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Descriptions and biological notes on three unusual mantellid tadpoles (Amphibia: Anura: Mantellidae) from southeastern Madagascar

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Abstract.—The morphologies of three unusual tadpoles from slow-flowing, sandy-bottomed, rain forest streams in southeastern Madagascar are described. The large oral apparatus of the tadpole of *Boophis picturatus* Glaw, Vences, Andreone, and Vallan, 2001 lacks all keratinized structures and has an elaborately-folded lower labium with five, radially oriented, flat-topped ridges.

The tadpole of *Mantidactylus guttulatus* (Boulenger, 1881) lacks all keratinized mouthparts and has three immense papillae where the upper jaw normally occurs.

The tadpole of *Mantidactylus lugubris* (Duméril, 1853) has an ornate oral apparatus involving greatly hypertrophied derivatives of jaw serrations and unique structures on the lower labium that resemble labial teeth.

The endemic anuran fauna of Madagascar is dominated by species in the genera *Boophis* and *Mantidactylus* (Mantellidae). Their described tadpoles typically have a common morphology, (i.e., 3–8 upper labial tooth rows, commonly three lower tooth rows, usually emarginate oral disc with a dorsal gap in the marginal papillae, sinistral spiracle, dextral vent, and dorsal eyes). Thus, the collection of three tadpoles with bizarre oral structures from streams in the vicinity of Ranomafana, Fianarantsoa Province, was completely unexpected. These tadpoles have characteristics that considerably expand our understanding of the diversity of tadpole oral morphology. The larvae also show extremes in modifications of typical oral morphology and differ strikingly from known tadpoles in their respective generic and species groups.

Materials and Methods

The oral terminologies of Altig & McDiarmid (1999) and the staging table of Gosner (1960) are used. Muscles of the oral apparatus were viewed with transmitted polarized light (e.g., Carr & Altig 1991), and translucent tissue was stained with Crystal violet to increase contrast. Formalin-preserved specimens were critical-point dried, sputter coated with gold-paladium, and viewed with a Ziess SMT Stereoscan 360. Measurements were made with an ocular micrometer (0.1 mm), and those involving the spiracular aperture, eyes, and nares were made from the centers of those structures. Body terminus was defined as the junction of the posterior body wall with the axis of the tail myotomes. Gut contents of one species were boiled in nitric acid to isolate

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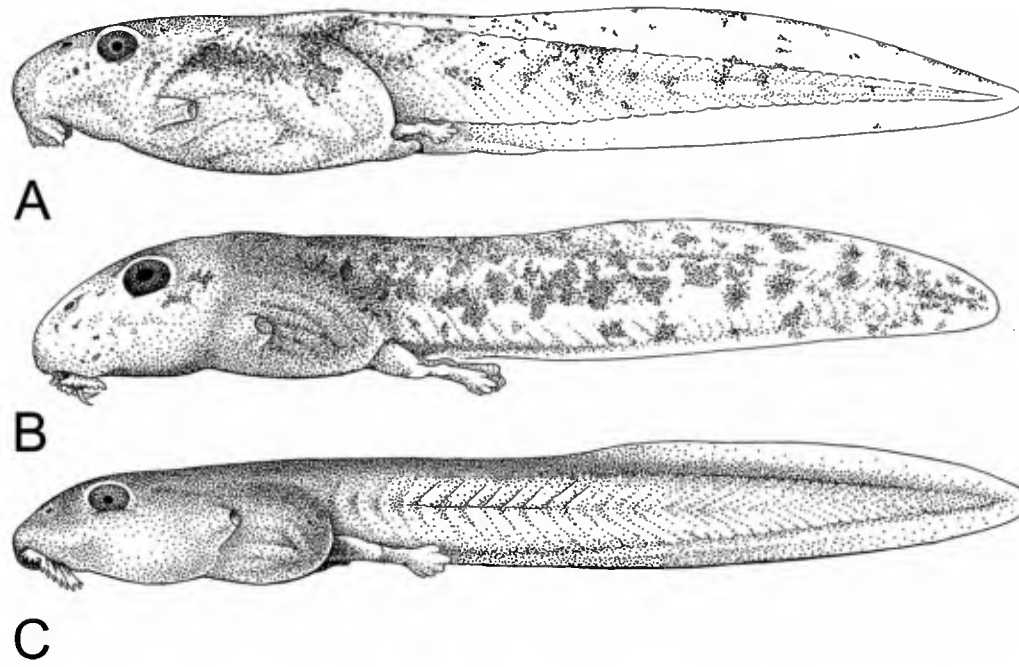

Fig. 1. Left, lateral views of tadpoles of (A) *Boophis picturatus*, (Stage 37, 35 TL, USNM 563654), (B) *Mantidactylus guttulatus* (Stage 27, 29.4 TL, USNM 563657), and (C) *M. lugubris* (Stage 30, 30.7 TL, USNM 563659).

the diatoms. All specimens were collected by RA from 15–30 Dec 1991 from small streams and rivers in Ranomafana National Park, ca. 6 km west of Ranomafana, Fianarantsoa Province, Madagascar, and representative samples are deposited in the collections of the National Museum of Natural History (USNM).

Tadpole Descriptions

Boophis picturatus Glaw, Vences, Andreone, & Vallan, 2001
Figs. 1A, 2A, D

Our identification of these tadpoles is based, in part, on data derived from a molecular survey of Malagasy tadpoles (e.g., Thomas et al. 2005). A tadpole with the morphology described here matched the mitochrondrial 16S RNA sequence of a tadpole from Ranomafana (ZSM 821/2004; Genbank accession DQ367352) which had more than 456 base pairs identical to the sequence of adults positively identified as *Boophis picturatus* Glaw et al., 2001 (ZSM 672/2003; Genbank accession AY848610; M. Vences, pers. comm.).

Twenty-two specimens (USNM 563652-54) in Gosner Stages 25–46 were available for study. Measurements (mm) of a Gosner Stage 37 specimen (USNM 563654) are: 35 total length (TL), 13 body length, 22 tail length, 4.4 tail muscle height at base, 3 tail muscle width at base, 2.4 maximum dorsal fin height located 13 from body terminus, 1.4 maximum ventral fin height located 11.2 from body terminus, 8.8 body width located 7.5 from snout, 6.3 body height located 7.9 from snout, 1.7 eye diameter, 0.8 pupil diameter, 4 interorbital distance, 0.4 narial diameter, 2.5 internarial distance, 2.1 snout to naris, 4.3 snout to eye, 7.5 snout to spiracle, 3.4 naris to eye, and 4.5 transverse diameter of oral disc. Other major characteristics are: oral

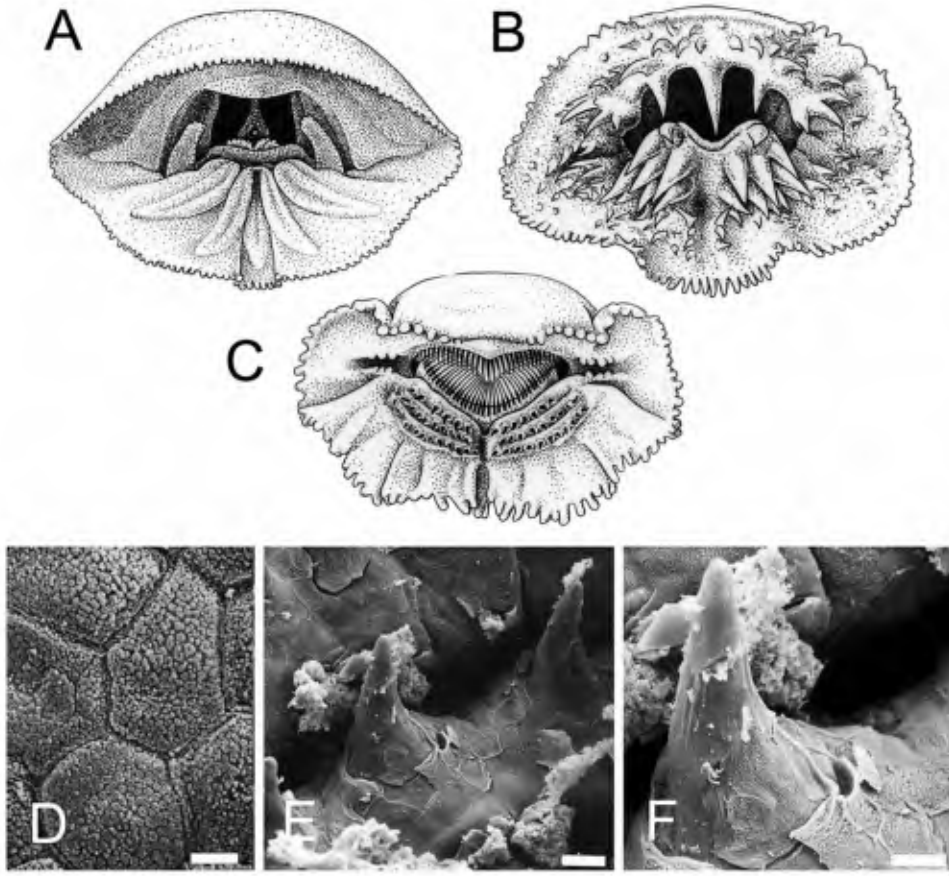

Fig. 2. Oral apparatus of (A) *Boophis picturatus* (transverse oral disc width = 4.5 mm), (B) *Mantidactylus guttulatus* (2.7), (C) *M. lugubris* (3.1) and (D) surface epithelial cells on the labial ridges of *Boophis picturatus* (scale bar = 20 μm) and labial tooth-like structures on the lower labium of *M. lugubris* at (E) low (scale bar = 89 μm), and (F) high (scale bar = 31 μm) magnifications.

disc large, ventral, and nonemarginate with complicated folds in the lower labium; marginal papillae tiny, uniserial, and complete around disc; eyes dorsal; spiracle sinistral; vent dextral; dorsal fin terminating at dorsal extent of body; neuromasts not visible; and all keratinized mouthparts absent.

The body is slightly depressed and flat ventrally; the buccopharyngeal area constitutes ca. 28% of the ventral body length. The snout is broadly rounded in dorsal view, and the nares have a pronounced pigmented rim with a slight dorsal protuberance. The low fins terminate in a point (Fig. 1A). The spiracular tube has a short medial wall. The large-diameter vent tube lies totally to the right of the ventral fin, and its ventral wall is large (2.5 mm long).

The jaws lack keratinized sheaths; in profile the suprarostral is medially convex (0.2 mm measured longitudinally), and the infrarostrals are broadly U-shaped. At rest, the large upper labium is somewhat bowl-shaped and nearly overhangs the mouth (Fig. 2A); typical submarginal papillae are lacking. A single, sagittal, low-profile ridge and two pairs of lateral ones extend radially from the mouth for most of the length of the labium but are not easily visible without staining; the cell margins (Fig. 2D) are well defined by intervening crevices that

abound with microvilli. At low magnifications, the cell surfaces appear slightly rougher than the surroundings; at higher magnifications the surfaces are uniquely textured.

A Gosner Stage 40 specimen (37.5 TL) has six myotomes in the rectus abdominis in parallel-sided bands. Fibers of the first three myotomes are tightly packed and the myosepta are closed; more anterior bundles have fewer fibers and open myosepta. The musculus mandibulolabialis superior is absent, and the musculus mandibulolabialis inferior is present as a small bundle beneath the most lateral part of the longitudinal ridges on the lower labium. Buccopharyngeal papillae include a small, medially located hemispherical flap with a crenulate edge, a profusion of tightly spaced buccal floor and buccal roof arena papillae with ornate, papillate tips, and a cluster of tightly spaced, similarly shaped papillae between the infrarostral cartilages. The longest buccal floor arena papillae are positioned laterally and lack ornamented tips. Two large lingual papillae with large, clavate, papillate heads are arranged transversely on the oval tongue anlage and bordered posteriorly by four smaller but similarly structured papillae.

In life the tadpoles are nearly transparent with a blotched pattern of melanophores and gold to silver iridophores. Patches of iridophores positioned in layers deeper than the skin form spots on the dorsolateral body wall adjacent to the base of the tail musculature and below the eye. In preservative, the body of the tadpole is colorless, except for scattered dots and small blotches of melanic pigment in the skin and deeper layers; dark pigment is concentrated over the brain and dorsolaterally in the abdominal wall directly behind the front limbs. The dorsum of the tail muscle has 4–5 diffuse bands, and the more prominent anterior band slants anteriorly. Small, scattered black blotches occur on the dorsal fin and tail musculature; the ventral fin and entire ventral surface are nonpigmented and transparent. The coloration provides amazing camouflage against the sandy substrate. Tadpoles resting in shallow depressions in the sandy bottom could not be seen even in 3–10 cm of water (1–2 m wide).

This tadpole surely is one of the most specialized foragers among known frog larvae. Tadpoles of *B. picturatus* were found only in a short (~10 m) section of a shallow, low gradient stream dubbed Sand Creek about 0.6 km (by trail) NE of the Research Camp. Live and preserved specimens (examined at 50×) contained nothing but large sand grains; no organic material was visible. The dry mass of sand (0.63 g) taken from the gut of a Gosner Stage 36 (40.8 TL) individual was 30% of the intact wet mass (2.1 g) of the tadpole. Fifteen of the largest sand grains ($\bar{x}$ = 4.1 ± 0.7 mm longest dimension) made up 54% of the weight of the gut contents, and many of the remaining grains were not especially small. The sizes and distribution of sand grains suggest that this tadpole ingests grains in a relatively narrow range. Whether the tadpoles feed preferentially in a part of the stream where currents have sorted the substrate by relative grain size, or whether they selected a narrow range of grain sizes from among those available is not known. For several reasons, we believe the former is true. Tadpoles were found only where the sandy substrate was relatively coarse, and grains in the gut were of similar size. The guts of several other kinds of tadpoles from other microhabitats on the same stream typically contained dark, organic debris. Second, these tadpoles were never found in microhabitats where tadpoles of other species were common or over substrates of finer or coarser sand than in the stream center. Examinations of the gut contents revealed diatoms in eight genera, but at least 90% were species of *Eunotia* (Eunotiaceae), a pennate form

that prefers acidic, oligotrophic waters. Because diatoms and other microalgae inhabit interstices among sand grains where light can penetrate, it seems likely that moderately large grains match that size and that the tadpoles feed in portions of the stream where diatoms are readily available and easily harvested.

Observations of specimens feeding in the lab showed the lower labium extending distally and laterally so that the sausage-shaped ridges separate in a radial pattern; during closure, the disc forms pleats as it retracts proximally and medially. At rest, the pleats on the lower labium are arranged so that the two parasagittal ridges lie next to each other and cover a depression in which the sagittal ridge sits.

Mantidactylus guttulatus
(Boulenger, 1881)
Figs. 1B, 2B

Several species in this group (subgenus *Mantidactylus*) likely occur near Ranomafana, and these tadpoles were identified tentatively by comparing the patterns of hand and foot webbing and the distinctive femoral gland of a stage 41 individual with syntopic adults.

Seven tadpoles in Gosner Stages 25, 27, and 41 (USNM 563655-57) from the Sakaroa River were available. Measurements (mm) of a stage 27 specimen (USNM 563657) are: 29.4 total length, 10.5 body length, 18.9 tail length, 3 tail muscle height at base, 2.6 tail muscle width at base, 1.4 maximum dorsal fin height located 11.3 from body terminus, 1 maximum ventral fin height located 13.7 from body terminus, 7.3 body width and 4.8 body height located 5 from snout, 1.5 eye diameter, 0.7 pupil diameter, 4 interorbital distance, 0.1 narial diameter, 2.5 internarial distance, 1.5 snout to naris, 4.1 snout to eye, 8 snout to spiracle, 2.6 naris to eye, and 2.7 transverse diameter of oral disc. Other important characteristics are: oral disc ventral, nonemarginate, and transversely oval; eyes dorsal; spiracle sinistral; vent dextral; dorsal fin terminating 1.0 mm before dorsal body terminus; ventral fin terminating at body; neuromasts not visible; and keratinized mouthparts absent.

Three immense, flexible, slightly curved, thorn-shaped papillae extend from the region where the upper jaw sheath normally occurs and lie in the spaces between the folds of the lower labium. The larger central papilla is ca. 0.6 mm. A similar papilla is situated lateral to the bases of the outside members of the trio. The oral disc (Fig. 2B) has prominent, widely spaced, uniserial, marginal papillae (16 per mm ventrally) that become less prominent or form wavy crenulations dorsolaterally. A dorsal median gap occurs in the marginal papillae. Spine-like, submarginal papillae are abundant and diverse; they include ca. 20 papillae that lie in a transverse line anterior to the three large papillae, a large, basally-trifid, lateral papillar unit on each side of the mouth, many smaller papillae scattered across the lower labium, and 5–6 large papillae that project from near the base of what appears to be a nonpigmented, nonkeratinized, nonserrate lower jaw sheath.

The body is flat ventrally and widest at about the plane of the eyes; a constriction occurs at the spiracular wall. A distinct rim on the posteromedial quadrant of the narial aperture is absent anterolaterally. The tail terminates in a narrow point (Fig. 1B). The nonpigmented spiracular tube has a free, projecting (ca. 0.3 mm) medial wall. The largely nonpigmented vent tube attaches to the right side of the ventral fin and has a large aperture; dusky pigment occurs in the ventral wall near the aperture.

In preservative, the entire, nonpigmented venter is transparent, as is a sizeable area around and anterior to the eye. The remainder of the dorsum is crudely

blotched with dark pigment. The dorsal fin and tail musculature are patterned with many distinct, irregular blotches formed from pigment in the skin and deeper layers near the muscles; the ventral side of the caudal musculature and the ventral fin are nonpigmented.

A Gosner Stage 41 specimen (40 TL; USNM 563656) has the major features described for the stage 27 specimen, and the oral disc shows no signs of metamorphic atrophy. Neuromasts are visible near the eye. The area in front of the eye is pigmented, the eyes face more laterally, and the nasolacrimal duct projects from the naris to near the front of the eye. The ventral marginal papillae also are larger (0.2 mm), and those on the lateral parts of the disc are more distinct and pointed than in the stage 27 tadpole. A golden tint on the surface of the distal two-thirds of the large, upper, middle papilla suggests light keratinization, and the ventral margin of the oral disc is not folded as orderly as in the smaller specimens.

Some tadpoles of *M. guttulatus* were found in leaf mats in the same small stream (Sand Creek) in which *B. picturatus* occurred. Those described here were taken under small, rather dispersed leaf mats in the shallows of the Sakaroa River.

Mantidactylus lugubris (Duméril, 1853)
Figs. 1C, 2C, E, F

Six specimens (USNM 563658–60) in Gosner Stages 25–43 were examined and identified by a metamorphic individual and figures in Glaw & Vences (1992). Unfortunately, multiple taxa are known by this name, and the ultimate name to be associated with the Ranomafana population(s) will require additional study (M. Vences, pers. comm.). Measurements (mm) of a Gosner Stage 30 specimen (USNM 563659) are: 30.7 total length, 9.6 body length, 21.1 tail length, 2.8 tail muscle height at base, 2.6 tail muscle width at base, 1.2 maximum dorsal fin height located 13.7 from body terminus, 1 maximum ventral fin height located 16.2 from body terminus, 5.9 body width located 4 from snout, 3.8 body height located 5.2 from snout, 1.1 eye diameter, 0.3 pupil diameter, 2.7 interorbital distance, 0.09 narial diameter, 1.8 internarial distance, 1.1 snout to naris, 2.9 snout to eye, 6.6 snout to spiracle, 1.7 naris to eye, and 3.1 transverse diameter of oral disc. Other pertinent characteristics include: oral disc almost ventral, nonemarginate, with median dorsal gap in marginal papillae; eyes dorsal; spiracle sinistral; vent dextral; dorsal fin terminating ca. 4.8 mm posterior to dorsal tail-body junction; ventral fin terminating at body; neuromasts not visible; and labial tooth row formula (LTRF) of 0/3(1). Glaw & Vences (1992:167, fig. 80, tad 29) provided small line drawings, a black and white photograph, and a brief description of this tadpole based on specimens from Tolagnaro in southern Madagascar. In the second edition of their field guide, they repeated the drawings and description and added scanning electron micrographs of the mouthparts (Glaw & Vences 1994:167, figs. 192, 193, tad 28).

The lower labium has widely spaced, conical, keratinized structures (12 per mm in left half-row of row P-1) in three rows, each with a median gap (Fig. 2C). These cones differ in structure and form from typical labial teeth (Fig. 2E, F), and each row sits atop a flat ridge that is delimited by crevices rather than being raised above the local terrain as with typical labial tooth ridges. Lateral views of hand-cut sections of the lower labium observed with polarized light show the cones sitting slightly within the epithelium, no replacement units beneath them, and their ridges lacking the massive amounts of connective tissue usually seen in tooth ridges. A demarcation on the upper third of the cone may indicate a fracture plane, but we have not seen a cone broken at this

point. The surface of the cone distal to this line differs from that of the basal two-thirds.

No such ridges or cones occur on the upper labium. The free margin of the upper labium stops dorsolaterally, and a series of short, uniserial "marginal" papillae situated directly on the surface of the snout extend medially for a short distance. The ventral marginal papillae are considerably longer than the lateral ones. Two short rows of stubby papillae lie immediately lateral to the jaws and appear to interdigitate when the labia come together during closure.

The plane of the upper jaw is recessed abruptly from the level of the papillae on the snout. The margins of the arms of the widely V-shaped upper sheath are slightly concave, and a single series of long (ca. 0.2 mm medially), slightly flattened, parallel, straight, lightly keratinized spikes (ca. 22 per mm) project posteriorly from it. The lengths of the spikes decrease laterally, so that a line connecting their tips mirrors the outline of the base of the upper jaw sheath. A series of about 54 long, recurved spikes (33 per mm) with rounded front faces project anteriorly and form the lower jaw armament. Even though the basal three-quarters of the jaw spikes are pigmented and project from the jaw tissue, there is no indication that these spikes are formed of repeating units like serrations of a typical jaw sheath. Live tadpoles can open their mouths far enough to clear the two sets of spikes. The track of the lower spikes forms a wide, flat-bottomed U-shape, and observations of live animals suggest that this curvature can be altered considerably.

The depressed body (Fig. 1C) is flattened ventrally and is widest slightly behind the plane of the eyes. The buccopharyngeal area constitutes ca. 46% of the ventral body length. The low, mostly nonpigmented fins join in a rounded point. The sinistral spiracular tube has a short, free medial wall. In dorsal and lateral view this tadpole resembles a darkly pigmented tadpole of *Hyalinobatrachium* sp. (Centrolenidae).

In life, all upper surfaces are uniformly black, although at 10× magnification, one can see a sparse but uniform peppering of bright green chromatophores throughout the dorsum. At about stage 40, the chromatophores begin to aggregate to form the transverse body band of the adult, and red gills are visible through the skin. The venter is mostly clear of pigment, and, except for a general fading to dark brown, no notable color changes occurred in preservative.

A Gosner Stage 25 (16.9 TL) preserved specimen agrees with the above description, except that it is more pale and less uniformly pigmented; row P-3 is only faintly developed, and the jaw spikes are less intensely pigmented. In a Gosner Stage 40 (31.8 TL) specimen, row P-3 is somewhat less prominent, neuromasts are visible near the eye, and the throat and belly have a scant suffusion of melanophores.

In a Gosner Stage 26 (37.5 TL) specimen, a feltwork of fine, criss-crossed collagen fibers gives a sheen to the throat and anterior part of the belly. The musculus mandibulolabialis is absent, and the rectus abdominis has six myotomes with closed myosepta.

In the buccopharynx, a small, subrectangular medial flap with irregular edges is present, and the immense internal naris sits in a reverse-comma shaped depression; the anterior end of the pocket is minutely papillate. A slight, U-shaped wall in the prenarial pocket has a minutely papillate border. A single, large prenarial papilla and one large, medial and several smaller, postnarial papillae are visible. A few buccal floor arena papillae occur in two sparse lines. Two, small, lingual papillae are arranged transversely and lean toward each other. Two transverse rows of papillae are positioned directly behind the lower jaw.

All tadpoles of this widespread species were collected in December above and below Riana Cascade of the Sakaroa River in Ranomafana National Park. This reach is ca. 5 m wide and 20–50 cm deep and flows at ca. 30 cm/sec. The river is a mosaic of quietly flowing water over a sandy bottom interspersed with riffles of cobble and rock. Specimens were captured primarily from the upstream ends of leaf mats (ca. 0.3–10 m^2) that formed immediately below riffles or cascades where the diurnally-active adults were abundant. Specimens in a dip net moved by serpentine undulations of the body and tail rather than by the rapid tail-beats typical of most tadpoles. Tadpoles (Gosner Stages 27–30) maintained in containers with fine sand never attempted to burrow but always hid under a leaf.

The arrangement of the dorsal marginal papillae, the presence of keratinized 'teeth' only on the lower labium, the nature of the tooth ridges, and the unusual jaw sheaths are unique. The spike-like jaw structures probably act in a sieving or winnowing action that allows selective gathering of small particles. Very specific flow rates may be important, and whether the tadpoles feed by active buccal pumping or passive filtration (e.g., *Otophryne robusta*, Wassersug & Pyburn 1987) is not known.

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