

Two new species of *Raorchestes* (Anura: Rhacophoridae) from the Silent Valley National Park in the Nilgiri Hills of the Western Ghats, India

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Manuscript received: 6 August 2015

Accepted: 2 February 2016 by MICHAEL F. BAREJ

Abstract. Two new species of rhacophorid bush frogs of the genus *Raorchestes* are described from the tropical montane wet forests in the Silent Valley National Park in the Nilgiri Hills, a high horst in the Western Ghats, India. Both species can be differentiated from their congeners by morphological and bioacoustic characters as well as differences in the mitochondrial 16S gene. Advertisement calls of the two new species are provided and tentative insights into the phylogenetic position discussed. Despite recent revisions of this genus from the Western Ghats, and the fact that the Silent Valley National Park is one of the most important and well-surveyed protected areas, the results of this study highlight the overlooked diversity in this area.

Key words. Amphibia, bush frog, species description, South Asia, protected area, montane habitat, taxonomy.

Introduction

The rhacophorid genus *Raorchestes* BIJU, SHOUCHE, DUBOIS, DUTTA & BOSSUYT, 2010 is a species-rich assemblage of bush frogs, which are found from the southern tip of the Indian Peninsula to northeastern India, Indochina, and mainland Southeast Asia (BIJU et al. 2010, FROST 2015). *Raorchestes*, which has been shown to be the sister clade of the primarily Sri Lankan *Pseudophilautus* by BIJU et al. (2010), is most diverse in India's Western Ghats, a highland region in which many novel endemic species are still being discovered (DINESH et al. 2015). A total of 51 *Raorchestes* species are known from the Western Ghats, including numerous taxa that are restricted to isolated sky islands (e.g., BIJU & BOSSUYT 2009, BIJU et al. 2010, ZACHARIAH et al. 2011, SESHADRI et al. 2012, PADHYE et al. 2013, VIJAYAKUMAR et al. 2014). Concerning relationships in the speciose genus *Raorchestes*, several recent phylogenetic studies indi-

cate the presence of two prominent clades and several subclades within the genus (BIJU et al. 2010, ABRAHAM et al. 2013, VIJAYAKUMAR et al. 2014). VIJAYAKUMAR et al. (2014) called one of the two prominent clades Clade 'N', which comprises species that are distributed predominately to the north of a prominent geological gap called the Palakkad Gap, while the other clade is composed of species primarily distributed to the south of this gap.

A recent expedition to the higher altitudes of the Silent Valley National Park in the Nilgiri Hills of the Western Ghats provided opportunities to observe and identify two novel taxa of *Raorchestes* in addition to two species that were previously described from the area (*R. signatus* and *R. tinniens*). While *R. signatus* inhabits forest habitats, *R. tinniens* is known from grasslands in the area of interest. Of the two novelties, one is a green, semi-arboreal taxon and the other a brown, ground-dwelling leaf litter taxon. Herein, we compare these two taxa to closely related and

sympatric *Raorchestes* species, and describe them as new to science.

Materials and methods

Study area

The Western Ghats form a long north–south hill range straddling the west coast of peninsular India, with a conspicuous gap that divides the range into two prominent parts. This geological gap, called the Palakkad Gap, has been recognized as a prominent biogeographical barrier for amphibians (VAN BOCXLAER et al. 2012), fishes (JOHN et al. 2013), mammals (RAM et al. 2015), and birds (ROBIN et al. 2015). The section to the north of the gap is technically the escarpment of a vast plateau, with several hill clusters and horsts dotting the western margin of the Ghats (NAIR 1991). The highest and most prominent amongst these is the Nilgiri Hills, a broad massif with an average altitude of 1,800 m a.s.l. with a western portion that rises to an average of 2,200 m a.s.l. (CANER & BOURGEON 2001). From this high western margin, the Nilgiri Hills descend steeply into the Malabar coastal plains (Fig. 1).

During the course of three field trips (May 2012, June 2013, May 2014) to the southwestern slopes of the Nilgiri Hills, which is part of the Silent Valley National park, two *Raorchestes* that were previously unknown from this area were observed and collected that could not be assigned to any described species in the genus. Morphological data were recorded from 2 specimens for each species; they were euthanised, fixed in 10% formalin, and later transferred to 70% ethanol. Measurements (rounded to 0.1 mm) and terminology follow those of ZACHARIAH et al. (2011) for adult frogs. These voucher specimens were deposited in the Natural History Museum, Trivandrum (TNHM), India.

Bioacoustics

Advertisement calls (vocalisations) were recorded with a TASCAM™ HDP2 portable stereo recorder in uncompressed WAV format at a sampling rate of 44.1 kHz and 16 bit resolution. Call parameters were measured using the R package of SEEWAVE (SUEUR et al. 2008, R Development Core Team 2015) and Raven® Pro 1.5 (Bioacoustics Research Group, Cornell Lab of Ornithology 2012). Call

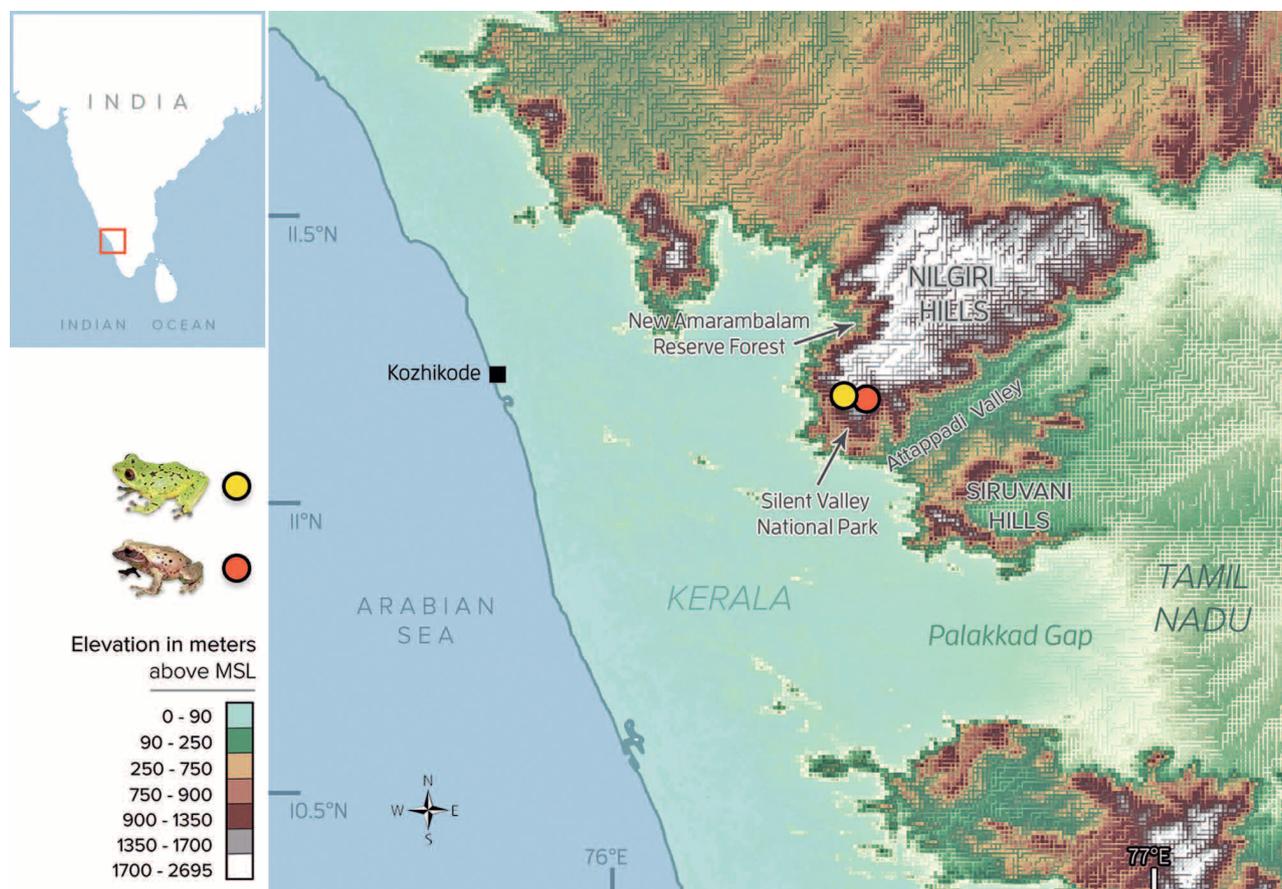


Figure 1. Map showing the type localities of *Raorchestes silentvalley* sp. n. (yellow dot) and *R. lechiya* sp. n. (red dot) in the Silent Valley National Park, Kerala, India.

analyses were standardized at a Fast Fourier transform (FFT) frame length of 512 Hz, a time-grid overlap of 99%, and Hann window type. A call is defined as the entire assemblage of acoustic signals produced in a given sequence (DUELLMAN & TRUEB 1994). A series of pulses (or notes) is defined as a call when pulses form temporally distinct segments that are separated by a return to the background noise between each note (HUTTER et al. 2015). Ambient temperature at the time of the recording was noted.

Molecular analysis

For molecular analyses, we extracted DNA from muscle tissue of four individuals of the two novel *Raorchestes* taxa with a Qiagen DNeasy kit. A ~570-bp segment of mitochondrial DNA, corresponding to the ribosomal subunit (16S rRNA) (FROST et al. 2006), was amplified through a standard polymerase chain reaction (PCR) (PALUMBI 1996). The sequences were deposited in GenBank under the accession numbers KT359622, KT359623, KT359628 & KT359629. The combined sequence data from GenBank (see Table 1; with the exception of *R. thodai* due to lack of sequences) were aligned with our sequencing outcomes using the default parameters of the MUSCLE algorithm (EDGAR 2004). Uncorrected p-distances for the partial 16S rRNA gene between included *Raorchestes* species were calculated with PAUP* 4.0 (SWOFFORD 2002) and visualized as a heatmap using R (for individual values, see Appendix).

To obtain preliminary insights into the phylogenetic position (tree not shown), we used the same *Raorchestes* dataset of our p-distance analysis and included *Pseudophilautus wynaadensis* as the outgroup taxon. A Maximum Likelihood (ML) tree was calculated using RAxMLv7.2.8 (STAMATAKIS 2006; substitution model: GTR + G; 200 independent best-tree searches; rapid-bootstrapping algorithm with 500 replicates). Calculations were based on an alignment of 542 bp after exclusion of ambiguous variable and hypervariable sections of the analysed gene fragment (1–7, 20–22, 54, 257–269, 563–570).

Morphological analysis

The following measurements were taken with a digital caliper (to the nearest 0.1 mm): snout–vent length (SVL, from tip of snout to vent); greatest head width (HW, at angles of jaws); head length (HL, from posterior edge of jaw to tip of snout); interorbital width (IUE, shortest distance between upper eyelids); upper eyelid width (UEW, greatest width of the upper eyelid); snout length (SL, from anterior border of eye to tip of snout); eye diameter (EL, horizontal distance between orbital margins of eye); forelimb length (FLL, from tip of elbow to proximal edge of palmar tubercle); hand length (HaL, distance from proximal edge of palmar tubercle to tip of third finger); thigh length (TL, from vent to external edge of knee); shank length (ShL,

from external edge of the knee to tip of heel); tarsus length (TaL, distance from heel to proximal edge of inner metatarsal tubercle); foot length (FoL, from proximal edge of inner metatarsal tubercle to tip of fourth toe). All measurements are given in millimetres. Because it has been shown that anurans have lost the first (medial) finger of the hand, the remaining four digits are numbered II through V, in contrast to the traditional I through IV (SCHUBIN & ALBERCH 1986).

Nomenclatural acts

The electronic edition of this article conforms to the requirements of the amended International Code of Zoological Nomenclature, and hence the new names contained herein are available under that Code of this article. This published work and the nomenclatural acts it contains have been registered in ZooBank, the online registration system for the ICZN. The LSID (Life Science Identifier) for this publication is: urn:lsid:zoobank.org:pub:1A96A10F-D5DC-46CA-B5F5-7FF4EE3A265D. The electronic edition of this work was published in a journal with an ISSN, has been archived, and is available from the following digital repository: www.salamandra-journal.com.

Results

In our exploratory phylogeny (results not shown), we find that both new species described here belong to the genus *Raorchestes*, which is diagnosed solely by its unique molecular phylogenetic position within Rhacophoridae (BIJU et al. 2010, ABRAHAM et al. 2013). Within *Raorchestes*, both species belong to the same major clade called Clade ‘N’ sensu VIJAYAKUMAR et al. (2014). This is corroborated by the results of the uncorrected p-distances calculated from the mitochondrial alignment, which demonstrates that species with highest genetic similarity to both novel taxa described here occur north of the Palakkad Gap (Fig. 2; Table 1). Consequently, we describe both taxa using morphological, molecular, and bioacoustics means and comment on their distribution and ecology.

Raorchestes silentvalley sp. n.

(Fig. 3, 4; Table 2)

ZooBank LSID: urn:lsid:zoobank.org:act:70D9E84F-C833-429D-B005-C51830931E15

Suggested common name: Silent Valley bush frog

Holotype: TNHM (H) 12.6.18/53 (Figs 3A, 4); adult male (27.1 SVL); collected from forest vegetation of 2 m in height at Sispara (11°12'7.34" N, 76°26'26.57" E) at 2,021 m a.s.l., Silent Valley National Park, Palakkad District, Kerala, by ANIL ZACHARIAH on 03 May 2012.

Table 1. Genetic similarity of *Raorchestes silentvalley* sp. n. and *R. lechiya* sp. n. with 51 congeners. Given are minimum (Min), maximum (Max), and mean (Mean) values of uncorrected pairwise sequence divergence, presented in % of ~570 bp of the 16S rRNA gene; corresponding GenBank accession numbers are provided alongside the species names, and species marked with an “*” belong to the Clade ‘N’; the species marked with a “+” is found outside of the Western Ghats. The analysed fragments of the 16S gene in *R. silentvalley* sp. n. (KT359628 & KT359629) were identical (uncorr. p-distance 0%) and almost identical (0.21%) for *R. lechiya* sp. n. (KT359622 & KT359623). See Appendix for uncorrected pairwise distance values (%) between all species.

No.	Species	<i>R. silentvalley</i> sp. n.			<i>R. lechiya</i> sp. n.		
		Mean	Min	Max	Mean	Min	Max
1	<i>R. charius</i> (JX092691)*	7.45	7.36	7.55	7.15	6.98	7.31
2	<i>R. griet</i> (JX092654)*	8.10	8.01	8.19	8.51	8.40	8.62
3	<i>R. coonoorensis</i> (JX092716)*	8.15	8.06	8.24	9.11	8.90	9.32
4	<i>R. ravii</i> (JX092720)*	7.96	7.86	8.05	8.26	8.06	8.45
5	<i>R. nerostagona</i> (JX092661)	8.62	8.62	8.63	9.21	8.85	9.57
6	<i>R. blandus</i> (JX092660)	9.98	9.89	10.08	9.54	9.25	9.83
7	<i>R. archeos</i> (JX092675)	8.88	8.87	8.89	10.09	9.72	10.46
8	<i>R. chotta</i> (JX092656)	8.22	8.21	8.23	8.82	8.61	9.02
9	<i>R. echinatus</i> (JX092696)*	5.86	5.86	5.86	6.04	5.77	6.30
10	<i>R. flaviventris</i> (JX092694)*	7.65	7.56	7.75	7.74	7.50	7.98
11	<i>R. hassanensis</i> (JX092673)*	8.23	8.14	8.33	7.91	7.67	8.16
12	<i>R. ponmudi</i> (JX092651)*	6.59	6.50	6.69	5.79	5.50	6.09
13	<i>R. bobingeri</i> (JX092680)	6.85	6.75	6.95	7.80	7.77	7.83
14	<i>R. glandulosus</i> (JX092665)	7.37	7.36	7.38	7.83	7.78	7.88
15	<i>R. jayarami</i> (JX092686)	7.07	6.97	7.16	8.00	7.78	8.22
16	<i>R. akroparallagi</i> (JX092650)	6.31	6.30	6.31	7.91	7.38	8.45
17	<i>R. ghatei</i> (JX092687)*	7.85	7.76	7.95	7.27	7.01	7.53
18	<i>R. leucolatus</i> (JX092669)*	8.00	7.99	8.01	8.40	8.39	8.40
19	<i>R. tuberothumerus</i> (JX092697)*	9.10	9.00	9.19	7.13	6.86	7.41
20	<i>R. bombayensis</i> (JX092657)*	8.75	8.65	8.84	7.26	6.99	7.54
21	<i>R. terebans</i> (JX092689)**	8.85	8.75	8.95	8.01	7.88	8.14
22	<i>R. luteolus</i> (JX092659)	5.42	5.42	5.43	6.70	6.21	7.19
23	<i>R. travancoricus</i> (JX092721)	6.49	6.48	6.50	6.66	6.18	7.14
24	<i>R. marki</i> (JX092719)*	5.28	5.18	5.37	4.74	4.66	4.83
25	<i>R. chromasynchysi</i> (JX092709)*	3.46	3.46	3.47	5.37	5.30	5.44
26	<i>R. indigo</i> (JX092678)*	5.46	5.37	5.56	4.08	4.00	4.17
27	<i>R. agasthyaensis</i> (JX092723)	5.65	5.65	5.66	6.03	5.76	6.31
28	<i>R. kadalarensis</i> (JX092702)	5.45	5.44	5.45	6.52	6.25	6.78
29	<i>R. aureus</i> (JX092672)*	4.90	4.80	4.99	2.39	2.22	2.55
30	<i>R. signatus</i> (JX092710)*	3.26	3.26	3.27	2.79	2.66	2.93
31	<i>R. tinniensi</i> (JX092715)*	4.11	4.11	4.12	3.55	3.55	3.56
32	<i>R. primarrumpfi</i> (JX092717)*	3.15	3.05	3.25	3.22	3.10	3.34
33	<i>R. montanus</i> (JX092690)*	3.45	3.44	3.46	3.12	3.10	3.14
34	<i>R. crustai</i> (JX092677)	7.33	7.32	7.34	7.42	7.08	7.75
35	<i>R. gramminirupes</i> (JX092649)	5.63	5.62	5.64	5.06	4.65	5.47
36	<i>R. johnceei</i> (JX092679)	4.62	4.61	4.63	4.50	4.09	4.90
37	<i>R. sushili</i> (JX092684)	6.32	6.22	6.42	7.06	6.79	7.32
38	<i>R. manohari</i> (JX092674)	5.24	5.14	5.34	6.27	6.23	6.32
39	<i>R. uthamani</i> (JX092722)	5.34	5.24	5.43	6.15	6.11	6.19
40	<i>R. chlorosomma</i> (JX092681)	5.18	5.09	5.28	6.43	6.38	6.49
41	<i>R. flavivularis</i> (JX092682)	4.30	4.29	4.30	5.54	5.25	5.82
42	<i>R. chalazodes</i> (JX092676)	5.45	5.35	5.54	6.36	6.31	6.42
43	<i>R. ochlandrae</i> (JX092666)	5.38	5.28	5.48	6.16	6.13	6.19
44	<i>R. kaikatti</i> (JX092718)	4.44	4.35	4.54	5.60	5.53	5.68
45	<i>R. kakachi</i> (JX092704)	5.21	5.20	5.22	5.82	5.54	6.10
46	<i>R. beddomii</i> (JX092653)	5.33	5.23	5.43	5.31	5.30	5.32
47	<i>R. theuerkaufi</i> (JX092693)	6.88	6.78	6.98	6.64	6.59	6.69
48	<i>R. munnarensis</i> (JX092655)	5.42	5.32	5.52	6.34	6.27	6.41
49	<i>R. resplendens</i> (JX092683)	5.04	4.94	5.13	5.88	5.85	5.91
50	<i>R. anili</i> (JX092708)	4.69	4.59	4.79	5.75	5.57	5.93
51	<i>R. dubois</i> (JX092668)	3.80	3.70	3.89	4.84	4.84	4.85

Paratype: TNHM (H) 12.6.18/54 (Fig. 3E); adult male (26.8 SVL), collected from foliage 1.5 m above the ground at the type locality by ROBIN KURIAN ABRAHAM on 03 May 2012.

Diagnosis: The genetic data (16S rRNA) identifies the new species as a member of the genus *Raorchestes*. *Raorchestes silentvalley* sp. n. differs from all known congeners by the following combination of characters: a medium-sized (SVL 26.9, N = 2) green bush frog with shagreened dorsal skin covered with small, horny, spiculate projections; bright yellow, granular ventral skin with large purplish black blotches interspersed towards the posteroventer and thighs; tympanum indistinct, supratympanic fold faint; palpebral membrane with pale green spots and blotches; palmar and plantar surfaces shagreened; fingers without webbing, toes half webbed; distinct advertisement call.

Description of holotype: An adult male of SVL 27.1 (Fig. 4); head wider (11.3) than long (9.8); snout rounded in ventral aspect, acuminate in lateral profile (Fig. 4C); snout length (4.0) slightly greater than diameter of the eye (3.8); canthus rostralis sharp and obtuse, loreal region flat; interor-

bit distance (4.8) greater than upper eyelid width (2.8); tympanum indistinct; supratympanic fold weak, extending from posterior margin of outer canthus to shoulder region; lanceolate tongue lacking lingual papilla, with small, hard region toward base; vomerine teeth absent; pupil horizontal; palpebral membrane ornamented with pale green spots (Fig. 4C); vocal sac single, median, and subgular.

Forelimb (6.2) shorter than hand (7.9); Fingers III, IV, and V with lateral dermal fringes; subarticular tubercles prominent, round, and single; supernumerary tubercles present on fingers III, IV, and V; inner palmar tubercles prominent, oval; discs well developed, expanded, and broadened; relative lengths of fingers II < III < V < IV; webbing between fingers absent.

Hind limb long, shank (14.3) subequal to thigh length (14.74); toes half webbed (Fig. 4D), extending from disc of toe V to distal subarticular tubercle of toe IV; lateral dermal fringes present on all toes; subarticular tubercles well developed, round, and single on all toes; supernumerary tubercles present but indistinct; inner metatarsal tubercle prominent, oval; outer metatarsal tubercle indistinct; relative lengths of toes I < II < III < V < IV.

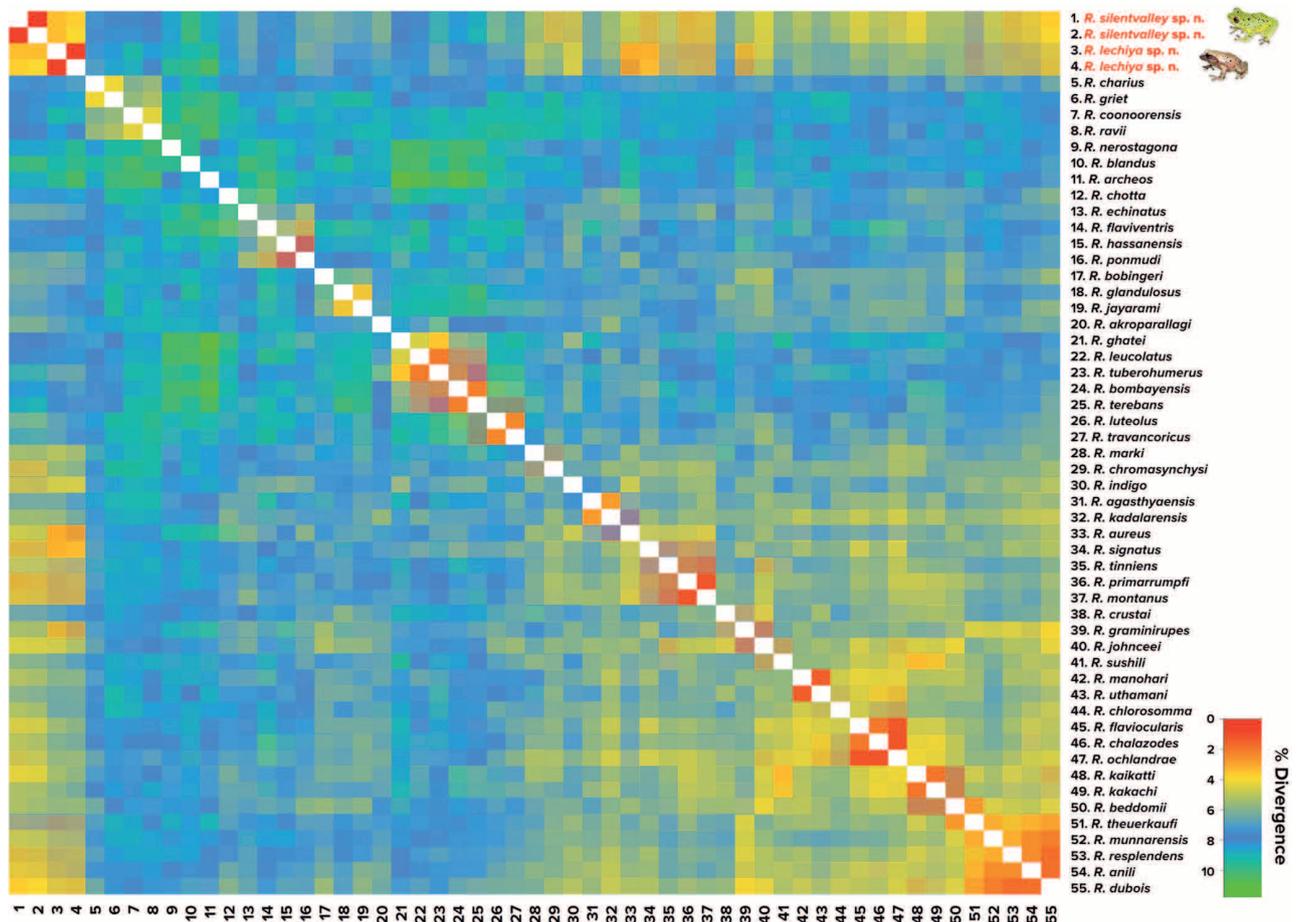


Figure 2. Heatmap of pairwise uncorrected p-distances of the 16S rRNA gene of *R. silentvalley* sp. n., *R. lechiya* sp. n., and 51 *Raorchestes* species from peninsular India. The colours red to green indicate low to high divergences. For respective Genbank accession numbers, see Table 1.

Table 2. Morphometric measurements (mm) of type specimens (all males). For abbreviations, see Materials and methods.

	SVL	HW	HL	IUE	UEW	SL	EL	FLL	HaL	TL	ShL	TaL	FoL
<i>Raorchestes silentvalley</i> sp. n.													
Holotype													
TNHM (H) 12.6.18/53	27.1	11.3	9.8	4.8	2.9	4.0	3.8	6.2	7.9	14.7	14.3	7.6	12.1
Paratype													
TNHM (H) 12.6.18/54	26.8	11.6	9.2	4.0	2.3	4.4	4.2	6.4	7.6	14.8	14.2	7.1	10.8
Mean	27.0	11.4	9.5	4.4	2.6	4.2	4.0	6.3	7.8	14.8	14.3	7.4	11.5
<i>Raorchestes lechiya</i> sp. n.													
Holotype													
TNHM (H) 12.6.18/55	19.8	8.4	8.2	3.2	2.1	3.5	3.3	4.3	5.6	11.1	11.2	6.3	7.2
Paratype													
TNHM (H) 12.6.18/56	21.1	8.6	7.3	3.3	1.7	3.7	3.2	5.3	5.0	10.3	10.6	6.3	7.7
Mean	20.5	8.5	7.6	3.2	1.9	3.6	3.3	4.8	5.3	10.7	10.9	6.3	7.5

Skin of dorsum strongly shagreened with small, horny spicules, and glandular projections that are more prominent towards the posterior and lateral sides; venter granular; dorsal surface of forelimbs and hind limbs shagreened; planar and plantar surfaces uniformly shagreened (Figs 4D, E). Small incision in the posterior surface of the right thigh made for tissue sample.

Coloration of holotype in life: Dorsum uniformly bright green with yellow-tipped glandular projections (Fig. 3A); venter bright yellow with irregular dark purplish and black patches along axillary margin of shoulder and on belly and groins, where the blotches are broader; small yellow spots or blotches interspersed in the broader purplish black patches in the posterior parts of the venter; anterior region

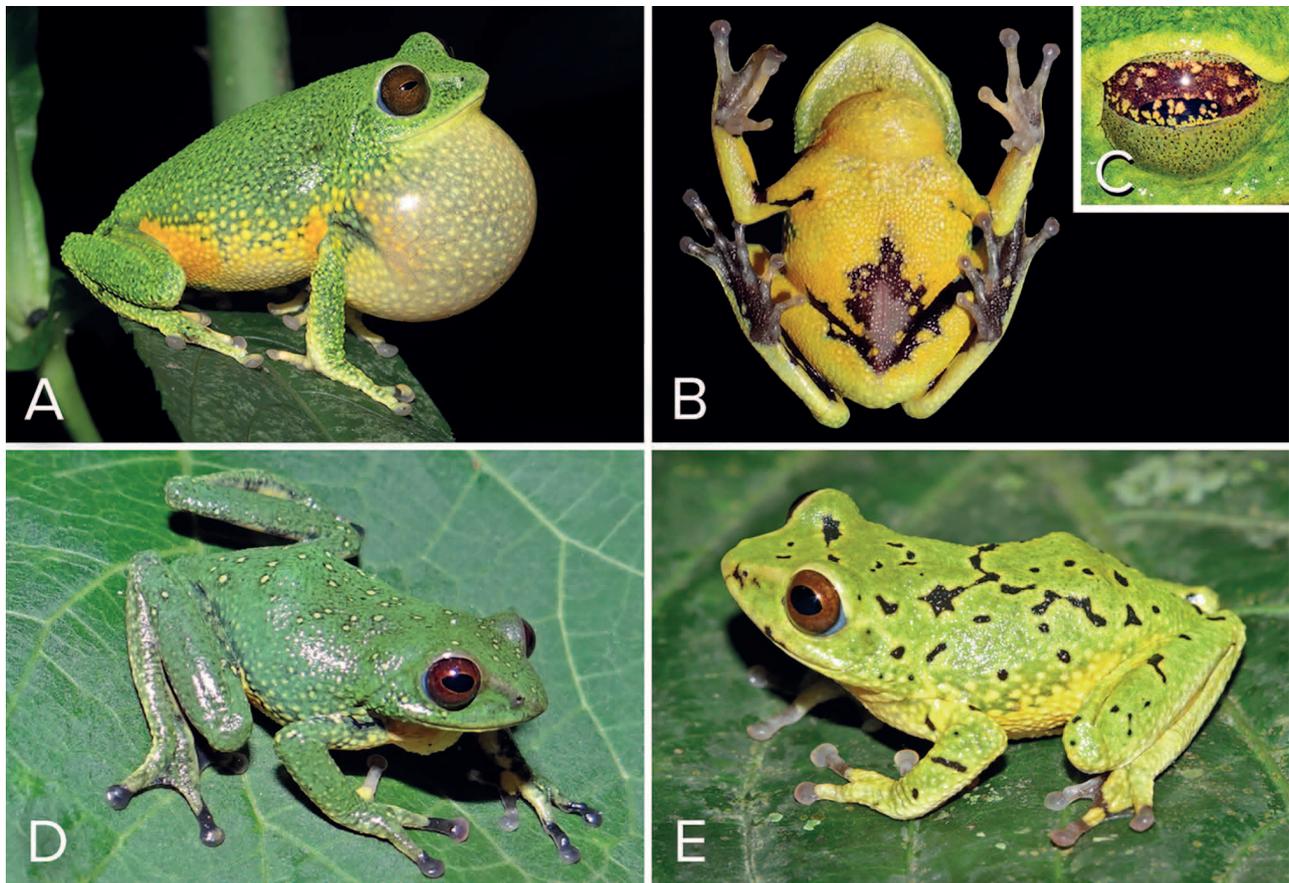


Figure 3. *Raorchestes silentvalley* sp. n. (in life): A) vocalizing male; B) ventral coloration of a live individual (Paratype); C) spotted pattern on palpebral membrane; D) and E) dorsal colour variation in other male individuals.

of thigh, posterior region of tibia, plantar surface grey to purplish black; outer faces of limbs green, inner faces yellow; discs and distal phalanges of hands and feet purplish grey; iris dark brown to cherry-red; palpebral membrane with pale green spots and blotches; vocal sac yellow; ventral surface of head greenish yellow.

Coloration of holotype in preservative (after three years of preservation): Dorsum bluish grey (Fig. 4A); venter off-white with irregular grey to black patches on belly and tibia (Fig. 4B); forelimbs grey to black above, fingers II and III off-white; hands and feet greyish black; flanks off-white; groin and posterior parts of thighs black.

Differential diagnosis: In general appearance, the green coloration of *Raorchestes silentvalley* sp. n. differentiates it from all closely allied species as demonstrated in our p-distance analysis (Fig. 2) such as *R. primarrumpfi*, *R. montanus*, and *R. signatus*, all of which show variable shades of brown coloration. It could however be confused with some other green *Raorchestes*, but can be differentiated as follows: It is distinguished from *R. beddomii* by hav-

ing contrasting purplish black blotches on a bright yellow venter (vs. whitish venter and yellow throat) and having a spiculate (vs. smooth) dorsum. It differs from male *R. chromasynchysi*, in having deep red eyes with green spots and blotches on palpebral membrane (vs. dark golden-brown eyes with a uniform transparent palpebral membrane) and in the shagreen (vs. smooth) plantar surfaces. *Raorchestes silentvalley* sp. n. is distinguished by its larger male size (mean SVL 27.0 mm, N = 2) from *R. bobingeri* (mean SVL 22.5 mm), *R. glandulosus* (mean SVL 25.2 mm) and *R. akroparallagi* (mean SVL 20.7 mm); uniform green colour in the loreal region and on dorsal surface of limbs (vs. yellow loreal region in *R. glandulosus*; yellow or orange colour on upper arms and posterior region of limbs in *R. bobingeri* and *R. jayarami*), horny spiculate dorsal skin (vs. smoothly shagreened dorsal skin in *R. bobingeri*, *R. jayarami*, *R. glandulosus*, and *R. akroparallagi*); half-webbed toes (vs. basally webbed in *R. akroparallagi*); and green blotches on palpebral membrane (vs. uniform transparent palpebral membrane in *R. bobingeri*, *R. glandulosus* and *R. akroparallagi*). *Raorchestes silentvalley* sp. n. dif-

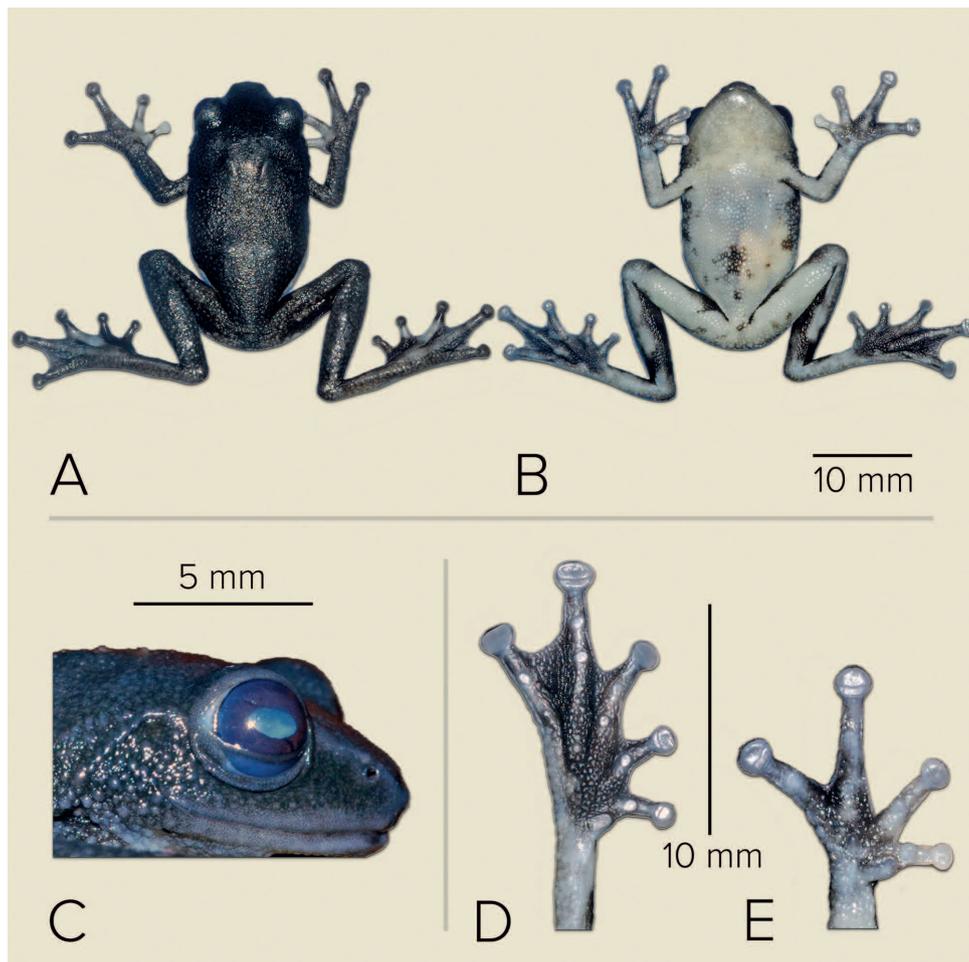


Figure 4. Holotype of *Raorchestes silentvalley* sp. n. (TNHM (H) 12.6.18/53): A) dorsal view; B) ventral view; C) lateral view of head; D) ventral view of right foot; E) ventral view of right hand.

fers from *R. flaviventris* by its smaller size (vs. large size of 36.5–50.5 mm), green blotches on palpebral membrane (vs. uniform palpebral membrane), absence of lingual papilla on tongue (vs. presence), and half-webbed toes (vs. basally webbed). *Raorchestes silentvalley* sp. n. can be distinguished from *R. indigo* by the presence of green blotches on palpebral membrane (vs. uniform palpebral membrane), and dorsal portions of fingers and toes grey (vs. dorsal faces of fingers and toes green). It differs from *R. tinniens* by being green with purplish grey phalanges (vs. brownish, with brown distal phalanges except for fingers II and III, which are yellow). *Raorchestes silentvalley* sp. n. can also be distinguished from the sympatric *R. signatus* primarily by its much smaller adult male size (vs. average adult male SVL > 30 mm), and in being a green frog (vs. variably mottled ranging from yellow to brown and red).

Variation: The colour pattern of *Raorchestes silentvalley* sp. n. varies markedly. This is apparent in the type specimens, and field observations of these anurans revealed variations of green colour across individuals. The holotype was uniform green dorsally in life. In contrast, the dorsum of the paratype (Fig. 3E) was bright green with black blotches and spots; some of the spots and blotches were connected in a reticulated pattern, and each blotch had a paler border. These markings extended onto the limbs as well. The dorsal face of a third individual (Fig. 3D) was darker green with pale green spots, and each spot had a dark border; the flanks were immaculate. The ventral sides of all individuals were yellow and bore greyish or purplish black blotches or streaks.

Etymology: The specific epithet *silentvalley* refers to the Silent Valley National Park in Kerala, India, where the species was observed and collected.

Distribution and Natural History: Thus far, *Raorchestes silentvalley* sp. n. has been recorded only from the montane wet forests (Figs 6A, B) of the southwestern slopes of the

Nilgiri Hills (Fig. 1). The species was observed at Sispara and the higher areas of Thudukki, both of which are situated in the Silent Valley National Park in Kerala at altitudes ranging between 1,850 and 2,200 m a.s.l. Vocalising individuals were perched 1–2.5 m above the forest floor on the branches and foliage of understorey shrubs (dominated by *Strobilanthes lawsonii* and occasionally by *S. lanata*) (Fig. 6C) and small tree saplings, in May. Several vocalising males were observed with blood-sucking leeches attached to them.

Genetics: The mean genetic difference in the sampled region of the 16S rRNA gene between *Raorchestes silentvalley* sp. n. and 52 Western Ghats congeners ranged from 3.15–9.98% (Table 1). The uncorrected pairwise sequence differences to the genetically most similar species were as follows: *R. primarrumpfi* (mean: 3.15%), *R. signatus* (3.26%), *R. montanus* (3.45%), and *R. chromasynchysi* (3.46%), respectively, all of which belong to Clade ‘N’ as demonstrated by VIJAYAKUMAR et al. (2014). Our preliminary phylogenetic analysis suggests *R. silentvalley* sp. n. to be sister to the *R. chromasynchysi* species complex identified in VIJAYAKUMAR et al. (2014).

Advertisement call: A total of four advertisement calls from three males were recorded at the type locality. Recordings were made at night immediately after light showers at an ambient temperature of 17.8–19°C. The frogs called in chorus at seemingly regular intervals. The advertisement call of *Raorchestes silentvalley* sp. n. (Fig. 5A) comprises of a series of high-frequency pulses (2,250 Hz), approximately 0.199 s apart on average, and the mean inter-call interval is 1.88 s (n = 4). There is an average of 6–8 rapid pulses in each call, increasing in amplitude from the initial pulse to the last. The mean duration of a call was 0.971 s (n = 4). All harmonics are multiples of the fundamental frequency. The note rate was 10.37/sec. In comparison, its sister taxon, *R. chromasynchysi*, has a call composed of 2–3 pulses (2,584 Hz) on average (Fig. 5B), with a call duration of 0.410 s (n = 5) and a mean inter-call interval of 1.44 s (n = 3).

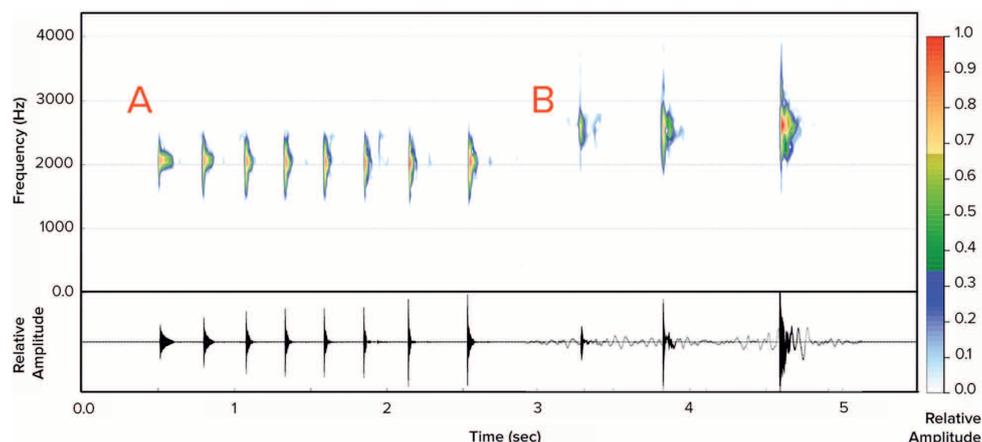


Figure 5. Comparative spectrograms (top) and corresponding oscillograms (bottom) of single notes of the advertisement calls on the same time scale of *R. silentvalley* sp. n. (A) and *R. chromasynchysi* (B).

***Raorchestes lechiya* sp. n.**
(Figs 7, 8; Table 2)

ZooBank LSID: urn:lsid:zoobank.org:act:1261A42B-BoCo-4571-87F4-8EC3B5381A88

Suggested common name: Lechiyappan's bush frog

Holotype: TNHM (H) 12.6.18/55 (Figs 7A, 8); adult male (19.8 SVL); collected from forest understorey vegetation at Sispara (11°12'5.70" N, 76°26'25.64" E) at 2,023 m a.s.l., Silent Valley National Park, Palakkad District, Kerala, by ANIL ZACHARIAH, 03 May 2012.

Paratypes: TNHM (H) 12.6.18/56; adult male (21.1 SVL); collected along with the holotype by ANIL ZACHARIAH.

Diagnosis: Its genetic data (16S rRNA) identifies the new species as a member of the genus *Raorchestes*. *Raorchestes lechiya* sp. n. can be distinguished from all known congeners by the following combination of characters: a small-sized (mean SVL 20.5, N = 2) pale brown bush frog with a granular dorsum bearing small, horny spicules; snout rounded in ventral view; distinct tympanum and supratympanic fold; dark brown loreal and tympanic regions; bicoloured eye with upper third copper-coloured and lower two-thirds dark brown; dark reticulated patterns on the venter; groin, anterior and posterior regions of thighs brown with white spots; basally webbed toes; distinct advertisement call.



Figure 6. Habitat of *Raorchestes silentvalley* sp. n. and *R. lechiya* sp. n.: A) Shola-grassland ecosystem (which is a climax matrix of tropical montane grassland and forests) in the upper Nilgiri Hills, at Sispara in the Silent Valley National Park; B) montane evergreen forest where both new species were located and collected; C) undergrowth of *Strobilanthes lawsonii*, perched upon which many vocalizing male individuals of both species were observed.

Description of holotype: An adult male of SVL 19.8 (Fig. 8); head wider (8.4) than long (8.2); snout rounded in ventral aspect, acuminate in lateral profile (Fig. 8C); snout length (3.5) subequal to horizontal diameter of the eye (3.3); canthus rostralis sharp, loreal region concave; interorbital distance (3.2) greater than upper eyelid width (2.1); tympanum distinct; supratympanic fold well developed, extending from the outer canthus to the region of forelimb; lanceolate tongue with lingual papilla; vomerine teeth absent; pupil horizontal; vocal sac single, median, and subgular.

Forelimb (4.3) shorter than hand (5.6); fingers III, IV, and V with lateral dermal fringes; subarticular tubercles prominent, round, and single; supernumerary tuber-

cles present on fingers III, IV, and V; inner palmar tubercles prominent, oval; discs well developed, expanded, and broadened; finger webbing absent; relative lengths of fingers $II < III < V < IV$.

Hind limb moderately long, shank length (11.2) subequal to thigh (11.1); toes basally webbed (Fig. 8D), extending from the distal subarticular tubercle of toe V to second subarticular tubercle of toe IV; lateral dermal fringes present on all toes; subarticular tubercles well developed, round, and single on all toes; supernumerary tubercles present but indistinct; inner metatarsal tubercle prominent, oval; outer metatarsal tubercle weakly developed; relative lengths of toes $I < II < III < V < IV$.



Figure 7. Male *Raorchestes lechiya* sp. n. (in life): A) holotype (TNHM (H) 12.6.18/55); B) vocalizing male (not collected); C) male in amplexus (TNHM (H) 12.6.18/55) with a female (not collected).

Skin of dorsum weakly shagreened with numerous small, weakly developed horny spicules, snout and head weakly shagreened, gradually turning more granular toward the posterior regions; venter highly granular; dorsal surfaces of fore- and hind limbs granular; planar and plantar surfaces weakly shagreened (Figs 8D, E). Small incision in the posteroventral surface of the right thigh made for tissue sample, partially obscured due to preservation.

Coloration of holotype in life: Dorsum pale brown, paler towards the head and snout; loreal and tympanic regions dark chocolate brown; a faint, dark, irregular band in the interorbital region; and two short, curved, parallel, brown bands on the dorsum (Fig. 7A); groin and anterior and posterior margins of the thighs with faint whitish spots; posterior face of thigh with dark reticulations suffused with pale whitish spots; dorsal face of thighs and shanks pale brown with dark brown transverse bands; venter pinkish white with brown vermiculations on throat, chest, and forelimbs, and, faintly, on the hind limbs; iris bicoloured with upper third copper-coloured and lower two-thirds dark brown.

Colour of holotype in preservative (after three years of preservation): Dorsum greyish brown (Fig. 8A); venter off-white with irregular brown vermiculations on throat, chest, and limbs (Fig. 8B); dorsal face of limbs greyish-brown;

fingers II and III white; hand and foot brown; flanks brown with darker and paler vermiculations and blotches; groin and anterior and posterior parts of thighs brown with pale whitish spots.

Differential diagnosis: *Raorchestes lechiya* sp. n. is a distinctly brown species; it could most likely be confused with *R. chlorosomma*, which occurs south of the Palakkad Gap (and belongs to the southern clade as identified by VIJAYAKUMAR 2014), and *R. archeos*, which occurs in the Agasthyamala Hills and on the Periyar Plateau, likewise south of the Palakkad Gap. The new species differs from *R. chlorosomma* by its small adult male size (vs. medium adult male SVL 25.9–27.5 mm), dark vermiculations on the throat and chest on a predominantly pinkish white ventrum (vs. pale, uniform whitish throughout), posterior margin of thigh with pale spots on a dark brown background (vs. uniform brown), dorsum granular, covered with spicules (vs. smoothly shagreened), presence of a lingual papilla on tongue (vs. no lingual papilla on tongue), and by the iris being lighter copper-coloured above and dark brown below (vs. metallic-green eyes in *R. chlorosomma*). *Raorchestes lechiya* sp. n. differs from *R. archeos* by its rounded, acuminate snout (vs. acutely pointed), dorsum granular, covered with several small horny

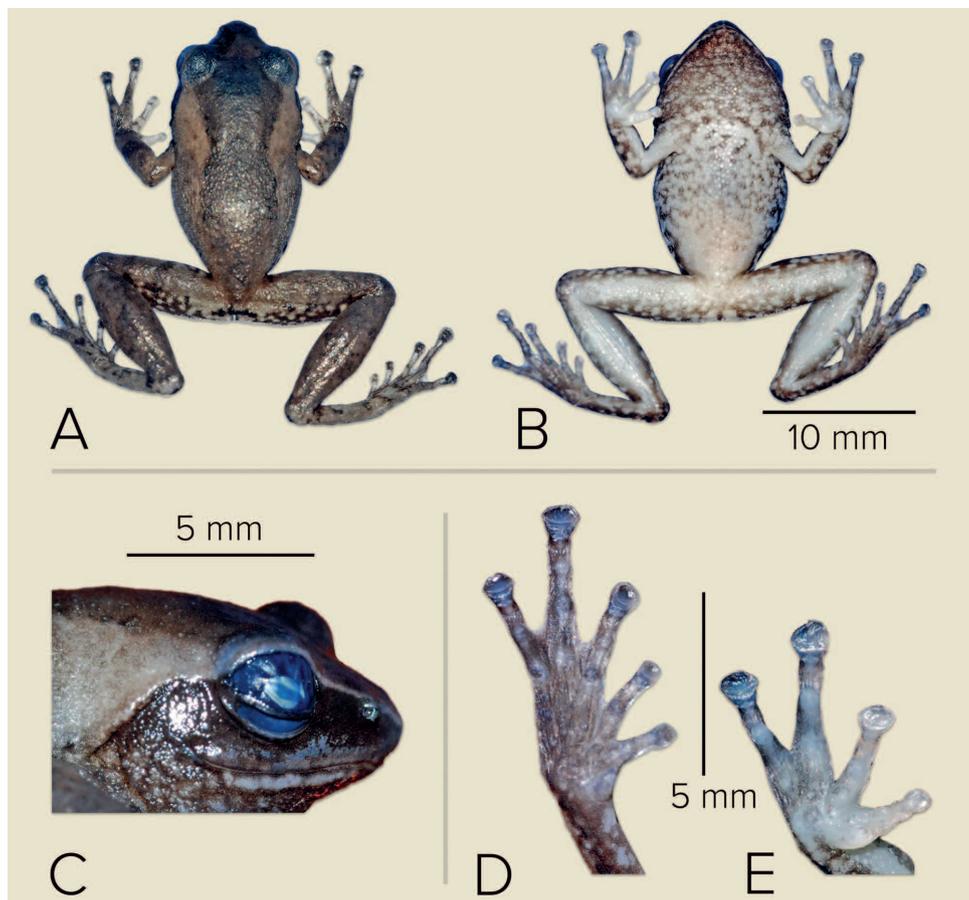


Figure 8. Holotype of *Raorchestes lechiya* sp. n. (TNHM (H) 12.6.18/55): A) dorsal view; B) ventral view; C) lateral view of head; D) ventral view of right foot; E) ventral view of right hand.

spicules (vs. sparsely spiculate shagreened), tongue with a lingual papilla (vs. without a lingual papilla), and bicoloured eyes (vs. uniformly golden-brown coloured). *Raorchestes lechiya* sp. n. could also be confused with *R. chromasynchysi*, *R. ravii*, *R. charius*, *R. coonoorensis* (all of which also occur in the Nilgiri Hills, but are not sympatric) and *R. tinniens*. It differs from *R. chromasynchysi* by having dark loreal and tympanic regions (vs. pale and similar to the overall body colour), and a mottled throat and chest (vs. plain), and bicoloured eyes (vs. uniformly gold-coloured). It differs from *R. ravii* by having a darker loreal region as compared to the dorsum (vs. uniform colour throughout the loreal region and dorsum), and by possessing bicoloured eyes (vs. uniformly golden-brown coloured). It can be differentiated from *R. charius* by its small adult size (vs. medium size) and from *R. charius* and *R. coonoorensis* by having a more granular dorsum with small spiculate projections and whitish spots in the groin and anterior and posterior regions of the thighs (vs. dorsum with prominent spiculate and tuberculated projections and large yellow or brown blotches on the groin and thighs in *R. charius* and *R. coonoorensis*). Furthermore, *R. lechiya* sp. n. is a robust frog (vs. slender elongate in *R. coonoorensis*). It differs from *R. tinniens* by having a pale venter with brown vermiculations and fingers II and III being white (vs. plain yellowish white ventrum and fingers II and III yellow). Apart from these species, it can be distinguished from the sympatric *R. signatus* primarily by its much smaller adult male size (vs. average adult male SVL > 30 mm), and by having a rounded snout in ventral view (vs. pointed). *Raorchestes lechiya* sp. n. differs from *R. aureus* primarily in possessing bicoloured iris (vs. uniformly golden iris) and in having differentially darker loreal and tympanic regions compared to the dorsal surface of the head (vs. uniformly coloured dorsal, loreal, and tympanic regions). Finally, *R. lechiya* sp. n. can be distinguished from *R. silentvalley* sp. n. by its brown coloration, extent of webbing (more developed in *R. silentvalley* sp. n.), smaller size, differences in call characteristics, and habitat use (forest understorey in *R. lechiya* sp. n. and midstorey in *R. silentvalley* sp. n.).

Variation: *Raorchestes lechiya* sp. n. exhibits minor differences in colour pattern among individuals. The paratype differs from the holotype by being dorsally covered with large black spots and lacking the two short parallel dark bands.

Etymology: The species name is a tribute to the late Mr. Lechiyappan of the Mudugar tribe, a forest tracker at Silent Valley National Park. He was instrumental in the early conservation undertakings of the Silent Valley Movement, which eventually led to the declaration of the area as a National Park.

Distribution and Natural History: *Raorchestes lechiya* sp. n. was found only in the upper reaches of the southwestern slopes of the Nilgiri Hills (Fig. 1) at altitudes ranging from 1,800–2,200 m a.s.l. We found the species inhabiting the leaf litter and understorey vegetation in montane wet forests (Figs 6A, B). We also observed individuals vocalising from branches of shrubs at an average height of 1 m from the ground, as well as an amplectant pair (Fig. 7C).

Genetics: The mean genetic difference in the sampled region of the 16S rRNA gene between *Raorchestes lechiya* sp. n. and 52 Western Ghats congeners ranged from 2.39–10.09% (Table 1). The uncorrected pairwise sequence differences to the genetically most similar species were as follows: *R. aureus* (mean: 2.39%), *R. signatus* (2.79%), *R. montanus* (3.12%), *R. primarrumpfi* (3.22%), respectively, and *R. chlorosomma* (6.43%) to which it is morphologically similar. Our preliminary phylogenetic analysis suggests *R. lechiya* sp. n. to be the sister species of *R. aureus* and nested in Clade 'N'.

Advertisement call: A total of three advertisement calls from two males were recorded at the type locality. Recordings were made at night immediately after light showers at an ambient temperature of 17.8–19°C. The advertisement call of *Raorchestes lechiya* sp. n. (Fig. 9A) is a low “treek”

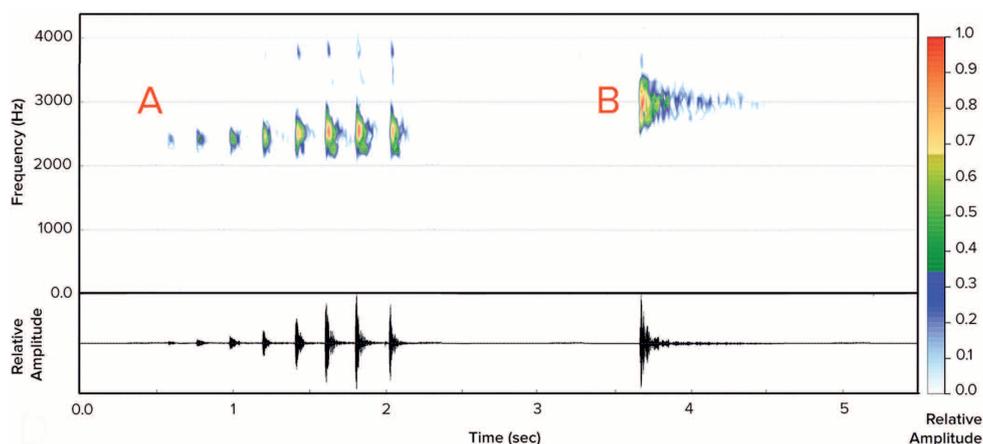


Figure 9. Comparative spectrograms (top) and corresponding oscillograms (bottom) of single notes on the same time scale of the advertisement calls of *R. lechiya* sp. n. (A) and *R. aureus* (B).

call, which is repeated at intervals of 1.30 s ($n = 3$) on average. A call on average comprised 7–9 rapid pulses, approximately 0.028 s apart on average. The mean duration of a call was 0.32 s ($n = 3$) and the fundamental frequency 2,625 Hz. The pulse rate was 25.16/s. In comparison, its putative sister taxon, *R. aureus*, has a call comprising a single pulse (3,000 Hz) with a mean duration of 0.18 s ($n = 3$) and an inter-call interval of 2.74 s (Fig. 9B).

Discussion

Our explorations in the montane wet forests around the peak of Sispara Betta (in the Walakkad section of the Silent Valley Forest Range) and the Thudukki section (Bhavani Forest Range) resulted in the discovery of two new species of bush frogs. This raises the number of described *Raorchestes* species in the Western Ghats to 53 (we have identified *R. emeraldi* as a junior synonym of *R. flaviventris*; ABRAHAM et al. 2015), making it the most speciose anuran genus in the region.

Preliminary phylogenetic results (tree not shown) place both species described here in the northern clade of *Raorchestes* (Clade 'N' of VIJAYAKUMAR et al. 2014), reflecting their distribution north of the Palakkad Gap. Even though both new species occur in the same habitat at altitudes above 1,800 m a.s.l., *R. silentvalley* occupies the upper layers of the forest understorey and lower midstorey on leaves and branches of small trees during the breeding season, whereas *R. lechiya* is found on lower vegetation and leaf litter on the forest floor. Both species are currently known only from within the Silent Valley National Park and likely endemic to the upper sections of the western and southwestern slopes of the Nilgiri Hills.

Genetically, *R. silentvalley* sp. n. has a divergence of > 3% in the 16S unpaired p-distance sequence from all other congeners (Table 1). It is most similar to *R. chromasynchysi*, which is found at lower altitudes without zone overlap and can be distinguished by morphological and vocal characters (the *R. silentvalley* sp. n. call is composed of 6–8 rapid high-frequency pulses at 2,250 Hz as opposed to 2–3 slower pulses at a slightly higher frequency of 2,584 Hz in *R. chromasynchysi*). *Raorchestes lechiya* sp. n. has a lower divergence compared to the most similar *R. aureus* (2.39%). Preliminary phylogenetic results place the new species sister to *R. aureus*, but *R. lechiya* sp. n. can be delimited by a combination of differences in male vocalisation characters (*R. lechiya* sp. n.: call comprising 7–9 rapid pulses at a frequency of 2,625 Hz versus a single pulse call at 3,000 Hz in *R. aureus*) and morphology. Moreover, both species differ in their distribution, with *R. lechiya* sp. n. being restricted to the higher reaches of the Nilgiri Hills, while *R. aureus* is found only in the upper reaches of the Siruvani Hills across the Attappadi Valley (see Fig. 1). Morphological, bioacoustic, distributional, and ecological data have been significant for diagnosing both novel species. Although neither has so far been recorded from outside the park, diligent survey efforts in neighbouring regions such as New Amarambalam,

Upper Bhavani, and the slopes to the south of the Kundah Range may provide supplemental distributional data.

The declaration of the area now known as Silent Valley National Park in 1985 followed in the wake of a landmark national and international debate on the relative importance of human development versus the protection of natural resources that ensued when the government proposed to construct a dam in the area in 1976 (MANOHARAN 1999). The scientific community responded quickly with a preliminary but thorough study of the area's biodiversity and fragile ecology, thereby providing information that enabled activists to rally favour for the area, its being exempted from human development, and leading to the abandonment of the Silent Valley hydroelectric project (GADGIL & GUHA 1994). The dedicated interest in the area resulted in the discovery and description of amphibian novelties such as *Micrixalus thampi*, *Duttaphrynus silentvalleyensis*, *Ghatophryne rubigina*, and *Ichthyophis longicephalus* (PILLAI 1981, 1986, PILLAI & PATTABIRAMAN 1981) from the area that is today classified as the Silent Valley National Park. Our addition of two new bush frog species underscores the importance of this national park and the region in general as a vital repository of irreplaceable biodiversity worthy of the struggle of the past, as well as preservation for the future.

Acknowledgements

We thank the Kerala Forest Department for providing access and collection permits to the first and last authors, respectively (WL 10-1965/12). We also are grateful to S. SIVADAS, former Divisional Forest Officer of the Silent Valley National Park, for making all necessary arrangements for this expedition. We are indebted to VIMAL M. (Assistant Conservator of Forests), PRAMOD KRISHNAN (Conservator of Forests), JOSHIL M. (Range Forest Officer), K. K. SUNIL KUMAR (Assistant Conservator of Forests), RENEY K. PILLAI (Wildlife Assistant), and all official and field staff who facilitated our stay in the park; RAVI CHELLAM, N. P. JAYAN, P. K. UTHAMAN and SETHUMADHAVAN for their unwavering support and encouragement provided during our survey; and to UMESH P. K. and SANDEEP DAS, whose support and photographs are very much appreciated. Thanks also due CARL HUTTER, TAYLOR BROADHEAD, and ANDRESSA DE MELLO BEZERRA for their help with the call analyses, to ARUN ZACHARIAH and VISHNU O. for help with sequencing, to KERRY COBB and KIN ONN CHAN for their support during phylogenetic analysis, and to LINDA TRUEB, MICHAEL F. BAREJ, GURURAJA K. V., and an anonymous reviewer who provided valuable and constructive suggestions on drafts of this manuscript.

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Supplementary material

Additional information is available in the online version of this article at <http://www.salamandra-journal.com>

1 Supplementary table. Uncorrected pairwise distances (%) for 16S mtDNA gene fragment between 53 species of *Raorchestes*.

