LETÍCIA SOARES FRAGA

PREENCHENDO LACUNAS SOBRE GIRINOS (AMPHIBIA: ANURA) DA ZONA DA MATA DE MINAS GERAIS

Dissertação apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Biologia Animal, para obtenção do título de *Magister Scientiae*.

Orientador: Renato Neves Feio

VIÇOSA - MINAS GERAIS
2023

Ficha catalográfica elaborada pela Biblioteca Central da Universidade Federal de Viçosa - Campus Viçosa

T
F811p
2023

Fraga, Letícia Soares, 1997-
Preenchendo lacunas sobre girinos (Amphibia: Anura) da Zona da Mata de Minas Gerais / Letícia Soares Fraga. – Viçosa, MG, 2023.
1 dissertação eletrônica (31 f.): il. (algumas color.).

Texto em português e inglês.
Orientador: Renato Neves Feio.
Dissertação (mestrado) - Universidade Federal de Viçosa, Departamento de Biologia Animal, 2023.
Inclui bibliografia.
DOI: https://doi.org/10.47328/ufvbbt.2023.160
Modo de acesso: World Wide Web.

1. Girino - Zona da Mata (MG : Mesorregião) - Morfologia. I. Feio, Renato Neves, 1960-. II. Universidade Federal de Viçosa. Departamento de Biologia Animal. Programa de Pós-Graduação em Biologia Animal. III. Título.

CDD 22. ed. 597.8139

Bibliotecário(a) responsável: Euzébio Luiz Pinto CRB-6/3317

LETÍCIA SOARES FRAGA

PREENCHENDO LACUNAS SOBRE GIRINOS (AMPHIBIA: ANURA) DA ZONA DA MATA DE MINAS GERAIS

Dissertação apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Biologia Animal, para obtenção do título de *Magister Scientiae*.

APROVADA: 27 de fevereiro de 2023.

Assentimento:

Letícia Soares Fraga
Letícia Soares Fraga
Autora

Renato Neves Feio
Orientador

AGRADECIMENTOS

Começo dedicando meus agradecimentos àqueles que me ensinaram a amar as coisas mais simples da vida. Meus pais, Maria Helena e José Roberto, obrigada pelo amor infinito e por serem meu exemplo de bondade e simplicidade, obrigada pelas manhãs de leite com achocolatado direto do curral, pelas noites na roça batendo papo, observando as estrelas, os vagalumes e ouvindo os sapos, são minhas memórias mais preciosas e me fizeram ser quem sou hoje.

Obrigada a minha irmã, Laura, minha fiel companheira e alma gêmea. A minha tia, Vanilda, exemplo de inteligência, sabedoria e dedicação. E aos meus avós, mestres que não lêem letras, mas que me ensinaram as lições mais valiosas dessa vida.

Ao Renato Feio, pela orientação e amizade. Serei sempre muito grata por me apresentar tantos lugares e bichos, pelas conversas, caminhadas, oportunidades e incentivos. Seguirei seu exemplo, como profissional e pessoa!

Ao Clodoaldo, profissional e pesquisador fenomenal, obrigada pelas inúmeras oportunidades e ensinamentos.

Ao Tiago Pezzuti por toda ajuda com a morfologia oral interna e por ser um exemplo e grande incentivador nessa jornada com os girinos.

Aos moonjenianos! Foi um privilégio dividir histórias com cada um de vocês, obrigada pelos campos, cafezinhos, cervejas, risadas e "pipipi-popopó" de sempre. Em especial, aos amigos que alocaram um pedacinho do meu coração, Leandro, Taluane e Samuli.

Aos meus amigos da biologia e da vida, por dividirem alegrias e tristezas comigo, crescemos demais juntos, isso é show de bola!

Ao Heitor, meu par e porto seguro. Obrigada pela parceria sem fim e por todo amor! E não posso esquecer, a Cacau, só por existir daquele jeitinho e completar nossa família.

Ao Museu de Zoologia João Moojen, por me receber de braços abertos desde a graduação. À Universidade Federal de Viçosa, pela oportunidade de realizar meus sonhos.

O presente trabalho foi realizado com apoio da Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Código de Financiamento 001.

A construção deste trabalho só foi possível porque sempre pude contar com pessoas incríveis em meu caminho, a todas essas serei eternamente grata.

“O silêncio do mato.
Uma árvore morta ainda de pé.
A última casa da rua. (...)
Fruta na altura das mãos.
Capim cortando as canelas da gente.
A primeira estrela do céu.(...)
Ser exato aquilo que a gente é.”

Alves

RESUMO

FRAGA, Letícia Soares, M.Sc., Universidade Federal de Viçosa, fevereiro de 2023. **Preenchendo lacunas sobre girinos (Amphibia: Anura) da Zona da Mata de Minas Gerais.** Orientador: Renato Neves Feio.

O conhecimento acerca da fase larval dos anfíbios ainda é limitado. Historicamente, há uma discrepância entre o número de descrições e estudos envolvendo anuros adultos quando comparado com aqueles relacionados aos girinos. Parte dessa desproporção pode ser explicada pela dificuldade de identificação das espécies durante a fase larval, já que muitas vezes girinos de espécies diferentes podem ser bastante parecidos. Existem combinações de características que, quando bem observadas, são ferramentas úteis para distingui-los, mas para que isto ocorra é necessário o acesso a descrições confiáveis. Evidencia-se assim a importância do desenvolvimento de estudos que contribuam para o reconhecimento das espécies e, consequentemente, possibilitem o crescimento do conhecimento envolvendo a fase larval dos anuros. Visto isto, no presente trabalho, são expostos novos dados sobre girinos da Zona da Mata de Minas Gerais, sudeste do Brasil. Apresentamos três artigos contendo a descrição larval das espécies: *Thoropa bryomantis*, *Physalaemus feioi* e *Leptodactylus barrioi*. A primeira espécie, *T. bryomantis*, se encontra distribuída através da região norte da Serra da Mantiqueira no domínio da Mata Atlântica, região sudeste do Brasil; anteriormente identificada como *T. lutzi* foi recentemente descrita com base em caracteres acústicos e morfológicos. A segunda espécie, *P. feioi*, com localidade-tipo "Mata da Biologia" no município de Viçosa, Minas Gerais, e com distribuição na Serra da Mantiqueira, era anteriormente associada à espécie *P. olfersii*. A última espécie, *L. barrioi*, foi coletada no município de Cataguases, cerca de 80km de sua localidade-tipo em Duas Barras, Rio de Janeiro; essa espécie pertence ao grupo de espécies *L. fuscus*, complexo *L. mystaceus*, foi descrita após uma revisão do complexo, porém o estudo não envolveu a fase larval. Para ambas descrições foram analisados caracteres morfológicos, morfométricos e coloração, e para *T. bryomantis* caracterizamos também a morfologia oral interna.

Palavras-chave: Larvas. Morfologia. Descrições.

ABSTRACT

FRAGA, Letícia Soares, M.Sc., Universidade Federal de Viçosa, February 2023. **Filling in gaps about tadpoles (Amphibia: Anura) from Zona da Mata de Minas Gerais.** Advisor: Renato Neves Feio.

Knowledge about the larval stage of amphibians is still limited. Historically, there is a discrepancy between the number of descriptions and studies involving adult anurans when compared to those related to tadpoles. Part of this disproportion can be explained by the difficulty in identifying the species during the larval stage, since tadpoles of different species can often look very similar. There are combinations of characteristics that, when well observed, are useful tools to distinguish them, but for this to happen, access to reliable descriptions is necessary. This highlights the importance of developing studies that contribute to the recognition of species and, consequently, enable the growth of knowledge involving the larval stage of anurans. Given this, in the present work, new data on tadpoles from the Zona da Mata of Minas Gerais, southeastern Brazil, are presented. We present three articles containing the larval description of the species: *Thoropa bryomantis*, *Physalaemus feioi* and *Leptodactylus barrioi*. The first species, *T. bryomantis*, is distributed across the northern region of the Serra da Mantiqueira in the Atlantic Forest domain, southeastern Brazil; previously identified as *T. lutzi* was recently described based on acoustic and morphological characters. The second species, *P. feioi*, with type locality "Mata da Biologia" in the municipality of Viçosa, Minas Gerais, and with distribution in Serra da Mantiqueira, was previously associated with the species *P. olfersii*. The last species, *L. barrioi*, was collected in the municipality of Cataguases, about 80km from its type locality in Duas Barras, Rio de Janeiro; this species belongs to the *L. fuscus* species group, *L. mystaceus* complex, it was described after a review of the complex, but the study did not involve the larval phase. For both descriptions, morphological and morphometric characters and coloration were analyzed, and for *T. bryomantis* we also characterized the internal oral morphology.

Keywords: Larval. Morphology. Description.

SUMÁRIO

1. INTRODUÇÃO GERAL ……..……..…….….. 08

2. ARTIGOS ……..…….……..……...….... 11

2.1 ARTIGO I - Fraga, L. S., Guimarães, C. S., Assis, C. L., Pezzuti, T. L. & Feio, R. N. (2023) The tadpole of *Thoropa bryomantis* (Anura: Cycloramphidae): external and oral internal morphology ……..... 12

2.2 ARTIGO II - Fraga, L. S., Assis, C. L., Guimarães, C. S., & Feio, R. N. (2022). The tadpole of *Physalaemus feioi* (Anura: Leptodactylidae) ……..……..….. 21

2.3 ARTIGO III - Fraga, L. S., Assis, C. L., & Feio, R. N. (2022). The tadpole of *Leptodactylus barrioi* from the Atlantic Forest of southeastern Brazil (Amphibia: Anura: Leptodactylidae) ……...…..…….. 26

3. CONCLUSÕES GERAIS 31

1. INTRODUÇÃO GERAL

No Brasil, encontra-se uma das maiores diversidades de anfíbios do mundo, com mais de 1144 espécies descritas, sendo cinco caudatas, 39 gymnophionas e mais de 1100 anuros (Segalla *et al.* 2021). Dentre as espécies de anuros descritas, cerca de 80% passam pelo processo de metamorfose, ou seja, possuem o estágio larval no seu ciclo de vida (Altig & McDiarmid, 1999; Andrade *et al.* 2007). No entanto, a fase larval dos anuros ainda é pouco conhecida quando comparada com a fase adulta, a exemplo das espécies brasileiras, estima-se que somente 60% das espécies possuem fase larval descrita (Provete *et al.* 2012; Dubeux *et al.* 2020; Guerra *et al.* 2020). Estes dados ainda podem estar subestimados, devido ao grande número de espécies descritas nos últimos anos sem caracterização da fase larval (Rossa-Feres *et al.* 2015).

O desequilíbrio entre o conhecimento da fase adulta e larval dos anuros pode ser explicado pela dificuldade na identificação correta das larvas (Pezzuti *et al.* 2021). Isto porque, em uma análise superficial, larvas de espécies diferentes podem apresentar uma morfologia muito similar. Por esse motivo, destaca-se a necessidade do desenvolvimento de estudos taxonômicos e descritivos de girinos que forneçam diagnósticos confiáveis para o reconhecimento das espécies (Rossa-Feres *et al.* 2015; Pezzuti *et al.* 2021). Dessa forma, tornando mais viável o desenvolvimento do conhecimento acerca desse estágio de vida dos anuros.

Estudos descritivos sobre morfologia e história natural de girinos são ferramentas importantes para elucidar processos ecológicos e evolutivos das espécies (Wogel *et al.* 2000). Os girinos também têm se revelado um importante complemento para o levantamento de informações sobre a biota local e estudos sobre a diversidade de espécies, isto devido ao fato de serem mais facilmente amostrados comparados à fase adulta (Mascarenhas *et al.* 2015, Pezzuti *et al.* 2021). Além disso, podem auxiliar na investigação e resolução de problemas taxonômicos e filogenéticos (Altig & McDiarmid, 1999; Dias *et al.* 2021).

O presente panorama, evidencia que o conhecimento sobre girinos ainda está em crescimento e que preencher as lacunas existentes é fundamental para compreensão biológica e taxonômica de muitas espécies. Nesse sentido, o presente trabalho tem como objetivo a descrição da fase larval de três espécies: *Thoropa bryomantis*, *Physalaemus feioi* e *Leptodactylus barrioi*. Contribuindo assim com conhecimento sobre os girinos da Zona da Mata de Minas Gerais.

2. REFERÊNCIAS BIBLIOGRÁFICA

Altig, R. & McDiarmid, R.W. (1999) Tadpoles: The Biology of Anuran Larvae. The University of Chicago Press, Chicago.

Andrade, G.V, Eterovick, P.C, Rossa-Feres, D.C. & Schiesari L.. 2007. Estudos sobre girinos no Brasil: histórico, conhecimento atual e perspectivas; pp.127-145. In: L.B. Nascimento & M.E. Oliveira (Eds.), *Herpetologia no Brasil II*. Sociedade Brasileira de Herpetologia, Belo Horizonte

Dias, P.H. dos S, Vera Candioti, F., Sabbag, A.F., Colaço, G., Da Silva, H.R., Haddad, C.F.B., Carvalho-e-Silva, A.M.P.T. & Grant, T. (2021). Life on the edge: Tadpoles of Cycloramphidae (Amphibia; Anura), anatomy, systematics, functional morphology, and comments on the evolution of semiterrestrial tadpoles. *Journal of Zoological Systematics and Evolutionary Research*, 59(6), 1297–1321.

Dubeux, M.J.M., Nascimento, F.A.C.D., Lima, L.R., Magalhães, F.D.M., Silva, I.R.S.D., Gonçalves, U., ... & Mott, T. (2020). Morphological characterization and taxonomic key of tadpoles (Amphibia: Anura) from the northern region of the Atlantic Forest. *Biota Neotropica*, *20*.

Guerra V., Jardim L., Llusia D., Márquez R., Bastos R.P. 2020. Knowledge status and trends in description of amphibian species in Brazil. Ecological Indicators 118:106754.

Mascarenhas, L., Tiso, C., Linares, A.M., de Moura, C.F.O., Pezzuti, T.L., Leite, F.S.F. & Eterovick, P.C. 2015. Improved local inventory and regional contextualization for anuran diversity assessment at an endangered habitat in southeastern Brazil. *Journal of Natural History* 50:1265–1281.

Pezzuti, T.L., Leite, F.S.F., Rossa-Feres, D.D.C., & Garcia, P.C.A. (2021). The tadpoles of the Iron Quadrangle, Southeastern Brazil: a baseline for larval knowledge and anuran conservation in a diverse and threatened region. *South American Journal of Herpetology*, 22(sp1), 1–107.

Provete, D.B., Garey, M.V., da Silva, F.R. & Jordani, M.X. 2012. Knowledge gaps and bibliographical revision about descriptions of freeswimming anuran larvae from Brazil. *North-Western Journal of Zoology* 8:283–286.

Rossa-Feres, D.C., Venesky, M., Nomura, F., Eterovick, P.C., Vera Candioti, M.F., Menin, M., Juncá, F.A., Schiesari, L.C., Haddad, C.F.B., Garey, M.V., dos Anjos, L.A. &

Wassersug, R. 2015. Taking tadpole biology into the 21st century: a consensus paper from the First Tadpoles International Workshop. *Herpetologia Brasileira* 4:48–59.

Segalla, M.V., Berneck, B., Canedo, C., Caramaschi, U., Cruz, C.A.G., Garcia, P.C.A., Grant, T., Haddad, C.F.B., Lourenço, A.C.C., Mângia, S., Mott, T., Nascimento, L.B., Toledo, L.F., Werneck, F.P. & Langone, J.A. 2021. List of Amphibians. *Herpetologia Brasileira* 10:121–216.

Wogel, H., Abrunhosa, P.A & Pombal JR., J.P. Descrição dos girinos de cinco espécies de anuros do sudeste do Brasil (Amphibia, Anura: Hylidae, Leptodactylidae, Microhylidae). *Boletim do Museu Nacional* (N.S.) Zoologia, Rio de Janeiro, v. 427, p. 1–16. 2000.

2. ARTIGOS

2.1 ARTIGO I - Fraga, L. S., Guimarães, C. S., Assis, C. L., Pezzuti, T. L. & Feio, R. N. (2023) The tadpole of *Thoropa bryomantis* (Anura: Cycloramphidae): external and oral internal morphology.

2.2 ARTIGO II - Fraga, L. S., Assis, C. L.Guimarães, C. S., & Feio, R. N. (2022). The tadpole of *Physalaemus feioi* (Anura: Leptodactylidae). *Zootaxa*, *5190*(3), 447–450.

2.3 ARTIGO III - Fraga, L. S., Assis, C. L., & Feio, R. N. (2022). The tadpole of *Leptodactylus barrioi* from the Atlantic Forest of southeastern Brazil (Amphibia: Anura: Leptodactylidae). *Zootaxa*, *5168*(1), 97–100.

2.1 ARTIGO I - Fraga, L. S., Guimarães, C. S., Assis, C. L., Pezzuti, T. L. & Feio, R. N. (2023) The tadpole of *Thoropa bryomantis* (Anura: Cycloramphidae): external and oral internal morphology.

The tadpole of *Thoropa bryomantis* (Anura: Cycloramphidae): external and oral internal morphology

LETÍCIA SOARES FRAGA[1*], CARLA SILVA GUIMARÃES[2], CLODOALDO LOPES ASSIS[1], TIAGO LEITE PEZZUTI[3] & RENATO NEVES FEIO[1]

[1] *Programa de Pós-graduação em Biologia Animal, Departamento de Biologia Animal, Museu de Zoologia João Moojen, Universidade Federal de Viçosa, Viçosa, MG, Brazil.*

[2] *Pós-graduação em Ecologia e Conservação, Instituto de Biociências, Universidade Federal do Mato Grosso do Sul, Campo Grande, MS, Brazil.*

[3] *Laboratório de Herpetologia, Departamento de Zoologia, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil.*

[*]*Corresponding author: leticiasoaresfraga97@gmail.com*

Thoropa Cope is a genus of frogs belonging to the family Cycloramphidae, whose larval stage is associated with rocky environments (Assis *et al.* 2021; Dias *et al.* 2020). Currently, seven species are recognized for this genus: *T. miliaris* (Spix, 1824), *T. taophora* (Miranda-Ribeiro, 1923), *T. megatympanum* Caramaschi & Sazima, 1984, *T. saxatilis* (Cocroft & Heyer, 1988) , *T. petropolitana* (Wandolleck, 1907), *T. lutzi* Cochran, 1938 and *T. bryomantis* Assis, Lacerda, Guimarães, Peixoto, Luna & Feio, 2021. The genus includes small and medium-sized species and are distributed in south, southeast and northeast Brazil (Frost, 2023). Among these species *T. bryomantis*, previously identified as *T. lutzi*, was recently described based on acoustic and morphological characters, occupying the northern region of the Serra da Mantiqueira in the Atlantic Forest domain in Southeast Brazil (Assis *et al.* 2021). However, the larval characteristics of this species are still unknown. Seeking to fill this gap, we describe here the tadpole of *T. bryomantis* and compare them with congener species.

The tadpoles were collected in the state of Minas Gerais, southeastern Brazil, in the type locality Serra do Sapecado, Glória district, Cataguases municipality (21° 14' 13.08" S, 42° 44' 08.04" W; 758 m a.s.l., Datum WGS 84) and Antônio Prado de Minas municipality (20° 58' 33.03" S, 42° 9' 56.88" W; 758 m a.s.l, Datum WGS 84). Species identity was confirmed in laboratory by raising the tadpoles until metamorphic stage. We euthanized the specimens with 5% lidocaine, preserved them in 10% formalin, and deposited them in the Coleção Herpetológica do Museu de Zoologia João Moojen (MZUFV 323–324) of the Universidade Federal de Viçosa (UFV).

External morphology descriptions, measurements, and proportions were based on 16 tadpoles in stages 32–42 (sensu Gosner 1960) (MZUFV 323–324). Measurements and terminology follows Altig & McDiarmid (1999) for TL (total length), BL (body length), TAL (tail length), IND (internarial distance), IOD (interorbital distance), and MTH (maximum tail height); Lavilla & Scrocchi (1986) for BW (body width), BH (body height), END (eye-nostril distance), ED (eye diameter), ND (narial diameter), and ODW (oral disc width); Grosjean (2005) for DFH (dorsal fin height) and VFH (ventral fin height). TL and TAL were measured using analogue caliper to the nearest 0.01 mm. Other measurements were made in a stereomicroscope with an ocular micrometer (Olympus SZ61).

For the description of internal oral morphology two tadpoles at stage 39 were dissected and prepared following the protocol presented in Conte *et al.* (2007). Images were obtained with a Zeiss model Leo 1430 VP scanning electronic microscope (SEM). Terminology for buccopharyngeal morphology follows Wassersug (1976). Comparison of external morphology with other described tadpoles of *Thoropa* was based on literature available (*T. miliaris*, *T. taophora*, *T. megatympanum*, *T. saxatilis*, *T. petropolitana* e *T. lutzi* – Dias *et al.* 2021; *T. megatympanum* – Caramaschi & Sazima, 1984, Eterovick & Sazima, 2020; *T. taophora* – Moura *et al.* 2019 and *T. miliaris* – Pezzuti *et al.* 2021). Comparison of internal oral morphology was based on Dias *et al.* (2021).

Tadpole description. Body depressed (BH/BW = 0.665–0.669), elliptical in dorsal view, round and depressed in lateral view (Fig. 1A–B). Snout oval in dorsal view and rounded in lateral view; nostrils rounded, dorsally located, without projections. Eyes medium-sized (ED/BL = 0.158–0.166), absent meniscus, dorsally located and dorsolaterally directed. Spiracle sinistral, with ventral opening in 2/3 of the body length, indistinct walls. Intestine coiled, with center of inflection away from the center of the body to the left side (Fig. 1C). Vent tube medial, posteroventrally directed, short, covered by the abdominal flap. Abdominal flap well-developed, extending from the spiracle region to the ventral tube, posterior margin slightly bilobate (Fig. 1C–D). Oral disc medium-sized (ODW/BW = 0.332–0.791), ventrally positioned, emarginated ventrally; a single row of elongated and conical marginal papillae laterally and in alternate disposition ventrally, with a wide anterior gap; submarginal papillae absent; LTRF 2(2)/3(1). All rows long, with approximately the same length (P3 is the smallest); jaw sheaths present, serrate, compressed laterally; anterior jaw sheath arc-shaped and posterior jaw sheath U-shaped (Fig. 1E). Tail narrow (MTH/BH = 0.51–0.629), robust musculature, acute termination. Dorsal and ventral fin very narrow (DFH/MTH = 0.036–0.169, VFH/MTH =

0.071–0.271); dorsal fin emerging on posterior third of the tail; ventral fin forming a groove along the entire tail. Measurements of the available developmental stages are shown in Table 1.

Coloration. In life, general pattern brownish with a background greenish. Body marble with irregular dark brown spots and triangular dark spot between eyes; iris black; ventral surface of the body translucent cream, with few melanophores (Fig. 1A–D). Tail musculature with about eight brown triangular spots (reversed) along its lateral length, and cream lozenges in dorsal view separated by brown bars. Fins with few scattered brown spots (Fig. 1A). In formalin 10%, the coloration becomes opaque.

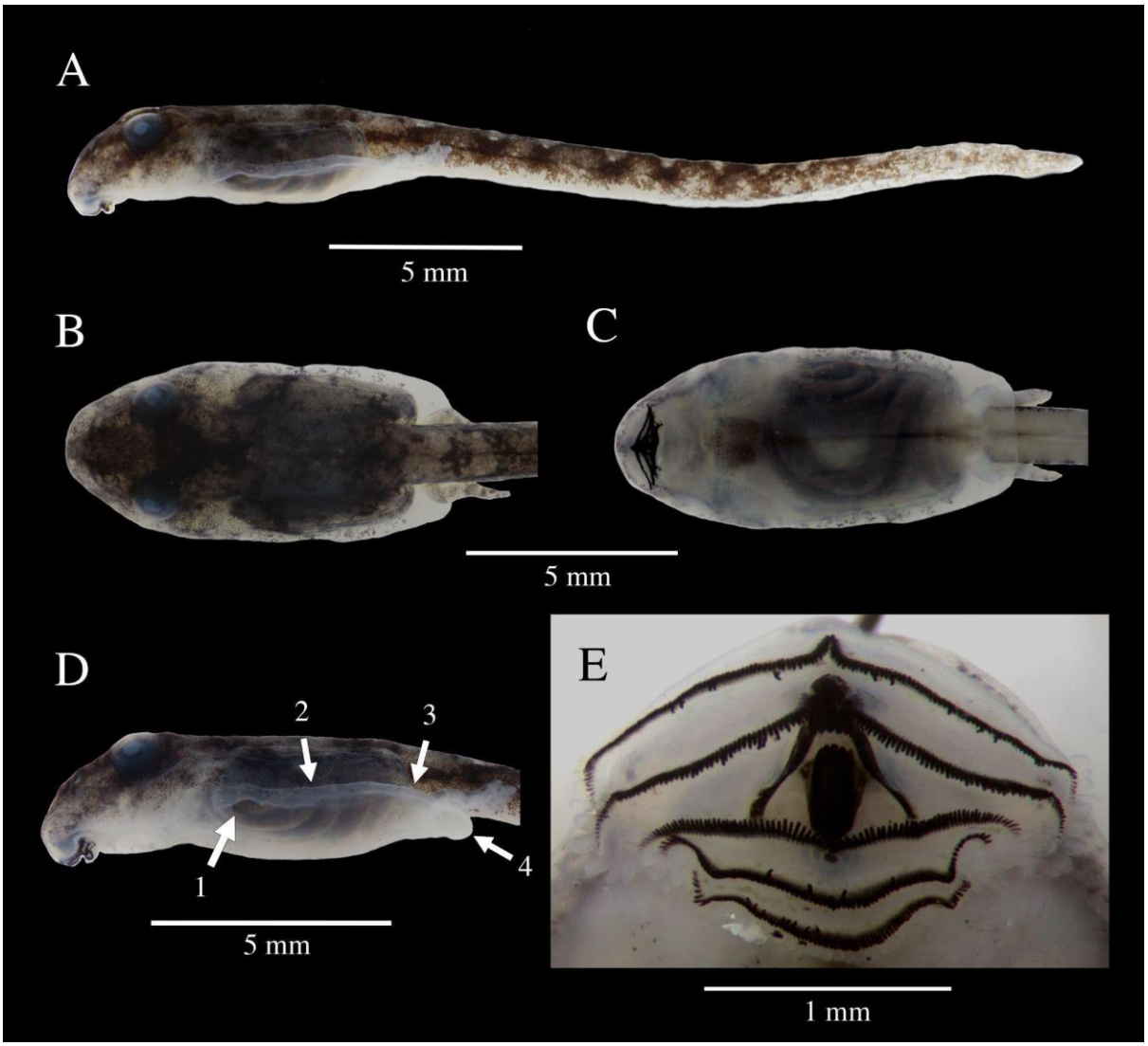

FIGURE 1. Tadpole of *Thoropa bryomantis* (MZUFV 324) at stage 36: (A) lateral, (B) dorsal, (C) ventral views; (D) details of the well-developed abdominal flap indicated by numbers 1–4; (E) oral disc.

Oral internal morphology. Buccal floor triangular shaped (Fig. 2A). Infralabial papillae arranged on a fleshy projection, an anterior pair of short, conical papillae and four posterior papillae, arranged side by side; the lateral papillae are long and bifid and the medial ones are conical and trifurcated (Fig. 2B). Lingual bud circular entirely covered by about 190 pustules; four large, conical lingual papillae, arranged medially on the lingual bud (Fig. 2B). Buccal floor arena rectangular, with approximately 48 pustulations concentrated posteriorly, delimited by seven pairs of large papillae; the three previous pairs larger and more robust; the third being trifurcated with a wide base (Fig. 2C). Buccal pockets narrow, shallow, arranged transversely. Ventral velum continuous and closed, laterally smooth, medially wavy with nine to 10 projections and median notch; glandular zone present laterally and in the projections of the velum (Fig.2C). Buccal roof triangular shaped (Fig. 2D). Prenarial arena narrow, without papillae. Choanae small, elliptical, oriented transversely, simple with thick and small borders (Fig. 2E). Postnarial arena triangular, with three pairs of small, rounded papillae. Median ridge trapezoid, low, upper margin with three to four papillae, the larger lateral ones with three small projections and the medial one in the shape of a peak; two pairs of papillae lateral to the medial ridge, branched with three to four small projections, transversely oriented (Fig. 2E). Buccal roof arena elliptical, filled with about 30 pustules and laterally delimited by seven to eight large, cylindrical lateral papillae with two to four projections; the arena is delimited posteriorly by small flattened and conical papillae and about 10 pustules that form an arch facing the anterior region of the buccal roof (Fig. 2F). Dorsal velum discontinuous and open, margins short and wavy with four projections on each side, medially interrupted; glandular zone present in the undulations and projections of the dorsal velum (Fig. 2F).

Variation. Tadpoles from both lots and localities are very similar, showing little intraspecific variation. The body in dorsal view may be ovoid in more advanced larval stages, three of 16 specimens (19%). The labial tooth row formula can be 2(1-2)/3(1), with a slight gap in A1, six of 16 specimens (38%). The lingual bud can vary from circular to oval and the medial ridge can vary from trapezoidal to elliptical, with three to four papillae.

Natural history notes. The tadpoles of *T. bryomantis* were found on humid rock walls with the presence of bryophytes in the middle of fields of granite outcrops above 600m. According to Assis *et al.* 2021 in the municipality of Antônio Prado de Minas, males were recorded during reproductive activity in areas adjacent to rocky outcrops, including pastures for cattle. The species occurs in sympatry with *Scinax cosenzai* Lacerda, Peixoto & Feio, 2012 and its congener *T. miliaris* (Assis *et al.* 2021).

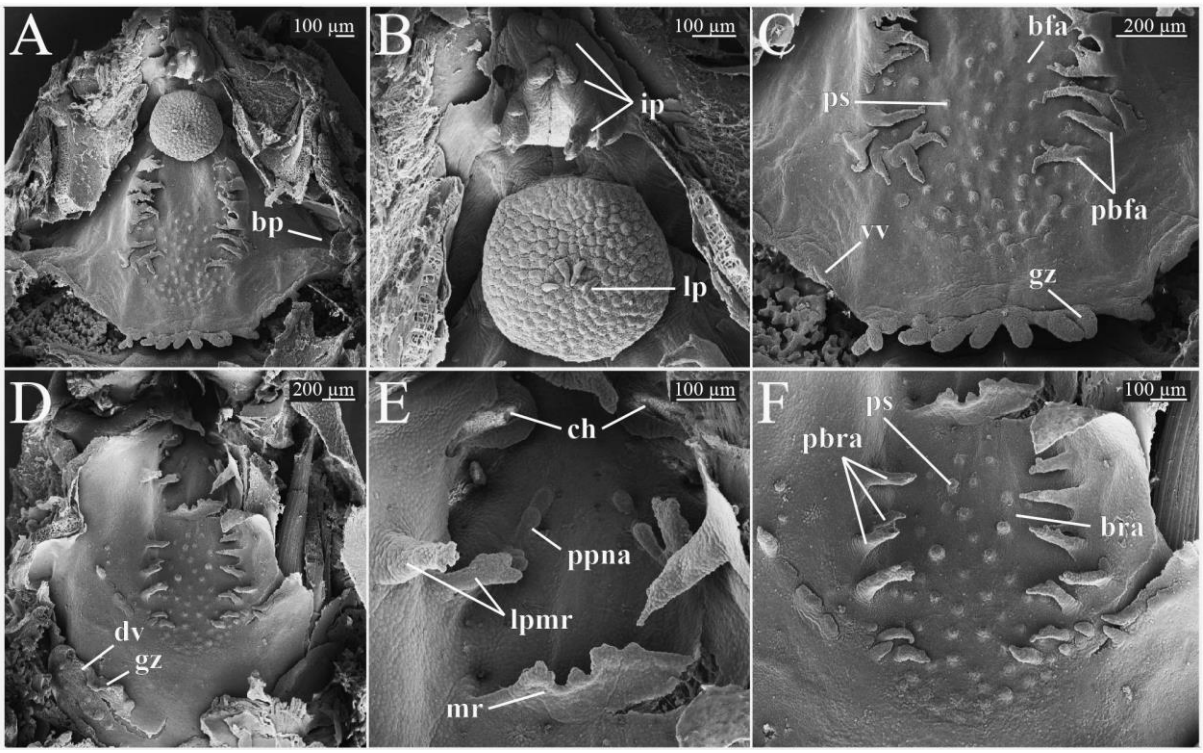

FIGURE 2. Internal oral morphology of *Thoropa bryomantis* tadpole (MZUFV 324) at stage 39: (A) Buccal floor; (B) Lingual region of buccal floor; (C) Buccal floor arena and posterior region; (D) Buccal roof; (E) Arena postnarial; (F) Buccal roof arena. bp = buccal pocket; ip = infralabial papillae; lp = lingual papillae; ps = pustulations; bfa = buccal floor arena; pbfa = papillae of buccal floor arena; vv = ventral velum; gz = glandular zone; dv = dorsal velum; ch = choanae; ppna = papillae of postnarial arena; lpmr = lateral ridge papillae; mr = median ridge; pbra = papillae of buccal roof arena; bra = buccal roof arena.

Comparison with other species. Tadpoles of the genus *Thoropa* have a very conservative morphology, with similarities such as body shape and snout; eyes prominent, dorsally located and dorsolaterally directed, nostrils dorsally located, without projections; shape and position of the intestine; presence of the abdominal flap; oral disc features, including labial tooth row formula and laterally compressed jaw sheaths; tail low with robust musculature; fins low and ventral modified into grooves. Such similarities also extend to the internal oral morphology in relation to the shape of the buccal floor and roof, characteristics of the buccal pockets, dorsal and ventral velum, prenasal arena, choanas, postnarial arena and median ridge. The distinction between congener species and *T. bryomantis* is possible because they have large eyes and meniscus present (medium eyes and meniscus absent in *T. bryomantis*); lateral spiracle (ventral in *T. bryomantis*); poorly developed abdominal flap in *T. miliaris* and *T.*

megatympanum, well-developed in *T. petropolitana* and *T. lutzi* with deeply bilobate posterior margin (well-developed and slightly bilobate in *T. bryomantis*); ventral tube free of ventral fin in *T. miliaris* and *T. megatympanum*; attached only to ventral fin in *T. saxatilis* and *T. petropolitana* (attached to ventral fin and abdominal flap in *T. bryomantis*); ventral fin insertion at the end of the tail in *T. taophora* (at the level of the ventral tube in *T. bryomantis*); dorsal-fin insertion mid-length of tail in *T. miliaris* and *T. megatympanum* (final third of tail in *T. bryomantis*). Regarding the internal oral morphology, congeneric tadpoles have two pairs of infralabial papillae (five pairs of papillae arranged under a fleshy projection in *T. bryomantis*); bell-shaped lingual bud (circular or oval in *T. bryomantis*) and the buccal floor arena delimited by 10 pairs of papillae (seven pairs in *T. bryomantis*). Congenerous larvae have a single pair of papillae lateral to the median ridge (two pairs in *T. bryomantis*); buccal roof arena without pustules and delimited by five to six pairs of papillae (arena filled with pustules and delimited by seven to eight pairs of papillae in *T. bryomantis*); dorsal velum without projections and gland zone not evident (four projections on each side and gland zone evident in *T. bryomantis*).

TABLE 1. Mean, standard deviation and range, for morphometric measurements (in mm) of tadpoles of *Thoropa bryomantis*, from Antônio Prado de Minas and Cataguases, Minas Gerais state, Brazil.

Characters	Stage 32–36 (n=7)	Stage 38–42 (n=9)
Total length	23.12 ± 03.88 (17.85–27.18)	26.23 ± 01.54 (23.44–28.36)
Body length	07.05 ± 00.72 (06.00–07.98)	08.05 ± 00.42 (07.10–08.44)
Body width	03.99 ± 00.61 (03.13–04.61)	04.87 ± 00.49 (04.02–05.41)
Body height	02.66 ± 00.32 (02.17–03.05)	03.09 ± 00.35 (02.46–03.60)
Tail length	16.22 ± 03.06 (12.10–19.53)	18.29 ± 01.27 (01.34–20.48)
Maximum tail height	01.55 ± 00.12 (01.36–01.73)	01.68 ± 00.09 (01.55–01.84)
Dorsal fin height	00.13 ± 00.07 (00.07–00.28)	00.19 ± 00.09 (00.05–00.31)
Ventral fin height	00.20 ± 00.09 (00.09–00.34)	00.29 ± 00.11 (00.13–00.50)
Eye diameter	01.09 ± 00.12 (00.95–01.26)	01.18 ± 00.09 (01.06–01.40)
Interorbital distance	02.40 ± 00.23 (02.07–02.64)	02.79 ± 00.32 (02.24–03.13)
Internarial distance	01.21 ± 00.25 (00.88–01.49)	01.15 ± 00.26 (00.77–01.59)
Eye-nostril distance	01.32 ± 00.11 (01.19–01.52)	01.55 ± 00.13 (01.28–01.75)
Oral disc width	02.12 ± 00.21 (01.80–02.48)	02.09 ± 00.18 (01.83–02.47)

Thanks to the National Center for Research and Conservation of Reptiles and Amphibians (RAN) for the collection permit (#54666–6); National Council for Scientific and Technological Development (CNPq) and Coordination for the Improvement of Higher Education Personnel (CAPES) for financial support and grants, and the Microscopy and Microanalysis Nucleus (CCB/UFV) for using the SEM and auxiliary equipment.

References

Altig, R. & McDiarmid, R.W. (1999) Body plan, development and morphology. In: Tadpole: The Biology of Anuran Larvae. University of Chicago Press, Chicago and London, pp. 24–51.

Assis, C. L., Lacerda, J. V. A., Guimares, C. S., Peixoto, M. A., Luna, M. C., & Feio, R. N. (2021). A new species of *Thoropa* Cope, 1865 (Anura, Cycloramphidae) from the Serra da Mantiqueira, Southeast Brazil. *Zootaxa*, *4995*(3), 505–522.

Caramaschi, U & Sazima, I. (1984) Uma nova espécie de Thoropa da Serra do Cipó, Minas Gerais, Brasil (Amphibia, Leptodactylidae). *Revista Brasileira de Zoologia*, v. 2, n. 3, p.139–146.

Conte, C.E., Rossa-Feres, D.C., Nomura, F., Haddad, C.F.B. & D'Heursel, A. & (2007). The tadpole of *Scinax catharinae* (Anura: Hylidae) with description of the internal oral morphology, and a review of the tadpoles from Scinax catharinae group. *Amphibia-Reptilia*, 28, 177–192.

Dias, P. H. dos S, Vera Candioti, F., Sabbag, A. F., Colaço, G., Da Silva, H. R., Haddad, C. F. B., Carvalho-e-Silva, A. M. P. T. & Grant, T. (2021). Life on the edge: Tadpoles of Cycloramphidae (Amphibia; Anura), anatomy, systematics, functional morphology, and comments on the evolution of semiterrestrial tadpoles. *Journal of Zoological Systematics and Evolutionary Research*, *59*(6), 1297–1321.

Eterovick, P. C., de Souza, A. M., & Sazima, I. (2020). Anuran Amphibians from the Serra do Cipó. *Gráfion Estúdio Editorial, Brazil*.

Frost, D.R. (2023) Amphibian Species of the World: an Online Reference. Version 6.0. Available from: http://research.amnh. org/herpetology/amphibia/index.html (accessed 18 Jan 2023).

Gosner, K.L. (1960) A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica*, 16, 183–190.

Grosjean, S. (2005) The choice of external morphological characters and developmental stages for tadpole-based anuran taxonomy, a case study in *Rana* (Sylvirana) *nigrovittata* (Blyth, 1855) (Amphibia, Anura, Ranidae). *Contributions to Zoology*, 74, 61–76.

Lavilla, E.O. & Scrocchi, G.J. (1986) Morfometría larval de los géneros de Telmatobiinae (Anura, Leptodactylidae) de Argentina y Chile. *Physis*, 44, 39–43.

Moura, P. H. A. G., SJ, D. R. A., Correa, C. N., & Nunes, I. V. A. N. (2019). Redescription of the tadpole of *Thoropa taophora* (Miranda-Ribeiro)(Anura: Cycloramphidae). *Zootaxa*, *4656*(2), zootaxa–4656.

Pezzuti, T. L., Leite, F. S. F., Rossa-Feres, D. D. C., & Garcia, P. C. A. (2021). The tadpoles of the Iron Quadrangle, Southeastern Brazil: a baseline for larval knowledge and anuran conservation in a diverse and threatened region. *South American Journal of Herpetology*, *22*(sp1), 1–107.

Wassersug, R.J. (1976) Oral morphology of anuran larvae: terminology and general description. *Occasional papers of the Museum of Natural History*, 48, 1–23

2.2 ARTIGO II - Fraga, L. S., Assis, C. L., Guimarães, C. S., & Feio, R. N. (2022). The tadpole of *Physalaemus feioi* (Anura: Leptodactylidae). *Zootaxa*, *5190*(3), 447–450.

Zootaxa 5190 (3): 447–450
https://www.mapress.com/zt/

Correspondence

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)
https://doi.org/10.11646/zootaxa.5190.3.9
http://zoobank.org/urn:lsid:zoobank.org:pub:B42FB1A2-EDB3-44A2-8975-E2A979D1EDB5

The tadpole of *Physalaemus feioi* (Anura: Leptodactylidae)

LETÍCIA SOARES FRAGA[1*], CLODOALDO LOPES ASSIS[1,3], CARLA SILVA GUIMARÃES[2] & RENATO NEVES FEIO[1,4]

[1]*Programa de Pós-graduação em Biologia Animal, Departamento de Biologia Animal, Museu de Zoologia João Moojen, Universidade Federal de Viçosa, 36570–900, Viçosa, MG, Brazil.*
[2]*Pós-graduação em Ecologia e Conservação, Instituto de Biociências, Universidade Federal do Mato Grosso do Sul, 79070–900, Campo Grande, MS, Brazil.* *sguimaraes.carla@gmail.com;* *https://orcid.org/0000-0001-6514-031X*
[3] *clodoassis@yahoo.com.br;* *https://orcid.org/0000-0001-5009-2706*
[4] *rfeio@ufv.br;* *https://orcid.org/0000-0002-3607-2896*
[*]*Corresponding author.* *leticiasoaresfraga97@gmail.com;* *https://orcid.org/0000-0001-7101-1231*

The Neotropical genus *Physalaemus* Fitzinger contains 50 species and is organized into two major clades: *P. cuvieri* and *P. signifer* (Lourenço *et al.* 2015; Frost 2021). The *P. olfersii* group belongs to the *P. cuvieri* clade and contains seven species: *P. olfersii* (Lichtenstein & Martens), *P. soaresi* Izecksohn, *P. maximus* Feio, Pombal & Caramaschi, *P. feioi* Cassini, Cruz & Caramaschi, *P. lateristriga* (Steindachner), *P. orophilus* Cassini, Cruz & Caramaschi and *P. insperatus* Cruz, Cassini & Caramaschi. Species of this group occur in the Atlantic Forest and have the advertisement call with a pulsed note, without frequency modulation (Giaretta *et al*. 2009; Lourenço *et al*. 2015). Within this group, *P. feioi* is the species that has the highest number of records in mountainous regions of southeastern Brazil (e.g., Serra da Mantiqueira), in the states of São Paulo, Minas Gerais, and Rio de Janeiro (Rocha *et al*. 2018). This species was described after a review of a species complex associated with *P. olfersii*, but the study did not include larval stages (Cassini *et al.* 2010). Here, we describe the tadpoles of *P. feioi* based on specimens from the type locality and compare them with the species of the *P. olfersii* group and other congeneric sympatric species.

We analyzed 12 tadpoles of *Physalaemus feioi* at Stage 36 (Gosner 1960), collected at Mata da Biologia (20°75′73″ S, 42°86′08″ W, 710 m a.s.l.; Datum WGS 84), Viçosa, Minas Gerais, southeastern Brazil. We reared three specimens in an aquarium until they completed metamorphosis for confirmation of the species (Fig. 1F). We euthanized the specimens with 5% lidocaine, preserved them in 10% formalin, and deposited them in the Coleção Herpetológica do Museu de Zoologia João Moojen (MZUFV 166, 332) of the Universidade Federal de Viçosa.

We follow Altig & McDiarmid (1999) for terminology and labial tooth row formula and Pezzuti *et al*. (2021) for morphological characterization. Measurements follow Lavilla & Scrocchi (1986): body height (BH), body width (BW), eye diameter (ED), nostril diameter (ND), eye-nostril distance (END), and eye-snout distance (ESD); Altig & McDiarmid (1999): total length (TL), body length (BL), tail length (TAL), maximum tail height (MTH), interorbital distance (IOD), internostril distance (IND), and tail muscle height (TMH); Grosjean (2005): dorsal fin height (DFH) and ventral fin height (VFH); Lins *et al*. (2018): spiracle height (SH) and spiracle width (SW). We measured specimens using a stereomicroscope with an ocular micrometer (Olympus SZ61), except for TL, BL and TAL, which we measured with calipers (precision 0.1 mm).

Morphology. Body depressed (BH/BW = 0.63–0.93) and ovoid (Fig. 1A–B). Snout rounded in lateral and dorsal view. Eyes dorsal, directed dorsolaterally. Nostrils circular, medium to large size (ND/BL = 0.03–0.06), placed dorsally, with a small fleshy projection on the medial margin. Spiracle sinistral, lateral, long (SL/BL = 0.16–0.47), opening at posterior third of body, inner wall of spiracle tube fused to body. Digestive tract circularly coiled, with inflection point central in the abdominal region (Fig. 1C). Vent tube dextral, directed posteriorly, with the end free of ventral fin. Oral disc ventral, laterally emarginate; upper lip with small marginal papillae arranged alternately in a single row and a large gap in the central portion; lower lip with densely packed alternate papillae. Labial teeth arranged in LTRF 2(2)/3(1); A1 slightly shorter than A2, P1 longer than P2 and P3, P2 slightly shorter than P3; jaw sheaths serrated on margins, upper sheath M-shaped, lower sheath V-shaped, upper wider than lower (Fig. 1D). Tail of intermediate height (MTH/TAL = 0.23–0.33), similar to body height (MTH/BH = 0.71–1.03), musculature slender (TMH/BH = 0.23–0.47) almost reaching tail tip, tail

Accepted by F. Candioti: 8 Sept. 2022; published: 29 Sept. 2022

tip rounded. Dorsal fin higher than ventral fin (DFH/VFH = 1.20–3.05), both with external margin slightly convex, the dorsal fin originating on posterior third of the body and the ventral fin at the level of the vent tube.

Coloration. In life, body light brown with melanophores scattered dorsally and laterally, venter opaque; spiracle translucent; eyes with black iris, gray pupil; tail musculature cream with scattered melanophores; fins translucent with irregular melanophores denser at margins (Fig. 1E). In formalin, coloration increasingly milder and translucent and irregular spots tending to disappear over time.

Measurements. Measurements (in millimeters) are given as range (average ± standard deviation): TL: 22.28–25.54 (23.96 ± 0.92); BL: 8.85–11.84 (10.22 ± 1.00); TAL: 11.40–15.28 (13.74 ± 1.17); MTH: 3.41–4.57 (3.82 ± 0.39); BH: 4.07–4.84 (4.54 ± 0.31); BW: 5.18–6.97 (6.20 ± 0.61); ED: 0.83–1.00 (0.90 ± 0.08); ND: 0.33–0.50 (0.46 ± 0.06); IOD: 1.33–1.75 (1.62 ± 0.17); IND: 0.75–1.00 (0.89 ± 0.09); END: 0.42–0.75 (0.63 ± 0.11); ESD: 1.75–2.33 (2.07 ± 0.19); DFH: 1.00–1.83 (1.31 ± 0.28); VFH: 0.42–1.16 (0.72 ± 0.25); TMH: 1.08–2.08 (1.69 ± 0.33); SH: 1.66–4.75 (3.70 ± 1.09); SW: 0.75–1.75 (1.33 ± 0.32).

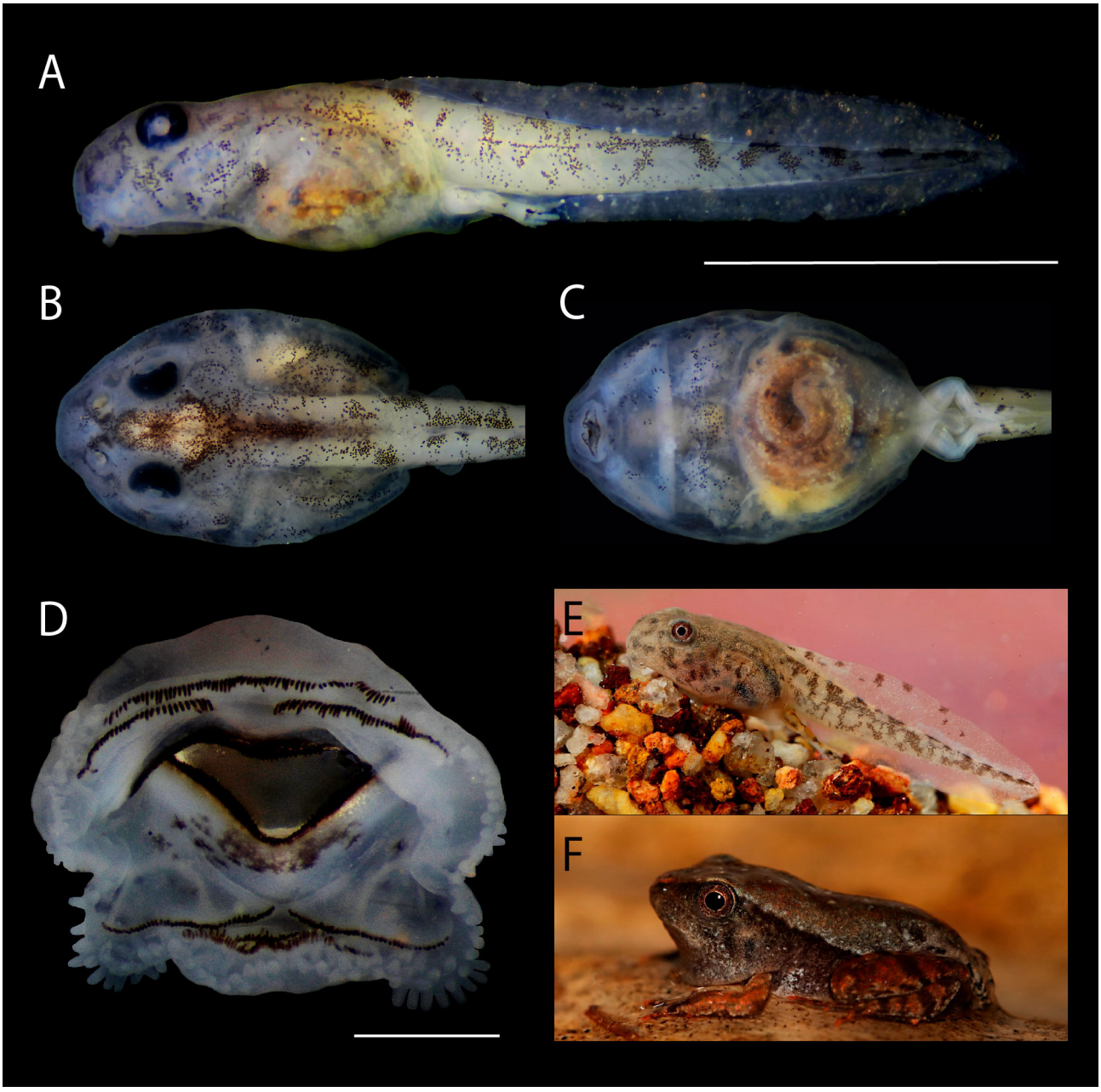

FIGURE 1. Tadpole of *Physalaemus feioi* (MZUFV332) at Stage 36: (A) lateral, (B) dorsal, (C) ventral views (scale bar = 10 mm); (D) Oral disc (scale bar = 0.5 mm). (E) Tadpole at Stage 38 photographed in life, and (F) newly-metamorphosed individual.

Comparison with other species. The tadpoles of *Physalaemus* are in general similar in shape and coloration of the body, shape and position of eyes and nostrils, arrangement of digestive tract and shape of spiracle (e.g., Nascimento *et al.* 2005). Major variations concern the oral disc, and tadpoles of the *P. olfersii* group share an oral configuration with LTRF 2/3 and ventral papillae complete with tadpoles of the *P. gracilis* group and the *P. signifer* clade (e.g., Vera Candioti *et al.* 2011). Some interspecific differences within the group are the snout shape (slightly truncated in *P. orophilus*), eye position (dorsolateral in *P. orophilus*, *P. soaresi*, and *P. maximus*), spiracle position (laterodorsal in *P. olfersii*;), oral disc position (anteroventral in *P. orophilus*, *P. maximus*, *P. olfersii*, and *P. lateristriga*), and caudal fin height (low fins in *P. olfersii* and *P. lateristriga*) (Weber *et. al.* 2005; Baêta *et. al.* 2007; Giaretta *et al.* 2009; Pezzuti *et al.* 2019; Mello *et al.* 2021). Tadpoles of *P. cuvieri*, *P. signifer* and *P. marmoratus* can be found in the same environments than *P. feioi* (Brassaloti *et al.* 2010; Moura *et al.* 2012; Lacerda *et al.* 2014), but larvae can be easily differentiated on the basis of oral disc configuration (ventral and ventrolateral gaps in lower papillae of *P. cuvieri*, only two lower tooth rows in *P. marmoratus*), and eye position (dorsolateral in *P. signifer*) (Weber & Carvalho-e-Silva 2001; Alves-Ferreira *et al.* 2021; Pezzuti *et al.* 2021).

The Centro Nacional de Pesquisa e Conservação de Répteis e Anfíbios (RAN) provided collection permit (#54666–6); Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) provided financial support and fellowship to LSF, CLA, CSG and RNF. We are grateful to Lucas Drummond for the translation, Eduarda Melo and Júlio Chaul for taking pictures of tadpoles.

References

Altig, R. & McDiarmid, R.W. (1999) Body plan: development and morphology. *In*: McDiarmid, R.W. & Altig, R. (Eds.), *Tadpoles: The Biology of Anuran Larvae*. *The* University of Chicago Press, Chicago, Illinois, pp. 24–51.

Alves-Ferreira, G., da Paixão, I.B.F. & Nomura, F. (2021) Morphological characterization and diversity of tadpoles (Amphibia: Anura) at Emas National Park and its surrounding, Goiás State, Brazil. *Biota Neotropica*, 21, 1–11. https://doi.org/10.1590/1676-0611-bn-2020-1178

Baêta, D., Lourenço, A.C.C., Pezzuti, T.L. & Pires, M.R.S. (2007) The tadpole, advertisement call, and geographic distribution of *Physalaemus maximus* Feio, Pombal, Caramaschi, 1999 (Amphibia, Anura, Leiuperidae). *Arquivos do Museu Nacional*, 65, 27–32.

Brassaloti, R.A., Rossa-Feres, D.D.C. & Bertoluci, J. (2010) Anurofauna da floresta estacional semidecidual da Estação Ecológica dos Caetetus, sudeste do Brasil. *Biota Neotropica*, 10, 275–291. https://doi.org/10.1590/S1676-06032010000100024

Cassini, C., Cruz, C.A.G. & Caramaschi, U. (2010) Taxonomic review of *Physalaemus olfersii* (Lichtenstein, Martens, 1856) with revalidation of *Physalaemus lateristriga* (Steindachner, 1864) and description of two new related species (Anura: Leiuperidae). *Zootaxa*, 2491 (1), 1–33. https://doi.org/10.11646/zootaxa.2491.1.1

Frost, D.R. (2021) Amphibian Species of the World: an Online Reference. Version 6.0. Available from: http://research.amnh.org/herpetology/amphibia/index.html (accessed 21 May 2021)

Giaretta, A.A., Martins, L.B. & Santos, M.P. (2009) Further notes on the taxonomy of four species of *Physalaemus* (Anura, Leiuperidae) from the Atlantic Forest of Southeastern Brazil. *Zootaxa*, 2266 (1), 51–60. https://doi.org/10.11646/zootaxa.2266.1.3

Gosner, K.L. (1960) A simplified table for staging anuran embryo and larvae with notes on identification. *Herpetologica*, 16, 183–190.

Grosjean, S. (2005) The choice of external morphological characters and developmental stages for tadpole-based anuran taxonomy: a case study in *Rana* (*Sylvarana*) *nigrovittata* (Blyth, 1855) (Amphibia, Anura, Ranidae). *Contributions to Zoology*, 74, 61–76. https://doi.org/10.1163/18759866-0740102005

Lacerda, J.V.A., Dayrell, J.S., Pires, D.M.V. & Feio, R.N. (2014) Anfíbios da Mata do Paraíso. *In:* Lima, G.S., Ribeiro, G.A., Gonçalves, W., Martins, S.V. & Almeida, M.P. (Eds.), *Ecologia de Mata Atlântica*. *Vol. 1. Estudos ecológicos na Mata do Paraíso.* Supremo, Viçosa, pp. 133–152.

Lavilla, E.O. & Scrocchi, G.J. (1986) Morfometría larval de los géneros de Telmatobiinae (Anura: Leptodactylidae) de Argentina y Chile. *Physis*, 44, 39–43.

Lins, A.C.R., De Magalhães, R.F., Costa, R.N., Brandão, R.A., Py-Daniel, T.R., Miranda, N.E.O, Maciel, N.M., Nomura, F. & Pezzuti, T.L. (2018) The larvae of two species of *Bokermannohyla* (Anura, Hylidae, Cophomantini) endemic to the highlands of central Brazil. *Zootaxa*, 4527 (4), 501–520. https://doi.org/10.11646/zootaxa.4527.4.3

Lourenço, L.B., Targueta, C.P., Baldo, D., Nascimento, J., Garcia, P.C.A., Andrade, G.V., Haddad, C.F.B. & Recco-Pimentel, S.M. (2015) Phylogeny of frogs from the genus *Physalaemus* (Anura, Leptodactylidae) inferred from mitochondrial and nuclear gene sequences. *Molecular Phylogenetics and Evolution*, 92, 204–216. https://doi.org/10.1016/j.ympev.2015.06.011

Mello, C.M., Gonçalves, D.D.S., Rossa-Feres, D.D.C. & Conte, C.E. (2021) The tadpole of *Physalaemus lateristriga* (Steindachner, 1864) (Anura: Leptodactylidae): external and oral internal morphology. *Zootaxa*, 4948, 447–450. https://doi.org/10.11646/zootaxa.4948.3.9

Moura, M.R., Motta, A.P., Fernandes, V.D. & Feio, R.N. (2012) Herpetofauna da Serra do Brigadeiro, um remanescente de Mata Atlântica em Minas Gerais, sudeste do Brasil. *Biota Neotropica*, 12, 209–235. https://doi.org/10.1590/S1676-06032012000100017

Nascimento, L.B., Caramaschi, U. & Cruz, C.G. (2005) Taxonomic review of the species groups of the genus *Physalaemus* Fitzinger, 1826 with revalidation of the genera *Engystomops* Jiménez-de-la-Espada, 1872 and *Eupemphix* Steindachner, 1863 (Amphibia, Anura, Leptodactylidae). *Arquivos do Museu Nacional*, 63, 297–320.

Pezzuti, T.L., Leite, F.S.F., Silva, D.H., Lourenço, A.C.C. & Baeta, D. (2019) The tadpole of *Physalaemus orophilus* from the Atlantic rainforest of southeastern Brazil (Amphibia, Anura, Leptodactylidae). *Zootaxa*, 4629 (1), 141–145. https://doi.org/10.11646/zootaxa.4629.1.11

Pezzuti, T.L., Leite, F.S.F., Rossa-Feres, D.D.C. & Garcia, P.C.A. (2021) The tadpoles of the Iron Quadrangle, Southeastern Brazil: a baseline for larval knowledge and anuran conservation in a diverse and threatened region. *South American Journal of Herpetology*, 22, 1–107. https://doi.org/10.2994/SAJH-D-20-00042.1

Rocha, L.C.L., Assis, C.L. & Pereira, E.A. (2018) Distribution extension and new state record of *Physalaemus feioi* Cassini, Cruz & Caramaschi, 2010 (Anura: Leptodactylidae: Leiuperinae). *Herpetology Note*s, 11, 893–896.

Vera Candioti, M.F., Haad, M.B., Baldo, J.D., Kolenc, F., Borteiro, C. & Altig, R. (2011) Different pathways are involved in the early development of the transient oral apparatus in anuran tadpoles (Anura: Leiuperidae). *Biological Journal of the Linnean Society*, 104, 330–345. https://doi.org/10.1111/j.1095-8312.2011.01727.x

Weber, L.N. & Carvalho-e-Silva, S.P.C. (2001) Descrição da larva de *Physalaemus signifer* (Girard, 1853) (Amphibia, Anura, Leptodactylidae) e informações sobre a reprodução e a distribuição geográfica. *Boletim do Museu Nacional*, 462, 1–6.

Weber, L.N., Carvalho-e-Silva, S.P.C. & Gonzaga, L. (2005) The tadpole of *Physalaemus soaresi* (Anura, Leptodactyliae), with comments on taxonomy, reproductive behavior, and vocalizations. *Zootaxa*, 1072 (1), 35–42. https://doi.org/10.11646/zootaxa.1072.1.3

2.3 ARTIGO III - Fraga, L. S., Assis, C. L., & Feio, R. N. (2022). The tadpole of *Leptodactylus barrioi* from the Atlantic Forest of southeastern Brazil (Amphibia: Anura: Leptodactylidae). *Zootaxa*, *5168*(1), 97–100.

Zootaxa 5168 (1): 097–100
https://www.mapress.com/zt/

Correspondence

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)
https://doi.org/10.11646/zootaxa.5168.1.9
http://zoobank.org/urn:lsid:zoobank.org:pub:D5E8F459-0F20-4FB1-93E6-98EAEAD074B4

The tadpole of *Leptodactylus barrioi* from the Atlantic Forest of southeastern Brazil (Amphibia: Anura: Leptodactylidae)

LETÍCIA SOARES FRAGA[1*], CLODOALDO LOPES ASSIS[1,2] & RENATO NEVES FEIO[1,3]
[1]*Programa de Pós-graduação em Biologia Animal, Departamento de Biologia Animal, Museu de Zoologia João Moojen, Universidade Federal de Viçosa, 36570–900, Viçosa, MG, Brazil.*
[2] *clodoassis@yahoo.com.br; https://orcid.org/ 0000-0001-5009-2706*
[3] *rfeio@ufv.br; https://orcid.org/0000-0002-3607-2896*
[*]*Corresponding author. leticiasoaresfraga97@gmail.com; https://orcid.org/0000-0001-7101-1231*

Leptodactylus is a genus of Neotropical frogs in the family Leptodactylidae, which has 83 species (Frost 2021). With the exception of *L. hylodes* (Reinhardt & Lütken) and *L. lauramiriamae* Heyer & Crombie, the remaining species are distributed in four major clades, based on molecular analysis, adult and larval morphology and behavioral data: *L. fuscus*, *L. latrans*, *L. melanonotus,* and *L. pentadactylus* groups (de Sá *et al*. 2014; Silva *et al*. 2020). Allocated in the *L. fuscus* clade, the *L. mystaceus* complex is composed of eight species: *L. didymus* Heyer, García-Lopez & Cardoso, *L. elenae* Heyer, *L. mystaceus* (Spix), *L. notoaktites* Heyer, *L. spixi* Heyer, *L. kilombo* Alves da Silva, Magalhães, Thomassen, Leite, Garda, Brandão, Haddad, Giaretta & Carvalho, *L. watu* Alves da Silva, Magalhães, Thomassen, Leite, Garda, Brandão, Haddad, Giaretta & Carvalho, and *L. barrioi* Alves da Silva, Magalhães, Thomassen, Leite, Garda, Brandão, Haddad, Giaretta & Carvalho. The species of this complex are widely distributed in forests and open formations of South America east of the Andes and are almost indistinguishable morphologically (Silva *et al*. 2020). Among the species in this complex, *L. barrioi* occurs in the Atlantic Forest of southeastern Brazil, in the states of Minas Gerais and Espírito Santo (Silva *et al*. 2020; Frost 2021). This frog was recently described based on acoustic and molecular characters (Silva *et al*. 2020), but the characteristics of its tadpoles are unknown. Thus, here we describe the tadpole of *L. barrioi*, and compare it to the species of the *L. mystaceus* complex.

In 2018 February, we collected tadpoles of *Leptodactylus barrioi* in the municipality of Cataguases, State of Minas Gerais, in southeastern Brazil (21º20’19 “S, 42º45’43 “W, 298 m a. s. l., Datum WGS 84). This locality is the same as the genetic voucher used in the description of this species (see Silva *et al*. 2020). We reared two tadpoles in an aquarium until the end of metamorphosis to confirm the identity of the species (Fig. 1E). We euthanized the specimens with 5% lidocaine solution, preserved in 10% formalin, and deposited them in the Museu de Zoologia João Moojen (MZUFV 334–35) at the Universidade Federal de Viçosa, Minas Gerais state, Brazil.

For measurements and description, we used eight individuals, six at Stage 36 and two at Stage 37 (Gosner 1960). We follow Altig & McDiarmid (1999) and Pezzuti *et al*. (2021) for morphological characterization. Measurements follow Lavilla & Scrocchi (1986): body height (BH), body width (BW), eye diameter (ED), nostril diameter (ND), eye-nostril distance (END), and eye-snout distance (ESD); Grosjean (2005): dorsal fin height (DFH) and ventral fin height (VFH); Altig & McDiarmid (1999): total length (TL), body length (BL), tail length (TAL), maximum tail height (MTH), interorbital distance (IOD), internostril distance (IND), and tail muscle height (TMH); Lins *et al*. (2018): spiracle height (SH) and spiracle width (SW). We measured the samples using a stereomicroscope with an ocular micrometer (Olympus SZ61), except TL, BL, and TAL, which we measured with calipers (0.1 mm precision). We obtained data on the morphological characteristics of the species of the *L. mystaceus* complex from the literature descriptions.

Tadpole description. We present the measurements of the tadpoles in Table 1. The tadpole presents a depressed body (BH/BW = 0.79–0.91), rounded depressed in lateral view and elliptical in dorsal view (Fig. 1A–B). Snout rounded in lateral view and elliptical in dorsal view. Eyes dorsal and dorsolaterally directed. Nostrils circular, small to medium sized (ND/BL = 0.02–0.03), dorsally and without internal projection. Spiracle sinistral, lateral, short (SL/BL = 0.10–0.17), opening at posterior third of body, inner wall of spiracle tube present, fused to body. Lateral lines visible. Digestive tract circularly coiled, with inflection point displaced from center of abdominal region (Fig. 1C). Vent tube medial, directed posteriorly, with a small free end of ventral fin. Oral disc positioned ventrally, not emarginate, marginal papillae in single

Accepted by F. Candioti: 29 Jun. 2022; published: 20 Jul. 2022

alternate row, wide anterior gap, submarginal papillae absent, labial tooth row formula –LTRF– 2(2)/3 or 2(2)/3(1); A1 slightly shorter than A2, P1, P2 and P3 equal sized; jaw sheaths serrated on margins, upper sheath arc-shaped, lower sheath V-shaped, upper wider than lower (Fig. 1D). Tail medium height (MTH/TAL = 0.30–0.32), approximate of height body (MTH/BH = 0.95–1.12), musculature slender to slightly robust (TMH/BH = 0.41–0.52) almost reaching tail tip, tail tip acute. Dorsal fin height intermediate (DFH/TAL = 0.08–0.10), originating on posterior third of body. Ventral fin height intermediate (VFH/TAL = 0.07–0.09), originating at the level of the vent tube. Dorsal and ventral fins with margins slightly convex, the dorsal fin higher than ventral fin (DFH/VFH = 1.05–1.25).

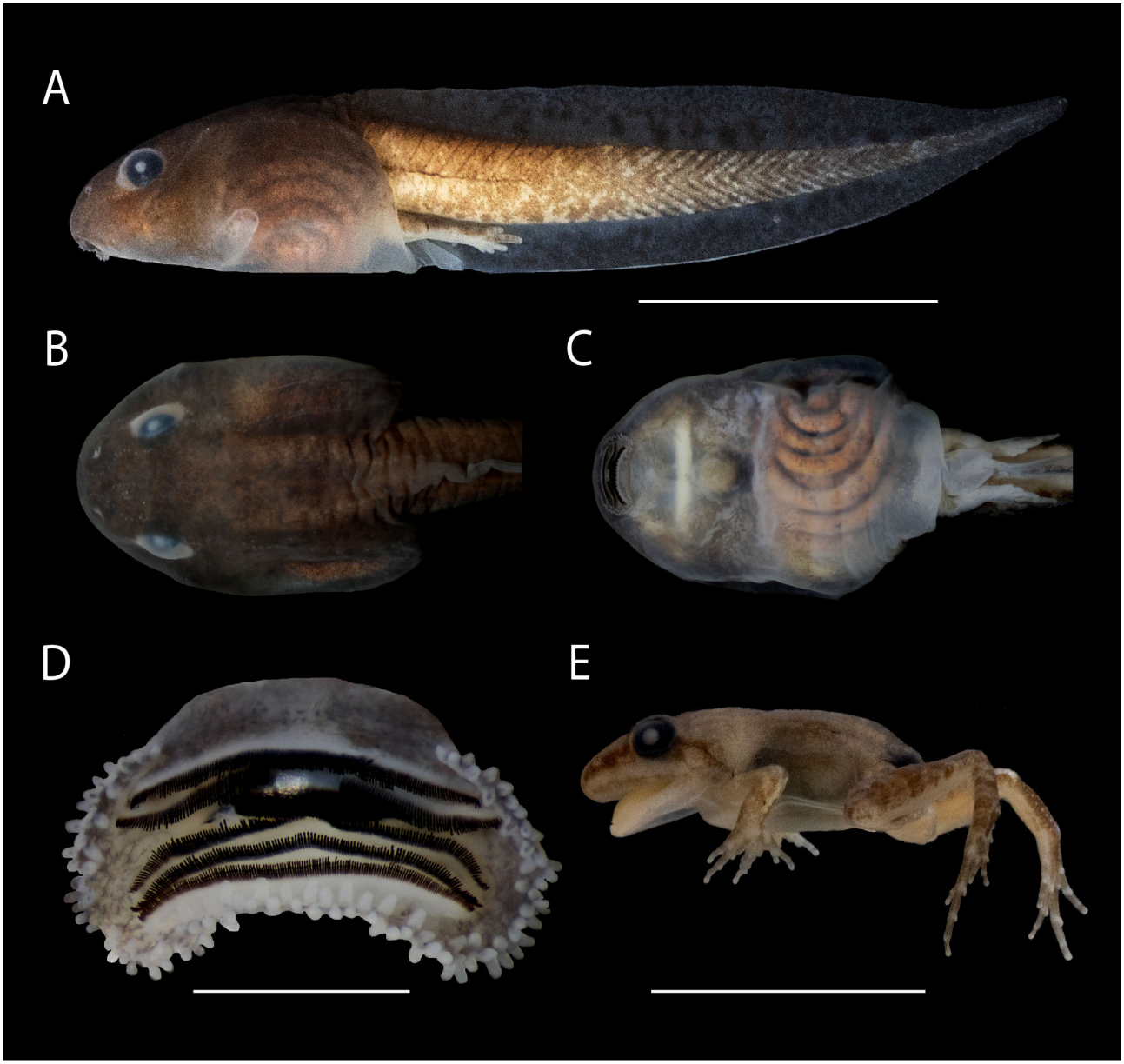

FIGURE 1. Tadpole of *Leptodactylus barrioi* (MZUFV 334) at Stage 36: (A) lateral, (B) dorsal, (C) ventral views (scale bar = 10 mm); (D) oral disc (scale bar = 1 mm); (E) newly metamorphosed individual (scale bar = 10 mm).

Coloration. In life, the body is homogeneously medium brown with small dark brown spots scattered dorsally and laterally; opaque gray venter; spiracle translucent; black iris with small gray dots, gray to white pupil. Tail musculature cream with many irregular dark brown spots, closely spaced; translucent fins with evenly dispersed dark brown irregular spots. In formalin, the coloration of the tadpoles becomes increasingly milder and translucent and over time the spots tend to disappear.

Comparison with other species of the group. The tadpoles of this genus have a very conservative morphology; tadpoles of the *L. mystaceus* complex exemplify this through similarities in body shape, vent tube position, spiracle morphology, oral disc features, and coloration. Some slight differences of *L. barrioi* regarding other species of the complex

include (1) rounded snout (truncated in *L. spixi*); (2) dorsal eyes (dorsolateral in *L. didymus*, *L. notoaktites* and *L. spixi*); (3) dorsal, rounded nostrils without projections (ovoid with projections in *L. elenae*, and dorsolateral in *L. notoaktites* and *L. spixi*; (4) ventrolateral spiracle (dorsolateral in *L. notoaktites* and *L. spixi*); (5) digestive tract coils on the left of the abdomen (central in *L. mystaceus*); (6) mid-high tail and dorsal fin (low tail and fin in *L. notoaktites*; and (7) ventral oral disc (anteroventral in *L. mystaceus*, *L. didymus*, *L. notoaktites* and *L. spixi*) (Hero 1990; Duellman 2005; Bilate *et al*. 2006; Prado & D'Heursel 2006; de Sá *et al*. 2007; Vera Candioti *et al*. 2007; Schulze *et al*. 2015). These features could be useful to distinguish tadpoles in this rather cryptic species-complex (Heyer *et al*. 1996; Silva *et al*. 2020).

TABLE 1. Mean, standard deviation and range, for morphometric measurements of *Leptodactylus barrioi* tadpoles from Cataguases, Minas Gerais state, southeastern Brazil.

Characters	Stage 36 (n = 6)	Stage 37 (n = 2)
Total length	28.67 ± 1.13 (26.68 – 29.95)	33.37 ± 2.65 (31.49 – 35.24)
Body length	9.87 ± 0.11 (9.69 – 10.02)	10.87 ± 1.32 (9.93 – 11.80)
Tail length	18.76 ± 1.03 (16.99 – 19.94)	22.57 ± 1.21 (21.71 – 23.42)
Maximum tail height	5.81 ± 0.34 (5.26 – 6.25)	6.93 ± 0.24 (6.76 – 7.10)
Body height	5. 46 ± 0.18 (5.17 – 5.72)	6.26 ± 0.14 (6.16 – 6.36)
Body width	6.53 ± 0.44 (5.97 – 7.15)	6.26 ± 0.25 (6.97 – 7.33)
Eye diameter	1.23 ± 0.11 (1.12 – 1.43)	1.49 ± 0.07 (1.44 – 1.54)
Nostril diameter	0.21 ± 0.04 (0.18 – 0.28)	0.24 ± 0.01 (0.23 – 0.24)
Interorbital distance	2.24 ± 0.12 (2.14 – 2.44)	2.74 ± 0.23 (2.57 – 2.90)
Internostril distance	1.65 ± 0.11 (1.51 – 1.78)	1.53 ± 0.06 (1.48 – 1.57)
Eye-nostril distance	1.34 ± 0.09 (1.26 – 1.48)	1.41 ± 0.10 (1.34 – 1.48)
Eye-snout distance	2.46 ± 0.13 (2.27 – 2.62)	2.26 ± 0.40 (1.98 – 2.54)
Dorsal fin height	1.78 ± 0.15 (1.60 – 1.93)	2.02 ± 0.15 (1.91 – 2.12)
Ventral fin height	1.57 ± 0.20 (1.31 – 1.82)	1.68 ± 0.03 (1.66 – 1.70)
Tail muscle height	2.47 ± 0.13 (2.26 – 2.58)	3.24 ± 0.06 (3.19 – 3.28)
Spiracle height	1.44 ± 0.12 (1.34 – 1.68)	1.11 ± 0.19 (0.97 – 1.24)
Spiracle width	0.81 ± 0.06 (0.73 – 0.89)	0.84 ± 0.16 (0.72 – 0.95)

We thank Centro Nacional de Pesquisa e Conservação de Répteis e Anfíbios (RAN) for the collection license (#54666–6); Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for financial support and fellowship conceded to LSF, CLA, and RNF. We are grateful to Eduarda Melo and Júlio Chaul for helping with the pictures of the tadpoles and Lucas Drummond for English review.

References

Altig, R. & McDiarmid, R.W. (1999) Body plan: development and morphology. *In*: McDiarmid, R.W. & Altig, R. (Eds.). *Tadpoles: The Biology of Anuran Larvae.* The University of Chicago Press, Chicago, Illinois, pp. 24–51.

Bilate, M., Wogel, H., Weber, L.N. & Abrunhosa, P.A. (2006) Vocalizações e girino de *Leptodactylus spixii* Heyer, 1983 (Amphibia, Anura, Leptodactylidae). *Arquivos do Museu Nacional, Rio de Janeiro*, 64, 235–245.

de Sá, R.O., Grant, T., Camargo, A., Heyer, W.R., Ponssa, M.L. & Stanley, E. (2014) Systematics of the Neotropical genus *Leptodactylus* Fitzinger, 1826 (Anura: Leptodactylidae): Phylogeny, the relevance of non-molecular evidence, and species accounts. *South American Journal of Herpetology*, 9, 1–128. https://doi.org/10.2994/SAJH-D-13-00022.1

de Sá, R., Langone, J.A. & Segalla, M.V. (2007) The tadpole of *Leptodactylus notoaktites* Heyer, 1978 (Anura, Leptodactylidae). *South American Journal of Herpetology*, 2, 69–75. https://doi.org/10.2994/1808-9798(2007)2[69:TTOLNH]2.0.CO;2

Duellman, W.E. (2005) *Cusco Amazónico – The lives of amphibians and reptiles in an Amazonian rainforest*. Cornell University Press, Ithaca, New York, xv + 33 pp.

Frost, D.R. (2021) Amphibian Species of the World: an Online Reference. Version 6.0. Available from: http://research.amnh.org/herpetology/amphibia/index.html (accessed 18 May 2021).

Gosner, K.L. (1960) A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica*, 16, 183–190.

Grosjean, S. (2005) The choice of external morphological characters and developmental stages for tadpole-based anuran taxonomy: a case study in *Rana* (*Sylvarana*) *nigrovittata* (Blyth, 1855) (Amphibia, Anura, Ranidae). *Zoology*, 74, 61–76. https://doi.org/10.1163/18759866-0740102005

Hero, J.M. (1990) An illustrated key to tadpoles occurring in the Central Amazon rainforest, Manaus, Amazonas, Brasil. *Amazoniana: Limnologia et Oecologia Regionalis Systematis Fluminis Amazonas*, 11, 201–262.

Heyer, W.R., García-Lopez, J.M. & Cardoso, A.J. (1996) Advertisement call variation in the *Leptodactylus mystaceus* species complex (Amphibia: Leptodactylidae) with a description of a new sibling species. *Amphibia-Reptilia*, 17, 7–31. https://doi.org/10.1163/156853896X00252

Lavilla, E.O. & Scrocchi, G.J. (1986) Morfometría larval de los géneros de Telmatobiinae (Anura: Leptodactylidae) de Argentina y Chile. *Physis*, 44, 39–43.

Lins, A.C.R., De Magalhães, R.F., Costa, R.N., Brandão, R.A., Py-Daniel, T.R., Miranda, N.E.O, Maciel, N.M., Nomura, F. & Pezzuti, T.L. (2018) The larvae of two species of *Bokermannohyla* (Anura, Hylidae, Cophomantini) endemic to the high lands of central Brazil. *Zootaxa*, 4527 (4), 501–520. https://doi.org/10.11646/zootaxa.4527.4.3

Pezzuti, T.L., Leite, F.S.F., Rossa-Feres, D.D.C. & Garcia, P.C.A. (2021) The tadpoles of the Iron Quadrangle, Southeastern Brazil: a baseline for larval knowledge and anuran conservation in a diverse and threatened region. *South American Journal of Herpetology*, 22, 1–107. https://doi.org/10.2994/SAJH-D-20-00042.1

Prado, C.P.A. & D'Heursel, A. (2006) The tadpole of *Leptodactylus elenae* (Anura: Leptodactylidae), with the description of the internal buccal anatomy. *South American Journal of Herpetology*, 1, 79–86. https://doi.org/10.2994/1808-9798(2006)1[79:TTOLEA]2.0.CO;2

Schulze, A., Jansen, M. & Köhler, G. (2015) Tadpole diversity of Bolivia's lowland anuran communities: molecular identification, morphological characterisation, and ecological assignment. *Zootaxa*, 4016 (1), 1–111. https://doi.org/10.11646/zootaxa.4016.1.1

Silva, L.A., Magalhães, F.M., Thomassen, M.H., Leite, F.S.F., Garda, A.A., Brandão, R.A., Haddad, C.F.B., Giaretta, A.A. & de Carvalho, T.R. (2020) Unraveling the species diversity and relationships in the *Leptodactylus mystaceus* complex (Anura: Leptodactylidae), with the description of three new Brazilian species. *Zootaxa*, 4779 (2), 151–189. https://doi.org/10.11646/zootaxa.4779.2.1

Vera Candioti, M.F., Brusquetti, F. & Netto, F. (2007) Morphological characterization of *Leptodactylus elenae* tadpoles (Anura: Leptodactylidae: *L. fuscus* group), from Central Paraguay. *Zootaxa*, 1435 (1), 1–17. https://doi.org/10.11646/zootaxa.1435.1.1

3. CONCLUSÕES GERAIS

O equilíbrio entre o conhecimento da fase adulta e larval dos anuros deve ser almejado para que seja possível a compreensão completa do ciclo de vida dos anuros e a abertura de novos caminhos para conservação de espécies e ambientes. Por esse motivo, o desenvolvimento de estudos taxonômicos e descritivos de girinos que fornecem diagnósticos confiáveis para o reconhecimento das espécies possuem um papel fundamental para o desenvolvimento do conhecimento acerca desse estágio de vida dos anuros. Cada vez mais estudos descritivos sobre morfologia e história natural de girinos se provam ferramentas importantes para elucidar processos ecológicos e evolutivos das espécies.

Os girinos têm se revelado um importante complemento para o levantamento de informações sobre a biota local e estudos sobre a diversidade de espécies, isto devido ao fato de permanecerem mais tempo no ambiente comparado à fase adulta e, consequentemente, são mais facilmente amostrados. Além disso, podem auxiliar na investigação e resolução de problemas taxonômicos e filogenéticos. Tudo isso, evidencia que o conhecimento sobre girinos ainda está em crescimento e que preencher as lacunas existentes é fundamental para compreensão biológica e taxonômica de muitas espécies. Nesse sentido, esperamos que o presente trabalho tenha agregado com o conhecimento sobre os girinos da Zona da Mata de Minas Gerais.