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### Presentation Abstracts (alphabetical by first author's last name)

Armbruster, Jonathan, Edward Burress, Shobnom Ferdous, and Paul Wieczorek. Auburn University, Department of Biological Sciences.

### The Evolutionary Ecology of Cyprinids

With over 260 species, the Cyprinidae is the largest family of freshwater fishes in North America. They occupy many different niches from deserts, to large rivers, to mountains. There are species with transcontinental ranges and species with very small ranges. 14 species or subspecies are considered extinct, accounting for 35% of the extinct North American fishes; thus, understanding the diversity of the Cyprinidae is imperative. We have been examining the evolutionary ecology of North American minnows and will discuss three topics: the ecology of minnows of a North Carolina assemblage, the changes in biodiversity from upstream to downstream habitats in Alabama, and the overall evolutionary history of shape change in North American minnows. Shape is being assessed using Geometric Morphometrics and is being used as a proxy for understanding overall ecology of the fishes. We have additionally examined diet through gut content analysis and stable isotope data for the North Carolina minnows. We explore the evolution of shape and diet through phylogenetic regression of characteristics, and find that many of the characters show phylogenetic signal. In Alabama, we find that as we go downstream, species are added within the main body of shape space (phenetic packing) vs. being added to the periphery (phenetic dispersal). Across North America, we find that basal clades occupy most of the fringe of shape space whereas the shiner clade fills in the space between all of the basal clades suggesting an initial burst of morphological diversity (phylogenetic phenetic dispersal) followed by further subdividing of shape space (phylogenetic phenetic packing). All these studies give us a greater understanding of the ecological place of cyprinids in North America, and will aid in future studies on conservation.

Bart Jr.<sup>1</sup>, Henry L., David A. Etnier<sup>2</sup>, and Wayne C. Starnes<sup>3</sup>. <sup>1</sup>Tulane University Biodiversity Research Institute; <sup>2</sup>University of Tennessee, Department of Ecology and Evolutionary Biology; <sup>3</sup>North Carolina Museum of Natural Sciences.

# Evidence for a New Species of Darter, Subgenus *Oligocephalus*, Genus *Etheostoma*, from Eastern Tributaries of the Lower Mississippi River, Mississippi and Tennessee

In the discussion of their description of the Gumbo Darter, *Etheostoma thompsoni*, Suttkus et al. (2012) commented that species-level recognition might be warranted for populations of *Etheostoma swaini* from eastern tributaries of the lower Mississippi River, because specimens from these populations differed markedly from specimens of *E. swaini* from the Pearl River (the species' type locality) in counts of lateral line scales, pored lateral line scales, scales around the caudal peduncle, pectoral fin rays and branched caudal fin rays. Suttkus et al. (2012) stopped short of describing the species because counts were not made for some southern Mississippi River populations and counts for other populations were thought to be intermediate. Here we present data gathered from specimens from the Obion River in Tennessee southward to the Buffalo River in Mississippi, which confirm convincingly that populations from eastern Mississippi River tributaries are diagnosable as a distinct species based their lower counts for lateral line scales, pored lateral line scales, and scales around the caudal peduncle. Our data do not confirm the differences in fin ray counts observed by Suttkus et al. (2012). Populations from the Lake Pontchartrain drainage and populations east of the Pearl River agree with *E. swaini*.

Bauer<sup>1</sup>, Eric, Brian Helms<sup>1</sup>, David Werneke<sup>1</sup>, Jason Zink<sup>2</sup>, and Greg Jennings<sup>3</sup>. <sup>1</sup>Auburn University, College of Science and Mathematics, Department of Biological Sciences; <sup>2</sup>Zink Environmental; <sup>3</sup>Jennings Environmental.

# Regional Reference Curves and Ecological Endpoints Using Fishes and Crayfishes for the Appalachian Plateau of Alabama

Eco-morphological stream design and assessment tools do not currently exist for the Appalachian Plateau ecoregion of Alabama. We sampled streams of the Bankhead National Forest in order to develop these tools in order to guide stream restoration efforts. These tools include: 1) stream geomorphological relationships with drainage area (regional curves) and 2) fish and crayfish assemblage metrics relationships with drainage area (ecological endpoints). Drainage area explained 94%, 97%, and 98% of the variation in channel depth, width, and crosssectional area of streams (N=40) in this region respectively. We collected 30 species of fish overall (N = 12 streams) with site-specific species richness ranging from 1 to 21. Drainage area explained 90%, 93%, 64%, 60%, and 39% of the variation in fish species richness, fish diversity (H'), community evenness, fish abundance, and fish biomass respectively. Conductivity never explained more than 30% of the variation in fish abundance or richness, but explained approximately 38% of fish biomass, nearly as much as that explained by drainage area. Three species of crayfish were collected with site-specific richness ranging from 1 to 3. Drainage area explained 58%, 68%, 18%, and 0% of the variation in species richness, average carapace length,

sex ratio, and crayfish abundance respectively (N=17 streams). Stream morphology, and fish and crayfish assemblage composition and metrics, to a lesser extent, change predictably with drainage area. These relationships can be used as guides for physical design and ecological endpoints for assessment of stream restoration projects in the Appalachian Plateau.

Burress<sup>1</sup>, Edward, Jordan Holcomb<sup>2</sup>, and Jonathan Armbruster<sup>1</sup>. <sup>1</sup>Auburn University, Department of Biological Sciences; <sup>2</sup>Fish and Wildlife Research Institute Florida Fish and Wildlife Conservation Commission.

### Body Size Is Negatively Correlated with Trophic Position among Minnows

Body size has many ecological and evolutionary implications for ecological networks, which extend across multiple levels of organization. Body size is often positively correlated with species traits such as metabolism, prey size, and trophic position. We use stable isotope analysis to quantify trophic position among minnows across multiple assemblages that differed in their species composition, diversity, and food web structure. Body size significantly predicted trophic position across different lineages and assemblages, and indicated a strong negative association. The observed negative relationship between body size and trophic position is contrary to conventional knowledge, and likely arose due to highly clade-specific patterns, such that clades consist of either large benthic species or small pelagic species. Body size and trophic position are likely coevolved in association with specialization along the benthic to pelagic resource axis.

Darden, Tanya, Forrest Sessions, Scott Lamprecht, Daniel Farrae, Jason Bettinger, and the Robust Redhorse Conservation Committee. South Carolina Department of Natural Resources.

### Evaluation of the Robust Redhorse (*Moxostoma robustum*) Restoration Effort in the Santee River Basin, South Carolina

Robust Redhorse (*Moxostoma robustum*) is a large-bodied and long-lived catastomid with a native range that encompasses the major rivers from the Carolina's Pee Dee River south to the Altamaha River (GA), with the notable exception of the Santee River basin. In an effort to increase the long-term viability of Robust Redhorse within its native range, we began a restoration effort to establishing a self-sustaining population in the Santee River Basin. Broodstock from the Savannah River were used to stock 72,000 juveniles from 9 year classes that involved 101 independent parental crosses during 2004-2013. Fingerlings were stocked in two Broad River segments and one Wateree/Congaree River section. Incidental electrofishing collection of adult fish began 2 years after stocking began. Males in spawning conditions were first observed in 2008 and the first spawning triad was encountered in 2010. To date, over adults have been collected and adults in spawning condition have been encountered in every stocked segment. Spawning behavior has been documented in two segments. Over 150 mature adult Robust Redhorse have been observed ascending a fish ladder on the Broad River preceding spawning season. Telemetry study has shown extensive movement and repeated seasonal use of

the specific river reaches. Genetic tagging evaluation and population characterization using a suite of 10 microsatellite markers (64 individuals) confirms that stocked fish are surviving in the new population, movement behaviors are similar to native populations, many year classes are present, genetic health metrics are optimistic and similar to the source population, and successful wild-spawned recruitment is occurring. Monitoring of the new Santee River population will continue, but all initial indications are optimistic for the restoration effort.

Elias, Diego, and Kyle Piller. Department of Biological Science, Southeastern Louisiana University, Hammond, Louisiana.

### Phylogeography of the Threadfin Shad (Dorosoma petenense, Gűnther, 1866)

The genus *Dorosoma* (Clupeidae) is composed of five recognized species: *Dorosoma cepedianum*, *D. smithi*, *D. anale*, *D. chavesi*, and *D. petenense*. The Threadfin Shad, *Dorosoma petenense* (Clupeidae), is the most widespread and generalist species of the genus possessing a neartic to neotropical distribution. However, the species is not continually distributed across its range, as a distributional break occurs within the Mexican Neo-volcanic Plateau in east central Mexico, in a region known as Punta del Morro. The taxonomic status of *D. petenense* has been problematic and others have recognized variation throughout its range, although there has not been a comprehensive taxonomic review of the species. The objectives of this study were to conduct a phylogeographic analysis of *D. petenense* throughout the range of distribution using molecular markers. Preliminary results show two clades of *D. petenense* in the northern and southern extremes of the distribution. Future work will focus on expanding the molecular data set and to add morphological data to better understanding taxonomic status of *D. petenense* and the phylogenetic relationships of the species within the genus *Dorosoma*.

Epstein<sup>1</sup>, Joshua M., Ben Baiser<sup>1</sup>, William E. Pine<sup>1</sup>, Christina M. Romagosa<sup>1</sup>, and Catherine Phillips<sup>2</sup>. <sup>1</sup>The University of Florida; <sup>2</sup>U.S. Fish and Wildlife Service.

#### **Functional Diversity of Southeastern U.S. Fish Communities**

Species represent a wide range and combination of traits, (i.e., behavior, feeding) some of which are unique and essential to ecosystem functioning, and some which are redundant within a community. This "functional diversity" (FD) is not always mirrored by traditional measures of species richness. Here we take a trait-based approach to explore patterns of FD in fish communities across the southeastern U.S. We obtained species presence/absence data from the MARIS, BISON, and USGS Nonindigenous Aquatic Species databases, and scored trophic and reproductive traits for the 386 fish species using the Virginia Tech FishTraits Database, primary literature, USFWS reports, and fish identification texts. We calculated species richness and FD (using the functional diversity throughout the eastern highland and Appalachian regions and lower functional diversity in the Coastal Plain areas. Our results suggest that sub-basins with

similar species richness but different FD values are likely a result of differences in river channelization, development, and habitat complexity. Going forward, we plan on relating FD to additional ecosystem health indicators (i.e., impervious surface and percent natural cover) to reveal potential correlations.

Eschenroeder, Jackman, and Dr. James Roberts. Georgia Southern University.

### Hybridization and Replacement of Roanoke Bass (*Ambloplites cavifrons*) with Invasive Rock Bass (*A. rupestris*) in Virginia: A Genetic Assessment of the Scope of the Problem

The Roanoke Bass (Ambloplites cavifrons) is a sport fish endemic to the Roanoke, Chowan, Tar, and Neuse drainages in Virginia and North Carolina. Virginia populations of this species have declined over time, presumably in part due to competition and hybridization with Rock Bass (A. rupestris), an invasive congener introduced from the Gulf slope throughout the early 20th century. Displacement and hybridization were reported as early as the 1950s, but the current status of this invasion and its impacts on A. cavifrons populations are unknown. Eleven nuclear DNA microsatellite markers were developed to discriminate between A. cavifrons, A. rupestris, and their hybrids, and in doing so to assess the current distribution of A. cavifrons in Virginia. The panel of markers provided a high degree of resolution among these groups, further allowing us to separate F1 hybrids from backcrossed individuals. Our results suggest a complex mosaic of invasion, displacement, and introgression patterns across the range of A. cavifrons in Virginia. In some populations all individuals appear to remain genetically A. cavifrons, whereas in other populations A. cavifrons has been partially or completely replaced by A. rupestris and/or hybrids. In the Pigg watershed, an ongoing invasion is apparent, with A. rupestris dispersing upstream and hybridizing with A. cavifrons, whereas in another watershed upstream invasion is prevented for now by a small dam. Of eight Virginia watersheds that historically contained A. cavifrons, only five now contain predominantly A. cavifrons, and three of these are susceptible to future invasion. This research helps to identify intact A. cavifrons populations that could be targeted for habitat restoration efforts, as well as vulnerable populations that could be targeted for programs seeking to prevent the spread of A. rupestris from adjacent waterways.

Farrae, Daniel J., Tanya L. Darden, and William C. Post. South Carolina Department of Natural Resources.

# Population Genetic Structure between Fall and Spring Spawned Atlantic Sturgeon (*Acipenser oxyrinchus oxyrinchus*) in the Edisto River, South Carolina

Along the Atlantic coast of the U.S., Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*) are managed as five distinct population segments (DPS). In 2012, the northernmost DPS was listed as Threatened and the other four were listed as Endangered. The Edisto River population (South Atlantic DPS), has a long sampling history with many juvenile Atlantic sturgeon captured. Evaluation of total length by capture day of year revealed a bimodal distribution, allowing fish to

be assigned to a fall or spring spawning event based on size. Fish assignments to year class resulted in fall year classes from 1996–2005 and 2011 (n=1,178) and spring year classes from 1996–1999 and 2001–2003 (n=209). We genotyped all sturgeon using a panel of 12 microsatellite markers to compare the genetic signatures of fall and spring spawned fish, assess the genetic "health" of the population(s), and estimate effective population size (Ne). Fall versus spring-spawned sturgeon were genetically different (FST=0.092); pairwise comparisons by year class basis supported these results. Additionally, STRUCTURE analyses indicated two populations (k=2) and identified fall and spring-spawned populations without any structure based on year class within a season. Observed heterozygosity for fall samples was 0.659 and for spring samples was 0.712; the inbreeding coefficient was effectively zero for both seasons. Overall estimates of Ne(LD) were 49.1 for fall and 14.0 for spring; Nb(LD) estimates by year class varied, but ranged from 20-30 per year class. Our results indicated that the Edisto River contains two genetically distinct populations of Atlantic sturgeon, one spawning in fall and the other in spring, and that both populations appear to have positive genetic "health" metrics. Although others have documented fall and spring spawning in several Atlantic river systems, this is the first evidence of genetic distinction of Atlantic sturgeon spawning groups within a single system.

Fraley<sup>1</sup>, Steve, Bryn H. Tracy<sup>2</sup>, and Andrea Leslie<sup>1</sup>. <sup>1</sup>North Carolina Wildlife Resources Commission; <sup>2</sup>North Carolina Division of Water Resources.

#### Status of Nonindigenous Fishes in North Carolina

Established populations of nonindigenous fishes are widespread in North Carolina (NC) and include 6 species from outside the USA, 13 species native to other states, as well as 59 indigenous species that now occur in systems outside their historical native range. Three tropical species are limited to artificially heated reservoirs with little potential for expansion into or impacts on native communities under present climate conditions, but most other species are extant in natural habitats and pose various threats to native communities and have the potential for further expansion. Twenty species were intentionally stocked by North Carolina Wildlife Resources Commission (NCWRC) and other resource managers for expanded game fishing opportunities or as forage for game fishes. Perhaps the most widespread and problematic species in this group is Flathead Catfish, Pylodictis olivaris, now common to abundant in eight Atlantic Slope river systems. Further threats include species presently known from adjacent states that occur in waters contiguous with NC waters (e.g., Northern Snakehead, Channa argus and related species, and Asian carps, Hypophthalmichthys spp.) and species with high commercial aquacultural interest. Vulnerability to and the establishment of nonindigenous species vary across river systems. The Catawba River system presently has the greatest number of nonindigenous species (35; 37% of the total number), and the New River system has the highest percentage of total species made up by non-indigenous species (29 spp., 52%), while the Lumber River system has the fewest (6 spp., 8%). NC's management and policies also vary considerably across regions and species. Historically, NC has not prioritized addressing the introduction and spread of non-native fishes. However, recently more attention has been focused to include the adoption of a statewide stocking permit requirement by the NCWRC, the development of a

statewide Aquatic Nuisance Species Management Plan, and a move to stock only sterile triploid trout and Grass Carp, *Ctenopharyngodon idella*.

Furtado, Brittany V., Loren Stearman, Jessie Green, Ginny Adams, and Reid Adams. University of Central Arkansas.

# Variation in Fish Assemblage Composition and Population Dynamics across a Gradient of Natural Gas Development on the Fayetteville Shale, Arkansas

The demand for natural gas has increased exponentially over the last 15 years and growth of unconventional natural gas development (UNGD) on the Fayetteville shale has occurred at a rate of 300-900 new wells/year. Using data collected each spring over five years (2010-2014), we tested for variation in both fish assemblage composition and population-level dynamics among three levels of UNGD (Control, Low, and High). Permutational multivariate analysis (PERMANOVA) indicated significant variation in fish assemblage composition among treatment groups in every year except 2014. Similarity percentage (SIMPER) analyses identified a suite of species that consistently contributed to greater than 10% of total dissimilarity among treatments. Analysis of length-frequency histograms for these species indicated fishes we have considered sensitive, due to specific reproductive and feeding requirements (e.g. Etheostoma spectabile, Etheostoma caeruleum, and Etheostoma whipplei), had an absence of young-of-year at High treatment sites in every year except for 2014. In contrast, more tolerant fishes (e.g. Lepomis cyanellus) had similar size distributions across all three treatments. Interestingly, *Notropis boops*, a species traditionally considered sensitive due to its intolerance to sedimentation, displayed trends in size-frequency distribution that closely mirrored Lepomis cyanellus. Analysis of both mean standard length (SL) and body condition showed both sensitive and tolerant species had significantly higher SLs and condition at High treatment sites which may be indicative of differences in resource allocation and/or some advantage for fishes at sites with greater UNGD that serves to mitigate the potential stressors. Both assemblage and population-level analyses showed variation linked to rapid UNGD, but due to a shift in the trends previously observed in the first four years of the study, also provided evidence for potential recovery in 2014 at sites that have seen greater levels of UNGD.

Gebhard<sup>1</sup>, Amy E., William G. Wells<sup>2</sup>, Thomas C. Johnson<sup>1</sup>, Robert T. R. Paine<sup>2</sup>, Lucas A. Hix<sup>1</sup>, Heather N. Ferrell<sup>1</sup>, Andrea N. Engle<sup>1</sup>, and Joshuah S. Perkin<sup>1</sup>. <sup>1</sup>Department of Biology, Tennessee Technological University; <sup>2</sup>School of Environmental Studies, Tennessee Technological University.

Loiters, Leavers, and Leptokurtosis: Measuring and Predicting Short-term Movement of Banded Sculpin, *Cottus carolinae*, in Tennessee

Fish movement is a critical component regulating population persistence and community structure in streams. Although there are numerous movement studies on *Cottus* species,

relatively little is known about the movement of Banded Sculpin (*Cottus carolinae*). Our goal was to determine if a newly developed leptokurtic dispersal modelling approach effectively predicted short-term temporal variability in movement of Banded Sculpin. We tracked 51 Banded Sculpin with passive integrated transponder tags for 46 days in a 600 m stretch of Little Creek, Tennessee to assess if a leptokurtic pattern predicted dispersal and if individuals exhibited "switching" behaviors (between stationary and mobile movement status). Leptokurtic models were developed using the *fishmove* package in Program R and correctly predicted dispersal patterns for 12 of 13 recapture occasions. Mean movement of stationary fish ("loiters") was 4 m (range 2-6 m) whereas mean movement of mobile fish ("leavers") was 75 m (range 45-150 m). The stationary proportion of the population (i.e., 1 – mobile proportion) averaged 0.45 (range 0.35-0.85). We found 28 of 51 fish exhibited switching behavior, and 18 of those 28 switched between stationary and mobile more than once. These findings collectively show that Banded Sculpin dispersal follows a leptokurtic pattern, is temporally dynamic even over short timescales, and individual dispersal histories include switching between stationary and mobile movement status. Validation of the leptokurtic dispersal model and increased resolution on the distances moved by Banded Sculpin benefit our understanding of benthic fish ecology in headwater tributary streams.

Greenwald, D. Noah, and Tierra R. Curry. Center for Biological Diversity.

### **Endangered Species Protection for Southeastern Aquatics**

In 2010, the Center for Biological Diversity petitioned for protection of 404 southeastern aquatic and wetland species under the Endangered Species Act, including 49 fish species. These fish species were selected with substantial input from the Southeastern Fishes Council. The Center developed an initial list of candidates based on NatureServe and American Fisheries Society rankings. Following a series of meetings between the Center and the Council, 23 species were eliminated from the petition, three were added and three were retained despite some disagreement among Council scientists. In 2011, the Center reached a settlement agreement with the U.S. Fish and Wildlife Service, under which the agency agreed to make an initial finding on the petition, as well as findings for hundreds of other species, resulting in a 2012 finding that 374 of the 404 species may qualify for endangered species protections. The agreement also resulted in listing of eight southeast fish species that had been awaiting protection on a list of candidate species in several cases for many years. In exchange for these findings, the Center agreed to limit the number of species for which it could file lawsuits to obtain listing decisions to no more than 10 species per year. The Center has been working with Southeastern Fishes Council scientists to identify the fish species from the 2010 petition that should be prioritized for listing and to date, has obtained dates for listing decisions for nine species. The Center's 2010 petition and 2011 settlement agreement have resulted in additional research on and protection of southeastern fishes.

*Gutzmer*<sup>1</sup>, Logan, Matthew Wagner<sup>2</sup>, Chad Kaiser<sup>1</sup>, Brian D.S. Graeb<sup>1</sup>, and Katie N. Bertrand<sup>1</sup>. <sup>1</sup>South Dakota State University; <sup>2</sup>Mississippi Museum of Natural Science.

# Interseasonal Diets, Critical Thermal Minimum, and Distribution of Jack Dempsey *Rocio* octofasciata in Fall River, South Dakota

Jack Dempsey *Rocio octofasciata* are endemic to Central America and were introduced into the Fall River Basin, South Dakota. In this system, Jack Dempsey is naturalized, abundant, and may have negative effects on co-occurring native species, including Mountain Sucker Pantoseus jordani. Overwinter persistence of Jack Dempsey in Fall River is made possible by headwater thermal springs. Headwater to confluence surveys and a thermograph of Fall River were used to assess the potential for range expansion and current distribution within Fall River. Relative abundance was seasonally assessed at eight sites throughout the basin. A thermograph was measured with temperature loggers at five sites to predict the ability of Jack Dempsey to overwinter in the Cheyenne River basin under present conditions, in conjunction with a lab based critical thermal minimum test. Jack Dempsey is primarily an insectivore in its native range, but no dietary information exists for the South Dakota population. Jack Dempsey diet surveys assessed the potential for niche overlap with native fishes and salmonids. Interseasonal diets were assessed by collecting fish seasonally in 2014/2015 and sorted and enumerated in the lab. Spring (N=50), summer (N=50), fall (N=50), winter (N=50) diets included 17 invertebrate orders and fish scales. Results from this study will provide biologists with an empirical assessment of the potential risk associated with Jack Dempsey in the Fall River Basin.

Harriger, Kate, John Knight, and Matt Wegener. Florida Fish and Wildlife Conservation Commission.

# Population Status of Harlequin Darters (*Etheostoma histrio*) in the Escambia River Watershed, Florida

Harlequin Darters (*Etheostoma histrio*) have been considered imperiled in Florida since 1977 due to a lack of knowledge about their population status and restricted range. A Biological Status Review determined that they should remain a species of special concern until more population demographics data is collected. In response, the Florida Fish and Wildlife Conservation Commission released a Species Action Plan (SAP) for Harlequin Darters with the objective to determine their population status. To address the objective of the SAP, our goal is to determine site abundance of Harlequin Darters, determine site-level darter density-habitat relationships, and use this information to extrapolate darter abundance to a stream-wide level. This work began in 2014 and is ongoing in Big Escambia Creek and Pine Barren Creek (tributaries to Escambia River). Site abundance of darters will be estimated for both creeks using mark-recapture and visual snorkeling techniques in 25-m stream reaches (sites). Two snorkelers will survey sites using small dip nets to capture darters. All captured darters will be marked with Visible Implant Elastomer paint during each sampling occasion. Each site will be sampled at least twice. Site abundance of darters will be calculated using a closed capture model in program MARK. Side scan sonar technology will be used to capture images of in-stream habitat (i.e. wood) throughout both creeks so that different habitat types (specifically percent composition of wood) can be quantified and delineated. Site-level darter density-habitat relationships will be determined for each creek using multiple regression analysis, and this information will be used to predict darter densities at unsampled sites and extrapolate dater abundance to a stream-wide level for both creeks. A final population assessment for each creek is expected by the end of 2016.

Harrington<sup>1</sup>, Richard C., Ethan France<sup>1</sup>, Matthew R. Thomas<sup>2</sup>, and Thomas J. Near<sup>1</sup>. <sup>1</sup>Yale University, Department of Ecology and Evolutionary Biology, Peabody Museum of Natural History; <sup>2</sup>Kentucky Department of Fish and Wildlife Resources, Fisheries Division.

# Molecular and Morphological Data Reveal Multiple Species Masquerading as *Etheostoma* kennicotti

The Stripetail Darter, *Etheostoma kennicotti* (Putnam), is distributed in a circle around the Nashville Basin in the Ohio, Green, Tennessee, and Cumberland River systems. There are two available names, but *Etheostoma cumberlandicum* Jordan and Swain is considered a synonym of *E. kennicotti*. Morphological differences discovered in a previous analysis were interpreted as clinal geographic variation, and *E. kennicotti* was treated as a single taxonomic unit. We collected DNA sequences from the mtDNA cytochrome b gene and two nuclear genes from 164 specimens of *E. kennicotti* sampled throughout the species' geographic range. In addition, we scored caudal pigmentation and collected meristic data from approximately 250 specimens. The phylogenetic analyses of the molecular data resolve populations of *E. kennicotti* into six major lineages that correspond to the Green River, Clarks and Ohio Rivers, lower Tennessee River system, upper Tennessee River system, the Cumberland River system that includes the Big South Fork, and the Laurel River system. The molecular phylogeny and morphological data support the recognition of *E. cumberlandicum* as a distinct species that is delimited to include all populations in the Cumberland River system exclusive of the Laurel River. We present analyses of morphological data for the five other species delimited in the molecular phylogenies.

Hart, Pamela B., and Jonathan W. Armbruster. Auburn University.

# Shape Variation among Lineages of the Southern Cavefish Species Complex in the Southeastern U.S.

The Southern Cavefish, *Typhlichthys subterraneus* Girard 1859, is one of the most fascinating stygobionts of the Amblyopsidae because it is a species complex (i.e., encompassing multiple species). Previous molecular analysis suggests the presence of 10 distinct lineages in the southeastern United States. Shape variation for this group had not been quantified previous to this study. We studied the morphological variation within the southeastern populations of the Southern Cavefish using geometric morphometric analysis. Our objective was to determine if there was a method able to morphologically distinguish the lineages. Subsequently, we examined if the morphological variation corresponded to the putative lineages. We applied the variable

reduction technique Principal Component Analysis to the geometric morphometric shape data. We then utilized Discriminant Function Analysis to determine if the lineages can be partitioned according to their shape. Many of the lineages were separated by shape; however, some lineages had considerable overlap. We will use the GM data to provide insight into characters that can be used to revise the taxonomy of the species complex.

Hayes<sup>1</sup>, Malorie M., Erika R. Krahl<sup>1,2</sup>, David C. Werneke<sup>1</sup>, and Jonathan W. Armbruster<sup>1</sup>. <sup>1</sup>Auburn University; <sup>2</sup>Southern Illinois University.

# Conservation Genetics of the Broadstripe Shiner, *Pteronotropis euryzonus*, an Endemic Species of the Middle Chattahoochee River

The Broadstripe Shiner, Pteronotropis euryzonus, is a freshwater minnow endemic to a small area that includes the tributaries of the middle and lower Chattahoochee River in Alabama and Georgia. Multiple hypotheses exist regarding structure and connectivity between the populations of P. euryzonus. The populations appear fragmented because individuals have not been detected in the main channel, suggesting limited dispersal potential and low gene flow between populations. In our study we found twenty-three sites in eleven tributaries which held populations of P. euryzonus. A total of 125 samples of P. euryzonus were collected for genetic analysis, and museum specimens were used for morphometric analyses. The mitochondrial genes cytochrome c oxidase I (COI) and cytochrome b (cytb) were used to assess genetic structure of P. euryzonus throughout its range. Geometric morphometrics were used to quantify shape variation among populations. Results suggest the presence of three genetically distinct populations: Northern, Pataula, and Southern, which also exhibit distinct morphologies. These populations support previous hypotheses of multiple forms of P. euryzonus in the Chattahoochee River. The presence of three genetically and morphologically distinct populations has significant conservation implications for *P. euryzonus*, which is currently listed as imperiled in both Alabama and Georgia. Three distinct populations, a restricted range, and recent disturbance to gene flow from local infrastructure may necessitate further protections to prevent extirpation.

Kaeser, Adam. U.S. Fish and Wildlife Service.

### From Mussels to Sturgeon, Side Scan Sonar Helps Advance the Conservation Mission

With access to low-cost, side imaging sonar and the development of tools and techniques for processing and analyzing data within a GIS framework, the opportunity to investigate and develop applications that address pressing conservation needs in navigable aquatic systems has never been greater. Across the Florida Panhandle the U.S. Fish and Wildlife Service is investigating the use of side scan sonar to detect and enumerate large sturgeons as an alternative approach for monitoring long-term trends in abundance as the species recovers. Within a meandering portion of the Apalachicola River, we have demonstrated the use of this technology to define suitable mussel habitat and assess changes in habitat over time, and have used easily

derived habitat metrics to model the distribution and abundance of an endangered species of mussel. In these cases, side scan sonar provided highly detailed, meso-scale level information about the subsurface environment; such information is critical to advancing our aquatic conservation mission in the 21st century.

Lewis, Mattie C., and Mollie F. Cashner. Austin Peay State University.

# Role of Nuptial Coloration in Interspecific Communication: Examining Color Vision in a Nest Host, the Creek Chub

Color vision plays a critical role in communication among fishes. In intraspecific mating interactions, bright hues are often seen in spawning males and serve to attract females or play a role in competition for mates. Nest association, the interaction in which one species utilizes the nest of another species to spawn, is common among minnows (Cyprinidae). Variation in host species likely influences interactions among nest associates. Pit-ridge builder Semotilus atromaculatus, the Creek Chub, is extremely territorial and males often defend nests by chasing away conspecific and heterospecific males. While the Creek Chub is defensive, there are some minnow species that it tolerates as nest associates, such as the federally protected Blackside Dace. All documented male nest associates of the Creek Chub exhibit a bright red hue during the spawning season. While this red coloration is likely important during intraspecific mating interactions, it could also play a role in intraspecific communication between nest host and associates. In order to begin to assess the possible role red vision may play in Creek Chub behavior, it is important to establish that they are capable of seeing red. Using classical conditioning, groups of three individuals were trained to identify red, orange, green, and blue. Each group was then tested to see if they could recall and recognize each color, and movements into target area were recorded. Data were analyzed with a one-way ANOVA. Red-trained fish responded to the red stimulus and did not respond to the other colors. These data will be compared to trials in which the fish were trained with orange and green. This is the first line of evidence that the Creek Chub is capable of seeing red, and knowing what colors the Creek Chub can see and differentiate between will give more insight into the complex behavior of nest association.

MacGuigan, Daniel J., and Thomas J. Near. Yale University.

### Phylogenomics of the Darter Clade Boleosoma

As the cost of DNA sequencing declines, we are increasingly able to generate genomic data for non-model organisms. Such large molecular datasets allow us to not only resolve contentious deep phylogenetic relationships, but also provide the detailed information necessary to study young species at a population level. Using RAD sequencing, we generated genomic data to investigate several taxonomic and systematic questions within the darter clade *Boleosoma*. This clade has posed significant challenges to ichthyologists, with substantial debate regarding

species-level diversity. Initial molecular phylogenetic hypotheses have relied on only a few loci, often with conflict amongst gene trees. By taking a genomic approach, we can increase our power to resolve the phylogeny of *Boleosoma*, investigate fine-scale genetic structure, and look for signatures of hybridization. Our preliminary data support several previously described relationships, such as the sister relationship between the morphologically similar Riverweed Darter (*Etheostoma podostemone*) and the Longfin Darter (*E. longimanum*), as well as the nested phylogenetic position of the Cumberland Darter (*E. susane*) within the Johnny Darter (*E. nigrum*). Additionally, our data support the hypothesis that the widespread Tessellated Darter (*E. olmstedi*) is actually composed of at least two species. Finally, our analyses offer insight into the controversial species status of the Waccamaw Darter (*E. perlongum*), suggesting little genetic divergence from the most geographically proximate populations of the Tessellated Darter (*E. olmstedi*).

### Mayden, Richard L. Saint Louis University, Department of Biology.

### The Making of the Fishes of Alabama

The making of the Fishes of Alabama was a long-held dream of the late Herbert T. Boschung. "Bo" retired early in 1987 to finish this book during his retirement years, after decades of study and sampling of both marine and freshwater fishes in Alabama. He dreamed of a book that pulled from the best qualities of all of the other "State" books at the time and succeeded in that effort. The final version of the book includes considerable general chapters at the beginning, an excellent, occasionally illustrated, index, and an outstanding compendium of references. The text is beautifully blended with outstanding illustrations by Joseph R. Tomelleri (Americanfishes.com) and pencil illustrations by several artists for the taxonomic keys. The book was published in 2004, and has continued to receive outstanding reviewers, and is said to have set a new mark as to standards for forthcoming state and regional books on fishes. Fishes of Alabama may not be considered a "field guide" or that easy to use in the field, but then what "State" book really is? Fishes of Alabama is an outstanding contribution to the field and should be on the desks of every ichthyologist, fisheries biologist, and anyone working with freshwater fishes. In fact, everyone should have a copy! From 1987 to the galley proofs the project included the help of many fantastic people and many great field experiences. Working mostly with Bo, Bernie, my students, and curators of many collections, I was honored to be part of the experience and a coauthor. Great thanks to all of these people for their efforts. Bo would want to give a special thanks to Joe (Tomelleri) for many great years of experiences and for leveraging the entire art collection for financial benefit to lower the price of the book for everyone!

*Meade*<sup>1</sup>, Mark, Linday White<sup>2</sup>, and Rahim Zettili<sup>1</sup>. <sup>1</sup>Jacksonville State University, Department of Biology; <sup>2</sup>Auburn University, School of Fisheries and Allied Aquaculture.

# Competition between Asiatic Weatherfish, *Misgurnus anguillicaudatus*, and Native Southeastern Fish Species: Biotic and Abiotic Factors

The Asiatic Weatherfish, *Misgurnus anguillicaudatus*, was first observed in the southeastern U.S. in the 1980s in Florida. Since that time the species has been observed in North Carolina, Tennessee, and Louisiana. Asiatic Weatherfish were first reported in Alabama in 2009. Based on recent survey data, Alabama populations are established in Logan Martin Lake (Pell City, AL) and appear to be migrating from the lake into nearby stream systems. Whether or not the species is injurious and competes with native species has been debated but no clear data is available and any assumptions are conjecture at time. Because the Asiatic Weatherfish has been reported coinhabiting regions with sensitive native species, such as the Coldwater Darter, Etheostoma ditrema, and the Banded Pygmy Sunfish, Elassoma zonatum, concern exists regarding potential competition. We examined abiotic competition between natives and Asiatic Weatherfish by determining changes in metabolism associated with changes in environmental temperatures. Asiatic Weatherfish were more tolerant to changing environmental temperature and likely can maintain growth and reproduction in conditions limiting to natives. In preliminary studies examining co-habitation and direct interaction with natives, Asiatic Weatherfish were the less tolerant species. When food supplies were limited, Asiatic Weatherfish were preved upon by natives and their numbers reduced substantially in a confined environment. In current studies we are examining adaptation of Asiatic Weatherfish to extreme temperatures. Further, because it has been suggested that U.S. populations of Asiatic Weatherfish are not of the same origin, we are making similar comparisons between natives and the exotic from other U.S. geographical regions.

### Mehrhoff, Loyal A. Center for Biological Diversity.

### **Recovery of Southeastern Aquatic Diversity**

The southeastern United States is considered a hotspot for aquatic biological diversity. We are exploring how to enhance conservation efforts aimed at the species in this hotspot and across the country. The Center for Biological Diversity has been cataloging population data on the 1,500 plus species that have been listed under the Endangered Species Act. We currently have 322 species, from across the nation, with trend data indicating an increase, decrease or stability in numbers. A review of this data shows that 61% of species with known trend data are increasing in number, 15% are stable and 24% are decreasing. For the 60 fish with trend data, 62% were increasing, 17% stable and 22% decreasing. Only eight fish from the southeast are currently in our database of trend data, this very small sample shows 4 species are increasing (50%), 1 is stable (13%) and 3 are decreasing (38%). We are continuing work to obtain trend data for more species, including southeastern fish, to identify where conservation is having success. Preliminary results suggest the Endangered Species Act is successfully saving species from extinction and putting them on the road to recovery.

Near, Thomas J., and Daniel J. MacGuigan. Yale University, Department of Ecology and Evolutionary Biology, Peabody Museum of Natural History.

### Next Generation Sequencing and the Phylogenetics of Darters

The resolution of darter phylogeny has increased dramatically over the past 15 years due to the use of molecular data. The emerging molecular phylogeny has aided in understanding the mechanisms of evolutionary diversification in darters and has resulted in a proposed change to their taxonomy. While the recommended changes to darter taxonomy are not universally accepted, they remain unchallenged with phylogenetic analysis of alternative datasets. In this talk we present phylogenetic analyses of a DNA sequence dataset comprising more than 3 million nucleotide sites sampled from 33,442 loci. The dataset was generated using restriction siteassociated DNA sequencing, RADSeq. The alignment contained data sampled from 142 specimens of more than 125 darter species. Perca flavescens and Sander vitreus were included as outgroup species to root the inferred phylogeny. Maximum likelihood analysis of this dataset, which includes 613,687 parsimony informative characters, presents an entirely resolved phylogeny of darters with very strong node support. Relationships among the major darter lineages include a clade consisting of Carnipellucida and Percina, which is resolved as the sister lineage of a clade containing Allohistium, Nothonotus, and Etheostoma. Analyses of individual loci indicate that Allohistium may have originated from hybridization between a lineage related to *Simoperca* and one that resolves outside of *Etheostoma*. Based on these phylogenetic analyses and previous morphological diagnoses, we recognize six genera of darters, Ammocrypta, Crystallaria, Percina, Allohistium, Nothonotus, and Etheostoma.

Neilson, Matthew E., and Pam Fuller. U.S. Geological Survey, Wetland and Aquatic Research Center, Nonindigenous Aquatic Species Database Program.

# Vectors of Aquatic Introductions in the Southeastern United States: Past and Present, Here and There

There are numerous vectors for introducing and spreading aquatic species, which may vary in importance and impact both spatially and temporally due to a variety of factors including species' biology, human behavior, demography, and geopolitical issues. Here we will examine spatial and temporal change in vectors of fish introductions in the southeastern United States, using data derived from the United States Geological Survey's Nonindigenous Aquatic Species Database. Most vectors show a general increase in number of species introduced over time. Sanctioned (i.e., stocking by natural resource agencies) releases, non-sanctioned (i.e., aquarium dumping) releases, and escape from captivity were the most important vectors. The relative importance of the vectors switched after 1950: more species were introduced via stocking before 1950 and more illegally-released species afterwards, with a sharp rise in captive escapes between 1923-1950 and 1951-1975. Vectors show differential geographic importance, with intentional

stocking the dominant pathway in all states except Florida, which has a high number of illegal releases. Governmental policy changes (e.g., altered natural resource management practices) and heightened awareness and education about impacts of introduced species may be driving a reduction in number of species introduced from 2001-present (relative to previous time period 1975-2000).

O'Donnell, Tim, Matt Walker, John Robinson, Mark Scott, Kevin Kubach, Brett Albanese, and Tanya Darden. South Carolina and Georgia Departments of Natural Resources.

### Development of an Edna Tool for Blackbanded Sunfish (Enneacanthus chaetodon)

Blackbanded Sunfish (Enneacanthus chaetodon) is a member of the North American centrarchid family 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 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 threatened, endangered, or invasive aquatic species because an organism can be detected by sampling its environment for DNA presence. We have developed a qPCR-based eDNA detection tool for Blackbanded Sunfish targeting a small fragment of the mitochondrial gene, cytochrome b. We have tested the eDNA tool for sensitivity and specificity in a series of laboratory experiments including qPCR with DNA isolations of Blackbanded Sunfish, several congeners, and other co-occurring species; aquaria with experimental densities of Blackbanded Sunfish ranging from 0-10 fish with and without co-occurring species; and at four known field sites where Blackbanded Sunfish were collected directly after taking water samples. All tests of the eDNA tool show that it is sensitive, specific, and effective at detecting Blackbanded Sunfish from environmental water samples.

Perkin<sup>1</sup>, Joshuah S., Thomas C. Boersig<sup>1</sup>, Amy E. Gebhard<sup>1</sup>, Lucas A. Hix<sup>1</sup>, Thomas C. Johnson<sup>1</sup>, and Natalie E. Knrop<sup>2</sup>. <sup>1</sup>Department of Biology, Tennessee Tech University; <sup>2</sup>School of Environmental Studies, Tennessee Technological University.

# Life History Theory Predicts Long-term Fish Assemblage Response to Impoundment Construction

Impoundment construction is a global ecological problem facing conservation and management of aquatic natural resources in lotic ecosystems. Myriad case studies document the long-term

effects of impoundments on stream fish assemblages, but there is a lack of theoretical guidance in terms of which species are predicted to respond negatively to impoundments. Life history theory has recently advanced as a framework for assessing mechanistic linkages between fish assemblage composition and flow regime gradients. Still, few studies have tracked multi-decadal trajectories of assemblage change to test life history predictions. We used fish collections from the Sabine River in Texas and Louisiana during 1950-2003 to assess how fishes employing different life history strategies responded to impoundment construction. Indicators of Hydrologic Alteration results included 18 of 33 flow regime metrics altered by impoundment construction, decreased coefficient of variation in flow (from 1.31 to 1.19), and increased consistency/predictability of flow (from 0.53 to 0.71). Canonical Analysis of Principal Coordinates revealed significant change (PMANOVA Psuedo  $F_{2,152} = 17.0, P < 0.01$ ) in taxonomic assemblage structure. Generalized estimation equations fitted to fish species probability of occurrence as a function of time revealed life history strategies (opportunist, periodic, and equilibrium) predicted taxonomic change. During the 53-year period, 68% of declining taxa were opportunistic strategists, 26% of declining taxa were periodic strategists, and 67% of increasing taxa were equilibrium strategists. Generalized Additive Models (GAMs) fitted to life history strategist richness as a function of time (expected change) were similar to GAMs fitted to strategist richness as a function of flow regime metrics (predicted change) based on overlapping 95% confidence intervals. Our study is among a few to apply life history theory to a multi-decadal trajectory of fish assemblage change and suggests predictions based on strategistsflow relationships are a useful framework for informing the science of environmental flows.

#### Pinder, Michael J., and Derek A. Wheaton. Virginia Department of Game and Inland Fisheries.

#### Past and Recent Efforts to Assess Blackbanded Sunfish Populations in Virginia

The Blackbanded Sunfish (Enneacanthus chaetodon) is a small (30-60 mm SL), deep-bodied centrarchid that ranges from the Coastal Plain of New Jersey to Florida. In Virginia, it is known in the Nottoway and Blackwater systems of the Chowan drainage where it inhabits densely vegetated swamps, beaver ponds, and small impoundments. The species is listed as endangered because of water pollution, hydrological alterations, vegetation removal, and introduction of exotic predators. Prior to 1999, four sunfish populations were known to exist in the Commonwealth. In 1999, we surveyed 123 sites in the Blackwater, Nottoway, James and Meherrin systems to assess species distribution and habitat use. We found three new populations, one in the Blackwater and two in the Nottoway drainages, bringing the total number of occupied sites to seven. Unfortunately, soon after their discovery, flooding by Hurricane Floyd resulted in dam failures at several known sites. Only two of the previous known locations had confirmed sunfish observations by 2013. Over the last two field seasons, we set forth to confirm sunfish presence at previously known sites, discover new locations, and determine sites for potential reintroduction. We used satellite imagery to categorize sites as excellent to poor based on the presence of vegetative cover, pond size and proximity to known sunfish populations. During the 2014-15 field seasons, we sampled 25 sites and documented five new and rediscovered one historic population. Today there are eight confirmed sunfish populations and seven potential

reintroduction sites. Future efforts will expand sampling at additional sites as well as monitoring known populations, conservation plan development and species reintroduction.

Rath, Mary, Kenny Jones, Lindsey Martin, Alexsis Mross, Starlene M. Loerch, Alexandra T. Hook, and Brook L. Fluker. Arkansas State University, Department of Biological Sciences.

# Saving the Endangered Dead: A Comparison of Rehydration Methods for Desiccated Preserved Fish Specimens

Natural history collections are valuable resources for studies in biodiversity, taxonomy, phylogenetics, ecology, and beyond. However, many institutions struggle to properly curate and maintain their collections due to budget constraints, curator turnover, and/or inadequate staff. Because of these challenges, nearly every collection has likely had to deal with desiccated fluidpreserved specimens. The Arkansas State University Museum of Zoology (ASUMZ) Ichthyology Collection is no exception. The collection houses approximately 13,000 specimen lots, but approximately 600 have experienced desiccation as a result of unsuitable storage containers. As a part of a larger effort to restore the ASUMZ Ichthyology Collection into a viable teaching and research resource, this project evaluated the efficacy of three specimen rehydration methods: (1) a surfactant-based method that used a 1% soap solution; (2) a previously published method that used distilled water in a high humidity chamber; and (3) a control method that involved a simple refill with 70% ethanol. The success of specimen rehydration was quantified as relative weight (grams/standard length) and as percent weight gain before and after rehydration procedures. Repeated measures ANOVA revealed significant differences in relative weight before and after rehydration (P < 0.0001); however, the three methods did not differ significantly from one another. One-way ANOVA yielded significant results for percent weight gain (P = 0.002): 102% for the soap method; 86% for the chamber method; and 83% for the control method. Pairwise comparisons indicated that the soap method differed from chamber (P = 0.014) and control (P = 0.003), but chamber and control methods were not different (P = 0.014) 1.000). Despite the success of each method, the surfactant-based method is preferred due to the rapid rate of rehydration. This project provides hope for saving desiccated fluid-preserved specimens that may otherwise be discarded and forever lost from the historical record.

Saco, Heather A., Ginny L. Adams, and S. Reid Adams. University of Central Arkansas.

# Changes in Land Use and Fish Assemblages in Three Ozark Highland Streams over Four Decades

Long-term studies evaluating land use and fish assemblage structure are often lacking but necessary to further understanding of anthropogenic influence on local fish assemblages. Land use is known to influence aquatic ecosystems, and alterations impact the biota. Sylamore, Jane's and Big creeks are in the White River drainage in north central Arkansas. During summers 2014 and 2015, we sampled fishes at 29 sites, with approximately 10 sites per creek. Sites, dates, and

sampling methodology were consistent with previous data collected during the 1970s from theses conducted at Arkansas State University. Morisita-Horn (M-H) similarity (0.72) and forest cover (83%) were highest in Sylamore Creek compared to Jane's (M-H 0.46, forest 72%) and Big (M-H 0.44, forest 45%) creeks. In Sylamore Creek, three species were found to have occurred in at least three sites in one collection (current or historic) but missing in the other. Both Moxostoma duquesnei and Cottus immaculatus were only found in historic collections, while Etheostoma blennioides was only found in the current collection. Four species showed this pattern in Jane's Creek including Gambusia affinis, Chrosomus erythrogaster and Micropterus salmoides in current collections and Ameiurus natalis in historic. Eleven species showed a distinct pattern in Big Creek, and several sensitive taxa (Hybopsis amblops, Noturus exilis, and Etheostoma autumnale) were found in current collections that were not present in historic. In Big Creek, there also appeared to be a complete replacement of Micropterus dolomieu by M. punctulatus and *M. salmoides*. Circumstantial evidence from local landowners suggests Jane's and Big creeks might be showing signs of recovery from historic nutrient enrichment from hog farms prior to the Clean Water Act; however, both systems may still be impacted by in-stream sedimentation. Fish metrics (including species richness, percent tolerant species, and percent intolerant species) in relation to land use composition will be discussed.

Schilling, Jennifer, Sam Borstein, Joel Corush, and Ben Keck. Ecology and Evolutionary Biology, University of Tennessee, Knoxville.

# Genetic and Morphological Patterns of Hybridization between *Nothonotus camurus* and *Nothonotus rufilineatus* in the Emory River, Tennessee

Hybridization and introgression are well documented in Darters. Over 25% of species hybridize with at least one other species, and over 12% of species have at least some genetic components from another species. These processes occur across the Darter Tree of Life and to varying degrees in different species pairs or specific drainage systems. In the Emory River system of Tennessee *Nothonotus camurus* and *Nothonotus rufilineatus* are hybridizing, and based on museum and TVA data they have been for at least several decades. *Nothonotus camurus* and *N. rufilineatus* are only sympatric in the lower Emory River, because *N. camurus* is restricted to that portion while *N. rufilineatus* occurs throughout most of the drainage. We are using mtDNA as a preliminary assessment to determine if introgression can be detected outside the area of sympatry. Additionally, we are quantitatively comparing the morphology of *N. rufilineatus* from populations that are sympatric vs. non-sympatric with *N. camurus*.

### Examining Isolation by Distance and Isolation by Environment for Six Headwater and Small Stream Resident Fishes within Six Drainages in Mississippi and Arkansas

Genetic Isolation by Distance (IBD) between populations is a common pattern across many different taxa. Genetic Isolation by Environment (IBE) between populations can also occur when they are separated by complete or incomplete environmental barriers to gene flow. Fish within dendritic river networks represent good systems for studying these patterns as they are mostly limited to movement and gene flow along network pathways. An important ecological gradient that affects species distribution within river networks is the stream size gradient, as summarized in the River Continuum Concept. Species that are adapted to reside in headwaters and small streams along this gradient theoretically have an increased potential for genetically isolated populations due to the hierarchical arrangement of dendritic networks. Network pathways connecting populations may cross larger streams which contain ecological conditions that are suboptimal or are outside of the ecological niche of these species, which has the potential to promote IBE. To examine this pattern, a comparative approach was used with 6 small stream and headwater resident fishes sampled across 6 river drainages (Pascagoula (PG), Pearl (PR), Bayou Pierre (BP), and Big Black (BB) in Mississippi, and Black (BL) and Little Red (LR) in Arkansas). A total of 2160 fish (144 populations consisting of 15 individuals per population) were caught, fin-clipped, and genotyped using 8 microsatellite markers. The species studied were Fundulus olivaceus, Semotilus atromaculatus, Erimyzon oblongus, Etheostoma parvipinne (PG and PR only), Etheostoma artesiae (BP and BB only), and Etheostoma whipplei (BL and LR only). Within each drainage, pairwise genetic, geographic, and ecological distance matrices were generated. These matrices were then combined for each species and a Multiple Matrix Regression with Randomization was performed to examine patterns of IBD and IBE. Results varied among the species, with some species having a higher influence of environment on genetic distance than others.

Stiles, Warren, and Carol Johnston. Auburn University.

### Effects of Hydrologic Change on Stream Fish Assemblages in Alabama

Past studies within single watersheds in the Southeast have found that unnatural changes in stream hydrology affect the fish assemblage negatively. With increases in land development and water withdrawals, significant declines in discharge have been seen in some Southeastern streams over the past 50 years. In this study, contemporary fish assemblage data collected in the summers of 2013 and 2014 were compared to historic collections in 6 unregulated watersheds across the physiographic regions of Alabama. Environmental variables such as hydrology and land cover were used to understand shifts in the assemblages. Shoal Creek in the Coosa River drainage is the least developed and most completely forested watershed in the study, and the assemblage in this piedmont stream showed the least interannual change based on Jaccard's and Morisita's similarity indices. Five Runs Creek, a slightly more developed tributary to the Yellow River on the lower coastal plain showed a significant relationship between changes in fish

assemblage and discharge over time with a positive relationship between discharge and time. Shoal Creek, a tributary to the Tennessee River in North Alabama located in the Highland Rim physiographic region also saw a significant relationship between hydrology, time, and assemblage; though in this watershed there was a negative relationship between discharge and time. Inclusion of more environmental variables will likely account for more variance in the data and strengthen the relationships seen in the data.

*Tan, Milton, and Jonathan W. Armbruster. Auburn University, Department of Biological Sciences.* 

### Minnows on the Edge: Incorporating Phylogeny into Conservation Biology

Phoxinin minnows are a diverse group of fishes distributed broadly in North America, and numbering over 300 species. Minnows face many threats, and some of these species are of great conservation concern in the Southeastern United States, such as the Slender Chub Erimystax cahni. With recent advances in phylogenetic inference of cypriniform fishes, we have generated an "All Cypriniformes Tree of Life" incorporating phylogenetic and phylogenomic data for over 2000 species of the order. Robust phylogenies can be utilized to inform conservation prioritization because they provide data on phylogenetic diversity and evolutionary distinctiveness. We calculated EDGE scores for phoxinin minnows, which incorporate phylogenetic data with IUCN Red List global rankings to assign extinction probabilities. These scores thus combine measures of evolutionary distinctiveness and global endangerment (EDGE) to allow for prioritization of conservation efforts. Of the species found in the Southeastern United States, numerous species of endangered and threatened minnows have EDGE scores within the range of critically endangered species of minnows, demonstrating that these species represent important evolutionary lineages despite a lower extinction risk. In addition, within each IUCN Red List ranking we found variation in EDGE scores due to evolutionary distinctiveness. For example *Chrosomus saylori* and *C. cumberlandensis* were both found to have higher EDGE scores than *Erimystax cahni*. Although all three of these species are listed as endangered in the IUCN Red List, Chrosomus species have greater evolutionary distinctiveness. By using a metric that incorporates phylogeny, information can be provided to prioritize conservation efforts towards species that represent evolutionarily distinct components of fish fauna.

#### Search for Blackbanded Sunfish, Enneacanthus chaetodon, Populations in Florida

The Blackbanded Sunfish, *Enneacanthus chaetodon*, is found from New Jersey to Central Florida, but is fragmented throughout the distribution and is considered rare or at risk throughout much of the range. The first collection in Florida was made at Mill Dam Lake in Marion County in 1936. It has since been collected at 16 additional systems from Central to Northern Florida,

Tuten, Travis, and Eric Nagid. Florida Fish and Wildlife Conservation Commission, Gainesville, Florida.

with most of these locations in or around the Ocala National Forest. Almost all of the Florida collections were made between 40 and 70 years ago, and more recent sampling trips to these sites where biologist targeted Blackbanded Sunfish had unsuccessful results. There are currently only two known populations of Blackbanded Sunfish in Florida and the State considers the species as rare and biologically vulnerable, but does not include them as a state listed taxa. Criteria that Florida uses to determine if species should be included as a state listed taxa include measures such as population size, population size reduction, limited geographical range (extent of occurrence or area of occupancy), and number of locations occupied. We are trying to identify current populations of Blackbanded Sunfish in Florida to better inform whether the species should be state listed. Our efforts are focused at historic collection sites and at some areas and waterbodies that have been sampled little or never before. Aerial imagery and museum records of Blackbanded Sunfish along with other fish species have been used to select potential sampling sites and areas. We experienced one early success identifying a new population, but have also had multiple unsuccessful sampling events including some at historic locations. Aerial imagery has also shown significant changes in the aquatic habitat availability at some of the historic collections sites.

Walker, Daniel J., and J. Brian Alford. The University of Tennessee, Knoxville, Department of Forestry, Wildlife, and Fisheries.

### Using Side Scan Sonar and GIS to Assess Spawning Habitat for Lake Sturgeon, *Acipenser fulvescens*, below TVA Dams on the Upper Tennessee River

The Lake Sturgeon (Acipenser fulvescens) was extirpated from the Tennessee River system in the 1960s. The species is now undergoing extensive restoration efforts across the Southeastern U.S. A milestone for the restoration effort is the occurrence of natural spawning, and a major hurdle potentially facing any natural spawning efforts by Lake Sturgeon in the Tennessee River is the fragmentation of the river system by large hydroelectric dams. However, Lake Sturgeon in other systems have been found to successfully spawn in the tailwaters of dams, when there has been sufficient environmental conditions and population sizes. Lake Sturgeon are lithophilic spawners, so one factor influencing the success of natural Lake Sturgeon spawning is the availability of coarse rocky substrate at the terminus of a migration. To assess the suitability of four Tennessee Valley Authority hydroelectric dam tailwaters on the Upper Tennessee River for future Lake Sturgeon spawning events, we utilized side-scan sonar and underwater imaging to collect data on the composition of the substrate. We then utilized GIS to categorize the substrate composition in 2-km reaches at each of the dam tailwaters with several methods. First, we performed "heads-up" digitization to classify the substrate depicted in the sonar imagery. We then randomly partitioned the real substrate imagery into training and validation sets. We used a training partition to perform maximum-likelihood estimation image classification of the sonar imagery, and another training dataset partition to perform inverse-distance weighted interpolation of the distribution of the substrate. Finally, we tested the accuracy of each of the three methods with a validation partition of the real substrate imagery, and compared the suitability of each of the four tailwaters based on the availability of good spawning habitat for

Lake Sturgeon. Our results may identify the need for artificial spawning reefs to support the ongoing restoration of the species.

Wellemeyer<sup>1</sup>, C. Juju, Joshuah S. Perkin<sup>1</sup>, Mary Liz Jameson<sup>2</sup>, and Katie H. Costigan<sup>3</sup>. <sup>1</sup>Department of Biology, Tennessee Tech University; <sup>2</sup>Department of Biological Sciences, Wichita State University; <sup>3</sup>School of Geosciences, University of Louisiana at Lafayette.

# Synthesizing Multiscale Processes Regulating Persistence of Arkansas Darter, *Etheostoma cragini*

Arkansas Darter, *Etheostoma cragini*, is listed as threatened by the Southeastern Fishes Council. Threats to the species include shrinking distributional range because of dewatering, destruction and fragmentation of habitat, invasion by exotic species, water pollution, and isolation of small populations. We studied multi-scale processes that regulate persistence of Arkansas Darter in Smoots Creek, Kansas, at mesohabitat  $(10^2 \text{ m})$ , reach  $(10^3 \text{ m})$ , segment  $(10^4 \text{ m})$ , and basin  $(10^5 \text{ m})$ m) scales to inform conservation and management of the species. We measured: (1) density  $(fish/m^2)$  of Arkansas Darter within mesohabitats during three summer months; (2) density near and away from stream confluences at the reach scale during two years; (3) persistence at sampling sites with varying hydrology during five years; and (4) occurrence in stream segments connected and disconnected to regional alluvial aquifers during 50 years. Analyses indicated Arkansas Darter density was not related to mesohabitat variables but was positively associated with water persistence at the mesohabitat scale. Arkansas Darter density was positively correlated with stream confluences at the reach scale and annual flow variability that promoted growth and reproduction at the segment scale. At the basin scale, occurrence was greatest among stream segments connected to groundwater outflows. Conservation approaches for Arkansas Darter might include, but are not limited to, maintaining sufficient surface flows to avoid stream desiccation among mesohabitats, preserving longitudinal stream connectivity along reaches, retaining or restoring natural flow regimes throughout segments, and sustaining hydrologic connectivity to groundwater sources throughout basins.

Wheeler<sup>1</sup>, Kit, Mary C. Freeman<sup>2</sup>, and Seth J. Wenger<sup>1</sup>. <sup>1</sup>River Basin Center and Odum School of Ecology, University of Georgia, Athens, Georgia; <sup>2</sup>U.S. Geological Survey Patuxent Wildlife Research Center, Athens, Georgia.

# **Rates and States in Flow Ecology: What Are They, How Are They Different, and Why Does It Matter?**

In most southeastern rivers, human modification (dams, withdrawals) of surface and ground water resources has extensively altered temporal discharge patterns. Throughout the region, these alterations now present substantial challenges to the long-term persistence of native fishes with evolutionary histories tied to fundamentally different hydrologic regimes. In response to these conservation concerns–and in the face of changing climatic conditions that threaten to exacerbate

patterns of water availability-there have been numerous attempts to develop quantitative predictive relationships (frequently called flow-ecology relationships) describing ecological responses of fishes and other aquatic biota to flow alterations. Attempts to generate flow-ecology relationships often fall into one of two approaches: (1) an ecological states approach that constructs flow-ecology relationships using time-averaged hydrologic conditions and ecological outcomes representing a single point in time, or (2) an ecological rates approach that relies on temporally coupled hydrologic conditions and ecological outcomes, both of which are collected over multiple time intervals. Here, we identify some key differences between rates and states approaches in flow ecology and advocate for increased use of rates approaches on the basis of two primary advantages they offer. Unlike approaches focused on ecological states, rates approaches permit evaluation and estimation of hypothesized links (either implicit or explicit) between ecological outcomes, flow characteristics, and underlying demographic processes, thereby deepening mechanistic understanding of flow-ecology relationships. Additionally, rates approaches enable generation of testable, temporally explicit predictions of ecological outcomes to flow alterations, which facilitate evaluation of alternative management and conservation scenarios. Ultimately, we believe a more nuanced understanding of flow-ecology relationships and the approaches used to generate them can help expedite successful application of scientifically-driven conclusions to complex issues in water resource management and freshwater biodiversity conservation.

Williams, Jim. Florida Museum of Natural History, Gainesville, Florida.

### Pleistocene River Drainages of Florida: Evidence from Fossil Fish and Mussels

During the Pleistocene, a span of 2.6 million years, the four glacial and three interglacial cycles drastically altered the landscape of Florida. During the most recent glacial maximum, about 18,000 years ago, the approximately 300 m drop in sea level exposed a vast land area off the west coast increasing the peninsular region to about twice its present width. The expanse of land off the east coast was far more limited especially in the southern portion of the peninsula. Examination of Pleistocene fossil freshwater fishes and mussels reveals the presence of a river drainage that connected Florida streams as far west as the Apalachicola and Ochlockonee drainages with the southern end of the peninsula. The presence of a large interconnected river drainage in peninsular Florida provided a dispersal route for aquatic organisms extending from the panhandle to the southern tip of the peninsula during the Pleistocene.

Zimmerman, Brian J., Marc R. Kibbey, and Marymegan Daly. Ohio State University.

### Fishes of Ohio Inventory and Distribution Project

Milton Trautman's classic book, "The Fishes of Ohio," was published in 1981 and did an excellent job presenting the distribution and status of Ohio's fish fauna at the time. In subsequent decades, fish communities of Ohio have changed in composition and distribution. In 2011, we

began an inventory of the current status of all fish species found in Ohio. Some of these changes we have documented are positive, including the large scale expansion of many species of riverine fish that have been characterized as sensitive to water quality. Other changes point towards declines, particularly in species reliant on wetland or glacial lake habitats. In addition to trends in distribution and abundance of native species, we see significant impact in the occurrence of non-indigenous species that were not documented by Trautman.

### **Poster Abstracts**

(alphabetical by first author's last name)

Alford, Brian, Joyce Coombs, Justin Wolbert, and Hayley Gotwald. The University of Tennessee-Knoxville.

# Fish Assemblages as Indicators of Agricultural Land Use Impacts in the Nolichucky River Watershed of Tennessee

We assessed the extent of current land use intensity (% of watershed area) and change from 2000 to 2015 in the Nolichucky River watershed and how spatiotemporal variability of land use influenced fish assemblages. During summer 2014-2015, we sampled fishes using TVA backpack electrofishing methods at riffle-run habitats in different stream size categories (5 tributary sites and 11 main stem sites). Site classification was based on levels of observed riparian land use impacts (quartiles from U.S. EPA rapid habitat assessment scores) and included least-impacted (n=7), moderately-impacted (n=4), and most-impacted (n=5) sites. Canonical correspondence analysis and SumF tests (F > 420; P < 0.01) revealed that fish assemblages were different based on site impairment classifications (344 samples, 50 species, 8,610 fish), and that elevation and temperature correlated the strongest with assemblage structure (CCA axis 1), followed by % current forest land cover, total dissolved solids, and specific conductivity (CCA axis 2). Results of blocked indicator species analysis suggested that *Notropis rubricroceus* was a strong indicator of the least-impacted condition (indicator values  $_{IV} = 35$ ; P < 0.01). Nothonotus *acuticeps* was a strong indicator of the moderately-impacted condition (IV = 32; P < 0.01), while Cottus carolinae, Etheostoma zonale, and E. simoterum were indicative of the most-impacted condition (IV = 26-29; P < 0.02). Additionally, abundances of fish with relatively more "equilibrium" functional and life history traits were indicative of least-impacted sites, while those with more "opportunistic" traits were indicative of most-impacted sites. Agricultural land use had stronger influences on tributary fish assemblages rather than on main stem assemblages, especially since N. rubricrosceus was never captured at most-impacted or moderately-impacted tributary sites. Fishes with traits such as cavity-spawning and lithophilic egg laying strategies, preferences for pools and aquatic vegetation were impacted most by agriculture, likely as a result of greater suspended sediments.

Andreoli, Joseph A. The University of Florida Department of Geography.

### Predicting the Geographic Ranges of Non-native Cichlids in Florida with Climate Change

Invasive species and climate change are two of the most pressing issues facing Florida today. The state is a hotspot for non-native fish introductions, including cichlids. Cichlids are a popular group of fish in aquaculture, and among anglers and aquarists, with many species having established populations in Florida. These species cause various environmental and socioeconomic impacts to the state. This study correlates the georeferenced presence points of six different cichlid species in Florida and the current bioclimatic (BIOCLIM) and hydrologic (HYDRO1K) variables at those sites using maximum entropy modeling (MAXENT), in a species distribution modeling (SDM) framework. These relationships are then extrapolated to two different representative concentration pathways (RCPs) for the years 2050 and 2070. The resulting maps give us predictions to where in Florida suitable habitat for a given species exists at a fine resolution of 1 square-kilometer. The geographic ranges vary from species to species, with a general trend of expansion throughout the state in the future given climate change. As eradication is difficult once a species becomes established, these models have use in informing risk assessments for sister taxa. There is predictive power in uncovering what parameters drive non-native cichlid ranges in Florida. These findings can be expanded to what variables are important for aquatic non-native species establishment at global and regional scales. For regions supporting rich fish diversity and endemism like the Southeastern United States, managers may use these findings in prioritizing effort and limited resources in controlling those non-native species causing the most negative impacts.

Bedal, Brooke A., and Mollie F. Cashner. Austin Peay State University, Department of Biology.

# Cytochrome *b* Lineage Assessment of Blackside Dace (*Chrosomus cumberlandensis*) from the Kentucky River System

The Blackside Dace (*Chrosomus cumberlandensis*) is a federally threatened minnow endemic to the Cumberland River system in Tennessee and Kentucky. Due to extensive survey work, a small number of populations have recently been encountered in river systems not previously known to contain *C. cumberlandensis*. There are three possible explanations for these discoveries: (1) these populations have recently been established due to anthropogenic movement of individuals across river systems; (2) these populations are the result of recent natural dispersal of *C. cumberlandensis*; (3) these populations represent genetically unique, small populations which have been undetected but represent distinct lineages. Using previously published cytb sequences of all species in the genus and sequences generated during the course of this study for individuals from a population in the center of the range of *C. cumberlandensis* and the recently discovered population found in the Kentucky River system, we have begun to assess whether this population is a distinct lineage or if it represents a recent range expansion. Future work will include a complete examination of population genetics with microsatellite markers to further elucidate patterns of gene flow within the main range and among the satellite populations.

Benesh, Kasey, Milton Tan, and Jonathan Armbruster. Auburn University.

### Urbanization and Range Stability of Etheostoma tallapoosae and Etheostoma stigmaeum

Alabama is one of the top states in terms of diversity of freshwater fish species; unfortunately it is also placed as one of the top states it terms of at risk fish species. The *Tallapoosa Darter*, *Etheostoma tallapoosae*, is a particularly unique species endemic to the Tallapoosa River System. We investigate if *E. tallapoosae*, despite its endemicity, is as stable as a widespread, common congener like *E. stigmaeum*. Due to the limited range *E. tallapoosae* it is vulnerable to

anthropogenic stressors such as an increase in development and conversion of land. As urbanization is a major threat to Alabama fishes, we predicted that HUC10s (hydrological units) with a higher percent urbanized land cover will have a greater time since last occurrence date of *E. tallapoosae*. We investigated the impact of urbanization on the range stability of *E. tallapoosae* and *E. stigmaeum*. We used the data for these species to test if range stability was different between a species limited in its range, *E. tallapoosae*, and a species with a wider range, *E. stigmaeum*, and if they differed in their response to anthropogenic stressors. This information can inform future conservation efforts and areas where more sampling or other conservation efforts should take place.

Brandt<sup>1</sup>, Stephanie L., Matthew R. Thomas<sup>1</sup>, Patrick L. Rakes<sup>2</sup>, J.R. Shute<sup>2</sup>, Crystal L. Ruble<sup>2</sup>, and Melissa A. Petty<sup>2</sup>. <sup>1</sup>Kentucky Department of Fish and Wildlife Resources; <sup>2</sup>Conservation Fisheries, Inc.

### Captive Propagation and Reintroduction of the Cumberland Darter, *Etheostoma susanae*, in the Upper Cumberland River Drainage, Kentucky

The Cumberland Darter, Etheostoma susanae, is endemic to a limited portion of Cumberland River drainage above Cumberland Falls in KY and TN. The species was federally listed as Endangered in 2011 due to range curtailment and fragmentation from habitat loss and degradation. In 2008, the Kentucky Department of Fish and Wildlife partnered with Conservation Fisheries, Inc. to develop spawning protocols for the species and produce the offspring needed to re-establish extirpated populations within its historic range. We selected Cogur Fork, a 10 sq km tributary of Indian Creek (upper Cumberland River drainage), as a suitable stream for reintroduction because habitat conditions were suitable but it did not contain a pre-existing population of *E. susanae*. A total of 4,045 captive spawned individuals marked with visible implant elastomer (VIE) tags have been stocked in Cogur Fork since 2009. During 2010-2014, we conducted periodic surveys following each annual release of tagged fish within a 1 km section of lower Cogur Fork. Using a combination of visual observation and seine hauls we have confirmed the survival of tagged fish released into Cogur Fork for periods exceeding one year. One survey during November 2010 detected a large number of stocked individuals (N=50) and extensive upstream dispersal of many fish, one as far as 700 m. A small but steadily increasing number of untagged individuals captured since 2012 suggests that some natural reproduction is occurring. However, it would be premature at this point to suggest that the project has been successful in restoring a wild population. A small number of untagged individuals present could indicate the early establishment of a wild population in Cogur Fork, but collection of larger numbers of untagged fish over several years after stocking ceases is the benchmark needed to support any strong argument for successful establishment of a reproducing population.

Buford, Jessica, Brittany L. McCall, and Brook L. Fluker. Arkansas State University, Department of Biological Sciences.

# The Importance of Georeferencing Small Natural History Collections and Their Contributions to Large Online Databases

The Arkansas State University Museum of Zoology (ASUMZ) Ichthyology Collection houses approximately 13,000 specimen lots from 23 of the contiguous states. Until recently, ASUMZ specimen data were only stored in handwritten catalogs, making query and locality mapping difficult and time consuming. As a part of a larger effort to restore the ASUMZ Ichthyology Collection into a viable teaching and research resource, we digitized and georeferenced the ichthyology collection data. Here, we use the ASUMZ georeferenced database as a case study to highlight the importance of georeferenced specimen data from small collections and their potential to contribute valuable information to large online databases such as FishNet2. Using georeference data from selected species, we show that small collections may not contribute new information in terms of known species range boundaries, but they can provide a very high resolution picture of distributional patterns on smaller scales (e.g., regional, county, and/or drainage level) proximate to the institution. We conclude that digitization and georeferencing of small collections, each with their own unique high resolution footprint, will contribute vastly to our knowledge of species distributions and our ability to accurately characterize biodiversity at fine scales.

Elkins<sup>1</sup>, Duncan, Anna L. George<sup>2</sup>, Sarah C. Hazzard<sup>2</sup>, Bernard R. Kuhajda<sup>2</sup>, and Seth J. Wenger<sup>1</sup>. <sup>1</sup>University of Georgia, Odum School of Ecology, River Basin Center; <sup>2</sup>Tennessee Aquarium.

### The Southeastern Aquatic Biodiversity Conservation Strategy

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, and these species are highly endemic. 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, will draw from the many excellent existing species-, state-, and watershed-level plans to synthesize an integrated conservation strategy at a scale and resolution appropriate for regional conservation Strategy, a synthesis that identifies management actions and policy recommendations for areas with high capacities where additional monies can be leveraged quickly as well as areas with higher needs and lower capacity where a longer-term strategy is necessary. This strategy will serve as a guide for NFWF and other foundations in making

investments in southeastern aquatic conservation. Phase 1 of this project, including Southeastern drainages east of the Mississippi, is slated for completion in Summer 2016.

### Davis, Johnathan. Young Harris College, Department of Biology.

### Should Fisheries Conservationists and Biologists Adopt a More "Fish-friendly" Diet?

Study of aquatic ecosystems and imperiled fishes has revealed several actions within watersheds that alter aquatic habitats, reduce fish diversity and impact fish health. Many of the identified culprits are directly or indirectly related to raising livestock for human consumption. Examples include the intensive grazing of livestock near streams that increases sedimentation, nutrient loads and bacteria levels, contamination of streams with concentrated animal waste from feedlot operations, greater release of methane, a known greenhouse gas, water withdrawals for watering livestock, destruction of riparian habitat for cattle access and increased suspension of in-stream sediments. Whereas many management actions have been proposed to limit the impact of livestock operations, such as preventing livestock access to streams, I suggest that a previously unconsidered factor is the increased consumption of meat in the American diet. Adoption of sustainable eating habits that reduce the amount of meat in the diet and support locally-grown, organically-produced foods may lessen observed impacts from livestock operations. Additionally, expected increase in human population size in the southeast coupled with increased meat consumption could further stress aquatic systems. Thus, I suggest that individuals in support of fish conservation who have food choice should consider supporting a more sustainable personal diet such as a vegetarian or organic-based diet and endorsing recent food movements that emphasize sustainably-produced foods.

Hazzard<sup>1</sup>, Sarah, Bernie Kuhajda<sup>1</sup>, Anna George<sup>1</sup>, Joe Powell<sup>2</sup>, Mack Lunn<sup>2</sup>, Justin Medley<sup>2</sup>, Daniel Samples<sup>2</sup>, and Coco Bennett<sup>2</sup>. <sup>1</sup>Tennessee Aquarium Conservation Institute; <sup>2</sup>Tennessee Technological University iCube.

# Freshwater Information Network (FIN): A Collaborative Effort to Create a Freshwater Species Database for the Southeastern United States

The Southeastern United States is a global biodiversity hotspot for freshwater aquatic organisms. Unfortunately, the Southeast also harbors some of the most imperiled aquatic species in the world. Thus the need for collaborative conservation efforts for southeastern aquatic organisms is more important now than ever. However, distributional and status data for these organisms are stored in a variety of platforms, including spreadsheets, museum records and databases, unpublished reports, and field notes. This can be exceptionally problematic for endangered species where recent sightings cannot be vouchered and accessed by others. The Freshwater Information Network (FIN) aims to combine all of these data sources into a single user friendly "living" database containing georeferenced locations and voucher photographs. We collected museum, institutional, and field note data and georeferenced records using the Guide to Best

Practices for Georeferencing. These records were then categorized based on year (Pre-1980, 1980-1999, 2000-2012) or by data collection type (ex. Negative, Museum, and Field Identification data). Currently FIN includes 61 of the most imperiled species of fishes within the Tennessee, Mobile, and Cumberland river drainages. This interactive website allows scientists to submit new locality data and field photograph vouchers. While access levels currently only include scientists, future tiers will include watershed groups and resource managers, as well as citizens. We are working to add over 300 other fishes found in the Tennessee, Cumberland, and Mobile drainages. Future additions to FIN will include other aquatic taxonomic groups in these drainages. With new addition of data from experts, conservation efforts in the Southeast can become a focused collaborative effort.

Hubbell, Joshua. The University of Southern Mississippi, Department of Biological Sciences.

# Utilizing Species Distribution Models to Prioritize the Habitat of the Yazoo Darter, *Etheostoma raneyi*, a Headwater Specialist

The Yazoo Darter, *Etheostoma ranevi*, is a species of Snubnose Darter within the clade Adonia and is endemic to the upper Yazoo River basin within the Little Tallahatchie, Yocona, and South Tippah Rivers in north-central Mississippi. Many streams within the Little Tallahatchie River drainage either originate, partially flow through, or flow parallel to federal or state managed land, and are afforded some protection. Contrastingly, streams harboring Yazoo Darter populations in the Yocona River drainage all flow through private land, and thus these populations are more threatened. During the summer of 2015, we selected 50 sites to sample for Yazoo Darters within the Upper Yazoo River basin. Specifically, 15 sites were selected from the known range of the species, 20 sites from the eastern portion of the range, and 15 sites were located on tributaries either downstream or that flowed into impoundments within the Yocona and Little Tallahatchie River basins. We used historic data in conjunction with the recent survey data, and species distribution models (MaxEnt) to assess the current distribution and habitat use of the Yazoo Darter. We used MaxEnt software to distinguish landscape scale factors associated with sites yielding Yazoo Darters. All models were built using MaxEnt version 3.3.3k. MaxEnt uses species presence data to approximate species distributions. MaxEnt uses presence-only data as well as environmental predictors (e.g., drainage area, stream order) across a predefined landscape that is split into grid cells, the software program then extricates a sample of locations that it compares to the presence sites. We built a model relating environmental factors (cumulative drainage area, stream order, stream slope, soil type, and land cover) to Yazoo Darter occurrences. The Maxent species distribution model predicts suitable habitat located within currently occupied and unoccupied stream segments throughout the Upper Yazoo River basin. Specifically, Maxent delineated 2nd and 3rd order stream reaches that had a drainage area ranging from 0-500 square km, and which flowed over soil types (predominately various sands and clays) as the best suitable habitat for Yazoo Darters. Land cover varies across the three sampled regions. The known range possesses the least amount of land that has been subjected to anthropogenic impacts (35%) while also possessing the highest percentage of land that is either forested or protected wetlands. Contrastingly, watersheds in the eastern range and within the

extent of the reservoirs possesses higher percentages of land subjected to anthropogenic impacts (52%, 48%), and less forested land and protected wetlands.

Jones<sup>1</sup>, Kenny, Brook L. Fluker<sup>1</sup>, and Bernard R. Kuhajda<sup>2</sup>. <sup>1</sup>Arkansas State University, Department of Biological Sciences; <sup>2</sup>Tennessee Aquarium Conservation Institute.

# Conservation Genetics of the Blueface Darter (*Etheostoma* sp. cf. *zonistium*), a Rare Undescribed Fish in Northwest Alabama

Previous morphological work revealed that select populations of the Bandfin Darter (Etheostoma *zonistium*) in northwest Alabama represent a rare, undescribed fish species; the Blueface Darter (E. sp. cf. zonistium). With the description of the Blueface Darter forthcoming, its rarity, extremely small distribution, and fragmented landscape will make it a potential candidate for listing under the Endangered Species Act shortly following its formal description. The Blueface Darter has a peculiar distribution in small 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 of Blueface Darters in tributaries of the upper Bear Creek system is potentially restricted by the Upper Bear Creek Reservoir. This study used mitochondrial (mt) cytochrome b DNA sequence data and microsatellite loci to test multiple hypotheses involving patterns of genetic variation and population fragmentation within the Blueface Darter. Our preliminary results suggest three main conclusions. (1) Both mtDNA and microsatellite data corroborate previous morphological analyses supporting the validity of the Blueface Darter as a distinct species; however, Blueface Darters in Bear Creek show evidence for retained ancestral microsatellite alleles. (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). Future work will focus on incorporating more samples and microsatellite loci to further evaluate these conclusions and population-specific conservation needs for the Blueface Darter.

MacGuigan, Daniel J., and Thomas J. Near. Yale University.

# Molecular and Morphological Species Delimitation in the Greenthroat Darter, *Etheostoma lepidum*

Discovering and describing biodiversity is one of the primary goals of systematic biology. The Greenthroat Darter (*Etheostoma lepdium*) is distributed primarily in southern Texas, but with a disjunct population in southeastern New Mexico. The species exhibits variation in morphology and male nuptial coloration, which has been considered clinal in nature. We investigated phylogenetic structure among populations of *E. lepidum* using DNA sequences from a mtDNA

gene and 14 nuclear DNA loci. We also investigated variation in meristic and male nuptial coloration traits in the context of the inferred molecular phylogeny to determine if *E. lepidum* comprises multiple undescribed species. Based on patterns of morphological variation and results from widely used molecular species delimitation approaches such as bGMYC, BPP, and Bayes factor delimitation, we found that *E. lepidum* appears to contain three distinct evolutionary lineages: one in Pecos River in New Mexico, one in the upper reaches of the Colorado River in Texas, and one in the lower Colorado River, Guadalupe River, and Nueces River in Texas. When interpreted in a phylogenetic context, morphology and coloration both support recognition of three species and provide several diagnostic characteristics. This research effectively demonstrates that molecular and morphological data can be used in tandem to delimit and describe new species of darters. Additionally, our results suggest roles for headwater capture, aridification, and vicariance as drivers of allopatric speciation in the *E. lepidum* species complex.

*McCoin*<sup>1</sup>, *Danielle R., John W. Johansen*<sup>2,3</sup>, and *Rebecca E. Blanton*<sup>1,2</sup>. <sup>1</sup>*Austin Peay State University, Department of Biology;* <sup>2</sup>*Austin Peay State University, Center of Excellence for Field Biology;* <sup>3</sup>*Tennessee Tech University, Department of Environmental Sciences.* 

### A Test of Intraspecific Size-dependent Variation in Male Intromittent Organs in a New Species of Crayfish (Cambaridae: *Orconectes*) from Tennessee

Crayfishes (Cambaridae) are a diverse, yet imperiled and understudied group of aquatic invertebrates of the southeastern U.S. Males of all species alternate between sexually active (Form I) and sexually inactive (Form II) states after reaching sexual maturity. The two forms differ in intromittent organ (gonopod) characteristics, including shape and length. Form I male gonopod characters have been used to distinguish species, subgenera, and genera of crayfishes and to support hypotheses of evolutionary relationships. An undescribed species of Genus Orconectes (Orconectes sp. nov.) was discovered in 2012 from the Caney Fork River system (Cumberland R.) of middle Tennessee. Preliminary investigations revealed considerable intraspecific variation in Form I male gonopod morphology including the length, shape, and presence of a spine-like projection on the mesial process. The goal of this study was to determine if the observed intraspecific variation in Form I male gonopods was related to individual size in Orconectes sp. nov. Measurements of total length for Form I males from multiple populations revealed the presence of two size classes. Measurements, including overall gonopod length, length and shape of the mesial and cephalic processes, and the presence or absence of the spinelike projection, were made on 10 randomly selected individuals from each size class. Multivariate size-corrected PCA analyses are ongoing. Results showing size-dependent intraspecific variation would have implications for the continued utility of Form I gonopod characteristics in crayfish systematics. Reliance on such characters has been questioned due to possible convergence of traits, but size-dependent variation has not been evaluated as a potential confounding issue. Additionally, intraspecific size-dependent variation may imply a novel mating strategy for this species that has not been previously documented for crayfishes.

Near<sup>1</sup>, Thomas J., and Matthew R. Thomas<sup>2</sup>. <sup>1</sup>Yale University, Department of Ecology and Evolutionary Biology, Peabody Museum of Natural History; <sup>2</sup>Kentucky Department of Fish and Wildlife Resources, Fisheries Division.

### Species Discovery and Delimitation of Barcheek Darters, Oopareia

Barcheek Darters, *Oopareia*, are a clade of eight described species that are distributed in the lower Tennessee River system, the Duck River system, the lower and middle portions of the Cumberland River system, and the upper portions of the Green and Barren river systems. Molecular phylogenetic analyses have resulted in the discovery and description of three species of *Oopareia, Etheostoma basilare, E. derivativum,* and *E. nebra.* We present an extensive molecular analysis that indicates *Oopareia,* may contain up to 20 species, 12 of which remain undescribed. The discovery of these new species is reflected by either deep genetic divergence or extensive paraphyly. For example, *E. basilare, E. obeyense,* and *E. derivativum* are reciprocally monophyletic, but exhibit deep genetic divergences that are characteristic of other closely related species of vertebrates. *Etheostoma smithi* and *E. striatulum* are each paraphyletic in mtDNA gene trees. Populations of *E. barbouri* from the Barren River system may be extirpated and are not available for genetic analyses, but published morphological data shows differences that are consistent with other interspecific contrasts among other Barcheek Darter species. In nearly all cases of deep phylogenetic divergence or paraphyly, there are morphological differences consistent with lineages identified in the molecular phylogenies.

Newburn, Tyler, Brittany Butcher, and Michael Sandel. The University of West Alabama, Department of Biological and Environmental Sciences.

# **Preliminary Characterization of the Dermal Mucosal Microbiome of Red Drum** (*Sciaenops ocellatus*)

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 "slime" remains poorly described. We provide the first description of a microbiome in the dermal mucosa of the Red Drum (Sciaenops ocellatus), a euryhaline sportfish of conservation concern in the southeastern United States. Red Drum became an important target for microbial surveillance, because (following the Deepwater Horizon Oil Spill) dermal lesions associated with a disease outbreak in Louisiana were reported to harbor Mycobacterium ulcerans. Throughout the tropics, M. ulcerans is responsible for Buruli Ulcer, a debilitating human disease that affects both humans and fishes. We hypothesized that M. *ulcerans* was carried east along the Gulf Loop Current during and after the Deepwater Horizon Oil Spill. To test the hypothesis, and to characterize the dermal mucosal microbiome of healthy Red Drum, we extracted whole genomic microbial DNA from the dermal mucosa of 60 specimens from coastal waters of Mississippi and Alabama. We used universal PCR primers to amplify a conserved genomic locus (16s). We applied Next Generation Sequencing and bioinformatic tools to quantify the proportion of DNA fragments assigned to known microbe genera, including gram negative and gram positive groups. We report substantial variation in

microbiome diversity among Red Drum specimens, which is poorly associated with geography. These results suggest that movement among adult Red Drum may serve as a mechanism for the spread of human pathogens, but that movement patterns are not strongly associated with predictions from the Gulf Loop Current. Ongoing investigations will determine the utility of intraspecific phylogeny of Red Drum as a predictor of microbiome similarity among individuals.

Patrick<sup>1</sup>, Genevieve, Emily Workman<sup>1</sup>, Kirsten Work<sup>1</sup>, Melissa Gibbs<sup>1</sup>, and Jonathan Freedman<sup>1,2</sup>. <sup>1</sup>Stetson University; <sup>2</sup>University of Florida.

### Diet Overlap by Fish Species in the Volusia Blue Spring Food Web

Springs in Florida have experienced a variety of disturbances, such as changes in flow rates, changes in nutrient quantities and ratios, and introductions of exotic species. Stable isotope analysis (SIA) of carbon and nitrogen is a good tool for constructing an ecosystem food web by examining shifts in isotopic ratios among different species. To examine the effect of these disturbances on the Volusia Blue Spring food web, we used SIA 1) to determine nutrient sources and trophic levels of organisms and 2) to construct a provisional food web. We collected seasonal samples of leaves, algae, amphipods, Sailfin Mollies (Poecilia latipinna), and Mosquitofish (Gambusia holbrooki) at three different locations: the spring boil (to capture primarily aquifer inputs), approximately midrun (to capture potential allochthonous inputs), and the St. Johns River just outside the run. We also collected fecal and/or muscle samples of Sailfin Suckermouth Catfish (*Pterygoplichthys disjunctivus*) and Florida Manatee (*Trichecus manatus*) from midrun. The isotopic signatures of Sailfin Mollies and Mosquitofish were similar, so we used gut content analysis to determine the degree of overlap between these species. Preliminary results revealed that for algae, there were seasonal and spatial differences in nitrogen signatures, possibly due to fertilizer inputs to the spring, and spatial differences in carbon signatures, possibly due to the use of bicarbonate from limestone as a carbon source in spring as opposed to organic sources in the river. For many organisms, the isotopic signatures differed spatially, with different signatures in the spring run and in the river and spatially. The isotopic overlap in Sailfin Mollies, Mosquitofish, and Sailfin Suckermouth Catfish appear to be due to similarities in diet.

*Ruble*<sup>1</sup>, *Crystal L., J.R. Shute*<sup>1</sup>, and Patrick L. Rakes<sup>1</sup>. <sup>1</sup>Conservation Fisheries Inc.; <sup>2</sup>U.S. Fish and Wildlife Service, Elkins, West Virginia.

# Life History of the Crystal Darter, *Crystallaria asprella*, as a Surrogate for the Federally Endangered Diamond Darter, *Crystallaria Cincotta*

Reproductive biology and early life history data are critical for the conservation and management of federally endangered fishes. In attempts to understand the life history of the federally endangered Diamond Darter, *Crystallaria cincotta*, the Crystal Darter, *Crystallaria asprella*, was used as a surrogate. The Crystal Darter is the only other member of the genus *Crystallaria*, and is widespread throughout the middle and eastern United States, making it an ideal candidate for

study in place of the Diamond Darter which is restricted to only the Elk River, WV. Crystal Darters collected from the Black River in Missouri spawned for 4 weeks (10 March - 13 April 2015). Water temperatures during spawning ranged from 13.7–20.0 C. Nuptial males differed from females in the length of and the development of tubercles on the anal fin during spawning time. Tubercles on the anal fins were not visible on the fish when the Crystal Darters were collected 13 September 2014. Females and males spawned with quick vibrations, burying eggs in fine sand in relatively swift clean depositional areas. Egg size was 1.7–1.9 mm. Embryo development lasted 7 to 10 days before hatch. Crystal Darter larvae were 6.8-7.0 mm total length (TL) at hatch, 7.2-7.4 mm TL at swim up, and 8.0-8.2 mm TL at time of volk sac absorption. Feeding larvae had relatively large mouth gapes. They were provided brine shrimp Artemia sp., Ceriodaphnia dubia neonates, marine Brachionus rotifers, and powdered foods (50-400 µm). Larvae preferred skimming prepared powders off the water surface rather than live foods as first feeding despite large gape size. Determining the ideal feeding conditions for this genus continues, but we have successfully reared 8 individuals from 2015 efforts. Refining techniques that worked with this species will hopefully yield a higher survivorship in the future and presumably be applicable to Diamond Darter propagation.

Saidak<sup>1</sup>, Christina Grace, Dr. J. Larry Wilson<sup>1</sup>, and Mark A. Cantrell<sup>2</sup>. <sup>1</sup>The University of Tennessee, Department of Forestry, Wildlife and Fisheries; <sup>2</sup>U.S. Fish and Wildlife Service.

### Movement Patterns for Reintroduced Lake Sturgeon in the Upper Tennessee River System

Lake Sturgeon, Acipenser fulvescens, are one of the slowest to reach sexual maturity and longestlived freshwater fish species in North America. These fish are a species of special concern by the U.S. Fish and Wildlife Service (USFWS), a threatened species by the American Fisheries Society in states where they occur, and a threatened species in Tennessee. They have been reintroduced into the Upper Tennessee River system since 2000. Since December 2013, 49 Lake Sturgeon have been implanted with ultrasonic acoustic transmitters, and 26 fixed-station receivers installed throughout the Upper Tennessee River System to monitor their movement. The objectives of this study were: 1) to determine dispersal and movement patterns of reintroduced Lake Sturgeon in the Upper Tennessee River system, 2) to identify and assess potential spawning habitats in the Upper Tennessee River system, and 3) to determine if dams inhibit upstream and downstream movements. Lake Sturgeon implanted with acoustic transmitters were detected and monitored throughout the study area in the Upper Tennessee (RM 427-632), Clinch (RM 0-5), Hiwassee (RM 5-501.9), Holston (RM 0-52.2), and French Broad Rivers (RM 0-32.3). There was a higher concentration of fish aggregating in Fort Loudoun Reservoir and smaller numbers in Watts Bar and Chickamauga Reservoirs. Movements varied with the majority of the fish traveling <5 km during the study period. Areas with aggregations of fish (>5 fish) were designated as "core use". During the study, there were 1,130,809 individual transmitter detection's recorded. Gaining a better understanding of the factors affecting Lake Sturgeon recruitment and survival will be critical in designing restoration or reintroduction programs in the upper Tennessee River system and areas like it.

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# Use of Pectoral Fin Rays to Test the Effect of Stream Discharge on Growth of Sicklefin Redhorse

The Sicklefin Redhorse (SFR) Moxostoma sp. is a rare catostomid fish in the upper Tennessee River watershed of western North Carolina and north Georgia whose listing status is currently under review. Status reviews rely on the best available scientific data, but age data, which is used to estimate population survival, growth rates, and recruitment for SFR, is difficult to collect and usually involves sacrifice of sampled individuals. We tested the utility of aging SFR with pectoral fin rays, which is a non-lethal procedure, from SFR collected in the spring of 2014 and 2015 from Brasstown Creek, GA. Pectoral fin rays were aged blindly and independently by two readers. Annual growth increments were measured with the use of ImageJ software to determine growth rates. Growth models constructed from growth curves were similar to previously published data. Captured SFR (n=48) were PIT tagged for future tracking and population estimation. Using age data, we compared growth rates of SFR to stream discharges. For spring 2014 data, yearly variation in annual growth varied significantly (P=0.03) due to mean annual and spring discharges with higher discharges resulting in decreased growth. SFR are long-lived (max observed age=21 years) and experience slow growth after reaching sexual maturity. Factors that alter stream discharges, such as climate change and landscape development, may increase discharge, possibly impacting SFR growth. Additional data from spring 2015 is currently being processed and will include evaluating changes in growth based upon maximum discharge and variability in discharge. Growth rates of other redhorses in Brasstown Creek will be constructed and compared to SFR.

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# Recent Discovery of the Ohio Shrimp, *Macrobrachium ohione*, in the St. Francis, White, and Arkansas Rivers

The Ohio Shrimp (*Macrobrachium ohione*) is known to exhibit amphidromy, a form of diadromy, meaning migration from freshwater to marine habitat is performed, but is for juvenile development and not linked to spawning. Migration range for Ohio Shrimp within the Mississippi River basin was historically from the Ohio River and Upper Mississippi River to the Gulf of Mexico, but shrimp abundance has reportedly declined, particularly upstream of Louisiana. Dams and channel flow alterations are hypothesized to have impacted upriver migrations of shrimp. Current range, abundance, and life history of Ohio Shrimp is relatively unknown in the Mississippi River basin in reaches distant from sea water. We report recent collections of Ohio Shrimp in Arkansas rivers that were notably greater than 800 km from the Gulf of Mexico. Most collections of Ohio Shrimp were made using baited wire-mesh traps set along shorelines and in deeper water when river conditions allowed. During May-September of 2012 and 2015, 49 Ohio Shrimp were caught in the lower Arkansas River, seven in the White

River, and one from the St. Francis River, for a total of 57 specimens. Total length (TL) was measured to assess age class using previous literature values. Samples contained 41 young of year (<50 mm TL), 16 age-1 (50-90 mm TL), and zero age-2 (>90 mm TL) individuals. Four age-1 females caught in the White River during late June and early July of 2015 had eggs visible between the swimmerets. Young of year encompassed 72% of the total and all were caught in the lower Arkansas River. The White and St. Francis river collections were previously undocumented locations, expanding the known range of Ohio Shrimp. Previous research noted the importance of main stem Mississippi River habitat to Ohio Shrimp, but our data suggest tributaries should also be considered in the conservation of this species.

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# NHC3: A Novel Approach to Encourage Student Participation in Natural History Collections Preservation, Research, and Outreach

The preservation and curation of natural history collections has lost much of the public interest in recent years. Some institutions view the maintenance of these collections as a financial burden and elect to destroy or donate entire collections. However, student involvement in collection maintenance can provide both collection maintenance services and valuable experience in a museum setting. The Natural History Collections Curation Club is a student organization that aids in the preservation of every biological collection at Arkansas State University and provides valuable volunteer experience to undergraduate and graduate students. Our work includes specimen preparation, collections, managed specimens within collections, and provided exciting educational and volunteer experiences to local schools and students. NHC3 has even branched out to other universities around the nation. Since our forming in the spring of 2013, clubs have formed at the University of California Santa Barbara, Missouri State University, and Pittsburgh State University. We believe that clubs like ours can be a previously untapped resource to help save many deteriorating collections and inform the public of the importance of having access to the rich biological and historical data contained within natural history collections.

Tracy, Bryn H. North Carolina Division of Water Resources.

### An Index of Biotic Integrity for Wadeable Streams in the North Carolina Sand Hills

Fish community data from wadeable Sand Hills streams have been collected by Division staff since 1990 following existing standard operating procedures. However, metrics and biocriteria were never developed specifically for these unique assemblages which have naturally low species diversity and low biological productivity. Fish assemblages were not rated accurately and consequently, the streams were classified as Not Rated until metrics and biocriteria could be developed. A 7-Metric Sand Hills Index of Biotic Integrity has been developed to identify

impairment and to statistically differentiate between highly impacted sites (rated Poor or Fair) and reference and near-reference sites (rated Good or Excellent). Relationships between the seven metrics versus specific conductance, pH, total habitat score, land use types, and land use disturbance classes were also evaluated. The metrics and biocriteria were further validated with additional reference and degraded sites data collected in 2013 and 2014. Based upon a dataset of more than 90 samples, reference sites and other least-impacted fish community sites in the Sand Hills should have low specific conductance, low pH, low abundances of fish, and low percentages of tolerant fish; high percentages of Key Sand Hills Species, Key Sand Hills fish, and Invertivore Cyprinids; at least two Intolerant Species; and high quality instream habitat characteristics. Impacted sites would be expected to have the converse of these characteristics. The Sand Hills NCIBI is an additional biological monitoring tool that will be used by the Division in evaluating wadeable streams, complementing the existing benthic macroinvertebrate assessments.

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## A Northern Fish in a Southern Land: Conservation Status of Trout-perch (*Percopsis omiscomaycus*) in Tygart's Creek and Lewis County, Kentucky

The Trout-Perch, Percopsis omiscomaycus (Walbaum), is a fish species widely distributed in northern North America, extending south to northeast Kentucky. The purpose of this study is to document the current species' distribution and population status in Kentucky and compare that to its historical distribution. We also seek to identify habitat variables associated with the present occurrence of Trout-Perch. The first phase of the study occurred from April to September 2014 and investigated four watersheds in Lewis County, Kentucky: Cabin Creek, Quicks Run, Salt Lick Creek, and Kinniconick Creek. The second phase took place in summer of 2015 and investigated the Tygart's Creek drainage, in Carter and Greenup counties. The sites in Lewis County and the tributaries of Tygart's Creek were surveyed for Trout-Perch by a semiquantitative protocol to quantify Trout-Perch abundance by intensively seining all pools in a 200-300 m stream reach. Habitat data were taken from three randomly selected pools at each site. Additional sites in the mainstem of Tygart's Creek were sampled qualitatively. We detected Trout-Perch at 5 of 22 sites in Lewis County, all in the Quicks Run and Salt Lick Creek watersheds. In the Tygart's Creek tributaries we detected Trout-Perch at 4 of 15 sites, three of which are new locality records. Trout-Perch were common to abundant in some of these sites. These are the first records for this species in the Tygart's Creek drainage since 2002. However, Trout-Perch was detected in only 1 of 7 Tygart's mainstem sites and was not detected in the Cabin Creek or Kinniconick Creek watersheds. Historical data indicate Trout-Perch have declined in Cabin Creek, Kinniconick Creek and the mainstem of Tygart's Creek. In sites with Trout-Perch present, Trout-Perch were associated with sites having small substrates and deep pools. We will continue surveying other historical localities in Kentucky in the future.

# Translocation of the Bluebreast Darter, *Etheostoma camurum*, to the Upper Licking River Basin of Ohio

The Bluebreast Darter (Etheostoma camurum) population in Ohio has suffered from human disturbance in the past but is currently experiencing a significant rebound. Widespread range expansion is thought to be the result of improvements in water quality. Despite expansion into much of their former distribution within Ohio, there are still three historical occurrence locations where anthropogenic impacts prevent re-colonization. For example, Bluebreast Darters were collected by S.F. Baird in 1853 in Yellow Creek, a tributary of the Mahoning River, and by R. C. Osburn in 1899 in the North Fork of the Licking River and in the Stillwater River near Dayton, but these locations have experienced large scale environmental disturbance and no longer harbor Bluebreast Darters. These locations have experienced large-scale environmental disturbance in the past, but currently have seen some recovery of the native fish communities. In particular, the Licking River basin presents an ideal opportunity for the reintroduction of this one time inhabitant. Bluebreast Darters have already naturally re-colonized the lower reaches of this river system, however the dam at Dillon Reservoir has prevented further expansion upstream. This project seeks to translocate Bluebreast Darters past this barrier, back into the upper Licking River basin, after over a century of absence. We intend to capture and tag Bluebreast Darters from the upper Muskingum River basin. Because the Licking River is a sub-basin of the Muskingum River watershed, we avoid concerns over locally adapted genetic divergence. Translocation events will occur once a year for five years, moving approximately 100 individuals to each of six reintroduction sites. Associated population monitoring will take place during and after these translocation events.