

AFS 2016044/17306

Helminth parasites and Prey composition in the stomach of *Ptychadena* species from Obazuwa Wetland in Edo State, Nigeria

A.A. Imasuen* and E.E. Enabulele

Department of Animal and Environmental Biology, University of Benin, P.M.B 1154 Benin City, Edo State, Nigeria.

*Corresponding author: abigail.imasuen@uniben.edu

(Received July 14, 2016)

(Accepted in revised form August 19, 2016)

ABSTRACT : This investigation was aimed at determining the helminth parasites and prey items in the stomach of *Ptychadena* sp. from Obazuwa wetland in Edo State Nigeria. Four *Ptychadena* sp. (*P. longirostris*, *P. bibroni*, *P. oxyrhynchus* and *P. mascareniensis*) were collected using the visual/acoustic encounter survey methods. The stomach contents of the frogs examined revealed a total of 127 identifiable items comprising of 108 arthropods, 16 nematodes and 3 cestodes. The arthropods recorded belonged to 9 taxa and percentage frequency of occurrence was: Araneae 5%, Blattaria 10%, Coleoptera 10%, Diptera 17.5%, Hymenoptera 47.5%, Lepidoptera 2.5%, Mollusca 15%, Myriapoda 7.5% and Orthoptera 7.5%. The Hymenoptera recorded in the four *Ptychadena* sp. only accounted for 23 (18.1%) of the total identifiable prey items. Most of the recorded helminths were recovered from the stomach of *P. oxyrhynchus* consisted of (cestodes and nematodes from 2 and 7 specimens, respectively while only nematodes were recovered from single specimens of *P. longirostris* and *P. bibroni* each. Percentage frequency of occurrence of the helminths was 5% for cestodes and 22.5% for the nematodes. The record of helminth parasites as part of the stomach contents of the frogs is a first report in Nigeria on dietary studies in anurans.

Key words: *Ptychadena*, diet, helminths, wetland, Nigeria

Introduction

The information derived from the study of anuran diet is essential to understanding their life history, population fluctuation and what impact habitat alteration may have on their diet (Anderson *et al.*, 1999). Research in Africa on anuran diet has increased in the last decade, and most of the commonly reported prey items on the menu are usually invertebrate orders such as Arachnida, Insecta, Collembola, Chilopoda, Gastropoda and Crustacea (Blackburn and Moreau, 2006; Luisei *et al.*, 2004; Kouame *et al.*, 2008; Enabulele and Aisien, 2012; Enabulele and Imasuen, 2012). Cannibalism has also been reported in some anurans (Knoepffler, 1976; Ogoanna and Uchedike, 2010; Hirschfeld and Rödel, 2011) and smaller vertebrate such as chameleon (D’Cruze and Sabel, 2005) and fish (Hirschfeld and Rödel, 2011).

Ptychadena species are frogs within the anuran family Ptychadenidae, they are distributed in tropical and subtropical countries (Rödel, 2000). In Nigeria, some *Ptychadena* species commonly reported

include; *P. bibroni*, *P. schubotzi*, *P. aequiplicata*, *P. longirostris*, *P. pumilio*, and *P. mascareniensis* (Aisien *et al.*, 2003; 2009; Onadeko and Rödel, 2009; Imasuen, 2012). Previous studies on the food and feeding habits of *Ptychadena* species has revealed that they are generalist feeders (mainly arthropods) with an active foraging behaviour (Rödel, 2000; Enabulele and Imasuen, 2012). *Ptychadena* species predation on conspecifics and other frogs has also been reported (McIntyre and Ramanamanjato, 1999; Fatroandrianjafinonjasolomiovazo *et al.*, 2011; Enabulele and Imasuen, 2012).

Although most studies on the stomach contents of anurans in general have revealed mainly a diversity of arthropod prey items, few investigations have reported helminth parasites in the stomach of anurans (Erftemeijer and Boeadi, 1991; Lima *et al.*, 2010). The diet of anurans may also play an important role in the parasite fauna they harbour particularly if their prey items serve as intermediate host of parasites. In this investigation, we report both the prey items and helminth parasites in the stomach of *Ptychadena* species collected from Obazuwa wetlands in Edo State of Nigeria.

Materials and Methods

Ptychadena species were collected from Obazuwa wetland (Latitude 5° and 6° N and Longitude 5° and 7° E) located in Ovia North East Local Government Area of Edo State (Figure 1). The wetland is located within an altered rainforest environment though anthropogenic activities around and in the wetland are strictly controlled. Vegetation in and around the wetland include; *Elaeis guineensis*, *Hevea brasiliensis*, *Alchomelaxiflora* sp, *Nephrolepis biserrata* and *Anacardium occidentale*.



Figure 1: Sampling site at Obazuwa wetland in Edo State, Nigeria.

Frogs were collected between the hours of 19:00 to 23:00 using both acoustic and visual encounter surveys (Crump and Scott, 1994; Rödel and Ernst, 2004). Captured frogs were euthanized by exposing them to chloroform in a jar and 5% formalin injected into their stomach to preserve the stomach contents. Protocols for the examination of the stomach contents were as previously described by Enabulele and Aisien (2012). Prey items in the stomachs were classified in categories representing taxonomic orders, except for the helminths parasites, which were separated into tapeworms (Cestoda) and roundworms (Nematoda). Two indices, frequency of occurrence of food item (FO) and rate of feeding of the frogs (RF) were determined as described by Sala and Ballesteros (1997). FO was calculated as number of stomachs with a particular prey/stomach item divided by total number of stomachs with identifiable items. RF was estimated as the percentage of stomachs containing identifiable items divided by the total number of stomachs examined. The RF varies from 0 (no feeding activity) to 100 (all individuals with food).

Results

Forty-five *Ptychadena* species comprising of 21 *P. longirostris*, 6 *P. bibroni*, 17 *P. oxyrhynchus* and 1 *P. mascareniensis* were collected from the study site. Examination of the stomach contents of the frogs revealed that 40 (88.89%) had identifiable prey items, 4 (8.89%) were empty and 1 (2.22%) had digested content (Table 1). Other stomach content recorded were plant materials in 12 (26.67%) frogs, cestodes from 2 (4.44%) *P. oxyrhynchus* while 9 (20%) comprising of 7 *P. oxyrhynchus*, 1 *P. longirostris* and *P. bibroni* respectively had nematodes (Table 1).

Table 1: Stomach contents of *Ptychadena* sp. from Obazuwa wetland in Edo State

Stomach contents	<i>P. longirostris</i> (n = 21)	<i>P. bibroni</i> (n = 6)	<i>P. oxyrhynchus</i> (n = 17)	<i>P. mascareniensis</i> (n = 1)	Total	%
Identifiable arthropods	19	4	16	1	40	88.89
Digested food	-	1	-	-	1	2.22
Empty	2	1	1	-	4	8.89
Plant material	4	1	7	-	12	26.67
Cestoda	-	-	2	-	2	4.44
Nematoda	1	1	7	-	9	20

A total of 127 identifiable stomach contents comprising of 108 arthropods, 16 roundworms and 3 tapeworms were recorded in the stomachs of the examined frogs (Table 2). The arthropods recorded belonged to 9 taxa and the %FO was: Araneae 5%, Blattaria 10%, Coleoptera 10%, Diptera 17.5%, Hymenoptera 47.5%, Lepidoptera 2.5%, Mollusca 15%, Myriapoda 7.5% and Orthoptera 7.5% (Table 2). The %FO for the helminth parasites were, 17.5% for roundworms while tapeworms was 5%. Among the arthropod prey items, only Hymenoptera was recorded in the four *Ptychadena* sp and accounted for 23 (18.1%) of the total identifiable items in the stomach of the frogs. Coleopterans, dipterans and molluscan items were recorded in three *Ptychadena* sp while prey items belonging to

araneae, blatteria, myriapoda and orthoptera were identified in two *Ptychadena* sp. (Table 2). Lepidoptera was only recorded in one *P. oxyrhynchus*. The rate of feeding in the frogs was determined to be 88.89%.

Table 2: Percentage frequencies (%FO) of identifiable items in the stomachs of *Ptychadena* sp. from Obazuwa wetland in Edo State

Stomach contents	Number of identifiable items in <i>Ptychadena</i> sp.				Total items	Stomach with items	%FO
	<i>P. longirostris</i> (n = 21)	<i>P. bibroni</i> (n = 6)	<i>P. mascareniensis</i> (n = 1)	<i>P. oxyrhynchus</i> (n = 17)			
Araneae	48	0	0	1	49	2	5
Blattaria	1	0	0	1	2	4	10
Coleoptera	2	1	1	0	4	4	10
Diptera	8	1	0	2	11	7	17.5
Hymenoptera	14	4	1	4	23	19	47.5
Lepidoptera	0	0	0	1	1	1	2.5
Mollusca	4	2	0	1	7	6	15
Myriapoda	2	0	0	2	4	3	7.5
Orthoptera	3	0	0	4	7	7	17.5
Nematoda	1	1	0	14	16	9	22.5
Cestoda	0	0	0	3	3	2	5

Discussion

Ptychadena species are considered to be ‘generalist feeders’ feeding mainly on an array of terrestrial invertebrates (Rödel, 2000). Analysis of the stomach contents of the four *Ptychadena* species (*P. longirostris*, *P. bibroni*, *P. oxyrhynchus* and *P. mascareniensis* from Obazuwa wetland revealed that 40 (88.86%) of the 45 stomach examined had identifiable contents (Table 1). The diversity of arthropod taxa reported in this study: Araneae, Blattaria, Coleoptera, Diptera, Hymenoptera, Lepidoptera, Mollusca, Myriapoda and Orthoptera (Table 2) were similar to the report by Enabulele and Imasuen (2012) from *Ptychadena* species sampled from a monoculture plantation and a rainforest habitat in southern Nigeria. However, Isopoda and Annelida reported by Enabulele and Imasuen (2012) were not recorded in this study. Previous studies on *Ptychadena* have also revealed their cannibalistic behaviour on both conspecifics and other frogs (McIntyre and Ramanamanjato, 1999; Fatroandrianjafinonjasolomiovazo *et al.*, 2011; Enabulele and Imasuen, 2012).

Hymenopterans (ants) appear to be a preferred prey items for *Ptychadena*, and was the only arthropods recorded in the stomach of the four *Ptychadena* species studied and also accounted for the highest frequency of occurrence 47.5% (Table 2). Enabulele and Imasuen (2012) also recorded high percentage of

hymenopterans (25%) in the diet of *Ptychadena*. It is possible that the ease by which hymenopterans can be eaten while crawling along the ground may be responsible for their high levels in the stomach of the frogs. Also, prey availability in a given environment may also be a contributing factor (Houston, 1973; Hirai and Matsui 1999; Maneyro and da Rosa, 2004). Interestingly, in this study an unusually high amount of prey item (48 spiders: Aranaea) was recorded in the stomach of one *P. longirostris* (Table 2). A possible explanation for the high number is that the frog may have encountered a nest of spiders.

Plants materials were also recorded in this study and accounted for 26.67% of items in the stomach of the frogs examined (Table 1). Enabulele and Imasuen (2012) reported plant seeds in the stomach of a single specimen of *P. oxyrhynchus*. Some authors are of the opinion that plants may not be food items in anurans but only an incidental ingestion during intake of prey (Evans and Lampo, 1996; Anderson and Mathis, 1999; Teixeira *et al.*, 1999; Van Sluys *et al.*, 2001). On the contrary, Anderson *et al.* (1999), suggested that the ingestion of plant material was a deliberate feeding behaviour aiding the breakdown of exoskeletons, elimination of intestinal parasites and as an extra source of water.

It is remarkable that a considerable amount of helminth parasites were recorded in the stomach of some of the frogs examined, nematodes and cestodes made up 20% and 4.44% of the identifiable items (Table 1). There is generally paucity of information on helminth recovered from the stomach of anurans during dietary investigation. It is possible that either helminthes are wrongly identified by investigators (perhaps grouped as larvae of coleopterans) or absent in most studies. In Nigeria, Imasuen *et al.* (2012) has however reported nematodes and cestodes cysts on the outer walls of some anurans including *Ptychadena* species. Erfstemeijer and Boeadi (1991) recorded nematodes in the stomach of *Microhyla heymonsi* and *Rana chalconota* with 3.9% and 1.9% frequency of occurrence respectively. Lima *et al.* (2011) also reported cycloneuralia nematode in *Phyllomedusa burmeisteri* which accounted for 6.76% frequency of occurrence of identifiable stomach contents. In the current study, the frequency of occurrence of the helminths was 5% for cestodes and 22.5% for the nematodes (Table 2). Most of the recorded helminths were recovered from the stomach of *P. oxyrhynchus* (cestodes and nematodes from 2 and 7 specimens respectively) while only nematodes was recovered from only one *P. longirostris* and *P. bibroni* (Table 1).

The nematode specimens recovered in this study belong to the genus *Physaloptera* which has been reported to occupy the stomach of reptiles, amphibians and mammals (Kung, 1948; Vrcibradic *et al.*, 2000; Ávila and Silva, 2010; Dorigo *et al.*, 2014). Aisien *et al.* (2009) first reported its presence in Nigeria from two amphibian hosts, *P. oxyrhynchus* and *P. bibroni*. However, the confirmation of the presence of this parasitic nematode in Nigeria was in the amphibian host *Phrynobatrachus plicatus* from a protected rainforest, Okomu National Park (Imasuen, 2012). In this investigation, *Physaloptera* was recorded in three *Ptychadena* species *P. oxyrhynchus*, *P. bibroni* and *P. longirostris*. The latter therefore enlarges the host range in the country. It's difficult to ascertain which organism serves as the intermediate host of *Physaloptera* considering its wide distribution across three vertebrate classes (amphibians, reptiles and mammals). Noteworthy however is that grasshoppers (orthopterans) were the only prey items recorded in the stomach of all *P. oxyrhynchus* harbouring *Physaloptera*.

Cestodes were recovered only from the stomachs of two *P. oxyrhynchus*. Previous parasitological studies on *Ptychadena* and other anurans in Nigeria have only revealed the occurrence of cestodes in the small intestine of infected frogs (Aisien *et al.*, 2003, 2009). *Cephalochlamys namaquensis* a cestode reported in the intestine of the aquatic frog *Silurana tropicalis* (Imasuen and Aisien, 2015) has been observed to occur in the stomach of *S. tropicalis* (personal observations by A.A. Imasuen). Although the cestodes recovered from *P. oxyrhynchus* could not be identified, its occurrence in the stomach may be due to its wandering movement in search of food or a reaction to stress factors on the frogs.

The findings in this investigation are consistent with previous studies on the diet of *Ptychadena* species and confirm the frogs as generalist feeders. The wetland environment appears not to have influenced the diversity of prey items available for the frogs in comparisons to similar studies on *Ptychadena* species from altered environment. The record of helminths particularly for the cestodes will require further investigations to confirm their occurrence in the stomach of *Ptychadena* and identify the species.

ACKNOWLEDGEMENT: The authors are grateful to the village head and people of Obazuwa community for permission to sample in Obazuwa wetland. Also appreciated is Mr. Festus Arijode for his assistance in field work.

References

- Aisien SO, Ajakaiye FB, Braimoh K: Helminth parasites of anurans from the savannah mosaic zone of South-western Nigeria. *Acta Parasit* 48(1): 47-54. 2003.
- Aisien AA, Imasuen AA, Ogoannah SO: Helminth Parasites of Amphibians from a Rainforest Reserve in Southwestern Nigeria. *Afr Zool* 44(1): 1-7. 2009.
- Anderson AM, Haukos DA, Anderson JT: Diet composition of three anurans from the Playa Wetlands of Northwest Texas. *Copeia* 1999(2): 515-520. 1999.
- Anderson MT, Mathis A: Diets of two sympatric Neotropical Salamanders, *Bolitoglossa mexicana* and *B. rufescens*, with notes on reproduction for *B. rufescens*. *J Herpetol* 33(4): 601-607. 1999.
- Ávila RW, Silva RJ: Checklist of helminths from lizards and amphisbaenians (Reptilia, Squamata) of South America. *J Venom Anim Toxins incl Trop Dis* 16(4): 543-572. 2010.
- Blackburn DC, Moreau CS: Ontogenetic diet change in the arthroleptid frog *Schoutheimella xenodactyloides*. *J Herpetol* 40: 388-394. 2006.
- Crump ML, Scott NJJ: Visual encounter surveys. In: *Measuring and monitoring biological diversity: standard methods for amphibians*. (Heyer WR, Donnelly MA, McDiarmid RW, Hayek LC, Foster MS. eds.), pp. 84-92. Smithsonian Institution Press, Washington. 1994.
- D’cruze NC, Sabel AJ: *Ptychadena mascareniensis* (Mascarene ridge frog): Predation on an endemic Malagasy chameleon. *Herpetological Bulletin* 93: 26-27. 2005.
- Dorigo TA, Maia-Carneiro T, Almeida-Gomes M, Siqueira CC, Vrcibradic D, Van Sluys M, Rocha CF: Diet and helminths of *Enyalius brasiliensis* (Lacertilia, Iguania, Leiosauridae) in an Atlantic Rainforest remnant in southeastern Brazil. *Braz J Biol* 74(1): 199-204. 2014.
- Enabulele EE, Aisien MSO: Diets of *Hemisis marmoratus* and *Leptopelis hyloides* (Order: Anura) from monoculture plantations in Southern Nigeria. *The Zoologist* 10: 48-52. 2012.
- Enabulele EE, Imasuen AA: Dietary constituents of *Ptychadena* species from a monoculture plantation and a rainforest habitat in southern Nigeria. *Nig J Life Sci* 2(1): 208-215. 2012.
- Erfemeijer P, Boeadi P: The diet of *Microhyla heymonsi* Vogt (Microhylidae) and *Rana chalconota* Schlegel (Ranidae) in a pond on West Java. *Raffles B Zool* 39(2): 279-282. 1991.
- Evans M, Lampo M: Diet of *Bufo marinus* in Venezuela. *J Herpetol* 30(1): 73-76. 1996.
- Fatroandrianjafinonjasolomiovazo TNL, Rasoamampionona NR, Vieites DR, Vences V: Diet of the Mascarene grass frog, *Ptychadena mascareniensis*, in Madagascar. *Malagasy Nature* 5: 68-74. 2011.
- Hirai T, Matsui M: Feeding habits of the Pond Frog, *Rana nigromaculata*, inhabiting rice fields in Kyoto, Japan. *Copeia* 1999: 940-947. 1999.
- Hirschfeld M, Rödel M-O: The diet of the African Tiger Frog, *Hoplobatrachus occipitalis*, in northern Benin. *Salamandra* 47(3): 125-132. 2011.
- Houston WWK: The food of the common frog, *Rana temporaria*, on high moorland in northern England. *J Zool* 171: 153-165. 1973.
- Imasuen AA: Investigation of the helminth parasitic infections and chytridiomycosis of amphibians in Okomu National Park, Nigeria. Ph.D Thesis, University of Benin, Nigeria. 2012.
- Imasuen AA, Ozemoka HJ, Aisien MSO: Anurans as intermediate and paratenic hosts of helminth infections in the rainforest and derived savanna biotopes of Southern Nigeria. *Inter J Zoo Article ID 823970*. 7pages, doi: 1155/2012/823970. 2012.
- Imasuen AA, Aisien MSO: Helminth parasites of *Silurana tropicalis* from the Okomu National Park, Edo State, Nigeria. *Nig J Parasit* 36(1): 61-66. 2015.
- Knoepffler L-P: Food habitats of *Aubria subsigillata* in Gabon. *Zool Afri* 11: 369-371. 1976.
- Kouame NG, Tohe B, Assemkin NE, Gourene G, Rödel M-O: Prey composition of two syntopic *Phrynobatrachus* species in the swamp forest of Bamco National Park, Ivory Coast. *Salamandra* 44: 177-186. 2008.
- Kung CC: On some new species of Spirurids from Terrestrial Vertebrates, with notes on *Habronema mansioni*, *Physaloptera paradoxa* and *Hartertia zuluensis*. *J Helminthol* 22(3-4): 141-164. 1948.
- Lima JE, Rödder D, Solé M: Diet of two sympatric *Phyllomedusa* (Anura: Hylidae) species from a cacao plantation in southern Bahia, Brazil. *N West J Zool* 6(1): 13-24. 2011.
- Luisseui L, Bikikoro L, Odegbune E, Wanboko SM, Rugiero L, Akani GC, Politano E: Feeding relationship between sympatric Afrotropical tree frogs (genus *Hyperolius*); The effects of predator body size and season. *Anim Biol* 54(3): 293-302. 2004.

- Maneyro R, da Rosa I: Temporal and spatial changes in the diet of *Hyla pulchella* (Anura, Hylidae) in southern Uruguay. *Phyllomedusa* 3: 101-113. 2004.
- McIntyre P, Ramanamanjato J-B: *Ptychadena mascareniensis mascareniensis* diet. *Herpetol Rev* 30(4): 223. 1999.
- Ogoanna SO, Uchedike E: Diet of two anurans (*Hoplobatrachus occipitalis* and *Bufo maculatus*) in Benin City, Nigeria. *Bio Res Comm* 22(4): 189-198. 2010.
- Onadeko AB, Rödel M-O: Anuran survey in South-western Nigeria. *Salamandra* 45(1): 1-14. 2009.
- Rödel M-O: *Herpetofauna of West Africa*. Vol. I. Amphibians of the West African Savanna, pp. 91-144, Frankfurt/M. (Edition Chimaira). 2000.
- Rödel M.-O, Ernst R: Measuring and monitoring amphibian diversity in tropical forests. I. An evaluation of methods with recommendations for standardization. *Ecotropica* 10: 1–14. 2004.
- Sala E, Ballesteros E: Partitioning of space and food resources by three fish of the genus *Diplodus* (Sparidae) in a Mediterranean rocky infralittoral ecosystem. *Mar Ecol Prog Ser* 152: 273–283. 1997.
- Teixeira RL, Schineider JAP, Giovanelli M: Diet of the toad *Bufo granulosis* (Amphibia, Bufonidae) from sandy coastal plain in southeastern Brazil. *Bol Mus Biol Mello Leitão* 10: 29-31. 1999.
- Van Sluys M, Rocha CFD, Souza MB: Diet, reproduction and density of the leptodactylid litter frog *Zachaenus parvulus* in an Atlantic Rain Forest of southeastern Brazil. *J Herpetol* 35(2): 322-325. 2001.
- Vrcibradic D, Cunha-Barros M, Vicente JJ, Galdino, CAC, Hatano FH, Van Sluys M, Rocha CFD: Nematode infection patterns in four sympatric lizards from a restinga habitat (Jurubatiba) in Rio de Janeiro state, southeastern Brazil. *Amphibia-Reptilia* 21(3): 307-316. 2000.