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Distribution, Status, and Life History Aspects of Two Rare Logperches, Percina burtoni and Percina apina

Jeffrey W. Simmons
Tennessee Valley Authority, jwsimmons0@tva.gov

Distribution, Status, and Life History Aspects of Two Rare Logperches, Percina burtoni and Percina apina

Abstract

The Blotchside Logperch, *Percina burtoni*, and Tennessee Logperch, *Percina apina*, are Tennessee-Cumberland River drainage endemics that have experienced range reductions due to anthropogenic influences. All known collection records were gathered to fully describe the historical distribution of these species for comparison to their currently inhabited range. Discussion of major impacts to rivers that contain or contained these species is included for an understanding of factors that may have influenced contemporary distributions. Extensive field surveys were conducted during 2014 to 2017 to aid in determination of the current status and distribution of populations. New information of previously unreported or undetected populations, population status, life history observations, and longevity estimates are presented.

Keywords

Tennessee-Cumberland Rivers, Percidae, natural history records, habitat use

Cover Page Footnote

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INTRODUCTION

The Tennessee and Cumberland River drainages are among the most biologically diverse aquatic ecosystems in North America and also rank as areas of highest imperilment (Elkins et al. 2019). Before extensive impoundment of these large river drainages, collectors were already documenting faunal declines due to stressors such as siltation and various forms of industrial pollution (Jordan 1889; Ortmann 1918). We are only able to surmise the previous distribution of many species within these systems by piecing together limited historical collection records. The Blotchside Logperch, *Percina burtoni*, is one of many fishes endemic to the Tennessee and Cumberland River drainages that has experienced a range reduction resulting from anthropogenic influences. Historical distributions obtained from museum records demonstrate that *P. burtoni* occupied a majority of the Tennessee River drainage and portions of the upper Cumberland River drainage. The last individual vouchered from the Cumberland River drainage was in 1947 and the last specimens obtained from several Tennessee River tributaries were during the mid to late 1800s and early 1900s. Recent studies identified two distinct lineages within *P. burtoni*, resulting in a new species, Tennessee Logperch, *Percina apina*, endemic to the Duck River system and Whiteoak Creek (George et al. 2006; Near et al. 2017). The discovery of *P. apina* as a distinct lineage created a further need for understanding the current status and threats to this narrowly distributed species, in addition to the already sparsely distributed populations of *P. burtoni* occurring in upstream Tennessee River tributaries.

Percina burtoni and *P. apina* are specialized insectivores that feed by flipping small cobbles with their padded snouts to prey upon attached or dislodged insects (Greenberg 1991; Heacock 1995). This feeding technique is beneficial because it takes advantage of a food source that is largely unexploited by other fishes (Jenkins and Burkhead 1994), but can also be detrimental if these habitats become degraded. Therefore, impoundments, unnatural sediment loads, or other pollutants that may alter feeding habitats or reduce prey availability are factors that can contribute to the loss of populations. In the Little River, *P. burtoni* was most often observed in flowing areas containing gravel and cobble substrates with an average water depth of 0.6 m (Heacock 1995; Jett 2010). Both of these studies observed that individuals were absent or occurred at lower densities in areas with increased turbidity or sediment accumulation. My observations, and those of others (Jenkins and Burkhead 1994), indicate that *P. burtoni* and *P. apina* may naturally occur at low densities, thus are considered uncommon or rare throughout their range. This may be due to their extremely narrow habitat preferences which are often separated by long stream reaches (pers. obs.).

Little information is available for aspects of life history such as spawning habitats, length of spawning period, differences in habitat use among sub-adults and reproductive and non-reproductive adults, and longevity. Jenkins and Burkhead (1994) discuss collection of gravid *P. burtoni* females and peak-colored nuptial males in an area with clean, loose gravel and moderate to strong flow in late April when the water temperature was 19 °C. Larvae of *P. burtoni* propagated in captivity were first collected during early April when water temperatures were near 15 °C (Ruble et al. 2009). Demersal juveniles were observed flipping small gravel similar to adult feeding behavior, indicating that clean rock substrates are important to survival at an early age (Rakes and Shute 2007). Etnier and Starnes (1993) and Jenkins and Burkhead (1994) suggested that *P. burtoni* lived at least four years.

The objectives of this paper are to: 1) Provide a summary of major impacts to river systems that contain or contained *P. burtoni* or *P. apina* that may have influenced the contemporary distribution of each species; 2) Present all known collection records to describe the historical and current distribution of these species; 3) Assess status of populations through spatial and temporal examination of these data; 4) Describe habitat use and life-history observations from extensive field surveys conducted from 2014 to 2017; and 5) Provide longevity estimates for both species. For ease of reading, the first four objectives are presented together for each river system of occurrence within the Tennessee and Cumberland River drainages.

METHODS

Occurrence data were obtained from an accumulation of collection records from natural history museums, the North Carolina Wildlife Resources Commission (NCWRC), the Virginia Department of Game and Inland Fisheries (VDGIF), and the Tennessee Valley Authority (TVA). Additional records were from published literature, theses, and credible unpublished reports which are cited as appropriate throughout the body of the manuscript. Remaining records were obtained from field sampling during this study. Institutional abbreviations follow Sabaj (2019) for all referenced museum records, except that UNAF refers to the University of North Alabama, Florence Zoological Collection.

Specifically, the following data were used: material examined by Near et al. (2017) which consisted of 221 specimens (117 lots) from 17 museum collections (these data are available from the Dryad Digital Repository, http://doi.org/10.5061/dryad.rm607); museum records (13) from the North Carolina State Museum of Natural Sciences (NCSM), Raleigh, North Carolina; Tulane University Museum of Natural History (TU), Belle Chase, Louisiana; and University of Alabama Ichthyological Collection (UAIC), Tuscaloosa, Alabama (last accessed through the Fishnet2 Portal, http://www.fishnet2.net, 2020-10-21); NCWRC records (18) from the South Toe River of the French Broad River system; VDGIF records from the Clinch River system (118) and Holston River system (45); and TVA records (130) from fish community surveys conducted from 1990 to 2019 throughout the Tennessee River drainage.

From the late 1980s until present, TVA has sampled fish communities in free-flowing streams and rivers throughout the Tennessee River drainage. Sample stations (874) were selected throughout each major river system within the drainage and have been consistently sampled on an alternating rotation. Sampling methods were designed to deplete habitats to maximize species detection at a site. Sampling was conducted with backpack-mounted electrofishers, seines, and boat electrofishers. Results of this sampling were quantified using Index of Biotic Integrity (IBI) methodology (Karr et al. 1986) that was adapted to the Tennessee River system. These surveys have provided much data on the distribution of *P. burtoni* and *P. apina*. Discussion of water quality conditions within the range of these species include results of these surveys.

During this study, surveys for *P. burtoni* and *P. apina* were conducted from 2014 to 2017, in addition to TVA fish community assessments in areas occupied by these species. Surveys for *P. burtoni* were primarily focused on determining its current status and distribution in streams and rivers where it had not been collected in recent times. Sampling for *P. apina* was conducted throughout the entirety of its range. This work provided updated status and distribution information, and allowed for acquisition of specimens and tissues used for its description (Near et al. 2017). Sampling was conducted by snorkeling, backpack electrofishing, and backpack electrofishing into a seine. Some locations were sampled on multiple occasions to increase probability of detection. During reproductive periods, water column velocity (cm/sec, 0.6 depth from the water's surface), bottom velocity (cm/sec) and water depth (m) were recorded with a portable flow meter and top-setting wading rod at locations of nuptial *P. burtoni* in some rivers. Life history and habitat use observations of both species were conducted while snorkeling. All voucher specimens collected were deposited in the Division of Vertebrate Zoology Ichthyology Collection (YPM ICH), Peabody Museum of Natural History, Yale University, New Haven, Connecticut.

A Geographic Information System, ESRI ArcGIS Version 10.5.1 software, was used for: spatial and temporal analyses of occurrence data; determination of ecoregions occupied by each species (USEPA 2017); and calculation of drainage area, determination of land cover (2016 National Land Cover Dataset for the United

States, Yang et al. 2018) and human population densities (2010 National Census data, U.S. Census Bureau 2020) for systems occupied by *P. apina*.

To estimate longevity of *P. apina* and *P. burtoni*, length data were analyzed using non-parametric kernel density estimation. Smoothed length distributions identified local maxima (modal peaks) for estimates of age classes using the "stats" package in R (R Core Team 2019). Kernel density estimation was conducted for three different time periods for modal comparisons: the entire data set, March through June, and July through December. Lengths were obtained from individuals examined in Near et al. (2017). These data are available from the Dryad Digital Repository (http://doi.org/10.5061/dryad.rm607).

RESULTS AND DISCUSSION

TENNESSEE RIVER DRAINAGE- *Percina burtoni*

Within the Tennessee River drainage, *P. burtoni* has been documented in 11 major tributary systems. Populations in three of these systems are presumed extirpated and the remainder are narrowly distributed. The following accounts are organized from northeast (upstream) to southwest (downstream) through the Tennessee River drainage in river systems that contain or historically contained *P. burtoni*.

Clinch River System

Clinch River--The Clinch River originates in southwestern Virginia and flows for 483 river kilometers (rkms) before its confluence with the Tennessee River. Of its total length, only 238 rkms remain free-flowing due to inundation by Norris, Melton Hill, and Watts Bar reservoirs (Figure 1). The Clinch River upstream of Norris Reservoir supports a tremendous amount of biodiversity and is considered to be one of the most ecologically important sections of river in North America (Zipper et al. 2014). This portion of the Clinch River harbors 20 mussel and 5 fish species federally listed as threatened or endangered, many of which represent the only remaining viable populations (Jones et al. 2014; Zipper et al. 2014). Threats to these have included several major point source pollution events and long term pollution from coal extraction and processing (Cairns et al. 1971; Hull et al. 2006; Zipper et al. 2014; Ahlstedt et al. 2016). Tributaries, particularly Copper Creek, have been important for sustaining fish diversity that could eventually recolonize the Clinch River following these perturbations (Jenkins and Burkhead 1994).

All historical and current records of *P. burtoni* from the Clinch River system are from areas upstream of Norris Reservoir. The first collections of *P. burtoni* from the Clinch River system were made during August 1967 in Scott County, Virginia from the upper Clinch River (CUMV 63173) and Copper Creek (CUMV 63131). Until the 1990s, most collections of *P. burtoni* were from these same general areas. Recent records demonstrate that it is much more widely distributed throughout the mainstem Clinch River and occupies many Clinch River tributaries, including a large reach of Copper Creek (Figure 1).

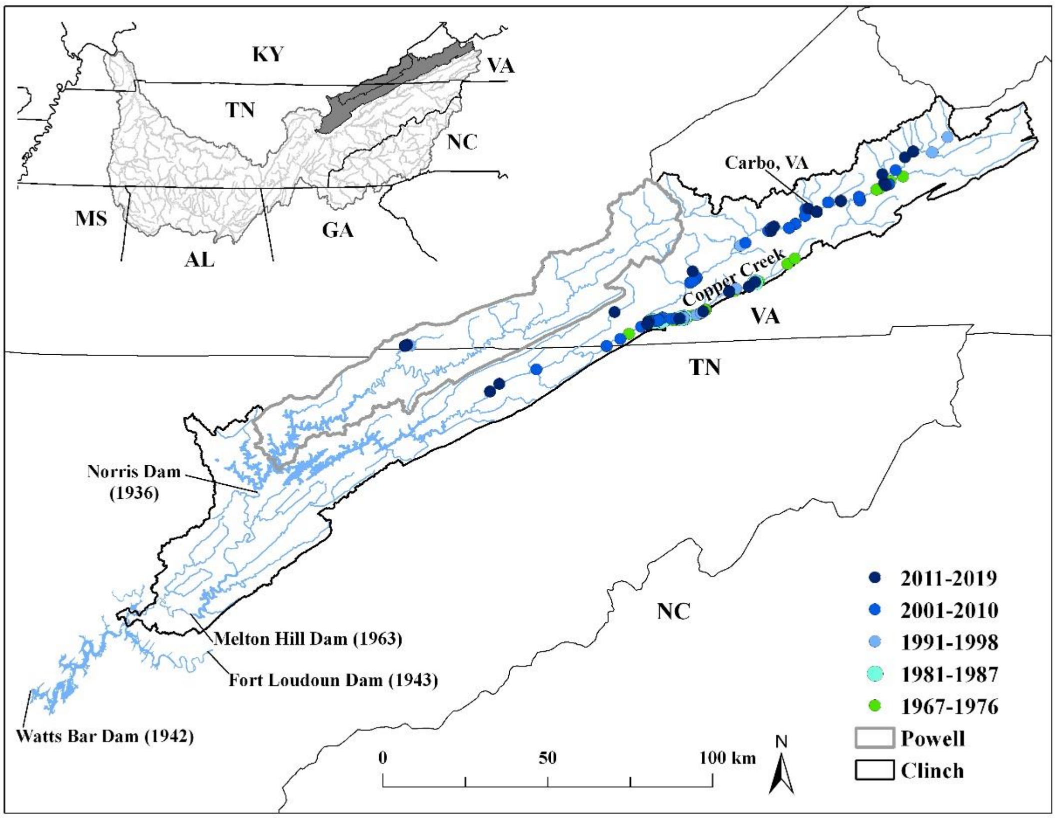

Figure 1. Historical and current distribution of *Percina burtoni* in the Clinch River system.

During this study, an individual (YPM ICH 028184) was collected from Dumps Creek near Carbo, Virginia, at an area that has a history of pollution. The world's largest coal processing plant was operated here and discharge from coal washing operations caused major sedimentation problems (Cairns et al. 1971; Jenkins and Burkhead 1994). One of the most significant point source pollution events that affected the upper Clinch River was collapse of a fly ash lagoon, which released approximately 492,000 m^3 of highly caustic alkaline slurry into Dumps

Creek. This entered the Clinch River at rkm 431 and resulted in a fish kill that extended about 145 rkms downstream (Cairns et al. 1971).

Powell River--Surface coal mining has negatively affected the Powell River, a major tributary of the upper Clinch River, since at least the 1950s (Ahlstedt et al. 2016). Mining operations disturbed one-third of the upper watershed area from 1984 to 2011, with coal production peaking from 2000 to 2005 (Zipper et al. 2016). Early documentation of the effects to fish communities were from a 1968 sample at Powell River km 299. Only eight fish species were collected using rotenone and no benthic macroinvertebrates were detected in lab processed benthic samples (TVA 1968).

The first collection of *P. burtoni* from the Powell River system was in 1997, where it was collected in Indian Creek, a tributary to the lower free-flowing portion of the Powell River (USNM 351110.5259). It was also encountered near this locality in 2008 during a TVA stream survey and most recently in 2017 and 2018 by VDGIF and TVA (M. Pinder, VDGIF, pers. comm. 2018; TVA unpub. data). This population was presumably more widely distributed within the Powell River system before water quality deterioration from coal mining. Because Norris Reservoir impounds more than 210 rkms between the lower free-flowing sections of the Clinch and Powell rivers, contemporary immigration from the Clinch River is unrealistic. However, high IBI scores in the Powell River near the confluence of Indian Creek indicate that conditions may be favorable for reintroduction efforts (TVA unpub. data).

Holston River System

Aquatic fauna in the Holston River system have been impacted by habitat loss and fragmentation from dams and chronic contamination from point sources. Important ichthyological collections made prior to these impacts only occurred in portions of the river system, leaving many areas under surveyed (Hughes 1994). Historical collections of *P. burtoni* before these changes were from the North and South Forks of the Holston River.

North Fork Holston River--The North Fork Holston River system is the only portion of the Holston River system that has not been altered by large impoundments (Figure 2). However, this river has experienced long term pollution. Mathieson Alkali Works began production of sodium carbonate alongside the North Fork Holston River at Saltville, Virginia in 1882 and eventually merged with Olin Industries in the 1950s (TVA 1983a). From 1950 to 1972, the plant at Saltville produced chlorine gas and sodium hydroxide through an electrolytic process that used mercury as an electrode (USEPA 2012). Electrolytic cells lost up to 45 kg of mercury per day during 22 years of operation (Carter 1977). Mercury contaminated wastewater, along with other chemical production wastes, were released into unlined settling ponds that discharged to the North Fork Holston River (Hildebrand et al. 1980). In 1970, elevated levels of mercury were detected in fish tissues in the North Fork Holston River and the plant ended operation in 1972 after deciding that it would be too costly to comply with water quality regulations (USEPA 2012). A 2005 study found mercury in sediments, interstitial water, and clam tissues up to 87 rkms downstream of Saltville, indicating that it was still entering the river from remediated holding ponds (Echols et al. 2009).

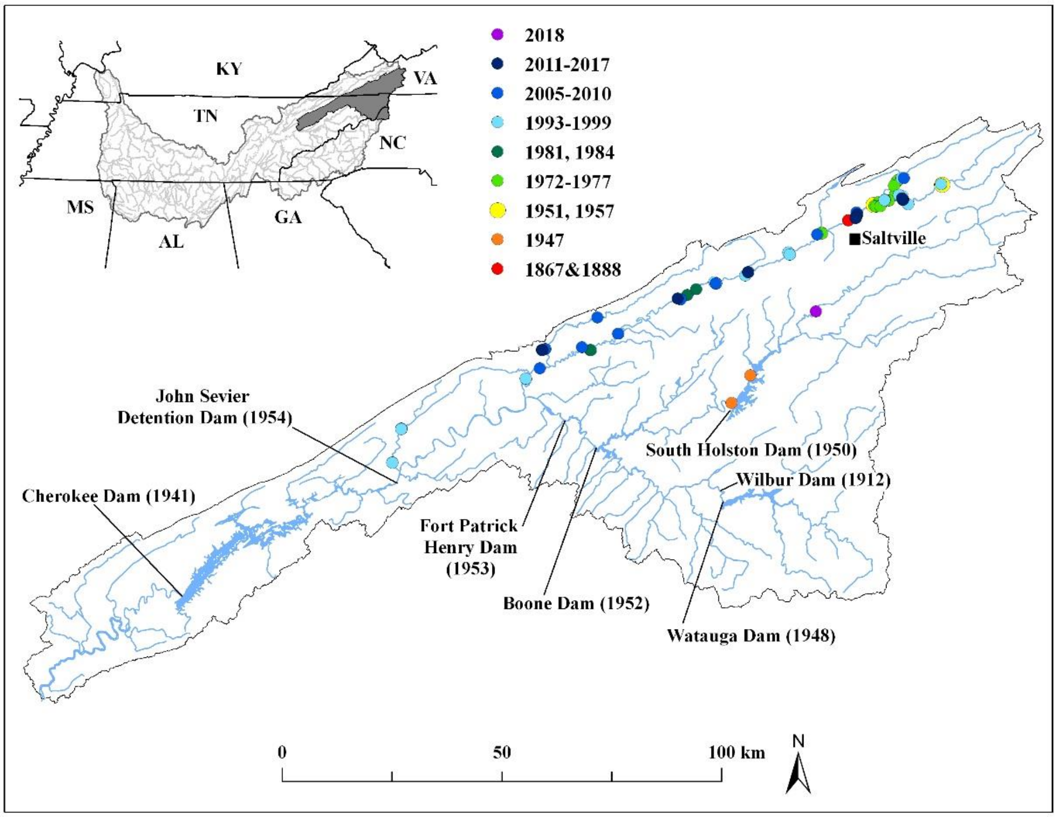

Figure 2. Historical and current distribution of *Percina burtoni* in the Holston River system.

The first collection of *P. burtoni* was from the North Fork Holston River at Saltville in 1867 (ANSP 13707, *paratype*; Cope 1869). Jordan (1889) subsequently collected *P. burtoni* at Saltville and noted that the river was swift but not very clear. Hill et al. (1980) assessed fish and benthic invertebrate communities in the North Fork Holston River from 1971 to 1977 at various stations spanning 173 rkms.

During the study period, *P. burtoni* was consistently collected at a reference site (rkm 147) upstream of the chemical plant at Saltville. Towards the end of the study, some faunal recovery was documented in reaches downstream of the retired chemical plant, including *P. burtoni* at rkm 125.5 (YPM ICH 28200).

Prior to 1977, *P. burtoni* had only been collected in the North Fork Holston River upstream of Saltville. During the last several decades, *P. burtoni* has been frequently collected from locations spanning 150 rkms in the mainstem North Fork Holston River and within three of its tributaries, demonstrating its ability to recolonize a river as conditions improved (Figure 2).

South Fork Holston River--Following collections by Cope and Jordan in the North Fork Holston River, *P. burtoni* was not collected again in the Holston River system until September 1947 when R.M. Bailey and others collected the first and last specimens from the South Fork Holston River (UMMZ 157579, 157611). These collection localities were inundated shortly thereafter by completion of South Holston Reservoir (Figure 2).

During July 2018, VDGIF biologists collected an adult *P. burtoni* from the Middle Fork Holston River (M. Pinder and T. Lane, VGDIF, pers. comm. 2018), representing the first record from this river and the first record from the South Fork Holston River drainage since completion of South Holston Dam in 1950 (Figure 2). Jordan (1889) noted that the Middle Fork Holston River flowed through pastureland and that the water was discolored due to turbidity. Cox (1986) listed excessive crop and pasture land erosion and input of animal waste from cattle operations as causes of high suspended solid loads in the Middle Fork Holston River, which caused turbidity even during base flow conditions. Thus, the July 2018 discovery of *P. burtoni* in the Middle Fork Holston River is significant and demonstrates that a population can persist in an impacted environment for long periods without detection.

Big Creek--An isolated population of *P. burtoni* was discovered in Big Creek by TVA and Tennessee Wildlife Resources Agency biologists during independent stream surveys in 1995. This stream flows into a small impoundment on the Holston River at rkm 175.5 that was constructed to maintain a cooling water supply for John Sevier Fossil Plant. This occurrence indicates a once larger and more connected population in the Holston River system. Although no individuals have been collected from the mainstem of the Holston River, a hybrid *P. burtoni* x *Percina sciera* (UT 91.6306) was collected during a TVA survey at Holston River km 190 in 1997. This could suggest periodic emigration from Big Creek or downstream immigration from the North Fork Holston River. During 1997, *P.*

burtoni was collected again in Big Creek during TVA stream surveys, but was not collected in five subsequent surveys from 2003 to 2016 (TVA unpub. data). During 2017, several locations in Big Creek were sampled unsuccessfully for *P. burtoni*. Big Creek flows through an area dominated by crop fields and pastures and lacks wooded riparian areas throughout a large portion of the drainage. At all locations sampled, substrates were embedded or covered by sediments.

French Broad River System

The French Broad River originates in the Blue Ridge Mountains of North Carolina and is fed by several large tributaries including the Nolichucky, Pigeon, and Little Pigeon rivers (Figure 3). Historically, water quality of the French Broad River was extremely poor due to tannery, paper mill, and rayon plant discharges (TVA 1945). Biochemical oxygen demands required to break down organics within these discharges resulted in low dissolved oxygen in the French Broad River from the confluence of the Davidson River downstream to the backwaters of Douglas Reservoir, a reach encompassed by > 180 rkms (TVA 1945).

The three major sub-basins of the French Broad River system have also experienced past water quality issues. The Pigeon River was severely polluted from paper mill effluents for most of the twentieth century and was unable to sustain fish in many reaches due to low dissolved oxygen levels (Hess and Tarzwell 1942; TVA 1945; Saylor et al. 1993). Rapid development of Gatlinburg, Pigeon Forge, and Sevierville led to degradation of the Little Pigeon River (Parmalee 1988; Doosey 2001). The Nolichucky River watershed was extensively mined for mica, feldspar, and kaolin and experienced long-lasting periods of extreme sediment loading (Duda and Penrose 1980). Three large impoundments have been constructed in the French Broad River system, none of which are in the vicinity of historical records of *P. burtoni* (Figure 3).

Swannanoa River--*Percina burtoni* was first collected within the French Broad River system by D.S. Jordan during August 1888 in the Swannanoa River near Asheville, North Carolina (USNM 40601). He described the river as clear with gravelly and rocky shoals which were excellent for collecting (Jordan 1889). The holotype was also collected from here in August 1934 (ANSP 70701) and was the last collection of *P. burtoni* from the Swannanoa River (Figure 3). Domestic sewage and sedimentation from increased urbanization were pollution concerns in the Swannanoa River shortly thereafter (TVA 1945).

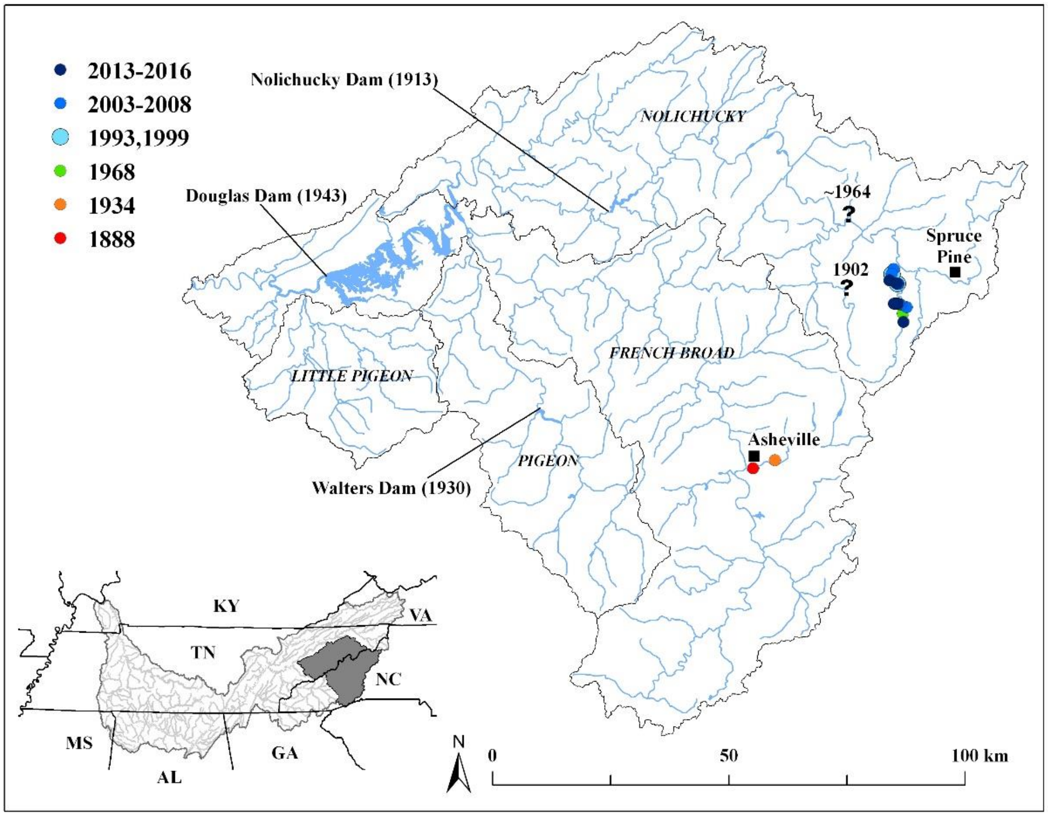

Figure 3. Historical and current distribution of *Percina burtoni* in the French Broad River system.

Nolichucky River sub-basin--The upper Nolichucky River sub-basin lies within the Spruce Pine Mining District where mica, feldspar, and kaolin extraction began in 1870 and escalated to large industrial operations by the 1940s (Duda and Penrose 1980). These operations were responsible for significant point source discharges of tailing wastes to the North Toe River near Spruce Pine. Biological surveys by Mullican et al. (1960) indicated that sediment loads in the Nolichucky River had reduced diversity of fish and benthic macroinvertebrates to the most tolerant taxa, all of which occurred in low densities. Extreme sediment loads entered the North Toe River from abandoned mines and waste tailing piles even after passing the 1965 Federal Water Pollution Control Act (Duda and Penrose 1980). Extensive reclamation efforts at mine sites during the 1980s greatly reduced sediment input (TVA 2002). TVA has documented significant recovery of the fish community during the past few decades from areas historically referred to as "biological deserts" within the Nolichucky River system (TVA 2002; TVA unpub. data).

Bean (1903) reported a collection of *Percina caprodes* by H.H. Brimley from the Cane River, a tributary to the Nolichucky River, Yancey County, North Carolina on 4 October 1902 (Figure 3). It is likely that the specimen was *P. burtoni* since *P. caprodes* has not been documented in the Nolichucky River sub-basin in North Carolina (TVA unpub. data). Menhinick et al. (1974) listed a logperch record from the North Toe River near Pigeon Roost from 1960s surveys conducted by the NCWRC. It is probable that this was also *P. burtoni*.

The first vouchered specimen of *Percina burtoni* from the Nolichucky River sub-basin was collected from the South Toe River in 1968 (NCSM 88804). The South Toe River flows into the North Toe River, which converges with the Cane River to form the Nolichucky River. Only areas within the lower portion of the South Toe River watershed were mined, thus a majority of this river system was spared from the resulting impacts (Duda and Penrose 1980). After initial collection of *P. burtoni* in 1968, no records existed until the 1990s, most likely due to lack of surveys. Recent surveys indicate that the only extant population of *P. burtoni* within the French Broad River system is stable (NCWRC unpub. data), where it is distributed throughout at least 20 rkms of the South Toe River (Figure 3). Expansion of this population is plausible if water quality conditions continue to improve in the North Toe River.

Little River System

The headwaters of the Little River are completely within the Great Smoky Mountains National Park (GSMNP) where they drain a portion of the Blue Ridge Mountains in Sevier and Blount counties, Tennessee. Before establishment of the National Park in 1940, the upper reaches of the watershed were logged, resulting in sedimentation and increased stream temperatures (King 1937). A tannery operated alongside the Little River at Walland from 1902 until it was destroyed by a fire in 1931. It was not rebuilt due to ongoing litigation over the use and pollution of the Little River (Stephan 1941). Current threats to water quality of the Little River include rapid human population growth in Blount County, which has intensified residential and commercial development within the watershed (Zhu and Li 2013).

The first collection of *P. burtoni* in the Little River was during 1968 at Walland (UT 91.272). Focused surveys for *P. burtoni* found it commonly distributed from Townsend, downstream throughout a reach spanning ~22 rkms (Heacock 1995; Jett 2010). During these surveys, the highest densities were within the upstream 13 rkms of its distribution. Both studies documented a negative correlation of *P. burtoni* with increasing turbidity levels and noted that water clarity of the Little River decreased downstream of Ellejoy Creek. Land use analyses of the Little River watershed determined that the greatest amounts of pasture, row crops, and beef production were in the Ellejoy Creek system (TVA 2003). Estimated soil loss from this landscape was 4,167 metric tons per year. Sediment loads from Ellejoy Creek are likely a factor in the paucity of *P. burtoni* observations in the Little River downstream (Figure 4). However, two sub-adults were collected in lower Ellejoy Creek in 2018 (TVA unpub. data), indicating that juveniles may tolerate somewhat degraded habitats.

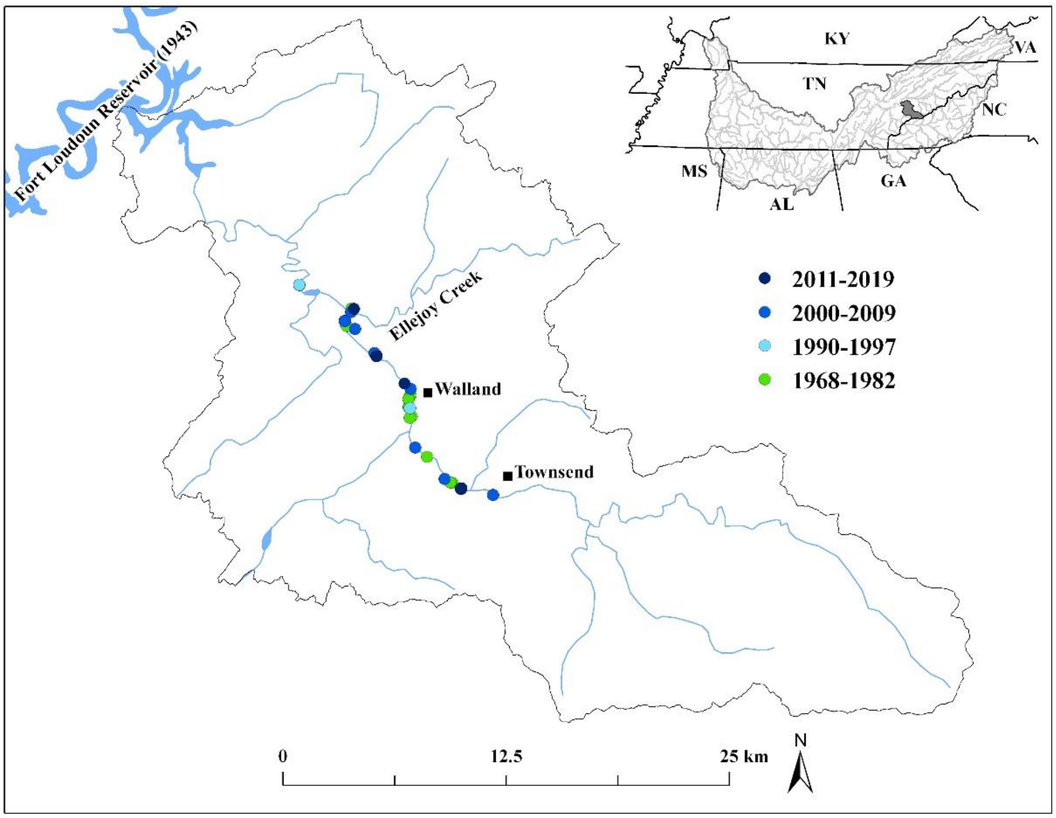

Figure 4. Historical and current distribution of *Percina burtoni* in the Little River system.

The Little River still supports relatively high densities of *P. burtoni*, where it has been observed on frequent occasions during recent years throughout a majority of its historically occupied range (Figure 4). Continued water quality protection afforded by the GSMNP will be beneficial in maintaining conditions suitable for persistence of *P. burtoni* and many other state and federally protected aquatic species.

Little Tennessee River System

The Little Tennessee River system is one of the most regulated tributaries within the Tennessee River drainage (Figure 5). Very few ichthyological collections were made in the Little Tennessee River system before a majority of the mainstem Little Tennessee River and several of its large tributaries were affected by impoundments (Winfield 1976). Several of the mainstem dams resulted in sporadic flow regimes and cold water discharges (Dendy and Stroud 1949; Boles 1968). During 1979, TVA closed the gates on the controversial Tellico Dam, which impounded the last remaining free-flowing section of the lower Little Tennessee River.

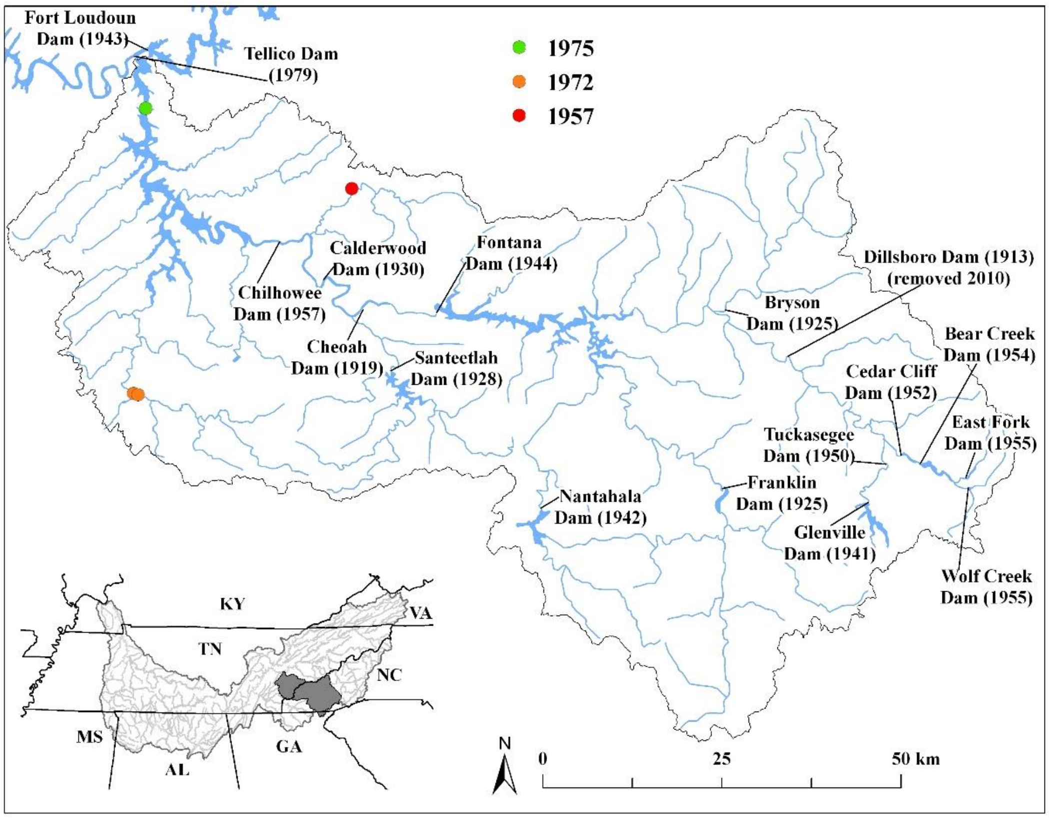

Figure 5. Historical distribution of *Percina burtoni* in the Little Tennessee River system.

Four records of *P. burtoni*, only one of which is represented by voucher specimens, have been reported from the Little Tennessee River system (Figure 5). Two specimens (UMMZ 183740) were collected during a 1957 rotenone treatment of Abrams Creek in an effort to eliminate sport fish competition from "rough fish" as detailed in Lennon and Parker (1959). This rotenone application occurred just before completion of Chilhowie Reservoir, which subsequently eliminated any potential recolonization of Abrams Creek from the Little Tennessee River. Winfield (1976) reported seeing individuals at two locations in the Tellico River while snorkeling during August 1972. The last record of *P. burtoni* from the system was observed during 1975 at Little Tennessee River km 10.9, which is now inundated by Tellico Reservoir (Etnier 1994).

Conservation Fisheries, Inc. (CFI) propagated *P. burtoni* from individuals captured from the Little River. Progeny were released into the Tellico River at the mouth of Quarry Creek during July 2008 and into Abrams Creek during 2017 in an effort to reestablish populations within the Little Tennessee River system (Petty et al. 2009; P.L. Rakes, CFI, pers. comm. 2017). Additional stocking of *P. burtoni* into Abrams Creek has been halted until further genetic resolution of brood stock sources are determined and translocation and monitoring plans are developed (M. Kulp, GSMNP, pers. comm. 2017).

Hiwassee River System

Aside from the Little Tennessee River system, the Hiwassee River system is the most impounded Tennessee River tributary (Figure 6). Virtually the entirety of the Hiwassee River and its major tributaries were altered by impoundments by 1943. Not only did these impoundments eliminate large sections of riverine habitats, they also altered remaining free-flowing areas through unnatural flow regimes, periods of low dissolved oxygen, diversion of water from sections of streambed, and cold-water discharges (Higgins and Brock 1999). The Ocoee River, the largest tributary to the Hiwassee, flows through the Copper Basin, an area considered as one of the most complicated environmental restoration sites in the United States due to the extent of impacts from mining and sulfuric acid production (Faulkner et al. 2006; Bell et al. 2007; Mathews and Harden 1999).

The only records of *P. burtoni* from the Hiwassee River system are from Spring Creek and the Hiwassee River near the confluence of Spring Creek (Figure 6). The first individual was collected in 1971 from Spring Creek (AUM 4938). *Percina burtoni* has only been seen twice in the mainstem of the Hiwassee River. The first collection was on 10 May 1975 downstream of the mouth of Spring Creek (YPM ICH 028199) as reported by Myhr (1977). The second was reported by Rakes and Shute (2007) who observed a few individuals in Hiwassee River just upstream of the confluence of Spring Creek in 2004. Additional individuals were seen within the lower 5.5 rkms of Spring Creek, most of which were near Long Ford (Rakes and Shute 2007). During May 2015, the lower few kilometers of

Spring Creek were surveyed while snorkeling and backpack electrofishing and no individuals were seen. Five individuals were located while snorkeling in June 2015 just downstream of Long Ford. These fish, three adults and two sub-adults, were observed in a slow run that flowed into a pool. Water depths were less than a meter and water willow beds bordered the stream margin. Substrates consisted of gravel and cobble, and were unlike other areas surveyed, which were predominantly bedrock, large cobble, and boulders. It appears that preferred habitats of this species are lacking in Spring Creek, thus their distribution appears to be quite patchy. Spring Creek flows through Cherokee National Forest which offers some level of watershed protection, although fine sediments were prevalent in slower flowing areas which indicated areas of erosion within the watershed.

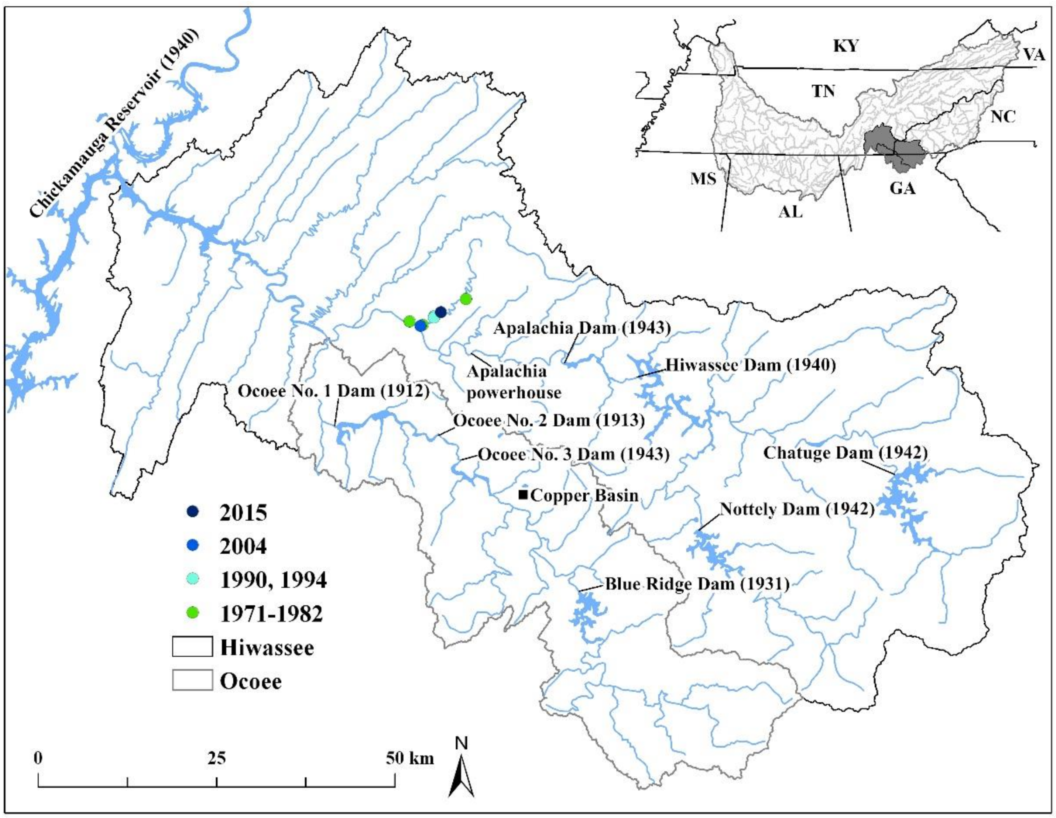

Figure 6. Historical and current distribution of *Percina burtoni* in the Hiwassee River system.

Spring Creek enters the Hiwassee River downstream of Apalachia powerhouse (Figure 6). A 20 km section of the Hiwassee River is diverted through a tunnel running through the mountain from Apalachia Reservoir to the downstream powerhouse. Streamflow in the bypass reach below Apalachia Dam is primarily maintained from tributary input and is a remnant of the warm-water stream environment that existed before construction of the dam. The powerhouse discharges cold water and the Hiwassee River downstream is now managed as a trout fishery. Cold water temperatures probably restrict *P. burtoni* from expanding to other areas within the Hiwassee River. Fish surveys have been consistently conducted by TVA at several stations on the Hiwassee River upstream and downstream of Spring Creek during the past 20 years and *P. burtoni* has not been detected. The few observations of this species in the mainstem Hiwassee at the mouth of Spring Creek may have been during periods of the year when natural stream temperatures were similar to that in the tailwater, thus allowing seasonal use of this portion of the river. Furthermore, no extant population of *P. burtoni* occurs within a regulated tailwater, further indicating its requirement for unregulated rivers.

South Chickamauga Creek System

A majority of the South Chickamauga Creek system flows through Catoosa County, Georgia before entering Tennessee where it flows into the Tennessee River at Chattanooga. Historical collection records from South Chickamauga Creek include *Moxostoma lacerum* (type locality), *Erimonax monachus*, *Noturus flavipinnis* and *Allohistium cinereum*, providing evidence that this stream once supported many more environmentally sensitive fishes than it does today (Jordan and Brayton 1878; Taylor 1969; Dahlberg and Scott 1971). Long term fish community monitoring at sites throughout the South Chickamauga Creek system indicate that some reaches have maintained good water quality while others have declined (TVA unpub. data).

While searching ichthyological collections for *P. burtoni*, a specimen was discovered from Whitfield County, Georgia (MCZ 24811). Streams in present day Whitfield County primarily drain to the Conasauga River (Coosa River system, Mobile Basin), but a portion of the northwest corner drains to the South Chickamauga Creek system. The specimen tag only included the county, state, and collector (A. Gerhardt). It also indicated that the specimen was received by the Museum of Comparative Zoology (MCZ) from the U.S. National Museum in 1861. Evidently, Alexander Gerhardt was a naturalist from Whitfield County, Georgia who frequently provided material to various taxonomists (Agassiz 1857; Lea 1862; Lea 1863). *Etheostoma caeruleum* (MCZ 24711) and *Cottus bairdii* (USNM 3185.5; MCZ 13646) were also from this collector and contained the same information, presumably because they were collected from the same locality and date. From this limited information, the only logical locale for these collections was from the South Chickamauga Creek system, since these species do not occur in the nearby Conasauga River.

Paint Rock River System

The upper Paint Rock River system has remained largely unaltered and unpolluted due to its isolation on the Cumberland Plateau Escarpment. Thus, it still supports a large number of rare fish and mussel species and is considered a refuge for biological diversity in the Tennessee River drainage (O'Neil et al. 2013). The Paint Rock River is formed by two large tributaries, Hurricane Creek and Estill Fork, where it flows for 96 rkms before its confluence with the Tennessee River (Figure 7). The lower 21 rkms of the Paint Rock River are impounded by Wheeler Reservoir, a Tennessee River impoundment that was completed in 1936. The upper portion of the watershed is predominantly forested while much of the Paint Rock River valley is utilized for various forms of agriculture. This river system was negatively affected in the past by small impoundments, stream channelization, erosion and agricultural runoff (Ahlstedt 1995; Fobian et al. 2014). However, recent biological assessments in the Paint Rock River system have demonstrated recovery due to improvements in water quality (O'Neil et al. 2013; TVA unpub. data). Elimination of many past perturbations can be attributed to ongoing habitat conservation and restoration efforts by numerous state, federal and non-profit collaborators (O'Neil et al. 2013; Fobian et al. 2014).

Percina burtoni was first collected from the Paint Rock River system in Estill Fork during 1980 (UAIC 6343.18, 6344.18). Surveys during the 1990s found it in Larkin Fork, the upper Paint Rock River, and Estill Fork (Metee et al. 2002; TVA unpub. data). In 2010, it was first collected in Hurricane Creek and from Clear Creek, which is the most downstream record within the Paint Rock River system (O'Neil et al. 2013). The collection in Clear Creek was 34 rkms from the next upstream record within the system. This may indicate recent downstream dispersal as habitat conditions have improved in the mainstem of the Paint Rock River and that *P. burtoni* is present between these localities. Recent collections during this study and by collaborators show that *P. burtoni* is widely distributed in the upper Paint Rock River system (Figure 7). It is also locally abundant as supported by collection of 18 individuals during an August 2019 survey downstream of Alabama Hwy 146 crossing of the Paint Rock River (C. Johnson, Alabama Department of Environmental Management, pers. comm. 2019).

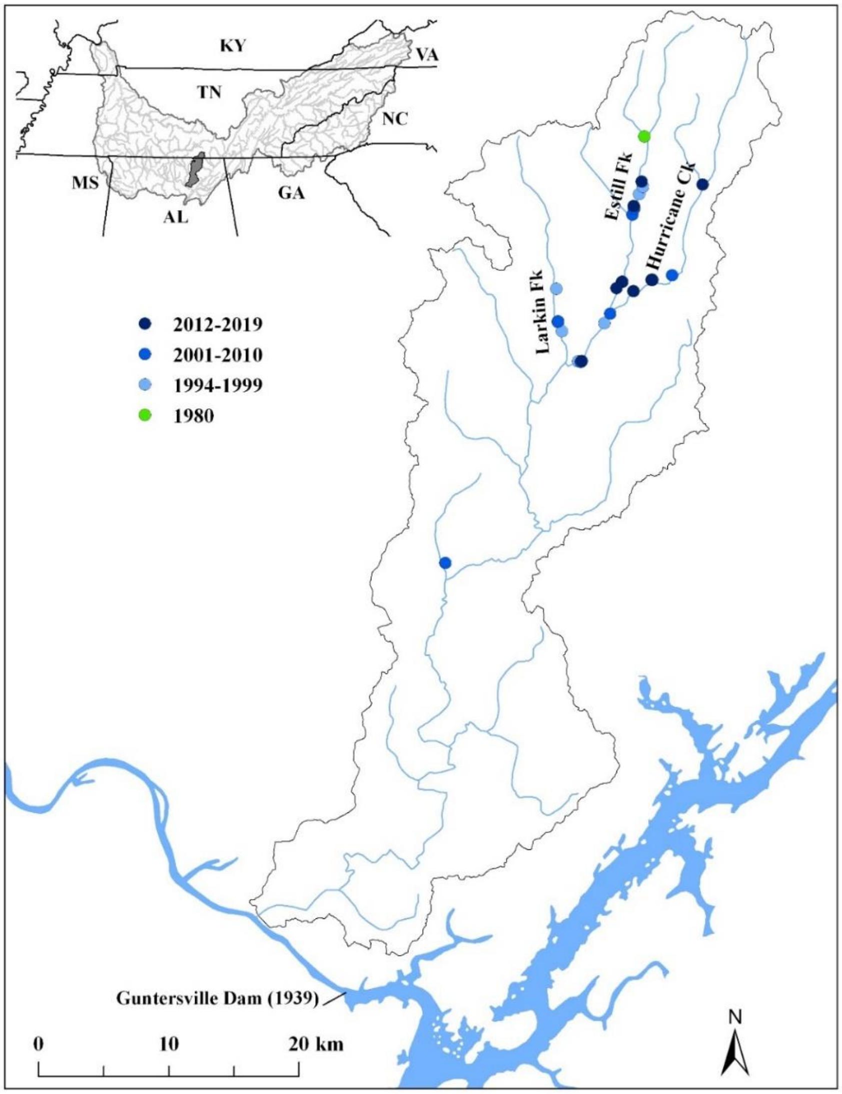

Figure 7. Historical and current distribution of *Percina burtoni* in the Paint Rock River system.

Elk River System

The Elk River flows for ~162 rkms downstream of Tims Ford Dam until it is inundated by Wheeler Reservoir (Figure 8). Present day land use in much of the Elk River watershed is dominated by row crops and pastureland which has resulted in heavy sediment loads in the lower portion of the river (Shepard et al. 2009; pers. obs.).

Prior to the first comprehensive fish survey during the late 1960s, only sporadic ichthyological collections had been made in the Elk River system (Jandebeur 1972). During recent decades, TVA and others have routinely surveyed portions of the basin (Mettee et al. 2002; Shepard et al. 2009; TVA unpub. data). The first discovery of *P. burtoni* in the Elk River system was by a TVA stream crew on 14 August 2007, where three individuals were collected in Big Creek, a tributary to Richland Creek, in Giles County, Tennessee (Figure 8).

Various locations on Big Creek were sampled during the course of this study to determine the current distribution of *P. burtoni*. The reach where *P. burtoni* was first collected in 2007 was sampled on several occasions during 2015 by snorkeling and backpack electrofishing into a seine. Additional upstream locations, spanning several kilometers, were surveyed while snorkeling. No individuals were detected during these efforts. Throughout this reach, row crops bordered the stream, many areas completely lacked riparian vegetation and sediments covered substrates.

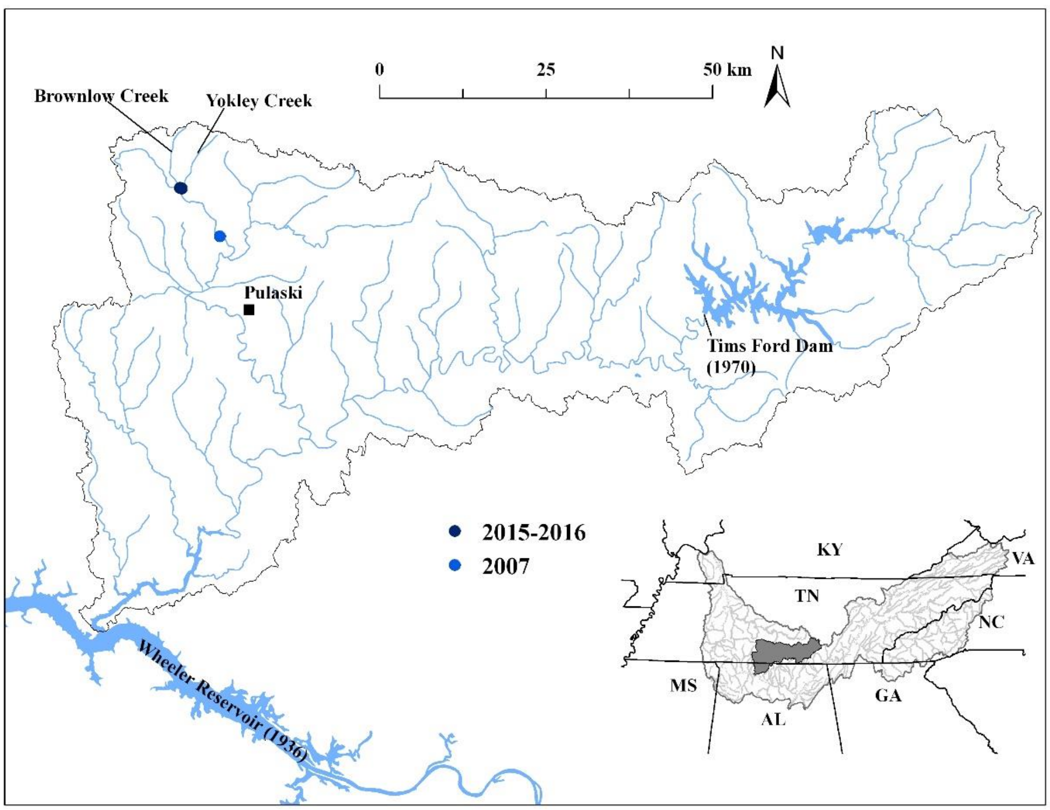

Figure 8. Historical and current distribution of *Percina burtoni* in the Elk River system.

During 2015, individuals were detected throughout a 650 m reach of Big Creek, from the confluence of Yokley Creek upstream to the mouth of Brownlow Creek (Figure 8). Within this reach, the stream was mildly turbid and substrates in low velocity areas were covered with sediment while swift areas maintained clean gravel substrates. On 6 August 2015, six individuals were seen while snorkeling a 220 m reach of Big Creek upstream of the confluence of Yokley Creek. One adult female and one sub-adult were observed in a slower flowing margin of a narrow gravel run surrounded by water willow. Upstream, the creek flowed over a small cascade into a deep run dominated by boulder, cobble and bedrock, and then transitioned into a long river-wide pool. Four adults were seen within this pool over clean gravel patches. On 1 September 2015, an individual was observed at the confluence of Brownlow Creek.

On 28 April 2016, flowing habitats were sampled by electrofishing downstream of the TN Hwy 245 crossing to the mouth of Yokley Creek and eight individuals were collected (YPM ICH 029438). A male and female were collected together in a swift run bordered by water willow with gravel and cobble substrates and were assumed to be a spawning pair. An additional female and two males were collected in consecutive efforts in similar habitats. Adult males all exhibited bold nuptial coloration and adult females had swollen abdomens indicating that they were gravid (see Figure 4 in Near et al. 2017). Males ranged from 121.6 to 131.2 mm standard length (SL) and females were 109.8 and 121.5 mm (SL). Mean water column velocity was 36.1 cm/s, mean bottom velocity was 21.9 cm/s and mean water depth was 0.47 m where these fish were collected. Water temperature was 18.8 °C. One female sub-adult, 75.3 mm (SL), was collected in a shallow riffle/run during these efforts. A male sub-adult, 74.8 mm (SL), was collected from the first run in Yokley Creek upstream from the confluence with Big Creek.

Although the majority of the Elk River systems drains through the Nashville Basin physiographic province, there are scattered remnants of the Highland Rim which are most numerous and best preserved on the divide between the Elk and Duck rivers (Theis 1936). The distribution of *P. burtoni* in the lower Tennessee River drainage is strongly tied to streams draining the Highland Rim. Big Creek represents a stream that drains a remnant section of the western Highland Rim and flows into the outer Nashville Basin. There are additional Elk River tributaries downstream of Richland Creek which flow from portions of the western Highland Rim that may provide suitable habitats for *P. burtoni*. Detection of additional occurrences in the Elk River system and proactive habitat conservation in Big Creek are imperative for conserving this narrowly distributed population.

Shoal Creek System

Shoal Creek flows south through the western Highland Rim into Alabama where the lower 21 rkms are inundated by Wilson Reservoir, a Tennessee River impoundment (Figure 9). Historical water quality issues have been noted in the Shoal Creek system but conditions improved following the implementation of the Clean Water Act (Morgan et al. 1997; U.S. Federal Register 2005).

The first comprehensive ichthyological survey of the Shoal Creek system was reported by Wagers (1974) and mentioned a recent collection of *P. burtoni* from Little Butler Creek, representing the first record from the system. This specimen (UNAF 287) was collected by M. Balch on 18 June 1971 (J. Ray, University of North Alabama, Florence, pers. comm. 2020). The locality label reads "Little Butler Creek, Wayne County, Tennessee". The exact locality of this collection is unknown since the section of Little Butler Creek in Tennessee is small and often intermittent, thus is not a place that would be typically inhabited by *P. burtoni*.

Shortly thereafter, *P. burtoni* was found in Butler Creek (MMNS 41436 and 41437 [1973]; UT 91.2306 [1981]) and from nearby locations in Shoal Creek (TVA 1983b) a few kilometers upstream of the backwaters of Wilson Reservoir (Figure 9). Morgan et al. (1997) conducted fish surveys throughout the Shoal Creek drainage at 85 different locations during 1995, resulting in collection of 78 species, but did not collect *P. burtoni*. Individuals collected during TVA stream surveys, this study, and from Ray et al. (2019) greatly expanded the known range of the species in the Shoal Creek system (Figure 9).

A male in spawning condition (YPM ICH 029437), 102 mm (SL), was collected from a gravel bottomed run in Knob Creek, a tributary to Shoal Creek, on 25 April 2016. Water column velocity was 92.2 cm/s, bottom velocity was 36.4 cm/s and water depth was 0.49 m. On 21 July 2015, an age-0 individual (YPM ICH 028335), 49.2 mm (SL), was collected in Shoal Creek while seine hauling a backwater pool. The pool was less than a meter deep, contained a gravel and sand bottom, and was completely surrounded by water willow beds. The downstream end of the pool connected to the main river via a small channel running through water willow, similar to habitat described for young-of-year in the Little River (Jett 2010).

In Chisolm Creek, on 22 July 2015, a reach greater than 300 m was snorkeled before detecting individuals. An adult was seen using a narrow gravel run bordered by water willow beds. Several *Percina evides* followed *P. burtoni* and opportunistically fed on insects that were dislodged after rocks were flipped by *P*. *burtoni*. Two additional adults occupied the upstream pool near the top of the run where water depths were shallower and substrates were clean. In the pool, *P. burtoni* were with *P. caprodes*.

Recent collections of *P. burtoni* demonstrate that they utilize appropriate habitats in both the mainstem of Shoal Creek and in portions of large tributaries several kilometers upstream from the mainstem. Areas snorkeled in Chisolm Creek indicated that the occurrence of *P. burtoni* was habitat specific and that snorkeling is an effective sampling technique for detecting individuals occurring at low densities.

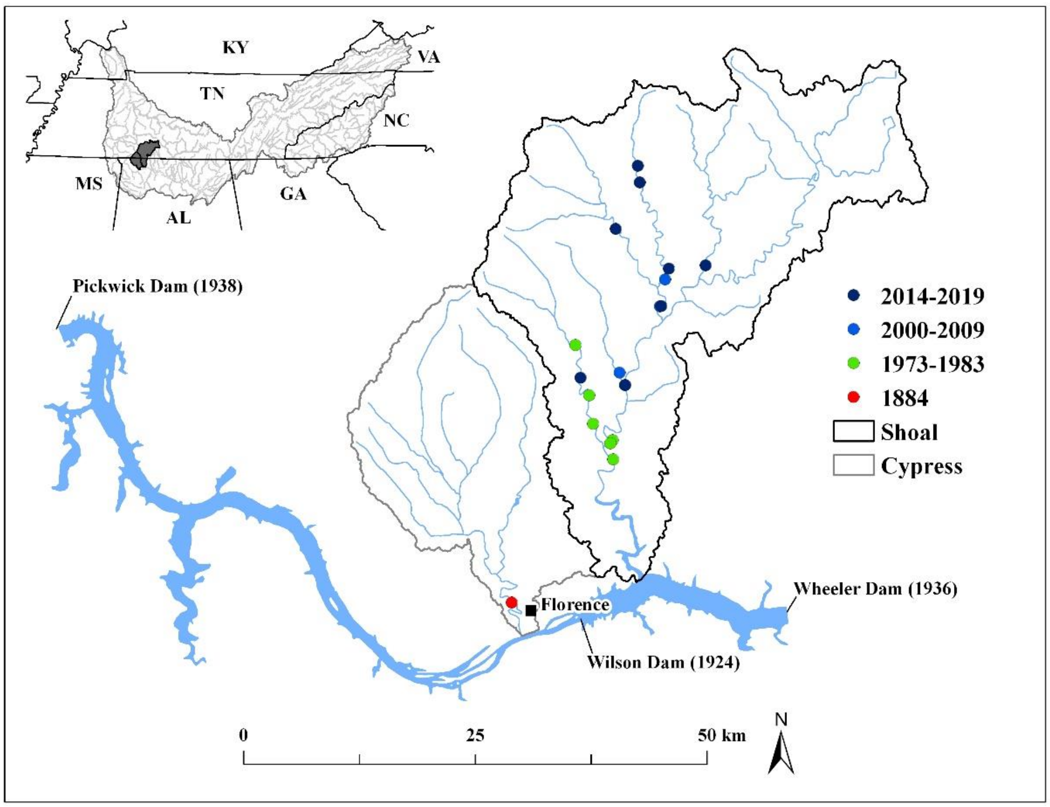

Figure 9. Historical and current distribution of *Percina burtoni* in the Shoal Creek system and the approximate location of the historical collection in Cypress Creek. The 1971 Little Butler Creek record was not plotted due to uncertainty of collection locale.

Cypress Creek System

A specimen from Cypress Creek (CAS 223384) was discovered during this study, representing a previously unreported occurrence for *P. burtoni* (Figure 9). No collection date was included, but collectors were listed as C.H. Gilbert and J. Swain. The only collections by Gilbert and Swain in this region were during 1884 and included type specimens of *Etheostoma blennius* from Cox's Creek, a tributary to Cypress Creek at Florence, Alabama (Gilbert 1887). Gilbert (1891) reported collections from Cypress Creek made by P.H. Kirsch, W.M. Andrews and E.O. Jones on 5 June 1889 and did not include *P. burtoni*. Therefore, this specimen was likely collected during their trip in 1884.

CUMBERLAND RIVER DRAINAGE- *Percina burtoni*

Within the Cumberland River drainage, *P. burtoni* was known from the Obey River system and from the Big and Little South Forks of the Cumberland River (Figure 10). The majority of vouchered collections were made in 1891 and were listed as *Etheostoma caprodes* in Kirsch (1893). These specimens have all been examined and confirmed as *P. burtoni* (Near et al. 2017). All Cumberland River drainage populations were considered extirpated until recent sightings in the Big South Fork of the Cumberland River.

Obey River System

A majority of the Obey River was impounded by Dale Hollow Reservoir in 1943. The lower reaches of the East and West Forks of the Obey River and Wolf River are also inundated by this reservoir. Of these, Wolf River is the only stream that was not subject to long-term pollution from coal mining. However, a rock washing plant for separation of barium ore resulted in extreme periods of turbidity in the Wolf River that extended downstream into the Obey River (Shoup et al. 1941).

The East Fork Obey River system has been most affected by coal mining pollutants which were first reported by Shoup et al. (1941). A contiguous 64 km reach from headwater tributaries to the backwaters of Dale Hollow Reservoir was found to be almost void of aquatic life due to acid mine drainage (Nichols and Bulow 1973). This pollution was still persistent during subsequent studies of water quality in the East Fork Obey River system (Baker 1989; Sasowsky and White 1993). Mining occurred to a lesser extent in the West Fork Obey River system where lasting effects of acid mine drainage were more isolated (Carrithers and Bulow 1973; O'Bara and Estes 1985).

Collections of *P. burtoni* from the Obey River and Eagle Creek (UMMZ 250221, 61515) were made by P.H. Kirsch in 1891 (Figure 10). These collections were made at the town of Olympus which no longer exists due to inundation by the reservoir. The last vouchers of *P. burtoni* from the Cumberland River system were from the Wolf River in 1947 (UMMZ 154611, 154612).

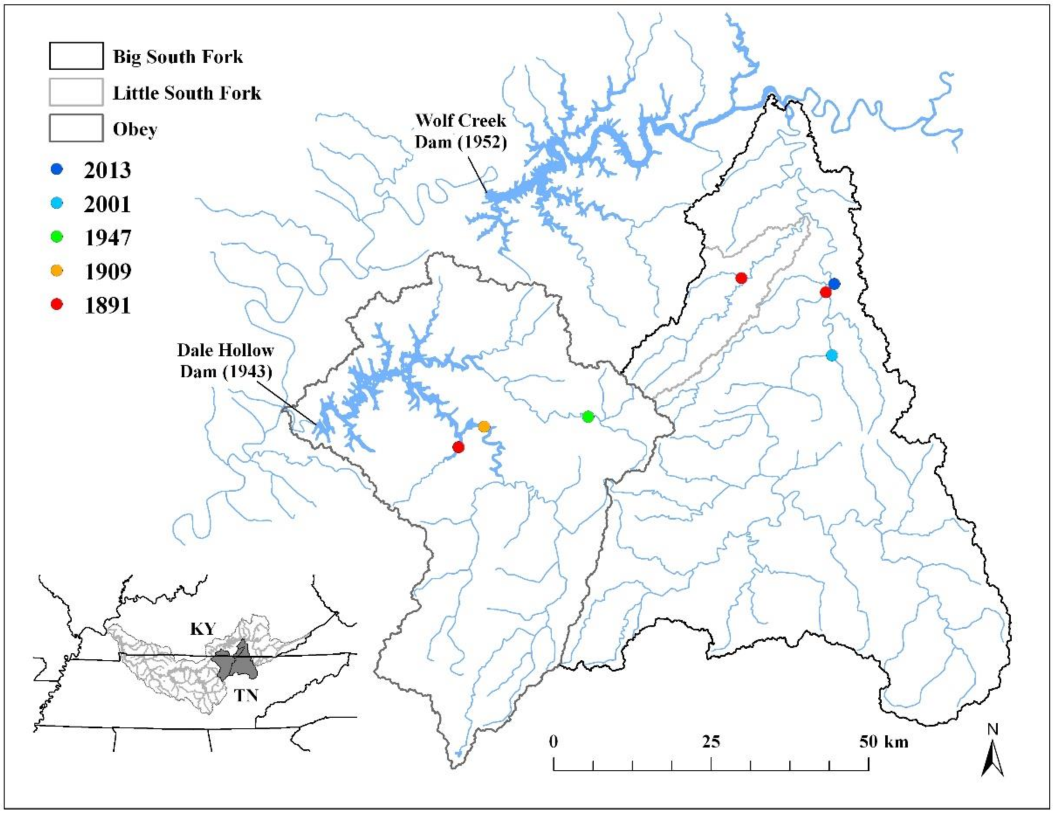

Figure 10. Historical and recent distribution of *Percina burtoni* in the Cumberland River drainage.

Two reaches of the Wolf River were snorkeled for *P. burtoni* during this study. Turbidity was low and substrates were clean of sediment but were dominated by limestone bedrock and large slab rock. These reaches mostly lacked gravel substrates in areas of flow, which seem to be important components of habitats used by *P. burtoni* in other rivers. The few flowing areas containing sand and gravel substrates were the only places where *P. caprodes* were seen. The fish community observed while snorkeling was diverse and included six species of darters.

Similarly, Moles et al. (2007) attributed lack of suitable habitat as a limiting factor for mussel abundance and distribution in the Wolf River. The stream segment with the highest diversity and abundance of mussels contained stable gravel bars, while most other reaches of the Wolf River were dominated by bedrock and unstable gravel pockets. An extant fish and mussel community containing several sensitive species provide justification for additional surveys for *P. burtoni* in the Wolf River. However, appropriate habitats utilized by *P. burtoni* may be limited.

Until recently, the Redlips Darter, *Allohistium maydeni*, was presumed extirpated from the Obey River system as a result of mining pollution (e.g. Powers et al. 2012), where the last reported occurrence was in 1970 from the West Fork Obey River (INHS 75541). During 2007, a single individual was collected in the East Fork Obey River and several more individuals were collected near this location during 2017 and 2020 (USFWS 2018; TVA unpub. data). In 2019, the author discovered a specimen of *A. maydeni* in the Tennessee Technological University fish collection (TTU 376) from the West Fork Obey River, collected in 1998. The author and others resurveyed this site during March 2020 and five individuals were collected. These recent collections indicate that the Obey River system has been under-surveyed, possibly due to an assumption that past pollution had resulted in it being of little biological value. These also provide rationale for continued surveys for *P. burtoni* in this system.

Big South Fork of the Cumberland River System

The only known collection of *P. burtoni* from the Big South Fork of the Cumberland River (UMMZ 250220) was from a shoal complex downstream of the confluence of Rock Creek on 7 September 1891, McCreary County, Kentucky (Kirsch 1893). In 1902, Stearns Coal and Lumber Company began mining coal in nearby areas along the Big South Fork (Butler 1922). This continued during much of the twentieth century and had severe impacts to aquatic fauna in the Big South Fork and its tributaries (Worsham et al. 2013). These were most notable in Bear Creek, Roaring Paunch Creek, and Rock Creek, all of which drain to the Big South Fork within a 15 rkm section (Hamilton and Turrini-Smith 1997; Worsham et al. 2013). Carter and Jones (1969) reported a pH of 3.6 near the mouth of Roaring Paunch Creek and mentioned this and Rock Creek as main sources of acid mine pollution to the Big South Fork. Comiskey and Etnier (1972) indicated that the lower portion of Rock Creek was so badly polluted by strip mining activities that many portions of the stream lacked a vertebrate or macroinvertebrate fauna. In 1981, no fish were detected during sampling efforts in the Big South Fork at the confluences of Bear and Rock creeks and only four pollution tolerant species were collected at the mouth of Roaring Paunch Creek (O'Bara and Estes 1984).

Completion of Wolf Creek Dam on the Cumberland River in 1952 further affected the Big South Fork by completely or seasonally inundating the lower 70 rkms, including the historical collection locality of *P. burtoni*. From January 2007 to January 2013, the elevation of Lake Cumberland was lowered due to concerns of dam failure. During this drawdown period, 14 rkms of the Big South Fork that had been inundated were free-flowing. Just prior to this extended reservoir draw down, acid mine drainage remediation had been conducted in Rock Creek (Carew 2002) and water quality in Bear Creek and Roaring Paunch Creek seemed to be improving (Worsham et al. 2013). Improved water quality from acid mine drainage remediation along with lower reservoir elevations, created a "perfect storm" for fish recovery in this reach of the Big South Fork. Repeated collections from 2013 to 2018 have resulted in 66 species of fish, including 17 species of darters (TVA unpub. data), demonstrating tremendous fish community recovery.

Although no one has been able to capture *P. burtoni* from the Big South Fork since 1891, two individuals have been observed during recent times (Figure 10). The first observation was made in the Big South Fork upstream of the confluence of Bear Creek during 2001. While conducting a mussel survey, three experienced and reputable biologists, S. Ahlstedt, R. Butler, and D. Hubbs, saw an individual while snorkeling. All three confirmed the sighting in email communications during 2016. The second observation was made on 21 November 2013, while snorkeling at a location located two shoal complexes downstream of Kirsch's 1891 collection (pers. obs.). This fish was followed for over a minute, allowing ample time for positive identification. This shoal is now degraded due to extended periods of inundation by Lake Cumberland since 2014. The location of the 2001 sighting was snorkeled during September 2016 and 2017. No individuals were observed, but appropriate habitats were present. Large cobble and boulder substrates are predominant in the Big South Fork. Both locations where *P. burtoni* was observed were unique in that they contained areas of smaller cobble and gravel substrates in flowing habitats.

Little South Fork of the Cumberland River System

Prior to the 1980s, the Little South Fork supported one of the most diverse and intact mussel assemblages in the Cumberland River system including two federally listed endangered species and many others considered imperiled throughout their range (Warren and Haag 2005). The Little South Fork also harbors one of the two extant populations of the endangered fish *Notropis albizonatus*

(Warren et al. 1994). In 1987, a survey of mussels in the Little South Fork revealed that unionid populations had been devastated by recent coal mining activities in the lower third of the river from near the confluence of Kennedy Creek and downstream (Anderson et al. 1991). Subsequent surveys during 1997 and 1998 documented a continued decline in mussels further upstream in the Little South Fork and was attributed to pollution from oil extraction (Warren and Haag 2005).

During September 2015 and 2016, several locations were snorkeled in the Little South Fork in search of *P. burtoni*. This included a reach above and below the confluence of Kennedy Creek where five specimens were collected in 1891 (FMNH 6798). Kirsch (1893) listed this locale as “Little South Fork at the mouth of Canada Creek”. Canada Creek no longer exists on contemporary topographic maps and was changed to Kennedy Creek at some point. This reach consisted mainly of pools, separated by a few small, shallow riffles. Substrates were free of silt, water clarity was good, the riparian zone was well established and numerous cool springs were felt flowing up from the river bottom. Limestone bedrock and cobble were predominant with interspersed patches of gravel and sand. On two occasions, the Little South Fork at Ritner Ford was snorkeled where C.E. Comiskey was convinced that a *P. burtoni* had escaped his dip net during a cyanide application in 1968 (Comiskey and Etnier 1972). The river bottom at this location was also dominated with limestone bedrock and slab rocks with few areas containing smaller substrates. This reach consisted of long pools with a few shallow riffles and runs. Several *P. caprodes* were seen in a small run with a gravel bottom, and this was the only area seen that may have been compatible with habitat preferences of *P. burtoni*. Water clarity was good, although some sediment deposition was observed in pools. Habitats at other locations snorkeled in the Little South Fork were similar to the sites described above.

No physical evidence of acid mine drainage or signs of oil contamination were noticed during 2015 or 2016, but each site could be described as a “mussel graveyard” due to the large number of relict shells. While snorkeling during September 2016, a live *Villosa taeniata* and *Pleurobema oviforme* were found at Ritner Ford and upstream of the confluence of Kennedy Creek, respectively. Numerous *N. albizonatus* were observed at Ritner Ford. These observations suggest that previous perturbations to the Little South Fork may have been curtailed. However, habitats frequently used by *P. burtoni* in other rivers appear to be lacking in areas surveyed in the Little South Fork. Access to the river is very limited and would require a long float trip to discover areas containing these habitats.

DUCK RIVER SYSTEM AND WHITE OAK CREEK- *Percina apina*

The Duck River system is the most biologically diverse river system in North America. It supports over 650 aquatic species, including at least 147 species of fish, 54 species of freshwater mussels, and 22 species of aquatic snails (Ahlstedt et al. 2017). The Duck River and its largest tributary, the Buffalo River, are high priority watersheds for imperiled fish conservation because of the high number of rare or endemic species (Butler 2002).

The upper and lower portions of the Duck River system, including the Buffalo River, are in the eastern and western Highland Rim physiographic province. Streams in the Highland Rim are characterized by moderate gradient with chert gravel bottoms. The middle portion of the system is within the Nashville Basin where streams are lower gradient and contain predominantly limestone bedrock and slab rock substrates. Due to these differences, many fish species that occur in the eastern and western Highland Rim are absent from the Nashville Basin (Starnes and Etnier 1986).

With the exception of impoundment by Normandy Dam near the headwaters, a majority of the Duck River system remains free-flowing. This is a significant factor responsible for maintenance of its high level of biodiversity. Normandy Dam lies within the outer Nashville Basin and the backwaters of the reservoir extend to the edge of the eastern Highland Rim where it impounds 26 rkms of the Duck River. Kentucky Reservoir, the lower most impoundment of the Tennessee River, inundates the lower 25 rkms of the Duck River. The Buffalo River remains one of the longest free-flowing rivers in Tennessee and flows into the Duck River just upstream of the backwaters of Kentucky Reservoir.

Percina apina is distributed within the western Highland Rim, almost exclusively in the lower portion of the Duck River system, with the exception of one population in Whiteoak Creek, a tributary to the Tennessee River downstream of the confluence of the Duck River (Figure 11). It is most widely distributed in the Buffalo River system and occurs in two other Duck River tributaries, Hurricane Creek and Big Swan Creek. It was historically distributed within the eastern Highland Rim in the upper Duck River.

The first collections of *P. apina* were during 1971 and 1972 in Buffalo River tributaries Forty-eight Creek (UT 91.618; AUM 4352) and Green River (AUM 4319), Big Swan Creek (UT 91.731) and from the upper Duck River (AUM 4377). Shortly thereafter, it was collected from Whiteoak Creek (TU 89496) in 1974 and from Hurricane Creek (TU 117994) in 1979. With the exception of the upper Duck

River, surveys conducted during this study continued to detect *P. apina* in all of these stream systems and at additional locations within the Buffalo River system (Figure 11).

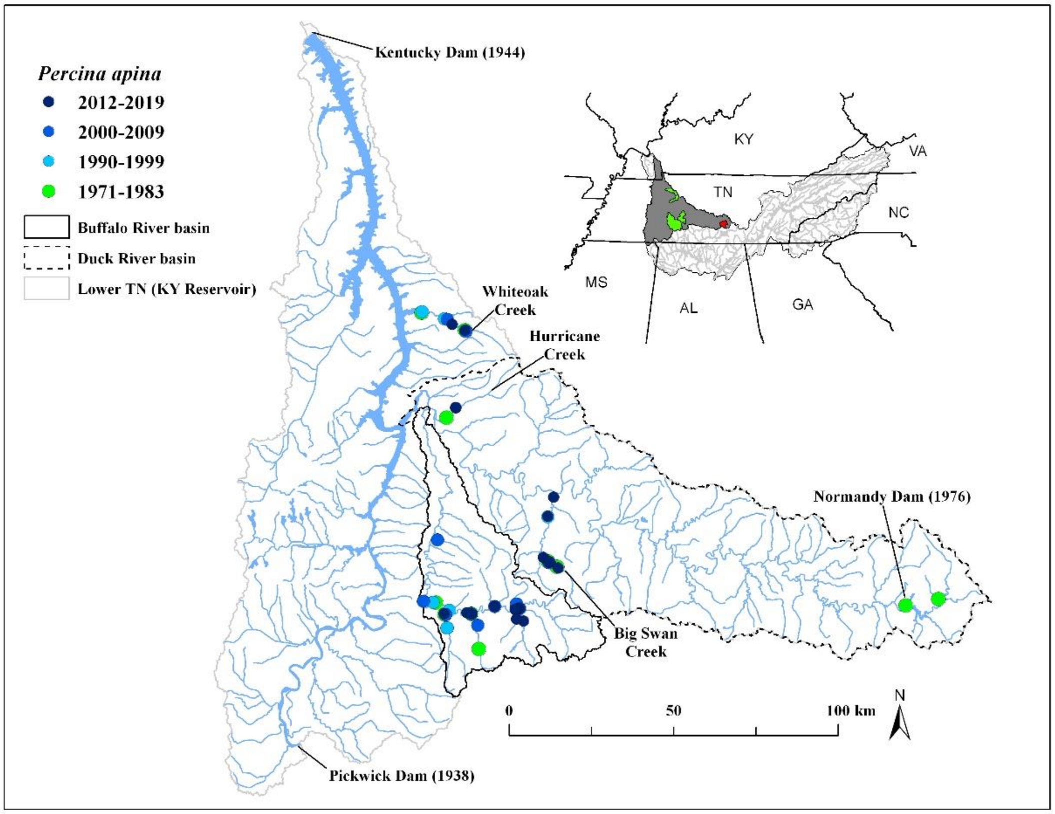

Figure 11. Historical and current distribution of *Percina apina*. The area shaded in red on the map inset represents an extirpated population while the areas shaded in green represent the currently occupied range.

The eastern most population is represented by two collections from the mainstem Duck River. The first was at rkm 403 on 7 October 1971 before inundation by Normandy Dam (AUM 4377). The second was at the mouth of Bashaw Creek upstream of the backwaters of Normandy Reservoir on 15 October 1977 (UT 91.1430), one year after completion of the reservoir. These collections also included the only specimens of *Percina macrocephala* (AUM 4379; UT 91.1544) from the Duck River system.

A reach of the Duck River from the mouth of Bashaw Creek upstream to the confluence of the Little Duck River was snorkeled on 21 May 2015. Additional snorkeling efforts were expended in the Little Duck and Duck Rivers upstream of their confluence. No individuals were seen, although *P. caprodes* was abundant. The most notable fish species occurrence within this reach was *Nothonotus aquali* (YPM ICH 028117). Just downstream of the confluence of the Little Duck River, the City of Manchester wastewater treatment discharge entered the river. Large cascades are present on the Little Duck and Duck Rivers about 1 km upstream of their confluence, and a small impoundment is present just upstream of the waterfalls on the Duck River, both of which serve as a barrier to upstream fish dispersal (e.g. Etnier and Bailey 1989). Therefore, after completion of Normandy Dam, *P. apina* was restricted to an approximate seven km reach from the backwaters of the reservoir upstream to the Duck River cascades.

In other portions of the mainstem Duck River, extensive surveys (snorkeling, scuba, rotenone, electrofishing, seining) have been conducted and no *P. apina* were observed (e.g. Koch et al. 1986; TVA 1975; TVA unpub. data). This may be due to avoidance of locations with large drainage areas. Where the Duck River enters the western Highland Rim, the drainage area is around 4,660 km^2 compared to < 600 km^2 where *P. apina* were collected in the upper Duck River. In the mainstem Buffalo River, *P. apina* have been collected where the drainage area is less than 1,200 km^2. None have ever been collected from the lower Buffalo River were the drainage area reaches 1,971 km^2, further indicating that larger stream size is not preferred by this species. Surveys conducted during this study found *P. apina* more common and abundant in major Buffalo River tributaries than in the mainstem. However, their presence in the mainstem indicates a largely connected population. Drainage area of locations occupied by *P. apina* elsewhere range from 211 km^2 (Hurricane Creek) and 277 km^2 (Whiteoak Creek) to 404 km^2 (Big Swan Creek).

Watersheds containing populations of *P. apina* are largely forested and have low human population densities. Whiteoak Creek, Hurricane Creek, and Big Swan Creek watersheds are greater than 78% forested, while much of the remaining land area is used for hay or pasture and very little is cultivated for other forms of agriculture. Human population density is $\leq$ 0.09 people per hectare in these watersheds. The Buffalo River watershed is predominantly forested (68%), but has a higher proportion of lands used for agriculture (19%) and contains areas of higher human population densities (0.14/ha).

The predominantly natural condition of watersheds occupied by *P. apina* has contributed to maintenance of good water quality. Most recent TVA IBI scores at locations occupied by *P. apina* ranged from 77 to 97% of the highest attainable score with a mean of 84% (N=19). Even though there are higher human population densities and more lands utilized for agriculture in the Buffalo River drainage, it appears that these factors have not yet reached a point that is preventing persistence of *P. apina*. Large riparian areas are maintained in a majority of the upper Buffalo River drainage and some lands are afforded protection by state and federal agencies. Most of the land along the Buffalo River and its tributaries is privately owned and timber companies hold most of the land on the upper slopes and ridges (Mast and Turk 1999). Poorly managed timber harvest operations along the ridges and increased development in the valleys are currently the greatest threats to *P. apina* throughout its range.

Life History Observations and Habitat Use

Snorkeling surveys for *P. apina* from late April to early September allowed for observations of behavior and seasonal differences in habitat use. Individuals were very habitat specific during both reproductive and non-reproductive periods. Although no spawning events were observed, males and females in reproductive condition were most often seen in swift runs where they occupied areas with water depths $\leq$ 1 m and with predominantly gravel and cobble substrates. Spawning may occur in shallow, swift riffles that could not be snorkeled. On numerous occasions, during late April to early June, adult males were collected in this habitat type with a backpack electroshocker.

Males maintained breeding colors through June, although peak breeding coloration was observed during April and May. A male with breeding colors was only seen using a pool habitat on one occasion. This was on the 27 April 2016 in the Little Buffalo River at a water temperature of 19 °C. His physical condition showed evidence of previous spawning interactions as his spinous dorsal fin contained several tears and a scrape wound was apparent on the left side of his dorsum (YPM ICH 029441). Pool habitats may be periodically used by spawning males as recovery areas. The physical toils of spawning were also observed on a large female from this locality that had a ragged and torn spinous dorsal fin (YPM ICH 029441). Her abdomen was not swollen, indicating that she had already laid her compliment of eggs. At this time, adults of both sexes were also observed in swift runs, which presumably included individuals that were still reproductively fit.

During summer months, both adult males and females were found in swift runs and pool areas containing silt free, gravel and cobble substrates. While snorkeling in swift habitats, individuals would frequently be pursued to the head of a run. They would then drift back downstream to the exact point of initial detection where bottom velocities were slower and the water was deeper. Those pursued into areas lacking clean gravel substrates would move back to cleaner areas. These behaviors suggest that their non-spawning habitat preference is strongly tied to clean rocky substrates and areas where they do not have to expend great amounts of energy maintaining position in swift flows. At locations in Whiteoak Creek and the Little Buffalo River, individuals were located at the head of pools where streamflow maintained clean substrates, compared to areas further into the pool where sediments had accumulated on the stream bottom. At some locations, silt was virtually absent, even in gently flowing pools. This was the case in Hurricane Creek where underwater visibility was exceptional due to the pristine nature of the stream.

In all streams surveyed, sub-adults were only seen in pool habitats and were often accompanied by adult females. In the Green River on the 3 June 2015, a "feeding troop" of ten individuals, consisting of females and sub-adults, stayed together as they moved upstream flipping rocks and feeding in a clean, rocky bottomed, flowing pool upstream of a shoal complex.

Longevity of *P. apina* and *P. burtoni*

Estimation of age-classes for each species required compilation of data from all years and rivers to allow for meaningful approximations due to limited sample sizes. Collections of *P. apina* were from March to June and August to October, while collections of *P. burtoni* were from March through November (Figure 12). Lengths of *P. apina* ranged from 34.7 – 141.3 mm SL (Figure 12A) and 40.5 – 142.0 mm SL for *P. burtoni* (Figure 12B).

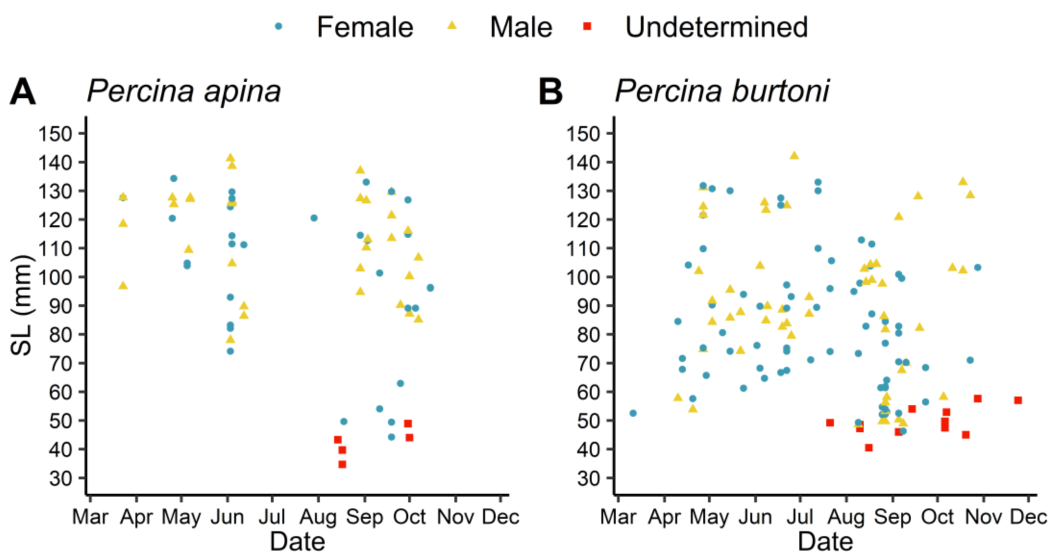

Figure 12. Length distribution of *Percina apina* (N=70) and *Percina burtoni* (N=137) by month from lengths obtained from museum specimens and individuals vouchered during this study.

Kernel density estimates identified four cohorts for *P. apina* and five for *P. burtoni* (Figure 13). Lower sample size for *P. apina* may have resulted in omission of at least one age class. Lengths ranging from 55 to 80 mm were mostly absent from the *P. apina* dataset but were present for *P. burtoni* and represented the age-1 cohort. Local maxima were not markedly different among time periods examined, with exception of lack of the age-0 cohort for spring/early summer.

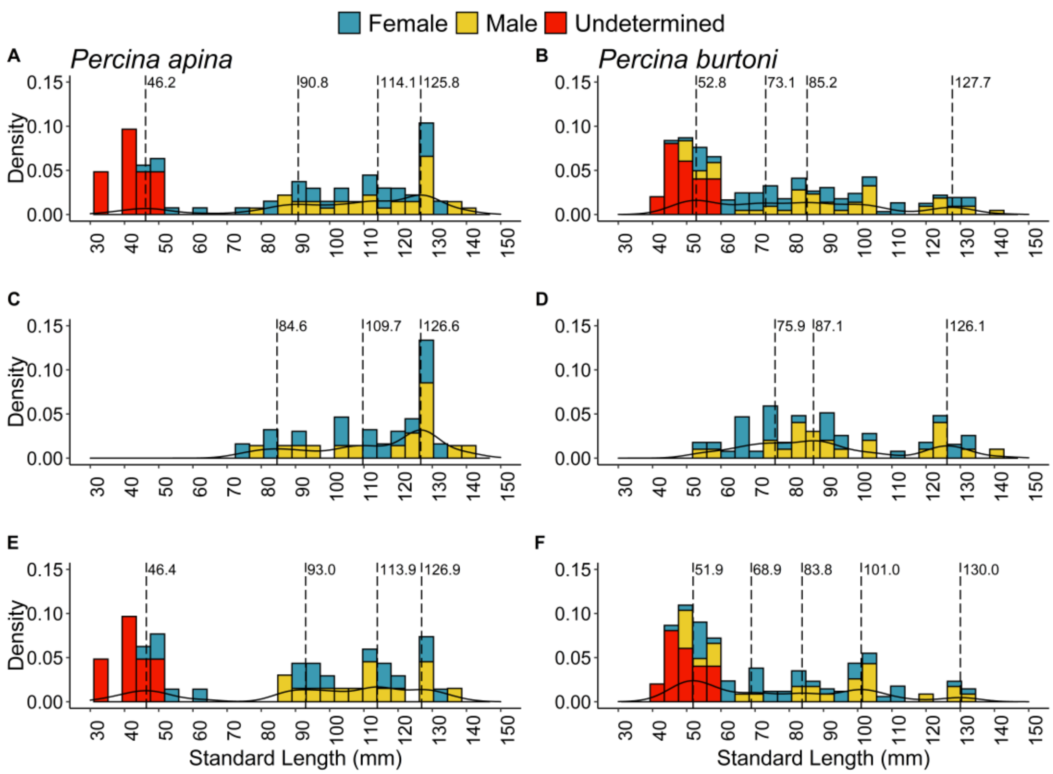

Figure 13: Smoothed kernel density estimates (bandwidth=5) with density histograms, and local maxima for *Percina apina* (A, C, E) and *Percina burtoni* (B, D, F). A (N=70) and B (N=137), all months combined. C (n=32) and D (n=56), individuals collected March-June. E (n=38) and F (n=81), individuals collected July-November.

Etnier and Starnes (1993) and Jenkins and Burkhead (1994) suggested that *P. burtoni* lived at least four years which is in agreement with this analysis. A closely related sister species, *Percina rex*, was estimated to live five years as determined from scale annuli (Rosenberger and Angermeier 2002). Growth of *P. rex* was rapid during the first two years and then slowed from age-3 to age-5 with considerable overlap of lengths. It is probable that an age-5 cohort was not detected for *P. apina* or *P. burtoni* due to reduced growth with age. Regardless, this is currently the best available data to provide insight into this aspect of life history for these rare species.

The largest, and presumably the oldest, individuals of both species were present during the spawning period but were not represented in subsequent months (Figure 13). Older fish may perish after spawning as was indicated by a large dead female *P. burtoni* (131.8 mm, SL) in Big Creek (Elk River system) during late April 2016. By July and August, age-0 *P. burtoni* and *P. apina* were present in collections. This age class did not appear to be sexually mature the following year as evidenced by the habitat use and behavior of many sub-adults of each species seen while snorkeling during the spring. Smaller nuptial males of both species collected during this study averaged ~100 mm (SL), while most reproductive females and males were larger. From these observations and subsequent age-at-length estimations, most reproducing adults of both species were three to four years old, while a smaller proportion of age-2 fish may also contribute to reproduction. When compared to more short-lived darter species, attaining sexual maturity at a later age could make these species vulnerable to extirpation if populations experienced conditions that limited survivability of older age-classes.

SUMMARY

Physical geography is an important factor associated with the distribution of *P. burtoni* and *P. apina*. Populations of *P. burtoni* occurring in the upper Tennessee River Basin are distributed within the Ridge and Valley and Blue Ridge physiographic provinces. The eastern edge of the Ridge and Valley is bounded by the steep Central Appalachian and Cumberland Plateau escarpments which serve as a divide between the Tennessee and Cumberland River drainages. Populations in the Clinch and Holston River systems, and the former South Chickamauga Creek population, are all within the Ridge and Valley. A large portion of the Holston River system lacks records for *P. burtoni*. Prior to impoundment and pollution, there may have been other areas that supported populations as evidenced by the few collections just before completion of South Holston Reservoir. The Little River and former Little Tennessee River populations occur at the western edge of the Blue Ridge and extend into the Ridge and Valley. Similarly, the isolated population in the Hiwassee River system inhabits the western margin of the Blue Ridge and may have occupied a portion of the Ridge and Valley. The current and historical distributions of this species within the French Broad River system are unique in that they are completely within the Blue Ridge. As in the Holston River system, these populations may have extended into the Ridge and Valley in some rivers.

Within the lower Tennessee River basin, the population in the Paint Rock River system is mostly contained to the Cumberland Plateau escarpment. This is also the case for the population in the Big South Fork of the Cumberland River. Populations in the Elk River system and Shoal Creek, and former populations in Cypress Creek and Obey River, are all within the Highland Rim.

Percina apina is mostly restricted to the Duck River system in streams completely contained within the western Highland Rim. Complete absence from the Nashville Basin and previous occurrence in the eastern Highland Rim, further demonstrates their preference for environmental attributes present in this ecoregion. Furthermore, it seems that this species avoids areas with large drainage areas, which may limit dispersal among populations. It is also important to note that neither species of logperch occurs in regulated rivers.

From the information presented, it appears that the Big Creek (Elk River system), Spring Creek (Hiwassee River system), Indian Creek (Powell River system), Middle Fork Holston River, and South Toe River (French Broad River system) are the most vulnerable *P. burtoni* populations due to their extremely limited distributions. I was only able to find *P. burtoni* throughout a 650 m reach of Big Creek where habitat conditions were degraded from sedimentation from surrounding agricultural lands. It seems that the Spring Creek population is mostly isolated and has little chance of expansion due to the regulated nature of the Hiwassee River below Apalachia powerhouse. Furthermore, habitats in Spring Creek are lacking and observed sedimentation may further degrade available habitats. Coal mining impacts have apparently reduced the distribution of *P. burtoni* to Indian Creek in the Powell River drainage where it is also disconnected from the nearby Clinch River population by Norris Reservoir. The recent discovery of *P. burtoni* in the Middle Fork Holston River warrants additional surveys to understand its status and distribution in this portion of the Holston River system. This may be an impetus for water quality improvement initiatives within this watershed. From recent records, it seems that the South Toe River population is stable and may have opportunity to expand if conditions continue to improve in the North Toe River.

The population occurring in the North Fork Holston River demonstrates the ability of *P. burtoni* to recolonize large sections of river following chronic impacts if a source population is present and the river recovers. Similarly, collection of *P. burtoni* in Dumps Creek, a stream that formerly received wastewater from a coal washing plant, shows positive signs of recovery. Present and future coal mining in the Clinch River watershed remains a threat to the long term persistence of *P. burtoni* and many other imperiled aquatic species. Effective regulation of mining activities are imperative to the conservation of these fauna. Water quality in the Little River watershed is somewhat secure due to headwater protection from the Great Smoky Mountains National Park. Continued poor agricultural land use practices and increasing residential development are threats to this river especially within the lower reaches. It is unknown whether a small population of *P. burtoni* still existed in the Tellico River prior to introduction of hatchery raised progeny of fish from the Little River. Ongoing efforts to purchase tracts and restore habitats in the Paint Rock River system may be able to maintain superior water quality throughout much of the occupied range of *P. burtoni*. Recent surveys revealed that *P. burtoni* is widely distributed and appears to exist as an unfragmented population throughout the Shoal Creek system. There are currently no major threats to the species in this system.

Recent *P. burtoni* sightings and documented fish community recovery in the Big South Fork of the Cumberland River are encouraging. An extant mussel fauna in the Wolf River and recent re-discovery of *A. maydeni* in the East and West Forks of the Obey River provide possibility of the existence of *P. burtoni* in the Obey River system. Focused future survey efforts are needed to further assess the status of *P. burtoni* in the Cumberland River system.

Presently, populations of *P. apina* are all stable and seem to exist with large numbers of individuals. Occupied areas are predominantly forested with low human population densities. Future large scale timber operations, conversion of forest to cropland, and human population growth are the greatest threats to this species.

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