



A new species of *Kalophrynus* (Amphibia, Anura, Microhylidae) from Southern Peninsular Malaysia

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Abstract

A new microhylid, *Kalophrynus limbooliati* **sp. nov.**, is described from the state of Johor, in the southern part of Peninsular Malaysia. Morphologically, the new species differs from all known congeners in the combination of medium body size; short fourth finger; two subarticular tubercles on fourth finger and none on the fifth toe; presence of light lateral stripe and dark inguinal spot; absence of nuptial pads and outer metatarsal tubercles. Acoustically, the new species clearly differs from all congeners whose calls have been reported, and resembles a syntopic ranid *Hylarana laterimaculata* with a long series of high-pitched whistle like notes.

Key words: *Kalophrynus*, advertisement call, cryptic species, new species, Southeast Asia, taxonomy

Introduction

The microhylid frog genus *Kalophrynus* Tschudi, 1838 has a wide range from Southern China to Java, the Philippines, and Assam, India (Matsui 2009). Peninsular Malaysia is situated in the center of this distribution and four species of *Kalophrynus* have been recorded from there (*K. pleurostigma* Tschudi; *K. robinsoni* Smith; *K. palmatisimus* Kiew; and *K. yongi* Matsui) (Berry 1975; Kiew 1984; Das & Haas 2003; Matsui 2009). Of these, *K. pleurostigma* was once thought to widely occur in the Peninsula (e.g. Berry 1975), but some records for this species later proved to include *K. palmatisimus* (Kiew 1984). Taxonomic problems amongst specimens referred to *K. pleurostigma* from Peninsula Malaysia, however, have never been completely resolved; in particular Dring (1979) noted morphological variation in this ‘species’, which is not attributable to misidentification of *K. palmatisimus* with well-developed toe webbing.

During fieldwork to the southern part of Peninsular Malaysia in 2009 we detected strange frog calls that resemble those of a ranid, *Hylarana laterimaculata* (Barbour & Noble) to the human ear (Leong *et al.* 2003). The calls, however, were heard from the forest litter, and not from near pools and streams where *H. laterimaculata* was actually found. Intensive searches in forest revealed that these calls were coming from a *Kalophrynus* species that superficially resembled *K. pleurostigma*.

The *Kalophrynus* in question is acoustically very unique and clearly differs from all congeneric species whose calls are known (see Matsui 2009; Dehling 2011). Because acoustic signals play an important role in reproductive isolation among frog species, unique calls alone justify recognizing the *Kalophrynus* from the southern part of the Peninsula as a distinct species. However, in addition to this difference, later detailed examination also revealed a suite of morphological characteristics that differentiate this taxon from all other *Kalophrynus*, including the superficially similar *K. pleurostigma*. Sequence divergences at the mitochondrial 16S rRNA gene, which is often utilized in elucidating cryptic species (e.g. Fouquet *et al.* 2007), were also very large between this species (as *Kalophrynus* sp.) and other *Kalophrynus* (Matsui *et al.* 2011). We herein describe this new species.

Material and methods

Frog surveys were made in the southern Peninsular Malaysia, in the states of Johor and Negeri Sembilan. We recorded frog calls in the field using digital recorders (Zoom H2 and Olympus LS-10), and measured air temperature at the time of recording with a quick-recording thermistor thermometer (Takara A 600). Calls recorded were analyzed with the SoundEdit 2 and SoundEdit Pro (MacroMind-Paracom, Inc) software packages on a Macintosh computer, as described elsewhere (Matsui 1997).

After taking tissues for phylogenetic analyses, specimens were fixed in 10% formalin, preserved in 70% ethanol, and are stored in Herpetological Collections at Universiti Kebangsaan Malaysia (UKMHC) and Graduate School of Human and Environmental Studies, Kyoto University (KUHE).

We took 18 body measurements following Matsui (1984): 1) snout-vent length (SVL); 2) head length (HL); 3) snout length (SL); 4) nostril-eye distance (N-EL); 5) eye length (EL, including upper eyelid, not superficial eyeball diameter); 6) tympanum-eye length (T-EL); 7) tympanum diameter (TD); 8) head width (HW); 9) internarial distance (IND); 10) interorbital distance (IOD); 11) upper eyelid width (UEW); 12) lower arm and hand length (LAL), from elbow to tip of third finger; 13) forelimb length (FLL); 14) tibia length (TL); 15) foot length (FL); 16) hindlimb length (HLL); 17) inner metatarsal tubercle length (IMTL); and 18) first toe length (1TL), from distal end of inner metatarsal tubercle to tip of first toe. We made all measurements to the nearest 0.1 mm with dial calipers under a binocular dissecting microscope. For the description of toe-webbing states we follow Savage (1975).

For morphological comparisons, we examined specimens of *Kalophrynus* stored at KUHE; The Natural History Museum, London (BM); Museo Civico di Storia Naturale, Genova (MSNG); National Museum of Natural History (Naturalis), Leiden (= Rijksmuseum van Natuurlijke Historie Leiden, RMNH); and Forschungsinstitut und Naturmuseum Senckenberg, Frankfurt am Main (SMF).

We examined the mitochondrial DNA genes 12S and 16S rRNA from a male holotype together with seven congeneric species. Molecular phylogenetic methods and results are reported in Matsui *et al.* (2011) (as *Kalophrynus* sp.).

Results

Systematics

Kalophrynus limbooliati sp. nov.

(Figs. 1–3)

Synonymy: *Kalophrynus pleurostigma*: Lim and Lim, 1992, p. 40, Bukit Timah, Singapore.

Holotype. UKMHC 705 (formerly KUHE 53284), adult male from Gunung (= Mt.) Pulai, Kpg. (Kampung = village) Sri, Kulai, State of Johor, Peninsular Malaysia (01°36' N, 103° 32' E, 457 m a.s.l.), collected by Kanto Nishikawa on 6 September 2009.

Paratypes. KUHE 53314, 53315, two adult males from Gunung Lambak (02°00' N, 103°22' E, 75 m a.s.l.), Kluang, Johor, collected 9 September 2009 by Kanto Nishikawa and Masafumi Matsui; KUHE 52061, a juvenile from Jalan Lombong, Panti (01°49' N, 103°50' E, 57 m a.s.l.), Kota Tinggi, Johor, collected 30 July 2008 by Amir Hamidy, Daicus Belabut, and Masafumi Matsui.

Etymology. The specific name is dedicated to Dr. Lim Boo Liat, Fellow of the Academy of Sciences Malaysia, who is a pioneer of field zoology in Malaysia.

Diagnosis. The new species is assigned to a member of *Kalophrynus* through mitochondrial DNA genealogy (Matsui *et al.* 2011) together with the following morphological characteristics: No spine-like projection of skin at heel and elbow; belly without a network pattern; tips of fingers not expanded; underside of fingers without greatly enlarged tubercles; snout less than twice diameter of eye; inner metatarsal tubercle low, not shovel-like; tympanum visible.

A medium-sized species that can be distinguished from all congeneric species by the following combination of characters; adult males 26.2–28.7 mm SVL (mean = 27.3, n = 3); snout pointed, directed downwards; third toe lon-

ger than fifth; projection of fourth finger from palm shorter than length of terminal phalanx of third finger; two sub-articular tubercles under fourth finger; no subarticular tubercles under fifth toe; indistinct gland dorsal to arm insertion; no nuptial pads or asperities; forelimb not very stout, without a humeral spine in males; outer metatarsal tubercle absent; light lateral stripe; usually with distinct inguinal dark spot without white rim or spotting; calls similar to *Hylarana laterimaculata* with a long series of high-pitched whistle like notes.

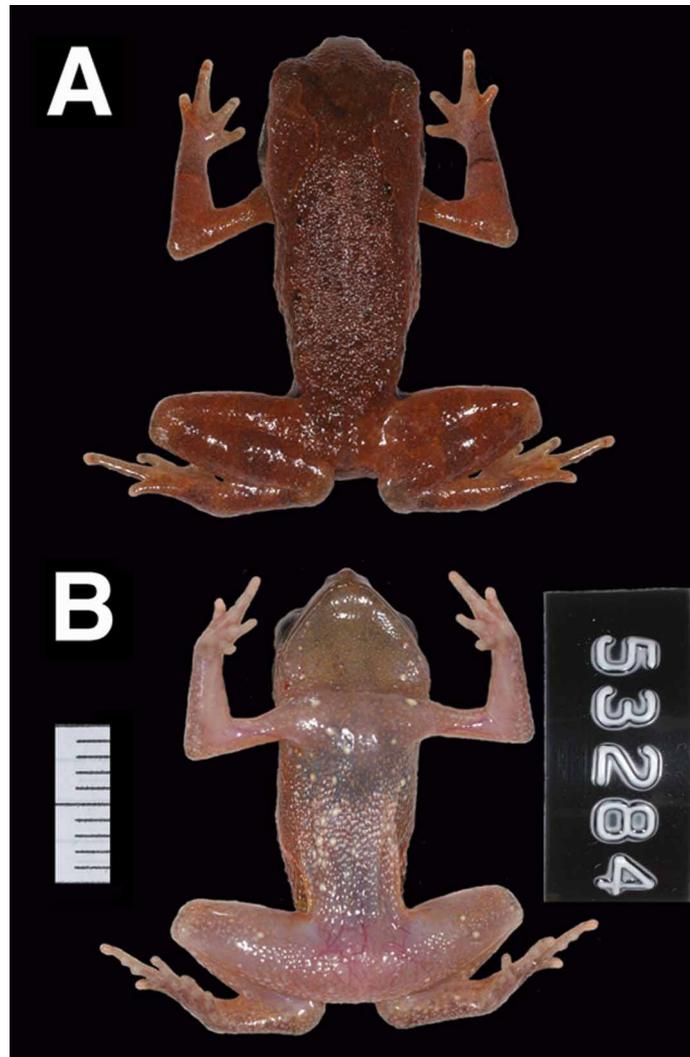


FIGURE 1. (A) Dorsal and (B) ventral views of the holotype of *Kalophrynus limbooliati* **sp. nov.** (male, UKMHC 705). Scale bar, 10 mm.

Description of holotype (measurements in mm). Medium sized (SVL 26.2); habitus moderately stocky (Figs. 1, 3); head triangular, slightly shorter (8.6) than wide (9.0); snout pointed, directed downwards in profile (Fig. 3), projecting beyond lower jaw; eye moderate, as long (3.5) as snout (3.5); canthus rostralis not sharply defined, straight; loreal region vertical, very slightly concave; nostril below canthus rostralis, directed laterally, much closer to tip of snout (1.1) than to eye (2.0); interorbital distance (3.7) much wider than internarial distance (2.1), the latter subequal to upper eyelid (2.2); pineal spot absent; tympanum distinct, diameter (2.3) more than three-fifths that of eye, and very close to eye (0.3); upper jaw edentate; tongue entire, without papillae; a crenulated ridge of skin on palate posterior to eye, preceded by a shorter, similarly notched one between posterior halves of eye; a median, subgular vocal sac; vocal openings posterior to rictus.

Forelimb long (18.0, 72% of SVL) and slender; fingers thick, basally slightly webbed; tips rounded, not dilated; relative length of fingers: $IV < I < II < III$; portion of fourth finger projecting from palm (0.9) shorter than length of terminal phalanx of third finger (1.4); outer palmar tubercle large and fleshy, inner indistinct; subarticular tubercles between finger tip and palmer tubercle rounded, two on fingers I, II and IV, three on III (Fig. 2A); humeral spine absent.



FIGURE 2. Ventral views of (A) hand and (B) foot of the holotype of *Kalophrynus limbooliati* **sp. nov.** (male, UKMHC 705). Scale bar, 5 mm.



FIGURE 3. Dorsolateral view of the holotype of *Kalophrynus limbooliati* **sp. nov.** (male, UKMHC 705) from Gunung Pulai, Johor.

Hindlimb moderately long (35.5, 135% of SVL) and slender; tibia moderately long (10.9, 42% of SVL), heels not overlapping when folded limbs held at right angles to body; tibiotarsal articulation of adpressed limb reaching to the center of tympanum when held alongside body; foot (8.7) much shorter than tibia (FL, 80% of TL); toe tips rounded; relative length of toes: $I < V < II < III < IV$; webbing poorly developed (Fig. 2B), formula: **I** 1–2 **II** 2–3 **III** 2–3 **IV** 3–1 **V**; subarticular tubercles numbering one on first and second toes, two on third toe, and three on fourth toe; distal tubercle on third toe and distal and middle ones on fourth toe prominent and rounded, others small and less distinct; no subarticular tubercles on fifth toe; inner metatarsal tubercle oval, length (1.0) more than half of first toe length (1.5); an indistinct outer metatarsal tubercle (Fig. 2B).

Skin coarsely granular dorsally, with small tubercles scattered posteriorly from behind upper eyelid to vent; tips of tubercles forming white asperities; an indistinct gland on side of head behind tympanum, not delimited by a sinuous groove; upper lateral surfaces of body with scattered minute tubercles; lower lateral surfaces of body, posterior half of chin, abdomen, and posterior side of thighs with large, flattened glandules; skin of gular region not modified for vocal sac; inner and outer margins of fourth finger without skin fringes; nuptial pads absent.

Color. In life, ground color of dorsum highly variable from light orange brown to dark chocolate brown, with several small dark brown spots and obscure hour-glass shaped marking from upper eyelid to shoulder; a light dorsolateral line extending from snout tip through margin of upper eyelid to groin, forming a boundary between lighter dorsum and darker sides of head and shoulder; tips of dorsal granules whitish; limbs dorsally brown with an obscure darker bar (Figs. 1A, 3); ventrum pinkish gray, with scattered white flecks (Fig. 1B); inguinal spot entirely black without lighter borders or pattern; iris golden with black pigmentation. In preservative, color and pattern have generally faded but not obviously changed.

Variation. Individuals of the type series are generally similar in morphology (Table 1). In both adult male paratypes (KUHE 53314, 53315), internarial distance is larger than upper eyelid width. They have darker throat and more developed asperities on dorsum than the holotype. The inguinal black spot varies in size, and is lacking on the left side in one individual (KUHE 53315). The single juvenile (KUHE 52061) has smooth dorsum, with several black spots scattered from shoulder, some continuing to the inguinal marking. Chin and anterior part of ventrum are dotted with dark spots in this juvenile paratype.

TABLE 1. Measurements of 18 characters (SVL in mm and medians of % ratios [R] of other characters to SVL) in the holotype and two paratypes (all adult males) of *Kalophrynus limbooliati*. See text for character abbreviations.

	SVL	RHL	RHW	RIND	RIOD	RUEW
UKMHC 705 (holotype)	26.2	32.6	34.3	8	13.9	8.2
KUHE 53314 (paratype)	27	32.6	32.2	9.6	12.2	7.8
KUHE 53315 (paratype)	28.7	31.9	32.6	9.1	11.7	7.3
mean \pm SD/median	27.3 \pm 1.28	32.6	32.6	9.1	12.2	7.8
	RSL	REL	RN-EL	RTD	RT-EL	RLAL
UKMHC 705 (holotype)	13.4	13.4	7.6	8.8	1	47.3
KUHE 53314 (paratype)	12.8	17.4	6.4	8.1	0.4	46.7
KUHE 53315 (paratype)	12	13.4	6.8	9.4	0.7	46
median	12.8	13.4	6.8	8.8	0.7	46.7
	RFLL	RTL	RFL	RHLL	RIMTL	R1TL
UKMHC 705 (holotype)	68	41.4	33.2	135.5	3.8	5.7
KUHE 53314 (paratype)	63	41.3	35.6	134.1	3.3	5.6
KUHE 53315 (paratype)	63.4	41.5	35.2	128.6	3	5.7
median	63.4	41.4	35.2	134.1	3.3	5.7

Range. Southern part of Western (Peninsular) Malaysia: Johor (Panti, Kota Tinggi; Gunung Pulai, Kulai [350–605 m a.s.l.]; Gunung Lambak, Kluang [02°00' E, 103°22' E, 75–185 m a.s.l.]; Trail Lagenda, Gunung Ledang, Tangkak [02°22' N, 102°37' E, 750–895 m a.s.l.: recorded by calls]) and Negeri Sembilan (Sungai Kenaboi, Kenaboi: recorded by calls). Singapore: Bukit Timah Nature Reserve (Lim & Lim 1992: see below, discussion). Possibly Janda Baik, Pahang, Western (Peninsular) Malaysia (Dring 1979: see below, discussion).

Natural history. On Gunung Pulai, Gunung Ledang, and Gunung Lambak, males were found calling in widely scattered choruses at dry nights (from 1930 h) in early September in the dense secondary broad-leaf forest. Calling males hid themselves among dead leaves and were very difficult to locate, but responded to playback of recorded calls and/or whistles imitating their calls. There were no large bodies of water at the calling sites. The associated anuran species observed at the nearest waters were *Hylarana laterimaculata* and *Microhyla heymonsi* Vogt.

At Sungai Kenaboi, calls were heard in secondary and bamboo forests on dry night. *Microhyla heymonsi*, *Fejervarya limnocharis* (Gravenhorst), *Humerana miopus* (Boulenger), *Hylarana labialis* (Boulenger), and *Polypedates leucomystax* (Gravenhorst) were observed at the same locality around pools along a forest road.

The single juvenile paratype from Pantı (KUHE 52061) was found walking at a dry night at the edge of secondary forest in late July. In early September, no further specimens of this species or calls were detected at this locality. Associated anuran species observed were *Phrynoidis aspera* (Gravenhorst), *Kaloula pulchra* Gray, *Micryletta inornata* (Boulenger), *Limnonectes blythii* (Boulenger), *H. miopus*, and *H. labialis*.

Call characteristics. We recorded the calls of *K. limbooliati* at Gunung Pulai, Kulai on 6 September 2009 (air temperature 23.9 C), Gunung Lambak, Kluang on 7 September 2009 (air temperature 27.2 C), and at Kenaboi, Negeri Sembilan on 10 and 11 September 2009 (air temperature 26.4 C). The advertisement call (Fig. 4) consists of a very long series of 24–61 (mean \pm SD = 41.1 \pm 11.2, n = 62/seven males) unpulsed notes and lasts about 6.3–15.2 (mean \pm SD = 10.9 \pm 2.8) s. The note repetition rate is 3.31–4.20 (mean \pm SD = 3.72 \pm 0.30) notes per s. Each note lasts about 61–76 (mean \pm SD = 70.0 \pm 6.0) ms, and time interval between two notes varies from 238–310 (mean \pm SD = 271.7 \pm 25.4) ms. The dominant frequency lies at 1632–2008 (mean \pm SD = 1909 \pm 156) Hz, and harmonics are at about 3000–4000 and 7000–8000 Hz (Fig. 4A). The call has marked frequency and intensity modulations, and frequencies abruptly increase to the middle and then decrease towards the end of a note.

Comparisons. Males of *K. limbooliati* **sp. nov.** with a SVL of 26.2–28.7 mm SVL (mean = 27.3 mm) are of similar size to *K. heterochirus* Boulenger (24.1–27.2 mm, mean = 26.3 mm), *K. eok* Das & Haas (26.3 mm), and *K. punctatus* Peters (28.3 mm); it can be readily differentiated from these species as follows: white spots in a black inguinal marking absent (present in *K. heterochirus*); second finger with two subarticular tubercles (with single subarticular tubercle in *K. eok*); fifth toe is not projecting as far as or farther than the third toe unlike *K. punctatus* (data from Matsui 2009).

Kalophrynus limbooliati **sp. nov.** is smaller in male body size than *K. yongi* (28.8–31.0 mm, mean = 30.3 mm), *K. calciphilus* Dehling (29.7–30.1 mm), *K. minusculus* Iskandar (32.2 mm), *K. palmatissimus* (31.2–38.8 mm, mean = 34.5 mm), *K. orangensis* Dutta, Ahmed & Das (35–38 mm), *K. intermedius* Inger (37.9–40.5 mm, mean = 39.2 mm), *K. baluensis* Kiew (34.8–39.0 mm), *K. stellatus* Stejneger (35.0–45.0 mm, mean = 39.3 mm), *K. pleurostigma* (35.0–50.4 mm, mean = 42.2 mm), and *K. interlineatus* Blyth (37.4–47.7 mm), and larger than *K. robinsoni* (16.8 mm), *K. nubicola* Dring (14.4–24.4 mm), *K. menglienicus* Yang & Su (19.8–23.4 mm, mean = 21.2 mm), *K. bunguranus* (Günther) (20.7–22.8 mm, mean = 21.8 mm), and *K. subterrestris* Inger (21.0–23.4 mm, mean = 22.6 mm) (data from Inger 1954; Matsui 2009; Dehling 2011).

Other than the body size differences, *K. limbooliati* **sp. nov.** can be differentiated from these species as follows: from *K. yongi* by having a normal humerus (strongly developed terminal ridges on humerus, and related skin modification in *K. yongi*); from *K. calciphilus* by the absence of outer metatarsal tubercle, presence of thinner dorsolateral stripe, without black margin dorsally, and dorsally more pointed snout (prominent tubercle present, dorsolateral stripe wide, bordered by black on both margins, and dorsally obtusely pointed in *K. calciphilus*); and from *K. intermedius* by the possession of light lateral stripe and inguinal black marking (stripe and marking absent in *K. intermedius*).

In *K. limbooliati* **sp. nov.**, the fourth finger from palm is shorter than length of the terminal phalanx of third finger, unlike *K. minusculus*, *K. palmatissimus*, *K. orangensis*, *K. interlineatus*, and *K. pleurostigma*, in which the fourth finger is longer. In addition, *K. limbooliati* **sp. nov.** has poorly developed webbing on fourth toe, which barely reaches median subarticular tubercle, sharply contrasting with the well-developed webbing in *K. palmatissimus*; lacks distinct dorsal marking unlike *K. minusculus* and *K. baluensis*, which usually have clear dark markings on dorsum; has black inguinal marking (yellow in *K. baluensis*). *Kalophrynus limbooliati* further differs from *K. stellatus* and *K. pleurostigma* by the lack of nuptial pads in males; and from *K. robinsoni* in the absence of spinous nuptial pads in the males.

Kalophrynus limbooliati **sp. nov.** differs from *K. nubicola* by the possession of distinct subarticular tubercles of fingers and toes (indistinct or absent in *K. nubicola*); from *K. menglienicus* by the possession of distinct tympanum and toe web (tympanum concealed and toe web absent in *K. menglienicus*); from *K. bunguranus* by the lack of light marking surrounding black inguinal mark (black inguinal marking in a light area in *K. bunguranus*); and from *K. subterrestris* by the presence of two subarticular tubercles on the fourth finger (a single subarticular tubercle in *K. subterrestris*).

The calls of six species of *Kalophrynus* have been analyzed (*K. baluensis*; *K. calciphilus*; *K. interlineatus*; *K. nubicola*; *K. pleurostigma*; and *K. yongi*; Matsui 2009; Dehling 2011). The call of *K. limbooliati* **sp. nov.** is unpulsed unlike the well pulsed call of *K. nubicola* (Dring 1984), and is composed of successive notes unlike the short, single note in *K. calciphilus* (Dehling 2011), *K. baluensis*, *K. heterochirus*, and *K. yongi* (Matsui 2009). *Kalophrynus limbooliati* **sp. nov.** has a call with successive notes like *K. pleurostigma* (Matsui *et al.* 1996), but the

note length is shorter (61–76 vs. 170–487 ms), the note repetition rate is larger (3.3–4.2 vs. 0.9–2.7 notes per s), and the dominant frequency is much higher (1632–2008 vs. 438–575 Hz) than *K. pleurostigma*. The call of *K. interlineatus* is a long trill unlike that of *K. limbooliati* (Matsui *et al.* 1996), and that of *K. palmatissimus* (a soft “ko-ko-ko”: Kiew 1984:150) is clearly very different from that of *K. limbooliati* **sp. nov.**

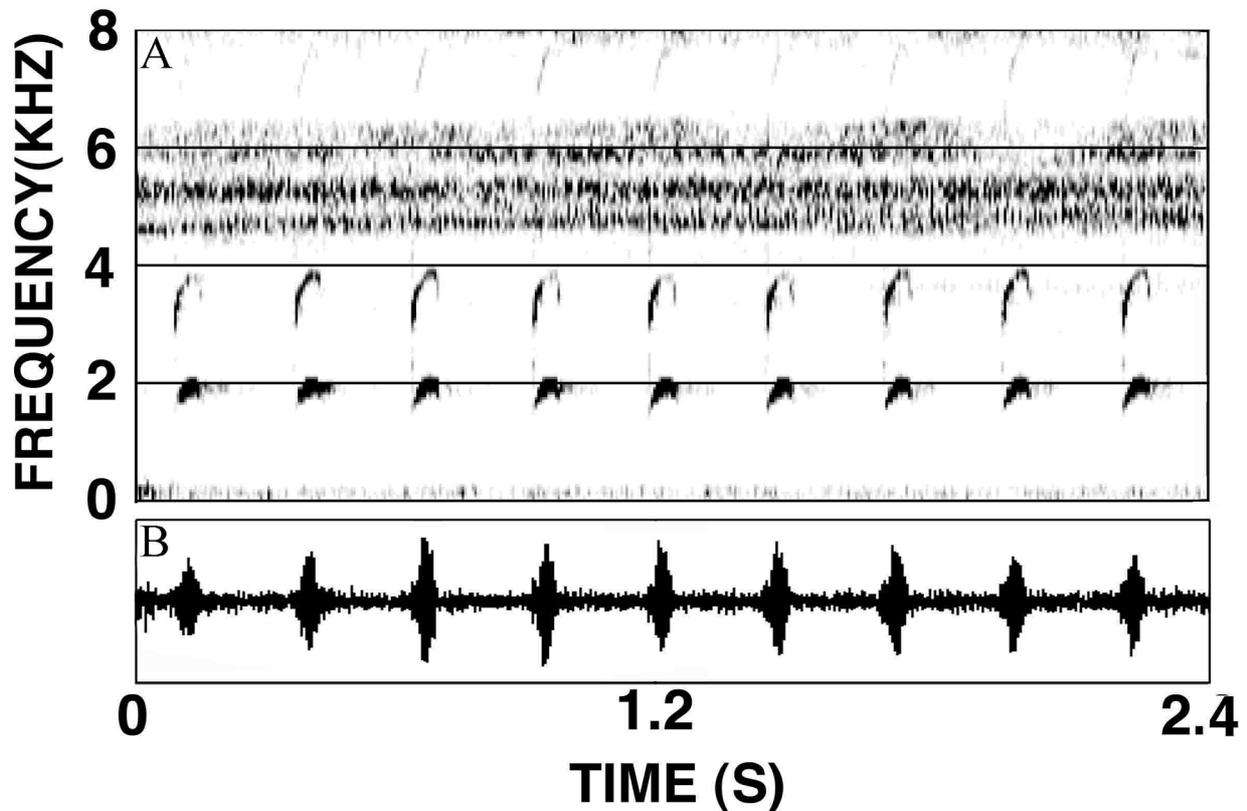


FIGURE 4. Sonagram (A) and wave form (B) showing a nine note portion of an advertisement call of *Kalophrynus limbooliati* **sp. nov.** from Gunung Lambak, Kluang, Johor, recorded at an air temperature 27.2 °C.

A ranid, *Hylarana laterimaculata* was found syntopic with *K. limbooliati* **sp. nov.** in Pulau and Lambak. They emitted long calls very similar to the human ear, but details clearly differ between the two species. Calls of *K. limbooliati* **sp. nov.** were longer than *H. laterimaculata* recorded at Lambak (mean±SD = 10.9±2.8 vs. 6.0±0.2 s in *H. laterimaculata*) and included larger number of notes (mean±SD = 41.1±11.2 vs. 12.5±0.6). Each note lasted shorter (mean±SD = 70.6±6.0 vs. 109.4±4.4 ms), with shorter time interval between two notes (mean±SD = 271.7± 25.4 vs. 520.3±18.2 ms) and larger note repetition rate (mean = 3.72±0.30 vs. 1.96 notes per s). In addition, the dominant frequency was evidently lower (mean±SD = 1909±156 vs. 2897±123 Hz) than that of *H. laterimaculata*.

Finally, the uncorrected pairwise sequence divergences of mitochondrial 16S rRNA gene between *K. limbooliati* and the seven congeners (*K. heterochirus*, *K. interlineatus*, *K. palmatissimus*, *K. pleurostigma*, *K. stellatus*, *K. subterrestris*, and *K. yongi*) are as large as 5.2–10.5%.

Discussion

It is highly probable that specimens of *K. limbooliati* have been historically identified as *K. pleurostigma* based on overall morphological similarity and the presence of black inguinal spot. It is likely that many records of *K. "pleurostigma"* in the Malay Peninsula (including states of Pahang, Selangor, Kelantan, Perak: Berry 1975), may also represent *K. limbooliati*. Dring (1979) noted that two female specimens of *K. "pleurostigma"* from Gunung Pulau and Janda Baik of Pahang (north of known distribution of *K. limbooliati*) were distinctly smaller than Bornean females. Because no particular remarks were made of toe web development in these females, Dring's (1979) *K. "pleurostigma"* seems to be not *K. palmatissimus*, which has characteristically well-developed toe webbing. Thus,

it is possible that these females, especially the specimen from Gunung Pulai (type locality of the new species) represent *K. limbooliati*.

Outside of Malaysia, *K. limbooliati* surely occurs in Singapore. A photograph of *K. "pleurostigma"* shown by Lim & Lim (1992: p. 40) very closely resembles the type series of *K. limbooliati* in body shape and coloration. The close geographical proximity of and absence of long-term biogeographical barriers between Singapore and known localities of *K. limbooliati* in southern Peninsula Malaysia supports this inference.

We heard calls of *K. limbooliati* in almost all lowland forest in southern Peninsula Malaysia that we visited but could not find eggs or larvae. Because there were no pools on the forest floors where males were calling, the species is suspected to have phytotelmic breeding preference. Specimens referred to *Kalophrynus "pleurostigma"* but which probably represent this new species, from Singapore have been reported to breed in cups of pitcher plant (Lim & Ng 1991; Lim & Lim 1992). We think the larvae from Singapore to be identical with the smaller larval form of *Kalophrynus* from the Malay Peninsula with a similar habitat (Berry 1972; Leong & Chou 1999; Matsui 2009). Thus, of the two larval types of *K. pleurostigma* reported from the Malay Peninsula (Berry 1972), the smaller form is most probably *K. limbooliati* and the larger form *K. pleurostigma*. The possibility that the larger form represents *K. palmatissimus* is very small because larvae of that species were reported to be even larger and inhabit in large pools (Kiew 1984). However, the population of *K. pleurostigma* from Peninsular Malaysia may be different from topotypic population from Sumatra, and further field surveys are needed to ascertain their taxonomic relationships.

As in many other parts of the world, the presence of previously unrecognized cryptic species has been revealed by distinctive call characteristics in many frog lineages in Southeast Asia (e.g. *Philautus mjobergi* Smith and *P. petersi* (Boulenger): Dring 1987; *Hylarana laterimaculata*: Leong *et al.* 2003; *Leptotalax melanoleucus* Matsui and *L. fuliginosus* Matsui: Matsui 2006). In the genus *Kalophrynus*, *K. interlineatus* has already been split from *K. pleurostigma* by the call difference (Matsui *et al.* 1996), and discovery of *K. limbooliati* by its unique call is the second instance in this genus to prove the importance of acoustic information in taxonomic studies. By this finding, the actual extent of so-believed wide ranging *K. pleurostigma* was further delimited. Further field surveys with recording calls will almost certainly uncover additional cryptic frog species and improve our understanding of many wide ranging species.

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