EASTERN

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25 September 2012

Diane Tecic
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Dear Ms. Tecic:

I am writing to thank you and the IDNR for helping offset the costs associated with conducting research on the herpetofauna of Illinois. I am pleased to include the enclosed Final Report for the portion of my study covered under the grant (#11-008W), "The community ecology associated with Hog-nosed Snakes." The report details the background of the project, and the information that was gathered during the grant period. I anticipate that at least two peer-reviewed publications will be produced based on the work completed under the aegis of this grant.

Based on my understanding of the herpetofaunal community at Thomson Sand Prairie (TSP) and IDNR/USFWS management objectives for this site, the population of *Heterdon nasicus* appears to be stable. Based on the number of individuals observed (including nesting females), I also think that the *Terrapene ornata* population is relatively stable at this site. Other species of reptiles appear to be doing well in this habitat, but were not observed as often on account of sampling protocols and detectability issues.

As you will read in my report, the scope of this project was restructured to provide a more detailed understanding of *H. nasicus* biology at TSP. Although I surveyed other sites within Illinois, I would not have been able to replicate the quality of scientific research about hog-nosed snakes by keeping other localities or species within the scope of this study.

If you have not already, I assume that you'll soon receive the Payment Request Form from EIU's Business Office. Please feel free to contact me should you have any questions concerning my report. Thanks again for your cooperation.

Cordially,

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Enclosure.

FINAL REPORT

Grant #11-008W

Grantee Name: Eastern Illinois University, Charleston, IL 61920; 217.581.2125 (R.W. Chesnut, Director of Research & Sponsored Programs)

Funded period: 1 November 2010 – 31 August 2012

The community ecology associated with Hog-nosed Snakes.

by

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15 September 2012

INTRODUCTION

This report details a 2-year study completed at Thomson Sand Prairie (TSA), Carroll County, Illinois, a compartment within the Upper Mississippi River National Wildlife & Fish Refuge. The original intent of the overall project was to compare ecological parameters of both Plains Hog-nosed Snakes (*Heterodon nasicus*) and Eastern Hog-nosed Snakes (*H. platirhinos*), with emphasis on the selection of habitat and diet. This report describes the research completed on *H. nasicus* at the TSP site based on the capture of 75 specimens, along with stable isotope analyses of blood and scale tissues from most of these snakes and multiple representatives of potential prey items sharing their habitat.

Objectives

This project focused on the biology of a snake species that occurs at relatively low density, *Heterodon nasicus*. The prey community for that species was also surveyed at TSP, providing a distributional occurrence of several amphibian and reptile species, as well as updated county records for these taxa. An additional objective of this project centered on student training and public education. Because high school, undergraduate and graduate students were involved in the completion of this project, they were exposed to various techniques in field biology. Through interactions with the general public at TSP, all personnel on this project were able impart positive understanding of amphibian and reptile species that occur in that region of Illinois.

Differences from original proposal

The scope of the original proposal included herpetofaunal surveys at three distinct sand prairie habitats in Illinois (Sand Road right-of-way [Madison Co.], Thomson Sand Prairie [Carroll Co.], and Nachusa Grasslands [Lee Co.]), as well as a detailed examination of the trophic ecology of a sister taxon, the Eastern Hog-nosed Snake, *Heterodon platirhinos*. For a variety of reasons, some of these aspects were not completed during the study period: 1. The number of *H. nasicus* specimens encountered at TSP was unexpectedly high, negating the need to sample other habitats for this species; 2. Preliminary surveys for *H. platirhinos* at all three sites revealed relatively low population densities, making equal sample sizes between species difficult to obtain (additional sites were also surveyed and, while present, sample sizes of *H. platirhinos* from a single locality were inadequate for comparisons with the *H. nasicus* at TSP); 3. In areas where *H. platirhinos* was encountered, the potential prey community included a few species for which I did not have state or federal T&E permits for specimen collection (an inadequate sampling of the prey community would have introduced bias into the research findings); and, 4. Given the relative success of encountering *H. nasicus* at TSP, it became logistically impractical to visit sand prairie habitats during the same sampling periods (which would be required for ecologically-relevant comparisons of diet and behavior). In summary, some of the objectives outlined in the original proposal (specifically interspecific and inter-site comparisons) were sacrificed in favor of the benefits available from pursuing a greater depth of understanding of the biology of the *H. nasicus* at TSP.

MATERIALS & METHODS

Study Site

Thomson Sand Prairie (TSP) is a 103.5 ha elongate unit of the Upper Mississippi River National Wildlife & Fish Refuge, located in Carroll County, Illinois (739790 E 4647614 N UTM Zone 15N) along the eastern border of the Mississippi River. Sand prairie vegetation consists mostly of bunch-grasses (e.g., *Dicanthelium* spp., *Panicum* spp., *Stypa* spp.) and herbaceous plants (e.g., *Tradescantia* spp., *Penstemon* spp., *Ionactis* spp.), but also includes the abundant cactus *Opuntia humifusa*, mosses, lichens, and a few woody shrubs and small trees. Of particular interest are the sand ‘blowouts’ present in the southwest portion of the prairie (Hart & Gleason 1907). These features are bowl-shaped, elliptical depressions of bare sand 260 - 1,400 m^2 in area. The blowouts are mostly oriented along a west-east axis (parallel to the prevailing wind), and aligned along the same longitude (Gleason 1910). Due to characteristics of their roots and seeds, a few species are able to colonize the bare sand in the blowout basin (e.g., *Acerates* spp., *Lespedeza* spp., *Tephrosia virginiana*), and these constitute an important microhabitat for *H. nasicus* (Thol 2008).

Field Methods

Specimen Collection – A 1.85 km U-shaped transect was established across the western portion of TSP, and surveyed for a total of 60 days between 19 September 2009 and 26 June 2011. Surveys occurred between 0830 and 1145 h, a period that corresponds with the daily maximum activity period for *H. nasicus* (Platt 1969). The time and location of all reptile and amphibian encounters on the transect, as well as standard measurements (sex, tail and snout-vent length [SVL; ±1 mm] and mass [±1 g]) were recorded. Scale and blood tissue was collected from 44 *H. nasicus*, and the guts of these snakes were palpated for food items.

To collect small vertebrates, a circular (200 m circumference) drift fence-pitfall trap array was established 225 m to the north of the transect route. The fence was constructed of 45-cm tall aluminum flashing and buried to a depth of ~15 cm, so that the resulting barrier was ~30 cm in height. Forty 4.5-L plastic jars were evenly-spaced along the inside of the fence such that their mouths (8.4-cm in diameter) were flush with the substrate surface. In a similar fashion, 12 evenly-spaced 18.9-L plastic buckets were buried along the outside of the fence. Prey species collected from the pitfall traps between 20 May and 24 June 2010 were weighed, euthanized, wrapped in aluminum foil and frozen. Opportunistic captures of anurans and racerunners throughout TSP during the study period were also weighed, euthanized, and stored in the same way as those captured in the pitfalls.

On 10 nights between 3 June and 24 June 2010, 44 small mammal traps were set in a line between the drift fence and the transect route (Wilson et al. 1996). The traps were baited with oats and peanut butter and any captured mammals were removed the following morning. During this same period, eggs from freshly-laid turtle nests (n = 20 *Chelydra serpentina* eggs and 8 *Chrysemys picta* eggs) were opportunistically collected at TSP. Eggs were weighed, wrapped in foil and frozen. Eggs of racerunners (n = 5) were obtained from the guts of *H. nasicus* on 24 - 26 June 2010. These samples were processed in a manner similar to that described for the specimens collected from the pitfall traps.

Prey taxa were grouped into six categories based on similarities between their carbon and nitrogen isotopic signatures: (a) toads (*Anaxyrus americanus*, n = 6); (b) racerunners and their eggs (*Aspidoscelis sexlineata*, n = 14); (c) painted turtle eggs (*Chrysemys picta*, n = 8); (d) snapping turtle eggs (*Chelydra serpentina*, n = 20); (e) leopard frogs (*Lithobates pipiens*, n = 7); and, (f) voles (*Microtus ochrogaster*, n = 7).

Behavioral Analyses – Snake behavior following its discovery was recorded using a Canon G10 digital camera. Specifically, the reaction of each snake to standardized harassment (tapping the snake twice per second on the head with a finger to simulate attack by a predator) was recorded. If a snake inverted its body, I ceased tapping to simulate the putative effect of the behavior on a startled or disinterested predator. If a snake subsequently ceased feigning death, I resumed tapping its head. Because *H. nasicus* are known to adjust the duration of their bodily inversions based on predator eye contact (Burghardt & Greene 1989), I kept my gaze on the screen of the camera throughout the duration of the filming (120 sec). From these recordings, the number of seconds to body inversion and the duration of inversion were measured (Gallup 1974). I compared these two dependent variables between sexes using an ANCOVA, using the covariates of time of capture and SVL; Kolbe 1999). I examined probability of death-feigning as a function of sex using a χ^2 test. I modeled latency to, and duration of, inversion as a function of SVL and time of capture using a multiple logistic regression.

Tissue Sampling, Preparation, and Analysis

Three tissue types from each *H. nasicus* were non-lethally sampled. Ventral scales from each snake were clipped and wrapped them in aluminum foil before freezing. I also drew blood from each snake's subcaudal vein and centrifuged the blood immediately to separate the red blood cells (RBCs) from the plasma. I drew off the plasma and deposited it in a tin capsule. I transferred the RBC pellet to a second tin capsule using forceps and placed the two closed capsules together into a microcentrifuge tube, which was subsequently frozen.

While frozen, whole prey items and tissue samples from snakes were freeze-dried for 24-48 h to remove water. Turtle eggs were punctured to facilitate complete drying. Once whole prey items were sufficiently brittle, each was placed in a steel canister with two ball bearings and agitated in a roll-grinding machine until the sample was homogenized. A small amount (~1.5 μg) of the resulting powder was placed into individual tin capsules. Snake blood and scale tissue samples were small enough to place directly into tin capsules without grinding. Isotope samples were processed by the University of Georgia Analytical Chemistry Laboratory using a Carlo Erba CHN Elemental Analyzer (Model NA1500) coupled to a Thermo Finnigan Delta V Isotope Ratio Mass Spectrometer. The output provided measures of total C, δC, percent 13C, total N, δN and percent 15N for each sample. I plotted δC versus δN for all samples to visually depict trophic relationships in the TSP vertebrate community. I compared differences among tissue types and between sexes. Correlation coefficients among tissue and isotope values within individuals were estimated, and these values were plotted against snake SVL and mass, as well as the day that the sample was obtained.

I used the Bayesian mixing model software MixSIR (Moore & Semmens 2008, Semmens et al. 2009) for MATLAB to analyze the relationship between the predator (*H. nasicus*) and its potential prey. A Bayesian approach to stable isotope mixing models is advantageous because it allows the inclusion of prior knowledge about diet (*e.g.*, gut contents) in the analysis and explicitly accounts for, and characterizes, any uncertainty in isotope values when estimating the contribution of sources to a mixture. The three tissue types from snakes of each age class (adults and young-of-year), and gender, were analyzed separately.

Synthesis of Dietary and Behavioral Data

Multiple regressions were used to estimate the explanatory value of each isotope value for each tissue type for on the SVL, mass, death-feigning latency and death-feigning duration of *H. nasicus*. For example, because toads were enriched in C13, snakes enriched in carbon were expected to be feeding more frequently on toads, with potential implications for their defensive behavior. The degree of individual specialization of *H. nasicus* in the TSP population was estimated using the ratio of the within-individual component (WIC) of niche width to the total niche width (TNW), expressed along a single continuous dimension (WIC/TNW; Ebenman & Nilsson 1982, Roughgarden 1972). I used only individual *H. nasicus* for which I had stable isotope values of carbon and nitrogen for the three tissue types (scale, RBC and plasma; n = 30). These values were adjusted using the average among-tissue variance within each individual. By analyzing sexes and age classes separately, as well as together, I isolated these components of inter-individual variation from other sources of variation (*e.g.*, ectomorph, Matthews & Mazumder 2004; behavioral syndrome, Sih et al. 2004).

RESULTS

The sex ratio of the 75 *H. nasicus* captured on TSP was slightly male-biased, with males representing 52% of captures. Young-of-year snakes comprised 19 (52.7%) of the 36 female and 29 (74.4%) of the 39 male snakes. Adult male SVL was smaller than in adult female snakes ($F_{1,26} = 1.45$, $p = 0.24$; Table 1). Male and female young-of-year snakes were of similar SVL ($F_{1,47} = 0.42$, $p = 0.52$). Regardless of age class, tail lengths were greater in males than in females ($F_{1,26} = 4.16$, $p = 0.05$ for adults; $F_{1,47} = 47.68$, $p < 0.0001$ for young-of-year snakes). Adult female mass tended to be greater than adult males ($F_{1,26} = 3.55$, $p = 0.07$) whereas the mass male and female young-of-year snakes was similar ($F_{1,47} = 0.28$, $p = 0.60$). The estimate for detection probability was 24.78 ± 0.05 %, meaning that *H. nasicus* could be expected to be found about once per hour when surveying the transect. “Deliverables” are included as 5 digital images in Appendix I. Records for species of herpetofauna observed during the completion of this study are listed in Appendices II and III.

Behavioral Analyses

From the 40 subjects encountered in May and June, 2010, I obtained 29 video recordings of *H. nasicus* behavior, of which 23 snakes feigned death within the first two minutes (an overall incidence of death-feigning of 79.3%). There was no relationship between snake gender and the probability of death-feigning ($\chi^2 = 0.10$, $p = 0.76$). Excluding subjects which never death feigned, the average latency to death-feigning was 23 sec, and the average duration of a death-feint was 39 sec. Mean latencies for males (33 sec) and females (16 sec) were similar ($F_{1,27} = 1.70$, $p = 0.21$; Fig. 1). The duration of inversion for female *H. nasicus* (49 sec) was similar to that for males (29 sec; $F_{1,21} = 1.77$, $p = 0.20$; Fig. 1). Including subjects which never death-feigned (*i.e.,* latency = 120 sec, duration = 0 sec) produced mean latencies to inversion for males and females that were different ($F_{1,27} = 4.11$, $p = 0.05$).

Larger snakes death-feigned for longer durations than small ones ($F_{1,21} = 21.82$, $p = 0.0002$ for mass; $F_{1,21} = 32.55$, $p < 0.0001$ for SVL), and a SVL-by-sex interaction ($F_{1,21} = 8.63$, $p = 0.009$, Fig. 2) indicated that this trend was driven by females. Large female snakes death-feigned for longer than small females ($R^2 = 0.82$), while male snakes did not follow this relationship as strongly ($R^2 = 0.51$). The time of capture did not influence the latency to inversion ($F_{1,21} = 1.60$, $p = 0.22$) or the duration of death-feigning ($F_{1,21} = 0.11$, $p = 0.75$), nor were there any interactions with time of capture and the morphometric variables ($F_{1,21} \leq 2.44$, $p \geq 0.15$). The model including time of capture and mass correctly predicted whether an individual subject would death-feign 77.3% of the time.

Diet and Stable Isotope Analyses

Gut contents obtained from 14 (11%) of *H. nasicus* captures included eggs of either *C. serpentina* or *C. picta*, and either juvenile or eggs of *A. sexlineata*. Isotopic signatures of the snakes varied among tissue types ($F_{2,64} \geq 9.59$, $p < 0.001$ for carbon, $p = 0.008$ for nitrogen; Fig. 3), as well as between sexes ($F_{1,64} \geq 12.30$, $p = 0.002$ for carbon; $p = 0.02$ for nitrogen). Females (mean carbon signature = -23.8 ± 0.04) were enriched in carbon relative to males (mean carbon signature = -24.0 ± 0.08). Both SVL and mass were correlated with plasma and scale carbon and nitrogen isotopic composition ($p < 0.001$ for both tissues and size measures), but not with RBC isotope signature. There was a sex-by-mass interaction for nitrogen for the same two tissue types ($p < 0.052$ for plasma, $p < 0.057$ for scale).

Hatchling *H. nasicus* isotope values reflected those of adult females (Fig. 4). As size increased, plasma isotopic content became enriched in carbon and depleted in nitrogen; this trend reversed between 285 and 330 mm snout-vent length. Above this range, plasma from larger snakes was more enriched in nitrogen and depleted in carbon. Nitrogen was about twice as variable as carbon for all three tissue types (Table 2).

Chelydra serpentina eggs were the prey type most enriched in nitrogen (Fig. 5), whereas voles and *A. sexlineata* were the most depleted in nitrogen. Voles were the prey type most depleted in carbon, while *Anaxyrus americanus* were the most enriched in carbon. Adult *Aspidoscelis sexlineata* had similar isotopic values to their eggs. Small *L. pipiens* showed some overlap with large voles; otherwise, all prey types occupied distinct regions within the trophic space. The mixing model predicted that *H. nasicus* fed on *C. serpentina* eggs with a posterior probability of 44.9-75.7%. This prediction was substantiated by gut content analyses of adult *H. nasicus* at TSP.

Although the guts of several juvenile *H. nasicus* contained *A. sexlineata* or their eggs, the mixing model did not predict that racerunners made up a substantial fraction of the diet of *H. nasicus* (posterior probability < 17.7%). Additionally, the model predicted that *Anaxyrus americanus* comprised 5.8-24.4% of the diet, although no *A. americanus* were recovered from guts of *H. nasicus* at TSP. The proportion of diet made up of *A. americanus*, *Aspidoscelis sexlineata*, and especially *C. serpentina* eggs, increased for RBCs over plasma and for scale tissue over RBCs. Concomitantly, the proportion of diet made up of voles decreased from 30.1-38.3% for plasma to < 8.1% for scale tissue. Leopard frogs and *C. picta* eggs were not predicted to comprise a substantial fraction of the diet (posterior probability < 3.6% in both cases). Adult *H. nasicus* of both sexes were predicted to have a specialized diet of *C. serpentina* eggs, whereas juvenile snakes were predicted to feed more broadly on voles, toads and racerunners, with a substantial maternal signature from turtle eggs.

Plasma nitrogen content was the best predictor of the duration of death-feigning in the regression model ($p = 0.04$, $R^2 = 0.95$). A quadratic term for plasma nitrogen was also a good predictor ($p = 0.04$). The nitrogen ratio of RBCs was a good predictor of snake body mass ($p = 0.03$, $R^2 = 0.79$). All other nitrogen terms and all carbon terms were not significant contributors to the model. The absolute values of niche width are relatively unimportant compared to the ratio of between-individual to within-individual resource use. Total niche width was similar for carbon and nitrogen. Female *H. nasicus* had more specialized diets than males (Table 3). Adult *H. nasicus* had more specialized diets than juveniles.

DISCUSSION

Natural History

Heterodon nasicus at TSP are morphologically within the range of known phenotypic variation for the species (Kolbe 1999, Platt 1969). The population appears to be stable, with many gravid females contributing to annual reproduction in 2010. *H. nasicus* at TSP appear to use sandy blowouts disproportionately to their availability, but whether this is due to higher snake abundance or higher detection probability in blowouts is unclear. For a snake, *H. nasicus* has a relatively high detection probability (Durso et al. 2011b). The ecosystem at TSP appears to be conducive to studies of *H. nasicus* due to their catchability and relatively high abundance (Kroll 1973, Platt 1969).

Behavioral Analyses

Studies of animals in the wild are critical to understanding the adaptive value of their behaviors in relation to the environment in which they evolved. Measures of behavior obtained from subjects in a natural setting are more reliable indicators of behavioral utility than those obtained in captive trials. Differences in latency to initiation and duration of death-feigning with sex and body size, and an interaction of sex and body size, which was driven mainly by female *H. nasicus*. Sexual dimorphism in behavior has rarely been documented in snakes, and has not been reported for *Heterodon*. Loss of death-feigning behavior in *Heterodon* in captivity has probably prevented researchers from detecting this effect.

Individuals of *H. nasicus* death-feigned for a relatively short time, compared to records for other members of the genus (*e.g*., Edgren 1955, Myers & Arata 1961). That male *H. nasicus* did not show as strong an ontogenetic change in behavior despite approximately equal sample sizes between the two sexes indicates that behavior of female hog-nosed snakes is more variable. Any influence of gravidity could not be isolated because all adult female *H. nasicus* in my study were gravid at the time of testing. Female *H. nasicus* typically reproduce in alternate years (Ernst & Ernst 2003); as such, in a given year I would expect that gravid females are most likely to be detected in visual encounter surveys, due to their increased frequency of movement and basking to provide an optimal thermal environment for embryo development (Gregory et al. 1987, Lillywhite 1987, Peterson et al. 1993). Future work should target non-reproductive adult females for comparison with gravid females.

Stable Isotope Analyses

Isotopic signatures provide information from all individuals captured instead of those having fed recently, thus improving the inferential power available to answer questions about dietary composition. Although scale tissue is replaced at a slower rate than blood, estimation of this turnover rate will require multiple years of isotope data. The apparent lack of influence of body size on RBC isotopic composition could be due to lower inter-individual variation in diet at the intermediate time scale.

Isotope analyses indicated that *H. nasicus* at TSP feed extensively on the eggs of *C. serpentina*. Turtle eggs are in high abundance in late spring and early summer, and *H. nasicus* might feed almost exclusively upon them during this time. Eggs represent low-risk sources of energy with low handling time and high temporal and spatial predictability.

Anaxyrus americanus probably represent only a small proportion of the diet of *H. nasicus* at TSP. That the *H. nasicus* in examined this study harbored no trematode parasites also indicates a low contribution of amphibians to their diet.

Plains Hog-nosed Snakes probably have a more generalized diet than either of their congeners. The diet of *H. nasicus* shifts as they age, and adults consume a wide variety of vertebrate prey, whereas small (<250 mm SVL) juveniles are restricted to a single prey type (*Aspidoscelis sexlineata* and their eggs). As these snakes grow, their dietary breadth increases, with a corresponding increase in growth rate until sexual maturity is attained (Platt 1969). Smaller snakes probably target different prey than large (>400 mm SVL) adults, but this is difficult to verify because smaller adults are rarely encountered. Large adults, especially females, probably feed almost exclusively on turtle eggs during the late spring and early summer. The mass-by-sex interaction was expected, because hatchling animals of both sexes carry the isotopic signature of their mothers, from whose diet all of the hatchling carbon and nitrogen is derived (Pilgrim 2007). For example, the presence of *A. sexlineata* in the diet of small *H. nasicus* is masked by the maternal isotope signature.

Human Impacts

This project was incorporated as part of the thesis research for a Master's of Sciences student in EIU's Department of Biological Sciences (EIU-BIO). In addition to that graduate student, five other students (two graduates and three undergraduates) from EIU-BIO assisted with the completion of this project. Portions of the fieldwork also received assistance or collaboration from members of the laboratory of Dr. Fred Janzen (Iowa State University) and participants in the Turtle Camp Research & Education in Ecology (TREE) program (mostly high school students). Regular interactions with the staff of the Upper Mississippi National Wildlife & Fish Refuge provided additional opportunities to educate on-site biologists about the study design and findings. Because the field work coincided with peak recreational use of the Thomson Causeway Recreational Area and visitors to TSP, individuals assisting with this research had multiple opportunities to inform the general public about the natural history of the TSP habitat, the vertebrate community that occurs at this site, as well as several techniques regularly employed in field ecology studies.

To date, the findings of this research have been incorporated into the following scientific presentations: (a) two papers at international conferences; (b) one poster at an international conference; (c) one poster at a regional conference; and, (d) one publication (Durso et al. 2011a). I anticipate that at least two more peer-reviewed articles will be generated from this research.

SYNTHESIS & RECOMMENDATIONS

Because toads were the most enriched in carbon of any vertebrate prey sampled, *H. nasicus* whose tissues were also more enriched in carbon were expected to death-feign more readily or for the longest durations, when compared to subjects with more depleted carbon ratios. This relationship was not supported by the data – female *H. nasicus*, which death-feigned for shorter durations, were depleted in carbon. Among adult snakes, however, females death-feigned for longer and initiated death-feigning sooner, which could be attributed to diet, reduced locomotory ability as a consequence of gravidity (Gregory & Gregory 2006), or both.

Limitations of the stable isotope method include the complications introduced by fractionation, ontogenetic and temporal variation in sources, and isotopic routing (Gannes et al. 1997, Pilgrim 2005). Mixing models are predictive, but limited in their ability to meet assumptions by the quality and quantity of laboratory data available, especially for fractionation coefficients. Other avenues for measuring diet could be pursued, such as amplifying and identifying prey DNA from predator feces or examining the parasite community of predators and prey so that the host-specific life cycles of certain trematode parasites might be used to track food chains (Pizzatto & Shine 2011).

Because populations of *H. nasicus* have decreased in size and number over much of its range (Phillips et al. 1999, Tennant 1985, Wright & Didiuk 1998), further study of this species is warranted. Conservation efforts can be aided by a better understanding of interactions of *Heterodon* with both their predators and prey. Hog-nosed snakes are good candidates for reintroduction programs, because of their lack of site fidelity to overwintering sites (Platt 1969, Plummer & Mills 2000). A thorough understanding of the biology of all *Heterodon* will help inform conservation decisions and elucidate the relationship between diet and behavior in toad-eating snakes.

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Table 1: Morphological characteristics (means ± one standard error) of 75 Plains Hog-nosed Snakes (*Heterodon nasicus*) from Thomson Sand Prairie (Carroll Co., Illinois) in 2010 and 2011.

Gender	Age class	n	Snout-vent length (mm)	Tail length (mm)	Mass (g)
Male	Young-of-year	29	178 ± 0.7	33 ± 0.2	6.70 ± 0.06
	Adult	10	451 ± 14.4	84 ± 2.8	111 ± 8
Female	Young-of-year	19	182 ± 0.9	25 ± 0.1	6.44 ± 0.09
	Adult	17	498 ± 6.6	68 ± 0.8	162 ± 5

Table 2: Mean (± one standard error) stable isotope ratios for blood plasma, red blood cell and scale carbon (δC^{13}) and nitrogen (δN^{15}) of 40 Plains Hog-nosed Snakes (*Heterodon nasicus*) from the Thomson Sand Prairie (Carroll Co., Illinois), as measured from samples collected in May and June, 2010.

		Carbon			Nitrogen		
		Plasma	Red blood cells	Scale	Plasma	Red blood cells	Scale
Male	Young-of-year	-24.315 ± .058	-23.512 ± .083	-23.147 ± 0.233	8.745 ± 0.276	10.989 ± 0.253	9.267 ± 0.457
	Adult	-23.658 ± 0.200	-23.431 ± 0.048	-22.822 ± 0.117	11.351 ± 0.284	11.592 ± 0.200	12.260 ± 0.196
Female	Young-of-year	-24.082 ± .085	-23.197 ± .067	-22.515 ± .152	8.804 ± 0.140	10.178 ± 0.153	9.496 ± 0.336
	Adult	-23.519 ± .108	-22.954 ± .134	-22.700 ± .145	11.099 ± 0.122	10.646 ± 0.155	11.182 ± 0.192

Table 3: Results of IndSpec estimation of degree of individual specialization for 30 Plains Hog-nosed Snakes (*Heterodon nasicus*) at Thomson Sand Prairie (Carroll Co., Illinois) in 2010.

Carbon

Model	Within-individual variation (WIC)	Between-individual variation (BIC)	Total niche width (TNW)	Relative degree of individual specialization (W/T)
All individuals	0.25	0.9511	1.201	0.2081
Males	0.2917	0.3422	0.6339	0.4602
Females	0.2374	1.219	1.456	0.163
Young-of-year	0.2732	0.5591	0.8323	0.3282
Adults	0.226	1.428	1.653	0.1367

Nitrogen

Model	Within-individual variation (WIC)	Between-individual variation (BIC)	Total niche width (TNW)	Relative degree of individual specialization (W/T)
All individuals	0.2698	0.8164	1.086	0.2484
Males	0.4188	1.084	1.503	0.2786
Females	0.1453	0.6435	0.7888	0.1842
Young-of-year	0.455	0.4897	0.9447	0.4816
Adults	0.06738	0.6676	0.735	0.09168

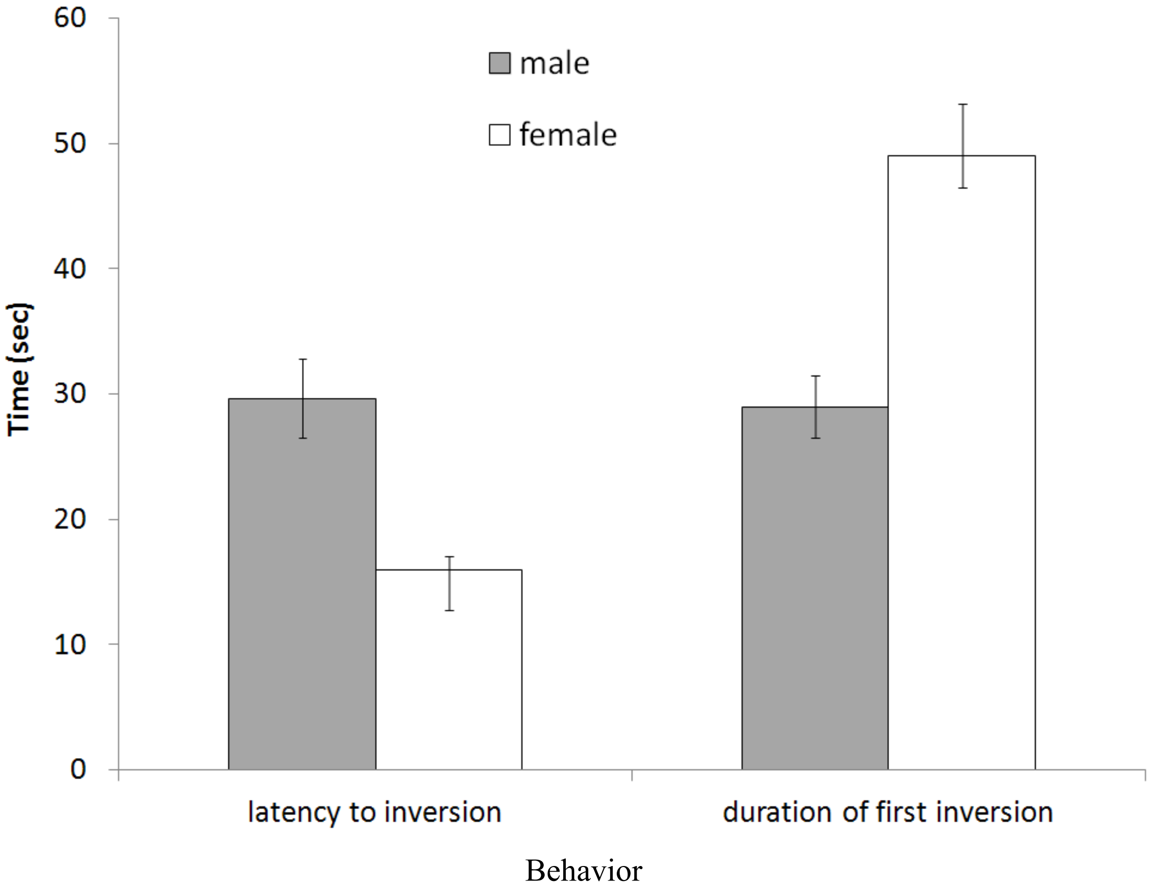

Figure 1: Differences in mean values for death-feigning behavior as a function of sex in Plains Hog-nosed Snakes (*Heterodon nasicus*) at Thomson Sand Prairie (Carroll Co., Illinois) in May and June, 2010. Bars represent ± 1 standard error.

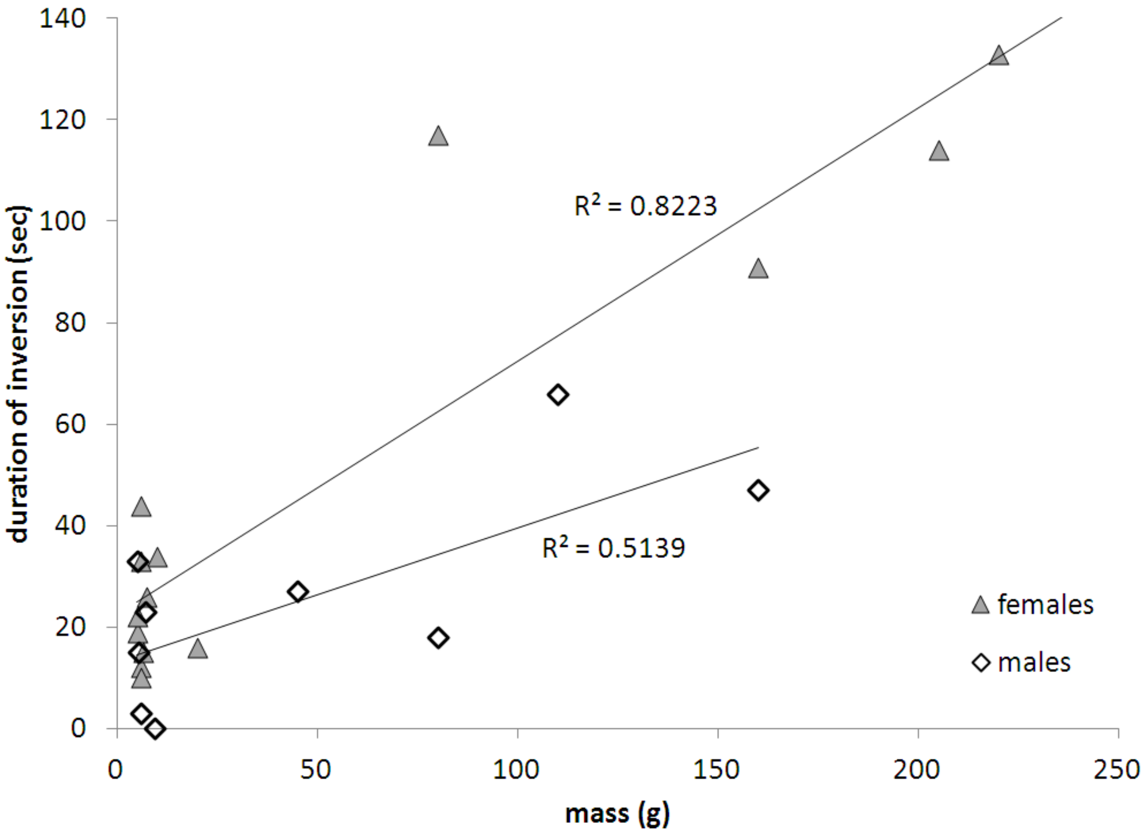

Figure 2: Relationship between intensity of death-feigning behavior and body mass for male (open diamonds) and female (shaded triangles) in Plains Hog-nosed Snakes (*Heterodon nasicus*) at Thomson Sand Prairie (Carroll Co., Illinois) in May and June, 2010.

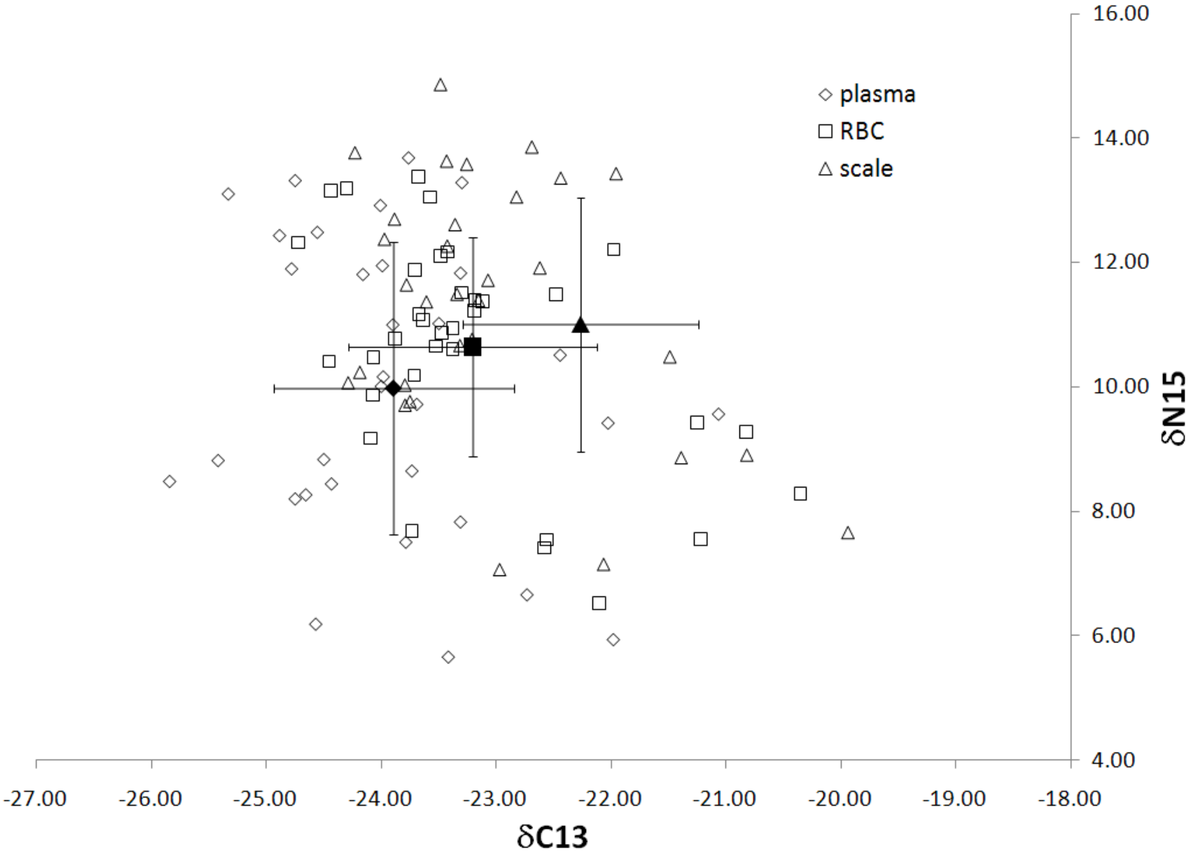

Figure 3: Mean stable isotope values for C^{13} and N^{15} in three tissue types of Plains Hog-nosed Snakes (*Heterodon nasicus*) at Thomson Sand Prairie (Carroll Co., Illinois) in 2010. Hollow symbols represent individual data points. Solid symbols represent means, bars represent ± 1 standard deviation. RBC = red blood cells.

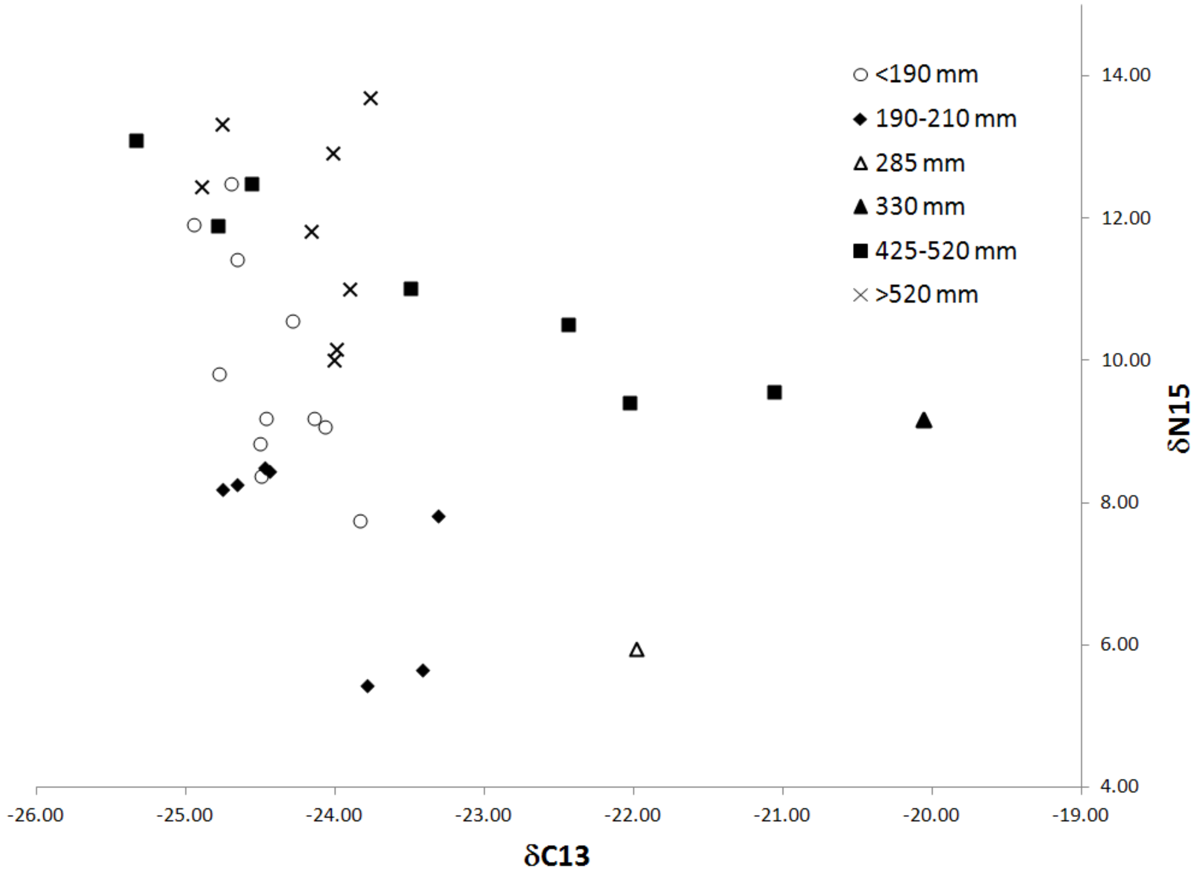

Figure 4: Stable isotope values for C^{13} and N^{15} for blood plasma of Plains Hog-nosed Snakes (*Heterodon nasicus*) at Thomson Sand Prairie (Carroll Co., Illinois) in 2010, as a function of size class.

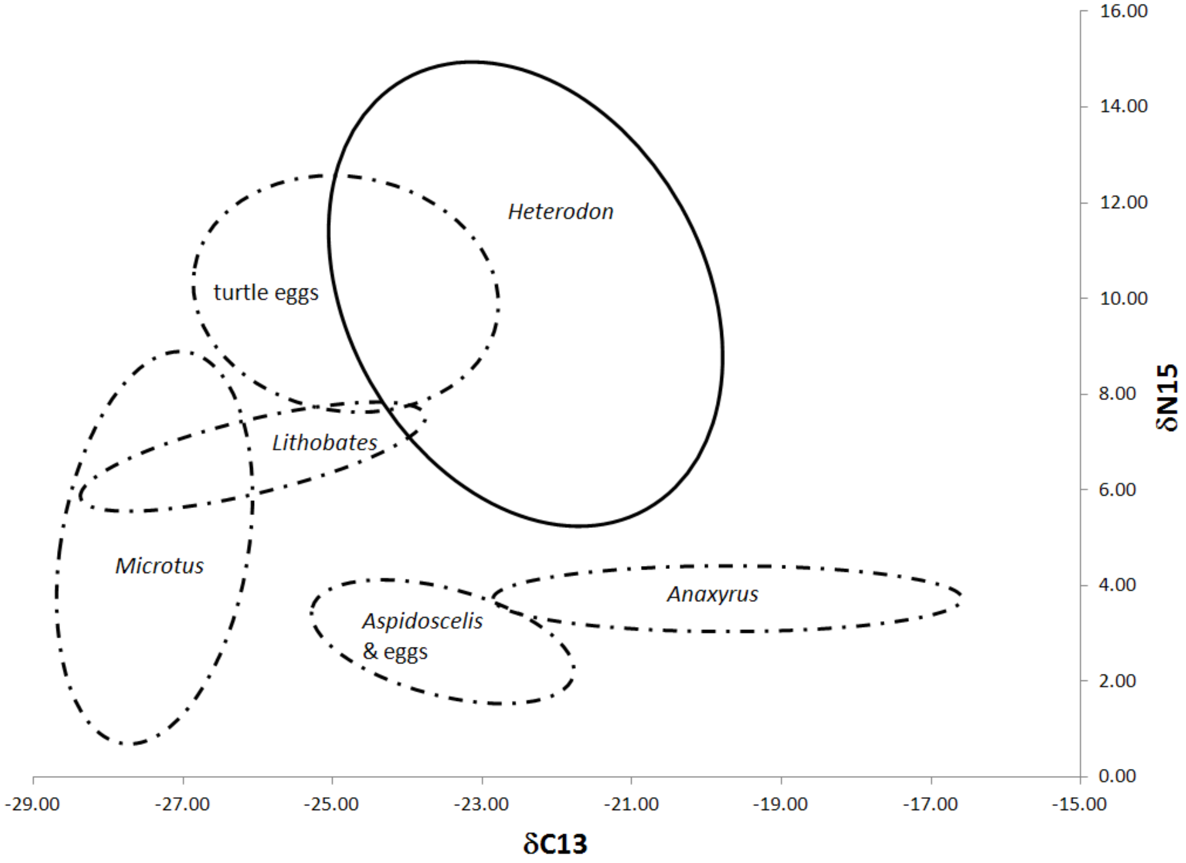

Figure 5: Structure of the predator-prey community of Thomson Sand Prairie (Carroll Co., Illinois) in 2010 as depicted in trophic space. Stable isotope values for C^{13} and N^{15} are presented for whole organisms (dotted circles) or Plains Hog-nosed Snake scale tissue (solid lines).

Appendix I : “Deliverables” of five digital images providing representative views of the research being conducted during the completion of this project.

Image A – Thomson Sand Prairie (Carroll County, Illinois), showing one of the alluvial blowout habitats where vegetation is sparse.

Image B – Two subadult *Heterodon nasicus* (Plains Hog-nosed Snake) at Thomson Sand Prairie (Carroll Count, Illinois); resting posture (left subject) and death-feigning posture (right subject).

Image C – Adult *Heterodon nasicus* depredating the eggs from a *Chrysemys picta* (Painted Turtle) nest on Thomson Sand Prairie (Carroll Co., Illinois).

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Image E – Adult female *Chelydra serpentina* (Snapping Turtle) constructing a nest at Thomson Sand Prairie (Carroll Co., Illinois).

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