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Larval description of *Leptobrachella murphyi* Chen, Suwannapoom, Wu, Poyarkov, Xu, Pawangkhanant & Che, 2021 (Amphibia: Anura: Megophryidae) from Chiang Mai province, northern Thailand, including genetic and morphological comparisons to other *Leptobrachella* of the region

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Abstract. We provide the first formal description of the tadpole of the megophryid frog, *Leptobrachella murphyi*, based on specimens collected from its type locality on Doi Inthanon, Chiang Mai Province, northern Thailand. The larvae were found in small mountain streams, inhabiting sandy substrates with accumulated leaf litter. Taxonomic confirmation was achieved using a combination of morphological characteristics and molecular sequencing of the mitochondrial 16S rRNA gene fragment. The tadpole of *L. murphyi* possesses the characteristic morphology of the genus, indicative of a semi-fossorial or fossorial lifestyle. The larva can be identified by a distinctive colour pattern on the body and tail, a cylindrical sinistral spiracle, a bevelled vent tube, and a specific tooth row formula of I:2+2/2+2:I. The study results also reveal key diagnostic differences between *L. murphyi* and its sympatric congener, *L. korifi*.

Key words. Asian leaf-litter toad, tadpoles, molecular phylogeny, genetic barcode, morphology, 16S rRNA

INTRODUCTION

Megophryid frogs of the genus *Leptobrachella* are widely distributed from southern China, northeastern India, and Myanmar through Thailand and Vietnam to Malaya, Borneo, and the Natuna islands (Frost, 2025). The genus *Leptobrachella* is understood broadly and in this work as a clade that also includes taxa formerly described as *Leptolalax* (Chen et al., 2018). The tadpoles of the genus *Leptobrachella* exhibit a distinctive morphology. Their bodies are dorsoventrally depressed and elongated, and they possess a cup-like expanded oral disc equipped with upper and lower jaw sheath and, in most species, tooth rows. This morphology is consistent with their fossorial ecology. They inhabit small streams, residing primarily beneath gravel beds or within interstitial spaces among larger rocks (Haas et al., 2006, 2022). However, the cryptic colour pattern and the scarcity of distinct larval morphological characteristics for interspecific diagnosis make the identification of larvae challenging among congeneric species (Ohler et al., 2011). The lack of data on intraspecific variation in larval features aggravates the problem.

Doi Inthanon, Chiang Mai Province, Thailand, is a high-altitude mountain ecosystem and a recognised priority area for biodiversity research, particularly as a crucial headwater region. Herpetological surveys of this area have recently confirmed the presence of two species of the genus *Leptobrachella*: *L. murphyi* (Chen et al., 2021) and *L. korifi* (Matsui et al., 2023). Both species are currently considered endemic to this montane location. Although the adult morphologies of both species were already known, the tadpole of *Leptobrachella korifi* was described only recently by Rongchapho et al. (2025).

To address the existing taxonomic gaps and the confusion surrounding *Leptobrachella* larval identification, the primary objectives of the present study were formulated as follows: (1) to provide the first formal morphological description of the tadpole of *L. murphyi*, and (2) to conduct a detailed comparative analysis of the larval morphology of *L. murphyi* and the sympatric *L. korifi* from their type locality.

MATERIAL AND METHODS

Specimens. All specimens were collected in June 2024 from stream sites in Doi Inthanon National Park in Chiang Mai,

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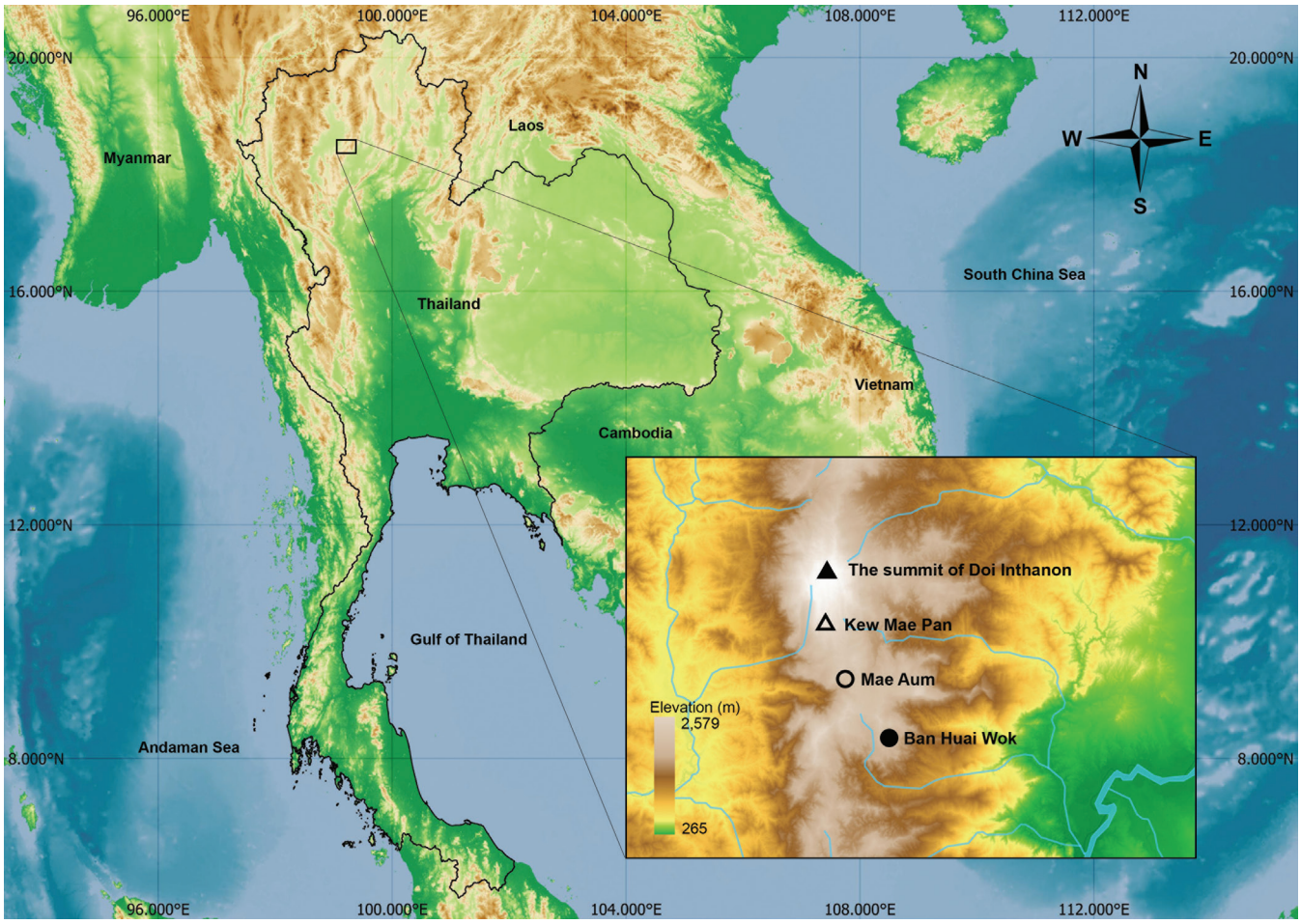

Fig. 1. Localities for specimens used in this study. The solid triangle represents the type locality of *Leptobrachella korii*. The clear triangle represents where *L. korii* were collected in this study. The solid circle represents the type locality of *L. murphyi*, and the clear circle represents where *L. murphyi* were collected in this study.

Thailand (Fig. 1). Three tadpoles of *Leptobrachella murphyi* were collected from Mae Oum (18° 30′ 29.11″ N, 98° 28′ 55.10″ E, 1,500 m a.s.l.), and three adults and two tadpoles of *L. korifi* were collected at the Kew Mae Pan Waterfall (18° 33′ 22.11″ N, 98° 28′ 54.25″ E, 2,200 m a.s.l.). The tadpoles were collected with dip nets by disturbing gravel along the stream margins and in riffles or by scooping accumulated leaf litter into the net. Adult frogs were captured at the same location.

Frogs and tadpoles were euthanised using an aqueous tricaine methanesulfonate (MS-222) solution. Before preservation, tissue samples were taken from the livers of the adult frogs and the tail muscles of the tadpoles and stored in 95% ethanol for molecular analysis. The specimens were fixed in 10% formalin and then transferred to 70% ethanol for preservation and deposited at the Thailand Natural History Museum (THNHM).

Morphological examination. The tadpoles were anesthetised in a separate plastic container with a solution of MS-222 (5 g/L) for approximately 5 min, as described by Heyer et al. (1994). After anesthetisation, the tadpoles were immediately photographed in a clear acrylic box in lateral, ventral, and dorsal views for morphological study. The images were processed to adjust the exposure and clean the background and were subsequently arranged into plates using Adobe Photoshop CS6. The taxonomic nomenclature described by Frost (2025) is used in the following discussion. The terms used to describe tadpoles were adopted from Ohler et al. (2011). Developmental stages were noted according to Gosner (1960). Inger's (1966) tooth row formula is used herein, and the work of Schulze et al. (2015) is referred to for the descriptions of colour patterns.

The tadpoles were photographed with a scale bar or ruler showing millimetre units for reference. The standard tadpole body measurements followed McDiarmid & Altig (1999) and Haas & Das (2011). Measurements were taken from photographs of live specimens using ImageJ software. They are represented by the following acronyms: BL, body length (body length from snout to the point where the axis of the tail myotomes meets the body wall); BW, maximum body width; ED, eye diameter (anterior point to posterior point of eye); ES, eye to snout distance (centre of eye from tip of snout); IND, internarial distance (centre to centre); IOD, interorbital distance (centre to centre); NE, naris to eye (centre of naris to centre of eye); SN, naris to snout distance (centre of naris from tip of snout); TAL, tail length (= TTL–BL); MTH, maximum tail height; and TTL, total length (tip of snout to tip of tail). The morphometric measurements are shown in Supplemental Table S1.

Molecular data and phylogenetic analyses. Total genomic DNA was extracted using the NucleoSpin Tissue Kit, according to the manufacturer's instructions. A fragment of the mitochondrial 16S rRNA gene of genomic regions was amplified and sequenced using specific primers 16S rRNA (Palumbi et al., 2002): the forward primer 16Sar (5′-CGCCTGTTTATCAAAAACAT–3′) and reverse primer 16Sbr (5′–CCGGTCTGAACTCAGATCACGT–3′). Each PCR reaction had a final volume of 50 µl, composed of 25 µl of MasterMix PCR, 2 µl of forward primer (10 pmol/µl), 2 µl of reverse primer (10 pmol/µl), Ultrapure water 16 µl and 5 µl of DNA (20–50 ng/µl). Ratios of ingredients were adjusted when necessary for the optimisation of PCR reactions. PCR amplification conditions followed: an initial denaturation of 95° C for 5 minutes, 35 cycles of denaturation at 94° C for 45 seconds, annealing at 55° C for 45 seconds, and extension at 72° C for 45 seconds, and a final extension step of 72 °C for 10 min.

For phylogenetic tree comparisons, the available sequence for congeners of *Leptobrachella* was downloaded from GenBank (Table 1). Corresponding sequences of *Leptobrachium smithi* (Matsui et al., 1999) and *Leptobrachium sylheticum* (Al-Razi et al., 2021) were used as an outgroup based on previous phylogenetic hypotheses (e.g., Wang et al., 2020; Matsui et al., 2023). All sequences, ranging from 493 to 1,412 bps, were aligned using ClustalW in MEGA version 11 (Tamura et al., 2021) with default parameters and subsequently trimmed to 490 bps for phylogenetic analysis. The best substitution model selected was TIM2+F+G4, using the minimum BIC score. Using a maximum likelihood (ML) analysis, an optimal tree was obtained, and branch supports were evaluated with 1,000 rapid bootstrapping replicates on an IQ-TREE web server (Trifinopoulos et al., 2016). Node values were considered well supported if ML bootstrap supports (BS) were ≥ 80%. The phylogenetic tree was adjusted on FigTree V. 1.4.4 (Rambaut, 2018). Uncorrected pairwise divergences (p-distance) were calculated in MEGA for a dataset containing only sequences of species of *Leptobrachella*.

RESULTS

Molecular results. The phylogenetic analysis was conducted based on 16S rRNA gene sequences, incorporating currently recognised species of the genus *Leptobrachella* (42 sequences representing 18 species) along with eight sequences of *Leptobrachella* from Chiang Mai Province, northern Thailand. All newly acquired sequences were grouped within the same clade as the type series for both species. Three sequences were identified as *L. murphyi*, and five as *L. korifi* (Fig. 2).

The three newly collected tadpoles of *Leptobrachella murphyi* showed an uncorrected p–distance of 0.11–0.46% (Table 2) from the type series KIZ034038, KIZ031200, KIZ034039 and K3427 (GenBank accession numbers: MZ710517, MZ710518, MZ710519, JN848455), which were collected from their type locality, Ban Huai Wok in Doi Inthanon. The five specimens of the sympatric congeners, *Leptobrachella korifi*, were confirmed to be in a clade with the holotype (KUHE 19134, GenBank accession number: LC741033) with insignificant genetic divergence (uncorrected p-distance of 0.00–0.11%) (Table 2).

TAXONOMY

Megophryidae Bonaparte, 1850

Leptobrachella Smith, 1925

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Material examined. Tadpole (THNHM 31137), Gosner's stage 25, Mae Uam Watershed Management Unit, Doi Inthanon, Chiang Mai, Thailand, coll. Danaisawadi P and Pramkasem S, June 2024; Tadpole (THNHM 31140), Gosner's stage 26, same collection data; Tadpoles (THNHM 31139), Gosner's stage 35, same collection data.

Larval morphology (Fig. 3). The following description of the tadpoles of *L. murphyi* in this study is based on three specimens, Gosner's stages 25, 26, and 35 (Total length = 41.71–52.91 mm.) and their photos in life. Body shape elliptical. Snout rounded in dorsal and lateral views. Nares slightly oval, framed with four lobed rims. Nares closer to the snout than to the eyes. Eyes dorsolaterally positioned; iris golden in life. Spiracle sinistral, cylindrical in shape, opening posterodorsally, tip of spiracle free from body wall. Vent tube cylindrical, with dextral opening, bevelled; vent tube entirely attached to lower fin. Tail represents 69.88–72.62% of total length; tail muscle very thick and strong, the highest at half of tail length, tail tip narrowly round, acute. Tail fin translucent; upper tail fin originates at approximately one-fifth of tail length, rises to mid-tail, becoming slightly convex in the middle third of the tail, and then gradually tapers in the posterior third of the tail. Lower tail fin starts low at the trunk-tail junction, increases in height towards middle tail section, reaches its maximum there and then gradually decreases posteriorly to tail tip.

Oral disc cup-shaped, wider than long, positioned and oriented ventrally. Upper labium continuous with deep median notch. Lower labium medially separated incompletely by a deep notch. Marginal papillae are conical in shape and arranged in a single row along the edge of the oral disc. The lateral papillae are round and slightly larger than the upper and lower papillae. Submarginal papilla present on left and right sides of jaw sheaths. Tooth row formula I:2+2/2+2:I, tooth rows located on keratodont ridges, upper uninterrupted tooth row shorter than lower uninterrupted tooth row, interrupted rows subdivided by beak. Jaw sheaths strong with coarsely serrated edge without a middle pointed. Upper sheath arched, covering some of the lower sheath, lower sheath V-shaped.

Table 1. Species and specimens used for molecular phylogenetic analyses for this study (* represents the tadpole specimens)

Species	Locality	Voucher No.	Accession No.	References
Ingroup				
Leptobrachella korifi	Chiang Mai, Thailand	THNHM 31146 *	PX461665	This study
Leptobrachella korifi	Chiang Mai, Thailand	THNHM 31144 *	PX461670	This study
Leptobrachella korifi	Chiang Mai, Thailand	THNHM 31141	PX461661	This study
Leptobrachella korifi	Chiang Mai, Thailand	THNHM 31142	PX461662	This study
Leptobrachella korifi	Chiang Mai, Thailand	THNHM 31143	PX461664	This study
Leptobrachella korifi	Chiang Mai, Thailand	KUHE 19134	LC741033 (Holotype)	Matsui et al., 2023
Leptobrachella murphyi	Chiang Mai, Thailand	THNHM 31137 *	PX461667	This study
Leptobrachella murphyi	Chiang Mai, Thailand	THNHM 31139 *	PX461666	This study
Leptobrachella murphyi	Chiang Mai, Thailand	THNHM 31140 *	PX461669	This study
Leptobrachella murphyi	Chiang Mai, Thailand	KIZ034038	MZ710517 (Paratype)	Chen et al., 2021
Leptobrachella murphyi	Chiang Mai, Thailand	KIZ031200	MZ710518 (Paratype)	Chen et al., 2021
Leptobrachella murphyi	Chiang Mai, Thailand	KIZ034039	MZ710519 (Holotype)	Chen et al., 2021
Leptobrachella murphyi	Chiang Mai, Thailand	K3427 *	JN848455	Chen et al., 2021
Leptobrachella bourreti	Lao Cai, Vietnam	ZMMU-A5636-02280	MH055872	Chen et al., 2018
Leptobrachella bourreti	Lao Cai, Vietnam	AMS R177673	KR018124	Rowley et al., 2015
Leptobrachella baluensis	Sabah, Malaysia	SPM 21604	LC056792	Eto et al., 2015
Leptobrachella dringi	Sarawak, Malaysia	KUHE 55610	AB847553	Matsui et al., 2014
Leptobrachella fuliginosa	Phetchaburi, Thailand	KUHE 20174	LC201987	Matsui et al., 2017
Leptobrachella fuliginosa	Phetchaburi, Thailand	KUHE 20197	LC201988	Matsui et al., 2017
Leptobrachella gracilis	Sarawak, Malaysia	KUHE 55624	AB847560	Matsui et al., 2014
Leptobrachella heteropus	Larut, Malaysia	KUHE 15486	LC202005	Matsui et al., 2017
Leptobrachella heteropus	Larut, Malaysia	KUHE 15490	AB847561	Matsui et al., 2014
Leptobrachella melanoleuca	Surat Thani, Thailand	KUHE 19720	LC201991	Matsui et al., 2017
Leptobrachella melanoleuca	Surat Thani, Thailand	KUHE 19694	LC201989	Matsui et al., 2017
Leptobrachella minima	Chiang Mai, Thailand	KIZ033745	MZ710526	Matsui et al., 2023
Leptobrachella minima	Chiang Mai, Thailand	KIZ031165	MZ710527	Matsui et al., 2023
Leptobrachella nyx	Ha Giang, Vietnam	AMNH A163810	DQ283381	Frost et al., 2006
Leptobrachella parva	Sarawak, Malaysia	KUHE 55308	LC056791	Eto et al., 2015
Leptobrachella pelodytoides	Vinh Phu, Vietnam	MVZ 223641	AY236797	García-París et al., 2003
Leptobrachella pelodytoides	Vinh Phu, Vietnam	MVZ 223642	AY236798	García-París et al., 2003
Leptobrachella petrops	Tuyen Quang, Vietnam	AMS R184826	KY459998	Rowley et al., 2017
Leptobrachella petrops	Tuyen Quang, Vietnam	AMS R184845	KY459999	Rowley et al., 2017
Leptobrachella sinorensis	Mae Hong Son, Thailand	KUHE 19809	LC741034	Matsui et al., 2023
Leptobrachella sinorensis	Mae Hong Son, Thailand	KUHE 19815	LC741035	Matsui et al., 2023
Leptobrachella sinorensis	Mae Hong Son, Thailand	KUHE 19816	LC741036	Matsui et al., 2023
Leptobrachella sola	Narathiwat, Thailand	KUHE 23261	LC202007	Matsui et al., 2017
Leptobrachella sola	Gombak, Malaysia	KUHE 52038	LC202009	Matsui et al., 2017
Leptobrachella ventripunctata	Yunnan, China	CIB 201307-062	LC201978	Matsui et al., 2017
Leptobrachella zhangyapingi	Chiang Mai, Thailand	KIZ07258	MH055864	Chen et al., 2018
Leptobrachella zhangyapingi	Chiang Mai, Thailand	KIZ07460	MH055865	Chen et al., 2018
Outgroups				
Leptobrachium smithi	Trang, Thailand	KUHE 23318	AB719246	Hamidy et al., 2012
Leptobrachium sylheticum	Sylhet, Bangladesh	JnUZool-A0120	MW377689	Al-Razi et al., 2021

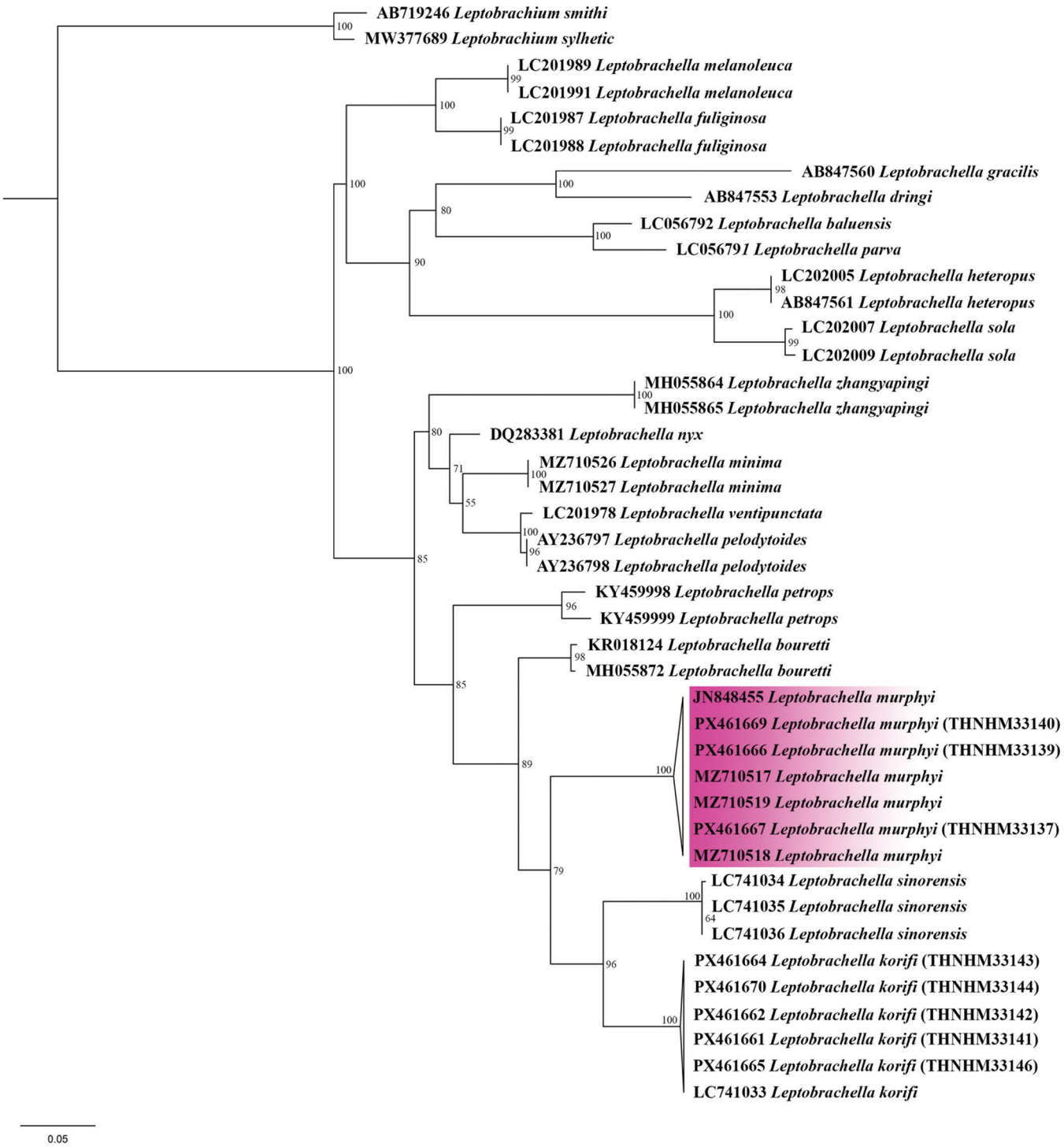

Fig. 2. Phylogenetic trees of *Leptobrachella*, based on mitochondrial 16S rRNA sequences, reveal a genetic match between the tadpoles of *Leptobrachella murphyi* with closely related recognised congeners.

Lateral line present and visible as golden dashes that surround the oral disc, body, and nares. It is positioned ventrolaterally on the body, curving downward to the trunk-tail junction and continues in a straight line along the middle of the muscular portion of the tail. At half the length of the tail, the line curves upward to follow the upper edge of the tail musculature toward the tip of the tail.

Colouration. In life, the body colouration is light brown with light golden blotches and dark spots on the dorsum, partially fusing to form a mottled pattern. In the lateral view, dark spots and light speckles of iridophores appear on the tail. Both start to become denser at approximately half of the tail's length and gradually become even denser at the tip.

In preservation, the body is pale brown, with small dark spots on the dorsum. Dark and light blotches are present at the tail and become denser from the mid-tail to the tail tip. The gut coil is visible. Lateral line visible but indistinct in preserved material.

Table 2. The uncorrected p-distances (percentages) were calculated for the examined 16S rRNA gene sequences of the genus *Leptobrachella*. The final dataset comprised a total of 497 nucleotide positions. Mean values of genetic distance are given in the lower half of the table. (*This study)

	Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1	*L. korifi* (n=5)*	0.00–0.11																	
2	*L. korifi* (n=1) [holotype]	0.00–0.11	0																
3	*L. murphyi* (n=3)*	5.52–6.30	5.66–6.30	0.00–0.11															
4	*L. murphyi* (n=4) [type series]	5.92–6.30	6.16–6.30	0.11–0.46	0.00–0.34														
5	*L. sinorensis* (n=3)	4.88–5.41	5.25–5.41	5.31–6.00	5.83–6.14	0.00–0.11													
6	*L. bourreti* (n=2)	6.35–6.77	6.63–6.77	5.29–5.95	5.62–5.81	5.88–6.10	0.00–0.34												
7	*L. petrops* (n=2)	6.69–7.94	6.95–7.76	7.60–8.75	8.12–8.75	7.79–8.71	8.10–8.27	0.00–1.53											
8	*L. zhangyapingi* (n=2)	10.03–10.26	10.24	9.23–9.89	9.59–9.74	8.18–8.35	7.27–7.32	9.21–9.30	0										
9	*L. nyx* (n=1)	8.73–9.13	9.13	8.23–8.77	8.63–8.77	8.34–8.52	6.02–6.17	7.54–7.62	5.73	0									
10	*L. ventripunctata* (n=1)	9.48–9.83	9.73–9.83	9.01–9.76	9.24–9.60	9.17–9.48	7.06–7.42	8.23–8.99	6.41–7.08	3.33–3.50	0.00–0.35								
11	*L. pelodytoides* (n=2)	9.76–10.02	10.01	9.14–9.65	9.21–9.50	9.44–9.62	7.06–7.20	8.70–9.11	6.58	3.32	0.35–0.69	0							
12	*L. minima* (n=2)	9.97–10.52	10.52	7.59–8.41	8.12–8.41	8.00–8.18	6.63–6.82	8.08–8.50	6.57	3.47	4.01–4.16	4.11	0						
13	*L. melanoleucus* (n=2)	11.56–11.97	11.94–11.97	10.52–10.91	10.89–11.13	10.00–10.21	8.97–9.31	9.63–10.59	9.15–9.17	8.13–8.15	10.25–10.57	10.58–10.61	8.90–8.93	0					
14	*L. fuliginosa* (n=2)	11.72–12.00	12.00	10.06–10.65	10.65–10.87	11.20–11.39	9.59–9.74	9.53–9.86	8.1	7.56	9.28–9.53	9.25	8.37	3.39–3.40	0				
15	*L. gracilis* (n=1)	14.03–14.38	14.16	11.83–11.97	11.97–12.50	14.09–14.29	13.10–13.14	13.76–13.83	13.9	13.11	13.52–13.60	13.53	13.45	12.44–12.47	11.58	0			

	Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
16	*L. dringi* (n=1)	10.96–11.44	11.23	11.53–11.93	11.61–11.77	12.05–12.24	11.06–11.09	12.01–12.07	12.36	10.24	10.74–10.87	10.68	11.28	10.72–10.274	10.6	7.42	0		
17	*L. heteropus* (n=2)	14.36–14.57	14.57	13.02–13.50	13.34–13.88	13.92–14.11	14.02	14.24–14.77	13.6	11.68	12.77–13.00	13.04	11.28	12.88–12.91	11.64	14.18	13.22	0	
18	*L. sola* (n=2)	14.14–15.29	14.71–15.29	13.26–14.51	13.94–14.58	13.78–14.54	14.04–14.69	14.65–15.92	14.00–14.26	13.05–13.23	13.71–14.39	13.8.–14.22	12.90–13.31	13.52–13.74	11.68–11.71	14.17–14.44	13.38–13.80	3.98–4.13	0.58

Variation. Tadpoles of *Leptobrachella murphyi* at different stages do not differ in their external features such as body shape or the structure of oral discs and tooth row formula. However, the larva at later stage (THNHM33139, stage 35) has a longer body and more extensive pigmentation on the body and tail, with more diffuse dark markings.

Leptobrachella korifi Matsui, Panha & Eto, 2023

Material examined. Tadpole (THNHM 31146), Gosner's stage 25, Kew Mae Pan, Doi Inthanon, Chiang Mai, Thailand, coll. Danaisawadi P and Pramkasem S, June 2024; Tadpole (THNHM 31144), Gosner's stage 27, same collection data. Two adult females (THNHM 31141, 31143), same collection data. One adult male (THNHM 31142), same collection data.

Larval morphology. (Fig. 4) The following description of *L. korifi* larvae is based on two specimens at Gosner's stages 25 and 27. Total length ranges are 49.72–51.18 mm: Body elliptical in dorsal view; slightly depressed in lateral view. Snout broadly rounded, and blunt in the dorsal view. Nares slightly oval, framed with a four-lobed rim, located almost dorsally, closer snout than eyes. Eyes dorsolaterally located; iris golden in life. Spiracle sinistral, conical, directed posterodorsally, tip of spiracle attached to the body wall. Vent tube short, cylindrical, with dextral opening, not bevelled and entirely attached to lower fin. Tail represents 69.35–72.31% of total length. Tail muscle well developed, reaching tail tip; tail tip moderately round, relatively blunt. Upper tail fin starts at one-sixth of tail length; in a slightly convex line the upper fin reaches maximum height at approximately one-third of tail length then gradually tapers in straight line to tail tip. Lower tail fin originating ventrally at trunk-tail junction with moderate height, reaching maximum height at the middle third of tail and then gradually tapering and curving towards tail tip.

Oral disc cup-like (Fig. 4F), wider than long, protruding ventrally, upper labium with shallow medial notch; lower labium incompletely divided by deep median notch. Uniserial row of marginal papillae along the edge or oral disc. Marginal papillae conical in shape, upper and lower papillae similar in size. Lateral papillae conical and pointed. Five submarginal papillae present on left and right sides of jaw sheaths. Tooth row formula: I:4+4/2+2:I in stage 25 and I:3+3/2+2:I in stage 27. Each of the tooth rows is carried by ridges. Uninterrupted tooth row on upper lip shorter than lower lip. Jaw sheaths black, strong, with coarsely serrated edges; upper sheath arched, median serrations slightly larger than lateral serrations. Upper sheath cover some of the lower sheath in resting position of the jaws; lower sheath V-shaped.

The lateral line system visible as a line of spots instead of dashes (see Fig. 4A, B). It surrounds the oral disc, body, and nares. The line curves downward to the junction of the trunk and tail, continuing at the mid-level of the tail muscle for one-third of the tail length (Fig. 4A).

Colouration. In life, the overall dorsal colouration on the head and trunk is a marbling of dark brown reticulation and

Fig. 3. The external morphology of *Leptobrachella murphyi* tadpole (THNHM 31137) at Gosner's stage 25 with A–C, colour markings on lateral view, dorsal view and ventral view; D, cylindical spiracle; E, beveled vent tube; and F, mouthpart structure.

golden blotches. In the lateral view, golden speckles are present on the tail fins, and there are small dark blotches and dark spots on the tail, with the pattern gradually becoming denser towards the tail tip in the posterior two-thirds of the tail length.

In preservation, the skin is pale brown with dark spots dorsally on the head and trunk, golden pattern absent and dark spots and a dark small blotch on the tail present. The gut coil is visible. A lateral line is visible but indistinct in preserved material.

Variation. The tadpole of *Leptobrachella korifi* in our study did not differ in body shape, except for the tooth row formula, as described above. For THNHM31144 (stage 27), the spiracle is slightly curved distally but remains essentially conical in shape. Moreover, the thickness of rectilinear markings and the size of golden blotches on the body vary between specimens.

In comparison to those examined by Rongchapho et al. (2025), the tadpoles of this species examined in this study exhibited no noticeable differences in most external morphological characteristics and in the tooth row formula, except for the spiracle shape and vent tube opening.

Habitat and ecological notes. All specimens of *Leptobrachella korifi* and *L. murphyi* were collected on

Fig. 4. The external morphology of *Leptobrachella korifi* tadpole (THNHM 31146) at Gosner's stage 25 with A–C, colour markings on lateral view, dorsal view and ventral view; D, conical spiracle; E, vent tube; and F, mouthpart structure.

a rainy day in June 2024. Tadpoles of *L. murphyi* were collected from a well-protected stream in a hilly evergreen forest approximately 1,500 m above sea level at the Mae Uam Watershed Management Unit. The tadpoles were collected from steep, fast-flowing streams 2–3 m wide and 0.2–0.3 m deep. The streambeds consisted mainly of gravel, sand, and rocky substrates, with no cascades. The tadpoles were found beneath small rocks on the banks of the streams and were recorded in syntopy with the tadpoles of *Megophrys* sp. and *Leptobrachium smithi*.

Specimens of both mature adult frogs and tadpoles of *Leptobrachella korifi* were collected at the Kew Mae Pan natural trail from rocky streams with cascades at an elevation of approximately 2,200 m above sea level. The tadpoles were discovered beneath rocks along the banks of the streams, syntopically with tadpoles of *Megophrys* sp., *Leptobrachium smithi,* and *Leptobrachium huachen*. The streams were fast-flowing, approximately 5 m wide and approximately 0.5 m deep. One adult female of *L. korifi* was found hiding under leaf litter at the same location.

DISCUSSION

Doi Inthanon National Park is the type locality for two congeneric species, *Leptobrachella korifi* and *L. murphyi*, whose tadpoles are sympatric, inhabiting different stream

Table 3. Characters of *Leptobrachella murphyi* and *L. korifi* at Stage 25 compared to *L. korifi* from previous study.

Compared characters	***L. murphyi* (THNHM 31137)**	***L. korifi* (THNHM 31146)**	***L. korifi***
	This study	This study	Rongchapho et al., 2025
Stage	25	25	26–35
Total length	52.91	49.76	21.5–48.7
BL/BW	2.17	2.06	1.75–2.29
TL/BL	2.65	2.26	1.36–3.13
Body shape in dorsal view	elliptical	elliptical	elliptical
Spiracle shape	Cylindrical	Conical	Flattened tubular
Colour pattern on the body	Dark brown and small golden spots on dorsal view	Dark brown reticulate with large golden blotches on dorsal view	Body brown with creamy spot. Chest grey
Colour pattern on the tail	Dark brown and dense golden spot dense at ⅔ of the tail length	Dark brown and dense golden spot dense at ⅔ of the tail length	Tail musculature grey with creamy spot
Lateral line visible	visible	visible	visible
Gut visible in life	visible	visible	visible
Gut visible in preservation	visible	visible	visible
Vent tube shape	Cylindrical, opening on right side, bevel, entirely attached to lower fin	Cylindrical, opening on right side, not bevel, entirely attached to lower fin	Large opening approximately conical, medial opening, entirely attached to lower fin
Tail tip	Narrowly round	Moderately round	Acute
Tooth row formula	I:2+2/2+2:I	I:4+4/2+2:I (stage 25) I:3+3/2+2:I (stage 27)	I:3+3/2+2:I (all are the same from stage 26–34)

branches that originate from the same primary watershed on Doi Inthanon. Tadpoles within the genus *Leptobrachella* typically require similar lotic habitats, characterised by fast-flowing water and gravel substrates, where they often shelter beneath the gravel layer (Inger, 1985; Haas et al., 2006). Given these shared ecological niches and their close phylogenetic relationships (Fig. 2), the external morphology of the two species is highly similar (e.g., the semi-fossorial body shape). While several characteristics (Table 3) permit differentiation in the field, the shape of the spiracle emerges as a robust distinguishing character: the tip of the spiracle in *L. murphyi* is cylindrical, while the spiracle is conical in *L. korifi* (Figs. 3D and 4D). The *L. murphyi* tadpole differs from that of *L. korifi* in several aspects of the oral disc. In *L. murphyi*, the marginal papillae gradually increase in size from the upper to the lower labium, whereas in *L. korifi,* they are of similar size across the entire oral disc. The tooth row formula in *L. murphyi* (I:2+2/2+2:I) comprises fewer rows than observed in *L. korifi* (I:4+4/2+2:I or I:3+3/2+2:I). In addition, the jaw sheaths of *L. murphyi* bear serrations that are approximately uniform in size, while in *L. korifi*, the jaw sheaths are coarsely serrated, with the central portion of the upper sheath deeper than the lateral serrations.

The morphological features of the *L. korifi* tadpoles examined in the present study were not fully consistent with the recent description by Rongchapho et al. (2025). The differences concerned body and tail proportions, spiracle morphology, tail tip shape, and the tooth row formula (Table 3). These observed morphological differences are due to different developmental stages examined in two studies, as ontogenetic larval growth and development are known to affect larval body proportions and features (Grosjean, 2005). However, according to raw data from Rongchapho et al. (2025), the low ratio of TL/BL reported differs from this study because damage occurred in the tails of three specimens, rather than being a natural trait. In terms of vent tube opening, while Rongchapho et al. (2025) described the vent tube of *L. korifi* as having a medial opening based on Stage 34 preserved specimens, our observations show a dextral opening. We suggest that this disagreement may arise from tissue distortion during preservation in the former study, as the vent tube is highly susceptible to collapse. Further studies with larger series of both live and preserved specimens are needed to clarify the extent of morphological variation in this trait.

In this study, the identification of *Leptobrachella* tadpoles was based exclusively on the external morphology and avoided colour characteristics. In field situations in daylight, all observed *Leptobrachella* tadpoles exhibited uniformly grey or brown colours, showing only minor differences in shading. However, subtle differences in tadpole colouration became apparent under controlled illumination, particularly flash photography.

We compared the morphological characteristics of fifteen candidate *Leptobrachella* tadpole species distributed across Northern Thailand and adjacent regions, including: *L. aerea* (Ohler et al., 2011), *L. bashaensis* (Lyu et al., 2020), *L. bidoupensis* (Vassilieva, 2021), *L. botsfordi* (Nguyen et al., 2020), *L. bourreti* (Ohler et al., 2011), *L. kalonensis* (Le et al., 2023), *L. mangshanensis* (Hou et al., 2018), *L. melanoleuca* (Rongchapho et al., 2024), *L. minima* (Ohler et al., 2011), *L. oshanensis* (Shi et al., 2021), *L. pelodytoides* (Smith, 1917; Kaewtongkum et al., 2014), *L. petrops* (Le et al., 2021), *L. rowleyae* (Nguyen et al., 2018), *L. ventripunctata* (Ohler et al., 2011), and *L. yeae* (Shi et al., 2021). See Supplementary table S2 for further details.

The tadpole of *Leptobrachella murphyi* differs from other known Northern Thailand *Leptobrachella* tadpoles in having a tail length approximately 72.6% of body length (vs. a tail length not exceeding 66% of the body length in *L. botsfordi*, *L. mangshanensis*, and *L. pelodytoides*), fine dark dots and golden speckles on the dorsum (vs. the absence of such a pattern in *L. botsfordi* and *L. bashaensis*, the presence of only golden speckles in *L. petrops*, and large golden blotches or dark reticulation in *L. korifi* and *L. melanoleuca*), dark dots and spots on the tail (vs. absence of such a pattern in *L. bidoupensis*, *L. botsfordi*, *L. bourreti*, *L. kalonensis*, *L. melanoleuca*, *L. oshanensis*, *L. rowleyae*, and *L. yeae*), an elliptical body shape in the dorsal view (vs. a slender elongated body shape in *L. botsfordi* and *L. bashaensis*), the cylindrical shape of the spiracle (vs. a conical shape in *L. bidoupensis*, *L. botsfordi*, *L. kalonensis*, *L. korifi*, and *L. rowleyae*), the cylindrical shape of the vent tube (vs. a conical shape in *L. bashaensis*, *L. melanoleuca*, *L. minima*, and *L. pelodytoides*) and an opening on the right (dextral) side (vs. a medial opening in *L. melanoleuca* and an opening on the left side in *L. botsfordi*).

In terms of the structure of the mouth parts, the oral discs of *Leptobrachella* tadpoles are usually oriented ventrally. The mouth parts are composed of the upper and lower lips and jaw sheath and tooth rows. The formula for the tooth rows varies significantly among species. However, *Leptobrachella murphyi* can be separated from other species by: presence of lower tooth rows (vs. absent in *L. bidoupensis* and *L. melanoleuca*); a single uninterrupted lower tooth row (vs. double uninterrupted lower tooth rows in *L. bashaensis* and *L. mangshanensis*); upper jaw sheath serrated without a pointed middle (vs. pointed middle section present in *L. korifi*, *L. aerea*, *L. bourreti* and *L. ventripunctata*).

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