

The tadpole of *Litoria multiplicata* (Anura: Hylidae) from Papua New Guinea

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ABSTRACT - We describe the tadpole of *Litoria multiplicata* from Papua New Guinea. It is readily distinguished from other *Litoria* tadpoles known from New Guinea by its distinctive pigment patterns consisting of bold black or dark brown stripes with scattered gold iridophores in the intervening pale areas. The oral disc is located ventrally and is completely surrounded by small papillae, adaptations for adherence to rocks in small streams where rapid increases in water flow are a common occurrence.

INTRODUCTION

The green treefrog, *Litoria multiplicata*, (Tyler, 1964) is a medium-sized species (to ~47 mm SVL) known from several localities between about 1,000 and 1,500 m a.s.l. along Papua New Guinea's central cordillera. The distribution and advertisement call of this species were described by Richards et al. (2009). Although the egg-deposition behaviour of *L. multiplicata* remains unknown, Menzies (2006) included the species in his '*L. iris* group', an assemblage of species that lay their eggs on leaves hanging over water, because a female examined contained only 'eight mature ova of slightly more than 2 mm diameter' suggesting it 'places its eggs in aerial situations'. He also provided a lateral drawing of a tadpole that he assigned to this species, with the brief description 'strikingly marked in black and gold, the fins moderately arched and evenly tapering to a narrow terminal ending' (Menzies, 2006: p146). No additional information on the tadpole of this species is available. Furthermore, the basis for the association of the tadpole illustrated by Menzies (2006) with *L. multiplicata* was not provided. Our collection of a small series of tadpoles that we assign to *L. multiplicata* confirms that the tadpole illustrated by Menzies (2006) is indeed likely to be that species, and provides the opportunity to present a detailed description of the body form and oral morphology of *L. multiplicata* tadpoles from Papua New Guinea.

METHODS

Collection details

Five tadpoles were collected from a small (1-3 m wide), shallow, rocky stream (Fig. 1a) in lower-montane rainforest at Tualapa, Southern Highlands Province, Papua New Guinea (05°18.245' S, 142°30.704' E, 1,438 m a.s.l.) between 11 and 26 July 2008. Tadpoles were placed in a tray and photographed in life, and then preserved in 5% formalin.

Measurements

Measurements of preserved specimens were taken in the laboratory using vernier calipers and an eye-piece micrometer attached to a Wild M5 stereoscopic microscope. The drawing of the oral disc was prepared with the aid of a drawing tube attached to the microscope. Tadpoles were staged according to Gosner (1960). Morphometric measurements used are explained in Table 1 and measurements are presented in Table 2. Description format, measurements and terminology follow Anstis (2013).

Identification

It was not possible to raise tadpoles to metamorphosis in the field to confirm identification, and all material was preserved in formalin precluding DNA matching. However we assign the tadpoles reported here to *L. multiplicata* on the basis that 1) males of this species were calling over the stream pools containing the tadpoles, 2) the only other *Litoria* species present during a week at the collection site was *L. modica*, a species with very different suctorial tadpoles (Richards, unpublished data), and 3) tadpoles identical to those described here were observed at another site further east in Papua New Guinea (Crater Mountain Wildlife Management Area, Eastern Highlands Province; 6°30.082' S, 145°01.977' E, 1,540 m a.s.l.), where again *L. multiplicata* adults were calling over the stream pools. At this latter site the only other *Litoria* species present along the streams over three weeks were *L. eucnemis* and *L. iris*, and neither of these species has tadpoles similar to those assigned here to *L. multiplicata* (Tyler, 1963; Anstis, 2013). We are therefore confident that the material described here represents *L. multiplicata*.

Character	Abbrev.	Description
Total length	TL	From tip of snout to tip of tail (lateral view)
Body length	BL	From tip of snout to tail-body junction at anterior ventral edge of tail muscle (lateral view)
Body depth	BD	Maximum vertical depth of body (lateral view)
Body width	BW	Maximum width of body across abdomen (dorsal view)
Eye-body width	EBW	Width of body in line with eyes (dorsal view)
Basal tail muscle depth	BTM	Maximum vertical depth of anterior tail muscle at tail-body junction (lateral view)
Snout to spiracle	SS	Distance from tip of snout to posterior dorsal edge of spiracular opening (lateral view)
Snout to eye	SE	Distance from snout to anterior edge of eye in (lateral view)
Snout to naris	SN	Distance from tip of snout to anterior edge of naris (lateral view)
Eye diameter	ED	Maximum eye diameter (lateral view)
Tail depth	TD	Maximum vertical depth of tail (lateral view)
Dorsal fin depth	DF	Dorsal fin vertical depth in line with TD (lateral view)
Tail muscle depth	TM	Tail muscle vertical depth in line with TD (lateral view)
Ventral fin depth	VF	Ventral fin vertical depth in line with TD (lateral view)
Inter-orbital span	IO	Distance between inner edge of each eye (dorsal view)
Inter-narial span	IN	Distance between inner edge of each naris (dorsal view)
Narial diameter	N	Maximum diameter of naris (dorsal view)
Oral disc width	ODW	Maximum width of oral disc (ventral view)
Snout-width	SW	Maximum width of snout in line with midline of oral disc (ventral view)
Labial tooth row formula	LTRF	after Altig (1970)

Table 1. Description of morphometric measurements of preserved tadpoles of *L. multiplicata*.

Stage	25	26	32	34	35
TL	34.5	38.5	46.6	45	45.5
BL	11.3	12.6	14.8	14.5	15.1
BD	5.2	5.5	7.0	7.0	7.2
BW	5.6	6.3	8.1	8.1	8.4
EBW	5.8	6.3	7.6	7.8	8.1
BTM	2.9	3.4	4.3	4.5	5.0
BTMW	2.7	3.2	4.3	4.7	4.8
SS	6.8	7.4	8.4	8.2	8.4
SE	3.6	3.5	4.4	4.2	4.2
SN	1.6	1.5	1.9	1.9	1.9
ED	1.0	1.2	1.5	1.6	1.9
TD	6.1	6.6	8.2	8.2	8.5
DF	1.9	1.9	2.6	2.6	2.7
TM	2.4	2.9	3.3	3.4	3.2
VF	1.8	1.8	2.3	2.3	2.6
IO	3.4	3.5	4.2	4.2	4.1
IN	2.4	2.7	3.1	3.2	3.2
N	0.3	0.3	0.4	0.5	0.5
ODW	3.2	3.4	4.5	4.3	4.3
SW	2.9	4.0	4.8	4.5	4.7

Table 2. Morphometric measurements of preserved tadpoles of *L. multiplicata*. Stages after Gosner (1960). See Table 1 for explanation of terminology.

RESULTS

Tadpole morphology

Five preserved tadpoles collected at stages 25, 26, 32, 34 and 35 were used for this description.

Maximum length

The latest developmental stage collected was stage 35, but the maximum total length of 46.6 mm was a specimen at stage 32. It is likely tadpoles may reach a slightly greater length than this in later stages. Limb development is delayed, and one tadpole at stage 25 (prior to the first emergence of a minute hind limb bud) is about three-quarters the body length and total length of the largest tadpole at stage 32.

Body

Body medium, oval to cylindrical and slightly depressed anteriorly. Abdomen slightly wider than deep in dorsal view. Snout broadly rounded in dorsal view, rounded in profile. Eyes appear near-lateral in life, tilted dorsally following preservation. The iris has a distinct gold ring around the pupil with a few fine iridophores scattered towards the margin. The nares are moderate in diameter (0.5 mm, stages 34 and 35), widely spaced and directed anterolaterally. The spiracle is narrow, relatively short and opens posteriorly or slightly dorsoposteriorly on or just below horizontal body axis, posterior to midpoint of body. The vent tube is dextral, broad, and opens diagonally down to edge of ventral fin (type b, Anstis 2013).

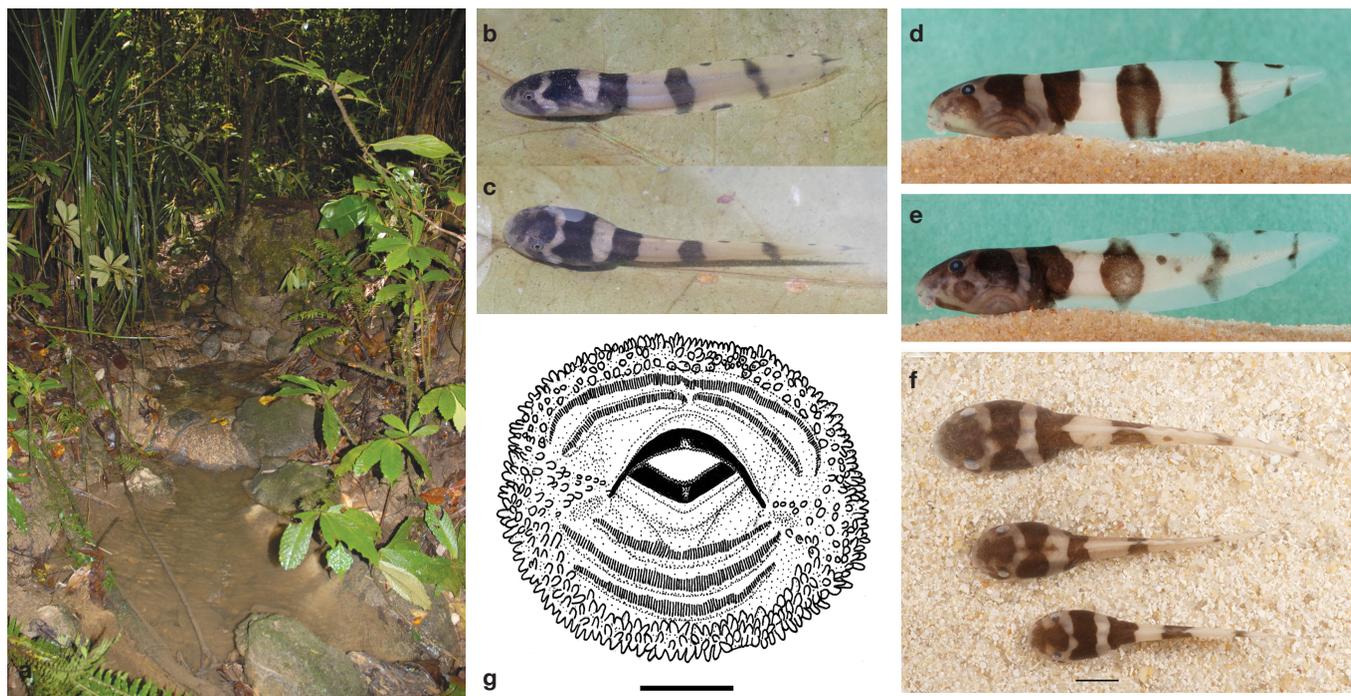


Figure 1a. Stream habitat of *L. multiplicata* in lower-montane rainforest at Tualapa, Southern Highlands Province, Papua New Guinea. **b** = dorsolateral and **c** = dorsal views of *L. multiplicata* tadpoles. **d** to **f** preserved tadpoles of *L. multiplicata* showing stage 26 (**d** lateral view), stage 32 (**b** lateral view) and **f** stages 32 (top) 26 (middle) and 25 (lower). **g** shows oral disc of *L. multiplicata* at stage 32. Bare represents 1mm.

Tail

Fins shallow. Dorsal fin begins at a point just proximal to body-tail junction and arches only slightly or not at all over midpoint, before tapering to a narrowly rounded tip. Ventral fin not arched. Muscle robust anteriorly, tapers posteriorly to a narrow point.

Colour in life

A strikingly beautiful species with bitonal pigmentation on body and tail (Fig. 1b to 1f). Three broad black or dark brown bands circumvent the dorsum and sides; one over the head from just posterior to the eyes to the snout, one across the mid-dorsal region and one over the base of the body and onto the tail. Between these bands are irregular pale areas with fine gold iridophores. The lateral line patterns are golden and clearly visible. As tadpoles grow, additional smaller dark spots may develop on the pale areas between the dark bands. In lateral view the abdomen becomes increasingly translucent towards the venter and intestines are partly visible. The tail is translucent milky-white with fine iridophores over the anterior dorsal muscle and fin and two prominent black or dark brown vertical bars, with a less prominent small vertical bar and or a few scattered dark spots posteriorly. Golden lateral line rows extend dorsolaterally along the muscle. The shape of the bands and other spots on body and tail is individually variable.

Colour in preservative.

Most pigment is retained in preservative except for the gold iridophores. The ventral surface is mostly translucent whitish to unpigmented in preservative but may have fine iridophores in life. The tail muscle is white and brown.

Oral Disc

Type 3 (Anstis, 2013), ventral. Three to four rows of small crowded papillae completely surround the disc, with additional submarginal papillae extending towards the jaw sheaths at each side. The disc is almost as broad as the snout (e.g. ratio of ODW to SW = 0.93, stage 32). Papillae reduced to two rows anteriorly in the medial margin of some specimens. Posterior papillae are generally larger than most other papillae. Two anterior and three posterior tooth rows, of which A¹ is the longest row and A² has a narrow medial gap. P¹ has a narrow medial gap in some specimens, but is fused in others. Jaw sheaths medium, upper sheath moderately arched with long sides. LTRF = 2(2)/3 or 2(2)/3(1). The oral disc is illustrated in Fig. 1g. It is likely that the oral disc of this tadpole is capable of maintaining adherence to rocks in flowing water, given its broad surface area and complete papillary border.

Behavioural observations

Tadpoles of *L. multiplicata* were always observed in slower-flowing, deeper stretches of streams (Fig. 1a). They were never encountered during sampling in shallow riffles and torrents in the same streams. Tadpoles appeared to spend most time on the rocky substrate, grazing from algae growing on the rocks; they were only occasionally seen swimming in the water column. Following heavy rainfall, tadpoles moved to the more sheltered edges of the stream pools, where they were observed to adhere to rock substrates with their oral discs to avoid being washed away.

DISCUSSION

Knowledge about the larval morphology of most New Guinean frogs with free-swimming tadpoles is lacking. However a number of tadpoles there have evolved highly specialised mouthparts with which they attach to rocks in fast-flowing water (e.g. Tyler, 1963; Menzies & Zweifel, 1974; Günther, 2006), a response to the steep terrain and torrential waterways typical of the island's rugged central cordillera. While considered 'suctorial' (*sensu* Anstis, 2013) because of their ability to adhere to rocks in flowing water, the tadpoles of *L. multiplicata* do not exhibit the extreme morphological adaptations seen in other New Guinean torrent-dwelling treefrogs such as the *L. arfakiana* group (e.g. Menzies & Zweifel, 1974), and the *L. nannotis* group in Australia (Richards, 1992; Anstis, 2013). These species have strongly dorso-ventrally depressed bodies and a mouth modified into a broad, ventrally located suctorial disc. The tadpole of *L. multiplicata* has an oral disc more typical of tadpoles occupying streams with moderate flow levels (Anstis, 2013). The body shape is oval to cylindrical rather than dorsally depressed, and the oral disc is much smaller than that of species occupying torrential habitats. The disc is, however, almost as broad as the snout (Table 2), completely surrounded by numerous oral papillae and is ventrally located, adaptations for clinging to the substrate in flowing water (Anstis, 2013). Indeed the general body form and oral morphology of *L. multiplicata* are typical of a number of congeners from Australia that are known to occupy small streams where flow rates may increase rapidly for short periods of time (e.g. *L. staccato*, *L. coplandi*, *L. personata* and members of the *L. lesueuri* complex; Anstis, 2013). These morphological features reflect the nature of the small, rocky streams where the tadpoles of *L. multiplicata* were found. Observations in the field indicated that these streams flow gently during periods of dry weather but flood rapidly when heavy rain falls, a common occurrence in the mountains of New Guinea.

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