BIOLOGIA (LAHORE)
June, 2020, 66 (I), 23-27

PKISSN 0006 – 3096 (Print)
ISSN 2313 – 206X (On-Line)

Diet composition of black rock Agama (*Laudakia melanura*)

MISBAH RAZZAQ, MUHAMMAD RAIS*, MUHAMMAD ARSLAN ASADI, SALEHA ABBASI & ABDULLAH IBRAHIM

Department of Wildlife Management, Faculty of Forestry, Range Management and Wildlife, Pir Mehr Ali Shah, Arid Agriculture University Rawalpindi, 46000, PAKISAN

ARTICLE INFORMAION

Received: 31-05-2019
Received in revised form: 25-04-2020
Accepted: 30-06-2020

***Corresponding Author:**

Muhammad Rais:
sahil@uaar.edu.pk

Short Communication

ABSTRACT

The empirical data on diet of many reptilian species of Pakistan are scarce. We aimed to document major dietary items of Black Rock Agama (*Laudakia melanura*) in rocky habitat, croplands, areas around human habitations and natural streams of Mori Said Ali Game Reserve (AJ&K) using stomach-content flushing and fecal analysis. Of the recorded 17 dietary items, ten (59%) were of plant origin and seven (41%) were of animal origin. *Pyrus pashia* (80%) and *Isodon rugosus* (0.37 ml) were recorded as the most frequent plant species in fecal samples and stomach contents, respectively. The majority of insects consumed by the lizard species were Coleopterans (0.22 ml) followed by Hymenopterans (0.18 ml) and Hemipterans (0.18%). The similarity index showed that the similarity in food items at sites "rocky habitat-human habitations" and "rocky-stream habitat" was highest (38% each) followed by "human habitations-stream habitat" (33%), rocky habitat-cropland habitat (30%), "cropland habitat- human habitations and cropland habitat- stream habitat (24% each) whereas the similarity in food items taxa using stomach content analysis at sites "rocky habitat-cropland habitat" was highest (87%) followed by "stream habitat-rocky habitat" and "stream habitat-cropland habitat" (66% each), "rocky habitat-human habitat" (25% each). Our findings established omnivory in the species for the first time. The lizard species feeds on non-woody vegetation such as grasses, herbs and shrubs and also ingests insects.

Keywords: Stomach content flushing, Fecal pellet analysis; *Pyrus pashia*

INTRODUCTION

The genus *Laudakia* currently encompasses 20 species distributed in parts of Asia (Iran, Afghanistan, Pakistan, India & Nepal) and Middle East. The species is distributed in western mountains, areas bordering Iran and Afghanistan and northern areas of Pakistan (Baig *et al.,* 1989 & 2012). The Black Rock Agama (*Laudakia melanura*) can be easily recognized by its dorso-laterally depressed head and body. It has a long segmented tail. The sides of head have spiny scales which are smooth or keeled on vertebral side. The callous glands are present in males at precloacal and abdominal positions (Baig, 1999). The Black Rock Agama inhabits cliffs and rocks of mountain ranges, vertical slopes of clay around bushes (Smith, 1935; Baig, 1999). The diet of other species of *Laudakia* has been studied to some extent elsewhere in the world. For instance, food items of Yellow-Headed Agama (*Laudakia nupta fusca*) were reported by Qashqaei & Ahmadzadeh (2015) and of Starred Agama (*Laudakia stellio*) by Ibrahim & El-Naggar (2013). However, there is a dearth of scientific literature on many lizard species of Pakistan including Black Rock Agama. We aimed to report diet composition of Black Rock Agama. Our findings would be helpful in understanding natural history of the lizard species. The results are also expected to shed light on the ecology of the study area through dietary items consumed by the lizard.

MATERIALS AND METHODS

Study area

We selected four representative habitats viz. rocky habitat, cropland habitat, habitat around human habitations and habitat around natural

Author's Contribution: M.N.K., Conducted experimental work; **N.R.,** Supervised whole investigation; **B.J.,** Helped in data analysis and blood sampling; **M.A.I.,** Helped in experimental work and wrote up the manuscript

streams of Mori Said Ali Game Reserve, Azad Jammu & Kashmir (AJ&K). The Mori Said Ali Game Reserve (33° 56.053 N; 74° 04.823 E) is located about 10 km in the south of Forward Kahuta City, District Haveli, AJ&K. The reserve covers an area of 273 ha with elevation ranges from 1666 m to 2500 m. The area mainly consists of hills with a very dense forest and cultivation of crops, mainly wheat and maize, on small ridges. The temperature remains cool throughout the year with minimum and maximum average temperature range of 2- 18 °C and 16- 30 °C in winter and summer, respectively. The average annual precipitation is 550 mm (Anwar *et al.,* 2015).

Sample effort, isolation of stomach and fecal content

We visited the four selected habitats on monthly basis from October, 2015 to May, 2016 to obtain fecal and stomach contents samples. We randomly laid out four transects (each having length and width of 100 and 30m, respectively) to standardize our search effort (Sutherlands, 1996). We captured specimens using a noose, and analyzed 21 stomach content samples (rocky habitat n=5, cropland habitat n=6, human habitat n=5 and stream habitat n=5) using stomach content flushing technique without administrating anesthesia while fecal droppings along the transects were collected. The jaws of the specimens were opened using sterilized spatula, stomach contents were flushed out by pumping water through an infusion tube of soft material connected with a syringe (as described by Legler, 1977). We repeated the procedure until no more stomach contents appeared, and released back the specimens at the capture site. The water containing stomach contents was decanted through a sieve, and food items were picked up with forceps, fixed and preserved in 70% ethanol and 10% formaldehyde. We identified the items under stereo-microscope (model specs) to the lowest possible taxonomic level.

We collected fecal samples from the field using forceps and kept in tagged sampling bags. The lizard species is not listed as a protected species in Azad Jammu and Kashmir Wildlife Act (2014). We analyzed 22 composite fecal samples, each composite sample contained 10 fecal pellets, (rocky habitat n=6, cropland habitat n=6, human habitat n=5 and stream habitat n=5) using micro-histological analysis following Sparks & Malecheck, 1968 with some modifications. We air dried the fecal pellets (samples) collected from the study area. The samples were crushed, using manual mortar and pestle, into small pieces and passed through sieve, tea strainer, to remove coarse material and dust. The samples were washed in flowing water and soaked in a soaking solution (distilled water, ethyl alcohol and glycerine in 1:1:1 ratio) for overnight. The samples were then grounded with Virtis Homogenizer. We transferred fifty percent of the samples to a labeled test tube and added five percent warm sodium hydroxide solution, and then heated the test tube in a boiling water bath for 4 to 6 minutes to allow particles to settle before removing the supernatant dark fluid. We repeated the procedure until we obtained a comparatively clear solution. We washed the material with warm distilled water for 2 minutes and dehydrated it through a series of 25 percent, 50 percent, 75 percent and 100 percent alcohol treatments, each for 10 minutes. The alcohol was removed through a series of xylene and alcohol mixtures (25 percent, 50 percent, 75 percent and 100 percent xylene) each for 10 minutes, except 100 percent for overnight. We transferred the material next day onto a clean glass slide, spread and mounted in dibutyl phthalate xylene (DPX) mounting medium under a cover slip. We prepared four slides for each sample and observed five microscopic fields per slide. The plant samples (leaves, stems, flowers) collected from the habitat were also treated in the same way to prepare reference slides for comparison with the photographs cells of plants from sample slides (Sparks & Malechek, 1968; Alipayo *et al.,* 1992).

Statistical and ecological analysis

We expressed the results as volume (ml), relative volume (%), total number of plant species, total number of animal species and total number of food items (plant and animal) (Windell & Bowen, 1978). We determined volume of each food item by water displacement using graduated cylinders to the nearest 0.01 ml. We calculated relative volume as volume of each food item divided by total volume of food items from each specimen multiplied by100 (Hynes, 1950). For each prey category, the frequency of occurrence (Fi%) was calculated using the formula: Fi= ni/n*100, where Fi= frequency of occurrence of i food item in the sample, ni= number of stomach content samples in which the i item was found, and n= total number of stomach contents samples. The food item taxa among the habitats were compared using Kruskal-Wallis test (α= 0.05) while number of food item taxa recorded through

fecal and stomach content analysis were compared using Wilcoxon test (α= 0.05). The Sørensen similarity index (S_S) was calculated using formula: S_S = 2C/(A+B) *100, where C = number of taxa common in both sites; A = number of taxa recorded from habitat 1 (four transects data pooled) and B = number of taxa recorded from habitat 2.

RESULTS

We recorded 17 food item taxa from the diet of Black Rock Agama from the study area (four habitat types). These included ten species of plants (59%) and seven insect orders (41%). *Pyrus pashia* (80%) was recorded as the most frequent plant species in fecal samples while *Isodon rugosus* (0.37 ml) was recorded as the most frequent plant species in stomach contents. The majority of insect taxa recorded from the diet belonged to order Coleoptera (0.22 ml) followed by Hymenoptera (0.18 ml) and Hemiptera (0.18%) (Table I). We recorded 11 (eight plant species and three insect orders) food item taxa from the diet of Black Rock Agama from rocky habitat; seven food items (four plant species and three insect orders) from cropland habitat; 11 food items (seven plant species and four insect orders) from habitats near human habitations and nine food items (five plant species and four insect orders) from the stream habitat (Table I). The Kruskil-Wallis test revealed that the number of food item taxa in the four sites did not differ significantly using fecal analysis (P= 0.12) and stomach content analysis (P= 0.36). However, insect remains were easily recognizable from stomach contents. The number of food item taxa did not differ significantly in the two methods (P> 0.05). The similarity index showed that the similarity in food item taxa at sites “rocky habitat-human habitations” and “rocky-stream habitat” was highest (38% each) followed by “human habitations-stream habitat” (33%), “rocky habitat-cropland habitat” (30%), “cropland habitat- human habitations” and “cropland habitat- stream habitat” (24% each) whereas the similarity in food item taxa using stomach content analysis at sites “rocky habitat-cropland habitat” was highest (87%) followed by “stream habitat-rocky habitat” and “stream habitat-cropland habitat” (66% each), “rocky habitat-human habitat” (25% each).

DISCUSSION

We documented the diet of the lizard species using the combination of stomach-content flushing and fecal analysis. Valido and Nogales (1994) suggested use of fecal pellets to get a good picture of interactions of lizards with other organisms. Angelici *et al.,* (1997) preferred fecal analysis to document diet of Green Lizard (*Lacerta bilineata*). Luiselli & Rugiero (1997) and Rosenberg & Cooper (1990) also suggested use of fecal analysis. Minton (1966) reported Black Rock Agama as a herbivorous lizard species. There is a dearth of literature on diet of Black Rock Agama (*Laudakia melanura*); therefore, studies on other Agamid lizard species are referred in the following section for the comparison. The findings of present study revealed that Black Rock Agama is an omnivorous lizard. It fed on insects such as beetles (Coleoptera), wasps, ants and bees (Hymenoptera), grasshoppers (Orthoptera), earwigs (Dermaptera), bugs (Hemiptera), termites (Isoptera), flies (Diptera) and various plant species. Qashqaei & Ahmadzadeh (2015) reported Yellow-Headed Agama (*Laudakia nupta fusca*) as an omnivorous lizard. As many as 9 food items (of which 6 were animal and 3 were plant items) were recorded with a shrub species- *Daphne oleoides*, beetles and ants constituting major portion of Yellow-Headed Agama diet. Ibrahim & El-Naggar (2013) reported that diet items of Starred Agama (*Laudakia stellio*) include insects (mainly coleopterans) and plant material (30% of the stomach content). Dietary analysis of (*Laudakia stello*) conducted by Cascio *et al.,* (2001) revealed 984 (857 spring; 127 summer) food items belonging to plant seeds, fruits, insects and mollusk. The lizard consumed more insects (Coleoptera and Hymenoptera) during spring while fruits of *Pistacia terebinthus* during summer.

The aforementioned studies report various Agamid lizard species as omnivorous with insects belonging to orders such as Coleoptera, Hymenoptera and Hemiptera as major food item though diet constitution differed due to difference in geographic areas, habitat and seasons.

The findings of the present study reports omnivory in Black Rock Agama. The species feeds on non-woody vegetation such as grasses, herbs and shrubs and also ingests insects such as Coleopterans, Hymenopterans and Hemipterans.

Table I: Frequency (F, as %) of food item taxa (total, plant species, and insect order) recorded from the fecal and mean volume (V, in ml) recorded using stomach content analysis of Black Rock Agama (*Laudakia melanura*) from Mori Said Ali Game Reserve, Azad Jammu and Kashmir.

	Taxa	Rocky Habitat		Cropland Habitat		Human Habitat		Stream Habitat		Total	
		F±S.E	V±S.E	F±S.E	V±S.E	F±S.E	V±S.E	F±S.E	V±S.E	F±S.E	V±S.E
Plant species	*Pyrus pashia*	74.1±17.7	0.28±0.04	87.5±8.80	0.23±0.10	88±9.08	0.25±0.05	73±30.94	0.03±0.02	80.65±13	0.19±0.05
	Isodon rugosus	9±2.23	-	-	-	-	0.37±0.17	-	-	9±2.23	0.37±0.17
	Oenothera rosea	15±8.94	-	-	-	15± 13.2	0.16±0.05	-	-	15±11.07	0.16±0.05
	Trifolium repens	7.5± 2.88	-	-	-	-	0.09±0.07	18.7±179	0.12±0.04	13.1±103	0.1±0.05
	Veronica persica	15±7.07	-	-	-	-	-	-	-	15±7.07	-
	Rumex hastatus	-	0.12±0.04	17±4.47	0.15±0.07	-	-	-	0.21±0.02	17±4.47	0.16±0.04
	Cynodon dactylon	-	0.08±0.04	17.5±8.6	0.15±0.07	-	-	-	-	17.5±8.6	0.11±0.05
	Berberius lycuim	-		-	-	17.5±8.6	-	12.5±9.7	-	15±9.11	-
	Sausseria spp	-	0.12±0.04	-	0.17±0.09	12.5±3.3	-	18.7±17.9	0.15±0.05	15.6±107	0.14±0.14
	Tagestes minuta	-		-		-	0.11±0.04	-	-	-	0.11±0.04
Insect orders	Coleoptera	-	0.28±0.07	-	0.28±0.07	-	0.17±0.02	-	0.16±0.03	-	0.22±0.04
	Homoptera	-	0.23±0.05	-		-	-	-	0.14±0.04	-	0.18±0.04
	Orthoptera	-	0.09±0.01	-	0.09±0.01	-	-	-	-	-	0.09±0.01
	Blattodia	-	-	-	-	-	0.12±0.02	-	-	-	0.12±0.02
	Isoptera	-	-	-	-	-	0.07±0.04	-	-	-	0.07±0.04
	Hymenoptera	-	-	-	0.21±0.02	-	0.20±0.07	-	0.20±0.10	-	0.20±0.06
	Hemiptera	-	-	-	-	-	-	-	0.18±0.05	-	0.18±0.05
Total	Mean	24.1±7.7	0.17±0.04	40.6±7.3	0.18±0.06	33.2±318	0.17±0.05	30.7±19	0.14±0.04	-	0.22±0.04
	Total No. of Plant species	5	4	3	4	4	5	4	4	9	8
	Total No. of insect Taxa		3		3		4		4		7
	Total No. of food Item Taxa	11		7		11		9		17	

REFERENCES

Anwar, M., Asadi, A. and Mustafa, N., 2015. Baseline studies on Wildlife Diversity in the Selected Protected Areas of Pakistan-Phase-11. Annual Progress Report (2014-2015 unpublished) HEC Project. Dept. of Wildlife Management, Pir Mehr Ali Shah Arid Agriculture University Rawalpindi.

Alipayo, D., Valdez, R, Holechek, R. J. L. and Cardenas, M., 1992. Evaluation of microhistological analysis for determining ruminant diet botanical composition. *J. Range. Manag*e., 45: 148-152.

Angelici, F. M., Luiselli, L. and Rugiero, L., 1997. Food habits of the green lizard, *Lacerta bilineata,* in central Italy and a reliability test of fecal pellet analysis. *Italian J. Zool*., 64: 267-272.

Baig, K. J., 1989. A new species of Agama (Sauria: Agamidae) from northern Pakistan. *Bull. Kitakyushu Mus. Nat. Hist.,* 117-122.

Baig, K. J., 1999. Description and ecology of a new subspecies of Black Rock Agama, *Laudakia melanura* (sauria: agamidae) from Balochistan, Pakistan. *Russian J. of Herpetol*., 6: 81 – 86.

Baig, K. J., Wagner, P., Natalia, B., Jeva, A. and BöHme, W., 2012. A morphology-based taxonomic revision of *Laudakia Gray*, 1845 (Squamata: Agamidae). *Vert. Zool*., 62: 213-260.

Cascio, P. L., Corti, C. and Luiselle, L., 2001. Seasonal variations of the diet of *Laudakia stello* from Nisyros island, Dodecanese (Greece). *Herpetol. J.,* 11: 33-35.

Ibrahim, A. A. and El-Naggar M. H., 2013. Diet and reproductive biology of the starred agama (*Laudakia stellio*) (Linnaeus, 1758) (Squamata: Agamidae), in the northern Sinai, Egypt. *Zool. Middle East.*, 59(2): 136 – 143.

Legler, J. M., 1977. Stomach flushing: a technique for chelonian dietary studies. Herpetologica, 33: 281-284.

Minton, S. A., 1966. A contribution to the herpetology of West Pakistan. *Bull. Am. Mus. Nat. Hist*., 134: 27-184.

Qashqaei, A. T. and Ahmadzadeh, A., 2015. Dietary Records of Yellow-Headed Agama in Hormozgan Province, Iran. *Russ. J. Herpetol.,* 22 (4):315-317.

Rosenberg, K. V. R. and Copper. J., 1990. Approaches to avian diet analysis. Studies in Avian Biol., 3: 80-90.

Smith, S. A., 1935. *The Fauna of British India including Ceylon & Burma*. Today & Tomorrow Publishing, New Dehli, India. 440 pp.

Sparks, D. R. and Malechek, C., 1968. Estimating percentage dry weight in diets using a microscopic technique. *J. Range Manag.*, 21: 264–265.

Sutherland, W. J., (ED.)., 1996. Ecological Census Techniques: A Handbook. Cambrige University Press. United Kingdom. 1-336.

Valido, A. and Nogales, M., 1994. Frugivory and seed dispersal by the lizard *Gallotia galloti* (Lactertidae) in a xeric habitat of the canary Islands. Oikos 70: 403-411.

Windell, J. T. and Bowen, S. H., 1978. Methods for study of fish diets based on analysis of stomach contents. *In Methods for the Assessment of Fish Production in Fresh Waters*. 3rd edn. (T. Bagnel, ed.). Oxford. Blackwell Scientific Publications. pp: 219-226.