

# A new species of Indigryllus (Orthoptera, Gryllidae, Eneopterinae, Xenogryllini) from Kerala, India, with first data on acoustics and natural habitat

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Ranjana Jaiswara, S. Sreebin, M. Monaal, Tony Robillard. A new species of Indigryllus (Orthoptera, Gryllidae, Eneopterinae, Xenogryllini) from Kerala, India, with first data on acoustics and natural habitat. Zootaxa, 2022, 5205 (6), pp.532-546. 10.11646/zootaxa.5205.6.2 . hal-03851340

# HAL Id: hal-03851340 https://hal.science/hal-03851340

Submitted on 14 Nov 2022

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Short title- Indigryllus sagani, a new species from India

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### Abstract

In India, the cricket subfamily Eneopterinae is represented by two known genera, *Xenogryllus* Bolívar and *Indigryllus* Robillard & Jaiswara. The latter was recently described and is known from a single species *I. kudremu* Robillard & Jaiswara, 2019 from Karnataka, India. The species was known exclusively from its morphological features and there existed no information on its biology and acoustic features. The present study is a step ahead in understanding the natural history and acoustic signalling in the genus. Based on a collaboration initiated by citizen science observations on the website iNaturalist, we describe a new species, *Indigryllus sagani* **sp. nov.** from Kerala, India, with details on its habitat and acoustic signals.

Keywords: calling song, citizen science, courtship, taxonomy, Western Ghats

#### Introduction

The recent taxonomic revision of the cricket tribe Xenogryllini (Jaiswara et al. 2018a, b & 2019) has led to the clarification of all the taxa within this clade and the description of several new taxa, including several new species from Africa in the genera *Xenogryllus* Bolívar, 1890 and *Pseudolebinthus* Robillard, 2006 (Robillard, 2006; Jaiswara et al. 2018a, b; Salazar et al. 2020; Robillard & Murphy, 2020), and the new genus *Indigryllus* Robillard & Jaiswara, 2019 from India. The only species of the genus, *Indigryllus kudremu* Robillard & Jaiswara, 2019 was described based on three type specimens, from an unclear locality. However, no biological information exists on this species and genus, including about the calling song.

This study takes its origin based on a few preliminary observations of the specimens of the genus on the website iNaturalist by one of us (SS). The specimens were identified as such by TR and RJ who initiated a collaboration to study these crickets in further detail in the field and semi-captivity. We present here the results of this collaborative work and describe a new species of *Indigryllus* based on the collected specimens, photographs, and acoustic data. Detailed information on the new species' morphology, natural habitat, behavior and acoustic signals is provided.

#### Material and methods

**Specimen collection and preservation:** The specimens studied here were collected in May and June, 2021 from Koottappuna, Nileshwar (a municipality in Kasaragod, Kerala, India). Individuals were found to be active after sunset in a rubber plantation, between 8:00–10:00 PM. Crickets were photographed and video and audio recorded in the field, before being collected using a sweeping net. Most individuals were kept in captivity for further observation and recordings (mating behavior and acoustic signals) and were later preserved in absolute ethanol and brought to the Entomology lab in the Department of Zoology, Panjab University, Chandigarh for further studies.

**Morphological structures and measurements:** Genitalia of male and female specimens were dissected by making a short slit between the sternite and tergite of the last abdominal segments. Dissected genitalia were cleared in 10% cold KOH solution and preserved in glass vials containing glycerine. External and internal morphological structures were studied under a stereo zoom microscope, Leica S9i (Leica Microsystems, Germany). Measurements of type

series of *Indigryllus sagani* **sp. nov.** were done using the analysis tool of Leica Application Suite version LAS V4.13.0 installed on a computer running on Windows 10 Operating system.

**Imaging:** Images of external and internal morphological features were taken either using a 10megapixel camera attached to the Leica microscope or using Canon EOS 80D, a Digital SLR camera attached with a macro lens, Canon EF-100mm F2.8L Macro IS USM in the Department of Zoology, Panjab University, Chandigarh, India. Two external light sources connected with 5 LED bulbs from the studio light were used to attain soft lighting on the specimen. Scanning Electron microscope imaging of stridulatory teeth was done at 1–2 kV resolution using JSM-6100 (JEOL, Japan) in the Department of Sophisticated Analytical Instrumentation Facility, CIL and UCIM, Panjab University, Chandigarh, India.

Acoustic recording and analyses: The calling song of 10 males were recorded in the field at night or in captivity between 8:00 pm and 01:30 am at 96 kHz in a 16-bit format using a Tascam, Linear PCM Recorder (DR-05X MK II, TEAC Professional, USA) with its in-built microphone. Courtship songs were recorded in captivity, by placing a couple in the terrarium. Acoustic analyses were performed using the computer software Avisoft-SASLab Pro version 4.40 (Specht 2011). Acoustic features were measured using the automatic commands of Avisoft-SASLab Pro. A sample of the newly recorded files is deposited in the Sound Library of the MNHN (https://sonotheque.mnhn.fr/ inventory number MNHN-SO-2022-997).

**Terminologies:** Terminologies for genitalia follow Desutter (1987), modified in Desutter-Grandcolas (2003) and Robillard & Desutter-Grandcolas (2004a). Terminologies for male FW venation follow Ragge (1955) and Robillard & Desutter-Grandcolas (2004b). The basic cricket song terminology follows Ragge & Reynolds (1998) a basic song unit is called syllable and corresponds to one opening-closure cycle of the male forewings, and a group of syllables, named echeme, forms a call unit in the context of acoustic communication.

## Abbreviations:

#### General morphology

FI, FII, FIII, fore, median, hind femur; FW, forewing; TI, TII, TIII, fore, median, hind tibia; Tarsomere I/II/III-1: basal segment of fore, median and hind leg tarsomere.

## Tegminal venation

1A–4A, first to fourth anal veins; CuA, anterior cubitus; CuA1, CuA2: first, second branches of CuA; CuP, posterior cubitus; M, median vein; R, radial vein; Sc, subcostal vein and its branches; c1-3, first to third cells of C alignment; d1 cell (mirror), first cell(s) of D alignment; d2, second cell of D alignment; e1, first cell of E alignment.

### **Depositories**

ZSI: ZSI Zoological Survey of India, Kolkata, India. MNHN: Muséum national d'Histoire naturelle, Paris, France. RJINS: Personal collection of Ranjana Jaiswara, India.

## Results

## Systematic part

Orthoptera

Gryllidae

Eneopterinae Saussure, 1874

Xenogryllini Robillard, 2004

## Genus Indigryllus Robillard & Jaiswara, 2019

Type species: Indigryllus kudremu Robillard & Jaiswara, 2019

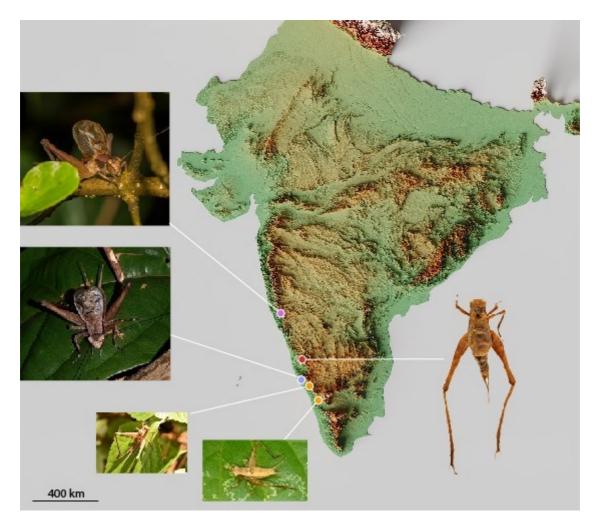
## Indigryllus sagani Jaiswara & Robillard sp. nov.

Figures 1-10

**Type material.** *Holotype*,  $\Diamond$ , **INDIA:** Kerala, Kasargod, Nileshwar, Koottappuna, ~19 m a.s.l., 06.VI.2021, 12°17'03.0"N 75°09'36.8"E, Sreebin S. leg., RJINS-163 (ZSI). *Allotype*–  $\heartsuit$ , RJINS-164, same locality as holotype (ZSI). *Paratypes* ( $6\Diamond$ ,  $3\heartsuit$ ), same information as holotype, V-VII.2021: 5 $\Diamond$  (RJINS-161, 165, 166, 171, 172);  $2\heartsuit$  (RJINS-169, 170);  $1\Diamond$ , ex-RJINS162 (MNHN-EO-ENSIF11149);  $1\heartsuit$ , ex-RJINS167 (MNHN-EO-ENSIF11150).

Additional specimens and observations. INDIA: Kerala, Kasargod, Nileshwar, Koottappuna, ~19 m a.s.l., 12°17'03.0"N 75°09'36.8"E, Sreebin, leg.: 1♀, on leaf, night, 28.V.2022, https://www.inaturalist.org/observations/119077595; 13, on leaf, night, 28.V.2022, https://www.inaturalist.org/observations/119077537; 1 juvenile, on leaf, night, 26.IV.2022, https://www.inaturalist.org/observations/113058320; 19, on soil, night, 6.VI.2021, https://www.inaturalist.org/observations/81891957; 12, on leaf, night, 29.V.2021, https://www.inaturalist.org/observations/80754108; 13, stridulating on shrub, night, 22.V.2021, https://www.inaturalist.org/observations/79899381; 1 juvenile, night, 9.V.2021, https://www.inaturalist.org/observations/78083841; 1 juvenile, on leaf, night, 2.V.2021, https://www.inaturalist.org/observations/76596447. Kerala, Thavinhal, 11.84881, 75.95213, 18, vegetation, 30.IV. 2017, M. Muzzatti on leg., https://www.inaturalist.org/observations/18431227. Kerala, 11.134528, 76.312889, 1 juvenile, 14XI.2020, Balakrishnan Peroth leg., https://www.inaturalist.org/observations/64936698.

Type locality. Koottappuna, Kasargod, Kerala, India.



**FIGURE 1.** Map showing the current distribution of *Indigryllus*: the red circle indicates the type locality of *I. kudremu*; the blue circle indicates the type locality of *Indigryllus sagani* **sp. nov.**; the orange circles indicate the localities corresponding to iNaturalist records of *Indigryllus* sp. for which no specimen could be studied tentatively identified as *I. sagani*; the pink circle indicates a iNaturalist record identified as *Indigryllus* sp which could belong to a new species. Map by Miguel Valenzuela, 2020, with permission (IG: @mappdoutofficial)

**Distribution.** In addition to type locality, the new species is known from two other districts of Kerala, i.e. Wayanad and Malappuram, based on observations from iNaturalist (see above, Fig. 1). The identification of these two specimens as *I. sagani* sp. nov. is tentative and will need confirmation based on specimen study.

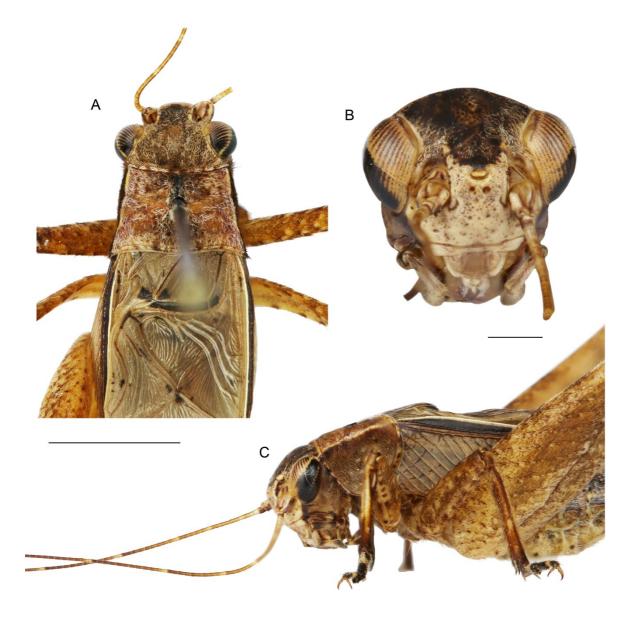
**Etymology**. The species is named after the American astronomer, Carl Edward Sagan in recognition of his significant contributions to scientific research on extra-terrestrial life.



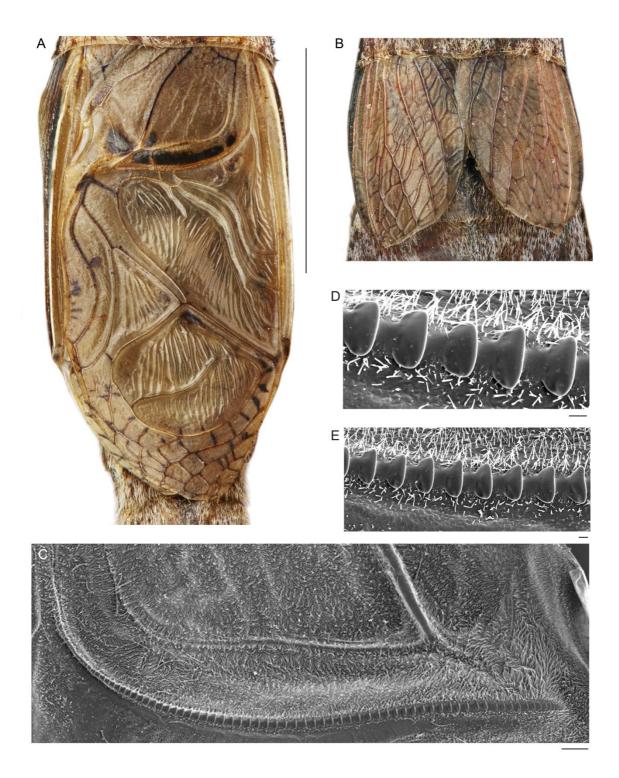
**FIGURE 2.** *Indigryllus sagani* **sp. nov.:** (A) dorsal view of male subgenital plate, (B) dorsal view of female subgenital plate and (C) dorsal view of male supra anal plate.

**Diagnosis.** *Indigryllus sagani* **sp. nov.** differs from *I. kudremu* by the male genitalia with pseudepiphallic lophi shorter and laterally winged, and by the following external characters: smaller body length; female subgenital plate posterior margin weak indentation, much wider than long (Fig. 2); 2A vein on male FWs forming obtuse angle; black patch between veins 1A and 2A extended more laterally towards plectrum.

**Description.** Body size average for the tribe. Most characters similar to *I. kudremu*, from which the new species differs as follows. Eyes slightly larger and more globular (Fig. 3B). Pronotum posterior margin slightly convex in the middle (Fig. 3A). TI with two inner and one outer apical spurs.

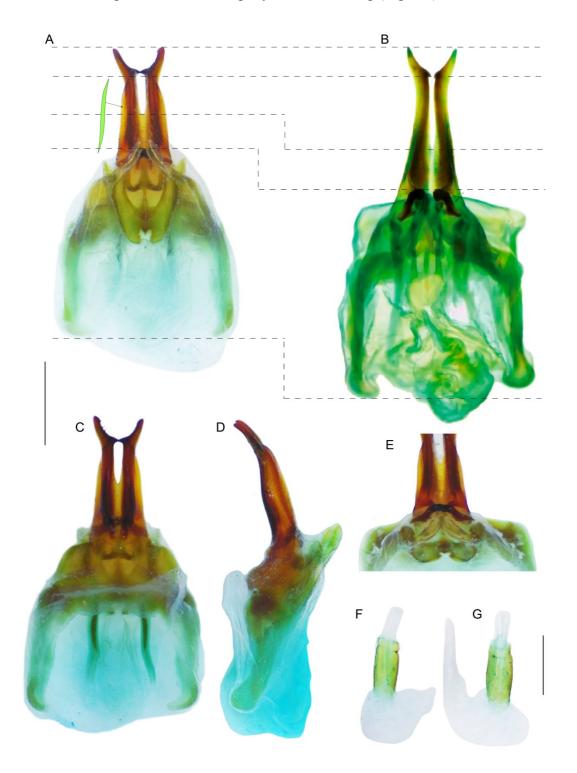


**FIGURE 3.** *Indigryllus sagani* **sp. nov.:** (A) dorsal view of male head and pronotum, (B) front view of face, (C) lateral view of male head and pronotum. Scale bars: A, C, 5 mm; B, 1 mm.



**FIGURE 4.** *Indigryllus sagani* **sp. nov.**: (A) male forewing in dorsal view, (B) female forewing in dorsal view, (C-E) scanning electron micrograph of the stridulatory teeth. Scale bars: A, B, 5 mm; C, 100 μm; D-E, 10 μm.

**Male.** FWs with somewhat parallel lateral margins (slightly widening posteriorly in *I. kudremu*). Vein 2A forming an obtuse angle; black patch between veins 1A-2A extended much more laterally towards plectrum; two harp veins (three occasionally); proximal part of first harp vein arising almost at 90° angle from CuA vein; cell d1 long, usually divided into two subcells (occasionally undivided or divided into three sub-cells); FW apex broader; apical field with 2-3 cell alignments. Mirror slightly wider than long (Fig. 4A).

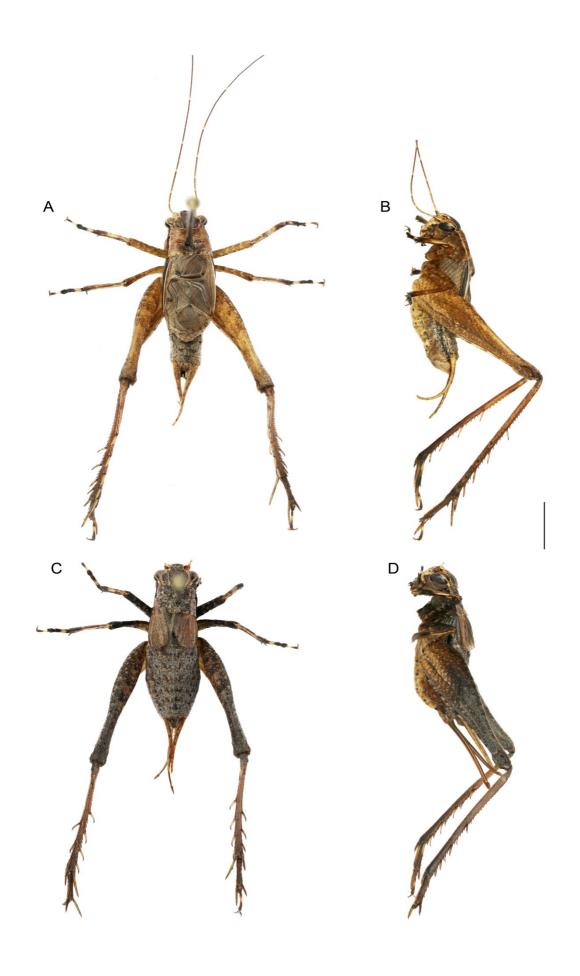


**FIGURE 5.** Genitalia of *Indigryllus sagani* **sp. nov**.: male genitalia in ventral (A), dorsal (C), lateral (D) and postero-ventral (E) views; comparison with *I. kudremu* in ventral view (B); female copulatory papilla of *Indigryllus sagani* **sp. nov.** in dorsal (F) and ventral (G) views. Scale bars: A-E, 1 mm; F-G, 0.5 mm.

**Male genitalia.** Pseudepiphallic lophi long, strongly sclerotized, bilaterally winged with less sclerotized structure, extending from anterior base upto 4/5<sup>th</sup> of posterior end; lophi apex with short and stout inner hook, converging towards each other, making U-shape appearance; pseudepiphallic parameres strongly sclerotized, short, slender, shallow bifurcation on posterior end, anterior end somewhat cone-shaped, sub-anteriorly short projection on inner side (Fig. 5A-E).

**Female.** FWs dorsal field with five strong longitudinal veins. 1A vein either bifurcated from 2/3<sup>rd</sup> or half of FW length. Lateral field triangular (Fig. 4B). FW apex broader compared to *I. kudremu*.

**Female genitalia.** Subgenital plate posterior margin weakly indented. Copulatory papilla with a basal cylindric sclerite and a narrower membranous apex (Fig. 5F-G).



**FIGURE 6.** *Indigryllus sagani* **sp. nov.**: male of light morph (A-B) and female of dark morph (C-D) in dorsal and lateral views, respectively. Scale bar: 5 mm.

**Coloration.** Individuals of this new species are found in two syntopic color morphs, a dark one (Figs 6C, 6D, 7, 8) and a paler one (Fig. 6A, 6B, 7, 8). The dark color morph is mainly dark gray brown, while the paler form is brown, with a median dark brown band along the whole body (Fig. 6A).



FIGURE 7. Indigryllus sagani sp. nov.: females of dark morph in their natural habitat.



**FIGURE 8.** *Indigryllus sagani* **sp. nov.:** male specimens of light morph (B, D) and dark morph (A, C) in their natural habitat; (B-D) showing wing positions while male is calling.

**Habitat.** Adults of *I. sagani* **sp. nov.** were seen during the months of May and June in Koottappuna, Nileshwar (a municipality in Kasaragod, Kerala), while juveniles were first observed in April. Individuals were found at low elevation, below 100 meters above sea level, adjacent to a rubber plantation with dense undergrowth. The region has dense vegetation (Figs 7 & 9), consisting of plant species such as *Camonea vitifolia, Ixora coccinea, Chromolaena adorata, Dioscorea bulbifera*, and over a hundred other species of wild plants (Figs 7, 8, 9).



FIGURE 9. Indigryllus sagani sp. nov.: natural habitat in type locality.

## Indigryllus sp.

(Fig. 1)

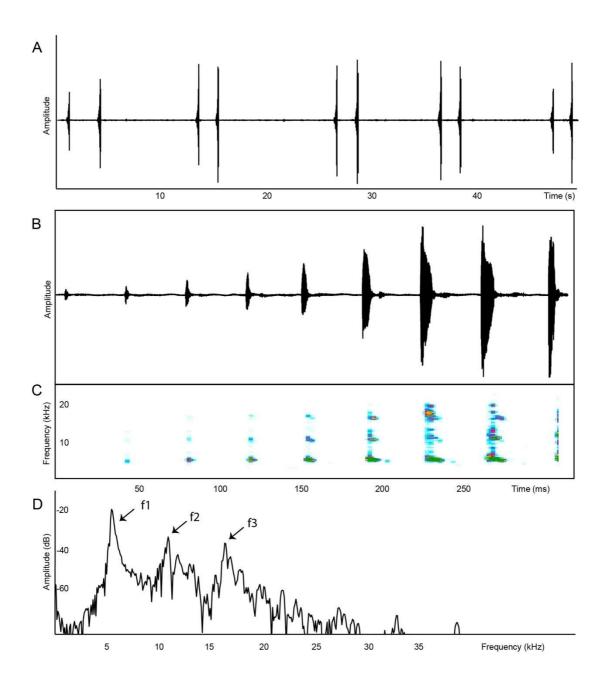
A male *Indigryllus* sp. was observed in Amboli in Pune, Maharastra, India based on the photographs uploaded on iNaturalist website (Fig. 1). Their details are as follows: INDIA, 1 adult male, Amboli, Maharashtra; 15.964716, 74.003571; June 2016; https://www.inaturalist.org/observations/57617074; observation by P. J. Ullal.

This record may correspond to a different species of *Indigrylllus*. Given the geographical complexity of the region and the subtle differences existing between *I. kudremu* and *I. sagani* **sp. nov.**, more information is necessary, including study of male genitalia.

**Table 1.** Measurements (in mm) of type specimens of *Indigryllus sagani* **sp. nov.** Abbreviations: BL, body length in dorsal view; FIIIL, length of FIII; FIIIW, width of FIII; TIIIL, length of TIII; FWL, forewing length; FWW, forewing width (at the level of maximal width at about 1/3 of FWL); Ias, inner spines on TIII dorsal side above the spurs; Ibs, inner spines on TIII dorsal side between the spurs; Oas, outer spines on TIII dorsal side above the spurs; Obs, outer spines on TIII dorsal side between the spurs; OL, ovipositor length; PronL, pronotum length; PronW, pronotum width; TaIIIs, spines of third hind tarsomere, not including the apical spines: Ids, inner dorsal spines; Ods, outer dorsal spines; Ols, outer spines on lateral side of TaIII.

	BL	PronL	PronW	FWL	FWW	FIIIL	FIIIW
Holotype Male	17.8	3.0	4.2	9.5	5.0	16.3	3.8
Male paratype RJINS-165	18.6	3.0	4.3	9.8	4.9	16.5	4.0
Male paratype RJINS-166	18.8	2.9	4.3	9.9	4.9	16.8	4.1
Allotype Female	17.9	3.2	4.4	3.8	2.9	16.1	4.5
Female paratype	16.3	3.0	4.3	4.0	2.6	15.5	4.3

	TIIIs				TallIs				
	TIIIL	Ias	Ibs	Oas	Obs	Ids	Ods	Ols	OL
Holotype Male	14.6	10	13	15	12	1	5	4	-
Male paratype RJINS-165	15.4	9	12	15	12	3	6	3	-
Male paratype RJINS-166	14.3	12	13	18	13	1	6	4	-
Allotype Female	13.8	10	12	16	11	1	7	5	8.2
Female paratype	12.9	8	11	14	11	1	5	4	8.4



**FIGURE 10.** Calling song of *Indigryllus sagani* **sp. nov.**: (A) oscillograms of 10 echemes grouped in doublets, (B) detailed oscillogram of one echeme and (C) corresponding spectrogram; (D) frequency spectrum showing harmonic frequencies.

## **Bioacoustics**

*Calling song.* Males of *Indigryllus sagani* **sp. nov.** were found calling from the top of low vegetation (Fig. 8B-D) between 7:00 pm – 4:00 am. At 30 °C, the calling song consists of a very short, relatively high-pitched echeme, widely spaced in time and irregularly arranged, sometimes grouped in doublets or triplets (Fig 10A), but not consistently across individuals. Echemes last for  $277 \pm 36$  ms and their irregular period ranges from ca. 2 to 10 s (mean= 5.3  $\pm$  3.2 s). Echemes are made of 6 to 9 indistinct syllables (mean=  $7.8 \pm 0.9$  syllables), although the first syllables are hardly distinguishable from the background noise and it lasts for approximately 1 ms. The next syllables increase progressively in amplitude and duration to become louder and lasts for ca. 7–10 ms. Average syllable duration in echeme is  $5.0 \pm 3.1$  ms, with an average period of  $38.2 \pm 2.4$  ms (Fig. 10A).

The frequency spectrum of the call shows significant variation within and between individuals. It consists of a noisy harmonic series with a dominant fundamental frequency of  $5.04 \pm 0.15$  kHz followed by more or less powerful harmonics 2 and 3 at ca. 10 and 15 kHz. In some individuals, the second harmonic frequency (f2) sometimes co-dominates the spectrum, although the dominant frequency is clearly the fundamental frequency. It is interesting to note that dominant frequency also tends to increase within echemes from initial to final syllables by ca. 200-400 Hz (Fig. 10C-D).

*Courtship song.* Courtship calls are produced by elevating the FWs to an approximate angle of  $45^{\circ}$  with respect to its abdomen. The courtship call is a shorter version of the calling song, lasts for  $147\pm28$  ms and includes 4-6 syllables (mean= 5.3), with a dominant frequency of  $4.3\pm0.15$  kHz. Courtship calls are usually accompanied by a tremulation of males.

### Discussion

The region of Kerala where the new *Indigryllus species* was discovered is rich in over 25 species of reptiles, 21 species of amphibians (including the critically endangered *Pseudophilatus amboli*), and over 700 species of insects were observed in this region. In terms of vegetation, the species *Camonea vitifolia* is particularly abundant in the places where *Indigryllus sagani* **sp. nov.** were observed.

The cricket genus *Indigryllus* was known previously from a single species, *I. kudremu*. The species was described from three specimens previously collected from Kudremukh, a locality

situated in Chikkamagaluru district of Karnataka, and there was no information on the ecology of the species. With the description of the new species *I. sagani* **sp. nov.**, the present study adds a new species to the genus collected from Kasargod district of Kerala, India, but also provides detailed information on the ecology and acoustic signals of the genus. Both Karnataka and Kerala, where the two species of the genus occur, are parts of the Western Ghats, one of the 34 biodiversity hotspots in the world. The southern part of the Western Ghats, that encompasses Karnataka and Kerala, is typically known for its tropical evergreen forests. Of the total forest cover of these areas, Karnataka bears 33% and Kerala 40% of open forests. These open forests have canopy cover between 10-40%, and *Indigryllus* seems to be abundant in these areas. In Koottappuna, the type locality of *I. sagani* **sp. nov.**, individuals were found calling from broad-leaved grasses, brambles and different shrub species present in the area, e.g. *Camonea vitifolia, Ixora coccinea, Chromolaena adorata, Dioscorea bulbifera*.

Interestingly, this study and the findings that it reports were initiated thanks to citizen science observations and photographs posted on the website iNaturalist. As previously demonstrated (Tan et al. 2021), for little studied insect groups, citizen science represents a rich source of knowledge which complements traditional methods for collecting data, and both rely on, and contribute to taxonomic studies.

In November 2021, a juvenile *I. sagani* photograph was uploaded on iNaturalist, which implies that eggs are being laid by adults somewhere between June-August and hatched out nymphs attain adulthood later in April. May-June might thus be the peak season of reproduction of the species, when males call actively from dusk to dawn. It is likely that most of the matings take place in this period. The peak calling season also coincides with the peak summer in Kerala, which is then followed by the monsoon. After the onset of monsoon, the calling activity reduces gradually.

As *I. kudremu* was never observed in living conditions, we lack information on its calling behavior to compare with that of the new species. However, the wide temporal window used by *I. sagani* seems similar to *Xenogryllus transversus*, a member of the sister genus of *Indigryllus*, which is also found in India. Like other members of the tribe Xenogryllini documented, males produce low-frequency calling songs. The males of *I. sagani* were also observed tremulating while producing the courtship call, which could be a behaviour shared by the whole tribe. Xenogryllini.

The genus *Indigryllus* is now known from two species, and photographs of *Indigryllus* uploaded on iNaturalist from two additional districts of Kerala, i.e. Waynad and Malappuram, suggest that it has a wider distribution in the state. *Indigryllus* has first been known from the southern Western Ghats, but a photograph on iNaturalist from Amboli, Maharashtra, suggest that they are also distributed in the northern parts as well. Examination of *Indigryllus* specimens from these additional locations is necessary in order to understand further the genus diversity and endemicity in India.

## Acknowledgements

We acknowledge the DST-INSPIRE Faculty scheme, Government of India for funding RJ to conduct this study. Thanks are also due to the Chairperson, Department of Zoology, Panjab University, Chandigarh, India, for providing infrastructural facilities. We thank Miguel Valenzuela (IG: @mappdoutofficial) for allowing us to use his map in Figure 1.

## Legend of figures

**FIGURE 1.** Map showing the current distribution of *Indigryllus*: the red circle indicates the type locality of *I. kudremu*; the blue circle indicates the type locality of *Indigryllus sagani* **sp. nov.**; the orange circles indicate the localities corresponding to iNaturalist records of *Indigryllus* sp. for which no specimen could be studied tentatively identified as *I. sagani*; the pink circle indicates a iNaturalist record identified as *Indigryllus* sp which could belong to a new species. Map by Miguel Valenzuela, 2020, with permission (IG: @mappdoutofficial)

**FIGURE 2.** *Indigryllus sagani* **sp. nov.:** (A) dorsal view of male subgenital plate, (B) dorsal view of female subgenital plate and (C) dorsal view of male supra anal plate.

**FIGURE 3.** *Indigryllus sagani* **sp. nov.:** (A) dorsal view of male head and pronotum, (B) front view of face, (C) lateral view of male head and pronotum. Scale bars: A, C, 5 mm; B, 1 mm.

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**FIGURE 6.** *Indigryllus sagani* **sp. nov.**: male of light morph (A-B) and female of dark morph (C-D) in dorsal and lateral views, respectively. Scale bar: 5 mm.

FIGURE 7. Indigryllus sagani sp. nov.: females of dark morph in their natural habitat.

**FIGURE 8.** *Indigryllus sagani* **sp. nov.**: male specimens of light morph (B,D) and dark morph (A,C) in their natural habitat; (B-D) showing wing positions while male is calling.

FIGURE 9. Indigryllus sagani sp. nov.: natural habitat in type locality.

**FIGURE 10.** Calling song of *Indigryllus sagani* **sp. nov.**: (A) oscillograms of 10 echemes grouped in doublets, (B) detailed oscillogram of one echeme and (C) corresponding spectrogram; (D) frequency spectrum showing harmonic frequencies.

**Table 1.** Measurements (in mm) of type specimens of *Indigryllus sagani* **sp. nov.** Abbreviations: BL, body length in dorsal view; FIIIL, length of FIII; FIIIW, width of FIII; TIIIL, length of TIII; FWL, forewing length; FWW, forewing width (at the level of maximal width at about 1/3 of FWL); Ias, inner spines on TIII dorsal side above the spurs; Ibs, inner spines on TIII dorsal side between the spurs; Oas, outer spines on TIII dorsal side above the spurs; Obs, outer spines on TIII dorsal side between the spurs; OL, ovipositor length; PronL, pronotum length; PronW, pronotum width; TaIIIs, spines of third hind tarsomere, not including the apical spines: Ids, inner dorsal spines; Ods, outer dorsal spines; Ols, outer spines on lateral side of TaIII.

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