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Identification of anuran species diversity of the Panna Tiger Reserve, Central India, using an integrated approach

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Abstract

We present a comprehensive inventory of amphibians from Panna Tiger Reserve in Madhya Pradesh based on morphological, molecular and bioacoustic data. Representatives of 15 anuran species were collected, corresponding to roughly four fifths of the known amphibian species of Madhya Pradesh. The main results of this study are: (1) Description of advertisement calls of eleven species, including the first-time description of advertisement calls of *Sphaerotheca pashchima*. (2) Identification of cryptic species using acoustic and molecular techniques. (3) Five new significant range extensions and new state records. (4) Description of geographical variation in call properties in three anuran species. This study also provides morphological descriptions with ecological and natural history notes for each species that may be useful in management planning for amphibian conservation in Panna Tiger Reserve.

Key words: Amphibia, Anura, diversity, bioacoustics, barcoding, morphology, protected area, Central India

Introduction

Systematic species inventories considerably expand our knowledge and understanding about species richness, diversity and conservation value of the surveyed sites (Jetz & Rehbek 2002). Some less prominent taxa, however, often remain undetected in biodiversity assessments which hinder in safeguarding the overall biodiversity at regional and global level, and in apparent "coldspots" of diversity (Ballesteros-Mejia *et. al*. 2013; Kareiva & Marvier 2003). Globally, amphibians are an underrepresented group of vertebrates in terms of scientific attention within protected areas (Nori *et al*. 2015), and this applies in India as well where amphibians have been rarely inventoried in the over 50 tiger reserves of the country. It is known that amphibians may go extinct even from the most intact and best protected areas where human disturbance is minimal (McMenamin *et al*. 2008).

Panna Tiger Reserve (henceforth PTR) in Central India has been studied extensively for large carnivore mammals (Sarkar *et al*. 2016), fishes (Johnson *et al*. 2012) and birds (Gurjar & Gawande 2011). The present study aims to address the gap in knowledge in amphibians using a combined approach of morphology, molecular and bioacoustics for this reserve.

Materials and methods

Study Site: Panna Tiger Reserve (24.73430 80.01472 to 24.46878 79.88694) lies in Panna and Chhatarpur districts

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of Madhya Pradesh. PTR comprises 1578.56 km^2 of combined area of core and buffer zones (Fig 1). The vegetation consists of tropical dry deciduous forest in Vindhya Rocky Mountains. Its landscape is characterized by highlands and canyons carved through the perennial river Ken that flows for 55 km through the Reserve (Qureshi *et al.* 2006). The area has extensive rocky plateaus that are separated from each other by the river valleys. Elevation varies from 204 to 540 m above sea level in Panna plateaus. This region is characterized as dry deciduous forest and chiefly depends on monsoon rainfall from June to September with an average amount between 600 and 1,100 mm (Jayapal *et al.* 2007). The maximum recorded temperature is 48 °C in summer and minimum is 6 °C in winter.

FIGURE 1. Amphibian habitats referred in the text: A—Seasonal rainwater puddles, B—Waterlogged paddy fields, C—Rocky streams and pools, D—Ponds and E—Waterlogged open grasslands.

Field Surveys: We employed nocturnal visual encounter surveys (VES). Each VES was conducted for one hour wherein two observers walked at slow pace scanning potential habitats (Clump & Scott 1994; locations marked yellow dots in Fig 2) in habitats such as temporary rainwater filled puddles, rocky streams, ponds and marshy grasslands, paddy fields in forest fringe areas in all six ranges (management areas of PTR) (Fig 1). We conducted 86 VES (each of one-hour × two observers) between July and August each year from 2017 to 2019. To provide a basic idea of abundance of species recorded in the study, we divided the category into two subjective classes i.e. common (> 50% encountered in VES) and uncommon (<50 encountered in VES). We excluded *Uperodon variegatus* from

any category as it was recorded only once. Fieldwork was conducted from 18:00 hours to 02:00 hours in night time following Indian Standard Time. Opportunistic sightings other than records of VES are marked as black dots in the map (Fig 2). We quantified species encounter rate per hour as number of individual of a species observed over a period of one hour during VES. It was calculated as number of individual of species recorded divided by one-hour of sampling effort per observer (Chandramouli *et al*. 2015; Table 1). Relative abundance of each species was represented as the proportion of the number of individuals of that species observed across all the samples (Chandramouli *et al*. 2015; Table 1).

TABLE 1. Amphibians recorded in the study area with species encounter rate, relative abundance determined by VES, and conservation status in PTR:

S. no.	Species Name	Encounter Rate/hour	Relative abundance	Conservation Status
	Anurans			
1	*Duttaphrynus melanostictus*	0.09	0.005	LC
2	*Duttaphrynus scaber* *	2.12	0.109	LC
3	*Duttaphrynus stomaticus*	0.21	0.011	LC
4	*Euphlyctis cyanophlyctis*	7.71	0.398	LC
5	*Fejervarya orissaensis* *	0.46	0.024	LC
6	*Minervarya caperata* *	1.16	0.060	NA
7	*Minervarya pierrei* *	4.66	0.240	LC
8	*Hoplobatrachus crassus*	0.01	0.001	LC, Schedule IV (WPA, 1972), Appendix II (CITES)
9	*Hoplobatrachus tigerinus*	0.26	0.013	LC, Schedule IV (WPA, 1972), Appendix II (CITES)
10	*Microhyla nilphamariensis*	1.13	0.058	NA
11	*Polypedates maculatus*	0.64	0.033	LC
12	*Sphaerotheca pashchima* *	0.62	0.032	NA
13	*Uperodon globulosus*	0.17	0.009	LC
14	*Uperodon systoma*	0.15	0.008	LC
15	*Uperodon variegatus*	Opportunistic record	-----	LC

*Indicates new state records. Abbreviations: LC–Least Concern, NA–Not Assessed. Conservation status includes International Union for Conservation of Nature (IUCN), Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) and Wildlife Protection Act of India, 1972 (WPA).

Molecular analyses: ***Sample collection and DNA extraction***—Tissue samples of representative specimen of *Fejervarya* sp., *Minervarya* spp. and *Sphaerotheca* sp. were collected from the PTR and stored in 100% ethanol until DNA extraction. We used the DNeasy Blood Tissue Kit (QIAGEN, Germany) for the isolation of genomic DNA in an elution volume of 60 µL. The extracted DNA was quantified by the QIAxpert system spectrophotometer, followed by 0.8% agarose gel electrophoresis to examine the quality of DNA. Isolated DNA was diluted in a final concentration of 40ng/µl for PCR amplification. ***PCR amplification and sequencing—***PCR amplification and sequencing of a fragment of the the 16S rRNA gene was performed using universal primers (Mitchell *et al*. 1993). PCR reactions were carried out in 20 µl reaction volumes using 1× PCR buffer (10 mM Tris–HCl, pH 8.3, and 50 mM KCl), 1.5 mM $MgCl_2$, 0.2 mM of each dNTPs, 3 pmol of each primer, 0.5 units of AmpliTaq Gold DNA Polymerase (Applied Biosystems) and 1µl (~30 ng) of template DNA. PCR conditions were: initial denaturation at 95 °C for 10 min, followed by 35 cycles of denaturation at 95 °C for 45 sec, annealing at 55 °C for 40 sec and extension at 72 °C for 75 sec. Final extension was at 72 °C for 10 min. PCR amplification was confirmed by electrophoresis on 2 % agarose gel stained with firefly dye and visualized under UV transilluminator. The amplified PCR products were treated with exonuclease-I and shrimp alkaline phosphatase (Thermo Scientific Inc.) at 37 °C for 20 min for the removal of any residual primer and dNTPs and followed by inactivation of enzymes at 85 °C for 15 min. The purified fragments were sequenced directly in an Applied Biosystems Genetic Analyzer 3500 XL from forward and

reverse direction using BigDye v3.1 kits. ***Molecular analysis***—Sequence chromatograms were checked visually and assembled in Sequencher®version 5.4.6 (Gene Codes Corporation, Ann Arbor, MI, USA) to obtain consensus sequences, and deposited in NCBI GenBank (accession numbers MN741152–MN741158). For phylogenetic analysis, 51 partial sequences of the 16S rRNA gene were downloaded from GenBank to reconstruct the phylogenetic relationships of PTR samples within the genera *Fejervarya*, *Minervarya,* and *Sphaerotheca* (Annexure III). Additionally, sequences of *Uperodon systoma* (EF017960) and *Nannophrys ceylonensis* (AF249047), *Euphlyctis cyanophlyctis* (AB488901) and *Hoplobatrachus tigerinus* (AB636619) were downloaded from GenBank and used as outgroups. Sequences were aligned using the Clustal W algorithm as implemented in BioEdit v7.1.3 (Hall 1999).

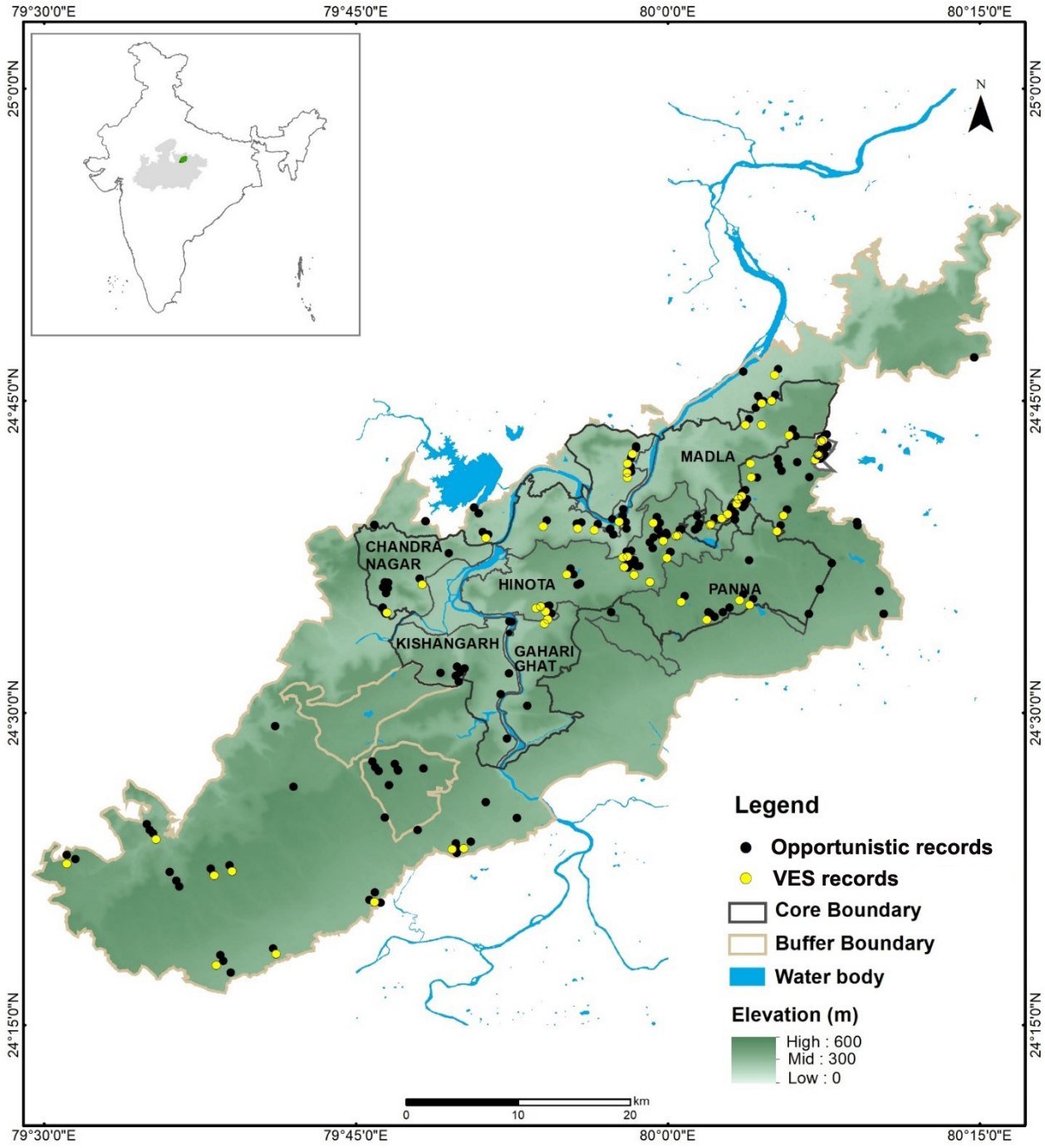

FIGURE 2. Map showing surveyed location for amphibians in PTR which includes 86 visual encounter survey (VES) localities marked in yellow dots and opportunistic sightings marked as black dots. Legend highlighting core and buffer boundary of Panna Tiger Reserve. In some cases, the dots appear overlapped due to the scale and the proximity of the localities. Inset images shows study site in Madhya Pradesh in map of India.

Out of eight extant Indian species of *Sphaerotheca*, we included the sequences of *S. pashchima, S. breviceps,*

S. dobsonii, S. pluvialis and *S. magadha* (Prasad *et al.* 2019). Whereas out of the 33 extant Indian species of *Minervarya*, we included the sequences of 19 species. In addition, we also included three *Fejervarya* species for the phylogenetic analysis (Garg & Biju 2017; Phuge *et al*. 2019). The alignment was checked manually and flanking regions were chopped for phylogenetic analyses. The data matrix was executed in MEGA X (Kumar *et al*. 2018) to determine an appropriate model for analysis. Best-Fit substitution model General time reversible with G+I have been executed in MEGA X with 2000 replicates to construct a phylogenetic tree under Maximum Likelihood (ML) criterion for *Fejervarya*, *Minervarya* and *Sphaerotheca* genera. The obtained best phylogenetic tree was then converted from FASTA to NEWICK format and generated file was then visualized using FigTree v.1.4.0 (http://tree.bio.ed.ac.uk/software/figtree/). Additionally, uncorrected pairwise genetic distance between the species was determined by using aligned file of both genera in MEGA X.

Bioacoustic recordings and analyses: ***Acoustic recordings***—Male advertisement calls were recorded in real-time (16-bit, 44.1 kHz) using Sennheiser MKH 416 unidirectional handheld microphones and Marantz PMD 620 MK–II digital audio recorders between 19:00 and 01:00 hours, mostly just after rains when male calling activity was at its peak. In the field, we distinguished the advertisement calls of focal male frogs by their distinct sound which were emitted regularly and separated from each other by silent intervals longer than call duration during breeding activity. These calls were most commonly heard during breeding period. In some cases, female frogs were seen approaching the calling males upon hearing these conspecific advertisement calls. The microphone was positioned straight at a distance of approximately 30–50 cm from inflating vocal sac of focal calling animals. We adjusted the recording level or gain settings before each recording to avoid overshoot and kept the gain settings constant during a recording. ***Acoustical analyses*—**We measured acoustic properties from total of 370 calls of 37 calling male individuals of eleven anuran species using Raven Pro 1.5 (Bioacoustics Research Program 2014). Measurements of call properties are given in Appendix 2. Ten advertisement calls per individuals were measured for analyses. The descriptions of acoustic properties are provided in Table 2. We derived definitions of acoustic properties used in this paper mainly from Köhler *et al*. (2017) and Thomas *et al*. (2014). In total nine different acoustic properties were selected for quantification of data (Appendix II). Mean, standard deviation (SD), range (minimum–maximum) and percent coefficient of variation (CV = SD/Mean X 100; for within species) were calculated using Microsoft Excel 2010. Number of individuals (n) of each species taken for measuring acoustic properties is mentioned after the species name in Appendix II.

TABLE 2. Descriptions of acoustic properties (after Köhler *et al*. 2017 and Thomas *et al*. 2014)

Properties of Calls	Description
Call duration (ms)	Duration between start of the first pulse and end of the last pulse of a call.
Call repetition rate (calls/minute)	Number of calls per minute. It is inverse of call period.
Inter-call interval (ms)	Interval between two consecutive calls measured from the end of the last pulse of a call to the start of first pulse of the next consecutive call.
Pulses per call (k)	Number of pulses in a call.
Pulse rate (pulses/s)	Number of pulses (k) minus 1, divided by duration between start of first pulse and start of last pulse.
Dominant frequency	The peak frequency of the call measured by producing a power spectrum from selection spectrum function (FFT size = 1024 pts, Hanning window, 43.1 Hz resolution) of Raven Pro Bioacoustics Research Program software over the duration of the entire call
Note duration	Duration between start of the first pulse in a note and end of the last pulse in a note within a call
Notes per call	Number of notes in a call
Pulse duration	Duration between start and end of the middle pulse in a call

We explain what we have considered a call, a note and a pulse in Fig 3 with the example of the advertisement call of *Duttaphrynus stomaticus* and two types of pulses in the call of *Euphlyctis cyanophlyctis*. The oscillograms, spectrograms and power spectra were prepared in Raven Pro 1.5 (FFT size = 1024 pts, Hanning window, 43.1 Hz resolution). Anurans included in the acoustic analyses are *Duttaphrynus melanostictus*, *D. scaber*, *D. stomaticus*, *Euphlyctis cyanophlyctis*, *Hoplobatrachus tigerinus*, *Fejervarya orissaensis*, *Minervarya caperata*, *M. pierrei*,

Sphaerotheca pashchima, *Microhyla nilphamariensis* and *Polypedates maculatus*. In the bioacoustics section of results, we mention values of call properties with standard deviation i.e. value ± standard deviation and range in parenthesis. All the original call measurements in the form an Excel File and call trace of 11 species are uploaded in the "Figshare" (DOI: https://doi.org/10.6084/m9.figshare.12161199.v2)

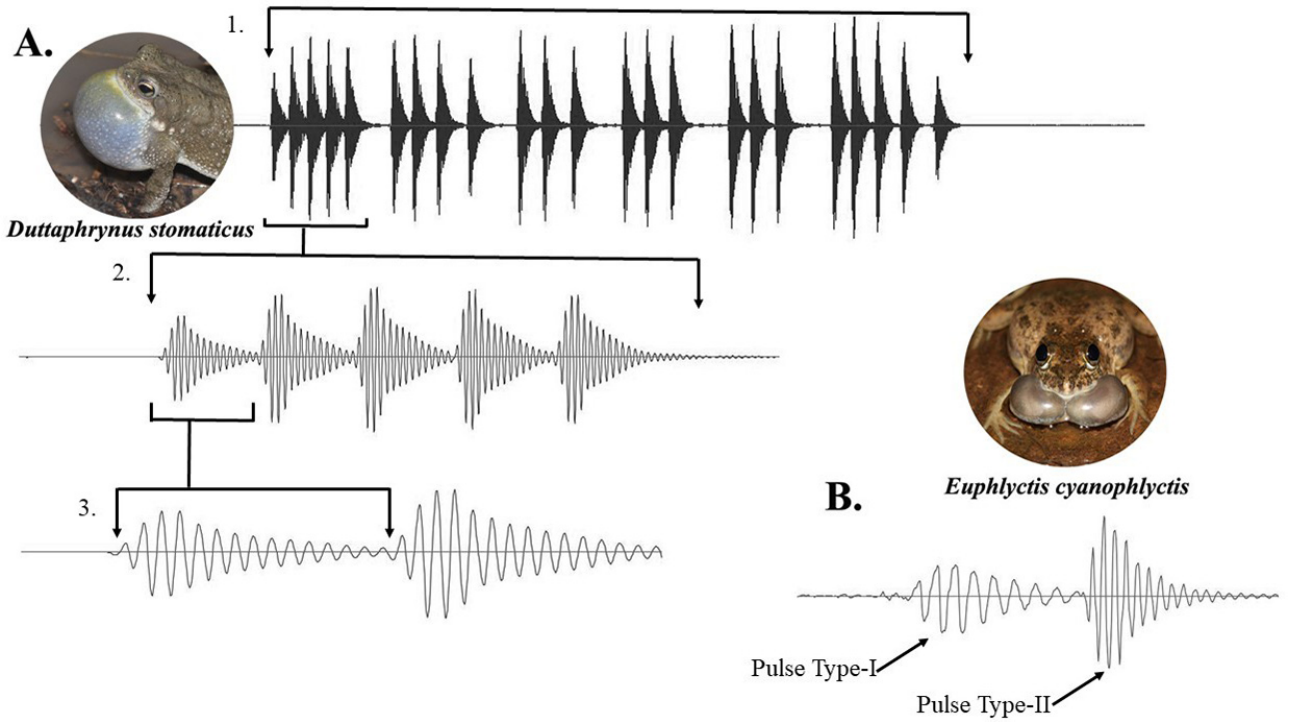

FIGURE 3. Method of measuring acoustic properties with the help of examples—A: Advertisement call of *Duttaphrynus stomaticus*: 1—Call, 2—Note, 3—Pulse. B: Pulse type I and pulse type II in the part of advertisement call of *Euphlyctis cyanophlyctis*.

Morphology: Voucher specimens were collected for taxonomic purpose only, fixed in formaldehyde (4%) and finally stored in 70% absolute ethanol at Wildlife Institute of India Museum (WIIAD). We performed morphometry of 56 voucher specimens from 15 anuran species collected during fieldwork. Males were identified based on nuptial pads, vocal sacs and when visible through skin (*Fejervarya, Minervarya, Sphaerotheca, Euphlyctis, Uperodons, Hoplobatrachus, Polypedates, Microhyla*) and by palpation and distinct size difference between male and female (*Duttaphrynus* spp.). We took the following measurements to the nearest 0.1mm using a Mitutoyo digital caliper: SVL = Snout-vent length from tip of snout to vent; HL = Head length from back of mandible to snout tip; HW = Head width (at the widest point, angle at the jaws); EL = Eye length (horizontally from the anterior to posterior corner of the eye); TD = Tympanum diameter (greatest horizontal width of the tympanum); FLL = Forelimb length (from the elbow to the base of the outer palmar tubercle); HAL = Hand length (from the base of the outer palmar tubercle to the tip of the III finger); TL = Tibia length (from outer surface of the flexed knee to the tibiotarsal inflection); FL= Femur length from vent to knee; FOL = Foot length (from the base of the inner metatarsal tubercle to the tip of the toe); IMT = The greatest length of the inner metatarsal tubercle. We used the term fejervaryan lines as a morphological character of species of *Fejervarya* and *Minervarya*. These are lines on the both sides of ventral body of frogs shared by members of *Fejervarya* and *Minervarya* (Sanchez *et al*. 2018). For the identification of species, we used literature data from Dubois & Ohler (1999); Kuramoto *et al*. (2007); Dinesh *et al*. (2015); Howlader *et al*. (2015); Padhye *et al*. (2017); Hasan *et al*. (2012); Gururaja (2012 & 2017); Garg *et al*. (2018 a & b). Taxonomic nomenclature follows Frost (2020).

Living specimens of representative species were photographed using Nikon D500 and Coolpix B500 cameras. GPS locations of each survey site were recorded with GPS MAP78 using the WGS84 datum, and maps were prepared in QGIS.

Results

We recorded 15 species of frogs belonging to nine genera and four families in this study. Advertisement call properties of 11 anuran species are described in the following. Identification of four species was confirmed using molecular data.

Morphology and Acoustic Descriptions

Family Bufonidae

***Duttaphrynus melanostictus* (Schneider)**
(Fig 4A)

Specimen examined: one male (WIIAD 242).

Morphological characters: Adult male SVL 58.12 mm; body stout; snout obtuse in dorsal and lateral view; prominent black cranial ridges over eyes; two large parotid glands behind eyes; pupil round; tympanum distinct and ¾ of the diameter of eye; dorsum skin warty, two longitudinal broken series of larger warts on dorsum; finger tips without discs; relative length of fingers II < I < IV < III; toes scarcely webbed, relative length of toes 1<2<3<5<4, inner metatarsal tubercle small and distinct. The calling male had single subgular highly distensible vocal sac. *Coloration in life*: Adult male pale yellow or brownish with reddish brown spots on dorsum, breeding males had orange colour on vocal sac region; edges of cranial ridge and upper lip black; series of larger warts on dorsum black; tips of fingers and toes black; underside pale white with fine white tipped spots.

Bioacoustics: We recorded a calling male (voucher WIIAD 242) near a stagnant waterbody at 22:30 hours in July at air temperature 27.2 °C. The male produced single type pulsatile calls. We analysed 10 advertisement calls. Calls were emitted mostly at regular intervals. The mean call duration was 64.1 ± 10.35 ms (44–70 ms) which was the shortest among all congeneric species recorded in PTR. Pulse rate was 112.39 ± 18.77 pulses/s (101.69–161.29 pulses/s) with mean number of pulses per call 6.8 ± 0.79 (5–8 pulses). The mean dominant frequency was 1701.15 ± 22.72 Hz (1679.6–1722.7 Hz) (Annexure II).

Remark: Common anuran in PTR landscape. Breeding recorded in seasonal puddles in July. Tadpoles supposedly belonging to this species were seen in July.

***Duttaphrynus scaber* (Schneider)**
(Figure 4B)

Specimens examined: two males (WIIAD 224 & WIIAD 236) and one female (WII AD 241).

Morphological characters: Adult males SVL 23.7–26 mm (n=2) and female SVL 31.9 mm; body small; head slightly wider than long (see table 3); distinct parietal ridges on head; snout obtuse from lateral view; pupil round; tympanum round, more than ½ the diameter of eye; dorsal skin rough with series of cornified warts on lateral sides; fingers relative length I = II < IV < III, finger tips rounded and without discs; relative length of toes 1<2<3<5<4, inner metatarsal tubercle elongated and distinct. Calling males had single subgular and highly distensible vocal sac. *Coloration in life*: Dorsum olive brown to fawn color with dark brown irregular spots. Prominent orange tipped warts on the lateral sides of body; iris golden; hind limbs cross-barred with dark brown color; throat and vocal sac is orange to dark yellowish (calling males); ventral side of body pale white.

Bioacoustics: We measured 30 advertisement calls from three males. They were observed calling from the edge of ponds and puddles between 23:30–02:00 hours in July at air temperature between 28.7–29.8 °C. All calling males produced single type pulsatile advertisement calls at mostly regular intervals. The mean call duration was 229.77 ± 30.24 ms (172–280 ms) which was longest among its congeners and mean pulse rate was 31.76 ± 1.89 pulses/s (28.69–34.48 pulses/s) with mean 7.6 ± 0.72 (6–9) pulses per call. The mean dominant frequency was 3514.2 ± 201.26 Hz (3230–3746.8 Hz).

Remark: Common in PTR landscape. Breeding was observed in seasonal ponds and waterfilled roadside ex-

cavations in July 2017. The present study represents the first confirmed record of *D. scaber* from Madhya Pradesh state. This is also its record of northern most range. Its earlier neighboring record is from Dang in Gujarat (Padhye *et al*. 2013) and Purba in West Bengal (Mahapatra & Ghorai 2019).

***Duttaphrynus stomaticus* (Lütken)**
(Fig 4C)

Specimen examined: one male (WIIAD 651).

Morphological characters: Adult male SVL 47.5 mm (n=1); head is wider than long; parotid glands elongated and flat; snout obtuse in lateral view; tympanum prominent, slightly more than half the diameter of eye; skin lacks horny warts, speculated on top and warty on lateral and hind side of body; fore limbs moderate and stout; fingers free of web, relative length of fingers II < IV < I < III; toes scarcely webbed, relative length of toes 1<2<5<3<4, inner metatarsal tubercle with sharp edges. Calling males had single subgular and highly distensible vocal sac. *Coloration in life*: Dorsum grey to olive brown with mixed grey blotches; upper lips white; upper surface of fore and hind limbs brown with light grey bands; fingertips black; ventral side of body pale white. Vocal sac of calling males was white to yellowish.

Bioacoustics: 30 advertisement calls from three males were analysed. Males were calling near the bank of puddles and calls were recorded between 22:30–00:30 hours in July (calling site air temperature was between 26.1°C–29.5°C). Single type pulsatile advertisement calls were delivered in mostly regular intervals. *D. stomaticus* had notes within call (sub unit of call) and had pulses within notes. This acoustic character was absent in other sympatric *Duttaphrynus* species found in PTR. The mean call duration was 259.6 ± 41.33 ms (169–351 ms) and mean note duration 25.47 ± 4.49 ms (21–34 ms). Mean pulse rate was 98.88 ± 8.09 pulses/s (83.33–114.16 pulses/s) with mean number of pulses 25.57 ± 4.26 (16–35) per call. The mean dominant frequency was 2571.07 ± 183.11 Hz (2411.70–2885.40 Hz).

Remark: Common in PTR landscape. Breeding and egg laying (strings of eggs) were seen in seasonal shallow puddles in agricultural fields in July.

Family: Dicroglossidae

***Euphlyctis cyanophlyctis* (Schneider)**
(Fig 4G)

Specimens examined: eight males (WIIAD 057, WIIAD 238, WIIAD 279–284), three females (WIIAD 055–056, WIIAD 061).

Morphological features: Males SVL 38.1–44.7 mm (n=8) and females SVL 55.2–59.5 mm (n=3); body robust; head large and triangular, head slightly longer than wide; snout obtusely pointed in dorsal view; eyes large with rhombus pupil; tympanum round and 4/5 the diameter of eye, supratympanic fold distinct; dorsal skin tubercular; fingers pointed and without discs, relative length of fingers IV < II < I < III; toes long and fully webbed with relative length of toes 1 < 2< 3 < 5 < 4. Calling males had paired subgular vocal sac. *Coloration in life*: Dorsum grey or greenish brown with darker rounded spots; flank with white band; hind limbs cross-barred with dark grey; males have black paired lateral vocal sacs; ventral of body white with dark blotches. Vocal sacs of calling males were black.

Bioacoustics: 20 advertisement calls from two males were analyzed. Calls were recorded between 21:00–02:30 hours in July and August; and calling site air temperature was between 26°C–27.1°C. Calling males produced calls while staying on the surface of water in stagnant waterbodies. Calls delivery was sporadic and with non-regular intervals. The advertisement calls of *E. cyanophlyctis* consisted of two types of pulses (Fig 3). The calls had a mean duration of 583.95 ± 209.57 ms (200–946 ms) with a mean pulse rate 30.74 2.08 pulses/s (28.69–35.90 pulses/s). The mean dominant frequency was 1886.30 ± 624.49 Hz (1507.30–3057.70 Hz).

Remark: Common in PTR. Breeding was observed in ponds and stream pools in July and August.

***Fejervarya orissaensis* (Dutta)**
(Fig 4D)

Specimens examined: two males (WIIAD 227 & WIIAD 249) and an individual with sex undetermined (WIIAD 066).

Morphological features: Adult males SVL 42.7–42.9 mm (n=2) and an individual (sex unknown) SVL 39.4 mm; body robust; head slightly longer than wide; snout obtusely pointed in dorsal view; eyes large; tympanum distinct, nearly ½ the diameter of eye, distinct supratympanic fold; tubercles present on dorsal and lateral body, short longitudinal ridges on mid-dorsum, fejervaryan lines on abdomen; fingers without disc and rounded, relative length of fingers II = IV < I < III; poorly developed toe webbing, toes tips rounded, relative length of toes 1<2<5<3<4, inner metatarsal tubercle elongated. Calling males had subgular paired vocal sac. *Coloration in life*: Brown dorsum with dark grey spots; fore limb and hind limb cross-barred with dark grey; upper and lower jaws marked with bands; pale yellow mottling on posterior thigh; ventral side of body white. Vocal sac of calling male was black.

Bioacoustics: We analysed 50 advertisement calls from five males. Calls were recorded between 22:40–00:20 hours and calling site air temperature was between 26.9°C–29.9°C. The calls were pulsatile and emitted mostly at regular intervals. The calling males produced single type calls with mean call duration of 98.76 ± 8.69 ms (82–109 ms). The pulse rate was 104.46 ± 2.03 pulses/s (94.59–107.53 pulses/s) with mean of 9.76 ± 0.98 pulses per call. The mean dominant frequency was 2412.59 ± 6.34.95 Hz (1162.80–2842.40 Hz).

Remark: Common in PTR. Breeding observed in seasonal rainwater puddles in paddy fields in July. The presence of this morphologically cryptic anuran is genetically confirmed in this study (Fig 8A). This is the north-westernmost confirmed record of this species in its range (780 kilometers aerial distance from type locality in Orissa), and a new state record for Madhya Pradesh state.

***Minervarya caperata* (Kuramoto, Joshy, Kurabayashi, and Sumida)**
(Fig 4E)

Specimens examined: three males (WIIAD 237, WIIAD 256–257), two individuals with sex undetermined (WIIAD 274, WIIAD 277).

Morphological features: Adult males SVL 27.5–36.1 mm (n=3) and individuals (sex unknown) SVL 25.8–25.9 mm (n=2); body slender with dermal ridges; head slightly longer than wide; snout slightly pointed in dorsal view; tympanum slightly more than ½ the diameter of eye, supratympanic fold distinct; dorsal skin glandular with series of broadly four longitudinal ridges, fejervaryan lines present on abdomen; fingers tip rounded, relative length of fingers IV < II < I < III; toes tips rounded, toes relative length 1<2<5<3<4. Calling males have bilobate subgular vocal sac. *Coloration in life*: Dorsum grey or brown with light brown tubercles; darker longitudinal ridges; flank covered with dark grey blotches; upper and lower jaws marked with dark bands; limbs cross barred with dark bands; groin region pale yellow; ventral of thighs pinkish; throat black (males during breeding season); abdomen white. Calling males had black vocal sac. *Variation*: WIIAD 237 had large bright orange mid-vertebral band.

Bioacoustics: We analysed 30 advertisement calls from three males, calls were recorded between 21:30–00:00 hours in July and August; and calling location air temperature was between 26.2°C–31.7°C. Males of *M. caperata* called in chorus hidden under the leaves and grass blades near the banks of muddy drains and puddles monsoonal rains. The mean call duration was 130.60 ± 25.49 ms (101–200 ms) with pulse rate of 113.37 ± 3.34 pulses/s (107.69–118.81 pulses/s) and mean of 14.63 ± 2.74 pulses delivered per call. The mean dominant frequency was 2094.48 ± 309.41 Hz (1938–3703.70 Hz).

Remark: *M. caperata* is a common frog in PTR landscape and was seen breeding in waterlogged open grasslands in July. It is a morphologically cryptic species so we performed genetic analysis and our 16S sequence matched with *M. caperata* from Karnataka in Western Ghats (Fig 8A). It was earlier known from its type locality in Karnoor in Karnataka. The present study provides its first state record for Madhya Pradesh. This new record is approximately 1070 km (aerial distance) northwards from its type locality.

***Minervarya pierrei* (Dubois)**
(Fig 4F)

Specimens examined: two males (WIIAD 245 & 246) and two females (WIIAD 059 & WIIAD 067).

Morphological features: Adult males SVL 26.6–27 mm (n=2) and females SVL 33–33.6 mm (n=2); head longer than wide; snout slightly pointed in dorsal view; tympanum marginally less than ¾ the diameter of eye, supratympanic fold visible; dorsal skin tubercular with conical warts, fejervaryan lines present on abdomen, skin on ventral side is smooth; fingers tip rounded, relative length of fingers IV < II < I < III; toes relative length 1<2<3<5<4. *Coloration in life*: Dorsum brown with dark brown spots, dark brown spots more prominent on the flank and upper groin; fore limbs and hind limbs with four dark brown bars; upper and lower jaws barred; groin and undersides of thighs pale yellow; ventral sides of thighs are pinkish; throat mild black (males during breeding season); abdomen white.

Bioacoustics: We recorded 30 advertisement calls from three males. Calls were recorded between 23:30–02:00 hours in July and calling location air temperature was between 27.4 °C–29.7 °C. Males produced single type calls. Mean call duration was 73.18 ± 9.86 ms (53–90 ms) with pulse rate of 113.37 ± 12.92 pulses/s (122.30–225.35 pulses/s) and mean of pulses per call was 14.70 ± 1.84. The mean dominant frequency was 3714.05 ± 67.58 Hz (3574.50–3832.90 Hz).

Remark: A common anuran in the region. Seen calling from waterlogged open grasslands in July and August. The result of our genetic analyses confirmed the presence of *M. pierrei* in PTR (Fig 8A). Its neighboring record is from Haryana, India (Frost 2020). This study provides a new state record of *M. pierrei* from Madhya Pradesh. This record also extends its southern most range to Panna in Central India.

***Sphaerotheca pashchima* Padhye, Dahanukar, Sulakhe, Dandekar, Limaye, and Jamdade**
(Fig 4J)

Specimens examined: five males (WIIAD 064, WIIAD 065, WIIAD 229, WIIAD 233 & WIIAD 275) and two females (WIIAD 225 & WIIAD 232).

Morphological features: Adult males SVL 32.1–45.5 mm (n=5) and two females SVL 45.8–47.1 mm (n=2); body stout; head wider than long; snout rounded dorsally as well as laterally; eyes large, pupil diamond shaped or round; tympanum nearly 2/3 the diameter of eye, supratympanic fold distinct; dorsal skin granular with some tubercles; fingers without web and round, relative length of fingers II < IV < I < III; relative length of toes 1<2<3<5<4, inner metatarsal tubercle large and very prominent. Calling males had bilobate subgular paired vocal sac. *Coloration in life*: Dorsum grey or brownish to pinkish with or without irregular dark spots; limbs cross barred with dark grey; upper lips marked with dark bands; yellowish marblings in groin region; ventral side of body white; calling males had black throat colour.

Bioacoustics: We analysed 30 calls from three males. Males called intermittently and delivered single type pulsatile calls. We recorded calls between 21:00–00:00 hours in July. The air temperature of calling site of frogs at the time of call recordings was between 26.5°C–29.3°C. The mean call duration was 377.60 ± 45.65 ms (319–455 ms). Mean pulse rate was 78.35 ± 3.03 pulses/s (72.43–83.08 pulses/s) with mean of 29.23 ± 2.87 pulses per call. The mean dominant frequency was 2074.37 ± 441.68 Hz (947.50–2368.70 Hz). This is the first-time description of advertisement calls of *Sphaerotheca pashchima* (Fig 7 & Annexure II).

Remark: Common in PTR and breeds in shallow puddles in grasslands and stream pools in early July. Occurrence of *S. pashchima* from PTR has been genetically confirmed in our study (Fig 8B). In earlier faunistic studies based on morphology only, *S. breviceps* was widely reported in literature. The present study provides the first record of *S. pashchima* from Madhya Pradesh. Now Panna region is the easternmost known locality for *S. pashchima*.

***Hoplobatrachus crassus* (Jerdon)**
(Fig 4H)

Specimens examined: one male (WIIAD 252) and one individual (sex undetermined, WIIAD 068).

Morphological features: A male with 70.2 mm SVL and one individual (unknown sex) SVL 92.9 mm; robust body; head longer than wide; snout obtusely pointed in dorsal view; tympanum distinct and nearly ¾ of the diameter of eye, supratympanic fold present; dorsal skin glandular with 5–6 broken longitudinal ridges; finger tips without discs, relative length of fingers IV < II < I < III; toes tips slightly rounded, relative toes length 1<2<3<5<4, Webbing fully developed in feet (I0–¼ II0–1/3 III0–1/3IV1/3–0V), distinct shovel shaped inner metatarsal tubercle. *Coloration in life*: Dorsum uniform grey with plump black spots; hind limbs cross-barred with dark grey bands; upper and lower jaws with dark bands; vocal sac blackish (calling males; unlike *H. tigerinus*), throat and gular region spotted with grey; ventral sides of body pale white.

Remark: Uncommon, recorded from two ranges in PTR. Sighted in waterlogged open grasslands after heavy rains in July.

***Hoplobatrachus tigerinus* (Daudin)**
(Fig 4I)

Specimens examined: one adult male (WIIAD 228).

Morphological features: An adult male, SVL 153.9 mm; body large and stout; head marginally longer than wide; snout obtusely pointed in dorsal view and projecting beyond mouth; tympanum distinct, nearly equal to the diameter of eye, prominent supratympanic fold; arms short and strong; dorsal skin glandular with broken longitudinal ridges; finger tips without discs, relative length of fingers II < IV < I < III; toes nearly full webbed, webbing formula I0–0II0–½ III½–½ IV ½–½V), relative toes length 1<2<3<5<4, inner metatarsal tubercle present. *Coloration in life*: Adult male pale brown or greyish (lemon yellowish during breeding season) with dark plump spots on dorsum; prominent greenish or pale yellowish stripe from behind the eye to the groin; a white streak on upper jaw; hind limbs cross-barred with four prominent bands; ventral sides pale white. Males had distinct blue paired subgular vocal sac which is likely to be an optical cue for attraction of females (Gomez *et al*. 2009; Fig 4I).

Bioacoustics: We analysed 50 advertisement calls from five males of *H. tigerinus*. The males were recorded calling between 23:30–03:00 hours in July and site air temperature was between 26°C–28.5°C. The mean call duration of analysed calls was 235.30 ± 48.65 ms (181–430 ms). Recorded mean pulse rate was 76.92 ± 5.32 pulses/s (61.82–87.18 pulses/s) with 17.36 ± 2.88 pulses (13–29 pulses) delivered per call. The mean dominant frequency was 1819.14 ± 388.87 Hz (1205.90–2239.50 Hz).

Remark: A common anuran in PTR. Observed breeding in seasonal water puddles in agricultural fields and waterlogged open grasslands in early July.

Family: Microhylidae

***Microhyla nilphamariensis* Howlader, Nair, Gopalan, and Merilä**
(Fig 4K)

Specimens examined: five males (WIIAD 234, WIIAD 235, WIIAD 262–264)

Morphological features: Adult males SVL 18.3–21 mm (n=5); head triangular, wider than long; snout nearly rounded in lateral view; pupil round; tympanum not visible, supratympanic fold present; dorsal skin smooth with small tubercles all over the upper and lateral side of body; fingers without webbing and flattened, relative length of fingers I < II < IV < III; toes long and thin, tips of toes rounded, rudimentary webbing in toes, relative length of toes 1< 2<5<3<4, small round inner metatarsal tubercle. This species is described from Bangladesh by Howlader *et al.* (2015) but Garg *et al.* (2018b) reported its widespread distribution in India and perhaps earlier records of *M. ornata* from central India should be referred to *M. nilphamariensis* (Garg *et al.* 2018b). Calling males had single subgular vocal sac. *Coloration in life*: Golden or pink dorsal with dark arrowhead markings on the back starting from eyes and widening towards hind limbs, tiny orange dots on dorsum; hind limbs cross-barred with dark brown; flank dark brown; throat black (calling males); ventral side of body white.

Bioacoustics: We analysed 50 advertisement calls from five males. The calls were recorded between 20:00–23:40 hours (July–September) and calling site temperature was between 25.7 °C–30.9 °C. Calling males produced

distinct pulsatile calls. The calls were delivered mostly at uniform intervals. Calls had mean duration of 415.22 ± 130.31 ms (195–625 ms). Mean pulse rate was 44.04 ± 3.4 pulses/second (38.17–50.76 pulses/second) with 18.36 ± 5.82 pulses delivered per call (8–26 pulses). The mean dominant frequency was 3067.18 ±104.52 Hz (2928.50–3273 Hz).

FIGURE 4. Photos of anuran species in life recorded during the study: A) *Duttaphrynus melanostictus*; B) *Duttaphrynus scaber*; C) *Duttaphrynus stomaticus*; D) *Fejervarya orissaensis*; E) *Minervarya caperata*; F) *Minervarya pierrei*; G) *Euphlyctis cyanophlyctis*; H) *Hoplobatrachus crassus*; I) *Hoplobatrachus tigerinus*; J) *Sphaerotheca pashchima*; K) *Microhyla nilphamariensis*; L) *Uperodon globulosus*; M) *Uperodon systoma*; N) *Uperodon variegatus*; O) *Polypedates maculatus*. Red star indicates new state records for Madhya Pradesh.

Remark: Commonly encountered frog in PTR. Observed breeding in seasonal pools in waterlogged open grasslands between July–September.

***Uperodon globulosus* (Günther)**
(Fig 4L)

Specimen examined: one male (WIIAD 231) & one sub adult (WIIAD 069)

Morphological features: An adult male SVL 51.9 mm and a sub-adult (sex unknown) SVL 36.9 mm; globular body shape; head narrow and small; snout obtuse in lateral view; eyes small, pupil round; tympanum hidden, supratympanic fold distinct; dorsal skin is smooth; finger tips rounded and without disc, relative length of fingers IV < I < II < III; hind limbs small, toes tips rounded, toes scarcely webbed, relative length of toes 1< 5< 2 < 3< 4, inner meta tarsal tubercle shovel shaped and very prominent, outer meta tarsal tubercle shovel shaped. Coloration in life: Grey or golden-brown dorsum speckled with tiny golden or pale white spots; dorsal surface of limbs light brown; upper and lower jaws dull white; throat (calling male) black; ventral side of body dull white, eggs bicolor.

Remark: Uncommon, recorded from three forest ranges. Seen breeding in temporary rainwater puddles in agricultural fields and grasslands in July. *U. globulosus* is widely distributed in Central India and has earlier been reported from Jabalpur (Garg *et al.* 2018a).

***Uperodon systoma* (Schneider)**
(Fig 4M)

Specimens examined: WIIAD 223 & WIIAD 230.

Morphological features: Two individuals (sex unknown) SVL 44.6–46.5 mm; globular body shape; head narrow, head width nearly same as head length; snout rounded in dorsal and lateral view; pupil round; tympanum hidden, supratympanic fold present; dorsum skin smooth; tips of fingers without disc and round, relative length of fingers I < IV < II < III; toes rounded, scarcely webbed, relative length of toes 1<5<2<3<4, inner metatarsal tubercle shovel shaped and very prominent, outer metatarsal tubercle shovel shaped. Coloration in life: Uniform chocolate brown dorsum with golden yellow marbling; upper eye lids golden yellow; eyes black; flank and groin lighter in color compared to dorsum; underside of toes and heels brown; lower lip (breeding males) white; throat and vocal sac (calling males) black.

Remark: Rare in PTR. Seen active for breeding in temporary rainwater puddles. Its explosive breeding lasted only for few days at the onset of monsoon in early July. Formerly recorded from Gwalior and Seoni districts in Madhya Pradesh (Chandra & Gajbe 2005; Garg *et al.* 2018a).

***Uperodon variegatus* (Stoliczkia)**
(Fig 4N)

Specimen examined: one female (WIIAD 291).

Morphological features: A female SVL 31.3 mm; body slender; head small and slightly wider than long; snout rounded to nearly truncated in dorsal view; tympanum hidden, supratympanic fold present; dorsum skin smooth; tips of fingers truncated, fingers relative length I < II < IV < III; tips of toes slightly truncated, relative length of toes 1< 2<5<3<4, webbing absent in toes, inner metatarsal tubercle distinct. Coloration in life: Dorsal surface of dorsum and limbs dark brown with irregular golden dots and marks; dorsal surface between of nose and eyelids predominantly spotted with golden color; ventral side of body dull white.

Remark: A female was recorded at night from agricultural fields near Amanganj (Panna district) in July 2019. Chandra & Gajbe (2005) had mentioned its presence in Madhya Pradesh without any precise locality. This study provides first confirmed record of *U. variegatus* from Madhya Pradesh. PTR represents its northernmost range.

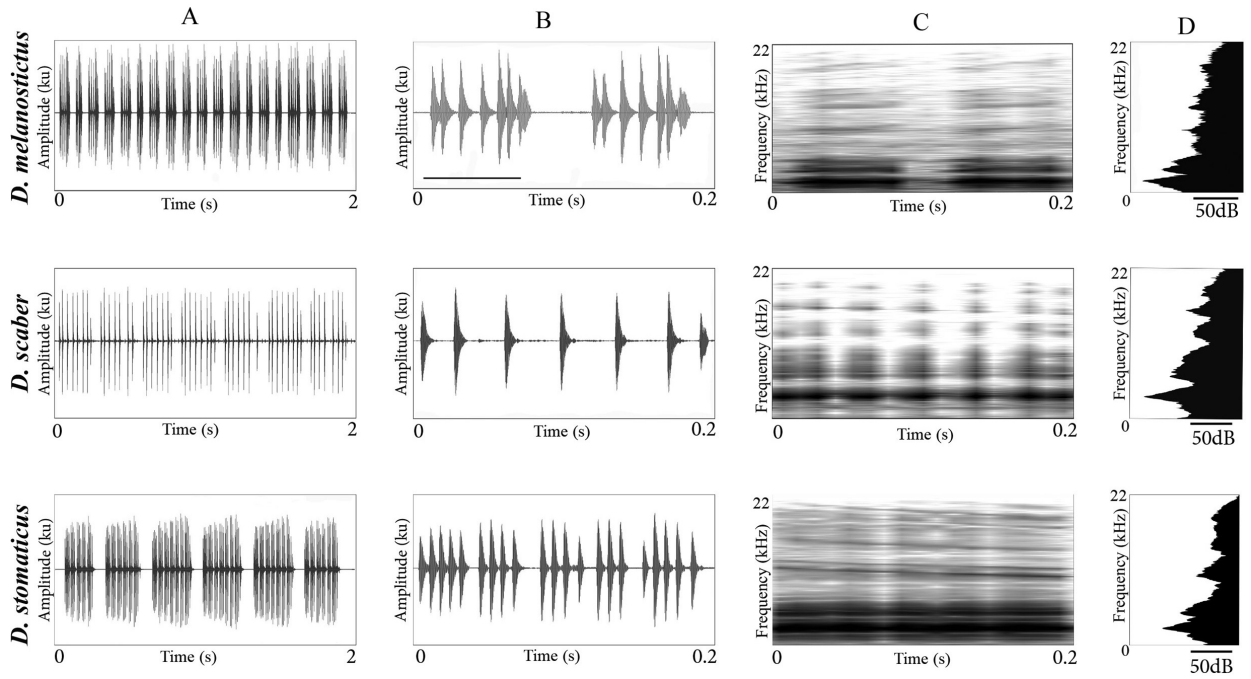

FIGURE 5. Advertisement calls of *Duttaphrynus melanostictus*, *Duttaphrynus scaber* and *Duttaphrynus stomaticus* recorded in PTR: From top to bottom A & B represent oscillograms, C—spectrogram, D—power spectrum, (FFT size = 1024 pts, Hanning window, 43.1 Hz resolution). In B of *D. melanostictus*, call is underlined for which power spectrum in D is produced. The call trace of each species is provided in supporting supplementary information.

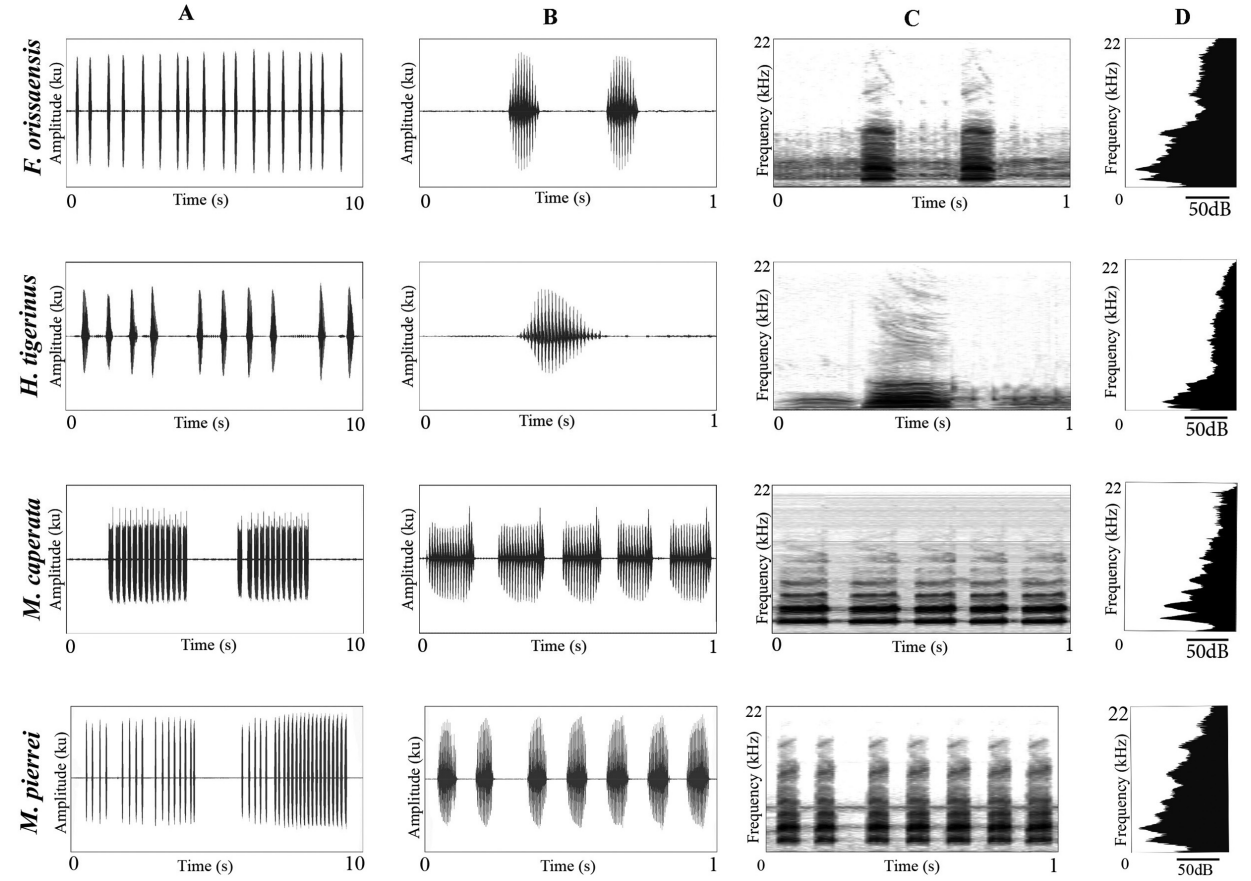

FIGURE 6. Advertisement calls of *Fejervarya orissaensis*, *Hoplobatrachus tigerinus*, *Minervarya caperata*, *Minervarya pierrei* recorded in PTR: From top to bottom A & B represent oscillograms, C—spectrogram, D—power spectrum, (FFT size = 1024 pts, Hanning window, 43.1 Hz resolution).

Family: Rhacophoridae

***Polypedates maculatus* (Gray)**
(Fig 4O)

Specimens examined: three males (WIIAD 226, WIIAD 250, WIIAD 271), one female (WIIAD 251) and four individuals (sex undetermined; WIIAD 062, WIIAD 063, WIIAD 272, WIIAD 273).

Morphological features: Males SVL 43.3–46.9 mm (n=3), female SVL 60.3 mm (n=1) and individuals (sex unknown) SVL 38.8–51.9 mm (n=4); body flattened and elongated; head longer than wide; eyes large, pupil horizontal; tympanum large and nearly 1/3 of the diameter of eye, supratympanic fold distinct; dorsum skin smooth, ventral side of body granular; tips of fingers with prominent discs, relative length of fingers I < II < IV < III; toes tips with discs, relative length of toes 1<2<3<5<4, webbing formula of toe I1–1II½–1III½–2IV1½–½V. *Coloration in life*: Dorsum uniform golden brown with dark brown blotches running from dorsal surface of head to rear body; a prominent dark brown streak running from tip of snout to covering tympanum and extending up to mid flank region; color of dorsal of limbs lighter than dorsum surface; limbs cross-bared with dark brown; upper lip white; yellow reticulation on brown background on posterior thigh; ventral side of body white.

Bioacoustics: 20 advertisement calls from two males were analysed in this study. We recorded calls between 17:40–00:30 hours and calling sites air temperature was between 27.1°C–29.7°C. The males were recorded calling from bushes and grassland near puddles in the months of July and August during monsoon. The calls were emitted at irregular intervals. The calls had a mean duration of 132.25 ± 25.54 ms (82–171 ms) with a mean pulse rate of 60 ± 5.9 pulses/second (50–74.63 pulses/s) and mean 5.75±1.12 pulses per call (4–9 pulses/call). The mean dominant frequency was 960.40 ± 125.06 Hz (818.3–1335.10 Hz).

Remark: Common in PTR. Calls from trees and shrub ~1m and above ground during July and August. Foam nests were seen in July among *Lantana* bush ca. 40 cm above water and also on the water surface.

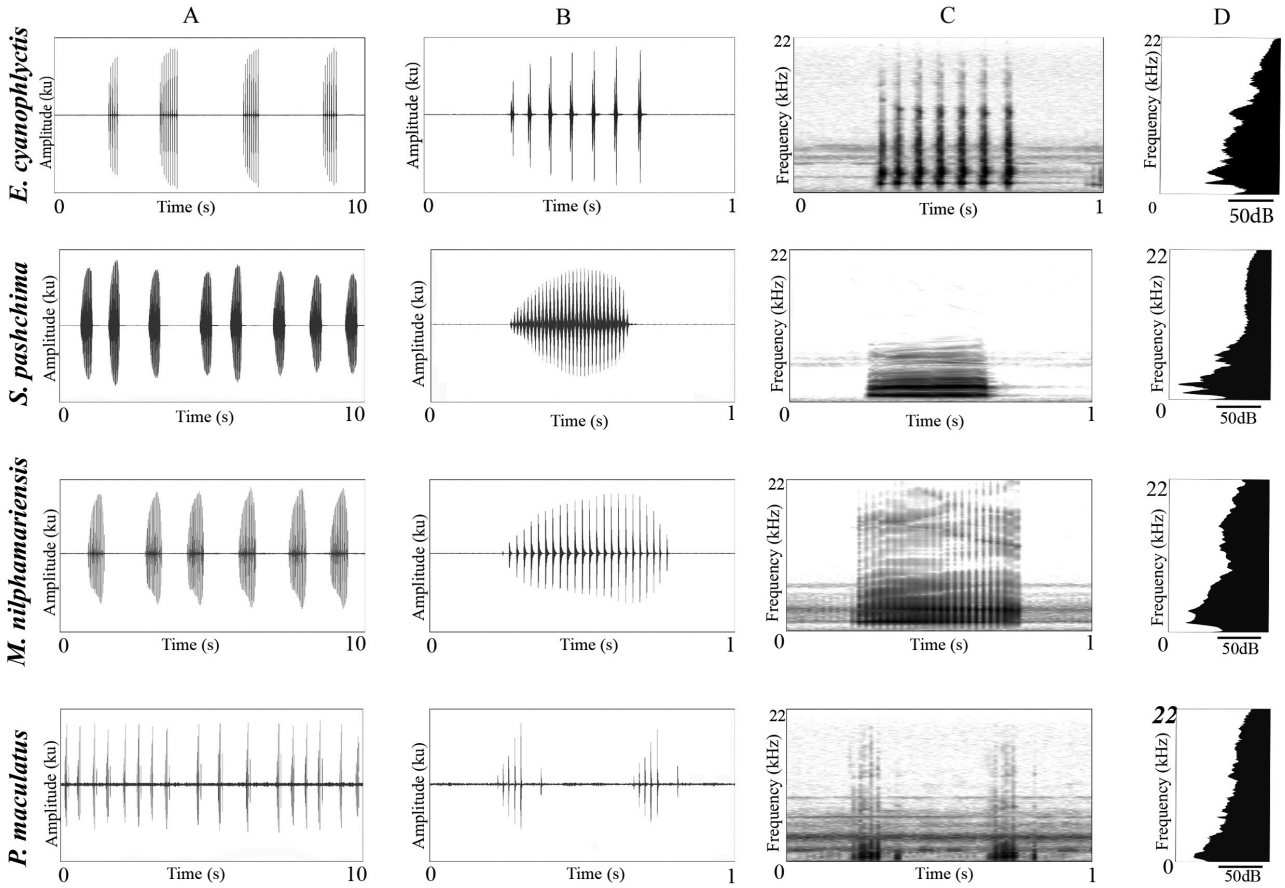

FIGURE 7. Advertisement calls of *Euphlyctis cyanophlyctis*, *Sphaerotheca pashchima*, *Microhyla nilphamariensis*, *Polypedates maculatus* recorded in PTR: From top to bottom A & B represent oscillograms, C—spectrogram, D—power spectrum, (FFT size = 1024 pts, Hanning window, 43.1 Hz resolution).

Molecular studies

We genetically identified four cryptic species, namely *Minervarya caperata*, *M. pierrei*, *Fejervarya orissaensis*, *Sphaerotheca pashchima*. *Fejervarya* and *Minervarya* are known to have conserved morphological characters (Dinesh *et al*. 2015; Phuge *et al*. 2019). *Sphaerotheca* members have morphological character variation with multiple colour morphs within population (Prasad *et al*. 2019). Because we could not ascertain these four species only on the basis of morphology, we relied upon molecular study for species identification as advised in previous taxonomic studies.

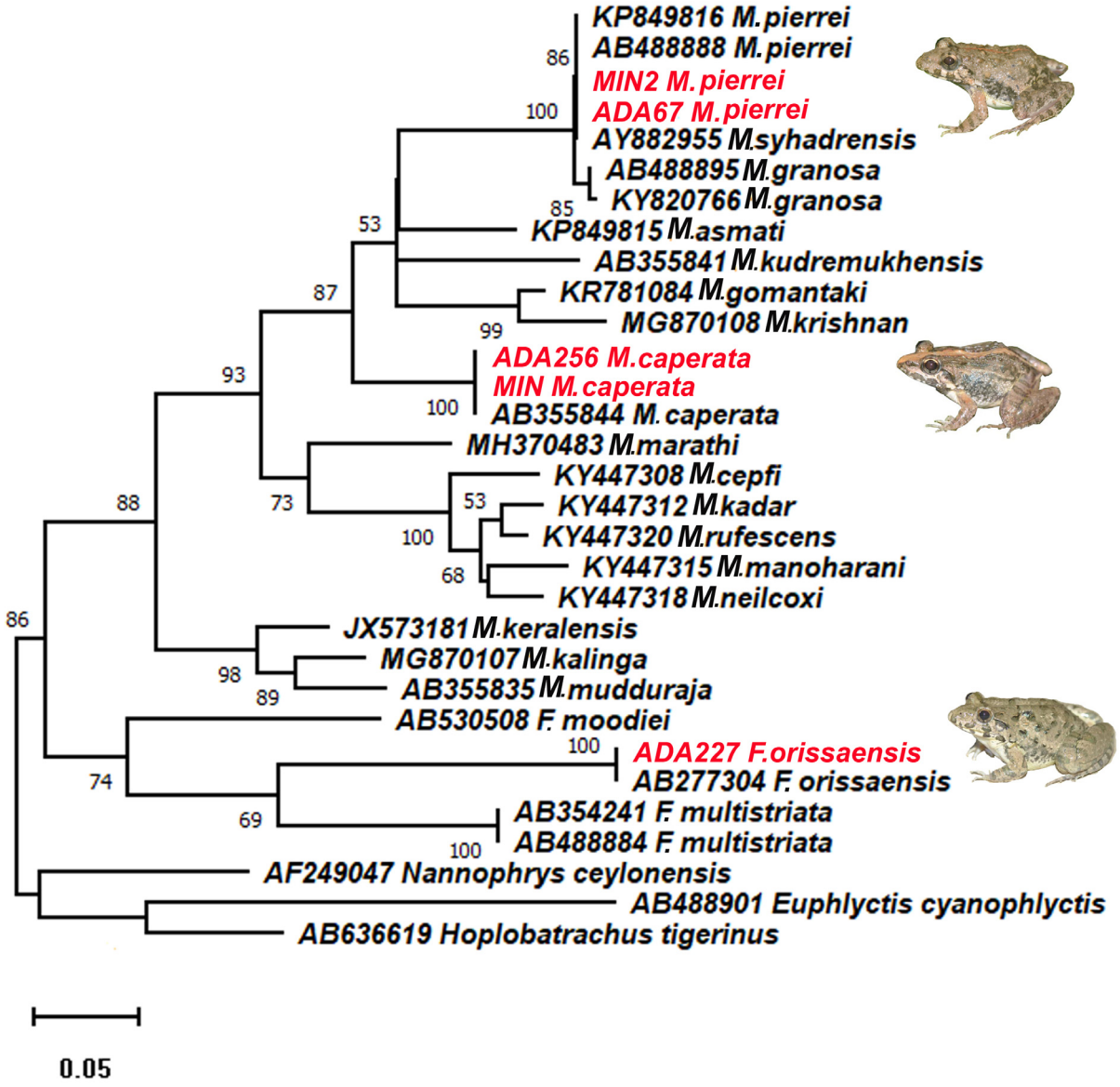

FIGURE 8A. Maximum Likelihood tree based on the 16S mitochondrial DNA dataset (nodes having >50% bootstrap values are shown) of genera **A.** *Fejervarya* and *Minervarya.*

We successfully obtained 551 bp DNA sequences for the 16S rRNA gene from all the samples collected from Panna Tiger Reserve. Subsequently, we performed BLAST (Altschul *et al*. 1997) searches with the generated sequence, which indicated unambiguous matching with the known sequences of the target species available in the NCBI GenBank (Appendix III). Out of seven specimens' samples, two each were of *Sphaerotheca pashchima*, *Minervarya caperata*, and *Minervarya pierrei* and one was of *Fejervarya orissaensis*. Furthermore, our phylogenetic trees (Fig 8A & 8B) were mostly in accord with prior studies (Garg & Biju 2017; Padhye *et al*. 2017; Prasad *et al*.

2018). Hence, we here provide the first records of *F. orissaensis*, *M. pierrei* and *M. caperata* from the PTR. Occurrence of *Sphaerotheca pashchima* was also confirmed by the phylogenetic tree (Fig 8B). The uncorrected pairwise genetic distance indicated that *F. orissaensis* (MN741152), *M. pierrei* (MN741156 & MN741155) and *M. caperata* (MN741153 & MN741154) are 100% similar with available GenBank sequence of *F. orissaensis, M. pierrei* and *M. caperata* respectively. Whereas *S. pashchima* (MN741157 & MN741158) shows 98.20% similarity with available GenBank sequence of *S. paschima*.

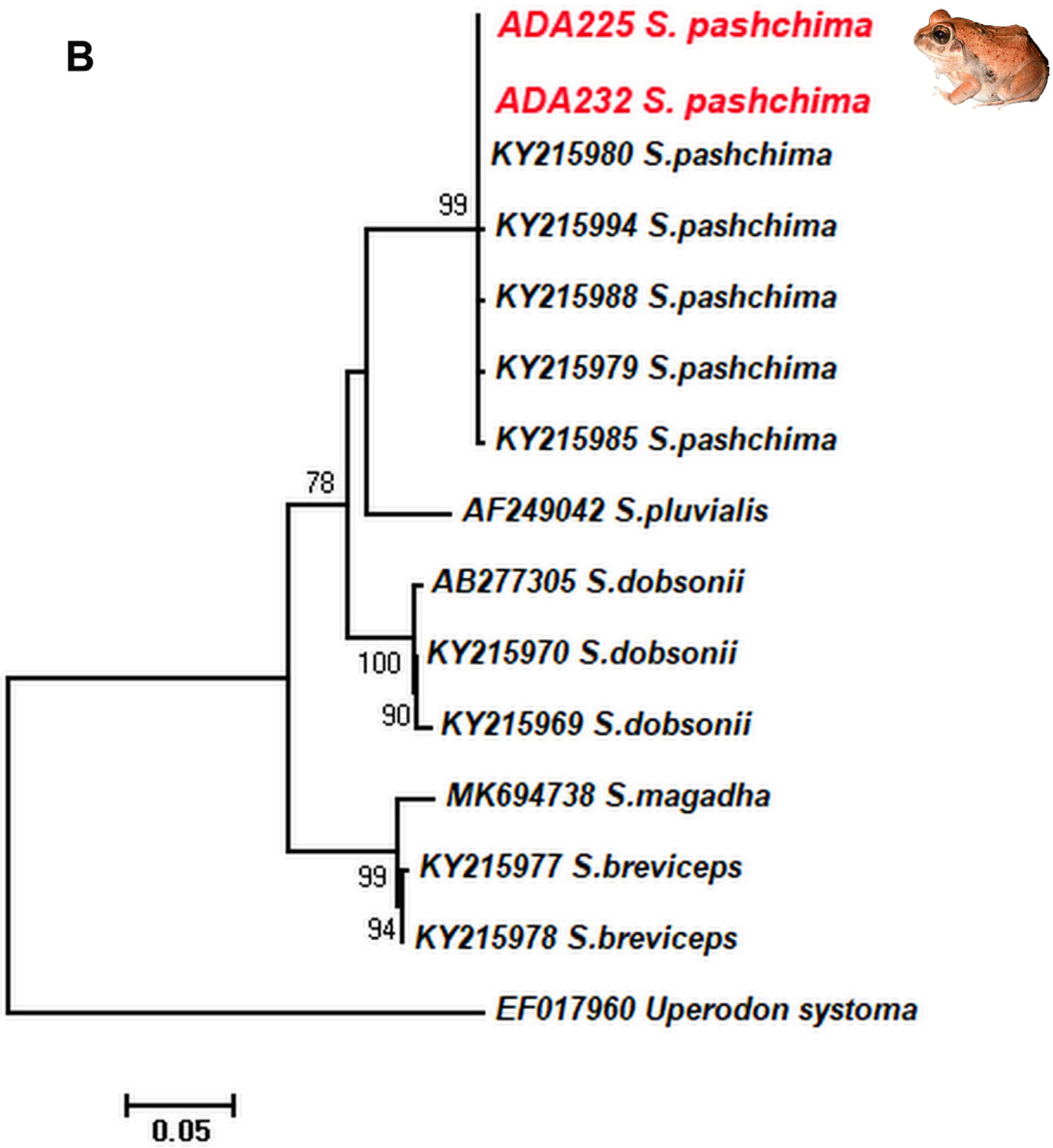

FIGURE 8B. Maximum Likelihood tree based on the 16S mitochondrial DNA dataset (nodes having >50% bootstrap values are shown) of genera **B.** *Sphaerotheca* (Red colour indicates sequences generated in this study).

Discussion

There are 19 species of amphibians reported from Madhya Pradesh by Chandra & Gajbe (2005) and Chandra & Ray (2007) based on morphological identification. Previous reports of *Duttaphrynus microtympanum*, *Indirana leithii*, *Fejervarya cancrivora, Fejervarya limnocharis, Sphaerotheca breviceps* and *Sphaerotheca rolandae* from Madhya Pradesh by Chandra & Gajbe (2005) and Chandra & Ray (2007) are doubtful and need further confirmation (Frost

2020). *Duttaphrynus microtympanum* and *Indirana leithii* are endemic to Western Ghats in South western India; *Fejervarya cancrivora* and *Fejervarya limnocharis* are not found in India; the presence of *Sphaerotheca rolandae* is doubtful in India (Frost 2020); *Sphaerotheca breviceps* is known from Karnataka, Tamil Nadu and Andhra Pradesh region and its records outside South India warrant genetic confirmation (Prasad *et al.* 2019).

Our study recognizes 15 amphibians from the PTR in Madhya Pradesh. Dicroglossidae is the dominant family (seven species) followed by Microhylidae (four species). The records of *Duttaphrynus scaber*, *Fejervarya orissaensis*, *Minervarya caperata, Minervarya pierrei* and *Sphaerotheca pashchima* constitute new state records. An integrated taxonomic approach helped us to identify cryptic species of *Fejervarya*, *Minervarya* and *Sphaerotheca*. Molecular analyses helped us confirm occurrence of *Fejervarya orissaensis*, *Minervarya caperata, Minervarya pierrei* and *Sphaerotheca pashchima* in the PTR. We placed *Fejervarya orissaensis* in the genus *Fejervarya* and *Minervarya caperata* and *Minervarya pierrei* in the genus *Minervarya* following Sanchez *et al.* (2018).

Bioacoustic techniques helped us to rapidly assess the distinctiveness of the species' advertisement call properties in field. In most of the cases, call characters or traits such as pulse rate within calls, number of pulses or note per call are more stable within the species despite of having individual peculiarity. Previous studies suggested that these call characters are important and useful for taxonomic inference because of their high interspecific variation and relatively narrow intraspecific variation (Köhler *et al*. 2017). The advertisement calls of eleven species (Annexure II) provided in this study will be helpful in future as a reference data. Bioacoustics analyses provided us reliable key to identify closely related species such as *Minervarya caperata* and *Minervarya pierrei*. This would help in delimiting cryptic species (Tishechkin 2014). We also provided information on geographic variation in calls, for example, the mean dominant frequency (3514.2 ± 201.26 Hz, temperature 29.8–28.7 °C, n=3) of the calls of *Duttaphrynus scaber* in our study was found to be lower as compared to the mean dominant frequency (3670 Hz, temperature 21.2 °C, n=2) of *D. scaber* calls recorded in Northern Western Ghats (Padhye *et al*. 2013); pulse rate (44.04 ± 3.4 pulses/s, n=5) of *Microhyla nilphamariensis* in our study was found to be higher than the pulse rate (37.9 ± 0.4 pulses/s, n=8) of calls from Bangladesh (Hasan *et al*. 2015); and mean call duration (145 ± 7 ms, temperature 22 °C, n=1) of *F. orissaensis* from Myanmar (Köhler *et al.* 2019) was longer compared to mean call duration (98.76 ± 8.69 ms, temperature 26.9–29.9 °C, n =5) of calls recorded of same species in our study and dominant frequency (1128 ± 22.7 Hz, n=1) was lower than the dominant frequency (2412.59 ± 634.95 Hz, n=5) of our recorded *F. orissaensis* calls; call duration of *E. cyanophlyctis* (1500 ms) from Coorg, Karnataka (Hampson & Bennett 2002) was longer than our calls (584 ± 209.57 ms, n=2) of same species in PTR and dominant frequency was found lower (1500 Hz) than the dominant frequency (1886.3 ± 624.5 Hz, n=2) of calls of *E. cyanophlyctis* in PTR. These variations may have occurred due to difference in temperature of comparative calls from literature recorded at lower or higher temperatures. Also, the difference in dominant frequencies in the references could be related to body size differences among the recorded individuals. Perhaps this indicates need of further study in these species. This study provides the first-time advertisement calls descriptions of *Sphaerotheca pashchima.* All these acoustic descriptions are focused on utility of anuran vocalizations for taxonomy and would be useful in monitoring and conservation of amphibians in the region. Hence this should serve as a good reference for future studies.

The drylands of PTR receive precipitation only through South west monsoon (July to September). The temporary rainwater puddles in grasslands and agricultural areas are found to be crucial breeding habitats for amphibians. Therefore, these seasonal habitats need to be mapped and included in the management plan of PTR. New confirmed distribution data will help assess the IUCN status better for Not Evaluated species such as *Minervarya caperata*, *Microhyla nilphamariensis* and *Sphaerotheca pashchima*. The present detailed inventory paper based on an integrated taxonomic approach significantly adds to the knowledge of diversity, distribution and conservation priorities for amphibians of Central India.

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APPENDIX I. Morphometric measurements (mm) of anuran specimens collected during the study in PTR (M = Male; F = Female; SU = Sex Unknown; SD = Standard Deviation):

Sex	Specimen Number		SVL	HL	HW	EL	TD	FLL	HAL	TL	FL	FOL	IMT
***Duttaphrynus melanostictus* (Schneider)**													
M	WIIAD 242		58.1	19.9	21.2	6.5	5.0	15.4	12.7	23.9	23.6	19.3	2.3
***Duttaphrynus scaber* (Schneider)**													
M	WIIAD 224		23.7	7.7	8.1	2.3	1.4	5.1	5.1	7.6	8.0	6.7	0.7
M	WIIAD 236		27.0	8.5	9.2	2.7	1.7	5.9	5.7	9.6	8.4	7.7	1.0
		Mean	**25.4**	**8.1**	**8.6**	**2.5**	**1.6**	**5.5**	**5.4**	**8.6**	**8.2**	**7.2**	**0.9**
		SD	**2.3**	**0.6**	**0.8**	**0.2**	**0.2**	**0.6**	**0.4**	**1.4**	**0.3**	**0.7**	**0.3**
F	WIIAD 241		31.9	10.7	11.0	3.2	2.1	7.6	6.2	10.8	11.4	9.1	1.4
***Duttaphrynus stomaticus* (Lütken)**													
M	WIIAD 651		47.5	18.1	19.0	4.6	2.6	12.0	10.7	19.3	19.1	16.6	2.0
***Euphlyctis cyanophlyctis* (Schneider)**													
M	WIIAD 238		40.4	15.5	14.8	5.3	4.0	7.3	10.5	19.6	19.9	20.1	2.8
M	WIIAD 279		43.4	16.5	15.6	4.9	4.3	8.1	11.3	22.7	21.3	21.7	2.7
M	WIIAD 280		44.7	17.1	17.3	5.6	3.8	8.8	12.2	24.1	23.3	23.0	3.0
M	WIIAD 281		44.4	17.3	17.9	5.3	4.2	9.1	12.3	23.7	23.3	23.7	3.4
M	WIIAD 282		44.4	16.5	16.9	5.2	4.6	8.9	12.8	22.3	23.1	23.1	2.8
M	WIIAD 283		43.1	17.2	16.5	4.6	3.9	7.5	10.7	21.6	20.2	21.5	2.7
M	WIIAD 284		39.2	15.2	15.8	5.3	3.6	8.0	11.2	20.1	19.6	16.5	2.5
M	WIIAD 057		38.1	14.0	12.8	4.7	3.3	8.3	9.2	19.1	17.9	18.2	2.3
		Mean	**42.2**	**16.2**	**15.9**	**5.1**	**4.0**	**8.2**	**11.3**	**21.6**	**21.1**	**21.0**	**2.8**
		SD	**2.6**	**1.2**	**1.6**	**0.3**	**0.4**	**0.7**	**1.2**	**1.9**	**2.0**	**2.5**	**0.3**
F	WIIAD 055		59.5	23.6	22.0	7.4	6.3	12.0	14.5	31.0	29.9	21.9	2.8
F	WIIAD 056		55.2	22.2	20.3	6.6	6.1	10.7	14.2	26.9	28.2	26.2	3.7
F	WIIAD 061		58.9	22.1	21.4	6.0	5.2	12.2	14.5	30.3	28.2	28.3	3.3
		Mean	**57.9**	**22.6**	**21.2**	**6.6**	**5.9**	**11.6**	**14.4**	**29.4**	**28.8**	**25.5**	**3.3**
		SD	**2.3**	**0.9**	**0.8**	**0.7**	**0.6**	**0.8**	**0.2**	**2.2**	**1.0**	**3.2**	**0.5**
***Fejervarya orissaensis* (Dutta)**													
M	WIIAD 227		42.7	19.3	16.1	4.9	2.6	8.2	8.7	19.5	17.7	17.6	2.3
M	WIIAD 249		42.9	15.2	17.0	4.9	2.5	7.9	8.7	20.2	18.7	17.5	2.7
		Mean	**42.8**	**17.2**	**16.6**	**4.9**	**2.5**	**8.1**	**8.7**	**19.9**	**18.2**	**17.6**	**2.5**
		SD	**0.1**	**2.9**	**0.6**	**0.0**	**0.1**	**0.2**	**0.0**	**0.5**	**0.7**	**0.0**	**0.3**
SU	WIIAD 066		39.4	15.2	13.4	4.6	2.1	6.6	8.0	19.4	16.7	17.7	2.2
***Minervarya caperata* (Kuramoto, Joshy, Kurabayashi, and Sumida)**													
M	WIIAD 237		36.1	13.3	10.3	3.7	1.9	6.0	6.2	16.4	16.2	16.1	1.5
M	WIIAD 256		27.5	9.8	7.8	3.2	1.9	5.3	5.4	12.5	12.3	13.0	1.2
M	WIIAD 257		28.0	10.8	8.7	3.5	1.9	5.6	5.7	12.9	11.8	14.1	1.2
		Mean	**30.5**	**11.3**	**8.9**	**3.5**	**1.9**	**5.7**	**5.8**	**13.9**	**13.4**	**14.4**	**1.3**
		SD	**4.8**	**1.8**	**1.3**	**0.3**	**0.0**	**0.4**	**0.4**	**2.1**	**2.4**	**1.6**	**0.2**

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APPENDIX I. (Continued)

Sex	Specimen Number		SVL	HL	HW	EL	TD	FLL	HAL	TL	FL	FOL	IMT
SU	WIIAD 277		25.9	10.1	9.0	3.1	1.9	4.7	5.6	13.7	12.4	13.1	1.2
SU	WIIAD 274		25.8	10.5	9.2	3.0	2.0	5.3	5.9	13.4	11.2	12.7	1.7
		Mean	**25.9**	**10.3**	**9.1**	**3.1**	**2.0**	**5.0**	**5.8**	**13.5**	**11.8**	**12.9**	**1.4**
		SD	**0.1**	**0.3**	**0.1**	**0.1**	**0.1**	**0.5**	**0.2**	**0.3**	**0.8**	**0.3**	**0.3**
***Minervarya pierrei* (Dubois)**													
M	WIIAD 245		26.6	9.5	7.5	2.5	1.7	4.9	5.1	11.6	12.6	11.8	1.3
M	WIIAD 246		27.0	10.8	8.4	2.7	1.6	4.7	5.5	11.7	11.8	12.5	1.4
		Mean	**26.8**	**10.2**	**8.0**	**2.6**	**1.6**	**4.8**	**5.3**	**11.6**	**12.2**	**12.1**	**1.4**
		SD	**0.3**	**0.9**	**0.6**	**0.1**	**0.1**	**0.2**	**0.3**	**0.1**	**0.6**	**0.5**	**0.1**
F	WIIAD 059		33.0	13.3	10.0	4.1	2.0	6.0	6.7	16.4	14.4	14.8	1.7
F	WIIAD 067		33.6	12.4	11.4	3.1	2.1	6.5	7.4	16.1	14.9	16.8	1.3
		Mean	**33.3**	**12.8**	**10.7**	**3.6**	**2.0**	**6.3**	**7.1**	**16.2**	**14.6**	**15.8**	**1.5**
		SD	**0.4**	**0.6**	**1.0**	**0.7**	**0.1**	**0.4**	**0.5**	**0.2**	**0.4**	**1.4**	**0.3**
***Sphaerotheca pashchima* Padhye, Dahanukar, Sulakhe, Dandekar, Limaye, and Jamdade**													
M	WIIAD 064		41.4	15.4	15.0	5.0	3.5	9.0	9.4	16.9	16.1	14.8	4.6
M	WIIAD 065		32.1	12.7	12.2	4.1	2.7	8.1	7.3	14.5	12.8	12.2	3.5
M	WIIAD 229		45.5	15.7	16.8	5.6	3.4	10.9	8.9	16.8	18.6	16.1	4.5
M	WIIAD 233		43.2	15.4	17.5	5.2	3.7	9.5	8.5	17.5	17.8	15.8	4.7
M	WIIAD 275		35.5	11.5	14.3	5.0	2.9	8.2	7.8	15.1	15.7	13.6	4.2
		Mean	**39.5**	**14.1**	**15.1**	**5.0**	**3.2**	**9.1**	**8.4**	**16.2**	**16.2**	**14.5**	**4.3**
		SD	**5.6**	**1.9**	**2.1**	**0.6**	**0.4**	**1.2**	**0.8**	**1.3**	**2.2**	**1.6**	**0.5**
F	WIIAD 225		45.9	17.7	16.3	6.0	3.2	9.1	9.1	17.6	17.9	15.7	4.3
F	WIIAD 232		47.2	17.5	19.0	6.2	3.3	9.8	10.3	18.1	18.7	16.4	5.4
		Mean	**46.5**	**17.6**	**17.7**	**6.1**	**3.3**	**9.5**	**9.7**	**17.8**	**18.3**	**16.1**	**4.9**
		SD	**0.9**	**0.1**	**1.9**	**0.2**	**0.1**	**0.5**	**0.9**	**0.4**	**0.6**	**0.5**	**0.8**
***Hoplobatrachus crassus* (Jerdon)**													
M	WIIAD 252		70.2	27.7	23.9	6.8	4.9	13.1	13.3	31.1	30.6	25.3	5.7
SU	WIIAD 068		92.9	35.9	34.6	8.9	6.6	17.8	18.9	51.3	47.0	40.7	5.3
***Hoplobatrachus tigerinus* (Daudin)**													
M	WIIAD 228		154.0	60.0	58.5	12.4	11.5	29.9	30.0	75.5	77.6	58.6	8.7
***Microhyla nilphamariensis* Howlader, Nair, Gopalan, and Merilä**													
M	WIIAD 234		18.7	6.5	5.6	1.3		3.1	4.2	8.4	7.2	7.0	0.7
M	WIIAD 235		18.3	5.0	5.5	1.7		2.7	4.5	9.0	7.7	7.9	0.7
M	WIIAD 262		21.1	5.7	6.3	1.7		3.7	4.5	9.5	8.7	8.1	0.6
M	WIIAD 263		19.2	5.4	5.6	1.6		3.5	4.5	9.2	8.3	8.1	0.8
M	WIIAD 264		19.5	5.1	6.3	1.4		3.3	4,41	8.9	8.1	7.4	0.8
		Mean	**19.3**	**5.5**	**5.9**	**1.5**		**3.3**	**4.4**	**9.0**	**8.0**	**7.7**	**0.7**
		SD	**1.1**	**0.6**	**0.4**	**0.2**		**0.4**	**0.1**	**0.4**	**0.6**	**0.5**	**0.1**
***Uperodon globulosus* (Günther)**													

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APPENDIX I. (Continued)

Sex	Specimen Number		SVL	HL	HW	EL	TD	FLL	HAL	TL	FL	FOL	IMT
M	WIIAD 231		52.0	12.5	15.4	3.5		9.8	14.7	17.9	21.4	16.2	5.6
SU	WIIAD 069		36.9	9.1	13.3	3.3		7.9	8.8	11.2	12.0	9.9	3.6
***Uperodon systoma* (Schneider)**													
SU	WIIAD 223		46.5	16.7	15.6	4.1		11.2	13.1	17.5	20.2	14.7	4.1
SU	WIIAD 230		44.6	13.1	14.2	5.1		9.7	12.8	16.7	18.2	15.2	4.2
		Mean	**45.6**	**14.9**	**14.9**	**4.6**		**10.4**	**13.0**	**17.1**	**19.2**	**14.9**	**4.1**
		SD	**1.4**	**2.6**	**1.0**	**0.7**		**1.0**	**0.2**	**0.5**	**1.4**	**0.3**	**0.1**
***Uperodon variegatus* (Stoliczka)**													
F	WIIAD 291		31.4	8.9	9.4	2.5		6.0	6.6	9.1	9.6	9.0	1.0
***Polypedates maculatus* (Gray)**													
M	WIIAD 226		47.0	19.8	15.5	5.6	3.8	9.8	11.7	21.5	21.4	15.6	1.7
M	WIIAD 250		44.0	16.3	14.5	5.4	3.4	9.4	12.3	21.4	21.8	16.7	1.5
M	WIIAD 271		43.3	17.5	13.9	4.9	3.3	9.1	12.2	22.3	22.3	17.1	1.7
		Mean	**44.8**	**17.8**	**14.6**	**5.3**	**3.5**	**9.4**	**12.1**	**21.8**	**21.8**	**16.5**	**1.6**
		SD	**1.9**	**1.8**	**0.8**	**0.3**	**0.3**	**0.4**	**0.3**	**0.5**	**0.4**	**0.8**	**0.1**
F	WIIAD 251		60.3	22.6	20.4	5.7	4.4	13.5	15.9	26.8	27.4	21.2	2.1
SU	WIIAD 272		38.8	14.5	11.7	4.4	3.0	8.4	10.2	20.1	18.8	14.4	1.6
SU	WIIAD 273		40.0	16.6	13.3	4.6	2.3	8.0	11.4	21.6	21.0	16.3	1.5
SU	WIIAD 062		51.9	18.0	19.2	5.0	4.0	10.7	14.3	26.3	25.8	20.0	1.7
SU	WIIAD 063		42.5	15.9	13.4	4.9	2.9	9.3	11.2	21.3	19.9	15.2	1.4
		Mean	**43.3**	**16.2**	**14.4**	**4.7**	**3.0**	**9.1**	**11.8**	**22.3**	**21.4**	**16.5**	**1.5**
		SD	**6.0**	**1.4**	**3.3**	**0.3**	**0.7**	**1.2**	**1.8**	**2.7**	**3.1**	**2.4**	**0.1**

APPENDIX II. Numerical values of call parameters based on data determined from a sample of 370 calls from 37 individual males of 11 species where "n" denotes number of individual of each species. 10 calls per individual were measured (except 18 calls used for the call repetition rate and inter-call interval for *Euphlyctis cyanophlyctis* where n = 2).

	Mean	SD	Range (Min–Max)	CV %
***Duttaphrynus melanostictus* (n=1)**				
Call Duration (ms)	64.1	10.35	44–70	16.15
Pulses per call (k)	6.8	0.79	5–8	11.6
Call repetition rate (calls/min)	9.21	1.14	8.47–11.76	12.32
Inter-call interval (ms)	45.9	2.28	41–49	4.97
Pulse rate (pulses/s)	112.39	18.77	101.69–161.29	16.7
Dominant frequency (Hz)	1701.15	22.72	1679.6–1722.7	1.34
Pulse duration (ms)	9.9	2.28	6–12	23.06
***Duttaphrynus scaber* (n=3)**				
Call Duration (ms)	229.77	30.24	172–280	13.16
Pulses per call (k)	7.6	0.72	6–9	9.53
Call repetition rate (calls/min)	3.31	0.37	2.79–4	11.17

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APPENDIX II. (Continued)

	Mean	SD	Range (Min–Max)	CV %
Inter-call interval (ms)	75.73	6.54	64–86	8.64
Pulse rate (pulses/s)	31.76	1.89	28.69–34.48	5.95
Dominant frequency (Hz)	3514.2	201.26	3230–3746.8	5.73
Pulse duration (ms)	7.9	0.88	6–10	11.2
Duttaphrynus stomaticus **(n=3)**				
Call Duration (ms)	259.6	41.33	169–351	15.92
Pulses per call (k)	25.57	4.26	16–35	16.65
Call repetition rate (calls/min)	3.01	0.4	2.23–4.24	13.46
Inter-call interval (ms)	78.13	5.69	67–90	7.28
Pulse rate (pulses/s)	98.88	8.09	83.33–114.16	8.18
Dominant frequency (Hz)	2571.07	183.11	2411.70–2885.40	7.12
Pulse duration (ms)	7.87	1.31	5–11	16.6
Note duration (ms)	25.47	4.49	21–34	17.61
Euphlyctis cyanophlyctis **(n=2)**				
Call Duration (ms)	583.95	209.57	200–946	35.89
Pulses per call (k)	18.4	5.57	8–28	30.25
Call repetition rate (calls/min)	0.42	0.13	0.11–0.64	32.15
Inter-call interval (ms)	2260.56	1688.84	1011–8603	74.71
Pulse rate (pulses/s)	30.74	2.08	28.69–35.90	6.75
Dominant frequency (Hz)	1886.3	624.49	1507.30–3057.70	33.11
Pulse duration (ms)	6.05	0.22	6–7	3.7
Pulse type 2 duration (ms)	4.3	1.03	3–6	23.98
Fejervarya orissaensis **(n=5)**				
Call Duration (ms)	98.76	8.69	82–109	8.8
Pulses per call (k)	9.76	0.98	8–11	10.05
Call repetition rate (calls/min)	2.32	0.81	1.02–5.13	34.95
Inter-call interval (ms)	382.24	171.03	99–895	44.74
Pulse rate (pulses/s)	104.46	2.03	94.59–107.53	1.94
Dominant frequency (Hz)	2412.59	634.95	1162.80–2842.40	26.32
Pulse duration (ms)	9.62	0.49	9–10	5.1
Hoplobatrachus tigerinus **(n=5)**				
Call Duration (ms)	235.3	48.65	181–430	20.68
Pulses per call (k)	17.36	2.88	13–29	16.57
Call repetition rate (calls/min)	1.04	0.31	0.45–1.81	29.66
Inter-call interval (ms)	818.88	386.99	318–2009	47.26
Pulse rate (pulses/s)	76.92	5.32	61.82–87.18	6.92
Dominant frequency (Hz)	1819.14	388.87	1205.90–2239.50	21.38
Pulse duration (ms)	10.96	1.44	9–15	13.16
Minervarya caperata **(n=3)**				
Call Duration (ms)	130.6	25.49	101–200	19.52
Pulses per call (k)	14.63	2.74	12–22	18.69
Call repetition rate (calls/min)	5.32	0.79	2.54–6.21	14.77
Inter-call interval (ms)	63.63	26.47	30–198	41.6

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APPENDIX II. (Continued)

	Mean	SD	Range (Min–Max)	CV %
Pulse rate (pulses/s)	113.37	3.34	107.69–118.81	2.95
Dominant frequency (Hz)	2094.48	309.41	1938–3703.70	14.77
Pulse duration (ms)	9.13	0.35	9–10	3.79
***Minervarya pierrei* (n=5)**				
Call Duration (ms)	73.18	9.86	53–90	13.48
Pulses per call (k)	14.7	1.84	11–18	12.49
Call repetition rate (calls/min)	7.14	1.41	3.07–9.62	19.74
Inter-call interval (ms)	75	41.7	43–253	55.6
Pulse rate (pulses/s)	204.96	12.92	122.30–225.35	6.3
Dominant frequency (Hz)	3714.05	67.58	3574.50–3832.90	1.82
Pulse duration (ms)	4.88	0.32	4–5	6.66
***Sphaerotheca pashchima* (n=3)**				
Call Duration (ms)	377.6	45.65	319–455	12.09
Pulses per call (k)	29.23	2.87	24–35	9.83
Call repetition rate (calls/min)	1.37	0.34	0.75–1.86	25.08
Inter-call interval (ms)	404.23	182.42	184–897	45.13
Pulse rate (pulses/s)	78.35	3.03	72.43–83.08	3.86
Dominant frequency (Hz)	2074.37	441.68	947.50–2368.70	21.29
Pulse duration (ms)	12.13	0.78	11–14	6.4
***Microhyla nilphamariensis* (n=5)**				
Call Duration (ms)	415.22	130.31	195–625	31.38
Pulses per call (k)	18.36	5.82	8–26	31.72
Call repetition rate (calls/min)	0.78	0.16	0.43–1.04	20.82
Inter-call interval (ms)	931.58	330.8	563–2030	35.51
Pulse rate (pulses/s)	44.04	3.4	38.17–50.76	7.72
Dominant frequency (Hz)	3067.18	104.52	2928.50–3273	3.41
Pulse duration (ms)	7.6	1.51	6–11	19.89
***Polypedates maculatus* (n=2)**				
Call Duration (ms)	132.25	25.54	82–171	19.31
Pulses per call (k)	5.75	1.12	4–9	19.44
Call repetition rate (calls/min)	1.01	0.75	0.17–2.37	74.13
Inter-call interval (ms)	1660.55	1399.24	280–5860	84.26
Pulse rate (pulses/s)	60	5.9	50–74.63	9.83
Dominant frequency (Hz)	960.4	125.06	818.3–1335.10	13.02
Pulse duration (ms)	4.9	1.21	3–7	24.69

APPENDIX III. List of DNA sequences included in the study

Species	Collection locality	Voucher number	Accession number	Reference
Sphaerotheca pashchima	PTR	WIIADA225 *S. pashchima*	MN741157	This study
Sphaerotheca pashchima	PTR	WIIADA232 *S. pashchima*	MN741158	This study
Sphaerotheca pashchima	Western India	ZSI-WRC A/1549	KY215980	Padhye *et al*. (2017)
Sphaerotheca pashchima	Western India	WILD-16-AMP-644	KY215994	Padhye *et al*. (2017)
Sphaerotheca pashchima	Western India	BNHS 6018	KY215988	Padhye *et al*. (2017)
Sphaerotheca pashchima	Western India	BNHS 6015	KY215985	Padhye *et al*. (2017)
Sphaerotheca pashchima	Western India	BNHS 6011	KY215979	Padhye *et al*. (2017)
Sphaerotheca Magadha	Koderma	ZSI/WRC/2179	MK694738	Prasad et al. (2019)
Sphaerotheca breviceps	Western India	BNHS 6005	KY215977	Padhye *et al*. (2017)
Sphaerotheca breviceps	Western India	WILD-16-AMP-645	KY215978	Padhye *et al*. (2017)
Sphaerotheca dobsonii	Western India	INHER Amphibia-86	KY215970	Padhye *et al*. (2017)
Sphaerotheca dobsonii	Bajipe, India	-	AB277305	Kotaki *et al.* (2008)
Sphaerotheca dobsonii	Western India	BNHS 6007	KY215969	Padhye *et al*. (2017)
Sphaerotheca pluvialis	-	-	AF249042	Bossuyt & Milinkovitch (2000)
Sphaerotheca pluvialis	-	-	AF215418	Vences (1999)
Minervarya caperata	PTR	WIIAD256 *M. caperata*	MN741153	This study
Minervarya caperata	PTR	MIN *M. caperata*	MN741154	This study
Minervarya caperata	Western India	BNHS_4659	AB355844	Kuramoto *et al*. (2007)
Fejervarya orissaensis	Orissa	haplotype_16S-ori	AB277304	Kotaki *et al.* (2008)
Fejervarya orissaensis	PTR	WIIAD227 *F. orissaensis*	MN741152	This study
Minervarya pierrei	Dhaka City	isolate_F1DSE_14	KP849816	Howlader *et al*. (2016)
Minervarya pierrei	PTR	WIIAD67 *M. pierrei*	MN741155	This study
Minervarya pierrei	PTR	MIN2 *M. pierrei*	MN741156	This study
Minervarya pierrei	Nepal_Chitwan	-	AB488888	Kotaki *et al.* (2008)
Fejervarya multistriata	Japan	Mult-Tai-1	AB354241	Djong *et al*. (2011)
Fejervarya multistriata	China_Husa	-	AB488884	Kotaki *et al.* (2008)
Fejervarya moodiei	Bangladesh	Fmod-Bd1	AB530508	Hasan *et. al*. (2012)
Minervarya granosa	India Mudigere	-	AB488895	Kotaki *et al*. (2010)
Minervarya granosa	BR Hills, Karnataka	SDBDU_2014.2541	KY820766	Garg & Biju (2017)
Minervarya asmati	Dhaka, Bangladesh		KP849815	Howlader *et al*. (2016)
Minervarya cepfi	Amboli, Maharashtra	ZSI/WGRC/V/A/938	KY447308	Garg & Biju (2017)
Minervarya gomantaki	Western Ghats, India	CES:F2289	KR781084	Dinesh *et al.* (2015)
Minervarya kadar	Thavalakuzhipara, Kerala	ZSI/WGRC/V/A/940	KY447312	Garg & Biju (2017)
Minervarya kalinga	Penninsular India	-	MG870107	Raj *et al.* (2018)
Minervarya keralensis	Karian Shola, Western Ghats	isolate_B-441	JX573181	Raj *et al.* (2018)
Minervarya krishnan	Western Ghats	-	MG870108	Raj *et al.* (2018)
Minervarya kudremukhensis	Western Ghats, India	BNHS_4656	AB355841	Kuramoto *et al*. (2007)
Minervarya manoharani	Chathankod-Bonnacad, Kerala	ZSI/WGRC/V/A/950	KY447315	Garg & Biju (2017)
Minervarya marathi	Western Ghats, India	-	MH370483	Phuge *et al.* (2019)
Minervarya mudduraja	Western Ghats, India	BNHS_4648	AB355835	Kuramoto *et al*. (2007)
Minervarya neilcoxi	Parambikulam, Kerala	ZSI/WGRC/V/A/951	KY447318	Garg & Biju (2017)
Minervarya rufescens	Kollur, Mookambika WLS, Karnataka	SDBDU_2009.4712	KY447320	Garg & Biju (2017)