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Diet and fecundity of *Physalaemus crombiei* (Anura: Leptodactylidae) from a sandy coastal plain of southeastern Brazil

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Abstract. Many ecological aspects of *Physalaemus crombiei* remain unstudied. Thus, the present study describes the diet and fecundity in a population of *P. crombiei* from southeastern Brazil. We analyzed the diet of 38 individuals of *P. crombiei* (25 males and 13 females). Sex ratio at reproductive habitat was slightly biased toward males. Females had greater size and mass compared to males. The diet of *P. crombiei* consisted only of small arthropods. Collembola dominated the stomach content in terms of frequency, number, and mass. Number of oocytes varied from 80 to 253 and showed positive relationship to size and mass of females. Our findings suggest *P. crombiei* reproduce throughout the year and have generalist diet. Apparently, this population is not threatened by local extinction, because these frogs feed upon any available prey that can be ingested, which can be linked to high fertility.

Key words: Atlantic forest, feeding ecology, predation, reproduction, sex ratio.

Amphibians occupy an important trophic position in terrestrial and aquatic ecosystems by controlling prey populations, especially invertebrates, and by functioning as prey for other animals (Toledo et al. 2007, Wells 2007). To understand the position of amphibians in the trophic network, it is important to know their diet composition (Gunzburger 1999).

Most amphibians are generalist consumers and feed upon invertebrates and small vertebrates (Duellman & Trueb 1994, Solé & Rödder 2010, Castro et al. 2016). On the other hand, some species have a narrow diet or even specialize on certain prey categories (Toft 1980, 1981, Ferreira et al. 2012, 2015). Intraspecific differences of prey consumed may be influenced by behavior, sex, and size and consequently influence fertility (Camilleri & Shine 1990, Donnelly 1991, Miranda et al. 2006). For instance, only large females of *Leptodactylus natalensis* Lutz, 1930 feed upon other anuran species and they also produce more oocytes (Ferreira et al. 2007). The differences in consumed prey by males and females may be related to activity patterns during forage, in which females are more active than males and consequently eat more mobile prey (Ferreira et al. 2007).

Anurans from the Atlantic Forest have almost 40 reproductive modes, varying from more basal (i.e. oviposition in water bodies) to more specialized ones (i.e. oviposition in foam nests) (Haddad et al. 2013). Generally, species that use basal reproductive modes have smaller eggs and higher fertility (i.e. larger clutches) than species with specialized modes (Prado et al. 2005, Pupin et al. 2010). However, anurans that use specialized modes have some advantages in environments in which fresh water is limited (Haddad & Prado 2005). For instance, some leaf litter anurans may use foam nests that maintain humidity, prevent desiccation and for predation (Heyer & Wolf 1989, Duellman & Trueb 1994, Giaretta & Menin 2004).

The 48 species of *Physalaemus* Fitzinger, 1826 are stream breeders (Haddad et al. 2013, Frost 2016). These frogs deposit few large eggs in foam nests on water or forest floor (Haddad & Prado 2005, Pupin et al. 2010). *Physalaemus* species have a broad food spectrum by consuming Formicidae, Isoptera, Colleoptera, Orthoptera, Araneae and Collembola (Falico et al. 2012, González-Duran et al. 2012, Menin et al. 2015). Most species are apparently generalist feeders. However, the life history of some species of *Physalaemus* is still unknown. For example, *P. crombiei* Heyer & Wolf, 1989 is an endemic species to the Atlantic Forest, with populations in Espírito Santo and Bahia states. Ecological aspects such as diet, sex ratio and fecundity remain unstudied for this species.

Thus, the goals of this study are: 1) describe the diet of *P. crombiei* in a sandy-coastal plain in Espírito Santo state, 2) test the relationship between prey items and sex, and 3) determine fecundity and relationship to female snout-vent length. These ecological aspects are particularly useful in providing practical interpretations of species-specific behavioral observations.

Fieldwork was carried out at a sandy-coastal plain (*restinga* habitat) in the Ubu Lagoon (20° 47' 22.63" S and 40° 35' 11.76" W) municipality of Anchieta, Espírito Santo state, southeastern Brazil. Restinga is an open xeromorphic habitat that covers the recent Quaternary sand-dunes of the coast, and is characterized by landscape dominated by shrubs, cactus, bromeliads, and grass patches, with extended areas of exposed sand (Lacerda et al. 1984). The study site has been severely disturbed in the last decades, and its original floristic composition has been replaced by non-native plants (Teixeira, R. L. pers. obs.).

The nocturnal samplings (18-22h) were conducted every two months from November 1999 to September 2000. The water bodies were surveyed and the individuals were hand-captured. All captured individuals were kept in water containing dissolved xylocaine. After this, individuals were transferred to a solution of formalin 5%. Then, they were washed and kept in a solution of alcohol 70%.

Laboratory procedures included measurement of snout-vent length (SVL) with caliper (0.01 mm precision) for each individual, weighing (0.1 g precision), and dissection for removal of stomachs and oocytes. All prey items were sorted, counted and identified to the lowest possible taxonomic level under a stereomicroscope. The prey items with a dubious identification were taken to invertebrate specialist for confirmation or correction. Food items were analyzed by their frequency, number and wet weight in an analytical balance (0.0001 g = 0.1 mg). Clutch size was determined for each female by manually counting the total number of mature oocytes. Mature oocyte is unpigmented and with mean diameter of 1.38±0.064 mm (range 1.25–1.49, Pupin et al. 2010).

Table 1. Frequency, number, and mass (g) of prey items in the diet of *P. crombiei* (N= 47).

Prey	Frequency (%)	Number (%); frog sex	Mass (%)
Arachnida			
Araneae	1 (3)	1 (2.1); male	2.1 (1.5)
Pseudoscorpionida	1 (3)	1 (2.1); female	3.4 (2.5)
Isoptera	2 (6.1)	4 (8.5); male	5.4 (4)
Insecta			
Coleoptera larvae	9 (39.4)	9 (19.1); 6 males, 3 females	23.5 (17.3)
Collembola	13 (51.1)	24 (51.1); 17 males, 7 females	37.5 (27.6)
Blattodea	1 (3)	1 (2.1); male	21.5 (15.8)
Formicidae	5 (15.2)	6 (12.8); 4 males, 2 females	9.8 (7.2)
Lepidoptera larvae	1 (3)	1 (2.1); female	32.7 (24.1)

Sex ratio was estimated using Pearson's chi-square exact test (χ^2) through a Monte Carlo simulation based on 999 replicates. The t-student test was employed to detect differences in SVL and weight between sexes. Linear regression was used to evaluate the relationship between number of oocytes and SVL of females. All analyses were performed in R 2.8.1 (R Development Core Team, 2004). Mean (±1 standard deviation) are provided throughout.

We collected 38 individuals of *P. crombiei* of which 13 (34%) were females and 25 (66%) were males. Sex ratio was 1.9:1 slightly biased toward males ($\chi^2 = 3.78$; $p = 0.07$). Mean SVL of females was 24.09 mm (±0.89) and ranged from 22.1 to 25.4 mm (Fig. 1). Mean SVL of males was 22.96 mm (±1.29) and ranged from 20.2 to 25.1 mm (Fig. 1). Females were larger than males ($T = 5.000$; $p < 0.05$).

Thirty-two (84%) individuals had at least one prey item in the stomach, of which 12 (37%) were females and 20 (63%) were males. A total of 47 prey items representing eight arthropod taxa were found in the stomach content. The mean number of prey items per stomach was 1.2 (±1.1). Collembola was the dominant food item of *P. crombiei* but Colleoptera larvae, Formicidae and Lepidoptera larvae were also important prey in terms of frequency, number and mass (Table 1). Males and females did not differ in frequency of prey ingested ($\chi = 7.35$, df = 7, $p = 0.39$), but five prey types (Araneae, Pseudoscorpionida, Isoptera, Blatodea and Lepidoptera larvae) were found only in one of the sexes (Table 1).

Females were found throughout the study period (January to September), except in May, when no individual of *P. crombiei* was captured. All females had well developed oocytes throughout the study period. The mean number of oocytes was 193 (±44) and ranged from 80 to 253. The smallest female having mature oocytes was 22.1 mm in SVL. The number of oocytes showed positive relationship to female SVL ($r = 0.82$; $p < 0.05$) (Fig. 2).

The diet of the studied population of *P. crombiei* was composed of arthropods, mainly Collembola and Coleoptera larvae. The high consumption of Collembola and Coleoptera by *P. crombiei* may be related to the high density of these preys in the leaf litter. The density of Collembola can reach 200 to 1800 individuals per dm³ and consequently influences the density of its predator (i.e. Coleoptera) (Bellinger et al. 2016). The high fraction of Collembola and Coleoptera in *P. crombiei* diet is similar to other congeners. For example, in the diet of *P. albonotatus*, Coleoptera and Collembola composed 47.2% and 47.1% of the total prey consumed, respectively (Falico et al. 2012). Collembola was more consumed prey in both warm (36.7%) and cold (47.8%) seasons for *P. gracilis*, Boulenger, 1883 (Rosa et al. 2002). Coleoptera was an important food item in the diet of *P. biligonigerus* Cope, 1861 and *P. cicada* Bokermann, 1966 (Santana & Juncá 2007, Rödder 2008). We did not evaluate prey availability in the environment but because species of the genus *Physalaemus* have local differences in the important consumed prey (Santos et al. 2004, Santana & Juncá 2007, Falico et al. 2012), it may suggest that these frogs consume similar proportion of prey to the abundance available in the environment.

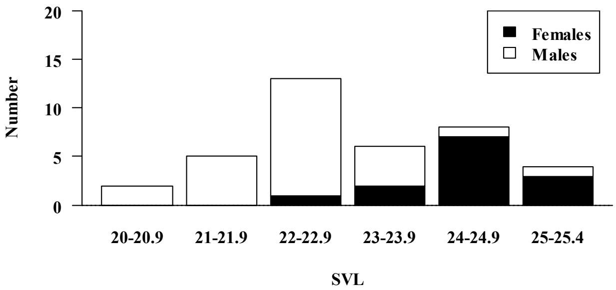

Figure. 1. Snout-vent length (mm) distribution frequency of males and females of *P. crombiei*.

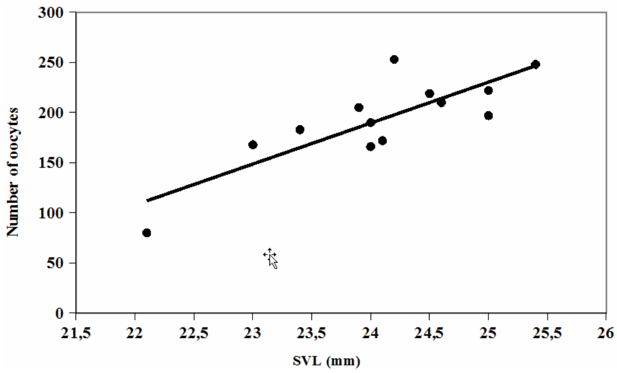

Figure 2. Relationship between the snout-vent length and mature oocytes in females of *P. crombiei*.

The larger size of females compared to males probably was not a consequence of small variation in diet, because there was no difference between the frequencies of the types of preys ingested by sex. However, the SVL of females showed a positive relation with the number of oocytes, which is also reported for other frogs (Berven 1988, Lemckert & Shine 1993, Lüddecke 2002, Pupin et al. 2010). This implies that larger females produce more eggs, which may result in higher reproductive success. Besides, during all of the study period, females showed a high number of mature oocytes, suggesting that this species has a broad reproductive period (Pupin et al. 2010).

Our study suggests that *P. crombiei* diet is not limiting population size because they can feed on any available prey that can be swallowed. Furthermore, females of *P. crombiei* produce large number of oocytes and reproduce throughout the year, which allows positive recruitment in the population. The slight difference in sex ratio is evidence of 'female chosen' breeding strategy (*sensu* Altmann 1974).

Based on the wide spectrum of food types, we suggest *P. crombiei* is a sit-and-wait generalist feeder. Toft (1981) stated that foraging behavior and the number of prey items in the stomach is correlated with the degree of specialization. The sit-and-wait strategy may be a population response to the most available prey type. Thus, we recommend future studies to investigate the available prey items in the environment to further understand feeding strategy of *P. crombiei*.

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References

Altmann, J. (1974): Observational study of behavior: sampling methods source. Behaviour 49: 227-267.

Bellinger, P.F., Christiansen, K.A., Janssens, F. (2016): Checklist of the Collembola of the World covered by collembola.org. <www.collembola.org>, accessed at 2016.06.30.

Berven, K.A. (1988): Factors affecting variation in reproductive traits within a population of wood frogs (*Rana sylvatica*). Copeia 3: 605-615.

Camilleri, C., Shine, R. (1990): Sexual dimorphism and dietary divergence: differences in trophic morphology between male and female snakes. Copeia 3:649-658.

Castro, I.M., Rebouças, R., Solé, M. (2016): Diet of *Dendropsophus branneri* (Cochran, 1948) (Anura: Hylidae) from a cocoa plantation in southern Bahia, Brazil. North-Western Journal of Zoology 12: 159-165.

Donnelly, M.A. (1991): Feeding patterns of the strawberry poison frog *Dendrobates pumilio* (Anura: Dendrobatidae). Copeia 723-730.

Duellman, W.E., Trueb, L. (1994): Biology of amphibians. 2rd Edition. The johns Hopkins University press, Baltimore.

Falico, D.A., López, J.A., Antoniazzi, C.E., Beltzer, A.H. (2012): Variación interpoblacional y ontogenética en la dieta de la rana llorona *Physalaemus albonotatus* (Anura: Leiuperidae). Revista Mexicana de Biodiversidad 83: 1187-1193.

Ferreira, R.B., Dantas, R.B., Teixeira, R.L. (2007): Reproduction and ontogenetic diet shift in *Leptodactylus natalensis* (Anura: Leptodactylidae) from sutheastern Brazil. Boletim do Museu de Biologia Mello Leitão 22: 45-55.

Ferreira, R.B., Schineider, J.A.P., Teixeira. R.L. (2012): Diet, fecundity, and use of bromeliads by *Phyllodytes luteolus* (Anura: Hylidae) in southeastern Brazil. Journal of Herpetology 46: 19-24.

Ferreira, R.B., Beard, K.H., Choi, R.T., Pitt, W.C. (2015): Diet of the Nonnative Greenhouse Frog (*Eleutherodactylus planirostris*) in Maui, Hawaii. Journal of Herpetology 49: 586-593.

Frost, D.R. (2016): Amphibian species of the world covered by American Museum of Natural History. <http://research.amnh.org/herpetology/amphibia/index.html>, accessed at 2016.05.30.

Giaretta, A., Menin, M. (2004): Reproduction, phenology and mortality sources of a 269 species of *Physalaemus* (Anura: Leptodactylidae). Journal of Natural History 38: 1711-1722.

González-Duran, G., Gutiérrez-Cárdenas, P.D.A., Escobar-Lasso, S. (2012): *Physalaemus pustulosus* (Tungara frog) Diet. Herpetological Review 43: 124-125.

Gunzburger, M.S. (1999): Diet of the red hills salamander *Phaeognathus hubrichti*. Copeia 2: 523-525.

Haddad, C.F.B., Prado, C.P.A. (2005): Reproductive Modes in Frogs and their unexpected diversity in the Atlantic Forest of Brazil. BioScience 55: 207-217.

Haddad, C.F.B., Toledo, L.F., Prado, C.P.A., Loebmann, D., Gasparini, J.L., Sazima, I. (2013): Guia dos Anfíbios da Mata Atlântica: Diversidade e Biologia. 2rd Edition. Anolis Books, São Paulo.

Heyer, W.R., Wolf, A.J. (1989): *Physalaemus crombiei* (Amphibia, Leptodactylidae), a new frog species from Espírito Santo, Brazil with comments on the *Physalaemus signifer* group. Proceedings of the Biological Society of Washington 102: 500-506.

Lacerda, L.D., Araújo, D.S.D., Cerqueira, R., Turco, B. (1984): Restingas. Origem, estruturas, processos. 1st Edition. Universidade Federal Fluminense, Niterói.

Lemckert, F.L., Shine, R. (1993): Costs of reproduction in a population of the frog *Crinia signifera* (Anura: Myobatrachidae) from Southeastern Australia. Journal of Herpetology 27: 420-425.

Lüddecke, H. (2002): Variation and trade-off in reproductive output of the Andean frog *Hyla labialis*. Oecologia 130: 403-410.

Menin, M., Santos, R.S., Borges, R.E., Piatti, L. (2015): Notes on the diet of seven terrestrial frogs in three agroecosystems and forest remnants in Northwestern São Paulo State, Brazil. Herpetology Notes 8: 401-405.

Miranda, T., Ebner, M., Solé, M., Kwet, A. (2006): Spatial, seasonal and intrapopulational variation in the diet of *Pseudis cardosoi* (Anura: Hylidae) from the Araucária plateau of Rio Grande do Sul, Brazil. South American Journal of Herpetology 1:121-130.

Prado, C.P.A., Uetanabaro, M., Haddad, C.F.B. (2005). Breeding activity patterns, reproductive modes, and habitat use by anurans (Amphibia) in a seasonal environment in the Pantanal, Brazil. Amphibia-Reptilia 26: 211-221.

Pupin, C.N., Gasparini, J.L., Bastos, R.P., Haddad, C.F.B., Prado, C.P.A. (2010). Reproductive biology of an endemic *Physalaemus* of the Brazilian Atlantic forest, and the trade-off between clutch and egg size in terrestrial breeders of the *P. signifer* group. Herpetological Journal 20: 147-156.

Rosa, I., Canavero, A., Maneyro, R., Naya, D.E., Camargo, A. (2002): Diet of four sympatric anuran species in a temperate environment. Boletín de la Sociedad Zoológica del Uruguay 13: 12-20.

Rödder, D. (2008): Diet of *Physalaemus biligonigerus* (Cope, 1861, "1860") and *Eleutherodactylus platydactylus* (Boulenger, 1903) (Anura: Leiuperidae, Brachycephalidae) from Bolivia and Paraguay. Russian Journal of Herpetology 15: 117-121.

Santana, A.S., Juncá, F.A. (2007): Diet of *Physalaemus* cf. *cicada* (Leptodactylidae) and *Bufo granulosus* (Bufonidae) in a semideciduous forest. Brazilian Journal of Biology 67: 125-131.

Santos, E.M., Almeida, A.V., Vasconcelos S.D. (2004): Feeding habits of six anuran (Amphibia: Anura) species in a rainforest fragment in Northeastern Brazil. Iheringia Série Zoológica 94: 433-438.

Solé, M., Rödder, D. (2010): Dietary assessments of adult amphibians. In: Dodd, K.Jr., editor. Amphibian ecology and conservation. Oxford: Oxford University Press.

Toft, C.A. (1980): Feeding ecology of thirteen syntopic species of anurans in a seasonal tropical environment. Oecologia 45: 131-141.

Toft, C.A. (1981): Feeding ecology of panamanian litter anurans: pat-terns in diet and foraging mode. Journal of Herpetology 15: 139-144.

Toledo, L.F., Ribeiro, R.S., Haddad, C.F.B. (2007): Anurans as prey: an exploratory analysis and the size relationships between predators and their preys. Journal of Zoology 271: 170-177.

Wells, K.D. (2007): The ecology and behavior of amphibians. 1st Edition. University Chicago Press, Chicago.