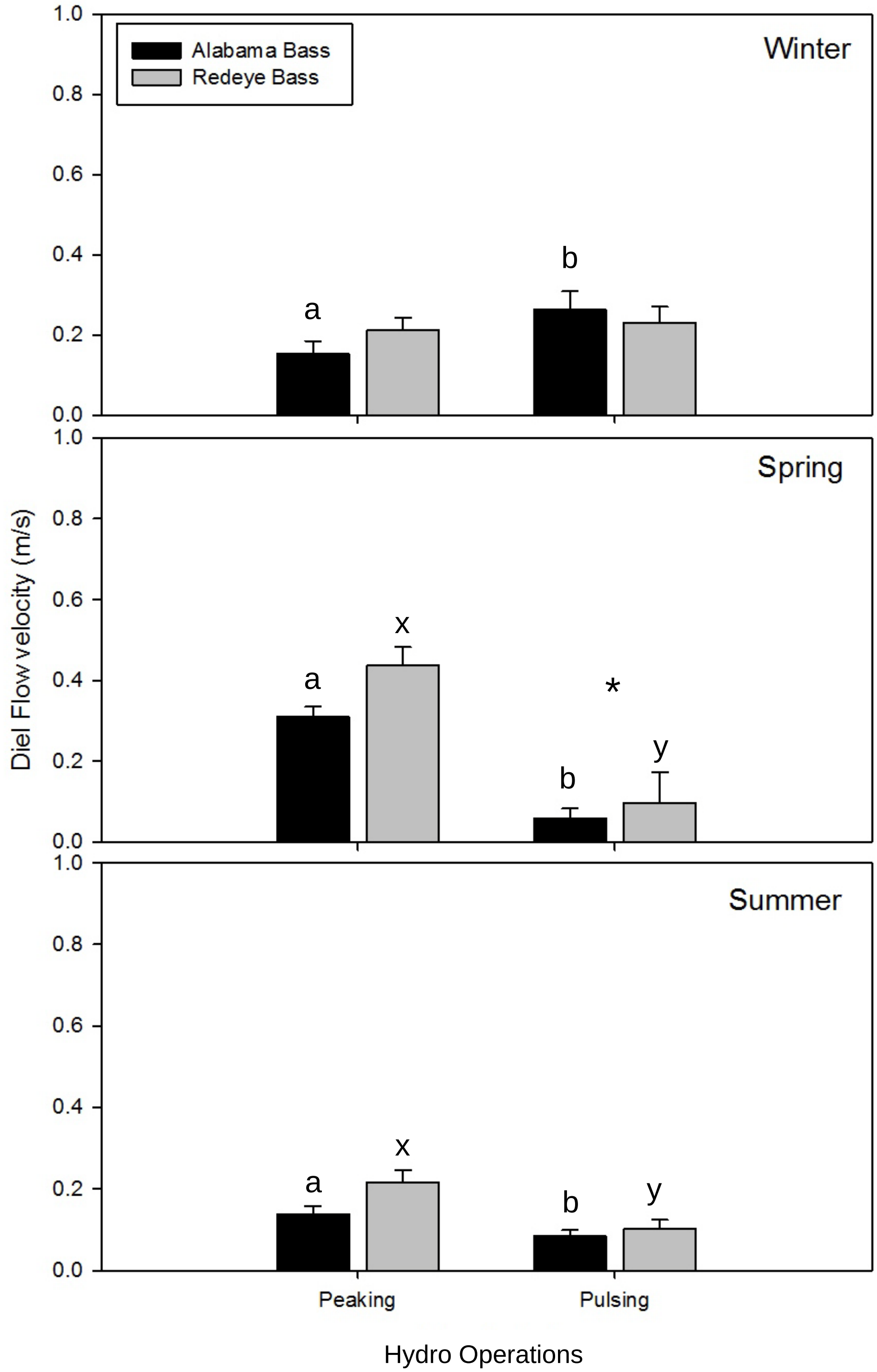

Figure 2-26. Flow velocity at the locations where Alabama Bass and Redeye Bass were observed during the two different dam operations in the Tallapoosa River, Alabama. Different letters represent a significant difference ($p < 0.05$) within the species and the asterisk represents a difference between the species ($p < 0.05$) and no notation means there was no significant difference.

3. An Investigation on the Physiological Condition of Alabama Bass and Redeye Bass, Based on Cortisol Levels and Leukocyte Profiles

Abstract. – Overall condition of fishes can be influenced by the amount of stress experienced. Stressors, such as alteration in temperature, oxygen or hydrology, can induce acute or chronic stress. Cortisol response is a good indicator of acute stress and additional measurements of stress include leukocyte profiles, with neutrophils increasing and lymphocytes decreasing (N:L). In this study I investigated whether the Hypothalamus-Pituitary-Interrenal (HPI) axis of Alabama Bass *Micropterus henshalli* and Redeye Bass *Micropterus coosae* was affected by hydropeaking operations at Harris Dam on the Tallapoosa River, Alabama. Fish were collected in fall on the Tallapoosa River and at two reference sites, Hillabee Creek and Saugahatchee Creek, which are two unregulated tributaries to the Tallapoosa River. Once collected, fish were tagged and blood samples were collected within five minutes of capture. Fish were transferred to a 113.5-L tub and remained in the tubs for a 1 h, whereupon another sample was collected. Blood smears were made for each sample. Blood was centrifuged and plasma was extracted and frozen until analysis. This confinement was expected to trigger the stress response in the HPI axis and cortisol concentrations and N:L should rise. The blood samples were assayed for plasma cortisol levels and blood smears were created and read. Both baseline cortisol levels and N:L were higher in the fish found at the disturbed location, and the percent change of cortisol was higher at the reference site. These results suggest that fish in the treatment site had an altered stress response that may have been due to the non-natural flow regime.

Introduction

Over the last several decades the field of physiological ecology has become more prevalent. Biologists are becoming more concerned with the impacts on animals exposed to anthropogenic disturbances, and are investigating how these disturbances affect the endocrine response (Romero 2004). When animals become chronically stressed, their physiology and behavior are often altered, with lower reproductive success and limited growth, being two of the major impacts (Barton et al. 1987; Van Weerd and Komen 1998).

Stress is best defined when an organism’s homeostasis is disturbed due to intrinsic or extrinsic stimuli, and these stimuli can be referred to as stressors (Chrousos and Gold 1992; Wendelaar Bonga 1997). There are two types of stressors, acute and chronic. Acute stressors are often associated with the “fight or flight” and other temporary situations; whereas, chronic stressors are events or factors that the animal is exposed to on a long-term basis. Once an animal has been stressed, it undergoes many physiological and behavioral reactions, which is known as an integrated stress response (Wendelaar Bonga 1997). The various reactions in the integrated stress response may occur within a few seconds to several minutes or hours after exposure to stressor (Sapolsky et al. 2000; Smith 2011). The impacts and the actual response of the organism is dependent on the type of stressor it’s enduring and the overall health of the organism (Romero 2004). Typically the stress response is an adaptive response, allowing for the animal to overcome the stressful situation; however, more intense or prolonged stresses, the response is maladaptive and possibly even physiological dysfunctional (Wendelaar Bonga 1997; Barton 2002).

The stress response begins with the activation of the nervous system and the adrenal medulla or, in teleostean fishes, the chromaffin tissue. This stimulus then allows for the rapid release of catecholamines, mainly epinephrine in fishes (Barton 2002). Next the Hypothalamic-Pituitary-Interrenal axis releases corticotropin-releasing hormone from the hypothalamus, resulting in secretion of adernocorticotropin (ACTH) by the anterior pituitary (Barton 2002). Once the ACTH begins circulation, the interrenal cells (adrenal cortex homologue) release glucocorticosteroids into circulation, and there is often a lag time associated to this steroid synthesis compared to catecholamine synthesis (Barton 2002). This initial process of the stress response is referred to as the primary stress response. The secondary stress response is when effects of hormones at the blood and tissue level, metabolic changes, changes in immune function, disturbance in osmoregulation and changes in hematological features are observed (Wendelaar Bonga 1997; Barton 2002). Lastly, the tertiary responses are the more direct effects on the whole organism. This is when changes are observed in behavioral patterns, performance characteristics and the organism may have a reduced response when exposed to future stressors (Wendelaar Bonga 1997; Barton 2002).

Typically when physiologists are investigating the stress response in organisms, glucocorticoid hormones, specifically corticosteroids (cortisol in teleostean fishes) levels in circulating plasma are the most common metric evaluated. This is due to the glucocorticoids increasing while the animal is responding to the environmental challenge (Bonier et al. 2009b), which allows for measurements to be collected during an acute stressor. The release of cortisol (CORT) is slower than catecholamines, but is much more prolonged, which allows for samples to be collected over a period of time (Waring et al. 1996; Martinez-Porchas et al. 2009). In most studies, a baseline CORT sample is collected and then the organism is exposed to an acute stressor and re-sampled to evaluate any changes in the CORT levels (Smith 2011). If there is little to no response after the animal has been exposed to the acute stressor, it is possible that the animal has been exposed to a chronic stressor and the HPI axis is no longer functioning at normal capacity (Hontela et al. 1992; Norris et al. 1999). Additionally, baseline levels have also been used to determine the health of an organism (Bonier 2009a). Higher baseline CORT levels have been associated to animals with poor reproductive fitness (Bonier 2009a).

Although CORT has been the most widely used metric for evaluating the stress response, there are some precautions with using this measure. There are many intrinsic and extrinsic factors that may impact CORT levels that need to be taken into consideration including; age, sexual maturity, species, previous exposure to stressors and exposure to chemicals (summarized in Martinez-Porchas et al. 2009). Baseline measurements of stress also need to be sampled quickly after capture. In fish, Maule and Mesa (1994) found that animals collected by electrofishing could be used for stress assessment if the fish are quickly sacrificed and sampled. Rainbow Trout *Oncorhynchus mykiss* showed a rise in CORT within 15 min of sampling (Barton and Grosh 1996), suggesting that there is a short window for these baseline samples to be collected. Additionally, CORT may not be the best indicator for chronic stress. As previously mentioned it is possible for the HPI axis to be exhausted or acclimated if the animals are exposed to a long-term stressor (Barton et al. 1985; Hontela et al. 1992; Norris et al. 1999; Romero 2004; Martinez-Porchas et al. 2009). Martinez-Porchas (2009) suggested that CORT still should be used as a bio-indicator but should be coupled with another marker, especially when evaluating chronic stressors.

There are several other bio-markers that can be used when assessing stress, such as white blood cell (WBC) counts, heat shock proteins, and measurements of other stress hormones.

Davis et al. (2008) reviewed the use of leukocyte profiles as method for evaluating stress. The use of white blood cell counts dates back to times prior to assay kits being readily available for evaluation of hormones (Dhabhar et al. 1996). Leukocyte profiles are altered when an animal is exposed to a stressor and is often related to levels of stress hormones (Davis et al. 2008). Vertebrates commonly have five types of WBC, which include lymphocytes, neutrophils (heterophils in some taxa), eosinophils, basophils and monocytes. However the two most common leukocytes, especially in fish, are lymphocytes and neutrophils. When an animal is exposed to a stressor the number of lymphocytes decrease, while neutrophils increase, allowing for the neutrophil:lymphocyte (N:L) ratio to be a measure of stress (Davis et al. 2008). Wedemeyer et al. (1990), suggested that in fish, N:L ratios could be one of the more sensitive markers for acute stress (Davis et al. 2008). Furthermore, when comparing both red and WBC counts in fish exposed to heavy metals, WBC counts were found to be more superior (Witeska 2005). This method has been shown to be reliable for all vertebrates and can be used to assess both acute and chronic stress.

In the field of fisheries, many studies have investigated the stress response, although the main focus of these studies has been in association with the aquaculture industry (reviewed by Wendelaar Bonga 1997). Outside of the review many studies have looked at the impacts of capture, handling, confinement and transportation (Carmichael et al. 1984; Davis and Parker 1986; Grutter and Pankhurst 2000). Additional studies have evaluated the stress response and its influence on behavioral traits (Barreto et al. 2009), how it is impacted by agricultural pollution (Miller et al. 2009), and its impacts on reproductive success (Gagliano and McCormick 2009). Other studies have investigated the differences between acute and chronic stressors (Pottinger et al. 1999), and how chronic stress impacts growth (Van Weerd and Komen 1998), physiological condition (Barton et al. 1987), feeding behavior, competitive ability and swimming performance (Gregory and Wood 1999) of fishes. The stress response in teleosts is similar to other vertebrate that are exposed to environmental challenges (Mazeaud et al. 1977; Barton and Iwama 1991; Wendelaar Bonga 1997). However, fish differ from terrestrial animals due to stressors inducing hydromineral disturbances and CORT and other hormones play an important role in controlling this disturbance (Wendelaar Bonga 1997). This highlights the importance of understanding stressors and the impacts on the CORT response in fish.

In light of the numerous dams around the world, researchers have been addressing how the alteration in both flow and temperature regimes impact the aquatic species that reside downstream of these instream structures. Hasler et al. (2009) suggested that researchers should attempt to use the ever expanding "toolbox" to investigate individual-level effects of hydropower operations on fish, including the use of endocrine measurements. Flodmark et al. (2002) investigated CORT and glucose responses in age-1 Brown Trout *Salmo trutta* that were subjected to simulated hydro-peaking operations in an artificial channel and found that these fish were able to rapidly habituate to the fluctuation in the flow. However, it has been noted in other taxa that when there is a habituation to a stressor, the animal may be in a state of allostasis and not homeostasis, which still has metabolic costs associated with it (Romero et al. 2009; McEwen and Wingfield 2010)

To determine if the altered flow regime impacts the stress response in Alabama Bass *Micropterus henshalli* and Redeye Bass *Micropterus coosae*, I choose to investigate the acute stress response of these animals. If these fish were chronically stressed, I expected to observe little to no response in the CORT levels to capativity, similar to previously studies that found fish to be chronically stressed when exposed to heavy metals (Hontela et al. 1992; Norris et al. 1999).

Additionally, I anticipated higher baseline levels in fish that were constantly exposed to the non-natural flows (Bonier et al. 2009a). As suggested in Martinez-Porchas et al. (2009), I examined these responses using plasma CORT but complemented this measure with WBC, due to the variability in CORT. By using both CORT and N:L ratios as bio-markers for stress I evaluated the baseline levels, the stress response, and the accuracy of the two methodologies. This study also allowed me to examine not only the differences between non-natural and natural flow regimes, but also evaluate stress responses between these two species, which have not been studied before.

Methods

In September 2011, a pilot study was completed on Largemouth Bass *Micropterus salmoides* at Auburn University's North Station research ponds. This study examined the best way to sample fish and allowed the methodologies of blood sampling and lab analyses to be established before field sampling in the fall. Ten fish were collected using a boom-mounted electrofishing boast on two occasions from pond S28 at the E.W. Shell Fisheries Center; blood was sampled from these fish within 5 min of capture. Blood was sampled from the caudal vein using 26-gauge heparinized needle; approximately 100 µL was collected for plasma assays and centrifuged on location, while approximately 50 µL was used in creating a blood smear. Fish were then floy tagged or fin clipped for identifying the fish in future samples. Fish were transported to a trough where they were confined with the other fish for 1 h. After 1 h samples were collected again, and fish were measured (total length, mm) and weighed (g) before being released.

In October and November 2011, three locations were sampled within the Tallapoosa watershed (Figure 3-1). The first site was on the regulated portion of the Tallapoosa River approximately 20 km downstream of Harris Dam (Figure 3-1). Due to time constraints, fish were collected for this study using the most efficient gear, even though all fish in the pilot study were collected using a boom-mounted electrofishing boat. Therefore, Alabama Bass were collected from the regulated site (N = 10) using a boom-mounted electrofishing boat but from the unregulated sites using angling. Likewise, Redeye Bass (N = 10 at each site) were collected from each site using angling. To ensure that stress responses of fish were not affected by capture method, an additional ten Alabama Bass were collected at the regulated site using angling and stress responses of fish collected with the two methods of capture and were compared using a student's t-test. When repeated sampling at the original sample site failed to collect the required sample size of Redeye Bass, additional fish were collected from Saugahatchee Creek, another unregulated tributary to the Tallapoosa River (Figure 1). Stress responses of fish from the two reference locations were compared using a student's t-test to ensure that fish from the two sites could be pooled into one reference location.

Once a fish was collected a stopwatch was started and time was recorded until sampling was complete. Blood was sampled from the caudal vein using the same method as in the pilot study, a blood smear was created, and then remaining blood was put on ice. Fish were floy-tagged and places in a 113.5-L cooler; biomass density did not exceed 50 g/L. The coolers were outfitted with an aerator and there were no water exchanges. Fish were than resampled 1 h after confinement. After the second sample was collected fish were euthanized with a 300 mg/L solution of Tricaine Methanesulfonate (MS-222) until expired and then placed on ice. Upon returning to the boat ramp, blood samples were centrifuged at 6,000 rpm for 7 minutes. Plasma was than extracted and on ice until returning to the lab where samples were frozen until they were assayed. Blood smears were fixed with methanol and stored until returning to the lab. Fish were measured (total length, mm), weighed (g), sexed, and otoliths were pulled for ageing. The coefficient of condition (Williams 2000) was calculated using the following formula:

$$K = \frac{100{,}000\, W}{L\text{^}3}$$

where: W = the weight of fish in grams;

L = the total length of the fish in millimeters.

Upon returning to the lab, blood smears were stained with a Hema-3 kit (Wright-Giemsa staining method; Fisher Scientific Company L.L.C., Middletown, VA). Slides were than dried, coverslipped, and stored until analysis. Each slide was read using a light microscope (100 x magnification). Relative counts of the blood cells were conducted and each type of WBC was tallied until 100 different leukocytes were identified (Smith 2011). The proportion of neutrophils to lymphocytes was calculated by dividing the number neutrophils by the number of lymphocytes. The blood plasma was assayed using commercially purchased enzyme-immunoassay kit designed for cortisol (Caymen Chemical, Ann Arbor, MI).

Evaluation of the CORT levels were completed by comparing mean CORT levels at each time period by reference and treatment site using a two-way analysis of variance (ANOVA). Similarly, N:L ratios were compared between times for each location. One-way and two-way ANOVAs were also completed to determine if there was a difference between sex, species, and age. To further investigate, the CORT response, percent change was calculated by taking the difference between the levels after 1 h and baseline and divided that by the baseline level. The percent change was then log transformed and analyzed with a two-way ANOVA to assess if there was a difference between location and species. The relation of fish condition, time sampled and CORT levels and N:L ratios were examined using Pearson correlations. All analysis was completed using Statview (version 5.0.1; SAS Institute Inc) and R (version 2.14.1; www.r-project.org).

Results

A total of 39 Alabama Bass and 11 Alabama Bass were collected from the Tallapoosa River and Hillabee Creek, respectively (Table 3-1). No significant differences existed between capture methods for the baseline samples of CORT (f = 2.91; *d.f.* = 1; p = 0.0976) or for the baseline N:L ratios (f = 0.21; *d.f.* = 1; p = 0.6470), thus all samples from the Tallapoosa site were pooled for Alabama Bass. A total of 11 Redeye Bass were collected from the Tallapoosa River; whereas 3 Redeye Bass were collected from Hillabee Creek and 5 from Saugahatchee Creek (Table 3-1). Samples from the two reference locations were pooled because stress responses of Redeye Bass were similar among locations for all time periods (baseline CORT: t = 0.23; *d.f.* = 6; p = 0.8258; baseline N:L ratios: t = -0.34; *d.f.* = 4; p = 0.7506; 1 h CORT: t = -2.50; *d.f.* = 5; p = 0.0544; 1 h N:L ratios: t = 1.20; *d.f.* = 5; p = 0.2826). Although fish of each sex were sampled at all locations for both species (Table 3-2), stress responses were similar among sexes (f=0.01; *d.f.* = 1; p=0.9279), and all sexes were pooled for all analyses (Table 3-2).

The condition of Alabama Bass was similar between the reference and treatment sites (t = 0.64; *d.f.* = 30; p = 0.5279). Condition of Redeye Bass was similar (t = -1.77; *d.f.* = 6; p = 0.1270) between the two reference locations, so all samples were pooled. There was no difference between condition of Redeye Bass at the reference site compared to the Redeye Bass from the Tallapoosa (t =-1.99; *d.f.* = 15; p=0.0650). Condition was not correlated to the four physiological parameters collected (Baseline CORT: r = 0.06; *d.f.* = 43; p = 0.6851; Baseline N:L ratios: r = 0.27; *d.f.* = 41 p = 0.0846; 1 h CORT: r = 0.16; *d.f.* = 42 p = 0.3181; 1 h N:L ratios: r = 0.11; *d.f.* = 43 p = 0.4796). Similarly, the amount of time (s) that it took to collect the first sample had no correlation to plasma CORT levels (*r* = 0.13; *d.f.* = 61; p=0.3140) or N:L ratios (*r* = 0.70; *d.f.* = 58; p=0.5968).

CORT levels were similar between species at each location across sample times (*f* = 0.168; *d.f.* = 1; p = 0.6840), but were different among locations for each species at both sample ,times (*f* = 5.7390; *d.f.* = 1 p = 0.0206), and there was no interaction between location and species (*f* = 1.4450; *d.f.* = 1,47 p = 0.2354; Figure 3-2). Plasma CORT levels were compared by location and species with a two-way ANOVA for the baseline levels (Figure 3-3) and levels after 1 h confinement (Figure 3-3). Baseline levels of CORT were higher at the treatment site than the reference site for both species (*f* = 8.3010; *d.f.* = 1; p = 0.0055) but were similar between species at each location (*f* = 0.2990; *d.f.* = 1; p = 0.5864), with no interaction detected between location and species (*f* = 0.2790; *d.f.* = 1,59; p = 0.5996; Figure 3-3). Levels of plasma CORT after 1 h of confinement were similar between locations (*f* = 3.5570; *d.f.* = 1; p = 0.0648) and species (*f* = 0.2570; *d.f.* = 1; p = 0.6146), with no interaction detected between location and species (*f* = 1.440; *d.f.* = 1,53; p = 0.2355; Figure 3-3). However, the percent change between the baseline and response CORT levels was higher at the reference site than the treatment site for both species (*f* = 4.9140; *d.f.* = 1; p = 0.0315), but was similar between species (*f* = 0.3110; *d.f.* = 1; p = 0.5795) and no interaction was detected between species and location (*f* = 1.4370; *d.f.* = 1,47; p = 0.2366; Figure 3-3).

The N:L ratios were similar between locations (*f* = 1.6570; *d.f.* = 1; p = 0.2042) and species (*f* = 0.1430; *d.f.* = 1; p = 0.7065), with no interaction detected between location and species (*f* = 0.1900; *d.f.* = 1; p = 0.6647; Figure 3-4). Similarly, baseline N:L ratios were similar between locations (*f* = 2.4140; *d.f.* = 1; p = 0.1259) and species (*f* = 0.2570; *d.f.* = 1; p = 0.6142), with no interaction detected between location and species (*f* = 0.3180; *d.f.* = 1; p = 0.5748). Also response sample N:L ratios were similar between locations (*f* = 1.1420; *d.f.* = 1; p = 0.2900) and species (*f* = 0.0170; *d.f.* = 1; p = 0.8958); no interaction was detected between location and species (*f* = 0.2800; *d.f.* = 1; p = 0.5992). Because there was no significance for the N:L ratios the ratio of change was not calculated and analyzed.

Discussion

In this study baseline CORT levels were higher in fish collected from the treatment site than those collected from the reference sites. The significance of higher baseline CORT levels to the overall health of organisms has been debated for years in the field of physiological ecology. In this study I found that there was not a difference in condition between the two locations for both species. Also it was observed that there was no major impact on growth of both Alabama Bass and Redeye Bass in the Tallapoosa River, which would also suggest that there was not a decrease in fitness (Chapter 1). Condition was not correlated to the baseline levels or response levels of CORT and there was not a difference between baseline CORT levels and species.

Baseline CORT levels were higher at the treatment site than the reference site for both species, and although no significant differences were detected in the response CORT levels between the two locations for either species, there was an apparent trend for CORT levels to be higher in treatment fish than reference fish. This is likely due to highly elevated levels of CORT already circulating in the treatment fish. Also, the percent change of CORT response was higher in the reference fish than in the treatment fish, suggesting that there is an altered response for the fish found in the treatment location. This could be due to acclimation to the chronic stress, fish in the treatment location trying to re-establish homeostasis, the fish are in a state of allostasis (Romero et al. 2009), and/or the fish are chronically stressed.

Acclimation can be best defined as when the animal no longer responds to the long-term stressor (Romero 2004). Typically, this is due to the animal no longer reacting to the stressor; thus, this acclimation could be considered habituation. Flodmark et al. (2002) concluded that fish exposed to experimentally fluctuating flows quickly habituated to the stressor. However, if the fish in the altered flow regime had acclimated or even become habituated to the stressor of an altered environment, it does not explain why the baseline levels were different. Romero (2004) further explains that when an animal becomes acclimated to a stressor, the HPI axis is altered. This alteration in the HPI axis enhances the response to other stressors, and is termed facilitation (Romero 2004). This facilitation of the stress response would help to explain why there was still a CORT response to the acute stressor, but again does not explain why the baseline levels of CORT were higher for Alabama Bass and Redeye Bass collected from the Tallapoosa River.

Allostasis was defined by McEwen and Wingfield (2003) as the concept of an on organism coping with an environmental threat (summarized in Romero et al. 2009). McEwen and Wingfield (2003) further describe the allostatic state to be when there are altered levels of the primary stress mediators, using glucocorticoids as an example. This could explain the high level of circulating CORT in the fish in the altered environment. However if the organism is in an allostatic state for a long period of time, it can result with the animal suffering from allostatic overload. This overload can lead to allostatic state independent of the environment and have major impacts on life history stages, such as sexual maturity and seasonal migrations (McEwen and Wingfield 2003).

Wingfield and Kitausky (2002) proposed another theory that is similar to that of allostasis, that there are emergency life history strategies to cope with stress, which they defined as the “leave-it” strategy, the “take-it” strategy, and the “take it first and then leave-it” strategy. The “leave-it” strategy is when an animal moves away from the unpredictable event, the “take-it” strategy is when the animal switches to a different set behavioral and physiological traits, and the “take it first and then leave-it” strategy is a combination of the two when the animal switches to the energy conserving mode and if the situation does not improve they leave (Wingfield and Kituasky 2002). They stated that higher baseline levels of CORT could represent a form of the "take-it" strategy, which may be why fish did not move out of the area during a telemetry study completed on the Tallapoosa River (Chapter 2). So, it is possible that the fish in the Tallapoosa River are currently in an allostatic state, experiencing allostatic overload, or they are functioning under an emergency life history strategy.

Alabama Bass and Redeye Bass in the Tallapoosa River could also be chronically stressed by the fluctuating flows. Generally, a symptom of chronic stress is elevated baseline levels of CORT (Rich and Romero 2005). Several studies also show that baseline CORT concentrations were higher in chronically stressed animals (Moore et al. 1991; Fowler et al. 1995). Rich and Romero (2005) suggested one way to attenuate to a chronic stressor is a downregulation of the HPA axis, where the animal no longer responds normally, which would help to minimize the effects of chronically high CORT levels. Other studies have shown an exhaustion of the HPI axis in animals exposed to chronic stress (Hontela 1997; Norris 1999). In my study, high baseline levels were observed and the stress-induced response was actually smaller for the fish in the Tallapoosa River based on percent change. The high baseline levels were similar to what has been observed in other studies of chronically stressed animals. The lesser response in the treatment fish was similar to what was observed in Rich and Romero (2005), which may indicate that there was a downregulation of the HPA axis in these fish in the Tallapoosa River.

In this study I did not find any significant evidence that N:L ratios could be used as a bio-marker for stress. Nevertheless, there was a trend in my data of the baseline N:L ratios being higher in the treatment groups. Additionally, after the 1 h confinement the N:L ratios were higher than baselines ratios for all the experimental groups. The change in leukocyte distribution can be considered to be induced with the release of glucocorticoids into the bloodstream (Dhabhar et al. 1996). However, this change in the leukocyte profile may not respond as quickly as other secondary stress responses. Davis et al. (2005) reviewed several studies that examined the variation in temporal sampling and concluded that it may be some time after exposure to a stressor for an increase in N:L (or H:L) ratios to be observed. It is possible that my 1 h sample was too soon to observe any effects on the N:L ratios. Additionally, the sample size for this study was relatively small and it is possible with a larger sample size the effects at the baseline level could have been observed. However, results from this study agreed with Müller et al (2011) that N:L ratio and plasma CORT concentrations cannot replace each other.

Conclusion

This study showed that there was difference in the CORT response between fish that are exposed to a natural flow regime and fish exposed to an alter flow regime. Although this study is only correlative and does not show causation that the non-natural flow regime is the reason for the altered stress response, I feel that this study provides good background information for further research to be conducted on the stress response of fish in hydrologically altered environments. Further field investigations may help to determine if this fish are in an altered state, i.e. allostasis, are chronically stressed or if there is a downregulation in the HPI axis. As Norris (2000) points out, laboratory studies may not adequately explain what is actually happening to fish in their aquatic habitats, which may explain the differing results from my study and Flodmark et al. (2002). Studies such as a 24 h stress test, an ACTH challenge (Norris 2000), or simply completing more field studies with a larger sample size and more locations, could allow for a more complete understanding of the stress response. The information from further work can then be combined with other data that is known about these fish. For example, if there is evidence of poor recruitment, it's possible that the altered stress response is affecting reproductive success.

As other researchers have pointed out CORT concentrations should be coupled with another bio-marker (Norris 2000; Martinez-Porchas et al. 2009). Although results from my study indicated that N:L ratios may not be useful to identify stress responses of Alabama Bass and Redeye Bass, further work should be conducted on this marker to evaluate whether higher sample sizes of longer time intervals between samples may affect the utility of this marker. Davis et al. (2005) points out that there are challenges that need to be further investigated in order for this bio-marker to be an accurate measure of stress. With the trends observed in this study there appears to be reason to include this bio-marker in future studies.

Table 3-1. The number of fish collected and the method of capture at the tree sampling sites, the treatment site on the Tallapoosa River, and the two reference sites on Hillabee Creek and Saugahatchee Creek.

Species	Method	Tallapoosa River	Hillabee Cr.	Saugahatchee Cr.	Total
Alabama Bass		**39**	**11**	**0**	**50**
	Electro-fishing	17	0	0	17
	Hook and Line	22	11	0	33
Redeye Bass		**11**	**3**	**5**	**19**
	Electro-fishing	0	0	0	0
	Hook and Line	11	3	5	19

Table 3-2. The number of species and the number of fish where gender, length and weight were collected at the three sampling sites, the treatment site on the Tallapoosa River, and the two reference sites on Hillabee Creek and Saugahatchee Creek.

Species	Sex	Tallapoosa River	Hillabee Cr.	Saugahatchee Cr.	Total
Alabama Bass		**39**	**11**	**0**	**50**
	Male	11	5	0	16
	Female	11	5	0	16
	Unknown	17	1	0	18
Redeye Bass		**11**	**3**	**5**	**19**
	Male	4	1	2	7
	Female	5	2	3	10
	Unknown	2	0	0	2

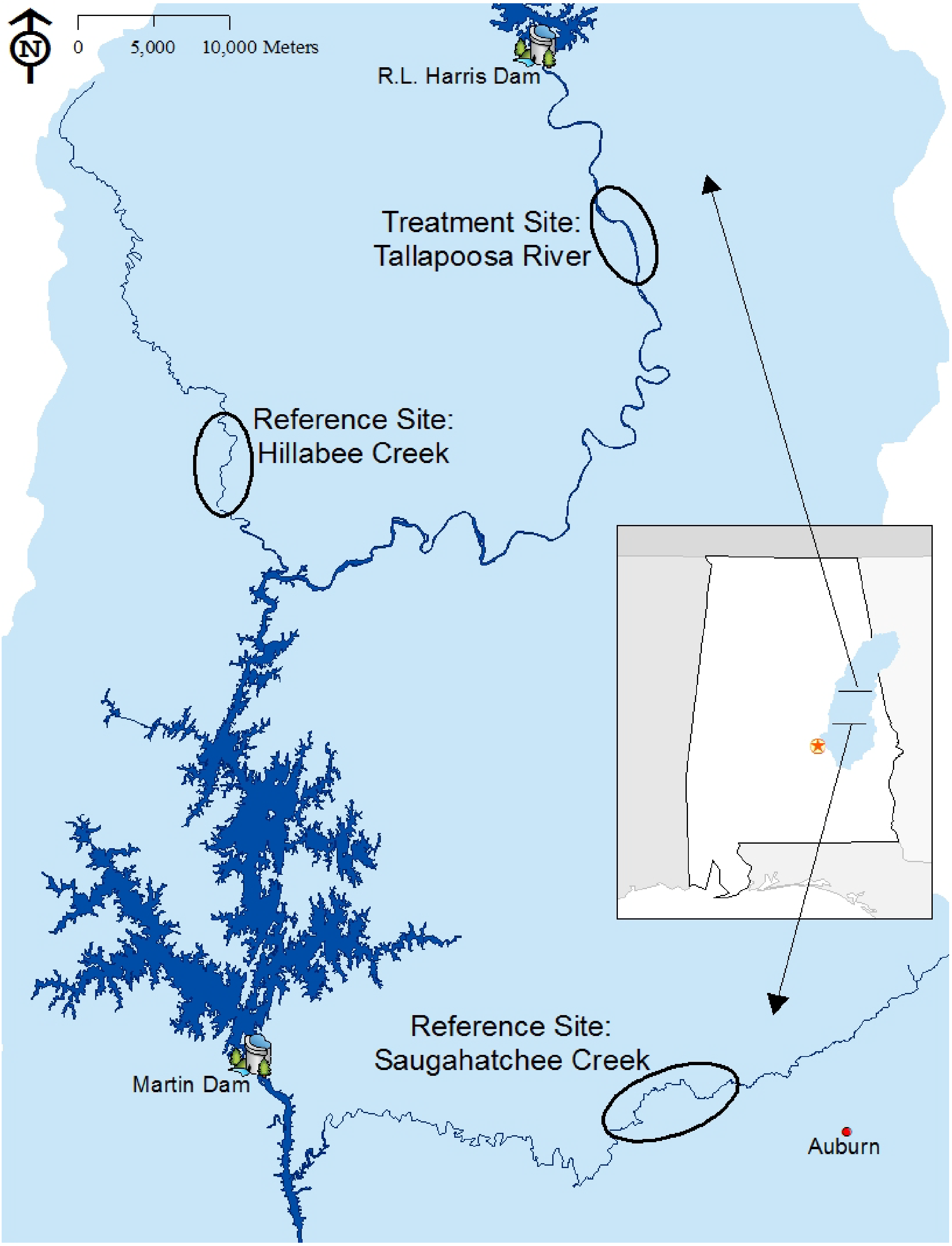

Figure 3-1. Overview map of the Tallapoosa Watershed and the three sampling locations, the treatment site on the Tallapoosa River and the two reference locations on Hillabee Creek and Saugahatchee Creek.

Baseline 1 h

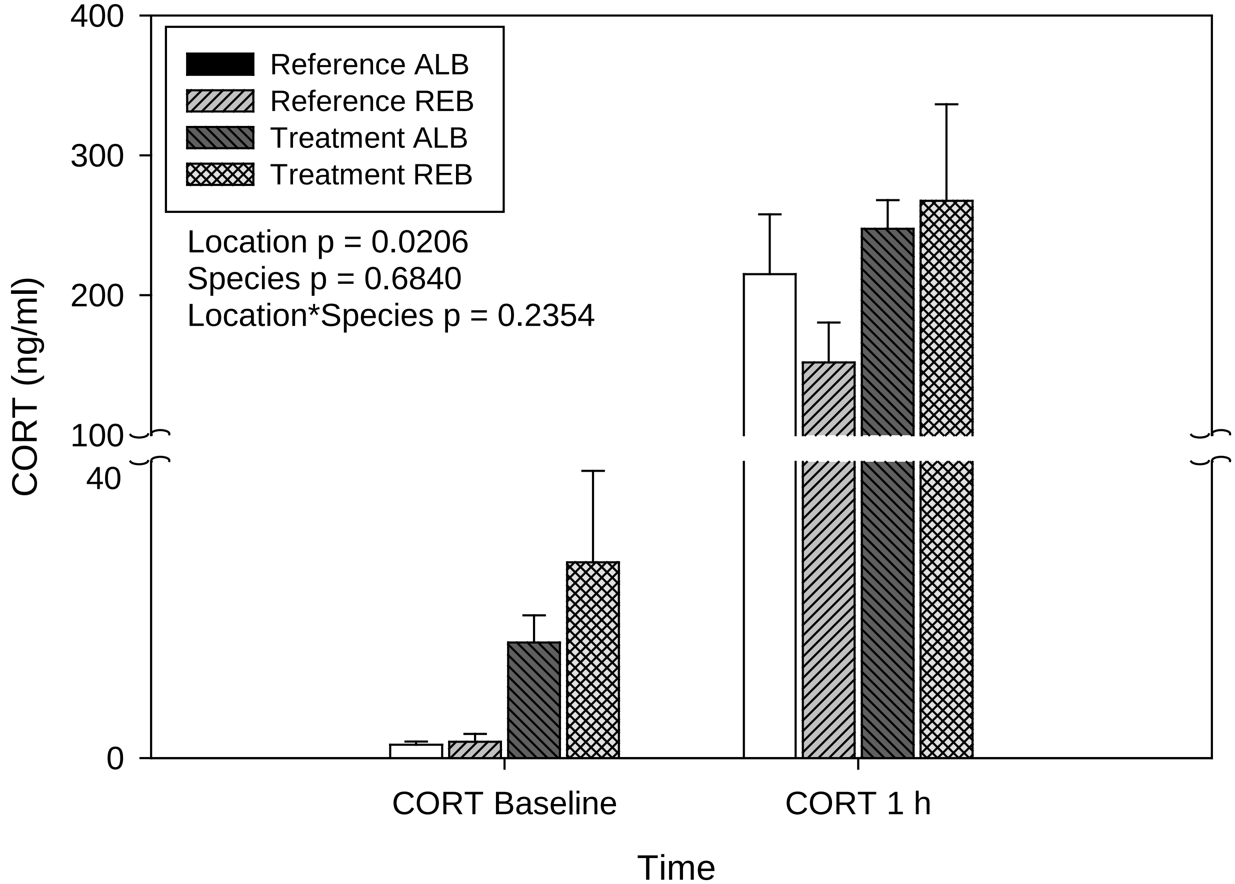

Figure 3-2. Cortisol levels increased in both the Alabama Bass (ALB) and the Redeye Bass (REB) at both the reference and treatment locations in response to confinement. The lines represent standard errors.

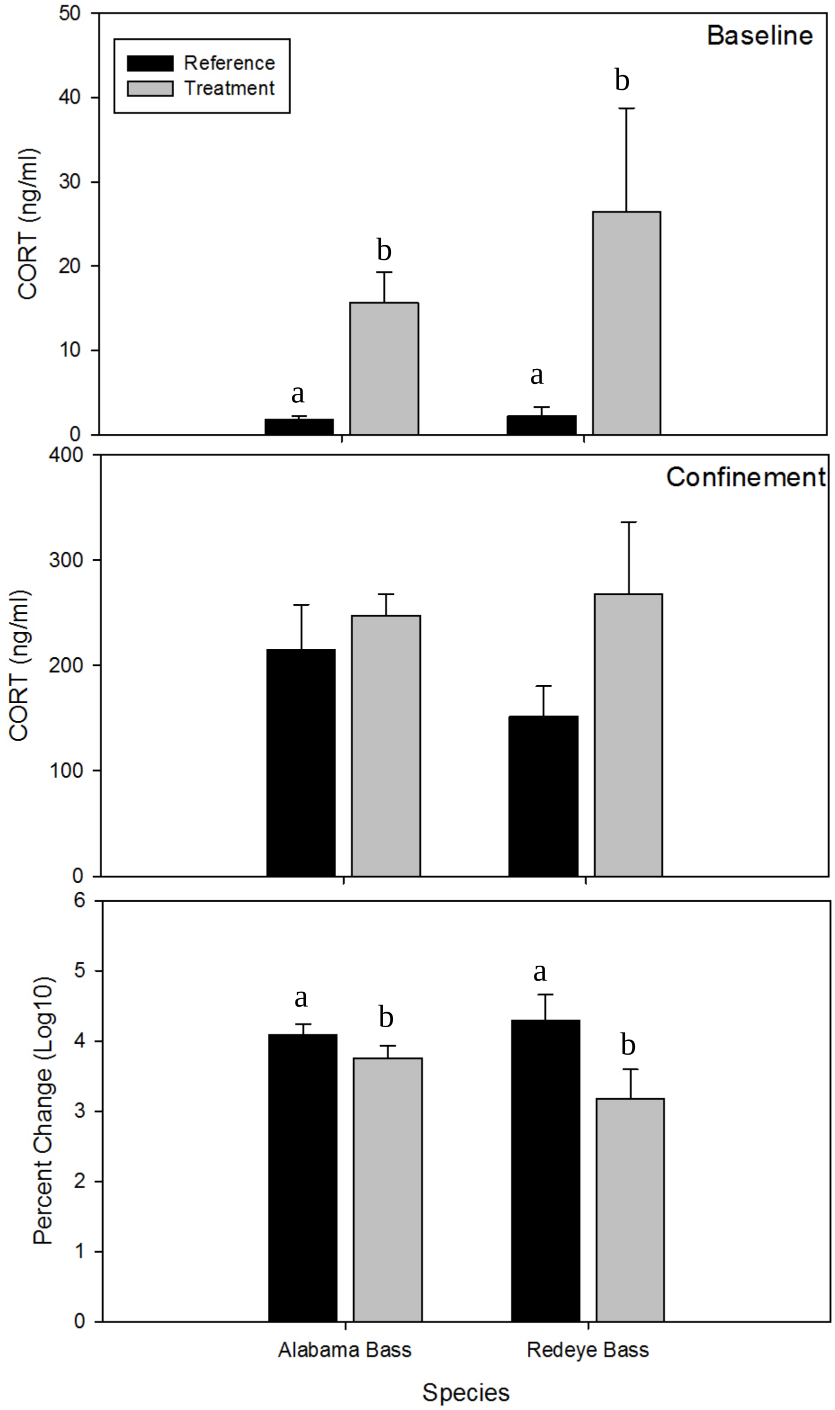

Figure 3-3. Baseline, response levels of plasma cortisol and the percent change between the baseline and response plasma cortisol levels for Alabama Bass and Redeye Bass at the reference and treatment locations. Identical letters represent non-significance and the lines represent standard errors.

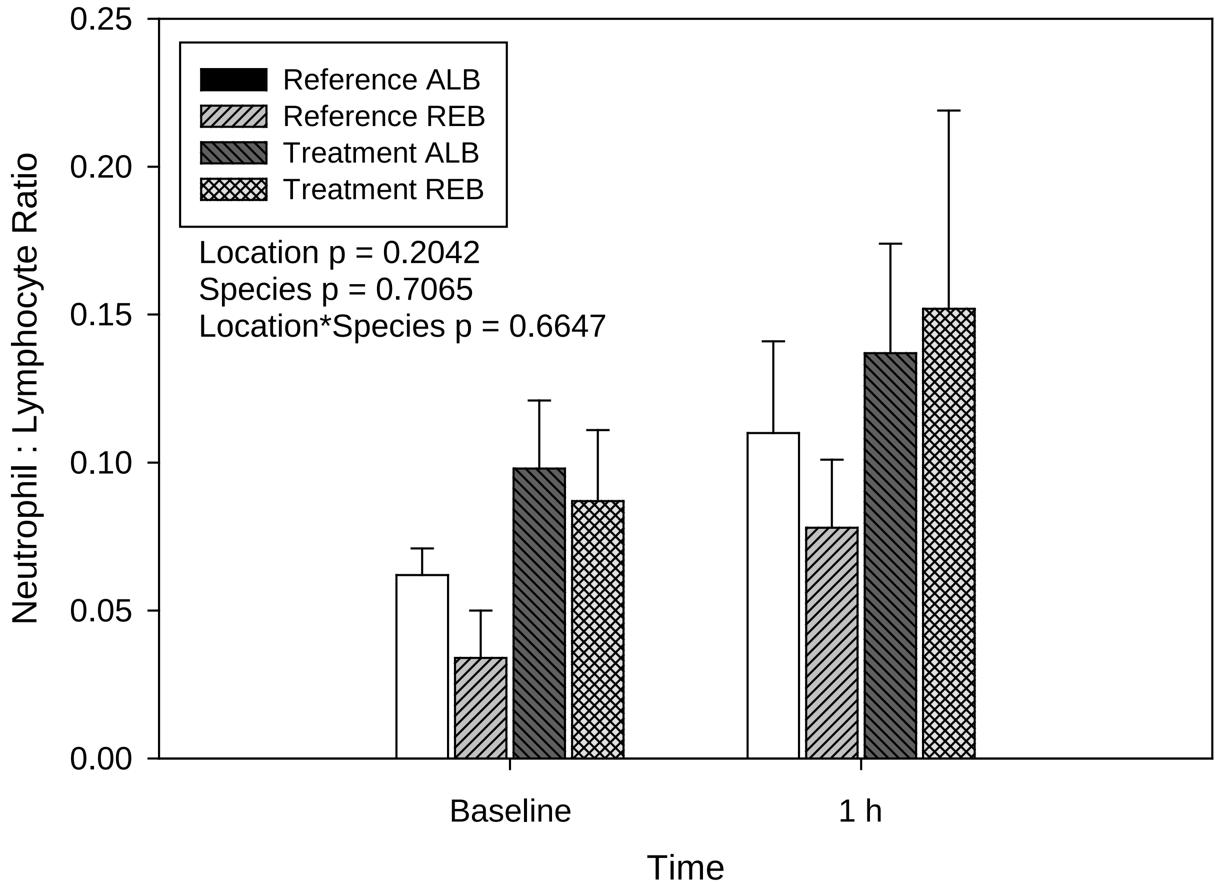

Figure 3-4. Neutrophil:Lymphocyte ratios showed a similar trend for the cortisol response for both Alabama Bass and Redeye Bass at the reference and treatment locations, however there was not a significant difference. The lines represent standard errors.

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