POPULATION ASSESSMENT OF REDHORSE (MOXOSTOMA SPP.) IN THE UPPER OCONALUFTEE RIVER, QUALLA BOUNDARY, NC

A thesis presented to the faculty of the Graduate School of Western Carolina University in partial fulfillment of the requirements for the degree of Master of Science in Biology.

By

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ABSTRACT

POPULATION ASSESSMENT OF REDHORSE (MOXOSTOMA SPP.) IN THE UPPER

OCONALUFTEE RIVER, QUALLA BOUNDARY, NC

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Western Carolina University (March 2018)

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The Oconaluftee River is a moderate to large sized stream located in the Little Tennessee River

basin of the southern Appalachian Mountains. The Little Tennessee River basin is home to six

different species of Redhorse, three of which, Black (Moxostoma duquesnii), Golden (M.

erythrurum), and Sicklefin (M. sp.), are thought to have historically occurred in the section of the

Oconaluftee River above Bryson Dam near the mouth of the River. Despite propagation efforts

and restorative stocking of the extirpated Sicklefin Redhorse population, there has been no sign

of success. Understanding of rare species (such as the Sicklefin Redhorse) has been a priority for

researchers; however, other Redhorse species are understudied in the southern Appalachian

Mountain region. Increasing the overall knowledge of other Redhorse populations in areas where

Sicklefin Redhorse have been extirpated provides vital information on ability of the habitat to

support Redhorse populations. From April to September of 2017, we used fyke netting and

mark-recapture electrofishing surveys to assess Redhorse population abundance and

demographics in the upper Oconaluftee River. Due to high flows and interference from otters, we

determined that the fyke net was unsuitable for sampling in this system. Using the raft

electrofishing system, we captured 624 Black Redhorse aged 1-12 and 138 Golden Redhorse

aged 1-10. No Sicklefin Redhorse were captured during our sampling events. Population

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abundance of the sampled reaches for Black Redhorse was estimated at n=2101 (Schumacher-Eschmeyer) or n = 1357 (Cormack-Jolly-Seber) and Golden Redhorse was estimated at n = 1084(Schumacher-Eschmeyer) and too few recaptures occurred to estimate using the Cormack-Jolly-Seber model. Two distinctive size classes were found for both species of Redhorse, which were grouped largely by age. Only one individual was aged at 4 years for Golden Redhorse, perhaps indicating a failed year class for the population or failure to capture Redhorse in that year class. Using life-table modeling, the Black Redhorse population in the upper Oconaluftee system was estimated to be slightly increasing. Electrofishing surveys were most efficient at lower discharge levels for capturing Redhorse. The von Bertalanffy growth models show similar parameters to those from other Southern Appalachian populations as well as Black Redhorse populations in Canada. Weekly larval fish samples were taken from April to August 2017 using drift nets and light traps to assess Redhorse reproductive occurrence and reproductive output. Larval catostomids were captured at all sampling locations with the majority (>70%) being identified as Moxostoma, indicating that there are reproducing Redhorse populations throughout the entire upper Oconaluftee system. There was a significant difference in sample location, with the middle sample site having the highest drift density (Mean drift densities for each site from 4/26/17-6/4/17: Up=7.54 x 10^{-3} larvae/m³, Mid=2.93 x 10^{-2} larvae/m³, Low=3.10 x 10^{-3} larvae/m³).

INTRODUCTION

Plant and animal diversity in the southern Appalachian Mountains is greater than the entire region of Northern Europe (Clark 2001). This high biodiversity is a result of large amounts of rainfall, varying topography, and a multitude of habitats present within the region (US Fish and Wildlife Service 2012). The age of southern Appalachian river drainages has contributes to a remarkably diverse fish assemblage with many endemic species (US Fish and Wildlife Service 2012, Cooke et al. 2005). Anthropogenic activity and urbanization of the southern Appalachian Mountains threatens water quality and habitat viability for many of these fishes. As streams run from the headwaters to lower elevations, construction, road, and agriculture runoff result in increased sediment and contaminant concentration throughout rivers (Eastern Band of Cherokee Indians, EBCI 2015). Historic land use has left some stream banks destabilized, and introduction of exotic or invasive species can result in ecosystem altering effects on both stream channels and their riparian buffers (EBCI 2015). Habitat fragmentation due to impoundments results in habitat loss, decreased population continuity, and impedes movement so that spawning grounds become unreachable (Cooke et al. 2005, Dynesius and Nilsson 1994). Impoundments also produce varied flow regimes and altered water quality (Freeman et al. 2001, Cooke et al. 2005, Rolls et al. 2013). Furthermore, fish passage mechanisms implemented to reduce the impact of dams only allow small fractions of targeted species to complete their migrations (Santos et al. 2006, Brown et al. 2013).

The southeastern United States is home to 44 species of Catostomidae (the Sucker family), of which the genus *Moxostoma* comprises 17 species (Cooke et al. 2005). *Moxostoma* were named as a misspelling of *Myzostoma*, which means, "sucking mouth," and their common

name of "Redhorse" is a testament to the large body size and red fins present on most of the species (Jenkins and Burkhead 1993). Redhorse are typically found in mid- to large-sized rivers that exhibit a moderate gradient (Jenkins and Burkhead 1993). As a potadromous fish, Redhorse rely on long stretches of continuous river systems to migrate upstream to viable spawning grounds (Favrot 2009, Jenkins and Burkhead 1993, Reid 2006a, Reid 2006b). Completion of their life cycle relies on availability of sediment-free, well-oxygenated substrate (Balon 1975, Reid 2006a).

Dams in riverine ecosystems have the potential to alter species richness and diversity, leading to changes of community assemblage composition (Santos et al. 2006, Reid et al. 2007). Reductions in habitat availability, population size, and fragmentation from dams have imperiled many Redhorse species (Favrot 2009, Fisk et al. 2015, Lamprecht and Scott 2013), increasing the potential for local extirpation (Morita and Yamamoto 2002, Reid 2006b). Species constrained to small or fragmented populations and habitat reaches are at an elevated risk of extinction due to genetic, demographic, and environmental stochasticity (Favrot 2009, Shaffer 1981).

Six species of Redhorse currently inhabit the Hiwassee and Little Tennessee River systems of the southern Appalachian Mountains (Favrot 2009). The Oconaluftee River, located on the Qualla Boundary in western North Carolina, is a tributary to the Tuckasegee River within the Little Tennessee River system (Figure 1). The Oconaluftee River hosts a diverse assemblage of fishes consisting of 33 native and 5 nonnative species (personal communication Mike LaVoie, EBCI). Five species of catostomid have been historically documented within the river: White Sucker (*Catostomus commersoni*), Northern Hogsucker (*Hypentelium nigricans*), Black Redhorse (*Moxostoma duquesnei*), Golden Redhorse (*M. erythrurum*), and Sicklefin Redhorse (*M. sp.*).

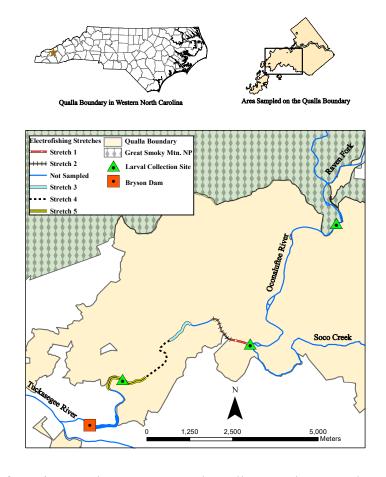


Figure 1: The Oconaluftee River, Jackson County and Qualla Boundary, North Carolina, with electrofishing sample reaches and larval collection sites identified.

All three Redhorse species purportedly feed on benthic invertebrates, algae, and detritus (Jenkins and Burkhead 1993, Jenkins 1999). In the Little Tennessee and Hiwassee drainages, spawning for each species typically begins in April and can last into June (Favrot 2009, Jenkins and Burkhead 1993). Spawning occurs on gravel, cobble, and small rubble within runs and riffles (Favrot 2009, Jenkins and Burkhead 1993). The largest abundance of Redhorse are present in clear streams with moderate to high gradients (Jenkins and Burkhead 1993, Jenkins 1999, Trautman 1981); however, some Redhorse have been found persisting in reservoirs (Jenkins and Burkhead 1993). Golden Redhorse occupy the broadest spectrum of warm water habitats and

persist in streams with moderate siltation (Jenkins and Burkhead 1993). Golden Redhorse however are less prevalent in cooler high gradient streams populated by Black and Sicklefin Redhorse (Jenkins and Burkhead 1993, Jenkins 1999). Sexual maturity and longevity varies among each species. Black Redhorse mature within 2-6 years and have been aged up to 17 years (Jenkins and Burkhead 1993, Reid 2009). Golden Redhorse reach maturity at 3-5 years can live up to 11 years (Jenkins and Burkhead 1993). Sicklefin Redhorse males reach maturity at 5-7 years and females at 7-8 years, with a lifespan of 17-20 years (Jenkins 1999). Sicklefin Redhorse use similar habitat as Black and Golden Redhorse and are thought to have similar life histories, but evidence of persistent populations above Bryson Dam on the Oconaluftee River can only be found for Black and Golden Redhorse.

Local extirpation of the Sicklefin Redhorse in the upper Oconaluftee River likely occurred due to habitat degradation and fragmentation resulting from the building of Bryson Dam. Duke Energy (EBCI 2015) currently operates this hydroelectric dam located at the mouth of the Oconaluftee River near its confluence with the Tuckasegee River. Bryson Dam prevents fish migration from the Tuckasegee River to the upper reaches of the Oconaluftee River, while simultaneously allowing outward migration through the dam's gates with no means of reentrance into the upper sections of the Oconaluftee River (Davis 2015, Stowe 2014).

Significance

Historically, sucker species were viewed as "rough fish" with low economic value, which led to them being overlooked for research and conservation efforts by state and federal agencies (Cooke et al. 2005). Gaps in knowledge of suckers are seen through the recent rediscovery of Robust Redhorse in 1980 and recognition of Sicklefin Redhorse as a distinct species in 1992 (Jenkins 1999, Cooke et al. 2005). The Sicklefin Redhorse had, however, been previously

recognized in Cherokee traditional ecological knowledge as a distinct fish species with the individual name "junghitla" (personal communication with Mike LaVoie, EBCI). Basic research, partnerships, political negotiation, and restoration programs are needed to preserve the diversity of suckers found in the southeastern United States (Cooke et al. 2005). Conservation and restoration of Redhorse population are lead by multiagency conservation committees (Candidate Conservation Agreement with Assurances 2002, Candidate Conservation Agreement 2015). These committees are composed of state, federal, and tribal agencies, private entities, and other non-governmental organizations that have provided political backing and funding to restore both Robust and Sicklefin Redhorse (CCAA 2002, CCA 2015).

Conservation committees and university researchers, together, can provide critical knowledge of Redhorse habitat needs, population genetics, and movement patterns (Davis 2016, Fisk et al. 2015, Davis 2015, Stowe 2014, Favrot 2009, Moyer et al. 2009). Stocking of Sicklefin Redhorse fingerlings is being used in an effort to establish new populations and supplement current populations (Rakes et al. 2015). Sicklefin Redhorse movement and habitat usage were also monitored using radio telemetry (Davis 2015, Favrot 2009, Stowe 2014). Fyke net trapping, seining, and visual observations have been used on the Hiwassee Drainage to assess spawning populations of Redhorse (Davis 2016, personal communication with Brett Albanese, Georgia Department of Natural Resources, GADNR). Larval sampling and analysis has been conducted on the Valley River within the Hiwassee River system to investigate the early life history of the family Catostomidae (Ivasauskas 2017).

As of 2015, the Sicklefin Redhorse conservation committee has stocked > 12,000 fingerlings 2-4 months old, along with a small number of both 6-8" (<20) juvenile and adult (<12) Sicklefin Redhorse into the upper Oconaluftee above Bryson Dam (Rakes et al. 2015,

personal communication with Michael LaVoie, EBCI). No Sicklefin Redhorse were stocked during 2016 or 2017. Radio telemetry has been used to track and monitor habitat usage of stocked individuals (Davis 2015, Stowe 2014). Radio telemetry studies have tracked transplanted adults and juveniles moving downstream to Oconaluftee Reservoir (the reservoir created by Bryson Dam), with some individuals passing through the gates of the Bryson Dam to the Tuckasegee River (Davis 2015, Stowe 2014). Despite stocking efforts, Sicklefin Redhorse populations have not become established based on limited sampling attemp (personal communication with Michael LaVoie, EBCI).

While the Black Redhorse is one of the more common Redhorse species in the southern Appalachian Mountains and within the Mississippi drainages, research on their population demographics in the southern Appalachian region is lacking (Davis 2016, Shumate 1988). Other Black Redhorse populations, such as those in the Mizzouri, Ozarks and southern Canada, have been comprehensively studied (Bowman 1970, Kwak and Skelly 1992, Howlett 1999, Reid 2006a, Reid 2006b, Reid 2009). One driving factor in research of the Canadian populations is the Black Redhorse's listing as threatened by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) (COSEWIC 2015). Population modeling has played a key role in assessing the health of Canadian populations, providing important estimates of reproductive rate and population dynamics (Young and Koops 2014, Vélez-Espino and Koops 2009).

Assessment of southern Appalachian Black Redhorse populations and their habitat could lead to comparisons among Black Redhorse populations and identify species needs and sensitivity.

The Cherokee people historically ate Redhorse as a source of protein during the spring of the year (Cantrell 2005). V-shaped rock weirs used to catch fishes can be found throughout the region historically occupied by the Cherokee (Cantrell 2005). The Redhorse was also commonly

prescribed as a cure for a breathing disorder thought to be caused by insects in the water (Cantrell 2005). Redhorse population assessment was identified as a priority research goal in a recent resource management plan (EBCI 2015). This assessment conducted in conjunction with EBCI provides quantitative population data on Redhorse population abundance, demographics, and reproductive output in the Oconaluftee River. This information is a necessary component to maintaining healthy populations and habitats on tribal lands (EBCI 2015).

This population assessment was done to (1) Evaluate Redhorse population health through population abundance estimates and population demographics, (2) Determine if a Sicklefin Redhorse population persists in the upper Oconaluftee River, and (3) Assess Redhorse reproductive output throughout the upper Oconaluftee River.

METHODS

The Oconaluftee River is formed by the confluence of Kephart Prong, Kanati Fork, and Smith Branch on the eastern side of Great Smoky Mountains National Park (Stowe 2014), and flows for approximately 30 km to join the Tuckasegee River near the community of Ela. The Oconaluftee River's gradient varies over the length of the river with its headwaters at an elevation of 1,611m above sea level falling to 544 m above sea level at its confluence with the Tuckasegee River. The headwaters are characterized by a moderately steep gradient with cool fast moving water amongst many large boulders, becoming less steep and containing more riffles, runs, and deep pool habitats following the confluence with Bradley Fork tributary (Stowe 2014).

Adult and Juvenile Population Survey

Fyke Net

We deployed a fyke net across the upper Oconaluftee River upstream of the confluence with Soco Creek on April 17, 2017. A 1.22 m x 1.52 m fyke net with multiple 7.62 m long x 1.22 m deep wing extensions, and a custom net holding box 0.76 m tall x 1.22 m wide x 1.22 m long (Duluth Nets, Duluth, MN) was placed in the river to capture migrating Redhorse as they made their spawning run upstream. The net contained two 12.7 cm exclusion rings on each side to prevent turtles from entering the box area. A similar fyke net setup was successfully used by Georgia Department of Natural Resources to survey Redhorse populations in 2016 and 2017 on Brasstown Creek, of the Hiwassee River Drainage in Georgia (personal communication with Brett Albanese, GADNR).

After initial deployment, I left the net overnight and checked it the next morning. Increased flows due to a thunderstorm event lifted the net from the bottom of the stream, decreasing the ability of the net to funnel fish into the trap section. Three large holes were also found in the net likely due to otters that I saw near the sampling site. One hole was located in the wing section of the net, another at the entrance to the trap section, and a third at the trap holding box. Due to high flows and a large predator population in the Oconaluftee River, the fyke net method was considered infeasible for sampling this stream. GADNR experienced similar difficulties when deploying fyke nets on larger streams such as the Ocmulgee and Nottley Rivers (personal communication with Brett Albanese, GADNR), indicating this technique may only be feasible in smaller streams that lack large predation risk.

Electrofishing

We conducted five electrofishing surveys from April to September 2017 using a twoperson cataraft electrofishing system. The cataraft was equipped with a 2.5 kW generator
powered pulsator (GPP) electrofisher (Smith-Root, Inc.). Due to equipment malfunctions, the
fourth and fifth reaches sampled during the fourth survey were done using a 5.00 kW GPP
electrofisher (Smith-Root, Inc.). Voltage was adjusted for water conductivity, flow, and depth to
allow for efficient capture, while preventing a fish frightened response or electrical-induced
mortality. During longer and hotter sampling runs, an aerator was mounted to the holding
container and ice was added to the water to reduce stress and mortality. Fish were netted using
an eight-foot fiberglass pole attached to a dip net with quarter-inch mesh for the first four
surveys and half-inch mesh during the fifth survey.

All surveys were done on the same reach of the Oconaluftee River from the confluence of Soco Creek to the lower end of the Qualla Boundary (Figure 1, Appendix A). This area was

chosen for sampling because it had adequate flow for the cataraft and lower amounts of recreational use. The portion of the Oconaluftee River from (35°28'28.02"N:83°20'15.18"W) to (35°28'23.48"N, 83°20'38.57"W) was not sampled due to a lack of streamside access. Each survey was completed in the shortest time period possible to reduce possibility of fish movement during each survey (3-17 days). Fish were processed streamside on average after approximately 30 minutes of electrofishing to reduce stress on captured Redhorse and lessen mortality. Nontarget species captured were removed from the raft holding container on-stream to reduce fish stress. Catch per unit effort (CPUE) was determined using active fishing time recorded by the electrofisher. CPUE was further analyzed in comparison to discharge during the sampling events. Water temperature and specific conductance was measured at processing sites using a YSI 85 meter (YSI Inc./Xylem Inc.). The United States Geological Survey (USGS) gauging station (03512000) located in Birdtown, NC provided other environmental data such as discharge.

Redhorse Handling and Processing

All Redhorse were processed streamside. At the end of each sampling event we portioned fish into separate containers. Each fish was identified to species and total length and wet weight was measured. Species was determined by presence of snout tubercles (present in reproductive male Golden Redhorse, absent in reproductive male Black Redhorse), lateral line scale counts (usually 44-47 for Black Redhorse, 40-43 for Golden Redhorse), and morphological variation in body shape and color (Jenkins and Burkhead 1993). Tubercle development, health condition, and reproductive condition were assessed and recorded. Sex was only determined if the fish was in a reproductive state. Gamete maturation was classified by stage (i.e. not ripe, hard squeeze ripe, slight squeeze ripe, running ripe) (Favrot 2009).

A tissue sample was taken from each fish for future use as a DNA sample. The small, approximately 5 mm³ caudal fin clip was placed in a 2 mL microcentrifuge tube containing non-denatured 95% EtOH (following the protocol provided by the U.S. Fish and Wildlife Service's regional Conservation Genetics Laboratory, Warm Springs, GA). The scissors used to collect the sample were wiped between samples to prevent contamination. The most anterior pectoral fin ray of each fish was cut as close to the body as possible using wire cutters and placed in a coin envelope to dry (Reid and Glass 2014, Reid 2007). Each fish was implanted with a 134.2 kHz 12 mm HDX Passive Integrated Transponder (PIT) tag (Oregon RFID, Biomark Inc.) near the base of the dorsal fin (Favrot 2009). If the fish was too small or appeared stressed it was excluded from being PIT tagged. Redhorse captured on subsequent electrofishing surveys were scanned for a PIT tag to determine recapture. If a fish was recaptured within the same survey event, it was excluded from the population abundance estimation. Fish were then immediately released upstream of electrofishing direction.

Redhorse Aging

Once each fin ray had dried completely, it was prepared for sectioning following the protocol described by Koch and Quist 2007. The cap from a 2 mL microcentrifuge tube was removed and the bottom conical part of the tube was removed to create a mold for each ray. I placed non-drying modeling clay into the cap of each 2 mL microcentrifuge tube to fix each fin ray vertically in the tube. The distal end of each fin ray was placed into the modeling clay with the proximal end exposed. I cut large fin rays at the distal end to prevent it from protruding out of the tube. After rays were fixed vertically, the body of the microcentrifuge tube was placed over the ray. Tubes were then filled from the bottom with an optically clear two-part epoxy (Epoxycure 2, Buehler Inc., Lake Bluff, IL) using a 30 mL syringe.

After the epoxy hardened, I used a three-blade low speed saw (IsoMet Low Speed Precision Cutter, Buehler Inc., Lake Bluff, IL) provided by the NC Wildlife Resources Commission (Balsam Depot), to cut two 1 mm sections from the proximal end of each fin ray. Sections were then mounted onto microscope slides using a toluene-mounting medium (Permount, Fisher Scientific, Hampton, NH). Fin rays were magnified under a dissecting scope (SMZ-168 Series, Motic Instruments Inc., Xiamen, China) using transmitted light. I then digitized each fin ray using a high-definition camcorder (Vixia HF M52, Canon Inc., Tokyo, Japan) attached to the dissecting scope. I used the Fiji distribution of imageJ (Schindelin et al. 2012) to adjust contrast and brightness of each image to clarify annuli (Figure 2). Annuli were distinguished as translucent bands following an opaque growth zone (Rude et al. 2013, Chalanchuk 1984). Due to uncertainty of identifying new growth after the edge annulus on each fin ray, each fish was aged with a (+) to account for development towards the next age class (Chalanchuk 1984). All aging was done blindly of fish size and species.

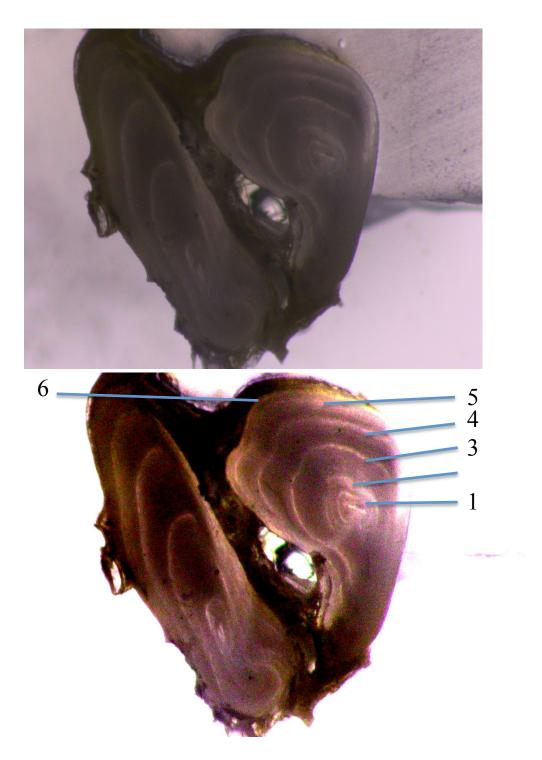


Figure 2: (Top) Redhorse fin ray viewed with transmitted light under dissecting scope, (Bottom) The same Redhorse fin ray enhanced using imageJ software (Fiji). Numbers represent age designation for each annulus.

Redhorse Population Modeling and Demographics

Abundance Estimates and Demographics

I conducted all analyses using R (v. 1.1.383, R Core Team 2016). The FSA: Fisheries Stock Assessment package (Ogle 2017) allowed me to estimate population abundance using both the Schumacher-Eschemeyer and Cormack-Jolley-Seber approach. I used both approaches to produce population abundance estimates for the Black Redhorse population, but only used the Schumacher-Eschemeyer approach for Golden Redhorse, as the Cormack-Jolly-Seber was unable to produce an estimate due to the low number of recaptures. The Cormack-Jolly-Seber model was considered a more appropriate approach for the sampling protocol used as it accounts for an open population. The assumption of a closed population in the Schumacher-Eschemeyer was violated for this system as immigration and emigration of Redhorse in the surveyed reaches was possible, however, this approach was still used for both species as a way to produce rough abundance estimates and to be able to compare Black and Golden Redhorse population size. A von Bertalanffy growth models (VBGMs) was produced for Black and Golden Redhorse using length-at-age data: $TL = L_{\infty}(1-e^{-K(t-t_{\theta})})$ (Ogle 2017). In this model L_{∞} is the mean asymptotic total length, t is the age, t_0 is the hypothetical age at length 0, and K is the Brody growth co-efficient (Ogle 2017).

Life Table Modeling

I built a static life-table model for the Black Redhorse population based on the 2017 electrofishing catch and age data. A weighted catch curve was created for Black and Golden Redhorse to estimate instantaneous (Z) and annual (A) mortality (Ogle 2017). Z was estimated using the slope of the catch curve and converted to A using the equation $A = I - e^{-Z}$ (Ogle 2017). Z was also estimated using the Hoenig (1983) empirical relationship between maximum

longevity of fishes and mortality: $Z = 4.22(T_{max})^{-0.982}$. T_{max} is the maximum age observed for the population during sampling. I produced a survivorship curve for both Black and Golden Redhorse for fish aged greater than or equal to one year of age based on 2017 electrofishing data. Survivorship (l_x) is the proportion of individuals surviving to each age group out of the original number of individuals. I then estimated the number of individuals alive at age 0 by multiplying n_x (number of surviving individuals at the start of age x) at age 1 by the proportion of individuals estimated to survive from age 0 to age 1 (0.00044) (Young and Koops 2014). The fecundity coefficient for age class x (m_x) is represented in Equation 1.

(1)
$$m_x = \sigma_x f_x$$

Where σ_x is the annual survival probability from age x-1 to age x, and f_x is the age specific annual number of female offspring produced by an individual by age x (Young and Koops 2014).

$$(2) \sigma_x = \frac{l_{x+1}}{l_x}$$

(3)
$$f_x = \eta_x \varphi \rho_x (\frac{1}{T})$$

To calculate f_x , η_x is the average fertility for a female at age x, φ is the proportion of females present in the population (assumed to be 50%), ρ_x is the proportion of reproductive fish at age x, and T is the periodicity of spawning for Black Redhorse (assumed to be 1) (Young and Koops 2014). The average fertility (total number of eggs produced) for a female at age x (η_x) was estimated using the size-at-age equation relating standard length (SL) to fertility (Young and Koops 2014):

(4)
$$\eta = (2.46 \times 10^{-6}) SL^{3.713}$$

Total length at age was estimated using the VBGM for Black Redhorse (Figure 8, Table 1). Standard length (SL) was estimated from total length using the equation TL = 1.19SL (Reid

2009). To determine the probability of a female reaching maturity at age x, a cumulative binomial distribution was used (Young and Koops 2014). Jenkins and Burkhead (1993) report that Black Redhorse reach maturity in two to six years in the Tennessee River Drainage. It was assumed that by age six all females in the population had reached maturity. The net reproductive rate (R_0), mean generation time (G), intrinsic rate of increase (r), and finite rate of increase (λ) were then estimated using the life table.

Larval Redhorse Sampling

Larval fish were sampled from April through August of 2017 using drift nets and light traps to assess Redhorse reproductive output and location within the upper Oconaluftee River. In the Valley River of the Hiwassee River Basin, Northern Hogsucker larvae were found from late April to early May, Black Redhorse from early to mid-May, and Golden and Sicklefin from middle to late May using similar methods (Ivasauskas 2017). Due to the lack of knowledge of larval emergence in the upper Oconaluftee River system, sampling was conducted over a longer time period than was expected for catostomid emergence. This was done to account for possible variance in emergence timing due to temperature differences. Three approximately equidistant sampling sites located along the length of the upper Oconaluftee River were used to assess variation in larval Redhorse occurrence and drift density at different reaches of the river (Figure 1, Appendix A).

Drift Nets

During each sampling event, two rectangular drift nets (0.26 m x 0.45 m opening, 1 m long; 500- μ m mesh) were placed side-by-side in relatively shallow zones (0.13 – 0.76m deep) near the shore (\leq 2m from shore) to capture drifting larva (LaVoie 2007, Reichard et al. 2004). I anchored nets to the substrate using rebar, with the top of each net set even with the water

surface (LaVoie 2007). If the water was shallower than the net, the height of the water column was recorded on both sides of the net. Flow velocity was measured using a Swoffer 2100 flow meter (Swoffer Instruments Inc., Federal Way, WA) set in the middle of the net opening at the beginning and end of each set. The mean velocity and area of net opening under the river's surface was then used to calculate the volume of water sampled during the sampling period (LaVoie 2007). Nets were set for only 30 minutes to prevent clogging and backflow (Zitek et al. 2004). Larval fish drift typically peaks during the first hours of darkness (Janáč and Reichard 2016), so all samples were taken between 20:00 and 24:00 (LaVoie 2007), with the exception of one net which was removed three minutes after the proposed sampling window. Drift net sampling was began 22 May 2017 and was not done the week of 9 June 2017 due to high flows and thunderstorms. Because all sites could not be sampled simultaneously, the order at which each site was sampled was determined randomly to remove temporal bias throughout each sampling occasion. For each larval fish sample site and date, water temperature and specific conductance was measured using a YSI 85 meter (YSI Inc./Xylem Inc., Rye Brook, NY). The United States Geological Survey (USGS) gauging station (03512000) located in Birdtown, NC provided stream discharge, as well as local weather conditions.

Light Traps

Light traps were used to sample larvae concurrent to drift net sampling. Light traps were constructed using 20.3 cm PVC pipe (Figure 3) as a slightly smaller variation of a design successfully used by Ivasauskas (2017). A 90° section of PVC pipe was removed, and acrylic sheets were glued in place to create a funnel with an opening approximately 1 cm wide. A LED flashlight (EO1, Fenix Lighting, Lone Tree, CO) was placed vertically in the back of the trap with an approximately 3.8 cm² aluminum reflector placed above the light. The top and the

bottom of the trap were made of plastic pot saucers painted black. The bottom saucer had drain holes to allow water to escape when the trap was removed from the stream and was lined with 1 mm mesh aluminum screen to retain larvae. The top of the trap was held in place using a bolt and wing nut system. This allowed the top to be removed when checking the trap. The trap bottom, screen, bolts and acrylic sheets were all fixed to the trap using Liquid Nails Brand *Fuze*It* All Surface adhesive (PPG Industries Ohio, Inc., Cleveland, OH). The bolts were also held against the side of the PVC body using cable ties. Two looped cable ties were attached to both sides of the trap for anchoring with rebar.

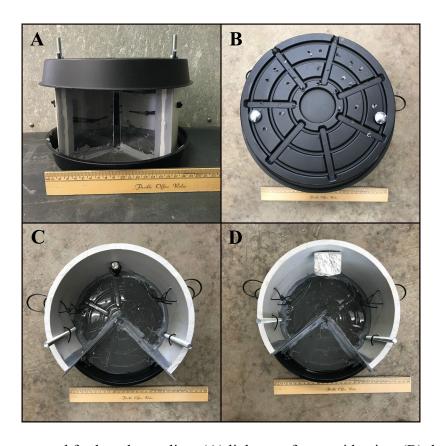


Figure 3: Light trap used for larval sampling, (A) light trap from a side view (B) the bottom of the light trap (C) interior of light trap without aluminum reflector (D) interior of light trap with aluminum reflector.

I placed one light trap at each of the three sampling sites (Figure 1) in still water areas to attract larvae through their phototaxis response (Peterson et al. 2008). A fourth light trap was added at the middle sample site the week of May 16, 2017 due to the loss of the still water side pool for the initial middle light trap due to high flows. Light traps were fished overnight and retrieved the following morning (Peterson et al. 2008). Water temperature and specific conductance were measured each time a trap was deployed or retrieved. Larvae collected from drift nets and light traps were immediately preserved in 95% ethanol.

Larval Fish Identification and Analysis

Drift net and light trap samples were stored in a refrigerator to slow down any tissue breakdown due to dilution of the 95% EtOH. Samples were filtered through a 500-µm sieve and the residue was inspected and all larval fishes were removed. Catostomids were then sorted from all other larval fish and identified to the lowest feasible taxon using keys in Wallus et al. (1990). Larval fish were projected onto a high definition monitor to aid in identification using a high definition camcorder (Vixia HF M52, Canon Inc., Tokyo, Japan).

Catostomid drift density over the sampling time was plotted to look for temporal peaks in drift to signify emergence of a species. A linear mixed effects model (lme4 and lmerTest packages, Bates et al. 2015 and Kuznetsova et al. 2017 respectively) was used to determine if there were differences in drift density of yolk sac larvae among sampling sites and the order in which sites were sampled with date as a random effect. Only yolk sac larvae data were used to in the linear mixed effects model to eliminate increases in larval drift that may be from the same species but in post-yolk sac absorption stage.

RESULTS

After five electrofishing surveys we captured 624 (81.9%) Black Redhorse and 138 (18.1%) Golden Redhorse. We captured the majority of fish just upstream or downstream of a riffle. Of those captured, we PIT tagged and released 460 Black Redhorse and 93 Golden Redhorse (Appendix B, Appendix C). We did not capture any Sicklefin Redhorse. We recaptured 47 (15.21% prior to last tagging event) Black Redhorse and 3 (4.54% prior to last tagging event) Golden Redhorse. We sampled all Golden Redhorse recaptures on the last survey event. CPUE decreased approximately exponentially as discharge increased (Figure 4).

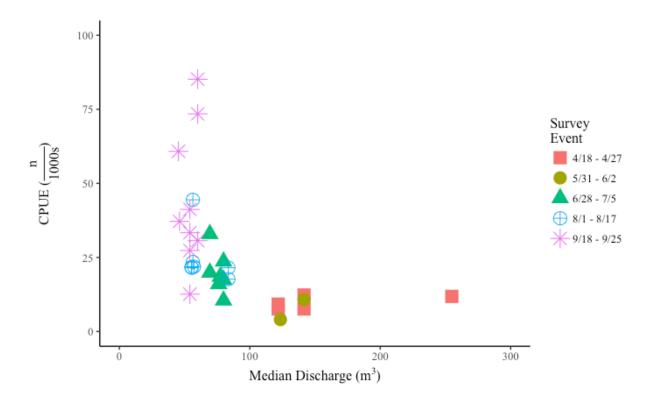


Figure 4: CPUE as a function of median discharge during 2017 electrofishing sampling, where n is the number of fish caught. Median discharge recorded by the USGS gauging station (03512000) during each sampling event.

Adult and Juvenile Population Demographics

The Black Redhorse population was estimated to be larger than the Golden Redhorse population. The Schumacher-Eschmeyer population estimate for Black Redhorse was N = 2101 (1540 – 3308) and for Golden Redhorse was N = 1084 (486 – 4697). The Cormack-Jolly-Seber population abundance, in which the population is seen as open due to immigration or emigration, estimate for the Black Redhorse population was N = 1357 (228-2485). Too few Golden Redhorses were recaptured to do a Cormack-Jolly-Seber estimate. Population abundance estimates using mark-recapture models had large confidence intervals due to the low number of recaptures of both Black and Golden Redhorse throughout the population surveys. Few recaptures occurred in different sections of the river indicating that throughout the summer these fish remained relatively stationary.

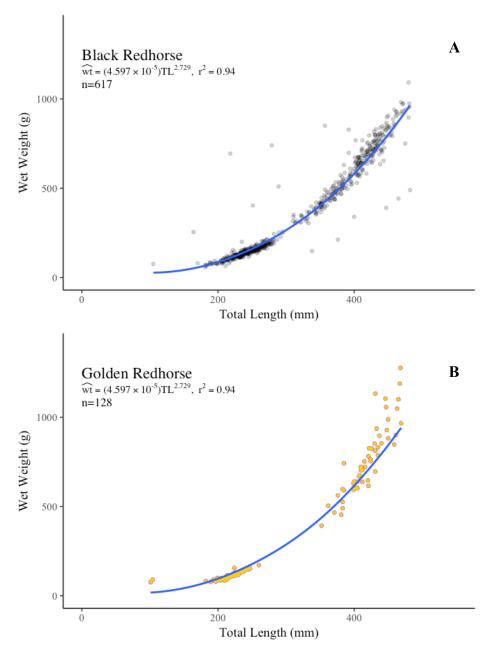


Figure 5: Total length and wet weight for Redhorse captured during the five 2017 electrofishing survey events of the upper Oconaluftee River. (A) Total length and wet weight for Black Redhorse. (B) Total length and wet weight for Golden Redhorse.

Total length of Black and Golden Redhorse were highly correlated with wet weight (Figure 5). Total lengths of fish with damaged caudal fins were not used. Captured Black Redhorse ranges of length, weight and age were 105 - 482 mm TL, 58 - 1092 g, and 1 – 12+

years respectively and 101 - 469 mm TL, 76 – 1276 g, and 1 - 10 years for Golden Redhorse. The majority of fish sampled were not in reproductive condition, however, the age at first reproduction found for Black Redhorse was 4+ for females and 5+ for males. The only reproductive female Golden Redhorse that was captured and aged was 7+ years. One reproductive male Golden Redhorse was captured but not aged due to errors made in mixing epoxy while preparing fin rays. As a result 588 out of 624 Black Redhorse and 127 out of 138 Golden Redhorse were aged.

Total lengths of both Black and Golden Redhorse conformed to a bimodal distribution separated largely by age (Figure 6). The lower mode consisted mostly of fish aged 1-3+ years, while the upper mode consisted of fish aged 4-12+ for Black Redhorse and 5-10 for Golden Redhorse. Only one 4+ Golden Redhorse was captured and aged (Figure 7).

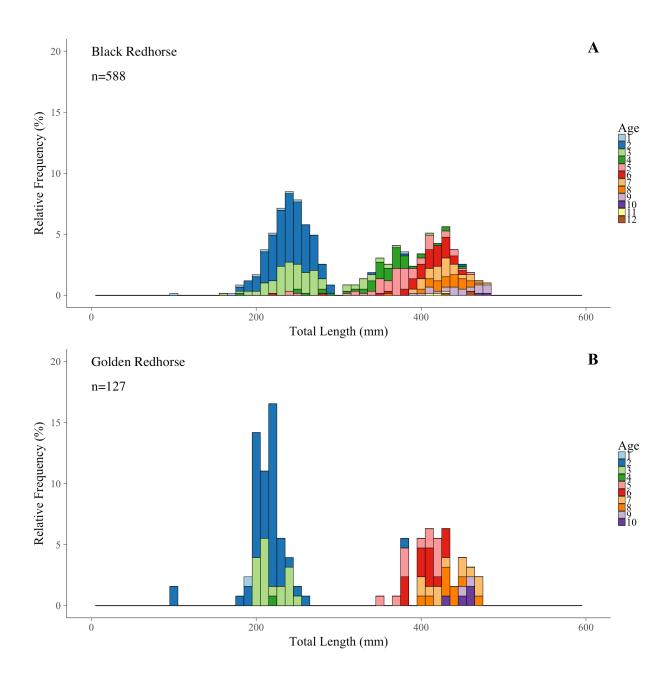


Figure 6: Total lengths of Black and Golden Redhorse captured form a bimodal distriburtion consisting mostly of younger and older age classes. (A) Stacked histogram of Black Redhorse total length by age. (B) Stacked histogram of Golden Redhorse total length by age.

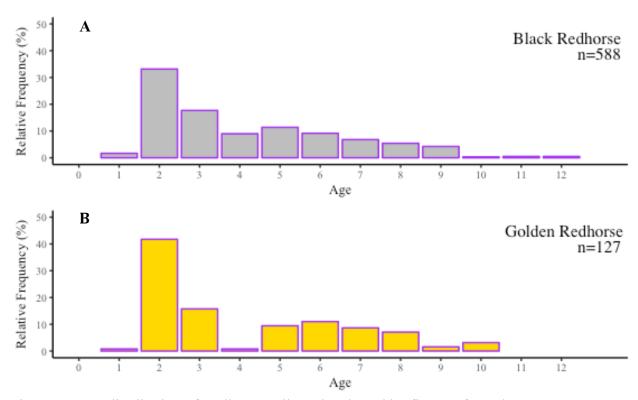


Figure 7: Age distribution of Redhorse collected and aged by fin rays from the upper Oconaluftee River in 2017. (A) Age distribution of Black Redhorse. (B) Age distribution of Golden Redhorse.

The von Bertalanffy growth model for both Black and Golden Redhorse begin to show a decrease in slope after age 2, which could represent species reaching sexual maturity and allocating less energy to growth and more to reproduction. (Figure 8, Figure 9). The asymptotic average length of Black Redhorse was less than that of Golden Redhorse, and Black and Golden Redhorse exhibited similar *K* values indicating that they grow in the same manner towards their asymptotic average length (Table 1).

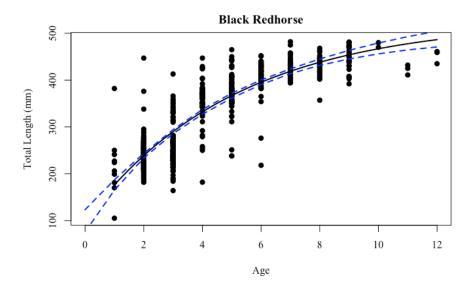


Figure 8: von Bertalanffy Growth Model for Black Redhorse captured during electrofishing surveys of the upper Oconaluftee River and 95% confidence intervals (dashed blue lines).

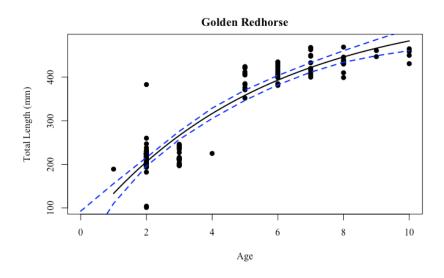


Figure 9: von Bertalanffy Growth Model for Golden Redhorse captured during electrofishing surveys of the upper Oconaluftee River and 95% confidence intervals (dashed blue lines).

Table 1: von Bertalanffy growth model parameter estimates for Black and Golden Redhorse sampled in the upper Oconaluftee River.

Species	<i>L</i> _∞ (95% CI)	K (95% CI)	t _θ (95% CI)
Black Redhorse	527 (488 – 566)	0.20(0.15-0.24)	-1.08 (-1.51 – -0.64)
Golden Redhorse	566 (464 - 668)	0.18(0.10-0.27)	-0.46(-1.13-0.21)

Life Table Modeling

Instantaneous mortality for Black Redhorse was estimated to be Z = 0.431 (0.294 – 0.568) and the annual mortality was found to be A = 35.0% (25.4% – 43.3%) when considering ages ≥ 2 (Figure 10). Instantaneous mortality for Golden Redhorse was Z = 0.318 (0.168 – 0.467) and the annual mortality was estimated at A = 27.2% (15.4% – 37.3%) for ages ≥ 2 . The ascending left limb from ages 0 to 2 indicate that these age classes were incompletely sampled using the cataraft electrofishing technique (Bowman 1970). Z was lower when using the Hoenig mortality estimate for Black Redhorse (Z = 0.368) and higher (Z = 0.440) for Golden Redhorse. The lower Hoenig mortality estimate for Black Redhorse is due to an older maximum age being sampled in the population than Golden Redhorse.

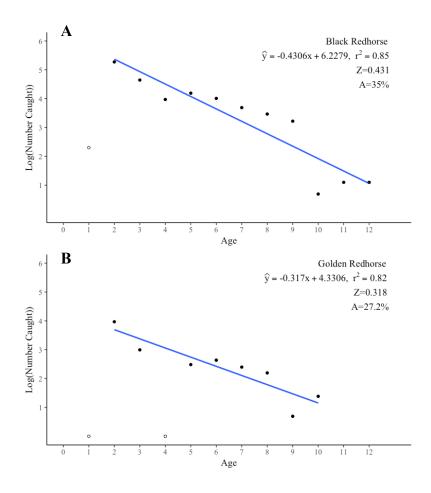


Figure 10: Catch curves of Redhorse over five electrofishing samples of the upper Oconaluftee River. (A) Catch curve for Black Redhorse. (B) Catch curve for Golden Redhorse. Age one was excluded for Black and Golden Redhorse to begin the regression line at the decreasing slope. Age 4+ was also excluded for Golden Redhorse to produce a curve with better fit.

The survivorship curves for Black and Golden Redhorse produced from electrofishing data both exhibit a Type 2 curve (Figure 11), suggesting an approximately constant mortality rate for these ages. This curve however does not include age 0 fish. Including age 0 fish would like produce a Type 3 curve typical of fish species due to high mortality rates early in life. The high survivorship from age 1 to age 2 for these data is due to the low number of age 1+ fish captured. It would be expected that there would be many more age 1+ fish than age 2+ fish, indicating that

the electrofishing technique may not have adequately sampled age 1 and younger year classes, or that age 1+ may be a failed year class.

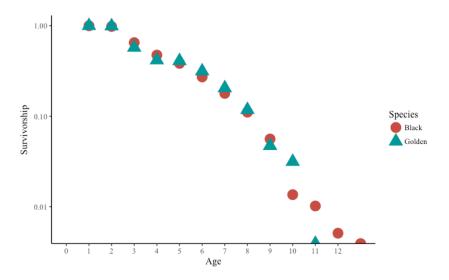


Figure 11: Survivorship curve for Black and Golden Redhorse captured using electrofishing in upper Oconaluftee River.

The net reproductive rate (R_0) for Black Redhorse based on survivorship and fecundity estimates (Table 2) was estimated to be 1.33, suggesting that the population was stationary to slightly increasing (Table 2). The resulting estimated mean generation time (G) was 5.99 yrs, finite rate of increase was estimated at $\lambda = 1.02$, and the estimated intrinsic rate of increase (r) was 0.02.

Table 2: Black Redhorse life-table constructed using 2017 upper Oconaluftee River electrofishing data.

Age	Total Length	Number of Individuals Surviving at Start of Age x	Proportion of Individuals Surviving at Start of Age x	Number of Eggs Produced for a Female at Age x	Proportion of Females Mature at Age x	Number of Female Offspring Produced by Individual at Age x	Fecundity Coefficient at Age x
(x)	(TL)	(n_x)	(l_x)	(η)	(ρ _x)	$(f_{\mathbf{x}})$	(m_x)
0	100	1336952 ¹	1	0.00	0	0	0
1	176	588	4.40×10^{-4}	0.00	0	0	0
2	239	578	4.32×10^{-4}	873.82	0.0625	27.31	26.84
3	290	383	2.86×10^{-4}	1791.90	0.3125	279.98	185.53
4	332	279	2.09×10^{-4}	2960.84	0.6875	1017.79	741.42
5	367	226	1.69 x 10 ⁻⁴	4295.70	0.9375	2013.61	1631.10
6	395	160	1.20 x 10 ⁻⁴	5644.09	1	2822.05	1997.91
7	419	105	7.85×10^{-5}	7026.03	1	3513.01	2305.42
8	438	65	4.86×10^{-5}	8283.67	1	4141.83	2563.99
9	454	33	2.47 x 10 ⁻⁵	9464.06	1	4732.03	2402.42
10	467	8	5.98 x 10 ⁻⁶	10510.00	1	5255.00	1273.94
11	478	6	4.49×10^{-6}	11458.96	1	5729.48	4297.11
12	486	3	2.24 x 10-6	12187.36	1	6093.68	3046.84
13	NA	0	0	NA	NA	NA	NA

Number of individuals surviving at age 0 was computed using survival estimate from age x to age x+1 from Young and Koops 2014.

Larval Catostomidae

Over the course of the sampling period, I captured 90 catostomid larvae out of 660 total larval fish sampled in drift nets (Appendix D). I only caught 6 catostomid larvae in light traps out of a total of 155 larval fish sampled (Appendix E). The majority of the catostomid larvae captured were in the yolk sac stage (n=56). Peaks in yolk sac larval drift densities occurred in late April to early May and mid- to late- May (Figure 11). Post yolk sac larval drift density peaked in early June with a small spike in drift density in early July (Figure 12). Four of the six catostomid larvae captured in light traps were captured in early June, concurrent with the spike in post yolk sac larval drift density.

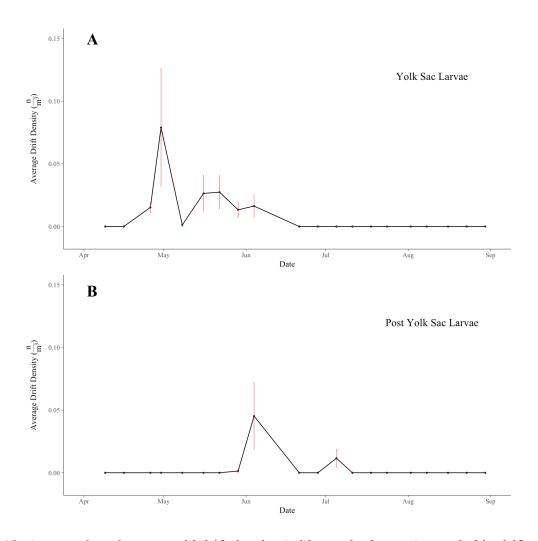


Figure 12: Average larval catostomid drift density (with standard errors) sampled in drift nets from the upper Oconaluftee River from April through August 2017. (A) Average drift density of yolk sac larvae. (B) Average drift density of post yolk sac larvae.

Of the yolk sac catostomids sampled during the peak in drift density in late April to early May, 74 % were identified as *Moxostoma*, and of the catostomids caught from mid-May to early June, 80% were identified as *Moxostoma*. Timing of larval drift was also consistent with Redhorse emergence reported by Ivasauskas (2017) in the Valley River. There is evidence to suggest a significant difference in order in which sites were sampled and among the three sites (Table 3, Figure 13, Figure 14). Similarity between larval drift due to sample order and larval drift differences between sites is likely due to the lack of randomization for the first 7 weeks of

the study. The highest drift density occurred at the middle sampling site and during the second sampling time, however the results were biased by one sampling event on 30 April 2017. Catostomid larvae were captured at all sites, with the highest density found at the middle sampling site (Figure 13).

Table 3: ANOVA summary for fixed effects for yolk sac catostomid larvae sampled using drift nets on the upper Oconaluftee River in 2017. Date was considered a random effect.

Fixed Effect	SS	MS	numerator df	denominator df	F	P
Site	0.00374	0.00187	2	96	2.543	0.0840
Order	0.00446	0.00223	2	96	3.039	0.0525

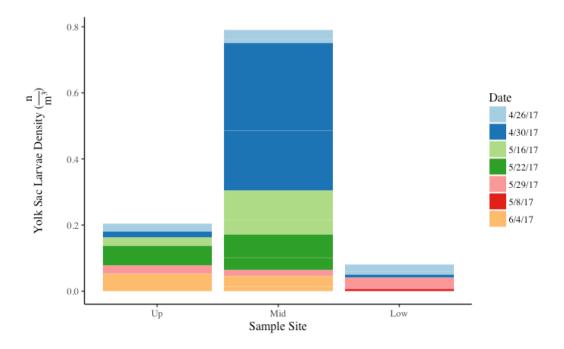


Figure 13: Yolk sac larval density sampled at each site. Bands across each bar represent different larval sampling dates. Two of the same color indicates captures by the two drift nets at each site for each date. Low is the furthest downstream site, Up is the furthest upstream sampling site, and Mid is the site found between the Low and Up sites (Figure 1).

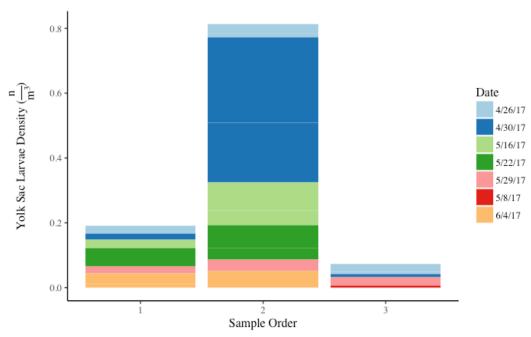


Figure 14: Yolk sac larval densities for the order sites were sampled during each sampling event. Samples during time 1, 2, and 3 were taken from 20:11- 21:15, 21:06-22:18, and 22:02-23:10, respectively, for all samples from 4/16/17 to 8/30/17. Samples on 4/9/17 were taken approximately one hour later than all other sampling times.

DISCUSSION

Adult and Juvenile Sampling

Sampling mid-sized, relatively high gradient streams such as the Oconaluftee River can prove challenging due to the flashiness of the system, width of the stream, and large variation in habitat types from shallow riffles to deep pools. While fyke nets have been used successfully in other streams (personal communication with Brett Albanese, GADNR), the use of a fyke net to sample the Oconaluftee was not feasible due to the stress on the wing sections that spanned over 30 meters. A relatively small rise in discharge from an afternoon thunderstorm began to foul the net and eventually lifted it from the bottom of the stream even though it was weighted with boulders and staked at approximately 2-3 m intervals. It would be a very time intensive task to maintain and clean extremely long wing sections during the sampling time frame. One solution to reducing the impact of flow on the net would be to stretch the fyke net across only a section of the river to avoid high flow areas (personal communication with Brett Albanese, GADNR). This strategy may not be suitable for sampling a system as large as the Oconaluftee, as it could result in missing perhaps a majority of moving fish. Predators also impact the effectiveness of a fyke net. While the net we used had turtle exclusion rings, animals (probably otters, which are common in the area) chewed through the mesh and entered the trap. Results of this attempt as well as other attempts to deploy fyke nets in mid to large size streams (Nottley and Ocmulgee Rivers) by the GADNR indicate that fyke nets are best suited for sampling smaller streams with lower discharge and lower mammal predator density.

Raft electrofishing was an effective method for sampling the large variety of habitat units within the upper Oconaluftee River. The cataraft system was more versatile than typical boat

electrofishing system because rafts can float over shallow riffles while still being able to sample deep holes. The cataraft electrofishing system captured Redhorse throughout the course of the entire survey period. We captured the majority of Redhorse upstream or downstream of riffles, which was also found for Ozark populations (Bowman 1970). Increased CPUE at lower discharge was likely due to easier raft maneuvering, slower fish drift rate, water clarity, and shallower pools. Sampler experience increased and discharge decreased as the summer progressed, so this result may be biased by an increase in sampler experience. Bowman (1970) found an increase in catch of Black Redhorse and lower discharge levels in Ozark Highland streams. Surveying in mid to late summer may allow for more efficient capture due to seasonally lower discharge levels, but spawning occurs in March, April, and possibly May. Sampling during springtime is still vital for understanding migration and reproductive capacity of the population is to be gained.

The recapture percentage for Black and Golden Redhorse was relatively low (15.2% Black, 4.5% Golden). This could indicate that cataraft electrofishing may be inefficient at capturing a large percentage of the Redhorse population. When sampling for a rare fish like Sicklefin Redhorse, low capture efficiency may result in false negatives for the presence of rare species. Recapture percentage could also be affected by extraneous variables such as fish movement outside the survey area or mortality between surveys. Increased effort of shocking throughout the surveyed reaches may allow for a greater number of recaptures. Using abundance estimation models that incorporate other variables such as amount of particular habitat available may also allow for a more accurate estimate of population size, but more complicated models may necessitate even larger sample sizes.

Population Assessment

Redhorse species composition from our samples within the upper Oconaluftee River differed considerably from those reported by Reid (2009) for the Grand River in Ontario. In the upper Oconaluftee River, Black Redhorse composed 81.8% of the catch while Golden Redhorse were only 18.1%, compared to the Grand River where Black Redhorse were 12.2% of the catch and Golden Redhorse were 33.2 % of the catch with all other *Moxostoma* captured consisting of Shorthead, Greater, River, and Silver Redhorse (Reid 2009). Catch curve instantaneous mortality estimates of Redhorse were higher than those in Canada (Z=0.26, Reid 2009). Hoenig instantaneous mortality estimates were also higher than reported for the Grand River population (Z=0.26, Reid 2009) but lower than estimated for the James and Elk Rivers, Missouri (MO) (Z=0.44-0.49, Howlett 1999) and the Roaring River, Tennessee (Z = 0.55, Shumate1988). Annual mortality for Golden Redhorse was lower than that reported by Quist and Spiegel (2012, Z=0.40%) in Iowa streams.

The lack of Golden Redhorse at age 4+ indicates there could have been a failed year class for the Oconaluftee population. No notable difference in discharge occurred on the USGS Oconaluftee River gauge (03512000) the year that the age 4+ fish would have been spawned to explain the gap. Other explanations of the two distinct size groups could be of biological significance such as reaching maturity or a new food source becoming available once a certain threshold size is reached causing a jump in growth rate. Age 4+ for Black Redhorse is also lower, which lends some support for an environmentally induced reduction in spawning. A low number of age 1+ could indicate another year class failure, however, it may also represent ineffectiveness of the gear to sample smaller fish.

The Grand River, Ontario Black Redhorse population was more similar to *Moxostoma* populations sampled in the southern Appalachians than those of Ozark streams in Missouri (Table 4). While age at first reproduction was higher than previously reported by Jenkins and Burkhead (1993), too few reproductive fish were sampled to determine variance of reproductive ages in the upper Oconaluftee River. Length and age distribution of both Black and Golden Redhorse were similar to those of the Grand River, Ontario reported by Reid (2009). A drop in size and age frequency was seen for Black Redhorse around 320 mm TL and at age 4+. There was a subsequent increase in frequency of larger than 320 mm and older individuals aged >4+. The age structure for Redhorse in the James River, MO (Beckman and Howlett 2013) varied from both Oconaluftee River and Grand River due to a low frequency of individuals after age 4+. Overfishing pressures in Missouri affecting larger individuals that are maturing within the population may contribute to this drop (Howlett 1999, Beckman and Howlett 2013). At maturation these fish begin to aggregate into shallow riffle habitat where they are more likely to fall prey to fishing (Beckman and Howlett 2013). The same drop in frequency at age 4+ can been seen in both Canadian populations and Oconaluftee River Black and Golden Redhorse populations, however, frequency of fish greater than 4 persist. This drop in frequency of individuals at age 3 to 4 could represent an age at maturation ontogenetic niche shift that causes this portion of the population to be less likely to be sampled through cataraft electrofishing. The drop in frequency of age 4+ fish in the James River, MO population could be a part of the species' typical life history, but fishing pressure could prevent fish from reaching older ages seen on the Oconlauftee and Grand Rivers.

The decrease in slope seen on the von Bertalanffy Growth Models for Black and Golden Redhorse after age 2 align with the age that both species are thought to reach maturity in the

Tennessee River system (Jenkins and Burkhead 1993). Diverting energy from growth to reproduction could produce the decrease in yearly growth rate exhibited in both species. A larger asymptotic average length for Golden Redhorse was expected as they have a larger maximum size in other populations (Jenkins and Burkhead 1993). Sicklefin Redhorse are also one of the larger Redhorse species with a maximum length approximately 650 mm (Jenkins 1999). A smaller population of the larger Golden Redhorse species and lack of Sicklefin Redhorse could indicate that the upper Oconaluftee River does not provide optimal habitat or forage for these larger bodied fishes to thrive.

The estimated L_{∞} and K values for Black Redhorse in the upper Oconaluftee River were similar to those estimated for the populations from Brasstown Creek, GA (Unpublished data from Dr. Jonathon Davis, Young Harris College) and Grand River, Ontario (Reid 2009), while the James River, MO population had a lower L_{∞} and higher K (Table 4). The estimated L_{∞} and K values for Golden Redhorse in the upper Oconaluftee River were similar to those found for the Brasstown Creek, GA population (Unpublished data from Dr. Jonathon Davis, Young Harris College), while the Iowa Rivers and James River, MO had lower L_{∞} and higher K values. The high L_{∞} and low K values of Golden Redhorse are also similar to those of Sicklefin Redhorse in Brasstown Creek, GA (unpublished data from Dr. Jonathon Davis, Young Harris College). The low K value for Golden Redhorse and Sicklefin Redhorse along with lower population densities could indicate that slower growth early in life may be a less advantageous strategy within the southern Appalachian drainages. Other variations in L_{∞} and K could be a result of varying habitat size and latitudinal climate differences from Georgia to Canada.

Table 4: von Bertalanffy growth model parameters for Sicklefin, Black, and Golden Redhorse. Variation among systems is likely due to variation in habitat as well as sample size.

Species	System	n	L_{∞}	K	Source
Sicklefin Redhorse	Brasstown Creek, GA	46	603	0.12	Unpublished data from Dr. Jon Davis
Black Redhorse	Grand River, Ontario	522	513	0.22	Reid 2009
	White/Arkansas River Tribs, MO	55	385	0.39	Beckman and Howlett 2013
	Brasstown Creek, GA	8	539	0.24	Unpublished data from Dr. Jon Davis
	Oconaluftee River, NC	588	527	0.20	This study
Golden Redhorse	Iowa Rivers	196	437	0.29	Quist and Spiegel 2012
	James River, MO	62	377	0.31	Beckman and Howlett 2013
	Brasstown Creek, GA	182	505	0.22	Unpublished data from Dr. Jon Davis
	Oconaluftee River, NC	127	566	0.18	This study

Other differences in life history could explain the lack of Sicklefin Redhorse in the upper Oconaluftee River. Black and Golden Redhorse begin to reach maturity from 2-6 and 3-5 years, while the Sicklefin Redhorse reaches maturity around 5-8 years of age (Jenkins 1999, Jenkins and Burkhead 1993). Reaching sexual maturity later in life may be a disadvantage in this system. Later age at first reproduction can increase generation time which is directly associated with net reproductive rate. While the Black Redhorse population was found to be stationary to slightly increasing, an increase in generation time without significant changes in other parameters such as fecundity could result in a declining population. Given the apparent almost constant mortality rate among juvenile and adult fish, longer time to maturation increases the probability of mortality of individuals before reaching maturity, which in turn would result in a drop in the number of females producing eggs. This may be of particular importance given that Redhorse populations have been found to be most sensitive to changes in survival of immature individuals (Young and Koops 2014). The generation time estimated from the life table model (G = 5.99 years) for the stationary/increasing southern Appalachian population was considerably

shorter than that estimated for the threatened Canadian population (G = 8.2 years) (Young and Koops 2014). This difference could be due to latitudinal climate differences, and may be at least partially responsible for the Canadian population's threatened status.

Cataraft electrofishing provided valuable information for Redhorse mostly aged age 2 or higher. The lack of information for age 0 and age 1 fishes may inhibit accurate population modeling. This was an issue for Young and Koops (2014) when estimating survival of aged 0 individuals. Their original model produced a finite rate of increase $\lambda = 2.2$, which was deemed higher than the likely maximum possible growth rate for the species of its size of $\lambda = 1.6$. Due to the high sensitivity of the Black Redhorse population to changes in the juvenile stage class (Young and Koops 2014), habitat use by juveniles should be of particular interest for Redhorse species within the southern Appalachians.

Sicklefin Redhorse are found only in streams of the Little Tennessee or Hiwassee drainages that have long reaches of un-impounded river, though both systems have large downstream reservoirs like Fontana and Hiwassee Reservoir (U. S. Fish and Wildlife Service 2016). Black and Golden Redhorse persist in in the upper Oconaluftee River, which has a small run-of-the-river reservoir, but Sicklefin Redhorse appears to have been extirpated. This could be due to differences in movement patterns among these species, where Sicklefin Redhorse requires longer unimpeded movements than do Black and Golden Redhorse. This hypothesis is somewhat corroborated by radio telemetry of Sicklefin Redhorse juveniles tracked passing through Oconaluftee Reservoir to the larger Tuckasegee River system (Davis 2015, Stowe 2014).

Larval Sampling

Larval catostomids were captured at all sampling locations indicating that catostomids spawn throughout the area sampled in the upper Oconaluftee system, including areas

considerably upstream of the upstream-most area of adult sampled via electrofishing. The highest drift density was found at the middle sampling site located just above the confluence of the upper Oconaluftee River and Soco Creek and just upstream of the upstream-most adult sample reach. Drift nets were the most efficient method for capturing larvae. Light traps were inefficient at capturing catostomid larvae at the sampling sites. Larger numbers of other families, other than catostomids, were captured in light trap samples. This may be due to the limited amount of still water at each site.

The two peaks in yolk sac larvae matched the timing found by Ivasauskas (2017) in the Valley River, and over 70% of catostomid larvae in each peak were identified to the genus *Moxostoma*. The use of DNA barcoding as suggested by Ivasauskas (2017) would allow for further identification of each specimen to the species level. However, unless species-specific identification is necessary, this may not be an economically feasible method of identification.

Conclusion and Suggestions for Future Research

Black and Golden Redhorse populations appear to be relatively healthy in the upper Oconaluftee River, but I did not find any Sicklefin Redhorse. The impact of habitat fragmentation resulting from Bryson Dam may prevent the upper Oconaluftee from supporting viable habitat, as has been found for other Redhorse species in other locations (Reid 2006a, Reid et al. 2007). Inability to move upstream after passing through Bryson Dam prevents any return migration. High gradient and small upstream drainage areas may also prevent some larger Redhorse species persisting in the upper Oconaluftee River (Reid 2006a).

Black Redhorse in the upper Oconaluftee River are more abundant than Golden Redhorse. A wide age range of Black Redhorse (ages 1-12) and Golden Redhorse (ages 1-10) were caught through cataraft electrofishing. For both Black and Golden Redhorse, total length

was represented by a bimodal distribution split around 320 mm with a reduction of age 4+ fish. Life-table modeling of Black Redhorse indicates that the population abundance is staying the same or slightly growing. Smaller population size and an apparently missing year-class may indicate that Golden Redhorse are doing less well. Identifying key factors that may have led to year-class failure and overall smaller population size in Golden Redhorse in this system than Black Redhorse may allow for better understanding of why similar species such as the Sicklefin Redhorse do not persist.

Redhorse were efficiently captured at low discharge levels using raft electrofishing.

Future sampling using this method should target low discharge periods for overall abundance estimates through mark-recapture analysis. Electrofishing should continue to be used to examine movement and survival of tagged individuals. Capture of drifting Redhorse larvae at sites higher in the system than those electrofished indicates that sampling for adult and juvenile Redhorse further upstream may produce a better understanding of the movements of Redhorse within the system.

Sampling higher in the system during spawning may allow for a better understanding of upstream movement patterns. Given that Redhorse are potadromous, population size may vary dramatically in certain areas of the river depending on time of year. Increasing sampling effort on smaller reaches over the length of the upper Oconaluftee River may also provide a better understanding of seasonal population fluctuations among river reaches. Deploying a PIT tag array across the river, to passively monitor movement of fish up and downstream could perhaps also collect this information. Increased effort should be made to capture Redhorse during spawning period to better estimate the number of spawning fish within the system, while other

efforts for estimating abundance of resident Redhorse within the systems should be focused on late summer when discharge is typically lower.

Future increased effort on a subset of sampled areas may allow for higher recapture rates in that area allowing for a more precise estimation of population size. Continued population surveys, studies of survival probability from age 0 to age 1, and population specific estimates of fecundity for southern Appalachian Black Redhorse populations would provide needed data to construct more complex models and use Population Viability Analysis to look at the stability of the population under the influence of changes in habitat or stochastic events that could cause population shifts.

Other possible sampling techniques for sampling Redhorse include the use of a fyke net in the tributaries and headwaters of the Oconaluftee system. Fyke nets could be used in smaller streams like Soco Creek or the Ravens Fork and Oconaluftee prior to their confluence. This technique may not be possible if high otter populations are present in those areas. Weirs (Favrot and Kwak 2016) may be another method for capturing migrating Redhorse that could be protected from mammal predators. Environmental DNA (eDNA) may be useful for detecting a rare fish such as the Sicklefin Redhorse; confounding factors may prevent eDNA from being used as a confirmation for presence of Sicklefin Redhorse within the upper Oconaluftee River. Stocking of fingerling, juvenile, and adult Sicklefin Redhorse in the past has rendered the use of eDNA negligible until recently. The upper Oconaluftee River is located within an area of high fishing pressure due to the abundance of trout streams in western North Carolina. Transport of Sicklefin Redhorse DNA from the Tuckasegee River just downstream of Bryson Dam from anthropogenic vectors or other vectors such as waterfowl could result in false positive reads presence of Sicklefin Redhorse.

The larval sampling time frame should be adjusted to begin early to mid-March and end in late July. Sampling during this time frame would sample peaks in larval drift of Northern Hogsucker prior to Redhorse drift (Ivasauskas 2017). Sampling higher in the Oconaluftee system may allow for determination of the upstream limits for the Redhorse spawning run. This could then provide researchers with an upper threshold for where to sample for adults and juveniles. If there is an indication that Sicklefin Redhorse persist within the upper Oconaluftee River, DNA barcoding would be a useful tool to identify larval fish to species and determine if Sicklefin Redhorse were reproducing.

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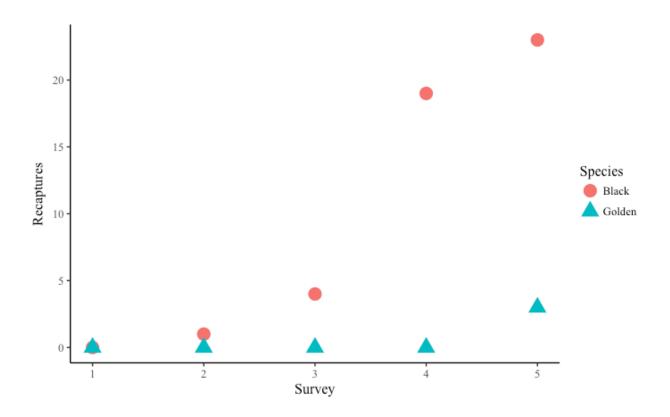
APPENDIX A: SAMPLING SITE LOCATIONS ON UPPER OCONALUFTEE RIVER IN 2017

Method	Site	Start Latitude	Start Longitude	End Latitude	End Longitude
Larval Drift Net / Light Trap	Upstream Larval Site	35°30'5.46"N	83°17'53.29"W	NA	NA
	Middle Larval Site	35°28'7.36"N	83°19'27.97"W	NA	NA
	Downstream Larval Site	35°27'28.29"N	83°21'53.42"W	NA	NA
Electrofishing	Stretch 1	35°28'7.36"N	83°19'27.97"W	35°28'10.60"N	83°19'51.09"W
<u> </u>	Stretch 2	35°28'10.60"N	83°19'51.09"W	35°28'27.78"N	83°20'15.12"W
	Stretch 3	35°28'23.48"N	83°20'38.60"W	35°28'7.53"N	83°21'1.67"W
	Stretch 4	35°28'7.53"N	83°21'1.67"W	35°27'31.56"N	83°21'27.35"W
	Stretch 5	35°27'31.56"N	83°21'27.35"W	35°27'18.36"N	83°22'8.14"W

APPENDIX B: ELECTROFISHING SUMMARIES FOR EACH SURVEY ON THE UPPER OCONALUFTEE RIVER 2017

Species	Survey Event	Number Capture d	Number Mortality	Number Tagged	Number Recapture s
Black Redhorse	4/18 - 4/27	70	6	40	0
	5/31 - 6/2	56	7	30	1
	6/28 - 7/5	193	15	111	4
	8/1 - 8/17	170	3	128	19
	9/18 - 9/25	187	8	151	23
	Total	676	39	460	47
Golden Redhorse	4/18 - 4/27	8	0	8	0
	5/31 - 6/2	9	0	7	0
	6/28 - 7/5	41	3	19	0
	8/1 - 8/17	49	2	32	0
	9/18 - 9/25	34	0	27	3
	Total	141	5	93	3

APPENDIX C: NUMBER OF RECAPTURES FOR BLACK AND GOLDEN REDHORSE FOR EACH SURVEY EVENT.



APPENDIX D: DRIFT NET SAMPLE SUMMARIES

Table D1: Upper larval site most midstream net catostomid captures and drift density during the larval sampling period from April through August 2017.

n sumpring pe		Yolk Sac	Post-Yolk Sac	Yolk Sac Drift Density	Post-Yolk Sac Drift Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	2	2	0	0.0234	0.0000
30-Apr	0	0	0	0.0000	0.0000
8-May	0	0	0	0.0000	0.0000
16-May	1	1	0	0.0264	0.0000
22-May	0	0	0	0.0000	0.0000
29-May	2	2	0	0.0257	0.0000
4-Jun	0	0	0	0.0000	0.0000
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	3	0	3	0.0000	0.0352
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

Table D2: Upper larval site, near shore net, catostomid captures and drift density during the larval sampling period from April through August 2017.

				Yolk Sac	Post-Yolk
			Post-Yolk	Drift	Sac Drift
		Yolk Sac	Sac	Density	Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	0	0	0	0.0000	0.0000
30-Apr	1	1	0	0.0173	0.0000
8-May	0	0	0	0.0000	0.0000
16-May	0	0	0	0.0000	0.0000
22-May	3	3	0	0.0581	0.0000
29-May	0	0	0	0.0000	0.0000
4-Jun	3	3	0	0.0528	0.0000
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	3	0	3	0.0000	0.0343
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

Table D3: Middle larval site most midstream net catostomid captures and drift density during the larval sampling period from April through August 2017.

				Yolk Sac	Post-Yolk
			Post-Yolk	Drift	Sac Drift
		Yolk Sac	Sac	Density	Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	3	3	0	0.0268	0.0000
30-Apr	5	5	0	0.1826	0.0000
8-May	0	0	0	0.0000	0.0000
16-May	1	1	0	0.0882	0.0000
22-May	3	3	0	0.0712	0.0000
29-May	0	0	0	0.0000	0.0000
4-Jun	2	1	1	0.0130	0.0130
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	0	0	0	0.0000	0.0000
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

Table D4: Middle larval site, near shore net, catostomid captures and drift density during the larval sampling period from April through August 2017.

				Yolk Sac	Post-Yolk
			Post-Yolk	Drift	Sac Drift
		Yolk Sac	Sac	Density	Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	1	1	0	0.0117	0.0000
30-Apr	14	14	0	0.2652	0.0000
8-May	0	0	0	0.0000	0.0000
16-May	1	1	0	0.0441	0.0000
22-May	1	1	0	0.0352	0.0000
29-May	2	2	0	0.0198	0.0000
4-Jun	2	2	0	0.0322	0.0000
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	0	0	0	0.0000	0.0000
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

Table D5: Middle larval site most midstream net catostomid captures and drift density during the larval sampling period from April through August 2017.

				Yolk Sac	Post-Yolk
			Post-Yolk	Drift	Sac Drift
		Yolk Sac	Sac	Density	Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	3	3	0	0.0221	0.0000
30-Apr	0	0	0	0.0000	0.0000
8-May	0	0	0	0.0000	0.0000
16-May	0	0	0	0.0000	0.0000
22-May	1	0	0	0.0000	0.0000
29-May	1	0	1	0.0000	0.0083
4-Jun	16	0	16	0.0000	0.1310
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	0	0	0	0.0000	0.0000
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

Table D6: Middle larval site, near shore net, catostomid captures and drift density during the larval sampling period from April through August 2017.

				Yolk Sac	Post-Yolk
			Post-Yolk	Drift	Sac Drift
		Yolk Sac	Sac	Density	Density
Date	Catostomids	Larvae	Larvae	(n/m^3)	(n/m^3)
9-Apr	0	0	0	0.0000	0.0000
16-Apr	0	0	0	0.0000	0.0000
26-Apr	1	1	0	0.0076	0.0000
30-Apr	1	1	0	0.0091	0.0000
8-May	1	1	0	0.0075	0.0000
16-May	0	0	0	0.0000	0.0000
22-May	0	0	0	0.0000	0.0000
29-May	4	4	0	0.0346	0.0000
4-Jun	9	0	9	0.0000	0.1276
21-Jun	0	0	0	0.0000	0.0000
28-Jun	0	0	0	0.0000	0.0000
5-Jul	0	0	0	0.0000	0.0000
11-Jul	0	0	0	0.0000	0.0000
18-Jul	0	0	0	0.0000	0.0000
24-Jul	0	0	0	0.0000	0.0000
2-Aug	0	0	0	0.0000	0.0000
8-Aug	0	0	0	0.0000	0.0000
16-Aug	0	0	0	0.0000	0.0000
23-Aug	0	0	0	0.0000	0.0000

APPENDIX E: LIGHT TRAP SAMPLING SUMMARY

Table E1: Total captures by each light trap throughout the sampling period. *Test-Middle was only deployed into the water during sampling from May 16 to August 30

Light Trap	Total All Larvae	Total Catostomid Larvae
Upper	72	1
Test-Middle*	20	0
Initial-Middle	28	0
Lower	35	5

Table E2: Captures for the light trap set at the upper larval collection site.

	• •	Catostomid
Date	All Larvae	Larvae
9-Apr	0	0
16-Apr	0	0
26-Apr	0	0
30-Apr	0	0
8-May	0	0
16-May	0	0
22-May	0	0
29-May	0	0
4-Jun	3	0
13-Jun	0	0
21-Jun	1	1
28-Jun	0	0
5-Jul	59	0
11-Jul	0	0
18-Jul	6	0
24-Jul	0	0
2-Aug	0	0
8-Aug	3	0
16-Aug	0	0
23-Aug	0	0
30-Aug	0	0

Table E3: Captures for the test light trap set at the middle larval collection site, this trap was added part of the way through the sampling period in response to loss of optimum sampling conditions at other <u>middle light trap</u>.

		Catostomid
Date	All Larvae	Larvae
16-May	0	0
22-May	0	0
29-May	0	0
4-Jun	10	0
13-Jun	1	0
21-Jun	1	0
28-Jun	7	0
5-Jul	0	0
11-Jul	1	0
18-Jul	0	0
24-Jul	0	0
2-Aug	0	0
8-Aug	0	0
16-Aug	0	0
23-Aug	0	0
30-Aug	0	0

Table E4: Captures for the initial light trap set at the middle larval collection site.

		Catostomid
Date	All Larvae	Larvae
9-Apr	0	0
16-Apr	0	0
26-Apr	0	0
30-Apr	0	0
8-May	0	0
16-May	0	0
22-May	0	0
29-May	0	0
4-Jun	0	0
13-Jun	3	0
21-Jun	1	0
28-Jun	11	0
5-Jul	13	0
11-Jul	0	0
18-Jul	0	0
24-Jul	0	0
2-Aug	0	0
8-Aug	0	0
16-Aug	0	0
23-Aug	0	0
30-Aug	0	0

Table E5: Captures for the light trap set at the lower larval collection site.

		Catostomid
Date	All Larvae	Larvae
9-Apr	0	0
16-Apr	0	0
26-Apr	0	0
30-Apr	0	0
8-May	0	0
16-May	0	0
22-May	21	1
29-May	0	0
4-Jun	7	4
13-Jun	1	0
21-Jun	0	0
28-Jun	0	0
5-Jul	4	0
11-Jul	1	0
18-Jul	0	0
24-Jul	0	0
2-Aug	0	0
8-Aug	1	0
16-Aug	0	0
23-Aug	0	0
30-Aug	0	0