Environment and Natural Resources Trust Fund 2015 Request for Proposals (RFP)

Project Title:	ENRTF ID: 021-A				
Conservation Genetics of Minnesotas Rare Lizards and Snakes					
Category: A. Foundational Natural Resource Data and Information					
Total Project Budget: \$ _224,341					
Proposed Project Time Period for the Funding Requested: <u>3 years</u> , J	luly 2015 - March 2018				
Summary:					
Populations of Minnesota lizard and snake SGCN species will be field sampled and genetically characterized to analyze genetic distinctiveness, diversity, and population connectivity to provide foundational data for conservation planning.					
Name: Matthew Heinicke					
Sponsoring Organization: University of Michigan (UM-Dearborn)					
Address: 3003 State St S					
Ann Arbor MI 48109					
Telephone Number: (734) 764-8566					
Email heinicke@umich.edu					
Web Address					
Location					
Region: Statewide					
County Name: Statewide					

City / Township:

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Alternate Text for Visual:

Map of Minnesota, showing county ranges of SGCN species and key regions targeted for sampling

Funding Priorities Multiple Benefits Outcomes Knowledge Base
Extent of Impact Innovation Scientific/Tech Basis Urgency
Capacity Readiness Leverage TOTAL



PROJECT TITLE: Conservation Genetics of Minnesota's Rare Lizards and Snakes

I. PROJECT STATEMENT

A total of 20 native lizard and snake species occur in Minnesota. Of these, 65% (11 of 17 snakes; 2 of 3 lizards) are listed as Species in Greatest Conservation Need (SGCN). This project aims to characterize the population genetics of these species to serve as baseline information which will inform conservation decisions.

Minnesota's 13 SGCN species share in common a dependence upon prairie and/or rock outcrop habitats, and have ranges that broadly overlap where suitable habitats exist. All these species depend on deep rock fissures or burrows in sandy/gravelly soils to hibernate successfully. Suitable habitats are found patchily across Minnesota, in locations including the southeastern bluff lands (11 species), the Upper Minnesota River Valley (5 species), and Lake Agassiz Beach Ridges (3 species). Conversely, none of these species now occur in south-central Minnesota from Jackson County to Freeborn County, where suitable prairie formerly existed but has since been converted for intensive agricultural use. Remaining habitat patches are fragmented by agriculture as well as roads, suburban development, aggregate mining, and, more recently, frac sand mining. Such fragmentation results in loss of genetic diversity, and ultimately is a major cause of local population extinctions. Some species (e.g., plains hognose snakes) are also impacted by direct collection or killing.

This project will use targeted field sampling to obtain individuals for genetic sampling. Genetic data obtained using emerging DNA sequencing technology (RAD-seq) will be used to identify unique, geographically-restricted genetic lineages, measure gene flow (connectivity) between populations, and estimate demographic parameters including breeding population size. Four focal SGCN species will be sampled densely (>10 individuals per population) in the project: the Five-lined Skink, Prairie Racerunner, Bullsnake, and Plains Hognose Snake. Data for these species will be extrapolated to make inferences about habitat associate SGCN species. The other nine SGCN species will still be sampled as feasible (<5 individuals/population) to identify unique genetic lineages.

The data generated by this project will be analyzed to provide guidance in a variety of conservation contexts. As examples, appropriate populations for species to be released as part of a prairie restoration project can be identified, and populations found to be genetically isolated from neighboring populations can be targeted for introduction of new individuals or linkage via habitat corridor development to boost genetic diversity.

II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: Field sampling

Tissue samples will be obtained during statewide surveys to be completed under standardized, reproducible scientific protocols. Tissues will be non-lethally harvested from collected individuals and preserved in cooled 95% ethanol to maintain DNA integrity. Survey locations will focus on, but not be limited to, the SE Minnesota bluff lands (Houston, Fillmore, Winona, Wabasha, Goodhue, Dakota counties), the Anoka Sand Plain (Anoka, Hennepin, Sherburne counties), upper St. Croix bluffs (Chisago Co.), upper Minnesota River valley (Renville, Yellow Medicine counties), and Agassiz beach ridges (Clay, Norman, Polk, Marshall counties). Some samples have already been collected by project partners and additional tissue samples will be obtained from museum repositories as feasible. Fieldwork will begin immediately upon receipt of funds and continue through a 2nd year.

Outcome	Completion Date
1. Previously-collected tissue samples are obtained from partner individuals and museums	August 2015
2. Complete field sampling and tissue collection at predetermined sites throughout MN	September 2016
3. Forward all samples to genetics lab for analysis	September 2016

Activity 2: Genetic data generation and analysis

Budget: \$82,187

Budget: \$142,154



Environment and Natural Resources Trust Fund (ENRTF) 2015 Main Proposal

Project Title: Conservation Genetics of Minnesota's Rare Lizards and Snakes

DNA will be extracted from tissue samples and sequenced using RAD-seq, a newly-developed technique in which Dr. Gamble is an expert and Dr. Heinicke is proficient. This method generates large volumes of data suitable for use in fine-scale genetic analyses without requiring pre-existing knowledge of study organisms' genomes. These data will be used to quantify genetic variation within and among populations, measure gene flow, and estimate population sizes. Work will occur in the lab of Dr. Heinicke, which he has fully equipped for DNA amplification and sequence analysis using non-ENRTF funds. Genetics work will begin in September 2015. Analysis results will be used to assess local population extinction risks and the likely efficacy of potential conservation actions.

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Outcome	Completion Date			
1. Data Generation: DNA sequences are obtained for all tissue samples being analyzed	March 2017			
2. Data Analysis: DNA sequences are quality checked and assembled, and analyzed using	September 2017			
relevant software (e.g. Stacks, STRUCTURE, etc.) to test for genetic variation and gene flow.				
3. Data dissemination: Results of genetic analyses are made available for use by the DNR	March 2018			
through a report to the nongame wildlife program and reported to the broader community				
through presentations at national conferences and publication in peer-reviewed journals.				
4. Recommendations: potential effectiveness specific conservation measures (e.g.	March 2018			
translocation, habitat corridor linkage, etc.) are assessed for local populations determined				
to be endangered by severe genetic bottlenecks.				

III. PROJECT STRATEGY

A. Project Team/Partners

<u>MN DNR (contact person Erica Hoaglund, nongame wildlife specialist)</u>. *ENRTF funds*: N/A. *Role*: Field sampling assistance, advising on site selection. <u>Christopher Smith</u>, M.Sc., AWB[®] (Wildlife Biologist & President, Minnesota Herpetological Society). *ENRTF funds*: N/A. *Role*: Minnesota 501(3)(c) nonprofit organization in support of this project. Possible non-ENRTF funding through small grants program (\$1,500). <u>Tony Gamble</u>, Ph. D. (Department of Genetics, Cell Biology, and Development, U. of MN; MN state amphibian and reptile SGCN advisory committee). *ENRTF funds*: N/A. *Role*: Field assistance, advising on genetic analysis. John Moriarty, CWB[®] (wildlife biologist, Three Rivers Park District). *ENRTF funds*: N/A. Role: field sampling assistance, advising on site selection.

B. Project Impact and Long-Term Strategy

This project will be of immediate use to the DNR in assessing appropriate actions to take in conserving Minnesota's lizard and snake species, from formulating genetically-sound translocation and reintroduction policies to identifying key prairie regions to protect which will maintain the greatest amount of genetic diversity in prairie-dependant species. Understanding the genetics of these species will by proxy provide data on probable genetic patterns for other, less-visible species that share the same habitats, including many prairie insects. The results of the project will be disseminated through multiple peer-reviewed research articles and national conference presentaions. The project as a whole is envisioned as the start of a long-term investigation into the effects of landscape changes at both long (e.g., glaciations) and short (recent urbanization) time scales on the genetic diversity of Minnesota reptiles and amphibians. Funding to sustain this research will be sought from the US National Science Foundation as well as the project director's institutional grants programs. Dr. Heinicke has already securedsome institutional funding that he will dedicate to the project.

C. Timeline Requirements

The 2015 plus 2016 field seasons (approximately May-September) will be adequate for the necessary level of sampling to occur. Genetic work and analysis will begin immediately upon completion of the 2015 field season and resume for new samples after the 2016 field season. Six months of time are budgeted after the 2016 field season to complete data generation, with a further six months budgeted for final data analysis. Reporting and writing are budgeted to take six more months. The result is a planned project period of 33 months.

2015 Detailed Project Budget

Project Title: Population Genetics of Minnesota's Rare Lizards and Snakes

IV. TOTAL ENRTF REQUEST BUDGET: 3 years BUDGET ITEM AMOUNT Personnel: Matthew Heinicke, Project Manager. Salary 79%, fringe benefits (rate is 26% of salaries) 21%; two Ś 19.893 years (one month salary/benefits per year) with 2% merit pay increase in year 2. Graduate Student Research Assistant (GSRA; lead lab worker, individual TBD). Salary 54%, fringe \$ 76,114 benefits (rate is 26% plus tuition remission in lieu of salary [University policy]) 46%, 36% FTE for two years or 15 hrs/week (based on University of Michigan-Dearborn current rates for graduate students) with 2% salary increase in Year 2. Undergraduate Students (TBD). Salary 93%, fringe benefits (rate is 7.65% of salary) 7%; 1,000 hours \$ 26,094 per year at a rate of \$12/hour with 2% salary increase in Year 2 Nongame Wildlife Biologists (individuals TBD). Salary 77%, fringe benefits (rate is 30% of salary) \$ 42,120 23%; 36 person-weeks or 1,440 FTE hours (to be divided among approximately 6 individuals based on field availability) over two years. Contracts: Illumina HiSeq DNA sequencing service (10 lanes, ~\$2,000/lane). To be provided by U. of MN or U. \$ 20,000 of Michigan sequencing core, or outside vendor, based on best quoted price. Equipment/Tools/Supplies: Field equipment: collecting tools (nooses, snake sticks, etc.), bags, sample vials, coolers, ethanol (to \$ 1,000 preserve tissues), genetic sampling insturments (scissors, forceps, etc.). Genetics lab reagents; DNA extraction kits, sequencing adapters, sample purification kits, restriction Ś 9.000 enzymes, etc. (to extract DNA from tissue samples and prepare for sequencing) \$ 1,000 Genetics lab consumables: pipette tips, tubes, gloves, etc. (used while extracting DNA from tissue samples and preparing for sequencing). Acquisition (Fee Title or Permanent Easements): N/A ¢ Travel: In-state travel to field sampling sites. Includes mileage (~30,000 miles), lodging, meals. \$ 29.120 Additional Budget Items: N/A ć TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND \$ REQUEST = \$ 224.341

V. OTHER FUNDS (This entire section must be filled out. Do not delete rows. Indicate "N/A" if row is not applicable.)

SOURCE OF FUNDS	AN	NOUNT	Status
Other Non-State \$ To Be Applied To Project During Project Period			
Professional Development Funds (to be awarded to Dr. Heinicke by the University of Michigan-			Pending
Dearborn Natural Sciences Department). To be used for presentation of results at conferences.			
Indirect Costs waived by the University of Michigan-Dearborn; 55% for MTDC, federally-negotiated			Approved
F&A rate.			
Other State \$ To Be Applied To Project During Project Period: N/A	\$	-	
In-kind Services To Be Applied To Project During Project Period: N/A	\$	-	
Funding History:	\$	-	
Start-up funds awarded to Dr. Heinicke: Funds spent on lab equipment to be used during this	\$	12,000	Spent
project			
Remaining S From Current ENRTE Appropriation: N/A	Ś	-	

Minnesota's Lizard and Snake Species in Greatest Conservation Need



Note that SGCN species' ranges are concentrated along major river valleys, in remaining patches of open habitat (bluff prairie, sandy prairie, beach ridge, rock outcrops, etc.).

Focal SGCN snakes and lizards



Snake



Five-lined Skink

*Photo credits: Wikimedia Commons (Bullsnake, Hognose Snake, Racerunner); Heather Heinz (Skink)

Racerunner

Project Manager Qualifications and Organization Description

Project Manager: Matthew P. Heinicke, Ph. D.

Matthew Heinicke is a Minnesota-native herpetologist who is Assistant Professor of Biology in the Department of Natural Sciences at the University of Michigan-Dearborn. At Michigan-Dearborn Dr. Heinicke teaches Introductory Organismal and Environmental Biology as well as upper-level Genetics. His research combines field and laboratory components and focuses on how geography and time interact to cause genetic divergence and diversification in amphibians and reptiles. Dr. Heinicke has expertise in both field herpetology and the laboratory and computational methods of genetics, and is intimately familiar with Minnesota reptiles, as are all project partners. In his research lab, Dr. Heinicke oversees ongoing reptile and amphibian genetics projects involving numerous undergraduate researchers (10 currently).

Education

University of Minnesota, B. S. in Biology (summa cum laude), 2003 Pennsylvania State University, Ph. D. in Biology, 2009

Appointments

Assistant Professor of Biology, University of Michigan-Dearborn (2012-present) Postdoctoral Researcher/Visiting Assistant Professor, Biology Dept., Villanova University (2010-2012)

Field Research Experience (since 2002)

United States (MN, PA), Dominican Republic, South Africa, Namibia, Botswana, Zimbabwe

Most Recent Peer-Reviewed Research Publication (out of >19 since 2007)

Heinicke, M.P., Daza, J.D., Greenbaum, E., Jackman, T.R., Bauer, A.M. 2014. Phylogeny, taxonomy, and biogeography of a circum-Indian Ocean clade of leaf-toed geckos (Reptilia, Gekkota), with a description of two new genera. *Systematics and Biodiversity* 12: 23–42.

Organization: University of Michigan-Dearborn

The University of Michigan-Dearborn is one of three institutions in the University of Michigan system (along with Ann Arbor and Flint). Founded in 1959 with a gift from the Ford Motor Company, the Dearborn campus has in the past 55 years grown to be a comprehensive university. The Department of Natural Sciences, with over 40 full-time faculty, and home to 600 undergraduate Biology majors, is now among the largest departments in the Dearborn campus. Environmental Science is a particular research strength within the department, and the campus maintains one of the largest natural areas for research and teaching in Metro Detroit. In addition to Dr. Heinicke's work on reptile genetics, other faculty research areas include butterfly conservation, spider behavior, bird migration, microbial bioremediation, and toxic metal accumulation, among others.