4th Hellbender Symposium

2009

Cumberland Falls State Park

Corbin, Ky

4TH HELLBENDER SYMPOSIUM

21-24 June 2009. Cumberland Falls State Resort Park, Corbin, Kentucky.

PROGRAM

(Note: all activities will take place in the Blair Building)

Sunday	**21 June 2009**
4:00 PM	**Check in for those lodging at Cumberland Falls State Resort Park**
7:30	**Welcoming Remarks & Field Trip Details**
8:00 -11:00	**Social/Mixer**
Monday	**22 June 2009**
8:30 AM	**Depart for field trips to local rivers**
5:00 PM	**Return to Cumberland Falls State Resort Park**
6:00	**Catered buffet dinner in Blair Building**
7:30	**Mapping the Range-Wide Status of Hellbender Populations** Jeff Humphries
8:00	**Social/Mixer**
Tuesday	**23 June 2009**
8:30 AM	**Opening Remarks** John MacGregor, Herpetologist, Kentucky Department of Fish & Wildlife Resources
	Session I: Surveys and Status Assessments
8:45	**A Status Summary Update of the Hellbender (*Cryptobranchus alleganiensis*) in Maryland** Daniel J. Feller and Edward L. Thompson
9:05	**Population Monitoring of Ozark Hellbenders in the Eleven Point River in Arkansas and Missouri** Kelly Irwin and Jeffrey T. Briggler
9:25	**The Decline of the Eastern Hellbender in Ohio** Gregory J. Lipps, Jr. and Ralph A. Pfingsten
9:45	**Landscape Factors Affecting Eastern Hellbender Distribution in the Susquehanna River Watershed of New York State** Sam Quinn
10:05	**Morning Break**
	Session I: Surveys and Status Assessments (continued)
10:25	**Conservation Status of the Hellbender in North Carolina** Jeff Humphries and Lori Williams
10:45	**'Round-the-Room State Status Updates *(Please limit to 5 minutes each)*:** Alabama, Arkansas, Georgia, Illinois, Indiana, Kentucky, Missouri, Mississippi, New York, Pennsylvania, Tennessee, Virginia, West Virginia
12:00 PM	**Lunch** (on your own)

	Session II: Genetics
1:30	**Preliminary Analysis of Genetic Variation of Hellbenders in the Tennessee Valley** Michael J. Freake and Eric J. Routman
1:50	**Genetic Sex Diagnosis of Cryptobranchid Salamanders** Paul M. Hime, Joshua S. Reece, and Jeffrey T. Briggler
2:10	**Conservation Genetics of Hellbender Salamanders in New York: The Use of mtDNA for Population Structure and Diversity Estimates** Noelle L. Rayman and Amy McMillan
2:30	**Afternoon Break**
	Session III: Health and Disease
2:50	**Health Assessment of Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*) Populations in Ohio and West Virginia** Rachael B. Weiss, DVM, Tiffany M. Wolf, Allan P. Pessier, Joe Greathouse, Gregory J. Lipps, Jr., and Barbara A. Wolfe
3:10	**Historic Occurrence of Chytridiomycosis in Hellbenders from Missouri** Catherine M. Bodinof, Jeffrey T. Briggler, Mary Duncan, Jeff Beringer, and Joshua J. Millspaugh
	Session IV: Ecology and Behavior
3:30	**Movement Patterns, Habitat Use, and Home Range Size in Eastern Hellbenders** Nicholas G. Burgmeier and Rod N. Williams.
3:50	**Predator Recognition in Larval Hellbenders from Missouri and the Problem of Introduced Trout** Alicia Mathis and Brian Gall
4:10	**Learned Responses of Larval Hellbenders (*Cryptobrancus alleganeinsis*) to Introduced Trout** Adam Crane and Alicia Mathis
4:30	**End of session**
6:00	**Catered buffet dinner in Blair Building**
7:00	**Video Night** (bring your Hellbender media clips and movies to share)
Wednesday	**24 June 2009**
	Session V: Recovery, Husbandry, and Headstarting
8:30 AM	**General Observations of Captive Reared Ozark Hellbenders Released in the North Fork of the White River, Missouri** Cathy Bodinof, Jeffrey T. Briggler, Randall E. Junge, Jeff Beringer, Mark D. Wanner, Chawna D. Schuette, and Joshua Millspaugh
8:50	**Update from the Ron Goellner Center for Hellbender Conservation** Chawna Shuette
9:10	**Hellbender Propagation Efforts at Shepherd of the Hills Hatchery, Branson, Missouri: Lessons Learned** Jeffrey T. Briggler , James A. Civiello, and Scott A. Crain
9:30	**Potential Impact of Capture and MS-222 Exposure on Hellbender (*Cryptobranchus alleganiensis*) Populations in the Ozarks** J. Kelly Byram and Max A. Nickerson

9:50	**Conservation Status and Farming of the Giant Salamander (*Andrias davidianus*) in China** Robert Browne
10:10	**Break**
	Session VI: Laws, Regulations, and Trade
10:30	**Federal Candidate Assessment for the Eastern Hellbender** Jeromy M. Applegate
10:50	**The Demand for Cryptobranchid Salamanders in Japan** Timothy Johnson
11:10	**CITES and the Implications of a CITES Listing for hellbenders** Craig Hoover
11:30	**Wrap-up:** The Cumberland Falls Resolution 5th Hellbender Symposium - Host, Location, and Date (19-22 June 2011?)
12:30 PM	**Adjourn**

ABSTRACTS

Session I: Surveys and Status Assessments

Mapping the Range-Wide Status of Hellbender Populations

Jeff Humphries (humphri2@yahoo.com)
North Carolina Wildlife Resources Commission, 404 Barclay Road, Chapel Hill, NC 27516

Where do the best hellbender streams still exist? Where are survey efforts lacking? How does the abundance of hellbenders relate to broad trends of watershed land-use and land ownership? These are fundamental questions that need to be answered over the hellbender's entire range if we hope to preserve healthy populations, restore degraded stream systems, and prioritize conservation efforts and funding. Over the past year, I have been compiling hellbender survey data from numerous researchers throughout the range of the species and mapping the relative abundance (number of hellbenders per person hour of searching) of hellbenders at individual study sites. Thus far, 674 study sites have been mapped with an associate abundance estimate. I welcome input and collaboration among researchers to improve upon this mapping effort so that it accurately reflects the conservation status of hellbenders.

A Status Summary Update of the Hellbender (*Cryptobranchus alleganiensis*) in Maryland

Daniel J. Feller (dfeller@dnr.state.md.us) and Edward L. Thompson
Maryland Department of Natural Resources, Natural Heritage Program, c/o Appalachian Lab, University of Maryland, 301 Braddock Road, Frostburg, MD 21532

In Maryland, the Hellbender (*Cryptobranchus allegheniensis*) has suffered dramatic declines, including a significant range reduction within the state and declining populations at extant sites. Historically, hellbenders inhabited the Youghiogheny River and Casselman River, both tributaries of the Ohio River, but also occurred within the Susquehanna River and its tributaries on the Atlantic slope. Surveys, results of a composite of reliable sighting reports, electrofishing, and intensive log peavey-aided rock lifting conducted from the 1980's to present have found the species current range limited to a 4 km section of the mainstem Casselman River, approximately 8 km of the Youghiogheny River, and absent from the Susquehanna River and its tributaries. New unconfirmed reports from the North Branch of the Casselman River may extend the species range upstream in that drainage. Densities are low in both extant populations, with only 31 individuals captured after lifting 7,300 rocks in a period between 1997 and present. Reproduction appears to be inhibited as the last observed nests were observed in the early to mid-1990's and all but one capture or sighting in recent decades have been of large adults. A proposed 3,000 acre coal deep mine along and under the North Branch Casselman River further threatens the continued existence of the entire Casselman River population.

Population Monitoring of Ozark Hellbenders in the Eleven Point River in Arkansas and Missouri

Kelly Irwin[1] (kirwin@agfc.state.ar.us) and Jeffrey T. Briggler[2]
[1]Arkansas Game & Fish Commission, 915 E. Sevier St., Benton, AR 72015
[2]Missouri Department of Conservation, PO Box 180, Jefferson City, MO 65102

In 2005, a long-term population monitoring program was established to provide the mechanism for standardized, quantifiable assessment of the Ozark Hellbender population in the Eleven Point River in Randolph County, Arkansas. Information from this program will be presented in conjunction with data from standardized surveys conducted in the Eleven Point River in Missouri.

The Decline of the Eastern Hellbender in Ohio

Gregory Lipps (GregLipps@aol.com) and Ralph A. Pfingsten
Gregory Lipps, LLC, 1473 County Road 5-2, Delta, OH 43515

Eastern hellbenders were historically found throughout the two-thirds of Ohio that drain to the Ohio River. Surveys in the mid-1980's resulted in the capture of 117 hellbenders in 16 eastern Ohio waterways. In 2006, with the support of the Ohio Division of Wildlife, surveys were initiated to verify the continued presence of these populations, as well as follow-up on anecdotal accounts of occurrence from additional locations. A total of 1149 person-hours have been spent searching for hellbenders in the past three seasons, resulting in 62 captures in 9 waterways. The relative abundance of hellbenders appears to have declined by over 78% in the past two decades. Furthermore, over 70% of the hellbenders observed have come from two watersheds, where individuals as small as 24 cm (total length) have been found. In the remainder of the state all captured individuals measured >47 cm. This shift in demographics to larger individuals indicates a lack of successful recruitment in nearly all of Ohio's extant Hellbender populations. Considering that Hellbenders have already been extirpated from the western-half of the state, identifying the causes of the declines and implementing effective strategies to safeguard Ohio's last remaining populations are the focus of current projects in the state.

Landscape Factors Affecting Eastern Hellbender Distribution in the Susquehanna River Watershed of New York State

Sam Quinn (saquinn@syr.edu)
State of New York College of Environmental Science and Forestry, 1 Forestry Drive, Syracuse, NY 13210

Eastern hellbender populations are declining range-wide and appear to be particularly imperiled in the New York State portion of the Susquehanna River watershed. In the summer of 2008 we surveyed 52 stream segments in the Susquehanna River watershed of New York including 22 historic population sites and successfully located only two individuals. Our research takes a broad-scale perspective to address the influence of the terrestrial landscape on eastern hellbender habitat quality. We are using Geographic Information System (GIS) software in conjunction with field data to quantify the relative importance of habitat elements known to be associated with eastern hellbender occurrence in order to build a better understanding of the causes of population persistence and extinction. Certain land use types such agriculture and developed impervious surface can influence stream conditions and in turn negatively affect the quality of eastern hellbender habitat by accelerating the amount of eroded sediments entering streams. In order to better inform management policies we are examining the association between landscape features and stream conditions at multiple scales in relation to population sites. Preliminary analyses indicate that the forested riparian buffers thought to be important to population persistence may have little influence on stream conditions in a watershed that has a high proportion of agricultural and developed land area. We feel that by taking this region-wide perspective we may best elucidate causes for population persistence and decline across the eastern hellbender's range and provide a basis for broad-scale habitat protection.

Conservation Status of the Hellbender in North Carolina

Jeff Humphries (humphri2@yahoo.com) and Lori Williams
North Carolina Wildlife Resources Commission, 404 Barclay Road, Chapel Hill, NC 27516

Hellbenders are state listed as a Species of Special Concern in North Carolina. Robust hellbender populations are well known from a few of the state's watersheds, however, exhaustive surveys have not been completed in most of North Carolina's streams until recently. We surveyed for hellbenders at 93 sites, on 35 streams, in western North Carolina during 2007-08 using a combination of rock lifting and snorkeling. Hellbenders were documented at 62 of the 93 sites surveyed. Though hellbenders still occur in most of their historic range in North Carolina, the majority of our surveys resulted in only one or a few individuals, despite intensive survey efforts. Detailed information about the status of hellbenders in North Carolina, as well as some interesting life history notes, will be discussed.

Session II: Genetics

Preliminary analysis of genetic variation of hellbenders in the Tennessee Valley

Michael J. Freake[1] (mfreake@leeuniversity.edu) and Eric J. Routman[2]
[1]Lee University, 1120 Ocoee St, Cleveland, TN 37311
[2]San Francisco State University 1600 Holloway Avenue, San Francisco, CA 94132

A preliminary analysis was conducted of mitochondrial genetic variation in eastern hellbenders, *Cryptobranchus alleganiensis alleganiensis*, from populations across the Tennessee Valley, in both Tennessee and North Carolina. We sequenced cytochrome b and cytochrome oxidase I regions of individuals from eight different rivers. When both regions were successfully sequenced for an individual, the data were concatenated. Our preliminary data suggest that the rivers differ in the level of within population genetic variation, and there is evidence of significant genetic differentiation between rivers, despite close geographic proximity.

Genetic Sex Diagnosis of Cryptobranchid Salamanders

Paul M. Hime[1] (herpkeepers@stlzoo.org), Joshua S. Reece[2], and Jeffrey T. Briggler[3]
[1]Saint Louis Zoological Park, Department of Herpetology and Aquatics, One Government Drive, Saint Louis, MO 63110
[2]Washington University in St. Louis, Department of Biology, Campus Box 1137, One Brookings Drive, Saint Louis, MO 63130
[3]Missouri Department of Conservation, 2901 West Truman Boulevard, P.O. Box 180, Jefferson City, MO 65102

This project seeks to identify genetic markers that reliably diagnose sex in *Cryptobranchus* and *Andrias*. Genetic sex diagnosis is well established for a wide variety of vertebrates and has significant implications for species conservation and research. To date, no method exists for genetic sexing of salamanders. Cryptobranchids are diploid organisms that exhibit a ZZ/ZW sex determining system in which females possess two different sex chromosomes and males possess an identical pair. By locating and characterizing regions of DNA unique to the W chromosome of females, one can diagnose sex by routine genetic techniques. Once sex-specific fragments of DNA are detected, those regions will be sequenced to generate primers for a reliable PCR-based assay for sex. In short, genomic DNA will be extracted from animals of known sex and screened for genetic regions that are polymorphic

between the sexes. Several potential avenues for generating polymorphic DNA fragments will be tested sequentially including RAPD, AFLP and homologous gene mapping. An initial sample from *Cryptobranchus alleganiensis bishopi* consisting of twenty males and twenty females will be used to locate female-specific fragments. After these fragments are analyzed and PCR primers are generated, a blind trial using coded samples of known sex from both currently recognized subspecies of *Cryptobranchus* will be conducted to assess the reliability of this technique. Another component of this study will test the effectiveness of these techniques in both species of *Andrias*. Given the close relationships within Cryptobranchidae, we expect this diagnostic to be widely effective across the family. The statistical significance of these results will be assessed using standard analyses. Once verified for *Cryptobranchus*, these primer sequences will be published, allowing other researchers access to this powerful tool.

Conservation Genetics of Hellbender Salamanders in New York: the Use of mtDNA for Population Structure and Diversity Estimates

Noelle L. Rayman (nlrayman@hotmail.com) and Amy McMillan
Buffalo State College, 1300 Elmwood Avenue, Buffalo, NY 14203

In New York State, the Eastern hellbender, *Cryptobranchus a. alleganiensis*, has been listed as special concern since 1983 due to small population size within the Allegheny and Susquehanna River Watersheds. Historic data indicate that hellbenders were once well-documented within these watersheds. Studies conducted in the Allegheny drainage within the last 20+ years have shown declines in the number of individuals, and age class distributions skewed towards older adults and few larvae/young adults. Polluted aquatic habitat, migration barriers, harvesting, and the loss of natural rock habitat used for nesting may be impacting these small populations. In order to protect this species, factors such as genetic diversity and population structure, in conjunction with demographic data, need to be better understood. This study is the first to investigate the genetic relationships of hellbender populations in New York. Thirty-eight tissue and blood samples collected from the Allegheny and Susquehanna River Watersheds in New York and Pennsylvania were used for DNA analysis of the mitochondrial cytochrome oxidase subunit I (COI) gene. Hellbender specific COI primers were used to sequence approximately 700 base pairs of this gene in order to evaluate genetic diversity and population structure of hellbenders within and between these watersheds. We hypothesize that genetic diversity within the drainages to be low and that genetic differentiation will exist between watersheds. However, initial results indicate similar haplotypes exist between the Allegheny and Susquehanna drainages. An understanding of genetic diversity and population structure may assist in determining if genetic factors are contributing to population declines and in developing a state recovery plan which includes possible reintroductions of the most genetically diverse individuals to historic locations or currently unoccupied stream reaches that contain optimal habitat.

Session III: Health and Disease

Health Assessment of Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*) Populations in Ohio and West Virginia

Rachael B. Weiss, DVM[1] (rweiss@thewilds.org), Tiffany M. Wolf, DVM,[2] Allan P. Pessier, DVM, DACVP,[3] Joe Greathouse,[4] Gregory J. Lipps, Jr.,[5] and Barbara A. Wolfe, DVM, PhD, DACZM[1]
[1]The Wilds, 14000 International Rd., Cumberland, OH 43732
[2]Minnesota Zoo, 13000 Zoo Blvd., Apple Valley, MN 55124

[3]Wildlife Disease Laboratories, Zoological Society of San Diego, P.O. Box 120551, San Diego, CA 92112
[4]Good Zoo at Oglebay Resort, Rt. 88 North, Wheeling, WV 26003
[5]1473 County Road 5-2, Delta, OH 43515

Health assessments were performed on hellbenders captured as a part of ongoing hellbender population surveys in Ohio and West Virginia during the natural breeding season in 2006-2008. Examinations were conducted on all individuals and included body weight, snout-vent length, blood collection for hematology and serum chemistry, skin swabs for chytridiomycosis (*Batrachochytrium dendrobatidis*) identification (PCR) and cloacal swabs for ranavirus (family iridoviridae) detection (PCR). To date, a total of 73 animals have been sampled in Ohio and West Virginia [n=32 (OH), n=41 (WV)]. Hellbender populations in OH and WV differ significantly with respect to hematologic profiles. Specifically, OH hellbenders demonstrate a higher proportion of lymphocytes (μ ± sem: 70.3 ± 2.5%) than WV hellbenders (60.4 ± 2.3%, $p < 0.01$), whereas WV hellbenders show a higher proportion of heterophils (30.2 ± 2.0 vs. 20.4 ± 2.1%, $p<0.01$), total protein (1.46 ± 0.13 vs. 1.01 ± 0.09 g/dl, $p<0.01$) and globulin levels (1.85 ± 0.11 vs.1.55 ± 0.06 g/dl, $p < 0.05$). Interestingly, during the sampling period of June to September, it appears that serum calcium levels are higher in female (9.9 ± 0.5 mg/dl), than in male hellbenders (7.3 ± 0.1 mg/dl), in both states ($p < 0.01$). *Batrachochytrium dendrobatidis* has been identified in both Ohio (n=1; 2007) and West Virginia (n=3; 2007) hellbender populations. These data provide a reference point that will be utilized to continue the health monitoring program and will help to direct further research to identify specific factors impacting hellbender populations in Ohio and West Virginia.

Historic Occurrence of Chytridiomycosis in Hellbenders from Missouri

Catherine M. Bodinof[1] (cmbodinof@mizzou.edu), Jeffrey T. Briggler[2], Mary Duncan[3], Jeff Beringer[4], and Joshua J. Millspaugh[1]
[1]University of Missouri, Department of Fisheries and Wildlife Sciences, 302 Anheuser-Busch Natural Resources Building, Columbia, MO 65211
[2]Missouri Department of Conservation, 2901 West Truman Boulevard, P.O. Box 180, Jefferson City, MO 65102
[3]Saint Louis Zoological Park, Department of Animal Health and Nutrition, One Government Drive, Saint Louis, MO 63110
[4]Missouri Department of Conservation, Resource Science Center, 1110 South College Avenue, Columbia, MO 65201

Recent declines in both Eastern and Ozark hellbender (*Cryptobranchus alleganiensis alleganiensis* and *C. a. bishopi* respectively) populations may be attributable to one or several causes. However, recent discovery of the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) in Missouri Rivers suggests that this pathogen may be one factor involved. Chytridiomycosis, infection caused by *Bd*, has been implicated as a major factor in numerous amphibian declines and local extinctions around the globe and was detected for the first time in hellbenders from Missouri in 2006. While hellbenders are known to carry *Bd*, its virulence for *C. alleganiensis* is poorly understood. Identifying length of time the fungus has persisted in Missouri streams may assist in identifying the severity of the threat *Bd* poses for hellbender populations. We evaluated over 200 archived hellbenders collected from 7 Missouri Rivers between 1896 and 1993 to identify minimum length of time *Bd* has occurred in hellbender populations. We collected epidermal tissue from each individual and analyzed samples through routine histology (hematoxylin and eosin staining). We positively identified *Bd* in hellbenders collected from 3 Rivers including the North Fork of the White,

Meramec and Current Rivers; earliest documentation of the pathogen came from hellbenders collected in 1969, 1975 and 1988, respectively. North Fork of the White River populations have therefore coexisted with the fungus for at least 4 decades. Although we did not detect *Bd* in samples from earlier dates or 4 other rivers, this does not preclude the possibility that *Bd* was present but undetected due to the limitations of our methods and sample availability.

Session IV: Ecology and Behavior

Movement Patterns, Habitat Use, and Home Range Size in Eastern Hellbenders

Nicholas G. Burgmeier (nburgmei@purdue.edu) and Rod N. Williams
Purdue University, 715 West State Street, West Lafayette, IN 47907

Eastern hellbenders (*Cryptobranchus alleganiensis alleganiensis*) are large aquatic salamanders that have suffered substantial population declines throughout their range. Many aspects of their biology have been studied, but little is known regarding habitat use, dispersal, and home range size within small isolated populations. We sampled for adult hellbenders at 40 locations along the Ohio River drainage during 2008. We surgically implanted radio transmitters in 17 hellbenders throughout a 112-km stretch of river. Individuals were tracked 2-3 times per week for up to 10 months. We discuss seasonal movement patterns, habitat use, and home range size in light of the physical characteristics associated with both macrohabitat and microhabitat types. Radio-tracking is expected to continue for an additional 12 months and will provide valuable insight into the breeding biology and movement patterns of eastern hellbenders within an agriculturally-dominated landscape.

Predator Recognition in Larval Hellbenders from Missouri and the Problem of Introduced Trout

Alicia Mathis[1] (AliciaMathis@MissouriState.edu) and Brian Gall[1,2]
[1]Missouri State University, Department of Biology, Springfield, MO 65897
[2]Utah State University, Department of Biology, Utah State University, 5305 Old Main Hill, Logan, UT 84322

Predator introductions pose a unique threat to prey in permanent aquatic habitats because predation is part of predator-prey evolutionary history, but innate predator recognition may make them incapable of recognizing the recently introduced predator. Here we present a study evaluating the ability of larval hellbenders (*Cryptobranchus alleganiensis*) to recognize chemical cues from native and introduced fish predators. We presented larvae from both subspecies of hellbenders with chemical stimuli from native and nonnative predatory fishes, a nonpredatory fish and a blank control. Compared to a nonpredatory fish (redhorse sucker, *Moxostoma* spp.), eastern hellbender larvae (*C. a. alleganiensis*) significantly reduced activity in response to chemical stimuli from native predators but did not do so in response to stimuli from nonnative trout. Responses of Ozark hellbender larvae to brown trout (*Salmo trutta*) were similar to that of the native fishes and different from the blank control, but responses to rainbow trout (*Oncorhynchus mykiss*) did not differ from the blank control. The general inability of larval hellbenders to recognize introduced predatory trout could lead to increased predation in the wild.

Learned Responses of Larval Hellbenders (*Cryptobranchus alleganiensis*) to Introduced Trout

Adam Crane (AdamCrane@missouristate.edu) and Alicia Mathis
Missouri State University, Department of Biology, Springfield, MO 65897

One potential contributor to the decline of the hellbender in Missouri may be the introduction of non-native trout, which may not be recognized as predators by larval hellbenders. Head-starting has been proposed as a conservation measure to increase recruitment into declining hellbender populations. However, success of head-starting could be low if released larvae do not take appropriate antipredator measures during encounters with trout. We used a classical conditioning protocol to train laboratory reared hellbenders to fear trout. Our protocols included exposing hellbender larvae to trout-scented water plus a hellbender distress secretion during training trials. In a subsequent test, these larvae responded to trout cues alone with a fright response. Learning was specific to trout because trained larvae did not respond to water that had been scented by a suckermouth catfish. Hellbenders did not exhibit learned responses in other trials where training consisted of exposure to the trout stimulus and control water. Although a number of details remain to be addressed concerning standardized procedures, we recommend that all head-starting programs for hellbenders, whose native habitat is outside of the natural range of trout, include trout-recognition training.

Session V: Recovery, Husbandry, and Headstarting

General Observations of Captive Reared Ozark Hellbenders Released in the North Fork of the White River, Missouri

Cathy Bodinof[1] (cmbodinof@mizzou.edu), Jeffrey T. Briggler[2], Randall E. Junge[3], Jeff Beringer[4], Mark D. Wanner[5], Chawna D. Schuette[5], and Joshua Millspaugh[1]
[1]University of Missouri, Department of Fisheries and Wildlife Sciences, 302 Anheuser-Busch Natural Resources Building, Columbia, MO 65211
[2]Missouri Department of Conservation, 2901 West Truman Boulevard, P.O. Box 180, Jefferson City, MO 65102
[3]Saint Louis Zoological Park, Department of Animal Health and Nutrition, One Government Drive, Saint Louis, MO 63110
[4]Missouri Department of Conservation, Resource Science Center, 1110 South College Avenue, Columbia, MO 65201
[5]Saint Louis Zoological Park, Department of Herpetology and Aquatics, One Government Drive, Saint Louis, MO 63110

Populations of Ozark hellbenders (*Cryptobranchus alleganiensis bishopi*) in Missouri have recently undergone dramatic declines. Hellbenders are ranked as a critically imperiled species of conservation concern (S1) and endangered within the state; and *C. a. bishopi* is a candidate for federal listing under the Endangered Species Act. While ongoing research is aimed at identifying the causes for observed declines, we are evaluating the viability of repatriation using captive-reared hellbenders to supplement wild hellbender populations. We collected eggs from the wild and reared hellbenders from 2003 to 2008 at the Saint Louis Zoo's Ron Goellner Center for Hellbender Conservation. Between May and October of 2008 we released 36 Ozark hellbenders (weighing 142 – 334 g, and measuring 29-36 cm total length) implanted with radio-transmitters at 2 sites on the North Fork of the White River, Missouri. We used radio-telemetry to relocate individuals and monitor survival, movements, accrual of physical abnormalities, parasites and infection of the amphibian chytrid fungus (*Batrachochytrium dendrobatidis*) since release. As of April 2009, we are monitoring 24

radio-tagged hellbenders; 22 of which are confirmed alive based on visual observations or recent movements. We have confirmed 9 mortalities and are unable to detect radio signals from 3 hellbenders, presumably due to transmitter failure. Recent recaptures revealed 15 of 27 hellbenders had gained weight and accrued few to no abnormalities since release. While success of repatriation relies on long term survival and reproduction of individuals, our preliminary findings suggest that captive rearing may be a valuable tool for producing hellbenders that can survive independently in the wild.

Update from the Ron Goellner Center for Hellbender Conservation

Chawna Schuette (herpgirlchawna@yahoo.com)
Saint Louis Zoological Park, Department of Herpetology and Aquatics, One Government Drive, Saint Louis, MO 63110

Captive propagation efforts at the Saint Louis Zoo have expanded since the last Hellbender Symposium in 2007. The Ozark hellbenders at the Saint Louis Zoo have oviposited at least three confirmed times from three different females over two breeding seasons. While viable, motile milt has been collected from three of the six adult males at the Saint Louis Zoo, no eggs have been fertilized. Some attempts at artificial fertilization have been made in 2008, but the attempts weren't successful for a variety of reasons which will be described in more detail during presentation. Twenty-four hour monitoring of the adult hellbender population is now being used during the breeding season to ensure that oviposition will be observed by staff so any necessary interventions can take place. Many interesting behaviors have been observed anecdotally which have led to a pilot study for a research project being developed to explore hellbender behavior and to determine the importance of social structure and behavior changes prior to and during breeding season. Heat treatment has been used to eradicate the amphibian chytrid fungus, *Batrachochytrium dedrobatidis* (*Bd*), from the juvenile hellbenders housed at the center. The animals treated under three different sets of treatments with variables that will be explained in presentation all show negative results when PCR tested for *Bd* for over one year in some animals post treatment. All animals treated are still negative. The method for this heat treatment will also be described in greater detail. Some interesting growth data has also been collected over the last several years with a large population of captive animals. We have seen trends in adult Ozark hellbender weight fluctuations over the course of the years that show consistency from year to year. These charts will be presented in the power point or will be available in poster format for people to view. There are also differences in the growth rates of Eastern hellbenders when compared to Ozark hellbenders which will be briefly reviewed.

Hellbender Propagation Efforts at Shepherd of the Hills Hatchery, Branson, Missouri: Lessons Learned

Jeffrey T. Briggler[1] (jeff.briggler@mdc.mo.gov), James A. Civiello[2] and Scott A. Crain[2]
[1]Missouri Department of Conservation, 2901 West Truman Boulevard, P.O. Box 180, Jefferson City, MO 65102
[2] Missouri Department of Conservation, 425 Hatchery Road, Branson, MO 65616

Declining numbers of wild hellbenders (*Cryptobranchus alleganiensis*) in Missouri have necessitated the use of captive propagation efforts to insure long-term recovery of this species. In the autumn of 2007, propagation efforts for hellbenders began at the Missouri Department of Conservation's Shepherd of the Hills Hatchery, Branson, Missouri. This facility, as well as the Ron Goellner Center for Hellbender Conservation at the St. Louis Zoo in Missouri, is dedicated to the long-term propagation needs of the species in Missouri. Propagation efforts have included hatching of Ozark (*C. a. bishopi*) and eastern hellbender

(*C. a. alleganiensis*) eggs, head-starting eastern hellbender young, and holding eastern hellbender adults for breeding. In addition, this facility provides opportunities for hellbender research. Here we discuss lessons learned from our captive breeding program pertaining to egg handling and incubation, larval diet preferences, spatial requirements, growth rates, and behavior. In addition, we will provide information on adult breeding hellbenders and challenges related to diseases. As with many propagation efforts, we continue to investigate options for future expansion needs and other uncertainties. Successful propagation is needed not only to augment wild populations, but also to provide larvae/juvenile hellbenders for research purposes to better address the reasons for the decline of the species for long-term recovery.

Potential impact of capture and MS-222 exposure on Hellbender (*Cryptobranchus alleganiensis*) populations in the Ozarks

J. Kelly Byram and Max A. Nickerson (maxn@flmnh.ufl.edu)
Florida Museum of Natural History, Gainesville, FL, 32611, USA

We surveyed field studies of hellbenders in Missouri from 1971 to present to determine how many animals had been captured and exposed to a sedative commonly used in field studies of amphibians, tricaine methanesulfonate (MS-222). While MS-222 is generally regarded as safe and is suggested by IACUC and USGS for use on a wide range of aquatic amphibian species, our survey of the literature found a number of potentially deleterious effects from the drug. In studies on multiple species, MS-222 has been shown to elevate fractional water absorption, impact vision and behavior, inhibit Gram-negative bacteria, and interfere with the accurate determination of parasite load of some protozoa, metazoa, rotifers, nematodes, and leeches. We conclude that, given the large numbers of animals captured and treated with MS-222 and the ample interdisciplinary research already available, there is reason for researchers to greatly reduce the use of MS-222 in the field in most situations.

Conservation Status and Farming of the Giant Salamander in China

Robert Browne (robert.browne@gmail.com)
IUCN/WAZA Amphibian Ark, Royal Zoological Society of Antwerp, Koningin Astridplein 26, 2018 Antwerp Belgium

The Critically Endangered, Chinese giant salamander (*Andrias davidianus*) is the largest extant amphibian. Natural populations of *A. davidianus* have suffered from both habitat loss and harvesting for consumption. However, over the last decade technologies have been developed that enable the reproduction of *A. davidianus* and their raising on farms. Since 1998 there has also been considerable improvement of natural habitat suitable for *A. davidianus* both through increased protection, reafforestation, and the development of eco-tourism in reserves. The farming of *A. davidianus* is mostly dependent on the use of artificial reproduction through hormonal induction, which reliably produces fertile eggs from the majority of females. There are a large number of adult wild caught broodstock *A. davidianus* in farms, however, their source populations are often uncertain. As *A. davidianus* will reproduce after 5 years F2 salamanders are already producing eggs. A mandate of the use for farms is the release of some individuals to rehabitate streams, rivers and lakes, and juveniles are already being conditioned for natural feed and released. Therefore, to avoid the loss of genetic diversity there is an immediate need to assess and manage the genetics of both relict natural populations of *A. davidianus* and wild caught broodstock in farms.

Session VI: Laws, Regulations, and Trade

Federal Candidate Assessment for the Eastern Hellbender

Jeromy M. Applegate (jeromy_applegate@fws.gov)
U.S. Fish and Wildlife Service, Ohio Ecological Services Field Office, 4625 Morse Rd., Suite 104, Columbus, OH 43230

The U.S. Fish and Wildlife Service is currently evaluating whether the status of the eastern hellbender (*Cryptobranchus alleganiensis alleganiensis*) warrants its designation as a candidate for listing under the Federal Endangered Species Act. A candidate species is one for which we have all the information needed to propose for listing as threatened or endangered, but do not propose because listing is precluded by other candidate species or listing actions that have higher priority. The current candidate evaluation updates and expands on the 2004 Eastern Hellbender Status Assessment using the best available data gathered from hellbender researchers and managers throughout its range. We are determining historical/current range and distribution and population estimates and status, and conducting a five-factor analysis of threats to the species. These factors include (1) the present or threatened destruction, modification, or curtailment of its habitat or range, (2) overutilization for commercial, recreational, scientific, or educational purposes, (3) disease or predation, (4) the inadequacy of existing regulatory mechanisms, and (5) other natural or manmade factors affecting its continued existence. The Fish and Wildlife Service anticipates a decision on eastern hellbender candidate status by the end of calendar year 2009.

The demand for Cryptobranchid salamanders in Japan

Timothy Johnson (timtim@gol.com)
Minami-Oya 1449-11, Machida-shi, Tokyo, Japan, 194-0031

CITES and the implications of a CITES listing for hellbenders

Craig Hoover (craig.hoover@fws.gov)
Branch of Operations, Division of Management Authority, US Fish and Wildlife Service, 4401 North Fairfax Drive, Arlington, VA 22203

The Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) is an international treaty involving 175 member nations or Parties. CITES works by subjecting international trade in specimens of selected species to certain controls. All import, export, re-export and introduction from the sea of species covered by the Convention has to be authorized through a licensing system. The species covered by CITES are listed in three Appendices, according to the degree of protection they need. Appendix I includes species threatened with extinction. Trade in specimens of these species is permitted only in exceptional circumstances. Appendix II includes species not necessarily threatened with extinction, but in which trade must be controlled in order to avoid utilization incompatible with their survival. Appendix III contains species that are protected in at least one country, which has asked other CITES Parties for assistance in controlling the trade. A specimen of a CITES-listed species may be imported into or exported (or re-exported) from a State party to the Convention only if the appropriate document has been obtained and presented for clearance at the port of entry or exit. Because there is international demand for hellbenders, CITES listing, and the trade control and monitoring that come with such a listing, may benefit the conservation of this species.

POSTERS

Population Dynamics of the Eastern Hellbender in the West Branch of the Susquehanna River

Andrew Llewellyn[1] and Peter J. Petokas[2] (petokas@lycoming.edu)
[1]Department of Biology, Susquehanna University, Selinsgrove, PA 17870
[2]Department of Biology, Lycoming College, Williamsport, PA 17701

We surveyed for Eastern Hellbenders (*Cryptobranchus alleganiensis*) in a major tributary of the Susquehanna River West Branch in north-central Pennsylvania from May through August 2007-2008. Two hellbender meta-populations, located 8 km apart, were surveyed to determine population size, density, and age and size structure. We lifted large cover rocks and captured at total of 242 adults and juveniles, and PIT-tagged most individuals >200mm total body length (TBL) as part of a long-term study of population dynamics. Using the Lincoln-Petersen method, we estimated population size to be 191 (95% CI=145-310) for the upstream population and 389 (95% CI=290-669) for the downstream population. Relative density was 1.47 hellbenders captured per $100m^2$ at the upstream site and 3.39 hellbenders captured per $100m^2$ at the downstream site. Hellbenders <300 mm total body length made up only 1.0% of all captures at the upstream site, but represented 15.9% of all captures at the downstream site. Mean hellbender size for the upstream site (TBL=399.5mm) did not differ significantly ($t=59.131$, $P<0.000$) from mean hellbender size for the downstream site (TBL=386.0mm). The male-to-female sex ratio did not differ significantly from 1:1 for the downstream site, but males outnumbered females 2:1 at the upstream site. The larger population size and higher density observed at the downstream site may be due to a higher density of large rock cover, while the smaller proportion of juveniles and sub-adults observed at the upstream site may be due to a low density of small cover rocks preferred by smaller individuals.

Ecology and Genetics of the Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*)

Shem Unger (sunger@purdue.edu) and Rod N. Williams
Department of Forestry and Natural Resources, Purdue University, West Lafayette, IN 47907

The eastern hellbender is a large paedomorphic salamander experiencing population declines throughout its range. Nationally, purported causes for declines include habitat destruction, degradation, and illegal harvesting. In Indiana, hellbenders are critically endangered and restricted to small areas located within the Ohio River drainages. Little is known about the reproductive status, population demography, or genetic composition of eastern hellbenders in Indiana. To address these areas of uncertainty, we will use a combination of field sampling, simulation modeling, and molecular tools to investigate the ecology and genetics of eastern hellbenders. Life history data collected during the field component of this project will be used to develop a stage-structured life history model and assess the long-term population viability of eastern hellbenders in Indiana. Questions regarding habitat quality and food availability will be evaluated by extensively sampling and comparing benthic macro-invertebrate communities from within Indiana and neighboring states. We will develop a suite of hypervariable genetic markers (microsatellites) to indirectly assess population size and gene flow as well as levels of genetic variation and structure at local and regional scales. By utilizing a combination of approaches, we can address questions that were previously intractable and thus further our knowledge with regards to hellbender ecology.

Directory of Registrants

Last Name	First Name	Company/Organization	Address Line 1	Address Line 2	City, State	Zip	Phone	Email
Applegate	Jeromy	U.S. Fish and Wildlife Service	4625 Morse Rd, Suite 104		Columbus, OH	43230	614-416-8993	jeromy_applegate@fws.gov
Armstrong	Mike	U.S. Fish and Wildlife Service	J.C. Watts Federal Building, Room 265	330 W. Broadway	Frankfort, KY	40601	502-695-0468	mike_armstrong@fws.gov
Baxley	Danna	Kentucky Dept. Fish and Wildlife Resources	1 Sportsman's Lane		Frankfort, KY	40601	502-564-7109	danna.baxley@ky.gov
Bodinof	Catherine	University of Missouri	302 ABNR	Dept. Fisheries & Wildlife	Columbia, MO	65211	540-357-0654	cmbodinof@mizzou.edu
Bonney	Steve	Ky. Dept. Fish & Wildlife Resources	2375 KY 801 South		Morehead, KY	40351	6067800854	stephen.bonney@ky.gov
Briggler	Jeffrey	Missouri Department of Conservation	2901 W. Truman Blvd		Jefferson City, MO	65109	573/522/4115	jeff.briggler@mdc.mo.gov
Brodman	Bob	Saint Joseph's College	Biology Department		Rensselaer, IN	47978	219 394-2539	bobb@saintjoe.edu
Browne	Robert	IUCN/WAZA Amphibian Ark, Royal Zoological Society of Antwerp	Koningin Astridplein 26		2018 Antwerp Belgium			robert.browne@gmail.com
Burgmeier	Nicholas	Purdue University	715 West State Street		West Lafayette, IN	47907	765-494-3568	nburgmei@purdue.edu
Byrnes	Chad	Gorman Nature Center	416 W 6th St		Mansfield, OH	44903	4196317246	potogold71@yahoo.com
Calfee	John	Missouri Department of Conservation	2901 W. Truman Blvd		Jefferson City, MO	65109	573/522/4115	john.calfee@mdc.mo.gov
Collins	Joseph	Kansas Biological Survey	University of Kansas		Lawrence, KS	66047	785-393-4757	jcollins@ku.edu
Collins	Suzanne	CNAH	1502 Medinah Circle		Lawrence, KS	66047	785-393-2392	scollins@ku.edu
Crane	Adam	Missouri State University	Department of Biology		Springfield, MO	65897	417-836-3185	adamcrane@missouristate.edu
Davis	Adam	The Wilds	14000 International Road		Cumberland, OH	43732	740-638-5030	adavis@thewilds.org
Doro	Sherri	Nashville Zoo	3777 Nolensville Road		Nashville, TN	37211	615-833-1534	sdoro@nashvillezoo.org
Fedele	Tim	Affiliate	201 Trauger Road		Latrobe, PA	15650	7248365429	tfedele@fedelelaborlaw.com
Feller	Daniel	MD DNR Natural Heritage	c/o Appalachian Lab., Univ. of MD	301 Braddock Rd.	Frostburg, MD	21532	301-689-7202	dfeller@dnr.state.md.us
Floyd	Mike	U.S. Fish and Wildlife Service	J.C. Watts Federal Building, Room 265	330 W. Broadway	Frankfort, KY	40601	502-695-0468	mike_floyd@fws.gov
Forestal	Gwen	Lycoming College	700 College Place		Williamsport, PA	18657	570-321-4006	forgwen@lycoming.edu
Freake	Michael	Lee University	1120 Ocoee St		Cleveland, TN	37311	423-6148282	mfreake@leeuniversity.edu

Directory of Registrants

Last Name	First Name	Company/Organization	Address Line 1	Address Line 2	City, State	Zip	Phone	Email
Greathouse	Joe	The Good Zoo at Oglebay	465 Lodge Drive		Wheeling, WV	26003	(304) 243-4029	jgreathouse@oglebay-resort.com
Gregory	Nathan	Kentucky Department of Fish and Wildlife Resources	1449 Cassidy Creek Rd.		Carlisle, KY	40311	859-289-8564	nathan.gregory@ky.gov
Groves	John	NC Zoological Park	4401 Zoo Parkway		Asheboro, NC	27205	336-879-7600	john.groves@nczoo.org
Hawkins	Laura	Affiliate	201 Trauger Road		Latrobe, PA	15650	7248580463	lhawkins32@zoominternet.net
Hayes	Thomas	Pittsburgh Zoo and PPG Aquarium	One Wild Place		Pittsburgh, PA	15206	(412)365- 2596	thayes@pittsburghzoo.org
Hedrick	David	Chattanooga Zoo	207 Hillcrest Ave.		Chattanooga, TN	37411	423-645-3796	d.hedrick@hotmail.com
Hime	Paul	Saint Louis Zoo	1 Government Drive		Saint Louis, MO	63110	314-646-4785	herpkeepers@stlzoo.org
Hohman	Jeff	East Kentucky Power	4775 Lexington Road		Winchester, KY	40392-0707	800-238-3443	jeff.hohman@ekpc.coop
Hoover	Craig	US Fish and Wildlife Service	4401 North Fairfax Drive		Arlington, VA	22203	703-358-2162	craig_hoover@fws.gov
Hopkins	William	Virginia Tech	Dept of Fisheries and Wildlife	100 Cheatham Hall	Blacksburg, VA	24061	540-231-7292	hopkinsw@vt.edu
Horchler	Doug	Marshall University	Dept. of Biological Sciences	One John Marshall Drive	Huntington, WV	25755		horchler@marshall.edu
Humphries	Jeff	NC Wildlife Resources Commission	404 Barclay Rd		Chapel Hill, NC	27516	919-928-4071	humphri2@yahoo.com
Irwin	Kelly	Arkansas Game & Fish Commission	915 E. Sevier St.		Benton, AR	72015	877.847.2690	kirwin@agfc.state.ar.us
Irwin	Lisa	Ouachita Technical College	Arts & Sciences Division	One College Circle	Malvern, AR	72104	501-332-0281	lirwin@otcweb.edu
Jackson	Rick	Chattanooga Zoo	McCallie Ave.		Chattanooga, TN	37411	423-503-3888	zoodesigns@aol.com
Jewell	David	Stowers Institute	7541 Wyoming		Kansas City, MO	64114	816-518-8639	daj@stowers.org
Johnson	Timothy	Oosanshouuo-no-Kai	Minami-Oya 1449-11	Machida-shi	Tokyo, Japan,	194-0031	81 (Japan) 90-3217-4250	timtim@gol.com
Klueh	Sarabeth	Indiana Department of Natural Resources	553 E. Miller Dr		Bloomington, IN	47401	812-334-1137	sklueh@dnr.in.gov
Lipps	Gregory	MW PARC / Gregory Lipps, LLC	1473 County Road 5-2		Delta, OH	43515	419-376-3441	greglipps@aol.com
Little	Raymond	Gulf Coast District, Everglades National Park	PO Box 5013		Everglades City, FL	34139	239-695-4219	Raymond_Little@nps.gov
MacGregor	John	KY Department of Fish and Wildlife	#1 Sportsmen's Lane		Frankfort, KY	40601	502-564-7109	john.macgregor@ky.gov

Directory of Registrants

Last Name	First Name	Company/Organization	Address Line 1	Address Line 2	City, State	Zip	Phone	Email
Maley	Abigail	Illinois Natural History Survey	804 Breen Dr.		Champaign, IL	61820	814-233-9843	ajmaley@gmail.com
Mathis	Alicia	Missouri State University	Department of Biology		Springfield, MO	65897	417-836-5699	aliciamathis@missouristate.edu
McGinnity	Dale	Nashville Zoo	3654 Bear Hollow rd.		Whites Creek, TN	37189	615-833-2248	dmcginnity@nashvillezoo.org
McMillan	Amy	Buffalo State College	1300 Elmwood		Buffalo, NY	14222	716-878-3756	mcmillam@buffalostate.edu
Moser	William	National Museum of Natural History, Smithsonian Institution	Department of Invertebrate Zoology, MRC 534	4210 Silver Hill Road	Suitland, MD	20746	301-238-1833	moserw@si.edu
Nichols	Barry	KTC Division of Environmental Analysis	200 MERO STREET		FRANKFORT, KY	40622	502.564.7250	barry.nichols@ky.gov
Nickerson	Max	Florida Museum of Natural History	PO Box 117800	University of Florida	Gainesville, FL	32611-7800		maxn@flmnh.ufl.edu
Noel	Krista	SCDNR	PO Box 2636		Murrells Inlet, SC	29576	843-289-1557	krisnoel1@yahoo.com
Petokas	Peter	Lycoming College	700 College Place		Williamsport, PA	18657	570-321-4006	petokas@lycoming.edu
Pfingsten	Ralph	ODNR	347 Pineview Cir		Berea, OH	44017	440-243-7568	rap347@wideopenwest.com
Pfingsten	Joan	ODNR	347 Pineview Cir		Berea, OH	44017	440-243-7568	rap347@wideopenwest.com
Quinn	Sam	SUNY ESF	150 Oakland Street		Syracuse, NY	13210	845-239-1297	saquinn@syr.edu
Rayman	Noelle	Buffalo State College	1300 Elmwood Ave		Buffalo, NY	14203	315-343-7397	nlrayman@hotmail.com
Saunders	Robin	National Zoo	3001 connecticut ave		washington, DC	20008	(202)633-3255	saundersr@si.edu
Schuette	Chawna	Saint Louis Zoo	1 Government Drive		Saint Louis, MO	63110	314-646-4655	herpgirlchawna@yahoo.com
Sisson	Mike	North Carolina Wildlife Resources Commission	P.O. Box 149		Hoffman, NC	28347	601-519-7726	sissonh@aol.com
Unger	Shem	Purdue University	715 West State Street		West Lafayette, IN	47907	765-494-3568	sunger@purdue.edu
Weaver	Harley	KY Fish & Wildlife	73 Lara Drive		Louisa, KY	41230	606.686.3642	harley.weaver@ky.gov
Williams	Rod	Purdue University	715 West State Street		West Lafayette, IN	47907	765-494-3568	rodw@purdue.edu
Wolfe	Barb	the Wilds	14000 International Rd		Cumberland, OH	43732	(740) 638-5030	bwolfe@thewilds.org
Wylie	Dan	Illinois Natural History Survey	804 Breen Drive		Champaign, IL	61820	815-341-4079	wylie.dan@gmail.com