

Are the males of *Dendropsophus minutus* (Peters, 1872) spider devourers?

João Lucas Pereira Ferreira¹, Kássio de Castro Araújo², Etielle Barroso de Andrade², Caio Vinícius Mira-Mendes³

¹ Programa de Pós-graduação em Zoologia, Departamento de Ciências Biológicas, Universidade Estadual de Santa Cruz-UESC, Rodovia Jorge Amado, km 16, Ilhéus, Bahia 45662-900, Brasil

² Grupo de Pesquisa em Biodiversidade e Biotecnologia do Centro-Norte Piauiense-BIOTECPI, Instituto Federal de Educação, Ciência e Tecnologia do Piauí-IFPI Campus Pedro II, Rua Antonino Martins de Andrade 750, Engenho Novo, Pedro II, Piauí 64255-000, Brasil

³ Departamento de Biologia, Universidade Estadual do Maranhão, São Luís, Maranhão 65055-310, Brasil

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ABSTRACT

In this study, we described the diet composition of males of the treefrog *Dendropsophus minutus* from the region of the Chapada das Mesas National Park, Maranhão, Northeastern Brazil. We collected 19 males, of which 17 had stomach contents. Of that, five prey categories were registered, with spiders being the most consumed prey item. We also observed that the snout-vent length of each male had a weak influence on prey selection. Notably, *D. minutus* consumed a substantial quantity of spiders. Nonetheless, further studies considering temporal variation, reproductive period, and prey availability, among others, are still needed to elucidate the role of spiders on the diet of *D. minutus*.

Key Words: Anuran; Amphibian; Diet; Feeding Habits; Trophic Ecology.

Dietary habits play a fundamental role in the natural history of a species, influencing its interactions with the environment and other organisms while also contributing to the dynamics of ecosystems (Moser *et al.*, 2017). Among vertebrates, anuran amphibians are a key group in studies of trophic interactions due to their role in energy transformation in the environment (Colón-Gaud *et al.*, 2009) and their intrinsic connection with aquatic and terrestrial ecosystems (Maneyro & Rosa, 2004). Therefore, through studies on the trophic ecology of frogs, it is possible to understand how energy flow and the cycling of matter occur in food chains, making conservation models more effective for both these animals and the ecosystems they inhabit (Núñez *et al.*, 2021).

The majority of anuran species are considered generalist predators, with their diet based on what is most available in the environment in which they

live (Eterovick & Sazima, 2004), with arthropods constituting their primary diet (Duellman & Trueb, 1994; Solé & Rödder 2010). Nevertheless, in various anuran groups, there has been observed a tendency toward diet specialization, often linked to behavioral, morphological, or physiological traits (e.g., Toft, 1980; Castro *et al.*, 2020; Carilo-Filho *et al.*, 2021).

The lesser treefrog *Dendropsophus minutus* (Peters, 1872) is a small-sized hylid ranging from 21 to 28 mm, presenting yellow, cream, or orange-brown tones of dorsal coloration pattern with dark dorsal and transverse bands that resemble an hour-glass shape (Andrade *et al.*, 2022; Frost, 2024). It is a widespread species in South America that inhabits typically open areas in different biomes, including disturbed areas and urban environments (Morais *et al.*, 2012; Abegg *et al.*, 2014; Vaz-Silva *et al.*, 2020). Likewise in Brazil as a whole, this species is also well

distributed in Maranhão state, Northeastern Brazil, being reported to the Amazon and Cerrado biomes, besides the coastal zone (Brasileiro *et al.*, 2008; Andrade *et al.*, 2017; Araújo *et al.*, 2020).

The feeding habits of *D. minutus* were already reported for localities in the North (Van Sluys & Rocha, 1998; Soeiro *et al.*, 2022), Northeast (Santos *et al.*, 2004), and South (Leivas *et al.*, 2018) Brazilian regions, which suggested a generalist and opportunistic diet. Nonetheless, as the diet anuran composition is usually influenced by different factors from environmental characteristics to morphological traits (Protázio *et al.*, 2019; Tupy *et al.*, 2021), the present study aimed to investigate the diet composition of males of *D. minutus* located at one Cerrado area from the region of the National Park Chapada das Mesas, Maranhão, Northeast Brazil.

During the fieldwork conducted on April 20, 2023, in the Chapada das Mesas National Park (CMNP), located in the state of Maranhão, Northeastern Brazil, we encountered an abundant population of *D. minutus* vocalizing in a temporary pond near to one the park entrances (7°09'15.0" W, 47°24'16.3"S, WGS84 datum, 315 m a.s.l.). The CMNP covers an area of about 160.000 ha, and the main plant physiognomy is Cerrado *sensu stricto* (Eiten, 1983; Ribeiro & Walter, 1998), but it is also influenced by adjacent Amazon and Caatinga biomes (Moraes & Lima, 2007), which makes it be considered a mosaic of vegetation ranging from open grasslands to dense forest formations and semideciduous patches on mountain tops (MMA, 2007; Silva *et al.*, 2019). The climate is tropically humid, with annual average temperatures of 23 °C and average annual rainfall ranging from 1500 to 1250 mm (MMA, 2007).

Of the uncountable vocalizing males observed around 20 h, we manually collected 19 adult males (collecting permits: SIS-BIO 82619/1), which were transported in humid plastic bags and, after that, euthanized following the ethical procedures of the Ethics Committee on the Use of Animals of the Universidade Estadual de Santa Cruz (CEUA/UESC - N° 024/21) and Brazilian Guide for the Production, Maintenance or Use of Animals in Teaching or Scientific Research Activities (Jared *et al.*, 2023). Collected specimens were fixed in formol 10%, preserved in alcohol 70%, and deposited in the Biological Collection of the Instituto Federal de Educação, Ciência e Tecnologia do Piauí – IFPI Campus Pedro II, state of Piauí, Brazil (CBPII 445–464).

In the laboratory, we dissected the collected individuals and remove their digestive tracts using a stereomicroscope (Lumen® LM310BZ). Each prey item found was counted and identified to the lowest taxonomic level (usually Order) using specialized literature (Triplehorn & Johnson, 2011). Plant material and unidentified diet fragments were not included in the statistical analyses. Using a digital caliper (0.01 mm precision) we measured the snout-vent length (SVL) and mouth width of each treefrog, besides the length and width of the prey items. We used the ellipsoid formula ($V = 4/3\pi (L/2) (W/2)^2$) to estimate the prey volume, where L and W represent the values of length and width, respectively (Dunham, 1983). In addition, we used the relative importance index (RII) to investigate how each prey item contributed to the species diet through the following formula: $RII = (F\% + N\% + V\%)/3$, where F, N, and V represent the percentual values of frequency of occurrence, abundance, and volume of each prey category (Pinkas *et al.*, 1971). The trophic niche breadth (prey volume) was measured using the inverse of Simpson's (1949) diversity index: $B = 1 / \sum_{i=1}^n p_i^2$, where p is the proportion of the resource category used and n is the number of resource categories adopted. The values can range from 1 (exclusive use of a single category) to n (equal use of all categories). Once our data violated the assumptions of simple linear regression ($p < 0.05$), we used the non-parametric Kernel regression test (Nadaraya-Watson, 1964) to investigate if the anuran snout-vent length and mouth width have a relationship with the prey size (estimated by prey volume). These analyses were performed using the “vegan” R package (Oksanen *et al.*, 2016).

Of the analyzed individuals, 17 anurans had at least one prey category in their stomachs. We found 20 prey items in the diet of *D. minutus* labeled in five categories, including plant pieces, which were not included in the statistical analyses. The average number of prey per stomach was 1.29, and the trophic niche breadth (prey volume) was 1.11. Araneae order was the most representative. Of the 20 prey, 70% were spiders, which represents about 95% of the total volume of consumed prey and 60% ($n = 10$) of the occurrence frequency (Table 1).

We found no support for the relationship between the prey size and anuran snout-vent length ($R^2 = -0.017$, $P > 0.05$, Deviance explained = 5.55%) and mouth width ($R^2 = -0.0256$, $P > 0.05$, Deviance explained = 4.76%). Although we suggest the

Table 1. Composition of the identified prey items consumed by *Dendropsophus minutus* males (n = 19) from the National Park Chapada das Mesas, Maranhão, Northeastern Brazil. Respective absolute and percentual values of abundance (N), volume (V), and frequency of occurrence (F), besides the relative importance index (RII).

Orders	N	N%	V	V%	F	F%	RII
Araneae	14	70	455.8	94.8	10	62.5	75.8
Coleoptera	2	10	8.6	1.8	2	12.5	8.1
Hymenoptera	2	10	2	0.4	2	12.5	7.6
Other items							
Larvae	2	10	14.3	3	2	12.5	8.5
Plant pieces	4	-	-	-	-	-	-
Total	20	100	480.7	100	16	100	100

morphological traits have a weak influence on prey selection by males of *D. minutus*, further studies with a large sampling size are needed to elucidate this assumption.

Our observations in this study show that *D. minutus* males consumed five different prey categories, displaying a notable dominance for a substantial quantity of spiders. This prey item is usually found in the diet of *Dendropsophus* species (e.g., Macale *et al.*, 2008; Castro *et al.*, 2016; Sanches *et al.*, 2021), which might be associated with their sit-and-wait foraging strategy (Toft, 1980, 1981; Castro *et al.*, 2016). Although some studies indicate a generalist and opportunistic diet for *D. minutus* populations in different environments, the diversity of prey consumed ranges from three to ten categories (Van Sluys & Rocha, 1998; Santos *et al.*, 2004; Leivas *et al.*, 2018; Soeiro *et al.*, 2022). For populations of *D. minutus* from an Araucaria Forest in the state of Paraná, Southern Brazil, Leivas *et al.* (2018) suggest it is a generalist predator that feeds on arthropods, primarily those in the orders Araneae, Lepidoptera, and Diptera. This pattern was also observed for populations in the Serra Norte, Carajás, state of Pará, Brazilian Amazon (Van Sluys & Rocha, 1998), Tapacurá Ecological Station, a rainforest enclave of the state of Pernambuco (Santos *et al.*, 2004), and in an Atlantic Rainforest urban fragment of the state of Paraíba, Northeastern Brazil (Leite-Filho *et al.*, 2017). Despite spiders being a prevalent component in the dietary compositions of *D. minutus*, none of these studies implied a specific pattern of Araneae specialization diet. Therefore, we reinforce the importance of dietary studies aiming to investigate the role of spiders in the *Dendropsophus* diet.

Regarding prey size selection, there is a literature consensus that larger frogs tend to eat more

volumetric prey (Toft, 1980; 1981). We found no support for the hypothesis that larger frogs tend to eat larger prey, at least within an intraspecific view. The positive relationship between the size of frogs and the prey size consumed by them seems easier to observe when we compare different species (Van Sluys & Rocha, 1998; Parmelee, 1999). Concerning intraspecific views, this hypothesis was already supported for some populations, such as *Adelophryne maranguapensis* from a mountain in the Brazilian semiarid (Araújo *et al.*, 2023); *Leptodactylus ocellatus* and *Dendropsophus microcephalus* in El Banco, Colombia; and Maldonado, Uruguay, respectively (Maneyro *et al.*, 2004; Muñoz-Guerrero *et al.*, 2007). In contrast, this relationship remains an unsolved issue because other studies found no support for this hypothesis (e.g., Solé *et al.*, 2019; Sant'Anna *et al.*, 2022; Santos-Souza *et al.*, 2022). Additionally, Blanco-Torres *et al.* (2020) suggest that predators using active search target low-mobility prey, whereas species using sit-and-wait strategies target highly nutritive prey that are difficult to manipulate.

Overall, we observed spiders were one important prey item for the diet of *D. minutus* males in the region studied. It could be because these prey items are more abundant and readily available for frogs perched in vegetation, not necessarily because they actively seek them out (specialization). In any case, further studies considering temporal variation, reproductive period, and prey availability, among others, are still needed to elucidate the role of spiders on the diet of *D. minutus*.

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