

**California-Nevada Amphibian Populations Task Force**  
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**ABSTRACTS**

ABBAS, GAR, ANNE POOPATANAPONG, and MARC STAMER  
USDA Forest Service, San Bernardino National Forest, San Bernardino, California; gabbas@fs.fed.us.

**Balancing Ranids and Recreation: Forest Service Efforts to Protect Mountain Yellow-Legged Frogs and Quality Recreation Opportunities**

The mountain yellow-legged frog (*Rana muscosa*, MYLF) was historically one of the most common frog species in Southern California and was found in virtually every perennial stream in the San Jacinto, San Bernardino and San Gabriel Mountains. They have dramatically declined statewide and the Southern California population segment was listed as endangered in 2002. The National Forest system houses most of the last known MYLF populations in Southern California and the Forest Service is actively pursuing measures to protect the remaining populations and promote species recovery. The San Jacinto Mountains, on the San Bernardino National Forest, are home to one of the largest remaining populations of MYLF and some of the best available habitat for potential recovery. The San Jacinto Mountains are also located within a two hour drive of the Los Angeles metropolitan area, and just outside Palm Springs, making these mountains a popular area for outdoor recreational activities. In 2001 the Forest Service started monitoring recreational use in the vicinity of MYLF habitats on the San Jacinto Ranger District to document any conflicts. This data and examples of management actions taken to address conflicts are presented here along with descriptions of other efforts the Forest Service is involved in to protect and restore MYLF populations and habitats.

BACKLIN, ADAM R.<sup>1\*</sup>, ROBERT N. FISHER<sup>2</sup>, and CINDY J. HITCHCOCK<sup>1</sup>

<sup>1</sup>United States Geological Survey, Irvine, CA, [abacklin@usgs.gov](mailto:abacklin@usgs.gov); <sup>2</sup>United States Geological Survey, San Diego, CA

**Status and Update on the Southern California Mountain Yellow-Legged Frog (*Rana muscosa*), Including Information Regarding the Captive Breeding Program**

The mountain yellow-legged frog (*Rana muscosa*) has declined considerably in southern California over the past several decades. It currently occupies less than 1% of its former range and is present at only 8 locations within the San Gabriel, San Bernardino, and San Jacinto Mountains. Each population is extremely small (between 10 -75 adult animals), with less than 200 adults remaining in the wild. A working group including state and federal agencies, academic institutions, zoological societies, and others are collaborating in recovering this species. Recently, a microsatellite analysis of the southern California populations shows a high degree of differentiation between the three southern California mountain ranges, which will help guide restoration efforts. Next steps for restoration of this species include implementing a captive breeding program to both protect current populations from catastrophic events and provide a source for reintroducing frogs into unoccupied sites within the frog's former range.

BOIANO, DANNY

Sequoia and Kings Canyon National Parks, Three Rivers, CA, [danny\\_boiano@nps.gov](mailto:danny_boiano@nps.gov)

### **Ecological Restoration of High Mountain Lakes in Sequoia and Kings Canyon National Parks, California**

Since 2001 Sequoia and Kings Canyon National Parks have been eradicating non-native trout from naturally fishless high mountain lakes to restore habitat for native fauna, with an emphasis on improving the status of a declining amphibian, the mountain yellow-legged frog (*Rana muscosa*). This frog was once a common inhabitant of high Sierra Nevada lakes, but has disappeared from about 94% historic localities, largely due to the widespread introduction of trout, and very recently due to an emerging infectious disease (chytrid fungus, *Batrachochytrium dendrobatidis*). To date we have removed nearly 22,000 trout, including eradication from six lakes, and made substantial progress toward eradication in five additional lakes. Mountain yellow-legged frog densities measured in the six restored lakes showed an average 18-fold increase between 2001 and 2006, while one lake showed a 62-fold increase. The ecological recovery in these lakes has attracted native predators, such as snakes, birds, and even mammals, which have been observed preying on the now-abundant frogs, tadpoles, and aquatic invertebrates. Due to this success, SEKI recently initiated planning to expand restoration to additional lakes and streams across these parks. Although chytrid fungus has recently extirpated many mountain yellow-legged frog populations in the Sierra Nevada, a few abundant populations occupying fishless sites have survived and reproduced after becoming infected. This finding further supports the importance of continuing to eradicate non-native trout from high mountain lakes.

BOYARSKI, VALERIE L.\* and MICHAEL J. SREDL

Arizona Game and Fish Department, 2221 West Greenway Road, Phoenix, AZ 85023; [vboyerski@azgfd.gov](mailto:vboyerski@azgfd.gov)

### **Opening the Toolbox: Using Candidate Conservation Agreements and Safe Harbor Agreements to Implement Recovery**

In addition to regulatory authority under the Endangered Species Act of 1973 (ESA), flexibility and landowner participation are essential for successful conservation of special status species. Several sections of the ESA are often regarded as regulatory ‘hammers’. However, Section 10, which permits incidental take when overall conservation actions result in a net benefit for the target species, allows greater flexibility for land managers and provides assurances for private landowners to conserve species. Increasing use of this section has resulted in the development and implementation of such tools as Candidate Conservation Agreements (CCA) and Safe Harbor Agreements (SHA). In Arizona, two closely related special status ranids, Ramsey Canyon leopard frogs (*Rana subaquavocalis*) and Chiricahua leopard frogs (*Rana chiricahuensis*), are being managed through active conservation. Ramsey Canyon leopard frogs, which are former candidates for federal listing, are being managed under a CCA that began in 1996. The taxonomic uncertainty surrounding this species required land managers and private landowners to plan for its potential reclassification. Chiricahua leopard frogs, which are listed as threatened, will be managed under the authority of the ESA and a recovery plan is currently in the final stages of development. A statewide SHA for Chiricahua leopard frogs was also developed and is now being implemented. This tool provides greater flexibility and assurances for private landowners in conserving special status species. It will also help maintain cooperation with landowners who signed on to the Ramsey Canyon leopard frog CCA, should the species be taxonomically subsumed into *R. chiricahuensis*. We will discuss how CCAs and SHAs can and do contribute to species recovery in the Southwest.

BRADFORD, DAVID\*, EDWARD HEITHMAR, NITA TALLENT-HALSELL, GEORGES-MARIE MOMPLAISIR, CHARLITA ROSAL, LEE RIDDICK, and KATRINA VARNER.  
U.S. Environmental Protection Agency, Environmental Sciences Division, Las Vegas, NV

### **Temporal Patterns of Airborne Pesticides in Habitat of the Mountain Yellow-legged Frog in the Southern Sierra Nevada.**

Airborne agricultural pesticides from the Central Valley of California have been implicated as a possible cause for recent, dramatic population declines of several amphibian species in remote mountain locations. To determine the temporal variation of pesticide levels in the habitat of one of these species, the mountain yellow-legged frog (*Rana muscosa*), we sampled water from four lakes at high elevation (2754-3475 m) in the southern Sierra Nevada. The lakes ranged between 45 and 85 km from the San Joaquin Valley (i.e., southern end of Central Valley). Lakes were sampled weekly or monthly from mid June to mid October, 2003. Nine of 45 target analytes were detected at least once among the four lakes. Four pesticides were found sufficiently frequently to evaluate for temporal patterns: two insecticide/acaricides (endosulfan I and propargite) and two herbicides (dacthal [DCPA] and simazine). Concentrations of these pesticides were extremely low, on the order of 1 ng/L (parts per trillion) or less. For endosulfan and propargite, temporal variation in their concentrations corresponded closely with application rates in the San Joaquin Valley, with a lag time of 1-2 weeks. In contrast, application of dacthal and simazine was practically nil in the Valley during the sampling period. Linear distance from the San Joaquin Valley alone did not appear to be an adequate predictor of contaminant levels in lake water. Mountain yellow-legged frog populations have largely disappeared from the vicinities of lakes with both the higher and lower pesticide concentrations observed in the study.

BREHME, CHERYL S., SARA L. SCHUSTER, CARLTON J. ROCHESTER, STACIE A. HATHAWAY, and ROBERT N. FISHER\*  
US Geological Survey, Biological Resources Discipline, San Diego, CA

### **Arroyo Toads (*Bufo californicus*) in Southern California; Findings and Trends from 3 to 10 years of Population Monitoring**

In 2003, we implemented a new monitoring program for the endangered arroyo toad (*Bufo californicus*) on Marine Corps Base Camp Pendleton (MCBCP). To address the problems associated with large variations in adult toad activity, we employed a spatial and temporal monitoring approach that tracks the presence of arroyo toad breeding populations by documenting presence of eggs and larvae. Sites are surveyed up to four times per year to calculate and account for imperfect detection probabilities. We also continued to conduct nighttime counts of adult toads from the monitoring program implemented by Dan Holland in 1996. In this presentation, we review the major trends and findings of the first three years of the spatial monitoring program and a decade of adult count transects. These include the findings that 1) toad activity has been highly variable among years, but relatively stable over the last decade, 2) associations between activity and rainfall are dependant upon hydroperiod, 3) proportion of wet area occupied appears to be the most stable monitoring metric, and 4) both proportion area occupied (PAO) and probability of detecting arroyo toads are negatively associated with the presence of non-native aquatic species.

BROWN, CATHY

USDA Forest Service, Stanislaus National Forest, PSW Research Station, PO Box 245, Berkeley, CA 94701; cathybrown@fs.fed.us

**Long-Term Monitoring for the Yosemite Toad (*Bufo canorus*) and Mountain Yellow-Legged Frog (*Rana muscosa*): The Sierra Nevada Amphibian Monitoring Program, an update.**

The USDA Forest Service Sierra Nevada Amphibian Monitoring Program is a long-term, bioregional monitoring program for two aquatic frog species, the mountain yellow-legged frog (*Rana muscosa*) and Yosemite toad (*Bufo canorus*), in the Sierra Nevada, CA. The monitoring assesses the status and change of populations and habitat for these two species at the scale of the species' ranges in the Sierra Nevada and provides information for the 10-year Forest Service planning cycle. The monitoring combines extensive and intensive components in one integrated design. Extensively, for each species, small watersheds (2-4 km<sup>2</sup>) are surveyed throughout the range of each species over a 5-year cycle, with 20% revisited annually. Population trends are measured by breeding occupancy and habitat trends are measured by attributes that assess hydrologic condition, habitat matrix, cover, water temperature, disturbance, and general characterization. Intensively, more detailed abundance, life history, and habitat data is collected in two small watersheds for the Yosemite toad. The extensive component was initiated during 2002 and 106 watersheds were surveyed over the past five years, with 26 re-surveyed for at least three years. Abundances of Yosemite toad adults, egg masses, tadpoles, and metamorphs were estimated in two intensive watersheds in 2006. Results will aid in management of these species providing information for making more informed management decisions.

BUSTEED, GARY.\*, SETH P. D. RILEY, and LANE CAMERON

<sup>1</sup>National Park Service, Santa Monica Mountains National Recreation Area, Thousand Oaks, CA, Gary\_Busteed@nps.gov

**Monitoring the Presence and Abundance of Aquatic Breeding Amphibians as Indicators of Ecological Change**

The National Park Service, in consultation with local and regional wildlife experts, are implementing field methods for monitoring the long-term reproductive success and population status of aquatic breeding amphibians (*Taricha torosa*, *Hyla cadaverina*, *Hyla regilla* and *Bufo boreas*) in the Santa Monica Mountains National Recreation Area (Los Angeles, California). Streams were selected for their value as amphibian habitat and their potential to provide information on amphibian populations in urban developed and parkland watersheds. A preliminary six year inventory was conducted by the NPS, in cooperation with Pepperdine University, USGS and the Resource Conservation District of the Santa Monica Mountains, documenting the presence and abundance of aquatic amphibians during their spring and early summer breeding season. Presence of invasive species was noted, the physical stream characteristics and basic water quality parameters were also measured. Urban streams have more water, less habitat diversity and a greater chance of having invasive predators. As a consequence these streams also have a lower abundance or absence of aquatic breeding amphibians. The results of the six year inventory effort has guided development of this long term monitoring strategy and is a part of the National Park Service's Vital Sign Monitoring Program.

FELLERS, GARY M. <sup>1\*</sup>, PATRICK M. KLEEMAN<sup>1</sup>, DONALD W. SPARLING<sup>2</sup>, and LAURA L. McCONNELL<sup>3</sup>

<sup>1</sup>Western Ecological Research Center, USGS, Point Reyes National Seashore, Point Reyes, CA 94956, [gary\\_fellers@usgs.gov](mailto:gary_fellers@usgs.gov); <sup>2</sup>Cooperative Wildlife Research Laboratory, Department of Zoology and Center for Ecology, Southern Illinois University, LS II, MS6504, Carbondale, IL 62901, [dsparl@siu.edu](mailto:dsparl@siu.edu); <sup>3</sup>U.S. Department of Agriculture, Agricultural Research Service, Environmental Quality Laboratory, Building 007, Room 225, BARC-W, Beltsville, Maryland 20705, [mcconnell@ba.ars.usda.gov](mailto:mcconnell@ba.ars.usda.gov)

### **Possible Factors in the Decline of California Amphibians: Contaminants and Disease**

For the last few years, we have been measuring concentrations of current use pesticides in the environment. Each of the most commonly used pesticides can be found in air, snow, or tissue samples, but it is unclear what the impact might be on local amphibians. We have addressed this problem by exposing native species of tadpoles to environmentally realistic concentrations of pesticides. Our experiments have run much longer than the typical 24 - 96 hr exposures used in most toxicity tests. We have found that the parent compounds are present in the Sierra Nevada in sufficient concentration to cause significantly depressed survival rates. Recently concluded lab experiments have shown that one of the breakdown products from organophosphorus pesticides is far more toxic than the related parent compound. Other research has involved the assessment of *Batrachochytrium dendrobatidis* (a chytrid fungus) in watersheds within two watersheds in Yosemite and one at Point Reyes National Seashore to determine the distribution of the fungus in tadpoles. Preliminary results from this work should be available for discussion.

GARCIA, ERNESTO

Partners in Amphibian and Reptile Conservation, US Fish and Wildlife Service, PO Box 2964, Weaverville, CA 96093; [ernest\\_garcia@fws.gov](mailto:ernest_garcia@fws.gov)

### **PARC and Habitat Management Guidelines for Amphibians and Reptiles**

Habitat alteration, fragmentation and loss are considered to be major challenges for the conservation of amphibians and reptiles (or herpetofauna). Herpetofaunal populations in the United States are declining, and as long as human populations expand, habitats will be modified. Thus, Partners in Amphibian and Reptile Conservation (PARC) recognized an opportunity to provide proactive guidance for improving the compatibility of land management practices with these animals. PARC has developed a series of regionally-specific best management practices, or Habitat Management Guidelines (HMGs). These guidelines use the best science available to produce recommendations that are easily understood and practical for land managers and private landowners. The objectives of these guidelines are to keep common species common, stem the decline of imperiled species, provide guidance on the management and restoration of amphibian and reptile habitats while benefiting many other wildlife species, and reduce the likelihood of species becoming listed as threatened or endangered. Landowners and land managers are presented with measures to help them maximize compatibility with their existing management objectives, or to optimize their management actions specifically for herpetofauna. I will present examples of the management recommendations from the HMGs, as well as information on PARC's development of accompanying HMG training modules.

GOFORTH, SUZANNE R.<sup>1\*</sup>, MICHAEL J. SREDL<sup>1</sup>, E. H. DUKE KLEIN<sup>2</sup>, WILLIAM P. BURGER<sup>1</sup>, and JEFF SERVOSS<sup>3</sup>

<sup>1</sup>Arizona Game and Fish Department, Phoenix, AZ, [sgoforth@azgfd.gov](mailto:sgoforth@azgfd.gov), [msredl@azgfd.gov](mailto:msredl@azgfd.gov), [bburger@azgfd.gov](mailto:bburger@azgfd.gov); <sup>2</sup>Tonto National Forest, Payson-Pleasant Valley Ranger Districts, Payson, AZ, [eklein@fs.fed.us](mailto:eklein@fs.fed.us); <sup>3</sup>U.S. Fish and Wildlife Service, Phoenix, AZ, [Jeff\\_Servoss@fws.gov](mailto:Jeff_Servoss@fws.gov)

### **Gentry Creek Management Area: Linking Local Conservation to Recovery**

The Chiricahua leopard frog (*Rana chiricahuensis*) was described in 1979 and is a member of the *Rana pipiens* complex. Its range extends along the Mogollon Rim in central Arizona into the mountains of west-central New Mexico, and from the sky islands of southeastern Arizona and southwestern New Mexico into the Sierra Madre Occidental of northeastern Sonora and western Chihuahua. Declines in the species began to be noted in the mid to late 1980s, and in June 2002 the Chiricahua leopard frog was federally listed as threatened under the Endangered Species Act. The draft Chiricahua leopard frog Recovery Plan recognizes the importance of tailoring recovery actions to the varying ecological and socio-political circumstances across the species' range. In the Gentry Creek Management Area, local partnerships form the backbone of efforts to re-build a viable metapopulation through augmentation of extant populations, establishment of new populations, habitat enhancement/restoration, threat alleviation, and monitoring. Nine historical Chiricahua leopard frog localities have been documented in the Gentry Creek vicinity. By the mid-1990s, frogs were known from only four sites, two of which had frogs that regularly reproduced. Cooperative conservation projects that began in 1998 to prevent extinction of the Gentry Creek metapopulation have gained support and momentum, and are beginning to show signs of success. By continuing to refine the approaches developed in the Gentry Creek Management Area and applying the model in other Management Areas across the species' range, we hope to make recovery and delisting of the Chiricahua leopard frog a reality.

GOLDSTEIN, JEFFREY<sup>1\*</sup>, KARIN HOFF<sup>2</sup>, and STANLEY HILLYARD<sup>3</sup>

<sup>1</sup>School of Life Sciences, University of Nevada, Las Vegas, [Goldstj1@unlv.nevada.edu](mailto:Goldstj1@unlv.nevada.edu); <sup>2</sup>Dept. of Biology, University of Nevada, Reno, <sup>3</sup>University of Nevada School of Dental Medicine, Las Vegas

### **Thermal Sensitivity of Growth, Development and Oxygen Consumption of the Relict Leopard Frog (*Rana onca*)**

The Relict Leopard frog (*Rana onca*) is a rare species that has been petitioned for endangered status under the Federal ESA. *Rana onca* was believed to have gone extinct sometime after 1950, but was rediscovered in the early 1990s. Currently there are five populations located near the Overton arm of Lake Mead and near Black Canyon on the Colorado River including thermally influenced springs with water temperatures in excess of 35°C at some sites. It is not known whether *R. onca* tadpoles acclimate to, or are adversely affected by, the high temperatures. Our experiments were designed to determine the effects of rearing temperature on size at metamorphosis, rate of development, and temperature preferences. We found some evidence of temperature acclimation in both oxygen consumption and temperature selection. We also found a very clearly defined optimal temperature for growth and development. We will discuss the implications of temperature optima and acclimation on tadpole survivorship and oxygen consumption.

GRIEGO, VINCENT, CATHY JOHNSON, KAREN LEYSE, and PETE TRENHAM\*  
US Fish and Wildlife Service, Sacramento Fish and Wildlife Office, 2800 Cottage Way, Sacramento, CA

### **Update on Recent DAPTF-Related Activities in the Sacramento Fish and Wildlife Service Office**

The Sacramento Fish and Wildlife Office is responsible for a number of ongoing and upcoming projects with relevance to declining amphibian populations. We will provide a brief summary of our work on the following topics: 1) efforts to conserve and recover listed amphibians in our jurisdiction (i.e., the California red-legged frog and California tiger salamander); 2) current status of candidate species (i.e., Sierra Nevada mountain yellow-legged frog and Yosemite toad); and 3) refuge surveys for malformed amphibians. We will also provide time for questions from the audience.

HEMMINGS, VIKTORIA<sup>1\*</sup>, JEF R. JAEGER<sup>1</sup>, MICHAEL J. SREDL<sup>2</sup>, MARTIN A. SCHLAEPFER<sup>3</sup>, RANDY D. JENNINGS<sup>4</sup>, CHARLES W. PAINTER<sup>5</sup>, DAVID F. BRADFORD<sup>6</sup>, and BRETT R. RIDDLE<sup>1</sup>

<sup>1</sup>School of Life Sciences, University of Nevada, Las Vegas, NV, [hemming2@unlv.nevada.edu](mailto:hemming2@unlv.nevada.edu); <sup>2</sup>Arizona Game and Fish Department, Nongame Branch, Phoenix, AZ; <sup>3</sup>Section of Integrative Biology, University of Texas, Austin, TX; <sup>4</sup>Department of Natural Sciences, Western New Mexico University, Silver City, NM; <sup>5</sup>Endangered Species Program, New Mexico Department of Game and Fish, Santa Fe, NM; <sup>6</sup>US Environmental Protection Agency, National Exposure Research Laboratory, Landscape Ecology Branch, Las Vegas, NV.

### **Phylogeography of *Rana yavapaiensis* and *Rana onca*: Preliminary Findings with Conservation Implications**

The closely related aridland frogs *Rana onca* (Relict Leopard Frog) and *Rana yavapaiensis* (Lowland Leopard Frog) have both experienced dramatic population declines. *Rana onca* currently occurs naturally at only 6 disjunct sites in southern Nevada. *Rana yavapaiensis* is present across central and SE Arizona, but it is patchily distributed in portions of its Arizona range and has disappeared from California and most of its New Mexico range. Although recent observations from Sonora, Mexico exist, the status of these populations is unknown. Our project will inform conservation strategies for both species by investigating phylogeography and population structure. In our current analysis of 202 frog samples, preliminary results from mitochondrial DNA (mtDNA) markers (ND2 and a portion of *cyt b* genes) recovered the previously observed phylogenetic break between *R. onca* and *R. yavapaiensis*. Levels of sequence divergence and applied rates of sequence evolution allow us to postulate that these species' mtDNA gene lineages separated during the early Pleistocene, possibly prior to the onset of major climatic oscillations. A recently discovered population of leopard frogs from the western Grand Canyon (i.e., Surprise Canyon) represents a *R. yavapaiensis* population with clear mtDNA distinction from other *R. yavapaiensis* populations in Arizona and Mexico. This disjunct population may have separated from Arizona and Mexico populations prior to the latest Pleistocene glacial period. Surprisingly, *R. yavapaiensis* from across its main distribution in Arizona and into Mexico shows very little genetic diversity, suggesting a recent range expansion from some unsampled location in Mexico.

HOBBS, BRIAN

Nevada Department of Wildlife, Las Vegas, NV, bhobbs@ndow.org

### **Amargosa Toad: Status Update and Habitat Restoration Success Story**

The ninth annual mark and recapture survey of the Amargosa toad, *Bufo nelsoni*, was completed in May and June 2006. The combined estimates for all of the surveyed parcels in Oasis Valley, Nevada increased from 1,669 adult toads in 2005 to 2,698 in 2006. At the Harlan/Keal parcel, which includes four separate sites, the mean population estimate from 2003 to 2005 was 42, but in 2006, the estimate increased to 362 individuals. This dramatic increase was due to the restoration and repair of the primary breeding pond on this parcel in fall 2003 and winter 2004. While toad captures in the vicinity of this pond have increased, very few, if any, toads have been captured at the other sites. Over the next few years we expect toad captures to increase at these surrounding sites as toads migrate from the breeding pond.

JACKSON, TINA<sup>1\*</sup>, AND DOREEN SUMERLIN<sup>2</sup>

<sup>1</sup>Colorado Division of Wildlife, Colorado Springs, CO, [Tina.Jackson@state.co.us](mailto:Tina.Jackson@state.co.us); <sup>2</sup>Arapaho National Forest, Granby, CO.

### **How Boreal Toad Conservation in Colorado is Saving the World**

The Southern Rocky Mountain Population of Boreal Toad (*Bufo boreas boreas*) was designated a candidate species by the USFWS in 1989. Colorado and New Mexico also listed the species within their respective states. In September 2005, the USFWS revised their earlier finding of warranted but precluded to not warranted thus removing the population from the federal candidate list. The Boreal Toad Recovery Team, originally created by the Colorado Division of Wildlife, continues to work towards recovery of the toad through research, monitoring, and education. We will discuss the various research projects and findings that will benefit the boreal toad in Colorado, Wyoming, and New Mexico as well as other amphibian species throughout the world. These projects include life history studies, reintroduction protocols, habitat requirements, and, most importantly, disease research.

JAEGER, JEF R.<sup>1,2\*</sup>, DAVID F. BRADFORD<sup>3</sup>, GARTH M. SPELLMAN<sup>4</sup>, and BRETT R. RIDDLE<sup>1</sup>

<sup>1</sup> School of Life Sciences, University of Nevada, Las Vegas, [jef.jaeger@unlv.edu](mailto:jef.jaeger@unlv.edu); <sup>2</sup>Currently also with the Public Lands Institute, University of Nevada, Las Vegas; <sup>3</sup>U.S. Environmental Protection Agency, National Exposure Research Laboratory, Landscape Ecology Branch, Las Vegas, NV; <sup>4</sup>Center for the Conservation of Biological Resources/West Core, Black Hills State University, Spearfish, SD.

### **Population Structure of the Red-spotted Toad, *Bufo punctatus*, in a Naturally Fragmented Desert Landscape**

We investigated the spatial scale at which genetic structure of *Bufo punctatus* within the Mojave Desert is organized by sequencing a portion of mitochondrial DNA control region for 831 toads collected from 43 sites around Las Vegas, Nevada. We grouped these collections *a priori* into seven geographic ranges based predominately on clusters of sites within mountain ranges. We used hierarchical analysis of molecular variance (AMOVA) in a series of nested procedures to assess genetic structure among mountain ranges, among sites within mountain ranges, and among individuals within sites. We also calculated pairwise  $F_{ST}$  among sites within mountain ranges, and inferred population processes within mountain ranges by applying neutrality test statistics. We identified 36 haplotypes that formed five groups using network analysis, and an additional haplotype at three sites that represented recently



colonized *B. punctatus* from the Colorado Plateau. The designated mountain ranges accounted for a significant amount (25.8%) of genetic variation, and we confirmed substantial genetic structure between most neighboring ranges. Within four mountain ranges, we found little genetic variation among collection sites, and inferred that a population bottleneck or range expansion likely explained a lack of diversity within two of these ranges. Within three mountain ranges we found significant genetic structure among sites; however, within two of these ranges only a few sites generally accounted for most of the pattern. Within the third range the observed structure appears to have resulted from a recent convergence of two divergent lineages. Our assessment supports the perspective that within the Mojave Desert, *B. punctatus* occurs primarily in patchy populations within mountain ranges that are currently isolated from similar populations in neighboring ranges.

JOHNSON, PAUL\* and ROBERT SAULINO  
National Park Service, Pinnacles National Monument, paul\_johnson@nps.gov

### **Investigating Chytrid Fungus in and around Pinnacles National Monument**

Pinnacles National Monument is located in the Inner Coast Ranges of Central California, near the southern edge of the current range of the foothill yellow-legged frog (*Rana boylei*). This species was recorded at Pinnacles in the first half of the 1900's, but none have been observed in recent decades. In 2005 we began a pilot study to evaluate methods for re-establishing the species. Early in 2006, as we were gearing up for full-scale re-establishment efforts, conversations with participants at the 2006 CA/NV DAPTF meeting in Arcata convinced us that we needed to take further precautions against spreading the chytrid fungus *Batrachochytrium dendrobatidis* (Bd).

In order to determine whether Bd was present in the area, we tested adults of all anuran species we could find in appreciable numbers at our re-establishment and donor sites (94 *R. boylei*, 127 *Pseudacris regilla*, and 12 *Bufo boreas*). All species sampled at all sites tested Bd-positive. After discussions with amphibian experts, we determined that the benefits of re-establishing the species did not outweigh the risk of possibly introducing a novel strain of Bd into Pinnacles. We then re-directed our efforts toward investigating the incidence, effects, and population genetics of Bd in and around Pinnacles. We tested sets of 30 metamorphs at several sites (90 *R. boylei*, 60 *P. regilla*, and 30 *B. boreas*) and then monitored the health and survival of the cohort. Results from the first year of the study will be presented.

JONES, THOMAS R.  
Arizona Game and Fish Department, Nongame Branch, 2221 West Greenway Road, Phoenix, AZ 85023;  
TJones@azgfd.gov.

### **Ten Years of Population Fluctuations in a Lowland Leopard Frog Population: Implications for Conservation.**

In the arid Southwest, ranid frogs often persist in fragmented landscapes in relatively small, isolated wetlands. These habitats might appear to be stable, and at times can support large populations of ranid frogs. I present a summary of 10 years of study of *Rana yavapaiensis* in the Sonoran Desert, occupying a relatively small watershed, and isolated from other nearby populations. Population dynamics have been influenced by episodic flooding and persistent drought coupled with disease and predation. Perceptions of stability might mislead conservation strategies. Long term monitoring programs at multiple sites are necessary to understand the effects of stochastic events and subsequent community-level processes on the persistence of local populations of ranid frogs.

KING, ABIGAIL D.<sup>1\*</sup>, MICHAEL J. SREDL<sup>1</sup>, JAMES C. RORABAUGH<sup>2</sup>, STEPHEN F. HALE<sup>3</sup>, and CRAIG IVANYI<sup>4</sup>

<sup>1</sup>Arizona Game and Fish Department, Phoenix, AZ, msredl@azgfd.gov, aking@azgfd.gov; <sup>2</sup>U.S. Fish and Wildlife Service, Phoenix, AZ, Jim\_Rorabaugh@fws.gov; <sup>3</sup>EcoPlan Assoc. Inc., Tucson, AZ, sfhale@comcast.net; <sup>4</sup>Arizona-Sonora Desert Museum, Tucson, AZ, civanyi@desertmuseum.org

### **Repatriation of the Tarahumara frog (*Rana tarahumarae*) to Arizona: 1992-2006**

Populations of the Tarahumara frog (*Rana tarahumarae*) declined in Arizona during the late 1970's and by 1983, the last wild *R. tarahumarae* in Arizona was found dead in Big Casa Blanca Canyon (BCBC), Santa Cruz County, Arizona. However, the species is still well-represented in parts of the northern Sierra Madre Occidental and adjacent sky islands in Sonora and Chihuahua, Mexico. Plans to repatriate *R. tarahumarae* to Arizona were initiated in 1992 and have been coordinated by the Tarahumara Frog Conservation Team. Stock for release was collected from Sonora, reared in captivity, and released to BCBC. Initial releases began in June 2004, and since then, over 900 individuals have been released to 3 areas of the canyon. We are using visual encounter surveys to document five, predetermined stages of success and evaluate the impact of potential threats (fire, drought, flooding, and disease). To date, we have seen a reasonably high percentage of individuals survive in the short-term (stage 1 success) and survive over winter (stage 2 success). Our monitoring efforts have also confirmed progress towards achieving our goals of three-year survival (stage 3), reproduction (stage 4), and recruitment (stage 5). A catastrophic wildfire in 2005, followed by flooding and sedimentation in late-summer of 2006 had a major impact on BCBC. In the coming field season, we plan to further evaluate impacts of these recent events and survey for additional sites to continue repatriation efforts in Arizona.

KUPFERBERG, SARAH<sup>1\*</sup>, AMY LIND<sup>2</sup>, and SARAH YARNELL<sup>3</sup>

<sup>1</sup>Questa Engineering Corp., 1220 Brickyard Cove Rd. Suite 206, Point Richmond, CA 94807, [skupferberg@pacbell.net](mailto:skupferberg@pacbell.net); <sup>2</sup>USFS, Sierra Nevada Research Center, 2121 Second St., Suite A-101, Davis, CA 95616, [alind@fs.fed.us](mailto:alind@fs.fed.us); <sup>3</sup>Geology Department, University of California, Davis, One Shields Avenue, Davis, CA 95616, [yarnell@geology.ucdavis.edu](mailto:yarnell@geology.ucdavis.edu).

### **Effects of Pulsed Flows on the Foothill Yellow-Legged Frog (*Rana boylei*)**

The foothill yellow-legged frog (*Rana boylei*) completes its life cycle within fluvial environments. The life stages occur along a continuum of susceptibility to flow fluctuation: from immobile eggs, to moderately mobile tadpoles, to highly mobile adults. We assessed the effects of pulsed flows on the early life stages, using existing data and new experiments. A review of California studies clearly indicated that eggs were scoured by aseasonal flow releases from dams, recreational boating flows and natural late spring rain events in an unregulated river. Stranding occurred when flows were suddenly reduced. Data regarding flow effects on tadpoles were problematic to interpret so we manipulated velocity in artificial channels in the laboratory and in the field. The primary short-term behavioral response of tadpoles to increased velocity was to seek refuge in the substrate. Critical velocity, the current speed at which tadpoles could no longer swim, nor maintain position in flow refugia varied with size, developmental stage, and population of origin. In field experiments, the long-term consequences of the behavioral response were increased risk of predation for small tadpoles and slowed growth of larger tadpoles. These results will be combined with hydrodynamic modeling and a population projection model to provide information for water managers on habitat conditions for early life stages under varying flow releases.

While conducting this research, we made incidental observations of the prevalence of the parasitic copepod, *Lernaea cyprinacea*, on *R. boylei* tadpoles and metamorphs. Limb deformities were also observed.

LOWE, JASON  
Bureau of Land Management, Hollister Field Office, CA

### **Chytrid Distribution in Foothill Yellow-legged Frogs and Pacific Tree Frogs in the Diablo Mountains and San Joaquin Valley Foothills of Central Coastal California**

In 2006, Pinnacles National Park and the Hollister BLM conducted skin swab sampling for the amphibian chytrid fungus in the Diablo Mountains, San Benito County, and western San Joaquin foothills, Fresno County, California. Initially, pooled samples, and later individual samples from foothill yellow-legged frogs (*Rana boylei*) and Pacific treefrogs (*Pseudacris regilla*) were found to be positive in nearly all sites sampled (10 of 12). Three streams (Laguna, Clear, Sawmill) and one river segment in the San Benito River watershed, two streams in the Salinas River watershed (Chalone, Sandy), two streams (Cantua, Arroyo Leona) in the Cantua Creek watershed, and one stream each in the Big Panoche (Silver) and Los Gatos Creek (White) watersheds tested positive for chytrid. The two sites not found to be positive had low sample sizes. Individual samples collected by BLM showed only nine of 49 (18%) *R. boylei* and one of seven (14%) *P. regilla* samples to be positive indicating that while chytrid may be widespread, the majority of the post-metamorphic population appears to be chytrid-free. The nine *R. boylei* that tested positive ranged in snout-vent length (SVL) from 25 to 40 mm, while no *R. boylei* over 40 mm SVL (n=15, range 41-65) tested positive for chytrid. Considering these sizes, all frogs known to be carrying the chytrid fungus were either young-of-the-year or in their first post-metamorphic year (sub-adults). Chytrid fungus was found in a variety of stream types including those dominated by Brewer's willow, salt cedar, and California sagebrush with water temperatures between 15 and 26°C. Nine of ten (90%) of chytrid-positive frogs were found in pools, even though pool microhabitats were represented by only 50% of the 57 samples. Population density varied among chytrid-positive streams from 92 adults and 435 sub-adults per kilometer at White Creek to only 8 adults and 33 sub-adults per kilometer at Sawmill Creek.

MARLOW, KARLA\*, JOE DRENNAN, RON JACKMAN, and KEVIN WISEMAN  
Garcia and Associates (GANDA), 2601 Mission Street, Suite 600, San Francisco, CA,  
kmarlow@garciaandassociates.com

### **Using Chin and Flank Photographs as an Identification Tool for Tracking Foothill Yellow-Legged Frog Movement, Breeding Behavior, Longevity and Breeding Population Size in the North Fork Feather River**

As part of a three-year study of foothill yellow-legged frog (*Rana boylei*) movement and breeding activities GANDA employed the use of pigment pattern identification of individual frogs for seven river breeding sites on the North Fork Feather River. In 2004, 2005, and 2006 we used chin and flank patterns for frog identification. In 2005, we also attached BD-2 radio transmitters to 46 females and 6 males to further address frog movements and verify identification methodologies. Identification of individual frogs was determined in the field by matching pigment shapes and patterns and, later, verified through photo comparisons by our team. To date, 528 individuals have been identified, a total of 334 females and 194 males. With photographs used directly in the field we were able to instantly document movements of individuals between tributaries and river breeding sites, as well as long-distance movement between river breeding sites of non-telemetered frogs. A subset of radio-tagged frogs showed similar movements.

We found chin and flank photographs useful for determining movement and breeding information, as well as longevity and population size estimates. Photographic data revealed 10 adult female frogs were captured in 3 consecutive years and 14 adult male frogs were captured in 2 consecutive years. Further fieldwork and longevity analysis is planned, but preliminary results indicate that females live at least 5 years in the wild and breed for at least 3 years. This method offers a reliable, non-invasive way to conduct mark-recapture studies on *R. boylei*, which may also be applied to other western ranid species in the field.

MENEKS, MAIJA

Humboldt-Toiyabe National Forest, Mountain City Ranger District, mmeneks@fs.fed.us

### **Columbia Spotted Frog Monitoring, NE Nevada**

Three Districts of Humboldt-Toiyabe National Forest in NE Nevada support populations of Columbia spotted frog (Great Basin Population), a Candidate species. Monitoring sites, one per District, were established in 2004 and 2005 to better understand local characteristics of this frog. This monitoring will assist in Forest project design; and monitoring is also a line item in the Nevada Columbia Spotted Frog Conservation Agreement. Monitoring includes PIT tagging and elastomere marking for mark-recapture. Other data taken includes GPS position, gender, snout-vent length, weight, and general observations of animal condition and capture location. Presented are two or three years of data, depending upon year of site establishment, with discussion as to potential relevance for local management.

PICCO, ANGELA M.\*, and JAMES P. COLLINS

School of Life Sciences, Arizona State University, Tempe, AZ, angela.picco@asu.edu

### **Amphibian Pathogen Movement Through the Bait Trade in North America**

Amphibians are moved commercially through the pet trade, food trade, bait trade, and as research organisms. The commercial transport of amphibians may enhance the spread of amphibian diseases. Emerging infectious diseases are implicated in the declines and extinctions of amphibian populations worldwide. One group of emerging infectious diseases, ranaviruses, is associated with amphibian die-offs in wild populations in North America, Europe, and South America, and in commercial populations in Asia and South America. To evaluate and manage the impact of commercially moved ranaviruses on amphibian populations, we must first understand the transport of amphibians and their pathogens through commercial trade. Our research uses the movement of amphibian pathogens through the tiger salamander bait trade in North America as a model for the transport of amphibians and their pathogens through commercial trade. Our results indicate that ranavirus-infected tiger salamanders are moved within the bait trade in Arizona, New Mexico, and Colorado. In addition, our results suggest a mechanism by which viable salamanders are released into wild populations through the tiger salamander bait trade. These results demonstrate that commercial trade is a possible mechanism for spreading amphibian disease. The goal of understanding EID movement in commercial trade is to monitor and control the spread of disease, predict future disease outbreaks, and take measures to protect declining populations of amphibians.

POPE, KAREN<sup>1,2\*</sup>, JUSTIN GARWOOD<sup>1,3</sup>, SHARON LAWLER<sup>2</sup>, and HARTWELL WELSH JR.<sup>1</sup>  
<sup>1</sup>USDA Forest Service, Pacific Southwest Research Station, Redwood Sciences Laboratory, 1700 Bayview Dr., Arcata, CA 95521; [kpope@ucdavis.edu](mailto:kpope@ucdavis.edu); <sup>2</sup>UC Davis Ecology, One Shields Ave., Davis, CA 95616; <sup>3</sup>Department of Wildlife Management, Humboldt State University, Arcata, CA 95521

### **Direct and Indirect Effects of Introduced Trout on Cascades Frogs in the Klamath Mountains of Northern California.**

We discuss the results of a 4-year basin-scale experiment assessing the effects of fisheries management options (stock, suspend stocking, remove fish) on distribution and abundance of amphibians in the Trinity Alps Wilderness. We found that non-native trout presence suppresses the numbers of the declining Cascades frog (*Rana cascadae*) and other amphibians. Three years after trout removals, Cascades frogs showed a marked increase in abundance. The increase in frogs seems to be due to both immigration and increased onsite recruitment.

Using the combination of three studies implemented at three spatial scales, we also assessed indirect effects of introduced trout on amphibians. In addition to being predators, introduced trout are a supplemental prey source to common aquatic garter snakes (*Thamnophis atratus*) in the Klamath Mountains and may further the decline of the Cascades frog by facilitating the increase and spread of *T. atratus*. We evaluated this hypothesis by comparing the diet, distribution, and density of *T. atratus* with another native sympatric garter snake species, the common garter snake (*T. sirtalis*), which appears to be a native local amphibian specialist in the Klamath Mountains. Diet composition differed between the two species: *T. atratus* preyed upon both fish and amphibians about equally whereas *T. sirtalis* preyed solely upon amphibians. The distribution and density of *T. atratus* was positively related to the distribution and density of trout while the distribution and density of *T. sirtalis* was positively associated with amphibians. When we compared the relative abundance of *R. cascadae* with and without *T. atratus*, we found fewer frogs in sub-basins where we also found *T. atratus*.

SCHOCK, DANNA M.<sup>1\*</sup>, TRENT K. BOLLINGER<sup>2</sup>, V. GREGORY CHINCHAR<sup>3</sup>, JAMES K. JANCOVICH<sup>1</sup>, and JAMES P. COLLINS<sup>1</sup>

<sup>1</sup>School of Life Sciences, Arizona State University, Tempe, AZ, [danna.schock@asu.edu](mailto:danna.schock@asu.edu); <sup>2</sup>Canadian Cooperative Wildlife Health Centre, Department of Veterinary Pathology, Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, SK; <sup>3</sup>Department of Microbiology, University of Mississippi Medical Center, Jackson, MS

### **Experimental Evidence that Amphibian Ranaviruses are Multi-host Pathogens.**

Infectious diseases, including those caused by ranaviruses (family *Iridoviridae*), are among the suspected causes of global amphibian declines. Like many pathogens, ranaviruses appear to infect multiple species. We examined several North American amphibian ranavirus isolates for the purpose of better understanding the effects these viruses have on host communities. Our present study had two objectives. The first was to characterize isolates from epizootics affecting wild amphibian populations and compare them to previously described isolates. The second was to investigate whether amphibian ranaviruses infect ecologically relevant heterologous species, and if so, document the outcome of infection. The combined results of molecular characterization and experimental challenge trials suggest that there are at least two distinct, widespread amphibian ranaviruses in North America but both appear capable of infecting several amphibian species that share breeding habitats. Our research also suggests that individual host populations may differ in their responses to ranavirus infection, a finding with complex

conservation implications. Ultimately, experiments that elucidate the dynamics of intra- and inter-specific transmission will be particularly important for understanding the roles that ranaviruses play in their host communities and the threat they pose to amphibian populations.

SJÖBERG, JON C\*<sup>1</sup>, CHAD MELLISON<sup>2</sup>, JAMES HARVEY<sup>3</sup>, JENNIFER NEWMARK<sup>4</sup>, JAMES MARBLE<sup>5</sup>, and KENT HATCH<sup>6</sup>

<sup>1</sup>Nevada Department of Wildlife, Las Vegas, NV; <sup>2</sup>US Fish and Wildlife Service, Reno, NV; <sup>3</sup>US Forest Service, Humboldt-Toiyabe National Forest, Reno, NV; <sup>4</sup>Nevada Natural Heritage Program, Carson City, NV; <sup>5</sup>Nye County Department of Natural Resources, Tonopah, NV; <sup>6</sup>Brigham Young University, Provo, UT.

### **Status of the Columbia Spotted Frog (*Rana luteiventris*) Toiyabe Sub-population; an Update on Three Years of Conservation Agreement and Strategy Implementation.**

A structured conservation program for the Toiyabe subpopulation of Columbia spotted frog in central Nevada initiated in 2004 has now completed three years of active conservation implementation. Using a partnership approach for implementing a long-term monitoring plan which incorporates annual multiple life-stage monitoring has been a successful strategy to better assess range wide amphibian populations; available data indicates improving population numbers at sentinel sites which may largely be attributable to reduced drought effects on occupied habitats. The interagency Toiyabe Spotted Frog Technical Team has had significant success in implementing the majority of conservation tasks identified in the Conservation Strategy (84%) at some level, including the active monitoring program, on-ground habitat improvements and prioritization and limited support for species research needs. Several impediments remain, including gaining access to historic and potential frog habitats on private and tribal lands and sourcing adequate funding for implementing priority research activities and management actions.

STEPHENS, MOLLY R.\*, AND BERNIE P. MAY

Genomic Variation Laboratory, University of California, One Shields Avenue, Davis, CA 95616, [mrstephens@ucdavis.edu](mailto:mrstephens@ucdavis.edu)

### **Conservation Genetics of the Yosemite Toad, *Bufo canorus***

The Yosemite toad (*Bufo canorus*) is an endemic and declining Sierra amphibian. Previous reports of population genetic structure in this species were limited to samples in Yosemite and Sequoia Kings Canyon National Parks, a subset of the species range. This study examines populations from throughout the entire range using 500 base pairs of mitochondrial DNA (mtDNA) control region. Analyses of population genetic structure revealed a paraphyletic *B. canorus* group, relative to its sister taxon, the western toad (*B. boreas*). *B. canorus* showed several clades, with the majority exhibiting a “southern” origin and several selected mtDNA haplotypes having affinities with “northern” *B. boreas*. This apparent gene flow with *B. boreas* in the northern part of its range is possibly indicative of secondary contact between the two species; alternatively, lack of monophyly for *B. canorus* might also suggest that the diagnostic characters of the montane *B. canorus* may have originated through either single or multiple speciation events. Phylogenetic analysis identified evolutionary significant units for both species at the clade level, as well as management units within these clades that inform systematics and management efforts for these taxa. The fact that *B. canorus* was found to be paraphyletic has implications for its conservation and also for our understanding of how to preserve not only species, but also speciation processes. Additional work is underway with nuclear markers to assist in determining whether the observed pattern is unique to mtDNA, or reflective of a true pattern of gene flow between species.

TATARIAN, TRISH

Wildlife Research Associates, Santa Rosa, CA; trish@wildliferesearchassoc.com

### **Movement Patterns of the California Red-legged Frog (*Rana aurora draytonii*) in an Inland California Environment**

During the fall, winter and spring of 1999-2000 and 2000-2001 I tracked 49 radio-transmitted individuals of the California red-legged frog (*Rana aurora draytonii*) at a series of 8 pools within the intermittent Round Valley Creek in Round Valley Regional Preserve, Contra Costa County, California. I found that there was no significant difference in body weight or length between those frogs that moved and those that didn't. The majority of frogs (57%) stayed at their source pool spending 47% of their time within specific locations within the pool. The remaining 42% moved away from their pool, either terrestrially or aquatically, on at least one occasion. I observed 43 terrestrial forays by 12 frogs and 18 aquatic forays were undertaken by 12 frogs within the aquatic environment. All movements started after the first 0.5 cm of rain in the fall, with more terrestrial movements being made in the fall pre-breeding season (57%) than in the winter breeding season (32%) or spring post-breeding season (11%). Frogs moved greater average distances aquatically (84.6 m) than terrestrially (27.7 m). Greater terrestrial distances were moved in the pre-breeding season (35.2 m) than in the breeding season (15.5 m) or post-breeding season (16.3 m) with the majority of movements occurring for only one of the 3-4 day survey periods. The majority of frogs (57%) were position faithful within a pool.

THOMPSON, STEVE\*, LARA RACHOWICZ, and JEFF MAURER

Yosemite National Park, Wildlife Management Branch, Yosemite, CA, steve\_thompson@nps.gov

### **Yosemite Mountain Yellow-legged Frog Restoration: Proceeding Carefully with Data Gaps and Expert Consultation**

Comprehensive surveys of mountain yellow-legged frog populations (*Rana muscosa*: RAMU) in 2000 – 2002 in Yosemite National Park provided the basis for planning an initial restoration program in the park. Possible source populations were identified, suitable sites vacant of fish for direct reestablishment of RAMU populations, and bodies of water for fish removal and subsequent RAMU reestablishment were located. To initiate this effort, a project to first re-evaluate RAMU populations was begun in 2005. Results from this project showed that over half of the populations found in 2000 – 2002 had disappeared, and many of the planned source populations were no longer large enough to support RAMU removal. In addition the fungal infection, *Batrachochytrium dendrobatidis*: B.d., was found to be nearly ubiquitous in the remaining frog populations. Such findings prompted a reassessment of the original restoration project, and presented park biologists with the dilemma of how to proceed with restoration without causing further damage to RAMU populations through removal of individuals from the few remaining source populations to uncertain fates, and possible further spread of B.d. To answer these questions and provide recommendations for Yosemite's management direction for RAMU restoration, a workshop of experts of various relevant disciplines was assembled over a 2-day period in January 2006. Guided by workshop discussions focused on the emerging population and pathogen constraints, a limited restoration management program commenced in Yosemite in summer 2006, including translocations to three water bodies. Their findings reflect the tradeoffs and risks we face in restoration of RAMU in a diseased landscape, against the risks of management paralysis; an untenable option given the continuing rapid decline of the species.

WELSH, HARTWELL H., JR., GARTH R. HODGSON, KAREN L. POPE\*, and DON T. ASHTON.  
USDA Forest Service, Pacific Southwest Research Station, Redwood Sciences Laboratory, 1700 Bayview  
Dr., Arcata, CA 95521, 707-825-2956; hwelsh@fs.fed.us

### **Amphibian (and Amphibious Reptile) Research in Northwest California: Using Science to Affect Changes in Amphibian Declines**

The Herpetology Group at Redwood Sciences Lab (USDA Forest Service, Pacific SW Research Station) with assistance from graduate students in the biology and wildlife departments at Humboldt State University has been studying the natural history, demography, and landscape ecology of amphibian assemblages in aquatic and terrestrial environments of northern California for 22 years. A primary focus of our research has been to examine the relationships between amphibian biology and human land management practices. Amphibian populations in this region have declined due to detrimental forestry practices, introductions of non-native predators, and manipulations of the natural flow regimes of north coast rivers. We summarize five recent studies that address how anthropogenic disturbances have altered age structure and body condition of plethodontid salamanders, how species assemblages distribute across environmental gradients, how flow regimes influence western pond turtle behavior and growth, and how we might track ecosystem recovery by monitoring amphibian communities. In addition, we will introduce the discovery of a unique redwood forest niche for an upland salamander.

WITTE, CARMEL L.<sup>1,2</sup>, MICHAEL J. SREDL<sup>3\*</sup>, ANDREW S. KANE<sup>4</sup>, LAURA L. HUNGERFORD<sup>5</sup>  
<sup>1</sup>University of Maryland, College Park, MD; <sup>2</sup>Zoological Society of San Diego PO Box 120551, San Diego, CA; <sup>3</sup>Arizona Game and Fish Department; 2221 W. Greenway Rd. Phoenix, AZ, msredl@azgfd.gov; <sup>4</sup>University of Maryland, College Park, MD; <sup>5</sup>Univ. of Maryland, Baltimore, MD

### **Examination of Risk Factors Associated with the Disappearance of Arizona Native Ranid Frogs (Family Ranidae)**

Populations of ranid frogs in western United States, including those in Arizona, have declined dramatically over the past 30 years. Researchers studying these declines have implicated numerous factors, including habitat loss and degradation, predation and/or competition by nonnative species, environmental contamination, diseases, climate change, and increased UVB radiation. These factors have been identified during lab or field experiments or field observations of a few, intensely studied populations over a relatively short period of time. In the arid western United States, extreme variation in local conditions across sites through space and time makes it particularly difficult to measure variables related to population declines in one locality and generalize them to the larger landscape. Therefore, investigating trends in risk across a large number of sites on a larger scale may help discriminate between site-specific factors that are important regionally versus those that are only important locally. If the same factors contribute to declines at multiple sites across a large geographic area, then surviving populations likely share traits that promote persistence, while disappearing populations share traits that increase their susceptibility to decline/disappearance. Identification of environmental and spatial risk factors is useful in understanding the degree and magnitude that different variables contribute to frog declines. Using case-control methods, we examined an observational dataset of ranid frog localities collected by Arizona Game and Fish Department to investigate risk factors associated with population disappearances over time. Elevation, nonnative predators (crayfish and nonnative fish), certain hydrological characteristics (lotic versus lentic), aspect, and effects of nearby sites had a significant effect on whether a population persisted or disappeared.

\*Indicates speaker in multi-authored presentation.