

**X-mas Bush Frog *Raorchestes shillongensis* (Pillai and Chanda, 1973) Conservation Project,
Meghalaya, India.**

Final Report



April 2017

Submitted by:

Abhijit Das (PI)

Bitupan Boruah (Researcher)



**صندوق محمد بن زايد
للمحافظة على
الكائنات الحية**

**The Mohamed bin Zayed
SPECIES CONSERVATION FUND**



Meghalaya Forest Department



**भारतीय वन्यजीव संस्थान
Wildlife Institute of India**

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Background of the Project

Globally amphibian fauna is represented by 7,642 species (amphibiaweb.org). A recent assessment of the entire group (iucnredlist.org/amphibians) found about 32% of amphibian species of the world are threatened, representing 1,856 species (amphibiaweb.org). In the last two decades nearly about 168 species are believed as extinct and approximately population of 2,469 species (43%) are declining (amphibiaweb.org). So amphibians are considered as the most threatened vertebrate group of the world. In the Oriental region out of total 1021 species of amphibians 41 species are extinct in wild or critically endangered and 266 species are in endangered or vulnerable stage.

The causes for amphibian declines are habitat loss, invasive species, pesticide pollution, an emerging disease called chytridiomycosis and global climate change. Habitat destruction is the most important factor leading to amphibian population declines. Habitats destruction is going on due various anthropogenic activities such as road construction, settlements and for cultivation etc (amphibiaweb.org).

Study on ecology and breeding biology of amphibians is of prime importance for the successful conservation the species along with their habitats (Gaitonde *et al.*, 2016). Out of 407 species of India (amphibian web 2017), only about 7-8% species were studied on their reproductive ecology, which are mostly from Western Ghats ranges. In India amphibian fauna of Western Ghats region have received a remarkable attention but in Northeast India still their precise phylogeny (Biju *et al.*, 2016) and ecology is very poorly studied.

Raorchestes shillongensis is a critically endangered and endemic bush frog of Meghalaya. To know its distribution and breeding biology, we carried out first ever focused study during May and August 2016 to supply information that can augment its conservation.

Major outcome of the Project

1. We added eighty two new localities in the distribution of the species, *Raorchestes shillongensis* (within East Khasi Hills) which covers 531 sq. km area. This distribution includes protected forests, community conserved sacred grooves, tourist spots and around human settlements.
2. The species is found to be occurring within the elevation range between 1000m and 1900m asl. Earlier it was known to occur up to an elevation of 1400m asl.
3. The species is found to be relatively common in and around Shillong City and is a peri-anthropic species restricted to Shillong plateau. The species frequently encountered mainly forest edges and human settlements where dense shrub is abundant.
4. Molecular phylogenetic study showed that the species closely related to South Asian Clade (*Raorchestes*) than East Asian Clade (*Philautus*).
5. The species is found to be highly polymorphic in its external morphology.
6. The axial amplexus of the species lasts for 9-11 hrs. Clutch size of the species is relatively smaller (8-17 eggs) and developmental period is longer (30-31 days) than other known species of Bush frogs in India.
7. The species lay eggs under leaf litters on moist soils. Female mix eggs with moist soil presumably to counter desiccation risk.
8. We observed male-male combat behaviour that lasted for 27 minutes.
9. Breeding biology of the species is found to be similar with Sri Lankan Bush frogs, *Pseudophilautus* species rather than Indian congeneric species.
10. This species can be regarded as a candidate “indicator species” for long term monitoring Shillong City Environment.
11. We recommend further research to know population trend of *Raorchestes shillongensis* over multiple years.

1. Introduction

The Old World tree frog (Rhacophoridae) currently contains over 404 species in two subfamilies Buergeriinae and Rhacophorinae, distributed throughout in sub-Saharan Africa, China, Southern Asia, Sri Lanka, Japan, Taiwan, Philippines, Indonesia and Greater Sunda Islands (Frost, 2017). Subfamily Buergeriinae has four species within a genus and subfamily Rhacophorinae contains 400 species with 17 genera (Frost, 2017). These frogs are found in a wide variety of microhabitats, ranging from the ground to the canopy level (Biju & Bossuyt, 2005a). Among the members of the Rhacophoridae, Bush frogs are distinguished by their direct-development (Bossuyt & Dubois, 2001; Grosjean *et al.*, 2008; Li *et al.*, 2009; Biju *et al.*, 2010; Yu *et al.*, 2010; Hertwig *et al.*, 2012) and are known to zoologist since 1822 (Bossuyt and Dubois, 2001). Bush frog clades are nested in the subfamily Rhacophorinae, those are distributed in south and Southeast Asia (Vijaykumar *et al.*, 2014, Frost 2016). The most significant revisionary work on Bush frog (*Philautus*) group has done by Bossuyt and Dubois in 2001. They recognized 84 valid species of *Philautus*. Earlier all Bush frogs are considered as monophyletic and assigned to genus *Philautus* Gistel 1848 (Hertwig *et al.*, 2012) on the basis of mainly morphological characters. Recent molecular studies resulted division of Bush frogs into three major clades *viz.* *Philautus*, *Pseudophilautus* and *Raorchestes*. Later two genera, *Pseudophilautus* and *Raorchestes* are sister clades and are polyphyletic with respect to genus *Philautus* (Li *et al.*, 2009). Out of these three clades, Sri Lankan clade assigned to genus *Pseudophilautus* Laurent 1943 (Li *et al.*, 2009; Yu *et al.*, 2010). Clade from Borneo and its adjoining Islands and few unstudied species from India are assigned to genus *Philautus* Gistel 1848 (Hertwig *et al.*, 2012). Most species of Western Ghats clade of Peninsular India are assigned to genus *Raorchestes* by Biju *et al.*, 2010 (type species: *Ixalus glandulosus*, Jerdon 1854) and coined the generic name in recognition of pioneering work on Batrachology of India by C. R. N. Rao. This genus ranges from the Western Ghats of India, through Northeast India to southern China, Laos and Vietnam (Biju *et al.*, 2010; Frost, 2016). These three Bush frog clades comprises about half (193 sp.) of the total Rhacophorid frogs (404 sp.) (Vijayakumar *et al.*, 2014, Frost 2016). Taxonomic sampling within Bush frog clade is still not complete. This is evident from the fact that ~ 100 species are added in India in recent one and half decades (Frost, 2016) mainly from Western Ghats and Sri Lanka, which is generally considered herpetologically ‘well studied’ (Bahir, M. M. 2005).

Now, there are 193 valid species of Bush frogs worldwide represented by three genera (*Raorchestes*, *Philautus* and *Pseudophilautus*) and 73 species are known from India (Frost, 2017). Out of which, genus *Raorchestes* is represented by 60 species, *Philautus* 9 species and *Pseudophilautus* by only four species (Frost, 2017). Northeast India harbours 10 species of Bush frogs with two genera (*Raorchestes*- 4 species and *Philautus*- 6 species) (Frost, 2017).

Northeast Indian region lies between two globally recognized biodiversity hotspots, the Indo-Burma hotspot and the Himalaya hotspot (Mittermeier *et al.*, 2004). The eight northeastern states, viz., Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland, Sikkim and Tripura are particularly interesting, because of unique hill ranges and different geological origin (Datta-Roy *et al.*, 2013). This region forms the transitional zone between the Indian subcontinent and South-East Asia and the climatic habitat range from the tropical valleys to the boreal mountain peaks (Pawar *et al.*, 2007, Biju *et al.*, 2016). The Himalayan range originated as a result of collision between the Indian plate and the Asian continent (Krishnan 1974). South of the eastern Himalayas, the Garo, Khasi and Jaintia Hills are dating back to the Cretaceous period (Hora 1944; Valdiya 2010). These complex topographical, physiographical and climatic factors are responsible for the rich diversity of flora and fauna of the Northeast region (Mathew & Sen 2010).

However amphibian fauna of the Northeast India have not received much attention in comparison to the Western Ghats (Biju *et al.*, 2016), specially information regarding the phylogeny and natural history of Bush frogs.

2. Project Objectives

1. To document the distribution and population status of *Raorchestes shillongensis*.
2. To study the habitat ecology and breeding ecology of the species.
3. To create awareness about the conservation significance of the species.

3. Review literature

Taxonomic studies on amphibian fauna of North East India can be traced back to Boulenger (1890-1920) and Annandale (1912) and Smith (1921-1935). Since then there had been a number of publications (Hora, 1923; Romer, 1949; Roonwal and Kripalani, 1961; Yazdani and Chanda, 1971; Chanda and Talukdar, 1973; Pillai and Yazdani, 1973; Pillai and Chanda, (1976-1981); Mansukhani *et al.*, 1976; Tilak and Mehta, 1977; Singh, 1977, 1995; Bhaduri and Saha, 1980; Mansukhani and Sarkar, 1981; Das 1984, 1988; Sahu, 1985; Sarkar and Sanyal, 1985; Chanda, 1986, 1990, 1990a,b, 1991, 1992, 1993, 1995, 2002, 2006, 2007; Kiyasetuo and Kher, 1986, 1987; Kiyasetuo, 1986; Chanda and Ghosh, 1989; Dasgupta, 1990; Roy and Elepfandt, 1993; Chanda and Sarkar, 1997; Dutta 1997; Mallick, 1997; Roy *et al.*, 1998; Ahmed and Goswami, 1999; Bordoloi and Borah, 1999; Choudhury *et al.*, 1999, 2001, 2001a, b; Dey and Gupta, (1999- 2002); Pillai and Ravichandran, 1999; Ahmed and Dutta, 2000, 2001; Bordoloi *et al.*, (2000- 2007); Das and Chanda, 2000; Deuti *et al.*, 2000; Dutta *et al.*, 2000; Ghosh and Sarkar, 2000; Saikia *et al.*, 2000; Sengupta *et al.*, (2000- 2008); Ahmed and Roy, 2001; Borah and Bordoloi, 2001; 2001a; Pathak *et al.*, 2001; Selim, 2001; Ahmed, 2002; Deuti and Dutta 2002; Hooroo *et al.*, 2002; Sarkar *et al.*, 2002; Dey and Ramanujam, 2003; Ao *et al.*, 2003; Mathew and Sen, (2003-2010); Sen and Mathew, (2003-2006); Das *et al.*, 2004; Grosslet *et al.*, 2004; Sen, 2004; Mathew *et al.*, 2005; Sarkar and Roy, 2006; Borthakur *et al.*, 2007; Ningombam and Bordoloi. 2007; Saha and Gupta, 2007; Talukdar *et al.*, 2007; Kamei *et al.*, 2009) which contributed significantly to the amphibian fauna of the region. Most comprehensive work done on systematics of the amphibian fauna of Northeast India is by Chanda (1994) covering 54 species. Mathew and Sen (2010) provide 119 species of amphibians in “Pictorial 'Guide to the Amphibians of Northeast”. In last five years and present year more than 10 species were added to the amphibian fauna of Northeast India by Das *et al.*, 2010; Sengupta *et al.*, 2010; Mahony *et al.*, 2011; Kamei *et al.*, 2012; Purkayastha and Matsui, 2012; Das *et al.*, 2013; Kamei *et al.*, 2013; Mahony *et al.*, 2013; and Biju *et al.*, 2016.

4. Study species

Raorchestes shillongensis was described as *Philautus shillongensis* by Pillai and Chanda in 1973 with eight specimens collected from Malki forest within the Shillong city below 1400m elevation. The species is listed in various publications as endemic but never recorded from elsewhere except the type locality. In a taxonomic review, Bossuyt and Dubois (2001) expressed their doubt regarding the taxonomy of the species and its possibilities to be a junior subjective synonym of *Ixalus annandalii*, Boulenger, 1906. However, this species is believed to be associated with tropical moist forest and probably undergoes direct development (Dutta et al. 2004). Biju *et al.* (2010) reallocated the species into genus *Raorchestes* by implication. In Conservation Actions section of IUCN evaluation team mentioned that it is not known whether this species occurs in any protected areas, but habitat protection and maintenance are urgent priorities for this species, and additional survey work is necessary to assess its current population status. This species is categorized as “critically endangered” by IUCN because of its very narrow distribution and its habitat declination. Like majority of amphibian species in Northeast India, information regarding ecology and biology of this species is also lacking.

5. Study Area

The type locality of *Raorchestes shillongensis* is Malki forest (25°33' 45" N and 91°53' 19" E) falls within Shillong city of East Khasi Hill district Meghalaya, Northeast India. This region lies at the junction of Himalayan and Indo-Burman biodiversity hotspots (**Figure 1**). Khasi Hills is situated on the middle part of Meghalaya and bounded on the West by Garo Hills, North by Assam, East by Jaintia hills district and South and the South by Bangladesh. Malki forest is an uninhabited strip of subtropical wet hill forest (Champion & Seth 1968) with some broadleaf trees (Mahony et al. 2013). Streams at these localities were approximately 1–3 m wide of moderate flow on a slight incline, primarily rocky with intermittent gravel bed, and with quiet pools that accumulate detritus, branches and leaf litter (Mahony et al. 2013). Furthermore, Malki forest is surrounded by thickly populated human dominated landscape. Some reserve forest like Upper Shillong, Riat-Laban, Shyrwart, Laitkor, Mawpat, Mawlai and Riat Khawan are located at the fringe areas of the Malki forest. Other minor forest fragments of the East Khasi Hills, designated as private and sacred forest (e.g. – Mawphlang), are maintained through community protection based primarily on traditional beliefs. Khasi Hills has about twenty amphibian name bearing taxa with specific and non specific type localities out of which the Shillong Plateau has ten taxa, the type localities of which lie specifically within its biogeographical limits (Mahony et al. 2013).

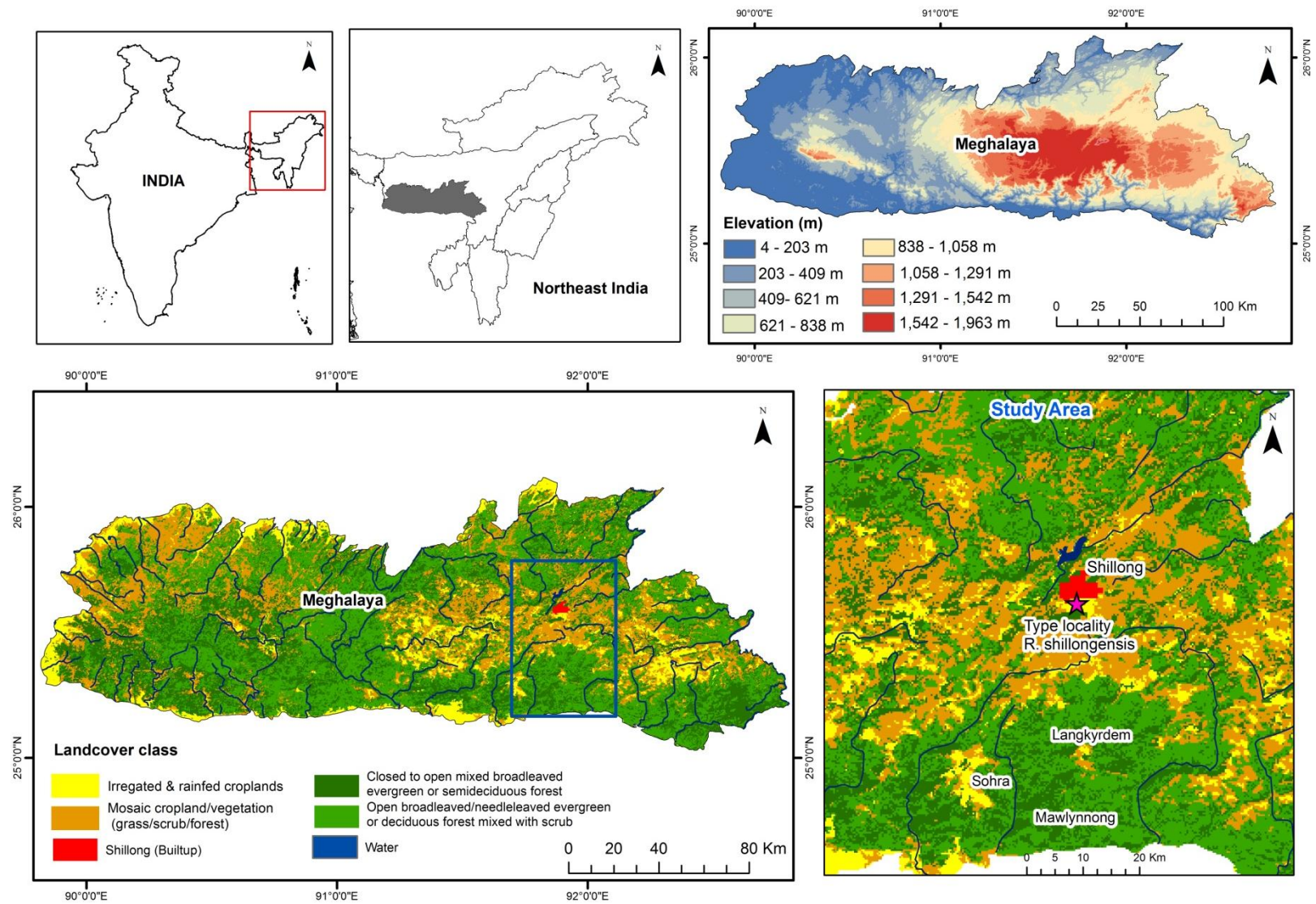


Figure 1: Map showing the study area.

6. Methodology

Survey was conducted during May and August 2016 in the East Khasi Hills district of Meghalaya, Northeast India.

6.1. Field survey

Surveys were made in different potential habitats such as streamside, near human habitations and bushes along roadsides and inside the forested area. We employed nocturnal visual encounter and acoustic search to locate calling aggregation. During daytime, we surveyed for resting individuals and egg clutch. Individuals were photographed and notes on habitat, microhabitat, perch height, variations, behavioural activities and GPS locations are recorded in a datasheet. Measurement (SVL) and weights are taken to the nearest 0.1 mm using a digital slide calliper (to the nearest 0.1) and Pesola Spring balance. Male and female were identified in the field based on presence or absence of vocal sac and externally visible ripe ova. Temperature and humidity was recorded using Hygrometer. Furthermore, to find out the status of the species at different habitat (e.g., forest edge, near human habitation, along stream, inside the pine forest) within the study area we counted the number of individuals within 5m×5m quadrat. Two persons counted the individuals inside the quadrates with the help of torchlight.

6.2. Breeding observation

We observed breeding biology of the species in five occasions in two different sites (Maliki forest and Risa colony) in natural condition. Observations were made from approximately 1m distance using torchlight. Each event was recorded in minutes and hours using a stopwatch. After completion of spawning clutch size, egg diameter, size (SVL) of the amplexant pair was taken. We evaluated the association between female's body size and clutch size and egg size. Development of the eggs continuously observed for two clutches in natural habitat (to avoid desiccation) with the help of magnifying glass. Two egg clutches were carried to the field station and kept in natural condition (with a maximum 1°C temperature difference from the outside) to see the percentage of successful hatchlings. Statistical analysis was done in Microsoft Excel (2013). Froglets after hatchling were released back to their natural habitat.

6.3. Call record and analysis

Call of a single uncollected male of *R. shillongensis* was recorded using a digital recorder (Sony IC recorder 7.4.0) in ZSI, Shillong Campus on 30th August at 19.41 hrs. Ambient air temperature of the calling site was taken with a digital thermometer.

Recorded calls were visualized and call characters were obtained using Raven Pro Ver. 1.5 (Charif, Waack & Strickman, 2010). We measured a total of five temporal properties that included call group duration, inter call group interval, intra call group interval, call duration and call rate of a call bout comprising of five call groups. One spectral property, i.e., peak frequency, was also measured over the entire series of calls. Terminologies and graphical representation of call properties analysed follow Bee, Suyesh & Biju (2013a) and Bee, Suyesh & Biju (2013b).

6.4. Sampling and preservation

We collected samples (adults) randomly using hand pick up method. Collected individuals were fixed in 10% Formalin and preserved in 70% Ethanol. Preserved specimens were studied in the laboratory of Wildlife Institute of India, Dehradun.

6.5. DNA analysis and phylogeny

6.5.1. DNA extraction

Total genomic DNA was extracted from liver tissues preserved in molecular grade Ethanol using a DNeasy blood and tissue kit (Qiagen, Germany) following the manufacturer's protocol. Partial gene fragments of 16S rDNA were amplified by polymerase chain reaction (PCR). Primers 5'-GCCTGTTTATCAAAAACAT-3' (16Sar-L) and 5'-CCGGTCTGAACTCAGATCACGT-3' (16Sbr-H) as forward and reverse for 16S (Palumbi *et al.* 1991), were used in the current study. Each PCR reaction was prepared in a 25 μ l volume amplification mixture consisting of 2.5 μ l MgCl₂, 2.5 μ l of Taq polymerase buffer, 2.5 μ l of dNTPs mix, 0.25 μ l of each primer (forward and reverse), 1 μ l of purified DNA, .67 μ l of Taq polymerase (Bangalore Genei Pvt. Ltd.) and Milli-Q water was used to make up the remaining 25 μ l volume. The amplification process for 16S rRNA gene sequences was conducted as follows:

An initial denaturing step at 94°C (4 min), 40 cycles of denaturing at 94°C (45 s), then annealing at 47.1°C (1 min) and extending at 72°C (1 min), and a final extension at 72°C (10 min). The PCR products thus generated were purified using spin columns. DNA sequences of

only the forward strand were obtained using corresponding forward primers. Sequences were later deposited in GenBank.

6.5.2. Sequence alignments and phylogenetic analyses

For incurring phylogentic position of the current species, homologous sequences for 16S rRNA gene for 10 species were downloaded from NCBI GenBank data base of closely related clades and genera belonging to Asian Bush frogs, from South and South East Asia. The species for which sequences were downloaded represent species from representative bush frog clades from the work of Vijay *et al.* (2016). *Kurixalus eiffingeri* was used as the out group for the analysis. Two sequences generated during the present study belonging to *Raorchestes shillongensis* collected from type locality (Malki forest) and Risa forest was added. Additionally sequences generated from one specimen from Riwai and one specimen from Mawlynnong, belonging to the genus *Raorchestes* was included to have a representation of taxonomic entity from different geographical entities within the distributional range of *Raorchestes shillongensis*. In total 14 sequences were used in the current analysis.

Program Muscle 3.6 (Edgar, 2004) implemented in MEGA 7 was used to align our new sequences against this data manually. The alignments were checked visually and corrected manually where necessary. Alignment gaps were treated as missing data. Accession numbers of new sequences generated in the study as well as those that were downloaded from GenBank for phylogenetic analysis. Phylogenetic analysis based on the combined data was performed by maximum likelihood (ML). For model to be used in ML analysis, we performed a partition model sensitivity analysis with Partition Finder to determine appropriate evolutionary models for the combined dataset. ML analyses were conducted in MEGA on the dataset. Model GTR was used for all subsets, and support for nodes of the resulting ML trees was assessed by analyses of 1000 bootstrap iterations.

6.6. Comparison

We compared the morphological characters of *R. shillongensis* with other species of Bush frogs occurring in Northeast India based on publications. We also compared our observation with the original description and holotype of *R. shillongensis*. Principal component analyses (PCA) was performed (*SPSS 16*) between *R. shillongensis* and *Raorchestes* sp. 1 using 15 morphometric measurements *viz.*, SVL, AG, HW, HL, AJS, ED, ES, IOS, UEW, FLL, F III, HLL, TBL, TL and TIV.

6.7. Map and distribution

Geographic range and point location map were generated using ArcGIS 10.3. Open source data from Global Administrative area (www.gadm.org) was used for an administrative boundary, and SRTM 90 m database (<http://srtm.csi.cgiar.org>) was used for elevation map. The area under minimum convex polygon (MCP) was computed by connecting the outermost occurrence points to estimate the extent of occurrence.

6.8. Abbreviations used for the morphometric measurements

SVL, snout-vent length (from tip of snout to vent); **SL**, snout length (from the anterior corner of eye to snout tip); **IOS**, inter-orbital space (least distance between upper eyelids); **ED**, eye diameter (horizontal diameter of the eyes); **UEW**, upper eyelid width; **AJS** (distance between angle of jaws and snout-tip); **HL**, head length (distance between mandible and snout tip); **HW**, head width (at angle of jaw); **FLL**, forelimb length (from proximal end of junction of arm with the body to tip of the 3rd finger); **F-I** to **F-IV**, length of 1st to 4th fingers (from the base of the inner metacarpal tubercle to the tip of the respective finger); **HLL**, hind limb length (from the mid-ventral line of attachment of legs with the body to tip of the 4th toe); **TL**, thigh length (distance from the mid-ventral line of attachment of legs with the body to surface of knee); **TBL**, tibia length (distance between surface of knee and surface of heel, with both tibia and tarsus flexed); **T-I** to **T-V**, length of 1st to 5th toes (from the base of the inner metatarsal tubercle to the tip of the respective toe). Toe web formula follows terminology applied in Savage and Heyer (1997) where each toe is represented by a Roman numeral and the extent of web between the digits by Arabic numerals denoting the number of segments and/or fractions free from the web); **TTA**, tibio-tarsal articulation.

Institutional acronyms: ZSI- Zoological Survey of India.

7. Results

7.1. Systematic of the species:

Philautus shillongensis. Pillai and Chanda, 1973.

Philautus (Philautus) shillongensis. Bossuyt and Dubois, 2001.

Pseudophilautus shillongensis. Li, Che, Murphy, Zhao, Zhao, Rao, and Zhang, 2009.

Raorchestes shillongensis. Biju, Shouche, Dubois, Dutta, and Bossuyt, 2010.

Philautus shillongensis. Mathew and Sen. 2010.

7.2. Holotype (Voucher No. - ZSI A6971)

Collector: R. Giri in 1991 from Malki forest (GPS point is not available in the description) about 3km east of Risa colony where Eastern Regional Station of ZSI is situated (**Figure 2**). Specimens were collected from hill slope that was cut for the construction of cutcha road and about a hundred meters away from a hilly stream. Condition: Lower mandible broken below the eye at left side. Eye broken. Toe discs not in good condition. Mouth open and tongue protruded out. Colour in preservation: Body colour light brown. Upper eyelids black. A dark line joining the upper eyelids. Bands on limbs dark brown.

We compared topotypic material collected from Laitkor, Malki, Risa colony and Cherrapunjee of the Shillong Plateau. We found similarities with original description by Pillai and Chanda as follows: relatively small body (both male and female SVL ranges between 14.21mm-21.26mm); nostril closer to snout than eye; snout pointed and little longer than the eye diameter; eye pupil horizontal; loreal concave; vomerine teeth absent; tongue without papilla and bifid; a dark line between upper eyelids; third finger longest; fourth toe longest; finger and toe discs rounded. We found the following difference with the original description- body with more or less warts; distinct supratympanic fold; tympanum absent; tongue deeply or weakly notched; circum marginal groove on fingers and toes; metatarsal tubercles absent. Lateral fold from axial to groin absent; tibio tarsal articulation reaching tympanic region;)-(or sand clock shape mark present on dorsum.

Specimens studied from Riwai, Mawlynnong and Nongkhylllem were found have distinct morphological, acoustic and genetic divergence from *Raorchestes shillongensis*.

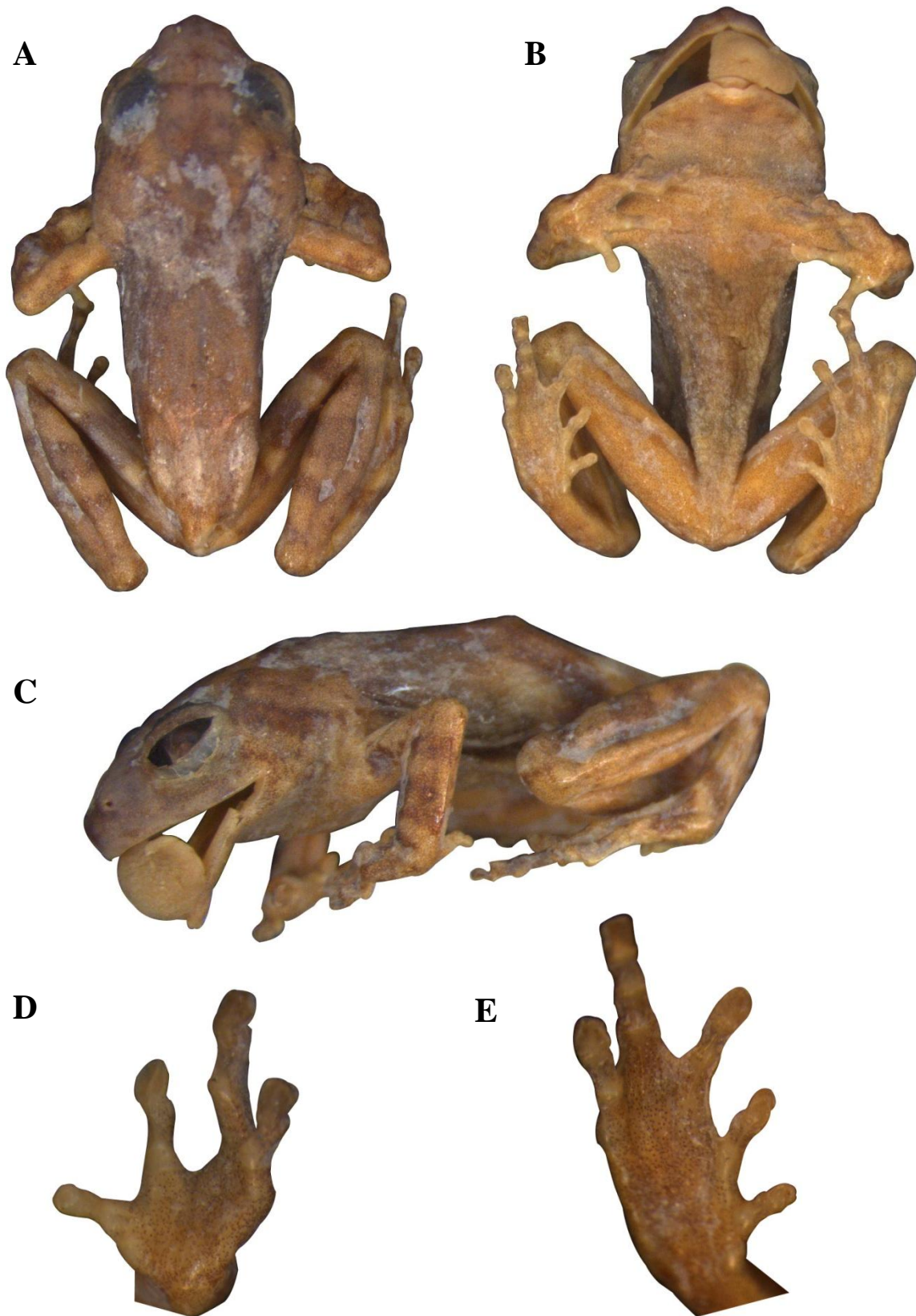


Figure 2: Holotype of *Raorchestes shillongensis* (SVL- 17mm). A. Dorsal view; B. Ventral view; C. Lateral view; D. Ventral view of hand; E. Ventral view of leg.

7.3. Redescription with fresh materials

Specimens were collected from Risa colony, ZSI Campus (25°33'41.8"N & 91°53'39.1"E, 25°33'24.4"N & 91°53'33.9"E), Malki forest edge (25°33'29.9"N & 91°53'3.8"E), Upper Shillong (25°33'55.4"N & 91°51'35.8"E) and Laitkor (25°32'39.7"N & 91°53'18.8"E), during study period. Specimens were deposited in Wildlife Institute of India, Dehradun.

7.3.1. Diagnosis: A very small sized frog (SVL of males: 14.21mm-19.01mm and SVL of females: 15.99mm-21.26mm). Head length slightly larger than width; snout pointed and slightly protruded beyond mouth in ventral view; loreal region concave, ES is slightly larger than; IOS is larger than ES; IOS is larger than UEW; eye pupil horizontal; tongue strongly or weakly notched; tympanum absent and supratympanic fold distinct from posterior corner of upper eyelid to near the shoulder; whole dorsal skin with numerous or less tiny warts; sometimes warts forms a mid dorsal ridge on head and forebody; ventrally and ventro-laterally smooth with black blotches; fingers webbed and without lateral dermal fringe; discs rounded; Tibia longer than thigh length; reduced webbing; subarticular tubercle prominent and rounded; metatarsal tubercles absent; toe tips with distinct rounded disks; circummarginal grooves present in fingers and toes.

7.3.2. Description of an adult female (Voucher no. RSA005F, **Figure 3**, all measurements provided in mm):

A small sized adult (SVL = 19.27); head longer than wide (HL = 7.14; HW = 6.75); snout pointed in both dorsal and ventral views; upper jaw protrudes slightly in ventral view; snout slightly longer the eye diameter (SL = 2.47; ED = 2.46); distance between angle of jaw to tip of the snout 6.04; canthus rostralis distinct; loreal region concave; inter-orbital space 1.9 times larger than upper eyelid width (IOS = 2.47; UEW = 1.3); nostrils oval shaped, without flap, closer to the tip of snout than to eye; tongue well notched; lingual papillae absent; vomerine teeth absent; tympanum absent; supratympanic fold distinct; eyes small (ED = 2.46), pupil-horizontal. Distance between axila and groin is 12.7.

Forelimb: Forelimb length is 12.12; fingers without lateral dermal fringe and web; subarticular tubercles prominent (finger: I=1, II=1, III=2, IV=1); relative length of fingers, I < II < IV < III (FI=2.26, FII= 4.17, FIII= 6.65, FIV= 5.28); discs rounded (finger: I DW= 0.73, II DW= 0.0.68, III DW= 1.3, IV DW= 0.9); circummarginal grooves present.

Hindlimb: Tibia slightly longer than thigh length (TBL= 8.68, TL= 8.33); Supernumerary tubercles and tarsal tubercle present (toe: I = 1, II = 1, III = 2, IV = 3, V = 2); toe V is longer than toe III (TV= 5.23, TIII= 4.47); relative length of toes I < II < III < V<IV (TI= 2.26, TII=

3.02, TIII= 4.47, TIV= 5.9, TV= 5.23); lateral dermal fringe along the toe V absent; webbing reduces, reaching upto the second subarticular tubercle at inside and below second subarticular tubercle at outside of toe IV; subarticular tubercle prominent and rounded; both inner and outer metatarsal tubercle absent; toe tips with distinct rounded disks (toe: I DW= 0.63, II DW= 0.59, III DW= 0.66, IV DW= 0.71, V DW= 0.73); circum marginal grooves present on all.

7.4. Colour in preservative: Dorsal colour grey or light brown to dark brown, pale to lateral side. Dorsal markings and bands on limb dark brown or black, prominent. Ventrally creamy white with black blotches. Anterior part of the head darker or rarely paler than rest of the body. A triangular dark mark directing to posterior between upper eyelids, not prominent in live. Eyelids and tympanic fold dark.

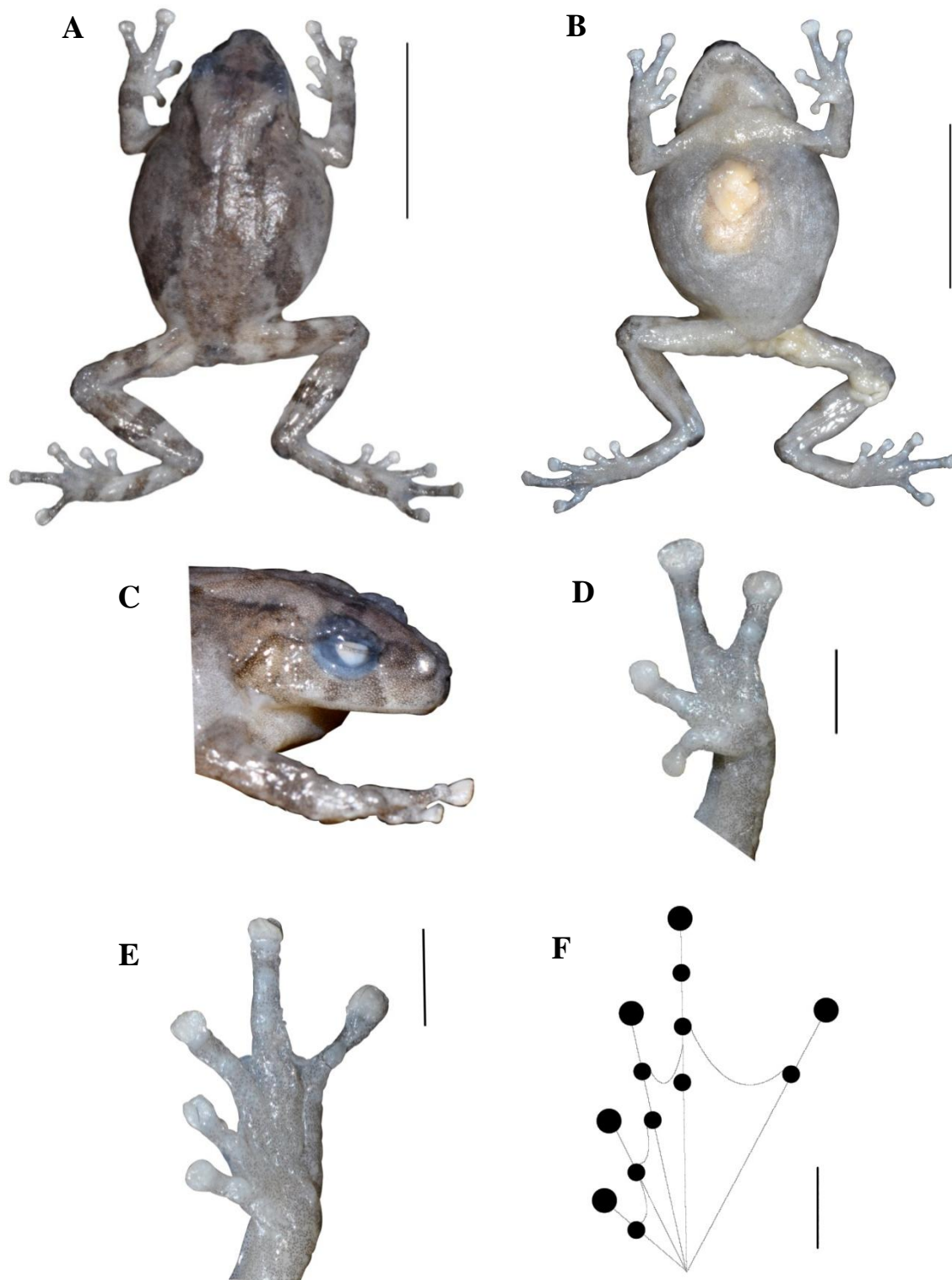


Figure 3: *Raorchestes shillongensis* (Voucher no. RSA005F). A. Dorsal view (scale bar 10mm); B. Ventral view (scale bar 10mm); C. Lateral view of head; D. Ventral view of hand (scale bar 2mm); E. Ventral view of foot (scale bar 2mm); F. Web pattern in foot (scale bar 2mm).

7.5. Colour in life: This species is found to be highly polymorphic in colour (**Figure 4**). However colour ventral side and vocal sac is same. Various polymorphs were described below.

Morph A: Dorsally brown. A dark line between the eyelids. A “)- (“ or “)(” shape dark mark from posterior corner of eyes to groin, extend to thigh and tibia. Sometimes a band from axilla also joins the dorsal band. Bands on limbs prominent. Region bellow the tympanic fold upto snout through eye is dark. Finger and toe disc reddish.

Morph B: Dorsum light brown or grey. No prominent “)- (“ shape mark. A slightly dark line joins the upper eyelids. Tympanic fold whitish. A mid dorsal ridge may present, prominent head and forebody. Groin whitish with dark patches. Bands on limbs light. Finger and toe discs whitish, sometime slightly reddish. Line from bellow the eye to upper lip may present.

Morph C: Dorsum yellowish or grey. The dark dorsal bands broad and outwardly dissolved. Region between these two bands is pale yellow or grey. Bands on limbs dark or pale. Finger and toe Discs transparent or slightly reddish.

Morph D: Dorsum creamy white with a dark brown patch of sand clock shape (from anterior edge of upper eyelids to groin). Posterior part of the dark patch extend to vent forming two bands. Dark patches also present in ventro-lateral region of belly. Bands on limbs dark brown and broad.

7.6. Sexual dimorphism: Male and female looks alike. There is no secondary sexual dimorphism other than body size and vocal sac. Male body size is relatively smaller than the body size of female (details are given in the **Table: 1**).



Figure 4: Polymorphism in *R. shillogensis*. 1- Morph A, 2- Morph B, 3- Morph C, 4- Morph D

7.7. Molecular phylogenetic position

The primers amplified approximately 550 bp long portions of the respective 16S gene. ML analyses on the dataset yielded similar topologies having good bootstrap support at major nodes. The following relations were inferred from the tree:

1. Trees from both the analysis showed that the current bush frog as a sister taxon to the south asian clade (*Rarorchestes*) than to the south east Asian clade (*Philautus*).

2. All four samples belonged to two different species *Raorchestes*_sp_Risa forest and *Raorchestes*_sp_Malki forest are closely related to *Raorchestes longchuanensis* from China.

3. In both the tree, monophyly of the Asian bush frogs was not supported. The results from ML analyses of concatenated gene sequences of 16S rRNA genes are shown in **Figur 5**.

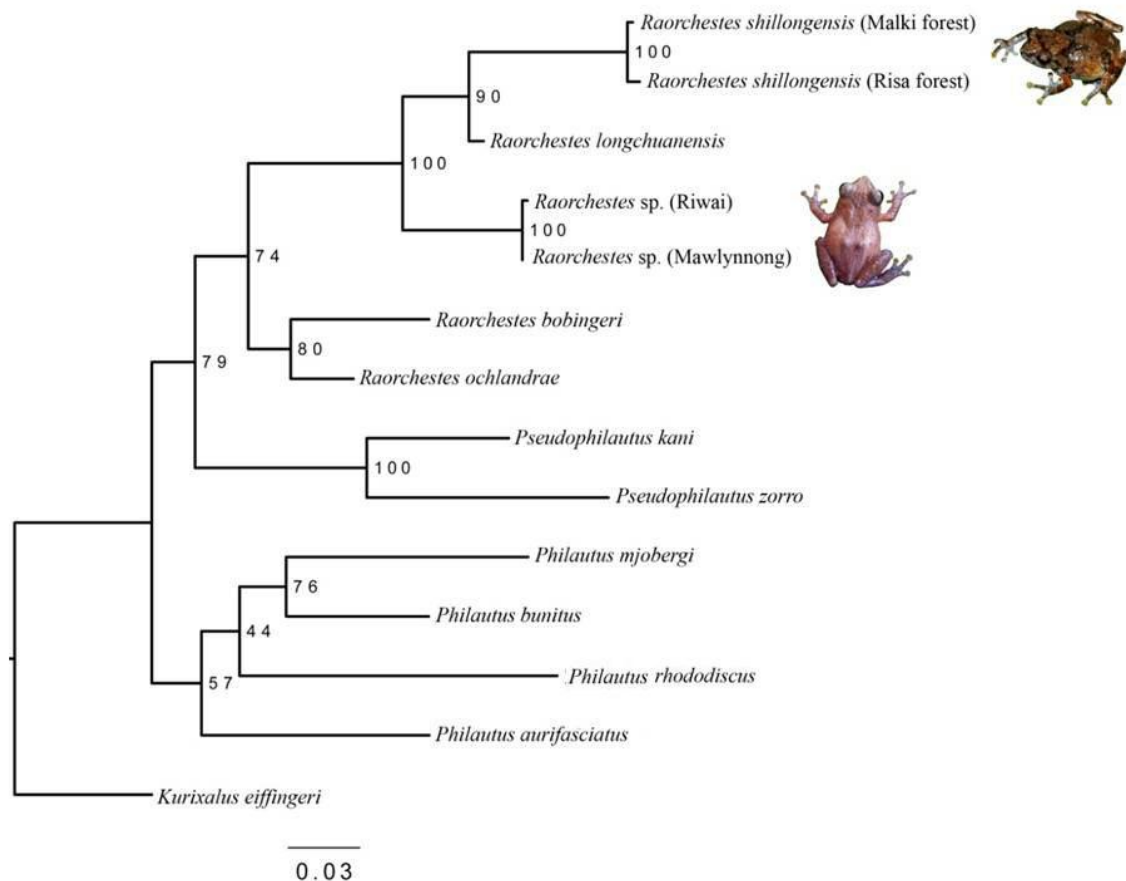


Figure 5: Maximum Likelihood tree for 12 Bush frog species and *Kurixalus eiffingeri* as an outgroup. Numbers on nodes indicates bootstrap support for Maximum Likelihood.

7.8. Differential diagnosis

We compared *R. shillongensis* with other species of Bush frogs occurring in Northeast India. It differs from *Raorchestes* sp. 1 collected from Riwai and Mawlynnong, being smaller in body size (only male individuals compared); tibia length is shorter than length of toe IV in *R. shillongensis* vs. tibia length is longer than toe IV in *Raorchestes* sp. 1; mid-dorsal ridge is absent in *R. shillongensis* (except in some individuals on head and forebody) vs. a distinct or less distinct mid-dorsal ridge from snout to vent in the *Raorchestes* sp. 1; no ridge on thigh and tibia present in *R. shillongensis* vs. present in *Raorchestes* sp. 1; dark patch on groin present in *R. shillongensis* vs. absent in *Raorchestes* sp. 1; TTA reaches tympanic region in *R. shillongensis* vs. TTA reaches the eye in *Raorchestes* sp. 1; *R. shillongensis* has more warts on body vs. less body warts in *Raorchestes* sp. 1. *R. shillongensis* is different from *Raorchestes* sp. 2 being small body size (only male individuals compared) and difference in call pattern; head longer than width in *R. shillongensis*, vs. head wider than long in *Raorchestes* sp. 2; nostril closer to snout than eye in *R. shillongensis* vs. nostril equidistant from snout and eye in *Raorchestes* sp. 2; supra-tympanic fold distinct in *R. shillongensis* vs. supra-tympanic fold absent in *Raorchestes* sp. 2; TTA reaches tympanic region in *R. shillongensis* vs. TTA reaches the eye in *Raorchestes* sp. 2;)-(or sand clock shape mark present on back in *R. shillongensis* vs. no markings present in *Raorchestes* sp. 2. *Raorchestes shillongensis* is different from *R. manipurensis*, having head length slightly larger than width (vs. head wider than length); tongue bifid in *R. shillongensis* vs. tongue lobed in *R. manipurensis*; tibia longer than thigh (femur) in *R. shillongensis* vs. tibia shorter than thigh (femur) in *R. manipurensis*; Nostril closer to snout than eye in *R. shillongensis* vs. nostril equidistant to eye and snout in *R. manipurensis*; TTA reaches tympanic region in *R. shillongensis* vs. TTA reaches nostril in *R. manipurensis*. *Raorchestes shillongensis* is different from *R. annandalii*, having nostril is closer to snout than eye (vs. nostril equidistant from snout and eye); TTA reaches tympanic region in *R. shillongensis* vs. TTA reaches tip of snout in *R. annandalii*; *R. shillongensis* is different from *R. sahai*, having pointed snout (vs. rounded snout); IOS larger than UEW in *R. shillongensis* vs. IOS less than UEW in *R. sahai*; nostril is closer to snout than eye in *R. shillongensis* vs. nostril equidistant from snout and eye in *R. sahai*; TTA reaches tympanic region in *R. shillongensis* vs. TTA reaches middle of eye in *R. sahai*. *Raorchestes shillongensis* different from *Philautus microdiscus*, having pointed snout (vs. rounded snout); IOS broader than ED in *R. shillongensis* vs. IOS is as wide as ED in *P. microdiscus*. *Raorchestes shillongensis* is different from *P. garo* as TTA reaches

tympanic region vs. TTA reaches anterior corner of the eye. In *R. shillongensis* IOS broader than ED vs. IOS smaller than ED in *P. kempiae*. *Raorchestes shillongensis* is different from *P. namdaphaensis*, having nostril is closer to snout in *R. shillongensis* vs. equidistant from eye and snout in *P. namdaphaensis*; TTA reaching tympanic region in *R. shillongensis* vs. TTA reaching between eye and nostril in *P. namdaphaensis*; no mid dorsal line present in *R. shillongensis* vs. present in *P. Namdaphaensis*; line on thigh and tibia absent in *R. Shillongensis* vs. present in *P. Namdaphaensis*. *Raorchestes shillongensis* is different from *P. dubius* in absence of papilla in tongue; snout pointed in *R. shillongensis* vs. snout rounded in *P. dubius*. *Raorchestes shillongensis* different from *P. kempii* having a distinct supra-tympanic fold (vs. absent supra-tympanic fold); snout pointed in *R. shillongensis* vs. snout rounded in *P. kempii*; TTA reaches tympanic region in *R. shillongensis* vs. tip of snout in *P. kempii*.

Table 1: Comparative morphometric measurements of *Raorchestes shillongensis*, *Raorchestes sp. 1* and *Raorchestes sp. 2*.

Body part	<i>R. shillongensis</i> (F), n= 12			<i>R. shillongensis</i> (M), n=25			<i>Raorchestes sp. 1</i> (M) n=15			<i>Raorchestes sp. 2</i> (M), n=2		
	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD
SVL	15.99-21.26	18.47	1.6	14.21-19.09	16.51	1.29	16.23-20.35	18.1	1.17	17.81-20.08	18.95	1.61
HW	5.75-7.23	6.45	0.53	4.33-6.68	5.81	0.53	5.8-7.57	6.67	0.58	6.71-7.32	7.02	0.43
HL	5.79-7.24	6.65	0.49	5.09-7.05	5.9	0.49	6.17-7.18	6.79	0.32	6.2-7.34	6.77	0.81
ED	2.07-2.76	2.39	0.2	1.56-2.68	2.08	0.3	2.22-3.21	2.66	0.33	2.41	2.41	0
SL	2.1-3.23	2.58	0.39	1.72-2.91	2.19	0.32	2.09-3.06	2.61	0.3	2.39-2.88	2.64	0.35
AJS	4.47-6.02	5.41	0.51	3.94-5.7	4.92	0.4	5.12-6.17	5.65	0.32	5.58-6.11	5.85	0.37
IOS	2.01-3.27	2.54	0.37	1.47-2.98	2.23	0.33	2.03-3.08	2.58	0.25	2.11-2.21	2.16	0.07
UEW	1.08-1.61	1.32	0.18	0.84-2.46	1.23	0.31	1-2.03	1.4	0.31	1.35-1.59	1.47	0.17
AG	7.74-12.7	9.92	1.25	6.44-10.62	8.39	1.11	7.86-10.67	9.03	0.63	9.56-12.46	11.01	2.05
FLL	10.61-12.94	11.59	0.75	8.16-12.91	11.34	1.27	9.83-18.75	12.71	2.05	12.77-14.21	13.49	1.02
FI	2.11-3.57	2.81	0.52	1.56-3.84	2.77	0.62	1.79-4.1	2.6	0.6	2.39-3.62	3.01	0.87

F I DW	0.43-1	0.75	0.16	0.31-1	0.63	0.17	0.47-0.77	0.6	0.1	0.72-0.82	0.77	0.07
F II	3.29-5.49	4.4	0.63	2.76-5.59	4.25	0.68	2.99-5.16	3.85	0.62	3.76-5.57	4.67	1.28
F II DW	0.45-1.46	0.98	0.3	0.4-1.43	0.81	0.31	0.56-1.05	0.79	0.14	0.87-1.15	1.01	0.2
F III	4.97-8.65	6.76	1.1	4.82-8.66	6.71	0.93	4.43-7.53	5.95	1.07	5.89-8.6	7.25	1.92
F III DW	0.81-1.75	1.31	0.29	0.38-1.75	1.13	0.41	0.91-1.53	1.16	0.19	1.31-1.99	1.65	0.48
F IV	4.21-7.87	5.9	1.08	3.76-7.34	5.68	0.9	3.48-6.27	4.94	0.86	5.05-6.88	5.97	1.29
F IV DW	0.56-1.84	1.18	0.37	0.4-1.76	1.01	0.36	0.72-1.4	0.99	0.18	1.35-1.42	1.39	0.05
TBL	7.8-9.68	8.73	0.58	6.37-8.66	7.76	0.57	8.5-10.41	9.3	0.52	10.65-11.02	10.84	0.26
TL	5.99-8.37	7.18	0.76	5.37-7.82	6.5	0.53	6.86	7.68	0.46	7.09-8.42	7.76	0.94
HLL	23.92-30.03	26.92	2.17	21.92-27.66	24.63	1.98	26.73-31.52	29.12	1.55	30.62-32.64	31.63	1.43
T I	1.61-4.26	2.72	0.79	1.64-2.98	2.47	0.4	1.66-3.34	2.24	0.49	1.9-2.03	1.97	0.09
T I DW	0.49-1.02	0.71	0.19	0.36-0.96	0.61	0.16	0.4-0.82	0.55	0.11	0.73-0.76	0.75	0.02
T II	2.96-5.93	4.08	0.89	2.61-4.47	3.73	0.54	2.56-4.82	3.5	0.64	3.38-3.76	3.57	0.27
T II DW	0.54-1.19	0.84	0.21	0.35-1.07	0.7	0.2	0.48-0.97	0.68	0.12	0.6-0.7	0.65	0.07
T III	4.47-8.9	6.45	1.17	4.52-7.12	5.87	0.85	4.23-7.69	5.47	0.98	5.93-6.75	6.34	0.58
T III DW	0.43-1.37	0.89	0.28	0.33-1.03	0.76	0.22	0.54-0.92	0.74	0.12	0.91-0.92	0.92	0.01
T IV	5.9-10.92	8.76	1.5	6.05-10.43	8.04	1.28	6.25-11.11	7.53	1.27	8.23-8.78	8.51	0.39
T IV DW	0.51-1.76	0.97	0.36	0.43-1.33	0.89	0.26	0.65-1.15	0.88	0.13	1.09-1.2	1.15	0.08
T V	5.23-9.64	7.07	1.22	4.74-8.37	6.57	0.91	4.75-8.55	6.04	1.01	6.4-6.52	6.46	0.08
T V DW	0.39-1.53	0.99	0.35	0.35-1.32	0.8	0.3	0.42-1.11	0.81	0.18	0.69-0.84	0.77	0.11

Table 2: Mensural and meristic data for adults of *R. shillongensis*, compared with other Bush frogs those occurs in Northeast India. References: **A.** Male and female SVL (range is given in mm), **B.** ED- less than IOS (0), more than IOS (1), equal to IOS (2); **C.** Line between eyelids- absent (0), faint (1), prominent (2); **D.** Nostril- equidistant from eye and snout (0), closer to snout (1); **E.** Snout- pointed (0), rounded (1); **F.** Tympanum- indistinct (0), distinct (1); **G.** Tympanic fold – absent (0), distinct (1); **H.** Tongue shape- lobed (0), notched (1); **I.** TTA reaching- tip of snout (0), nostril (1), middle of eye (2), anterior corner of eye (3), between nostril and eye (4), tympanic region (5); **J.** Mid-dorsal line/ridge- absent (0), present on head and forebody (1), present on snout to vent (2); **K.** Line/ridge on thigh and tibia- absent (0), present (1). **L.** Inner meta-tarsal tubercle- absent (0), present (1); **M.** Outer meta-tarsal tubercle- absent (0), present (1).

Species	A	B	C	D	E	F	G	H	I	J	K	L	M
<i>R. shillongensis</i> (M)	14.24-19.09 (25)	0	0,1	1	0	1	1	1	5	0,1	0	0	0
<i>R. shillongensis</i> (F)	15.99-21.26 (12)	0	0,1	1	0	1	1	1	5	0,1	0	0	0
<i>Raorchestes</i> sp. 1 (M)	16.23- 20.35 (15)	1	0,1	1	0	0	1	1	2	2	1	0	0
<i>Raorchestes</i> sp.2 (M)	17.81-20.08 (2)	1	1	1	1	0	1	1	2	0	0	0	0
<i>Raorchestes</i> sp.2 (F)	22.14 (1)	1	1	1	1	0	1	1	2	0	0	0	0
<i>R. sahai</i> *	25-26	1	0	0	1	1	-	1	2	0	0	1	0
<i>R. manipurensis</i> *	25	2	0	0	0	1	1	0	1	0	0	1	0
<i>R. annandalii</i> *	16-20	0	1	0	0	1	1	-	0	0	0	0	0
<i>P. garo</i> *	13-16	0	0	0	0	1	1	-	3	0	0	1	0
<i>P. kempiae</i> *	17-18	1	2	0	1	0	1	-	0	0	0	0	0
<i>P. namdaphaensis</i> *	28	0	2	0	0	0	1	-	4	0	1	1	0
<i>P. dubius</i> *	43	-	0	0	1	1	1	-	2	0	0	1	0
<i>P. kempii</i> *	15	-	0	1	1	1	0	-	0	0	0	0	0
<i>P. microdiscus</i> *	29	2	0	1	1	1	1	-	2	0	0	1	0

‘*’ taken from available literature, ‘-’ not mentioned.

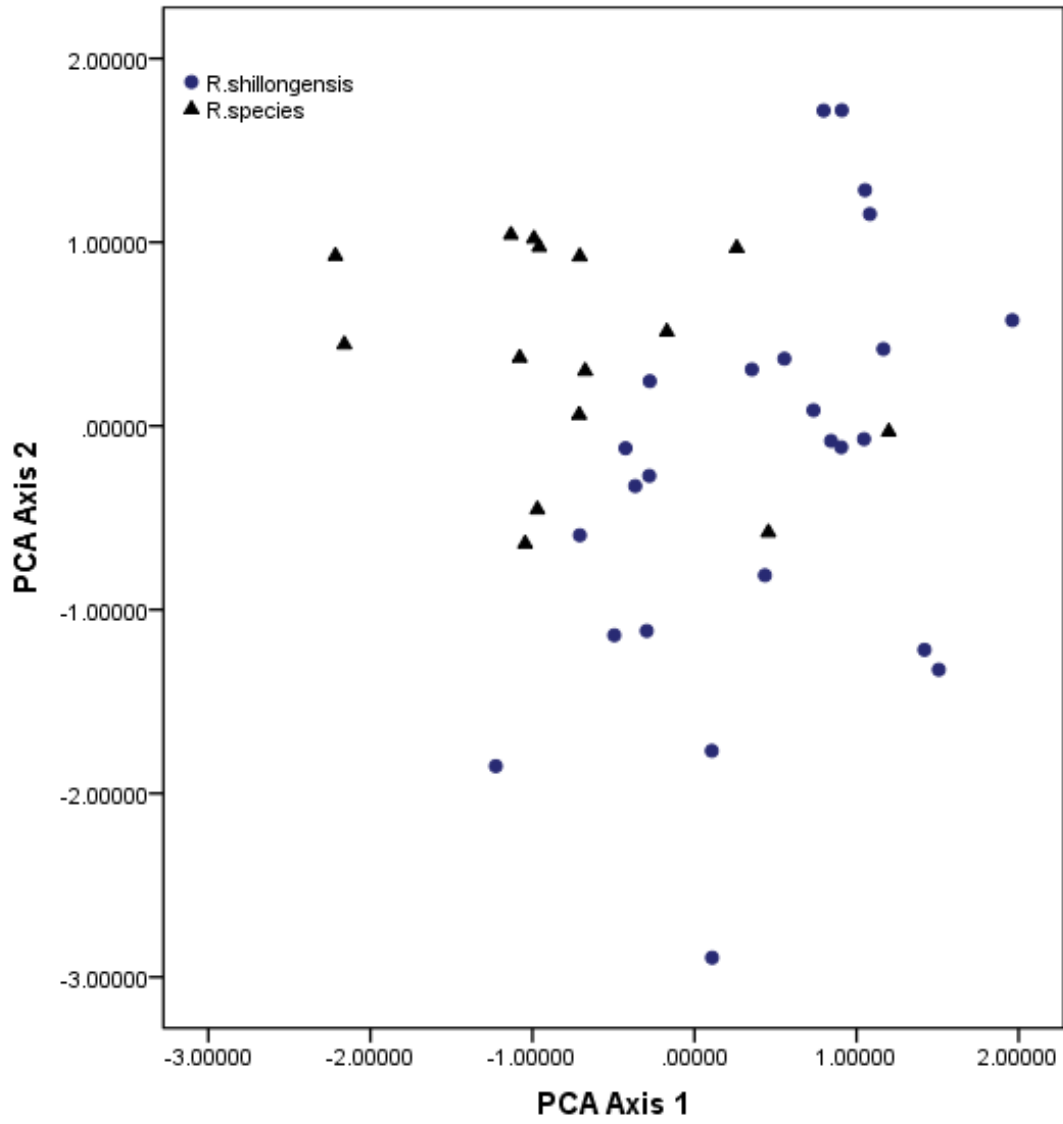


Figure 6: Result of PCA of morphometric data for *Raorchestes shillongensis* and *Raorchestes sp.1*.

7.9. Reproductive Biology

7.9.1. Breeding period: The species comes out with the onset of raining in April and their decreases towards August. Habitat of the species varies from human habitation to forested areas. We observed their breeding from ends of May to July during the study period. We made total five observations on breeding behaviour of *R. shillongensis* (29.05.2016/ 02.06.2016/ 06.06.2016/ 25.06.2016/ 07.07.2016). All observations were carried out in natural habitat in two different sites of Malki forest. Four observations made near ZSI Shillong, Risa Colony and one is 1km distant from it towards east. Details of the observations are given in the **Table 4**.

7.9.2. Male vocalization: Calling males come out just beginning of dusk (18.30 hrs) and calling is more frequent up to 24.00 hrs. Calling of males also can be heard from the bushy thickets by day. Advertisement calls delivered in call groups (**Figure 7**) consists of two to multiple calls. Duration of the call bout analysed was 22.62s and number of call group was five with call numbers varying 3-5. Duration of call group was 0.72 ± 0.21 s (n=5). Duration of call group varies with the number of calls. Inter call group interval was 4.76 ± 0.76 s (n=4). Intra call group interval was 0.19 ± 0.02 s (n=14). Call had a single pulse (non-pulsatile) for a short moment of 0.05 ± 0.01 s (n=19). Overall peak frequency of the calls was 3.62 kHz.

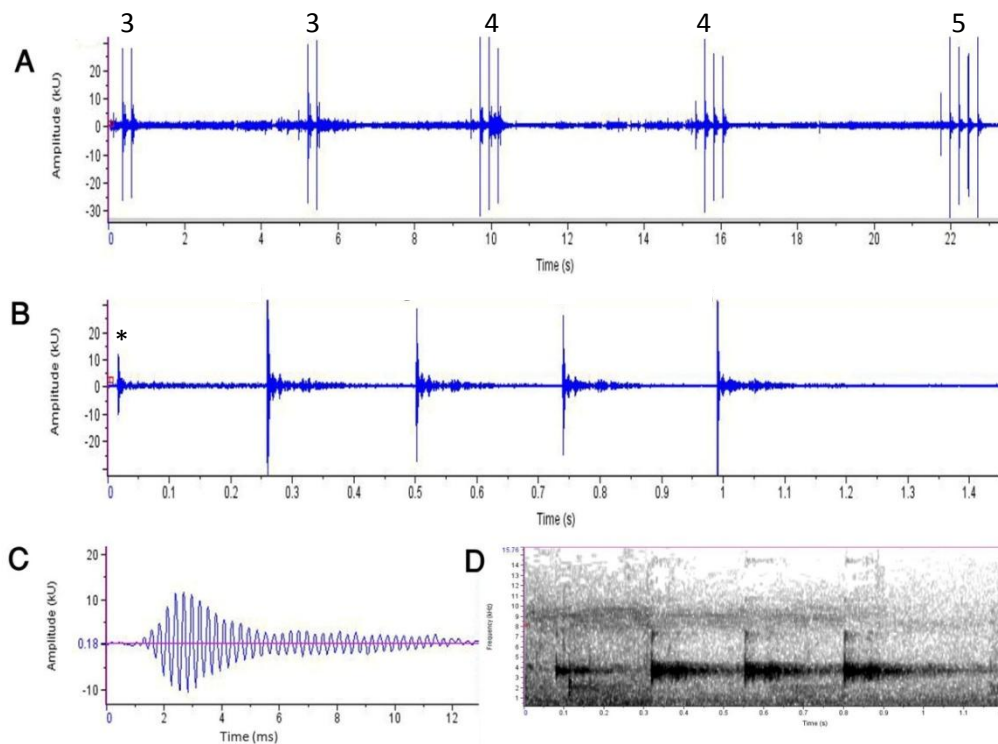


Figure 7: Advertisement call of *R. shillongensis* at ambient air temperature 24.1°C. **A.** A call bout comprising of five call groups (digits indicated number of calls in the respective call groups). **B.** Showing of 5th (last) call group with five calls depicted in A. **C.** Showing 1st call (non-pulsatile) depicted in B (indicated by asterisks). **D.** Spectrogram of 4th call group with four calls depicted in A.

7.9.3. Reproductive strategy: Description of the reproductive behaviour is based on all five observation. At the evening males aggregated and started coarse in the bushes of human habitation upto forested areas. They calls intensively after the dusk (18.00-18.30hrs) and declines towards late night (after 24.00 hrs). Males perch and calls from branches or leaves of shrubs. Male's perch height is $69.2 \pm 38.73\text{cm}$ (n=73) and female's perch height is $52.53 \pm 42.02\text{cm}$ (n=15) above ground (**Figure 8**). Males are arboreal and descend with the amplexant female only to fertilize the eggs. However, we did not observe any competition of among males during their amplexus. They starts amplexus in the arboreal habitat and lays egg on ground under leaf litters in the early morning. Out of seven recognized amplexus mode of anurans (Willaert *et al.*, 2016), amplexus mode of *R. shillongensis* it is axillary.



Figure 8: Comparison of perch height between male and female individuals of *R. shillongensis*.

7.9.4. Amplexus: Courtship commences with the calling male from a perch and female slowly moves towards a calling male. Females perch usually lower substrates of shrubs. Large ripe ova in female are externally visible from lateral and ventral side. When the female reach very close to the calling male, the male quickly grasp the female and engages in axillary amplexus (**Figure 9**). The hind limb of the amplexant male remains attached on the hind limb of the amplexant female. Amplexus starts at late evening between 21.05 hrs to 23.40 hrs. Once the pair comes into physical contact the amplexant male ceases calling, but in one case amplexant male continued calling with low pitch for initial four minutes. The amplexus pair remains in the arboreal habitat for 4.30-8 hours. Between 4.00 hrs to 5.45 hrs the amplexant descends to the ground and moves in search of a suitable place to lay egg.

After 1-2 hours, they enter under leaf litters. By the time, the body colour of the amplexant becomes dark and camouflage with the moist soil. This change is prominent in female rather than male. When the female finds a suitable place in the moist soil she stops moving. The female starts to rotate with her male at a small angle at either sense keeping the cloacae at a constant point and besides the rotation, she moves her head in up and downward direction. This is probably to free the egg laying place, but no pit observed at that place. In one instance, we observed the amplexant pair fell down while moving in arboreal habitat and in another case, amplexant male dislodged while entering into the leaf litters, but in both cases they resumed the amplexus quickly.

7.9.5. Oviposition and egg clutch: The amplexus lasts for 9-11 hours. Female lay 8-17 eggs on moist soil under leaf litters, early in the morning between 7.30hrs-10.10 hrs. Eggs are rounded, unpigmented and with a transparent jelly coat (**Figure 11**). Eggs lay in clump and diameter of eggs ranges 2.42-5.2mm (n=47). At the time of egg laying the female slightly lifts her body and male fertilizes the eggs. As soon as fertilization completed the male dislodges and leaves the place. The male cleans its body with limbs while leaving the place. As male left the place female starts dragging moist soil from her surrounding, rotating either side keeping cloaca constant near the laid eggs. She uses both hands to gather soil alternatively with a very short interval. After that, she mixes the egg mass with gathered soil by her legs alternatively. During this process, eggs do not get separation. Completing the process of mixing the female cleans her body as male do and leaves the place. In one case female stayed near the eggs for 4 minutes. Time taken to complete the egg laying is 20 minutes to 4.55 hours and 11-19 minutes to mix the eggs with soil. One more egg clutch containing nine eggs was found at a slope along forest trail under leaf litter on 30.07.2016.

Regression analysis showed that the female SVL is strongly correlated with clutch size ($R^2=0.81$, n=5) but correlation with egg size is not so much significant ($R^2=0.49$, n=4) (**Figure 10**).

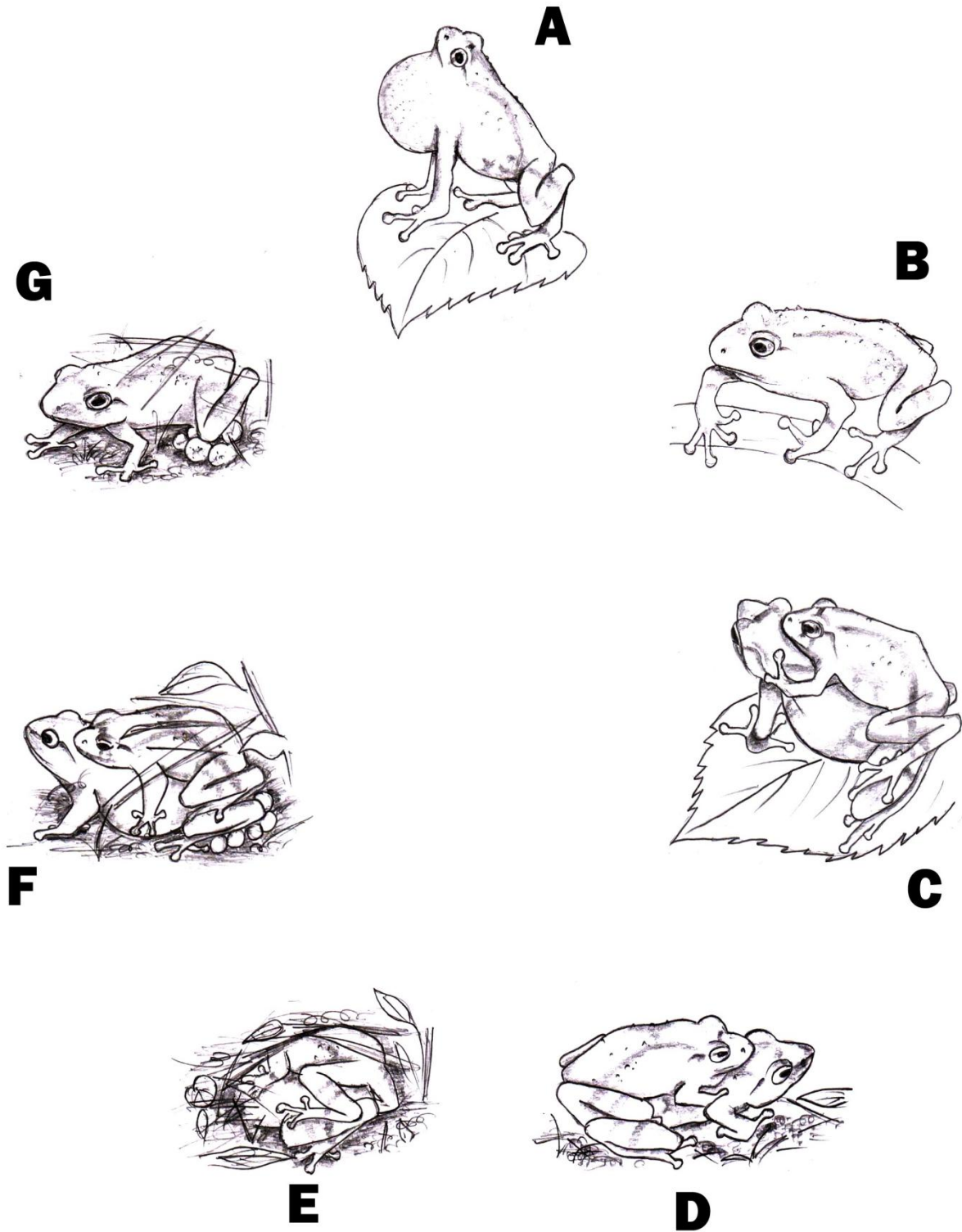
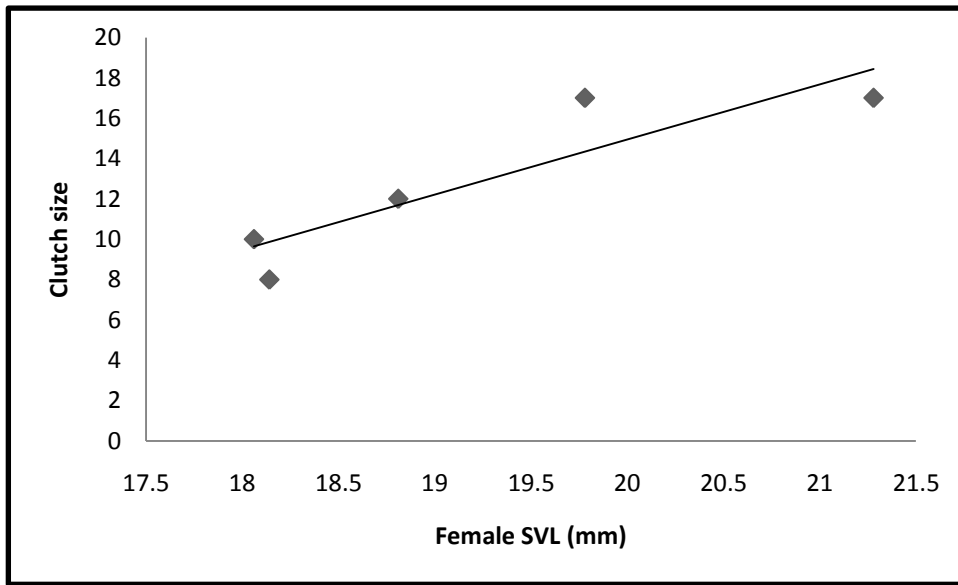
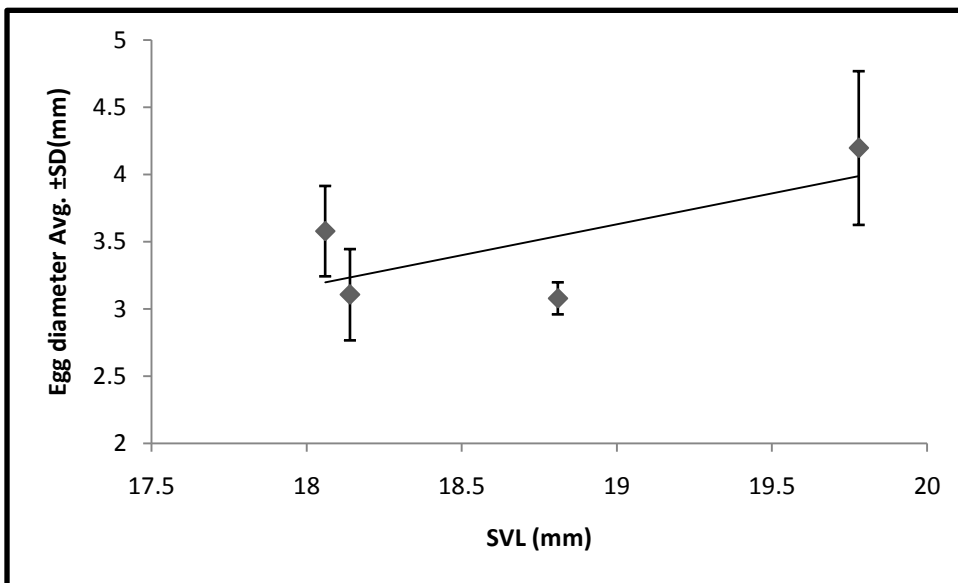


Figure 9: *R. shillongensis*. **A-** A calling male from its perch; **B-** A gravid female slowly approaching towards the calling male; **C-** Male & female in axial amplexus; **D-** Amplexant pair on ground in search of a suitable place; **E-** Amplexant pair entering under leaf litter for egg lay; **F-** Egg laying and fertilization under leaf litter; **G-** Mixing of fertilized eggs with moist soil by the female after the male dislodged.



(A)



(B)

Figure 10: **A.** female SVL shows a strong positive correlation with number of eggs ($R^2 = 0.81$) and **B.** egg size ($R^2 = 0.49$).

Table 3: Developmental stages of embryos of *R. shillongensis* (Based on clutched laid on 08.06.2016). Stages are in comparison to *Pseudophilautus viridis* from SriLanka (Bahir *et al.*, 2005).

Date	Days from the date of laid	Stage	Characteristics
19.06.2016	12	Stage 2-3	Limb buds clearly visible, unpigmented subdermal eyes, tail elongated, calcium deposition visible.
22.06.2016	15	Stage 4	Eyes large and black, pigment on dorsal side of the yolk.
24.06.2016	17	Stage 5-6	Toe demarcation initiated, eye pupil visible.
28.06.2016	21	Stage 10	Limbs fully developed, toes and fingers visible, pigmentation covers limbs and spread towards lateral side of the yolk.
05.07.2016	28	Stage 14	Tail almost absent, little amount of yolk present, looks like adult.

7.9.6. Development: As in other *Raorchestes* species, development in *R. shillongensis* is direct; i.e. without free swimming tadpole stage (**Figure 11**). Incubation or developing period is 30.5 ± 0.71 days (n=2) (**Table 5**). Rate of successful hatchlings is 100% (n=2). Body colour of the froglet is brown and slightly blackish towards lateral side of the belly or brown with dark)-(shape mark on back as in **Morph A**. Three more Froglets were found at base of shrubs (ground level) at evening time on 06.07.16.

7.9.7. Male –male combat: We did not observe any competition of males for amplexus. However, we got one male-male combat pair on 10.06.2016, which lasted for 27 minutes at edge of a forest trail (**Figure 12**). Initially they were at a perch height of 70cm above the ground. The aggression call was different from the advertisement call with low pitch rapidly repeated notes. After few minutes they fell down on the clays and litters. Again, they stood on their hind limbs and grasped each other by touching chest and sometime in reverse direction. One grasped on head while another one grasped at the axial portion and called continuously with the puffing vocal sac. Beside this, they avoid each other and release advertisement call and resume fighting. However, this could be territorial behaviour because we did not see any female in their surroundings.

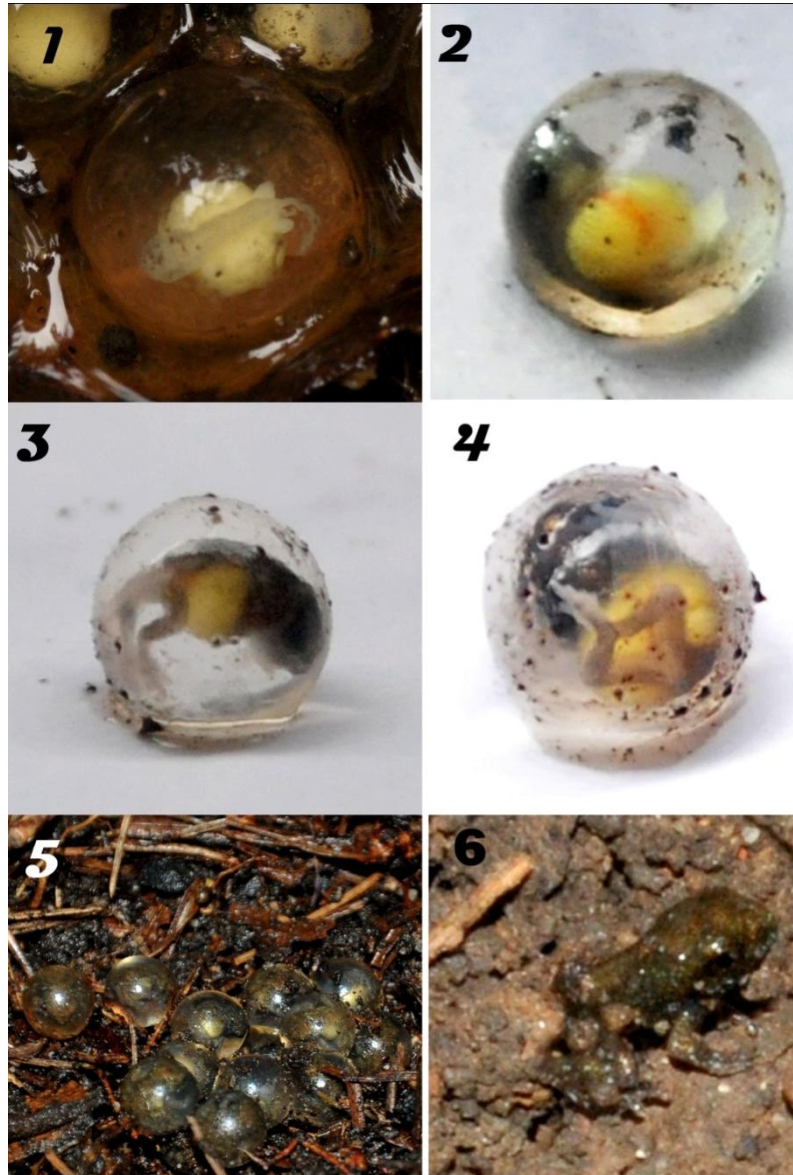


Figure 11: Developing embryo of *R. shillongensis* 1. Stage 2-3; 2. Stage 5-6; 3 & 4. Stage 10; 5. Stage 14; 6. A newly hatched froglet.



Figure 12: Male-male combat of *R. shillongensis*. **1.** Mounting of one by another male touching the bellies and in reverse direction; **2.** One male (above) grasping the leg of the other (below); **3.** Aggression call with the buffing vocal sac; **4.** Grasping one another by standing on foot and touching the chests.

Table 4: Observations on breeding of *R. shillongensis* during the study period.

Date	Temp/ Humidity (°c/%)	Perch height (M/F) (cm)	SVL (M/F) (mm)	Distance between M & F (cm)	Amplexus start time	Time of descending to ground	Time of entering under leaf litters	Time egg laying completion	Male dislodge time	Time of completion of egg mixing process	Female Leaving time	Clutch size	L/B of clutch (mm)
29.05.16	17.1/ 93	51/120	17.56/18.06	100	21.05	05.00	07.05	08.00	08.02	08.20	08.23	10	12.58/10.23
02.06.16	17.8/ 92	63/40	16.37/18.81	23	21.50	05.45	07.35	07.55	07.57	08.10	08.13	12	19.44/16.67
07.06.16	18.3/89	70 (same leaf)	18.23/21.28	very close	21.15	05.15	06.55	07.30	07.31	07.45	07.50	17	19.01/11.51
25.06.16	20.8/92	52/78	17.73/18.14	30	22.25	05.30	07.20	08.10	08.13	08.37	09.05	8	14.51/9.04
06.07.16	21.6/82	77/50	16.85/19.78	60	23.40	04.00	05.15	10.10	10.14	10.25	10.27	17	15.66/13.72

Table 5: Different reproductive modes in Bush frogs of India and Sri Lanka.

Species name	SVL (mm)	Clutch size	Days to hatch	Mode	Habitat	Parental care	Egg diameter (mm)	Locality
<i>R. shillongensis</i>	♀: 18.47 ±1.6 (12) ♂: 16.5 ±1.31 (25)	8-17	30-31 (2)	17	On moist soil under leaf litters	Male leaves after fertilization and female leaves after egg mixing with soil	3.59 ±0.63 (47)	Malki Forest Shillong, Meghalaya
<i>Philautus cf. leucorhinus</i> *	♀: 33.7 ♂: 28.9	51	19	20	Above ground, (10cm) on wet leaves, between rocks	Pair separates after spawning	3.5 ±0.16 (51)	Karnataka
<i>P. glandulosus</i>	♀: 24.5-26 ♂: 20-22.9	41	28	20	Above ground (1.5-3m) on wet leaves	Pair separates after spawning	4.4 ±0.2 (48)	Waynaad, Kerala
<i>P. variabilis</i> *	♀ and ♂: 30.0 ±4.5	54-62		20	Above ground	Eggs beneath abdomen of female, chasing intruding males	4.1 ±0.2 (30)	Karnataka
<i>P. nerostagona</i>	♀: no report ♂: 30.1-34	41	20	20	Above ground (10m) in tree hole (10cm deep)	-	4.5 ±0.3 (41)	Waynaad, Kerala
<i>P. tinniens</i>	♀: 25 ♂: no report	-	-	17	On ground	-	-	Nilgiri hills, Tamil Nadu
<i>R. bombayensis</i>	♀: no report ♂: no report	26-27	-	20	Above ground on wet leaves	-	-	Karnataka
<i>P. bobingeri</i>	♀: 23.5-26 ♂: 21.3-24.8	24	18	20	Above ground (4m) on Acacia tree	-	3.9 ±0.4 (24)	Ponmudi hills, Kerala
<i>P. graminirupes</i>	♀: 27.3-29.4 ♂: 21.4-22.6	30-38	24	17	On ground, grass clump, rocky crevice	-	4.9 ±0.5 (38)	Ponmudi hills, Kerala

<i>R. resplendens</i>	♀: 25.2–28.3 ♂: 22.7–24.5	18-28		17	under moss cover forest floor	Leaves after spawning	4.1 ±0.4 (24)	Eravikulam National Park, Western Ghats
<i>R. ochlandrae</i>	♀: 23.3 ♂: 22.1–25.6	6	-	20	Above ground, inside bamboo internode	Male attends the eggs until hatch	4.94 ± 0.06 (developing embryo)	Kerala
<i>R. chalazodes</i>	♀: 25.2 ♂: 23.7 ±2.66 (3)	5-8		20	Above ground (25cm), inside bamboo internode	Male attends the eggs until hatch	5.73 ±0.66 (28)	Kalakad Mundanthurai Tiger Reserve, Western Ghats
16 species of <i>Pseudophilautus</i> #	♀: ♂:	6-155	24-68	17	In soil cavity (1.5-50 cm ³)	Male voluntarily departures, female abandons after concealing the eggs	3.7-5.7	Sri Lanka
<i>Pseudophilautus femoralis</i>	♀: ♂:	7-22	37-49	20	on underside of leaf above ground (0.3-2m)	after male dislodged female sits on the eggs for 1-3 hrs	-	Sri Lanka
<i>P. regius</i>	♀: ♂:	17	-	17	Soil cavity	Female burrows the eggs and leaves	3.1 (17)	Sri Lanka

Reproductive mode follows Duellman & Trueb 1994, * now referred to *Pseudophilautus wynaadensis* (see Frost 2016), # see Bahir *et al.* (2005).

7.10. Habitat ecology

The species (*Raorchestes shillongensis*) started emerging with the onset of raining in the last week of April. During this period of survey the species is found to be quite common in all habitats of the region, mainly in the Shillong city. Individuals were encountered from human settlements, garden, road side, stream sides to forested area. Adults were recorded perching from 8cm to a maximum height of 183 cm above ground. Individuals perches on leaves, climbers, branches of small tress and trunk of Pine trees. We plotted total twenty quadrates (5m×5m) in four different habitats. Sampling habitat types were forest edge, near human habitation, along stream and inside the pine forest in Malki forest. A total 129 individuals of *R. shillongensis* were observed from four habitat types during the survey. The highest number of individuals encountered in forest edge (34.88%) followed by human habitation (30.23%), stream side (24.03%) and lowest in inside forest area (10.85%) (**Table 6**).

Study shows that the most preferred habitat of the species is forest edge and human habitation. In these habitats plant like Eupatorium, Melostoma are abundant. These habitats provide a significant canopy cover and thick leaf litter layer for this species, which avoids the desiccation of direct developing eggs and keeps moisture. In many species, vocal advertisement represents the most important characteristic of males during the adult phase of the life cycle (Ryan 1983; Pough et al.1992), especially in amphibian group. Furthermore, calls increase the probability of being exposed to predators (Dissanayake and Wellapuli-Arachchi 2012). The canopy cover (>70%) and a moist thick leaf litter layer (20 mm) are important to resist desiccation of their direct developing eggs (Bahir et al. 2005; Karunarathne and Amarasinghe 2007, Dissanayake and Wellapuli-Arachchi 2012). Stream edges are mostly rocky and area deep into the forest (mainly top regions) have less shrubs, mainly occupied by ginger, thorny climbers and some tall grass of narrow leaf. So, due to lack of good canopy cover, moist soil and thick leaf litter layer, these habitats are less preferred by the species.

Table 6: Individual encounter in different habitat types.

Habitat type	Individuals encounter range	Average individuals
Forest edge	8-11	9 ±1.22
Near human habitation	2-12	7.8 ±3.77
Along stream	3-10	6.2 ±2.77
Inside pine forest	0-8	2.8 ±3.11

7.11. Distribution

We added about 82 new localities in the distribution range of the species (**Appendix: 1**) which covers 531 sq. Km (**Figure 13**). The present distribution include Mawlai reserve forest, Laitkor reserve forest, Mawpat reserve forest, Upper Shillong reserve forest along with some community conserved and semi protected forest like Mawphlang Sacred Grove and North Eastern Hill University (NEHU) campus. Some tourist spots within the distribution range of *R. shillongensis* are Cherrapunjee, Elephant falls, Shillong Peak, Happy Valley, Dynkhlyin. The species is restricted to an altitudinal range from 1087m to 1924m asl within the study area.

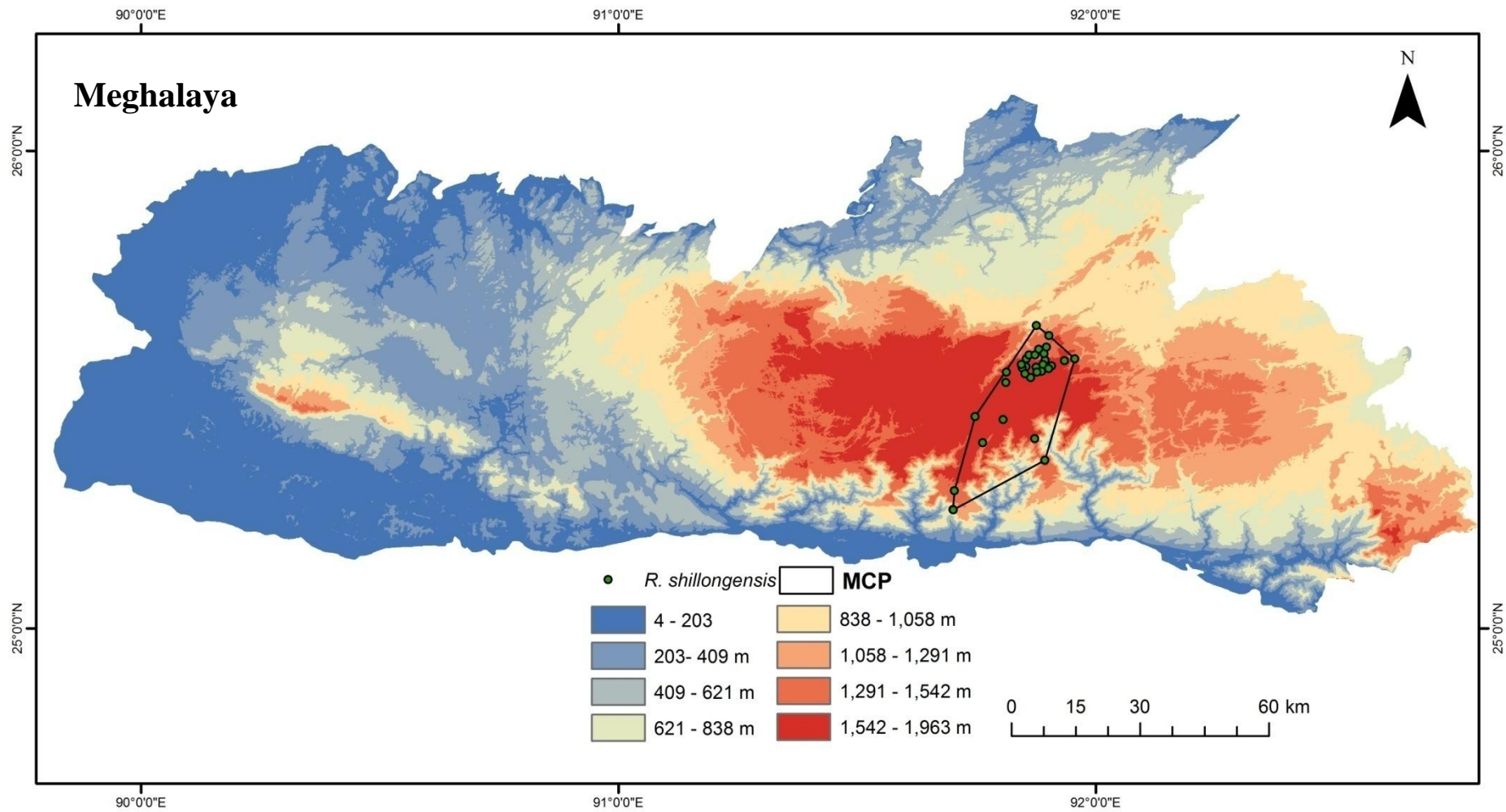


FIGURE 8: Current distribution map of *Raorchestes shillongensis*.

8. Discussion

This study reconfirms *R. shillongensis* based on morphological, acoustical characteristics, field observation along with molecular analysis. Description includes presence or absence of morphological characters, which are not mentioned in the original description of the species. Moreover, our study provides first ever information on distribution of the species. Study limits the current distribution of the species as endemic to the East Khasi Hills district. Within its political boundary Mawlai reserve forest and Cherrapunjee limits the species as its northern and southern boundary respectively within an elevation range 1000m to 1900m, as species from low elevation sites, Riwai and Mawlynnong (extreme south of the Khasi hills) and Nonkhyllem wildlife sanctuary and Badapani (Ri-voi district) are found completely different from species *R. shillongensis*. Mathew and Sen (2010) showed its distribution in Mizoram, but there is no clear explanation. Therefore, we have not included this locality in the distribution of the species. Again, Mathew and Sen (2009) reported *P. garo* based on morphology from the ZSI, Risa Colony, Shillong, which is near the type locality of *R. shillongensis*. It could be misidentified because the population of *R. shillongensis* highly polymorphic and some morph looks similar to *P. garo*. Our molecular study shows that the population from the Shillong is *R. shillongensis*. Therefore, we assigned the specimen identified by Mathew and Sen to *R. shillongensis*.

Molecular phylogenetic study revealed that *R. shillongensis* is closely related to South Asian clade (*Raorchestes*) than East Asian Clade (*Philautus*). Within the *Raorchestes* clade *R. shillongensis* is nested with *Raorchestes longchuanensis* of China rather than other congeneric species of Western Ghats of India.

The reproductive biology of the Rhacophorid group comprised of a diverse reproductive modes, ranging from foam nests and free-feeding tadpoles to lecithotrophy and direct development (Brown & Alcalá 1982, 1994; Grosjean et al. 2008). Terrestrial direct development in the genera *Philautus* and *Raorchestes*, evolved independently, i.e., the eggs undergoes direct development and hatch into tiny froglets avoiding the free swimming tadpole stage (Bossuyt & Dubois 2001; Bahir et al. 2005; Gururaja and Ramachandra 2006; Grosjean et al. 2008; Li et al. 2009; Biju et al. 2010). There are 40 types of globally recognized reproductive mode in amphibians (Gururaja 2010). Recently Willaert *et al.* described a new mode of reproduction in *Nyctibatrachus humayuni* from Maharashtra. The present study documents the breeding behaviour of *R. shillongensis* in the wild and first report for this group of frogs from Northeast India. According to Duellman and Trueb (1994),

breeding mode of *R. shillongensis* belongs to type 17. Clutch size of *R. shillongensis* is almost equal to *R. chalazodes* and *R. ochlandrae* but developmental duration is longer than that of so far known Indian bush frogs. It could be due to cold and continuous rainfall in the region. Amplexus mode which is axillary, egg colour and size is same with other Bush frogs. Reproductive mode *R. shillongensis* is slightly similar (mode 17) with *R. tinniens*, *R. graminirupes* and *R. resplendens*. Again, *R. resplendens* lay eggs under moss covered forest floor, deep inside the base of bamboo clumps (Biju *et al.*, 2010), *R. graminirupes* lay eggs on ground, grass clump, rocky crevices, *R. tinniens* lays in deep hole on ground (Gururaja and Ramachandra, 2006), while *R. shillongensis* lays eggs on moist under leaf litters. However, mixing of eggs with moist soil is not previously observed in any bush frogs of India. Breeding behaviour of *P. variabilis* and *P. cf. leucorhinus* by Kanamadi *et al.* (1996) and Gururaja and Ramachandra (2006) respectively which are now referred to *Pseudophilautus wynnadensis*. In both description they mentioned that the species lay eggs in vegetation above ground. However, there are some difference between the descriptions as in *P. variabilis*, female guarded the eggs and chase the males (Kanamadi *et al.*, 1996) while in Gururaja and Ramachandra's description there is no any parental care in *P. cf. leucorhinus*. *R. gladulosus* and *R. bombayensis*, *R. nerostagona*, *R. bobingeri* deposits eggs above the ground on arboreal habitats (mode 20). We did not observe any parental care in *R. shillongensis* like in *R. chalazodes* and *R. ochlandrae* as they stay with the eggs until hatched. Breeding behaviour of *R. shillongensis* shows some similarity with some ground nesting *Pseudophilautus* sp. of Sri Lanka as they mix the eggs with soil, probably for better distribution of sperms (Bahir *et al.*, 2005). However, *R. shillongensis* do not excavate soil for egg lay and egg separation does not occur during mixing with soil as in Sri Lankan *Pseudophilautus* (Bahir *et al.*, 2005). Clutch size of *R. shillongensis* is relatively small than other bush frogs except *R. chalazodes*, *R. ochlandrae* and Sri Lankan bush frog *Pseudophilautus regius*. The body colour of *R. shillongensis* changes to dark camouflaging with soil colour during ground dwelling for egg lay, which is not reported in any Indian bush frogs except Sri Lankan ground nesting bush frogs (Bahir *et al.*, 2005). The colour change of ground-nesting *Philautus* during egg laying is probably to reduce the predation risk, especially these frogs often nest in daytime (Bahir *et al.*, 2005). As in some other frog, in *R. shillongensis* has also significant positive correlation between egg diameter and female's body size (SVL).

However, *R. shillongensis* is distributed within a small range (approximately 530 sq. km) in the East Khasi Hills and relatively abundant in the backyards and forest edges. During the

study period we observed that the habitat of the Malki forest and adjacent habitats are rapidly degrading due to various anthropogenic activities like fire wood collection, intentional forest fire, excessive use of detergent for cloth washing in the forest streams (Mahony *et al.*, 2013), garbage deposition by the local people as well as in the tourist spots etc. Cleaning of forest as well as bushes and use of chemicals for cultivation in the surroundings of Malki and Upper Shillong forests are responsible for the viable survival and declination of this endemic bush frog.

9. Conservation and awareness

Shillong Plateau is well known for high endemism of amphibian species, but national reserves and other government-protected forests of the East Khasi Hills district (2,752 km²) represent only 18.65 km² in seven locations, the largest forest is the Upper Shillong Protected Forest with only 7.66 km² area (Mahony *et al.*, 2013). Additionally, during the survey period we recorded other 31 species of herpetofauna from the region (Appendix: 2). However, Illegal tree felling, unregulated mining in protected and unprotected areas in the state (Gilbert, 2012), as well as in the Shillong Plateau may have considerable affect in the survival of amphibian fauna of the region (Mahony *et al.* 2013). Therefore, awareness among the local people and conservation of these last remaining forests is urgent, which are the typical habitats of the threatened endemic species of the region.

We made a short documentary on the species, *Raorchestes shillongensis*, which is the first documentary on amphibian from Northeast India. It is hoped that this will be helpful for the awareness among the common people and young generations and towards the conservation of the habitats as well as the species.

10. Conclusion

Recently several phylogenetic studies indicate the presence of two prominent clades and several subclades within the genus *Raorchestes* (Biju et al. 2010, Abraham et al. 2013, Vijayakumar et al. 2014). So future studies should focus on the phylogeny and taxonomy of the Bush frog clades *Philautus*, *Pseudophilautus* and *Raorchestes* (Li, J. 2011). Moreover, Northeast India being recognized as one of the major centres of amphibian diversity, but only the caecilian fauna of this region has been studied within a phylogenetic framework so far (Kamei et al. 2012). However, this finding demonstrates the poor scientific knowledge we have about biogeography and taxonomy of the genus *Raorchestes* in the Indo Chinese region. Future research on this group from the region could uncover many undescribed species of this group.

11. Way ahead

1. A long term study to estimate the population size, document the fluctuation of *R. shillongensis*.
2. Projecting it as a habitat indicator species for Shillong City.
3. Citizen Science approach in conservation and monitoring of this species.

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Appendix 1: Gazetteer list of distribution of *Raorchestes shillongensis* in Meghalaya with locality records, GPS coordinates, elevation and locality status (**P**- Protected forest, **UP**- Unprotected area, **TP**-Tourist place, **CP**- Community protected forest, **SP**- Semi-protected area).

Sl. No.	Latitude	Longitude	Elevation m (asl)	Locality	Status of the locality
1.	25°32'20.2"	91°51'3.9"	1853	Shillong Peak	P
2.	25°32'22.3"	91°51'2.5"	1800	Upper Shillong forest	P
3.	25°32'23.8"	91°50'59.1"	1791	Upper Shillong forest	P
4.	25°32'26.8"	91°50'59.7"	1803	Upper Shillong forest	P
5.	25°32'42.2"	91°51'14.4"	1682	Upper Shillong forest	P
6.	25°32'43"	91°51'13.4"	1711	Upper Shillong forest	P
7.	25°32'54"	91°51'14.6"	1694	Upper Shillong forest	P
8.	25°32'53.5"	91°51'3.9"	1676	Upper Shillong forest	P
9.	25°32'44.7"	91°50'46.1"	1781	Upper Shillong forest	P
10.	25°32'51.9"	91°51'1.1"	1747	Upper Shillong forest	P
11.	25°33'41.8"	91°53'39.1"	1577	Risa Colony	UP
12.	25°33'34.2"	91°53'42.8"	1600	Risa Colony	UP
13.	25°33'16.5"	91°53'59.1"	1707	Motinagar Forest	P
14.	25°33'24.7"	91°53'56.4"	1634	Motinagar Forest	P
15.	25°33'33.6"	91°53'51.8"	1606	Motinagar Forest	P
16.	25°33'23.7"	91°53'46.3"	1625	Motinagar Forest	P
17.	25°33'25.8"	91°53'54.2"	1607	Motinagar Forest	P

18.	25°33'19.8"	91°53'50.4"	1659	Motinagar Forest	P
19.	25°33'16.3"	91°53'51.7"	1675	Motinagar Forest	P
20.	25°33'14.6"	91°53'51.5"	1707	Motinagar Forest	P
21.	25°33'10.8"	91°53'52.7"	1747	Motinagar Forest	P
22.	25°33'6.5"	91°53'46.6"	1783	Forest Near Durdarshan centre	P
23.	25°33'39.3"	91°53'7.7"	1593	Malki Forest Office	P
24.	25°33'29.9"	91°53'3.8"	1588	Malki Stream	P
25.	25°33'25.8"	91°53'4.7"	1611	Malki Stream	P
26.	25°33'22.4"	91°53'5.5"	1613	Malki Stream	P
27.	25°33'31.7"	91°53'0"	1599	Malki Stream	P
28.	25°33'31.2"	91°53'6.6"	1621	Laitkor forest	P
29.	25°33'33.7"	91°53'11"	1599	Laitkor forest	P
30.	25°33'18.4"	91°53'15.6"	1656	Laitkor forest	P
31.	25°33'9.5"	91°53'22.6"	1785	Laitkor forest	P
32.	25°32'59.7"	91°53'20.8"	1807	Laitkor forest	P
33.	25°32'50.4"	91°53'16.3"	1882	Laitkor forest	P
34.	25°32'39.7"	91°53'18.8"	1884	Near Laitkor Nursing Centre	P
35.	25°33'24.4"	91°53'33.9"	1589	Risa Forest Stream	P
36.	25°33'15.8"	91°53'38.9"	1639	Risa Forest Stream	P
37.	25°33'11.3"	91°53'35"	1667	Risa Forest Stream	P
38.	25°33'12"	91°53'33"	1700	Risa forest	P
39.	25°33'18.2"	91°53'36.2"	1655	Risa forest	P
40.	25°33'10.6"	91°53'27"	1770	Risa forest	UP
41.	25°33'42"	91°53'2.4"	1559	Malki	P
42.	25°34'50.6"	91°53'17.5"	1418	Orchid hotel (Polo)	UP
43.	25°33'13"	91°53'32.3"	1719	Risa forest	P
44.	25°33'11"	91°53'26.2"	1762	Risa forest	P
45.	25°33'9.4"	91°53'22.6"	1784	Risa forest	P
46.	25°33'13.6"	91°53'23.4"	1753	Risa forest	P

47.	25°14'56"	91°42'3.1"	1171	Sohra	P
48.	25°14'54.6"	91°42'6.2"	1101	Sohra	P
49.	25°33'52.9"	91°51'15.4"	1607	Mawphlang Sacred Groove	CP
50.	25°33'10"	91°50'39.6"	1723	Mawphlang Sacred Groove	CP
51.	25°26'36.2"	91°44'50.7"	1815	Mawphlang Sacred Groove	CP
52.	25°26'14.5"	91°48'22"	1855	Cherrapunji road	UP
53.	25°23'19.7"	91°45'47.4"	1757	Cherrapunji road	UP
54.	25°16'31.67"	91°41'10.03"	-	Nohkalikai falls	TP
55.	25°23'51.4"	91°52'20.4"	1570	Mawlynnong road	UP
56.	25°21'8.4"	91°53'36.5"	1555	Pynursla	UP
57.	25°30'54.2"	91°48'42.4"	1709	Upper Shillong	UP
58.	25°32'11.9"	91°48'46"	1675	Sado, Upper Shillong	P
59.	25°32'49.9"	91°52'29.5"	1912	Shillong Peak	P
60.	25°33'55.4"	91°51'35.8"	1557	Upper Shillong	P
61.	25°34'8.5"	91°51'50.8"	1497	Upper Shillong	P
62.	25°34'21.3"	91°51'36.3"	1536	Nongsehrim	UP
63.	25°34'21.6"	91°52'22.5"	1507	Relebang	UP
64.	25°34'21.6"	91°52'41.4"	1500	Police Bazar	UP
65.	25°34'29.5"	91°52'46"	1464	Police Bazar	UP
66.	25°35'4.1"	91°52'50.9"	1431	Lawmali	UP
67.	25°34'31.3"	91°53'30"	1480	Municipal	UP
68.	25°34'48.4"	91°53'25.3"	1431	Polo	UP
69.	25°34'43.8"	91°53'35.2"	1440	Polo	UP
70.	25°35'21.3"	91°53'47.8"	1425	Golf Link	UP
71.	25°36'46.6"	91°54'7.2"	1400	Northeast Hill University	SP
72.	25°17'17.1"	91°42'13.6"	1418	Sai-Mika resort, Dynkhlyin	UP
73.	25°38'4"	91°52'32.6"	1087	Mawlai Forest	P
74.	25°33'53.6"	91°57'23.1"	1475	Sweet Fall	TP
75.	25°33'37.3"	91°56'3.2"	1660	Everliving Museum	UP
76.	25°32'58.3"	91°54'27"	1754	Laitkor	UP

77.	25°32'17.4"	91°53'12.5"	1872	Laitkor	UP
78.	25°32'38.2"	91°54'5.2"	1829	Laitkor	UP
79.	25°32'11.8"	91°52'37.3"	1879	Laitkor	UP
80.	25°31'31.2"	91°51'49.7"	1847	Laitkor	UP
81.	25°32'0.5"	91°51'6.3"	1924	Shillong Peak	TP
82.	25°32'13.23"	91°49'20.85"	-	Elephant Falls	TP

Appendix 2: Herpetofaunal records during the study period from the East Khasi Hills with their IUCN status and location.

Taxonomic group	Family	Scientific name	Location	IUCN status
Toads	Bufonidae	<i>Duttaphrynus melanostictus</i>	Malki forest, Mawlai forest	
		<i>Duttaphrynus himalayanus</i>	Mawphlang Sacred Groove	
		<i>Bufoides meghalayanus</i>	Riwai	
Frogs	Megophryidae	<i>Leptobrachium smithi</i>	Riwai	
		<i>Xanophrys major</i>	Mawphlang Sacred Groove	
		<i>Xanophrys oropedion</i>	Malki forest, Mawlai forest, Laitkor, Upper Shillong, Mawphlang Sacred Groove	
	Dicroglossidae	<i>Fejervarya sengupti</i>	Malki forest, Mawphlang Sacred Groove, Mawlai forest, Laitkor, Upper Shillong, NEHU campus	
		<i>Fejervarya</i> sp.	Riwai	

		<i>Hoplobatrachus tigerinus</i>	Riwai	
	Ranidae	<i>Amolops gerbillus</i>	Mawlai forest	
		<i>Odorrana mawphlangensis</i>	Malki forest, Mawphlang Sacred Groove	
		<i>Odorrana chloronata</i>	Risa colony	
		<i>Hylarana cf. leptoglossa</i>	Throughout the study area	
	Rhacophoridae	<i>Raorchestes</i> sp.	Riwai	
		<i>Nasutixalus jerdonii</i>	Mawphlang Sacred Groove	
		<i>Polypedates himalayensis</i>	Mawphlang Sacred Groove	
		<i>Polypedates</i> sp.	Riwai	
		<i>Rhacophorus bipunctatus</i>	Throughout the study area	
Lizards	Agamidae	<i>Calotes jerdoni</i>	Laitkor	
		<i>Ptyctolaemus gularis</i>	Malki forest, Upper Shillong, Laitkor, Mawlai forest, NEHU campus	
		<i>Japalura planidorsata</i>	Sohra	
	Gekkonidae	<i>Cyrtodactylus khasiensis</i>	Mawlai forest, Riwai	
		<i>Hemidactylus platyurus</i>	Risa colony	
	Scincidae	<i>Eutropis</i> sp.	Throughout the study area	
		<i>Sphenomorphus</i> sp.	Throughout the study area	
Snakes	Colubridae	<i>Pareas monticolus</i>	Risa colony, Malki forest, Laitkor	
		<i>Pseudoxenodon macrops</i>	Mawphlang Sacred Groove	

		<i>Trachischium monticola</i>	Cleve Colony, Shillong; Malki forest	
		<i>Dendrelaphis</i> sp.	Dawki	
	Viperidae	<i>Trimeresurus albolabris</i>	Malki forest, Mawlai forest, Pynursla	
		<i>Ovophis monticola</i>	Malki forest, Upper Shillong	