Hydrological Regimes, Pond Morphology, and Habitat Use: Predicting the Impact of an Emerging Aquatic Pathogen

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Chytridiomycosis, a newly-identified fungal disease of amphibians, is leading to rapid die-offs of mountain yellow-legged frog populations in some parts of the Sierra Nevada, but not in other sites where frogs persist with only mild fungal infections. Our data indicate that this spatial difference in disease outcome is not due to differences in fungal virulence or frog susceptibility, and now we are exploring other potential causative factors including characteristics of the physical environment such as pond morphology and hydrologic regime.

Declines in amphibian populations have been reported throughout the world in recent years. A number of factors have contributed to these population declines, including disease, introduced species, habitat loss and alteration, and climate change. Chytridiomycosis is a potentially fatal disease of amphibians caused by the chytrid fungus, *Batrachochytrium dendrobatidis*, which has appeared recently in the aquatic habitats of California and throughout the world. In portions of the Sierra Nevada mountains of California, the disease is causing rapid die-offs of mountain yellow-legged frogs, *Rana muscosa*, a threatened native frog species. In other areas of the Sierra, infected populations of *R. muscosa* appear to be persisting with the fungus. In this study we are investigating why the fungal pathogen is having different outcomes on frog populations in different California watersheds.

We are exploring a number of factors, including: (a) differences in pond morphology or topography of the landscape in different areas that result in the frogs using the habitat differently at the different sites, altering their risk of acquiring and succumbing to the disease; (b) differences in the transmission, infectivity, and/or virulence of the fungal strains at the different sites; or (c) differences in susceptibility of the frog genotypes at the different types of sites which could lead to the observed differences in the impact of the fungal pathogen. As part of (c) we are investigating the effects of antimicrobial peptides, which are released from the frog skin, which may determine the frogs' susceptibility to disease.

During the summers of 2004 and 2005 we conducted detailed surveys at sites in the Sierra Nevada experiencing *R. muscosa* die-offs due to chytridiomycosis, and sites with *B. dendrobatidis* present with persistent *R. muscosa* populations. We used a newly-developed, non-destructive, quantitative PCR protocol to determine the infection status (presence/absence of *B. dendrobatidis*) and infection level (fungal loads) of *R. muscosa individuals*. We marked adult *R. muscosa* and recaptured and re-swabbed the same individual repeatedly. We found that infected frogs at die-off sites carried very high fungal loads, while at persistent sites the infected frogs were experiencing only low to moderate infections. Interestingly, at the persistent infected sites, some adult *R. muscosa* were found to lose the infection through time, and some infected individuals survived over the long overwintering period. Data are still being gathered on hydrological regime, pond morphology, and habitat use.

Prior laboratory experiments found no evidence that differences in fungal strains are responsible for the different population-level impacts of the disease at the different sites.

In recent experiments we investigated variability the susceptibility of *R. muscosa* to *B. dendrobatidis*, concentrating on the impacts of antimicrobial peptides in the frogs' defense against disease. We exposed subadult *R. muscosa* to different quantified doses of *B. dendrobatidis*. In some treatments we removed the antimicrobial peptides from the frogs prior to exposure to *B. dendrobatidis*, and in other treatments the peptides were not removed. It was predicted that if antimicrobial peptides served to defend the frogs against the disease, then individuals from which peptides had been removed prior to exposure would be more likely to become infected. However, we found absolutely no difference between the fractions of individuals that became infected after peptide removal versus those that were exposed to the same dose without prior peptide removal. This suggested that the

antimicrobial peptides are not sufficient to protect *R. muscosa* from chytridiomycosis in even a simple experimental situation, and therefore that differences in antimicrobial peptide production between sites is unlikely to be responsible for the observed differences in the population-level impacts of this disease.

Publications

Briggs, C.J, V.T. Vredenburg, R.A. Knapp, and L. J. Rachowicz., Investigating the population-level effects of chytridiomycosis, an emerging infectious disease of amphibians, Ecology, in press.

Rachowicz, L. J., Transmission of an emerging infectious disease in a declining amphibian species: *Batrachochytrium dendrobatidis* in the mountain yellow-legged frog (*Rana muscosa*), Ph.D. Dissertation, University of California, Berkeley, 2005.

Professional Presentations

Briggs, C. J., Investigating the population-level effects of chytridiomycosis, a fungal disease of amphibians, Invited Seminar, Oregon State University, Zoology Department, Corvallis, OR, October 17, 2004

Briggs, C. J., Investigating the population-level effects of chytridiomycosis, a fungal disease of amphibians, Invited Seminar, Arizona State University, Department of Mathematical Biology, Tempe, AZ, November 10, 2004

Briggs, C. J., Host-pathogen population dynamics, of chytridiomycosis and mountain yellow-legged frogs in the Sierra Nevada Mountains, California, Integrative Research Challenges in Environmental Biology grant meeting, Tempe, AZ, November 11, 2004

Briggs, C.J., Host-pathogen population dynamics, of chytridiomycosis and mountain yellow-legged frogs in the Sierra Nevada Mountains, California, California/Nevada Declining Amphibian Population Task Force Meeting, Berkeley, CA, January 14, 2005

Briggs, C. J., Investigating the population-level effects of chytridiomycosis, a fungal disease of amphibians, Invited Seminar, San Francisco State University, Biology Department, San Francisco, CA, May 3, 2005

Briggs, C. J., Investigating persistence of a chytrid fungal disease of amphibians, Invited Seminar, Ecology and Evolution of Infectious Diseases Meeting, Fort Collins, CO, May 19, 2005

Briggs, C. J., "Investigating the population-level effects of chytridiomycosis in mountain yellow-legged frogs", Invited Symposium Presentation
International Union of Microbiological Societies,
San Francisco, CA, July 25, 2005

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