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ABSTRACTS

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Restoration of Mountain Yellow-legged Frogs in Sequoia and Kings Canyon National Parks

To help reverse the decline of the mountain yellow-legged frog (*Rana muscosa*), Sequoia and Kings Canyon National Parks (SEKI) began removing introduced trout in 2001 from six lakes and adjacent streams using gill nets and backpack electrofishers. Over 1,700 introduced trout were removed from these sites in 2003, and a total of over 7,700 introduced trout have been removed since the project inception in 2001. Complete eradication of introduced trout is expected from five of the six lakes by the middle of the 2004 season. To assess frog recovery, shoreline visual encounter surveys also were conducted in the restoration areas. The average number of frogs and tadpoles detected per restoration lake was 16 times greater in 2003 versus 2001. The change observed in the strongest responding frog population was 33 times greater in 2003 versus 2001. SEKI recently received a funding award to hire an additional frog restoration crew for three seasons. As a result, removal of introduced will begin in five additional SEKI lakes in 2004. A programmatic environmental document is also being planned that will guide management of all threats to mountain yellow-legged frogs across the entire park.

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Synopsis of Current Amphibian Projects Supported or Conducted by the California Department of Fish and Game

During 2003, the California Department of Fish and Game (CDFG) supported or conducted a variety of activities that benefited native amphibians. CDFG sampled 1,984 waters during its third year of amphibian/fish surveys in the Sierra Nevada. Of these waters, 989 contained amphibians – 195 with *Rana muscosa* and 32 with *Bufo canorus*. Six sites contained *R. muscosa* and trout. CDFG conducted habitat restoration (trout removal) for *R. muscosa* in ten lakes on the east side of the Sierra Nevada, and in one stream in southern California. CDFG sponsored three research contracts including *R. muscosa* monitoring in southern California (Fisher USGS), *R. muscosa* habitat use and movement in northern Sierra Nevada (esp. stream habitats) (Vredenberg, UC Berkeley), and responses of *R. cascadae* and other wildlife to changes in fishery management in the Trinity Alps (Pope UC Davis). CDFG continued work on aquatic biodiversity basin management plans to guide amphibian conservation and angling opportunities. Basin plans provide long-term aerial fish stocking criteria; interim short-term criteria, developed in 2001, are based on presence of *R. muscosa* and evaluating stocking need based on sound fishery

management practices. CDFG began working with the State Water Resources Control Board to develop and implement amphibian survey protocols and requirements for FERC relicensing. CDFG and the USFWS produced a joint, interim survey guidance for the California tiger salamander.

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Factors Implicated in Amphibian Population Declines in the United States

Factors known or suspected to be adversely affecting native amphibian populations in the US were identified using information from 267 species accounts written in a standardized format by multiple authors in the forthcoming book, "Status and Conservation of U.S. Amphibians." Specific adverse factors were identified for 53 (58%) of 91 anurans and 93 (53%) of 176 caudates. Land use was the most frequently implicated adverse factor for both anurans and caudates. The predominant land use factors were agriculture, urban development, and timber harvest/silviculture, followed by road construction/use, livestock grazing, altered fire regime, recreational use/development, and mining. Exotic animal species were the second most frequently implicated adverse factor for anurans, and third for caudates. Exotic taxa implicated consisted of a number of introduced fishes, American bullfrogs, crayfish, defoliating insects, and other amphibians. Chemical contamination ranked third for anurans and second for caudates, and included acid precipitation, pesticides/herbicides, and mine water pollution. A regional analysis revealed that exotic species were implicated significantly more frequently among anurans in the western US than elsewhere, whereas chemical contamination was implicated significantly more frequently among caudates in the non-western US. Less frequently implicated factors were disease, water source modification, collecting/harvesting, and UV-B radiation.

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Metapopulation Processes or Infinite Dispersal?: Habitat Patch Occupancy by Toads *(Bufo punctatus*) in a Naturally Fragmented Desert Landscape (Poster)

Amphibians are often thought to have a metapopulation structure, which may render them

vulnerable to habitat fragmentation. The red-spotted toad (*Bufo punctatus*) in the southwestern USA and Mexico commonly inhabits wetlands that have become much smaller and fewer since the late Pleistocene. This study tests two predictions based on metapopulation theory --- the incidence of habitat patch occupancy is directly related to patch size and inversely related to patch isolation --- and a third, potentially competing hypothesis that patch occupancy is influenced by local environmental conditions. In a 20,000 km^2 area of the eastern Mojave Desert, 128 potential habitat patches (primarily springs) were identified and surveyed for local environmental characteristics and presence/absence of *B. punctatus*. Patch isolation metrics were based on nearest-neighbor distances, calculated both as Euclidian distance and distance via connecting drainage channels. *B. punctatus* was found at 73% of the sites, including all of the 15 historic (pre-1970) sites. Based on stepwise multiple logistic regression, the incidence of patch occupancy increased significantly with patch size, and was also significantly related to elevation, latitude, and four metrics that were associated with rocky terrain, periodic scouring water flows, and ephemeral water. In contrast, incidence of patch occupancy was not significantly related to patch isolation. These findings are consistent with a "patchy population" model, rather than the classical equilibrium metapopulation model, implying frequent dispersal among patches and virtually no local extinctions. Implicated dispersal distances of many kilometers are large for an amphibian.

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Using the ARMI Metric, Proportion Area Occupied, for Monitoring Arroyo Toads in Southern California

ARMI (Amphibian Research and Monitoring Initiative) has adopted proportion of area occupied (PAO, MacKenzie et al. 2002) as a standardized metric for midlevel monitoring of amphibian populations. In 2003, we implemented a new monitoring program using this metric for the endangered arroyo toad (*Bufo californicus*) on Marine Corps Base Camp Pendleton (MCBCP). Using the PAO approach, we are tracking the presence of breeding arroyo toads over 89 km of potential habitat. Because they have a much higher probability of detection, we are using the presence of tadpoles to document the presence of adult breeding populations. We implemented the first year of a 5 year rotating panel design by comprehensively surveying 120 randomly stratified survey lengths (250 km each). The survey lengths were visited from 1 to 4 times throughout the breeding season. We then used the loglinear modeling program PRESENCE to model the data and correct for imperfect detection probabilities. In 2003, 87.4% (se = 9.5) of wet habitat was occupied by breeding arroyo toads. We also evaluated over 14 habitat and survey specific variables in the models. These included landscape variables, environmental variables, and the presence of nonnative plant and aquatic vertebrate species. Results showed that the absence of crayfish was the single most significant predictor of the presence of arroyo toad tadpoles. At this time, we do not know if this relationship is causative or correlative.

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Chytridiomycosis in Mountain Yellow-Legged Frogs (*Rana muscosa*)

A collaborative project is underway at the University of California, Berkeley to study the impact of chytridiomycosis on *Rana muscosa* populations. The project includes large scale surveys of distribution and temporal progression of the disease through the Sierra, laboratory and field experiments studying transmission and the disease process, genetic work investigating the origin and spread of the disease, and dispersal studies to parameterize spatially-explicit models aimed at understanding the dynamics of the disease.

Laboratory and field mesocosm experiments were performed to estimate the mortality rate of post-metamorphic *Rana muscosa* and to test if infected tadpoles retain their infection as they metamorphose. In the lab at 17□C, 100% of infected tadpoles metamorphosed and died of chytridiomycosis, and 100% of uninfected tadpoles metamorphosed successfully with no mortality. Mesocosm experiments during the summer of 2003 at four infected lakes and three uninfected lakes in the Sierra Nevada, California followed tadpoles through metamorphosis. Field results mimicked lab results; infected tadpoles metamorphosed and experienced very high mortality, and uninfected experienced very low mortality.

In attempt to characterize the disease processes involved with chytridiomycosis, two factors, ambient temperature and hydration status, were found to contribute to morbidity and mortality of frogs infected with *Batrachochytrium*. Laboratory experiments conducted at UCB support previous findings of ambient temperature affects on survival rate. In *Rana muscosa,* temperatures of 22□C result in a 50% mortality rate while temperatures of 17□C result in 100% mortality. Furthermore, observational evidence collected at UCB support dehydration and hypovolemia to be present during, and contribute to, the ultimate demise of infected animals.

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Population Status, Threats, and Conservation Efforts for the Amargosa Toad (*Bufo nelsoni*)

The Amargosa toad is an amphibian endemic to the Oasis Valley in Nye County, Nevada. Due to the cooperation and efforts of several agencies, organizations and local residents, placing the toad on the Endangered Species Act list was found to be "not warranted" by the U.S. Fish and Wildlife Service in 1994. The Conservation Agreement for the Amargosa Toad (*Bufo nelsoni*) and co-occurring species in Oasis Valley, Nye County, Nevada was signed in 2000. An intensive mark and recapture survey, using PIT tags, began at various sites throughout the valley in 1998. These surveys have resulted in estimates for groups or complexes of survey sites using the computer program MARK. Valley-wide, the population estimates over the past 6 years have ranged from

1774 to 2401 toads. Based on these annual visits, the cooperators have been able to assess and address habitat needs at each survey site or complex of sites. Such needs include rehabilitation and maintenance of spring sites and non-native species removal and/or control. Upcoming projects include a pond restoration at the Harlan/Keal site and habitat rehabilitation in the Amargosa River just north of town.

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Status of the Relict Leopard Frog (*Rana Onca*): Our Limited Understanding of the Distribution, Size, and Dynamics of Extant and Recently Extinct Populations

The relict leopard frog (*Rana onca)* was once thought to be extinct, but has recently been shown to comprise a valid taxon with extant populations. Here, we discuss research from several studies, conducted between 1991 and 2001, that represent the basis for our understanding of the distribution, size, and dynamics of extant and recently extinct relict leopard frog populations. We review phylogeographic findings that have been used to identify this taxon, discuss the delineation of a minimum historical range, report the findings from mark-recapture studies and visual encounter surveys, and describe the extinction of 2 of 7 populations extant in the 1990s and speculate on the causes for these extinctions. A minimum historical range for this taxon was based on records from 24 localities (> 1 km apart) along the Virgin and Muddy River drainages and adjacent portion of the Colorado River drainage in southern Nevada, northwestern Arizona, and southwestern Utah. These frogs currently exist naturally at only 5 spring sites distributed in 2 general areas: near the Overton Arm of Lake Mead, and in Black Canyon along the Colorado River below Lake Mead, Nevada. The loss of populations in the 1990's greatly reduced the extant distribution of this frog. These population extinctions occurred concomitantly with encroachment of emergent vegetation into pools. A rough estimate for the total number of frogs at all sites in 2001 was approximately 1100 adults (range 693-1833). Annual adult survivorship from a mark-recapture study at one site was estimated at 0.27. The limited number and distribution of populations, the low estimated total population size, the high estimated population turnover, and the observations of recent population extinctions are reasons for concern about the continued existence of the relict leopard frog.

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Re-Establishing California Red-Legged Frogs to Their Historic Range Within Pinnacles National Monument

California red-legged frogs (*Rana aurora draytonii*) were once abundant in the streams and reservoir at Pinnacles National Monument, California. In the early 1980's they were extirpated from the reservoir due to an infestation of exotic catfish and the draining of the reservoir. By the late 1990's the remaining stream-breeding populations had reached very low levels. We believe that the reservoir supported the primary source population in the area, and that the stream may not provide adequate habitat to maintain self-sustaining populations. We began a project in 2001 to re-establish a breeding population at the reservoir, which is currently free of exotic predators. We moved portions of egg masses, just prior to hatching, from streams into secured holding pens in the reservoir. The holding pens were designed to protect tadpoles from predation while providing adequate food supply and water circulation. After tadpoles had reached sufficient size, they were released into the reservoir at large and their numbers were tracked using minnow traps. At the end of the third year of the project, there were at least 7 one-year-old and 20 two-year-old frogs at the reservoir. Over 400 metamorphs were produced in the third year, indicating that frogs may have already begun breeding in the reservoir. Without this project, we believe the status of California red-legged frogs in our watershed to be precarious at best. Specific methodology will be presented.

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Use of habitat by Adult Amargosa Toads (*Bufo nelsoni*)

The Amargosa toad (*Bufo nelsoni*) is narrowly a precinctive species to the Oasis Valley in Nye County, NV. This species occupies the riparian areas of the ephemeral Amargosa River, and associated springs in the Oasis Valley. Using radio telemetry, adult toads were tracked over the course of one year to determine how they use available habitat. Approximately 10 males and 10 females were tracked at two study sites. Home range sizes did not differ between male and female toads. Distances moved by individuals varied by sex and month of the year. Movements were significantly larger after rain. The frequency of animal movements declined during the fall and winter. The relative distance toads were found relative to water differed by gender, with males observed closer to water than were females. Toads were rarely found greater than 50 m from the water.

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Negative Effects of Introduced Trout on *Ambystoma macrodactylum* in the Sierra Nevada, California: Results from "Historic Site" Surveys, Watershed Surveys, and Experimental Data

Ambystoma macrodactylum sigillatum, the long-toed salamander, is thought to be experiencing local declines in areas with fish but is still common in the Sierras. I combined three methods to assess the effects of introduced trout on *Ambystoma* presence in California's Sierra Nevada. I surveyed 20 lakes that historically supported these salamanders, comparing sites with and without introduced trout to see if local extinctions were correlated with trout introductions. I also completed multiple surveys of 57 lakes and ponds in two watersheds to assess landscape patterns of amphibian and trout occurrence. Finally, I used experimental enclosures to quantify trout impacts on salamander recruitment. Failure to detect *A. m. sigillatum* larvae in sites where they were historically present was correlated with trout introductions. In the watershed survey, trout presence was negatively correlated with presence of *A. m. sigillatum* larvae. To supplement surveys, I tested trout effects on larval *A. m. sigillatum* in a lake, using eight 5 m by 5 m enclosures. Trout negatively affected larval survival and body size. In high elevation lakes and ponds, *A. m. sigillatum* larvae are known to overwinter before reaching metamorphosis. Surveys suggest that larvae are negatively affected by trout presence and are seldom present in lakes with trout. *A. m. sigillatum* populations may persist at the watershed scale because some larvae reach metamorphosis in one season when small ponds dry during drought conditions. A comparison of survey methods highlights the importance of using methods such as seining in addition to visual surveys.

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Reintroduction of a Declining Amphibian: Determining an Ecologically Feasible Approach Through Analysis of Decline Factors, Genetic Structure, And Habitat Associations

Species reintroductions provide a model for integrating practical and theoretical aspects of conservation biology and developing suitable sets of analytical tools. Reviews have suggested that research is needed in several areas to improve the success of reintroductions: (1) causes of species declines, (2) genetic relationships of source populations, (3) social dynamics, (4) habitat associations, and (5) monitoring designs. Amphibians present distinct challenges due to unique ecological, genetic, and demographic characteristic such as low mobility and patchy distributions. This study focuses on a declining amphibian in California (the foothill yellow-legged frog, *Rana boylii*) and includes three components: (1) spatial analysis to determine the primary causes of decline, (2) description of range-wide genetic variation, and (3) quantification of habitat associations. One recent study suggests that agricultural chemicals and climatic factors may be involved in the decline. My work includes more detailed analyses of these threats and adds information on dams and water flow modifications - key influences on this stream-dwelling frog.

Initial study of habitat associations shows that a narrow set of stream conditions are necessary for reproduction. Genetic data also indicate low variation throughout the range. I propose a model for integrating this and other information into reintroduction programs.

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Amphibian Diseases! Why Didn't We Think Of This Before?

Potential factors in the worldwide decline of amphibians have been hot topics for the last 25 years. Disease has now been implicated in a number of amphibian declines on 6 continents and it is likely that this is just the beginning of our understanding. Amphibian chytridiomycosis and Iridoviruses have been the primary culprits identified so far, but other diseases are also present and acting on amphibian populations. Educating ourselves as professional biologists about a variety of amphibian diseases, what to be aware of when in the field and how to minimize the risk of spreading disease pathogens need to be our primary concerns, as we continue to investigate the causes of declines in our amphibians on local and global levels.

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Amphibian Chytridiomycosis and California Herpetofauna

Amphibian chytridiomycosis has been detected in over 13 species of amphibians in the State of California. *Batrachochytrium dendrobatidis* is species-specific in its effects and even these effects can be influenced by environmental conditions. The primary order of business is determining which species are mortally affected and across which geographic ranges the mortality occurs; and also which species can be carriers and across which geographic range this occurs. Amphibians generally occur as a suite of species which will interact with each other at various points of their life history. My study site at Grant County Park, San Jose, California has 7 species of amphibians with aquatic life stages within the park boundaries. Three species are currently special status. Amphibian chytridiomycosis has been detected in 3 species, including one special status species and further monitoring over the next 5 years will assist in determining how widespread the disease is both geographically and taxonomically and what the overall effects of the pathogen is on the diverse amphibian populations within the park.

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Metapopulation Structure in Two Species of Rare Toads (*Bufo nelsoni* and *Bufo exsul*) with Implications for Conservation.

Recognizing metapopulation structure in amphibian populations is tremendously important to conservation efforts as it significantly alters our expectations with respect to population and species persistence. At the same time, misidentifying populations as metapopulations can have dire results. We review the metapopulation concept as it applies to amphibian populations, and present applications of this concept to the conservation of two species of toads (*Bufo nelsoni* and *Bufo exsul*). We used both direct and indirect methods to determine that both species meet stringent criteria as metapopulations. There are many differences in the metapopulation processes (e.g. migration rates, levels of differentiation, etc.) in each species. These differences may be best explained by the differences in the degree of physical isolation among subpopulations, and differences in climate, between the ranges of the two species. The differences in metapopulation dynamics also have implications for our expectations with respect to conservation of these rare toads.

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Amphibian Conservation Planning in Nevada: The Efficacy of Conservation Agreements and Strategies

Three of Nevada's endemic amphibians, the Amargosa toad *Bufo nelsoni*, Columbia spotted frog *Rana luteiventris* and relict leopard frog *Rana onca* are species of concern classified as protected amphibians under Nevada Administrative Code. These species have been the subject of recent listing petitions and the US Fish and Wildlife Service has determined that listing under the ESA for Great Basin populations of the Columbia spotted frog is warranted but precluded. Effective conservation for these species is challenging because of limited resources, complex agency jurisdictions and land ownerships, gaps in biological knowledge and limited understanding of potential threats.

Cooperative inter-agency efforts to implement conservation actions for these species in Nevada have focused on the development of species-specific Conservation Agreements and Strategies (CAS) which commit signatory participants to the implementation of adaptive conservation actions and the maintenance of field-level, collaborative working groups that coordinate on-ground activities, evaluate the efficacy of conservation actions and can adaptively modify initial conservation strategies based on findings, changed conditions and new information. A key element for success of this conservation approach has been the aggressive participation of the species working groups, which have identified and implemented interim conservation actions during the frequently tedious CAS development processes.

The first CAS, for Amargosa toad, was developed over a five-year period and implemented beginning in October 2000. Because the Columbia spotted frog in Nevada occurs in two distinct,

geographically disparate population segments, and conservation planning and implementation for each population segment involves unique participants, two separate but associated CASs have been developed and were implemented in September 2003. A relict leopard frog CAS, which also involves the states of Arizona and Utah, is under development and should be completed by mid-year 2004.

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Foothill Yellow-Legged Frog (*Rana boylii*) Predation by the Introduced Signal Crayfish (*Pacifasticus leniusculus*) in the North Fork Feather River, CA

As part of ongoing monitoring of foothill yellow-legged frog (*Rana boylii*) populations on the North Fork Feather River (Butte and Plumas Co., CA) for Pacific Gas and Electric's FERC relicensing projects, we employed an underwater video camera to document potential predation of *R. boylii* embryos and tadpoles. Between 16 and 27 June 2003, we operated an Aqua-Vu® underwater video camera at two known *R. boylii* breeding sites. Analysis of over 92 hours of videotape revealed that the introduced signal crayfish (*Pacifasticus leniusculus*) prey upon the egg masses of *R. boylii*. Five other species were also observed around egg masses and tadpole groups including four species of fish (two non-native species) and Sierra garter snakes (*Thamnophis couchii*). Results of visual encounter surveys suggested that *P. leniusculus* also prey upon larval stages of *R. boylii*, based upon observations of tail injuries. *Pacifasticus leniusculus* is present in large numbers in this drainage and has been observed in other systems where *R. boylii* are found, including the Pit and Stanislaus Rivers. Although the extent of predation upon *R. boylii* by crayfish is not known, other studies demonstrate that crayfish can have devastating impacts upon all life stages of native amphibians. Further research is needed to explore both the indirect and direct effects of signal crayfish on *R. boylii* populations in order to assess the potential impact this exotic species may have on the ecology and distribution of the foothill yellow-legged frog.

* Denotes speaker in multi-authored presentations