

Taxonomic reassessment of the poorly known microhylid, *Kalophrynus menglienicus* Yang & Su, 1980

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Abstract

The taxonomic status of *Kalophrynus menglienicus* Yang & Su, 1980 was evaluated based on newly collected topotype specimens. Phylogenetic analysis showed that this species should be assigned to the genus *Micryletta* Dubois, 1987. In addition, morphological diagnosis and descriptions based on the newly collected topotype specimens were provided.

Key Words

16S rRNA, Menglian County, Paddy Frog, taxonomy

Introduction

Kalophrynus menglienicus, a species of Microhylidae, was described by Yang and Su (1980) from Menglian County, Puer City, southwestern Yunnan Province, China, based on morphology. This species was previously known only from its type locality, and the taxonomic status of this species has not been well resolved due to the lack of molecular data.

Fei (2020) allocated *Kalophrynus menglienicus* to the genus *Micryletta*, but did not provide any explanations.

During our fieldwork in southern Yunnan Province, China, in 2021, five specimens of *Kalophrynus menglienicus* were collected from its type locality. The results of morphological comparison and molecular analysis showed that these specimens belong to the genus *Micryletta*. After the examination of the type specimens of *K. menglienicus*, we reassessed the taxonomic status of *K. menglienicus*.

Materials and methods

Specimens were collected by hand at night. Photographs were taken to document the color pattern in life prior to euthanasia. Liver tissues were stored in 99% ethanol. Specimens were fixed and preserved in 75% ethanol and deposited at Kunming Natural History Museum of Zoology, Kunming Institute of Zoology, Chinese Academy of Sciences (**KIZ**).

Measurements were taken to the nearest 0.1 mm with digital calipers. The methodology of measurements followed Liu et al. (2021). **SVL**: snout–vent length, measured from the tip of the snout to cloaca; **HL**: head length, measured from the tip of snout to hind border of jaw angle; **SL**: snout length, measured from the anterior corner of eye to the tip of snout; **EL**: eye length, measured as the distance between anterior and posterior corners of the eye; **NEL**: nostril–eye length, measured as the distance between the anterior corner of the eye and the nostril center; **HW**: head width, measured

as the maximum width of head on the level of mouth angles in ventral view; **IND**: internarial distance, measured as the distance between the central points of nostrils; **IOD**: interorbital distance, measured as the shortest distance between the medial edges of eyeballs in dorsal view; **UEW**: upper eyelid width, measured as the maximum distance between the medial edge of eyeball and the lateral edge of upper eyelid; **TMP**: Tympanum length, measured as the horizontal tympanum diameter; **FLL**: forelimb length, measured as the length of straightened forelimb to the tip of third finger; **LAL**: lower arm and hand length, measured as the distance between elbow and the tip of third finger; **HAL**: hand length, measured as the distance between the proximal end of outer palmar (metacarpal) tubercle and the tip of third finger; **1FL**: first finger length, measured as the distance between the tip and the distal end of inner palmar tubercle; **IPTL**: inner palmar tubercle length, measured as the maximum distance between proximal and distal ends of inner palmar tubercle; **MPTL**: median palmar tubercle length, measured as the maximum diameter of median palmar tubercle; **OPTL**: outer palmar tubercle length, measured as the maximum diameter of outer palmar tubercle; **3FDD**: third finger disk diameter; **HLL**: hindlimb length, measured as the length of straightened hindlimb from groin to the tip of fourth toe; **TL**: tibia length, measured as the distance between the knee

and tibiotarsal articulation; **FL**: foot length, measured as the distance between the base of the inner metatarsal tubercle to the tip of the fourth toe; **IMTL**: inner metatarsal tubercle length, measured as the maximum length of inner metatarsal tubercle; **1TOEL**: first toe length, measured as the distance between the distal end of inner metatarsal tubercle and the tip of first toe; **4TDD**: fourth toe disk diameter. We compared the newly collected specimens with the type series of *Kalophrynus menglienicus* deposited at KIZ.

Total genomic DNA was extracted from liver tissues using the standard phenol-chloroform method (Hillis et al. 1996; Sambrook and Russell 2001). A fragment of the mitochondrial 16S rRNA gene was amplified and sequenced. The primers L2188 (Matsui et al. 2006): 5'-AAAGTGG-GCCTAAAGCAGCCA-3' and 16H1 (Hedges 1994): 5'-CTCCGGTCTGAACTCAGATCAGTAGG-3' were used in amplification and cycle sequencing. The experiment protocols used in this study are the same as Liu et al. (2021). Purified PCR products were sequenced by Davis Sequencing using BigDye terminator 3.1 and sequences were edited and manually managed using SeqMan in Lasergene 7.1 (DNASTAR Inc., Madison, WI, USA) and MEGA X (Kumar et al. 2018). All new sequences have been deposited on GenBank, other sequences used in this study were downloaded from Genbank (Table 1).

Table 1. Sequences used in molecular analyses of this study.

Species	Voucher	Locality	Accession No.
<i>Micryletta aishani</i>	SDBDU 3920	India: Assam, Cachar district, Subhong	MK889218
<i>Micryletta dissimulans</i>	AUP01690	Thailand: Songkla Prov., Saba Yoi district	MT573414
<i>Micryletta dissimulans</i>	AUP01691	Thailand: Songkla Prov., Saba Yoi district	MT573415
<i>Micryletta dissimulans</i>	AUP01696	Thailand: Songkla Prov., Saba Yoi district	MT573416
<i>Micryletta dissimulans</i>	AUP01698	Thailand: Songkla Prov., Saba Yoi district	MT573413
<i>Micryletta erythropoda</i>	ZMMU A4721-1533	Vietnam: Dong Nai, Ma Da (Vinh Cuu) N.R.	MH756146
<i>Micryletta erythropoda</i>	ZMMU A4721-1542	Vietnam: Dong Nai, Ma Da (Vinh Cuu) N.R.	MH756147
<i>Micryletta hekouensis</i>	KIZ20210510	China: Honghe, Hekou	MZ536627
<i>Micryletta hekouensis</i>	KIZ20210511	China: Honghe, Hekou	MZ536628
<i>Micryletta immaculate</i>	KFBG 14270	China: Hainan, Exian	MW376736
<i>Micryletta immaculate</i>	KFBG 14271	China: Hainan, Exian	MW376737
<i>Micryletta inornata</i>	MZB Amph 23949	Indonesia: Sumatra, Deli Serdang	LC208135
<i>Micryletta inornata</i>	MZB Amph 23947	Indonesia: Sumatra, Deli Serdang	LC208136
<i>Micryletta inornata</i>	MZB Amph 23948	Indonesia: Sumatra, Deli Serdang	LC208137
<i>Micryletta inornata</i>	MZB Amph 27242	Indonesia: Sumatra, Aceh	LC208138
<i>Micryletta inornata</i>	USNM 587625	Myanmar: Tanintharyi	MT609033
<i>Micryletta inornata</i>	USNM 587901	Myanmar: Tanintharyi	MT609034
<i>Micryletta lineata</i>	KUHE 23858	Thailand: Ranong	AB634695
<i>Micryletta lineata</i>	CAS 247206	Myanmar: Tanintharyi Div., Kawthaung dist.	KM509167
<i>Micryletta nigromaculata</i>	ZMMU A5947	Vietnam: Hai Phong, Cat Ba N.P.	MH756148
<i>Micryletta nigromaculata</i>	ZMMU A5937	Vietnam: Hai Phong, Cat Ba N.P.	MH756149
<i>Micryletta nigromaculata</i>	ZMMU A5946	Vietnam: Hai Phong, Cat Ba N.P.	MH756151
<i>Micryletta nigromaculata</i>	DTU 301	Vietnam: Ninh Binh, Cuc Phuong N.P.	MH756154
<i>Micryletta steinegeri</i>	KUHE 35937	China: Taiwan, Yunlin	AB634696
<i>Micryletta steinegeri</i>	ZMMU A5336-1	China: Taiwan, aohsiung	MW376732
<i>Micryletta steinegeri</i>	ZMMU A5336-2	China: Taiwan, Kaohsiung	MW376733
<i>Micryletta steinegeri</i>	ZMMU A5336-3	China: Taiwan, Kaohsiung	MW376734
<i>Micryletta sumatrana</i>	/	Indonesia: Sumatra Selatan	MN727065
<i>Micryletta menglienicus</i>	KIZ20210708	China: Menglian, Jingmao	OK335183
<i>Micryletta menglienicus</i>	KIZ20210709	China: Menglian, Jingmao	OK335184
<i>Micryletta menglienicus</i>	KIZ20210710	China: Menglian, Jingmao	OK335185
<i>Micryletta menglienicus</i>	KIZ20210711	China: Menglian, Jingmao	OK335186
<i>Micryletta menglienicus</i>	KIZ20210712	China: Menglian, Jingmao	OK335187
<i>Glyphoglossus yunnanensis</i>	2015000386	China: Yunnan, Kunming	MN860400
<i>Kalophrynus interlineatus</i>	KUHE 33787	Myanmar: Chatthin	AB634698
<i>Kaloula pulchra</i>	KUHE 35171	Thailand: Kanchanaburi	AB201194
<i>Microhyla fissipes</i>	KUHE 32943	China: Anhui, Huangshan	AB201185
<i>Uperodon systoma</i>	SDBDU 2005.4723	India: Tamil Nadu, Kunnappattu	MG557949

Sequences were aligned using ClustalW (Thompson et al. 1994) integrated in MEGA X (Kumar et al. 2018) with default parameters. Genetic divergences (uncorrected p-distance) were calculated in MEGA X (Kumar et al. 2018). The best substitution model GTR+F+I+G4 was selected using the Akaike Information Criterion (AIC) in ModelFinder (Kalyaanamoorthy et al. 2017). Maximum Likelihood analysis was performed in RaxmlGUI 2.0 (Silvestro and Michalak 2012) and nodal support values were estimated by 1,000 rapid bootstrap replicates. Bayesian Inference was performed in MrBayes 3.2.7 (Ronquist et al. 2012) based on the selected substitution model. Two runs were performed simultaneously with four Markov chains starting from a random tree. The chains were run for 1,000,000 generations and sampled every 100 generations. The first 25% of the sampled trees was discarded as burn-in after the standard deviation of split frequencies of the two runs was less than a value of 0.01, and then the remaining trees were used to create a 50% majority-rule consensus tree and to estimate Bayesian posterior probabilities.

Results

Phylogenetic analysis showed that all the newly collected specimens were homogeneous and nested in the genus *Micryletta* but not *Kalophrynus* (Fig. 1). This indicates that these specimens should be assigned to the genus *Micryletta*. The specimens from the type locality of *K. menglienicus* formed a distinct clade sister to *M. immaculata* with strong support. The genetic divergences between the newly collected specimens and other species of *Micryletta* ranged from 3.3% (with *M. immaculata* and *M. steinegeri*) to 7.7% (with *M. nigromaculata*) (Table 2).

In order to confirm that our new collections are conspecific with *Kalophrynus menglienicus*, we compared our new collections with the type series of *K. menglienicus* and the original description by Yang and Su (1980). Morphological comparisons between the newly collected specimens and the type series of *K. menglienicus* are pre-

sented in Table 3. There is no significant morphological difference between our collections from the type locality of *K. menglienicus* and the type series of *K. menglienicus*. For coloration, some of the new collections agree with the original description by Yang and Su (1980) while others show more variations (Fig. 4), as all the new collections are homogeneous, so we consider these variations are intraspecific. Hence, we conclude that our new collections from the type locality of *K. menglienicus* are conspecific with *K. menglienicus*.

According to the original description (Yang and Su 1980) of *Kalophrynus menglienicus*: body size small; forelimbs thin and slender; tympanum indistinct; sub-articular tubercles on fingers and toes present; supernumerary tubercles on palm present; three metacarpal tubercles; finger I shorter than finger II; foot longer than tibia; webbing between toes absent; no outer metatarsal tubercle. These characteristics tally with the diagnosis (Dubois 1987; Fei et al. 2009; Yang and Poyarkov 2021) of *Micryletta* but do not tally with the diagnosis (Fei et al. 2009; Matsui et al. 2017) of *Kalophrynus*.

In conclusion, we agree with Fei (2020) to transfer *Kalophrynus menglienicus* to the genus *Micryletta*. However, as the Latin generic name *Micryletta* is in a feminine gender, the specific epithet *menglienicus* needs its gender changed to feminine, so the new combination should be *Micryletta menglienica*. We propose “Menglian Paddy Frog” for the common English name and “孟连小姬蛙” (Mèng Lián Xiǎo Jī Wā) for the common Chinese name of this species.

Taxonomic account

Micryletta menglienica (Yang & Su, 1980)

Figures 2–4

Type material. *Holotype*. KIZ 751377, adult male. *Paratypes*. KIZ 751333–751339, KIZ 751371–751376, KIZ 751378–751385, KIZ 751387–751389, KIZ 751409–751416, 32 adult males.

Table 2. Uncorrected p-distances (%) of 16S rRNA sequences among *Micryletta* species and outgroups.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1 <i>Micryletta menglienica</i>															
2 <i>Micryletta aishani</i>	3.5														
3 <i>Micryletta dissimulans</i>	5.4	4.4													
4 <i>Micryletta erythropoda</i>	6.7	4.7	7.4												
5 <i>Micryletta hekouensis</i>	3.8	3.5	5.0	6.4											
6 <i>Micryletta immaculata</i>	3.3	4.5	6.4	7.2	4.6										
7 <i>Micryletta inornata</i>	6.7	5.1	6.1	7.7	5.7	7.2									
8 <i>Micryletta lineata</i>	5.0	3.2	6.0	2.9	4.6	6.6	6.5								
9 <i>Micryletta nigromaculata</i>	7.7	4.7	5.2	8.3	8.0	8.0	6.7	7.1							
10 <i>Micryletta steinegeri</i>	3.3	3.5	4.8	6.6	3.1	4.2	5.6	5.1	7.1						
11 <i>Micryletta sumatrana</i>	6.7	5.9	5.1	9.1	6.7	8.3	8.2	7.4	5.5	6.0					
12 <i>Glyphoglossus yunnanensis</i>	14.8	10.0	9.3	15.3	14.5	21.2	10.4	17.3	12.7	14.1	10.7				
13 <i>Kalophrynus interlineatus</i>	17.4	13.5	14.1	18.3	17.4	17.4	12.9	17.1	15.6	16.7	15.7	17.6			
14 <i>Kaloula pulchra</i>	14.1	10.4	9.4	13.5	13.8	14.6	10.2	12.9	12.5	13.9	11.9	15.1	18.5		
15 <i>Microhyla fissipes</i>	16.1	10.1	10.4	17.0	16.4	16.7	11.8	16.1	14.2	15.8	12.6	15.1	18.4	16.2	
16 <i>Uperodon systoma</i>	10.3	10.1	10.7	12.5	10.1	9.9	13.2	10.3	10.3	9.8	11.3	11.7	15.8	8.1	11.4

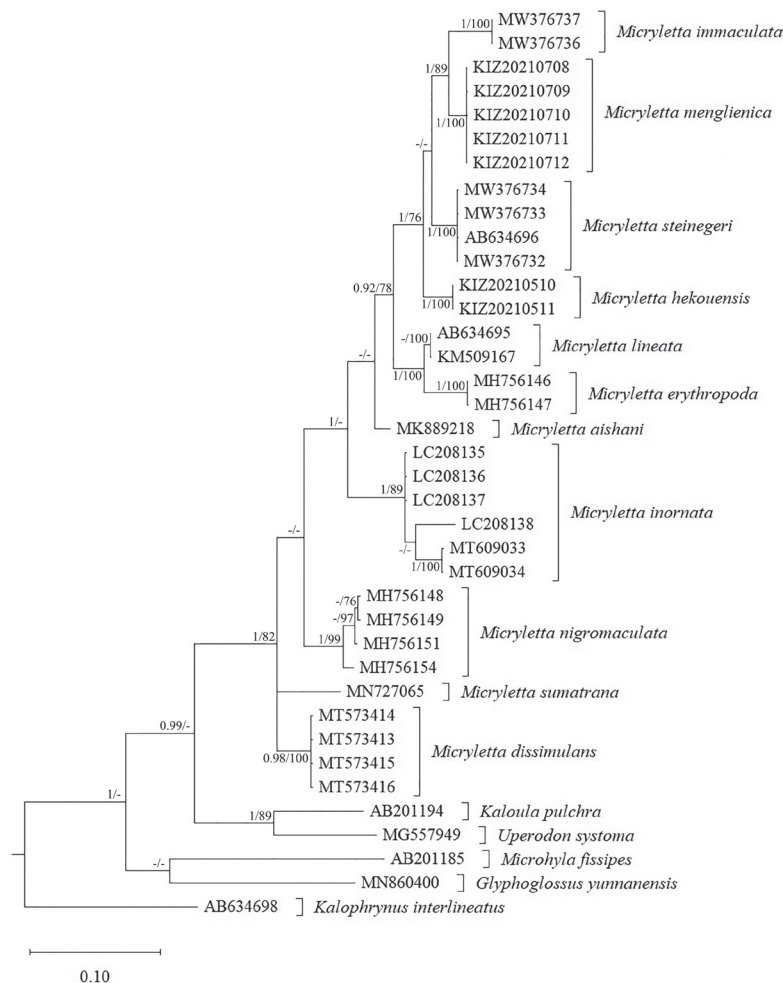


Figure 1. Bayesian Inference tree of *Micryletta* reconstructed on the base of 16S rRNA gene sequences. Values before slashes correspond to Bayesian posterior probabilities (>0.9 remain), and values after slashes correspond to Maximum Likelihood bootstrap replicates (>70 remain).

Table 3. Comparisons between the type specimens of *Micryletta menglienicus* and the newly collected specimens.

	Holotype KIZ 751377	Paratypes n = 32	Topotype KIZ20210708	Topotype KIZ20210709	Topotype KIZ20210710	Topotype KIZ20210711	Topotype KIZ20210712
Sex	Male	Males	Male	Male	Male	Male	Male
SVL	19.6	18.0–21.9	19.4	18.7	21.8	21.7	20.1
HL	6.5	5.3–7.1	6.7	6.2	7.1	7.1	6.9
SL	2.7	2.2–3.1	3.0	2.7	3.1	2.9	2.9
EL	2.4	1.7–2.4	2.2	2.1	2.3	2.4	2.4
NEL	1.6	1.2–1.9	1.7	1.6	1.7	1.7	1.7
HW	6.5	5.8–7.3	6.7	6.2	7.0	7.0	6.9
IND	2.1	1.5–2.2	1.8	1.7	2.1	2.2	2.2
IOD	2.4	2.1–2.7	2.6	2.4	2.7	2.7	2.5
UEW	1.4	1.0–1.6	1.2	1.3	1.6	1.5	1.6
TMP	1.2	0.7–1.5	1.1	1.2	1.5	1.4	1.2
FLL	13.5	12.0–15.4	14.0	13.2	15.4	15.5	14.4
LAL	9.6	9.1–10.5	9.8	9.6	10.8	11.0	10.4
HAL	5.6	4.7–5.8	5.3	5.3	6.1	6.1	5.4
IFL	2.3	1.9–2.5	2.4	2.3	2.6	2.4	2.2
IPTL	0.4	0.3–0.6	0.4	0.4	0.4	0.5	0.3
MPTL	0.6	0.5–0.8	0.5	0.5	0.6	0.7	0.5
OPTL	0.8	0.6–0.8	0.7	0.6	0.8	0.8	0.7
3FDD	0.5	0.4–0.5	0.4	0.4	0.5	0.5	0.5
HLL	28.6	26.5–32.6	28.5	27.4	33.3	33.1	29.8
TL	9.0	8.2–9.7	8.9	8.3	10.5	10.5	9.3
FL	9.5	8.6–10.5	9.6	9.4	10.6	10.9	10.0
IMTL	0.7	0.4–0.7	0.6	0.4	0.7	0.5	0.5
1TOEL	2.3	1.8–2.6	2.3	2.2	2.7	2.6	2.5
4TDD	0.6	0.4–0.6	0.4	0.4	0.5	0.5	0.6

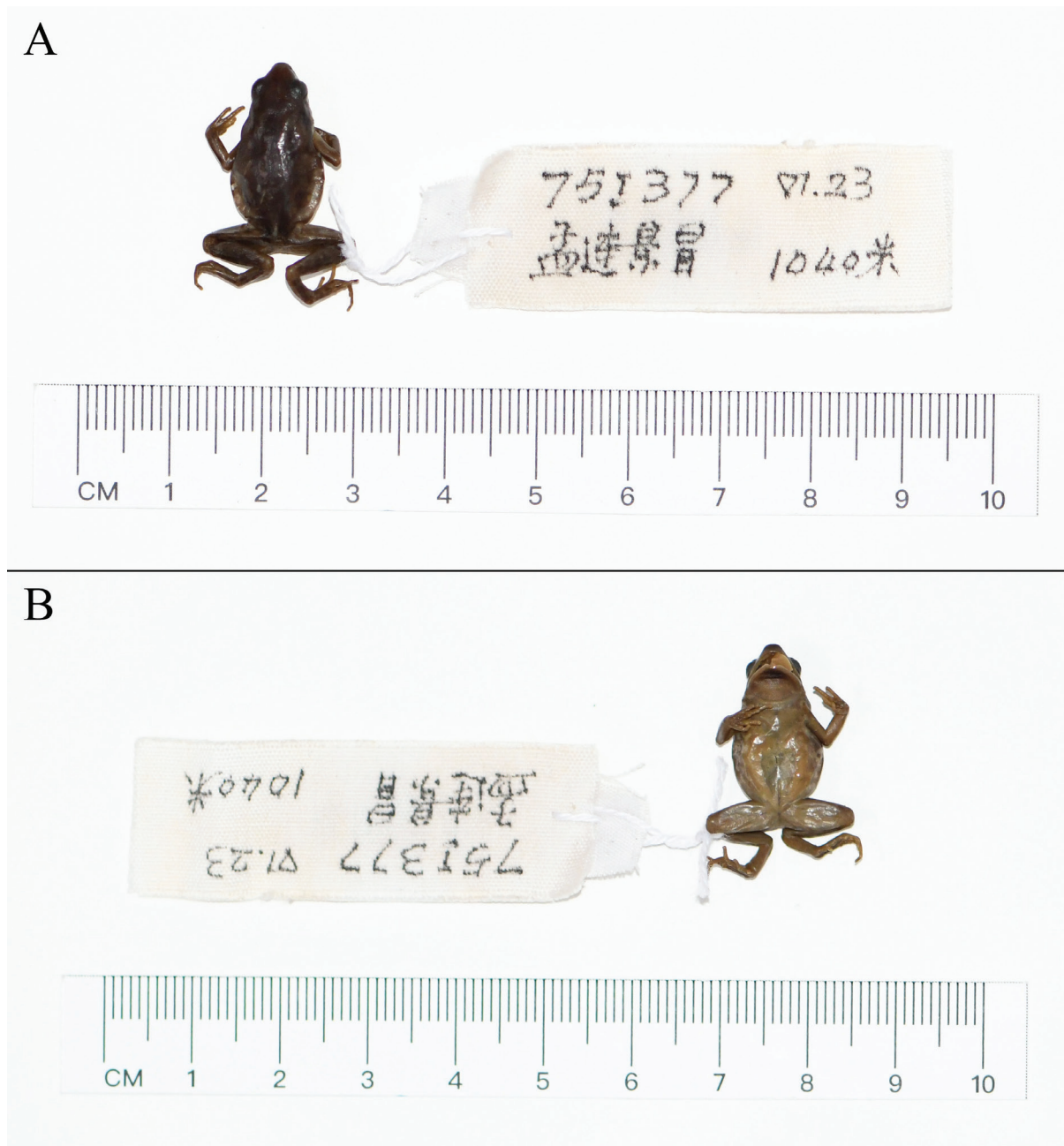


Figure 2. The holotype (KIZ 75I377) of *Micryletta menglienica* in preservative. **A.** Dorsal view; **B.** Ventral view.

Type locality. Jingmao Village, Jingxin Township, Menglian County, Puer City, Yunnan Province, China.

Distribution. Menglian County, Puer City, Yunnan Province, China (Fig. 5).

Diagnosis. Small body size; head width approximately equal to head length; tympanum small and indistinct; supratympanic fold distinct; vomerine teeth absent; tongue oval, with no notch at posterior tip; forelimbs slender and long, hindlimbs slender and relatively short, tibiotarsal articulation of adpressed limb reaching eye or level of between eye and tympanum; subtle longitudinal median ridge present on dorsum; dorsolateral fold absent; dorsum of body purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern; no

bands on dorsum of limbs; a black streak extending from tip of the snout to crotch; upper lip white; Ventral side of head, body, and limbs grayish brown or purple gray, white marbling patterns on chest and belly, some white spots on chin region and ventral side of limbs.

Description of the topotype specimens. *Specimens examined.* KIZ20210708–KIZ20210712, five adult males, collected by Shuo Liu on 17 July 2021 from Jingmao Village, Jingxin Township, Menglian County, Puer City, Yunnan Province, China (22°29'16"N, 99°40'20"E; at an elevation of 1050 m).

Morphological description. SVL 18.7–21.8 mm; habitus relatively slender; head small and triangular, width approximately equal to length (HW/HL 0.99–1.00); snout

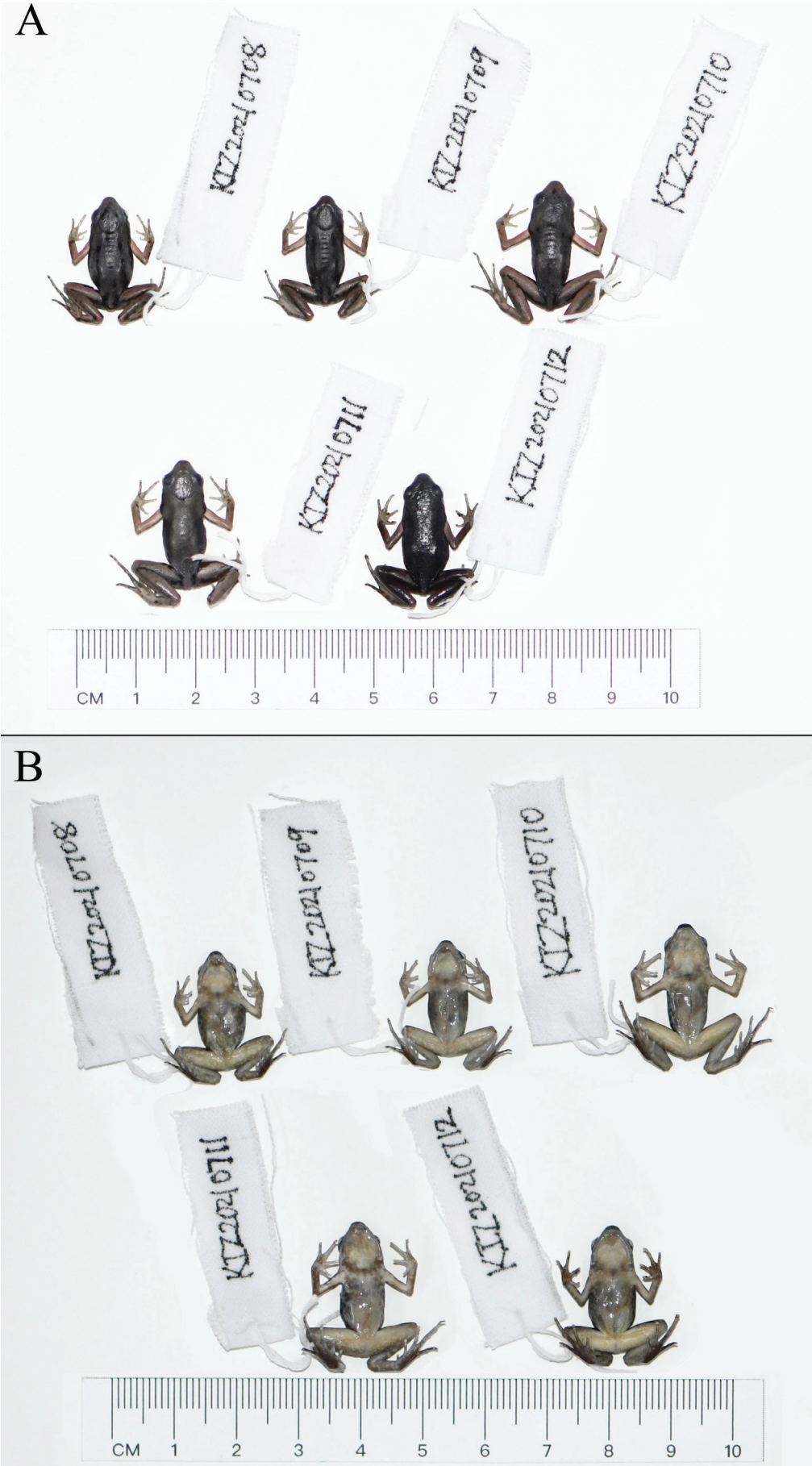


Figure 3. The newly collected specimens of *Micryletta menglienicus* in preservative. **A.** Dorsal view; **B.** Ventral view.

abruptly rounded in dorsal view and slightly acuminate in profile, projecting beyond margin of lower jaw; eyes relatively small, slightly protuberant, pupil oval, transverse, eye diameter slightly smaller than interorbital distance (EL/IOD 0.85–0.96). Top of head flat, canthus rostralis rounded and distinct; loreal region weakly concave; nostril round, closer to tip of snout than to eye; interorbital distance greater than internarial distance (IOD/IND 1.14–1.44) and upper eyelid width (IOD/UEW 1.56–2.17). Tympanum rounded, small (TMP/HL 0.16–0.21) and indistinct; supratympanic fold distinct. Choanae rounded; vomerine teeth absent; opening of vocal sac long cleft; tongue oval, with no notch at posterior tip.

Forelimbs slender and long (FLL/SVL 0.71–0.72). Fingers slender with no webbing, rounded in cross-section, no lateral fringes; relative finger lengths: I<II<IV<III; tips of fingers round and not dilated; subarticular tubercles on fingers distinct, rounded and prominent, formula 1, 1, 2, 2; supernumerary tubercles on palm present and developed; three metacarpal tubercles, inner one rounded and smallest, median one rounded and almost directly in front of oval outer one; two rounded and one elongated prominent supernumerary palmar tubercles on the base of fingers II–IV, respectively; nuptial pad absent.

Hindlimbs slender and relatively short (HLL/SVL 1.47–1.53); tibiotarsal articulation of adpressed limb reaching eye; foot slightly longer than tibia (FL/TL 1.01–1.13). Relative toe lengths: I<II<V<III<IV; tarsal fold absent; tips of toes round and not dilated, slightly wider than those of fingers; webbing between toes absent; subarticular tubercles on toes oval and prominent, formula: 1, 1, 2, 3, 2; dermal ridges present under 2nd to 4th toes but indistinct; inner metatarsal tubercle rounded, prominent, and small; outer metatarsal tubercle absent.

Dorsal skin scattered with small tubercles on dorsum of body, flanks, and hindlimbs, dorsal skin of forelimbs smooth; subtle longitudinal median ridge present on dorsum; dorsolateral fold absent; lateral sides of head smooth; ventral skin of body and limbs smooth.

Coloration in life. Coloration varies greatly, dorsum of body purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern. Dorsum of forelimbs light yellow, dorsum of hindlimbs the same color as dorsum of body, no bands on dorsum of limbs. A black streak extending from tip of the snout to crotch, lower part of the streak on flank grayish white with some black spots. Upper lip white. Ventral side of head, body, and limbs grayish brown or purple grey, white marbling patterns on chest and belly, some white spots on chin region and ventral side of limbs. Iris bi-colored, with upper third bronze and lower two-thirds brownish black.

Natural history. All specimens were found under the dead leaves on the ground at night (Fig. 6). Once startled, they jumped out from under the dead leaves. The collection site is surrounded by broad-leaved forest and bamboo, and there is a river nearby. No reproductive behavior was observed.

Morphological comparison. *Micryletta menglienica* differs from *M. aishani* by head width approximately equal to head length (vs. head wider than long); snout abruptly rounded in dorsal view and slightly acuminate in lateral view (vs. snout shape nearly truncate in dorsal view and acute in lateral view); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching armpit).

Micryletta menglienica differs from *M. dissimulans* by dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum reddish brown with merging irregular shaped brown blotches edged in beige); a black streak extending from tip of the snout to crotch (vs. no black streak extending from tip of the snout to crotch); white stripes on upper lips present (vs. absent); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching tympanum).

Micryletta menglienica differs from *M. erythropoda* by relatively smaller body (SVL 19.4–23.4 mm vs. up to 30 mm); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum gray or beige to saturated ochre or brick red, dark contrasting round or irregular shape spots irregularly scattered throughout the dorsum); outer metatarsal tubercle absent (vs. present); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching posterior edge of tympanum).

Micryletta menglienica differs from *M. hekouensis* by head width approximately equal to head length (vs. head wider than long); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. areas above canthus rostralis, upper eyelids, areas posterior to eyelids, and dorsum of upper arms golden, other parts of dorsum almost solid black or yellowish gray with brownish black stripes); supratympanic fold distinct (vs. supratympanic fold indistinct); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching front of eye).

Micryletta menglienica differs from *M. immaculata* by relatively smaller body (SVL 19.4–23.4 mm vs. up to 23.3–30.1 mm); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum bronze brown to reddish brown without dark patterns); flank with black streak (vs. flank with no streak); webbing between toes absent (vs. basal and poorly developed); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching tympanum).

Micryletta menglienica differs from *M. inornata* sensu stricto from Sumatra, Indonesia, and from Tanintharyi, Myanmar, by head width approximately equal to head length (vs. head wider than long); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum brownish gray with a silver tinge and irregular blackish brown blotches of variable size); supratympanic fold distinct (vs.

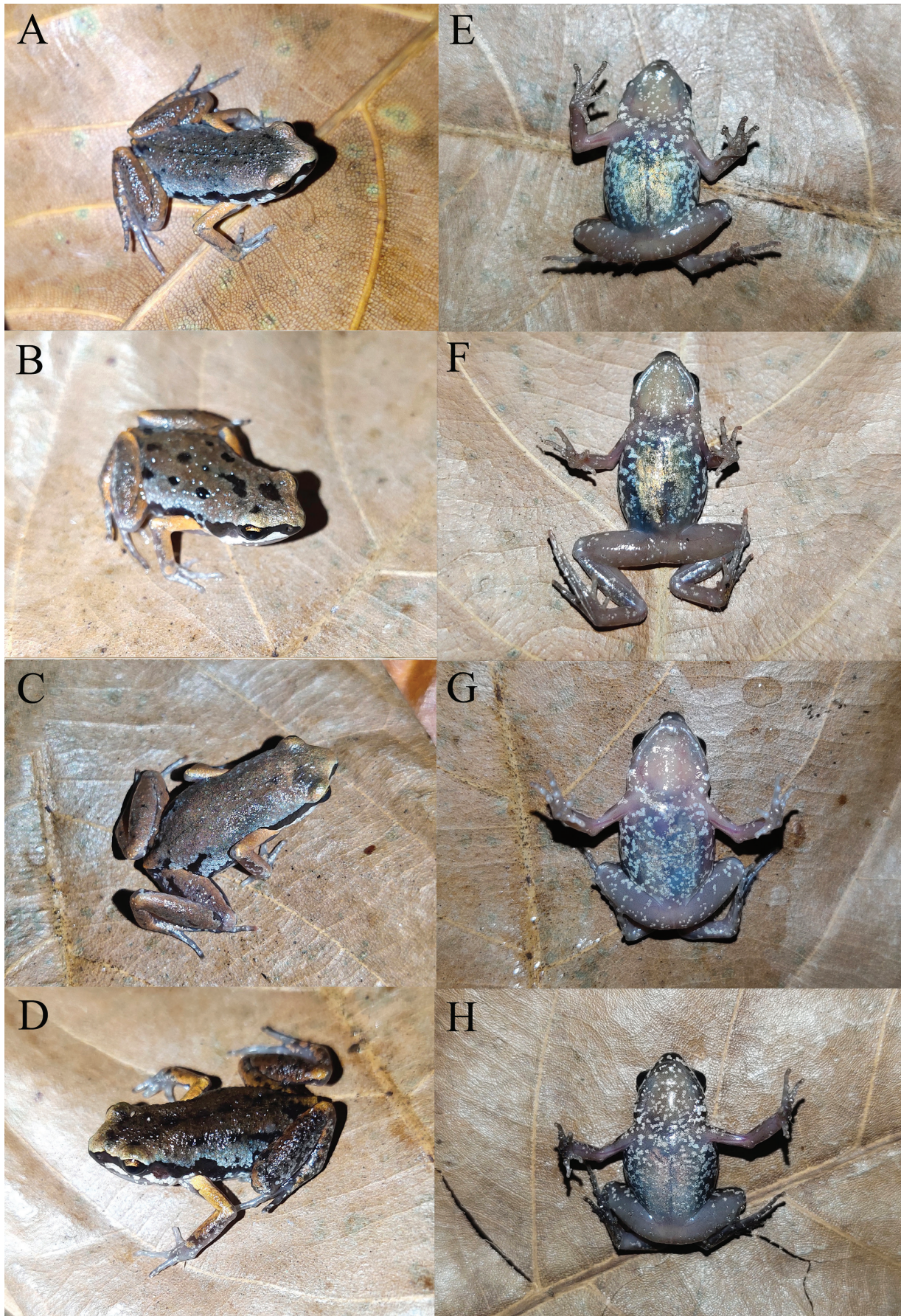


Figure 4. *Micryletta menglienicus* in life from Jingmao Village, Jingxin Township, Menglian County, Puer City, Yunnan Province, China. A–D. Dorsal view; E–H. Ventral view.

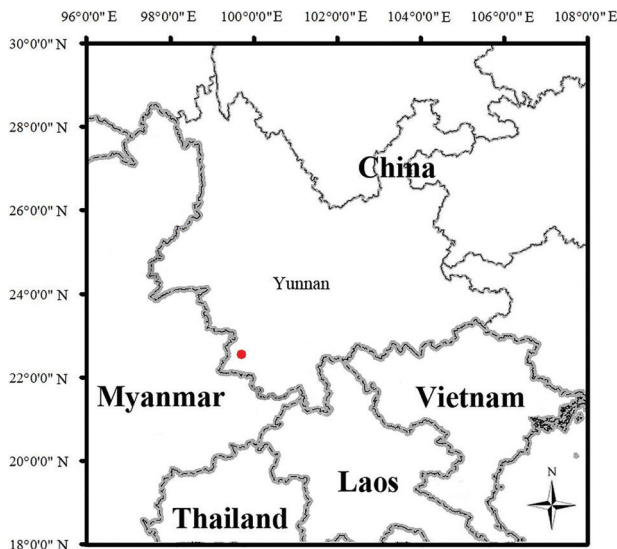


Figure 5. Map showing the type locality (red dot) of *Micryletta menglienica* in Menglian County, Puer City, Yunnan Province, China.

supratympanic fold weakly developed); no dark bands or spots on dorsum of limbs (vs. indistinct dark bands or irregular dark spots and blotches on dorsum of limbs).

Micryletta menglienica differs from *M. lineata* by relatively larger body in males (SVL 19.4–23.4 mm vs. 19.0–19.2 mm); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum brownish grey with three straight continuous or broken lines); Ventral side of head, body, and limbs grayish brown or purple grey, white marbling patterns on chest and belly, some white spots on chin region and ventral side of limbs (vs. venter beige with light brown mottling along throat).

Micryletta menglienica differs from *M. nigromaculata* by dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum brown to reddish brown with dark brown irregular hourglass shaped pattern and two large dark inguinal spots); a black streak extending from tip of the snout to crotch (vs. no black streak extending from tip of the snout to crotch); white stripes on upper lips present (vs. absent); chin region in males brownish black (vs. whitish with light gray marbling).

Micryletta menglienica differs from *M. sumatrana* by relatively larger body in males (SVL 19.4–23.4 mm vs. 17.4 mm); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum golden brown scattered with small black spots); flank with black streak (vs. flank with irregular blackish patches with cream mottling); dark cross bands on tibia and tarsus absent (vs. present); a few small white spots on ventral side of limbs (vs. dark brown and cream mottling on ventral side of limbs); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching front of eye).

Micryletta menglienica differs from *M. steinegeri* by relatively smaller body in males (SVL 19.4–23.4 mm vs.



Figure 6. Habitat of *Micryletta menglienica* at Jingmao Village, Jingxin Township, Menglian County, Puer City, Yunnan Province, China.

up to 24.3 mm); dorsum purple brown, blueish gray, or dark brown, with small or large black spots, black stripes, or no pattern (vs. dorsum dark gray to violet with irregular dark blotches or speckles); supratympanic fold distinct (vs. supratympanic fold weak and indistinct); webbing between toes absent (vs. rudimentary webbing); tibiotarsal articulation adpressed limb reaching eye or between eye and tympanum (vs. reaching tympanum).

Discussion

In Yang and Su (1980), the type locality of *Kalophrynus menglienicus* was given as Menglian, but more precise locality was not given. Menglian is a county, and covering a large area. We checked the type specimens of *K. menglienicus* and found that the more precise toponym was written on the original labels: “孟连景冒”. This toponym refers to Jingmao Village, Jingxin Township, Menglian County, Puer City, Yunnan Province, China. The topotype specimens of *K. menglienicus* we collected are from the exact site of the type locality of *K. menglienicus*.

According to the original description of Yang and Su (1980), the skin is scattered with small horny granules, and there is an inverted triangle transparent area in the center of the belly. However, we found that some individuals have both of these two characters while others have one or none of these two characters among the specimens from the same locality. It means that these characters are not stable, so they cannot be used as diagnosis characters.

Micryletta inornata, another species of Microhylidae, was widely reported from mainland Southeast Asia. However, recent phylogenetic studies have indicated that *M. inornata* sensu stricto is restricted to Indonesia and southern Myanmar, and the populations of *M. inornata* sensu lato contain several undescribed paraphyletic lineages with respect to other named taxa (Alhadi et al. 2019; Das et al. 2019; Munir et al. 2020; Miller et al.

2021). Previously, Yang and Rao (2008), Fei et al. (2009), Fei et al. (2012), Fei (2020), and AmphibiaChina (2021) all recorded *Micryletta inornata* distributed in Xishuangbanna Prefecture, Yunnan Province, China. We suspect that the population recorded as *M. inornata* in Xishuangbanna is likely to be *Micryletta menglienicus*; more specimens are needed to be collected to verify this inference.

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References

- Alhadi F, Hamidy A, Farajallah A, Munir M, Atmaja VY, Garg S, Biju SD, Smith EN (2019) Rediscovery of *Micryletta inornata* (Boulenger, 1890) from Sumatra: redescription, molecular identity and taxonomic implications. *Zootaxa* 4613(1): 111–126. <https://doi.org/10.11646/zootaxa.4613.1.5>
- AmphibiaChina (2021) The database of Chinese amphibians. Electronic Database. <http://www.amphibiachina.org> [Accessed on 4 August 2021]
- Boulenger GA (1890) List of the reptiles, batrachians, and freshwater fishes collected by Professor Moesch and Mr. Iversen in the district of Deli, Sumatra. *Proceedings of the Zoological Society of London* 1890: 30–39.
- Das A, Garg S, Hamidy A, Smith EN, Biju SD (2019) A new species of *Micryletta* frog (Microhylidae) from Northeast India. *PeerJ* 7: e7012. <https://doi.org/10.7717/peerj.7012>
- Dubois A (1987) *Miscelanea taxinomica batrachologica*, II. *Alytes* 6: 1–9.
- Fei L (2020) *Atlas of Amphibians in China* (Field Edition). Henan science and Technology Press, Zhengzhou, 837 pp.
- Fei L, Hu SQ, Ye CY, Huang YZ (2009) *Fauna Sinica* (Vol. 2). Amphibia Anura. Science Press, Beijing, 957 pp.
- Fei L, Ye CY, Jiang JP (2012) *Colored Atlas of Chinese Amphibians and Their Distributions*. Sichuan Publishing House of Science and Technology, Chengdu, 620 pp.
- Hedges SB (1994) Molecular evidence for the origin of birds. *Proceedings of the National Academy of Sciences of the United States of America* 91(7): 2621–2624. <https://doi.org/10.1073/pnas.91.7.2621>
- Hillis DM, Moritz C, Mable BK (1996) *Molecular Systematics*. Second Edition. Sinauer Associates, Sunderland, 655 pp. <https://doi.org/10.2307/1447682>
- Kalyaanamoorthy S, Minh BQ, Wong TKF, von Haeseler A, Jermiin LS (2017) ModelFinder: fast model selection for accurate phylogenetic estimates. *Nature Methods* 14: 587–589. <https://doi.org/10.1038/nmeth.4285>
- Kumar S, Stecher G, Li M, Knyaz C, Tamura K (2018) MEGA X: Molecular Evolutionary Genetics Analysis across computing platforms. *Molecular Biology and Evolution* 35: 1547–1549. <https://doi.org/10.1093/molbev/msy096>
- Liu S, Hou M, Mo MZ, Rao DQ (2021) A new species of *Micryletta* Dubois, 1987 (Anura, Microhylidae) from Yunnan Province, China. *Herpetozoa* 34: 131–140. <https://doi.org/10.3897/herpetozoa.32.e69755>
- Matsui M, Eto K, Belabut DM, Nishikawa K (2017) A New *Kalophrynus* (Amphibia, Anura, Microhylidae) from Peninsular Malaysia. *Current Herpetology* 36(2): 75–86. <https://doi.org/10.5358/hcj.36.75>
- Matsui M, Shimada T, Liu W-Z, Maryati M, Khonsue W, Orlov N (2006) Phylogenetic relationships of Oriental torrent frogs in the genus *Amolops* and its allies (Amphibia, Anura, Ranidae). *Molecular Phylogenetics and Evolution* 38(3): 659–666. <https://doi.org/10.1016/j.ympev.2005.11.019>
- Miller AH, Zug GR, Wogan GOU, Lee JL, Mulcahy DG (2021) Phylogeny, Diversity, and Distribution of *Micryletta* (Anura: Microhylidae) in Myanmar. *Ichthyology & Herpetology* 109(1): 245–257. <https://doi.org/10.1643/h2020100>
- Munir M, Hamidy A, Matsui M, Kusri MD, Nishikawa K (2020) A new species of *Micryletta* (Amphibia: Anura) from Sumatra, Indonesia. *Zoological Science* 37(3): 295–301. <https://doi.org/10.2108/zs200006>
- Ronquist F, Teslenko M, Van Der Mark P, Ayres DL, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539–542. <https://doi.org/10.1093/sysbio/sys029>
- Sambrook JF, Russell RW (2001) *Molecular Cloning: A Laboratory Manual*. 3rd edn. Cold Spring Harbor Laboratory Press, New York, 2345 pp.
- Silvestro D, Michalak I (2012) raxmlGUI: a graphical front-end for RAXML. *Organisms Diversity and Evolution* 12(4): 335–337. <https://doi.org/10.1007/s13127-011-0056-0>
- Thompson JD, Higgins DG, Gibson TJ (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Research* 22: 4673–4680. <https://doi.org/10.1093/nar/22.22.4673>
- Yang DT, Rao DQ (2008) *Amphibia and Reptilia of Yunnan*. Yunnan Publishing Group Corporation, Yunnan Science and Technology Press, Kunming, 411 pp.
- Yang DT, Su CY (1980) A new species of the family Microhylidae frog from Yunnan. *Zoological Research* 1: 257–260.
- Yang JH, Poyarkov NA (2021) A new species of the genus *Micryletta* (Anura, Microhylidae) from Hainan Island, China. *Zoological Research* 42(2): 234–240. <https://doi.org/10.24272/j.issn.2095-8137.2020.333>