California/Nevada Amphibian Populations Task Force 2025 Meeting

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ABSTRACTS

California Newt (*Taricha torosa*) from Ramirez Canyon, Los Angeles or Ventura County, California. Photographed by Sean Anderson.

ORAL PRESENTATIONS

* Indicates presenter in multi-authored presentation

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Untangling the Complex Relationship between Mountain Yellow-Legged Frogs and Wildfire in Southern California

Recurrent wildfires are a natural component of the ecosystems found in southern California's forest and scrubland. These fires are essential to maintaining overall ecological health of these systems; however, the frequency and severity of wildfires has increased in recent times. These high intensity wildfires adversely impact stream health by increasing sediment deposition, altering nutrient loads, and reducing canopy cover. Thus, wildfires both directly and indirectly impact a wide variety of aquatic wildlife. One species that is dependent on pristine stream habitat is the endangered mountain yellow-legged frog (*Rana muscosa* - MYLF). MYLF have declined in southern California since the 1970s and currently occupy a small number (<10) of fragmented sites across three mountain ranges in southern California. These remaining sites are regularly impacted by wildfire which has also coincided with reduced or extirpated frog populations. Specifically, we have observed a pattern whereby MYLF populations increase in abundance immediately following a wildfire and then catastrophically crash three to five years post-fire. The question remains: What processes are responsible for this pattern? We propose a new study to test the following alternative hypotheses: habitat, food, water quality limitations and disease. We hope this study can be used to inform future efforts.

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Lessons in Telemetry: A Study Using Two Satellite GPS Systems on Southwestern Pond Turtles on Marine Corps Base Camp Pendleton

The USFWS's proposal to list the southwestern pond turtle as Threatened under the ESA prioritizes *Actinemys pallida* habitat conservation. To aid Marine Corps Base Camp Pendleton in managing the upland habitat surrounding known populations of southwestern pond turtles, we studied upland habitat use by nesting female turtles at two sites in 2023. We used two types of GPS units; at Cockleburr Lagoon the units were cellular enabled, while the units at San Mateo Creek were satellite only. We affixed transmitters to 13 turtles at Cockleburr Lagoon and eight turtles at San Mateo Creek. Unfortunately, the GPS units did not work as anticipated. Only three of the Cockleburr Lagoon units were functional by the end of the study, and two of the San Mateo Creek units worked at the end of the study. Despite the faulty units, we were able to obtain some GPS data from both sites. At Cockleburr Lagoon 11 of the units returned locations and at San Mateo Creek six of the units returned locations. We were not able to determine potential nesting habitat for the San Mateo Creek turtles, but two turtles at Cockleburr Lagoon used overlapping terrestrial locations, suggesting that they may have nested.

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Recovering an Endangered Frog Species through Integrative Reproductive Technologies

Integrative reproductive technologies are revolutionizing the conservation of amphibians, exemplified by the endangered mountain yellow-legged frog (*Rana muscosa*). This talk will outline a decade of advancements in ex situ breeding and genetic management, focusing on the integration of assisted reproductive technologies (ARTs) such as hormone monitoring, genome resource banking, and artificial fertilization. By addressing challenges like low fertility and offspring survivability, the program has achieved the translocation of over 8,500 individuals while preserving genetic diversity. Key findings include the role of brumation in enhancing reproductive success and post-release survival, as well as the impact of genetic screening on breeding strategies. These efforts highlight the potential of multidisciplinary approaches to mitigate amphibian population declines and promote sustainable reintroduction programs.

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Dipnet Surveys Provide Accurate Abundance Estimates for California Tiger Salamander and Pacific Treefrog Larvae

Several aquatic sampling techniques are commonly used to detect and quantify amphibian larvae. Although many techniques have been standardized, comparisons among methods have been poorly studied. We employed a removal design to investigate the detection rate of amphibian larvae using box enclosures and evaluated the reliability of timed dipnet sampling relative to the more quantitative box enclosure sampling across 13 vernal pools. For these comparisons, we studied larvae of the endangered California Tiger Salamander (*Ambystoma californiense*; CTS) and the smaller and more abundant tadpoles of the Pacific Treefrog (*Pseudacris regilla*; PTF). Based on N-mixture model estimates, box enclosure sampling captured only 5% more of the CTS larvae present than PTF tadpoles, despite disparities in their sizes and estimated abundances. Dipnet sampling was slightly more effective at detecting CTS larvae than enclosure sampling, while both methods equally detected PTF tadpoles. We found a strong relationship ($R^2 = 0.92$) between the densities of CTS larvae estimated from dipnet sampling and those calculated from the more quantitative enclosure sampling, indicating that dipnet sampling is a reliable measurement of relative abundance. We conclude that timed dipnet surveys can be an important tool for monitoring populations of CTS, and possibly other lentic-breeding amphibians, especially given its relative efficiency that can be replicated across both time and space.

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Controlled Propagation to Save the Santa Barbara Population of California Tiger Salamanders

Habitat loss and fragmentation often leave small isolated populations of amphibians that are subject to inbreeding depression. The California tiger salamander has experienced extensive habitat loss, particularly in the Santa Barbara Distinct Population Segment. These endangered California amphibians are vital to the vernal pool ecosystems in which they breed and are often a keystone predator in these vulnerable

communities. Small population sizes and poor genetic health have drastically reduced the fitness of these populations, which now face extinction without genetic interventions. Here we present two years of data from a project designed to increase the genetic health and population stability of this species. We successfully bred 9 pairs of adults from different populations to increase genetic diversity and produce healthier individuals with a greater chance of survival and lifetime success. We raised the offspring in mesocosm tanks, producing over 1,300 metamorphs that were released back into the wild. In this talk we will describe our results from the first two years and present data on the survival and size differences between genetic crosses.

DELANEY, KATY S.[1]*, SARAH M. WENNER[2], HEIDI B. CUTIA[1], MARK MENDELSOHN[1], ADAM BACKLIN[3], ELIZABETH GALLEGOS[3], ROBERT N. FISHER[3], and SETH P.D. RILEY[1].
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Ten Years in the Santa Monica Mountains: California Red-legged Frog (*Rana draytonii*) Reintroduction Successes and Challenges

Five years into the National Park Service's California red-legged frog reintroduction project, Santa Monica Mountains National Recreation Area experienced the largest wildfire ever recorded in the park. The Woolsey fire grew to 100,000 acres, burning the source population of frogs, as well as every reintroduction site. Now, 6 years later, some sites have recovered, and some have not. In this talk, we will present data on the translocation project status, as well as information about reproduction, survival, and population establishment in the Santa Monica Mountains.

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Chileno Valley Newt Brigade: 2024 Update

A short film by Lori Eanes presents the story of the Chileno Valley Newt Brigade and their ongoing mission to help California and rough-skinned newts (*Taricha torosa, T. granulosa*) survive the annual migration across Chileno Valley Road between their terrestrial habitat and their aquatic breeding habitat in Laguna Lake, Petaluma, California. The film focuses on the importance of amphibian wildlife road crossings at these critical migration points. In addition to their teams of extraordinarily dedicated volunteers who brave rainy winter nights to move newts, CVNB has embarked on a project to install road crossings for the newts and fellow amphibians of Chileno Valley. The film will be followed by a short update on the crossing project and the rescue effort.

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Effect of Wetland Removal and Creation on *Rana draytonii* (California Red-Legged Frog) in a Dune Complex of Central California, USA: Evidence from over Two Decades of Research.

The loss of natural wetlands is often offset through the creation of mitigation wetlands. However, uncertainty remains if created wetlands adequately replace the vertebrate habitats that were originally lost. Between 2000-2023, we measured the relative abundance of California red-legged frogs (*Rana draytonii*) in removed

(n = 43), created (n = 13), and unaltered (n = 5) wetlands in San Luis Obispo County, California. Throughout our study, juvenile and adult abundances declined in removed wetlands, increased in created wetlands, and remained stable in unaltered wetlands, but juvenile and adult abundances between the three wetland types were not statistically significant. Throughout our study, 77% of the 13 created wetlands were colonized by adults and juveniles, and oviposition was detected in 54% of these wetlands. Further, we investigated the biotic and abiotic factors that were positively and negatively associated with adult and juvenile counts in the three wetland types, and we specifically investigated what factors make some created wetlands better at supporting the species than others. The results of our study indicate created wetlands as important features that can support all life stages of California red-legged frog, and created wetlands should be considered a viable conservation strategy for this federally threatened species.

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Amphibian Update for Yosemite National Park – Success of New Management Actions and Future Planning Efforts

In 2024, a newly reintroduced population of California Red-legged frog (*Rana draytonii*) successfully reproduced and recruited new individuals in the presence of invasive American bullfrog (*Lithobates catesbeianus*). Although preliminary, *R. draytonii* population re-establishment may be possible with concurrent *L. catesbeianus* removal. Second, we will present on future planned management actions for Foothill Yellow-legged frog (*R. boylii*). Third, we will present on reversing the effects of drought and decline of Sierra Nevada Yellow-legged frog (*R. sierrae*) in a single meadow population. Lastly, we will present on the planning, results, and future actions of the first attempt (pilot) reintroduction of the Yosemite toad (*Anaxyrus canorus*) to a formerly occupied meadow.

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Monitoring Methods for Tidal Marsh Populations of Northwestern Pond Turtle (*Actinemys marmorata*, NPT) within Suisun Bay, CA

Although NPT are often referred to as a freshwater species, a robust population is present within the brackish tidal marshes of the Suisun Bay. VNLC monitored NPT for five years (2020-2024) at the 5,733-acre Military Oceanic Terminal Concord (MOTCO) military base which supports tidal marsh habitat. Both visual encounter surveys (VESs) and basking platform camera trap stations (BPCTSs) were employed to better understand the relative distribution and density of turtles at the site. VESs were primarily conducted by kayak. Repeated VESs provided suitable data regarding presence, distribution, and abundance of turtles but individual surveys provided inconsistent data. We deployed BPCTS from June through August, and collected photos every half hour during the daytime. The BPCTS setup was challenging to perfect in the dynamic tidal system and will be discussed more thoroughly in the presentation. BPCTS monitoring produced consistent results about the abundance of turtles in certain parts of the slough, with as many as 22 turtles on the platform in a single photo, and with some basking platforms experiencing daily use. Many questions remain about this population with regards to the location of nesting habitat, habitat preferences and suitability, and potential establishment of non-native turtle populations.

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15 Years of Mountain Yellow-legged Frog Reintroductions: Lessons Learned

The mountain yellow-legged frog (*Rana muscosa*, *Rana sierrae*) has disappeared from more than 90% of its historical range due in part to disease caused by the amphibian chytrid fungus (*Batrachochytrium dendrobatidis* - Bd). We conducted a 15-year frog reintroduction study in California's Sierra Nevada that indicated that reintroduction of resistant frogs can allow reestablishment of extirpated populations. This research provided insights into several aspects of reintroductions that have relevance for ongoing and future efforts to recover mountain yellow-legged frogs across their range. These include outcomes from frog translocations versus reintroductions of captive-reared individuals, effect of captive rearing on diurnal versus nocturnal frog activity, fitness differences between donor populations, effectiveness of "immune priming" methods to reduce frog susceptibility to Bd, and the predictability of reintroduction success. Although research into some of these issues is ongoing, I will briefly discuss our current understanding of each one and their implications for frog reintroduction efforts.

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Divergence Time Estimation and Paleobiogeography of the Salamander Subgenus *Plethopsis*

The disparate global range of Plethodontid salamanders in the *Batrachoseps* subgenus *Plethopsis* is unique, and implicates perplexing historical biogeographic scenarios. Using uncorrelated relaxed molecular clock methods and fossil-calibrated divergence estimates from Shen et al. (2016), we present a time-scaled phylogeny for the genus *Batrachoseps* in order to test hypotheses concerning the diversification of the subgenus *Plethopsis.* Our estimated divergence time intervals detract support from a hypothesis that *Batrachoseps robustus* diverged as the flow of the Owens River changed course at ca. 3.2 Ma, as evidenced by sediment deposits at Searles Lake (Phillips, 2008). Instead, our estimates support that diversification of the known *Plethopsis* species began earlier, in the late Miocene to early Pliocene, as extensional activity formed the proto-Owens Valley and led to ensuing hydrological and climatic changes, driving vicariance between populations in the proto-southern Sierra Nevada and Inyo Mountains. Moreover, our estimates support a scenario wherein *B. campi* and *B. wright* diverged in the Pliocene as ancestral populations of *B. wright* expanded northward to Oregon via a corridor of relatively mesic habitat in the western Great Basin.

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Occurrence and Surface Availability of Siskiyou Mountains Salamanders (*Plethodon stormi*) and Scott Bar Salamanders (*P. asupak*) in Northern California

Estimating the distributions of cryptic species is essential for conservation, yet understanding distributions is hampered by animal behavior and imperfect detection. We developed and implemented a multi-scale occupancy survey protocol to estimate the probability of occurrence, probability of being active on the surface, and detection probability of two range-restricted terrestrial salamanders, Scott Bar (*Plethodon*

asupak) and Siskiyou Mountains salamanders (*P. stormi*), in northern California. We established survey sites near locations of historical occurrence of these salamanders and surveyed each site on 1–6 occasions in the late fall of 2023 and spring of 2024. We used WAIC to compare models with different environmental variables for predicting salamander occurrence and surface activity. Much model selection uncertainty in the effects of covariates existed, but we were able to estimate occurrence probability (0.29 [95% equal-tailed interval = 0.12–0.59]), probability of activity on or near the surface (0.76 [0.37–0.99]), and detection probability (0.34 [0.17–0.60]) using our sampling protocol. Our study highlights the difficulty of quantitatively sampling behaviorally cryptic salamanders and suggests that substantial survey effort may be required to estimate the effects of environmental conditions on terrestrial salamander occurrence and activity.

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Inter-Toad Gene Flow: A Genomic Assessment of Hybridization Between Western and Yosemite Toads (*Anaxyrus boreas* × *canorus*)

For nearly seven decades there has been intrigue and confusion over whether Yosemite Toads (*Anaxyrus canorus*) hybridize with the more widespread Western Toad (*Anaxyrus boreas*). Although morphology has provided convincing evidence in some instances, identifying hybrids through morphological characteristics can be difficult as hybrids can express a mosaic of parental phenotypes or even be indistinguishable from one parental taxon. Morphology also does not allow for the determination of the degree of genetic introgression, hybrid class, or time since hybridization. The listing of the Yosemite toad as Threatened under the ESA highlights the need for conservation management, and a definitive approach for identifying *A. boreas* × *canorus* hybrids. We developed a genomic hybrid panel by coupling new and existing DNA sequence data for Western and Yosemite Toads, and putative hybrids from the Blue Lakes region of Eldorado National Forest (Alpine County, CA). We discovered that ~90% of samples were backcrossed hybrids, containing up to 20% Yosemite Toad ancestry, up to 30% inter-species diversity, originating as recently as 5–6 generations ago. Interestingly, more ancient introgression is evident within several Yosemite Toad populations in Yosemite and Kings Canyon National Parks. Future work should first fill in sampling gaps to perform hybrid cline analysis, and ideally assess hybrid fitness directly using common garden experiments. This study was focused on a small region of the Northern Contact Zone between these two species but has applicability across the entire range of the Yosemite Toad.

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Yosemite Toad Reintroduction: Survival, Behavior, and Predation at 'Toad Island' in Yosemite National Park

Yosemite toads (*Anaxyrus canorus*) were once abundant but now occupy less than 50% of their historical range. Climate change-related factors, including reduced snowfall, higher temperatures, and increased fire frequency, have disrupted toad phenology and hindered connectivity between seasonal habitats. At Toad Island, a site that previously supported a healthy population, Yosemite toads have not been observed since the 2013 Rim Fire. To support species recovery and develop reintroduction methodologies, a pilot project was initiated to re-establish Yosemite toads at this location. In 2022, tadpoles were collected from three robust populations, and the San Francisco Zoo headstarted a cohort for reintroduction. In June 2024, 118

adult toads were released at Toad Island. To assess dispersal, habitat use, and survival, we equipped 39 toads with radio transmitters and tracked their movements from June to September, conducting weekly visual surveys to monitor untagged individuals. Here, we present initial findings on movement patterns, morphometrics, predation risks, and lessons learned from the 2024 reintroduction effort.

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Hanging On in the Canyon: Status Update on the Kern Canyon Slender Salamander (*Batrachoseps simatus*)

The slender salamanders (genus *Batrachoseps*) are a group of small lungless salamanders ranging from Oregon to Baja California. Notably, 8 species are found in the southern Sierra Nevada and adjacent Tehachapi Mountains, although instances of sympatry are rare. Four of these species are regional endemics with especially small ranges. Among these is the Kern Canyon Slender Salamander (*Batrachoseps simatus*), found only in the lower Kern River Canyon. Historically, this species has presented challenges to researchers given its very limited periods of surface activity in a region that often experiences multi-year droughts. In the last few decades, *B. simatus* has become increasingly difficult to detect, suggesting population declines or even extirpations. Identifying causes of these declines has been elusive, but in 2022 the U.S. Fish and Wildlife Service proposed *B. simatus* for federal listing as Threatened. Despite apparent declines in the core of its range, in recent years new populations have been discovered in unexpected areas; these discoveries might have implications for the listing status and long-term management of the species.

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Post-fire Update on the Relictual Slender Salamander (*Batrachoseps relictus)*

The genus *Batrachoseps* comprises a species-rich group of 22 small, mostly elongated, terrestrial salamanders restricted to the Pacific Coast. As a group, they are characterized by a strongly conserved morphology, low vagility, and generalized habitat requirements. Several species in the genus are considered micro endemic. One of these, the Relictual Slender Salamander (*B. relictus*), is found only at 13 sites on Breckenridge Mountain at the southern end of the Sierra Nevada. *B. relictus* stands out from other members of the genus in having adopted a semi-aquatic lifestyle. These factors led the USFWS to propose *B. relictus* for listing as Endangered in 2022. Subsequently, in July 2024 the Borel Fire burned 59,288 acres on Breckenridge Mountain and adjoining Piute Mountains, becoming the second largest California wildfire of 2024. The fire burned through 9 of 13 *B. relictus* sites with many experiencing moderate to high severity fire. Post-fire surveys by our team and USFS biologists revealed positive detection of salamanders at 3 of 9 sites. However, the long-term sustainability of these sites is uncertain given this species' unique natural history. We offer suggestions for innovative management strategies for the conservation of *B. relictus*.

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Hiring a Molecular Bounty Hunter: A CRISPR-based Assay for Rapid Detection of Chytrid Fungus

Chytridiomycosis, caused by the fungus *Batrachochytrium dendrobatidis* (Bd), has been associated with declines in amphibian population worldwide. Non-invasive diagnoses rely on swabbing and quantitative PCR (qPCR) assays. However, processing samples via qPCR requires molecular biology experience and expensive instruments. Outsourcing to laboratories can further increase project costs and wait times. CRISPR-based diagnostics have enabled the development of novel methods for pathogen detection. Specific High-sensitivity Enzymatic Reporter unLOCKing (SHERLOCK) assays use the CRISPR-Cas13a enzyme complex, a molecular "bounty hunter" that detects target nucleic acids and produces a measurable fluorescent signal. These assays are rapid (< 1 hr) and sensitive at low DNA concentrations. Additionally, non-geneticists with minimal training can perform SHERLOCK reactions. We developed a SHERLOCK assay to detect a molecular barcode of the Global Panzootic Lineage of Bd (BdGPL) from swabs and eDNA samples. Our assay positively detected culture-derived DNA from BdGPL and three additional Bd strains within 35 minutes of SHERLOCK reaction initiation in the laboratory. Our assay detected down to 5 copies of BdGPL DNA during initial sensitivity testing. With further developments, our assay will facilitate the monitoring of Bd in captive and wild environments. Findings are preliminary and provided for timely best science.

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Mapping Invasive Bullfrogs across California: Insights on Future Directions for Monitoring and Management

One of the most pervasive world-wide invaders is the American bullfrog (*Lithobates catesbeianus*). Currently, there is limited cohesive information about the locations of bullfrogs in California. To help prioritize and inform management efforts, there are ongoing efforts to collate bullfrog observations from disparate data sources into a single map/dataset that visualizes bullfrog distribution and expansion in the state. These data sources include community-generated observations from platforms like iNaturalist, a CDFW bullfrog observation app, and opportunistic sound recordings of bullfrogs that were collected via bird monitoring efforts. Leveraging on-the-ground and sound-based observations provides a unique dataset that we can use to better understand and mitigate the drivers of bullfrog expansion, including analysis of relationships with covariates like landscape disturbance and biodiversity. Further, having a web-based platform will help streamline reporting requirements and identify early invasions.

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A Year in the Thermal Life of the Relict Leopard Frog (*Rana onca*) at Hot and Cold Springs

The relict leopard frog is a species of conservation concern that occupies a narrow range in the eastern Mojave Desert. Hot springs have been perceived as favorable habitat for the species, which seems reasonable given that all remaining historical (relictual) populations occupy hot springs. On the other hand, populations at some of the hottest springs are generally small with limited recruitment, while several large, robust

populations have been established at cold springs through translocations. We aimed to understand the thermal ecology of this species by comparing temperature profiles of frogs at hot and cold springs, especially during the critical seasons of summer and winter when ambient temperatures are at extremes. We collected continuous body temperature data from adult frogs in the wild using implanted data loggers, documenting patterns in the populations over a year. We also placed loggers within habitat to record general ambient temperatures. In the laboratory, we used a thermal gradient to determine seasonal temperature preferences of adult frogs captured from both hot and cold springs during summer and winter. The intent of this research was to provide managers with information to assess habitat quality for the relict leopard frog and better target efforts to expand critical habitat.

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Habitat Use and Partitioning by Foothill Yellow-Legged Frogs and Red-Bellied Newts in their Aquatic Life Stages

How do amphibians share stream habitat in their aquatic life stages? The answer is: we have a lot to learn! This study investigates two obligate stream-breeding amphibians whose ranges, breeding seasons, and habitat overlap spatially and temporally in first order streams. We 1) collected data on the aquatic life stages of Foothill Yellow-Legged Frogs (*R. boylii*) and Red-Bellied Newts (*T. rivularis*), 2) applied the existing body of knowledge about *R. boylii* that is currently based in riverine and higher order stream systems to small first order streams, and 3) compared and contrasted it with habitat use between these two species. We collected data at the meso-, sub-, and microhabitat scale, including canopy closure and fish presence, and looked at spatial distribution throughout aquatic life stages. Key findings highlight the value of runs at the mesohabitat level, low gradient riffles and pocket water runs at the subhabitat level, and patterns in substrate type, water flow rate, and canopy cover among species and life stage. We also identify a novel subhabitat designation that is highly influential to larval life stages: protected side channel pools. Based on these results, we offer specific management recommendations that can support egg and larval development for *R. boylii* and *T. rivularis*.

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Habitat Use and Movement of the California Red-legged Frog (*Rana draytonii*) in the Rock Creek Off Highway Vehicle Area in the Eldorado National Forest, California

Habitat use and movement patterns of the federally threatened California red-legged frog (*Rana draytonii*) are not well understood in the Sierra Nevada foothills. The Eldorado National Forest conducted a study in 2021 using radio telemetry and Passive Integrated Transponder (PIT) tags to advance the current knowledge of how these cryptic frogs move around the Rock Creek Trail System in the Georgetown Ranger District. We designed our study to address the concern for migrating adults during the rainy seasons of spring and fall in the Rock Creek OHV area, when wet conditions may trigger closures of the OHV area. We put radio trackers on 15 adults in a known breeding pond on the Forest and in nearby Bear Creek during spring and fall, with the aim of evaluating habitat use when frogs are expected to be migrating overland during the wet season. The breeding pond and the creek are 360 m straight-line distance apart through upland habitat. None of these frogs were tracked moving overland when the radio tracker belts were on them. However, useful information was observed by following the PIT tagged individuals during breeding (when females were too gravid to put radio tracking belts on them) and the dry season. We PIT tagged 31 individual frogs: 4 subadults, 11 adult males, and 16 adult females. Five individuals were recorded in both the pond and the creek, 11 individuals were only found in the pond, and 15 individuals were only found in the creek. This does provide evidence that some adults are moving between the creek and the pond, though it is uncertain if

they are moving through the upland habitat a straight-line distance of 360 m or through the hydrologically connected ephemeral drainage that empties downstream into Bear Creek, a total distance of up to 805 m.

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Landscape Genetics of the Western Pond Turtle: Complex Hybridization in a Complex Landscape

The western pond turtles, *Actinemys marmorata* and *A. pallida*, historically occurred over much of western North America, including the majority of California and northern Baja. The group has been in a state of taxonomic flux for decades, both in terms of its generic allocation and the recognition of species/subspecies-level lineages. In 2014, we published the first range-wide analysis of single nucleotide variation across western pond turtles, found little evidence for introgression, and recognized two distinct species. Since then, we have completed a more extensive RADseq analysis of 1393 individuals from across the species range. Our final data set came in at over 230,000 high-quality SNPs after stringent quality filtering, and approximately 41,000 SNPs after thinning of physically proximate SNPs along each chromosome. Our results indicate that there is more admixture in the central coast than previously thought, and that there is additional evidence of strong population structure within each lineage. We are in the final phase of sequencing an additional >200 individuals at the whole genome level as part of the California Conservation Genomics Project. Our goal is to use both the RAD and whole genome resequencing data to reevaluate the taxonomy and history of the clade.

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Range-wide Landscape Genomics of the California Tiger Salamander

The California tiger salamander (*Ambystoma californiense,* "CTS") is a Federally and State listed species, and faces myriad threats across its range in California. The primary drivers of decline, habitat degradation and destruction, may lead to fragmented and isolated populations, which in turn lead to population declines due to reduced genetic diversity and inbreeding depression. Therefore, an understanding of distribution of genetic diversity and patterns in connectivity is essential to adequately manage populations to support recovery. Here we generated a genetic dataset from 2,529 native CTS from more than 400 localities across all three Distinct Population Segments. We examined population structure across the range and identify potential management units. We then modeled metrics of genetic diversity as a function of landscape attributes to identify variables that may promote genetic diversity and healthy populations. We found that ponds that occur in complexes with several ponds within dispersal distance tend to have higher diversity than those in isolation and that landscapes with fewer ponds exhibit higher resistance to gene flow. These findings suggest that management of CTS at the landscape level should place a very high value on connected networks of ponds within the dispersal distances of CTS.

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Private Property, Pot Grows, and Putative Species: Microendemism in Stanislaus River Canyon *Batrachoseps*

Slender salamanders (genus *Batrachoseps*) have presented an intriguing challenge to researchers attempting to study them. This speciose group of small plethodontid salamanders includes taxa that occupy difficult terrain with low numbers of surface-active animals encountered during surveys. Highly conserved morphology across deeply diverged species groups has historically masked the extent of diversity in this genus; in the last 70 years the genus has expanded from 2 to 22 described species. Our current work on the Hell Hollow slender salamander (*B. diabolicus*) highlights many of the realities of research on this genus of enigmatic amphibians. *B. diabolicus* is found in the lowlands of the Sierra Nevada foothills, a fire-prone region which is also rapidly growing in terms of human population. When described, this species was noted to encompass staggering genetic diversity, with highly divergent populations found in the central portion of the species' range. In this presentation, we will share some recent results and progress from current work on this species, as well as some of the logistical realities of researching *Batrachoseps*. Our expanded sampling and modern genetic approaches have allowed new insight about habitat preferences, range extent, and population diversity for this member of an at-risk salamander group.

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Assessment of Wildfire Impacts on Foothill Yellow-legged Frogs (*Rana boylii*) from the North Fork Feather River, California

Wildfires within the range of the Foothill Yellow-legged Frog (*Rana boylii*) have increased in frequency and severity in recent years, particularly in the Sierra Nevada, yet the long-term effects of these fires on frog populations are poorly understood. The Camp Fire of 2018 was the deadliest and most destructive fire in California history, burning over 150,000 acres. For the past 21 years (2004-2024), we have conducted annual egg-mass and mark-recapture surveys on the North Fork Feather River, where the Camp Fire burned occupied frog tributary habitat with moderate-to-high burn severity. While we did not observe any direct mortality on post-metamorphic frogs, preliminary analysis of our survey results suggests that this species has some tolerance to large wildfires, six years-post fire; however, other indirect effects such as siltation of breeding and rearing habitat remain unknown. Assessment of impacts such as wildfire to amphibian species are facilitated by long-term population data, highlighting the importance and value of obtaining these types of data over multiple decades.

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Can We Save Them All? A Framework for Successful Anuran Head-Start Programs

The San Francisco Zoo's Conservation Department has reared over 7,500 threatened and endangered subadult anurans for release over the past decade. We reflect on the outcomes of cohorts of Mountain and Sierra Nevada Yellow-legged Frogs and California Red-legged Frogs raised in our facility. We share results

on population trends in *ex situ* survival, metamorphosis, and disease to provide a framework for future head-start programs in California. We highlight opportunities for research between zoos and universities, consulting firms, and state and federal agencies to collaborate on saving local species.

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Effects of Invasive American Bullfrogs and their Removal on Northwestern Pond Turtles

The American bullfrog is an invasive species known for its voracious diet, and native turtles are among those eaten by bullfrogs. We examined the effects of bullfrogs and their removal on Northwestern pond turtles at four sites in Yosemite National Park. From 2016–2022, we monitored turtle populations in two sites where bullfrogs were present and two where they have been absent. We removed 12,317 bullfrogs, larvae, and whole egg masses from one site and 4,067 from the other. We captured just large adult turtles where bullfrogs were present compared with all sizes where bullfrogs were absent. Before nearly complete eradication, juvenile turtles were only found with bullfrogs when they were recovered from bullfrog stomachs. Turtles at bullfrog-present sites were 26–36% larger and 76–97% heavier than turtles from bullfrog absent sites. Turtle abundance and densities were also 2–100 times higher at bullfrog absent sites. We captured the first juvenile turtles at bullfrog-present sites only after reaching nearly complete bullfrog eradication in 2019. Our study shows a prolonged lack of juvenile turtle recruitment where bullfrogs were present but offers hope that bullfrog control may succeed in recovering turtle populations by easing predation pressure on small turtles.

POSTERS

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Developing a Bioacoustic Workflow with Audiomoth, Kaleidoscope, and RavenPro for Anuran Conservation

The emergence of affordable automated recording units (ARUs), machine learning-based automated detection, and sound analysis tools has brought about a surge of interest in bioacoustics for conservation applications. As the CSU Channel Islands campus lies adjacent to the Santa Monica Mountains National Recreation Area (SMMNRA), this is an ideal setting to deploy ARUs to monitor local anuran species assemblages. With this in mind, we sought to develop a low-cost, scalable workflow for bioacoustic monitoring of local anurans. Our methodology integrates AudioMoth recorders with automated call detection in Kaleidoscope and fine-scale acoustic measurement capabilities of RavenPro, enabling comprehensive species detection and characterization. Kaleidoscope has pre-dominantly applied machine learning to bird and bat species-specific identification, and there remains a need to improve anuran species detection on the platform. The workflow leverages Kaleidoscope Pro's noise filtering preprocessing and cluster analysis to automatically group similar vocalizations within extensive audio datasets while RavenPro software allows for acoustic measurements of vocalizations such as call duration, peak frequency, and bandwidth, and to export these measurements for statistical analyses. By combining these affordable bioacoustic hardware with

advanced audio processing and measurement tools, we will demonstrate a workflow for cost-effective, anuran population monitoring.

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Identifying Methods for Eradicating African Clawed Frogs (*Xenopus laevis*) in the Santa Monica Mountains

African Clawed Frogs (ACF, *Xenopus laevis*) are a growing ecological concern as they have impacted native amphibians and are a host for diseases. The National Park Service (NPS) first detected this species in Santa Monica Mountains National Recreation Area on June 8, 2022 at the Rancho Sierra Vista (RSV) Pond. Since the first detection, NPS has consulted with local, state, and federal natural resource agencies and other institutions in determining methods for reducing the established population and preventing the spread of ACF in the park. Over the past couple of years, we have implemented methods of control including seining followed by euthanasia. We are considering drift fencing and pitfall traps around the pond to capture any individuals dispersing. In 2022, approximately 200 were removed from a culvert near the RSV pond. In 2023 and 2024, we caught and euthanized thousands of frogs at all life stages. Although we have considered and implemented methods of action, we are looking to identify the source location and develop a *Xenopus* task force to discuss future methods of control. As our efforts to mitigate this species from spreading continue, we invite suggestions and collaborations in order to protect our native herpetofauna.

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Investigating Terrestrial Habitat Use and Movement of Western Pond Turtles (*Actinemys marmorata*) during a Montane Meadow Restoration Project in Yosemite National Park

California's only native turtle, the western pond turtle species complex *(Actinemys marmorata* and *A. pallida*), are candidates for listing under the federal Endangered Species Act due to declines throughout their range. To better inform conservation and management actions, several studies have investigated western pond turtle terrestrial habitat use throughout California, including the lower elevations of the Sierra Nevada, which is considered as a stronghold for the species. However, little is known about upland habitat for estivation and/or overwintering use associated with various aquatic habitat types (i.e., ponds, small streams, seasonally flooded pools, and meadow habitats). Ackerson Meadow, located at 4,600 feet elevation, rests close to the western pond turtle's eastward and elevational range boundary. Additionally, it is the site of the largest meadow restoration project, by fill volume, to date within the Sierra Nevada. Since 2022, Yosemite biologists have been monitoring turtle movements at the site to understand the natural estivation/overwintering habits of *A. marmorata* in the montane zone of the Sierra Nevada, and document turtle responses to restoration actions. We tracked the timing of departure from, and return to, their aquatic habitats, overwintering site distance to nearest water, and characterized overwintering site selection. This poster will present radio-telemetry data on terrestrial habitat use for estivation and overwintering at the site. It will also give an update on findings from this project that build knowledge of the western pond turtle's natural history in the montane zone and further the development of protections for the species during projects that impact occupied habitats.

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Removal of Introduced American Bullfrogs (*Lithobates catesbeianus*) in Yosemite Park, California, 2019-2024

Introduced American bullfrogs (*Lithobates catesbeianus*) threaten freshwater ecosystems by predating on native fauna, competition, disease spread, and other direct and indirect mechanisms. Bullfrogs were first officially recorded in Yosemite National Park in 1955. During ten years of targeted bullfrog eradication (2005-2015), the National Park Service (NPS) removed the last observed signs of bullfrog breeding in Yosemite Valley in 2012, began reintroducing historically native California red-legged frogs (*Rana draytonii*) to the valley in 2016, and removed the last observed bullfrog from the valley in 2019. Following the success of eradication and reintroduction efforts in Yosemite Valley, the NPS began removing bullfrogs from four lentic sites near Hetch Hetchy Reservoir: Gravel Pit Lake and Swamp Lake, located in park-designated wilderness, Mud Lake (USFS), and one adjacent site. These sites contain historic red-legged frog habitat and may facilitate further bullfrog invasion without proper management. Across all four sites, the NPS removed over 15,000 bullfrogs of varying life stages from 2019-2024. This poster summarizes these six years of targeted bullfrog eradication, including removal techniques, population and size class fluctuations over time, and important lessons learned.

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Aquatic Prey in an Introduced Population of African Clawed Frogs (*Xenopus laevis*) in the Santa Monica Mountains

African clawed frogs (*Xenopus laevis*) are known to feed on different types of prey including invertebrates, fishes, and amphibians, and potentially impact the structure of aquatic communities. Populations of this non-native species are distributed in freshwater habitats throughout southern California and have been recorded in several protected areas. We conducted a survey of aquatic prey of *X. laevis* found in a freshwater pond in Rancho Sierra Vista/Satwiwa, on the western portion of the Santa Monica Mountains National Recreation Area. We performed dissections to examine the stomach contents of 60 specimens and (where possible) identified arthropod prey items to order or family level. The most frequent prey items in our sample were Ephemeroptera (mayfly nymphs), Odonata (dragonfly nymphs), and aquatic Hemiptera (Notonectidae, backswimmers). We compare our findings with those of previous studies of trophic ecology of *X. laevis* in southern California and discuss its potential impact on local aquatic ecosystems.

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Polymorphism as a Precursor to Speciation in a California Endemic Salamander

The salamander species *Batrachoseps stebbinsi* is an underrepresented subject in morphological studies. Authors have noted morphological differences and genetic divergence between northern and southern populations. This has led to speculation as to whether or not the northern and southern populations are separate species due to their observed morphological differences and the apparent gap in their distribution.

To date, no formal analysis of range-wide morphological data has been published. In addition, this species contains distinct color morphologies that appear to be correlated with habitat (Figure 1). This study aims to quantify the color morphology and shape differences (body and limb proportions) between northern and southern populations of *B. stebbinsi*. The role of sexual dimorphism and size at maturity will be included in the analysis. Whether selection is driving the observed variation in color and shape differences is not yet known.

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Elevational Patterns of Body Size, Limb Size, and Trait Loss in Terrestrial-breeding Frogs

Terrestrial-breeding frogs (Strabomantidae) belong to a diverse clade distributed throughout the Andes and Amazon region. Species living at high elevations experience colder temperatures than those at lower elevations and may experience different selection pressures promoting morphological change. We investigated patterns of morphological change in the context of two hypotheses, Bergmann's rule (organisms living in cooler climates tend to have larger body size than those in warmer climates) and Allen's rule (organisms living in cooler climates tend to have shorter appendages than those in warmer climates). We tested if species at high elevations tend to be larger than those at low elevation, and if they have relatively shorter limbs than those at low elevation. We compiled a dataset for ~300 species representing six genera and used elevation as a proxy for temperature to assess the relationship between morphological traits and elevational midpoint. Additionally, we used an X-ray micro-computed tomography (micro-CT) system to obtain images (micro-CT scans) of the skull and skeleton of selected species and characterize their osteology. Bergmann's rule was supported in one of the six examined genera, while Allen's rule was supported in three of the six examined genera. We also observed the loss of skeletal elements in some of the frog species analyzed and discuss trait variance among closely related species.