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Sick and Stressed in the City? Disease Susceptibility of Australia's Frog Species in the Anthropocene

Brittany Mitchell

For animals, disease transmission is usually higher in urbanised areas than non-urbanised areas due to animals living in closer proximity to one another, increases in novel species interactions because of changed community assemblages, and greater toxin and stress exposure leading to immune suppression. As such, wildlife diseases are emerging more frequently in the Anthropocene and are a massive threat to the persistence of biodiversity.

One taxon particularly affected by wildlife disease are amphibians. Globally, forty-one percent of all amphibian species are threatened, and hundreds of species are already thought to be extinct. A major driver of amphibian declines is chytridiomycosis, a disease caused by the amphibian chytrid fungus. The disease infects the skin of

Top: Brittany Mitchell is a PhD Candidate at the Centre for Ecosystem Science at the University of New South Wales and the Australian Museum Research Institute.



Like this green tree frog (*Litoria caerulea*), frogs have been found dead en masse across eastern Australia. The exact cause is still unknown. Image: Suzanne Mcgovern, The Conversation.

amphibians and disrupts electrolyte balance, resulting in cardiac arrest and eventually death. A crucial component of a frogs innate immune system is contained within its glands within its skin. These glands produce a range of bioactive peptides, including antimicrobial peptides, which are documented to be an effective mechanism against a range of pathogens. Various studies have demonstrated the importance of antimicrobial peptides against amphibian chytrid fungus in addition to their traditional immune system.

Stressful life events, such as limited access to food resources and habitat disturbance, can increase stress hormones in frogs. When stress hormones are produced, they can suppress both innate and adaptive immunity, potentially making frogs more susceptible to disease. Despite the link between anthropogenic activity and increased stress hormone production, limited research exists examining the effects of anthropogenic pressures, such as noise and light pollution, on frogs' immune function. Furthermore, no research currently exists for Australian frog species. As a result, we have a limited understanding of how frogs and other animals in an ecosystem may be impacted in a world that is becoming increasingly more urbanised. Thus, a component of my research aims to determine the effect stressful anthropogenic activity has on the production of amphibian chytrid fungus and subsequent immune function in Australia's frog species. More specifically, it is hypothesised that frogs in more urbanised areas will:

- 1. Have higher levels of the stress hormone corticosterone,
- 2. Have impaired immune function, and
- 3. Have different amphibian chytrid fungus profiles than frogs from natural areas, putting them at greater risk of infection and subsequent mortality.

Frogs across an urban gradient, potentially the Peron's tree frog (*Litoria peronii*), will have a small blood sample collected, in addition to two minimally invasive assays. Blood samples will measure the corticosterone levels as an indicator of a stress response in urbanised areas; and secondly, count the immune cells present. Next, the frogs will be injected with an immune stimulant in the foot, and swelling responses will be measured to gauge immune strength. Lastly, a skin sample will assess the frogs' amphibian chytrid fungus profile. The profiles between frogs living in urban and non-urban sites will be compared via a specialised form of mass spectrometry (MALDI-TOF).

This study will be the first in Australia to address the interaction between disease and urbanisation in frogs. It will allow us to focus our conservation efforts on mitigating threats and developing crucial methods in Australia, where frogs need it most. This study is especially relevant to the mass mortality events we are currently seeing across the eastern coast of Australia. It is highly applicable for threatened frog species currently persisting in highly modified environments such as the green and golden bell frog (Litoria aurea) and growling grass frog (Litoria raniformis). With this founding understanding, we can begin to answer other questions like whether the stress of urbanisation leads to reduced immunity and antimicrobial peptide production? Which species are likely to be most vulnerable in the future? What modifications can be made to the urban habitat to minimise impacts on immunity and better conserve our frog species?

Funds provided by the Australian Wildlife Society will be used to analyse antimicrobial peptides and allocated towards associated fieldwork costs.



The anticipated study species is a Peron's tree frog [Litoria peronii]. Image: Doctor Jodi Rowley.