

Conservation genetics of the Ozark pocket gopher

Project Summary

The Ozark pocket gopher (*Geomys bursarius ozarkensis*) is a range-restricted subspecies of the broadly distributed plains pocket gopher (*G. bursarius*) with documented populations only in Izard Co., Arkansas. The goal of this study is to assess the genetic diversity and divergence of the Ozark pocket gopher in order to guide conservation priorities and future management decisions. Specific objectives include:

1. To determine its genetic uniqueness (and taxonomic validity) of the Ozark pocket gopher
2. To determine levels of connectivity among regions of documented occurrence
3. To assess levels of genetic diversity in this small, isolated subspecies

Project Leader

Russell Pfau, PhD
Professor
Department of Biological Sciences
Tarleton State University
Stephenville TX 76402
pfau@tarleton.edu
254-968-9761

Project Partner

Matthew Connior
Bentonville, AR
mconnior@nwacc.edu

Project Budget

SWG amount requested:	\$28,858
Match:	\$10,100
Total:	\$38,958

Project Statement

Need

The Ozark pocket gopher (*Geomys bursarius ozarkensis*) is a range-restricted subspecies of the broadly distributed plains pocket gopher (*G. bursarius*) with documented populations only in Izard Co., Arkansas (Connior and Risch, 2009a; Connior et al., 2010). This isolated population of gophers was documented to be *G. bursarius* by Elrod et al. (1996) and described as a subspecies by Elrod et al. (2000) based on mitochondrial DNA sequences and cranial morphology. However, the latter study examined only one DNA sequence from each subspecies. Larger sample sizes are required to distinguish between genetic diversity within and among subspecies or populations--a critical component necessary to identify genetic uniqueness worthy of taxonomic recognition and conservation concern. Therefore, *there is a need to better understand the genetic uniqueness of this subspecies and to clarify its phylogenetic and phylogeographic affinities with the two nearest subspecies of the plains pocket gopher (G. b. missouriensis and G. b. illinoisensis).*

Specific locations of occurrence were documented by Connior and Risch (2009a), and I have recently documented additional occurrences by visually identifying mounds from satellite imagery in Google Maps (accessed December 2017; Fig. 1). Based on these occurrence data, it appears that the distribution of the Ozark pocket gopher is not continuous, but consists of multiple, potentially disjunct populations. Herein, I refer to these as populations simply because they appear discontinuous based on the available occurrence data--their status as true, disconnected populations is a hypothesis that I am proposing herein. These hypothesized populations are delimited in Figure 1, and consist of one large and three smaller populations. Three of the four hypothesized populations appear to be largely restricted to narrow, riparian corridors (B, C, and D). The larger one (A) has extensions along corridors, but also has broad areas not closely tied with riparian habitats. These four populations are not connected by riparian corridors, but the potential exists for gene flow between the hypothesized populations because apparently suitable soil habitat exists between documented occurrences (Fig. 1).

Using radiotelemetry and drift fences, Connior and Risch (2009a) did not document any instances of long-distance dispersal beyond home range sizes of $\sim 290 \text{ m}^2$. However, radiotelemetry in gophers can only assess dispersal over a portion of an individual's lifespan, and can rarely capture instances of infrequent, long-distance dispersal which may occur only among a few individuals within a population. Such dispersal events also are not likely to be detected by other methods. Infrequent, long-distance dispersal events could be sufficient to maintain genetic connectivity among populations and to allow gophers to recolonize areas from which they have been extirpated (due to natural or human causes). *There is a need to document the population genetic structure and level of genetic connectivity within the Ozark pocket gopher in order to better understand the impacts of population extirpation and potential for recolonization.*

Small populations are subject to genetic drift causing loss of genetic diversity which could potentially lead to inbreeding depression and loss of evolutionary potential. *A need exists to compare genetic diversity of the Ozark pocket gopher with the two nearest subspecies of the plains pocket gopher to provide a baseline for monitoring future changes in genetic diversity.*

Purpose and Objectives

The purpose of this study is to assess the genetic diversity and divergence of the Ozark pocket gopher in order to guide conservation priorities and future management decisions. Specific objectives include:

1. To determine the genetic uniqueness (and taxonomic validity) of the Ozark pocket gopher
2. To determine levels of connectivity among regions of documented occurrence
3. To assess levels of genetic diversity in this small, isolated subspecies

Location

Izard County, Arkansas (Ozark Highlands; Ozark-Ouachita Riparian and Pasture Land habitats)

Approach

Population genetic techniques have the potential to shed light on the population structure of the Ozark pocket gopher. Mitochondrial DNA sequencing and multilocus genetic techniques such as microsatellites and amplified fragment length polymorphisms have been used extensively over the past several decades to assess population structure. With the advent of high-throughput DNA sequencing methods, even more powerful approaches have been developed. Specifically, reduced-representation sequencing, led by RADseq, or restriction site-associated DNA sequencing (Miller et al., 2007) provide a means to obtain data from thousands of nuclear loci across the genome of non-model organisms. The large number of loci assayed increases the statistical power to detect population structure when it exists.

My approach will use two genetic methods: mitochondrial DNA sequencing to address Objective 2 and RADseq to address Objectives 1 and 3.

Gophers will be live- and kill-trapped (depending on population size and density), with Matthew Connior leading these efforts. For live-trapped gophers, a toe snip will be obtained from the hind foot. For kill-trapped gophers, tissues will be obtained by dissection and the skin, skull, and skeleton will be prepared as vouchers for deposition (along with tissues) into the Texas Tech Natural Science Research Laboratory collection. Live-trapping will be conducted using traps developed by Connior and Risch (2009b) and kill-trapping by standard gopher traps. For comparative purposes, tissue will be subsampled from the two nearest subspecies (*G. b. missouriensis* and *G. b. illinoensis*) from tissues of specimens already in collections (no additional field work necessary).

DNA extraction and polymerase chain reaction (to amplify mitochondrial DNA) will be conducted in my laboratory. DNA sequencing of mitochondrial DNA will be performed by the Genomics Core Lab at Texas A&M University—Corpus Christi. Resulting DNA sequences will be analyzed from a phylogenetic and phylogeographic context to determine how divergent the Ozark pocket gopher is from the nearest two subspecies.

Preliminary steps for RADseq will be performed in my laboratory. RADseq sequencing libraries will be prepared and sequenced at the Genomics and Bioinformatics Service Laboratory, Texas A&M AgriLife Research Genomics and Bioinformatics Service (College Station, TX). A PyRAD software pipeline will be used to prepare raw data for analysis. The resulting multi-locus genotypes will be used to identify population structure and measure population connectivity

using methods such as STRUCTURE (Pritchard et al., 2000), discriminant analysis of principal components (DAPC; Jombart et al., 2010), analysis of molecular variance (AMOVA), and F_{ST} .

Both mitochondrial and RADseq datasets will be used to quantify genetic diversity relative to the nearest two subspecies by measuring haplotype diversity, heterozygosity, and effective number of alleles.

Timeline:

- October 2018 - May 2019: Field work to obtain specimens
- June - December 2019: Lab work to obtain genetic data
- January - September 2020: Data analysis and drafting of report

Expected Results and Benefits

The Ozark pocket gopher is ranked as critically imperiled in Arkansas due to its restricted range and nuisance control by landowners. Previous genetic studies were very limited in scope, only being able to determine that the Izard County population was *G. bursarius* and not *G. breviceps* (as occurs elsewhere in the state). Because they did not quantify the genetic uniqueness of this population by taking into account diversity among individuals within the population versus between subspecies, the genetic uniqueness of this subspecies is not well documented. The proposed study will be the first to thoroughly quantify genetic uniqueness of the subspecies, genetic connectivity across its distribution, and genetic diversity within the subspecies. These three results will be important for guiding future conservation decisions.

Literature cited available upon request.

Budget

PI salary and benefits (one month)	\$13,623
Travel/room/board for PI	\$1,135
Contracted services for fieldwork (Matthew Connior)	\$6,000
Wages for student assistants	\$720
Shipping costs (specimens from Matthew Connior to Russell Pfau)	\$180
Mitochondrial DNA sequencing (80 individuals)	\$1,200
RADseq (80 individuals)	\$6,000
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Total SWG amount requested	\$28,858
Matching funds (35%; includes lab equipment)	\$10,100
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Total amount of project	\$38,958

Qualifications

The project leader has 23 years of experience in the field of population and evolutionary genetics, with a focus on mammals. Recently, two Master's students completed their theses on the population and evolutionary genetics of pocket gophers: *Geomys arenarius* and *Geomys breviceps*. These studies are being prepared for submission to peer-reviewed journals. A third project, currently underway, seeks to understand the impacts of population isolation of *Geomys bursarius* along the Colorado River of Texas.

Matthew Connior, project partner, studied the Ozark pocket gopher for his Master's thesis, has extensive hands-on experience with this subspecies, and has developed good relationships with landowners in the area. He has studied pocket gophers throughout Arkansas, Oklahoma, Louisiana, and Texas, resulting in numerous publications.

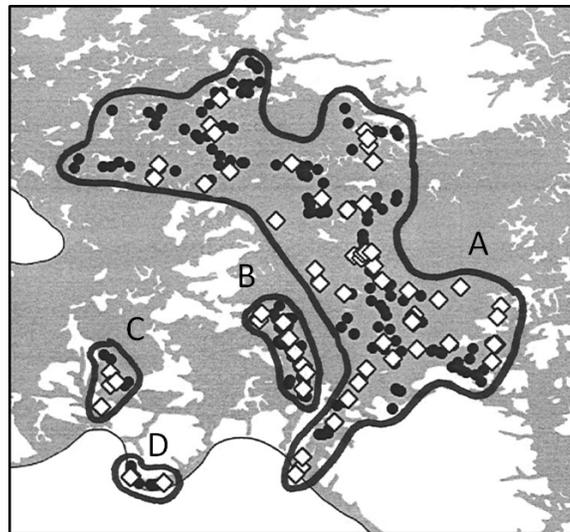


Figure 1. Occurrence locations and hypothesized populations of the Ozark pocket gopher in Izard County, AR. Open diamonds are occurrences documented by Connior and Risch (2009) and Connior et al. (2010). Closed circles are additional occurrences documented by visually identifying mounds from satellite imagery in Google Maps (accessed December 2017). Gray polygons are named soil types inhabited by gophers based on GIS mapping of mounds (closed circles only) and SSURGO soil classifications.