



Baseline Fungal Community Analysis and Investigation for *Pseudogymnoascus (=Geomyces) destructans* at Fort Hood, Coryell County, Texas

Project # 14-766

Background:

In order to further the mission of DoD's investment into bat management in the face of the threat from white-nose syndrome (WNS), the Legacy program has funded a novel research study to investigate early detection protocols through environmental sampling for *Pseudogymnoascus destructans* (*Pd*), the pathogenic fungus that causes the development of WNS in affected bat species. White-nose syndrome has caused the decline of millions of North American bats since *Pd* was discovered here in the winter of 2006. In March and May of 2015, the Arkansas Game and Fish Commission (AGFC) confirmed WNS-infected tri-colored bats in several caves in the Ozark Mountains of northern Arkansas, as well as *Pd* in mines of the Ouachita Mountains in the west-central part of the state (AGFC 2015a; AGFC 2015b). The implications of the new geographical records of *Pd* and WNS in Oklahoma and Arkansas are consequential to management interests for bats and caves in Texas because of the potential migratory connection to hibernacula in those adjacent states. This study was conducted on Fort Hood, a 214,968-acre active training facility in Coryell County, Texas, that hosts several hibernacula and maternity colonies for the migratory tri-colored bat (*Perimyotis subflavus*) and the cave myotis (*Myotis velifer*). Tri-colored bats are susceptible to WNS and have been documented to be affected by the disease in adjacent states; and may introduce *Pd* to Texas hibernacula. Cave myotis have not tested positive for the disease, but remain at risk of infection and may function as a vector of *Pd* to other vulnerable species due to its highly colonial, cave-dwelling nature. The DoD Legacy Resource Management (Legacy) Program funded this study in an effort to determine the status of *Pd* and to better understand the composition of the fungal communities in Fort Hood caves, and to develop protocols applicable to other DoD installations. The Legacy Program assists the DoD land managers in protecting and enhancing resources while supporting military readiness.

Objective:

The primary objective of this study was to determine the status of *Pd* and other fungi within known hibernacula and maternity colonies in Fort Hood caves. Monitoring for changes in the fungal community composition may help detect early warning signs of *Pd* invasion. *Pd* monitoring can inform management recommendations for at-risk species to help the DoD Natural Resources Conservation Compliance Program meet its own objective of maintaining unrestricted access to training and testing areas at Fort Hood. Specifically, our approach included 1) filling in regional information gaps regarding the presence of *Pd* and baseline characterization data on the fungal communities in bat-occupied caves; 2) developing a methodology for non-invasive detection of *Pd*; 3) testing the detection methodology by implementing substrate sampling at select Fort Hood caves; and 4) developing a baseline status report detailing the methodology and results of the assessment.

Summary of Approach:

In 2015, we collected 38 samples of substrate from four Fort Hood caves and 30 environmental swabs from one of those caves or redundant verification by an independent laboratory. Permanent sampling stations were established within each cave based on a variety of factors including selecting areas with a suitable amount of media for sampling; an adequate distribution of samples based on cave size, formation, and substrates; and specific areas of interest such as directly below or on known roosts, areas of differential temperature gradients, within flowpaths of water, or locations of visible fungal growth. DNA was isolated from the substrate samples and the isolated DNA was used for PCR amplification of the 18S rRNA IGS region using two different sets of primers described in Muller et al. (2013) and synthesized by Integrated DNA Technologies at the Veterinary Diagnostic Laboratory at the University of Illinois at Urbana-Champaign. The environmental swab samples were analyzed by the U.S. Geological Service National Wildlife Health Center Diagnostic Microbiology Laboratory in Madison, Wisconsin.



Benefit:

The development and implementation of this study is broadly applicable across other DoD installations with bats inhabiting cave roosts, especially those harboring protected species, and will maintain mission readiness in the event of positive detection by demonstrating proactive management that will facilitate communication and planning by maintaining active monitoring efforts. The detection of *Pd* before bats exhibit symptoms of WNS will allow resource managers to implement robust monitoring programs and prevention measures, thus advancing the DoD's goals described in the national plan to assist research, surveillance, disease management, diagnostic testing, and funding for WNS programs within individual DoD districts. In addition, we hope to contribute to the understanding of this emerging disease and fill regional information gaps on fungal communities in caves.

Accomplishments:

This phase of Legacy funding has enabled the first step in understanding cave fungal community composition on Fort Hood before invasion by *Pd*. Although substrate samples from all four caves and environmental swab samples were negative for *Pd* detection, the substrate sample suite did lead to the identification of several fungal taxa in these caves, including several that intriguing for a variety of reasons. Establishing permanent sampling stations within the caves and establishing laboratory relationships and methods were key accomplishments during this first year; these accomplishments will facilitate longer term monitoring of fungal communities and a baseline understanding of the current status of *Pd* and threat of WNS at Fort Hood.

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