

**Optimizing a Standard Sampling Program to Evaluate Fish Community Structure for  
Non-wadeable Rivers in Alabama**

by

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## Abstract

Alabama's non-wadeable rivers support exceptionally high biodiversity of fishes but no formal sampling program has been developed to monitor the fish community in these systems. Recent developments in Alabama have caused increased interest by Alabama Department of Conservation and Natural Resources biologists to develop such a sampling program, but studies determining adequate sampling effort are lacking. This study was developed to compare two different boat-based electrofishing methods (continuous bank-line and point sampling) to sample the fish community and determine the effort necessary to accurately represent the fish communities present. Furthermore, a comparison of day and night electrofishing was done in two rivers with the continuous bank-line method. Four rivers of various sizes and locations in Alabama were sampled along two 100-mean-stream-width transects. Because habitat complexity can affect sampling effort, substrate was mapped using side-scan sonar within select reaches of each river, and low and high complexity transects were identified for electrofishing sampling. Sampling was done in summer and fall of 2015 and 2016. This study had three objectives: 1) Determine the optimal amount of sampling required to collect 50% and 75% of the species expected to be encountered, with 95% confidence, using bank-line and point sampling in each river, 2) Assess the effects of season and habitat complexity on sampling optimization for bank-line and point sampling in each river, and 3) Determine the effect of day/night sampling on sample optimization for bank-line sampling in two rivers. At 95% confidence, the bank-line method caught 50% of species estimated within 30 to 50 mean-stream-widths, and 75% of species estimated within 90 to 130 mean-stream-widths during fall. Point sampling was the least effective method and did not capture enough species to determine effort necessary to meet sampling objective. Fall was generally a more efficient season than summer to meet sampling objectives. Species richness and sampling effort was typically not affected by habitat complexity category. Night shocking was more efficient than day in the larger river, but similar in the smaller river.

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## List of Abbreviations

ADCNR	Alabama Department of Conservation and Natural Resources
BLE	Bank-Line Electrofishing
CPE	Catch-per-effort
HHC	High Habitat Complexity
LHC	Low Habitat Complexity
PASE	Point Abundance Sampling by Electrofishing

## **I. INTRODUCTION**

Non-wadeable rivers are important features of the landscape and often support popular fisheries for a variety of species, yet have often been overlooked in sampling programs. In recent years there has been increased interest in monitoring the status and trends of fishes in these systems, but studies determining adequate sampling effort are lacking. Agencies responsible for monitoring thousands of river km need consistent (standardized) and cost-efficient methods for making assessments. Oversampling causes excessive labor costs, while undersampling produces inaccurate and imprecise estimates of fish species richness and abundance – important indicators of fish assemblage integrity and diversity (Angermeier and Smoger 1995; Peterson and Rabeni 1995; Hughes et al. 2002; Dauwalter and Pert 2003; Vehanen et al. 2013). Therefore, guidelines for optimization of fish community sampling in non-wadeable rivers are needed by agencies to design efficient surveys (Seegert 2000).

Riverine ecosystems typically support high biodiversity (Sowa et al. 2007), and Alabama has the greatest fish diversity of any state in the U.S., with over 300 species of fish found within its borders (Boschung and Mayden 2004), and the vast majority of this diversity is found within riverine ecosystems. Despite this diversity, little attempt has been made to monitor changes of the fish communities in lotic habitats within the state, especially in non-wadeable rivers (S. Rider, ADCNR, personal communication). Sweeping land-use changes have been occurring throughout the southeastern U. S. in recent decades, including Alabama (Warren et al. 2000), and biologists fear that concomitant loss of fish biodiversity may go unnoticed with no monitoring program in place.

### **I.1. Monitoring Programs**

Standardization of monitoring programs have been used on lakes and reservoirs for decades and have proven essential to gain an understanding of the interactions between spatial and temporal patterns and variability of populations (Yeh 1977; Boxrucker et al. 1995; Kubecka et al. 2009). Proper monitoring programs are critical for making informed management decisions, inferences, or testing for responses to ecosystem variability. Without monitoring, science-based decisions are often not possible (McClelland et al. 2012). Standardization of electrofishing methods that provides data of equivalent quality for comparison across waterways has proven difficult for lotic systems. Effort is recorded by either time or distance, with distances either fixed or proportional to river size. Data can be collected over one long transect per river, but are more commonly collected over a set number of subsamples for replication. However, standardization and statistical problems, in addition to the other difficulties associated with river sampling, led Tomanova et al. (2013) to state “the study of fish communities in large, non-wadeable rivers remains one of the most difficult problems in freshwater ecology”.

The most direct and effective measure of integrity of an aquatic system is the status of life in the water (Karr and Chu 2000). The Index of Biotic Integrity (IBI) was developed to assess the ecological quality of lotic systems (Karr 1981). The original index was designed for midwestern U.S. streams and consisted of 12 fish community metrics, divided into categories of species richness, trophic structure, and fish abundance and condition. It was created as a result of the 1972 Federal Water Pollution Act, and its 1977 and 1987 amendments that were enacted to restore and maintain biological integrity of the nation’s waters (Miller et al. 1988). The IBI has become one of the most commonly used biological monitoring approaches for assessing the status of fish communities and the overall ecological condition of streams (Karr and Chu 2000). The original IBI has been adapted to numerous geographic regions in the United States (Miller et

al. 1988; Whittier et al. 2007) and beyond (Belpaire et al. 2000; Van Liefferinge et al 2010), usually modified to account for indicator species/families found in each area. Although the original IBI and many of its later adaptations were created for small streams, some have successfully used it in larger systems (Minns et al. 1994; Bowen et al. 1998; Lyons et al. 2001; Hughes and Herlihy 2007; Maret et al. 2007). The use of the IBI and other simplified multi-metric indices is more difficult in systems that contain high species diversity and habitat heterogeneity (Bowen et al. 1998; Dauwalter and Pert 2003). One reason for this is that the IBI is greatly affected by rare species, which tend to be distributed in a patchy manner (Dauwalter and Pert 2003; Meador 2005; Kanno et al. 2009). Therefore, sampling effort should be sufficient to collect these rare species, while minimizing labor cost.

Compared with the biological assessment of wadeable streams, assessment of non-wadeable riverine fish communities has lagged (Meador 2005). Many factors that have been shown to dictate sampling effort in small streams (e.g. Patton et al. 2000; Lyons et al. 2001) are likely also relevant for larger, non-wadeable rivers. If increased precision and accuracy in estimates of species richness are desired, adjustments in sampling effort relative to species richness are required (Paller 1995; Hughes et al. 2002). Compared to smaller streams, many large rivers support more fish species that are difficult to sample because of their association with deep, turbid, or swift moving waters. Although sampling all habitat types is necessary to adequately represent fish assemblage structure and species richness (Angermeier and Smoger 1995); this is rarely possible in larger, non-wadeable rivers (Hughes et al. 2002).

## **I.2. Sample Design Issues in Lotic Systems**

Insufficient sampling effort increases the variability for estimates of species richness and abundance, largely because of rare species that are distributed in a discontinuous manner (Kanno

et al. 2009). High species diversity is often associated with a greater number of rare species, which may not be collected until large area of suitable habitat have been searched (Paller 1995). Fish species can be considered rare because their required habitats are rare in the sample reach, their population density is low, or were inadequately collected by the chosen sampling gear (Kanno et al 2009). Many authors suggest removing rare species from data sets when two or less individuals are collected from an entire reach, or when a species composes less than 1% of relative abundance (e.g., Paller 1995; Kanno et al 2009). Attempts to collect all rare species would substantially increase sampling effort well beyond the means of most managing agencies. For instance, Hughes et al. (2002) predicted that an average of 286 mean wetted channel widths (MWCW) would have to be sampled to collect all species present in non-wadeable Oregon rivers.

Electrofishing is generally considered to be most effective gear over a variety of conditions and for a wide range of species (Seegert 2000; Flotemersch and Blocksom 2005). Numerous studies have shown electrofishing to collect the highest number of species with the least amount of effort (e.g., Wiley and Tsai 1983; Patton et al. 2000; Poos et al. 2007). Much of the early electrofishing efforts in lotic systems used multiple-pass depletion techniques, but many studies have found that single pass electrofishing methods have similar precision and accuracy, with much less effort (Paller 1995; Simonson and Lyons 1995; Odenkirk and Smith 2005; Bertrand et al. 2006; Vehanen et al. 2013).

A comprehensive river monitoring program will use several different gears to adequately sample all available habitats and attempt to balance specific gear biases, as all fish sampling gears are species and size selective (Miranda and Schramm 2000; Patton et al. 2000; Schramm et al 2002). The main channel of navigable rivers is an important habitat for larger specimens and

certain fluvial fish species, as well as a pathway for migratory species (Boschung and Mayden 2004). However, this habitat cannot be efficiently sampled by electrofishing (Dauwalter and Fisher 2007) because this gear only effectively samples fishes in shallow (<1.5 m) and near-shore habitats (Poos et al. 2007). Thus, detecting all species present in navigable rivers likely requires multiple gears fished over several seasons, and would only be accomplished with a great amount of effort (Galat et al. 2005). Some other commonly used gears to survey rivers include gill nets, fyke nets, trawls, trammel nets, seines, and hoop nets (Herzog et al. 2005; Bonar et al. 2009; Welker and Drobish 2011; Neebling and Quist 2011). Standardizing sampling effort and comparing data pooled from different reaches becomes difficult when using multiple gears to assess the overall assemblage (Lyons et al. 2001). However, electrofishing at night may substitute for additional sampling gears in large rivers because main-channel species often move inshore during nighttime hours (Flotemersch et al. 2011).

The amount of sampling effort required to adequately estimate fish community metrics is often dictated by habitat complexity (Simonson et al. 1994; Angermeier and Smoger 1995; Zorn et al. 2011). As river size increases (i.e. order, width), the length of river required to encounter the same number available habitats increases proportionally (Pearson et al. 2011). Additionally, as systems increase in size, the proportion of deep, open-water habitats that are difficult to sample may also increase, thus potentially increasing the site length or number of gears required to adequately sample the site (Flotemersch et al. 2011; Pearson et al. 2011). There is no single site length that addresses all research and applied questions; instead the appropriate site length for stated objectives will be a compromise between the overall survey design, the intensity of data collection for a particular sampling event, the resource constraints present, and the sampling

gear used (Bowen and Freeman 1998, Bertrand et al 2006; Fischer and Paukert 2009; Flotemersch et al. 2011).

Some studies support the use of a fixed-distance sampling site length (Flotemersch and Blocksom 2005) while others propose site lengths as a function of multiples of the mean wetted channel width (MWCW) (e.g. Lyons 1992; Hughes et al 2002; Dauwalter and Pert 2003). Using the MWCW approach increases sampling effort with river size. This approach can complicate comparisons between sites because sampling lengths and effort are not equivalent. Also, wide rivers confer extremely long sampling sites that could require excessive effort and cost (Flotemersch et al. 2011), which can be overcome by setting a maximum site length (Meador and McIntyre 2003; Flotemersch and Blocksom 2005). The fixed-distance approach is easier to implement in the field and involves less logistical planning (Patton et al. 2000; Lyons et al. 2001; Flotemersch and Blocksom 2005), but can miss or under-represent habitats that occur at longer intervals, especially in larger rivers. Fixed-distance surveys have unequal sampling effort relative to river size which could result in greater variability from reference condition, reducing overall accuracy and precision of the data (Hughes and Herlihy 2007; Maret et al. 2007). In studies of non-wadeable lotic habitats, site lengths used for fish sampling varied from 500 m (Blocksom et al. 2009) to 100 MWCW (Hughes et al. 2002).

Site-scale sampling design includes specifying the spatial scale over which the samples will be collected (site length), the amount and types of habitats that will be sampled within that site, the field sampling methods to be used, and the estimated number of person-hours (Flotemersch et al. 2006). Estimates of species richness are sensitive to site-scale design and sampling effort, because riverine habitat is heterogeneous, with irregular distribution of fishes among habitat types (Angermeier and Smoger 1995; Kanno et al. 2009). As a result, the number

of species collected at a given site will increase with sampling effort, but will also vary with biogeography, behavior, and abundance of the species being sampled, as well as the patchiness of the macrohabitat types (Flotemersch et al. 2011). Species richness tends to reach an asymptote much sooner in homogenous rivers as site length increases; whereas, heterogeneous rivers with discontinuously distributed species requires much greater effort for species richness to reach an asymptote (Hughes et al. 2002; Kanno et al. 2009; Flotemersch et al. 2011). The ideal sampling effort is the minimum that will allow the stated objectives to be precisely and accurately addressed (Angermeier and Smoger 1995; Patton et al. 2000; Dauwalter and Pert 2003). Estimates of species relative abundances have been shown to require less sampling effort for a given accuracy than estimates of the absolute number of species (Angermeier and Smoger 1995; Dauwalter and Pert 2003).

When sampling rivers with diverse macrohabitat types it is important to carefully identify and consider the geographic extent of interest (e.g. river segment to microhabitat) before selecting and setting the sampling site length (Flotemersch et al. 2011). Maximizing habitats sampled can be accomplished by either adding different gear types, targeting specific habitats, or increasing site length (Seegert 2000; Bonar et al. 2009). When a long sampling site is warranted for estimating spatially extensive characteristics, sampling several shorter sub-sites with separate data is better than one large data set for one site, to generate multiple data points that can be used to determine conditions at a smaller geographic range (Hughes et al. 2002; Fischer and Paukert 2009; Kanno et al. 2009). A split-geographic design will provide data at two geographic extents; the smaller also provides a means of estimating variability within the larger geographic extent, assuming data are recorded by sub-site. Data from multiple shorter sub-sites can be used with randomization methods such as Monte Carlo analysis, standard error estimates, and similarity

indices. Data collected in this manner is also needed to determine the effect of site length and sample size (Flotemersch et al. 2011).

Flotemersch et al. (2011) recommends an approach to sample large navigable rivers by sampling many smaller sites based on the relative occurrence of major macrohabitat types that are present in the system. The optimal amount of effort that would be necessary to sample all fish species present with 90-95% confidence within each macrohabitat could be determined and adjusted based on the availability of these macrohabitats. This approach could increase the ability to detect habitat-specific relations that might be concealed by sampling designs that include longer reaches without separation. Quantifying habitats present in a system could lead to an increase in sampling efficiencies, resulting in greater overall spatial coverage of the resource for the same level of effort expended (Flotemersch et al. 2011).

Many fish species exhibit diel patterns of habitat use for feeding and migration, which could influence catchability and detectability of some species in standardized surveys (Flotemersch et al. 2011). Because these patterns are often species- or size-specific, their effects on standardized surveys can be difficult to quantify (Baumgartner et al. 2008). Species or sizes that might be considered rare or absent during the day could be more commonly collected with night collections and give a better representation of abundance, size structure, or species composition (Flotemersch and Blocksom 2005). Night electrofishing has been found to yield more species and greater sizes and biomass than daylight sampling (Lyons et al. 2001), but logistical and safety concerns may limit adoption of this approach by agencies in lotic systems regardless of concerns over data quality (Hughes et al 2002). In lotic systems with channel depths greater than 4 m, diel fish movements could significantly impact the quality of daytime

electrofishing results enough that consideration of night electrofishing could be justified (Flotemersch and Blocksom 2005).

### **I.3. Point Abundance Sampling by Electrofishing**

Although most agencies in the U.S. typically conduct electrofishing in rivers by sampling along the bank, a different method has been developed and used in other parts of the world, especially in Europe. This technique, called point abundance sampling by electrofishing (PASE), appears to overcome some of the standardization problems of traditional bank shocking. Using the PASE method, electrofishing is conducted at a series of fixed points for a set amount of time (e.g. 30 s). This sampling method was originally designed in France in the late 1970's then spread to numerous studies throughout Europe (Tomanova et al. 2013) and more recently been employed in North America (Lapointe et al. 2006; Trumbo 2016). Tomanova et al. (2013) found PASE to produce a greater number of samples with less effort or time compared to continuous bank sampling. Many small samples often have less variability and more statistical power than a few large ones (Miranda et al. 2000; Lapointe et al. 2006). The PASE design provides a relatively accurate estimate of common species density, but often fails to capture rare species (Lapointe et al. 2006; Tomanova et al. 2013). Thus, like other electrofishing designs, it provides only an incomplete picture of community structure. Although PASE designs usually are used with random points, Tomanova et al (2013) suggested a systematic strategy that ensures proportional sampling of all fishable habitats. They also suggested collecting 10 additional sampling points from rare habitats to improve estimates of species occurrence and to gain valuable local knowledge on the habitat preferences of rare species. This technique is not commonly used in the United States and has not been assessed in rivers of the southeastern states, which are characterized by high habitat heterogeneity and species richness.

#### **I.4. Habitat Classification using Side-Scan Sonar**

It is difficult to assess the effectiveness of sampling efforts in lotic systems without knowing the habitat types present and the relative area these habitats occupy within a system (Zorn et al. 2011). Quantification of the available habitat types within a significant sampled length of river has been difficult in the past (Simonson et al. 1994). Habitat classification have typically only been recorded on a macrohabitat level, or when taken on a finer scale (such as substrate parameters) has not been adequately quantified for an entire reach. Cost associated with acquiring these data has been prohibitive for state and federal agencies, because it has traditionally required either very expensive equipment, highly trained personnel, excessive person-hours of labor, or tools that are not suitable for use in shallow waters (Kaeser and Litts 2010).

In 2005, Humminbird (Eufaula, AL) released relatively low-cost (< \$2,000) side-scan sonar units that can produce high resolution images of substrates, woody debris, submerged objects, and depth. This system can be used to map sub-surface habitat features in any water body large enough to transport a small watercraft. Soon after the release of these Humminbird side-scan sonar units, Kaeser and Litts (2008) developed techniques to use this technology to record images and locate deadhead logs submerged in small streams. This grew into developing techniques to classify the substrates from these images at a high rate of accuracy and speed using very little effort in person-hours (Kaeser and Litts 2010) and has progressed to relate these habitats to ecology of fish and mussel species (Goclowski et al. 2013; Smit 2014). Given the importance of habitat distribution to designing sampling surveys, this technique shows great promise to delineate the exact habitat composition of large reaches of river and allow sample

reaches to be chosen based on maximizing habitat diversity or in proportion to available habitats. However, to date, little if any of this work has been attempted.

Ultimately, the quantity and quality of information desired and the funding and personnel available dictates the level of effort that is expended for monitoring each site. Quantification of species richness and relative abundance using multiple sub-site sampling and data recording with randomization-based analyses and testing of the capacity of the sampling design to meet these objectives should drive the appropriate site length. Increased standardization of these monitoring study designs is needed to facilitate data exchange, indicate data quality, improve credibility and make assessments more comparable within and among political jurisdictions (Flotemersch et al. 2011).

#### **I.5. Research Goal and Study Objectives**

This study evaluated the ability of boat electrofishing to sample the fish communities of four non-wadeable Alabama rivers using standard bank-line electrofishing (BLE) and PASE. Each method conducted was along transects characterized by low and high habitat complexity to explore the effects of this variable on sampling efficiency. Furthermore, a comparison of day and night electrofishing was done in two of the four study rivers, ones that differed considerably in size and geomorphology (Tallapoosa River and Alabama River). This study had three objectives: 1) Determine the optimal amount of sampling required to collect 50% and 75% of the species expected to be encountered, with 95% confidence, using BLE and PASE in each river, 2) Assess the effects of season and habitat complexity on sampling optimization for BLE and PASE in each river, and 3) Determine the effect of day/night sampling on sample optimization for BLE sampling in two rivers.

## **II. Methods**

### **II.1. Study Area**

This research was conducted in four non-wadeable rivers within Alabama (Figure 1). The four rivers and approximate sampling locations were chosen by ADCNR personnel (Steve Rider, personal communication) to be a representative sample of non-wadeable rivers in various parts of the state. The Sipsey River originates by the Marion and Fayette county lines near Glen Allen, Alabama, and flows 148 km across northwestern Alabama before entering the Tombigbee River near Vienna, Alabama. It is the second longest free-flowing river in Alabama, and flows through an estimated 123,500 ha of bottomland swamps and wetlands (Hopper et al. 2012). The Sipsey River tranverses both the Southwestern Appalachian Plateau and Southeastern Plains ecoregion and is considered a diversity “hotspot” for freshwater mussels (42 species) and fishes (102 species). Sampling was conducted seven miles south of Aliceville, Alabama, off of state route 14 along the border of Pickens and Greene County (Figure 1).

The Alabama River forms at the confluence of the Coosa and Tallapoosa rivers, and flows 512 km towards the Gulf of Mexico, draining approximately 58,500 km<sup>2</sup> of eastern Alabama, northwestern Georgia, and a portion of southeastern Tennessee (Boshung and Mayden 2004). The Alabama River is joined by the Tombigbee River in southwestern Alabama to form the Mobile River before flowing into the Mobile-Tensaw Delta and eventually into Mobile Bay. As many as 96 species of fish have been collected from the mainstem Alabama River, although evidence points to a steady decline in species diversity over the last four decades (Freeman et al. 2005). This river was sampled in the reach downstream of Claiborne Lock and Dam below U.S. Highway 84 Bridge (Figure 1).

Another free-flowing river, the Choctawhatchee River begins in Barbour County, Alabama, and flows 227 km across the Coastal Plain region through Florida, before entering the Gulf of Mexico. Considered another diversity hotspot, the Choctawhatchee River contains 70 species of fish (Boshung and Mayden 2004). Only the upper 87 km of the Choctawhatchee River is within Alabama's borders and sampling efforts on the Choctawhatchee River took place just north of the Florida border, near Geneva, Alabama (Figure 1).

The Tallapoosa River begins in Paulding County, Georgia, and flows southwesterly 426 km across the Piedmont Upland and East Gulf Coastal Plain regions before its confluence with the Coosa River to form the Alabama River. The Tallapoosa River has been impounded four times, mostly in the lower portion of the Piedmont and the Fall Line. Although at least 126 species of fish once occurred in the Tallapoosa River, loss of habitat and flow alterations resulting from impoundment have reduced the native fish fauna (Irwin and Freeman 2002). The study site on the Tallapoosa River was 19 km northeast of Dadeville, Alabama, near the Germany's Ferry Road crossing (Figure 1). This area of the river undergoes frequent fluctuations in flows and water levels from Harris Dam, approximately 53 km upstream (Earley 2012).

In each river, boat ramps were located and rivers were scouted to determine a distance in either direction that could be accessed by boat. Some rivers had natural barriers such as shoals or shallow sand flats that would restrict boat access beyond these points, especially during periods of low water. Other rivers sampling endpoints were determined by reasonable distance from ramp alone. Efforts were made to eliminate dam effects of sample results as much as possible by sampling a minimum of two miles below a dam or before a reservoir effect of river widening was noticeable above a dam.

## **II.2. Habitat Surveys**

### **II.2.1. General Methods**

During spring 2015, after determinations of endpoints were made, the selected reaches of each river were surveyed using side-scan sonar during high-flow events to maximize streambed area surveyed. Survey lengths were from 11 to 40 km long depending on mean wetted channel width (MWCW) and navigability (Table 1). The wetted channel width was measured at 10 random points on the river using a Nikon RifleHunter 1000 laser range finder. Points where MWCW measurements were taken were recorded on the GPS. After actual sampling areas were determined, measurements were then taken at 10 evenly spaced intervals along the entire survey areas, compared to satellite images and the mean was calculated as the MWCW for each river.

Sonar surveys were taken using a boat outfitted with a Humminbird 1197c Side Imaging system coupled with a GPS; sonar images of the riverbed were collected using a frequency of 455 kHz and a range of up to 50 m per side. Rivers with a MWCW greater than 100 m were surveyed using multiple passes down each side, with resulting images stitched together in ArcGIS (Smit 2014). After sonar images and GPS data were collected, they were processed using SonarTRX Pro software to geo-reference and curve images that were taken on the bends of the river to facilitate proper placement on ArcMap. Processed sonar images were then placed over satellite images within ArcMAP. River bank lines were outlined, then substrates were identified, outlined, and labeled into four categories, based on size (Table 2; modified from Kaeser and Litts [2010]). The areas of each substrate type were measured to determine total area, locations, and habitat availability in each river system. Percent substrate type was calculated for each river (Table 3).

### II.2.2. Transect Selection

Two transects were chosen for sampling in each river using the habitat maps. One transect represented high habitat complexity and the other represented low complexity. Habitat complexity of each transect was quantified by counts and area of each substrate category. High-habitat complexity (HHC) transects were chosen first from possible transect lines which passed through the most number of substrate polygons. Low-habitat complexity (LHC) transects were then chosen from a transect line on the opposite shoreline which passed through the fewest number of substrate polygons. Each transect was 100 MWCW long except in the Alabama River, where they were capped at 10 km. Each transect was divided into 10 subtransects of equal length (i.e. 10 MWCW or 1 km in the Alabama River; Table 1). Because transects were so long (i.e. 4.0 – 10.0 km), it was decided *post hoc* that habitat complexity was unlikely to be similar across the entire transect. Therefore, after each transect was divided into 10 equal subtransects, each subtransect was then categorized as HHC or LHC based on the number of substrate polygons a line 7.5 m from shore (i.e. representing the electrofishing boat transect) passed through. Due to varying levels of habitat complexity across rivers, river-specific criteria were used to designate high and low complexity. For the Alabama, Choctawhatchee, and Sipsey Rivers, the LHC category consisted of subtransects that only passed through a single substrate polygon. The HHC category from these three rivers consisted of subtransects that passed through two or more polygons. The Tallapoosa River had more diversity in terms of percent area of non-sand substrate and count of non-sand polygons than the other rivers (Table 3). Therefore, LHC was classified as a subtransect that passed through three or fewer substrate polygons, and subtransects that passed through four or more substrate polygons were considered

to be HHC. The final placement and locations of each subtransect for each river is listed in Appendix 5-8.

### **II.3. Species Surveys**

#### **II.3.1 General Methods**

Each river was sampled four times, in both summer and fall of 2015 and 2016. The summer samples were conducted during July and August and the fall samples were conducted in October and November. Fish were collected using electrofishing on a boat with dual boom-mounted electrodes, and was standardized in all rivers by setting the control box at 5.0 amps with pulsed DC current at a rate of 60 pulses per second. Stunned fish were collected by a single crew member using a long-handled dip net from the bow of the boat. All collected fish were identified and counted. Additionally, all sportfish and large-bodied nongame species were measured to total length (mm) and weighed (g). All fishes that were not able to be positively identified in the field were preserved in a 10% buffered formalin solution and returned to the lab. A representative sample of voucher specimens of most species (i.e., those not of conservation concern) were also preserved in a 10% buffered formalin solution for reference and verification. Fish species that were too large or state-listed were vouchered with photographs and released unharmed.

Due to varied sizes of the rivers sampled, appropriate sized boats and electrofishing gear were used for each river. The Alabama River was by far the largest river sampled with a MWCW of over 200 m, more than twice as wide as the next largest river. For this river a 6.1-m boat was used with a Smith Root 7.5 GPP electrofisher system. The other three rivers were sampled with a 4.3-m boat equipped with a Midwest Lakes Infinity electrofishing system, paired with a 5000-watt generator. Summer 2015 sampling encountered a major outboard failure early

on with the smaller boat and a 4.3-m boat equipped with a Smith Root 2.5 GPP electrofisher system was used for all sampling on the smaller rivers. A similar problem occurred in fall 2016 with the large river boat and all sampling on the Alabama River was done using a 5.5-m boat equipped with a Smith Root 5.0 GPP electrofisher system.

### **II.3.2. Bankline Methods**

Transects in all rivers were sampled between the hours of sunrise and sunset using the BLE method. These transects were sampled again at night using BLE on the Alabama and Tallapoosa Rivers; these samples were conducted between 30 min after sunset and 30 min before sunrise. These rivers were chosen to evaluate night BLE due to their differences in size, substrate composition, and geomorphology. Day and night BLE on these rivers was separated by at least two weeks, and the order that samples were done was chosen at random.

All BLE sampling was conducted from start line to stop line on the GPS for each subsample, keeping the boat in water roughly 1.0 - to 1.5m deep. Start and stop times, date, subsample number, and “pedal down” timer readings were recorded for each subsample. Transect length was the same for all subsamples within a river, and electrofishing times were maintained as close as possible among subsamples. Species data from each subsample was recorded immediately after completion of electrofishing (Appendix 4). Physicochemical parameters (temperature, pH, conductivity, and total dissolved solids) were recorded at the beginning of each day and at a sampling midpoint for the day. Fish were processed in a separate work up boat with two crew members on the Alabama and Tallapoosa Rivers to maximize sampling efficiency. Due to lower numbers of fish from shorter transects, sampling on the Choctawhatchee and Sipsey Rivers was able to be accomplished with a three-person crew on the electrofishing boat.

### **II.3.3. Point Sampling Methods**

Sampling using the PASE method was conducted using the same gear and along the same transects described above. This sampling occurred one day before daytime BLE sampling, as it was considered to be unobtrusive and unlikely to affect catch rates of the more intensive BLE sampling. A total of 100 points was evenly spaced along each transect 1 MWCW apart, except for the Alabama River where points were evenly spaced at 100 meters apart. Thus, each subsample reach from BLE sampling contained 10 PASE samples. Points were selected and spaced using GIS in the lab; during sampling the electrofishing boat was navigated to by approaching the bank from the channel, and when depth approximated those of the BLE transect, the boat was pointed upstream and position maintained against the current while electrofishing began and continued for 30 s. A GPS point and depth measurement was recorded at each exact location. In addition, at every 10th point physiochemical parameters (temperature, pH, conductivity, and total dissolved solids) were recorded. Fish were collected and identified in the same manner as described for BLE sampling. All 200 points for PASE sampling was done using a single boat with a three-person crew in one day for each river.

### **II.4. Data Analysis**

All statistical data analyses were conducted using Program R statistics software (R Core Team 2015). Species richness was quantified as a count of species present for each reach, also referred to as  $S_o$  for species observed. Diversity was calculated for each reach using Shannon's Diversity Index using the "diversity" function in R with the "vegan" package.

Species accumulation curves were created for both HHC and LHC transects in each river using the community ecology package (i.e. vegan) in R with the function "specaccum" using the classic method "random", which finds the mean species accumulation curve and its standard

deviation from 1,000 random permutations of the data, or subsampling without replacement. Species accumulation curves were used to evaluate the influence of increasing number of samples on estimates of species richness (Oksanen et al. 2008).

Estimates of species richness was calculated for both HHC and LHC transects in each river by using the “specpool” function in R. For this analysis, abundance data of each species captured in each subsample were used with the Chao method. The Chao method of estimating species richness uses the equation:

$$S_P = S_0 + \frac{a_1^2}{2a_2} \frac{N-1}{N}$$

where  $S_p$  is the extrapolated richness,  $S_0$  is number of species observed,  $a_1$  represents the number of species captured in only one subsample (i.e. singletons),  $a_2$  is the number of species captured in only two subsamples (i.e. doubletons), and  $N$  is the number of subsamples. When no doubletons were present, a bias-corrected version was used. In that case the Chao equation simplifies to:

$$S_P = S_0 + \frac{1}{2} a_1 (a_1 - 1) \frac{N-1}{N}$$

Proportional abundance was calculated by dividing the number of each species by the total abundance of all species combined (Guy and Brown 2007). Proportional abundance was used for a version of Morisita’s similarity index modified by Horn (1966):

$$C_H = \frac{2 \sum_{i=1}^S x_i y_i}{\left( \frac{\sum_{i=1}^S x_i^2}{X^2} + \frac{\sum_{i=1}^S y_i^2}{Y^2} \right) XY}$$

where  $x_i$  is the number of times species  $i$  is represented in the total  $X$  from one sample and  $y_i$  is the number of times species  $i$  is represented in the total  $Y$  from another sample and  $s$  is the number of unique species in each sample. This index was used to compare species composition

between HHC and LHC transects, between seasons, and to compare day and night BLE sampling (Guy and Brown 2007).

In the two rivers where night-time BLE sampling was conducted, species accumulation curves were created for each diel period and analyzed as described above for daytime BLE sampling. Species composition was compared between day and night sampling in each river using Morisita's similarity index. Values of 0.6 and above are considered very similar and values below 0.4 are considered low similarity (Haley and Johnston 2014).

To evaluate species accumulation curves, a function was created in R to calculate the number of samples needed to capture a percentage of the species estimate with a percent of confidence. This function was written as follows:

```
samp_func2=function(x,proportion_of_max,confidence_level,max_species){  
  threshold=proportion_of_max*max_species  
  x1=ifelse(x>threshold,1,0)  
  samps=1:(dim(x)[1])  
  probs=apply(x1,1,mean)  
  min(samps[probs>confidence_level])  
}
```

Where `proportion_of_max` was a value between 0 and 1 representing percent species, `confidence_level` was a value between 0 and 1 representing percent confidence, and `max_species` was a value representing the species estimate. The output was a number representing the minimum number of samples needed to obtain a specified percent of total estimated species with the level of confidence specified. The species richness estimate used for each river was a mean of estimates from all sampling events from day BLE and night BLE (when applicable). Species

estimates from PASE sampling was not included in this mean because richness was vastly underestimated or had extremely high standard errors from the estimate.

The effects of season, habitat complexity, diel period, and gear on number of samples necessary to achieve 50% of species estimate with 95% confidence were assessed using ANOVA's (R Core Team 2015). Each river was analyzed separately because of the differences in geomorphology, equipment used, and definitions of habitat heterogeneity. Forward stepwise selection was used to fit the best model to the data for each river. All statistical comparisons were considered significant at P-value of 0.1 because of the low number of samples that could be taken on a two year project.

### **III. RESULTS**

#### **III.1. Habitat Surveys**

Sample reaches in all rivers were dominated by sand which included all fine substrate as defined in Table 2. The Tallapoosa River sample reach had the least percentage of sand compared to the other three rivers (Table 3). Bedrock was the second most predominate substrate in sample reaches of all rivers, ranging from 6.1% to 14.4%. Gravel and boulder categories composed less than 4% of the substrate in all rivers except the Tallapoosa River which had a combined total of 13.0%.

Based on MWCW, transect lengths were set at 4.0, 5.8, and 7.8 km in the Sipsey, Choctawhatchee, and Tallapoosa rivers respectively; transect length in the Alabama River was fixed at the 10-km maximum length as noted previously. After placing subtransects in their final categories, the Sipsey River had equal numbers of subtransects in HHC and LHC categories. The Alabama River and Choctawhatchee River had 11 LHC and 9 HHC subtransects. The Tallapoosa River had 9 LHC and 11 HHC subtransects. Since most rivers had unequal numbers

of subtransects in these categories, comparisons between these groups were done at the highest common efforts (i.e. 9 subtransects or 90 points).

### **III.2. Total Fish Collection**

In total, 60,998 individual fish were collected across rivers, representing 106 different species in 24 different families. Most fish were collected from the Alabama River with 35,161 individuals representing 65 species within 21 different families. A total of 14,267 individual fish was collected in the Tallapoosa River representing 45 species in 11 different families. In the Choctawhatchee River, 6,731 fish were collected representing 50 different species from 18 families. The Sipsey River yielded 4,839 individual fish representing 60 species from 17 different families.

### **III.3. Bankline Electrofishing Method**

The Alabama River yielded a total of 11,576 individual fish of 56 different species in 21 families with 51.4 h of pedal time using 273.2 person-hours. The Tallapoosa River yielded a total of 5,788 fish of 40 unique species in 9 families with 47.7 h of pedal time using 240.8 person-hours. In the Choctawhatchee River, 5,323 fish were sampled from 48 species in 18 families with 33.7 h of pedal time using 152.4 person-hours. In the Sipsey River, 4,020 fish were sampled from 55 species in 16 families with 30.8 h of pedal time using 142.5 person-hours of labor.

#### **III.3.1 Season Effects in BLE Sampling**

Species accumulation curves combined across habitats and transects (i.e. 20 subtransects per sampling event) for the Alabama River showed higher species richness from both 2016 samples than 2015 samples (Figure 2). Species richness was identical in both seasons in 2015 (38) and only differed by one between summer (46) and fall (47) of 2016. All curves started to

level out and approach an asymptote as subtransects increased. Species richness estimates using the Chao equation ranged from 41 to 54 across seasons and years (Figure 2). Morisita's similarity index was calculated (Table 4) and indicated a larger difference in fish community structure between seasons (0.57 and 0.71) than between years (0.93 and 0.87). When separating species accumulation curves by habitat, mean species richness was lower in summer than fall (Table 5). Mean catch per subtransect was also lower in summer (105) than in fall (184), though there was a large year difference in fall with CPE's being much higher in 2015 than 2016 (Table 5). Mean Shannon diversity index was higher in summer (2.73) than in fall (2.48).

In the Tallapoosa River, combined species accumulation curves (20 subtransects per sampling event) showed consistently higher species richness in fall over summer (Figure 3). Species richness was 26 and 27 for summer and 31 and 33 for fall. Species richness estimates from the Chao equation ranged from 28 to 63, with much higher estimates and standard errors from fall as these curves were still rising, whereas summer curves more closely reached an asymptote (Figure 3). Morisita's similarity index (Table 4) indicated similar fish community structure between seasons and between years among seasons with all values above 0.80. When separating by habitat, mean species richness was lower in summer than fall (Table 6). Mean catch per subtransect was much lower in summer than in fall as were Shannon diversity index means.

In the Choctawhatchee River, combined species accumulation curves (20 subtransects per sampling event) leveled out and approached an asymptote as subtransects increased in fall, but showed steeper slopes in summer samples (Figure 4). Species richness was 32 and 39 from summer sampling events and 36 and 40 from fall sampling times. Species richness estimates using the Chao equation ranged from 38 to 43 with low standard errors. Morisita's similarity

index (Table 4) indicated similar communities between seasons and between years among seasons with all values above 0.60. When separating by habitat, mean species richness was lower in summer than fall (Table 7). Mean catch per subtransect was much lower in summer (48) than in fall (85) with CPE's being much higher in 2015 than 2016. Mean Shannon diversity indices were lower in summer than in fall (Table 7).

In the Sipsey River combined species accumulation curves (20 subtransects per sampling event) showed species richness was higher in both fall sampling events (39 and 44) than in summer (33 and 37). These curves (Figure 5) start to approach an asymptote as subtransects increase but were still gaining species at the end of sampling, with fall curves rising more steeply and ending closer to an asymptote than summer curves. Species richness estimates using the Chao equation ranged from 50 to 75. Standard error for these estimates was highest in summer 2015, which had the lowest species richness and highest species estimate. Morisita's similarity index (Table 4) indicated similar communities between seasons and between years among seasons with all values between 0.75 and 0.84. When separating by habitat (Table 8), mean species richness was lower in summer (27.8) than fall (34.5). Mean catch per subtransect was also much lower in summer (28) than in fall (72). Mean Shannon diversity index was lower in summer (2.37) than in fall (2.46).

### **III.3.2. Habitat Effects in BLE Sampling**

In the Alabama River, mean species richness was virtually identical between the LHC (34.9) and the HHC (35.5; Table 5). Shannon diversity indices were likewise identical between the categories. Not surprisingly, Morisita's similarity index values were very high between the habitat categories, with values ranging from 0.86 to 0.91 (Table 9) across samples. Mean catch per subtransect (all species) was 150 in LHC and 138 in HHC.

More of a difference was observed between habitat categories in the Tallapoosa River. Mean species richness was 22.5 in the LHC category and 25.4 in the HHC category (Table 6). Shannon diversity index was also lower in the LHC compared to the HHC category. However, Morisita's similarity index values were very high between the habitat categories with values ranging from 0.94 to 0.99 across samples (Table 9) for each sampling event. Mean catch per subtransect (all species) was 75 in LHC and 70 in HHC.

Similar to the Alabama River, habitat complexity had less of an effect on fish community structure in the Choctawhatchee River. Mean species richness was 30.1 in the LHC category and 32.8 in the HHC category (Table 7). Shannon diversity indices were very similar between habitat categories. Morisita's similarity index values were likewise high between the habitat categories with values ranging from 0.95 to 0.99 across samples (Table 9). Mean catch per subtransect (all species) was 67 in LHC and 66 in HHC.

Habitat complexity also had little effect on the estimates of fish community structure on the Sipsey River. Mean species richness was virtually identical between the LHC and HHC categories (Table 8), however, the mean Shannon diversity index was higher in the LHC (2.49) than the HHC category (2.34). Regardless, Morisita's similarity index values indicated high similarity between the habitat categories (i.e. > 0.90) except for summer of 2015 which had a value of 0.78 (Table 9). Mean catch per subtransect (all species) was 56 in LHC and 45 in HHC.

### **III.3.3. Day BLE Required Effort**

Results were mixed for the significance of season and habitat on the number of samples needed to acquire 50% of the species estimate with 95% confidence for each river. In the Alabama River, the number of samples needed to reach this objective was similar between seasons (summer 4.5, fall 3.5) and between habitat categories (HHC 4.3, LHC 3.8;  $F_{3,7} = 0.8$ ;  $P$

= 0.5551). But in the Tallapoosa River, sampling required more effort to reach this objective in the summer (7.0 samples) than in the fall (4.0); sampling in LHC took 6.5 samples compared to 4.5 samples from HHC ( $F_{3,7} = 18.67$ ;  $P = 0.0081$ ). In the Choctawhatchee River, more samples were required to meet the sample objective in the summer (5.5 samples) than in the fall (3.3 samples,  $F_{1,7} = 6.23$ ;  $P = 0.0670$ ) but was similar between habitat categories (HHC 4.0, LHC 4.8,  $F_{1,7} = 0.69$ ;  $P = 0.4522$ ). A similar relation was observed in the Sipsey (summer 8.0, fall 4.8;  $F_{1,7} = 18.78$ ;  $P = 0.0123$ ; HHC 4.0, LHC 4.8;  $F_{1,7} = 0.11$ ;  $P = 0.7.556$ ).

#### **III.3.4. Night BLE on the Alabama and Tallapoosa Rivers**

Even though it was only conducted on two rivers, more fish were caught from the night BLE samples than any other method. The Alabama River collected a total of 21,260 individual fish of 59 different species with 51.2 h of pedal time using 280.0 person-hours. The Tallapoosa River yielded 7,367 individual fish representing 37 species with 46.6 h of pedal time using 261.6 person-hours.

Species accumulation curves combined across habitats and transects (i.e., 20 subtransects per sampling event) from night BLE in the Alabama River showed higher species richness from both 2016 samples than 2015 samples, and higher species richness in fall compared to summer within each year (Figure 6). All summer curves started to level out and approach an asymptote as subtransects increased but the fall 2016 sample had a steeper slope and higher asymptote in comparison. Mean species richness estimates using the Chao equation ranged from 47 to 66.

Species accumulation curves comparing day with night BLE in the Alabama River were very similar between diel periods for each sampling event, with trends repeating both years from each season (Figure 7). The summer season graphs showed the night curves started with higher species richness and ascended at a faster rate than day curves. As effort increased, night curves

began to approach an asymptote while day curves crossed above them and ended at a steeper slope with a higher species count. Fall season graphs showed a consistent pattern of night curves starting with more species and maintaining a similar slope as day curves, with night curves always remaining above day curves.

Nine species of fish were collected only at night in the Alabama River; whereas, five were only caught during the day (Table 10). Usually these were caught in one or two sampling events in low numbers, and in many cases only a single specimen was collected. Ten species of fish had catch rates at night that were double those during the day, and three species had catch rates during the day that were double those at night (Table 10). Fish community structure collected by these two methods were most similar in summer 2015 and fall 2016 (Morisita index 0.81 and 0.77, respectively), but similarity was noticeably lower in the other two sample events (Table 11).

Results from night-time bank-line sampling on the Alabama River were also compiled separating habitat categories (Table 12). Mean species richness had little difference between the LHC category (36.9) and the HHC category (35.5). Overall mean Shannon diversity index was slightly higher in the LHC (2.47) than the HHC samples (2.23). Overall mean number of individuals per subtransect was 276 in the LHC and 254 in the HHC category. Mean species richness was 34.8 in summer and 37.7 in fall. Mean number of individuals per subtransect was also lower in summer (246) than in fall (286), although there was a large year-to-year difference in fall, with CPE's being much higher in 2016 than 2015. Mean Shannon diversity index was nearly identical between summer (2.36) and fall (2.34). Fish community structure between day and night samples across habitat types followed a similar pattern to that already described for the

overall samples, with higher similarity in summer 2015 and fall 2016 and lower in the other samples (Table 11). The sole exception was in HHC from fall of 2016.

Species accumulation curves combined across habitats and transects (i.e., 20 subtransects per sampling event) from night BLE in the Tallapoosa River showed higher species richness in summer compared to fall within each year but had very little difference among sampling events (Figure 8). All curves started to level out and approach an asymptote as subtransects increased but the summer 2015 sample had a steeper slope and higher asymptote in comparison. Mean species richness estimates from the Chao equation ranged from 29 to 68.

Species accumulation curves comparing day with night BLE in the Tallapoosa River were very similar between diel periods for each sampling event, with trends repeating both years in each season (Figure 9). The summer season graphs showed a consistent pattern of night curves that started with more species and maintained a similar slope as day curves, with night curves always remaining above day curves. In fall, day curves started at the same number of species as night curves but ascended at a faster rate and maintained a position above the night curves throughout the rest of sampling.

Five species of fish were collected only at night in the Tallapoosa River; whereas, eight were only caught during the day (Table 13). Usually these were caught from a few sampling events in low numbers, and in many cases only a single specimen was collected. Nine species of fish had catch rates at night that were double those during the day, and four species had catch rates during the day that were double those at night (Table 13). Fish community structure collected by these two methods were very similar, with Morisita index values all above 0.90 with the exception of fall 2015 which was 0.80 (Table 11), suggesting very little difference in the fish communities sampled between diel periods for each sampling event.

Results from night-time bank-line sampling on the Tallapoosa River were also compiled separating habitat categories (Table 14). Mean species richness had little difference between the LHC category (23.8) and the HHC category (25.5). Overall mean Shannon diversity index was slightly lower in the LHC (2.00) than the HHC samples (2.14). Overall mean number of individuals per subtransect was 98 in the LHC and 87 in the HHC categories. Mean species richness was 25.3 in summer and 23.9 in fall. Mean number of individuals per subtransect was also lower in summer (86) than in fall (98). Mean Shannon diversity index was similar between summer (2.12) and fall (2.02). Fish community structure between day and night samples across habitat types followed a similar pattern to that already described for the overall samples, with all Morisita index values above 0.80 (Table 11). The sole exception was in LHC from fall of 2015 with a value of 0.73.

In terms of the number of samples necessary to capture 50% of the number of species estimated with 95% confidence, the Alabama River required more samples from day sampling (4.0) than night (2.5) to reach the sampling objective ( $F_{1,15} = 12.00$ ,  $P = 0.0061$ ). However, regardless of diel period, number of samples were similar between seasons ( $F_{1,10} = 1.33$ ,  $P = 0.2751$ ) and habitat categories ( $F_{1,10} = 0.33$ ,  $P = 0.5765$ ). The Tallapoosa River had two different interactions; one between diel periods and season ( $F_{1,15} = 25.00$ ,  $P = 0.0005$ ), and the other between diel periods and habitat categories ( $F_{1,15} = 9.00$ ,  $P = 0.0133$ ), so the effects of diel periods was determined separately by season and habitat categories. More samples were required during the day than at night to reach the sampling objective in the summer (day 7.0, night 3.8;  $F_{1,7} = 11.79$ ;  $P = 0.0139$ ), but not during the fall (day 4.0, night 4.5;  $F_{1,7} = 1.0$ ;  $P = 0.3559$ ). Likewise, more samples were required to meet the sampling objective during the day than at

night in LHC (day 6.5, night 4.0,  $F_{1,7} = 3.95$ ,  $P = 0.0941$ ), but not in HHC (day 4.5, night 4.3,  $F_{1,7} = 0.13$ ,  $P = 0.7304$ ).

### **III.4. Point Sampling Method**

The Alabama River yielded a total of 2,325 individual fish of 45 different species in 18 families using 107.1 person-hours. In the Tallapoosa River, 1,112 fish of 30 unique species in 9 families were collected using 105.9 person hours. In the Choctawhatchee River, the PASE samples totaled 1,408 fish from 40 species in 15 families using 101.4 person hours. In the Sipsey River this method yielded 819 fish of 41 species in 12 families using 93.0 person-hours. A total pedal-down effort time of 6.7 h was used in each river. The majority of PASE samples did not meet the minimum 50% of the species richness estimate thresholds required to analyze the amount effort necessary to meet the objective, therefore, PASE samples were not able to be included in these analyses.

#### **III.4.1. Season Effects in PASE sampling**

Overall species accumulation curves across habitat types (i.e., 200 points per sampling event) in the Alabama River were fairly consistent and showed little variation between sampling events (Figure 10). There were no obvious trends between seasons or years. All of these curves had a steep slope that never approached an asymptote as the number of points increased. Mean species richness ranged only from 29 to 33 across samples. Species richness estimates using the Chao equation ranged from 35 to 91. Standard error of these species richness estimates were extremely high for summer 2015 and fall 2016.

The quantities of all species captured from Alabama River PASE sampling are listed by sampling event in Appendix 11 alongside the day BLE sampling results. Several species were not detected with point sampling that were sampled from BLE methods (Table 15), but all these

species were relatively rare. Only a single specimen of one species was captured with point sampling that was not detected with BLE methods (*Lepomis auritus*) which is not native to the Alabama River and may represent a new location for the species. Morisita's index comparing PASE samples to day BLE samples (Table 16) indicated that fish community structure was similar between methods in the summer samples (0.96 and 0.94). Fish community structure was less similar between years in the fall samples, with values of 0.78 and 0.70.

When separating samples by season and habitat, mean species richness in the Alabama River was 24.3 in summer and 23.3 in fall (Table 17). Mean catch per subsample was lower in summer (2.42) than in fall (3.40), although there was a large year-to-year difference in fall with CPE's being higher in 2015 than 2016. Mean Shannon diversity index was higher in summer (2.58) than in fall (2.26). On average, slightly fewer points collected no fish in summer (22.3%) than in fall (27.3%).

In the Tallapoosa River, overall species accumulation curves (i.e., 200 points per sampling event) were always higher for fall than summer (Figure 11). Observed species richness ranged from 16 to 24 from all sampling events, and estimated species richness from the Chao equation ranged from 24 to 26, which were approximately half that encountered from all methods combined (45). All of these curves had a steep slope that did not approach an asymptote as points increased (Figure 11).

The quantities of all species captured from Tallapoosa River PASE samples are listed by sampling event in Appendix 12 alongside the daytime BLE sampling results. Several species were not detected with point sampling that were sampled from BLE methods (Table 15), but all these species were relatively rare. Species that were caught only during point sampling were a single specimen of *Dorosoma cepedianum* and two *Ichthyomyzon gagei*. Morisita's similarity

index values comparing PASE samples to day BLE samples (Table 16) were all high (0.85 to 0.99), suggesting fish community structure sampled by these two methods was close to identical.

When separating samples by season and habitat, mean species richness in the Tallapoosa River was 11.6 in summer and 16.8 in fall (Table 18). Mean catch per point was lower in summer (0.96) than in fall (1.84). Mean Shannon diversity index was lower in summer (1.69) than in fall (1.90). On average, more points collected no fish in summer (56.0%) than in fall (32.5%).

Overall, species accumulation curves in the Choctawhatchee River (i.e., 200 points per sampling event) showed no discernable trends among seasons or years (Figure 12). All of these curves ended with a steep slope and did not level out or approach an asymptote as subsamples increased. Species richness ranged from 22 to 31 across all sampling events. Species richness estimates using the Chao equation ranged from 26 to 70. Standard error of these species richness estimates were extremely high for 2016 sampling events.

The quantities of all species captured from Choctawhatchee River during point sampling are listed by sampling event in Appendix 13 alongside the day BLE sampling results. Several species were not detected with PASE sampling that were sampled from BLE methods (Table 15), but all these species were relatively rare. Only a single specimen of one species was captured with point sampling that was not detected with BLE methods (*Alosa chrysochloris*). Morisita's similarity index values (Table 16) were all very high (0.92 to 1.00) comparing PASE samples to day BLE samples, suggesting similar communities were sampled with these two methods.

When separating samples by season and habitat, mean species richness in the Choctawhatchee River was 17.0 in summer and 21.4 in fall (Table 19). Mean catch per

subsample was lower in summer (1.56) than in fall (2.37). Mean Shannon diversity index was lower in summer (1.78) than in fall (2.15). On average, more points collected no fish in summer (48.5%) than in fall (36.5%).

Overall, species accumulation curves across habitat types (i.e., 200 points per sampling event) in the Sipsey River were steeper in fall than summer (Figure 13). All of these curves ended with a steep slope and did not level out or approach an asymptote as subsamples increased. Species richness in the fall was almost double that in summer, with 24 and 28 species observed in fall and 15 and 16 species observed in summer. Mean Chao species richness estimates was 21 in both summer samples with higher values of 38 and 40 in fall. Standard error for these estimates was highest in fall sampling events.

The quantities of all species captured from Sipsey River PASE samples are listed by sampling event in Appendix 14 alongside the day BLE sampling results. Several species were not captured during PASE sampling that were caught during the day BLE sampling (Table 15), but all these species were relatively rare. Species that were caught by PASE samples but not sampled during BLE sampling were *Campostoma oligolepis*, *Notemigonus crysoleucas*, *Notropis amplamala*, *Esox americanus*, and *Crystallaria asprella*. All of these species were only represented by a single specimen. Morisita's similarity index values comparing PASE samples to day BLE samples (Table 16) were all very high (0.96 to 0.99), suggesting that fish community structure sampled by these two methods was virtually identical.

When separating samples by season and habitat, mean species richness in the Sipsey River was 10.9 in summer and 19.0 in fall (Table 20). Mean catch per subsample was also much lower in summer (0.38) than in fall (1.67). Mean Shannon diversity index was similar in

summer (2.03) and fall (2.15). On average, more points collected no fish in summer (72.8%) than in fall (42.8%).

#### **III.4.2. Habitat Effects in PASE Sampling**

Mean species richness in the Alabama River PASE samples was lower in the LHC category (22.8) than the HHC (24.8), but the reverse was true for mean Shannon diversity index (Table 17). Mean catch per point (all species) was 1.0 fish/point lower in the LHC than HHC category. On average, more points collected no fish in the LHC (28.9%) than HHC category (19.7%).

Mean species richness in the Tallapoosa River PASE samples was lower in the LHC category (13.0) than the HHC category (15.4). Mean Shannon diversity index was also lower in the low habitat (1.72) than high habitat samples (1.86). Mean catch per point (all species) was 1.33 for low habitat and 1.45 for high habitat. On average, points that collected no fish were similar among habitat categories (Table 18).

Mean species richness in the Choctawhatchee River PASE samples was 18.6 in the LHC category and 19.8 in the HHC category. Mean Shannon diversity index was nearly identical from low habitat (1.96) than high habitat samples (1.97). Mean catch per point (all species) were 2.02 for low habitat and 1.90 for high habitat. On average, points that collected no fish were similar among habitat categories (Table 19).

Mean species richness in the Sipsey River PASE samples was higher in the LHC category (16.3) than the HHC category (13.6). Mean Shannon diversity index was also higher in low habitat (2.18) than high habitat samples (2.01). Following this same trend, mean catch per point (all species) was 1.27 for low habitat and 0.78 for high habitat. On average, more points collected no fish in the HHC (63.8%) than LHC category (51.8%; Table 20).

## **IV. DISCUSSION**

### **IV.1. PASE Sampling**

The PASE method has some advantages over the more commonly used BLE method. One advantage is exact location data, which coupled with side-scan data could be useful for a more detailed data set. Flow, depth, and other very site-specific parameters could be measured as well. This method is based on the statistical principle that many small samples are better than a few large samples (Copp 2010). Using PASE, many samples can be collected in a short amount of time compared to continuous bank-line sampling methods (Tomanova et al. 2013); for example, in my study 200 points were collected with a similar amount of on-water time as 10 BLE samples. This strategy of acquiring many samples is very useful for developing accumulation curves such as species or number of individuals. This method would have useful applications to specific habitat or a single species study and fish community studies in areas with low richness or where rare species are not a concern of the study. It has been shown to be an effective method for generating IBI type of fish community metric data with less effort and numbers of fishes as BLE methods (LaPointe et al. 2006; Tomanova et al. 2013)

However, the PASE method has some disadvantages over the BLE method. This method of sampling is essentially a small subset of the area that would be covered by a BLE sample, which could affect estimates of species richness and number of individuals, as demonstrated in my study. The fish community structure estimated in this study were very similar between the two methods according to the Morista's similarity index values, but, this index is resistant to undersampling, tends to overestimate similarity, and is highly sensitive to the most abundant species with the relatively rare species having little effect (Chao et al. 2006); therefore, it may not have captured all the differences between these sample methods. The PASE method

captured the more common species in similar proportional abundance to the BLE method when an adequate amount of samples were analyzed (>100 points). However, rarer species are often excluded from PASE samples compared to BLE samples from the same area (LaPointe et al. 2006; Tomanova et al. 2013). Thus estimations of species richness in my study were often significantly underestimated or had very high standard errors when compared to BLE samples from the same transect. If documenting the presence of rare species, maximizing number of species captured, or accurately estimating species richness in a diverse system are among the goals of a study, then sampling should be conducted using a BLE versus a PASE design.

One of the biggest limitations of PASE sampling is the high proportion of points with zero catches. Catch-per-effort distributions can often deviate from normal when zero catches are numerous, as may be the case when samples are short and fish abundance is low (Miranda et al. 1996). Thus, high proportion of points with zero catch must be accounted for when analyzing data. Zero catches in the PASE samples in my study ranged from 12.2% to 77.0% with a mean of 42.4%. The mean was much lower in the Alabama River (24.8%) than the other three rivers (42.5% to 57.8%), which could be due to higher densities of fish or lower water clarity. However, the Alabama River was sampled with a larger electrofishing boat than the other rivers, thus fish were subjected to a larger electrical field, resulting in each point effectively sampling a greater area. Any long-term monitoring program should standardize not only the amperage or power of the electrical field, but also the size of it by specifying boat size, length of booms, and distance between anodes used across rivers to reduce as much variability as possible for the inevitable equipment failure or future replacements (Bonar et al. 2009).

The PASE sampling technique originated in Europe in the 1970's and has since been developed and widely used for monitoring and sampling programs in some European countries

(Copp 2010). Tomanova et al. (2013) studied effects of different efforts (25, 50, 75, and 100 points) using the PASE method in French rivers on biotic indices and found 75 points to be adequate effort to provide a reproducible representation of fish community structure in most medium and large rivers that they sampled. The majority of the species accumulation curves in this study closely approached an asymptote at 100 points. In contrast, the species accumulation curves in Alabama rivers did not approach an asymptote at the highest effort level of 200 points. This difference was likely due to higher species richness and greater number of rare species present in southeastern United States rivers compared to French rivers (Kanno et al. 2009). Also mean catch per point was higher in the Tomanova et al (2013) study than my project (8.88 fish/point vs. 1.83 fish/point). Several possibilities could explain this difference, including higher fish densities, more shoaling fishes encountered, or methodology differences which targeted sampling locations more likely to hold fishes in their study. Though they didn't report specific numbers of fishless samples, they did place their rivers in two categories for data analysis; those with > 30% fishless points and < 30% fishless points, suggesting overall means from my rivers of 24% to 57% were likely similar.

Few studies employing the PASE method in the United States have been published. A recent study in the southeastern United States (Trumbo et al. 2016) compared PASE and BLE methods in a Louisiana river floodplain. A comparison of PASE species richness in their study sampled 86.5% of the number of species captured by the BLE method while my rivers captured a range of 72.0% to 83.3% from each river. Methods were slightly different between the studies, including a sampling effort of 60 s per point (vs. 30 s for my study). Both studies had an increased CPUE using the PASE method with Trumbo et al. (2016) averaging 3.75 times greater CPUE over the BLE method while my study averaged 1.34 times greater CPUE for all fish

combined. This was attributed to a decreased fright bias over continuous BLE methodology where fish might sense the electrical field approaching and escape capture before it overtakes them. Trumbo et al. (2016) discussed a reduced effort of 30 s or less per point as a more appropriate sampling time and my preliminary testing and the results of Tomanova et al. (2013) concluded this as well.

## **IV.2. Equipment failure**

Usually projects that evaluate sampling methods attempt to use the same gear throughout to minimize potential bias (Hughes et al. 2002). However, this project had to cope with numerous equipment failures that caused samples to be collected with multiple electrofishing arrays. Because each sample sequence required a large amount of time and manpower, timelines were limited and samples had to be collected with the best available gear. When a different boat was used, all sampling for that sampling event was conducted with that alternative boat.

All four rivers were affected by equipment failure. The large boat used for three of the four Alabama River samples was an older boat that ADCNR planned to decommission after this project. Thus it was not feasible to replace the steering system when it broke prior to the fall 2016 sample. The smaller boat used for sampling the other three rivers had a recurring problem with the outboard motor that rendered it inoperable for the first sampling event (summer 2015). This outboard was replaced for the remaining three sampling events.

The two replacement boats were of similar size to the original boats, but had smaller generators and lower peak output power. When using these alternative boats, all other standardizations of sampling were upheld, including the use of appropriate voltage to maintain five amps of electrofishing power. Though both original boats had higher peak watt generators, their power was never maxed out to maintain the standardized amperage, and the alternative

boats had sufficient power to meet this threshold. Though electrofishers connected to dissimilar electrode arrays cannot produce identical fields, even if waveform, voltage, and amperage are held constant (Miranda and Dolan 2003), one study (Henry et al. 2001) found that in-water voltage gradients was similar among 10 electrofishing boats in Alabama.

Comparing Morisita's similarity index among sampling events showed that all affected sampling event comparisons maintained a value above the highly similar benchmark of 0.60 (Haley and Johnston 2014) with most comparisons above 0.80. Morisita index values were often lower between unaffected sampling events than values comparing affected and unaffected events, indicating that the change in gears likely had little effect on results. Values of number of individuals, Shannon index, and species richness from affected sampling events were all within the range of unaffected sampling events for all rivers. Although using different equipment likely had some effect, there was no obvious evidence suggesting it was significant.

### **IV.3. Diel Effects**

The effects of diel periods on sampling results were different among the two rivers examined in this study. Flotemersch and Blocksom (2005) suggested that size of the river, specifically mean channel depth, determined the significance of diel effects on sampling. This appears to be true of my study as well. The larger Alabama River required fewer samples at night to achieve sampling objectives regardless of season and habitat sampled. In contrast, the smaller Tallapoosa River had mixed results depending on season and habitat complexity.

There are three main movement patterns that can account for changes in fish assemblage structure over the diel cycle: vertical migrations between the river bottom and surface, lateral migrations between the bank and the channel, and longitudinal migrations into or out of the study area (Baumgartner et al. 2008). The nature and extent of these migrations can be species-

specific and vary with respect to season (Wolter and Freyhof 2004), which can make it difficult to predict their effects on sample designs aimed at collecting a wide range of fish species.

Vertical and lateral migrations towards the shore would make fishes more vulnerable to BLE sampling as it is most effective at shallower depths and conducted along the bank. The Alabama River is larger, deeper, and has a more diverse fish community than the Tallapoosa River (Boschung and Mayden 2004), which may offer more opportunities for all three types of diel migrations to occur (Haley and Johnston 2014). The more homogenous substrate of the Alabama River might also make lateral diel movements a more typical pattern for many species than a river such as the Tallapoosa River that is shallower with more complex habitats to travel between. This could explain why the LHC category in the Tallapoosa River required fewer samples to reach the objective at night but the HHC category did not. The Tallapoosa River had the highest substrate complexity of the four study rivers. The HHC category in the Tallapoosa River contained subtransects that had more rock substrates, including shoal areas, with associated shallower depths and often faster flow compared to the LHC category. This could restrict daily longitudinal movements for many species and reduce the possibility for vertical and lateral movement into areas of increased vulnerability to this gear.

Seasonal differences in the Tallaposa River showed that summer required more samples during the day to meet objectives, whereas, fall showed no difference. Seasonal variation was possibly driven by lateral movements, mainly in LHC habitats. Some temperate species may seek refuge from the low flow, warmer portions near the banks during the day in the summer and move into these areas only at night, but lower water temperatures in the fall may not cause fish to leave these areas during the day. Copp (2010) found that fish move into shore at night during the

summer of their first year of life, but in other seasons, the patterns of diel migration may be more complex.

Several studies have noted an increase in species richness and numbers of fish sampled when collections take place at night (Lyons et al. 2001; Wolter and Freyhof 2004; Flotemersch and Blocksom 2005; Baumgartner et al. 2008; Copp 2010). Catch rate of all species combined was higher in night samples than day samples in my study with one exception during a fall season from each river. Species richness in the Alabama River was higher in night samples in the fall but not in the summer. Sampling objectives were reached with less effort at night in the Alabama River regardless of season and habitat complexity due to higher number of species collected per subtransect at night, even though total species accumulated by the end of the samples were not always higher. Night samples always began at a higher number of species, ascended at a faster rate in the beginning, but summer curves then tended to more closely reach an asymptote toward the end. If the sampling objective was modified to evaluate a higher percentage of the species estimate, then summer estimates would eventually require less effort to reach the objective during day BLE sampling in the Alabama River. In other words, night and day species accumulation curves from summer eventually crossed as effort approached the peak. Samples in the Tallapoosa River consistently had higher species richness at night during summer and lower species richness at night during fall. Species accumulation curves were more similar in shape among samples than the Alabama River therefore, interpretation of results were more straightforward.

My results indicated that sampling these two rivers at night would in most cases require less effort to reach reasonable sampling objectives. However this reduced effort comes with added difficulties and safety concerns associated with night sampling. Electrofishing at night

causes personnel concerns due to added fatigue and may be fiscally unreasonable (Flotemersch and Blocksom 2005). Additional safety concerns arise from limited visibility, particularly in in reaches with numerous obstructions or fast, turbulent water (Lyons et al. 2001). However, modern LED lighting has improved in recent years, to the point of nearly eliminating visibility concerns at night, with very low power consumption.

#### **IV.4. Habitat Effects**

The importance of habitat when sampling fish communities has been emphasized in many studies, particularly within wadeable streams (Karr 1981; Simonson et al 1994; Peterson and Rabeni 1995; Simonson and Lyons 1995; Freeman et al. 1997; Bayley and Peterson 2001; Pearson et al. 2011; Zorn et al. 2011; Stanfield et al 2013). Different habitat complexities had been found to host a different suite of species and rare species may only be found with adequate effort within their habitat preference. However, habitat complexity did not appear to be an important factor determining species accumulation in three of the four rivers in my study. Habitat complexity was only a factor in the Tallapoosa River which was the most heterogeneous river of the four in terms of bottom substrate. Thus habitat complexity may be more important in rivers characterized by greater amount of rocky substrate and shoals than in sandy coastal plain rivers.

Habitat complexity may have not been adequately quantified in my study due to the large scale over which sampling was conducted. Transects were 100 MWCW long, broken into 10 subtransects of equal length following protocol established in previous studies (i.e., Hughes et al. 2002). This allowed enough samples to build a species accumulation curve and for them to be of equal length to compare to each other. Since each subtransect was 1/10 of a transect long, they ranged from 0.4 to 1.0 km in length across study rivers. Long sample reaches may mask small

scale habitat variability that may be of interest to resource managers (Flotemersch et al. 2006). In many cases, these lengths may have been too long to encapsulate just a high or low habitat category. Habitat assessments in the stream studies mentioned above are generally more descriptive of characteristics and only described effort as being of adequate distance to cover these different areas. Sampling more subtransects of shorter length and higher frequency, such as 5 MWCW or 1/20 of a transect may have allowed a better resolution of habitat complexity at a finer scale.

Finally, habitat complexity in this study was based solely on substrate. Although this is an important factor of habitat complexity, there are many more aspects to habitat such as depth, flow, submerged or emergent vegetation, woody debris, and many other elements that would influence species composition within an area (Simonson et al. 1994). The large scale of this project made finer-scale measurements of these variables impractical, but omission of these may also have contributed to the lack of habitat effects on sampling efficiency observed in this study. Future studies examining these relationships in non-wadeable rivers should attempt to sample habitat and fish at finer scales to better evaluate relations between the two. However, manpower and resource demands of sampling at this scale on rivers of this size examined in this study may be hard to meet for most agencies.

#### **IV.5. Seasonal Effects**

Fall was the most efficient season to sample for three of the four rivers surveyed. The species accumulation curves in these rivers showed a trend of steeper and higher curves in fall compared to summer. Juveniles of many species often congregate in the shallow areas of rivers near the bank (Peterson and Rabeni 1995). Due to their small size during summer, these fish likely do not fully recruit to the gear used until the fall (Bonar et al. 2009). Migrations from the

study area would have more impact on species compositions in summer, because most seasonal fish migrations in North America take place in spring and summer and therefore species composition would be more stable in fall.

Unlike the other rivers, the Alabama River had similar effort requirements to meet the sampling objective for both seasons sampled. A study in the Alabama River that collected species exclusively on gravel and sand bars (Haley and Johnston 2014), had Morisita values comparing summer and fall on fish community estimates ranging from 0.03 to 1.00 across sampling sites. A single species that often dominated the catch had a very significant impact on these numbers and values were given both with and without this species. Fish assemblages were found to vary seasonally and some species were detected in greater numbers during fall samples, which were attributed to lower water levels. A review of species captured in my study from each sampling event showed a similar amount of species that were collected exclusively in either season. The high number of species present and complicated dynamics of each species seasonal habitat preferences may vary their vulnerability to boat electrofishing and shift the community structure sampled. Overall this shift did not seem to affect numbers of species collected, therefore did not affect the efficiency of effort necessary to meet the sampling objective seasonally.

A study from the Tallapoosa River on optimizing boat electrofishing sampling on centrarchids (Sammons 2015) found some seasonal differences, though results were mixed for each species in the study. Overall, fall was recommended as the most efficient season among the three examined (spring, summer, and fall) from this study. Results were attributed mainly to water levels in fall from this region, which typically are lowest during this season and result in better efficiency of this gear. My study indicated that fall was a more efficient season than

summer to meet the sampling objective in three of the four study rivers. Effects of migrations are minimized during fall and typically water levels are lower during this season, which has been correlated to increased vulnerability to this gear. Therefore, fall was the most efficient season to meet the sampling objective overall for non-wadeable rivers in Alabama.

## **V. Conclusions and Management Implications**

Alabama's diversity of fishes is higher than that of any other political unit in North America and one of the largest faunas for any comparable area of the temperate world (Boschung and Mayden 2004). However, most non-wadeable rivers in the state have never been monitored for biodiversity (S. Rider, personal communication). Long-term monitoring of these high diversity rivers should focus on collecting as many species as fiscally reasonable instead of a lower standard of sampling the minimum species to obtain a biotic integrity score. Focusing on maximizing species collected will supply the necessary data to calculate a biotic score or index once one is developed for Alabama's non-wadeable rivers, while also providing quality data on rare species, including Alabama endemics and species of concern.

The heterogeneous shallow Tallapoosa River had limited additional data and benefits from night sampling. This coupled with the numerous underwater hazards, fast flows, and hard to navigate shoal areas, makes night sampling an inadvisable option. However, the homogenous, deep Alabama River had greater effects of night sampling efficiency and was much safer to navigate. Night sampling on the Alabama River would add valuable data to a long-term monitoring program and should be considered, perhaps in addition to day sampling on a less frequent schedule. This is in agreement with conclusions of Flotemersch and Blocksom (2005) that at depths greater than 4 m, the diel movements of fish significantly impact the quality of results to the extent that the consideration of night electrofishing is justified.

The rivers chosen for this study spanned the widest geographical area, size, and geomorphology possible to allow the widest application of results across the state. Though each river was analyzed separately, there were some clear indications of overall guidelines for a general monitoring program for non-wadeable rivers in Alabama. The PASE method was not suitable for use in a program which places value on collecting a sufficient percentage of species with such high richness as Alabama's rivers contain. The BLE method consistently met sampling objectives posed and results by season are listed in Table 21. The fall season proved to be a better time for sampling than summer. Habitat complexity generally did not affect sampling efficiency in coastal plain rivers but should be examined further in more structurally heterogeneous rivers common to the Piedmont and other upland physiographic provinces. Nine subtransects of sampling (i.e., 90 MWCW or a maximum of 9 km) would collect 75% of the species estimated with a probability between 50% and 95% and could be completed with a single day of sampling. The following is a breakdown of the findings for each river based on my results.

1) **Alabama River** – The largest and most specious of the study rivers, the fish community is influenced by euryhaline species because of its close proximity and connectivity to the gulf (Haley and Johnston 2014). Despite its large size and diverse fish community, the amount of effort necessary to meet sampling objectives was not affected by season or habitat. The subtransect size in this river was 1 km. At 95% confidence, four subtransects (i.e., 4 km) were needed to capture 50% of the species estimated during the day and 12 subtransects (12 km) were necessary to capture 75% of these species. Night sampling was more efficient and added several species; night sampling with 95% confidence required 2.5 subtransects (2.5 km) to capture 50% of the species estimated

and 9.5 subtransects to collect 75% of these species. There is minimal additional risk from navigational hazards due to the deep open nature of this section; thus night sampling should be considered.

2) **Tallapoosa River** – A typical Piedmont stream, the Tallapoosa River is more heterogeneous than the other rivers, with a smaller percentage of sand and more rocky areas including shoal complexes. This section of the river also has a daily influence of flow alterations from controlled dam releases and has been the subject of numerous studies on the effects of flow alteration on the fish community (e.g., Travnicek and Meceina 1994; Irwin and Freeman 2002). Habitat complexity was a significant factor in the amount of effort required to meet the sampling objective although all habitat types should be sampled to more accurately represent the fish community. Sampling in fall was more efficient than summer, and at 95% confidence required four subtransects (i.e., 3.12 km) to capture 50% of the species estimated and 13 subtransects (9.36 km) to capture 75% of the species estimate during the day. Night sampling was not more efficient than day sampling, and with the many navigational hazards present in the river, is not recommended.

3) **Choctawhatchee River** – A smaller coastal plain river, the fish community in the Choctawhatchee River is influenced by euryhaline species due to its close proximity and connectivity to the gulf. Like the Alabama River, habitat complexity did not affect the amount of effort required to meet the sampling objective, but fall was the most efficient season to sample over summer. At 95% confidence, fall sampling required three subtransects (i.e., 1.74 km) of effort to capture 50% of the species estimated and nine subtransects (5.22 km) to capture 75% of these species.

4) **Sipsey River** – The smallest study river, the Sipsey River had similar characteristics to the Choctawhatchee, though without the presence of euryhaline species. Like the Choctawhatchee, habitat complexity did not affect the amount of effort required to meet the sampling objective, but fall was the most efficient season to sample. At 95% confidence, fall sampling with 95% confidence required 4.5 subtransects (i.e., 1.8 km) of effort to capture 50% of the species estimated and 11 subtransects (4.4 km) to capture 75% of these species.

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## VII. TABLES

Table 1. Side-scan sonar total distance scanned, upper and lower points scanned in decimal degrees, mean wetted channel width (MWCW), and resulting subtransect length for each river. Note that subtransect length was maximum length of 1.00 km for Alabama River.

River	Distance (km)	Upper		Lower		MWCW (m)	Subtransect Length (km)
		Lat	Long	Lat	Long		
Alabama	39.8	31.552	-87.515	31.404	-87.698	200	1.00
Tallapoosa	12.3	33.017	-85.611	32.957	-85.691	78	0.78
Choctawhatchee	19.3	31.096	-85.839	30.998	-85.830	58	0.58
Sipsey	11.0	33.045	-88.082	33.037	-88.145	40	0.40

Table 2. Classification scheme and associated definitions for substrate mapping.

Substrate Class	Definition
Sand	> 75% of area composed of particles < 2 mm diameter (sand silt, clay or fine organic detritus).
Gravel	>25% of area composed of rocks > 2 mm, but < 500 mm diameter across the longest axis.
Boulder	Rock or groups of rocks > 500 mm across the longest axis within 1.5 m of next boulder.
Bedrock	> 75% of area composed of bedrock as upper layer of substrate.

Table 3. Percent of each substrate present within study area of each river from sonar survey interpretations.

Substrate	Alabama	Tallapoosa	Choctawhatchee	Sipsey
Sand	90.40	72.56	88.67	86.60
Gravel	0.12	7.72	0.48	1.22
Boulder	3.35	5.31	1.64	1.05
Bedrock	6.13	14.40	9.20	11.13

Table 4. Morisita's similarity index comparing seasons within a year and comparing years for each season using the day BLE method.

River	Summer-Fall 2015	Summer-Fall 2016	2015 - 2016 Summer	2015-2016 Fall
Alabama	0.57	0.71	0.93	0.87
Tallapoosa	0.81	0.92	0.90	0.96
Choctawhatchee	0.88	0.63	0.67	0.98
Sipsey	0.83	0.75	0.75	0.84

Table 5. Day BLE sampling subtransects (N), species richness (S<sub>o</sub>), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Alabama River. Comparable values were also averaged by season and habitat.

Season	Habitat	N	S <sub>o</sub>	S <sub>o</sub> @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>	
Summer	2015	Low	11	35	32.7	1058	96	2.78
		High	9	27	27.0	1033	115	2.54
	2016	Low	11	37	35.4	1116	101	2.81
		High	9	42	42.0	1004	112	2.81
	Mean				34.3		105	2.73
Fall	2015	Low	11	32	30.9	2816	256	2.49
		High	9	33	33.0	1905	212	2.56
	2016	Low	11	43	40.4	1601	146	2.33
		High	9	40	40.0	1043	116	2.53
	Mean				36.1		184	2.48
Overall	Mean	Low			34.9		150	2.60
		High			35.5		138	2.61
		Total			35.2		145	2.61

Table 6. Day BLE sampling subtransects (N), species richness (S<sub>o</sub>), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Tallapoosa River. Comparable values were also averaged by season and habitat.

Season	Habitat	N	S <sub>o</sub>	S <sub>o</sub> @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>	
Summer	2015	Low	9	19	19.0	583	65	1.93
		High	11	27	26.2	682	62	2.31
	2016	Low	9	19	19.0	326	36	1.84
		High	11	23	22.1	419	38	2.07
	Mean				21.6		50	2.04
Fall	2015	Low	9	24	24.0	944	105	2.24
		High	11	29	27.5	1307	119	2.25
	2016	Low	9	28	28.0	835	93	2.13
		High	11	27	25.9	692	63	2.28
	Mean				26.4		94	2.22
Overall	Mean	Low			22.5		75	2.03
		High			25.4		70	2.23
	Total				24.0		72	2.13

Table 7. Day BLE sampling subtransects (N), species richness (S<sub>0</sub>), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Choctawhatchee River. Comparable values were also averaged by season and habitat.

Season		Habitat	N	S <sub>0</sub>	S <sub>0</sub> @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>
Summer	2015	Low	11	26	24.9	405	37	2.70
		High	9	28	28.0	238	26	2.68
	2016	Low	11	30	28.9	714	65	1.68
		High	9	35	35.0	572	64	1.66
	Mean				29.2		48	2.18
Fall	2015	Low	11	38	36.5	1152	105	2.69
		High	9	38	38.0	1072	119	2.57
	2016	Low	11	31	30.2	672	61	2.58
		High	9	30	30.0	498	55	2.60
	Mean				33.7		85	2.61
Overall	Mean	Low			30.1		67	2.41
		High			32.8		66	2.38
		Total			31.4		67	2.40

Table 8. Day BLE sampling subtransects (N), species richness (So), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Sipse River. Comparable values were also averaged by season and habitat.

Season	Habitat	N	So	So @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>	
Summer	2015	Low	10	24	23.1	306	31	2.39
		High	10	27	25.8	274	27	2.17
	2016	Low	10	31	29.7	262	26	2.50
		High	10	29	28.0	297	30	2.41
	Mean			27.8	26.7	284.75	28	2.37
Fall	2015	Low	10	36	35.1	899	90	2.58
		High	10	38	36.6	580	58	2.40
	2016	Low	10	32	31.1	763	76	2.48
		High	10	32	31.2	639	64	2.36
	Mean			34.5	33.5	720.25	72	2.46
Overall	Mean	Low		30.8	29.8	557.5	56	2.49
		High		31.5	30.4	447.5	45	2.34
	Total			31.1	30.1	502.5	54	2.41

Table 9. Morisita's similarity index comparing high and low habitat heterogeneity samples using the day BLE method.

River	Summer 2015	Fall 2015	Summer 2016	Fall 2016
Alabama	0.88	0.86	0.90	0.91
Tallapoosa	0.96	0.99	0.99	0.94
Choctawhatchee	0.95	0.98	0.99	0.97
Sipsey	0.78	0.93	0.92	0.97

Table 10. Species collected with the BLE method in the Alabama River exclusively during a single diel period and species which had catch rates which more than doubled between diel periods. Catch rate lists excludes rare species.

Species only collected		Catch rate	
Night	Day	> Double at Night	> Double at Day
<i>Menidia beryllina</i>	<i>Aphredoderus sayanus</i>	<i>Moxostoma poecilurum</i>	<i>Pomoxis nigromaculatus</i>
<i>Lepomis humilis</i>	<i>Lepomis cyanellus</i>	<i>Lepomis macrochirus</i>	<i>Cyprinella venusta</i>
<i>Pomoxis annularis</i>	<i>Campostoma oligolepis</i>	<i>Micropterus henshalli</i>	<i>Cyprinus carpio</i>
<i>Brevoortia patronis</i>	<i>Percina nigrofasciata</i>	<i>Macrhybopsis storeiana</i>	
<i>Notemigonus crysoleucas</i>	<i>Gambusia affinis</i>	<i>Notropis candidus</i>	
<i>Morone chrysops</i>		<i>Notropis edwardraneyi</i>	
<i>Percina kathae</i>		<i>Pimephales vigilax</i>	
<i>Percina shumardi</i>		<i>Anchoa mitchilli</i>	
		<i>Lepisosteus osseus</i>	
		<i>Polydon spathula</i>	
		<i>Ictalurus furcatus</i>	
		<i>Ictalurus punctatus</i>	
		<i>Pylodictis olivaris</i>	

Table 11. Morisita's similarity index comparing night BLE samples to day BLE samples.

River	Samples	Summer 15	Fall 15	Summer 16	Fall 16
Alabama	All	0.81	0.65	0.63	0.77
	High	0.85	0.67	0.63	0.69
	Low	0.72	0.65	0.57	0.83
Tallapoosa	All	0.91	0.80	0.91	0.94
	High	0.89	0.86	0.90	0.95
	Low	0.92	0.73	0.92	0.90

Table 12. Night BLE sampling subtransects (N), species richness (S<sub>o</sub>), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Alabama River.

Comparable values were also averaged by season and habitat.

Season		Habitat	N	S <sub>o</sub>	S <sub>o</sub> @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>
Summer	2015	Low	11	34	32.7	2886	262	2.53
		High	9	30	30.0	1935	215	2.35
	2016	Low	11	41	39.3	2766	251	2.45
		High	9	37	37.0	2242	249	2.12
	Mean				34.8		246	2.36
Fall	2015	Low	11	33	31.9	1697	154	2.65
		High	9	35	35.0	1589	177	2.37
	2016	Low	11	46	43.7	4777	434	2.24
		High	9	40	40.0	3368	374	2.09
	Mean				37.7		286	2.34
Overall	Mean	Low			36.9		276	2.47
		High			35.5		254	2.23
		Total			36.2		266	2.35

Table 13. Species collected with the BLE method in the Tallapoosa River exclusively during a single diel period and species which had catch rates which more than doubled between diel periods. Catch rate lists excludes rare species.

Species only collected		Catch rate	
Night	Day	> Double at Night	> Double at Day
<i>Dorosoma cepedianum</i>	<i>Campostoma oligolepis</i>	<i>Ambloplites ariommus</i> ,	<i>Hypentelium etowanum</i>
<i>Notemigonus crysoleucas</i>	<i>Cyprinella gibbsi</i>	<i>Lepomis auritus</i>	<i>Moxostoma poecilurum</i>
<i>Ictalurus furcatus</i>	<i>Notropis baileyi</i>	<i>Lepomis gulosus</i>	<i>Pomoxis nigromaculatus</i>
<i>Marone saxatilis</i>	<i>Fundulus bifax</i>	<i>Lepomis macrochirus</i>	<i>Fundulus olivaceus</i>
<i>Ichthyomyzon gagei</i>	<i>Morone chrysops</i>	<i>Lepomis microlophus</i>	
		<i>Micropterus tallapoosae</i>	
		<i>Cyprinus carpio</i>	
		<i>Ictalurus punctatus</i>	
		<i>Pylodictis olivaris</i>	

Table 14. Night BLE sampling subtransects (N), species richness (So), species richness at highest comparable sampling effort (N=9) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=9) of each reach and sampling event for the Tallapoosa River. Comparable values were also averaged by season and habitat.

Season		Habitat	N	So	So @N <sub>9</sub>	# Individuals	CPE (fish/sample)	H' @N <sub>9</sub>
Summer	2015	Low	9	25	25.0	735	82	2.15
		High	11	29	27.6	880	80	2.25
	2016	Low	9	23	23.0	862	96	2.04
		High	11	27	25.7	975	89	2.02
	Mean				25.3		86	2.12
Fall	2015	Low	9	23	23.0	1199	133	1.84
		High	11	24	23.4	1233	112	2.17
	2016	Low	9	24	24.0	734	82	1.96
		High	11	26	25.1	749	68	2.12
	Mean				23.9		98	2.02
Overall	Mean	Low			23.8		98	
		High			25.5		87	
		Total			24.6		92	

Table 15. Species collected from BLE samples that were not collected by the PASE method by river.

Alabama	Tallapoosa	Choctawhatchee	Sipsey
<i>Aphredoderus sayanus</i>	<i>Minytrema melanops</i>	<i>Acipenser oxyrhynchus</i>	<i>Amia calva</i>
<i>Menidia beryllina</i>	<i>Pomoxis annularis</i>	<i>Strongylura marina</i>	<i>Anguilla rostrata</i>
<i>Moxostoma duquesnei</i>	<i>Campostoma oligolepis</i>	<i>Dorosoma cepedianum</i>	<i>Strongylura marina</i>
<i>Ambloplites ariommus</i>	<i>Ctenopharyngodon idella</i>	<i>Ctenopharyngodon idella</i>	<i>Carpionodes velifer</i>
<i>Lepomis cyanellus</i>	<i>Cyprinella gibbsi</i>	<i>Notropis maculatus</i>	<i>Erimyzon tenuis</i>
<i>Lepomis humilis</i>	<i>Notemigonus crysoleucas</i>	<i>Esox americanus</i>	<i>Ictiobus bubalus</i>
<i>Pomoxis annularis</i>	<i>Notropis baileyi</i>	<i>Esox niger</i>	<i>Minytrema melanops</i>
<i>Alosa chrysochloris</i>	<i>Notropis texanus</i>	<i>Ictalurus furcatus</i>	<i>Moxostoma carinatum</i>
<i>Brevoortia patronus</i>	<i>Fundulus bifax</i>	<i>Etheostoma davisoni</i>	<i>Ambloplites ariommus</i>
<i>Campostoma oligolepis</i>	<i>Ictalurus furcatus</i>	<i>Etheostoma swaini</i>	<i>Pomoxis annularis</i>
<i>Hiodon tergisus</i>	<i>Morone chrysops</i>		<i>Dorosoma petenense</i>
<i>Ameiurus natalis</i>	<i>Morone saxatilis</i>		<i>Notropis ammophilus</i>
<i>Lepisosteus spatula</i>	<i>Etheostoma chuckwachatte</i>		<i>Lepisosteus osseus</i>
<i>Morone chrysops</i>	<i>Perca flavescens</i>		<i>Morone chrysops</i>
<i>Crystallaria asprella</i>	<i>Gambusia affinis</i>		<i>Ammocrypta meridiana</i>
<i>Percina kathae</i>			<i>Etheostoma artesia</i>
<i>Percina nigrofasciata</i>			<i>Etheostoma histrio</i>
<i>Percina shumardi</i>			<i>Percina suttkusi</i>
<i>Polyodon spathula</i>			<i>Ichthyomyzon gagei</i>

Table 16. Morisita's similarity index comparing PASE samples to day BLE samples.

River	Samples	Summer 15	Fall 15	Summer 16	Fall 16
Alabama	All	0.96	0.78	0.94	0.70
	High	0.96	0.50	0.96	0.78
	Low	0.91	0.86	0.92	0.59
Tallapoosa	All	0.99	0.93	0.95	0.85
	High	0.99	0.92	0.96	0.86
	Low	0.96	0.94	0.82	0.83
Choctawhatchee	All	0.93	0.94	1.00	0.92
	High	0.92	0.97	1.00	0.94
	Low	0.95	0.90	0.98	0.91
Sipsey	All	0.96	0.97	0.99	0.98
	High	1.00	0.92	0.90	0.99
	Low	0.84	0.96	1.00	0.96

Table 17. Number of PASE subsamples (N), percentage of points with no catch, species richness (S<sub>o</sub>), species richness at highest comparable sampling effort (N=90) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=90) of each reach and sampling event for the Alabama River. Comparable values were also averaged by season and habitat.

Season	Habitat	N	% Points w/no fish	S <sub>o</sub>	S <sub>o</sub> @N <sub>90</sub>	# Individuals	CPE (fish/point)	H' @N <sub>90</sub>		
Summer	2015	Low	110	22.7	23	21.9	246	2.24	2.58	
		High	90	12.2	25	25.0	275	3.06	2.49	
	2016	Low	110	28.2	28	26.2	252	2.29	2.63	
		High	90	24.4	24	24.0	194	2.16	2.61	
	Mean			22.3		24.3		2.42	2.58	
Fall	2015	Low	110	25.5	22	20.8	408	3.71	2.26	
		High	90	15.6	25	25.0	614	6.82	1.85	
	2016	Low	110	39.1	24	22.3	175	1.59	2.44	
		High	90	26.7	25	25.0	161	1.79	2.50	
	Mean			27.3		23.3		3.40	2.26	
Overall	Low			28.9		22.8		2.46	2.48	
	Mean	High			19.7		24.8		3.46	2.36
	Total			24.8		23.8		2.91	2.42	

Table 18. Number of PASE subsamples (N), percentage of points with no catch, species richness ( $S_o$ ), species richness at highest comparable sampling effort (N=90) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=90) of each reach and sampling event for the Tallapoosa River. Comparable values were also averaged by season and habitat.

Season		Habitat	N	% Points w/no fish	$S_o$	$S_o$ @N <sub>90</sub>	# Individuals	CPE (fish/point)	H' @N <sub>90</sub>
Summer	2015	Low	90	62.2	9	9.0	51	0.57	1.58
		High	110	50.0	17	15.4	136	1.24	2.08
	2016	Low	90	58.9	10	10.0	89	0.99	1.54
		High	110	54.5	13	11.8	106	0.96	1.56
	Mean			56.0		11.6		0.96	1.69
Fall	2015	Low	90	30.0	17	17.0	169	1.88	2.20
		High	110	24.5	21	19.8	253	2.30	2.15
	2016	Low	90	30.0	16	16.0	171	1.90	1.57
		High	110	44.5	16	14.5	141	1.28	1.66
	Mean			32.5		16.8		1.84	1.90
Overall		Low		45.3		13.0		1.33	1.72
	Mean	High		43.4		15.4		1.45	1.86
		Total		44.3		13.9		1.40	1.79

Table 19. Number of PASE subsamples (N), percentage of points with no catch, species richness (S<sub>o</sub>), species richness at highest comparable sampling effort (N=90) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=90) of each reach and sampling event for the Choctawhatchee River. Comparable values were also averaged by season and habitat.

Season		Habitat	N	% Points w/no fish	S <sub>o</sub>	S <sub>o</sub> @N <sub>90</sub>	# Individuals	CPE (fish/point)	H' @N <sub>90</sub>
Summer	2015	Low	110	64.5	21	19.2	85	0.77	2.49
		High	90	72.2	12	12.0	38	0.42	2.09
	2016	Low	110	30.9	16	14.8	282	2.56	1.18
		High	90	26.7	22	22.0	218	2.42	1.38
	Mean			48.5		17.0		1.56	1.78
Fall	2015	Low	110	30.0	26	24.2	359	3.26	2.17
		High	90	32.2	26	26.0	238	2.64	2.31
	2016	Low	110	46.4	18	16.3	162	1.47	2.00
		High	90	36.7	19	19.0	190	2.11	2.11
	Mean			36.5		21.4		2.37	2.15
Overall		Low		43.0		18.6		2.02	1.96
	Mean	High		41.9		19.8		1.90	1.97
		Total		42.5		19.2		1.97	1.97

Table 20. Number of PASE subsamples (N), percentage of points with no catch, species richness (So), species richness at highest comparable sampling effort (N=90) calculated from the species accumulation curves, number of individuals captured, catch per effort, and Shannon Index at highest comparable sampling effort (N=90) of each reach and sampling event for the Sipse River. Comparable values were also averaged by season and habitat.

Season		Habitat	N	% Points w/no fish	S <sub>o</sub>	So @N <sub>90</sub>	# Individuals	CPE (fish/point)	H' @N <sub>90</sub>
Summer	2015	Low	100	68.0	12	11.3	46	0.46	2.01
		High	100	74.0	12	11.4	39	0.39	2.09
	2016	Low	100	72.0	13	12.3	34	0.34	2.22
		High	100	77.0	9	8.6	31	0.31	1.81
	Mean			72.8	11.5	10.9		0.38	2.03
Fall	2015	Low	100	30.0	19	18.4	224	2.24	2.19
		High	100	51.0	16	15.5	121	1.21	2.18
	2016	Low	100	37.0	24	23.1	203	2.03	2.30
		High	100	53.0	20	18.9	121	1.21	1.94
	Mean			42.8	19.8	19.0		1.67	2.15
Overall		Low		51.8	17.0	16.3		1.27	2.18
	Mean	High		63.8	14.3	13.6		0.78	2.01
		Total		57.8	15.6	14.9		1.02	2.09

Table 21. Number of samples needed to reach each sampling objective shown by season using the BLE method. Species estimates ( $S_{est}$ ) that the sampling objectives are based on are listed for each river. Means are listed for each season with standard errors in parenthesis.

Diel Period	River	$S_{est}$	2015		2016		Mean	
			Summer	Fall	Summer	Fall	Summer	Fall
Samples to accumulate 50% of species estimate with 95% probability								
Day	Alabama	45.9	5	4	3	4	4 (1.0)	4 (0.0)
	Tallapoosa	33.6	4	4	8	4	6 (2.0)	4 (0.0)
	Choctawhatchee	40.2	7	2	5	4	6 (1.0)	3 (1.0)
	Sipsey	42.4	11	4	8	5	9.5 (1.5)	4.5 (0.5)
Night	Alabama	45.9	3	3	2	2	2.5 (0.5)	2.5 (0.5)
	Tallapoosa	33.6	6	5	4	5	5 (1.0)	5 (0.0)
Samples to accumulate 75% of species estimate with 50% probability								
Day	Alabama	45.9	14	11	6	7	10 (4.0)	9 (2.0)
	Tallapoosa	33.6	8	9	18	8	13 (5.0)	8.5 (0.5)
	Choctawhatchee	40.2	17	4	8	9	12.5 (4.5)	6.5 (2.5)
	Sipsey	42.4	18	7	13	9	15.5 (2.5)	8 (1.0)
Night	Alabama	45.9	16	9	5	4	10.5 (5.5)	6.5 (2.5)
	Tallapoosa	33.6	11	15	10	13	10.5 (0.5)	14 (1.0)
Samples to accumulate 75% of species estimate with 95% probability								
Day	Alabama	45.9	18	15	9	9	13.5 (4.5)	12 (3.0)
	Tallapoosa	33.6	12	13	20	13	16 (4.0)	13 (0.0)
	Choctawhatchee	40.2	20	6	11	12	15.5 (4.5)	9 (3.0)
	Sipsey	42.4	20	9	16	13	18 (2.0)	11 (2.0)
Night	Alabama	45.9	19	12	8	7	13.5 (5.5)	9.5 (2.5)
	Tallapoosa	33.6	18	19	16	18	17 (1.0)	18.5 (0.5)

## VIII. FIGURES



Figure 1. Study area showing site locations for each river. Upper bounds of sampling areas are indicated with a green line and lower bounds are shown with a red line.

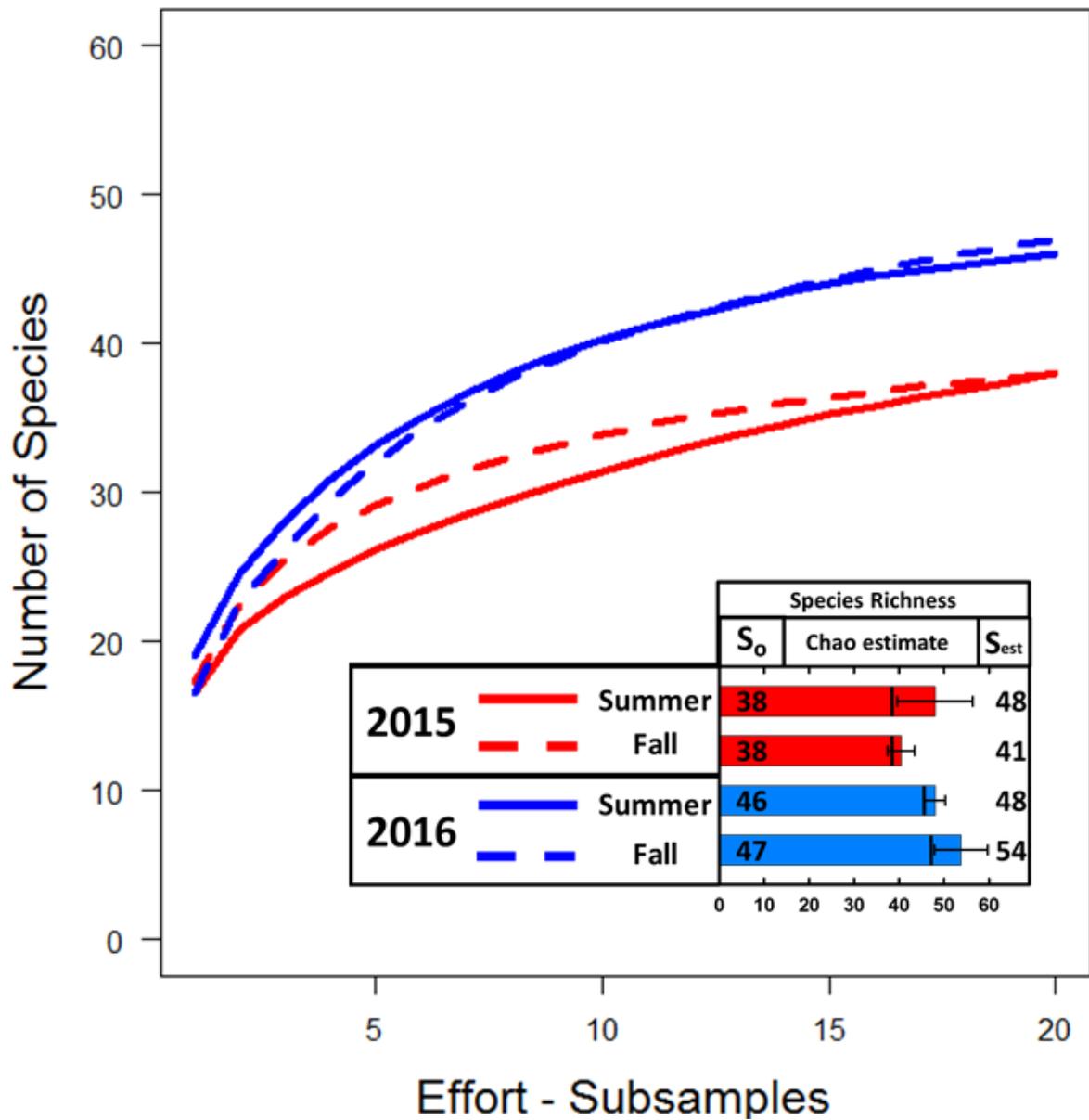


Figure 2. Species accumulation curves for Alabama River day BLE with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

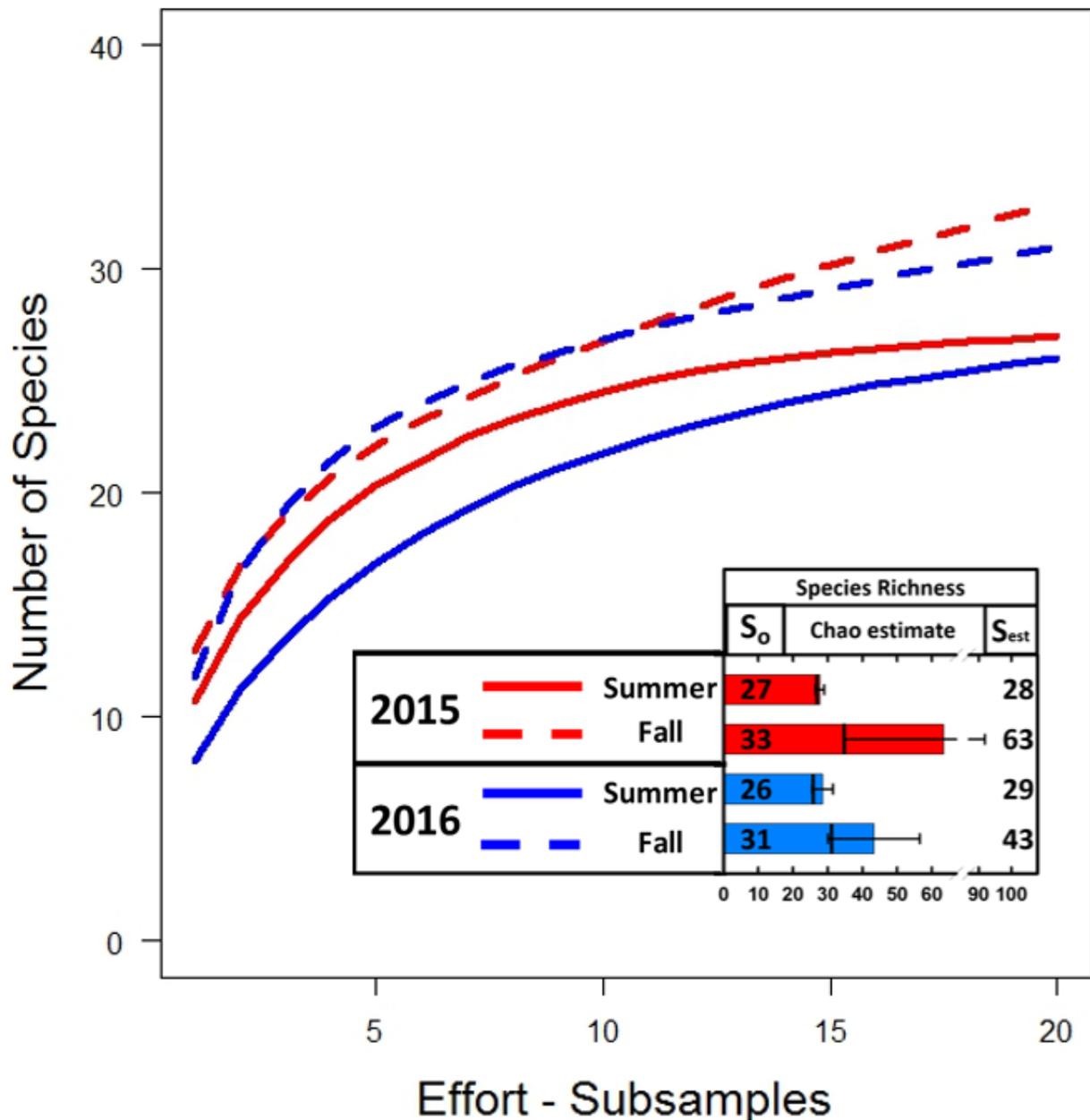


Figure 3. Species accumulation curves for Tallapoosa River day BLE with habitats combined. Each sampling event is shown with species richness ( $S_0$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

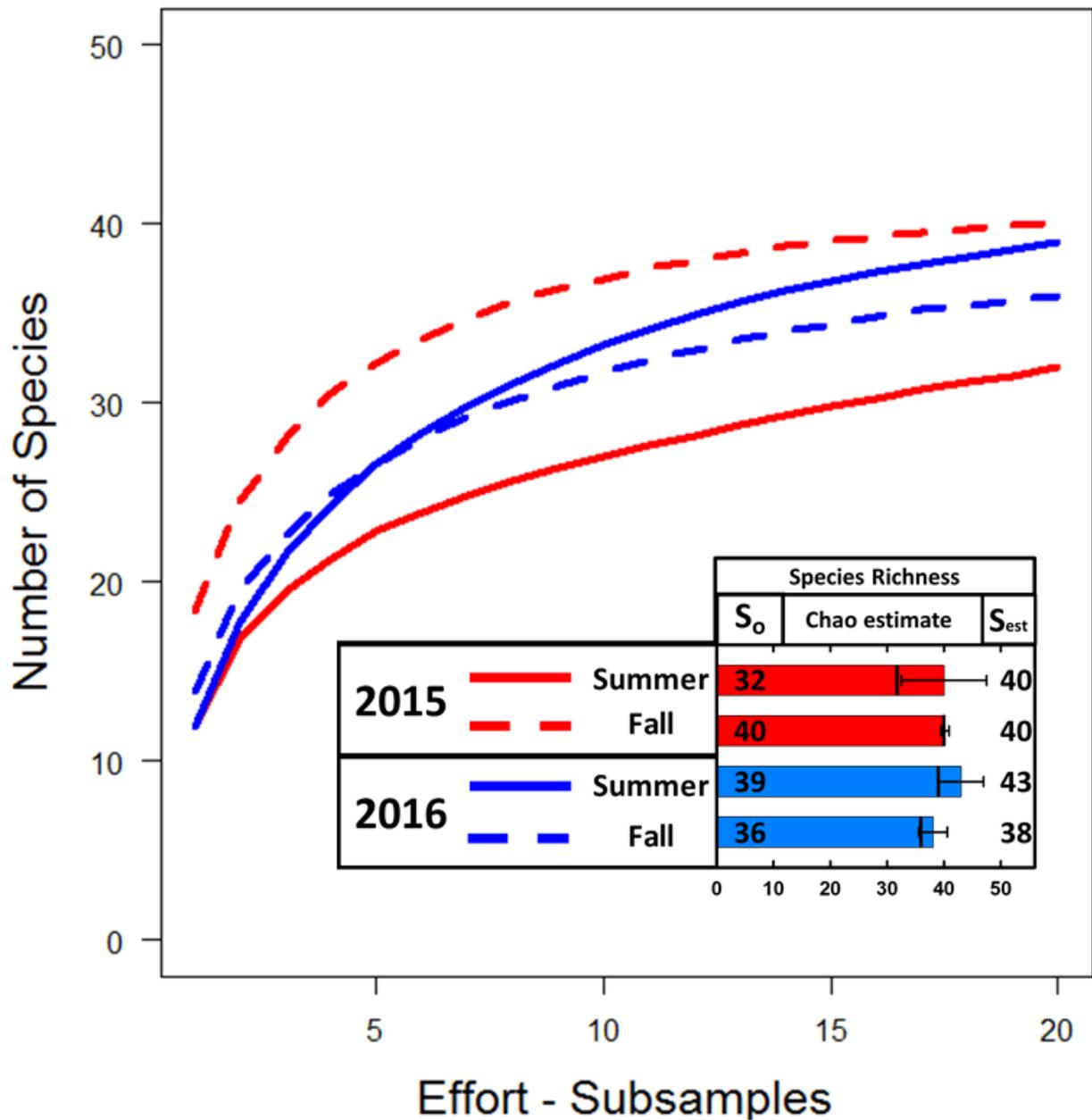


Figure 4. Species accumulation curves for Choctawhatchee River day BLE with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

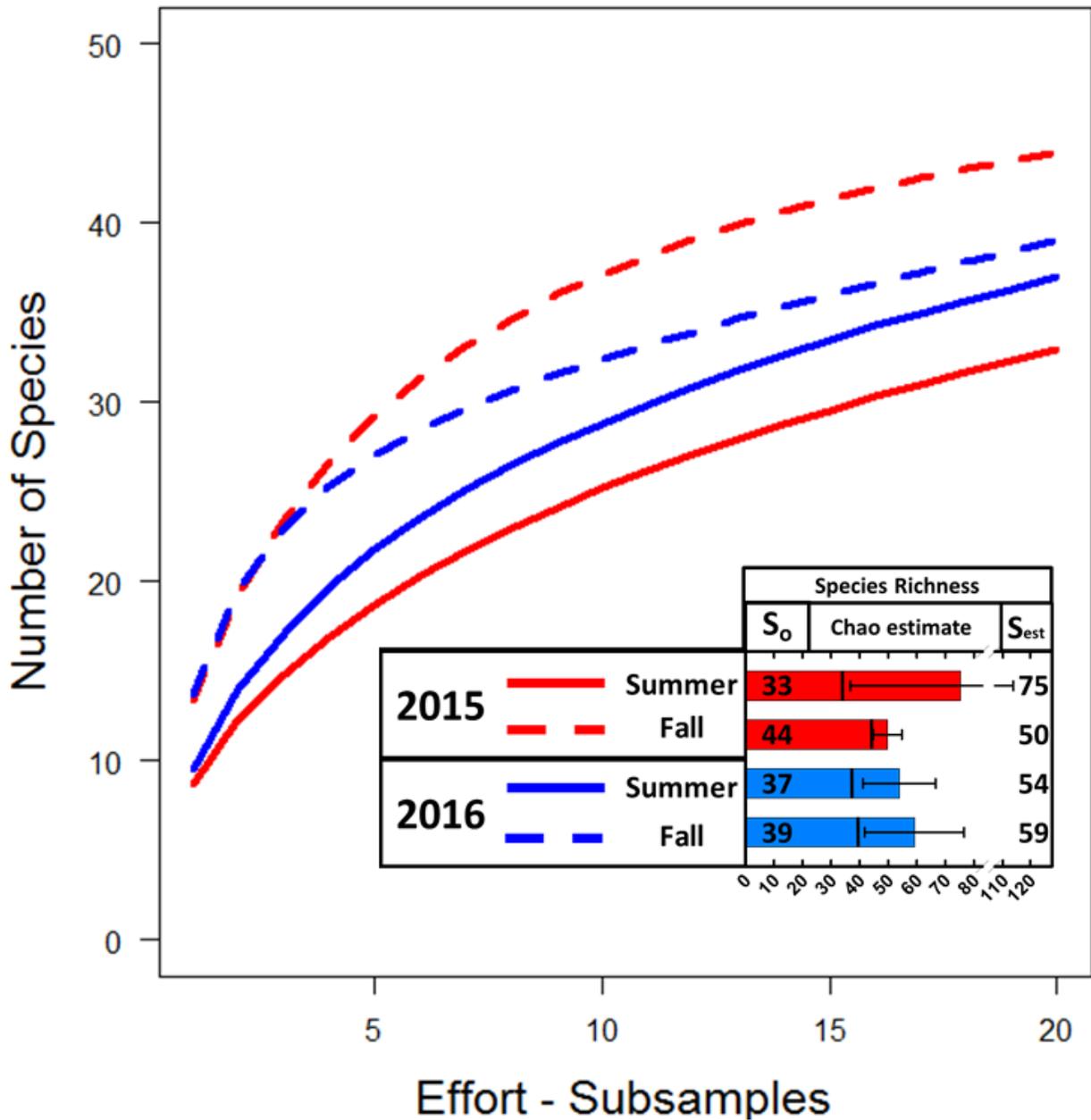


Figure 5. Species accumulation curves for Sipsey River day BLE with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

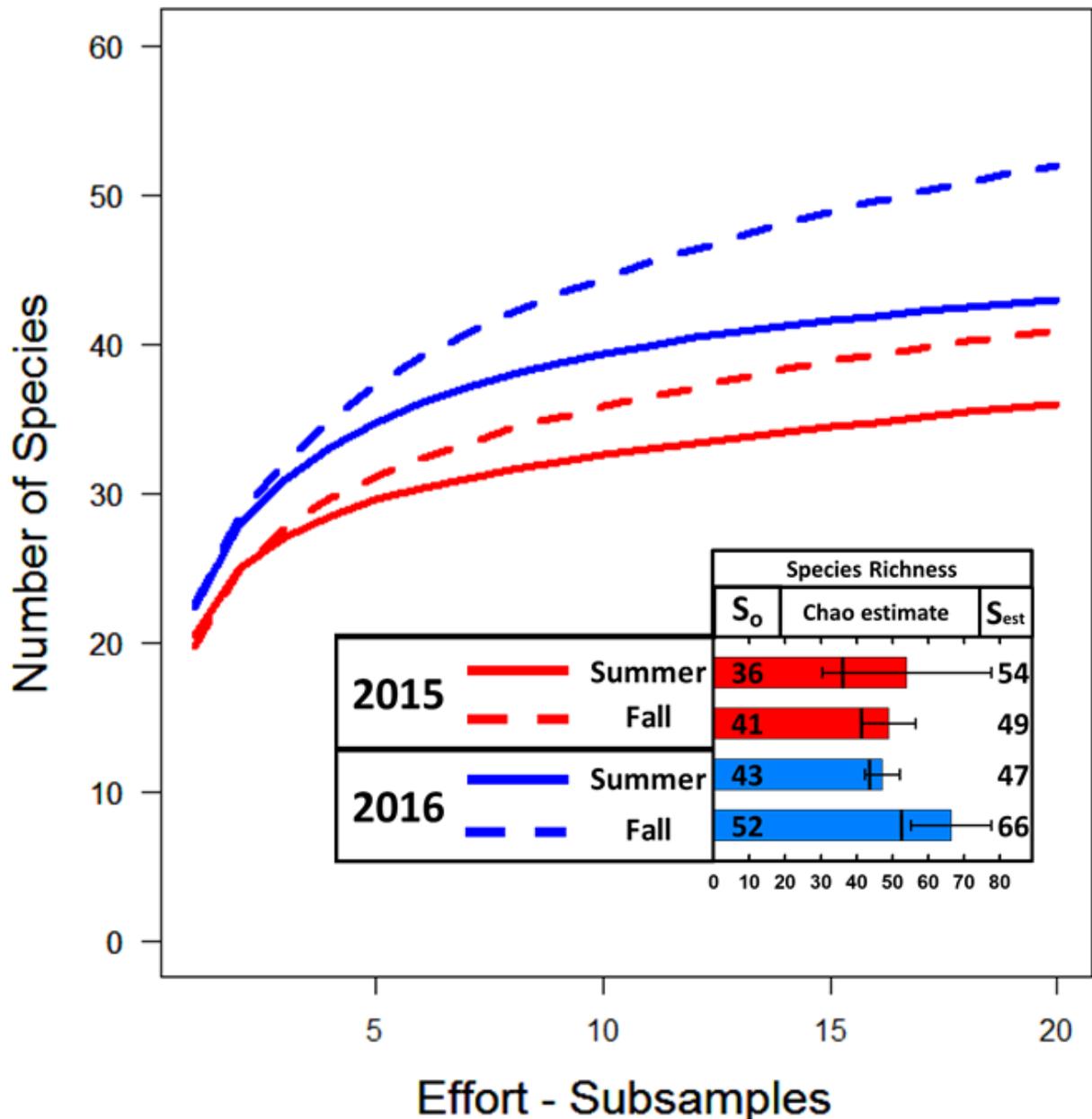


Figure 6. Species accumulation curves for Alabama River night BLE with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

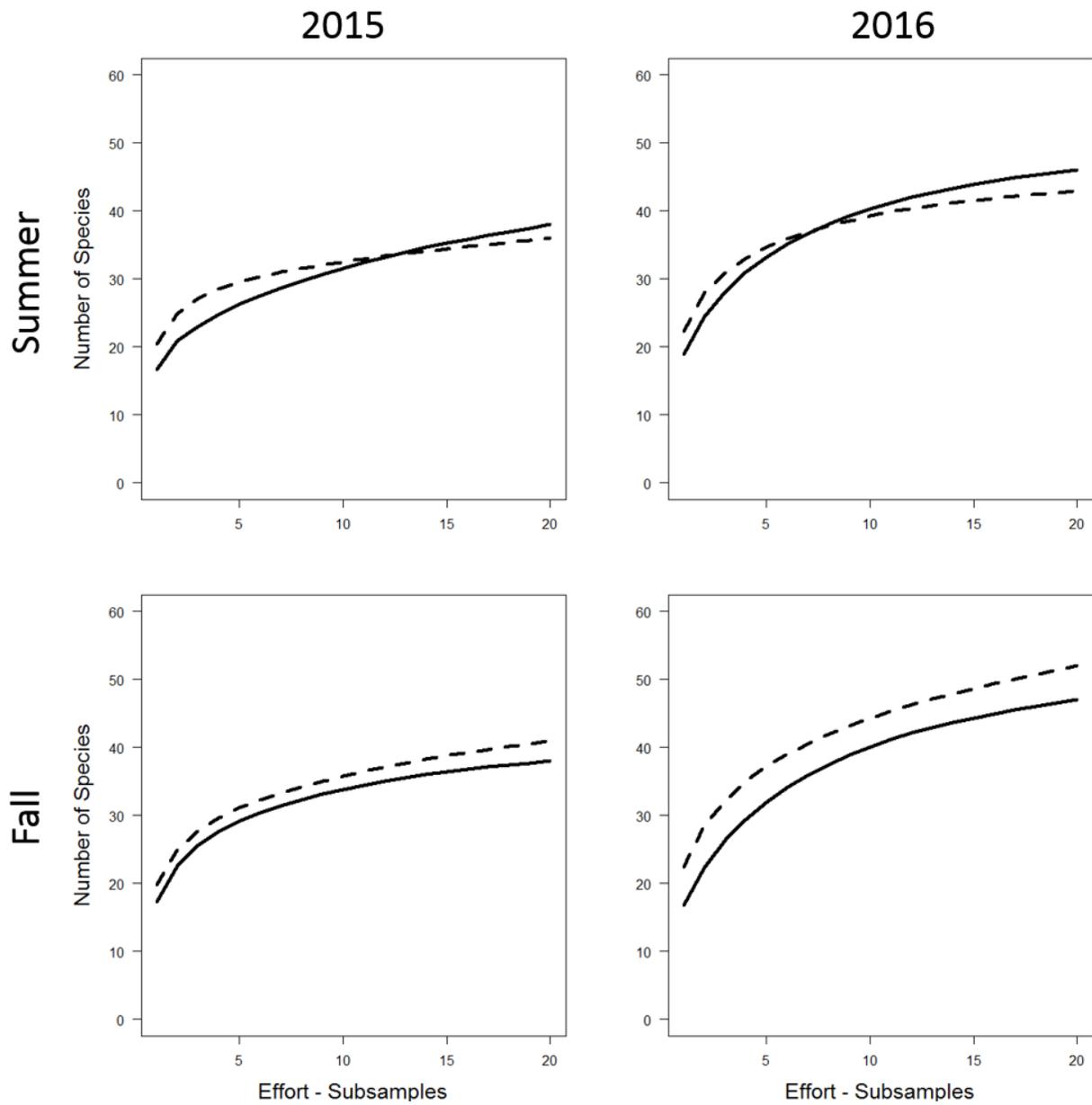


Figure 7. Species accumulation curves comparing day (solid line) with night (dashed line) BLE samples from each of the four sampling events on the Alabama River.

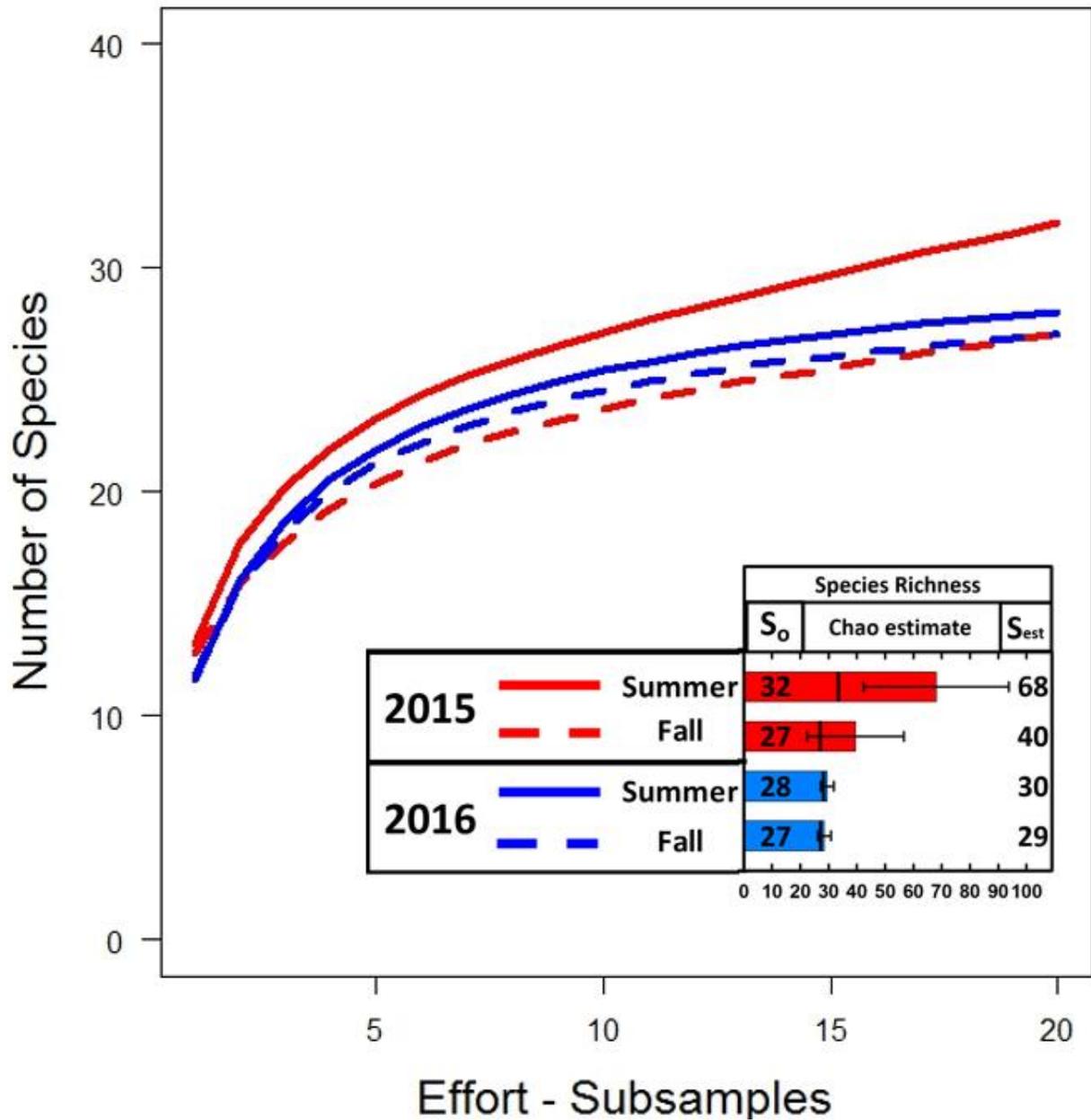


Figure 8. Species accumulation curves for Tallapoosa River night BLE with habitats combined. Each sampling event is shown with species richness ( $S_0$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

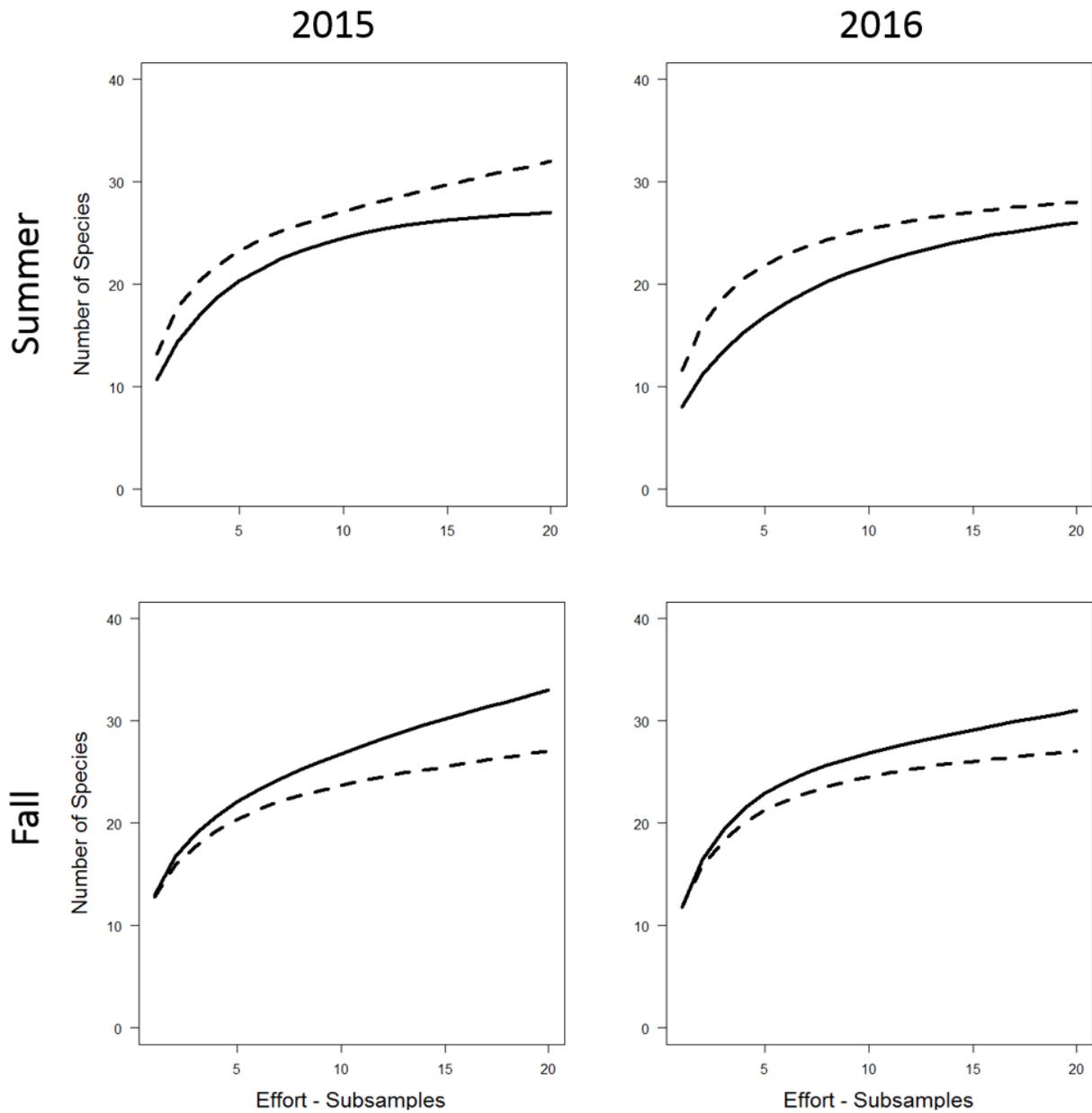


Figure 9. Species accumulation curves comparing day (solid line) with night (dashed line) BLE samples from each of the four sampling events on the Tallapoosa River.

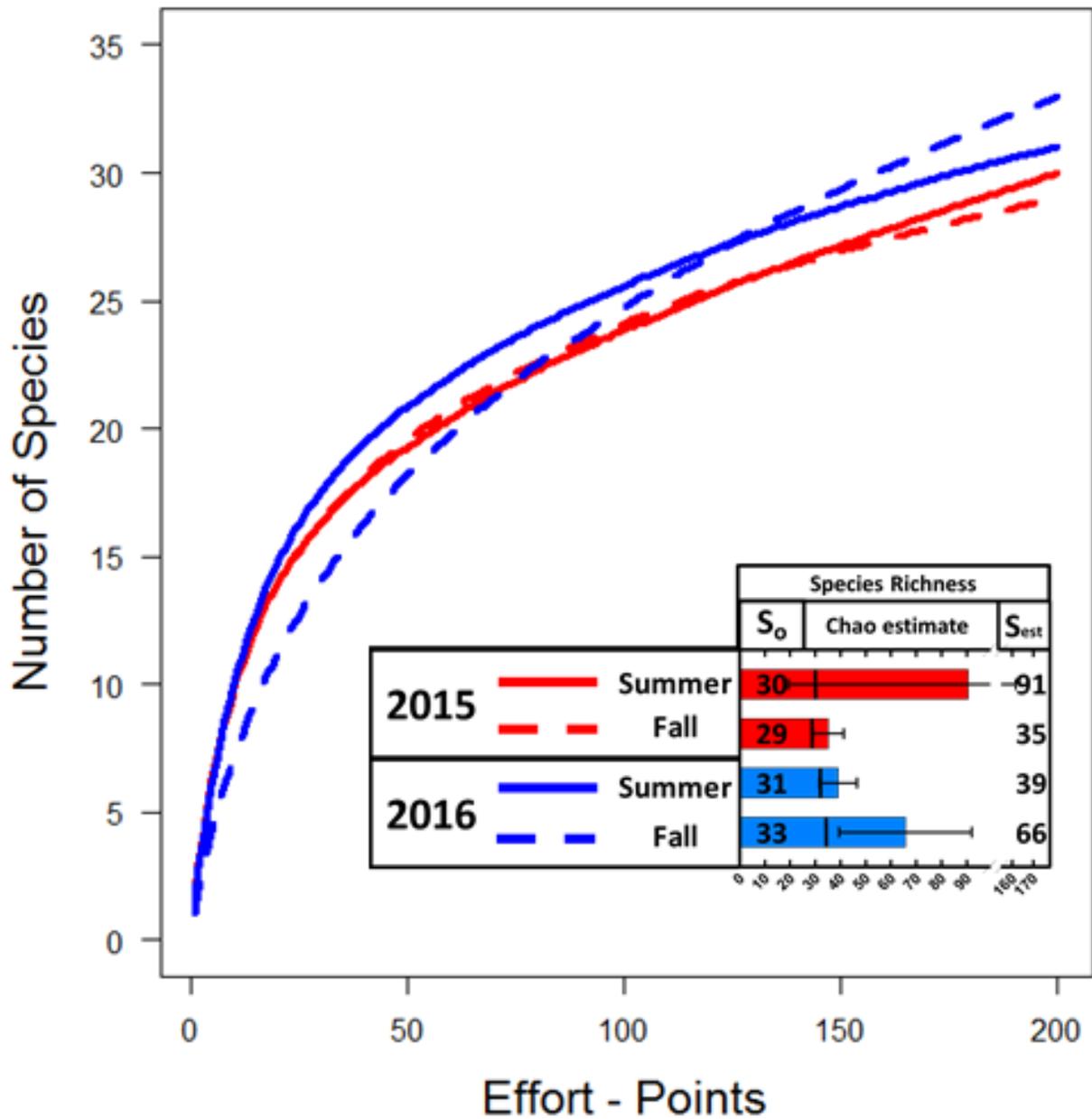


Figure 10. Species accumulation curves for Alabama River PASE sampling with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

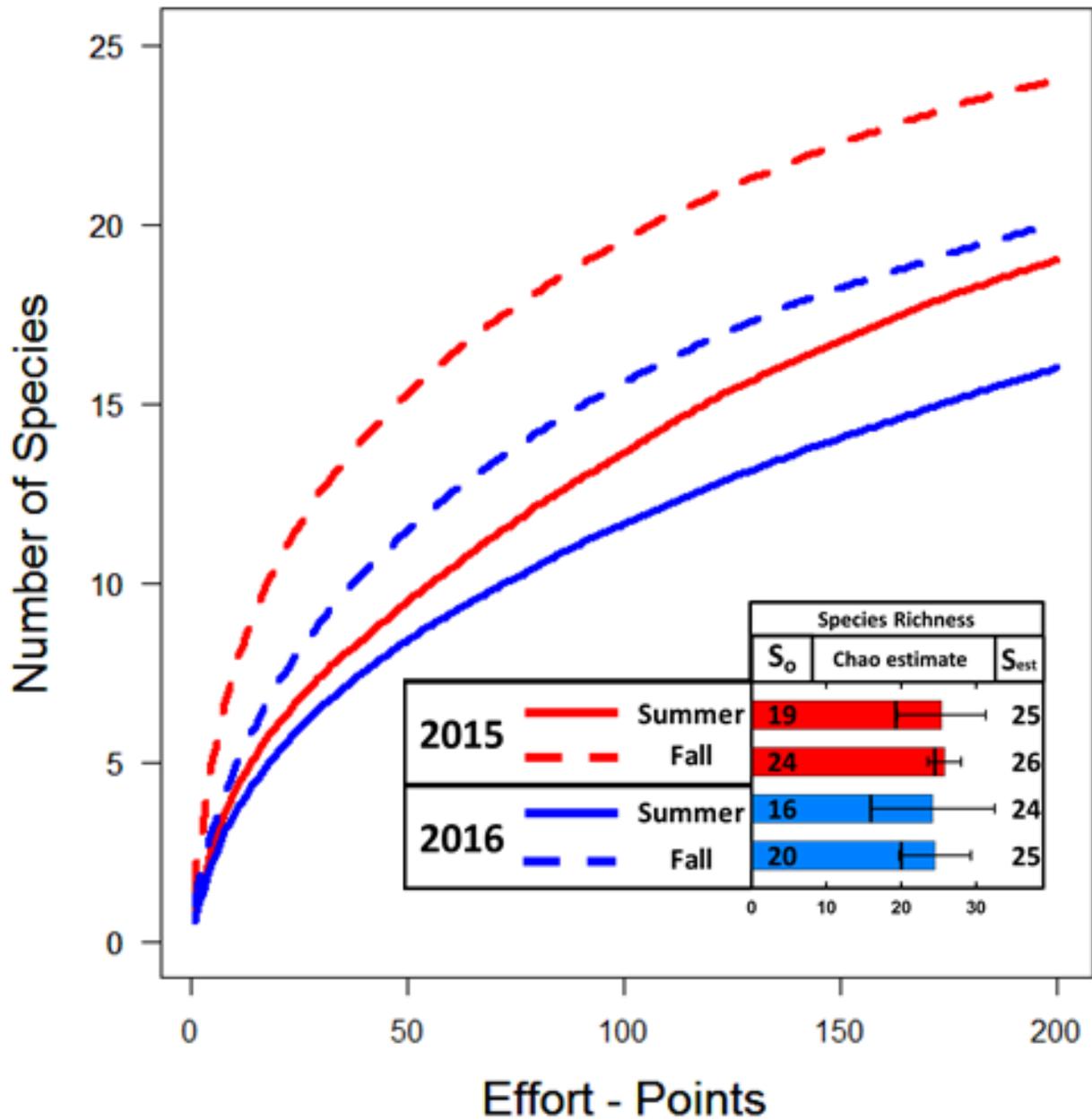


Figure 11. Species accumulation curves for Tallapoosa River PASE sampling with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

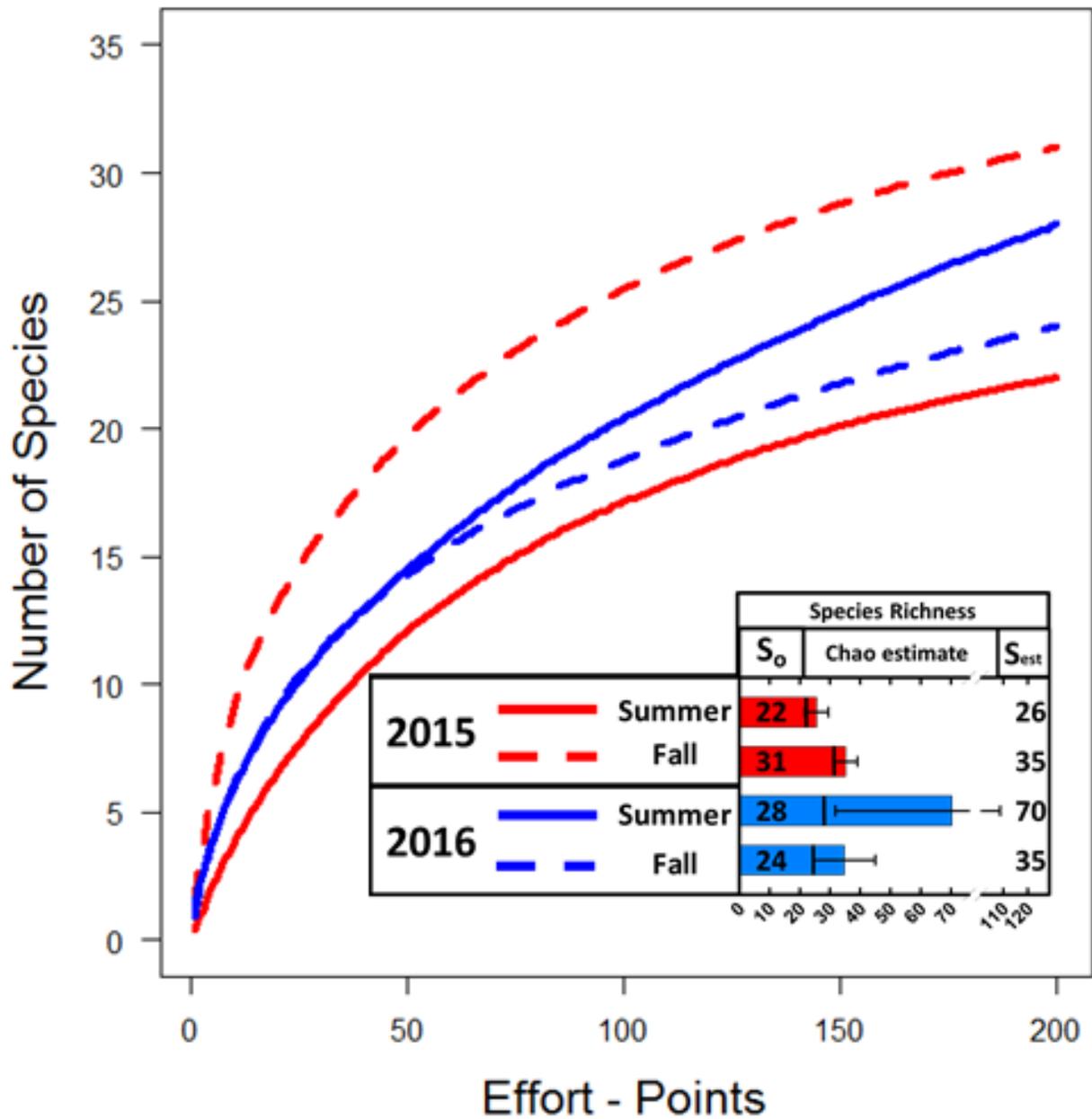


Figure 12. Species accumulation curves for Choctawhatchee River PASE sampling with habitats combined. Each sampling event is shown with species richness ( $S_0$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

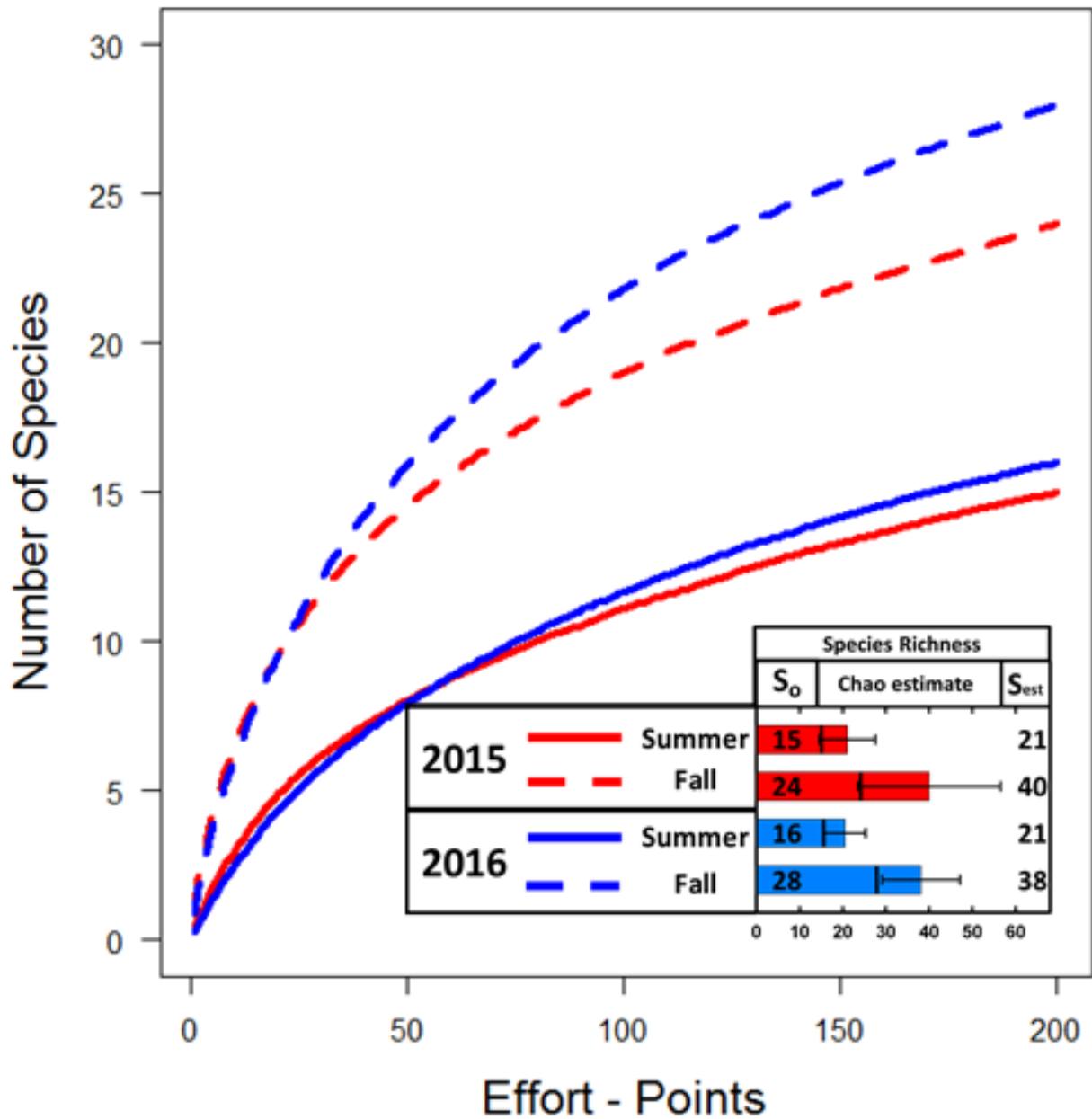


Figure 13. Species accumulation curves for Sipsey River PASE sampling with habitats combined. Each sampling event is shown with species richness ( $S_o$ ) and the species estimate ( $S_{est}$ ) using Chao's equation. Error bars represent standard error from the species estimate.

## **IX. APPENDICES**

**IX.1. PASE Form – Auxiliary Data**

<b>Point Sampling</b>		River _____	Seg _____		Date _____	
Point #	Depth (m)	Waypoint #	pH	Temp	Conductiv	Substrate
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**Point Sampling** River \_\_\_\_\_ Seg \_\_\_\_\_ Date \_\_\_\_\_

Point #	Depth (m)	Waypoint #	pH	Temp	Conductiv	Substrate
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53						
54						
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IX.4. Coordinates for start and stop locations in decimal degrees for each day and night BLE subtransect in the Alabama River. Bank indicates which side of the river was sampled when traveling in a downstream direction.

Subtransect	Start		Stop		Bank
	Lat	Long	Lat	Long	
Low 1	31.546	-87.519	31.542	-87.528	Right
Low 2	31.542	-87.528	31.547	-87.536	Right
Low 3	31.547	-87.536	31.553	-87.544	Right
Low 4	31.553	-87.544	31.556	-87.553	Right
Low 5	31.546	-87.580	31.540	-87.588	Right
Low 6	31.540	-87.588	31.533	-87.595	Right
Low 7	31.533	-87.595	31.527	-87.603	Right
Low 8	31.494	-87.591	31.491	-87.582	Left
Low 9	31.491	-87.582	31.487	-87.573	Left
Low 10	31.455	-87.566	31.446	-87.568	Left
Low 11	31.446	-87.568	31.437	-87.571	Left
High 1	31.556	-87.553	31.555	-87.563	Right
High 2	31.555	-87.563	31.551	-87.572	Right
High 3	31.551	-87.572	31.546	-87.580	Right
High 4	31.487	-87.573	31.482	-87.565	Left
High 5	31.482	-87.565	31.474	-87.562	Left
High 6	31.474	-87.562	31.464	-87.563	Left
High 7	31.464	-87.563	31.455	-87.566	Left
High 8	31.437	-87.571	31.430	-87.578	Left
High 9	31.430	-87.578	31.425	-87.586	Left

IX.5. Coordinates for start and stop locations in decimal degrees for each day and night BLE subtransect in the Tallapoosa River. Bank indicates which side of the river was sampled when traveling in a downstream direction.

Subtransect	Start		Stop		Bank
	Lat	Long	Lat	Long	
Low 1	33.010	-85.612	33.004	-85.618	Right
Low 2	33.004	-85.618	32.998	-85.621	Right
Low 3	32.998	-85.621	32.993	-85.626	Right
Low 4	32.987	-85.631	32.980	-85.633	Right
Low 5	32.973	-85.633	32.969	-85.638	Right
Low 6	32.966	-85.645	32.968	-85.652	Right
Low 7	32.968	-85.652	32.971	-85.660	Right
Low 8	32.983	-85.632	32.976	-85.634	Left
Low 9	32.966	-85.650	32.969	-85.657	Left
High 1	32.993	-85.626	32.987	-85.631	Right
High 2	32.980	-85.633	32.973	-85.633	Right
High 3	32.969	-85.638	32.966	-85.645	Right
High 4	32.976	-85.634	32.970	-85.635	Left
High 5	32.970	-85.635	32.966	-85.642	Left
High 6	32.966	-85.642	32.966	-85.650	Left
High 7	32.969	-85.657	32.971	-85.665	Left
High 8	32.971	-85.665	32.971	-85.672	Left
High 9	32.971	-85.672	32.966	-85.678	Left
High 10	32.966	-85.678	32.960	-85.683	Left
High 11	32.960	-85.683	32.957	-85.690	Left

IX.6. Coordinates for start and stop locations in decimal degrees for each day and night BLE subtransect in the Choctawhatchee River. Bank indicates which side of the river was sampled when traveling in a downstream direction.

Subtransect	Start		Stop		Bank
	Lat	Long	Lat	Long	
Low 1	31.059	-85.847	31.056	-85.851	Right
Low 2	31.051	-85.850	31.047	-85.849	Right
Low 3	31.047	-85.849	31.044	-85.852	Right
Low 4	31.044	-85.852	31.039	-85.852	Right
Low 5	31.039	-85.852	31.033	-85.853	Right
Low 6	31.033	-85.853	31.029	-85.855	Right
Low 7	31.029	-85.855	31.024	-85.857	Right
Low 8	31.011	-85.843	31.012	-85.838	Left
Low 9	31.012	-85.838	31.011	-85.832	Left
Low 10	31.011	-85.832	31.010	-85.827	Left
Low 11	31.007	-85.831	31.002	-85.830	Left
High 1	31.065	-85.843	31.063	-85.848	Right
High 2	31.063	-85.848	31.059	-85.847	Right
High 3	31.056	-85.851	31.051	-85.850	Right
High 4	31.018	-85.853	31.013	-85.853	Left
High 5	31.013	-85.853	31.010	-85.848	Left
High 6	31.010	-85.848	31.011	-85.843	Left
High 7	31.010	-85.827	31.006	-85.823	Left
High 8	31.006	-85.823	31.004	-85.827	Left
High 9	31.004	-85.827	31.007	-85.831	Left

IX.7. Coordinates for start and stop locations in decimal degrees for each day and night BLE subtransect in the Sipsy River. Bank indicates which side of the river was sampled when traveling in a downstream direction.

Subtransect	Start		Stop		Bank
	Lat	Long	Lat	Long	
Low 1	33.042	-88.119	33.041	-88.123	Left
Low 2	33.041	-88.123	33.041	-88.126	Left
Low 3	33.041	-88.126	33.038	-88.126	Left
Low 4	33.038	-88.126	33.037	-88.129	Left
Low 5	33.037	-88.129	33.037	-88.133	Left
Low 6	33.037	-88.136	33.040	-88.137	Left
Low 7	33.036	-88.085	33.038	-88.089	Right
Low 8	33.038	-88.089	33.038	-88.092	Right
Low 9	33.038	-88.092	33.036	-88.095	Right
Low 10	33.037	-88.102	33.036	-88.106	Right
High 1	33.037	-88.109	33.039	-88.112	Left
High 2	33.039	-88.112	33.042	-88.115	Left
High 3	33.042	-88.115	33.042	-88.119	Left
High 4	33.037	-88.133	33.037	-88.136	Left
High 5	33.040	-88.080	33.037	-88.081	Right
High 6	33.037	-88.081	33.036	-88.085	Right
High 7	33.036	-88.095	33.035	-88.099	Right
High 8	33.035	-88.099	33.037	-88.102	Right
High 9	33.036	-88.106	33.033	-88.106	Right
High 10	33.033	-88.106	33.035	-88.108	Right

IX.8. Number of individuals sampled of each species for each sampling event comparing day and night BLE sampling on the Alabama River. Species are listed by family in alphabetical order. Rows containing all zeros indicate a species that was captured only during point sampling.

	Day				Night			
	2015		2016		2015		2016	
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall
<b><u>Achiridae</u></b>								
<i>Trinectes maculatus</i>	0	0	0	19	0	0	2	2
<b><u>Amiidae</u></b>								
<i>Amia calva</i>	66	58	79	48	32	19	44	43
<b><u>Anguillidae</u></b>								
<i>Anguilla rostrata</i>	2	1	6	1	0	1	3	2
<b><u>Aphredoderidae</u></b>								
<i>Aphredoderus sayanus</i>	0	0	0	3	0	0	0	0
<b><u>Atherinopsidae</u></b>								
<i>Labidesthes sicculus</i>	1	5	7	6	0	0	4	8
<i>Menidia beryllina</i>	0	0	0	0	0	1	0	0
<b><u>Belonidae</u></b>								
<i>Strongylura marina</i>	4	0	6	0	5	0	12	0
<b><u>Catostomidae</u></b>								
<i>Carpiodes cyprinus</i>	0	3	2	0	4	26	0	3
<i>Carpiodes velifer</i>	68	11	73	6	86	82	30	16
<i>Cycleptus meridionalis</i>	4	1	2	4	0	2	0	1
<i>Ictiobus bubalus</i>	56	102	26	54	86	199	31	25
<i>Moxostoma duquesnei</i>	0	0	0	4	0	1	0	3
<i>Moxostoma poecilurum</i>	24	16	22	19	62	67	75	97
<b><u>Centrarchidae</u></b>								
<i>Ambloplites ariommus</i>	0	0	0	2	0	2	0	1
<i>Lepomis auritus</i>	0	0	0	0	0	0	0	0
<i>Lepomis cyanellus</i>	0	2	2	3	0	0	0	0
<i>Lepomis gulosus</i>	3	2	1	3	1	4	3	5
<i>Lepomis humilis</i>	0	0	0	0	0	0	0	1
<i>Lepomis macrochirus</i>	473	467	362	452	1466	954	1825	2829
<i>Lepomis megalotis</i>	160	111	193	134	23	86	83	177
<i>Lepomis microlophus</i>	1	2	3	6	3	1	4	12
<i>Lepomis miniatus</i>	15	6	6	9	0	7	0	10
<i>Micropterus henshalli</i>	71	87	120	122	314	221	325	502
<i>Micropterus salmoides</i>	212	180	146	82	155	69	105	99
<i>Pomoxis annularis</i>	0	0	0	0	0	1	0	0

<i>Pomoxis nigromaculatus</i>	117	34	81	57	76	25	47	17
<b><u>Clupeidae</u></b>								
<i>Alosa chrysochloris</i>	1	0	1	0	0	0	1	1
<i>Brevoortia patronus</i>	0	0	0	0	0	0	0	13
<i>Dorosoma cepedianum</i>	125	262	166	72	299	44	88	179
<i>Dorosoma petenense</i>	76	399	29	25	271	47	23	40
<b><u>Cyprinidae</u></b>								
<i>Campostoma oligolepis</i>	0	0	1	0	0	0	0	0
<i>Ctenopharyngodon idella</i>	0	4	2	2	1	4	0	0
<i>Cyprinella venusta</i>	113	558	231	256	15	19	136	217
<i>Cyprinus carpio</i>	27	44	16	8	12	14	7	7
<i>Hybognathus nuchalis</i>	0	1	23	19	0	1	13	25
<i>Macrhybopsis storeriana</i>	1	31	9	1	49	11	114	156
<i>Notemigonus crysoleucas</i>	0	0	0	0	1	0	0	1
<i>Notropis atherinoides</i>	96	1141	184	857	270	527	162	1560
<i>Notropis candidus</i>	0	121	17	23	407	149	523	1092
<i>Notropis edwarddraneyi</i>	0	113	15	2	92	68	750	334
<i>Notropis texanus</i>	2	41	3	9	0	1	2	16
<i>Opsopoeodus emiliae</i>	0	2	0	0	0	0	1	3
<i>Pimephales vigilax</i>	1	92	3	3	40	41	135	218
<b><u>Engraulidae</u></b>								
<i>Anchoa mitchilli</i>	9	546	9	63	523	39	175	130
<b><u>Fundulidae</u></b>								
<i>Fundulus olivaceus</i>	1	5	9	4	1	0	4	1
<b><u>Hiodontidae</u></b>								
<i>Hiodon tergisus</i>	1	0	0	0	0	0	0	1
<b><u>Ictaluridae</u></b>								
<i>Ameiurus natalis</i>	0	0	0	1	0	0	0	1
<i>Ictalurus furcatus</i>	4	3	4	3	6	11	6	6
<i>Ictalurus punctatus</i>	55	32	61	32	147	86	104	142
<i>Pylodictis olivaris</i>	9	0	5	5	7	2	20	7
<b><u>Lepisosteidae</u></b>								
<i>Lepisosteus oculatus</i>	169	185	93	16	255	137	79	41
<i>Lepisosteus osseus</i>	4	1	4	1	32	39	16	3
<i>Lepisosteus spatula</i>	1	0	1	0	1	0	0	0
<b><u>Moronidae</u></b>								
<i>Morone chrysops</i>	0	0	0	0	0	0	1	3
<i>M. chrysops x M. saxatilis</i>	0	0	0	2	0	0	1	7
<i>Morone saxatilis</i>	1	0	5	5	1	0	2	9
<b><u>Mugilidae</u></b>								
<i>Mugil cephalus</i>	26	42	10	153	30	5	6	16

**Percidae**

<i>Crystallaria asprella</i>	0	0	4	0	0	0	1	1
<i>Percina kathae</i>	0	0	0	0	0	0	0	1
<i>Percina nigrofasciata</i>	0	0	0	3	0	0	0	0
<i>Percina shumardi</i>	0	0	0	0	0	1	0	0
<i>Percina suttkusi</i>	0	0	2	3	0	0	6	6

**Poeciliidae**

<i>Gambusia affinis</i>	0	0	0	1	0	0	0	0
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**Polyodontidae**

<i>Polyodon spathula</i>	1	0	1	0	24	6	5	0
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**Sciaenidae**

<i>Aplodinotus grunniens</i>	91	10	75	41	24	266	34	55
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IX.9. Number of individuals sampled of each species for each sampling event comparing day and night BLE sampling on the Tallapoosa River. Species are listed by family in alphabetical order. Rows containing all zeros indicate a species that was captured only during point sampling.

	Day				Night			
	2015		2016		2015		2016	
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall
<b><u>Amiidae</u></b>								
<i>Amia calva</i>	4	1	0	1	0	0	1	0
<b><u>Catostomidae</u></b>								
<i>Hypentelium etowanum</i>	22	43	10	19	3	7	6	6
<i>Minytrema melanops</i>	14	1	0	2	9	8	4	2
<i>Moxostoma duquesnei</i>	10	9	0	1	1	38	2	1
<i>Moxostoma poecilurum</i>	242	174	81	88	55	166	61	35
<b><u>Centrarchidae</u></b>								
<i>Ambloplites ariommus</i>	6	26	6	29	23	64	14	40
<i>Lepomis auritus</i>	363	470	264	460	457	923	410	608
<i>Lepomis cyanellus</i>	18	11	5	10	20	27	13	12
<i>Lepomis gulosus</i>	2	1	2	6	10	9	7	7
<i>Lepomis macrochirus</i>	165	221	95	170	289	369	314	200
<i>Lepomis megalotis</i>	3	0	3	0	1	0	0	0
<i>Lepomis microlophus</i>	3	2	7	7	15	8	8	10
<i>Lepomis miniatus</i>	0	2	4	3	0	1	0	0
<i>Micropterus henshalli</i>	200	314	57	168	291	387	263	128
<i>Micropterus salmoides</i>	13	42	22	6	23	31	16	16
<i>Micropterus tallapoosae</i>	12	30	5	28	38	73	19	36
<i>Pomoxis annularis</i>	0	1	0	0	1	0	0	0
<i>Pomoxis nigromaculatus</i>	9	13	3	9	4	4	0	1
<b><u>Clupeidae</u></b>								
<i>Dorosoma cepedianum</i>	0	0	0	0	4	0	16	2
<b><u>Cyprinidae</u></b>								
<i>Campostoma oligolepis</i>	0	0	1	3	0	0	0	0
<i>Ctenopharyngodon idella</i>	0	0	1	0	9	0	0	0
<i>Cyprinella callistia</i>	0	16	0	12	1	0	3	4
<i>Cyprinella gibbsi</i>	0	0	1	0	0	0	0	0
<i>Cyprinella venusta</i>	79	408	152	216	188	182	522	210
<i>Cyprinus carpio</i>	39	11	1	8	54	56	18	19
<i>Notemigonus crysoleucas</i>	0	0	0	0	0	1	0	0
<i>Notropis baileyi</i>	0	1	0	0	0	0	0	0
<i>Notropis stilbius</i>	3	345	3	208	21	14	63	57

<i>Notropis texanus</i>	0	4	0	0	2	2	0	0
<i>Pimephales vigilax</i>	0	46	0	1	9	0	1	2
<b><u>Fundulidae</u></b>								
<i>Fundulus bifax</i>	0	1	0	0	0	0	0	0
<i>Fundulus olivaceus</i>	3	23	2	18	1	1	3	7
<b><u>Ictaluridae</u></b>								
<i>Ictalurus furcatus</i>	0	0	0	0	1	0	0	0
<i>Ictalurus punctatus</i>	26	26	4	16	47	43	23	37
<i>Pylodictis olivaris</i>	6	1	3	1	13	5	8	9
<b><u>Moronidae</u></b>								
<i>Morone chrysops</i>	0	1	0	0	0	0	0	0
<i>Morone saxatilis</i>	0	0	0	0	1	0	0	0
<b><u>Percidae</u></b>								
<i>Etheostoma chuckwachatte</i>	0	0	0	1	0	0	0	0
<i>Etheostoma stigmaeum</i>	4	1	0	5	0	1	6	3
<i>Perca flavescens</i>	1	0	0	0	0	0	0	0
<i>Percina kathae</i>	10	1	6	7	15	3	12	5
<i>Percina palmaris</i>	6	1	4	10	8	1	17	12
<i>Percina smithvanizi</i>	2	4	3	13	0	0	6	14
<b><u>Petromyzontidae</u></b>								
<i>Ichthyomyzon gagei</i>	0	0	0	0	1	8	1	0
<b><u>Poeciliidae</u></b>								
<i>Gambusia affinis</i>	0	0	0	1	0	0	0	0

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IX.10. Number of individuals sampled of each species for each sampling event comparing day BLE and PASE sampling on the Alabama River. Species are listed by family in alphabetical order. Rows containing all zeros indicate a species that was captured only during night-time sampling.

	Day				Point			
	2015		2016		2015		2016	
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall
<b><u>Achiridae</u></b>								
<i>Trinectes maculatus</i>	0	0	0	19	0	0	0	2
<b><u>Amiidae</u></b>								
<i>Amia calva</i>	66	58	79	48	9	8	7	6
<b><u>Anguillidae</u></b>								
<i>Anguilla rostrata</i>	2	1	6	1	0	1	0	1
<b><u>Aphredoderidae</u></b>								
<i>Aphredoderus sayanus</i>	0	0	0	3	0	0	0	0
<b><u>Atherinopsidae</u></b>								
<i>Labidesthes sicculus</i>	1	5	7	6	0	2	3	1
<i>Menidia beryllina</i>	0	0	0	0	0	0	0	0
<b><u>Belonidae</u></b>								
<i>Strongylura marina</i>	4	0	6	0	0	0	1	0
<b><u>Catostomidae</u></b>								
<i>Carpiodes cyprinus</i>	0	3	2	0	1	0	0	0
<i>Carpiodes velifer</i>	68	11	73	6	12	3	11	0
<i>Cycleptus meridionalis</i>	4	1	2	4	0	0	0	1
<i>Ictiobus bubalus</i>	56	102	26	54	24	33	16	6
<i>Moxostoma duquesnei</i>	0	0	0	4	0	0	0	0
<i>Moxostoma poecilurum</i>	24	16	22	19	4	3	1	1
<b><u>Centrarchidae</u></b>								
<i>Ambloplites ariommus</i>	0	0	0	2	0	0	0	0
<i>Lepomis auritus</i>	0	0	0	0	0	0	0	1
<i>Lepomis cyanellus</i>	0	2	2	3	0	0	0	0
<i>Lepomis gulosus</i>	3	2	1	3	0	1	1	1
<i>Lepomis humilis</i>	0	0	0	0	0	0	0	0
<i>Lepomis macrochirus</i>	473	467	362	452	142	74	94	88
<i>Lepomis megalotis</i>	160	111	193	134	39	22	30	35
<i>Lepomis microlophus</i>	1	2	3	6	1	0	1	2
<i>Lepomis miniatus</i>	15	6	6	9	1	0	4	1
<i>Micropterus henshalli</i>	71	87	120	122	24	10	19	16
<i>Micropterus salmoides</i>	212	180	146	82	25	17	19	11
<i>Pomoxis annularis</i>	0	0	0	0	0	0	0	0

<i>Pomoxis nigromaculatus</i>	117	34	81	57	27	21	16	12
<b><u>Clupeidae</u></b>								
<i>Alosa chrysochloris</i>	1	0	1	0	0	0	0	0
<i>Brevoortia patronus</i>	0	0	0	0	0	0	0	0
<i>Dorosoma cepedianum</i>	125	262	166	72	17	51	38	3
<i>Dorosoma petenense</i>	76	399	29	25	22	310	2	0
<b><u>Cyprinidae</u></b>								
<i>Campostoma oligolepis</i>	0	0	1	0	0	0	0	0
<i>Ctenopharyngodon idella</i>	0	4	2	2	0	1	2	0
<i>Cyprinella venusta</i>	113	558	231	256	15	113	28	36
<i>Cyprinus carpio</i>	27	44	16	8	5	7	0	0
<i>Hybognathus nuchalis</i>	0	1	23	19	0	0	5	0
<i>Macrhybopsis storeriana</i>	1	31	9	1	1	0	5	0
<i>Notemigonus crysoleucas</i>	0	0	0	0	0	0	1	0
<i>Notropis atherinoides</i>	96	1141	184	857	27	225	72	25
<i>Notropis candidus</i>	0	121	17	23	1	51	7	14
<i>Notropis edwarddraneyi</i>	0	113	15	2	2	6	0	1
<i>Notropis texanus</i>	2	41	3	9	7	4	10	26
<i>Opsopoeodus emiliae</i>	0	2	0	0	0	1	0	0
<i>Pimephales vigilax</i>	1	92	3	3	1	5	0	1
<b><u>Engraulidae</u></b>								
<i>Anchoa mitchilli</i>	9	546	9	63	3	16	0	9
<b><u>Fundulidae</u></b>								
<i>Fundulus olivaceus</i>	1	5	9	4	3	1	1	3
<b><u>Hiodontidae</u></b>								
<i>Hiodon tergisus</i>	1	0	0	0	0	0	0	0
<b><u>Ictaluridae</u></b>								
<i>Ameiurus natalis</i>	0	0	0	1	0	0	0	0
<i>Ictalurus furcatus</i>	4	3	4	3	1	0	1	1
<i>Ictalurus punctatus</i>	55	32	61	32	29	1	10	8
<i>Pylodictis olivaris</i>	9	0	5	5	0	0	2	0
<b><u>Lepisosteidae</u></b>								
<i>Lepisosteus oculatus</i>	169	185	93	16	28	24	14	1
<i>Lepisosteus osseus</i>	4	1	4	1	0	0	2	0
<i>Lepisosteus spatula</i>	1	0	1	0	0	0	0	0
<b><u>Moronidae</u></b>								
<i>Morone chrysops</i>	0	0	0	0	0	0	0	0
<i>M. chrysops x M. saxatilis</i>	0	0	0	2	0	0	0	0
<i>Morone saxatilis</i>	1	0	5	5	3	0	0	5
<b><u>Mugilidae</u></b>								
<i>Mugil cephalus</i>	26	42	10	153	2	9	0	11

**Percidae**

<i>Crystallaria asprella</i>	0	0	4	0	0	0	0	0
<i>Percina kathae</i>	0	0	0	0	0	0	0	0
<i>Percina nigrofasciata</i>	0	0	0	3	0	0	0	0
<i>Percina shumardi</i>	0	0	0	0	0	0	0	0
<i>Percina suttkusi</i>	0	0	2	3	0	0	0	1

**Poeciliidae**

<i>Gambusia affinis</i>	0	0	0	1	0	0	0	1
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**Polyodontidae**

<i>Polyodon spathula</i>	1	0	1	0	0	0	0	0
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**Sciaenidae**

<i>Aplodinotus grunniens</i>	91	10	75	41	45	2	23	5
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IX.11. Number of individuals sampled of each species for each sampling event comparing day BLE and PASE sampling on the Tallapoosa River. Species are listed by family in alphabetical order. Rows containing all zeros indicate a species that was captured only during night-time sampling.

	Day				Point			
	2015		2016		2015		2016	
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall
<b><u>Amiidae</u></b>								
<i>Amia calva</i>	4	1	0	1	1	0	0	0
<b><u>Catostomidae</u></b>								
<i>Hypentelium etowanum</i>	22	43	10	19	6	4	0	1
<i>Minytrema melanops</i>	14	1	0	2	0	0	0	0
<i>Moxostoma duquesnei</i>	10	9	0	1	1	1	0	0
<i>Moxostoma poecilurum</i>	242	174	81	88	37	28	8	1
<b><u>Centrarchidae</u></b>								
<i>Ambloplites ariommus</i>	6	26	6	29	0	6	0	7
<i>Lepomis auritus</i>	363	470	264	460	55	133	71	168
<i>Lepomis cyanellus</i>	18	11	5	10	2	3	1	2
<i>Lepomis gulosus</i>	2	1	2	6	3	2	0	1
<i>Lepomis macrochirus</i>	165	221	95	170	37	46	27	47
<i>Lepomis megalotis</i>	3	0	3	0	0	1	1	0
<i>Lepomis microlophus</i>	3	2	7	7	1	2	0	0
<i>Lepomis miniatus</i>	0	2	4	3	0	1	0	3
<i>Micropterus henshalli</i>	200	314	57	168	20	52	7	18
<i>Micropterus salmoides</i>	13	42	22	6	0	10	0	0
<i>Micropterus tallapoosae</i>	12	30	5	28	1	6	2	3
<i>Pomoxis annularis</i>	0	1	0	0	0	0	0	0
<i>Pomoxis nigromaculatus</i>	9	13	3	9	0	2	0	0
<b><u>Clupeidae</u></b>								
<i>Dorosoma cepedianum</i>	0	0	0	0	0	1	0	0
<b><u>Cyprinidae</u></b>								
<i>Campostoma oligolepis</i>	0	0	1	3	0	0	0	0
<i>Ctenopharyngodon idella</i>	0	0	1	0	0	0	0	0
<i>Cyprinella callistia</i>	0	16	0	12	2	2	1	0
<i>Cyprinella gibbsi</i>	0	0	1	0	0	0	0	0
<i>Cyprinella venusta</i>	79	408	152	216	8	60	66	32
<i>Cyprinus carpio</i>	39	11	1	8	0	8	0	1
<i>Notemigonus crysoleucas</i>	0	0	0	0	0	0	0	0
<i>Notropis baileyi</i>	0	1	0	0	0	0	0	0
<i>Notropis stilbius</i>	3	345	3	208	0	30	2	9

<i>Notropis texanus</i>	0	4	0	0	0	0	0	0
<i>Pimephales vigilax</i>	0	46	0	1	0	2	0	0
<b><u>Fundulidae</u></b>								
<i>Fundulus bifax</i>	0	1	0	0	0	0	0	0
<i>Fundulus olivaceus</i>	3	23	2	18	1	14	1	5
<b><u>Ictaluridae</u></b>								
<i>Ictalurus furcatus</i>	0	0	0	0	0	0	0	0
<i>Ictalurus punctatus</i>	26	26	4	16	4	3	1	6
<i>Pylodictis olivaris</i>	6	1	3	1	0	1	1	1
<b><u>Moronidae</u></b>								
<i>Morone chrysops</i>	0	1	0	0	0	0	0	0
<i>Morone saxatilis</i>	0	0	0	0	0	0	0	0
<b><u>Percidae</u></b>								
<i>Etheostoma chuckwachatte</i>	0	0	0	1	0	0	0	0
<i>Etheostoma stigmaeum</i>	4	1	0	5	0	0	1	2
<i>Perca flavescens</i>	1	0	0	0	0	0	0	0
<i>Percina kathae</i>	10	1	6	7	2	0	2	1
<i>Percina palmaris</i>	6	1	4	10	3	0	3	2
<i>Percina smithvanizi</i>	2	4	3	13	1	0	0	2
<b><u>Petromyzontidae</u></b>								
<i>Ichthyomyzon gagei</i>	0	0	0	0	2	0	0	0
<b><u>Poeciliidae</u></b>								
<i>Gambusia affinis</i>	0	0	0	1	0	0	0	0

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IX.12. Number of individuals sampled of each species for each sampling event comparing day BLE and PASE sampling on the Choctawhatchee River. Species are listed by family in alphabetical order.

	Day				Point				
	2015		2016		2015		2016		
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall	
<b><u>Achiridae</u></b>									
<i>Trinectes maculatus</i>	15	34	5	12	6	7	0	0	
<b><u>Acipenseridae</u></b>									
<i>Acipenser oxyrhynchus</i>	0	0	1	0	0	0	0	0	
<b><u>Amiidae</u></b>									
<i>Amia calva</i>	10	27	10	15	0	3	1	1	
<b><u>Anguillidae</u></b>									
<i>Anguilla rostrata</i>	5	15	14	2	2	0	1	0	
<b><u>Aphredoderidae</u></b>									
<i>Aphredoderus sayanus</i>	0	6	1	7	0	4	0	7	
<b><u>Atherinopsidae</u></b>									
<i>Labidesthes sicculus</i>	2	5	3	0	0	10	2	0	
<b><u>Belonidae</u></b>									
<i>Strongylura marina</i>	1	0	3	0	0	0	0	0	
<b><u>Catostomidae</u></b>									
<i>Carpionodes cyprinus</i>	13	5	3	0	1	0	0	0	
<i>Carpionodes velifer</i>	7	7	3	5	0	1	1	1	
<i>Minytrema melanops</i>	1	30	1	7	1	2	0	0	
<i>Moxostoma poecilurum</i>	30	111	9	48	3	6	3	5	
<b><u>Centrarchidae</u></b>									
<i>Ambloplites ariommus</i>	1	5	3	3	0	2	0	0	
<i>Lepomis auritus</i>	20	34	6	13	1	5	1	2	
<i>Lepomis cyanellus</i>	2	10	5	5	1	2	0	1	
<i>Lepomis gulosus</i>	1	11	2	0	0	2	1	3	
<i>Lepomis macrochirus</i>	46	119	111	107	7	31	29	22	
<i>Lepomis megalotis</i>	63	215	59	60	4	43	18	14	
<i>Lepomis microlophus</i>	32	31	13	28	5	1	6	2	
<i>Lepomis miniatus</i>	0	0	0	0	0	0	1	0	
<i>L. miniatus x L. punctatus</i>	1	14	1	3	0	2	4	1	
<i>Micropterus haiaka</i>	7	27	11	19	1	3	4	3	
<i>Micropterus salmoides</i>	29	74	33	24	2	10	1	1	
<i>Pomoxis nigromaculatus</i>	0	3	0	2	0	0	1	1	
<b><u>Clupeidae</u></b>									
<i>Alosa chrysochloris</i>	0	0	0	0	0	0	1	0	

<i>Dorosoma cepedianum</i>	0	0	0	23	0	0	0	0
<b><u>Cyprinidae</u></b>								
<i>Ctenopharyngodon idella</i>	0	4	0	2	0	0	0	0
<i>Cyprinella venusta</i>	151	450	805	261	34	173	205	128
<i>Cyprinus carpio</i>	19	2	3	1	1	0	0	0
<i>Hybopsis winchelli</i>	4	256	24	142	6	115	17	51
<i>Macrhybopsis aestivalis</i>	0	2	0	2	0	1	0	0
<i>Notropis amplamala</i>	1	0	0	0	0	6	0	9
<i>Notropis longirostris</i>	5	20	6	7	8	12	4	9
<i>Notropis maculatus</i>	0	13	0	0	0	0	0	0
<i>Notropis texanus</i>	58	407	69	186	21	108	17	62
<b><u>Esocidae</u></b>								
<i>Esox americanus</i>	0	2	2	0	0	0	0	0
<i>Esox niger</i>	0	0	1	0	0	0	0	0
<b><u>Fundulidae</u></b>								
<i>Fundulus olivaceus</i>	0	9	2	4	0	0	1	0
<b><u>Ictaluridae</u></b>								
<i>Ictalurus furcatus</i>	0	6	4	10	0	0	0	0
<i>Ictalurus punctatus</i>	48	130	13	73	4	13	3	14
<i>Noturus leptacanthus</i>	0	9	8	9	0	3	1	1
<i>Pylodictis olivaris</i>	8	6	1	2	0	0	2	0
<b><u>Lepisosteidae</u></b>								
<i>Lepisosteus oculatus</i>	21	18	19	18	2	2	6	1
<i>Lepisosteus osseus</i>	2	0	0	2	0	0	1	0
<b><u>Mugilidae</u></b>								
<i>Mugil cephalus</i>	26	43	8	18	8	8	0	4
<b><u>Percidae</u></b>								
<i>Ammocrypta bifascia</i>	0	0	3	2	0	1	0	0
<i>Etheostoma davisoni</i>	0	0	1	0	0	0	0	0
<i>Etheostoma swaini</i>	0	4	0	2	0	0	0	0
<i>Percina austroperca</i>	2	8	2	0	2	1	1	0
<i>Percina nigrofasciata</i>	12	51	18	46	3	19	3	9
<b><u>Poeciliidae</u></b>								
<i>Gambusia holbrooki</i>	0	1	0	0	0	1	0	0

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IX.13. Number of individuals sampled of each species for each sampling event comparing day BLE and PASE sampling on the Sipsey River. Species are listed by family in alphabetical order.

	Day				Point			
	2015		2016		2015		2016	
	Sum	Fall	Sum	Fall	Sum	Fall	Sum	Fall
<b><u>Amiidae</u></b>								
<i>Amia calva</i>	0	2	0	0	0	0	0	0
<b><u>Anguillidae</u></b>								
<i>Anguilla rostrata</i>	0	0	0	2	0	0	0	0
<b><u>Aphredoderidae</u></b>								
<i>Aphredoderus sayanus</i>	0	3	0	14	1	0	0	2
<b><u>Atherinopsidae</u></b>								
<i>Labidesthes sicculus</i>	0	34	9	25	0	16	0	6
<b><u>Belonidae</u></b>								
<i>Strongylura marina</i>	1	0	5	0	0	0	0	0
<b><u>Catostomidae</u></b>								
<i>Carpiodes velifer</i>	0	4	4	0	0	0	0	0
<i>Erimyzon tenuis</i>	0	2	0	0	0	0	0	0
<i>Hypentelium etowanum</i>	1	3	0	1	0	0	0	1
<i>Ictiobus bubalus</i>	1	2	4	2	0	0	0	0
<i>Minytrema melanops</i>	0	13	2	1	0	0	0	0
<i>Moxostoma carinatum</i>	0	1	0	0	0	0	0	0
<i>Moxostoma erythrurum</i>	4	1	1	0	0	0	1	0
<i>Moxostoma poecilurum</i>	61	174	29	57	9	37	3	10
<b><u>Centrarchidae</u></b>								
<i>Ambloplites ariommus</i>	4	2	4	7	0	0	0	0
<i>Lepomis cyanellus</i>	0	0	1	0	0	4	0	0
<i>Lepomis gulosus</i>	1	9	0	3	0	1	0	0
<i>Lepomis macrochirus</i>	77	244	58	197	13	42	8	40
<i>Lepomis megalotis</i>	164	317	111	475	29	82	12	121
<i>Lepomis microlophus</i>	9	35	4	25	1	6	0	6
<i>Lepomis miniatus</i>	1	9	5	24	0	3	0	1
<i>Micropterus henshalli</i>	43	58	34	50	4	10	0	9
<i>Micropterus salmoides</i>	28	36	21	30	4	6	4	5
<i>Pomoxis annularis</i>	5	7	1	1	0	0	0	0
<i>Pomoxis nigromaculatus</i>	0	6	1	2	0	1	0	1
<b><u>Clupeidae</u></b>								
<i>Dorosoma cepedianum</i>	66	25	28	6	2	3	2	0
<i>Dorosoma petenense</i>	0	0	2	0	0	0	0	0

**Cyprinidae**

<i>Campostoma oligolepis</i>	0	0	0	0	0	0	0	1
<i>Cyprinella venusta</i>	48	238	140	151	5	74	19	35
<i>Hybopsis winchelli</i>	0	56	1	73	0	0	0	4
<i>Notemigonus crysoleucas</i>	0	0	0	0	0	0	0	1
<i>Notropis ammophilus</i>	0	0	1	0	0	0	0	0
<i>Notropis amplamala</i>	0	0	0	0	0	0	0	1
<i>Notropis atherinoides</i>	7	24	39	69	2	1	0	36
<i>Notropis stilbius</i>	1	5	0	4	0	1	0	0
<i>Notropis texanus</i>	10	58	9	39	0	34	0	6
<i>Notropis volucellus</i>	1	1	0	1	0	1	0	0
<i>Opsopoeodus emiliae</i>	0	3	1	10	0	0	0	3
<i>Pimephales vigilax</i>	1	45	1	31	0	10	1	16

**Esocidae**

<i>Esox americanus</i>	0	0	0	0	1	0	0	0
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**Fundulidae**

<i>Fundulus notatus</i>	1	6	1	11	0	1	0	5
<i>Fundulus olivaceus</i>	4	10	9	17	1	5	5	3

**Ictaluridae**

<i>Ictalurus furcatus</i>	0	1	0	0	0	1	0	0
<i>Ictalurus punctatus</i>	4	7	8	10	2	0	2	3
<i>Noturus leptacanthus</i>	0	0	3	1	0	0	1	1
<i>Pylodictis olivaris</i>	8	4	10	1	9	1	2	0

**Lepisosteidae**

<i>Lepisosteus oculatus</i>	10	4	1	2	2	0	1	0
<i>Lepisosteus osseus</i>	2	1	1	1	0	0	0	0

**Moronidae**

<i>Morone chrysops</i>	0	0	0	1	0	0	0	0
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**Percidae**

<i>Ammocrypta meridiana</i>	0	1	0	0	0	0	0	0
<i>Crystallaria asprella</i>	0	0	0	0	0	0	1	0
<i>Etheostoma artesia</i>	1	0	0	0	0	0	0	0
<i>Etheostoma histrio</i>	0	0	0	1	0	0	0	0
<i>Etheostoma stigmaeum</i>	0	1	0	13	0	0	0	2
<i>Percina kathae</i>	3	0	2	13	0	0	0	2
<i>Percina lenticula</i>	1	0	0	0	0	0	0	1
<i>Percina nigrofasciata</i>	4	13	2	26	0	0	2	2
<i>Percina sciera</i>	0	3	1	5	0	2	0	0
<i>Percina suttkusi</i>	1	2	0	0	0	0	0	0

**Petromyzontidae**

<i>Ichthyomyzon gagei</i>	0	1	0	0	0	0	0	0
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**Sciaenidae**

*Aplodinotus grunniens*            7    8    5    0            0    3    1    0