

## **Ozark Cavefish Distribution and Life History Related to Mining in Jasper and Newton Counties**

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### **Executive Summary**

Parts of Jasper and Newton counties fall within the Tri-State Mining District, an area that was mined extensively for lead and zinc resulting in significant contamination of groundwater resources. To address mining contamination problems and complement ongoing Superfund remediation activities, the U.S. Department of the Interior (DOI) and Missouri Department of Natural Resources (MDNR) began a Natural Resource Damage Assessment (NRDA) to evaluate injuries to resources associated with the release of hazardous substances and determine appropriate compensation for these injuries. It was recognized that the known and potential distribution of the Ozark cavefish (*Amblyopsis rosae*), a federally listed threatened and Missouri listed endangered species, overlaps this region. However, there is only limited information available to describe the species' distribution, biology, or basic life history including the toxic effects of metals on the organisms that inhabit groundwater, or the role that chronic contamination has played in shaping species' distributions.

The goal of this project is to increase our understanding of the distribution and basic life history of Ozark cavefish to improve the effectiveness of conservation and recovery efforts for the species and the ecosystems it occupies by informing the NRDA process. We propose surveys for aquatic animals and water quality in unconfirmed/potential cavefish sites in and near areas that have been impacted by mining in the Tri-State Mining District in addition to using easily accessible, active sites (sites inhabited by Ozark cavefish) to study fish growth, sighting frequency, and physiological status through time. We will emphasize comparisons between impacted and unimpacted sites to inform management and mitigation decisions. Specific objectives include:

1. Perform surveys of active and unconfirmed/potential Ozark cavefish sites in and adjacent to Tri-State Mining District affected areas to document occurrence of cave-adapted organisms such as cavefish, cave crayfish, and other aquatic species.
2. Perform water quality analyses to describe baseline chemical conditions and test for concentrations of metals.

3. Measure life history characteristics of Ozark cavefish including growth, condition, reproductive status, and persistence/immigration in active sites in and adjacent to mining affected areas using mark/recapture methods.

The project will involve funding two hourly employees based in Neosho, Missouri, to perform surveys and life history studies, and a contract with MDNR to analyze water samples for contaminants. The duration of the project will be for FY10 and FY11 with a total budget of \$83,080, with \$55,328 (67%) requested from the NRDA via a USFWS grant application.

Information from this project will be directly applicable to conservation management of the Ozark cavefish and the habitat areas/systems associated with sites occupied by the species (i.e., caves, recharge zones, aquifers). The project will thereby meet several state and federal conservation goals/objectives. Data collected will enhance the ongoing NRDA process by providing information related to injury determination and quantification in groundwater impacted by mining. Because Ozark cavefish are a primary link between the NRDA and U.S. Fish & Wildlife Service (USFWS) and Missouri Department of Conservation (MDC), there are potentially significant funds that could be recovered to restore the quality of groundwater as a drinking source that would, at the same time, benefit conservation of Ozark cavefish by supporting work performed by MDC. This project addresses MDC Next Generation Goals for conserving animals and their habitats (Goal 1) and protecting clean and healthy waters (Goal 2). Further, active Ozark cavefish sites and their recharge zones are recognized collectively as a Conservation Opportunity Area, and are therefore, a priority for conservation effort.

### **Information Need and Justification**

Parts of Jasper and Newton counties fall within the Tri-State Mining District, an area that was mined extensively for lead and zinc from c.1850 to 1960 (IEC 2008). As a result of this mining and related activities, large amounts of metals including cadmium, lead, zinc, and nickel were released into the environment, including into groundwater and associated sediments in subterranean areas potentially occupied by unique aquatic organisms such as cavefish, cave crayfish, and numerous other invertebrates. Cadmium, lead, zinc, and other metals associated with mining are hazardous substances that are potentially toxic to a wide variety of plants and animals.

To address these contamination problems and complement ongoing Superfund remediation activities by the Environmental Protection Agency (EPA), the DOI and MDNR began a NRDA. The NRDA process evaluates injuries associated with the release of hazardous substances and determines appropriate compensation for those injuries. The NRDA has completed an initial preassessment phase where it determined that metals were released as a result of mining operations at a level sufficient to cause potential injury to natural resources. During the ensuing assessment planning phase, a plan was produced to outline routes of investigation that would be pursued related to injury determination and quantification (IEC 2008). The plan recognized the likelihood that natural resources associated with groundwater were negatively affected by contamination from mining wastes. However, there is limited information about the toxic effects of metals on the organisms that inhabit groundwater or the role that chronic contamination of underground habitat has played in shaping species' distributions.

The Ozark cavefish (*Amblyopsis rosae*) is a federally listed threatened and Missouri listed endangered species that occurs in groundwater of southwest Missouri, including Jasper and Newton counties. Despite the high profile status, relatively little remains known of the species' distribution, biology, or basic life history (population dynamics, growth), particularly in Missouri. The lack of such basic information is largely attributable to the species' ecology:

- (i) few sites are occupied by the species,
- (ii) in-ground habitats are minimally accessible to humans, and
- (iii) population densities appear to be relatively low where the animals do occur.

Given the importance of life history information in formulating conservation and management actions, obtaining such information is critical. A significant portion of the cavefish's potential range in southwest Missouri (Jasper and Newton counties), including a recently discovered active cavefish site, lies within areas that have been negatively impacted by historical mining activities. Groundwater contamination by metals may lead to reduced health or mortality of Ozark cavefish and other aquatic organisms.

Our records indicate that eight sites are inhabited by Ozark cavefish in Jasper and Newton counties, with an additional 41 sites regarded as either having unconfirmed reports of Ozark cavefish or conditions that make the site a potential location to search for cavefish (Figure 1). Of the eight active sites, one site just east of Seneca lies within a Designated Area, three sites are adjacent to impacted areas and streams, but outside recognized land surface boundaries, and three sites are further removed from areas known to be impacted by mining. Of the 41 unconfirmed or potential sites, two are within waste area boundaries, 11 are adjacent to impacted areas and streams, and the remaining 38 sites are further removed from impacted areas. Because the boundaries of most shallow aquifers are unknown and it is not uncommon for surface streams in karst regions to lose some flow to adjacent shallow aquifers, the degree to which any of these sites is or was impacted by contamination from mining activities is not known. In addition, the locations of several potential sites are inexact and need to be more precisely determined. For example, 33 potential sites could only be located to the middle of a quarter section. Access to several unconfirmed and potential sites may be restricted by landowners or by unsafe conditions. Therefore, significant effort will be required to locate and survey several sites; however, we are hopeful that during the course of investigations new sites will be discovered.

There is limited published information available describing life history of the Ozark cavefish. The only studies were conducted in Logan Cave, Arkansas, by three graduate students of Dr. J. Johnson of the Arkansas Cooperative Fish and Wildlife Research Unit at the University of Arkansas (Fayetteville).

In the first study (Means and Johnson 1995), investigators attempted to determine the extent and patterns of movement of Ozark cavefish in Logan Cave with a secondary objective of evaluating the accuracy of standard visual observation methods for measuring numbers and status of cavefish populations. Investigators tagged cavefish using a visual implant tag inserted in the caudal peduncle region of the fish and performed several surveys over a little more than a year to recapture and measure cavefish for length and note locality. During the study, investigators

tagged 80 cavefish with several recaptures and new, with untagged cavefish observed at a relatively consistent rate on each trip. Cavefish were usually found in quiet waters and moved net distances of up to 906 m (mean 142 m). Smaller cavefish were recaptured more frequently and moved less than larger cavefish. Conclusions of the study were that standard visual census methods do not provide useful information on the size or status of the cavefish population due to immigration into the human-accessible portion of the cave stream from inaccessible portions or from the surrounding aquifer; consequently, standard census methods would be expected to underestimate population sizes. Extrapolated movement rates could be several kilometers during the typical cavefish lifetime. Investigators argued that the evidence for immigration and significant movement support protection of the recharge zone and karst aquifer in addition to the cave stream.

A second study (Brown and Johnson 2001) aimed to determine survival, growth, and further study movement of Ozark cavefish in Logan Cave. Methods similar to Means and Johnson (1995) were used, with this study occurring immediately following the first and lasting approximately two years. An additional 140 cavefish were tagged, and of that total, 68 were recaptured at least once. Based on recapture data, investigators produced a persistence curve showing that half of the fish disappeared from collections after three months, with 28 months the longest time an Ozark cavefish was known to persist. As in the first study, untagged fish were captured at a relatively consistent rate throughout the study, with the exception of a significant increase in the number of smaller fish during July of each year. Growth in length was highest during April through October, with smaller fish growing more rapidly than larger fish. Net distances moved were as high as 673 m, with larger fish moving further than smaller fish. Based on recapture data, the investigators calculated an annual immigration rate of 20 Ozark cavefish.

The third study (Adams and Johnson 2001) focused on evaluating the metabolic rate and body condition of Ozark cavefish in Logan Cave. For this study, investigators measured in-situ oxygen consumption, length, and weight (mass) of cavefish during time periods corresponding to each of the four seasons coincident with minor fluctuations in temperature and flow. Sample sizes were five or six fish during each season. Investigators found that the relationship between mass and metabolic rate was positive during summer and autumn, and negative during winter and spring (a negative relationship is highly unusual and could be an artifact of the low sample sizes). Also, body condition (the combination of length and weight) was higher during summer and autumn than during winter and spring. Investigators noted that a gray bat maternity colony was present from late March to early September and likely increased nutrients and associated food availability for cavefish during that time. Four female cavefish were collected during late August that had large, visible ova in their abdomen, observed by shining a bright light through the translucent body wall. Young fish (approximately 10 mm long) were seen in late July.

The information to be collected by this project is important to species recovery and meeting the goals of the NRDA to determine the extent of injury to natural resources as a result of contamination by mining wastes. Both federal and state plans describe degraded water quality as a primary threat to Ozark cavefish. The federal plan (USFWS 1989) specifically describes water quality in the western portion of the species' range as threatened by toxic metals in the area of old lead and zinc mines of the Tri-State Mining District and calls for water quality monitoring of heavy metals and population surveys in possible cavefish sites. The state plan (MDC 1999)

states that groundwater pollution may be the single greatest threat to the continued existence of the Ozark cavefish with mine leachates the first item included in a list of examples. Water quality monitoring, including analyses for metals contamination, is a primary objective listed for species recovery with additional information needs addressed by our proposed project: investigate reported sightings and potential sites; conduct additional research into the biology, life history and habitat requirements of the Ozark cavefish; and further develop and refine census methods.

Because Ozark cavefish are potentially threatened by groundwater contaminated by mining wastes, the species is an important link between the NRDA, USFWS, and MDC. Significant funds could be recovered to address improvements to the quality of groundwater as a drinking source and at the same time benefit conservation of Ozark cavefish (and other aquatic organisms) by supporting work performed by MDC including, habitat improvement (reducing erosion and sediment transport into cave systems), site protection (cave gates, well covers), research (recharge delineation, surveys), and outreach (public meetings to increase awareness, educational materials). Because MDC already does most of these activities, there could be an opportunity to fund ongoing work that is regarded as high priority without initiating new programs. Therefore, this could be a cost-effective opportunity to meet several state and federal goals for Ozark cavefish recovery.

The goal of this project is to increase our understanding of the distribution and basic life history of Ozark cavefish to improve the effectiveness of conservation and recovery efforts for the species and the ecosystems it occupies by informing the NRDA process. We propose surveys for aquatic animals and water quality in unconfirmed/potential cavefish sites in and near areas that have been impacted by mining in the Tri-State Mining District in addition to using easily accessible, active sites (sites inhabited by Ozark cavefish) to study fish growth, sighting frequency, and physiological status through time. We will emphasize comparisons between impacted and unimpacted sites to inform management and mitigation.

## **Decision Elements**

### Objectives:

1. Perform surveys of active and unconfirmed/potential Ozark cavefish sites in and adjacent to Tri-State Mining District affected areas to document occurrence of cave-adapted organisms such as cavefish, cave crayfish, and other aquatic species.
2. Perform water quality analyses to describe baseline chemical conditions and test for concentrations of metals.
3. Measure life history characteristics of Ozark cavefish including growth, condition, reproductive status, and persistence/immigration in active sites in and adjacent to mining affected areas using mark/recapture methods.

As data allow, we will test hypotheses that predict a negative effect of mining (increased groundwater contamination) on Ozark cavefish life history characteristics (reduced growth and condition, lower egg counts in reproductive females, occurrence of abnormalities such as lesions and eroded fins). We anticipate that much of the data we collect will be descriptive with limited

applicability of statistical analyses to interpretation of results due to unavoidably low sample sizes. Nevertheless, descriptive data may play an important role in informing the NRDA and other conservation management decisions.

## **Expected Benefits**

Information from this project will be directly applicable to conservation management of the Ozark cavefish and the habitat areas/systems associated with sites occupied by the species (caves, recharge zones, aquifers). The project will thereby meet several state and federal conservation goals/objectives already listed above. Data we collect will enhance the on-going NRDA process by providing information related to injury determination and quantification in groundwater impacted by mining. Because Ozark cavefish are a primary link between the NRDA, USFWS, and MDC, there are potentially significant funds that could be recovered to address improvements to the quality of groundwater as a drinking source that at the same time benefits conservation of Ozark cavefish by supporting work performed by MDC. This project addresses MDC Next Generation Goals for conserving animals and their habitats (Goal 1) and protecting clean and healthy waters (Goal 2). Further, active Ozark cavefish sites and their recharge zones are recognized collectively as a Conservation Opportunity Area and are therefore a priority for conservation effort.

## **Approach**

This project will involve funding two hourly employees, a lead assistant and field assistant, to perform surveys focused on sampling for aquatic organisms and water quality in approximately 20 to 40 sites, depending on accessibility, in Jasper and Newton counties. Multiple visits (two – four) will likely be necessary in order to survey sites under low and high water conditions. The assistants will be based in Neosho or other reasonably close MDC office and would use an MDC truck.

Surveys for animals will employ standard visual census methods currently used for population monitoring. Water chemistry analyses will be performed as a routine part of each site visit. Some parameters will be measured in the field with portable instruments and test kits. These include temperature, conductivity, pH, dissolved oxygen, turbidity, hardness, alkalinity, nitrate, ammonia, and phosphate. Water samples will be collected for laboratory analyses of dissolved heavy metal concentrations including cadmium, zinc, and lead. These analyses will be done through a contract to MDNR. Sediment samples also will be collected, dried, and analyzed by MDC staff for metal concentrations using equipment available from USFWS. Sites will be surveyed at least two times, preferably under different flow conditions (low, high) and in different seasons. Sites that appear to have a high potential to be inhabited by Ozark cavefish, denoted for example by the presence of cave crayfish, may be visited more frequently.

Depending on the outcome of water quality analyses, we will evaluate sites within the boundaries of mining Designated Areas or proximate to impacted surface streams as “treatment” sites, with sites outside the boundaries of Designated Areas constituting “reference” sites if water chemistry analyses support minimal metals contamination. There is currently one active Ozark cavefish monitoring site within the boundaries of a waste area (potential treatment site), and

eight active sites near, but not within waste area boundaries (potential reference sites). A priori determination of how many sites will be regarded as impacted vs. reference is difficult because the extent of contamination in and around waste areas is only approximately known. Also, there were widespread, small-scale mining operations in the region that could have resulted in contamination of groundwater outside of Designated Areas.

In sites occupied by Ozark cavefish, we will use established mark/recapture methods to study species' life history characteristics. Sites will be visited once every four to eight weeks. Fish will be captured using dip nets or baited traps. Following capture, fish will be measured for total and standard length, weighed, inspected for reproductive status, and if total length > 30 mm injected with a Visible Implant Alpha tag (Northwest Marine Technology Inc.) in the caudal peduncle region. These pliable, injectable tags are 1 x 2.5 mm in size and utilize combinations of three character alphanumeric codes with three fluorescent colors thereby allowing for individual and batch marking. Detection of tags is enhanced through use of an LED light. Previous studies (Means and Johnson 1995, Brown and Johnson 2001) used similar methods to tag Ozark cavefish in Logan Cave, Arkansas. Fish will be measured and tagged without use of anesthetic, and following handling will be briefly held in a cooler to ensure recovery to a vigorous state. From these data we will calculate growth rates, body condition as a combination of length and weight, and frequencies of tagged and untagged fish in a particular site to estimate persistence and immigration. We do not anticipate opportunity to investigate movement within a site because most of the active Ozark cavefish sites we will be studying are wells or short cave stream segments. However, in one site there are two wells separated by approximately 140 m (460 feet) with movement between the wells a possibility.

Tag retention and handling effects will be assessed using commonly available aquarium cavefishes (will choose one of several species depending on availability and consultation with Bill Elliott). Approximately 30 aquarium cavefish will be handled and tagged to simulate the protocol used in the field. The fish will be held in a facility in Columbia or Neosho (FWS National Fish Hatchery) and monitored daily for general health and mark status.

Inspection of female Ozark cavefish for reproductive status and counts of eggs in the abdomen will be attempted using handheld backlighting behind a thin aquarium in the field.

### **Duration and Schedule**

The project will be conducted during MDC FY10 and FY11 (July 1, 2009 - June 30, 2011). Work during the first quarter of FY10 will involve hiring staff, buying gear, and planning surveys. Surveys will begin near the end of the first quarter and continue for the duration of the project. Potential/unconfirmed sites would be visited at least two times during the study, and active sites visited once every four to eight weeks.

## Budget

|                              | FY10     | FY11     |                        | Total per Category |
|------------------------------|----------|----------|------------------------|--------------------|
| <b>Budget Category</b>       |          |          |                        |                    |
| <b>Expense</b>               | \$19,000 | \$17,700 |                        | \$36,700           |
| <b>Temporary Labor</b>       | \$23,190 | \$23,190 |                        | \$46,380           |
| <b>Equipment</b>             | \$0      | \$0      |                        | \$0                |
| <b>Funds Requested</b>       |          |          |                        |                    |
| <b>Total per Fiscal Year</b> | \$42,190 | \$40,890 | <b>Total all Years</b> | \$83,080           |
| <b>Outside Funding*</b>      | \$27,914 | \$27,414 |                        | \$55,328           |
| <b>% Outside Funds</b>       | 66%      | 67%      | <b>%Outside Funds</b>  | 67%                |

See Resource Science budget spreadsheet in Appendix A for additional details. Expense includes field gear, tagging supplies, water quality test kits and portable instruments, and contract to MDNR for metals analyses. Temporary Labor includes one lead assistant \$9.32/hr for 1600 hrs and one field assistant \$8.49/hr for 975 hrs.

## Potential Funding Sources

Funding would be provided by USFWS through competitive award of NRDA funds, MDC Resource Science budget, and application for MDC Wildlife Diversity Funds.

## Literature Cited

- Adams, G.L. and J.E. Johnson. 2001. Metabolic rate and natural history of Ozark cavefish, *Amblyopsis rosae*, in Logan Cave, Arkansas. *Environmental Biology of Fishes* 62:97-105.
- Brown, J.Z. and J.E. Johnson. 2001. Population biology and growth of Ozark cavefish in Logan Cave National Wildlife Refuge, Arkansas. *Environmental Biology of Fishes* 62:161-169.
- Industrial Economics, Incorporated (IEC). 2008. Damage assessment plan for Jasper and Newton counties, Missouri. Draft Final Report for State of Missouri.
- Means, L.M. and J.E. Johnson. 1995. Movement of threatened Ozark cavefish in Logan Cave National Wildlife Refuge, Arkansas. *The Southwestern Naturalist* 40(3):308-313.
- Missouri Department of Conservation. 1999. An action plan for the Ozark cavefish (*Amblyopsis rosae*) in Missouri. 26 pp.
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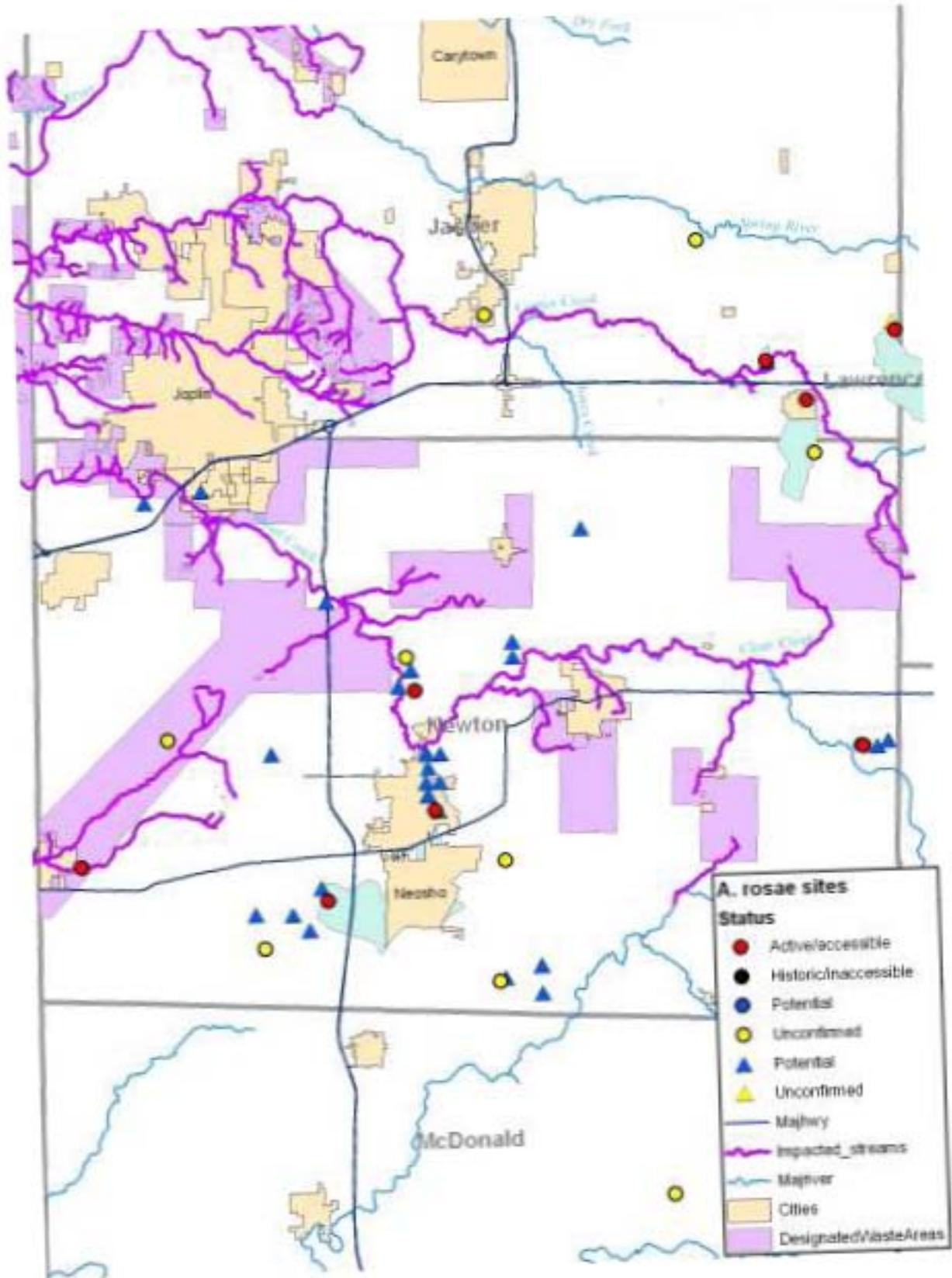


Figure 1. Locations of Ozark cavefish sites and mining impacted areas (Designated Waste Areas) in Jasper and Newton counties.

Appendix A. Resource Science budget worksheet.

## Resource Science Project or Management Evaluation Proposal Budget Projection

**Name of Project / Management Evaluation:**

Account number: \_\_\_\_\_ WPI (if known): \_\_\_\_\_ File Name: \_\_\_\_\_

|                                | FY10        | FY11        |  |  |  |                        | Total per Category |
|--------------------------------|-------------|-------------|--|--|--|------------------------|--------------------|
| <b>Budget Category</b>         |             |             |  |  |  |                        |                    |
| 1 <b>Expense</b>               | \$19,000.00 | \$17,700.00 |  |  |  |                        | \$36,700.00        |
| 2 <b>Temporary Labor</b>       | \$23,190.00 | \$23,190.00 |  |  |  |                        | \$46,380.00        |
| 3 <b>Equipment</b>             | \$0.00      | \$0.00      |  |  |  |                        | \$0.00             |
| <b>Funds Requested</b>         |             |             |  |  |  |                        |                    |
| 4 <b>Total per Fiscal Year</b> | \$42,190.00 | \$40,890.00 |  |  |  | <b>Total all Years</b> | \$83,080.00        |
| 5 <b>Outside Funding*</b>      | \$27,914.00 | \$27,414.00 |  |  |  |                        | \$55,328.00        |
| 6 <b>% Outside Funds</b>       | 66.16%      | 67.04%      |  |  |  | <b>%Outside Funds</b>  | 66.60%             |
| 7 <b>Objectives</b>            | 1,2         | 1,2         |  |  |  |                        |                    |

8 **Outside Funding\*:** Natural Resource Damage Assessment (USFWS)

9 **In-kind contributions\*\*** \_\_\_\_\_

10 **Expense Needs:** Contract to DNR \$12,000 each year, field gear and supplies \$7,000 FY10 and \$5,700 FY11.

11 **Equipment Needs:**  
**Existing:** 1 GPS unit to be checked out from RS  
**New or Additional:** \_\_\_\_\_

12 **Additional Computer/Office Equipment:** none

13 **Additional Laboratory/Storage Needs:** none

14 **Additional Office Space Needs:** none

15 **Location of Temporary Position:** Southwest Region (Neosho)

16 **Details for Temporary Labor by Fiscal Year:**  
FY10 Need 1 / 975 hr @ 8.49/hr = \$8,278 and 1 / 1600 hr @ \$9.32/hr = \$14,912  
FY11 Need 1 / 975 hr @ 8.49/hr = \$8,278 and 1 / 1600 hr @ \$9.32/hr = \$14,912

17 **Vehicle Needs by Fiscal Year:**  
1 1/2 ton, 4x4, with camper shell 12 months each year

**Comments:** \_\_\_\_\_

\* Outside funded partners are those contributing money to the project and must be budgeted for in RSD budget.  
 \*\* In-kind contributions are not dollars, but are other means of supporting the project and are not budgeted for in the RSD budget.