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Comparison of Two Diet Collection Methods in the Lizards, *Sceloporus jarrovii* and *Sceloporus virgatus*

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The diet of the Striped Plateau Lizard (*Sceloporus virgatus)* has been described as insectivorous (Cole 1963; Smith 1996, 1999), although a conclusive diet analysis has not been done. Several experimental studies have shown the lizard's willingness to eat mealworms (*Tenebrio*), crickets (*Acheta*), and acridid grasshoppers in both laboratory and field-based feeding experiments (Ballinger and Holscher 1983; Phelan and Niessen 1989; Weiss 2001). A stomach content analysis on individuals from Sonora, Mexico showed a high percentage of the diet composed of Hymenoptera (ant: 52%, non-ant: 18%) and adult Coleoptera (14%) (G. R. Smith, unpubl. data). Smith (1999) noted anecdotally that *S. virgatus* has also been observed attempting to eat a centipede (30–40 mm in length).

The diet of the Yarrow's Spiny Lizard (*Sceloporus jarrovii*) is much more extensively known than that for *S. virgatus*. *Sceloporus jarrovii* feeds on a wide variety of insects, spiders, and plant matter. Stomach content analyses have shown that adults eat more Coleoptera, large Heteroptera (formerly Homoptera), and Lepidoptera, whereas neonates feed predominantly on small Heteroptera and Hymenoptera (ants). Both age classes eat approximately equal amounts of Orthoptera, Heteroptera (formerly Hemiptera), Diptera, Hymenoptera, and Arachnida (Bursey and Goldberg 1993). In a separate study, Barbault et al. (1985) observed that the bulk of adult stomach contents were made up of Coleoptera, Orthoptera, and Lepidoptera. There were also observed differences in the size of prey items present between neonates and adults; however, it has been suggested that this could result from differences in gape size and microhabitat use (Bursey and Goldberg 1993). These differences in prey size captures may help to reduce competition among different ages and sizes of *S. jarrovii* (Simon 1976).

A diet analysis based on behavioral observations has not been completed on either *S. virgatus* or *S. jarrovii*. All previous diet studies of *S. jarrovii* have been conducted by stomach content analysis via dissection (Ballinger and Ballinger 1979; Barbault et al. 1985; Bursey and Goldberg 1993; Goldberg and Bursey 1990; Simon 1975, 1976; Simon and Middendorf 1985). Diet studies of *S. virgatus* have not been completed by any method. Diet analysis of other *Sceloporus* species have also only been completed using stomach contents (Barbault et al. 1985; Ballinger et al. 1977; Rose 1976). In order to test the validity of determining diet through behavioral observations, I compared prey selection in observations with stomach contents as part of a long-term study of foraging behavior.

METHODS

The foraging study was conducted at two sites (Herb Martyr and John Hands Campgrounds) near the American Museum of Natural History's Southwestern Research Station (SWRS), located outside Portal, Arizona (USA) inside the Coronado National Forest of the Chiricahua Mountains from May to July in 2004 and 2005. Both sites are located in seasonally dry creek beds adjacent to two USDA Forest Service Campgrounds a few miles from SWRS and have ample sympatric populations of both *S. jarrovii* and *S. virgatus*.

The Herb Martyr (HM) site was located at 31.8724167°N, 109.2352667°W (elev. 1787 m). This study area was 200 m long and spanned the width of the creek bed (3–7 m) and ca. 7 m on each side of the creek bank. The transect was demarcated at the northwest end by a barbed wire fence, delineating the end of the campground property, and on the southeast end by a dam. The John Hands (JH) study area was 320 m in length and was located at 31.8782333°N, 109.22295°W (elev. 1714 m). The site was bordered by a 0.5 m tall concrete dam at the top and a 5 m tall dam at the lower end. The width of the creek bed varied between 10 and 15 m, and up to 20 m of creek bed bank was also used. Within the first few weeks of the study, the creek beds located at both sites were dry, with no water flowing. Both sites had an upper canopy primarily of Alligator Juniper (*Juniperus deppeana*), Arizona Sycamore (*Platanus wrightii*), and Ponderosa Pine (*Pinus ponderosa*), with an understory of willow (*Salix* sp.), *Yucca* sp., and seedlings of the larger trees.

Behavioral Assays.—During the first week of research in both 2004 and 2005, up to 30 individuals of each age class and species at both sites were caught by handheld nooses. Captured lizards were identified to species, sexed, and placed into an age category (neonate or adult). Neonate and adult *S. jarrovii* were easily distinguishable based on size because the neonates were less than half the size of the adults. Lizards were individually marked using toe clips (Tinkle 1967) and paint markers (Jones and Ferguson 1980; Simon and Bissinger 1983) and then released.

Feeding behavioral assays were then conducted May to July. Lizards were observed daily from 0830 h to 1200 h, and often from 1400 h to 1600 h, in half-hour increments. Observations were conducted from at least a 1 m distance to avoid disturbing the lizards. I recorded the size and type (description and taxonomic classification) of prey captured. A total of 106 *S. jarrovii* adult, 70 *S. jarrovii* neonate, and 73 *S. virgatus* adult successful foraging observations were made. At least one observation was made per marked lizard, and in some cases, up to four observations were made per lizard.

Stomach Content Collection.—Stomach flushes, using the methods of Legler (1977), Legler and Sullivan (1979), and James (1990), were performed on ten unmarked lizards of each group (*S. jarrovii* adults, *S. jarrovii* neonates, and *S. virgatus* adults) at each site to compare behavioral observations of prey choice with stomach content analysis. Lizards captured for stomach flushing were collected in creek beds, upstream and downstream of the boundaries of each site to avoid disrupting the study populations. All lizards were collected between 1100 h and 1200 h and returned to the sites by 1700 h the same day. Stomach flushing was done in a laboratory at the Southwestern Research Station.

Stomach contents were collected using the following method. The lizard's mouth was propped open using a stiff plastic ring. Then a thin catheter (inner diameter = 1.19 mm, outer diameter = 1.70 mm) was passed down the esophagus until reaching the end of the stomach, as felt by external palpation (Legler and Sullivan 1979). To further reduce the risk of over-insertion, the length of the tube was measured against the outside of the lizard body to just before the forelimbs, which corresponds to the midpoint of the stomach (James 1990). Once the tube was inserted it was connected to a 10cc hypodermic syringe and enough room-temperature water was pumped into the stomach to enable the food to rise to the mouth. Early trials suggested that cold water was more likely to put the lizard into shock during stomach flushing. Food was removed either by forceps or by suspending the lizard upside-down over a fine mesh net to let the items flow out with the water (James 1990; Legler 1977; Legler and Sullivan 1979). While the stomach was full of water, the abdomen was gently massaged to break up the food bolus (Middendorf 2005). This entire flushing process was repeated until no food came out of the stomach (2–4 times total).

After stomach flushing completion, the lizards were held in small plastic terraria and monitored for any adverse effects for at least one hour. If the lizard stopped breathing after the procedure was completed, it was briefly immersed in a bucket of ice water to "jump-start" its system (James 1990). Any lizards that did not survive the procedure were dissected to determine cause of death and any residual stomach contents were removed and preserved (N = 18). These contents were combined with the prey items obtained through stomach flushing for analysis.

All food items were collected in a fine mesh net, transferred to a 45 ml sample vial, and stored in 75% ethanol until analysis in Fall 2005. Stomach contents were sorted and identified to the lowest classification level possible, usually to the Order level (Bland and Jaques 1978; Jackman 1997; Milne and Milne 1980; Roth 1985). The abundance of prey items was identified by counting the number of complete organisms or by using the number of heads, legs, or wings to estimate total number of items eaten of each prey type.

RESULTS

Diet content in 2004 and 2005 foraging assays differed from that of stomach content analysis (Table 1). The predominantly observed prey types for *S. jarrovii* adults were Formicidae (2004: 27%, 2005: 63%), Diptera (24%, 23%), and Isoptera (44%, 2004 only). In contrast, the stomach contents for this lizard group primarily consisted of Formicidae (55%) and Hymenoptera (16%). The stomach contents contained several taxa that were never observed in the behavioral observations: Hymenoptera (as stated previously), Arachnida (3%) and Heteroptera (8%). Also, the high prevalence of Diptera in the observed diet was not confirmed by stomach content analysis, as Diptera were not present in any stomachs of *S. jarrovii* (Table 1).

Neonates of *S. jarrovii* showed similar differences in diet based on the source of the data. The dominant prey taxa in behavioral assays were Formicidae (2004: 27%, 2005: 77%) and Diptera (2004: 62%, 2005: 16%). Diptera were not found in any neonate stomachs and the most prominent prey types were Formicidae (38%) and Heteroptera (35%), a taxon not observed in the foraging assays. Several additional taxa found in the stomach contents, but lacking in the behavioral data were Arachnida (6%), Hymenoptera (non-ant; 6%), Scorpiones (1%), Isopoda (3%), and unidentifiable fleshy larvae (4%) (Table 1).

Diet comparisons of stomach contents and behavioral observations for *S. virgatus* revealed similar differences to both adult and neonate *S. jarrovii*. The dominant prey items from behavioral observations were again Formicidae (2004: 54%, 2005: 52%) and Diptera (2004: 34%, 2005: 31%). Dipterans were notably absent from stomach contents. The dominant food items found in the stomachs were: Formicidae (36%), Heteroptera (19%), and Coleoptera (23%). Heteroptera was not present in the behavioral diet and Coleoptera were only present in small amounts in 2005 (5%). Arachnida were not present in the behavioral diet, but made up 8% of the stomach content diet (Table 1).

DISCUSSION

Observations of prey attacked revealed that the dominant food items in the diet of adult *S. jarrovii* were Formicidae, Diptera, and Isoptera (Table 1). This contrasts somewhat with the stomach content diet, which consisted predominantly of Formicidae and Hymenoptera (non-ant) (Table 1). Previous stomach content analyses have shown that the adult *S. jarrovii* eat mainly Coleoptera, Heteroptera, Lepidoptera, and Orthoptera (Barbault et al. 1985; Bursey and Goldberg 1993). Differences in dominant food items between this study and previous work may be due to the differences in study locations, which undoubtedly have differing available prey bases. The Bursey and Goldberg (1993) study occurred in Pima Co., Arizona (southwest of Tucson) and the Barbault et al. (1985) study occurred in northern Mexico, whereas this study was limited to the early summer in the Chiricahua Mountains (southeast of Tucson). In addition, the high incidence of Isoptera in the observational diet was an isolated incident in 2004. A single individual that I was observing found a hole in the ground from which reproductive termites were emerging, enabling the lizard to eat a large number of termites (32) with very little effort.

For neonate *S. jarrovii*, the diet was very similar to adult *S. jarrovii* in that it was dominated by Formicidae and Diptera in the behavioral observation diet (Table 1). The stomach content diet was high in Formicidae, but also Heteroptera (Table 1). For *S. virgatus* the dominant prey items in the observational diet are Formicidae and Diptera, similar to both age classes of *S. jarrovii* (Table 1). The stomach contents, however, are made up mainly Formicidae, Heteroptera, and Coleoptera (Table 1).

Observed diet analysis discrepancies may be an artifact of the methodology surrounding stomach content analysis in general. The stomach flushing technique may not be as exhaustive as dissection, as some prey items may not be successfully removed from the lizard's stomachs. It is possible that the items least likely to be removed were large-bodied organisms that could not be washed out by water. Of the 18 individuals (3 *S. jarrovii* adults, 11 *S. jarrovii* neonates, and 4 *S. virgatus*) that died during the procedure and were later dissected, only one individual (an adult *S. virgatus*) had a completely empty stomach, so it appears that this method did not disgorge all items. Although not all prey items were extracted from the stomachs, those items that were extracted are still useful for comparison with the behavioral observations.

A second explanation for the diet discrepancy is that many prey items may have been partially digested due to the length of time since eating, resulting in an underestimate of easily digested organisms (Vitt and Zani 1998). All of the lizards in this study were kept for a minimum of two hours before stomach flushing. Lepidopterans, which are known to be dominant in the diet of adult *S. jarrovii* (Barbault et al. 1985; Bursey and Goldberg 1993), were not present in high numbers in the stomach samples (Table 1), possibly because these soft-bodied organisms are digested fairly quickly (Bullock et al. 1993). The only evidence of Lepidoptera in the stomach contents were wings, which I counted to get an estimate of the number of individuals eaten. This technique may have resulted in an underestimate of the number of Lepidoptera present in the diet, though behavioral observations were consistent with this low prevalence (Table 1). Perhaps completing stomach flushes immediately upon capture would alleviate some of these issues, however, it still does not resolve the problem of lizards that have been foraging for hours prior to capture.

Diptera were a major dietary component for all three lizard groups according to the behavioral observations, but were not present in any of the stomach contents (Table 1). Diptera are soft-bodied, with very little chitin, making them an easily digested prey source. However, unlike the Lepidoptera, there was no evidence of Diptera wings or other identifiable body parts in the stomach contents. Unpublished data of *S. virgatus* stomach contents also show no evidence of Diptera eaten, though "unidentifiable" organisms made up 9% of the diet and perhaps partially digested Diptera were contained therein (G. R. Smith, unpubl. data). Another alternative is that no Diptera were actually ingested, although this conflicts with the behavioral observations, and seems very unlikely due to heavy prevalence of Diptera at the site (pers. obs.).

The contradiction between the diet sources within this study may also be an artifact of observer bias. Since behavioral observations occurred at a distance of over one meter, and often over two meters, it is entirely possible that what the observers thought was a "fly" may simply have been another small flying insect that could not be distinguished at that distance. This bias may also explain why Heteroptera and Hymenoptera were either rare or completely lacking in the observational diet of all three lizards groups, yet are common in the stomach contents of this study (Table 1) and other previous studies (Bursey and Goldberg 1993). However, I am confident that the behavioral methodology was sound and may need only further refinement to improve accuracy.

While both the stomach flushing method and behavioral observations provide valuable insight into the diet of *S. jarrovii* and *S. virgatus*, it is unclear which method is truly representative of their diets. Likely, both methods are only pieces of the whole picture, but by looking at the data from both methods together, we gain a much greater understanding of the diet of these two lizard species. Further research comparing behavioral observations and stomach contents will need to be conducted in order to determine if an observational method alone can be used as a representation of lizard diet. This experiment is also the first time that this particular behavioral methodology has been used, so repetition of

TABLE 1. Total diet composition from stomach contents and behavioral observations. Data for each prey item are represented as the percentage of the entire diet from that source.

Lizard Group	Data Source	N	Arachnida	Chilapoda	Coleoptera	Diptera	Formicidae	Heteroptera	Hymenoptera	Isoptera	Lepidoptera adult	Orthoptera	Scorpiones	Unidentified larvae
Sceloporus	stomach	20	3.3	0.7	9.8	0	54.9	8.5	15.7	0	3.3	0	0	2.6
jarrovii	behavior 04	76	0	0	0	24.7	27.3	0	0	44.2	0	0	0	1.3
adult	behavior 05	30	0	0	6.7	23.3	63.3	0	0	0	6.7	0	0	0
Sceloporus	stomach	20	6.1	0	6.1	0	37.8	35.0	5.6	0	1.7	0	1.1	3.9
jarrovii	behavior 04	27	0	0	0	61.5	26.9	0	0	7.7	3.9	0	0	0
neonate	behavior 05	43	0	0	7.0	16.3	76.7	0	0	0	0	0	0	0
Sceloporus	stomach	20	8.8	0	23.4	0	35.8	19.0	4.4	0	5.8	0	0	2.9
virgatus	behavior 04	38	0	0	0	34.3	54.3	0	0	0	2.9	2.	0	2.9
adult	behavior 05	35	0	0	5.6	30.6	52.8	0	2.8	0	0	5.6	0	2.8

the methodology may be necessary. Other than keeping lizards in enclosures and feeding them all possible prey items, the two methods in this study are our best hope of determining lizard diets while providing minimum impact. In addition, it is important to develop a method to determine diet through behavioral observations, since this is the only method by which we can also record prey species-specific data on pursuit time, search time, handling time, or prey capture success rate in the field.

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