

Reproductive notes and larval development of *Macrogenioglottus alipioi* (Anura: Odontophrynidae) from the northern Atlantic forest

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Abstract

Reproductive notes and larval development of *Macrogenioglottus alipioi* (Anura: Odontophrynidae) from the northern Atlantic forest. The neotropical monotypic genus *Macrogenioglottus*, described to enclose *M. alipioi*, is a forest leaf litter frog with a wide geographic distribution in eastern Brazil, despite being considered a rare species. Herein, we present some reproductive data of this species from a population of the north of the São Francisco River, Northeastern Brazil. We present information on the amplexus, egg clutch, advertisement, and release calls, along with a description of its larval development. Adults were observed only after heavy and continuous rains, in a temporary pond located inside the forest. Males call in chorus at the edge of the pond. The advertisement call consists of a pulsatile single note, with no substantial variation in relation to the other population of the species, but which promptly differs this genus from the remaining Odontophrynidae. The amplexus is axillary. One female deposited 298 pigmented eggs in a plastic bag. The tadpole reaches its larger size at Stage 39, being the largest tadpole within the family. At the end of metamorphosis, the froglet snout–vent length is equivalent to tadpole total length at stage 26–27, indicating substantial postmetamorphic growth until adulthood. Data on body size of males and females are also provided. The uniqueness of some larval traits and mainly on some call attributes supports the systematic distinction of *Macrogenioglottus*.

Keywords: Call, Egg clutch, Morphology, Ontogeny, Reproduction, Tadpole.

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Resumo

Notas reprodutivas e desenvolvimento larval de *Macrogenioglottus alipioi* (Anura: Odontophrynidae) no norte da Mata Atlântica. *Macrogenioglottus alipioi* é uma espécie rara, alocada em um gênero monotípico, habitante da serapilheira de florestas da região Neotropical, com ampla distribuição no leste do Brasil. Apresentamos alguns dados reprodutivos dessa espécie, a partir de uma população encontrada ao norte do rio São Francisco, no nordeste brasileiro. Incluímos informações a respeito do amplexo, desova, cantos de anúncio e de soltura, em conjunto com a descrição do seu desenvolvimento larval. Indivíduos adultos foram observados apenas após chuvas fortes e contínuas, em poça temporária no interior da mata. Machos cantam em coro na margem da poça. O canto de anúncio consiste em uma única nota pulsátil sem diferenças significativas das demais populações da espécie, mas que prontamente difere *Macrogenioglottus* dos demais Odontophrynidae. O amplexo é do tipo axilar. Uma fêmea depositou 298 ovos pigmentados em um saco plástico. O girino adquire o seu maior tamanho do estágio 39, sendo o maior girino dentre aqueles das demais espécies na família. No final da metamorfose o comprimento rostro–cloacal juvenil é equivalente ao comprimento total do girino em estágio 26–27, indicando um crescimento posmetamórfico substancial até atingir a idade adulta. Também fornecemos dados dos tamanhos dos machos e fêmeas. A peculiaridade de alguns caracteres larvais e, principalmente, de alguns atributos do canto, corrobora a distinção sistemática de *Macrogenioglottus*.

Palavras-chave: Canto, Desova, Girino, Morfologia, Ontogenia, Reprodução.

Introduction

Amphibians are the tetrapods with the greatest reproductive diversity documented so far (Duellman 1985, Wells 2007, Crump 2015). Regarding anurans specifically, one conspicuous characteristics of their reproductive behavior concerns the acoustic intraspecific communication. Bioacoustic data have been largely applied in taxonomic evaluation of anuran species with wide geographical distribution, corroborating their current taxonomic status (Mângia *et al.* 2019) or revealing an important cryptic diversity (Fouquet *et al.* 2014, Ron *et al.* 2018, Lopes *et al.* 2020). In fact, most of the anuran calls have primarily reproductive functions (see Toledo *et al.* 2015 for a revision). The most common example here is the advertisement call, usually emitted by males to attract co-specific females during the breeding season (Wells 2007, Toledo *et al.* 2015, Kohler *et al.* 2017), being used for species delimitation in taxonomy due to its role in pre-zygotic isolation (Kohler *et al.* 2017, Carvalho *et al.* 2020). However, there are other types of less documented calls that can be emitted in social

contexts and can permeate anuran reproduction, as well as the release calls, which can be produced by both males and non-receptive females when grabbed by another male (Duellman and Trueb 1994, Toledo *et al.* 2015). Just like the advertisement calls, release calls have been useful to differ between close related species (Castellano *et al.* 2002, Grenat and Martino 2013).

Another striking reproductive aspect of anurans is the presence of a distinctive larval phase during its life cycle (Wassersug 1975, Harris 1999). The larval phase reaches a crucial point during the transition to adulthood through metamorphosis. Anuran postembryonic ontogeny encompasses three processes: (1) the development and disappearance of larval structures, (2) the remodeling of larval structures into adult structures, and (3) the development and differentiation of adult structures (Fabrezi *et al.* 2012). New information on this subject is important general developmental patterns, understand ontogenetic diversity and clarify questions about systematic relationships (e.g., Hall *et al.* 1997, Fabrezi *et al.* 2012, Barrionuevo 2018). Thus, developmental changes need to be

well documented and go beyond the information available on standardized ontogenetic tables (e.g., Taylor and Kollros 1946, Gosner 1960), which despite being very useful for different comparative purposes and being well suited for many species, may not take into account interspecific variations (most of them heterochronic processes) that may have played important roles on frog diversification (Chuliver and Fabrezi 2019, Mongin and Carvalho-e-Silva 2013, Silva *et al.* 2018, Cordioli *et al.* 2019).

The neotropical monotypic genus *Macrogenioglottus* Carvalho, 1946 belongs to the family Odontophrynidae (sensu Pyron and Wiens 2011); it is the sister taxon of *Odontophrynus* Reinhardt and Lütken, 1862, and together they are the sister clade of *Proceratophrys* Miranda-Ribeiro, 1920 (Amaro *et al.* 2009, Pyron and Wiens 2011, Martino *et al.* 2019). The genus was described to include *M. alipioi*, a forest leaf litter frog with wide geographic distribution in eastern Brazil, despite being considered a rare species (Toledo *et al.* 2014). *Macrogenioglottus alipioi* occurs in regions of Atlantic Forest from Pernambuco state, in the northeast region, to São Paulo state, in the southeast region (Haddad *et al.* 2013, Frost 2022). The species shows the reproductive mode 2 or 3 (Abravaya and Jackson 1978, sensu Nunes-de-Almeida *et al.* 2021) and its tadpoles are Type IV according to Orton's (1953) classification (Figure 1).

There are some reports on the biology and natural history of *Macrogenioglottus alipioi* (such as tadpole morphology, mating behavior, call and karyotype) that come from populations of the southeastern region of Brazil (Abravaya and Jackson 1978, Silva *et al.* 2003), in the southern limit of its distribution. There are also data about the tadpole from the extreme north distribution, in the area north of the São Francisco River (Lisboa *et al.* 2011, Dubeux *et al.* 2020); and recently, Mira-Mendes *et al.* (2020) published the calls and morphometric data from populations close to and from the type locality (Ilhéus, Bahia state), south of the São Francisco River. In order to know additional

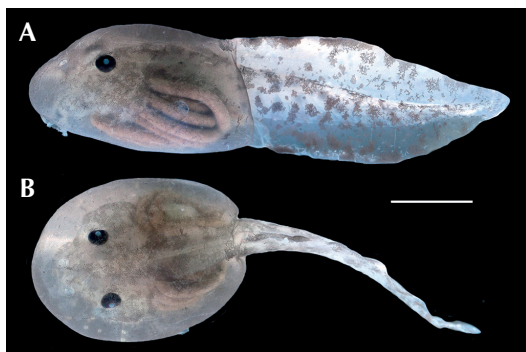


Figure 1. Tadpole of *Macrogenioglottus alipioi* (MHNUFAL 13952) at Stage 26 from Mata do Catolé, Maceió, Alagoas state, northeastern Brazil. (A) Lateral and (B) dorsal views (scale bar = 5 mm). Photo by Marcos Dubeux.

aspects of the population from the northern part of its distribution that may help to establish a more complete scenario of the taxonomic status of the genus and its relationships with the other members of the family Odontophrynidae, we present new reproductive data, including egg clutch, advertisement and release calls, and data on its larval development, from a population of *M. alipioi* in the north of the São Francisco River, northeastern Brazil.

Materials and Methods

We conducted field surveys at Mata do Catolé (09°35' S, 35°49' W; datum WGS84, 47 m a.s.l.), an Atlantic Rain Forest fragment located at the Environmental Protection Area of Catolé and Fernão Velho, in Alagoas state, northeastern Brazil. In this area, vegetation type is ombrophilous and the regional climate varies from humid to sub-humid, with a rainy season (March–August) and a dry season (September–February). The mean annual rainfall varies from 1500 to 2000 mm, and temperature varies from 23 to 25°C (Assis 2000).

Reproductive observations were made during visual encounter surveys (VES; Heyer *et al.* 1994) at night, in a temporary pond on 12 July and 21 August 2011 (17:30–23:00 h). After heavy rains

at night (21:00–22:00 h, air temperature = 20.8–20.9°C, relative air humidity = 100%), we recorded vocalizations of four males of *Macrogenioglottus alipioi* on 12 July 2011 (one collected, MHNUFAL 10942, SVL = 82.2 mm) at the edge of ponds, using a digital recorder Sony PCM-D50R (sampling rate of 44.1 kHz and resolution of 16 bits) coupled to a unidirectional microphone Sennheiser ME66/K6. The microphone was positioned about one meter from calling males. Vocalizations were classified as advertisement call according to the social context of the observed individuals (Toledo *et al.* 2015). To obtain the release calls, a false amplexus was simulated on one male using the thumb and the forefinger to push him until the emission of calls.

A total of 200 advertisement calls (50 calls per male) and 50 release calls (one male) were analyzed in Raven Pro 1.5 (Bioacoustics Research Program 2014) with the following settings: window type Hann, DFT size 256 samples and overlap 90%. The following temporal and spectral parameters were analyzed: call duration (s), interval intercall (s), call rate (calculated per minute, call/min), dominant frequency (Hz), and minimum and maximum frequency (Hz). The dominant frequency was obtained through the Peak Frequency function, minimum and maximum frequencies through Frequency 5% and Frequency 95%, respectively. Acoustic terminology and analysis follow Köhler *et al.* (2017). Sound figures were produced with Seewave package, version 1.7.3 (Sueur *et al.* 2008), in R 3.1.0 (R Core Team 2018), with the following settings: window Hann, DFT size 1024 samples and overlap 90%. We also included information about snout–vent length (SVL) of females ($N = 2$) and males ($N = 10$) from this locality and from Murici Ecological Station (09°12' S, 35°52' W, datum WGS84; 593 m a.s.l.), municipality of Murici, also in Alagoas State, to assess sexual dimorphism. Specimens are deposited in the herpetological collection of the Museu de História Natural, Universidade Federal

de Alagoas (MHNUFAL; Appendix I), Brazil. The snout–vent length of females and males was measured with calipers to the nearest 0.1 mm.

One amplexant pair was collected (MHNUFAL 10940–41) and placed in a plastic bag where the female spawned. We count the total number of eggs in the spawn (MHNUFAL 10982), measuring their maximum diameter (without considering the gelatinous layers) using an ocular micrometer installed on a Coleman NSZ 405 stereomicroscope.

We collected 78 tadpoles between May 2008 and August 2011 (Appendix I). All tadpoles came from the same pond, being posteriorly fixed and preserved in 10% formalin. Average rainfall and temperature data were similar over three of the four years along the collection period. In addition, to reduce a hypothetical bias of the environmental conditions of successive years on the development/growth of the tadpoles, we analyzed specimens from the same developmental stages collected in different years to take into account possible variations. Species identification was made by direct comparison with samples from the same locality used by Lisboa *et al.* (2011) in the description of the larvae. We selected specimens to address all stages of development (*sensu* Gosner 1960), from the emergence of the spiracle (Stage 25) to complete atrophy of the tail (Stage 45), and then we grouped them following the developmental periods of Etkin (1968): premetamorphosis, prometamorphosis and metamorphosis. Morphometric variations were recorded through 16 measurements, six from Altig and McDiarmid (1999): body length (BL), maximum tail height (MTH), tail length (TaL), tail muscle height (TMH), tail muscle width (TMW), and total length (TL); eight from Lavilla and Scrocchi (1986): eye diameter (ED), eye–nostril distance (END), intranarial distance (InD), intraorbital distance (IoD), maximum body height (MBH), maximum body width (MBW), oral disc width (ODW, taken with the oral disc partially opened), and snout–nostril distance (SND); and two from Grosjean (2005): dorsal fin height (DFH) and

ventral fin height (VFH). Most of the measurements were taken using an ocular micrometer installed on a Coleman® NSZ 405 stereomicroscope, except for TL, BL, MBH, and MBW which were measured with calipers (0.1 mm accuracy). The terminology of external morphology follows Altig and McDiarmid (1999). Data are presented as mean \pm standard deviation ($\bar{x} \pm SD$).

Results

Observations on Reproduction

Males and females of *Macrogenioglottus alipioi* were only observed after heavy and continuous rains, in a temporary pond inside the

forest. Males were found calling in chorus at the edge of the pond (Figure 2A) where they were separated from each other by at least 3 m. Calling activity began after sunset (17:30 h) and continued during the night. We observed three mating events around 21:00 h, all of them occurring in the leaf litter near the pond, and about 1 m apart from each other. The amplexus is axillary (Figure 2B). In one occasion, we separated an amplexant pair and left them 1 m apart from each other. The male began to call immediately, the same female moved towards him, and the amplexus restarted.

Snout-vent length of females were 93.5 and 101.1 mm ($N = 2$) and SVL of males ranged from 77.7–90.8 mm (84.3 ± 4.1 mm, $N = 10$).

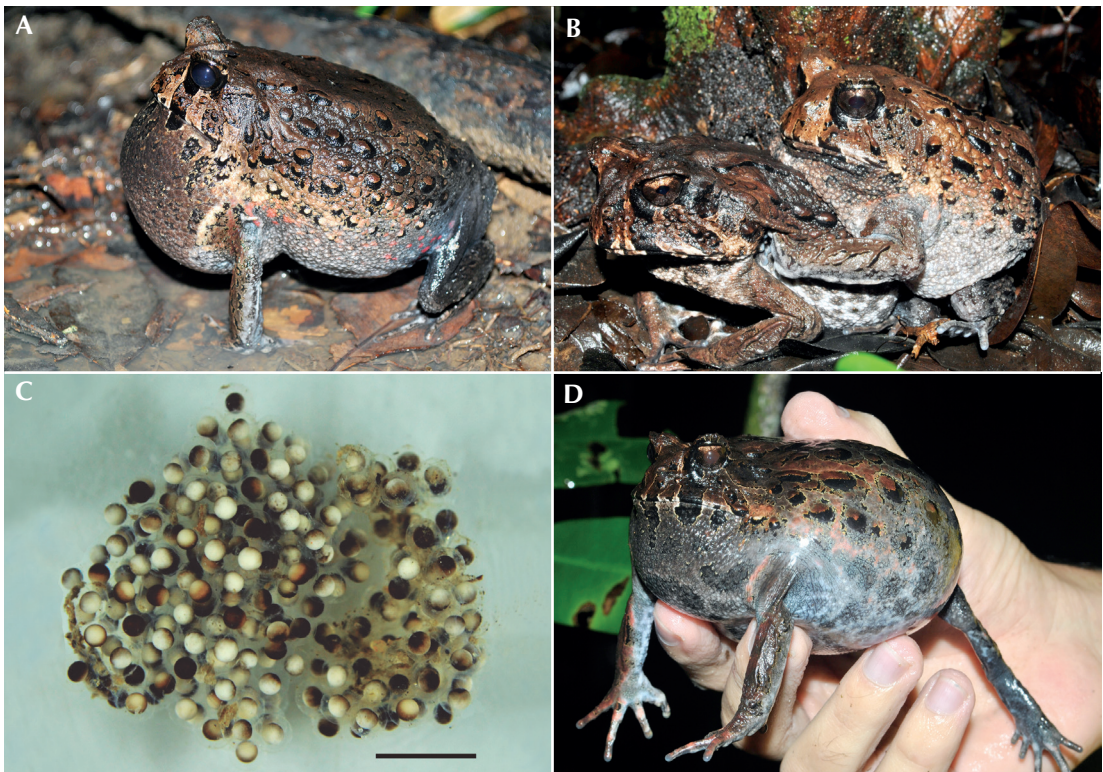


Figure 2. Breeding activity of *Macrogenioglottus alipioi* from Mata do Catolé, Maceió, Alagoas state, northeastern Brazil. (A) Male calling on the leaf litter, (B) couple in amplexus, (C) egg clutch (scale bar = 10 mm), and (D) manipulation of a male to emit the release call.

Spawning.—The spawn contained 298 eggs, free or slightly adhered to one another by a gelatinous mass. The eggs had a diameter between 1.8 and 2.6 mm (2.2 ± 0.1 mm, $N = 298$) and were pigmented in the animal pole, with color ranging from light to dark brown (Figure 2C).

Advertisement call.—The advertisement call (Figure 3A–B) of *Macrogenioglottus alipioi* consists of a pulsatile single note that lasts 0.3–0.6 s (0.4 ± 0.07 s), with intervals between calls varying from 0.3–5.5 s (0.9 ± 0.6 s). Call repetition rate was of 10.3–98.3 calls/min (53.9 ± 18.3 calls/min), dominant frequency ranged from 344.5–430.7 Hz (390.4 ± 43.1 Hz), maximum frequency 430.7–516.8 Hz (492.1 ± 39 Hz) and minimum frequency was 258.4 Hz.

Release call.—The release call (Figure 3C–D) was emitted sporadically by one male during handling (Figure 2D). The call consists of one single low-frequency note, as the advertisement call, but with a shorter duration (0.3 ± 0.02 s) and higher call repetition rate (23.5–77.6 calls/min, 57.6 ± 12.2 calls/min). Inter-call interval ranged from 0.5–2.3 s (0.8 ± 0.4 s), with a dominant frequency of 430.7 Hz. Maximum frequency 775.2–1292.0 Hz (902.7 ± 89.1 Hz) and minimum frequency was 344.5 Hz.

Larval Development

Premetamorphosis (stages 25–35; Figure 4A–D).—This phase is characterized mainly by body and tail growth and early development of the hind limbs. The tadpoles have an ovoid body in dorsal view (MBW/BL 0.6–0.9, 0.71 ± 0.07 , $N = 46$) and depressed laterally (MBH/MBW 0.6–0.8, 0.7 ± 0.06 , $N = 46$). At Stage 25, the mouthparts are already formed, with the typical labial tooth row formula (LTRF) 2(2)/3(1), a single row of marginal papillae and one or two rows of submarginal papillae on the lower labium; the pigmentation of the jaw sheaths is

more restricted to the distal margin, which gradually increases as the larva grows until become fully pigmented at about Stage 32 (Figure 5A–B). At Stage 25, the gills are already completely covered by the operculum; there are remnants of the yolk sac, which disappears in the next Stage, and the lateral spiracle is present on the middle third of the body, with the inner wall free, longer than the external wall. At Stage 33, the stylopodium and zeugopodium begin to differentiate. Coloration in preservative: at Stage 25, the body is translucent with two groups of melanophores visible, the first one is filiform in shape, located on the dorsal and lateral integument; the second group is stellate, visible on the connective tissue that covers the chondrocranium and viscera, also forming speckles on the caudal musculature and fins. From Stage 26 to Stage 30 these melanophores become more numerous, with those of the first group also arising ventrally and around the nostrils, and those of the second group forming dark spots on the tail. At Stage 27, there are also small subepidermal whitish punctuated chromatophores on the eyes and dorsum, but more easily seen on the viscera. From Stage 31 the filiform melanophores become denser, interlaced, leaving the tadpole with a dark brown coloration. Light brown stellate spots begin to appear on limb bud at Stage 29. At Stage 28, the lateral line system become apparent.

Prometamorphosis (stages 36–40; Figure 4E).—Along this phase the body is still growing, following the limb development and the beginning of metamorphosis. The body shape does not change (MBW/BL 0.7–0.8, 0.7 ± 0.04 , $N = 15$). At Stage 36, the forelimbs are visualized by transparency inside the gill chambers; as they develop, press the overlying skin. At Stage 38, a slight depression appears around each nostril, the spiracle inner wall becomes smaller than the external one, and the nasolacrimal duct begins to appear in some individuals as a shallow groove from the

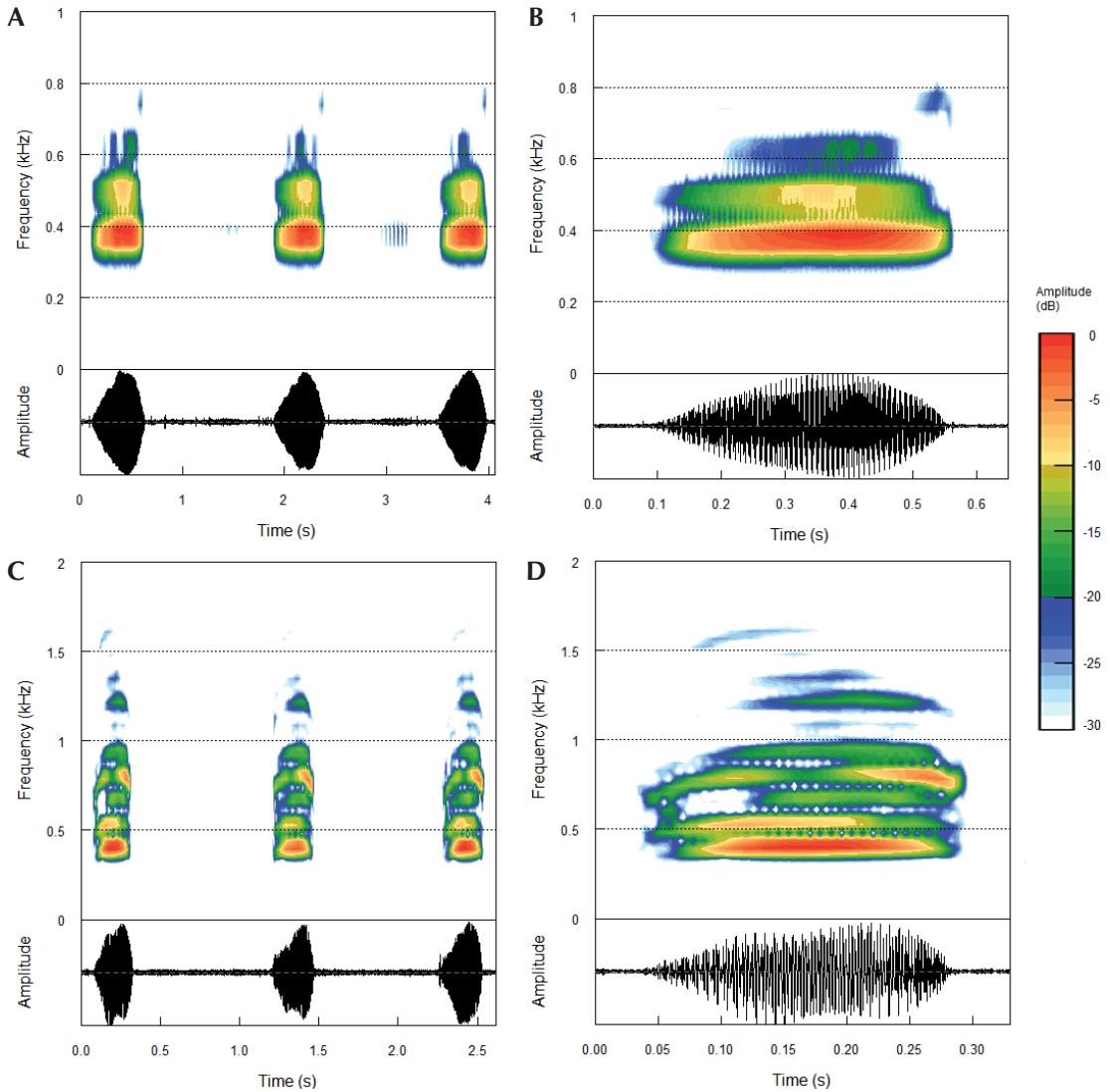


Figure 3. Calls of *Macrogenioglottus alipioi* from Mata do Catolé, Maceió, Alagoas state, northeastern Brazil. (A) Series of three advertisement calls (MHNUFAL 10942); (B) detail of an advertisement call; (C) series of three release calls (specimen not collected); and (D) detail of a release call. In all figures, spectrogram above and oscillogram below.

anterior region of the eye to the nostrils. At Stage 39, the tadpole acquires its larger size (TL = 69.9 mm, $N = 1$; Figures 4E and 6A), decreasing progressively in the following

stages. The eye–nostril distance begins to decrease gradually (Figure 6B), and the relative size of the eyes starts to increase gradually in relation to the body size. At Stage 40, the

snout–nostril distance also starts to decrease, but in a more abrupt way than the other measures (Figure 6B). Coloration in preservative: from Stage 39 the filiform melanophores form irregular spots on the dorsum. At Stage 38, the brown stellate spots of the limb bud become darker, forming agglomerations. At the following Stage, the pattern of spots on the phalanges begins to differentiate, highlighting the subarticular regions and external metatarsal tubercle.

Metamorphosis (stages 41–46; Figure 4F–I).—This last phase is characterized by an abrupt decrease in size, mostly due to the decrease of tail length, and loss of most larval characters. This overall larval reduction is directly reflect in the continuous reduction of eye–nostril and snout–nostril distances, and in the increase of the intracocular distance in relation to body width (eyes will be positioned laterally), shaping the future adult head (Figure 6B–C). At Stage 41, the tadpole body acquires a more oval shape, with the central dorsal region more prominent, although the bodily relationship almost does not change ($MBW/BL = 0.7$, $N = 4$). In the region where the forelimbs will emerge the skin becomes thinner and lighter. The upper and lower eyelids appear (Figure 5E–F). The nostrils aperture become narrower. In the oral disc, labial teeth in A1, A2, P1, and P2 rows becomes progressively vestigial; the P3 row begins to degenerate; the upper jaw acquires a more frontal position while the lower jaw exhibits broken parts (Figure 5C); the mouth is getting bigger (oral disc width/body width ratio increases; Figure 6C). The vent tube degenerates, remaining some remnant connected to the ventral fin, but which disappears completely in the following stages. The caudal fins begin to decrease in height. The skin becomes thicker, tubercles appear on the dorsum, on the eyelids and in the inner region of the thighs (next to the cloaca). There are also small elevations (tubercles or warts) that are darker in the

interocular region, along the medial region of the dorsum, and laterally behind the eyes (resembling the adult pattern). At Stage 42, the head begins to differentiate, becoming wider than the rest of the body. The eyelids and eyelid tubercles become more evident (Figure 5F–H). The mouth position becomes gradually more anterior, remaining only a few marginal papillae laterally on the upper lip; there are no more vestiges of the labial teeth and jaws (Figure 5D). In some individuals, the tongue is already visible. The lateral line system disappears. The spiracle and the jaw sheaths have disappeared, and the caudal fins begin to decrease in height. The forelimbs have already emerged, and the ruptured opercular membrane is still visible. At Stage 43, the larva begins to take on the adult appearance, the tubercles are more evident, the *cantus rostralis* are already visible, and the eyes are more extrinsic, with the pupil acquiring an angular shape. The nostrils are positioned more dorsofrontally and there is no record of the larval oral apparatus in the mouth. The caudal fins are atrophied in advanced state, along with the caudal musculature (Figure 5I–K), resulting in an abrupt decrease in the larval total length (37.6 ± 2.2 mm, ranging from 29.6–49.0 mm, $N = 5$, Figure 6A). The epidermis is much thicker, with the interocular tubercles blackened; the intestines can no longer be seen by transparency. At stages 44–45, there is already an adult configuration (Figure 4I), the eyes are positioned laterally, the nasal cartilage forms a protrusion where the nostrils open, the tail is reduced to a bud (Figure 5L), and the total length is equivalent to that of the tadpoles in Stage 25–26 (17.9 and 18.6 mm, $N = 2$). Coloration in preservative: body dark brown, with darker spots surrounding the tubercles; belly with a whitish appearance, with light brown spots.

A summary of the main events of postembryonic external development is shown in Table 1, and the morphological measurements of the larvae in Appendix II.

Table 1. Summary of the main external larval developmental features of *Macrogenioglottus alipioi*. Stages are according to Gosner (1960).

Stage	N	Main external developmental features	Total length ($\bar{x} \pm SD$) (mm)
25	5	Mouthparts fully formed, pigmentation of the jaw sheaths restricted to the distal margin; no trace of the yolk sac; spiracle with inner wall free and longer than the external wall; body translucent with filiform and star-shaped melanophores.	14.4 ± 1.4
26	5	Jaw sheaths fully formed; hind limb appears as a small bud ($L < \frac{1}{2} D$); chromatophores more numerous, grouping and forming spots on body and tail, nostrils with pigmented margins.	25.3 ± 1.9
27	5	Hind limb bud slightly longer than half its diameter ($L \geq \frac{1}{2} D$); whitish chromatophores on dorsum, eyeballs and viscera; abdomen iridescent.	26.9 ± 5.3
28	4	Lateral line system visible; chromatophores in high density; hind limb bud slightly longer than its diameter ($L \geq D$).	33.7 ± 6.1
29	2	Hind limbs longer than one and a half times its diameter ($L > 1 \frac{1}{2} D$) and with rare light brown punctuations.	39.7 ± 4.6
30	4	Hind limb bud with a length equivalent to twice its diameter ($L = 2D$).	35.5 ± 2.5
31	5	Submarginal papillae close to emarginations of the oral disc in some individuals; hind limb bud paddle shaped (autopodium with indentations); larva with coloration dark brown due to higher density of chromatophores.	41.9 ± 2.7
32	3	Beginning of indentation between toes IV and V; larva darker, punctuation on limbs forming spots similar to those of the tail.	49.5 ± 1.1
33	4	Beginning of indentation between toes III and IV (estilopodium and zeugopodium differentiated).	45.6 ± 2.7
34	4	Beginning of indentation between toes II and III.	48.4 ± 7.8
35	5	Beginning of indentation between toes I and II.	49.9 ± 7.1
36	2	Visualization of the forelimbs within the branchial chambers; separation of toes III and V.	58.8 ± 1.2
37	4	All five toes separated.	53.9 ± 8.4
38	3	Emergence of a depression around each nostril; spiracle with inner wall longer than the external wall; emergence of the nasolacrimal duct and the spot pattern on the phalanges.	59.8 ± 10.5
39	1	Appearance of subarticular patches and on external metatarsal tubercle; filiform chromatophores agglomerated forming spots on dorsum; hind limbs almost entirely covered by spots.	69.9
40	5	Forelimb larger and clearly visible ventrolaterally within the branchial chamber; appearance of metatarsal tubercles.	65.1 ± 11.3
41	4	Body ovoid, dorsum with medial region prominent; eyes directed laterally, appearance of the upper and lower eyelids; forelimb form a lateral bulge under the skin; advanced degeneration of the mouthparts; disappearance of vent tube; appearance skin tubercles.	61.7 ± 7.3

Table 1. Continued.

Stage	N	Main external developmental features	Total length ($\bar{x} \pm SD$) (mm)
42	5	Disappearance of lateral line system; head differentiation; disclosure of the tuberculous pattern of adult skin; mouth commissure does not reach the anterior limits of the nostrils; disappearance of spiracle; emergence of forelimbs; decrease of caudal fins; skin thicker, appearance of the tongue.	41.7 \pm 3.1
43	5	<i>Cantus rostralis</i> evident; skin tubercles more evident; nostrils dorsofrontal; mouth commissure reaches the anterior limits of the eyes; tail and fins in an advanced state of atrophy; skin thicker and darker.	37.5 \pm 7.0
44	1	Acquisition of adult form, tail remains as a stub; dark spots surround the tubercles; mouth commissure below the eye.	18.6
45	1	Mouth commissure exceeding the posterior limits of the eye; skin darker.	17.9

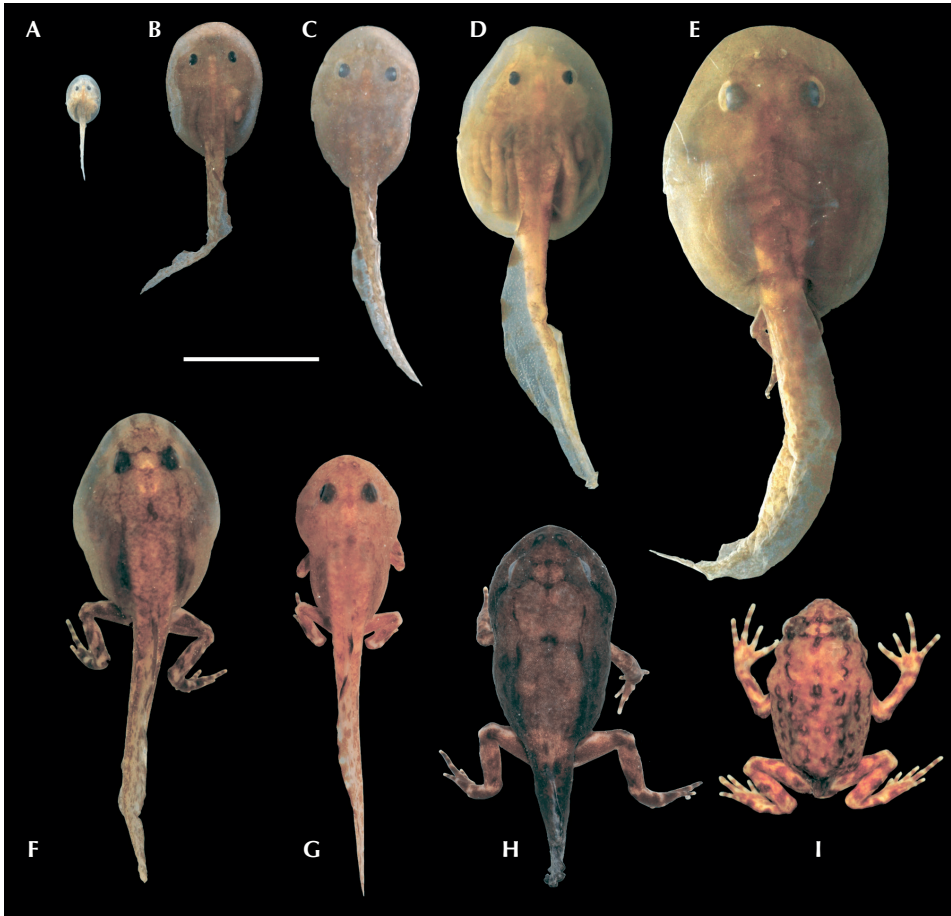


Figure 4. Representative series of *Macrogenioglottus alipioi* development. (A) Stage 25, (B) Stage 29, (C) Stage 32, (D) Stage 35, (E) Stage 39, (F) Stage 41, (G) Stage 42, (H) Stage 43, and (I) Stage 45 (scale bar = 20 mm).

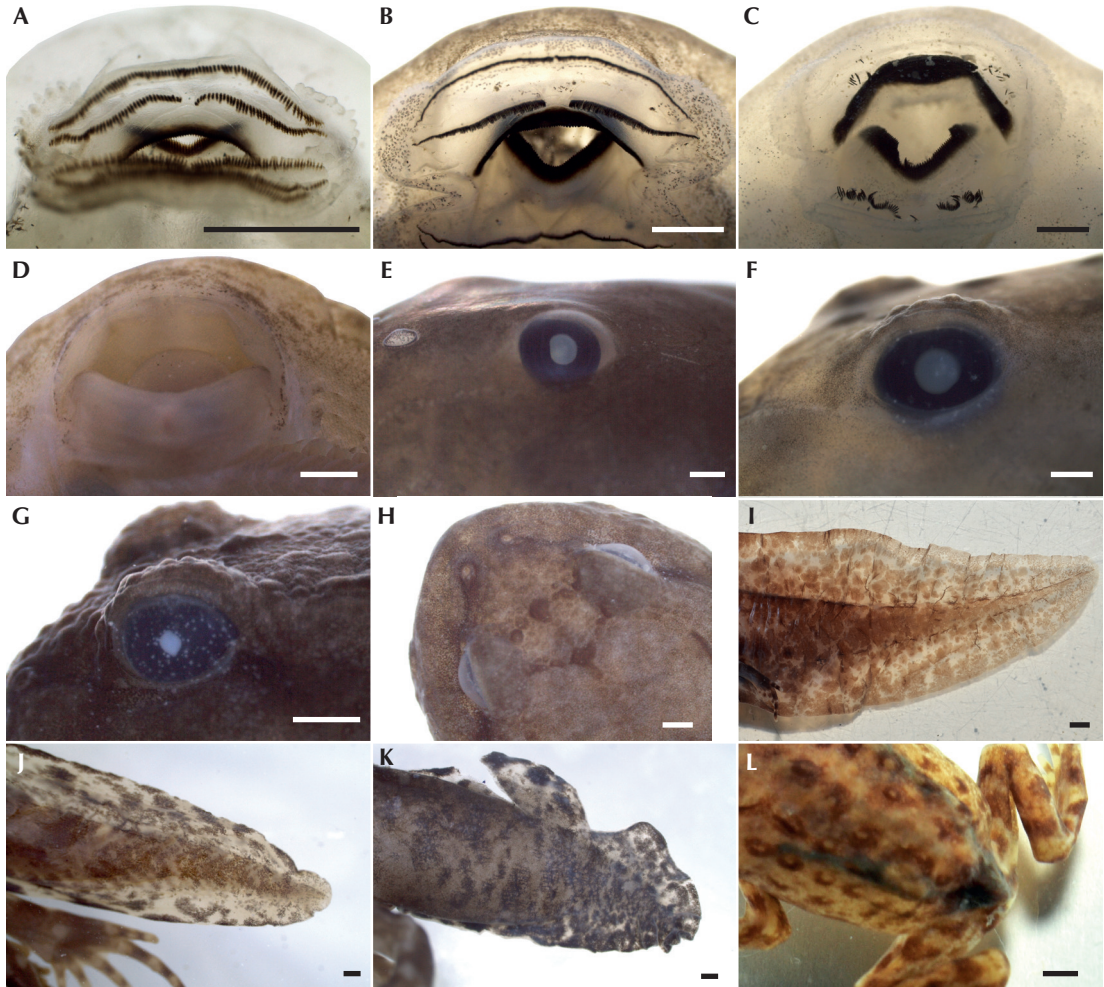


Figure 5. Changes in external morphology during larval development of *Macrogenioglottus alipioi*. (A–D) Mouth transformations in ventral view: (A) Stage 25, (B) Stage 32, (C) Stage 41, and (D) Stage 42. (E–G) Transformations in the region of the eyes in lateral view: (E) Stage 32 (F) Stage 42, and (G) Stage 43. (H) Region of the eyes and nostrils in dorsal view at Stage 43. (I–K) Tail transformation in lateral view: (I) Stage 40, (J) Stage 42, (K) Stage 43. (L) Posterior region of the froglet in dorsal view (Stage 45; scale bar = 1 mm).

Discussion

Reproductive Observations

Abravaya and Jackson (1978) and Mira-Mendes *et al.* (2020) observed reproductive activity of *Macrogenioglottus alipioi* from Santa Teresa (Espírito Santo state) and Uruçuca (Bahia

state), respectively, and reported that individuals breed only after heavy rains in ponds. Herein, we corroborated this observation, adding the information that the activity occurs usually in the middle of the rainy season, and suggesting that the temporal reproductive pattern of *M. alipioi* is explosive (*sensu* Wells 1977), similar to almost all species in the family Odontophrynidae (e.g.,

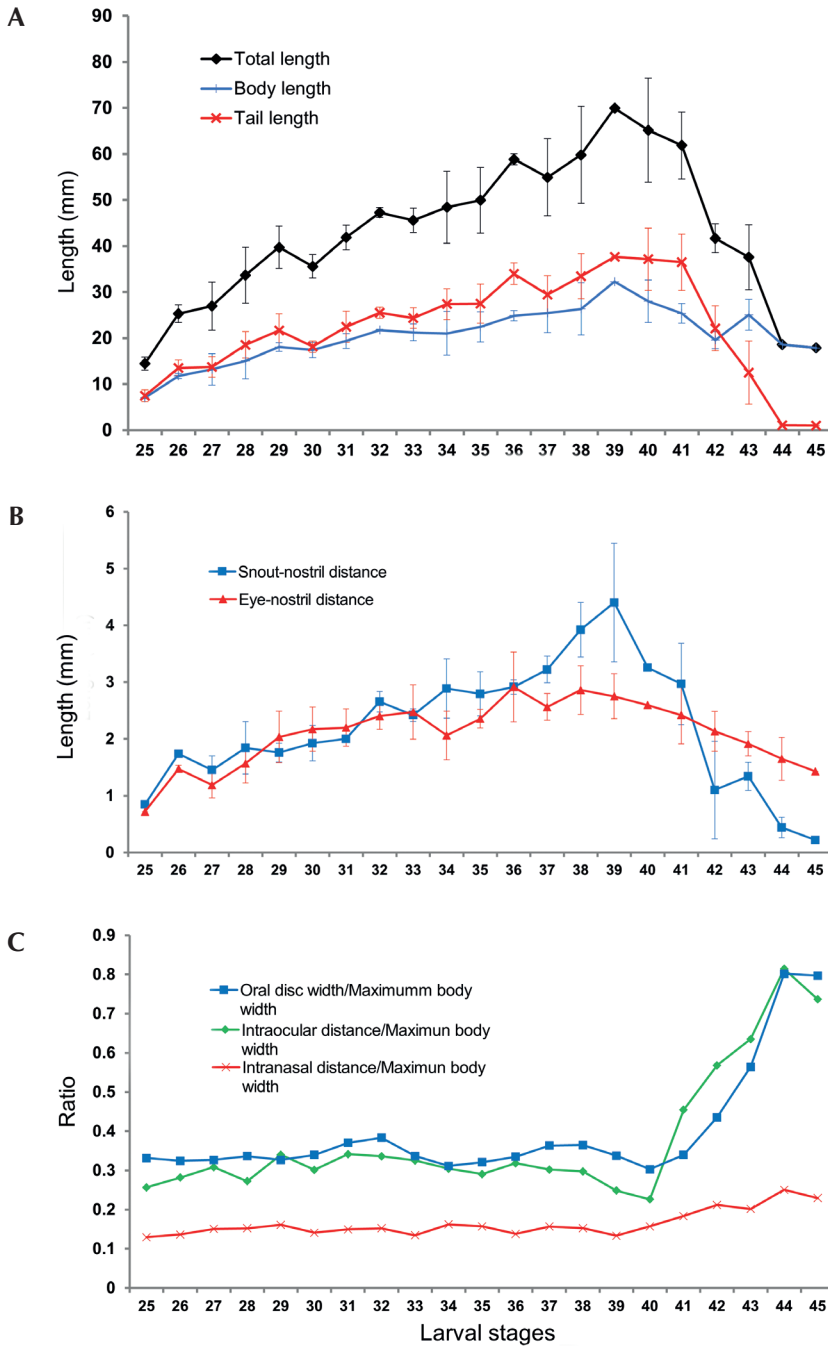


Figure 6. Some morphometric and ratios values of *Macrogenioglottus alipioi* during larval development from stages 25 to 45. (A) Mean and standard deviation values of body, tail, and total length; (B) mean and standard deviation values of snout–nostril distance and eye–nostril distance; and (C) ratio of oral disc width, intraocular distance, and intranasal distance in relation to maximum body width.

Santos *et al.* 2008, Araújo *et al.* 2009, Godinho *et al.* 2013, Valdez and Maneyro 2016, Malagoli *et al.* 2016), which could explain why this species is rarely seen in the field.

Abravaya and Jackson (1978) reported a gravid female of *Macrogenioglottus alipioi* with about 3650 eggs in their ovaries. The authors also stated that the numbers of eggs oviposited on each oviposition event although not determined, would be small, probably less than 20 eggs, an unrealistic hypothesis for a large species with free-living tadpoles (see Crump 1974, Prado and Haddad 2005). Hartmann *et al.* (2010) found 2900 eggs in a *M. alipioi* clutch from Picinguaba (São Paulo state, southeastern Brazil). Our collected spawning from an amplexant pair containing 298 eggs must be considered with caution once it was obtained inside a plastic bag (an artificial situation). Nonetheless, clutch size can vary considerably between females from the same population (Lemckert and Shine 1993, Prado and Haddad 2005) due to numerous factors, such as environmental conditions and female nutritional status (Ryser 1989, Jørgensen 1992).

Sexual dimorphism in snout-vent length occurs in most species of *Odontophrynus* (see references in Mira-Mendes *et al.* 2020) and *Proceratophrys* (Boquimpani-Freitas *et al.* 2002, Giaretta *et al.* 2008, Santana *et al.* 2010, Ávila *et al.* 2011, Nunes *et al.* 2015). Hartmann *et al.* (2010) found significant sexual dimorphism in SVL of *Macrogenioglottus alipioi* from São Paulo state, with females bigger than males. On the other hand, Mira-Mendes *et al.* (2020) did not observe significant differences in the SVL in a sample of *M. alipioi* from Bahia state (although the authors found sexual differences in the head and hind limb size). Despite we found that females of *M. alipioi* are bigger than males, we could not test the existence of sexual dimorphism in our population because our small female sample ($N = 2$), nevertheless the data suggest that sexual dimorphism is a peculiar feature in the genus.

The advertisement call traits of *Macrogenioglottus alipioi* provided here are

similar to those described by Mira-Mendes *et al.* (2020) for a population from Bahia state with only the dominant frequency range slightly higher in our sample (430.7–516.8 Hz here, against 344.5–430.7 Hz from the Bahia sample). Although geographic variation in advertisement call of anurans was demonstrated in conspecific population of several species (e.g., Castellano *et al.* 2000, Pröhl *et al.* 2007, Baraquet *et al.* 2015), we could not determine if this is the case here with only these data. Conversely, the pulsatile character of the note emitted by *M. alipioi* promptly distinguishes *Macrogenioglottus* from the pulsed note found in *Odontophrynus* and *Proceratophrys* species (Nascimento *et al.* 2019 and references therein, Magalhães *et al.* 2020, Mira-Mendes *et al.* 2020 and references therein, Simões *et al.* 2020, Rosset *et al.* 2021, Santana *et al.* 2021, Moroti *et al.* 2022). The release call was also similar to that described by Mira-Mendes *et al.* (2020), with the spectral and temporal parameters overlap (dominant frequency 387.60–559.90 Hz against 430.7 Hz in our sample and duration of the call of 0.032–0.314 s against 0.2–0.3 s in our sample) also suggesting that they belong to the same nominal taxon.

Larval Development

When Abravaya and Jackson (1978) described the larval external morphology of *Macrogenioglottus alipioi*, they made only a brief comparison with some species of *Odontophrynus*. In addition, Lisboa *et al.* (2011) reported some differences in relation to the population studied by Abravaya and Jackson but did not discuss it in the context of the family Odontophrynidae. Ten of the 11 species of *Odontophrynus* have their larvae described (Nascimento *et al.* 2013 and references therein, González *et al.* 2014, Menegucci *et al.* 2016, Costa *et al.* 2017, Rocha *et al.* 2017, Santos *et al.* 2017a, Moroti *et al.* 2022). In *Proceratophrys*, on the other hand, for 21 of the 42 known species the larvae have been described (Provete *et al.*

2017 and references therein, Santos *et al.* 2017b, Dias *et al.* 2018, 2019).

The odontophrynid tadpoles have a slightly depressive and ovoid body in dorsal view. The average total length ranges from 33 mm in *Proceratophrys* (stages 34–38), 54 mm in *Odontophrynus* (stages 36–38), and up to 58 mm in *Macrogenioglottus* (Stage 35). *Macrogenioglottus* also presented a much more robust body than the other genera (i.e., larger body size, for example, in relation to the length of the eyes and the oral disc). The nostrils of *Macrogenioglottus* are circular while in *Odontophrynus* they may also be oval/elliptical (with a small cutaneous extension on its medial margin in *O. occidentalis* (Berg, 1896), *O. lavillai* Cei, 1985, *O. maisuma* Rosset, 2008, and *O. monachus* Caramaschi and Napoli, 2012; Lavilla and Srocchi 1991; Grenat *et al.* 2009, González *et al.* 2014 Menegucci *et al.* 2016); in some *Proceratophrys* species it also can be reniform [e.g., *P. appendiculata* (Günther, 1873), *P. bigibbosa* (Peters, 1872), *P. dibernardoii* Brandão, Caramaschi, Vaz-Silva, and Campos, 2013, and *P. renalis*; Nascimento *et al.* 2010, Dias *et al.* 2014, 2018, Santos *et al.* 2017a]. The oral disc is usually surrounded by a single row of marginal papillae (a double row in the lower or lateral labium can be observed in *O. maisuma*, and *O. occidentalis*; Cei 1987, Borteiro *et al.* 2010). The labial tooth row formula is 2(2)/3(1), but may occur 2/3(1) [e.g., *O. carvalhoi*, *P. appendiculata*, *P. izecksohni* Dias, Amaro, Carvalho-e-Silva, and Rodrigues, 2013, and *P. tupinamba* Prado and Pombal, 2008; Caramaschi 1979, Fatorelli *et al.* 2010, Dias *et al.* 2014] with individual variations, 2(2)/3, 2/3(1), and 2/3 in *O. americanus* (Duméril and Bibron, 1841) and *O. maisuma*; Fernández and Fernández 1921, Echeverría and Montanelli 1992 (1995), Borteiro *et al.* 2010].

The knowledge about external postembryonic development is virtually absent in the family Odontophrynidae, with information only for *Odontophrynus cordobae* Martino and Sinsch, 2002 (Grenat *et al.* 2011). In general, the development phases of *O. cordobae* are

quite similar to those of *M. alipioi*. However, some important events seem to occur later in *O. cordobae*. For example, at Stage 25, *M. alipioi* has the mouthparts already fully formed while in *O. cordobae* only the jaws sheaths and the first two tooth rows are present. The tadpole acquires its larger size at Stage 39 in *M. alipioi*, while in *O. cordobae* it occurs later at Stage 41. Lastly, the major modifications of metamorphosis occur at Stage 43 in *O. cordobae*, while in *M. alipioi* they occur at stages 41–42. Before assuming that these differences reflect interspecific variation, we must consider that the larval series used by Grenat *et al.* (2011) came from captive bred specimens and were reared in an artificial environment, which may influence the time of development of the species in comparison to natural population (see Sheil *et al.* 2014).

In most tadpoles, the larval growth that occurs until the onset of metamorphosis follows exponentially on a sigmoid curve (Di Cerbo and Biancardi 2010). This phase corresponds to a period of maximum growth and minimum development (Altig and McDiarmid 1999, Grosjean 2005). In *Macrogenioglottus alipioi* this phase of almost gradual somatic growth occurs until Stage 39, where the larva reaches the maximum total length. In *O. cordobae*, as said before, this peak occurs a little later at Stage 41. In the majority of anuran species this peak occurs between these two stages [e.g., *Bokermanohyla circundata* (Cope, 1871) - Mongin and Carvalho-e-Silva 2013; *Phasmahyla cruzi* Carvalho-e-Silva, Silva, and Carvalho-e-Silva, 2009 - Costa *et al.* 2010; *P. guttata* (Lutz, 1924) - Costa and Carvalho-e-Silva 2008; *Physalaemus biligonigerus* (Cope, 1861) - Chuliver and Fabrezi, 2019], but we can find species that this event take place earlier [e.g., *Chiasmocleis lacrimae* Peloso, Sturaro, Forlani, Gaucher, Motta, and Wheeler, 2014, Stage 38; Cordioli *et al.* 2019; *Dermatonotus muelleri* (Boettger, 1885), Stage 37; Fabrezi *et al.* 2012; and *Megaelosia goeldii* (Baumann, 1912), Stage 38; Silva *et al.* 2018].

At Stage 25 the mouthparts of *M. alipioi* is already fully formed, as the labial tooth rows reached their definitive number, like most species with a 2/3 tooth row pattern (Gosner and Black 1954, Sedra and Michael 1961, Tubbs *et al.* 1993, Gómez *et al.* 2016). Although the pigmentation of the jaw sheaths is still limited to their edges, they are completed at the next Stage. The first sign of degeneration of larval mouthparts is the loss of labial teeth. In *Dryophytes chrysolcelis* (Cope, 1880), Thibaudeau and Altig (1988) noted that the loss of labial teeth seems to occur haphazardly in all rows; however, in this species the P3 row is absent well before other rows, exactly the last labial teeth row to arise in early postembryonic development. These findings suggested the disappearance could occur in the reverse order of appearance. Our results for *M. alipioi* were not in agreement with this suggestion, since P3 was at the beginning of degeneration when there were only traces of A1, A2, P1 and P2.


Downie *et al.* (2004) found two patterns of tail loss during metamorphosis of ten anuran species: (1) during the first day, the tail reduction is dramatic (to less than 60% of the maximum), then slower over the final stages; and (2) the tail remains at over 80% of its full length for the first day after forelimb emergence, with variable rates of reduction thereafter. In all these cases, tail retention during the early part of metamorphosis was associated with remaining in the water and rapid tail loss with early emergence from the water. Despite the absence of data on developmental time, *Macrogenioglottus alipioi* seems to fit on the first pattern, from Stage 40 there is an abrupt decrease in total length, mainly due to tail atrophy, although body length also decreases during this period. The species left the water soon after forelimb emergence. At the end of the metamorphosis, the snout-vent length is equivalent to the tadpole total length at stages 26–27 (~18 mm) which indicates that substantial post-metamorphic growth until adulthood will occur (~70–100 mm).

The emergence of the forelimbs is usually pointed to as the initial event of the metamorphic climax (Etkin 1936, Gosner 1960). However, according to Hall *et al.* (1997), the abrupt decrease in tail and body sizes may indicate the onset of the metamorphic climax if it occurs clearly before the emergence of the forelimbs. In *Macrogenioglottus alipioi*, the reduction in size of the tadpoles, which starts at Stage 40, was the event that preceded all typical changes of the beginning of metamorphic climax, suggesting the initial sign of metamorphosis, similarly to what occurs in *Odontophrynus cordobae* (Grenat *et al.* 2011) and quite different from other species, such as *Dermatonotus muelleri*, in which body size increases until the emergence of the forelimbs (Fabrezi *et al.* 2012). These facts indicate that the initial event and the sequence of changes are variable among anurans (Fabrezi *et al.* 2012) and corroborate the model proposed by Smith-Gill and Berven (1979), which suggests that larval growth and development are decoupled events.

Macrogenioglottus has an interesting taxonomic history. This genus has been already included in its own family (Macrogenioglottidae; Reig 1972) and has been also considered a junior synonym of *Odontophrynus* (Lynch 1971). Despite the scarcity of data on larval development in Odontophrynidae, the peculiarity of some larval traits, such as the larger size and robustness of the larva at similar developmental stages, and mainly on call characteristic, as the nature pulsatile of the note, pointed out herein supports the taxonomic distinction of *Macrogenioglottus* evidenced previously by molecular data (Amaro *et al.* 2009, Pyron and Wiens 2011, Teixeira-Junior *et al.* 2012).

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Appendix I. *Specimens examined of Macrogenioglottus alipioi from northeastern Brazil. Localities (Alagoas state): MC, Mata do Catolé, Maceió; EEM, Estação Ecológica de Murici, Murici.*

MHNUFAL No.	Stage	N	Locality	MHNUFAL No.	Stage	N	Locality
10982	clutch	-	MC	8432	36	1	MC
9343	25	4	MC	9059	36	1	MC
11276	25	1	MC	10166	36	1	MC
11276	26	1	MC	10987	37	3	MC
11084	26	4	MC	11049	37	1	MC
10165	27	2	MC	11038	38	2	MC
9343	27	1	MC	10165	38	1	MC
11084	27	1	MC	11049	39	1	MC
10811	27	1	MC	11049	40	3	MC
10811	28	1	MC	10985	40	1	MC
9059	28	1	MC	3784	40	1	MC
11276	28	2	MC	10811	41	4	MC
8435	29	1	MC	8430	42	1	MC
10811	29	1	MC	9788	42	4	MC
10166	30	1	MC	10166	43	2	MC
10811	30	3	MC	10985	43	1	MC
10811	31	3	MC	9059	43	2	MC
10166	31	2	MC	2771	44	1	MC
9059	32	1	MC	2770	45	1	MC
10987	32	2	MC	10940	adult male	-	MC
8430	33	1	MC	10941	adult female	-	MC
10165	33	1	MC	8165	adult male	-	MC
10987	33	1	MC	8166	adult male	-	MC
10811	33	1	MC	4948	adult male	-	MC
10811	34	1	MC	5493	adult male	-	MC
8430	34	1	MC	5494	adult male	-	MC
10166	34	1	MC	10942	adult male	-	MC
8432	34	1	MC	5483	adult male	-	MC
9345	34	1	MC	12074	adult female	-	EEM
8433	35	1	MC	12075	adult male	-	EEM
11038	35	1	MC	12076	adult male	-	EEM
8432	35	3	MC				

Appendix II. Measurements (in mm) of *Macrogenioglottus alipioi tadpoles*. Data presented as mean \pm SD. Stages are according to Gosner (1960).

Measurements	Stage 25 (N = 5)	Stage 26 (N = 5)	Stage 27 (N = 5)	Stage 28 (N = 4)	Stage 29 (N = 2)	Stage 30 (N = 4)	Stage 31 (N = 5)	Stage 32 (N = 3)	Stage 33 (N = 4)	Stage 34 (N = 4)	Stage 35 (N = 5)
Total length	14.4 \pm 1.4	25.3 \pm 1.9	26.9 \pm 5.3	33.7 \pm 6.1	39.7 \pm 4.6	35.5 \pm 2.5	41.9 \pm 2.7	49.5 \pm 1.1	45.6 \pm 2.7	48.4 \pm 7.8	49.9 \pm 7.1
Body length	7.0 \pm 0.2	11.8 \pm 0.6	13.2 \pm 3.4	15.1 \pm 3.9	18.1 \pm 0.9	19.2 \pm 1.7	19.4 \pm 1.6	22.4 \pm 0.2	21.2 \pm 1.8	21.0 \pm 4.7	22.4 \pm 3.2
Tail length	7.5 \pm 1.3	13.5 \pm 1.7	13.7 \pm 2.2	18.5 \pm 2.9	21.6 \pm 3.6	16.3 \pm 1.2	22.5 \pm 3.4	27.1 \pm 1.2	24.3 \pm 2.2	27.4 \pm 3.4	27.5 \pm 4.2
Tail muscle height	1.7 \pm 0.1	2.7 \pm 0.2	3.3 \pm 0.6	3.8 \pm 0.8	4.7 \pm 0.5	5.0 \pm 0.3	5.6 \pm 0.3	5.3 \pm 0.3	5.4 \pm 0.8	5.1 \pm 1.0	6.0 \pm 0.5
Maximum tail height	4.4 \pm 0.2	7.2 \pm 0.5	7.7 \pm 1.8	8.3 \pm 1.9	10.3 \pm 1.2	10.4 \pm 1.2	10.6 \pm 0.4	10.7 \pm 2.1	10.6 \pm 0.7	11.8 \pm 1.3	11.8 \pm 2.8
Maximum body height	3.9 \pm 0.2	7.4 \pm 0.8	7.6 \pm 1.6	8.2 \pm 2.3	10.6 \pm 0.6	10.6 \pm 1.0	9.9 \pm 1.3	10.6 \pm 0.9	10.2 \pm 1.9	10.6 \pm 3.8	11.8 \pm 2.1
Maximum dorsal fin height	1.8 \pm 0.1	2.7 \pm 0.2	2.7 \pm 0.8	3.1 \pm 0.6	4.1 \pm 0.5	3.9 \pm 0.4	4.5 \pm 0.4	4.6 \pm 0.9	4.6 \pm 0.8	5.1 \pm 0.8	4.7 \pm 1.8
Maximum ventral fin height	1.3 \pm 0.1	2.1 \pm 0.1	2.1 \pm 0.7	2.4 \pm 0.7	3.1 \pm 0.1	3.0 \pm 0.4	3.4 \pm 0.4	3.6 \pm 0.6	3.4 \pm 0.6	3.5 \pm 0.8	3.3 \pm 1.4
Snout–nostril distance	0.8 \pm 0.0	1.7 \pm 0.2	1.4 \pm 0.5	1.8 \pm 0.2	1.8 \pm 0.3	1.9 \pm 0.1	2.0 \pm 0.2	3.0 \pm 0.1	2.4 \pm 0.5	2.9 \pm 0.4	2.8 \pm 0.1
Eye–nostril distance	0.7 \pm 0.1	1.5 \pm 0.2	1.2 \pm 0.3	1.6 \pm 0.4	2.0 \pm 0.4	2.2 \pm 0.3	2.2 \pm 0.2	2.3 \pm 0.5	2.5 \pm 0.4	2.1 \pm 0.2	2.3 \pm 0.6
Eye diameter	0.8 \pm 0.1	1.3 \pm 0.1	1.4 \pm 0.4	1.7 \pm 0.4	2.0 \pm 0.2	1.8 \pm 0.1	1.9 \pm 0.2	2.5 \pm 0.1	2.0 \pm 0.3	2.4 \pm 0.5	2.5 \pm 0.3
Intraocular distance	0.7 \pm 0.0	1.0 \pm 0.1	1.4 \pm 0.3	1.0 \pm 0.2	1.5 \pm 0.2	1.5 \pm 0.1	1.6 \pm 0.1	1.8 \pm 0.3	1.6 \pm 0.1	1.8 \pm 0.8	1.6 \pm 0.4
Intraocular distance	1.6 \pm 0.1	2.8 \pm 0.3	2.9 \pm 0.6	3.0 \pm 0.9	4.3 \pm 0.5	3.9 \pm 0.7	4.4 \pm 0.4	5.0 \pm 0.6	4.8 \pm 0.6	4.6 \pm 1.4	4.6 \pm 1.3
Tail muscle width	1.0 \pm 0.1	1.8 \pm 0.2	2.3 \pm 0.3	2.6 \pm 0.5	3.4 \pm 0.1	3.5 \pm 0.5	3.7 \pm 0.1	4.6 \pm 0.6	4.0 \pm 0.4	4.1 \pm 0.6	4.4 \pm 0.4
Maximum body width	6.2 \pm 0.3	9.8 \pm 0.7	9.5 \pm 2.0	11.2 \pm 2.4	12.6 \pm 1.6	12.9 \pm 1.9	12.9 \pm 0.8	14.3 \pm 0.6	14.7 \pm 1.4	15.1 \pm 4.5	15.8 \pm 3.2
Oral disc width	2.1 \pm 0.2	3.1 \pm 0.2	3.1 \pm 0.9	3.8 \pm 1.0	4.1 \pm 0.4	4.3 \pm 0.3	4.8 \pm 0.4	6.4 \pm 0.3	4.9 \pm 0.4	4.7 \pm 0.4	5.0 \pm 0.5

Appendix II. *Continued.*

Measurements	Stage 36 (N = 2)	Stage 37 (N = 4)	Stage 38 (N = 3)	Stage 39 (N = 1)	Stage 40 (N = 5)	Stage 41 (N = 4)	Stage 42 (N = 5)	Stage 43 (N = 5)	Stage 44 (N = 1)	Stage 45 (N = 1)
Total length	58.8 ± 1.2	53.9 ± 8.4	59.8 ± 10.5	69.9	65.1 ± 11.3	61.7 ± 7.3	41.7 ± 3.1	37.5 ± 7.0	18.6	17.9
Body length	24.9 ± 1.1	25.6 ± 4.3	26.3 ± 5.7	32.3	28.0 ± 4.6	25.4 ± 2.2	19.6 ± 1.8	25.0 ± 3.4	18.6	17.9
Tail length	34.0 ± 2.3	28.3 ± 4.1	33.5 ± 4.9	37.7	37.1 ± 6.7	36.5 ± 6.1	22.1 ± 4.9	12.5 ± 6.8	1.1	1.0
Tail muscle height	6.0 ± 0.4	5.8 ± 0.9	6.5 ± 1.3	8.1	6.8 ± 1.2	5.9 ± 0.7	4.5 ± 0.4	5.8 ± 1.0	-	-
Maximum tail height	13.1 ± 1.3	11.8 ± 2.8	15.1 ± 4.8	18.1	15.8 ± 2.3	13.4 ± 0.8	7.1 ± 1.3	7.1 ± 2.1	-	-
Maximum body height	12.2 ± 0.9	13.5 ± 3.4	12.7 ± 3.9	19.6	14.6 ± 3.6	12.5 ± 1.4	8.2 ± 1.0	9.4 ± 1.3	7.3	6.0
Maximum dorsal fin height	5.9 ± 0.3	5.2 ± 1.5	6.7 ± 1.8	7.9	7.2 ± 1.0	6.1 ± 0.5	2.8 ± 0.5	1.6 ± 1.1	-	-
Maximum ventral fin height	3.7 ± 0.5	3.6 ± 0.7	3.7 ± 0.7	6.5	4.8 ± 0.9	4.3 ± 0.4	1.6 ± 0.4	0.9 ± 0.6	-	-
Snout–nostril distance	2.9 ± 0.2	3.3 ± 0.5	3.9 ± 1.0	4.4	3.2 ± 0.7	3.0 ± 0.9	1.1 ± 0.2	1.3 ± 0.2	0.4	0.2
Eye–nostril distance	2.9 ± 0.2	2.5 ± 0.4	2.9 ± 0.4	2.7	2.6 ± 0.5	2.4 ± 0.3	2.1 ± 0.2	1.9 ± 0.3	1.7	1.4
Eye diameter	2.3 ± 0.0	2.8 ± 0.5	2.8 ± 0.5	3.3	3.2 ± 0.4	3.3 ± 0.0	2.4 ± 0.2	2.5 ± 0.5	2.2	2.1
Intranarial distance	1.9 ± 0.1	1.8 ± 0.2	2.0 ± 0.2	2.3	1.8 ± 0.4	1.7 ± 0.2	1.3 ± 0.1	1.6 ± 0.2	1.5	1.1
Intraocular distance	5.3 ± 0.2	5.2 ± 1.3	5.5 ± 1.9	6.2	4.6 ± 1.0	8.1 ± 0.1	6.4 ± 0.2	7.9 ± 0.7	7.1	6.7
Tail muscle width	4.1 ± 0.3	4.4 ± 0.7	5.4 ± 1.3	6.5	5.9 ± 1.0	5.0 ± 0.4	3.9 ± 0.4	5.2 ± 0.9	-	-
Maximum body width	16.7 ± 1.2	17.5 ± 3.4	18.5 ± 3.9	24.7	20.6 ± 4.2	18.0 ± 1.7	11.3 ± 1.4	12.4 ± 1.6	8.8	9.1
Oral disc width	5.6 ± 0.1	6.5 ± 1.0	6.7 ± 1.1	8.3	6.3 ± 1.3	6.1 ± 0.6	4.9 ± 0.5	7.0 ± 1.8	7.0	7.3