# Feeding ecology of *Amazophrynella teko* (Anura: Bufonidae) in the eastern Brazilian Amazonia

Carla Juliana SANTOS-SOUZA <sup>1</sup>, Fillipe PEDROSO-SANTOS <sup>1</sup>, Karla REBELO-SILVA <sup>1</sup>, Maria Jeovana LIMA-MARTINS <sup>2</sup> and Carlos Eduardo COSTA-CAMPOS <sup>1, 3, \*</sup>

 Laboratório de Herpetologia, Departamento de Ciências Biológicas e da Saúde,
Universidade Federal do Amapá, Campus Marco Zero do Equador, 68903-419, Macapá, Amapá, Brazil.
Laboratório de Arthropoda, Departamento de Ciências Biológicas e da Saúde,
Universidade Federal do Amapá, Campus Marco Zero do Equador, Macapá, Amapá 68903-419, Brazil.
Programa de Pós-graduação em Biodiversidade e Biotecnologia da Rede Bionorte, Universidade Federal do Amapá Federal do Amapá, Macapá, Brazil.
\*Corresponding author: C. E. Costa-Campos, E-mail: dududueducampos@gmail.com

Received: 27 April 2023 / Accepted: 29 October 2023 / Available online: December 2023 / Printed: December 2023

**Abstract.** Diet is one of the most important dimensions of the trophic niche, contributing to understanding intra- and interspecific interactions in food webs. *Amazophrynella teko* was described in 2018, and given the scarcity of information about its feeding ecology, we describe here the diet of this species in a population from eastern Amazonia based on stomach flushing. We analyzed the stomach contents of 68 *A. teko* (18 juveniles, 13 females, and 37 males). We found 360 prey items from 23 prey categories from males (Shannon-Wiener index: H' = 2.36, N = 29), 17 for females (H' = 1.53, N = 12), and 14 for juveniles (H' = 2.35, N = 11). Formicidae were the most abundant prey items (284 prey items), Myrmicinae were the most abundant subfamily, and Crematogaster was the most abundant genus in all age/sex groups. Females had a narrower niche amplitude ( $B_A = 0.08$ ) than juveniles ( $B_A = 0.58$ ) and males ( $B_A = 0.21$ ). Trophic niche overlap between males and females ( $O_{jk} = 0.92$ ), and between males and juveniles ( $D_{jk} = 0.77$ ), higher than the overlap between females and juveniles ( $O_{jk} = 0.55$ ). The prey category accumulation curve, in relation to the number of stomachs flushed, did not reach an asymptote. The relationship between the SVL and JW of the toads with the volume of prey was not significant for males, females, and juveniles. The larger jaw width of females observed in our study may be related to the greater volume of prey they consumed. There were no significant differences among the diets for males, females, and juveniles based on the volumetric composition of prey. *Amazophrynella teko* feeds largely on ants but opportunistically will forage on other invertebrates such as spiders, beetles, and mites. The present study provides relevant information about the feeding ecology of *A. teko*, a poorly known species from north Brazil, eastern Brazilian Amazonia.

Keywords: diet, food items, natural history, foraging, trophic niche.

### Introduction

Diet is one of the most important dimensions of the trophic niche of species, contributing to understanding intra- and interspecific interactions in food webs (Toft 1985, Huckembeck et al. 2014). Bufonid frogs are usually considered generalists (Duellman & Trueb 1994), although some studies report dietary specialization (Solé et al. 2017, Núñez et al. 2021). This variation in diet may be caused by differences in habitat-prey composition or prey-selection behaviors (Duellman & Trueb 1994) and differences in size and type of prey related to the foraging mode (Toft 1981). Although the feeding ecology of several anuran species has been identified as paramount to unveil their natural history and the dynamics of food webs (e.g., Baía et al. 2020, Brandão et al. 2020, Plaza et al. 2021), basic feeding ecology data remain limited and have not been collected for anurans species widely distributed in the Neotropical region.

One such example is *Amazophrynella teko* Rojas-Zamora, Fouquet, Ron, Hernández-Ruz, Melo-Sampaio, Chaparro, Vogt, Carvalho, Pinheiro, Ávila, Farias, Gordo & Hrbek, 2018, a bufonid frog widely distributed throughout Amazonia. It is a small-sized toad with conspicuous sexual dimorphism, with males much smaller than females (Rojas et al. 2018). This species inhabits the forest leaf litter, breeds in seasonal pools, and has diurnal and crepuscular habits (Rojas et al. 2018).

Amazophrynella teko was described in 2018, and given the scarcity of information about its feeding ecology, the diet of this species in a population from eastern Amazonia is described herein. The specific aims of this study were to

(1) identify the most important prey items present in the diet of males, females, and juveniles of *A. teko*; (2) analyze the niche breadth and the level of niche overlap between males, females, and juveniles; (3) test significance of the changes in prey volume with the Snout vent-length, jaw width and sex (male, female and juvenile); (4) verify differences in the volumetric composition of prey, and the number of prey consumed, among sexes (males, females and juveniles).

#### Material and methods

#### Study site

The study was carried in two conservation units at Amapá state, Brazil: (1) Parque Natural Municipal do Cancão (0.9008°N, 52.0134°W; datum WGS84), municipality of Serra do Navio, and (2) Reserva Extrativista Municipal Beija-Flor Brilho de Fogo (0.7918°N, 51.9783°W), municipality of Pedra Branca do Amapari. All sites were mainly *terra firme* forests, characterized by a forest with seasonal flooding, also within the Equatorial (Am) climate domain according to the Köppen-Geiger classification, with mean air temperatures > 27.0 °C and annual rainfall of 2,850 mm (Alvares et al. 2013).

### Data sampling

Individuals of *A. teko* were sampled in four diurnal surveys (March 2019, October 2019, June 2021, and September 2021) during rainy seasons. All samples had a duration of 5 consecutive days per month, with samples between 8:00 and 11:00 h. The sampling effort was carried out by three people each day for a total sampling time of 180 effective hours. We located the toads through active search (Crump & Scott Jr. 1994) and captured them manually. Following capture, we applied stomach-flushing (Solé et al. 2005) to obtain the stomach contents, which were preserved in 70% ethanol. To prevent recapture,

all captured individuals were kept in moist plastic bags until the end of sampling and were then released. Individuals were classified as juveniles if they had no obvious sexually dimorphic characters and SVL less than the minimum size of adult males, 12.9–15.8 mm, following Rojas et al. 2018). Adults were sexed by examining the external morphology (presence of vocal sacs in males and readily visible eggs through the ventral skin of females). We measured the snout-vent length (SVL) and the jaw width (JW) of each specimen using a digital caliper (0.1 mm precision) and used the Student t-test to evaluate the relationship only between JW of males and females.

## Dietary analysis

Prey items were counted and identified to the lowest possible taxonomic level under a Zeiss Stemi SV 11 stereomicroscope, following the identification keys of Rafael et al. (2012) for insect orders and Baccaro et al. (2015) for subfamilies and genera of Formicidae. The length and width of each prey item were measured with calipers (0.1 mm precision), and the volume was estimated using the ellipsoid formula (Dunham 1983):  $V = (4\pi/3)(length/2)(width/2)^2$ . The index of relative importance was estimated using the formula *IRI* = (N% + % V%)F%, in which N = numerical percentage, V = volumetric percentage, F = frequency of occurrence percentage of each prey category in relation to the total samples (see Pinkas et al. 1971).

Food niche breadth was calculated using the formula proposed by Levins (1968)  $B = 1/\Sigma pi^2$ , where B = niche breadth and pi =proportion of item *i* in the diet. The values of niche breadth were standardized ( $B_A$ ) using the formula  $B_A = (B - 1)/(n - 1)$ , where  $B_A =$ standardized Levins index and n = number of possible resources. Values close to zero are attributed to a specialist diet, while values close to one are attributed to a generalist diet. The food niche breadth was analyzed with the software Ecological Methodology (Krebs 2014).

To analyze if the trophic niches overlap between males, females, and juveniles, with regards to the degree of similarity between their diets, we used the Trophic Niche Overlap Index of Pianka ( $O_{jk}$ ) (Pianka 1974):  $O_{jk} = \sum P_{ij} P_{ik} / \sqrt{\sum P_{ij}^2} \sum P_{ik}^2$ , where  $O_{jk}$  is the niche overlap index between the species j and k;  $P_{ij}$  is equivalent to the proportion of the resource type i relative to the total of resources used by the species j;  $P_{ik}$  is the proportion of resource i relative to the total of resource categories used by the species j; and n is the total number of resource categories used by the species j and k. The index ranges from 0 to 1 when there is no overlap or a complete overlap between the species' diets, respectively (Krebs 2014). We used the niche overlap module from the EcoSim program (version 7.0) (Gotelli & Entsminger 2005) with 1,000 randomizations.

To estimate the prey category richness in the diet, we made rarefaction curves based on samples with the software EstimateS 8.2.0 (Colwell 2009), with 1,000 random permutations. In this analysis, stomachs were treated as samples, and prey types were richness categories (curves were made for each sex). In addition, we used the Shannon Diversity Index to quantify diet diversity  $H' = -\sum pi \ln pi$ , in which pi is the numeric proportion of prey i in the total number of individuals of n prey types.

We also evaluated the relationship between snout-vent length (SVL) and the jaw width of each specimen (juvenile, female, and male) with the volume of the prey consumed through Spearman's correlation. We transformed variables to their natural log when necessary to meet assumptions of normality (Zar 2010). The significance of the changes in prey volume was tested using a General Linear Mixed Model (GLMM), with the SVL and JW as independent quantitative variables; sex (male, female, and juvenile) as an independent categorical variable; and collection period (March 2019, October 2019, June 2021 and September 2021) as a random effect variable. We inspected model residuals to confirm normal distribution and homoscedasticity to fit the linear model. Only the response variable was log-transformed to meet the assumption of normal distribution. We included only JW as a response variable because SVL, JW, and weight were highly correlated (Pearson R > 0.7).

We performed a permutational multivariate analysis of variance (PERMANOVA) through Euclidean distance (Anderson 2001) to test for differences in the volumetric composition of prey among sexes (males, females, and juveniles), using order-level taxonomic resolution, except for Formicidae, which was classified by genus. In addition, we evaluated whether the number of prey consumed differed among sexes through Mann-Whitney tests. Pairwise PERMANOVA was performed with the software R (R Core Team 2020), using the "vegan" (Oksanen et al. 2015) and "PairwiseAdonis" (Martinez Arbizu 2020) packages, and the Mann-Whitney tests were performed with the "car" package (Fox & Weisberg 2019).

# Results

We analyzed the stomach contents of 68 *A. teko* (18 juveniles, 13 females, and 37 males). Of the 68 toads analyzed, 58.8% (n = 40) were captured in March 2019, 2.9% (n = 3) in October 2019, 13.2% (n = 9) in June 2021, and 23.5% (n = 16) in September 2021 (Fig. 1). Of the 68 analyzed individuals, 16 (23.5%) had empty stomachs (8 males, 1 female and 7 juveniles), while 52 (76.5%) contained stomach contents (29 males, 12 females and 11 juveniles).

We found 360 prey items from 23 prey categories for males, 17 for females, and 14 for juveniles (Table 1). The number of prey items per stomach varied from 1–63 items (mean =  $7.3 \pm 12.9$ , for males), 1–94 items (mean =  $8.5 \pm 22.2$ , for females), and 1–8 items (mean =  $3.4 \pm 2.7$ , for juveniles). The most important category in the diet was Formicidae (IRI = 76.7%), followed by Araneae (IRI = 7.8%) (Fig. 2). Formicidae (284 items of 5 subfamilies – Ectatomminae, Formicinae, Myrmicinae, Paraponerinae and Ponerinae, and 17 genera) was the most abundant for males (121 prey items), females (133) and juveniles (30 prey items), representing 78.9% of the stomach contents (see Table 2). Myrmicinae was the most abundant subfamily, and *Crematogaster* was the most abundant genus in all age/sex groups (Fig. 3).

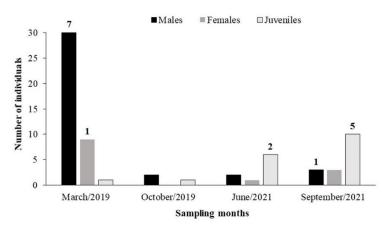
Females had a narrower niche amplitude ( $B_A = 0.08$ ) than juveniles ( $B_A = 0.58$ ) and males ( $B_A = 0.21$ ). Trophic niche overlap between males and females was 0.92, and between males and juveniles was 0.77, higher than between females and juveniles (0.55). The prey category accumulation curve, in relation to the number of stomachs flushed, did not reach an asymptote and indicated a higher taxonomic richness in the diet of males (Fig. 4). The number of prey categories of males (23 prey types) and relative abundance of prey categories (Shannon-Wiener index: H' = 2.36) of males and juveniles (14 prey types, H' = 2.35) was similar and more diverse than females (17 prey types, H' = 1.53).

The relationship between the SVL and JW of the toads with the volume of prey was not significant for males (SVL: *rs* = 0.05, p = 0.79, n = 29; JW: *rs* = 0.09, p = 0.62, n =29), females (SVL: *rs* = 0.32, p = 0.31, n =12) and juveniles (SVL: *rs* = 0.11, p = 0.75, n = 11; JW: *rs* = 0.06, p = 0.86, n = 11), but there was a statistically significant relationship between jaw width and volume of prey for females (*rs* = 0.61, p = 0.04, n = 12). The JW of females was greater than males (*t* = -4.93, p < 0.001). Prey volume values varied significantly according to the JW (Mixed model:  $F_{2.46}$  = 4.70; p = 0.03) but not according to the sex of individuals (Mixed model:  $F_{2.46}$  = 0.25; p = 0.70). No significant interaction effect was observed between the JW and males, females, and juveniles (Mixed model:  $F_{2.46}$  = 0.31; p = 0.73) on prey volume.

There were no significant differences among the diets for

the volumetric composition of prey between males and females (PERMANOVA: F = 1.95, p = 0.21), males and juveniles (F = 0.820, p = 1.00), and females with juveniles (F = 1.34, p = 0.59). Likewise, considering only Formicidae, there are no significant differences among the diets between males and females (F = 1.51, p = 0.17), males and juveniles (F = 1.85, p = 0.20), and females and juveniles (F = 0.95, p = 0.54).

Regarding the number of prey consumed, we did not find significant variations between males and females (Mann-Whitney test: U = 157, p = 0.63), males and juveniles (U = 159, p = 0.50), and females and juveniles (U = 57, p = 0.58). The average number of prey items consumed by males was 5.8 (SD = 8.3, range: 1–45), by females was 12.1 (SD = 21.7, range: 1–78), and by juveniles was 4.3 (SD = 2.7, range: 1–9).



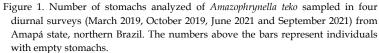


Table 1. Food items found in the stomach of 52 specimens (males, females and juveniles) of *Amazophrynella teko* collected in two conservation units at Amapá state, North of Brazil. N = number of items consumed, N% = numerical percentage of items, F = frequency of items consumed, F% = frequency of occurrence, V = volume of items consumed, V% = numerical percentage of items, IRI = index of relative importance for each category of items in the diet, (L) larvae.

Prey	Males (N = 29)				Females (N =12)				Juveniles (N = 11)				
	N (%)	F (%)	V (%)	IRI	N (%)	F (%)	V (%)	IRI	N (%)	F (%)	V (%)	IRI	
Acari	4 (2.38)	4 (5.63)	0.52 (0.61)	2.88	2 (1.38)	1 (3.33)	0.33 (0.37)	1.69	1 (2.13)	1 (4.17)	0.02 (0.04)	2.11	
Araneae	17 (10.12)	10 (14.08)	2.44 (2.87)	9.02	5 (3.45)	4 (13.33)	1.91 (2.12)	6.30	6 (10.77)	3 (12.50)	0.83 (1.61)	8.96	
Coleoptera	7 (4.17)	4 (5.63)	7.61 (8.94)	6.25	-	-	-	-	4 (8.51)	3 (12.50)	1.06 (2.05)	7.69	
Collembola	3 (1.79)	3 (4.23)	0.20 (0.23)	2.08	1 (0.69)	1 (3.33)	0.01 (0.01)	1.34	1 (2.13)	1 (4.17)	0.01 (0.02)	2.10	
Diplopoda	5 (2.98)	1 (1.41)	1.03 (1.21)	1.86	2 (1.38)	1 (3.33)	2.33 (2.59)	2.43	-	-	-	-	
Diptera	1 (0.60)	1 (1.41)	0.19 (0.22)	0.74	-	-	-	-	1 (2.13)	1 (4.17)	0.04 (0.08)	2.12	
Formicidae	121 (72.0)	39 (54.9)	72.0 (84.5)	70.5	133 (91.7)	21 (70.0)	85.5 (94.80)	85.5	30 (63.80)	12 (50.0)	49.35 (95.62)	69.82	
Hemiptera	1 (0.60)	1 (1.41)	0.16 (0.19)	0.73	-	-	-	-	1 (2.13)	1 (4.17)	0.24 (0.47)	2.25	
Hymenoptera (L)	1 (0.60)	1 (1.41)	0.31 (0.36)	0.79	-	-	-	-	-	-	-	-	
Ixodida	6 (3.57)	5 (7.04)	0.49 (0.58)	3.73	1 (0.69)	1 (3.33)	0.06 (0.07)	1.36	3 (6.38)	2 (8.33)	0.06 (0.12)	4.94	
Psocoptera	1 (0.60)	1 (1.41)	0.10 (0.12)	0.71	1 (0.69)	1 (3.33)	0.01 (0.01)	1.34	-	-	-	-	
Siphonaptera	1 (0.60)	1 (1.41)	0.15 (0.18)	0.73	-	-	_	-	-	-	-	-	

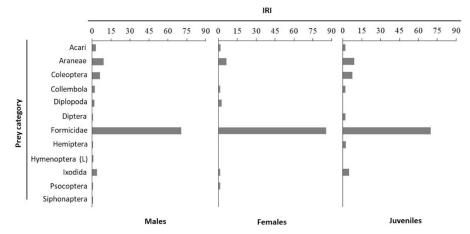


Figure 2. Diet of *Amazophrynella teko* from Amapá state, northern Brazil. Bars represent the Index of Relative Importance (IRI).

Table 2. Genera and subfamilies of Formicidae found in the stomach of 52 specimens (males, females, and juveniles) of *Amazophrynella teko* collected at Amapá state, northern Brazil. N = number of items consumed, N% = numerical percentage of items, F = frequency of items consumed, F% = frequency of occurrence, V = volume of items consumed, V% = numerical percentage of items, IRI = index of relative importance for each category of items in the diet, (n.i) = non-identified Myrmicinae.

		Males (	N = 29)	Females (N = 12)				Juveniles (N = 11)				
Prey	N (%)	F (%)	V (%)	IRI	N (%)	F (%)	V (%)	IRI	N (%)	F (%)	V (%)	IRI
	Formicinae											
Camponotus	-	-	-	-	2 (1.50)	1 (4.76)	1.15 (1.35)	2.54	-	-	-	-
Nylanderia	5 (4.13)	4 (10.26)	1.69 (2.35)	5.58	-	-	-	-	2 (6.67)	2 (16.67)	0.65 (1.32)	8.22
	Ectatomminae											
Gnamptogenys	-	-	-	-	1 (0.75)	1 (4.76)	5.41 (6.33)	3.95	2 (6.67)	1 (8.33)	32.08 (65.01)	26.67
	Myrmicinae											
Apterostigma	-	-	-	-	2 (1.50)	1 (4.76)	3.63 (4.25)	3.50	-	-	-	-
Blepharidatta	8 (6.61)	4 (10.26)	5.19 (7.21)	8.03	3 (2.26)	1 (4.76)	0.51 (0.60)	2.54	-	-	-	-
Carebara	6 (4.96)	2 (5.13)	0.64 (0.89)	3.66	-	-	-	-	-	-	-	-
Crematogaster	63 (52.07)	10 (25.64)	36.57 (50.82)	42.84	94 (70.68)	5 (23.81)	24.55 (28.72)	41.07	8 (26.67)	2 (16.67)	4.95 (10.03)	17.79
Cyphomyrmex	1 (0.83)	1 (2.56)	2.31 (3.21)	2.20	-	-	-	-	-	-	-	-
Myrmicinae (n.i.)	5 (4.13)	4 (10.26)	4.37 (6.07)	6.82	6 (4.51)	4 (19.05)	11.28 (13.20)	12.25	-	-	-	-
Myrmicocrypta	2 (1.65)	1 (2.56)	3.78 (5.25)	3.16	-	-	-	-	-	-	-	-
Pheidole	11 (9.09)	4 (10.26)	2.56 (3.56)	7.63	4 (3.01)	2 (9.52)	2.20 (2.57)	5.04	8 (26.67)	3 (25.00)	3.07 (6.22)	19.30
Solenopsis	14 (11.57)	5 (12.82)	1.38 (1.92)	8.77	-	-	-	-	7 (23.33)	2 (16.67)	2.28 (4.62)	14.87
Strumigenys	1 (0.83)	1 (2.56)	0.04 (0.06)	1.15	4 (3.01)	2 (9.52)	0.69 (0.81)	4.45	2 (6.67)	1 (8.33)	0.42 (0.85)	5.28
Trachymyrmex	3 (2.48)	2 (5.13)	11.10 (15.43)	7.68	9 (6.77)	2 (9.52)	27.26 (31.89)	16.06	1 (3.33)	1 (8.33)	5.90 (11.96)	7.87
						Parapo	onerinae					
Paraponera	-	-	-	-	7 (5.26)	1 (4.76)	5.25 (6.14)	5.39	-	-	-	-
						Pon	erinae					
Anochetus	-	-	-	-	1 (0.75)	1 (4.76)	1.15 (1.35)	2.54	-	-	-	-
Hypoponera	2 (1.65)	1 (2.56)	2.33 (3.24)	2.48	-	-	-	-	-	-	-	-
Total	121	39	71.96	100	133	21	85.47	100	30	12	49.35	100

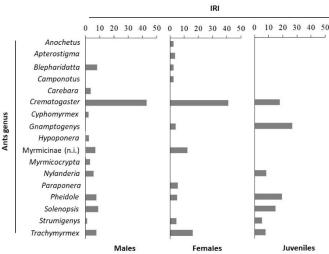
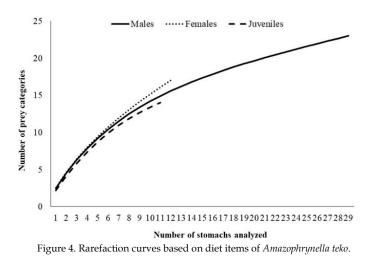


Figure 3. Genera of Formicidae found in the stomach of of *Amazophrynella teko* collected in Amapá state, northern Brazil. Bars represent the Index of Relative Importance (IRI).



# Discussion

Many studies have shown that some bufonids species are antspecialists for different reasons. For instance, Clarke (1974) suggested that ants and coleopterans are frequently present in the diet of leaf litter species, such as bufonids, due to the availability and abundance of these prey in the soil. In addition, Vences et al. (1998) pointed out that bufonids may have phylogenetic constraints to eat ants because basal bufonid species would have been small myrmecophagous. In line with these statements, our results indicate that the bufonid A. teko, a species from the leaf litter, feeds on ants that can consume substantial prey (e.g., Araneae, Coleoptera, and Acari) to supply nutritional demands. The higher IRI values for beetles by juveniles of A. teko in this study may be related to energy demands due to this prey being highly chitinous and requiring long digestion, suggesting that the gradual supply of nutrients helps in the development/growth of juveniles. Indeed, Taveira et al. (2021) observed that individuals of A. manaos (a conspecific with A. teko; see Taucce et al. 2022) older than 15 mm had dietary restrictions (i.g., increased consumption of soft-bodied prey and decrease in chitinous prey).

The high incidence of Myrmicinae ants in the diet of A. teko is in accordance with what is recognized for species of Amazophrynella, such as A. bokermanni (Travassos 2003) and A. manaos (Taveira et al. 2021), as well as for other species of bufonids such as Rhinella abei (Moser et al. 2022), R. alata (McElroy & Donoso 2019), R. castaneotica (Figueiredo et al. 2022), R. crucifer and R. icterica (Sabagh & Carvalho-e-Silva 2008), R. major (Oliveira-Souza et al. 2022), R. ornata (Flynn et al. 2020) and R. proboscidea (Borges et al. 2019). Indeed, the Myrmicinae ants are the most numerous and frequently in the diet of bufonids (Oliveira-Souza et al. 2022) due to the higher diversity of genera and habitats of these ants in the environment (e.g., leaf litter and arboreal habitats) (Fernández 2003). Furthermore, the higher abundance of arboreal ants that forage in leaf litter (e.g., Crematogaster) in the diet of A. teko suggested that this species displays climbing behavior on understory vegetation. Such behavior has been noted for small-sized bufonid species, such as R. castaneotica, a sympatric species with A. teko in our study area (Oliveira-Souza et al. 2020, Costa-Campos & Pedroso-Santos 2022, Figueiredo et al. 2022), but this behavior has not yet been reported for species of Amazophrynella. Therefore, further field observations are needed to support our hypothesis.

The niche breadth of *A. teko*, considered narrow in our study, suggests a specialist feeding behavior. This condition is common in small-sized tropical bufonids with no teeth, such as *A. teko* (Isacch & Barg 2002). In addition, the positive relationship between morphological characteristics (e.g., snout-vent length and jaw width) and the volume of prey consumed by toads is more common in generalist species, which consume fewer but larger prey, than ant-specialist species that feed on larger proportions but on small-sized prey (Lima & Magnusson 2000). Therefore, the largest jaw width of females observed in our study may be related to the greater volume of prey they consumed. Taveira et al. (2021) noted the same pattern for *A. manaos* but did not present data on these relationships. The relationship is poorly described for leaf-litter anuran species (see *Adenomera marmorata;* 

# Santos-Pereira et al. 2015).

We conclude that A. teko feeds largely on ants but opportunistically will forage on other invertebrates such as spiders, beetles, and mites. Spiders and mites may be important items in the diet of small species that forage in the leaf litter (Lima & Magnusson 2000, Santos-Pereira et al. 2015, Moser et al. 2022). Beetles are often reported as an important prey in the diet of bufonids (De Oliveira et al. 2017, Oliveira-Souza et al. 2022), and due to the high species richness of this group, as well as their abundance in the environment, are easy to find and thus consumed by several species (Moser et al. 2022). Our study provides relevant information about the feeding ecology of A. teko, a poorly known species of north Brazil, eastern Brazilian Amazonia. Thus, further studies are needed to disentangle these observations in ground-dwelling and leaf-litter anurans so that we can learn more about aspects related to the natural history of this species.

#### Acknowledgments

We thank Aline Oliveira-Souza, Jessica Anaissi, and Fernanda Melo for their field assistance. Instituto Chico Mendes de Conservação da Biodiversidade by the collection permits (ICMBio/SISBIO #48102-4). We thank Yurii Kornilev and the anonymous reviewers for their valuable comments on a previous manuscript draft. Carlos Eduardo Costa-Campos thanks the Conselho Nacional de Desenvolvimento Científico e Tecnológico – CNPq for the research grant (Proc. 307697/2022-3).

## References

- Alvares, C.A., Stape, J.L., Sentelhas, P.C., Gonçalves, J.L.M., Sparovek, G. (2013): Köppen's climate classification map for Brazil. Meteorologische Zeitschrift 22: 711–728.
- Anderson, M.J. (2001): A new method for non-parametric multivariate analysis of variance. Austral Ecology 26(1): 32–46.
- Baccaro, F.B., Feitosa, R.M., Fernandez, F., Fernandes, I.O., Izzo, T.J., Souza, J.L.P., Solar, R. (2015): Guia para os Gêneros de Formigas do Brasil. Manaus, Editora INPA.
- Baía, R.R.J., Sanches, P.R., Pedroso-Santos, F., Florentino, A.C., Costa-Campos, C.E. (2020): Diet overlap of three sympatric species of *Leptodactylus* Fitzinger (Anura: Leptodactylidae) in a protected area in the Brazilian Amazon. Cuadernos de Herpetología 34: 175-184.
- Borges, A.C.R., Santos, T.F., Frazão, L., Marques-Souza, S., Menin, M. (2019): Food habits of *Rhinella proboscidea* (Anura: Bufonidae) in terra firme forests of central Amazonia. Phyllomedusa: Journal of Herpetology 18: 37–46.
- Brandão, R.A., Fenker, J., Lopes, B.E.P.C., Sena, V.M.A., Vasconcelos, B.D. (2020): Diet of terrestrial anurans in an ephemeral and simplified habitat during the dry season in the Brazilian Cerrado. Ethology Ecology & Evolution 32(6): 1– 24.
- Clarke, R.D. (1974): Food habits of toads, genus *Bufo*. American Midland Naturalist 91: 140-147.
- Colwell, R.K. (2009): EstimateS: Statistical estimation of species richness and shared species from samples. Version 8.2.0. < Available from: http://viceroy.eeb.unconn.edu/estimates, accessed on 18.01.2023>
- Costa-Campos, C.E., Pedroso-Santos, F. (2022): Antipredator behaviour employed by *Rhinella castaneotica* (Bufonidae) during climbing behaviour. Revista Latinoamericana de Herpetología 5(3): 9–11.
- Crump, M.L., Scott Jr., N.J. (1994): Standard techniques for inventory and monitoring. pp. 84-92. In: Heyer, W.R., Donnelly, M.A., Mc Diarmid, R.W., Hayek, L.C., Foster, M.S. (eds.), Measuring and monitoring biological diversity: standard methods for amphibians. Washington and London, Smithsonian Institution Press.
- De Oliveira, M., Avila, F.R. de, Tozetti, A.M. (2017): Diet of *Rhinella arenarum* (Anura, Bufonidae) in a coastal habitat in southern Brazil. Herpetology Notes 10: 507–510.
- Duellman, W.E., Trueb, L. (1994): Biology of Amphibians. The Johns Hopkins University Press, Baltimore.
- Dunham, A.E. (1983): Realized niche overlap, resource abundance, and intensity

of interspecific competition. pp. 261-280. In: Pianka, R.B., Schoener, T. (eds.), Lizard Ecology: studies of a model organism. Harvard University Press.

- Fernández, F. (2003): Introducción a las hormigas de la región Neotropical. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, Bogotá.
- Figueiredo, V.A.M.B., Santos, E.M., Pedroso-Santos, F., Sanches, P.R., Costa-Campos, C.E. (2022): Diet and clutch size of *Rhinella castaneotica* (Anura: Bufonidae) from a forest area in Serra do Navio. North-Western Journal of Zoology 18: 35–39.
- Flynn, C.N, Araújo, P.G., Rocha, C.F.D. (2020): Diet and microhabitat use by juveniles *Rhinella ornata* (Anura, Bufonidae) in an insular Brazilian Atlantic Rainforest area. Brazilian Journal of Biology 23: 1–3.
- Fox, J., Weisberg, S. (2019): An R Companion to Applied Regression. Third edition. Sage, Thousand Oaks CA.
- Gotelli, N.J., Entsminger, G.L. (2005): EcoSim: Null Models Software for Ecology. Version 7.72. <Available from: http://together.net/~gentsmin/ecosim.htm, accessed on 20.09.2022>
- Huckembeck, S., Loebmann, D., Albertoni, E.F., Hefler, S.M., Oliveira, M.C., Garcia, A.M. (2014): Feeding ecology and basal food sources that sustain the Paradoxal frog *Pseudis minuta*: a multiple approach combining stomach content, prey availability, and stable isotopes. Hydrobiologia 740: 253–264.
- Isacch, J.P., Barg, M. (2002): Are bufonids toads specialized ant-feeders? A case test from the Argentinian fooding pampa. Journal of Natural History 36: 2005–2012.
- Krebs, C.J. (2014): Ecological methodology. University of British Columbia, Vancouver.
- Levins, R. (1968): Evolution in changing environments: some theoretical explorations. Princeton University Press, Princeton.
- Lima, A.P., Magnusson, W.E. (2000): Does foraging activity change with the ontogeny? An assessment for six sympatric species of postmetamorphic litter anurans in Central Amazonia. Journal of Herpetology 34: 192–200.
- Martinez Arbizu, P.R. (2020): Package Version 0.4: PairwiseAdonis: pairwise multilevel comparison using adonis. <Available from: https://github.com/pmartinezarbizu/pairwiseAdonis, accessed on 20.03.2023>
- McElroy, M.T., Donoso, D.A. (2019): Ant morphology mediates diet preference in a Neotropical toad (*Rhinella alata*). Copeia 107(3): 430–438.
- Moser, C.F., Farina, R.K., Dudczak, A.C., Tozetti, A.M., Lignau, R. (2022): Feeding ecology of endemic frogs of the Atlantic Forest in southern Brazil. Anais da Academia Brasileira de Ciências 94(2): e20210282.
- Núñez, K., Duré, M., Zárate, G., Ortiz, F., Mendoza, M. (2021): Diet of *Melanophryniscus paraguayensis* (Anura: Bufonidae): an endemic species to Paraguay. Herpetological Conservation and Biology 16(2): 251–258.
- Oksanen, J.F., Blanchet, G., Kindt, R., Legendre, P., Minchin, P.R., O'hara, R.B., Wagner, H. (2015): Vegan: community ecology, R package. < Available from: http://CRAN.R-project.org/package=vegan, accessed on 20.03.2023>
- Oliveira-Souza, A.E., Brito, T.M., Santana, M.M.S., Reis, T.S., Soeiro, A.L.S., Costa-Campos, C.E. (2020): Atelopus hoogmoedi (Hoogmoed Harlequin Frog), Rhinella castaneotica (Pará Toad), Rhinella lescurei and Ameerega pulchripecta (Silverstone's Poison Frog). Climbing Behavior. Herpetological Review 51(4): 813.
- Oliveira-Souza, A.E., Santana, M.M.S., Martins, M.J.L., Anaissi, J.S.C., Sanches, P.R., Costa-Campos, C.E. (2022): Diversity of ants in the diet of *Rhinella major* (Anura: Bufonidae) in an urban area in North Brazil. Herpetology Notes 15: 663–670.

- Pianka, E.R. (1974): Niche overlap and diffuse competition. Proceedings of the National Academy of Sciences of the United States of America 71(5): 2141– 2145.
- Pinkas, L., Oliphant, M.S., Iverson, I.L.K. (1971): Food habits of albacore, bluefin tuna and bonito in Californian waters. California Department of Fish and Game: Fish Bulletin 152: 1–105.
- Plaza, J.L., Metillo, E.B., Sanguila, M.B. (2021): Trophic ecology of syntopic anurans of tropical stream communities. Journal of Tropical Ecology 37: 109– 117.
- R Development Core Team (2020): A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <Available from: https://www.R-project.org/, accessed on 20.03.2023>
- Rafael, J.A., Melo, G., Carvalho, C., Casari, C.A., Constantino, R. (2012): Insetos do Brasil: Diversidade e Taxonomia. Ribeirão Preto, Brazil, Holos.
- Rojas, R.R., Fouquet, A., Ron, S.R., Hernández-Ruz, E.J., Melo-Sampaio, P.R., Chaparro, J.C., Vogt, R.C., Carvalho, V.T., Pinheiro, L.C., Avila, R.W., Farias, I.P., Gordo, M., Hrbek, T. (2018): A Pan-Amazonian species delimitation: high species diversity within the genus *Amazophrynella* (Anura: Bufonidae) PeerJ 6: e4941.
- Sabagh, L.T., Carvalho-e-Silva, A.M.P.T. (2008): Feeding overlap in two sympatric species of *Rhinella* (Anura: Bufonidae) of the Atlantic Rain Forest. Revista Brasileira de Zoologia 25: 247–253.
- Santos-Pereira, M., Almeida-Santos, M., Oliveira, F.B., Silva, A.L., Rocha, C.F.D. (2015): Living in a same microhabitat should means eating the same food? Diet and trophic niche of sympatric leaf-litter frogs *Ischnocnema henselii* and *Adenomera marmorata* in a forest of Southern Brazil. Brazilian Journal of Biology 75: 13–18.
- Solé, M., Beckmann, O., Pelz, B., Kwet, A., Engels, W. (2005): Stomach-flushing for diet analysis in anurans: an improved protocol evaluated in a case study in Araucaria forests, southern Brazil. Studies on Neotropical Fauna and Environment 40: 23–28.
- Solé, M., Rocha, M.S., Decarli, C., Santos, C.R., Pereira, C.K. (2017): Diet of postmetamorphic *Rhinella icterica* (Spix, 1824) from the Araucaria Plateau of Rio Grande do Sul, Brazil (Anura: Bufonidae), Herpetology Notes 10: 443–448.
- Taucce, P.G., Costa-Campos, C.E., Carvalho, T.R., Michalski, F. (2022): Anurans (Amphibia: Anura) of the Brazilian state of Amapá, eastern Amazonia: species diversity and knowledge gaps. European Journal of Taxonomy 836: 96–130.
- Taveira, R., Barros, A.L., Silva-Forsberg, M.C. (2021): Variação temporal da dieta de Amazophrynella manaos (Anura: Bufonidae) em Manaus, Amazonas, Brasil. pp. 90-104. In: Silva-Forsberg, M.C., Magalhães, L.C.S., Aguiar, D.P.P., Rebêlo, G.H. (eds.), Biodiversidade, invasões biológicas, educação científica e relações com o entorno do Parque Estadual Sumaúma. Manaus, Editora UEA.
- Toft, C.A. (1981): Feeding ecology of Panamanian litter anurans: Patterns in diet and foraging mode. Journal of Herpetology 15: 139-144.
- Toft, C.A. (1985): Evolution of diet specialization in poison-dart frogs (Dentrobatidae). Herpetologica 51: 202–216.
- Travassos, A.E.M. (2003): Biologia reprodutiva e hábito alimentar de Dendrophryniscus minutus (Melin, 1941) (Amphibia: Bufonidae) na Floresta Nacional de Caxiuanã, Pará. Dissertação de Mestrado, Universidade Federal do Pará, Museu Paraense Emílio Goeldi.
- Vences, M., Glaw, F., Böhme, W. (1998): Evolutionary correlates of microphagy in alkaloid-containing frogs (Amphibia: Anura). Zoologischer Anzeiger 236: 217–230.
- Zar, J.H. (2010): Biostatistical analysis. 5th ed. Upper Saddle River, Prentice Hall.