

## Diet of three sympatric *Physalaemus* (Anura: Leptodactylidae) from the Brazilian semiarid region

Dieta de três *Physalaemus* simpáticos (Anura: Leptodactylidae) do semiárido brasileiro

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**Abstract:** Studies on the use of food resources and trophic niche of different species can help understand ecological relationships. In this study, we evaluated the diet of three sympatric species of *Physalaemus* from the Brazilian semiarid. We analyzed a total of 264 individuals, being 100 specimens of *Physalaemus albifrons*, 93 of *P. cicada*, and 71 of *P. cuvieri*. Seven prey categories were recorded: three for *P. cicada* and *P. albifrons*, and six distinct categories for *P. cuvieri*. The orders Coleoptera, Isoptera and the family Formicidae were the most representative categories in the diet of *P. cicada*, *P. cuvieri*, and *P. albifrons*, respectively. Although there is a high trophic niche overlap, differences in preference for food categories may reduce potential competition in these frog species from the Caatinga.

**Keywords:** Amphibia. Food niche. Arthropods. *Physalaemus albifrons*. *Physalaemus cicada*. *Physalaemus cuvieri*.

**Resumo:** Estudos sobre o uso de recursos alimentares e nicho trófico em diferentes espécies podem ajudar na compreensão das relações ecológicas. Aqui, avaliamos a dieta de três espécies simpáticas de *Physalaemus* do semiárido brasileiro. Analisamos um total de 264 indivíduos, sendo 100 espécimes de *Physalaemus albifrons*, 93 de *P. cicada* e 71 de *P. cuvieri*. Foram registradas sete categorias de presas: três para *P. cicada* e *P. albifrons*, e seis categorias distintas para *P. cuvieri*. As ordens Coleoptera, Isoptera e a família Formicidae foram as categorias mais representativas na dieta de *P. cicada*, *P. cuvieri* e *P. albifrons*, respectivamente. Embora haja uma elevada sobreposição de nichos tróficos, as diferenças na preferência das categorias alimentares podem reduzir a potencial competição nestas espécies de anuros na Caatinga.

**Palavras-chave:** Anfíbios. Nicho alimentar. Artrópodes. *Physalaemus albifrons*. *Physalaemus cicada*. *Physalaemus cuvieri*.

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## INTRODUCTION

The genus *Physalaemus* Fitzinger, 1826, currently comprises 43 species (Segalla et al., 2019), distributed from Mexico to southern South America (Nascimento et al., 2005; Grant et al., 2006; Frost, 2019). Species of this genus inhabiting the Atlantic Forest domain and the phytophysiognomies of the *Cerrado*, *Pantanal*, Amazonia, and *Caatinga* in Brazil (Lisboa & Haddad, 2009; Caldas et al., 2010; Frost, 2019), occupying a wide variety of microhabitats (Heyer et al., 1990).

Studies with *Physalaemus* species have been conducted regarding their taxonomy (Heyer & Wolf, 1989; Feio et al., 1999; Ron et al., 2005; Pimenta et al., 2005), phylogenetic and phylogeography (Lima, 2012). Biological aspects are known mainly from Atlantic Forest and *Cerrado* areas, and include reproduction (Brasileiro & Martins, 2006; Pupin et al., 2010), parasitology (Toledo et al., 2013; Campião et al., 2014; Aguiar et al., 2015; Leivas et al., 2018; C. Oliveira et al., 2019), and diet (Santana & Juncá, 2007; Moser et al., 2017; Leivas et al., 2018). From the Brazilian semiarid, ecological data, especially on diet, are scant.

Dietary studies are important for understanding trophic niche and ecological relationships (Bianchi, 2009; M. Oliveira et al., 2015). Several factors may influence the diet of anurans, including seasonal changes in food availability, competitive relationships, body size, and ecological tolerance of different species (Duellman & Trueb, 1994).

In this context, the use of different food resources can explain coexistence between species. Thus, our objective is to evaluate the diet of the sympatric species: *Physalaemus cicada* Bokermann, 1966, *Physalaemus cuvieri* Fitzinger, 1826, and *Physalaemus albifrons* Spix, 1824, from Brazilian semiarid.

## MATERIAL AND METHODS

The study was carried out in a *Caatinga* area in the municipality of Farias Brito ( $06^{\circ}51'55.7''S$ ;  $39^{\circ}32'08.0''W$ ), Ceará state, Brazil. The predominant climate of the region is tropical semiarid, with average rainfall of

896.5 mm (IPECE, 2015). The samplings were carried out from January to March, 2017, for two days a month.

Specimens were collected by hand in two temporary ponds during visual searches, which consist of walking and visually exploring all possible microhabitats (Bernarde, 2012). Specimens were euthanized following ethical procedures (CFMV, 2013) right after being captured, fixed in 10% formalin (Calleffo, 2002), and deposited at Coleção Herpetológica of Universidade Regional do Cariri (URCA-H).

A midventral incision was made in all specimens and the contents of the gastrointestinal tract were analyzed under a stereoscopic microscope. All prey items were counted and identified to the lowest possible taxonomic level using specialized literature (Oliveira-Costa, 2011). Fragments that could not be identified due to the high level of digestion were grouped into the category unidentified arthropods (UA).

The length and width of the intact items found in the stomachs were measured with the aid of a digital caliper. Their respective volumes were estimated through the ellipsoid formula:

$$V = \frac{4\pi}{3} \left( \frac{\text{length}}{2} \right) \left( \frac{\text{width}}{2} \right)^2$$

To determine the relative contribution of each prey category, the relative importance index was calculated using the following formula (Powell et al., 1990):

$$\text{RI} = \frac{F\% + N\% + V\%}{3}$$

To demonstrate the relative level of specialization in the diet, we calculate the amplitude of trophic niche, according to Levins' index (Krebs, 1998), which was standardized through the mathematical expression of Hurlbert (1978). To calculate  $P_j$ , we applied the volume values of each food item disregarding the value of unidentified Arthropods (UA). The food overlap was estimated according to Pianka index (Pianka, 1973) considering the RI values.



$$B = \frac{1}{\sum b_j^2}$$

$$B_A = \frac{B - 1}{n - 1}$$

We performed a Correspondence Analysis to evaluate prey categories that contributed most to the diet of each species, considering the RI values, disregarding the value of unidentified arthropods (UA), using the PAST 4.02 software.

## RESULTS

We analyzed 264 individuals of *Physalaemus*, being 100 specimens of *Physalaemus albifrons*, 93 of *P. cicada*, and 71 of *P. cuvieri* (243 males and 21 females in total, all adults). All specimens captured were in reproductive period. Only 10 individuals of *P. albifrons* (10%), 13 individuals of *P. cicada* (14%), and 15 individuals of *P. cuvieri* (21%) analyzed presented stomach contents (36 males and two females in total). The stomach contents identified were grouped into seven prey categories.

For *Physalaemus albifrons*, three prey categories were identified: Formicidae, Coleoptera and Dermaptera. The presence of plant material was also observed. Formicidae was the most frequent (45%) and important (42%) prey category. For *Physalaemus cicada* only Coleoptera was identified, representing 66.8% of the total volume found. *Physalaemus cuvieri* presented the highest prey diversity, with six prey categories identified.

Unidentified arthropods (UA) represented the largest volume (90%). Isoptera was the second most consumed category, with 32.7% of the number of prey ingested and Relative Importance Index (RI) (14.5%; Table 1).

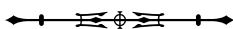
Formicidae, Coleoptera, and Isoptera contributed most to the diet of the three species. The correspondence analysis indicated that each species had a greater contribution from a specific category of prey (Figure 1). Overlap between *P. cicada* with *P. albifrons* was 0.67, and between *P. cicada* with *P. cuvieri* was 0.60, higher than overlap between *P. albifrons* and *P. cuvieri* 0.40. *Physalaemus cicada* presented narrow niche amplitude ( $B_A = 0.01$ ), than *P. albifrons* ( $B_A = 0.80$ ) and *P. cuvieri* ( $B_A = 0.25$ ).

## DISCUSSION

The mating system of anurans includes competition between males (Arak, 1983) or choice of females (Ryan, 1985), in which females are considered a limiting resource to the reproductive success of males (Robertson, 1986; Wogel & Pombal Jr., 2007). Duellman & Trueb (1994) stated that many anuran males could remain fasted during the reproductive period, which may explain the low number of specimens with stomach contents in our study. Thus, during the reproductive period males of *Physalaemus* tend to focus their energy expenditure on reproduction, reducing other activities, such as feeding.

Table 1. Prey items found in the diet of three sympatric *Physalaemus* in Brazilian semiarid. Legends: V% = volume; F% = frequency; N% = number; RI = relative importance index.

Prey items	<i>P. albifrons</i>				<i>P. cicada</i>				<i>P. cuvieri</i>			
	V%	F%	N%	RI	V%	F%	N%	RI	V%	F%	N%	RI
UA					32.9	47	32.4	37.4	90	55	39.4	61.5
Coleoptera	49.7	22.2	11.8	27.9	66.8	41.2	23.5	43.9	1.5	5	1	2.5
Dermaptera	23.2	22.2	11.8	19								
Formicidae	23.5	45.4	58.8	42.2					6.8	15	12.5	11.4
Isoptera									0.9	10	32.7	14.5
Arthropoda eggs					0.3	11.8	44.1	18.7	0.1	10	11.5	7.2
Scolopendromorpha									0.8	5	2.9	2.9



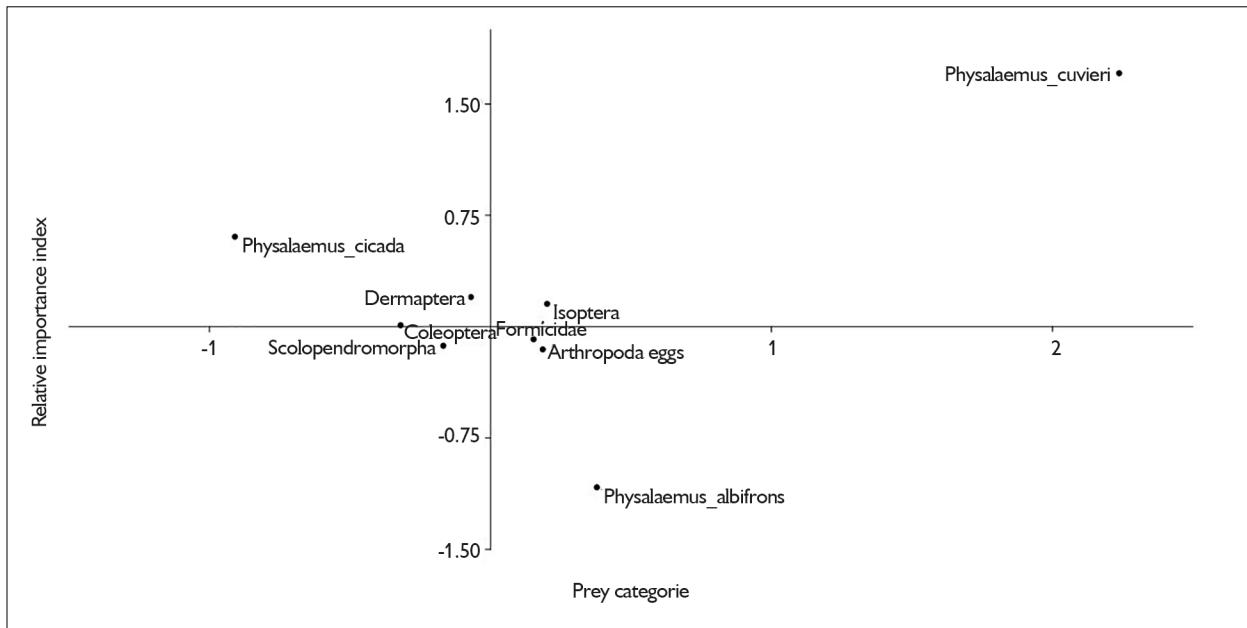


Figure 1. Correspondence analysis for prey categories in the diet of three sympatric *Physalaemus* in Brazilian semiarid.

Ants are considered of unpleasant taste and metabolically difficult to digest (Hirai & Marsui, 2000). However, they are frequently consumed by leiuperines, such as *Engystomops*, *Pleurodema* and *Physalaemus* (Narváez & Ron, 2013; Pincheira-Donoso, 2002; Sousa & Ávila, 2015; this study). Thus, the preference of Formicidae as the main food observed for *P. albifrons* and *P. cuvieri* is a well documented common pattern within the group, including for *P. cicada* (Santana & Juncá, 2007; M. Oliveira *et al.*, 2015; Leivas *et al.*, 2018). In a dietary study of *P. cicada* in the state of Bahia, Northeastern Brazil, Formicidae was the prey category with the highest volume and frequency (Santana & Juncá, 2007). In our study, *P. albifrons* consumed three prey categories, being Formicidae the most frequent (45%) and important (42%) prey, corroborating the study of Cruz (2000) from São Francisco River sand dunes in the state of Bahia. Moreover, consumption of ants by *P. cuvieri* and *P. albifrons* (present study; M. Oliveira *et al.*, 2015), *P. cuvieri* from Atlantic Forest (Santos *et al.*, 2004; Leivas *et al.*, 2018), *P. ephippiger* Steindachner, 1864,

from the Eastern Amazon (Rodrigues & Santos-Costa, 2014), and *P. albonotatus* Steindachner, 1864 (Falico *et al.*, 2012), and *P. riograndensis* Milstead, 1960 (López *et al.*, 2003), from Argentina, reinforces the importance of this prey category for this genus.

Foraging behavior reflects the availability of local resources (Leivas *et al.*, 2018). This way, individuals tend to have a high predation rate of organisms with terrestrial habits such as ants and termites (Toft, 1980). Moser *et al.* (2017), in their study of *Physalaemus lisei* Braun and Braun, 1977, stated that the availability of a given prey could impose opportunistic foraging behavior. The high amount of ants available in the environment enabled a higher comsumption. This can lead to an erroneous interpretation, confusing availability of the item with preference for this food item (Becker *et al.*, 2007).

Herein, the most consumed category for *P. cuvieri* was Isoptera (32.7%), whereas for *P. albifrons* was Formicidae (58.8%), and for *P. cicada* was Coleoptera (23.5%). Niche amplitude for *P. albifrons* (*BA* = 0.80)

and *P. cuvieri* ( $BA = 0.25$ ) were higher than *P. cicada* ( $BA = 0.01$ ). Also, overlap was higher between *P. cicada* and *P. albifrons* ( $Oik = 0.67$ ) and between *P. cicada* and *P. cuvieri* ( $Oik = 0.60$ ). These differences in diet may indicate distinct foraging tactics in the analyzed *Physalaemus* (M. Oliveira et al., 2015). Exploring different items decrease interspecific competition, which allows the coexistence of species in the same niche (Azevedo-Ramos et al., 1999; Hero et al., 2001).

Among the six categories consumed by *P. cuvieri*, Isoptera was the most representative arthropod order ( $RI = 14.5\%$ ), which is in accordance with the studies of Pinto (2011) and Leivas et al. (2018) from Atlantic Forest fragments. Leivas et al. (2018) states that trophic interactions for this species indicate low variation in the diet along its geographic distribution, feeding mainly on arthropods (Formicidae, Isoptera and Araneae). This seems to be a common pattern in the diet of *Physalaemus* species occurring in Brazil (Santos et al., 2004; M. Oliveira et al., 2015; Moser et al., 2017).

Santana & Juncá (2007) reported five prey categories for *P. cicada*, with Formicidae being the most representative. In our results, *P. cicada* consumed only Coleoptera ( $V = 66.8\%$  and  $RI = 43.9\%$ ). This dissimilarity may be result of its small size, which prevents competition with larger congeners in our study area, reducing the diversity of ingested items.

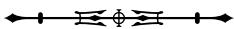
Despite difference between prey types for the anuran species in the present study, there is a considerable niche overlap. Although limited food resources may lead to competition, historical factors are relevant to the configuration of their current foraging tactics (M. Oliveira et al., 2015). Moreover, according to Bassar et al. (2017), the co-occurrence of species initiates a process of rapid evolutionary adaptation in both species, which may minimize potential competition. However, further studies on the food ecology of sympatric amphibians, especially congeners, are necessary to elucidate the mechanisms that allow coexistence.

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