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Is Out of Sight Out of Mind? The Use of Environmental DNA as a Tool for the Discovery and Monitoring of Rare Groundwater Organisms

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New faculty member participating in the RCEU program for the first time.

Project Summary:

Is Out of Sight, Out of Mind? The Use of Environmental DNA as a Tool for the Discovery and Monitoring of Rare Groundwater Organisms

Biodiversity is being lost at an alarming rate because of several factors associated with ever increasing human population growth and economic activities. Severely hampering our abilities to effectively manage and conserve biodiversity during the current extinction crisis is a fundamental lack of knowledge on distributions and abundances of species, particularly those that are small and that live in habitats extremely difficult to survey and study. One such group are stygobionts—obligate subterranean species that inhabit groundwater aquifers. Despite the importance of groundwater aquifers to humans, our knowledge of groundwater biodiversity is extremely limited, as subterranean habitats are particularly challenging to access and study. In the eastern U.S., over 300 obligate subterranean species have been described. Most have highly restricted distributions, and, consequently, are of conservation concern and at an elevated risk of extinction. The analysis of environmental DNA (eDNA) from water samples offers an exciting and potentially effective way to detect and monitor groundwater biodiversity that might otherwise be difficult or impossible to survey using traditional approaches. However, the efficacy of eDNA as a tool for groundwater biodiversity monitoring has not been thoroughly assessed. We propose to develop and examine the effectiveness of eDNA assays in groundwater habitats of the eastern United States. Specifically, we will (1) develop a reference DNA sequence database that leverages previously collected specimens in addition to sequence data available in existing databases; (2) develop and test species-specific eDNA assays using quantitative PCR (qPCR); and (3) compare the performance of eDNA versus traditional survey methods. This project will provide a foundation for future studies to gather vital insights into the distributions of rare and threatened groundwater organisms, including cavefishes, salamanders, snails, amphipods, isopods, and crayfishes, and inform conservation and management decisions for these species.

Student Prerequisites:

The RCEU student should have completed the Principles of Biology and Organismal Biology sequence (BYS 119 and 120), Genetics and Evolution (BYS 219), and the Chemistry sequence (CH 121/125 and 123/126).

Student Duties:

The RCEU student will be responsible for assisting in all phases of the project, including (1) field collection of eDNA water samples from caves, springs, and wells in northern Alabama, northwestern Georgia, and central and eastern Tennessee; (2) generation of a reference sequence database using traditional PCR and Sanger sequencing from previously collected specimens; (3) development of species-specific assays involving primer and probe *in silico* design and testing and *in vivo* testing; and (4) extraction and screening of eDNA samples using qPCR. The RCEU student will learn how to extract DNA from tissue and environmental samples, setup and run traditional PCR and qPCR reactions, use gel electrophoresis to visual DNA extraction and PCR results, and other molecular laboratory techniques. The student will learn how to pipette, use sterile technique, and other essential molecular laboratory skills that are highly marketable. The student also will learn how to use software to design and test PCR primer and probe assays and align and analyze sequencing data. The student will have the opportunity to experience and learn caving techniques and how to collect environmental samples. The student will participate in data analyses and report/manuscript preparation. A manuscript(s) will be submitted to a refereed journal based on the results of this project, with the RCEU student as first author or coauthor.

Mentor Supervision and Interaction:

The RCEU student will be taught the field and laboratory techniques directly by Dr. Niemiller, who will work with the student daily as they learn the procedures. The student on occasion will be assigned scientific papers to read regarding eDNA techniques and protocols and will review these papers each week with Dr. Niemiller. The RCEU student will meet with Dr. Niemiller daily at his office (SST 302M) and/or laboratory (SST 355) while learning techniques then transitioning to a less frequent meeting schedule as the project progresses but no less than at least once per week. The student is expected to attend bi-weekly lab meetings. The student will produce a report and presentation of the results of his/her project at the end of the summer that will be evaluated by Dr. Niemiller.