

Chytrid fungus and *Ranavirus* in Long Island Frog Populations



Possible leads on the disappearance of the Southern Leopard Frog from Long Island.



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Abstract:

Amphibian populations worldwide have been devastated by Chytridiomycosis, a disease caused by Chytrid fungus (*Batrachytrium dendrobatidis*). Having likely originated in Africa, it has been found on nearly every continent and can be spread by a number of hosts. Many amphibian species do not have any tolerance to the fungus and populations can quickly disappear following its introduction to the area. However, some can survive the disease and will act as carriers to spread it. Iridovirus is also a threat to local populations and was found at the Brookhaven National Lab last year. As such, this project was conducted to investigate the potential link that these diseases may have with a recent island wide decline and possible extirpation of the southern leopard frog (*Rana sphenocephala*). In their absence, several common and related potential vector frog species were sampled from various Long Island wetlands. Frogs were swabbed and the swabs were tested for Chytrid and Iridovirus. PCR and gel electrophoresis will be used to confirm the presence or absence of the diseases. The presence of chytrid or iridovirus in local populations would serve as a possible lead in the decline of Southern Leopard Frogs on Long Island. This was a data collection study and will not include the final results.

Introduction:

Chytrid:

Chytridiomycosis, a disease caused by the fungus *Batrachochytrium dendrobatidis* has been correlated with the decline and extinction of many amphibian species worldwide. Chytrid fungus was brought to the forefront of amphibian decline research following the extinction of the Golden Toad and Harlequin Frog in Monteverde, Costa Rica (Pounds, 1994). It has been found in North and Central America, Australia and Europe (Ouellet, 2003; Pounds, 1994; Obendorf, 2005; Stuart, 2004). While other elements such as climate change seem to factor into the disappearance of species, Chytrid fungus has been indicated as the main cause in many cases.

The fungus is found in the keratinized skin of amphibians (Longcore, 1999). In adults this is contained to the stomach, legs and toe pads of the animal. In larvae the mouth parts are usually the only tissue affected. The fungus is not normally lethal to larval stages; however in metamorphosing and adult stages, it can be fatal (Blaustein, 2004). The exact mechanism by which *B. dendrobatidis* kills is still largely unknown. The only consistent symptoms of the fungus are excessive skin cell loss and lesions in animals with heavy infections. Although *B. dendrobatidis* is lethal in some species, other populations survive with the fungus and remain relatively unharmed by it. These species may act as carriers to spread the disease to other areas and other populations. North American bullfrogs have been shown to survive with active infections (Garner, 2006). Bullfrogs and other surviving species can be tested to determine if the fungus is in an area.

Ranavirus:

Iridoviruses of the family *Iridoviridae* and genus *Ranavirus* are also a threat to amphibian populations. There are several different types of Iridoviruses which affect different groups of organisms. *Chloriridovirus*, and *Iridovirus* affect only invertebrates, while *Ranavirus* and *Lymphocystivirus* affect vertebrates (Williams, 2000). They are also responsible for the decline of some species such as the tiger salamander (*Ambystoma tigrinum diaboli*) (Bollinger, 1999). Iridovirus is also involved in the decline and disappearance of some species of turtle (De Voe, 2004; Chen, 1999). *Ranavirus* affects different species in different ways.

Both Chytrid Fungus and *Ranavirus* are possible contributors to the decline and likely extirpation of the Southern Leopard frog from Long Island. *Ranavirus* has been found in box turtles at Brookhaven National Laboratory; but it is unknown how far its range reaches. It is still uncertain whether or not Chytrid fungus has infiltrated Long Island frog populations, however it is known for its rapid spread and nearly unstoppable movement.

Discussion:

Chytrid fungus is a threat to Amphibian species and biodiversity worldwide. It has been found in many species on different continents (see Figure 1). While not all of these species are killed by the fungus, they can continue to carry it. It is unknown whether or not some species are susceptible to the fungus. Leopard Frogs are thought to be killed by the fungus and Bullfrogs and Green frogs are not. However, there is still no conclusive evidence at this time proving that Chytrid fungus has been introduced anywhere on Long Island.

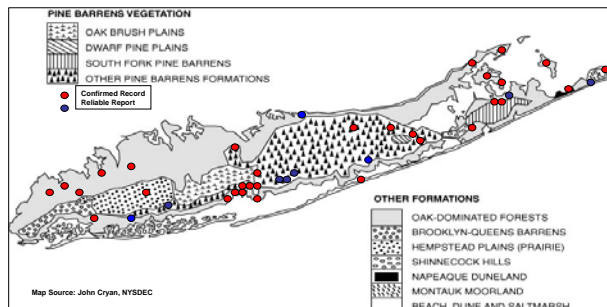
Ranavirus has been shown to be lethal to other reptile and amphibian species. It may play a role in the decline of Leopard Frogs. However there is still no conclusive evidence proving whether or not it is present in frog populations. Disease is a very likely contributor to the extirpation of Southern Leopard frogs from the region. Their sudden disappearance is indicative of a catastrophic event rather than a slow change. Other possibilities include interspecific competition and invasive species introduction. An ongoing project will look into these possibilities along with the effects of chemicals and pesticides in the environment and habitat loss.

Methods and Materials:

Initial data collection included surveying ponds with historic leopard frog sightings. These surveys were conducted by a minimum of two researchers at a time. At all the sites where there were green frogs, bullfrogs or pickle frogs, individuals were collected. These individuals were measured and swabbed. Swabs were collected for samples of both Chytrid fungus and *Ranavirus*. Chytrid swabs were collected from the ventral skin surfaces. The *Ranavirus* swabs were collected from the mouth and lips of the frogs. Following the collection, all swabs were kept in vials and transported to a freezer as soon as possible for long term storage.



Potential leopard frog sites on long island.



Results and Conclusion:

This study was an initial data collection for further analysis at a later time. Swabs were stored in a freezer and DNA extractions were performed. However, until PCR analysis can be done, the study will not be completed. While it is possible that chytrid fungus is present in long island populations, it will not be known until further tests are done. Since *Ranavirus* has been found on Long Island, it is likely that it is present in some populations. Its presence or absence has not been confirmed in Long Island frogs at this point.

Acknowledgements:

I would like to thank the Department of Energy and the Brookhaven National Laboratory Office of Science for this opportunity. I would also like to extend my gratitude to the Office of Educational Programs and the S.U.L.I. program. Finally, I would like to thank Chris Camacho and Valerie Titus for their help in the field and in the lab.