Title: Status survey and population characteristics of the Paleback Darter (Etheostoma pallididorsum) and the Caddo Madtom (Noturus taylori)

Project Summary:
This project seeks to provide updated information on the status and population characteristics of two of Arkansas’ Species of Greatest Conservation Need, the Paleback Darter and Caddo Madtom. The main objectives of this project include: (1) conducting surveys of historic localities throughout their range; (2) obtaining size-distribution (estimating age classes) data and relative abundance for both species at select locations on a seasonal basis; and (3) generating population characteristics for each species via non-lethal genetic methods (e.g. genetic variation, effective population sizes, patterns of migration). The overall objective is to provide data for both species that will inform potential listing decisions under the Endangered Species Act by the U.S. Fish and Wildlife Service, in addition to state ranking decisions by the Arkansas Game and Fish Commission.

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   Phone: (501)-961-1419 email: joharris@astate.edu

Project Budget:
SWG Amount Requested (64.8%): $56,855
Match Amount Provided (35.2%): $30,897
Total Amount of Project: $87,752
Need
The Paleback Darter (*Etheostoma pallididorsum*) and Caddo Madtom (*Noturus taylori*) are both endemic to the upper Caddo and Ouachita rivers in the Ouachita Highlands of southwestern Arkansas. While neither species is protected under the Endangered Species Act (ESA), they are Species of Greatest Conservation Need (SGCN) in Arkansas due to their restricted range, unique life-history traits, and limited suitable habitat. In 2010, the Center for Biological Diversity released a petition to list 404 aquatic species from the southeastern U.S. under the ESA, including *E. pallididorsum* and *N. taylori*.

Status surveys have not been conducted for *E. pallididorsum* in 11 years (Robison 2004). Turner and Robison 2006 sampled *N. taylori* throughout its range, however, it is unclear what year(s) these samples were conducted. Although levels of population genetic diversity, effective population size, and migration have been estimated for both species using allozymes (Turner and Robison 2006), a fine-scale genetic analysis using highly variable microsatellite DNA markers, may provide more detailed estimates of these population characteristics for both species. Given both species’ rarity, vulnerability, and inclusion in the recent ESA listing petition, current information on status, distribution, genetic variation, and life-history are needed to guide conservation decisions for these enigmatic species.

Purpose and Objectives
The purpose of this project is to obtain updated information on the status and population characteristics of *E. pallididorsum* and *N. taylori* to inform future conservation and listing decisions for these SGCN. The desired outcome of this project will be achieved by the following objectives: (1) conduct surveys for both species at historic localities throughout their range; (2) obtain size-distribution (estimate age class) data and relative abundance for both species at select locations on a seasonal basis; and (3) generate population characteristics for each species via non-lethal genetic methods (e.g. genetic variation, effective population sizes, patterns of migration).

Location
Sample locations will include historic localities in the Caddo and Ouachita rivers (both species) and in the upper Little Missouri River (*N. taylori*) in southwestern Arkansas (Fig. 1). Historic localities of *E. pallididorsum* include selected localities of Robison (2004) in the mainstem of the Caddo River and relevant tributaries (Collier, Gap, Lick, and Polk creeks), in addition to tributaries of the Ouachita River (Big Fork, Kate’s, and Mazarn creeks). Historic localities of *N. taylori* include selected localities of Robison and Harris (1978) in the mainstem of the Caddo, South Fork Caddo, Ouachita, and Little Missouri rivers.
Approach
Sampling will be conducted at historic localities for both species at least once during the duration of the project. Three localities each in the Caddo and Ouachita rivers where both species are historically known will be sampled once each season (winter, spring, summer, and fall) for the duration of the project in order to provide both size-distribution (age-class) estimates and a relative abundance at these locations. For *E. pallididorsum*, select breeding sites (if accessible) will be sampled during the spawning season (December-March) to evaluate basic movement patterns and timing of spawning migrations. Although *N. taylori* has not been found in the upper Little Missouri River in recent times despite intensive sampling (Turner and Robison 2006; B. Crump and M. Cole, personal communication), we will be sample the historic locality at least once during the project to confirm absence/presence.

Sampling will be conducted using a 10 or 20 ft. seine, and effort (in time and number of seine efforts) will be recorded to obtain relative abundance in terms of catch per unit effort. Upon capture, target species will be held in suitable containers, sexed (when possible), enumerated, and measured for Standard Length. Select individuals will be fin-clipped for genetic analysis and all individuals will be returned live to the point of capture. All other fishes (species associates) will be identified, enumerated, and released at the point of capture. Descriptive habitat characteristics will be noted and photo-documented when possible.

Tissue samples for genetic analyses will be taken via fin clips with fine-tip forceps and scissors, and fishes will be released live at their point of capture (as approved by the ASIH guidelines, Nickum et al. 2002). Geographic coverage of samples for genetic analyses for both species will attempt to include up to 30 individuals per site from up to five localities per drainage (i.e., Caddo and Ouachita) and one locality from the Little Missouri River for *N. taylori*. DNA will be extracted from tissues using the DNeasy Kit (Qiagen) and individuals will be genotyped for up to
10 microsatellite DNA markers following previously published protocols (Fluker et al. 2010; Williams and Moyer 2012; Fluker et al. 2014). Microsatellites are highly variable, short regions of DNA with simple 2-6 base pair repeats and have been used extensively for understanding genetic characteristics of similar imperiled species (e.g. Fluker et al. 2010; Austin et al. 2011; Williams and Moyer 2012; Fluker et al. 2014). Microsatellite DNA fragments will be amplified from individuals via polymerase chain reaction and prepared samples will be read on an ABI 3730 DNA analyzer. The complete genetic dataset for both species will be analyzed using a number of population genetic methods to estimate: (1) significance of genetic structure among populations; (2) amount of genetic variability within each population; (3) migration among populations; (4) effective population sizes; and (5) recent changes in effective population sizes (i.e. genetic bottlenecks).

Expected Results and Benefits
Results of this project will provide the Arkansas Game and Fish Commission, U.S. Fish and Wildlife Service, and U.S. Forest Service with current data on which to base conservation and management decisions for E. pallididorsum and N. taylori. The expected results are timely as both of these SGCN have been petitioned by the Center for Biological Diversity to be listed under the ESA. Additionally, genetic data generated from this project will provide a snapshot of current levels of population connectivity, genetic diversity, and effective population sizes, in addition to providing baseline levels of genetic variation that can be compared to similar studies in the future. Levels of genetic variation at the population level will provide measures of “genetic health” which may help to prioritize conservation needs on a population-by-population basis. Results of this study will be disseminated in the form of presentations at professional meetings, written reports, and/or peer-reviewed journal publications.

References
Qualifications

Mitzi Cole: Received M.S. in Biology from the University of Arkansas. Has over 23 years of experience sampling freshwater fishes. Has worked with the Paleback Darter since 1990, resulting in several technical reports and publications.

Betty Crump: Received M.S. in Biology from Arkansas State University. Has over 25 years of experience sampling freshwater fishes. Has worked with the focal species of this project since the late 1980s, resulting in several technical reports and publications.

Brook L. Fluker: Received Ph.D. in Biology from the University of Alabama in 2011 with an emphasis in phylogenetics, population genetics, and conservation of freshwater fishes. Has 11 years of experience sampling, handling, and collecting tissues from freshwater fishes in North America, including experience assisting with surveys of freshwater mussels and snails and seasonal abundance surveys for several federally protected fishes. This work has resulted in six publications, 10 technical reports, and 30+ presentations at professional conferences and meetings.

John L. Harris: Received Ph.D. in Zoology from University of Tennessee in 1986 with emphasis in taxonomy and systematics of aquatic fauna concentrating on fish and mussels. Has 30+ years experience in performing mussel surveys and impact analyses resulting in numerous peer-reviewed publications and/or agency reports. Has co-directed or been a committee member for 15 graduate students researching distribution and/or life history aspects of freshwater mussels in Arkansas.
<table>
<thead>
<tr>
<th>Budget Justification</th>
<th>Year 1</th>
<th>Year 2</th>
<th>Total</th>
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<td><strong>PERSONNEL</strong></td>
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<td>Graduate Student</td>
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<td>Travel to study sites</td>
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<td>Lodging</td>
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<td>DNA processing, extraction, reactions, and runs</td>
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**Description of Match from Arkansas State University**

- Waived Indirect costs (26% [ASU rate is 36%]) $13,438
- Faculty Salary (8.5% time per month for 18 months) $9,350
- Faculty Fringe (37.3% of matched salary) $3,488
- 36% of salary and fringe waived $4,622
- **Total Match provided** $30,897 (35.21%)

*mileage: Jonesboro to Norman, AR roundtrip (460 miles), plus 80 local miles per trip; **Lodging rate; Norman, AR