Influence of Thermal Habitat on Survival and Genetic Structure of Prairie Lizards

By
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Abstract

Extensive studies of the life history traits of lizards have revealed that temperature influences body size, reproduction and survival (ex. Smith and Ballinger 2001; Adolph and Porter 1993). In previous studies, Alyssa Bangs (a graduate student) and I found that environmental temperature did not influence morphology of prairie lizards in central Arkansas (Bangs 2016). That result is particularly shocking in light of a large body of work that documents influences of temperature on body size in *Sceloporus* (see review in Bangs 2016). Consequently, I initiated additional research to explain why lizards in Central Arkansas did not conform to expected norms. My focus in follow-up work is on survival and genetic isolation. As mentioned above, temperature also influences survival. In turn, survival influences body size because older lizards are larger. Equal survival in the two habitats would lead to similar age- and size-structured populations. Another possible explanation that could explain why lizards in Central Arkansas do not conform to expectations is that lizards move freely between the warmer rocky and cooler forested habitats and thus, do not spend their entire life in one thermal environment. Consequently, I initiated work to determine whether lizards in the rocky habitats were genetically distinct, which would only be possible if the populations were isolated. If lizards moved between habitats their genes would mix and the populations would be genetically similar.

In spring and summer of 2016 and 2017, several students and I captured and photographed 863 lizards. The scale pattern on their heads is distinct and the photographs allowed us to identify individuals (see Kellner et al. 2017). I compared survival among rocky (hot), forested

(cool) and edge (hot and cool) environments. Median proportion of recaptures were 0.15 for rocky, 0.12 for forest, and 0.10 for edge habitats. Those differences are not statistically different ($\chi^2 = 0.783$, df=2, p=0.676).

We evaluated the genetics of 701 lizards (the other 162 captures were recaptured individuals). We found that populations in most of our rocky habitats were genetically distinct from other populations. The pattern of isolation suggests that large bodies of water and roads represent barriers to movement.

Purpose/Objectives

Our objectives were to determine whether differences existed in survival and genetic structure of prairie lizards in relatively isolated rocky habitats, contiguous forested habitats and edge habitat which contains both rocky and forested habitat in proximity. We expected to find a difference in survival because activity patterns differed among habitats (Bangs 2016) which should lead to a difference in survival (reviewed by Adolph and Porter 1993). Lizard populations in environments in which lizards restrict their activity periods generally have higher survival (ex. Li et al. 2011; Adolph and Porter 1993). In our system, daily activity of lizards in the summer is shorter in the rocky habitats, in which temperatures exceed lethal levels by midday, than in the forested and edge sites in which cooler temperatures are always available.

A second objective was to compare genetic differences among lizard populations in different habitats. Populations that are genetically distinct are isolated, because movement of individuals would cause genetic mixing that would create more genetic homogeneity. Total isolation of lizards in rocky and forested habitats would mean that lizards in those habitats were consistently exposed to a single thermal environment. We have never observed lizards from our

study sites moving across large expanses of grasslands or across roads. Consequently, we expected to find differences in genetic structure between isolated rocky and forested habitats.

Introduction

One of us (C.K) has been studying lizards in rocky and forested habitats near Russellville since 2011. Surprisingly, those lizards have not exhibited expected size differences that are associated with thermally different habitats (Smith and Ballanger 2001, Adolph and Porter 1993). Consequently, our goal in this research was to gain some insight into why lizards near Russellville have not responded to temperature differences to the degree that other populations have (the magnitude of our temperature differences is similar to that found in other studies). Many other researchers have found differences in survival of lizards living in thermally different habitats (reviewed by Smith and Ballinger 2001). We expected to find similar differences in survival in our system.

An underlying assumption of the previous research conducted by C. Kellner is that lizards in rocky habitats do not shuttle between forested and rocky habitats because the rocky habitats are isolated by water, large expanses of open grassy habitats or roads. Consequently, we did not expect lizards to move between the two habitats, and in many hours of observation we never observed a lizard exit one of the rocky habitats. However, some degree of movement is possible. So, the information gained by evaluating the genetic structure of lizard populations will help to confirm the degree of isolation for lizards in rocky habitats.

In addition, comparing the genetic structure of populations of lizards in rocky and forested habitats has merit beyond confirming whether the populations are isolated. In particular, our proposed research will add to our knowledge concerning the interactions between habitat

fragmentation and genetic diversity (Primack 2014) which may predispose populations to extinction. Populations of lizards in our study sites have exhibited extreme population fluctuations during the past 5 years (C. Kellner pers. obs). Some populations have declined significantly over the period and probably have experienced a loss in genetic diversity (they have undergone genetic drift). Genetic drift and limited gene flow are important processes that alter genetic structure in isolated populations (Whiteley et al. 2010, Kikuchi and Isang 2002, Frankham 1997). Consequently, our research will address an important issue in conservation biology because loss of genetic diversity can lead to extinction (Primack 2014).

Methods

Study Sites: We used sites located along the shoreline of Lake Dardanelle, at Old Post Park, Bona Dea Nature Trail in Russellville, and Mount Nebo State Park. Lizards were captured in twenty-two sites in 2016 and 2017 (6 edge, 7 forested, and 9 rocky).

Methods: We used nooses to capture, to the degree possible, all the lizards at each site. A small noose of dental floss was tied to the end of a stick. After capture, a small dot of yellow paint was placed on the base of each lizard's tail. The mark is temporary but is sufficient to identify most individuals for several weeks (until lizards shed their skin). Paint marks allowed us to focus capture efforts on unmarked and presumably uncaptured individuals. To determine survival, captured lizards in each habitat were photographed for individual identification. The pattern of head scales on prairie lizards is unique for each individual (Kellner et al. 2017). We used the Interactive Individual Identification System software, I^3S (Van Tienhoven et al. 2007) to match images that we acquired in 2017 to those from 2016. Thus we determined which, and how many, individuals survived between 2016 and 2017.

We used microcapillary tubes to collect blood from the post-orbital sinus for each individual as described by MacLean et al. (1973). The collected blood was transferred to microcentrifuge

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tubes containing an acid citrate dextrose (ACD) solution, and those tubes were stored in a freezer until we extract the DNA.

From the blood samples, we extracted DNA and amplified 13 microsatellite markers, which we previously found to be suitable for our work. To estimate genetic diversity between rocky and forest populations of lizards, we calculated the average individual heterozygosity across all markers by using two computer programs: GenAlEx 6.5 and Arlequin. These data would also provide evidence for gene flow (Chistiakov et al. 2006). Consequently, we could determine if populations are completely isolated or if gene flow exists, which would help explain the lack of morphological differences among populations. Program GENEPOP (http://genepop.curtin.edu.au/) was used to group individuals that had similar genetic makeup. GENEPOP, uses an iterative Bayesian process to group individuals based on their genetic makeup. The program, determines the most parsimonious number of population groups. We assigned each lizard into the population that was most similar genetically and then examined the distribution of the populations over the landscape to determine which habitats contained isolated populations.

RESULTS & DISCUSSION

Survival

Only 49 of 356 lizards captured in 2016 were recaptured in 2017, or 0.13. In other words, we observed approximately 87% population turnover between years. Our field observations support this estimate; at the end of the field season, most adult lizards disappeared at about the time the hatchlings appeared.

Survival among habitat types did not differ significantly ($\chi2 = 0.783$, df=2, p=0.676) among rocky (0.15), forested (0.12) and edge (0.10) habitats. The distribution of estimates among the sites suggest that survival is extremely similar among habitats (Figure 1). This does

help explain why lizards in our habitats did not differ in size because most of the individuals in all of the populations are young of the year that have experienced only one growing season. However, we still expect a difference in size because lizards in rocky habitats are only active for a few hours each during the hottest part of the summer whereas lizards in forested habitats are active throughout the day (Bangs 2016). Consequently, lizards in the rocky habitats only forage for a few hours each day. Also, lack of a significant difference in survival is also surprising since differences in activity patters are usually associated with differences in survival.
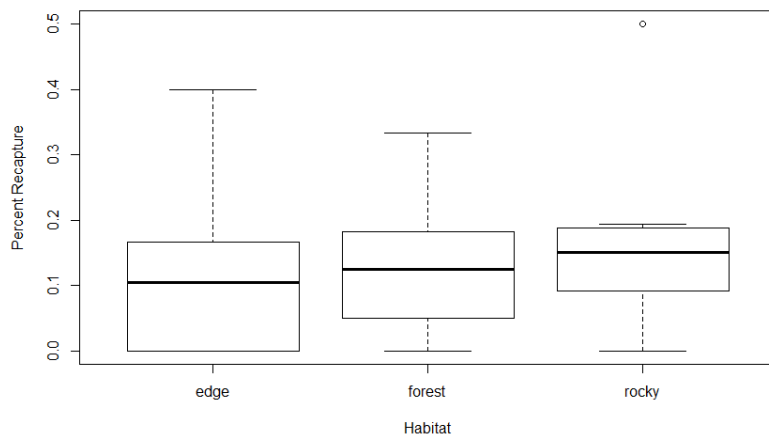

Figure 1. Boxplots of percent recaptures among habitats. The Outlier for rocky habitat is from a site in which we captured 2 individuals in 2016 one of which was captured in 2017 (thus a recapture rate of 0.5).

Genetic Structure

We extracted DNA from 793 lizards. We extracted DNA from all first time captures within a year, and also a few lizards that were recaptured within a season (the paint mark is lost after shedding and occasionally we recaptured a previously marked individual). The DNA was processed and each individual's microsatellites were sequenced so that we could discern the genetic structure within each population. Figure 2 represents our data divided into five somewhat distinct genetic units, where each

unit is represented by a different color. Homogeneous color bands represent more consistent and distinct populations. We assigned each individual lizard to one of those five populations, based on how similar their genetic makeup was to the group and then mapped out their locations to see whether the populations in our sites were genetically distinct.
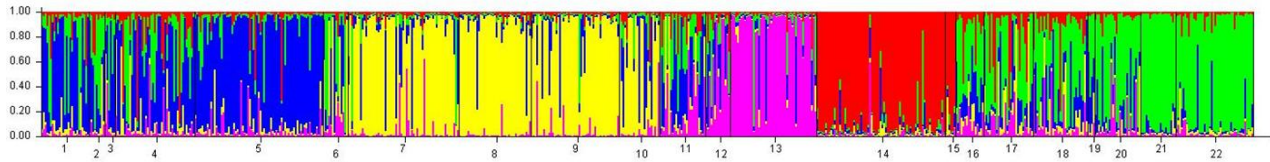

Figure 2. Genetic structure of the data compiled by GENEPOP for the lizards captured in 2016 and 2017. Each bar represents an individual lizard and bars that are composed of multiple colors indicate the proportion of times the program assigned an individual to a particular population group. The program works iteratively and relies on a Bayesian assignment algorithm. The numbers on the x-axis represent our 22 study sites and the y-axis represents a proportional scale that indicates the proportion of times a lizard is assigned to each of the 5 color populations.

Populations in a few of our sites were genetically distinct, which suggests that those populations are isolated. However, most of our sites were not genetically isolated. Figure 3 suggests that the lizard population structure along lakeshore is different from the other three populations on the photo. In contrast, populations on the north and south side of the Lock and Dam, are not distinct from nearby forested area (Figure 4). We found that lizards in forested areas were not genetically distinct. For, example, lizards captured among 5 sites at Mount Nebo State Park were not genetically distinct from one another (Figure 5).

We conclude that some movement is occurring between most of the rocky and forested habitats. We cannot ascertain the frequency of such movements without intensively following individuals throughout their lifetime. However, we suspect that movements between rocky and forested habitats are infrequent in our populations. For example, the north Lock and Dam population is not distinct from the forested area to the north. However, lizards moving between those habitats would have to traverse

several hundred meters over mown lawns. Further, we were usually successful in capturing and painting most lizards in our study sites. Although, most of the individuals disappeared during the spring and summer (see survival analysis above), if the disappearance was due to movements, unmarked and previously uncaptured individuals would likely have shown up in our study sites, and this was not observed. So, while results of our genetic analysis suggest that movements occur between some of the rocky and forested habitats, we do not think that the frequency of such movements would be sufficient, to eliminate the expected differences in morphology between lizards in cooler forested and warmer rocky habitats.

Figure 3. Aerial photo showing Lake Dardanelle. The bottom of the photo shows the area along Lakeshore Drive and also included Bona Dea Reserve. The top left of the photo shows the Boat Ramp area and the top right shows the Disc Golf area. Each colored dot represents a single lizard and the color indicates the population to which the lizard was assigned most frequently. The population colors come from Figure 2.
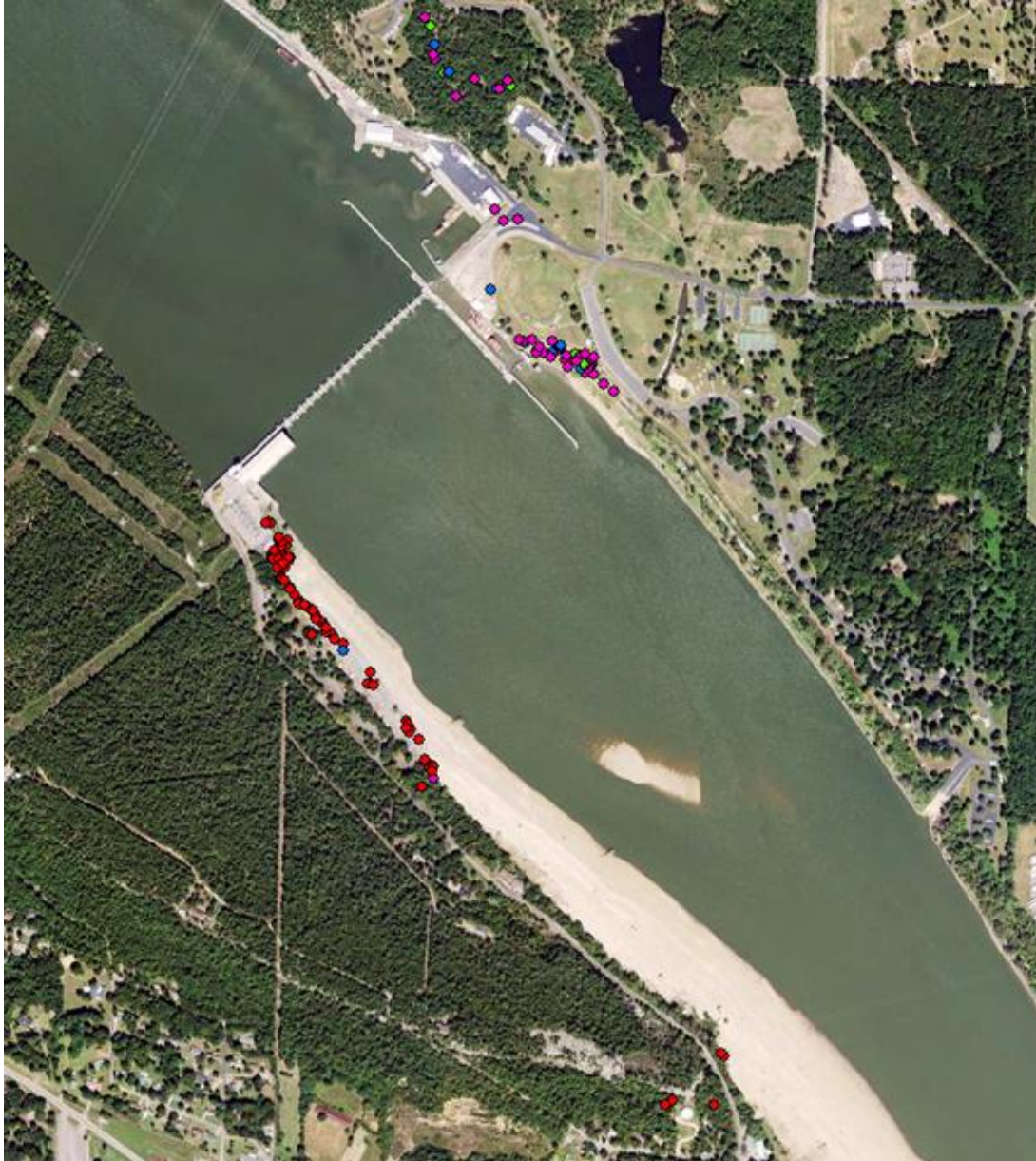
Figure 4. Aerial photo of the Arkansas River Lock and Dam near Pilot Rock. Each colored dot represents one lizard. Again, the colors and populations are derived from figure 2.

Figure 5. Mount Nebo State Park including three areas where we captured lizards. Each dot represents one lizard.

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