# **Research Proposal For**

## **Graduate Student Research and Professional Development Fund**

To the School of Graduate Studies Western Illinois University 1 University Circle Macomb, IL 61455

Submitted By XXXXXX Department of Biological Sciences Waggoner Hall 372 1 University Circle Macomb, IL 61455

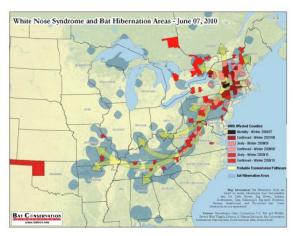
Date Submitted: September 15, 2009 Title of Research: **THE HUNT FOR WHITE NOSE SYNDROME IN ILLINOIS** Amount Requested: \$750 Advisor: Dr. Andrea Porras-Alfaro

XXXX XXXXXX Graduate Student

### Title: THE HUNT FOR WHITE NOSE SYNDROME IN ILLINOIS

#### Background

Bats are fundamental ecosystem components for insect suppression, pollination and seed



dispersal (Jones et al. 2009). White Nose syndrome (WNS) is a new disease that has been affecting bat populations in the United States and Canada. With the spread of WNS, bats which aid in these processes will face extinction e.g. *Myotis sodalis* (Indiana bat). To date, White Nose Syndrome (WNS) has killed at least 75% of surveyed bats within the

Figure 1. The progression of WNS in the US as June 2010

Northeastern United States. Figure 1 shows that WNS has slowly made its way across the Eastern United States from New York, where it was first documented in the winter of 2006 (Blehert et al. 2009), to Tennessee. Presently, WNS is on the doorstep of the Midwest, with likely cases already seen in Missouri (Figure 1).

The cause of White Nose Syndrome is presumed to be a newly described species of fungus, *Geomyces destructans* (Gargas et al. 2009). To date there is a lack of literature about extremophiles like *G. destructans*, a psychrophilic (cold-loving) fungus with optimal growing temperatures at 6 °C. By studying the psychrophilic fungi associated with bats we can monitor the presence of local psychrophilic species that can be used as a mechanism to control *G. destructans*.

#### **Objectives**

1) The primary goal of this research is to document the psychrophilic fungi associated with Illinois bats such as Indiana bats (*Myotis sodalis*) and little brown bats (*Myotis lucifugus*). My hypothesis (based on preliminary studies) is that bats carry a high diversity of psychrophilic fungi and these fungi are not pathogenic. These isolated fungi could also potentially be used as biocontrol agents for *G. destructans*.

2) The secondary goal of this study is to document the appearance of *G. destructans* within Illinois since the spread of WNS is nearing the state. We will use *Geomyces* specific primers and culturing techniques to accomplish the goal of identifying the psychrophilic fungi.

# Methodology

Forest Service personnel, the Illinois Department of Natural Resources (IDNR) and University of Florida (Dr. Robert McCleery) are supporting all fieldwork of this research project. I collected all the samples of this research project with the support and under the permits of our collaborators during the summer and fall of 2010. We currently have more than 200 samples from 10 caves in southern Illinois.

Using bat-trapping and sampling techniques outlined by the USGS National Wildlife Center to limit bat-to-bat transmittance of WNS, the bats will be swabbed and samples will be inoculated *in situ* on petri plates. Samples will be incubated at 6 °C to only isolate psychrophilic fungi. As Figure 2 shows this technique was tested last semester and it was very successful producing about 200 pure cultures of cold-loving fungi.

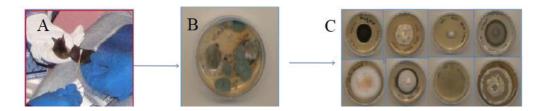


Figure 2. Bat swabbing technique to isolate psychrophilic fungi that are on bats. A: Swabbing of bat wing. B: Swabbed petri plate showing fungi cultured from bat wing. C: Plates of isolated fungus from plate swabbed from bat.

Direct isolations from wing biopsy will be conducted. In a preliminary study, last semester, this

technique was also evaluated (see Figure 3).

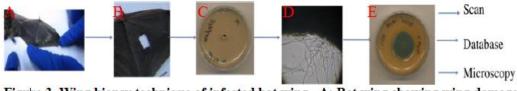


Figure 3. Wing biopsy technique of infected bat wing. A: Bat wing showing wing damage. B: Section of the damage wing cut. C: The cut damaged bat wing inoculated on a petri plate. D: Microscopic view of fungal hyphae growing from the bat wing tissue. E: The entire fungus grown from the bat wing.

DNA techniques such as PCR and sequencing will be used to identify psychrophilic fungi using fungal internal transcribed spacer (ITS) rRNA primers (Gargas et al. 2009).

## **Benefits of Research**

This research is of fundamental importance to monitor the presence of WNS in the state of Illinois. My research will also increase the knowledge of psychrophilic fungi associated with bats, being the first study of its kind in Illinois to my knowledge. The study will catalog the fungi bats had before the appearance of WNS and changes in community structure after the arrival of *G. destructans*.

## References

- Blehert DS, Hicks AC, Behr M, Meteyer CU, Berlowski-Zier BM, Buckles EL, Coleman JTH, Darling SR, Gargas A, Niver R, Okoniewski JC, Rudd RJ, Stone WB. 2009. *Bat white-nose syndrome: an emerging fungal pathogen?* Science 323 (5911): 227.
- Gargas, A., Trest, M.T., Christensen, M., Volk, T.J. & Blehert, D.S. 2009. *Geomyces destructans sp. nov.* associated with bat white-nose syndrome. Mycotaxon, 108, 147–154.
- Jones, G., Jacobs, D.S., Kunz, T.H., Willig, M.R. & Racey, P.A. (2009) Carpe noctem: the importance of bats as bioindicators. Endangered Species Research, 8, 93–115.

# Budget

My research will be conducted in Dr. Porras-Alfaro's lab at Western Illinois University. We are working in collaboration with Forest Service, IDNR and University of Florida. The supplies that will be acquired with this award will allow me to maintain the collections and start the identification of cultures using molecular techniques. My advisor has also applied for external funding that will partially cover the cost of this research (pending). This research project is also supported by partial funds provided by IDNR, Forest Service and University of Florida (Dr. Robert McCleery).

Supplies	Amoun t	Price	Catalog Number	Amount Requeste d from SoGS	Amount Requested from CAS
60x15mm Petri Dish W/Breathinglug Stackable	3	\$93	25384-090	\$93	\$185
ExoSAP-IT® For PCR Product Clean-Up	1	\$93.00	78200	\$93.00	0
PCR Master Mix, Promega	1	\$75.00	PAM7502	\$75.00	0
Malt Extract Agar 500 g	1	\$103.0 0	EM1.05398. 0500	\$103.00	0
Sequencing cost (\$4 per sequence)	1	\$137	University of New Mexico	\$137.00	\$64
Total				\$500	\$250