



Oral Presentations:

1. USFWS STATUS OF AT-RISK AQUATIC SPECIES

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On September 1, 2016, the US Fish & Wildlife Service announced a seven-year work plan for ESA listing and critical habitat decisions. This plan is intended to improve effectiveness and implementation of the ESA and provide for best possible conservation of our nation's imperiled wildlife, known as at-risk species. In the southeast, three Service programs are cooperating in the conservation of at-risk species: 1) Ecological Services is conducting status assessments and making recommendations for listing species, 2) Fish and Aquatic Conservation (FAC) assess populations, and utilize unique skills from the Fish Technology and Health centers, and 3) National Wildlife Refuges assist with acquiring data on at-risk species as well as managing those species that occur on NWR lands.

FAC, formerly known as the Fisheries Program, is composed of 2 subunits: National Fish Hatcheries (NFHs) and Fish & Wildlife Conservation Offices (FWCOs). The NFHs maintain ark populations, develop propagation techniques and life history requirements, and work with the FWCOs and the Fish Technology Center to assist with the development of cryopreservation techniques and genetic management plans. FWCOs conduct habitat and population assessments to guide federal trust species management. Additionally, the Southeast Region FAC, through its region wide fish habitat and fish passage programs, has developed a Regional Aquatic Habitat Team to implement conservation delivery associated aquatic habitat fragmentation and overall aquatic habitat condition. Active involvement in several key components of Strategic Habitat Conservation position the FAC program to improve information gathering for at-risk species and quickly deliver conservation actions such as habitat improvement and species propagation, if appropriate. Examples of successful projects will be presented.

Keywords: At Risk Species; USFWS; fish passage; fish habitat; ESA
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2. YELLOWFIN MADTOM REINTRODUCTION INTO THE NORTH FORK HOLSTON RIVER

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Species reintroduction is a prominent task in many recovery plans. Here we highlight the six year effort to reintroduce the Yellowfin Madtom (*Noturus flavipinnis*) into the North Fork Holston River (NFHR) of southwestern Virginia. Because the river was known historically for the species, the US Fish and Wildlife Service (USFWS) designated it as Non-essential/Experimental for Yellowfin Madtom in 1988. However, no effort was made to reintroduce the species until the formation of a Yellowfin Madtom Conservation Committee in 2010. The purpose of the committee was to provide guidance on a reintroduction strategy. Before any releases could be considered, the committee determined the need for habitat, threat, species presence, and genetics assessments. Because reintroduction involved the propagation of an endangered species, the USFWS required the development of a controlled propagation plan. Administratively, an intensive effort was made to inform federal, state, and local government officials and agencies of our intent. Finally, we were able to incorporate an education/outreach component by involving local school children to assist in the release.

Keywords:madtom; reintroduction, assessment, conservation
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3. COMPARING SCIENTIFIC UNCERTAINTIES AMONG SEVEN AT-RISK DARTERS

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Of the 374 aquatic-dependent southeastern species that the U.S. Fish and Wildlife Service deemed potentially warranting federal protection in 2011, 43 of these are freshwater fishes. Even though seven families of fishes are represented, one-half of the 43 species (22) are darters. An comparison of scientific knowledge of seven at-risk darters (*Etheostoma bellator*, *E. cinereum*, *E. maydeni*, *E. forbesi*, *E. trisella*, *E. tuscumbia*, and *Percina brevicauda*) endemic to the Cumberland, Tennessee, and Mobile Basin drainages indicates that there are varying levels of knowledge on the taxonomy, distribution, abundance, genetics, ecology, life history, and threats to these species. While a paucity of these data can make listing decisions difficult, many species warrant listing even if some data are lacking.

Keywords:Percidae; *Etheostoma*; *Percina*; conservation; USFWS
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4. CONSERVATION GENETICS OF THE UNDESCRIBED BLUEFACE DARTER (*ETHEOSTOMA* SP. CF. *ZONISTIUM*), A RARE FISH IN NORTHWEST ALABAMA

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Previous morphological work revealed that select populations of the Bandfin Darter (*Etheostoma zonistium*) in northwest Alabama represent a rare and undescribed species; the Blueface Darter (*E. sp. cf. zonistium*). With the description of the Blueface Darter forthcoming, its rarity, restricted distribution, and fragmented landscape will make it a potential candidate for listing under the Endangered Species Act following its description. The Blueface Darter has a peculiar distribution in tributaries of two distinct drainage basins; the Tennessee River (Bear and Little Bear creeks) and the Black Warrior River (Hubbard Creek of the Sipsey Fork). Further, population connectivity among tributaries of the upper Bear Creek system is potentially restricted by the Upper Bear Creek Reservoir. This study used mitochondrial (mt) DNA sequence data and microsatellite loci to evaluate genetic distinctiveness of the Blueface Darter compared to the Bandfin Darter and to test multiple hypotheses involving patterns of genetic variation within the Blueface Darter. Our results suggest three main conclusions that will help prioritize conservation needs for the Blueface Darter. (1) Both mtDNA and microsatellite data corroborate previous morphological analyses supporting the validity of the Blueface Darter as a distinct species; (2) mtDNA data suggests that the Hubbard Creek population is the result of a very recent inter-drainage transfer from Bear Creek. Hubbard Creek also exhibits a signature of founder effect based on microsatellite data (i.e., significant genetic structure and lower genetic variation compared to the Bear Creek population). (3) Our data shows possible reservoir-fragmentation effects for the Little Bear Creek population (i.e., significant genetic structure and lower genetic variation compared to the Bear Creek population). Based on these results, we define appropriate conservation units and priorities for the Blueface Darter, which will be useful to state and federal agencies following the species' formal description.

Keywords: microsatellite DNA, mtDNA, population structure, genetic diversity
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5. IS IT TIME TO UTILIZE ANALYSIS OF WHOLE GENOME SEQUENCES AS A TOOL FOR MAKING CONSERVATION DECISIONS?

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Genetic population structures are generally deduced from analysis of mitochondrial DNA sequences and/or nuclear intron sequences and/or microsatellite sequences. Based on the geographic distribution of variation of these sequences, the populations may then be designated as either management units (MU) or evolutionarily significant units (ESU). While most of these designations are based on neutral variation, the underlying assumption is that such divergent populations are likely harboring different adaptive variants which should be conserved. However, it is not possible to know what degree of neutral variation partitioned among populations correlates with partitioning of any adaptive variation. Whole genome sequence

analysis of populations should be able to identify the presence of adaptive variation among populations, if any, and thus decisions about conservation of specific populations could be based on this criterion directly. The Tallapoosa darter (*Etheostoma tallapoosae*) has been shown to be divided into at least two genetically distinct populations (MUs) by segregation of unique mitochondrial and nuclear gene sequences. This variation is mainly neutral variation. In an effort to eventually assess if these populations harbor unique adaptive gene variants, the genome of the Tallapoosa darter has been sequenced and partially assembled and preliminary annotation of some genes has been carried out. In order to carry out a genomic population analysis of this species, the genome will first have to be fully assembled and annotated and genomic sequencing cost will need to decrease below current levels. In the meantime, the genomes of *Percina crypta*, *Etheostoma chuckwachatte* and *Etheostoma jessiae* have also been sequenced and orthologs of the annotated *E. tallapoosae* genes are being assessed for the presence of adaptive variation as a first step to determining the degree to which adaptive gene variants are partitioned among recognized species prior to the analysis of adaptive variation within a species.

Keywords: darter; genomic sequencing; adaptive variation; conservation
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6.

7. CONSERVATION STATUS, HABITAT USE, AND PHYLOGEOGRAPHY OF THE IMPERILED TENNESSEE-ENDEMIC EGG-MIMIC DARTER (*ETHEOSTOMA PSEUDOVULATUM*)

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Restricted to six tributaries of the Duck River system of Tennessee, *Etheostoma pseudovulatum* is state endangered and has been petitioned for federal listing. Additionally, the mainstem Duck River may act as a barrier isolating each tributary population. A status survey has not been conducted since 1995 and genetic diversity has never been evaluated. Thus, objectives were to: 1) examine the distribution, abundance, population size, and overall genetic diversity; 2) describe general habitat use; and 3) assess phylogeographic patterns of genetic diversity to evaluate whether the Duck River acts as a barrier to gene flow among tributary populations. Twenty-five localities representing all historical localities of *E. pseudovulatum* were sampled in spring and fall of 2014 using standard seining techniques to assess presence and estimate population size using the Petersen mark-recapture method. Habitat variables were measured and analyzed for association with *E. pseudovulatum* presence. Range-wide genetic diversity was examined using the mitochondrial ND2 gene. *Etheostoma pseudovulatum* was present at all 25 localities sampled and population-size estimates ranged from 5 to 258 individuals per 75-meter reach, comparable to those observed historically. The species was significantly associated with low flow, a range of greater depths, and presence of undercut banks, organic debris, and root wads. Eleven haplotypes were detected with one haplotype shared across all tributaries except Beaverdam Creek, which had a unique haplotype assemblage compared to all tributaries except Little Piney Creek. Haplotypes were recovered in two clades: 1) Beaverdam Creek and two individuals from Little Piney Creek; and 2) the other five tributary populations.

These results suggest historical gene flow among all tributaries except Beaverdam Creek, which has potentially been isolated from other tributaries. Overall, the species was locally abundant and appears stable. However continued future monitoring is recommended, with focus on smaller tributaries and the genetically distinct Beaverdam Creek population.

Keywords:conservation status; phylogeography; habitat use; darter
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8. RELATIVE ABUNDANCE AND HABITAT USE BY SICKLE DARTERS (*PERCINA WILLIAMSI*) IN TENNESSEE

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During summer 2016, we surveyed historic and new sites in the Emory and Little Rivers for the ESA-petitioned Sickle Darter (*Percina williamsi*). Using snorkeling, backpack electroshocking, and seining techniques, relative abundance was measured as catch per unit effort and density. Snorkeling was determined to be the most effective sampling technique, and relative abundance was greater for the Emory River population (7.2 fish/snorkeler-hr; 7.8 fish/m²) for Little River (2.5 fish/snorkeler-hr; 4.9 fish/m²). In Little Rock Creek (tributary to Emory), the dominant two substrates used by individuals were sand then boulder, whereas in Emory River and Little River it was gravel followed by sand. Sickle Darters were observed in low-gradient areas of streams, almost entirely in pool habitats (mean depth was 41-51 cm) with mean current velocities measuring 0.5-0.6 m/s (timed neutrally bouyant object method). Canopy cover of riparian vegetation measured in the stream by a densiometer averaged 82% at Emory River darter locations, 35% at Little River, and 90% at Little Rock Creek. Water quality was considered good at all sites, with total dissolved solids (TDS) less than 63 ppm at all sites. Channel margins containing beds of Water Willow (*Justicia americana*), Illinois Pondweed (*Potamogeton illinoensis*), American Pondweed (*P. epihydrus*), and small woody debris (SWD) were heavily used by Sickle Darters, and in some instances fish were seen selectively foraging on dipteran larvae attached to SWD.

Keywords:conservation; percidae; habitat; snorkeling
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9. MICROHABITAT SELECTION AND PARTITIONING AMONG DARTERS IN THE NOLICHUCKY RIVER, TENNESSEE

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The Nolichucky River is a 185 km long tributary of the French Broad River that arises from the Blue Ridge Mountains in western North Carolina and flows into eastern Tennessee. This river is home to a diverse assemblage of darters, including species of conservation concern. Because

darther species can be habitat specialists and exhibit high levels of niche segregation, we undertook snorkel surveys of 4 riffle-run sites on the mainstem of the Nolichucky River to investigate the degree of microhabitat partitioning by syntopic darters. We conducted the surveys during August-September 2015 at low flows. We encountered 244 darters, of which five species occurred in abundances great enough for analyses (Banded Darter *Etheostoma zonale*, Gilt Darter *Percina evides*, Greenside Darter *Etheostoma blenniodes*, Sharphead Darter *Nothonotus acuticeps*, and Olive Darter *Percina squamata*). We measured depth, velocity, and recorded substrate occurrences in a 0.25x0.25 m quadrat centered on each individual's location. We used non-metric multidimensional scaling to graphically investigate the general patterns of microhabitat use among the five species of darters analyzed. We then used multinomial logistic regression to identify individual habitat variables that significantly differentiated the microhabitat of each species, using the cosmopolitan Banded Darter as the reference case. The NMDS indicated that the most abundant and wide-ranging species were more generalistic, utilizing a diverse range of microhabitat variable gradients, while the two species of conservation concern appeared to be more specialized in their habitat use. Specifically, the logistic regression indicated that the Sharphead Darter utilized significantly more shallow, faster velocity habitat while the Olive Darter used primarily deeper habitat with less bedrock than the reference. In light of recent attempts by industrial stakeholders to withdraw water and expel treated water back to the river, our results should be considered in any future conservation actions undertaken in the Nolichucky River watershed.

Keywords: Sharphead Darter; Olive Darter; habitat partitioning; non-metric multidimensional scaling; multinomial logistic regression
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10. INTRODUCTION OF SMALL WOOD IN A DEGRADED STREAM RESULTS IN GREATER FISH SPECIES AND FUNCTIONAL DIVERSITY

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The introduction of wood to streams for the benefit of fish communities has usually meant the installation of large wood (LW) in complex installations. Such installations are prohibitively costly and beneficial results are often lacking, especially in lowland Coastal Plain streams. However, our experience in the field and previous work has indicated the importance of small wood (SW) as habitat for stream fishes. The installation of SW has promise as an economical alternative to the installation of LW though the effects of SW installation on fish communities are largely unknown in degraded lowland streams. We tested the response of a fish community in a highly degraded stream to the introduction of SW over a three year study. We estimated species richness, evenness, and dominance as well as functional richness, evenness, and divergence. We then used permutation methods to test for differences in these estimates among Control reaches (unmanipulated reaches mostly devoid of wood), reaches with patchy wood installations (Patchy), and reaches with dense wood installations (Dense). Test results show no differences between Patchy and Dense treatments. However, test results indicate that woody reaches have higher species richness and evenness and lower dominance. Functional evenness

and divergence did not show any changes among treatments but functional richness was higher for the Dense and perhaps the Patchy treatments compared to the Control reaches. Ordinations, models, and rank abundance indicate that darters and catfishes show the most pronounced positive response to the introduction of SW. Our results indicate that the introduction of SW is beneficial to fish communities and ecosystem function. Though our study needs to be replicated, the introduction of SW to degraded lowland streams may prove to be a way forward in restoring stream habitat.

Keywords: stream restoration; Coastal Plain; in-stream wood; fish community; functional diversity

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11. SINGLE-SPECIES AND SPECIES-INTERACTION OCCUPANCY MODELING OF THE YAZOO DARTER AND GOLDSTRIPE DARTER WITHIN THE UPPER YAZOO RIVER BASIN

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Understanding patterns of occurrence are important for ecologists to make inferences about environmental predictors and their roles in the determination of occupancy of a species. Also, patterns of co-occurrence are important for understanding how species with similar ecological traits interact and if these similarities may lead to overdispersion, or the occupancy of different sites due to competition. Occupancy is a metric for evaluating populations, and occupancy models that account for the undetectability of a study's target species are useful tools providing solutions to these problems. In this study, I used site occupancy and two species occupancy models to assess the occupancy and co-occurrence of the Yazoo Darter *Etheostoma raneyi* and Goldstripe Darter *Etheostoma parvipinne* within the Upper Yazoo River system. The modeling approach in this study enabled comparisons of covariates that influenced occupancy at two spatial scales (catchment and site) for all single species occupancy models. Across their established ranges in the Upper Yazoo River basin, the probability of occurrence (i.e., Ψ) of Yazoo darters appears to be 0.38, whereas for goldstripe darters Ψ appears to be 0.49. At the site scale, results suggest that the heterogeneity of stream depth (SD) is the most important habitat component that influences the probability of occurrence of Yazoo Darters while SD and the heterogeneity of substrate appear to be the most important site scale variables influencing the occurrence of goldstripe darters. Furthermore, drainage area (km²) appears to be the important hydrological component that influence the probability of occurrence at the catchment scale. Finally, occupancy models suggest that Yazoo darters are more dependent on groundwater flow for their occurrence in relation to goldstripe darters. Both darters appear to co-occur less frequently than would be expected by chance ($\phi = 0.96$), and the probability that the area is occupied by both species (i.e., Ψ^{AB}) is 0.44.

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12. FISH-HABITAT ASSOCIATIONS IN THE SCENIC KINGS RIVER, ARKANSAS

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The Kings River is located in the upper White River basin in northwest Arkansas and is designated as an Extraordinary Resource Waterbody by Arkansas Game and Fish Commission. It is a free-flowing river and home to 63 species of fish including several species endemic to the Ozark Mountains, *Notropis nubilus*, *Noturus albater*, *Noturus flavater*, *Etheostoma juliae*, and a Species of Greatest Conservation Need, *Notropis ozarcanus*. Despite being recognized as a regionally important aquatic resource; no basin wide studies of fishes have been conducted. We collected a total of 13,928 individuals encompassing 43 species across 21 sites via seines. Total abundance ranged from 187 to 1189 and species richness ranged from 3 to 29 species at a given site. *N. ozarcanus* was found at 9 of 21 sites in low numbers (relative abundance <2% at any given site). Principal component analysis was used to create a composite habitat axis for correlations with fish metrics. We classified 27 of the 43 species collected as intolerant. Percent intolerant ranged from 12% to 96% at a given site and was positively correlated with Habitat Axis 1 ($\rho=0.56$, $p=0.02$) indicating increasing pool depth, riffle velocity and decreased pool length are important. Evenness ($\rho=-0.62$, $p=0.008$) and diversity ($\rho=-0.81$, $p<0.001$) were negatively correlated with Habitat Axis 1. At eight of the sites we also conducted snorkel surveys to examine community structure prior to a planned restoration by The Nature Conservancy. We found variation between the sites designated as degraded compared to reference sites, with a decrease in relative abundance of sensitive cyprinid species using seine data. Snorkel surveys revealed the complexities surrounding fish use of run/pool reaches with species distributions related to instream characteristics including depth, stream width, and large wood structure. Baseline data will be used in future assessments of fish-habitat relationships.

Keywords: fish-habitat; conservation; restoration
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13. SCALE SHAPE VARIATION IN A SPECIOSE AND HIGHLY IMPERILED GROUP OF FISHES, THE DARTERS (PERCIDAE: ETHEOSTOMATINAE)

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Fish scales play an important role in reducing drag and facilitating movement. However, little research has focused on the relative impacts of environment, phylogeny, life history, sex, or behavior on scale phenotype. For example, though the species-rich darter clade (Percidae: Etheostomatinae) has a vast research history and scale counts are commonly used to diagnose species, studies of variation in scale phenotype have been limited to a small number of species. Additionally, none have evaluated scale shape in the context of modern hypotheses of diversity and phylogeny. In a pilot study to determine if scale shape varied among darter species and clades, straight-line measurements, which archived scale shape, and counts of radii and ctenii

were taken from 236 individuals, representing 80 species, all genera, 26 subgenera, and 29 darter clades. Variation in number of ctenii and radii was observed among genera, subgenera and clades (*Percina ctenii*, radii: =19.9, 9.3; *Nothonotus*: =25.2, 14.3; *Etheostoma*: =24.9, 13.3; *Crystallaria*: =11.0, 6.3; *Ammocrypta*: =8.1, 6.9). Principle components analysis revealed *Ammocrypta* and *Crystallaria* have shorter scale lengths and shorter ctenii relative to other genera. Variation among species and clades within each genus, particularly *Percina* and *Etheostoma*, was also noted. Subsequent data were collected to elucidate the effects of sex on scale shape and meristics using geometric morphometrics for a subset of darter taxa. Sexual dimorphism in scale shape was not observed for the species examined. Because scales aid in reducing drag, future work will investigate the impact of environment, behavior, and phylogeny on darter scale shape. Understanding the relative impact these factors have on the darter phenotype will provide insight on forces that contributed to their overall diversification and potential responses of this group of fishes to environmental changes.

Keywords:darters; geometric morphometrics; scales
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14. OCCUPANCY MODELING OF BLACKSIDE DACE (*CHROSOMUS CUMBERLANDENSIS*) AND CUMBERLAND ARROW DARTERS (*ETHEOSTOMA SAGITTA*) USING CONDUCTIVITY AS A COVARIATE

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The upper Cumberland River system has a unique assemblage of fishes with many endemic species that are imperiled, such as Blackside Dace and Cumberland Arrow Darter. Natural resource extraction such as coal mining and logging have been shown to negatively impact fish and other aquatic life, because of water quality degradation. Conductivity is a parameter of water quality that is frequently measured to gauge stream health, and used as a potential physiological stressor to aquatic life. A recent study by Hitt et al (2016) indicated that conductivity had the strongest effect on the occurrence and abundance of Blackside Dace, where a significant threshold response was determined at 343 $\mu\text{S}/\text{cm}$. The goal of our study was to provide an independent test of the threshold response value to determine if the occupancy of Blackside Dace was different among sites with conductivity above and below the respective threshold response conductivity value. Estimated occupancy of Blackside Dace in streams below 343 $\mu\text{S}/\text{cm}$ was 0.19 ± 0.08 , while in streams with higher conductivities estimated occupancy was 0.07 ± 0.07 . We also compared the estimated occupancy of other fishes to Blackside Dace to show relative tolerance to elevated conductivities. Cumberland Arrow Darter had an estimated occupancy of 0.51 ± 0.10 in streams below 343 $\mu\text{S}/\text{cm}$, while in streams with higher conductivities estimated occupancy was 0.14 ± 0.09 . Tolerant species such as the Creek Chub (*Semotilus atromaculatus*) do not exhibit such divergent occupancy values; below 343 $\mu\text{S}/\text{cm}$ estimated occupancy was 0.90 ± 0.05 , while in higher conductivities estimated occupancy was 0.81 ± 0.10 . These results support the threshold response values determined by Hitt et al. (2016). Implications of the two studies provide resource managers with a useful and defensible benchmark for determining

impairment, with Blackside Dace

Keywords: Blackside Dace; Occupancy Modeling; Conductivity; Cumberland Arrow Darter
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15. ECOLOGICAL DIVERSIFICATION ASSOCIATED WITH THE BENTHIC-TO-PELAGIC TRANSITION BY NORTH AMERICAN MINNOWS

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Ecological opportunity is often regarded as a key factor that explains why diversity is unevenly distributed across life. Colonization of novel environments or adaptive zones may promote diversification. North American minnows exhibit an ancestral benthic to pelagic habitat shift that coincided with a burst in diversification. Here, we evaluate the phenotypic and ecological implications of this habitat shift by assessing craniofacial and dietary traits among 34 species and testing for morphology-diet covariation, convergence, and adaptive optima. There were several instances of morphology-diet covariation such as correlations between mouth angle and the consumption of terrestrial insects and between relative gut length and the consumption of algae. After accounting for size and phylogenetic non-independence, benthic species had longer heads, longer snouts, eyes positioned higher on their head, smaller mouth angles, and longer digestive tracts than pelagic minnows. Benthic minnows also consumed more algae but less terrestrial insects, by volume, than pelagic minnows. Lastly, there were three distinct evolutionary regimes and more convergence in morphology and dietary characteristics than expected under a Brownian motion model of evolution. These findings indicate that colonization of the pelagic zone by minnows involved myriad phenotypic and dietary changes associated with exploitation of terrestrial subsidies. Thus, minnows exhibit phenotype-dietary covariation, an expansion of ecological roles, and a burst in diversification rates in response to the ecological opportunity afforded by the colonization of a novel habitat.

Keywords: Adaptive radiation; craniofacial shape; Cyprinidae; diversification; morphology
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16. PHYLOGEOGRAPHY OF THE ROSYSIDE DACE (*CLINOSTOMUS FUNDULOIDES*)

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The genus *Clinostomus* (Teleostei: Cyprinidae) is an understudied genus of fishes in the family Cyprinidae. *Clinostomus* has a large geographic range occupying much of eastern North America. Currently, only two species are recognized within *Clinostomus*: *C. funduloides* and *C. elongatus*. A previous morphological study identified multiple subspecies within *C.*

funduloides and later, one additional undescribed taxon was proposed (Smoky Dace). Given the broad distribution and disjunct geographic range of this genus, a phylogenetic assessment of *Clinostomus* is needed to further resolve the taxonomic diversity within the genus. In particular, we focused on assessing diversity within the Rosyside Dace (*C. funduloides*). Multiple tissue samples of *C. funduloides* were obtained across the range. Mitochondrial and nuclear DNA sequence data was obtained, and data was analyzed using Bayesian phylogenetic methods to investigate the diversity and phylogenetic relationships within the species. Results of this study will give insight on the phylogenetic relationships, genetic differentiation, and taxonomic status of the populations within *C. funduloides*.

Keywords: Cyprinidae; Phylogenetics

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17. MOLECULAR RELATIONSHIPS WITHIN *TYPHLICHTHYS SUBTERRANEUS* (PERCOPSIFORMES: AMBLYOPSIDAE) AND CONSERVATION IMPLICATIONS

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Cryptic morphology coupled with collection difficulties and recent divergence of genetic lineages can inhibit the designation of species in stygobiont taxa. By designating Evolutionarily Significant Units (ESUs), the conservation community can create management plans to conserve the genetic diversity within a group. The lineages of *Typhlichthys subterraneus* were designated as ESUs and given conservation ranks; however, newly sampled populations had not been previously investigated as to their association with existing lineages. Genomic DNA was extracted from fin clips and three genes were amplified (ND2, S7, Rhod). Sequences were aligned and edited manually in Geneious, and Maximum Likelihood phylogenies were generated for each gene as well as a concatenated dataset. Newly sampled populations were recovered within existing lineages but unique topologies of two previously designated caves were found. Conservation ranks utilizing both the NatureServe and IUCN Red List criteria were recalculated for each lineage. One lineage was downgraded to Vulnerable due to population sizes and another newly designated lineage was given a rank of Critically Imperiled (NatureServe) and Critically Endangered (IUCN Red List). Nonmonophyletic relationships within the phylogeny may be a result of gene flow between aquifers or low genetic resolution due to incomplete lineage sorting.

Keywords: Cavefish; conservation status; phylogeny; subterranean biology

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18. CONSERVATION PLANNING FOR SOUTHEASTERN AQUATIC BIODIVERSITY

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The southeastern United States is a global hotspot of freshwater biodiversity, supporting almost two-thirds of the country's fish species, over 90% of the U.S. total species of mussels, and nearly half of the global total for crayfish species. Many of these species are narrow endemics. Unfortunately, this region is also a hotspot for imperilment; the number of imperiled freshwater fish species in the Southeast has risen 125% in the past 20 years. While the causes of this imperilment, including habitat modification or loss, development, and introduced species, have been extensively documented, efforts to reverse these trends have been hampered by limited funding and lack of public awareness. This project, funded by the National Fish and Wildlife Foundation, used over 500,000 point records to compile range maps for 592 species of freshwater fishes. We used these ranges, combined with endemism and imperilment scores, to derive a set of priority areas that form the basis of an integrated conservation strategy at the watershed scale (8-digit HUC). This project's outcomes include a Southeastern Aquatic Biodiversity Conservation Strategy, a synthesis that identifies management actions, and policy recommendations appropriate to the threats and conservation capacity for 10 priority areas. Nine of the top ten Huc-8 sub-basins were in the Tennessee River or Alabama River drainages, but based on species complementarity the final set was adjusted to include one sub-basin each from the Green and Cumberland systems.

Keywords: Southeast, fish, mussels, crayfish, conservation
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19. IDENTIFYING PHYLOGEOGRAPHIC BREAKS IN DARTERS ACROSS THE GULF COASTAL PLAIN OF THE SOUTHEASTERN UNITED STATES

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Recent phylogeographic studies of darters distributed throughout the Gulf Coastal Plain in the southeastern United States have revealed unique patterns of diversification. However, concordant phylogeographic breaks among darters inhabiting this region have not yet been well identified. One obvious and well-tested phylogeographic break among coastal plain species is the division between populations east and west of the Mississippi River. This study used mitochondrial and nuclear DNA sequence data from several Gulf Coastal Plain darter species including *Etheostoma chlorosoma* (Bluntnose Darter), *E. gracile* (Slough Darter), and *E. parvipinne* (Goldstripe Darter) to evaluate concordance of the east-west Mississippi phylogeographic break, and to gain a better understanding of factors that shape genetic structure in darters across the Gulf Coastal Plain. Preliminary analyses revealed that only one of the three species (*E. parvipinne*) exhibits a sharp phylogeographic break east and west of the Mississippi River. For *E. gracile*, specimens from the Colorado River exhibit pre-Pleistocene (approx. 4 mya) divergence from all other populations, and more recent (approx. 0.5 mya) divergence between two well-supported groups subdivided by the Mississippi embayment in Arkansas. Phylogeographic breaks were more complex for *E. chlorosoma*, with relatively deep divergence between a western clade (Ouachita, Red, Neches, and Trinity rivers) and an "eastern" clade (Hatchie, Tennessee, Wabash, and Sabine rivers). The recovery of individuals from the Sabine River in the "eastern" clade reflects

the need for additional sampling of *E. chlorosoma* to understand the nature of this phylogeographic pattern. Ongoing work with additional nuclear DNA markers and increased sampling will be incorporated into this framework, shedding light on these unique phylogeographic patterns in the Gulf Coastal Plain.

Keywords: mtDNA, nuclear DNA, Mississippi River Discontinuity
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20. PERSISTENCE AND STABILITY OF PEARL DARTER POPULATIONS

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One of the original Desperate Dozen, the Pearl darter (*Percina aurora*) is historically known from only the Pearl and Pascagoula River systems of Mississippi and Louisiana. However, it appears that populations remain only in the Pascagoula River drainage, likely extirpated from the Pearl River drainage (last specimen taken in 1973) after installation of flow control structures. Proposal for listing was initiated in 1999 with a recent recommendation for Threatened status. Throughout 2013-2016 we surveyed sites within the Pascagoula River and its major tributaries (Chickasawhay and Leaf Rivers) to assess long-term patterns of population persistence in reference to surveys conducted over a decade ago. While yearly variation in abundance (CPUE) was observed, frequency of occurrence remained stable throughout the surveyed years indicating persistence and stability in the Pascagoula populations. Among the sampled rivers, CPUE and occurrence rates were highest in the Chickasawhay and Pascagoula Rivers. Furthermore, Pearl darter presence during coarse spatial sampling in other tributaries within this drainage (Okatoma and Black Creeks, Bouie River) have indicated available habitat remains and populations still persist in these relatively under-sampled major tributaries.

Keywords: population stability; pearl darter; Pascagoula River; conservation; occupancy
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21. PRELIMINARY ANALYSIS OF FISH ASSEMBLAGE COMPOSITION IN THE UPPER SALINE RIVER, ARKANSAS OVER MULTIPLE DECADES

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The mainstem of the Saline River is one of the longest unregulated rivers in Arkansas, flowing 325 km to the Ouachita River. This region supports high fish species richness (n=109), and 13 taxa are designated as Species of Greatest Conservation Need. Rapid development within the watershed began in the 1970's, and anthropogenic alteration is known to have negative effects on stream fish assemblages. Understanding persistence of stream fish assemblages and shifts in composition following landscape alterations is of interest to fish ecologists to guide conservation

efforts. We used rarefaction and a combination of multivariate analyses (non-metric multidimensional scaling, permutational analysis of variation, and similarity percentage analysis) to examine fish assemblage structure between period I (1981-1982) and period II (2016) at 10 sites. Preliminary results show rarefied richness was significantly lower during period II based on non-overlapping 95% confidence intervals. Furthermore, fish assemblage composition shifted between the two periods (PERMANOVA: $P < 0.01$). Average community dissimilarity (SIMPER) between periods was 58% and three species (*Notropis boops*, *Lepomis megalotis*, and *Hybognathus nuchalis*) each contributed to >10% total dissimilarity. Persistence (T) of fish assemblages averaged 0.63 (range = 0.35 to 0.76) across paired sites. Despite moderate persistence of assemblages between periods, we found a notable decline in fish species richness. Ultimately, fish species intolerant to siltation declined or were not detected during recent collections, whereas tolerant species increased in distribution and abundance. Our results suggest fish assemblages have changed through time in the upper Saline River basin indicated by a loss of richness and high degree of community dissimilarity.

Keywords: assemblage structure; upper Saline River; watershed disturbance; species richness; historical data
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22. DOES PHYSIOGRAPHY INFLUENCE STREAM FISH RESPONSE TO HYDROLOGIC VARIABILITY?

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Broadly distributed taxa present a unique conservation challenge due to spatial variation in geologic, topographic, and climatic factors that may influence biotic responses to changing conditions. Many Southeastern stream fishes have distributions that overlap multiple physiographic provinces, yet drivers of population and community dynamics are frequently examined at spatially restricted scales, which limits understanding of interactions between physiography and key drivers. We addressed this issue by examining stream fish response to hydrologic variability in the Apalachicola-Chattahoochee-Flint River Basin, a large watershed encompassing multiple physiographies. We assessed stream fish response by estimating persistence and colonization rates as a function of physiography, a measure of high flow frequency (number of high flow days, HFD), and their interaction. We used periodic survey data from 40 sites distributed throughout the basin; 23 sites were below the Fall Line in the Coastal Plain ecoregion, while the remaining sites were in either the Piedmont or Blue Ridge ecoregion, both of which are above the Fall Line. To minimize confounding effects, we restricted our analysis to a subset of 19 taxa that were detected in multiple streams above and below the Fall Line. Rates were estimated by a multi-species dynamic occupancy model fitted using a hierarchical Bayesian framework that accounts for incomplete detection. Our results suggest an important role of physiography, with colonization rates much higher at Coastal Plain sites than at sites above the Fall Line. Additionally, we found that persistence was positively related to HFD regardless of physiography, whereas the effect of HFD on colonization appeared more likely to

vary by region. Collectively, our results support the hypothesis that demographic processes underpinning changes in stream fish community dynamics can vary among different physiographic regions, thus highlighting the need to consider spatially variable relationships in conservation efforts.

Keywords:flow ecology; Coastal Plain; Fall Line; colonization; persistence
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23. HISTORIC CHANGES IN FISH ASSEMBLAGE PATTERNS IN THE LITTLE MISSOURI RIVER, ARKANSAS.

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Long-term data sets are required to understand links between land use change and persistence and stability of fish assemblages. Data from fish faunal surveys conducted in the 1970s and 1980s by students from University of Louisiana at Monroe were available to begin examining changes in fish assemblages and relationships with land use in the Little Missouri River watershed. Jaccard Index, Morisita-Horn Index, and Bray-Curtis Index were used to determine similarity between historic (1970s and 1980s) and current (2016) fish assemblages. Fish assemblage persistence across sites ranged from 22% to 91%. *Notropis boops* (39% and 28%) had highest relative abundances during both collection periods, respectively. Fish assemblages between time periods had low to moderate similarity based on three indices (Jaccard's: 0.44, Morisita-Horn: 0.51, Bray-Curtis: 0.43). *Micropterus punctulatus* increased in frequency of occurrence across sites by 75% in the Little Missouri River from early 1970s to 2016. Similarly, *Lepomis megalotis* increased by 42%, *Etheostoma blennioides* 25%, *Lepomis cyanellus* 33%, *Gambusia affinis* 25%, and *Hybognathus nuchalis* 17%. *Etheostoma collettei* decreased in frequency of occurrence by 33% and *Etheostoma chlorosoma* by 25%. There was an apparent increase in lowland, pool-adapted species across the 12 study sites. There were correlations between current percent land use in the upstream catchment (determined from NWALT dataset) and Jaccard's fish similarity among time periods at 10 sites. Fish assemblage similarity was negatively correlated with development and agricultural land use ($\rho = -0.77$ and -0.40 , $p < 0.05$) and positively correlated with forested land use ($\rho = 0.50$, $p < 0.05$). Moving forward, we will use environmental variables to evaluate changes in fish assemblage patterns in the Little Missouri River.

Keywords:persistence:stability:land use
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24. MIGRATION DYNAMICS OF OHIO SHRIMP, *MACROBRACHIUM OHIONE*, IN ARKANSAS

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The Ohio shrimp (*Macrobrachium ohione*) is amphidromous, where a marine environment is required for early juvenile development, but other life stages are completed in freshwater. In the Mississippi River system, upriver adult habitat historically included the Ohio River and Upper Mississippi River, but abundance has reportedly declined, particularly upriver of Louisiana. Migration has been hypothesized to be impacted by dams and channel flow alterations. Current distribution, abundance, and life history of Ohio shrimp are relatively unknown upriver of Louisiana. We collected shrimp monthly, beginning in March 2016, with wire-meshed traps deployed at three sites along the mainstem Mississippi River and at sites in the Lower Arkansas, White, and St. Francis rivers. Age-0 to age-2 shrimp were collected routinely at study sites on the Mississippi River; however, study reaches within tributaries varied seasonally for catches of age-0 shrimp. Total catch drastically increased during late May/early June (20.5 – 25.4 °C), peaked during late June/July (30 °C), and declined during August/September. Preliminary examination of shrimp collected at McAllie Access on the Mississippi River in July suggests sex ratios of migrants varied with size/age class. Age-0 migrants (TL range 21.8 – 43.3 mm) were female-skewed (370 females: 202 males), whereas older shrimp, apparently continuing their upstream migration, were male-skewed (5 females: 35 males). Migration of age-0 shrimp was further than 600 river kilometers from the Gulf of Mexico, and preliminary data suggest a number of older Ohio shrimp, primarily males and inexplicably some females, continue to move upriver after their first overwintering. Despite declines from historic densities, Ohio shrimp likely remain important components of food webs in the lower Mississippi River and its tributaries.

Keywords: Amphidromy; migration; *Macrobrachium*; Arkansas; Mississippi
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25. PRELIMINARY GENETIC INVESTIGATIONS INTO THE DIFFERENTIATION OF *PROCAMBARUS PENNI* AND *PROCAMBARUS CLEMMERI*, INCLUDING CLARIFICATION OF DISTRIBUTIONS IN MISSISSIPPI

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Effective conservation begins with the ability to differentiate species and describe species distributions. The Pearl blackwater crayfish (*Procambarus penni*) is a Mississippi Species of Greatest Conservation Need (SGCN) due to rarity. Yet, uncertainty surrounds *P. penni* in terms of distribution and abundance. Furthermore, uncertainty exists in its differentiation from the Cockscomb crayfish (*P. clemmeri*). To clarify distribution and differentiation between these two species, we collected 48 specimens from 16 sites across the Pearl, Pascagoula, and Coastal Rivers drainages in southern Mississippi where putative *P. penni* and *P. clemmeri* are known to exist. Specimens were assigned a species identification using 1) morphological traits (i.e. gonopod) and 2) patterns in genetic relationships based on mitochondrial DNA (16s and CoxI).

Preliminary results suggest that *P. penni* and *P. clemmeri* are genetically distinct species. However, results also indicate that taxonomic identification via gonopod morphology is not always consistent with observed genetic relationships. Additional work is forthcoming to resolve these discrepancies.

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26. AQUATIC HABITAT RESTORATION VIA REMOVAL OF JIM WOODRUFF DAM ON THE APALACHICOLA RIVER: A PROPOSAL

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The Apalachicola River is 106 miles long and is formed by the confluence of its two major tributaries, the Flint and Chattahoochee Rivers, at the town of Chattahoochee, Florida. Its drainage basin includes portions of east Alabama, west Georgia, and north Florida. It is the second largest Gulf Coast river basin east of the Mississippi River. Jim Woodruff Dam was constructed on the Apalachicola River in the mid-1950s. Justification for the dam was barge navigation and power generation. While power is still generated at the dam, there has been no channel maintenance and almost no barge traffic for the past 15 years. Removal of Jim Woodruff Dam would allow restoration of approximately 80 miles of riverine habitat plus additional miles of tributaries to the lower Chattahoochee and Flint Rivers. This would contribute significantly to the recovery of many of the 100+ species of native fishes and 34 mussels in the basin. Dam removal would especially benefit the five diadromous fishes, one of which is listed as federally threatened (Gulf Sturgeon, *Acipenser desotoi*) and one as a candidate species (Alabama Shad, *Alosa alabamae*). Restoration of riverine habitat would also contribute significantly to the recovery of six federally listed mussels—four endangered and two threatened—plus one candidate species. The major issues involved in removal of Jim Woodruff Dam (e.g., hydrologic, ecologic, biologic, and economic) will be discussed.

Keywords: Apalachicola River; habitat restoration; dam removal; Jim Woodruff Dam
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Poster Presentations:

1. GROWTH AND DIET OF THE BROWN MADTOM (NOTURUS PHAES) IN FORT POLK, LOUISIANA

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The brown madtom, *Noturus phaeus*, is a moderately sized nocturnal madtom species found occupying dense woody cover in streams of the Mississippi River tributaries, the Sabine River,

and the Bayou Teche drainages. Of 27 madtom species, over half have been state or federally listed, and currently make up six of seven Ictalurid species protected under the federal Endangered Species Act. The length of the brown madtoms collected from Fort Polk, LA were recorded, then the pectoral spines were aged by counting annuli. These data were then analyzed by comparing length-at-age histograms. The proportion of invertebrates found in gut contents and field were used to formulate Strauss's Linear Index. According to the von Bertalanfy growth model, *N. phaeus* follows the standard growth curve. According to Strauss's Linear Index, their foraging habits tend to reverse between winter 2004 and summer 2015. Understandably, fewer insect orders were collected in the winter of 2004. Basic life-history information is not only useful for management of *N. phaeus*, but might also provide insight into life histories of related madtom species, ultimately leading to improved practices for future madtom conservation. This work will continue in summer 2016 so better comparisons can be made across years.

*Keywords: Madtom; von Bertalanfy; diet
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2. EFFECTS OF WAVE EXPOSURE ON THE STRUCTURE OF FISH ASSEMBLAGES ACROSS AN EXPOSURE GRADIENT

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Wave exposure can have a significant effect on the structure of fish assemblages of any coastal area. Few studies have explored the extent of this in salt marsh estuarine habitats, which is vulnerable to coastal erosion. Therefore, a study to better understand the possible effects of increased wave exposure on fish assemblages is needed as the threat of coastal erosion becomes increasingly prevalent. The Mississippi coastline has salt marsh habitat with many small bays and estuaries that provide shelter and produce gradients of wave exposure ideal for this study. We sampled fish assemblages monthly for 5 years at several coastal sites to address questions about how wave action impacts fish assemblages. The sites were categorized based on the intensity of exposure (open/intermediate/sheltered). We predict that sites with similar wave exposures will exhibit similar trends in both species richness and fish abundance. We hypothesize that the sites with intermediate wave exposure will have the most fish diversity in accordance with the Intermediate Disturbance Hypothesis. These samples were taken on days with minimal wave action for the safety of the collection crew; Therefore, we plan to further this study by sampling these sites on waver days to determine if these trends continue.

*Keywords: fish communities, wave action, ecology, salt marsh
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3. A COMPARISON OF HOST GENETICS AND ENVIRONMENTAL EFFECTS ON THE DERMAL MICROBIOME OF BANDED SCULPIN (*COTTUS CAROLINAE*)

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The “slime” layer of fishes is of ecological and evolutionary importance, as it is inhabited by a co-evolving community of microbes, including commensals and potential pathogens. The diversity and dynamics of this community are influenced by the host genetics and the environment, but the relative contributions of these two factors remain largely unexplored. We are applying high-throughput molecular techniques to test for differential host and environment effects on the microbiome of Banded Sculpin (*Cottus carolinae*), a species with strong population structure and a broad geographic range. We focus on the Mobile River Basin, where the species occupies both upland and lowland habitats. One population is found in Rabbit Creek, Clarke County, AL; which is part of the Tombigbee River drainage. The other is found in Reedy Creek, Clarke County, AL; which is part of the Alabama River drainage. While these populations are within 15 miles of each other, and nearly 100 miles from upland populations, they do not share a recent common ancestor. Genetic evidence allies the Rabbit Creek population to species found in the Black Warrior drainage while the Reedy Creek population is more similar to sculpins found in the Cahaba and Tallapoosa Rivers. The microbiome analysis reveals significant direct effects of genes (clades) and environment, as well an interaction effect. We discuss the relative importance of each, along with future directions involving the application of phylogenetic comparative methods to microbiome analysis.

Keywords: microbiome; slime; pylogenetics
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4. BODY SHAPE VARIATION WITHIN AND AMONG LINEAGES OF THE RAINBOW DARTER, *ETHEOSTOMA CAERULEUM*

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The Rainbow Darter (*Etheostoma caeruleum*) is distributed widely throughout the eastern United States, with several disjunct populations in the lower Mississippi River drainage. An unpublished morphological study of *E. caeruleum* suggested several potentially distinct species in the White River drainage and in tributaries of the lower Mississippi River drainage. However, published phylogeographic studies are not concordant with morphological data. This study used geometric morphometrics to evaluate body shape differences of *E. caeruleum* across its range. Specifically, we asked whether differences in body shape corresponded to previously identified lineages or putative species based on meristic data. Preliminary results based on populations from loess habitats of Crowley’s Ridge in Arkansas (St. Francis River drainage) and upland habitats of the Ozark Highlands (White River drainage) revealed some overlap in body shape between distinct lineages. However, disjunct populations from Crowley’s Ridge were the most distinct, with reductions in body and head depth relative to body length. Results from this study will provide valuable information about the distinctiveness of potentially unrecognized diversity within *E. caeruleum*.

Keywords: Geometric morphometrics, Crowleys Ridge, polytypic species
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5. ANALYSIS OF BODY SHAPE VARIATION AMONG RESTRICTED AND WIDESPREAD POPULATIONS OF THE SOUTHERN REDBELLY DACE, *CHROSOMUS ERYTHROGASTER*

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The Southern Redbelly Dace (*Chrosomus erythrogaster*) has a widespread distribution throughout most of the eastern United States, with several disjunct populations in midwestern states and in the lower Mississippi River drainage. Of particular interest are potentially isolated populations on Crowley's Ridge in Arkansas and the Bluff Hills in Mississippi. Currently, it is unclear whether these populations share connections with larger core populations in the nearby Ozark and Eastern highlands, or represent isolated populations on these "upland islands" within the Gulf Coastal Plain. This study used geometric morphometric techniques to evaluate body shape differences among *C. erythrogaster* from loess habitats of Crowley's Ridge and the Bluff Hills and upland habitats of the Ozark and Eastern Highlands. In conjunction with an ongoing population genetic study, results from this study will provide valuable information about the distinctiveness of potentially isolated populations in Arkansas and Mississippi.

Keywords: Geometric morphometrics, Crowleys Ridge, Bluff Hills
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6. DEVELOPMENT OF MICROSATELLITE LOCI FOR THE THREATENED BLACKSIDE DACE (*CHROSOMUS CUMBERLANDENSIS*) AND OTHER CO-OCCURRING CYPRINID SPECIES

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Allelic diversity was assessed for sixty microsatellite loci developed for the Blackside Dace (*Chrosomus cumberlandensis*) using two populations (Big Lick Branch and Right Fork Maces Creek). Overall, twenty polymorphic microsatellite loci were identified, with allelic diversity ranging from two to 13 alleles. An additional three loci were determined to be monomorphic. These are the first microsatellite loci reported for this species and will allow us to further examine the genetic variation within and among populations throughout its range. Additionally, potential cross-amplification and allelic variation was assessed for co-occurring cyprinid species known to hybridize with the Blackside Dace (*Semotilus atromaculatus* and *Chrosomus erythrogaster*), which will be useful when determining the degree of hybridization in the federally threatened Blackside Dace.

Keywords: Conservation; Genetics; *Chrosomus cumberlandensis*; Blackside Dace; Cyprinid
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7. CHANGES IN FISH ASSEMBLAGES OF THE YAZOO RIVER DRAINAGE BASED ON ANTHROPOGENIC EFFECTS

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Many aquatic systems are subject to fish assemblage changes as a result of anthropogenic alteration. Reservoirs result in increased homogeneity of communities relative to stream communities lacking obligate stream species. Community and environmental data, including land usage factors, were collected from 100 sites representing 116 species in the Yazoo River drainage around the Sardis and the Enid reservoirs. It is hypothesized patterns of community structure associated with reservoirs, increased homogeneity and a lack of stream specialist taxa, are observed in this system. Sites near the reservoir are expected to have a subset of the species seen further upstream while catering to fishes adapted to lentic environments. A possible confounding variable may be the relative abundance of channelized streams as a result of anthropogenic effects which could have a similar effect on community structure, increased homogeneity, as a reservoir. It is hypothesized sites with anthropogenic land alteration will also have increased homogeneity in community structure.

Keywords: community; reservoir; land-use; anthropogenic; Yazoo
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8. ENVIRONMENTAL DNA VS. TRADITIONAL SAMPLING: A CASE STUDY USING THE FEDERALLY THREATENED LEOPARD DARTER, *PERCINA PANTHERINA*

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Using traditional methods to sample for rare or endangered species may not always be reliable due to their low population numbers. However, the recent development of environmental DNA (eDNA) techniques represents a valuable tool to help counteract inefficiency associated with this traditional sampling. One example for which eDNA may be valuable in the detection of a rare species of fish is the Leopard Darter, *Percina pantherina*. This federally threatened fish, endemic to the Little River system in Arkansas and Oklahoma, is experiencing a decrease in genetic variation due to drastic population decline. Traditional sampling has failed to detect *P. pantherina* in the Robinson Fork since 2006 and in the Cossatot River since 2011. The objectives of this project are as follows: 1. to compare the effectiveness of two different eDNA sampling techniques, water and substrate, for detecting *P. pantherina*; 2. to compare potential differences in detectability of *P. pantherina* among eDNA, seining, and snorkeling; and 3. to determine if there are extant populations of *P. pantherina* in the Cossatot River and Robinson Fork. Species-specific primers that amplify a 137bp fragment of the cytochrome b gene were designed and in silico PCR screens using Primer Blast revealed a high degree of specificity for *P. pantherina*. These primers were used with PCR and assays of eDNA samples to compare *P.*

pantherina detectability between traditional and eDNA sampling for winter, spring, and summer samples. Traditional sampling for these periods resulted in zero detection of *P. pantherina* within our focal systems; however, *P. pantherina* eDNA has been detected in both winter and spring samples. These preliminary findings suggest that *P. pantherina* is not extirpated from the Cossatot River and Robinson Fork in Arkansas, but the species is likely undetectable when using traditional sampling methods in these systems.

Keywords: *Percina pantherina*; Leopard Darter; eDNA
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9. ISLANDS IN THE STREAM, IS THAT WHAT WE ARE: ASSESSING THE SPATIAL SCALE OF GENETIC DIVERSITY IN THE ENDANGERED TUXEDO DARTER, *ETHEOSTOMA LEMNISCATUM*.

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The federally endangered Tuxedo Darter, *Etheostoma lemniscatum*, is known from a 38.6 km reach of the Big South Fork of the Cumberland River where it is associated with shallow, slow-moving water with cobble or slabrock substrate, generally located adjacent to and upstream of riffles. This habitat type is patchily distributed and separated by long sections (generally over 1 km) of deep pools that are thought to be unsuitable for the species. Since *E. lemniscatum* is a small-bodied benthic darter with no swim bladder and benthic larvae, the species may have limited dispersal and, thus, limited gene flow among optimal habitat patches. This study will use genetic data to examine whether optimal habitat patches are functionally islands (or metapopulations), resulting from restricted dispersal and gene flow among patches separated by intervening pool habitats. Fin clips were taken from *E. lemniscatum* collected while snorkeling. To document the spatial genetic structure of the species, four sets of GPS coordinates were taken to delineate each habitat patch where *E. lemniscatum* individuals were observed or captured. A total of 232 individuals were observed; of these 165 individuals were captured and fin clipped between August-October 2015. Fish were collected from 18 distinct habitat patches encompassing the entire range of the species. Sixteen variable microsatellite loci were identified and optimized for PCR amplification from an initial survey of 45 species-specific microsatellite primers. A subset of individuals (n=42) from 12 habitat patches from across the range of the species have been genotyped. From this, preliminary results describing the genetic diversity and structure of the species will be estimated using standard methods for evaluation of microsatellite variation. Efforts to genotype all individuals at all target loci are ongoing.

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10. TRANSLOCATION OF BLUEBREAST DARTERS INTO THE UPPER LICKING RIVER, OHIO: INITIAL STEPS

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Aquatic ecosystems of Ohio historically supported diverse and abundant stream and river fish communities. Loss and fragmentation of high-quality aquatic habitat and impairments in water quality have led to significant alterations in the diversity, composition, and productivity of native fish communities. The bluebreast darter (*Etheostoma camurum*), for example, was extirpated from many Ohio systems over a century ago. In late spring 2016, 974 adult bluebreast darters were translocated from the greater Muskingum River basin into the upper Licking River. Translocated individuals were marked with visible implant elastomer (VIE) tags; in fall 2016, ~12% of translocated individuals were recaptured, as well as four young-of-year. VIE tags revealed that two individuals had moved greater than nine river kilometers following translocation. Complementing our translocation activities, we are also investigating the influences of hydrogeomorphology, water-chemistry, and resident fish communities on the ecology and reproduction of reintroduced darters.

Keywords: reintroduction, translocation, bluebreast darter *Etheostoma camurum*
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11. SPATIOTEMPORAL POPULATION DYNAMICS OF THE CADDO MADTOM (*NOTURUS TAYLORI*)

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The Caddo Madtom (*Noturus taylori*), an endemic to the Ouachita Mountain Ecoregion, is one of Arkansas' Species of Greatest Conservation Need and has recently been petitioned for federal listing under the Endangered Species Act. The restricted distribution of *N. taylori* in the upper reaches of the Caddo and Ouachita rivers, limited suitable habitat, and a knowledge gap in life-history characteristics necessitates further research to determine the current status and long-term population dynamics of the species. A previous study using allozymes revealed that *N. taylori* exhibited small effective population sizes and provided evidence for historical between-drainage differentiation, in addition to more recent reservoir-induced within-drainage differentiation. Here, we use microsatellite DNA loci and mitochondrial (mt) cytochrome *b* DNA sequence data, in conjunction with intensive sampling, to test multiple hypotheses involving patterns of within and among drainage connectivity and spatiotemporal population dynamics. In addition, seasonal sampling is being conducted to evaluate the current status and preferred habitat characteristics of *N. taylori*. Bayesian phylogenetic analysis of mtDNA using coalescent models resolve the Caddo and Ouachita river drainages as reciprocally monophyletic with recent divergence in the late Pleistocene (approx. 125,000 years ago). Preliminary analyses of the microsatellite data suggest similar pattern of significant between drainage differentiations ($F_{st} = 0.14$). Of 15 historical localities 11 have been thoroughly sampled for presence/absence, and 8 of 11 localities were successful. Five of the 15 historical localities were included in the survey where an average increase in relative abundance was observed from spring to summer with 0.29 to 9.05 relative abundance in the Caddo river and 0.09 to 4.75 relative abundance in the Ouachita river. Results from this study will aid in future listing decisions by U.S Fish and Wildlife, and

state rankings by the Arkansas Game and Fish Commission.

Keywords:conservation, habitat fragmentation, phylogeography
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12. GENETIC STRUCTURE AND DIVERSITY OF DISJUNCT POPULATIONS OF RAINBOW DARTERS (*ETHEOSTOMA CAERULEUM*) AND SOUTHERN REDBELLY DACE (*CHROSOMUS ERYTHROGASTER*) THROUGHOUT THE MISSISSIPPI CORRIDOR

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Studies focusing on the conservation of North America's freshwater fish fauna mostly focus on species with recently fragmented distributions or those species with naturally small ranges. Our grasp of how recent, natural isolation affect the migration and genetic diversity of common and widespread freshwater fishes is still limited. *Etheostoma caeruleum* and *Chrosomus erythrogaster* are widespread with overlapping distributions, highly abundant, and show comparable patterns of naturally disjunct populations on their range margins. Both species have potentially isolated populations on Crowley's Ridge in Arkansas and the Bluff Hills in Mississippi. These regions are relatively recent geological formations that would have been colonized by these species within the last 10,000 years making these species excellent subjects for studying the impacts of recent, natural genetic isolation. In this study, we analyze microsatellite DNA loci from both species, with an emphasis on the tributaries of the lower Mississippi river, to evaluate genetic diversity and connectivity among populations on Crowley's Ridge, the Bluff Hills, and larger core populations. Using DNA sequence data, we also test hypotheses about the pattern and timing of colonization of Crowley's Ridge and the Bluff Hills. The results of this study will provide valuable information about the biogeographic and demographic history as well as the conservation status of potentially disjunct populations of *E. caeruleum* and *C. erythrogaster* from Crowley's Ridge and the Bluff Hills.

Keywords:Isolation; Microsatellites; *Etheostoma caeruleum*; *Chrosomus erythrogaster*; genetic diversity
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13. INVASION OF THE BASIN SNATCHERS: DETECTION OF THE SPREAD OF SILVER CARP IN THE LAKE PONTCHARTRAIN BASIN USING ENVIRONMENTAL DNA (EDNA)

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Silver Carp have become a major threat to aquatic diversity in the United States. The species is now recorded from 15 states, including Louisiana, and is predicted to spread to many more. The

Bonne Carré Spillway is a flood control structure connecting the Mississippi River and Lake Pontchartrain Basins. The spillway was constructed to relieve flooding pressures during high water periods to protect New Orleans by releasing Mississippi River flood waters into Lake Pontchartrain. On average, the spillway is opened once every ten years, most recently in 2011 and 2016. We utilized environmental DNA sampling techniques and species-specific primers for D-loop to assess the occurrence of Silver Carp across the Lake Pontchartrain Basin before and after the opening of the spillway in January 2016. We collected water samples from nine sites throughout the basin and six sites from within the spillway's floodplain for eDNA detection. Positive detections were determined by the return of confirmed Silver Carp sequences from qPCR amplifications. Pre-opening sampling yielded positive eDNA hits from 5/9 lake and up-river sites and 6/6 floodway sites. The spillway was opened for 22 days, and the same sites were visited for repeat sampling in early and late summer 2016. Results from the early summer sampling yielded positive hits at all (9/9) lake and up-river and fewer (4/6) floodway sites, whereas in late summer a fewer number of positive hits were recovered from the lake and up-river (4/9) and floodway sites (1/6). The results of this study clearly indicate that eDNA sampling is an effective approach for detecting Silver Carp. Unfortunately, the results also suggest that despite the salinity of Lake Pontchartrain, Silver Carp distribution across the basin was greatly facilitated by the influx of freshwater from flood prevention measures.

Keywords: environmental DNA, invasive species, Louisiana
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14. EFFECT OF MARSH FRAGMENTATION AND PATCH AREA ON SPECIES RICHNESS IN MISSISSIPPI

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Fragmentation divides continuous habitat into smaller patches. Fragmentation can also produce smaller population of species, because fragmentation can split a population into smaller groups. Both terrestrial and aquatic ecosystems suffer from fragmentation. There are a variety of fishes that live along marsh, each depending upon the marsh for protection, food, and sometimes even competition. Species richness of fishes can be altered due to marsh fragmentation. We predict islands with larger patch index should have a higher species abundance and species richness of fishes. For this study, 19 islands of marsh were sampled in the months of June and July with two 5m pulls of a 30 foot wide bag seine per island of marsh. Fish assemblages were sampled at each island. A total of 3305 fishes were captured and 33 species were identified. A drone was used to take aerial images of each island of marsh to determine the patch index (the ratio of patch size and perimeter) of each island to determine how much each island has fragmented.

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15. STREAM BIOMONITORING OF FORT POLK, LOUISIANA

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Since 2001, macroinvertebrate and fish communities have been monitored periodically at headwater streams on the Ft. Polk Army Installation and the Peason Ridge Artillery Range in Louisiana. To improve water quality, there have been improvements made to culvert designs, water treatment, and waste disposal practices. Researchers have been returning to the site yearly to compile data in order to assess the influence of the changes and determine the need for further enhancements.

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16. CHARACTERIZATION OF THE DERMAL MUCOSAL MICROBIOME ACROSS POPULATIONS OF RED DRUM (*SCIAENOPS OCELLATUS*)

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The teleost dermal mucosa is a host-mediated ecosystem for commensal microbes, and the first line of defense against pathogens. Despite advances in microbial taxonomy and DNA sequencing technology, microbial diversity within fish

Keywords: Red Drum; Microbiome
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17. LAUREL DACE (*CHROSOMUS SAYLORI*) MICROHABITAT PREFERENCES AND DIET COMPETITION WITH NON-NATIVE SPECIES IN IT'S HISTORICAL RANGE

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Laurel Dace (*Chrosomus saylori*) are an endangered species endemic to the Tennessee River Basin in Tennessee. Historically, Laurel Dace were known to occupy only seven streams up on Walden Ridge, TN. Recent sampling has indicated a decrease in the range of this species. Due to their small isolated range, microhabitat variables and diet competition with non-native species were studied in order to understand possible drivers of their small population size. For the microhabitat study, we sampled a total of 66 sites (pools) in Bumbee Creek and Lick Branch. Each pool was measured for water volume, water depth, substrate type, canopy cover and silt depth. We demonstrated a significant correlation ($p < .05$) in mean water depth and mean water volume between pools where Laurel Dace were present versus absent. Substrate type, canopy cover, nor silt depth appeared to determine presence or absence of Laurel Dace ($p > .50$). The diet

of Laurel Dace and its non-native competitors, Green Sunfish (*Lepomis cyanellus*) and Bluegill (*Lepomis macrochirus*), were examined as well. Five sites within the range of Laurel Dace were sampled. Any invasive species caught at these sites, were preserved for stomach content analysis. Laurel Dace were not collected from these sites, but were obtained from the already preserved specimen collections at the University of Tennessee. Stomach content analysis was performed by dissecting these preserved specimens. Preliminary results from the *C. saylori* stomach contents revealed that during warmer months, their diet consisted primarily of terrestrial and aquatic invertebrates and in colder months, their diet appeared to be primarily plant matter, such as algae. In order to protect this unique species, it is imperative that we continue these studies in order to understand more about the Laurel Dace and how it interacts with both abiotic and biotic factors within its environment.

Keywords: Microhabitat, Diet, Laurel Dace,
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18. BUTTAHATCHEE RIVER INDEX OF BIOTIC INTEGRITY ASSESSMENT

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The Buttahatchee River is a 125 mile tributary to the Tombigbee River (Mobile River Drainage) flowing through northern Alabama and Mississippi. It is considered a priority strategic habitat unit by Alabama and Mississippi due to its high aquatic species diversity. In both states it contains populations of Frecklebelly Madtom *Noturus munitus*, a species currently petitioned for federal listing. This project served to complete an Index of Biotic Integrity for the river system in Mississippi in 2016 by collecting fishes, while data for fishes from the Buttahatchee River in Alabama was collected by Geological Survey of Alabama in 2015. Additionally, it served as an update on the status of *Noturus munitus* in the Buttahatchee River in Mississippi. A total of 26 sites were collected in 2016 Buttahatchee River in Mississippi during this project and resulted in 6026 fishes comprised of 71 species. Although *Noturus munitus* was found at four sites (n=18) in AL in 2015 and at five sites in MS (n=54) in 2016, relative abundance was much lower than historical collections in Mississippi. Data from this project will be used to continue long term monitoring of the river system and will help target both impaired areas that may benefit from in depth habitat assessments and associated conservation actions or high quality areas where habitat protection may need to be focused.

Keywords: Buttahatchee; IBI; *Noturus munitus*
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19. SURVEYS FOR THE DIAMOND DARTER (*CRYSTALLARIA CINCO*), AN ENDANGERED SPECIES KNOWN HISTORICALLY FROM THE GREEN RIVER IN KENTUCKY

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The Diamond Darter formerly occurred in the Ohio River basin in Kentucky, Tennessee, Ohio, and West Virginia; however, it is now extant only within a 22-mile section of the Elk River in West Virginia. Due to its decline and currently restricted range, the Diamond Darter was federally listed as endangered in 2013. In Kentucky, the species is known only from six pre-1930 records: lower Cumberland River (1), upper Green River (3), and Ohio River (2). It was last collected in the Green River near Cave Island, Edmonson County, in 1929. Extensive sampling for fishes in the Green River during the past 30 years using seines and electrofishing has failed to detect the species. The Diamond Darter is difficult to collect using standard sampling methodologies because it is nocturnally active and can occur in depths and current velocities not easily worked with a seine or electrofisher. Because the upper Green River contains habitat similar to that occupied by the species the Elk River, a 95-mile section from Cave Island (Mammoth Cave National Park) upstream to Greensburg has been designated a critical habitat unit. During 2012-2015, we completed sampling within the critical habitat unit using a benthic trawl at 38 sites and nocturnal sampling with seines and spotlights at six sites. Our objective was to determine if the species still persists in the Green River and document fish community composition, habitat, and water quality variables. We documented a total of 55 fish species, but the Diamond Darter was not encountered. Updated distributional data were obtained for six state-listed species of conservation concern, as well as a general inventory of the fish fauna and habitat conditions. This information is intended to help guide future Diamond Darter recovery actions (e.g., reintroduction).

Keywords: endangered; Diamond Darter; distribution; survey;
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20. TEMPORAL AND SPATIAL PATTERNS OF FISH DISTRIBUTION AND DIVERSITY IN THE NOXUBEE RIVER

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The Mobile Basin has undergone extensive channel modification with corresponding declines in the distribution and abundance of native aquatic fauna. However, many of the declining aquatic species of the Mobile Basin may persist within unmodified sub-basins. The Noxubee River is a sub-basin of the Mobile Basin that is relatively unaltered throughout its watershed and we hypothesized that the system could serve as a refugium for declining riverine species of the Mobile Basin. We characterized species richness and assemblage structure of fishes in the Noxubee River and its tributaries to determine whether or not contemporary fish assemblages resembled the historical assemblages as described before construction of the Tennessee-

Tombigbee Waterway. We found that fish assemblages of the contemporary river are similar to those of the historical river and many species now absent from another portion of the Mobile Basin are still present and maintain similar distribution and abundance patterns. Our results are informative because there is little existing information available regarding fish assemblages in the Noxubee River since major alterations were imposed upon the Mobile Basin through construction of the waterway. These results are discussed relative to findings from another Mobile Basin tributary that exhibited a marked change in many fish assemblage properties following construction of the waterway. Variable resiliency among tributaries to altered main-channel rivers illustrates the need for predictive tools that can be employed to prioritize conservation efforts.

Keywords: Mobile Basin; Tennessee-Tombigbee Waterway; Noxubee River; regulated rivers, fish assemblage structure
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21. STATUS SURVEY AND CONSERVATION GENETIC ASSESSMENT OF THE PALEBACK DARTER, *ETHEOSTOMA PALLIDIDORSUM*

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The Paleback Darter (*Etheostoma pallididorsum*) is endemic to the upper Caddo and Ouachita rivers in the Ouachita Highlands of west-central Arkansas. It is a Species of Greatest Conservation Need in Arkansas and, in 2010, was included in a mega-petition for federal listing by the Center for Biological Diversity. To better inform federal listing decisions, the objectives of this project were as follows: (1) conduct a range wide survey of all historic localities; (2) obtain size-distribution data and relative abundance at select localities on a seasonal basis; and (3) generate population characteristics via non-lethal genetic methods (e.g. genetic variation, effective population sizes, patterns of migration). Average relative abundance (defined as capture number/number of sample efforts) of all four seasonal sites was similar between winter and spring (0.36 and 0.39, respectively), with highest values in the summer (0.94). Over all seasons, relative abundance was similar between Caddo and Ouachita drainages (0.59 and 0.54, respectively). Preliminary analysis of microsatellite DNA loci revealed low, but significant genetic structure among localities ($F_{st} = 0.054$, $P < 0.001$), with the highest levels of structure observed between river drainages. Population genetic diversity is relatively low (mean $H_e = 0.59$; mean alleles per locus = 5.06), but comparable to close relatives *E. boschungii* (mean $H_e = 0.67$; mean alleles per locus = 6.74) and *E. tuscumbia* (mean $H_e = 0.57$; mean alleles per locus = 5.53). While preliminary, these results suggest that *E. pallididorsum* is relatively stable and abundant at historic localities sampled thus far. Preliminary genetic data suggests low, but significant levels of structure between the Caddo and Ouachita river drainages. Future analyses will provide updated information on range-wide presence/absence at historic localities and seek to better understand the historical and contemporary aspects of connectivity within and among Caddo and Ouachita river drainages.

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22. ENVIRONMENTAL DNA DETECTION OF BLACKBANDED SUNFISH (ENNEACANTHUS CHAETODON) IN SOUTH CAROLINA AND GEORGIA

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Blackbanded Sunfish (*Enneacanthus chaetodon*) is a member of the North American Centrarchid family that is distributed along the Atlantic coast from New Jersey to Florida, and is typically associated with shallow and heavily-vegetated blackwaters of herbaceous and forested wetlands. Blackbanded Sunfish have occurred historically in four distinct population centers including central Florida, southern Georgia, the Carolinas, and New Jersey. Despite repetitive efforts to sample Blackbanded Sunfish over the past decade throughout their southern range using traditional methods (seines, traps, electrofishing), surveys have provided evidence of only a few extant populations in GA and FL and a potentially fragmented distribution along the upper coastal plain in SC. The results of these surveys suggest that Blackbanded Sunfish populations are in decline in their southern range and that the habitat preference of the species prohibits effective sampling using traditional fisheries methods. The collection of environmental DNA (eDNA) has become a popular approach for monitoring rare or invasive aquatic species because an organism can be detected by sampling its environment for DNA presence using relatively rapid and efficient techniques, compared with traditional labor-intensive sampling methods. We have developed a qPCR-based eDNA detection tool for Blackbanded Sunfish targeting a small fragment of the mitochondrial gene, cytochrome b. Laboratory and field tests of our eDNA tool demonstrate robust detection probabilities. Here we present preliminary results for the second phase of this collaborative, multistate project: collecting and analyzing eDNA at sites with apparently suitable habitat for Blackbanded Sunfish in SC and GA with hopes of discovering new populations that have not been detected through traditional sampling. A total of 60 sites were visited in both GA and SC (30 sites per state) during the fall of 2015 and the spring of 2016, respectively. All samples collected from GA have been processed and our results show eDNA detections of Blackbanded Sunfish in four locations where traditional sampling was ineffective. Analyses are ongoing for SC sites, but Blackbanded Sunfish eDNA has been detected for nearly 1/3 of these sites. Our preliminary results detecting new populations of Blackbanded Sunfish in the southeastern US are encouraging for conservation effort of this species. The next phase of our project will be to return to all locations that had positive eDNA detections and rigorously sample these highly vegetated, blackwater systems using traditional sampling methods (i.e. seines, dip nets, electrofishing, etc.) to collect enough individual tissue samples for a large scale population genetic health assessment.

Keywords: Blackbanded Sunfish; eDNA; environmental DNA; blackwater; qPCR
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23. FISH SURVEY AND BIOASSESSMENT OF CYPRESS CREEK IN THE TENNESSEE RIVER DRAINAGE

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Cypress Creek is a northern tributary to the Tennessee River (Pickwick Lake) in Lauderdale County, Alabama and Wayne County, Tennessee with a drainage area of ~650 km² in the Western Highland Rim Ecoregion. Although sampled for fishes such as *Hemitremia flammea* and *Etheostoma tuscumbia* over the past several decades, the last comprehensive fish survey of the watershed was conducted in 1971, and changes in the fish fauna may have occurred. Historical data (1884–2015) containing over 3,600 records was compiled and distribution maps were generated for each previously documented species. Sampling was conducted in 2009–2016 (primarily 2015–16) at 62 localities and utilized the 30+2 method of the Geological Survey of Alabama (GSA). Community composition was analyzed with the Index of Biotic Integrity (IBI) using parameters for Tennessee Valley streams developed by the GSA. Collections from all major tributaries and previously unsampled stream reaches yielded ~1,600 records representing 80 species. Several species (e.g. *Aphredoderus sayanus*, *Noturus exilis*) that were historically rare in collections were more frequently encountered when sampling appropriate habitats in multiple tributaries. Species that had not been encountered for 70 years or more (e.g. *Notropis ariommus*, *Etheostoma cinereum*) were not rediscovered in this study and are likely extirpated from Cypress Creek. The current fish fauna is 96 species, which represents high species diversity for a moderate-sized stream in this region, and ranks Cypress Creek among the most species-rich streams in the southern bend of the Tennessee River. IBI scores varied among tributaries, suggesting differing anthropogenic impacts on fish community composition across the watershed. Land use changes within the watershed should be closely monitored to ensure the aquatic biodiversity of this stream is not compromised.

Keywords: Biodiversity; Extirpation; Fish Community; Index of Biotic Integrity; Tennessee River
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24. CONSERVATION AND RESTORATION OF OHIO WETLAND AND GLACIAL-LAKE FISHERIES

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Fishes associated with wetland and glacial lake habitats are highly imperiled in Ohio. Here, we present the initial stages of a plan to restore these fishes through a combination of captive propagation, research, and reintroduction activities. In Year 1, we successfully propagated 225 Blacknose Shiners *Notropis heterolepis* and 140 Lake Chubsuckers *Erimyzon sucetta* at rearing facilities of the Schiermeier Olentangy River Wetland Research Park at The Ohio State

University. Subsequently, propagated fish were released into a protected constructed wetland. These propagation and reintroduction efforts are complemented by research that will help identify the environmental conditions that affect success of reintroductions and the establishment of stable populations, as well as evaluate the influences of native wetland fish reintroductions to resident aquatic communities and food webs. Year 2 activities will include ongoing research and the propagation and reintroduction of additional wetland species.

Keywords: reintroduction, propagation, wetland, glacial lake
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25. ONE FISH? TWO FISH? HOW MANY SPECIES COMPOSE THE LONGEAR SUNFISH (*LEPOMIS MEGALOTIS*)?

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Determine if color variations and geographic isolation (as determined by our DNA analysis) in populations of *Lepomis megalotis* are sufficient to split this species in two or more species. Materials and methods used were taxon sampling, genes and alignment and phylogenetic analysis. Examined 63 samples from 33 locations and recovered a well resolved phylogenetic tree. We find three major lineages of *Lepomis megalotis* but they do not correspond to distinct geographic areas nor do they correspond to distinct eco-morphotypes. The variation that we find within *Lepomis megalotis* is less than we find between other species of sunfishes. We reject the alternative hypothesis (H_1) that *Lepomis megalotis* corresponds to more than one species and fail to reject the null hypothesis (H_0) that *Lepomis megalotis* is a single species that is highly morphologically variable with several eco-morphotypes.

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27. CONSERVATION STATUS OF THE SMALLSCALE DARTER, *NOTHONOTUS MICROLEPIDUS*

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The Smallscale Darter (*Nothonotus microlepidus*) occurs in only four tributaries to the middle and lower Cumberland River System in Tennessee and Kentucky (Little, Harpeth, Red, and Stones rivers). Due to its limited range, it is recognized as a “Species of Greatest Conservation Need” in Kentucky and “Deemed in Need of Management” in Tennessee. It has been petitioned for federal listing, but insufficient information is available to evaluate listing criteria. Using standard seining and backpack electrofishing techniques, we sampled 16 historical localities in Tennessee for Smallscale Darter to determine presence and abundance. Additionally, reach-scale habitat assessments were performed for each site, and flow and depth was measured for a subset of localities to characterize species-habitat associations. At two sites from each river system, the

Jolly-Seber mark recapture method was used to estimate population size. All remaining sites were surveyed in one or more attempts to determine presence/absence and population estimates were generated using regression and first pass collection data. The Smallscale Darter was present at 12 of 16 sampled localities. The Harpeth River had the largest population size estimates. The Stones River contained 3 of the 4 localities at which the focal species was not detected and showed a trend of lower population sizes and habitat quality scores, indicating that populations in this system may be particularly vulnerable. Smallscale Darters were found in habitat patches with an average flow of 1.72 cm/s and an average depth of 31.01 cm. We are currently surveying historical localities in Kentucky, using GIS to examine occurrence and abundance of the species in relation to land use activities, and analyzing genetic structure across the range of the species to provide a robust basis for conservation actions for the Smallscale Darter.

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